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Human Health Risk Assessment for Biodiesel Production, Distribution and Use in Canada

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2-EHN	2-ethyl hexyl nitrate
AAFC	Agriculture and Agri-Food Canada
AARI	Alberta Agricultural Research Institute
ACC	American Chemistry Council
AM	Alveolar Macrophages
APTT	Activated partial thromboplastin time
AQBAT	Air Quality Benefits Assessment Tool
AQMAS	Air Quality Modelling Applications Section
ARDD	Alberta Renewable Diesel Demonstration
ARS	Agricultural Research Service
ASTM	American Society for Testing and Materials
ATSDR	Agency for Toxic Substances and Disease Registry
AURAMS	A Unified Regional Air quality Modelling System
BAL	Bronchoalveolar lavage
BALF	Bronchoalveolar lavage fluid
BP	Blood pressure
BSE	Bovine Spongiform Encephalopathy
BTEX	Benzene, Toluene, Ethylbenzene, Xylenes
B*X*	Biodiesel blends where *X* indicates the per cent content per volume of biodiesel
CAC	Criteria Air Contaminant
Cal-EPA	California Environmental Protection Agency
CARB	California Air Resources Board
CaME	Canola oil methyl ester
CCME	Canadian Council of Ministers for the Environment
CD	Census Division
CEPA	Canadian Environmental Protection Act
CFIA	Canadian Food Inspection Agency
CFR	Code of Federal Regulations
CFSPH	Center for Food Security and Public Health
CGSB	Canadian General Standards Board
CHD	Coronary Heart Disease
CI	Compression ignition
CJD	Creutzfeldt-Jacob disease
CMIT	5-Choro-2-methyl-3(2H)-isothiazolone
CO	Carbon monoxide
CO_2	Carbon dioxide
CONCAWE	Conservation of Clean Air and Water in Europe
CRF	Concentration Response Function

CWS	Canada Wide Standard
C*AA*:*B*	Hydrocarbons where *AA* indicates the number of C in the chain and *B* the number of double bounds
DA	Dissemination Area
DCM	Dichloromethane
DE	Diesel exhaust
DEP	Diesel Exhaust Particulates
DF	Diesel fuel
DMBA	Dimethylbenzanthracene
DMSO	Dimethyl sulphoxide
DNA	Deoxyribonucleic acid
DOC	Diesel Oxidation Catalyst
DPF	Diesel Particulate Filter
DSL	Domestic Substances List
DTT	Dithiothreitol
EC	Elemental carbon
ECE	Economic Commission of Europe
ECE-EUDC	Economic Commission of Europe Europe-Exhaustion Driving Cycle
EFSA	European Food Safety Authority
EGR	Exhaust Gas Recirculation
ERMS	Emissions Research and Measurement Section
FAAE	Fatty Acid Alkyl Ester
FAME	Fatty Acid Methyl Ester
FiME	Fish oil methyl ester
FrAME	Animal frying oil methyl ester
FTA	Federal Transit Administration
FTP-75	Federal Test Procedure City Test
GEM	Global Environmental Multiscale model
GFAP	Glial fibrillary acidic protein
GHG	Greenhouse gas
GREET	Greenhouse Gases, Regulated Emissions, and Energy Use in Transportation model
GTL	Gas to liquids
GVWR	Gross vehicle weight rating
HC	Hydrocarbon
HCTPCT	Haematocrit percentage
HDDV	Heavy-duty diesel vehicle
HDGV	Heavy-duty gasoline vehicle
HDV	Heavy-duty vehicle
HEI	Health Effects Institute

HF	High frequency
Hb	Haemoglobin
%ΔHI	Percent change in health impact due to biodiesel use relative to health impact due to above-background base-case air pollutant concentration
HI ABBC	Health impact due to above-background base-case concentration
ΔHI BD	Incremental health impact due to biodiesel
HR	Heart rate
HRV	Heart rate variability
HSSM	Hydrocarbon Spill Screening Model
HVO	Hydrotreated vegetable oil
IARC	International Agency for Research on Cancer
IL	Interleukin
KOH	Potassium hydroxide
kPa	Kilopascal
LDDV	Light-duty diesel vehicle
LDDT	Light-duty diesel truck
LDGT	Light-duty gasoline truck
LDGT	Light-duty gasoline vehicle
LDH	Lactate dehydrogenase
LDV	Light Duty Vehicle
LED	Low emission diesel
LF	Low frequency
LF/HF	Low frequency/high frequency ratio
LNAPL	Light NAPL
LOAEL	Lowest observable adverse effect level
LSD	Low sulphur diesel
LS-DF	Low sulphur diesel fuel
MBM	Meat and bone meal
MC	Motorcycle
MCH	Mean corpuscular haemoglobin
MCHC	Mean corpuscular haemoglobin concentration
MCV	Mean corpuscular volume
MIP	Macrophage inflammatory protein
MIT	2-Methyl-3(2H)-isothiazolone
MK 1	Swedish low-sulphur fuel
Mn	Manganese
MPa	Megapascal
MTT	3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide
Na^+	Sodium ion
NAC	NO_X adsorber catalyst

NaOH	Sodium hydroxide
NAPL	Non-aqueous phase liquid
NBB	National Biodiesel Board
ND	Non detected
NEDC	New European Driving Cycle
NH_4^+	Ammonium ion
NMOC	Non-methane organic compound
NO_2	Nitrogen dioxide
NO_3^-	Nitrate ion
NOAEL	No observable adverse effect level
NO_X	Nitrogen oxides
N-PAH	Nitro-polycyclic aromatic hydrocarbons
NPRI	National Pollutant Release Inventory
NREL	National Renewable Energy Laboratory
O_2	Oxygen
O_3	Ozone
OC	Organic carbon
OIE	The World Organisation for Animal Health
PAH	Polycyclic aromatic hydrocarbon
PCPA	Pest Control Products Act
PDD	Pollution Data Division of Environment Canada
PHC	Petroleum hydrocarbon
PM	Particulate Matter
$PM_{2.5}$	Particulate Matter with a maximum aerodynamic diameter of 2.5µm
PM_{10}	Particulate Matter with a maximum aerodynamic diameter of 10µm
PMN	Polymorphonuclear neutrophil
PMRA	Pest Management Regulatory Agency
POC	Particle oxidation catalyst
POM	Polycyclic organic matter
PPO	Pure plant oil
PPM	Parts per million
PrP	Prion proteins
PrP^C	Prion proteins – cellular
PrP^{Sc}	Prion proteins – scrapie
PT	Prothrombin time
RBC	Red blood cell
REE	Rapeseed ethyl ester
RFS2	US Renewable Fuels Standard Program
RIA	Regulatory Impact Assessment
RME	Rapeseed Methyl Ester

RMSSD	Root mean square of SDNN
RNA	Ribonucleic acid
ROS	Reactive oxygen species
RSO	Rapeseed oil
SAE	Society of Automobile Engineers
SCR	Selective Catalytic Reducer
SDNN	Standard deviation of normal beats
SEE	Soy ethyl ester
SME	Soy methyl ester
SMOKE	Sparse Matrix Operator Kernel Emissions processing system
SO_2	Sulphur dioxide
SO_4	Sulphate
SOF	Soluble organic fraction
SRM 1975	Standard reference material
SRM	Specified risk material
sULSD	seasonal ULSD
TBHQ	*tert*-Butylhydroquinone
THC	Total hydrocarbon
TNF-α	Tumour necrosis factor-α
TPH	Total Petroleum Hydrocarbon
TPM	Total particulate matter
TSE	Transmissible spongiform encephalopathy
TT	Thrombin time
TUV	Toxicity unit per litre exhaust sampled
TUW	Toxicity unite per µg soluble fraction of particulate
UFP	Ultrafine particles
ULSD	Ultra Low Sulphur Diesel
US06	Supplemental Federal Test Procedure
USDA	United States Department of Agriculture
US EPA	United States Environmental Protection Agency
US FTP	United States Federal Test Procedure
UST	Underground storage tank
vCJD	Variant Creutzfeldt-Jacob disease
VIN	Vehicle identification number
VKT	Vehicle kilometres traveled
VOC	Volatile organic compound
VOF	Volatile organic fraction
WHO	World Health Organization

Executive Summary

Health Canada assessed the potential human health implications of the widespread use of biodiesel in Canada, considering the production, distribution, storage and use stages in the lifecycle of biodiesel fuel. The general approach employed is as comprehensive as the available information allows and is comparative in nature, i.e., the impacts of biodiesel blends are compared to those of conventional ultra low sulphur diesel (ULSD) and presented as relative risks and benefits. The primary consideration of this analysis is the potential impact of biodiesel use on mobile sector emissions and atmospheric concentrations of air pollutants.

The Government of Canada put in place a 2% renewable content requirement in diesel and heating oil on July 1st, 2011, as outlined in the *Regulations Amending the Renewable Fuels Regulations* (P.C. 2011-795 June 29, 2011) and published in the *Canada Gazette*, Part II,[1] on July 20, 2011. The Regulation does not specify the use of biodiesel fuel in distillate or heating fuels. Rather, any liquid fuel meeting the definition of renewable fuel as per the Regulation, produced from one or more of the designated feedstocks, and complying with the maximum content specified may be acceptable.[2]

Biodiesel is a mixture of fatty acid alkyl esters produced from vegetable oils and animal fats via transesterification with an alcohol (generally methanol). The combinations of fatty acids in fats and oils can vary substantially depending on the source material and influence the resulting biodiesel properties. Biodiesel is usually blended with ULSD and the resulting blends are denoted by B*X*, where *X* indicates the percent of biodiesel in a blend, on a volume basis (e.g., B5 is a blend comprised of 5% by volume biodiesel and 95% by volume ULSD). Biodiesel blends up to B20 can generally be used in most compression ignition engines without any modifications.

Biodiesel production facilities rely on technologies and processes that vary according to the type of feedstock and the level of integration or complexity of a facility. Biodiesel production activities can lead to a variety of emissions or releases to water, air, and soil. Risks and hazards associated with biodiesel facilities are common to other industrial sectors (e.g., combustion emissions, fugitive emissions, and spills) and can be limited by mitigation strategies, both behavioural and technological. The most common air emissions from biodiesel production plants are methanol (during transesterification), hexane (during oil extraction), and criteria air contaminants (CACs) (e.g., particulate matter (PM) emissions from fuel-powered generators). Total amounts emitted are expected to be relatively low and to meet regulatory requirements,

[1] Vol. 145, No. 15 - July 20, 2011. Available at http://canadagazette.gc.ca/rp-pr/p2/2011/2011-07-20/html/sor-dors143-eng.html

[2] See the definitions of *renewable fuel* and *renewable fuel feedstock* in the Renewable Fuels Regulations at http://canadagazette.gc.ca/rp-pr/p1/2010/2010-04-10/html/reg1-eng.html

based on information from the National Pollutant Release Inventory[3] and environmental assessments of various Canadian biodiesel production facilities. Ambient air concentrations near facilities are expected to meet air quality guidelines. Emissions of heavy metals and air toxics are generally expected to be minimal, as specific activities that would lead to significant emissions of these pollutants have not been identified.

No extensive database or tool has been specifically developed to predict the environmental fate and transport of biodiesel releases, and empirical values for numerous physical and chemical properties of biodiesel fuel components are not available. Health Canada conducted screening level environmental fate and transport modelling of different biodiesel fuel spill scenarios to identify key potential impacts. The modelling results of neat ULSD, neat biodiesel, and biodiesel blends show that biodiesel fuel components are projected to travel less than ULSD fuel components, as expected based on biodiesel's physical and chemical characteristics, notably the greater biodegradation rate of biodiesel fuel components compared to diesel fuel fractions. Notwithstanding the modelling uncertainties (i.e., assumptions and input data), the limited mobility of biodiesel fuel components can be considered a benefit since the soil and groundwater contamination is expected to be relatively contained and, consequently, have less impact on environmental and human health than petroleum fuels following releases.

With regards to the use of biodiesel fuels in on-road heavy-duty diesel vehicles (HDDVs) and its impact on exhaust emissions compared to ULSD, the following trends are noted:

- considerable reductions in PM, CO, hydrocarbon, volatile organic compound, and polycyclic aromatic hydrocarbon emissions;
- no net impact or a slight increase in NO_X emissions; and
- no significant impact on the efficiency of after-treatment devices.

For the current assessment, the impacts of biodiesel use on Canadian fleet-wide mobile source emissions were estimated with the MOBILE6.2C model in collaboration with Environment Canada. It was assumed that biodiesel affects emissions of on-road HDDVs only. Table ES.1 shows the percent change in fleet-average HDDV emissions estimated for B5 and B20 in comparison to ULSD, for 2006, 2010, and 2020.

Biodiesel is projected to have less impact on HDDV exhaust emissions in 2020 due to the turn-over of the Canadian HDDV fleet, as 2010 and beyond model-year HDDVs are equipped with new engine technologies and exhaust emission controls in order to meet more stringent emission standards.

[3] http://www.ec.gc.ca/inrp-npri/Default.asp?lang=En&n=B85A1846-1

Table ES-1 Percent change in Canadian fleet-average HDDV emissions from MOBILE6.2C for B5 and B20 compared to ULSD for 2006, 2010 and 2020

Pollutants	B5			B20		
	2006	2010	2020	2006	2010	2020
1,3-Butadiene/Acetaldehyde/ Acrolein/Formaldehyde	-4	-3	-1	-18	-14	-3
Benzo[a]pyrene	-3	-3	-2	-14	-12	-9
Benzene	-4	-3	-1	-18	-14	-4
CO	-3	-3	-2	-11	-10	-7
Elemental carbon/ Organic carbon	-3	-3	-2	-13	-12	-9
NO_X	1	1	1	4	4	3
$PM_{10\ exhaust}/PM_{2.5\ exhaust}$	-3	-3	-2	-13	-12	-9
SO_2/ NH_3	0	0	0	0	0	0
Toluene/Ethylbenzene/Xylene	-4	-3	-1	-18	-14	-3
VOC_{total}/ THC_{total}	-4	-3	-1	-18	-14	-3

Air quality modelling was undertaken to investigate the impact of biodiesel blends on air pollution in Canada. Specifically, MOBILE6.2C results of Canadian mobile source emissions for the basecase and biodiesel scenarios were used as input to the air quality modelling.[4] Photochemical modelling for the current project was conducted with *A Unified Regional Air quality Modelling System* (AURAMS) in collaboration with Environment Canada (see Table ES.2 for scenarios). National scenarios of biodiesel use were modelled on a 22.5-km grid covering the whole country. In addition, a 2-week high pollution episode was modelled at high resolution (3-km grid) over the Montréal region.

Table ES-2 Timeframes of photochemical modelling (AURAMS) scenarios

Years	Fuel	Canada (22.5-km grid)	Montréal (3-km grid)
2006 and 2020	B0 (ULSD)	Annual	June 12 to 23
	B5	Annual	June 12 to 23
	B20*	Annual	June 12 to 23
* National B20 scenarios include the assumption that B20 is used during the summer months only (May-September, inclusive) and B0 is used during the winter months (October to April, inclusive) due to technical requirements.			

It is predicted that the national use of B5 and B20 under 2006 conditions would result in small (less than 1%) but non-negligible changes in air quality compared to ULSD use. In general,

[4] Upstream emissions associated with biodiesel fuel (i.e., fuel production, transportation, and distribution) are not considered in the emissions inventory and air quality modelling.

$PM_{2.5}$ and O_3 concentrations decrease in urban areas and increase in surrounding areas. CO concentrations are expected to decrease in all regions. For the 2020 projections, changes in predicted air quality are very small (less than 0.5%) and often close to model detection limits. Ozone and $PM_{2.5}$ concentrations are generally reduced in urban centres, but increase slightly in surrounding areas. CO concentrations are reduced in most areas. The smaller air quality impacts of biodiesel use in 2020 are due to the significant reductions in basecase fleet emissions in 2020 compared to 2006, as a result of the introduction of cleaner vehicles.

Short-term high resolution modelling of the Montréal urban area revealed similarly small changes in air quality. High-resolution modelling provided enhanced spatial resolution of air quality impacts, bringing to light different air quality phenomena caused by smaller scale meteorological regimes and a more detailed distribution of mobile emission sources, such as the impacts of major bridges and highways.

Health Canada's Air Quality Benefits Assessment Tool (AQBAT) was used to quantify Canadian morbidity and mortality risks/benefits from changes in CAC concentrations associated with the use of B5 or B20 compared to ULSD in the on-road HDDV fleet, in either 2006 or 2020. In 2006, annual B5 or summertime B20 use are associated with a reduction of about five to seven premature mortalities as well as minimal reductions in hospital admissions, emergency room visits and other morbidity outcomes, due primarily to minor reductions in $PM_{2.5}$ and O_3 levels. The health benefits associated with biodiesel use are expected to be reduced by 2020 due to the incorporation of new emission control technologies in the HDDV fleet.

Qualitative consideration of the available mobile source air toxics emissions data indicate that minimal reductions are expected for air concentrations of benzene, 1,3-butadiene, acetaldehyde, formaldehyde, acrolein and PAHs in association with the use of biodiesel, which may translate into very minor reductions in human exposure to these pollutants, particularly near roads that are heavily trafficked with HDDVs. However, the emissions benefits and any associated reductions in human exposures are expected to diminish by 2020.

A toxicological review of biodiesel exhaust was conducted with two objectives: to determine if biodiesel exhaust has a similar, reduced or greater impact than diesel exhaust in terms of specific health effects; and to attribute any difference in the magnitude of effects observed (between biodiesel and diesel exhaust) to a change in the level of a specific physicochemical parameter(s) in the exhaust.

A review of several studies determined that biodiesel exhaust is unlikely to exceed diesel exhaust in terms of respiratory effects. Only two studies were reviewed that examined cardiovascular effects of biodiesel exhaust. Based on this limited data set, it was not possible to draw any

conclusions as to how biodiesel and diesel exhaust compare with respect to cardiovascular effects.

A review of outcomes relevant to the initiation of carcinogenesis indicated that biodiesel and diesel exhaust are similar in terms of clastogenicity, biodiesel exhaust has a similar or lower effect on biochemical events (reactive oxygen species, inflammation) associated with genetic instability, and biodiesel is equal to or exceeds diesel with respect to cytotoxicity. The majority of studies investigating mutagenicity demonstrated that PM extract from biodiesel exhaust is potentially less mutagenic than diesel exhaust PM extract.

Only one inhalation study considered reproductive and developmental effects, neurological effects, and systemic effects resulting from exposure to biodiesel exhaust. Given that this study did not include a diesel treatment, it was not possible to draw any comparison between biodiesel and diesel exhaust. Dermal exposure to biodiesel was also considered because of potential exposure during refuelling. However, skin irritation, a potential outcome of this type of exposure, was not considered in the study reviewed.

No information was available for immunological effects resulting from exposure to biodiesel exhaust.

Regarding the second objective, it was determined that toxicological studies investigating respiratory, cardiovascular, and outcomes associated with initiation of carcinogenesis increasingly reflect efforts to ascribe differences in biological responses between biodiesel and diesel exhaust to differences in physicochemical characteristics between the two fuels. However, for most studies, differences in individual pollutant levels between biodiesel and diesel exhaust have not been specifically linked to changes in a given biological response.

A review was conducted to examine the risk that inhalation exposure of the Bovine Spongiform Encephalopathy (BSE) infectious agent may occur in the general population as a result of the combustion of biodiesel made from Specified Risk Material (SRM) derived tallow. The risk was considered negligible provided that SRM and tallow destined for biodiesel production are processed to achieve a tallow purity standard of not more than 0.15% insoluble impurity content, as per Canadian Food Inspection Agency directives. In a second scenario, in which the insoluble content of the SRM-derived tallow exceeds 0.15% and contains BSE agents, it is expected that biodiesel manufacturing and combustion processes would contribute to a reduction in the risk of inhalation exposure to BSE agents.

The potential for allergic reactions in the general population following inhalation exposure to exhaust from soy-based biodiesel was investigated due to the fact that soy is one the main foods eliciting allergic reactions. It was concluded that denaturation and hydrolysis of proteins during biodiesel production as well as purification processes are likely to reduce the allergenicity of

biodiesel. In the event that allergenic proteins survive the latter processes, it is highly probable they would be destroyed during the combustion process given that temperatures in diesel engines are significantly higher than those which cause significant alterations in protein structure, thus eliminating the potential for allergic reactions.

A review of the major fuel additive categories that are likely to be used in biodiesel fuels in Canada was carried out. The review included key background and toxicity information for different types of additives as well as specific products. There is a relatively high level of uncertainty associated with additives due to the fact that it is difficult to predict which products will be used on a consistent basis in biodiesel blends and because there is relatively limited toxicological and exposure information available for these compounds.

Overall Conclusion

Although the scenarios examined in this assessment do not replicate specific existing Canadian biodiesel use policies, they were selected in order to provide an overall picture of potential health impacts of biodiesel use in Canada. Overall, the use of B5 or B20 nationally is expected to result in very minimal air quality and health benefits/risks, and these are likely to diminish over time. Although substantial modelling and data limitations remain, the currently available evidence suggests that the incremental health impacts associated with the widespread use of low level biodiesel blends in Canada as compared to the use of ULSD are expected to be minimal.

Chapter 1. Biodiesel Fuel in Canada: Background and Scope of the Risk Assessment

Diesel-like fuels from biomass-derived feedstocks have been produced and used for more than a century (Knothe 2005; Mittelbach and Remschmidt 2004). Biodiesel, a mixture of fatty acid alkyl esters (FAAEs), is produced from vegetable oils and animal fats (composed of triacylglycerides), via transesterification. This chemical reaction leads to a fuel with properties similar to those of petroleum diesel that can be used in most compression ignition engines without any modifications. Because methanol is the most common alcohol used in this process, biodiesel fuel is often referred to as a fatty acid methyl ester (FAME). The combinations of fatty acids in fats and oils can vary substantially depending on the source material and influence the resulting biodiesel properties.

Biodiesel fuels are currently produced in many countries from a variety of feedstocks. They are usually used in the form of blends of 5% to 20% by volume with conventional diesel fuel. Blends containing biodiesel are generally denoted by B*X*, where *X* indicates the percent of biodiesel in a blend, on a volume basis (e.g., B5 is a blend comprised of 5% by volume biodiesel and 95% by volume petroleum diesel).

Straight vegetable oil and animal fats used directly as fuels are not considered biodiesel, nor are fuels from animal or vegetable feedstocks produced via processes other than transesterification (e.g., hydrotreated diesel[5]) (Knothe 2005).

1.1 Biodiesel Fuel Regulations

The Government of Canada put in place a 2% renewable content requirement in diesel and heating oil on July 1st, 2011, as outlined in the *Regulations Amending the Renewable Fuels Regulations* (P.C. 2011-795 June 29, 2011) and published in the *Canada Gazette*, Part II,[6] on July 20, 2011. The Regulation does not specify the use of biodiesel fuel in distillate or heating fuels. Rather, any liquid fuel meeting the definition of renewable fuel as per the Regulation, produced from one or more of the designated feedstocks, and complying with the maximum content specified may be acceptable.[7]

The national 2% requirement adopted in 2011 will create a demand for up to 650 million litres of renewable diesel fuel (MJ Ervin and Associates 2008; NRCan 2007).[8]

[5] Hydrotreated vegetable oil is renewable diesel produced via hydrotreatment and isomerization processes. This renewable diesel is indistinguishable from diesel derived from fossil fuels.

[6] Vol. 145, No. 15 - July 20, 2011. Available at http://canadagazette.gc.ca/rp-pr/p2/2011/2011-07-20/html/sor-dors143-eng.html

[7] See the definitions of *renewable fuel* and *renewable fuel feedstock* in the Renewable Fuels Regulations at http://canadagazette.gc.ca/rp-pr/p1/2010/2010-04-10/html/reg1-eng.html

[8] Estimated demand volume varies according to reference year.

1.2 Biodiesel Feedstocks

The biodiesel industry in Canada relies on various feedstock suppliers, such as farmers and renderers. Canadian biodiesel production is based on three major feedstocks: virgin vegetable oils, animal tallow, and recycled frying oils or yellow grease. Feedstock selection is important for economic, environmental, and technical reasons. It is also highly dependant on the regional or national availability of oils and fats.

Tallow and yellow grease are low-value or waste products from other activities. These feedstocks are relatively economical and distributed in rural and urban areas. As they are characterized by relatively finite volumes, growth in the availability of these feedstocks is expected to be limited.

Virgin vegetable oils originate from 'new' feedstocks, some of which can be intentionally grown for biofuel production. Canola is the most favourable oilseed crop for biodiesel production in Canada, with canola seeds containing around 44%[9] oil. The remainder of the canola seed can be processed into a high protein meal mainly for use as livestock feed.

Presently, the majority of biodiesel production is based on tallow and recycled grease and there is only limited domestic production based on canola and soy oils. Estimates for 2009 show that 49% of the biodiesel production capacity was tallow-based (up from 36% in 2008), 37% was yellow grease-based (down from 59% in 2008), and 14% was canola-based (up from 6% in 2008) (GAIN 2009).

1.3 Biodiesel Demand and Production

The Canadian biodiesel fuel market is currently hindered by various factors, mainly the limited demand for diesel fuels due to the small number of light-duty diesel vehicles (e.g., passenger cars, light-duty trucks) and low market penetration beyond a normal trucking range from the production facilities (PPD Technologies Inc. 2008). However, the federal requirement of 2% renewable content in diesel fuel and heating oil in 2011, amounting to 650 million litres, and the construction of large capacity biodiesel plants will likely alter some characteristics of the Canadian market to meet demand.

Biodiesel production facilities are generally distributed from British Columbia to Québec. Canola oil is assumed to be the primary future feedstock in Canada due to the abundance of production in western provinces, while eastern provinces are expected to rely on yellow grease and recycled cooking oil for about half of their biodiesel production.[10] According to the Canadian

[9] http://www.grainscanada.gc.ca/canola/harvest-recolte/2010/hqc10-qrc10-6-eng.htm (accessed March 14, 2011)
[10] Canada Gazette Part II, Vol. 145, No. 15 – July 20, 2011. [http://canadagazette.gc.ca/rp-pr/p2/2011/2011-07-20/html/sor-dors143-eng.html].

Renewable Fuels Association, approximately thirteen biodiesel facilities are currently in operation in Canada.[11]

Industry information indicates that around 500 million litres of biodiesel could be produced from cheaper feedstocks (e.g. tallow, yellow grease) (GAIN 2009). Hence, tallow and yellow grease feedstocks could potentially be sufficient to meet most of the demand resulting from the *Renewable Fuel Regulations* (i.e., 650 million litres). A survey of existing and planned biodiesel production facilities showed that biodiesel production capacity in Canada could, in the near future, be substantially above the volume necessary to meet the federal 2% renewable content requirement in diesel and heating oil fuel. If low-cost feedstocks like tallow and yellow grease are available for biodiesel production, then virgin vegetable oils, such as canola oil, could be relatively expensive for use in biofuel production within and beyond the regulatory requirements. This also underlines the issues of feedstock availability and prices, and access to competitive international biodiesel markets.

Despite the Government of Canada's requirement, the growth in biodiesel production has not increased significantly until very recently. As such, it is anticipated that the federal requirement is unlikely to be met entirely by domestic biodiesel production (GAIN 2009), at least in the early period of the national mandate. Concurrently, Natural Resources Canada (NRCan) estimated that in 2011 about 70% of biodiesel demand due to the federal requirement was met by domestic production and the remaining 30% by imports from the US. In 2012, it is assumed that 90% of incremental biodiesel demand will be met by Canadian products and 10% by imports of renewable diesel (i.e., hydrotreated oils and fats).[12]

1.4 Biodiesel Distribution and Fuel Specifications

Regarding storage, blending, and distribution of biodiesel fuel, fuel stability and quality are important factors. Fuel properties must remain stable throughout the storage and distribution stages and always meet industry specifications to allow for the proper operation of equipment. Biodiesel stability is generally dependant on environmental conditions, the fatty acid composition of the feedstock oil, the age of the biodiesel fuel, the presence of additives, and the quality of the base petroleum diesel fuel (Karavalakis et al. 2010). Fuel additives range from oxidative stabilizers to biocides and appear to provide benefits during fuel storage by inhibiting chemical degradation of the fuel (e.g., the formation of acids, oxidation) or by inhibiting the growth of micro-organisms. It has also been observed that lower blends, such as B5 and B20, have better storage stability than B100 (McCormick and Westbrook 2010).

[11] http://www.greenfuels.org/en/industry-information/plants.aspx (accessed on December 16, 2011)

[12] Canada Gazette Part II, Vol. 145, No. 15 – July 20, 2011. [http://canadagazette.gc.ca/rp-pr/p2/2011/2011-07-20/html/sor-dors143-eng.html].

Storing neat biodiesel at primary terminals[13] is considered the most practical approach and a good way to ensure maximum quality of the fuel. With this option, stages downstream of a primary terminal are not required to handle neat biodiesel fuels, but only specific blends. Another possibility is for B100 to be stored at secondary terminals. Biodiesel would thus be blended just prior to distribution to retail outlets (NRCan 2007).

Various options are available for blending biodiesel with diesel fuel. These generally vary according to market demands, environmental conditions (e.g., temperature), quality standards, and industry best practices. Ideally, B100 would be blended by in-line injection[14] at primary terminals and transported downstream to retail outlets, clients, or secondary terminals in blended form, mostly by truck (NRCan 2007).

Aside from the in-line injection blending at primary terminals, splash-blending[15] at primary or secondary terminals is also possible. Sequential blending is a variation of splash blending that relies on metered pumps to accurately determine the amount of fuels required for specific blends (NRCan 2007). Another option is in-tank blending.[16] Although less costly and requiring less infrastructure compared to in-line injection, these blending procedures are less precise and could cause improper blending, thus leading to irregular fuel properties between batches.

Another challenge for biodiesel is that it is currently transported by truck and rail, which makes transportation and distribution of the finished product more difficult, costly, and energy-intensive than if pipelines were used. Biodiesel is not transported by pipeline partly because there is a risk of cross-contamination of jet fuel with biodiesel components when both fuels are shipped in multi-product pipelines (McCormick et al. 2009).

As is the case for conventional petroleum fuels, biodiesel fuels produced or imported for the Canadian market must meet specifications. Standards have been developed to ensure the quality of finished biodiesel fuel for each of the various stages of the production process. Notably, undesired transesterification by-products and impurities (e.g., glycerine, methanol, free fatty acids, and salts) must be measured as they affect the performance of biodiesel fuels.

In Canada, petroleum and biodiesel fuel specifications are developed and published by the Canadian General Standards Board (CGSB). CAN/CGSB 3.520-2011, *Automotive Diesel Fuel*

[13] There are roughly 76 primary terminals in Canada.

[14] In-line blending is the addition of biodiesel to diesel fuel as it travels through a pipe or hose providing sufficient turbulence to thoroughly mix both fuels. This approach ensures maximum homogeneity and has been demonstrated to function in cold conditions. This method is currently used to blend fuel additives.

[15] Splash-blending is a procedure whereby biodiesel and diesel fuel are loaded into a tank (e.g. distribution truck, vehicle fuel tank) separately with relatively little mixing.

[16] This technique involves loading the different fuels at the same time into a tank. The fill rate is assumed to be sufficient to create a homogenous mixture, without the need for additional agitation. Large storage tanks could also be equipped with agitators to provide additional mixing.

Containing Low Levels of Biodiesel (B1-B5) (December 1st, 2011) is a standard for automotive low-sulphur diesel fuel containing low levels of biodiesel esters (B1-B5) intended for use in high-speed diesel engines that require low sulphur diesel fuel to meet emission control regulations and high-speed diesel-powered equipment.

Internationally, comparable standards include the American Society for Testing and Materials (ASTM) and the European Committee for Standardization (EN): ASTM-D6751 for neat biodiesel or B100, ASTM-D975 for blends up to B5, ASTM-D7467 for B6-B20 blends, ASTM-D396 for blends up to B5 intended for use in various fuel-oil burning equipment, and EN 14214 for blends up to B7.

1.5 Scope of the Health Risk Assessment

The health impacts analysis detailed in this report evaluates the potential human health implications of the widespread use of biodiesel in Canada, considering the production, distribution, storage and use stages in the lifecycle of biodiesel fuel. The general approach employed is as comprehensive as the available information allows and is comparative in nature, i.e., the impacts of biodiesel are compared to those of conventional ultra low sulphur diesel and presented as relative risks and benefits. Because of the ubiquitous exposure of the general population to air pollution and the very significant health effects associated with air quality, a primary consideration of this analysis is the potential impact of biodiesel use on mobile sector emissions and atmospheric concentrations of air pollutants.

Potential atmospheric emissions from the production of biodiesel in Canada are presented in Chapter 2, which includes a review of the key processes involved and available emissions data.

Chapter 3 addresses environmental fate and transport and the potential effects of biodiesel on contaminant movement through aquifers following fuel spills.

Chapter 4 presents a review of published literature regarding the relative change in vehicle emissions resulting from the use of biodiesel blends compared to emissions from conventional diesel fuel.

Modelling of the Canadian on-road heavy-duty diesel fleet was undertaken and the relative impacts of biodiesel compared to conventional diesel use on regulated and unregulated emissions are presented in Chapter 5. Scenarios considered in this analysis include the relative impact of nationwide use of B0, B2, B5, B10 and B20 in 2006, 2010, 2015 and 2020.

The relative impact of biodiesel use on air quality, estimated through photochemical modelling of the B0, B5 and B20 scenarios in 2006 and 2020, is presented in Chapter 6.

Chapter 7 provides a review of the literature regarding the relative toxicity of biodiesel emissions compared to diesel emissions. Consideration is given to cardiac, respiratory, neurological, mutagenic/carcinogenic, reproductive/developmental, immunological and systemic health outcomes.

Health impacts associated with the estimated changes in Canadian air quality due to biodiesel use are presented in Chapter 8.

Possible benefits of renewable fuel use resulting from reductions in greenhouse gas emissions, occupational health risks associated with the renewable fuels industry, potential health impacts from the agricultural production of feedstock materials, and the potential impacts of renewable content in home heating fuel on emissions from burners and heaters, air quality and health were not considered in this assessment.

Scientific documentation and non-scientific publications published or available before August 1st, 2011 were reviewed and considered in the conduct of this health risk assessment.

1.6 References

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Chapter 2. Biodiesel: Upstream Activities and Emissions

Biodiesel production facilities rely on technologies and processes that generally vary according to the type of feedstock and the level of integration or complexity of a plant. Some facilities integrate oilseed crushing, oil extraction, and biodiesel production, while others are limited to the transesterification process. Key stages considered in this assessment include feedstock handling, biofuel production, fuel storage, and transportation. Farming activities and practices were excluded from the analysis. The emissions from biodiesel production plants were compared to those of petroleum facilities or other industries when data was available and relevant.

Upstream stages have very different impacts and influences on the whole life cycle. Farming and oil seed crushing activities lead to significant amounts of particulate matter emissions, while oil extraction can lead to emissions of air toxics as a result of the use of solvents such as hexane to optimize oil recovery. The largest portion of hydrocarbon emissions resulting from the production of biodiesel fuel are expected to come from oilseed extraction steps (Pang et al. 2009).

2.1 Transesterification Process

Biodiesel production includes two distinct steps: i) oil extraction, and/or purification and/or rendering; and ii) transesterification. Oil extraction/purification/rendering relies on physical (e.g., heating, crushing, pressing) and chemical (e.g., solvents) processes. It may be completed off-site and product trucked to the biodiesel plant or conducted on-site in fully integrated facilities. Transesterification is a multi-step process involving raw oil, alcohols, and catalysts, generally in closed reactors and under vacuum conditions, as a batch or continuous process.

Essentially, one unit (by weight) of vegetable oil mixed with one-tenth unit of alcohol will yield one unit of biodiesel fuel and one-tenth unit of glycerine (Huo et al. 2008). Glycerine has a non-negligible economic value. It can be collected, purified and sold for use in other industrial sectors (e.g., pharmaceuticals, cosmetics, and animal feed).

2.2 Biodiesel production facilities: Activities and Emission Sources

Biodiesel production activities can lead to a variety of emissions or releases to water, air, and soil. Although most hazards are limited to the process facility (i.e., mainly occupational), some releases or accidents could impact nearby communities (e.g., air emissions and waste water discharges).

Hazards (physical, chemical, biological) associated with biofuel production facilities are generally common to other industrial sectors such as: transportation accidents, fires, explosions, spills, and exposure to biological agents (e.g., moulds, bacteria). The risks and impacts of these hazards are known and mitigation strategies, both behavioural and technological, are designed to

limit their occurrence (Resource Environmental Associates Limited (REA) 2010). Further, economic incentives usually limit the loss of marketable or reusable co-products through emissions.

Some process equipment used for biodiesel production, such as reactors, decanters, wash tanks, stripper columns, and distillation columns, are expected to release criteria air pollutants and air toxics into the atmosphere. Combustion emissions (NO_X, CO, SO_2, PM_{10}, $PM_{2.5}$, VOCs, polycyclic aromatic hydrocarbons (PAHs)) will also be emitted from steam boilers, generators, and backup equipment. These emissions will vary depending on the fuel type and technologies selected.

Additional VOC, air toxic, and PM emissions may also result from activities and installations such as storage tanks, biodiesel and glycerine shipments, equipment leaks, cooling towers and haul roads (US EPA 2008). Although VOCs can originate from many stages of biodiesel production, the main concerns are methanol emissions during the transesterification stage and hexane emissions from oil extraction. However, most methanol emissions should be recovered in modern installations (e.g., vapour recovery systems, sealed storage containers, physical separation methods and distillation processes) and reused (Agriculture and Agri-Food Canada (AAFC) 2008).

Other emissions, including heavy metals and air toxics, are generally not of concern since no specific activities have been identified that would lead to significant emissions of these pollutants (REA 2010).

Biodiesel production facilities generally use water for feedstock preparation, cooling and washing biodiesel. The most probable contaminants to water and soil from biodiesel production facilities are methanol, hexane, catalysts (potassium hydroxide and sodium hydroxide), phosphoric acid, glycerine, and biodiesel (AAFC 2008).

Apart from the production process, soil and water quality can also be impacted by spills during transportation, and storage, either of neat biodiesel fuels, biodiesel blends, or other products. An analysis of the environmental fate and transport of biodiesel fuel compounds in soil and groundwater under different Canadian scenarios is presented in Chapter 3 of this report.

The most common air emissions from biodiesel production plants are limited to methanol, hexane, and criteria air contaminants (CACs). Total amounts emitted are expected to be relatively low and to generally meet regulatory requirements, based on information available from the National Pollutant Release Inventory (NPRI)[17] and environmental assessments and environmental impact statements of various biodiesel production facilities (all reports are

[17] http://www.ec.gc.ca/inrp-npri/Default.asp?lang=En&n=B85A1846-1

publicly available and were provided by the Environmental Assessment Division of Health Canada and AAFC). Ambient air concentrations near facilities are expected to meet air quality guidelines. In comparison, petroleum refineries report greater levels of CACs released to air (refer to the NPRI pollution data library). However, the scale of petroleum refineries is important when comparing biofuels with conventional fuels, as some refineries produce more fuel in a few days than most biofuel facilities do in a whole year. Converting the emissions per year data to emissions per litre of fuel or energy (Btu) produced allows for a more relevant evaluation.

Exploratory estimates of air emissions per litre of fuel produced for two large petroleum refineries and a biodiesel plant currently in operation in Canada, based on NPRI emissions data and publicly available corporate information, were conducted (see Table 2-1). Emissions per litre of fuel were determined by dividing 2009 NPRI facility emissions by the throughput capacity. The estimates did not consider the production slate (e.g., gasoline, distillates, petrochemical products) for refineries or temporary shutdowns during the reporting year.[18] Furthermore, the selected biodiesel facility was not necessarily representative of the Canadian industry.

Table 2-1 Tentative comparison of air emissions per litre of fuel produced at Canadian refining and biodiesel production facilities, based on 2009 NPRI data

Facility	Refinery 1		Refinery 2		Biodiesel	
Air emissions	**tonnes/yr**	**kg/L**	**tonnes/yr**	**kg/L**	**tonnes/yr**	**kg/L**
methanol	0.017	$1.15*10^{-9}$	--	--	14	$3.11*10^{-4}$
NO_X (as NO_2)	1642	$1.11*10^{-4}$	1840	$2.45*10^{-4}$	36	$8.00*10^{-4}$
PM_{10}	276	$1.87*10^{-5}$	102	$1.36*10^{-5}$	3.7	$8.22*10^{-5}$
$PM_{2.5}$	245	$1.66*10^{-5}$	69	$9.17*10^{-6}$	1	$2.22*10^{-5}$
TPM	387	$2.63*10^{-5}$	136	$1.81*10^{-5}$	11	$2.44*10^{-4}$
SO_2	4284	$2.91*10^{-4}$	5091	$6.77*10^{-4}$	25	$5.56*10^{-4}$
VOCs	690	$4.68*10^{-5}$	855	$1.14*10^{-4}$	11	$2.44*10^{-4}$
Refinery 1: Ultramar, St-Romuald, QC; 42.1 ML/day capacity; overseas crude Refinery 2: Suncor, Edmonton, AB; 21.5 ML/day; oil sands-based feedstocks Biodiesel facility: Rothsay-Laurenco, Sainte-Catherine, QC; 45 ML/year; tallow feedstocks						

For the major CACs and methanol, the results showed that biodiesel plants were expected to emit more emissions per unit of fuel output than petroleum refineries. Even for an oil sands-based feedstocks refinery, emissions were generally lower compared to the biodiesel facility, except for SO_2. However, petroleum refineries report emissions of various air toxics with human health implications, such as PAHs, benzene, toluene, ethylbenzene, xylenes, and heavy metals. Emissions of these chemical species did not meet the NPRI reporting requirements at biodiesel facilities (refer to the NPRI pollution data library), so they could not be assessed. Nonetheless, not meeting NPRI reporting requirements implies that emissions are substantially lower at

[18] Yearly production was calculated using daily throughput capacity multiplied by 350 activity days.

biodiesel production facilities. In general, the available emissions data for existing or planned biodiesel facilities suggest relatively low total emissions compared to other industrial activities.

For emissions to soil and water, emissions from biodiesel production facilities appear negligible under normal operating conditions based on NPRI data, environmental assessments and environmental impact statements.

2.3 Regulatory Framework for Biodiesel Industrial Projects

Although monitoring emissions data from most biodiesel facilities are not readily available, it is possible to ascertain the federal, provincial, and even municipal regulations with which biodiesel production plants would need to comply. In addition, environmental licences and certificates of approval from government agencies were consulted.

According to available reports and approvals for biodiesel production facilities, emissions from the production stages are expected to be relatively minor. Emissions of concern, such as the CACs and air toxics, are regulated and biodiesel plants are required to meet the required standards set by the federal and provincial governments. In addition, although monitoring data is lacking, regulations and guidelines exist to avoid deleterious environmental and human health impacts via environmental assessments prior to the approval of new facilities in a designated location.

2.4 Biodiesel Production Emissions Modelling

The GHGenius model has been used to predict CAC emissions associated with biodiesel production (e.g., $(S\&T)^2$ Consultants Inc. 2008). GHGenius, developed for Natural Resources Canada (NRCan), is capable of analyzing lifecycle emissions on a mass emitted per distance travelled basis for a variety of diesel vehicle types using petroleum diesel and biodiesel fuels. It considers GHGs, energy consumption, and a number of CACs associated with the production and use of transportation fuels.

This model covers most life cycle stages of a fuel, is flexible, focuses on releases to air, covers GHGs and air pollutants, and has Canadian and US data ($(S\&T)^2$ Consultants Inc. 2008). Calculations within the model are completed on a per unit of energy basis and the model uses a carbon-based approach for GHG estimations. The criteria air contaminants emissions are based in part on the US EPA AP-42[19] emission factors to which modifiers have been applied. Based on a comparative analysis with the *Greenhouse gases, Regulated Emissions, and Energy use in Transportation* (GREET) model, a similar model used by the US EPA, it was determined that GHGenius was a suitable model to predict upstream lifecycle emissions from biodiesel production facilities in Canada ($(S\&T)^2$ Consultants Inc. 2008).

[19] http://www.epa.gov/ttnchie1/ap42/

In the model, process emissions are broken up into emissions that represent the oil extraction and the transesterification stages since they can occur at different facilities. The *production* emissions are also presented distinctly from the *use* emissions.

Production emissions combine emissions from the combustion of fuels used in oil production and refining and the non–combustion process emissions. Control factors are applied so that actual emissions are aligned with those of the NPRI in Canada ((S&T)^2 Consultants Inc. 2008). Common values for the transesterification step are determined for all biodiesel pathways, while individual values are allocated to each oil feedstocks for the oil preparation stages (e.g., crushing, purification).

GHGenius output from previous runs completed by (S&T)^2 Consultants Inc. (2008) using version 3.12 show that process-related emissions for the transesterification stage are generally minor, due in part to the low volatility of feedstocks and end-products. Methanol is the only volatile element for which air emissions are expected ((S&T)^2 Consultants Inc. 2008).

In contrast, the modelling results indicate that the oil extraction step is more emissions-intensive than the biodiesel production stage. PM and VOCs are the primary process emissions related to vegetable oil processing. PM emissions of 32 g/GJ, 30 g/GJ and 0 g/GJ of oil produced were estimated for soy and canola, tallow, and yellow grease (this stage is absent for yellow grease), respectively.

Solvents, generally hexane, used for oil extraction contribute to VOC emissions. Hexane emissions equivalent to 30 g/GJ of fuel produced were expected for all feedstocks ((S&T)^2 Consultants Inc. 2008).

GHGenius (version 3.18) was also run by Health Canada to estimate biodiesel upstream emissions. Test runs were completed for ultra low sulphur diesel (ULSD) and B5, B20, and B100 from soy and canola for the years 2008, 2010, 2015, and 2020. The goal is to identify potential key areas of the upstream activities that may impact emissions. The focus is limited to activities regarding the production per se of biodiesel fuels.

The results for light-duty vehicles (LDVs) and heavy-duty vehicles (HDVs) were analyzed. Results for HDVs are presented in Table 2-2 and Table 2-3. In general, a decrease in upstream emissions of CO and SO_X is expected with increasing canola biodiesel content (e.g., 81% and 11% decrease in CO and SO_X, respectively, with B20) compared to ULSD. Although fertilizer manufacturing leads to considerable SO_2 emissions, overall SO_2 emissions from the production stages are much lower for biodiesel compared to petroleum fuels. Displacement of emissions by

biodiesel production co-products (e.g., glycerine, protein meal) and vehicle operation benefits[20] are responsible for a large share of the CO emissions decrease (see Table 2-3).

Table 2-2 HDV (buses and trucks combined) results sheet for full lifecycle emissions by pollutant in g/km, for the year 2010, using GHGenius version 3.18

Buses and Trucks combined	Petroleum	Biodiesel blends		Neat biodiesel
2010	**Diesel**	**Canola B5**	**Canola B20**	**Canola**
	g/km			
CO				
Vehicle operation	0.18	0.18	0.17	0.09
Upstream	*0.26*	*0.21*	*0.05*	*-0.85*
Vehicle material and assembly	0.10	0.10	0.10	0.10
Total	0.54	0.49	0.32	-0.66
NO_X				
Vehicle operation	0.40	0.40	0.42	0.44
Upstream	*1.06*	*1.33*	*2.19*	*6.89*
Vehicle material and assembly	0.07	0.07	0.07	0.07
Total	1.53	1.80	2.69	7.40
VOC-Ozone weighted*				
Vehicle operation	0.11	0.11	0.10	0.04
Upstream	*0.14*	*0.15*	*0.19*	*0.37*
Vehicle material and assembly	0.03	0.03	0.03	0.03
Total	0.29	0.29	0.32	0.44
SO_X				
Vehicle operation	0.07	0.07	0.06	0.06
Upstream	*0.85*	*0.83*	*0.76*	*0.39*
Vehicle material and assembly	0.10	0.10	0.10	0.10
Total	1.01	0.99	0.93	0.54
PM				
Vehicle operation	0.02	0.02	0.02	0.01
Upstream	*0.09*	*0.12*	*0.20*	*0.68*
Vehicle material and assembly	0.05	0.05	0.05	0.05
Total	0.16	0.18	0.26	0.73
* VOCs are weighted according to their ozone forming potentials.				

[20] Although GHGenius uses an algorithm to mimic MOBILE6.2C results, the vehicle operation emission estimates in GHGenius differ from the MOBILE6.2C data presented in Chapters 5, 6, and 8 of this report.

Table 2-3 HDV (buses and trucks combined) full-cycle CO emission breakdown for the year 2010, in g/km, using GHGenius version 3.18

Buses and Trucks combined Total CO (g/km)	Petroleum	Biodiesel blends		Neat biodiesel
2010	**Diesel**	**B5**	**B20**	**Canola**
Vehicle operation	0.18	0.18	0.17	0.09
Fuel dispensing	0.001	0.001	0.001	0.001
Fuel storage and distribution	0.004	0.005	0.007	0.02
Fuel production	**0.082**	**0.081**	**0.080**	**0.074**
Feedstock transport	0.02	0.02	0.02	0.01
Feedstock and fertilizer production	0.15	0.16	0.19	0.36
Land use changes and cultivation	0.00	0.00	0.00	0.00
CH4 and CO2 leaks and flares	0.00	0.00	0.00	0.00
C in end-use fuel from CO2 in air	0.00	0.00	0.00	0.00
Emissions displaced by co-products	**-0.002**	**-0.06**	**-0.25**	**-1.31**
Sub total (fuel cycle)	0.44	0.39	0.22	-0.76
% changes (fuel cycle)	--	-12.5	-50.7	-271.1
Vehicle assembly and transport	0.01	0.01	0.01	0.01
Materials in vehicles (incl. storage)	0.10	0.10	0.10	0.10
Grand total	0.54	0.49	0.32	-0.66
% changes (grand total)	--	-10.2	-41.3	-221.6

Upstream PM, NO_X, and VOC emissions are projected to increase in association with an increase in biodiesel use. PM emissions[21] are expected to increase by 122% with B20 because of the impact of fuel production and feedstock recovery on overall upstream emissions. Results for NO_X emissions show a large increase (107% with B20), mostly due to land-use changes, cultivation and fertilizer manufacture. For example, as the fertilizer goes through the de-nitrification cycle after it is applied to the soil, it releases NO_X. Upstream VOC emissions are estimated to increase by 36% with B20, mostly due to methanol (transesterification stage) and hexane (oil extraction stage) emissions.

Modelling results of upstream emissions for all target years are similar (i.e., decrease in CO and SO_X, and increase in PM, NO_X, and VOC emissions), as shown in Table 2-4 (in g/GJ) and Table 2-5 (in g/km). However, a general decrease in emissions in all pollutants is noted for both conventional diesel and biodiesel fuels between 2007 and 2020. These projected reductions are likely due in part to modifications in fuel characteristics, energy use, and emissions factors that

[21] PM emissions are not broken down by size fractions for upstream activities in GHGenius; estimates of impacts on $PM_{2.5}$ and PM_{10} emissions are unavailable.

are specific to target years in GHGenius. These can mirror improvements in various domains, such as agricultural yields, biodiesel processing, and emission control measures. However, biodiesel fuel appears less beneficial for CO in 2020 (-42.2 g/GJ) than in 2008 (-68.21 g/GJ). An analysis of the detailed output shows that co-product benefits are reduced by almost 30% in 2020.[22]

Table 2-4 Emissions over the whole upstream fuel cycle per unit of energy delivered to end users in g/GJ (low heating value) for canola biodiesel for the years 2007, 2008, 2010, 2015, and 2020 using GHGenius versions 3.12 and 3.18

Year	**2007**		**2008**		**2010**		**2015**		**2020**	
Model	**version 3.12 [B]**		**version 3.18**							
Fuel [C]	**Hwy diesel**	**B100**	**Hwy diesel**	**B100**	**Hwy diesel**	**B100**	**Hwy diesel**	**B100**	**Hwy diesel**	**B100**
	g/GJ									
NMOC [A]	11.8	25	10.5	26.2	9. 9	25.9	9.0	25.4	8.5	25.0
CO	15.9	-72	18.1	-68.1	17.8	-59.2	17.8	-47.3	17.7	-42.2
NO_2 [A]	72.5	237	81.2	501.6	74.1	480.9	70.5	453.1	68.2	430.3
SO_X [A]	73.6	47	68.0	35.7	59.1	27.0	49.1	17.4	44.0	12.9
PM	6.3	42	6.9	48.4	6.3	47.3	5.4	45.4	4.8	44.2

[A] For GHGenius version 3.12, data are presented in VOC, NO_X, and PM_{10}
[B] $(S\&T)^2$ Consultants 2008
[C] Hwy diesel: *highway diesel*, refers to ULSD (in comparison to *off-road diesel*)

Table 2-5 Upstream NO_X emission per activity for buses and trucks combined for the years 2010, 2015, and 2020 in g/km, using GHGenius version 3.18

Year	**2010**		**2015**		**2020**	
Fuel	**Diesel**	**Canola biodiesel**	**Diesel**	**Canola biodiesel**	**Diesel**	**Canola biodiesel**
	g/km					
Vehicle operation	0.401	0.441	0.400	0.440	0.399	0.439
Fuel dispensing	0.004	0.004	0.003	0.003	0.003	0.003
Fuel storage and distribution	0.025	0.129	0.027	0.110	0.029	0.095
Fuel production	0.312	0.169	0.297	0.150	0.287	0.137
Feedstock transport	0.194	0.016	0.189	0.016	0.185	0.015
Feedstock and fertilizer production	0.534	0.865	0.501	0.707	0.480	0.606
Land use changes and cultivation	0	7.043	0	6.855	0	6.673
Emissions displaced by co-products	-0.008	-1.336	-0.010	-1.366	-0.010	-1.395
Fuel cycle Sub-total*	1.463	7.332	1.408	6.916	1.371	6.574
Fuel cycle % change	--	**401.3**	--	**391.3**	--	**379.4**
* *Some stages are not represented in the table.*						

[22] GHGenius does not provide information why co-product benefits are reduced. It is possible that production processes in other sectors, for products that biodiesel co-products are displacing, are expected to improve, thus reducing CO emissions per product output.

Basic assumptions for activities such as farming, fertilizer use, and land use changes can vary significantly between models and lead to different results. The general lack of available emissions data from the biofuels industry also reduces the reliability of individual emission factors for upstream activities. Nonetheless, life cycle models remain practical tools for identifying the main drivers of emissions at different stages of fuel production or use.

Results from GHGenius life cycle model runs demonstrate that, depending on which life cycle stages are taken into consideration, biodiesel fuels can have positive or negative impacts on air pollutant emissions. However, where emissions occur is also important, but the spatial distribution of emissions is not determined in GHGenius. Most atmospheric emissions from farming, fertilizer manufacturing, and biofuel production plants are expected to occur in rural areas; whereas exhaust emission benefits from the use of biofuels are expected to decrease urban emissions. To assess impacts on population exposure and health, changes in pollutant emissions should be weighted correctly to reflect the geographic distribution of emissions relative to that of populations.

Huo et al. (2009) and Pang et al. (2009) emphasized the importance of accounting for both the location and sources of emissions to achieve a fair comparison between petroleum and renewable fuels. The life cycle analyses of Huo et al. (2009) and Pang et al. (2009) showed that tailpipe emissions contributed most of the life cycle emissions for biodiesel blends and petroleum diesel; hence they represent the most sensitive input to the life cycle emission estimates. This underlines the fact that any air quality benefits from biofuels will depend on when and where a fuel is produced or consumed, in addition to baseline air quality conditions.

A life cycle assessment of first generation biofuels by the French research group ADEME (2010) also arrived at similar conclusions for five distinct indicators: non-renewable energy use, GHG emissions, photo-chemical oxidation potential, human toxicity potential, and eutrophication potential.[23] The main contributor to the human toxicity potential indicator was vehicle use. The report states that the emission benefits at the vehicle stage of the life cycle compensate up to twenty times the upstream agricultural activities, such as fertilizer and pesticide use to protect crops or enhance yields. The benefits are mostly due to a decrease in PAH exhaust emissions with the use of biodiesel, which significantly lowers its toxicity potential (ADEME 2010).

2.5 Discussion and Limitations

Preliminary analysis of emissions data from the Canada's NPRI suggests that biodiesel facilities may emit more CACs than conventional petroleum refineries on a fuel unit basis, while air toxic emissions are considerably lower. Based on the GHGenius model, results of upstream biodiesel

[23] ADEME used the CML (*Centrum voor Milieukunde Leiden*) life cycle model developed by Leiden University to assess environmental indicators.

emissions between 2007 and 2020 are similar, as are the upstream diesel emissions. A general decrease in emissions in all pollutants is noted over time for both conventional diesel and biodiesel fuels. When biodiesel is compared to diesel, a decrease in CO and SO_X emissions, and an increase in PM, NO_X and VOC emissions are observed, both on a g/GJ and a g/km basis. However, limited publicly available data of key pollutant emissions from biodiesel facilities (e.g., PM, VOCs, solvents) do not allow for precise emissions modelling. The use of emission factors based on inaccurate data may not reflect actual emission rates and result in increased uncertainties.

Regionally or provincially, specific life cycle analyses may be more useful to governments, industry, and stakeholders in assessing the impact of biodiesel production's expansion across Canada. This approach would take into consideration:

- Emissions inventories from fuel production equipment;
- Potential release scenarios (water/air/soil impacts);
- Location- or facility-specific factors;
- Specific fuels and/or power source used by facilities;
- Impacts of raw material production;
- Transportation modes and distances for raw materials and final products; and
- Cost analysis of health benefits and overall economic impact of biodiesel.

Greater availability of emissions data, notably for air pollutants, would allow for more relevant assessments of the impacts of biodiesel facilities on their surroundings. This could lead to the development and application of mitigation measures for the most significant emission sources and a better appreciation of the potential benefits or impacts of biodiesel production on human populations.

2.6 References

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Chapter 3. Environmental Fate and Transport of Biodiesel Fuel

Environmental and health risks from accidental releases and spills of bulk biodiesel fuels can be expected in Canada. Because biodiesel and biodiesel blends are transported and delivered via truck, rail, and barge, recognized as the riskiest ways of carrying large volumes of liquid fuels, these risks are increased. Biodiesel is not likely to be moved through pipelines due to pipeline compatibility and fuel quality issues (e.g., contamination of jet fuel with biodiesel components). With a 2% renewable content requirement in diesel fuel and heating oil, it is estimated that an equivalent of 20,000 to 40,000 litres of neat biodiesel per year could be released via spills into the environment (Hollebone 2008).

This chapter outlines the drivers of risk and potential exposure pathways relevant for environmental releases, the principles of subsurface contamination by petroleum and biodiesel fuels, and the main parameters expected to influence pollutant migration in the event of a spill or a large release of biodiesel and diesel fuels. A review of fuel biodegradation is also included. Lastly, results from preliminary subsurface modelling of diesel, neat biodiesel, and biodiesel blend releases are presented. The main objective of this exploratory modelling is to gain some knowledge regarding the differences in subsurface transport of biodiesel and petroleum diesel. As the impacts of biodiesel spills have not been investigated extensively and the contamination potential of biodiesel fuels remains to be elucidated, another objective is to identify major data gaps for the development of more realistic modelling approaches. The focus of this chapter is limited to subsurface water quality, which is often a major health concern associated with releases of petroleum products.

3.1 Risk Drivers and Exposure Pathways for Petroleum and Biodiesel Fuel Contamination

Petroleum hydrocarbons are a concern for a number of reasons, such as their reduced chemical nature (i.e., propensity to oxidize), volatility, toxicity, carcinogenicity, environmental mobility, persistence, and their ability to degrade soil quality (CCME 2008b). The contamination profile at a fuel spill is a function of several factors, including source, fuel type, site characteristics, and time since release (CCME 2008b). With regards to biodiesel fuels, it is expected that contamination will depend on feedstock characteristics, chemicals added during the production process and by-products, in addition to the blend level.

In Canada, guidelines for the remediation of contaminated sites are based on the Canada Wide Standards (CWS) for Petroleum Hydrocarbon in Soil (CCME 2008b). Because petroleum fuels are a mixture of numerous substances and not a single compound, total petroleum hydrocarbon (TPH) concentrations cannot be used to directly assess human health risks (Park and Park 2010).

Several methods have been developed to simplify health risk evaluations of fuel contaminations. For example, fuels can be divided into aromatic and aliphatic fractions based on the equivalent carbon number, for which various parameters are defined using correlations with the equivalent carbon number. Fuel fractions are then considered as single compounds in fate and transport models (Park and Park 2010). For the purpose of the current health assessment, the fractions determined by the Canadian Council of Ministers of the Environment (CCME) based on the equivalent carbon number were used: F1 (C6-C10), F2 (C10-C16), F3 (C16-C34), and F4 (C>34).

As shown in Table 3-1, the fatty acid methyl esters (FAMEs) that compose biodiesel are predominantly in the F3 (C16 to C34) range defined by CCME (2008a, b), with a lesser amount in the F2 (C10 to C16) range.

Table 3-1 Equivalent carbon number estimation for the most common individual FAME compounds[24]

Component	Structure*	Equivalent carbon number	CCME fraction
Methyl myristate	C14:0	16.6	F3
Methyl palmitate	C16:0	17.5	F3
Methyl palmitoleate	C16:1	17.7	F3
Methyl stearate	C18:0	19.7	F3
Methyl oleate (cis) and elaidate (trans)	C18:1	19.3	F3
Methyl linoleate	C18:2	19.5	F3
Methyl α- and γ-linolenate	C18:3	19.7	F3
Methyl arachidate	C20:0	20.6	F3
Methyl eicosinate	C20:1	20.6	F3
Methyl behenate	C22:0	22.2	F3
Methyl erucate	C22:1	22.4	F3
Methyl tetracosanate	C24:0	23.9	F3
* The chemical structure is designated by the notation C*AA*:*B*, where *AA* indicates the number of carbon atoms in the carbon chain and *B* the number of double bonds in the carbon chain. AA does not include the carbon in the ester group.			

Source: Meridian 2009a, Tables 5A and 5B

For human health risk assessments, it is important to determine the key fractions among the fuel components, as some can have a greater effect on human health (Park and Park 2010). In addition, the impact of fuels fractions on human health may vary depending on the exposure route (e.g., inhalation, groundwater ingestion). For example, F1 and F2 fractions are generally mobile and can be volatile, while F3 and F4 are not considered volatile or mobile in soil and groundwater but have a tendency to be environmentally resilient.

[24] Calculated from boiling point (BP) using procedures from Gustafson et al. (1997) [$EC=4.12 + (0.02*BP) + (6.5*10^{-5} * BP2)$].

Health risks from inhalation of volatile chemicals and ingestion of contaminated water can be significant if these pathways are operable (mostly F1 and some F2 fractions). However, because biodiesel fuel components are generally equivalent to F3 compounds (see Table 3-1), inhalation and groundwater contamination are not identified *prima facie* as major pathways.

Different human health and environmental soil quality guidelines are published for carcinogenic compounds such as benzene, toluene, ethylbenzene, and xylene (BTEX) and carcinogenic petroleum hydrocarbons (e.g., PAHs) (CCME 2010, 2011) as they are generally analyzed separately from the petroleum fractions (CCME 2008b). However, BTEX and PAHs are not considered in the modelling of biodiesel spills due to data and model limitations.

3.2 Subsurface Soil and Water Contamination by Fuels

The behaviour and movement of contaminants in subsurface media depend on their chemical and physical properties, the nature of the spill event (e.g., volume, area, rate, duration), as well as the environmental conditions (Mercer and Cohen 1990). Fuel contaminants in subsurface media can exist in four phases: vapour in the air contained in pore spaces of unsaturated soil; sorbed to solids; dissolved in water; and as a separate, immiscible, non-aqueous phase liquid (NAPL) (FRTR 1997). Contaminant transport or migration occurs in the aqueous (water), vapour (gaseous), and NAPL phases (Yoon et al. 2009). As for relevant soil and groundwater properties impacting the transport of contaminants, these include the composition of the geologic materials, the moisture content, the particle size distribution, the depth of the water table, the hydraulic gradient, the hydraulic conductivity, and the soil and groundwater geochemistry.

In summary, subsurface fuel contamination occurs in the following way. If a sufficient volume of petroleum fuel is released at the ground surface, for example from an accidental spill, it will infiltrate into the soil and migrate downward through the unsaturated soil before reaching the water table. The soluble compounds contained in the fuel may then be slowly dissolved into the groundwater. Once the contaminants are dissolved into groundwater, various processes determine the speed and distance these contaminants migrate away from the original source. In the case of small release volumes, or very large depths to the water table, the fuel may not reach the groundwater table.

3.3 Biodegradation of Petroleum and Biodiesel Fuels

Biodegradation by natural populations of microorganisms is a primary mechanism for the removal of petroleum products from the environment. However, some components of petroleum hydrocarbons, such as branched aromatics, are recalcitrant to microbial degradation or are simply non-biodegradable (Stolz et al. 1995).

As a general rule, a fuel is considered biodegradable more than 90% of it degrades within 21 days (Sendzikiene et al. 2007). Biodiesel fuels from a variety of feedstocks (e.g., rapeseed, soy) have been classified as readily biodegradable compounds (Zhang et al. 1998 *in* US EPA 2008; NESCAUM 2001). Some biodiesel fuels have been observed to exceed the 90% degradation benchmark under aerobic conditions after 21 days, while petroleum diesel fuel is limited to around 60% (Sendzikiene et al. 2007; Makareviciene and Janulis 2003). In terms of energy requirements, biodiesel appears as a much better carbon source to support microbial growth (Owsianiak et al. 2009).

Horel and Schiewer (2011) investigated the effects of temperature on biodegradation in a sandy soil contaminated with diesel fuel, fish oil biodiesel, and several blends of the two. Generally, it was observed that biodegradation rates increased with temperature (from 6^oC to 20^oC) and biodiesel content.

Differences have been reported between vegetable and animal-based biodiesel fuels due to the fatty acid profile and inherent components of those feedstocks. For example, because of the presence of natural antioxidants in vegetable oil-based biodiesel fuels, degradation under aerobic conditions was only initiated once the activity of antioxidants had ceased (Sendzikiene et al. 2007). Further, unsaturated fatty acid compounds are more prone to oxidation.

Peterson and Möller (2005) indicated that biodiesel fuels contain significantly more biodegradable organic matter than petroleum diesel reference fuels, reflected by higher biological oxygen demand (BOD_5) values. Yet, a high BOD_5 can impact soil and aquatic organisms locally if, for example, it leads to the depletion of oxygen and anaerobic conditions in the subsurface. Nonetheless, under anaerobic conditions, which are common in the saturated zone (i.e., below the water table), studies suggest that while biodegradation may be slower, neat biodiesel can be biodegraded under anaerobic conditions in the presence of a variety of electron acceptors (e.g., sulphate, nitrate) (Cyplik et al. 2011; Atkas et al. 2010).

Concerns have been raised about the presence of chemical antioxidants and additives in biodiesel fuels and their potential impact on biodegradation, but few relevant studies have investigated this issue.

3.3.1 Co-metabolism between petroleum and biodiesel products

No clear consensus exists regarding co-metabolism[25] or synergistic effects between biodiesel and petroleum products in a spill (Owsianiak et al. 2009). Biodiesel fuels have been observed by some authors to speed the rate at which biodiesel-petroleum blends can biodegrade (Zhang et al. 1998; von Wedel 1999; Mudge and Pereira 1999; Peterson & Möller 2005; Pasqualino et al. 2006; Prince et al. 2008). In contrast, some authors suggest that the higher biodegradation rate is

[25] As used in the text, co-metabolism is defined as an increase in the degradation rate and extent of a *product A* (e.g., petroleum diesel) in the presence of a *product B* (e.g., biodiesel), compared to that of *product A* alone.

not due to co-metabolism effects, but possibly to commensalism[26] between active microorganisms (Mariano et al. 2008; Cyplik et al. 2011). Adding biodiesel to regular diesel can enhance degradation of the latter, presumably by helping to establish a more active microbial culture (Horel and Schiewer 2011).

3.4 Subsurface Fate and Transport Modelling of Diesel and Biodiesel Fuels Under Specific Scenarios

Due to a lack of relevant experimental and/or case study data under Canadian conditions, Health Canada undertook modelling to provide a preliminary assessment of the potential environmental and human health issues associated with releases of biodiesel (Meridian 2009a, b, c; 2010a, b).

3.4.1 Fraction and compositional approaches

Two approaches are considered for fate and transport modelling of fuels in the subsurface. The *compositional approach* uses the values of chemical and physical parameters of individual fatty acid alkyl esters (FAAEs) to calculate a weighted average based on the average composition of a specific biodiesel fuel. Modelling the fate and transport of individual components of biodiesel may be a practical option because, in general, fewer than a dozen FAMEs comprise more than 99% of biodiesel fuels (Singh and Singh 2010). Table 3-2 presents the typical composition of petroleum diesel and neat canola biodiesel.

The *fraction approach* is used for petroleum hydrocarbons because they contain a large number of individual compounds (numbering in the hundreds) with insufficient fate and transport and toxicological data, making the analytical and computational requirements excessive (Gustafson et al. 1997). It relies on the relationship between the equivalent carbon value of fuel components and various other properties to derive values for chemical and physical characteristics based on the CCME PHC fractions (CCME 2008a). For consistency, biodiesel and biodiesel blends are also assessed via the fraction approach or via a combined approach. For biodiesel blends, the mass fractions within each fraction are adjusted accordingly (e.g., B20 is composed of 20% biodiesel sub-fractions and 80% petroleum sub-fractions). The FAMEs present in canola-based biodiesel are predominantly in the F3 (C16 to C34) range (see Table 3-1). Therefore, to include all diesel and biodiesel fuel fractions, only the F2 and F3 are evaluated.

[26] In mixed cultures, commensalism plays an important role since each species may have a specific function in the enzymatic reaction sequences, responsible for the breakdown of more complex molecules.

Table 3-2 Composition of unblended petroleum and canola-based biodiesel fuels

Component	Percent by weight	Reference
ULSD		
PHC F2		
C10-C12 aliphatic	18	CCME 2008b, Tables C.4 and D.10
C10-C12 aromatic	4.5	
C12-C16 aliphatic	22	
C12-C16 aromatic	5.5	
PHC F3		
C16-C21 aliphatic	28	CCME 2008b, Tables C.4 and D.10
C16-C21 aromatic	7	
C21-C34 aliphatic	12	
C21-C34 aromatic	3	
BIODIESEL - Canola		
16:0 methyl palmitate	4.5	Meridian 2009a, Table 9
16:1 methyl palmitoleate	0.2	
18:0 methyl stearate	1.9	
18:1 methyl oleate (cis) and elaidate (trans)	60.8	
18:2 methyl linoleate	22.2	
18:3 methyl α- and γ- linolenate	8.8	
20:0 methyl arachidate	0.1	
20:1 methyl eicosinate[27]	0.5	
22:0 methyl behenate	0.1	
22:1 methyl erucate	0.8	
24:0 methyl tetracosanate	0.1	

Source: adapted from Meridian 2010a.

3.4.2 Scenarios

Scenarios are designed to represent plausible, common biofuel incidences under Canadian conditions (i.e., based on the current biofuel production and distribution infrastructure and practices; and frequent types of fuel spills), namely releases from underground storage tanks (USTs) and accidental spills from a tanker fuel truck (CCME 2006, 2008b).

Releases from USTs take place at the depth the tanks are buried and are often slow leaks occurring over long periods of time due to the difficulty in detecting these leaks. The UST scenario assumes that all fuel and biofuel components are dissolved completely in groundwater and does not address partitioning from a light NAPL (LNAPL). To allow sufficient contrast in flow and transport between fuels, a worst case scenario is defined. For example, the initial source concentration of fuel in groundwater and the hydraulic conductivity are set to 10 mg/L and

[27] Eicosenic acid methyl ester

32,000 metres per year,[28] respectively (i.e., greater than expected values). Also, an infinite source mass is assumed.

The Truck spill scenario simulates a release from a tanker truck in the event of a vehicle accident or truck rollover. This type of release could occur at virtually any location in Canada where fuels are transported by truck, most likely on or adjacent to a roadway. In this event, a large amount of fuel (potentially the entire contents of a truck; 10,000 L for the simulation) may be released on the ground surface over a very short period of time. In most cases, it is expected that residual contamination remaining after an initial spill response could be evaluated using the same assumptions as for environmental quality guideline derivation.

For the current assessment, biodiesel fuel is assumed to be exclusively canola-based from transesterification with methanol (i.e., FAMEs) and scenarios are modelled with both coarse and fine-grained soils. Initial groundwater concentrations are either based on HSSM modelling results (see Section 3.4.3) or based on a selected total dissolved diesel, biodiesel or fuel blend concentration at the source. Consequently, the initial concentrations of individual components differ depending on the fuel blend.

3.4.3 Model selection and applicability

Two models (out of 34 reviewed) with possible applications for biodiesel modelling are used for this assessment (Meridian 2009c): BIOSCREEN/BIOSCREEN-AT and the Hydrocarbon Spill Screening Model (HSSM). The scarcity of experimental physical-chemical and fate and transport data for biodiesel fuel components favours the use of simple, transparent, and widely used models, with lower input requirements.

BIOSCREEN (US EPA 1996) assesses three-dimensional transport for dissolved phase hydrocarbons. The BIOSCREEN model only allows for limited input of physical and chemical properties: the model cannot address multiple fuel components with different chemical and physical properties simultaneously. Hence it is more appropriate and applicable for initial site screenings of biodiesel contamination. A related model, BIOSCREEN-AT, is more appropriate for non steady-state conditions, large distances or high biodegradation rates.

HSSM was developed by the US EPA and is adapted to NAPL transport, simulating NAPL movement during the initial stages of a spill. Although the LNAPL is generally composed of several distinct chemical compounds, HSSM can only trace the transport of one chemical at a time, as selected by the user. As it approximates or excludes some processes, HSSM is considered a screening model (e.g., assumes subsurface homogeneity, excludes some chemical and hydrologic phenomena, short computing time) (Meridian 2009c) and is intended only to provide order-of-magnitude results (Weaver et al. 1994). The HSSM model assumptions, input

[28] Approximate value for a sorted sand and gravel medium.

parameters, modelling modules, and source codes are detailed in Weaver et al. (1994) and Charbeneau et al. (1995).

Fuel mixtures can be modelled if they are considered as a single substance with a unique set of values for their physical and chemical properties. Alternatively, the simulation programs currently available allow modelling each fuel components and fuel fractions individually. No modelling system with reasonable data requirements and the capability to simulate the transport of complex mixtures was identified during this assessment. Hence, the modelling activities described in this Chapter assume that there are no interactions between fuel components (e.g., changes in effective solubility, co-metabolism effects). It is uncertain how this assumption impacts the modelling results (i.e., more or less conservative results).

3.4.4 Input data: requirements, availability, and surrogates

CCME inputs are used for both coarse- and fine-grained soils because they are considered representative of Canadian conditions (CCME 2006; 2008a,b[29]). Environmental parameters are adjusted to reflect different biodiesel release scenarios. Scenario specific input parameters are presented in Table III-1 of Appendix III.

Fuel composition and physical-chemical parameters are assembled from previous scoping studies (Meridian 2009a, b, c). The composition of unblended ULSD and canola-based biodiesel is shown in Table 3-2. The PHC fractions include sub-fractions of aliphatic and aromatic components within a specified equivalent carbon range (mostly between 9 and 20 carbon atoms), each having defined values for physical and chemical properties (CCME 2001, 2008a). Canola biodiesels typically have carbon chains of 16, 18, or 20 with an overall range of 14 to 24 carbons (Chhetri et al. 2008 *in* Demirbas 2009).

Several key properties are not available in the literature for whole biodiesel. To fill data gaps, the *Estimation Programs Interface* (EPI) *Suite* (US EPA) and the *SPARC Performs Automated Reasoning in Chemistry* (NREL - Athens, GA) software programs are used. These programs generate values for physical and chemical properties for individual compounds based on molecular structure and basic information about the environment. Results from EPI Suite and SPARC are generally similar. When results differ considerably, preference for one program is based on support from alternative sources of information on physical and chemical properties of biodiesel fuel compounds, such as literature data, or professional judgement.

Where no data can be found regarding the physical-chemical properties of biodiesel-diesel blends, but are available for neat biodiesel fuels, a linear relationship based on the blend concentration and properties of neat fuels is used to extrapolate values (Meridian 2009a).

Specific NAPL inputs required for the HSSM model are presented in Table III-2 of Appendix III. Physical and chemical parameter values of fuel components used in the modelling are presented for ULSD fuel and canola biodiesel fuel in Tables III-3 and III-4, respectively, of Appendix III.

[29] Refer to Tables C.1 and C.2 in CCME 2008b.

3.5 Results

3.5.1 Underground storage tanks

A first series of runs for ULSD, neat biodiesel, B5, and B20 are completed using BIOSCREEN-AT to determine the maximum distance at which an assumed measurable concentration of 0.001 mg/L for all fuel components[30] is predicted after 1, 2, 3, 4, and 5 years.[31] A second series of runs models the concentrations following a transport time of 1 and 5 years, at 3 m increments from 0 to 30.5 m. These distances and increments are chosen to capture the full extent of the plume and allow a fair characterization of its progression in time.

As expected, plume modelling results show that lighter ULSD aromatic compounds are generally transported the furthest, followed by light end FAMEs such as palmitic and palmitoleic acid methyl esters (see Figures III-1 and III-2 in Appendix III). Heavier biodiesel and ULSD compounds prove to be relatively immobile. Light-end ULSD components such as C10-C12 hydrocarbon chains migrate up to 250 m after 5 years, while biodiesel components remain within 50 m of the source area due in part to their increased biodegradation rates. Regarding the biodiesel fuel blends, minor differences are noted between the behaviour of biodiesel components in the B5 and B20 plumes, while the ULSD subfractions behave similarly to the neat ULSD simulation.

Predicted concentrations of biodiesel fuel components decrease more rapidly than diesel fuel compounds in relation to distance from the source, partly because they are assumed to degrade much faster. In contrast, heavier hydrocarbons are less soluble and are adsorbed to soil particles, two factors that limit their migration.

Plume composition relative to distance from source area after 5 years, as estimated with BIOSCREEN-AT for the UST scenario, is represented in Figure III-3 (see Appendix III).[32] Groundwater concentrations for individual fuel components are represented and summed per fuel type for each receptor point (or distance from source). As the plume moves away from the source, the total groundwater concentration decreases and the composition profile eventually includes only the more mobile elements. Beyond the source area, light end aromatic compounds comprise most of the ULSD plume. As for biodiesel plume composition, only palmitic acid is expected to travel more than 20 m from the source contamination after 1 year (data not shown; Meridian 2010b), while oleic and linoleic acid methyl esters compose most of the plume near the source.

[30] Each carbon fraction and biodiesel component is simulated individually.
[31] The 0.001 mg/L concentration value was selected by Meridian (2010b) and considered appropriate to describe plume evolution. It is not based on a health or a groundwater quality standard.
[32] *Source*: Meridian 2010b, Figure 8

3.5.2 Truck spill

The migration of the NAPL plume is modelled with HSSM for the Truck spill scenario. Modelling results indicate that the NAPL plume does not reach the water table with fine-grained soil conditions (data not shown; Meridian 2010a), so only coarse-grained soil conditions and modelling results are considered. In fact, the fuel type has less influence than hydrogeological input parameters in determining the NAPL plume size. Modelling is conducted with BIOSCREEN-AT to determine the predicted concentrations at 1.5 m increments over a distance of 15 m, at 5 weeks, 6 months, and 1 year after the initial release of fuel. Similar inputs to the UST scenario are used for the truck spill scenario, such as contaminant concentrations set to the maximum solubility in water limits (Table III-1, Appendix III). Source depletion is based on the total content of the transport truck, estimated at 10,000 L.

Results regarding plume composition and concentration relative to distance from source area after one year for the truck spill scenario show that dissolved biodiesel compounds from the LNAPL plume are predicted to migrate at a lower rate than ULSD components and be restricted to shorter distances (data not shown; Meridian 2010b). After one year, composition and concentrations are predicted to continue evolving (data not shown). [33] These changes are due to the advancing biodiesel plume as the NAPL source is dissolved.

For ULSD, the composition of the plume outside the source area appears to stabilize over a short period of time and concentrations of aromatic compounds begin to decrease within a year with depletion of the source. For blends, dissolved FAMEs released from the LNAPL plume are expected to be minimal, partly because of biodiesel's higher biodegradation rate.

3.6 Discussion

Many of the processes that influence the fate and transport of the dissolved contaminants derived from diesel and biodiesel are the same. Biodegradation is notably important because it removes organic mass from the contaminated aquifers. For the diesel fuel components considered in the modelling, all have a half-life of 1750 days (see Table III-3 of Appendix III). Biodiesel fuel is assumed to degrade at twice the rate of diesel fuel fractions (half-life of 875 days; see Table III-6), which has a significant impact on the modelling results as biodiesel mass in the subsurface is removed more rapidly, limiting the potential for fuel compounds to travel away from the source unaltered.

Regarding biodegradation rates, it must be emphasized that the model uses first-order decay values, i.e., first-order aerobic degradation rates. For this assumption to be valid sufficient oxygen is required in the subsurface to avoid the onset of anaerobic conditions, which can alter the biodegradation rates and possibly lengthen the groundwater plumes. If this modelling condition is violated or incorrect, the fate and transport of biodiesel blends and petroleum fuels

[33] See Meridian 2010b, Figure 9 to Figure 11

could be much different. However, even under anaerobic conditions, biodiesel is expected to degrade faster and lead to similar migration trends as exposed in Section 3.3. In addition, biodegradation supposes that pollutants are available for biological degradation. Aqueous compounds are usually more bioavailable than compounds sorbed to soil media. Because petroleum fuel components have a tendency to adsorb strongly to subsurface media, hindering the biodegradation processes, differences can be expected between petroleum and biodiesel fuels.

The sorption coefficient (K_{OC}) and corresponding retardation factor are fuel parameters that significantly impact pollutant migration. These parameters explain why palmitic acid is expected to travel more than other biodiesel fuel components. In fact, the K_{OC} and retardation factor values for palmitic acid are much less than for longer chained components, such as oleic, linoleic, and eicosenic acid methyl esters (see Table III-4, Appendix III). They also explain why aromatic compounds in diesel fuel are expected to travel faster than aliphatic compounds.

3.7 Conclusion

Novel and marginal fuels like biodiesel do not benefit from an extensive database pertaining to environmental releases and no tool has been specifically developed to predict environmental fate and transport.

For this health risk assessment, a review of the literature was conducted to evaluate the quantity and quality of chemical and physical data available regarding biodiesel fuels. The aim of this review was to identify the relevant data to populate fate and transport models and allow for the modelling of biodiesel releases. Key findings from this review indicated that physical and chemical properties of biodiesel fuel components are significantly different from petroleum fuels. For example, the data showed that biodiesel has greater viscosity (especially at lower temperatures), greater solubility, increased biodegradability, and a lower sorption coefficient. However, data gaps were identified for several physical and chemical parameters necessary for fate and transport modelling (e.g., Henry's Law Constant, vapour pressure, water solubility), either for whole biodiesel fuels and/or individual biodiesel components. These values were estimated using the EPI Suite and SPARC programs, or determined based on physical and chemical relationships (e.g., equivalent carbon number). Ultimately, it was concluded that sufficient information was available to conduct preliminary modelling with simple, low input requirement models.

The modelling results of neat ULSD, neat biodiesel, and biodiesel blends show that biodiesel fuel components are expected to travel less than ULSD fuel components, as expected based on biodiesel's physical and chemical characteristics. The reduced migration is assumed to result in part from the greater biodegradation rate of biodiesel fuel components compared to diesel fuel fractions. In addition, the variety of biodiesel compounds expected to migrate away from a

source area is quite limited. Only palmitic acid methyl ester demonstrated a propensity to migrate a significant distance (approximately 42 m from the source after 5 years) because of its relatively low sorption coefficient. In contrast, modelling results indicate that diesel fuel fractions composed of short-chained aromatic compounds travel much greater distances (nearly 250 m from the source after 5 years).

The limited mobility of biodiesel fuel components can be considered an environmental benefit since the contamination plumes is expected to be contained within a relatively small volume of soil and groundwater. Notwithstanding the modelling uncertainties (i.e., assumptions and input data), it is reasonable to conclude that biodiesel fuels would have less impact on the environment and human health than petroleum fuels following an uncontrolled release in a natural or urban environment. However, no relevant conclusion can was drawn regarding spills of biodiesel fuel blends, as interactions between fuel components are not considered in the modelling. In addition, the relative toxicity of individual FAMEs is not readily available.

According to a review of the biodiesel literature, it does not appear that such an assessment of biodiesel subsurface transport via modelling has been completed and published in North America; although reports on ethanol releases have been published (e.g., US EPA 2010b, NESCAUM 2001) and preliminary results of a multimedia assessment of biodiesel are available[34] (Cal-EPA 2010).

3.7.1 Limitations and uncertainties

Various data gaps and limitations can be identified, such as the presence and impact of additives, biodiesel-diesel interactions in soil and groundwater, and the limited experimental data regarding physical and chemical parameters of individual biodiesel fuel components.

Access to more detailed and relevant physical and chemical data would allow for the use of more complex models that consider interactions in soil and groundwater between fuel components, additives, and evolving environmental conditions (e.g., aerobic and anaerobic conditions), and therefore more realistic modelling. Although this lack of data and use of simplistic models are considered major limitations to the modelling conducted as part of this assessment, they are unavoidable as no modelling system with reasonable data requirements and the capability to simulate the transport of complex mixtures is available.

Another limitation is the absence of reports focusing on biodiesel spills and subsurface transport of biodiesel fuel components. Subsequently, it is not possible to compare the current modelling results with other simulations or existing field data. Several field studies have demonstrated that biodiesel can biodegrade at a greater rate than diesel fuels (see Section 3.3), but as Cyplik et al. (2011) note, findings from one study cannot be directly extrapolated. Essentially, the authors

[34] See http://www.arb.ca.gov/fuels/multimedia/multimedia.htm

note that the characteristics of a local microorganism community (e.g., bacterial groups present in contaminated soil) can vary significantly from one location to another and that environmental conditions (e.g., aqueous media, porous media, aerobic conditions) may affect the response of these communities in the presence of biodiesel and diesel fuels.

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Chapter 4. Impacts of Biodiesel Use on Vehicle Emissions – Review of the Literature

The transportation sector is responsible for emissions of several pollutant types, such as criteria air contaminants[35] (i.e., PM, CO, NO_X, SO_2, VOCs, and NH_3) and air toxics (e.g., polycyclic aromatics hydrocarbons and aldehydes). Mobile sources are the largest contributing sector to NO_X and VOC emissions, and diesel vehicles are responsible for a large share of the NO_X and PM emissions from this sector.

The introduction and use of biodiesel fuel in Canada will inevitably modify the profile of vehicle exhaust emissions in comparison to petroleum diesel fuel. A literature review by the US EPA (2002a) underlined that several biodiesel fuel characteristics could have an impact on exhaust emissions. The emissions, for example, were found to vary in relation to the biodiesel feedstock (i.e., fuel chemistry), the biodiesel blend, the reference and/or blending diesel fuel, the engine/vehicle type and technology, and the drive cycle sequence. Thus, various biodiesel parameters need to be considered in a review of emission impacts.

This Chapter will address the main factors that influence exhaust emissions of regulated (NO_X, PM, hydrocarbons and CO) and unregulated (e.g., VOCs, PAHs) pollutants for vehicles and engines using biodiesel fuels.[36]

4.1 Engine Type, Calibration, and Test Cycles

The majority of published studies have investigated the impact of biodiesel on exhaust emissions from heavy-duty vehicles, mainly because they represent a greater share of the diesel fuel market in North America and also are considered important emitters of PM and NO_X. A more limited number of reports, mainly European studies, have looked at emissions from light-duty vehicles.

Although general trends for regulated emissions and other air pollutants are observed when using biodiesel blends in compression ignition engines, exhaust emissions remain engine specific (Graboski et al. 2003). Various engine parameters, such as power capacity and fuel injection technologies, can vary and it highlights the importance of using identical, or at least similar, engines when comparing exhaust emissions between conventional and biodiesel fuels. In a review of biodiesel emission data published up to 2001, the US EPA (2002a) reported that the use of B20 in heavy-duty engines led to considerable reductions in PM, HC, and CO emissions and a slight increase in NO_X emissions. It was suggested that the magnitude of changes varied according to the engine manufacturer, model, and design (US EPA 2002a; McCormick et al. 2006).

[35] As defined by Environment Canada (http://www.ec.gc.ca/inrp-npri/default.asp?lang=En&n=9264E929-1#c)

[36] A comprehensive and all-inclusive review of biodiesel impacts on emissions represents a non-trivial task, beyond the scope of the current assessment.

Modern engines are calibrated to meet regulatory standards when using reference fuels, usually ultra low sulphur diesel (ULSD), meeting specific fuel quality criteria (e.g., ASTM D975). These fuel criteria differ for biodiesel fuels (e.g., ASTM D7467 for B6-B20 and ASTM D6751 for B100) and depend on the physical and chemical nature of the fuel components. Engine calibration can significantly affect emissions when fuelled with either biodiesel or ULSD fuels (Peterson et al. 2009; Ireland et al. 2009). For example, the lower energy density of biodiesel fuel implies that a greater volume of fuel is required to produce an equivalent amount of energy. A shift from the optimized factory engine settings can influence combustion and impact emissions of regulated pollutants (Tat et al. 2003).

Emissions from internal combustion engines are tested on different test cycles. These cycles are a sequence of speed and load variations within a determined timeframe, designed to replicate highway, city and suburban driving conditions, or standard, passive, and aggressive driving behaviours. These and other test cycle variables (e.g., engine load) will have an influence on emissions (Durbin and Norbeck 2002; Rosenblatt et al. 2008). For example, differences in biodiesel effects on NO_X emissions reported from different test cycles can be reconciled when the analysis is carried out on the basis of load (Sze et al. 2007; Eckerle et al. 2008; US EPA 2009).

Cold starts can also influence overall emissions significantly. Martini et al. (2007) observed that HC and CO emissions were greater with biodiesel fuels, but only for the first 200 to 300 seconds. Similar findings were also observed for PM emissions by Fontaras et al. (2009). It was suggested that the higher boiling point of biodiesel fuels influences the evaporation process when the engine is still cold. Once the engine was warm, emissions from petroleum fuels and biodiesel fuels were similar.

4.2 Regulated Air Pollutants

The regulated air pollutants refer to chemical species for which tailpipe emission standards exist and include PM, CO, NO_X, and HC. Analysis of heavy-duty engine and chassis data generally show that NO_X emissions increase slightly, while PM, CO, and HC emissions decrease with the use of biodiesel in comparison to reference petroleum fuels (US EPA 2002a, 2009).

4.2.1 Particulate matter

Several authors have reported that biodiesel fuels yield lower PM emissions than petroleum diesel (Morris et al. 2003; McCormick et al. 2003; Souligny et al. 2004; McCormick et al. 2006; McCormick 2007). A linear relationship was determined between PM mass emission reductions and increasing concentrations of biodiesel (US EPA 2002; Durbin et al. 2007).

Lapuerta et al. (2008a) identified several reasons for the documented trend of PM reductions with the use of biodiesel in both light-duty and heavy-duty vehicles:

- the oxygen content of biodiesel and the related lower stoichiometric[37] need for air;
- the quasi absence of sulphur and aromatics, both considered soot precursors;
- the greater oxidation of biodiesel PM; and
- the lower final boiling point of biodiesel.[38]

These factors generally promote better fuel combustion and a reduction in soot when biodiesel is used.

While reductions in total PM mass are consistent in HDVs (US EPA 2002a), some studies have reported variations in the response of LDVs over different load conditions (Kousoulidou et al. 2009) or test cycles (Karavalakis et al. 2009a, 2009b). CONCAWE (2010) reported reductions in PM emissions from the testing of three modern LDVs over the New European Drive Cycle (NEDC) regulatory test cycle.

Particulate matter (PM) is generally measured by mass for regulatory purposes, but it can be measured or evaluated in different ways, such as by size and/or number. When comparing biodiesel fuels to a reference petroleum diesel fuel, studies report diverging results for the distribution of emitted PM size fractions. Results either demonstrate an increase (Bunger et al. 2000), a reduction (Jung et al. 2006; Tsolakis 2006; Bennett et al. 2008; Lapuerta et al. 2008; Ballasteros et al. 2010; Fontaras et al. 2010; Kim and Choi 2010; Zhu et al. 2010), or no change (Bunger et al. 2000) in the particle size distribution. Reasons for these discrepancies vary between studies.[39]

Similarly for PM number, reports indicate either a reduction in the number of emitted particles with the use of biodiesel fuels (Chen and Wu 2002; McCormick 2007; Bennett et al. 2008; Lapuerta et al. 2008a), which according to Bennett et al. (2008) correlates well with lower PM mass emissions, or an increase in PM number (Di et al. 2009; Turrio-Baldassarri et al. 2004; Lue et al. 2001). Turrio-Baldassarri et al. (2004) attributed the increase in PM count to a slight shift in the fraction of fine to ultrafine particles translating into a significant increase in particle number. Alternatively, Di et al. (2009) noted that biodiesel combustion yields greater number concentrations of PM because the reduction in particle mass lowers the agglomeration and coagulation rate of smaller particles. This results in lower soot mass, but greater particle number concentration in comparison to ULSD. The decreased surface area of solid particles is thought to promote the formation of nanoparticles by homogeneous nucleation, resulting in increased total particulate numbers (Fontaras et al. 2010).

[37] Stoichiometric ratio: An ideal air-fuel mixing ratio when all fuel and air is consumed.
[38] A lower final boiling point lowers the probability of soot or tar being formed from heavy hydrocarbon fractions unable to vaporize.
[39] For example, a decrease in the size fraction can result from an increase in smaller-sized particles and/or a decrease in larger-sized particles.

Differences in PM size distribution and count seem to depend on the fuels, engines, drive cycles, and exhaust sampling methods (Bunger et al. 2000; McCormick 2007). The latter factor is very important when reviewing study findings as no standard exhaust dilution and PM measurement methods exist. The large number of measurement approaches is currently a limiting factor to understanding the impact of biodiesel fuels on size particle size distribution, and this must be taken into consideration for the findings presented below.

4.2.2 Hydrocarbons

Hydrocarbon compounds are the product of incomplete combustion and the evaporation of hydrocarbon fuels. Biodiesel fuel combustion generates lower HC emissions than petroleum diesel (Morris et al. 2003; McCormick et al. 2003; Souligny et al. 2004; McCormick et al. 2006; McCormick 2007; Rosenblatt and Rideout 2007; Bennett et al. 2008), especially under low-load conditions where absolute total HC (THC) emissions are much higher (Lapuerta et al. 2008b). A linear relationship was determined between HC emissions reductions and biodiesel concentration in fuel (EPA 2002a; Durbin et al. 2007).

According to Lapuerta et al. (2008a), reductions in total HC emissions with the use of biodiesel fuels are attributable to better fuel combustion because of:

- the greater oxygen content;
- the higher cetane number;
- the higher *final* distillation points for diesel fuels;
- the advanced injection and combustion timing of biodiesel;
- lower sensitivity of THC emission detection devices for biodiesel oxygenated compounds; and
- sampling procedure artefacts with the higher molecular weight biodiesel fuel.

A few studies show increases or no significant differences in HC emissions with biodiesel fuel use (see Turrio-Baldassarri et al. 2004; Holden et al. 2006). These divergent results may be due to THC levels near the detection limits, as are typical of diesel engines (Lapuerta et al. 2008a).

Literature data for carbonyls and light aromatics are conflicting (e.g., Sharp et al. 200b; Correa and Arbilla 2006; Di et al. 2009; Ratcliff et al. 2010). It is generally proposed that differences between various studies are due to the composition of the vegetable or animal oils, combustion reactions, and the complexity of light HC formation in diesel engines fuelled with biodiesel.

4.2.3 Carbon monoxide

Biodiesel fuel yields lower CO emissions than petroleum diesel (Morris et al. 2003; McCormick et al. 2003; Souligny et al. 2004; McCormick et al. 2006; McCormick 2007; Rosenblatt and Rideout, 2007; Bennett et al. 2008). A linear relationship was determined between CO reductions and the biodiesel concentration (US EPA 2002a; Durbin et al. 2007).

Lapuerta et al. (2008a) considered a decrease in CO emissions, either linear or non-linear, as the general trend when substituting diesel with biodiesel fuel due to better fuel combustion, enabled by:

- the additional oxygen content and increased cetane number of biodiesel blends; and
- the advanced injection and combustion timing of biodiesel.

A few studies do not report a decrease in CO emissions (Turrio-Baldassarri et al. 2004; Mayer et al. 2005; Holden et al. 2006; Yang et al. 2007; CONCAWE 2010; Kousoulidou et al. 2010).

4.2.4 Nitrogen oxides

Based on correlations of emissions with biodiesel blend percentage in a 2002 literature review by the US EPA (2002a), only NO_X emissions showed an increasing trend with increasing biodiesel content in the fuel blends. Increases in NO_X emissions from the use of biodiesel fuels are reported mostly for heavy-duty engines or vehicles (McCormick et al. 2001; US EPA 2002a; Morris et al. 2003; Tsolakis 2006; Durbin et al. 2007). A review of biofuels by Verbeek et al. (2008) also mentioned that NO_X emissions increased in light-duty diesel vehicles (LDDVs), but results have been observed to differ for LDDVs (Kousoulidou et al. 2009; Pelkmans et al. 2009; CONCAWE 2010).

In a 2008 review, Lapuerta et al. concluded that a slight increase in NO_X emissions from biodiesel combustion is the most common observation in the scientific literature. Mueller et al. (2009) determined that the biodiesel NO_X increase results from a number of coupled mechanisms whose combined effects under different conditions may vary, depending on specific combustion and fuel characteristics. Based on these reports (Lapuerta et al. 2008a; Mueller et al. 2009), the biodiesel NO_X emission increase could likely be explained by factors leading to higher in-cylinder temperatures and greater thermal NO_X formation:

- injection-related issues;
- reduced soot radiant heat transfer;
- mean carbon chain length and saturation;[40]
- advances in combustion phasing; and
- mixtures closer to stoichiometric during ignition and in the standing premixed auto-ignition zone.

The effect of biodiesel use on NO_X emissions appears to vary between different feedstocks. For example, authors have noted that NO_X emissions are lowest for saturated and long-chained fatty acids (McCormick et al. 2003; Lapuerta et al. 2008a). However, Moser et al. (2009) observed that a reduction in double bond content of a biodiesel blend is not necessarily enough to result in significant NO_X emission differences. Engine model year does not appear (qualitatively) to have an impact on NO_X emissions (Hoekman et al. 2009).

[40] It is suggested that molecular differences influence NO_X indirectly by altering physical fuel properties such as density and compressibility, which in part influence injection and NO_X emissions.

Since the 2002 US EPA review, new engine or vehicle testing studies with biodiesel fuels have been completed. Yanowitz and McCormick (2009) analyzed the heavy-duty vehicle emissions data available regarding the effect of B20. Over all tests reported, a statistically significant average increase in NO_X emissions of 2% was found when using B20. However, when two-stroke engines were removed from the database, as they are not representative of the on-road fleet or newer engines, the increase compared to petroleum fuel was lowered to 1%. This value was not statistically significant.

4.3 Non-regulated Air Pollutants

Non-regulated air pollutants are not regulated by specific tailpipe emission standards, but can have an impact on air quality and human health. The following sections detail the emission changes of key non-regulated pollutants reported in the literature.

4.3.1 Sulphur compounds

Because biodiesel fuels are virtually sulphur free, reductions in sulphate emissions (a PM fraction) are expected. Sulphate (SO_4) emission was reduced when using biodiesel in comparison to conventional diesel fuel and results were generally similar across the heavy-duty engines tested (Souligny et al. 2004; Rosenblatt and Rideout 2007). SO_2 emissions were also reduced when comparing biodiesel to petroleum diesel emissions (Lue et al. 2001). In contrast, Cheung et al. (2009) observed higher sulphate emissions in a LDV powered by biodiesel fuel in comparison to petroleum diesel. This increase was presumably linked to biodiesel's greater dilution of lubrication oils in the engine, which can increase the sulphur content of the combustion mixture.

4.3.2 PAH, N-PAH, and MAH

Polycyclic aromatic hydrocarbons (PAHs) originate from PAHs initially present in the fuel and the products of incomplete combustion reactions (Ballasteros et al. 2010), and are present both in the particulate and gaseous phases in diesel exhaust emissions. PAH formation from combustion reactions is relevant for fuels such as biodiesel that do not contain any aromatic compounds, as reductions in PAHs and nitro-PAHs are expected. Sharp et al. (2000b) even reported that reductions appear proportional to the biodiesel content, although more recent reports suggest otherwise (e.g., Karavalakis et al. 2010a; Martini et al. 2007). The presence of the oxygenated ester group could contribute to the formation of PAH and oxy-PAH species (Karavalakis et al. 2010a), and potentially increase emissions of certain toxic compounds (Martini et al. 2007).

Low molecular weight PAHs are generally found in higher concentrations, but the higher molecular weight PAHs (e.g., benzo[a]pyrene, benzo[a]anthracene) associated with particles contribute more to the total carcinogenic potential of emitted PAHs (Ravindra et al. 2008 and Li et al. 2003 *in* Ballasteros et al. 2010). Ratcliff et al. (2010) observed that emissions of 3 and 4-ring PAHs decreased substantially with biodiesel use in a heavy-duty diesel engine. In LDDVs, Karavalakis et al. (2010a) observed that heavier PAHs were reduced for all biodiesel fuel and

test cycle combinations in comparison to low sulphur diesel. However, the greatest impact was observed on low molecular weight PAHs, which increased with the use of biodiesel. Zou and Atkinson (2003) also tested biodiesel fuels in two LDDVs, targeting 18 PAHs. They observed mostly non-linear reductions of gaseous PAH emissions with the use of biodiesel compared to petroleum diesel. As for the particulate phase PAH emissions, no clear trend was observed. Zou and Atkinson (2003) concluded that from a health standpoint, biodiesel fuel blends did not significantly lower the emissions of the more toxic particulate PAHs compared to petroleum diesel.

Although the feedstock origin and fuel type are important factors, PAH emissions are also dependent on the test cycle and various engine parameters (Zou and Atkinson2003; Karavalakis et al. 2010b). Notably, the level of unsaturation of the fuel, cold-starts, and low speed and load conditions appeared to favour the formation and emission of PAHs (Karavalakis et al. 2010b).

4.3.3 Elemental carbon, organic carbon, and soluble organic fraction

Elemental carbon (EC) is associated with soot formed during combustion while organic carbon (OC) is associated with condensed phase compounds on the soot core. The latter compounds are generally soluble in organic solvents and are referred to as the soluble organic fraction (SOF) of particulate matter. However, data reported for SOF and OC are not equivalent since OC reports only the carbon while SOF reports total mass for the soluble compounds.

Most studies concur that the EC fraction is reduced with biodiesel fuels, while the SOF increases (Bunger et al. 2000; Sharp et al. 2000a; Souligny et al. 2004; Zhu et al. 2010). The increase in SOF is probably due to a lower volatility of the unburned biodiesel hydrocarbons, which favours their condensation and adsorption on particle surfaces (Dwivedi et al. 2006; Lapuerta et al. 2008a; Karavalakis et al. 2009c; Zhu et al. 2010).

The ratio of organic to elemental carbon varies considerably depending on vehicle technology, test mode, and fuel type (Holden et al. 2006). Zhang et al. (2009) observed that the differences in the OC/EC fractions when using biodiesel fuel, at all engine speed and load test conditions, mainly resulted from a sharp decrease in EC, demonstrated by a larger OC/EC fraction for B20 in comparison to petroleum diesel. Higher OC emissions were also observed by Durbin et al. (2000) in most vehicles tested with biodiesel fuel blends.

4.3.4 Volatile organic compounds

Volatile organic compounds (VOCs) are organic chemicals that include aldehydes, ketones, and other toxic hydrocarbons. Generally, the makeup of aldehyde emissions is similar for petroleum or biomass-based fuels. Low molecular weight carbonyl compounds (CCs), such as formaldehyde, acetaldehyde, and acrolein make up around 90% of the emitted aldehydes and ketones (Sharp et al. 2000b; Turrio-Baldassarri et al. 2004; Holden et al. 2006; Peng et al. 2007; Rosenblatt et al. 2008; Karavalakis et al. 2010b).

Variations in emissions differ considerably among the individual aldehyde compounds, but most seem to decrease with biodiesel fuel use in heavy-duty diesel applications (Sharp et al. 2000b; US EPA 2002a; Souligny et al. 2004), especially with biodiesel blends greater than B20 (Rosenblatt and Rideout 2007; Rosenblatt et al. 2008; Di et al. 2009).

Many studies report increases, decreases or insignificant differences in VOC emissions with the use of biodiesel fuels (Lapuerta et al. 2008a). Some authors have proposed that differences between studies may be accounted for by different engine technologies, testing procedures, and fuel chemistry (Graboski et al. 2003; Holden et al. 2006; da Silva and Pereira 2008). For example, Ratcliff et al. (2010) observed that the overall effect of biodiesel on carbonyls was minimal. The absence of a considerable effect in this study was probably due to the use of a modern engine equipped with exhaust gas recirculation (a device that reduces combustion temperatures) or to low-speed and low-load operation conditions.

Also, Guarieiro et al. (2008) demonstrated that sampling methods could significantly influence emission results of CC. Lastly, the presence of impurities or additives (e.g., ethanol) in the biodiesel fuel may influence aldehyde emissions (Pang et al. 2006).

4.3.5 Ozone forming potential

Ground-level ozone formation depends on a multitude of chemical and physical factors such as ambient temperature and insolation, atmospheric concentrations of NO_X and VOCs, and the atmospheric chemical reactivity of different VOCs. Studies have reported that the use of biodiesel blends can lower the ozone forming potential by up to 50% (Souligny et al. 2004; Dincer 2008). The reduction in ozone potential is attributed to lower HC emissions, notably aldehydes, and the lower reactivity of hydrocarbon compounds emitted from biodiesel blends compared to petroleum diesel (Souligny et al. 2004; Dincer 2008; Peng et al. 2008; He et al. 2009).

4.3.6 Metals

Diesel tailpipe emissions are reported to have relatively low metal concentrations in comparison to other vehicle sources (Schauer et al. 2006) and the individual emission rates of metals generally show great variations within classes of vehicles and test cycles.

The metal content in diesel, biodiesel and lubricating oil plays a significant role in metal exhaust emissions (Dwivedi et al. 2006; Wang et al. 2003). Greater concentrations of metals in the liquid fuel samples coincide with higher concentrations in the exhaust emissions.

Many metals in tailpipe PM emissions are expected to be from lubricating oil additives and engine wear fragments accumulated in the engine oil (Lough et al. 2005; Schauer et al. 2006). The presence and concentration of metals in the engine oil is important because biodiesel has

greater engine oil dilution properties (Dwivedi et al. 2006), which could lead to more engine oil-borne metals in combustion emissions.[41] However, overall, Dwivedi et al. (2006) concluded that biodiesel is likely to reduce metal exhaust emissions.

4.3.7 Other

In a recent publication by Ratcliff et al. (2010), products of incomplete combustion unique to biodiesel were recognized as FAME fragments. Methyl acrylate and methyl 3-butenoate were detected as significant species in B20 and B100 exhaust. However, because this is the only study reviewed that has identified these FAME fragments (limited to one test engine and one soya-based biodiesel fuel), these findings cannot be generalised and their significance is unclear at this time. More research is necessary to better characterize potential emissions of FAME fragments from engines or vehicles operating on biodiesel fuels and their impacts on air quality.

4.4 After-Treatment Devices

After-treatment devices can, in principle, alter the size, density, oxidative potential, and composition of particles from diesel exhaust emissions (Maricq 2007; Biswas et al. 2009). The mechanisms by which these devices lower emissions differ greatly and are based on specific physical and chemical reactions prior to or following fuel combustion. It is assumed that diesel vehicles will only be able to meet the 2010 tailpipe standards with the use of after-treatment devices (Maricq 2007; Peterson et al. 2009). The effects of after-treatment methods on regulated PM and gaseous emissions (Frank et al. 2004), ultrafine particle size distributions (Frank et al. 2004), and unregulated emissions (Tang et al. 2007) have been shown to be greater than the effect of changing fuel type.

4.4.1 Exhaust gas recirculation

Exhaust gas recirculation (EGR) serves to limit the generation of NO_X by recirculating a portion (25% to 40%) of an engine's exhaust gas back to the engine cylinders. The physical properties of diesel and biodiesel soot and the reactivity of the soot are altered as a consequence of EGR (Al-Qurashi and Boehman 2008). A reduction of both regulated and unregulated emissions can be achieved, especially when coupled with a diesel particulate filtration device (Muncrief et al. 2008).

Because of its oxygen content and subsequent better combustion, biodiesel appears to be less impacted by EGR than ULSD,[42] resulting in overall better emission reductions (Tsolakis et al. 2006; Park et al. (2009).

4.4.2 Diesel particulate filtration

Diesel particulate filters (DPFs) were developed to reduce diesel exhaust particulate matter emissions by essentially physical trapping. Various materials and pore sizes are used to capture

[41] http://www.biodiesel.org

[42] By lowering the intake of ambient air and oxygen, EGR can increase PM emissions when used alone.

the solid fraction of diesel particulate matter and DPFs can attain filtration efficiencies above 90%.

Mayer et al. (2005) observed that there is no discernable difference in DPF efficiency between petroleum fuels and biodiesel blends. However, some authors report that biodiesel fuels have a beneficial effect on DPF operability since particles from biodiesel combustion are more easily oxidized (Jung et al. 2006), facilitating DPF regeneration.

The combination of EGR and DPF with the combustion of biodiesel fuels can reduce both NO_X and PM emissions (Muncrief et al. 2008). The combination of EGR and biodiesel resulted in a beneficial effect on DPF regeneration and operation. The decrease in PM and corresponding increase in the NO_X/PM ratio is favourable for DPF operability, because proportionally more NO_2 (an oxidant) is available for DPF regeneration.

4.4.3 Diesel oxidation catalysts

Catalytic converters primarily reduce CO and HC emissions, but they can also generate NO_2 for oxidizing soot on a continuous basis or for improving the low-temperature performance of selective catalytic reduction (SCR) devices (Johnson 2011). They also serve the purpose of protecting other after-treatment devices from excessive PM or unburned fuel (Theinnoi et al. 2008).

Diesel oxidation catalysts (DOCs) are effective with conventional diesel fuel and alternative fuels like biodiesel up to B20 (provided the biodiesel meets biodiesel and sulphur content specifications)[43] and can reduce CO and HC by around 90% (MECA 2009; Peterson et al. 2009). Emission levels of carbonyls, benzene, PAHs, n-PAHs, and EC and OC combined mass were similar for B20 and ULSD in a heavy duty diesel engine equipped with a DOC (Tang et al. 2007). Catalytic conversion efficiency of total particulate matter is generally improved with increased biodiesel content. The fuel-derived volatile organic fraction (VOF) associated with particulates from unburned biodiesel appears to be easier to oxidize than particulates associated with unburned diesel fuel. Less carbon soot in PM emissions of biodiesel fuels could also create a more favourable environment for treatment by a DOC.

Issues have been raised regarding the use of biodiesel and DOC durability. It appears that operating on biodiesel meeting ASTM specifications for allowable content of alkali and alkaline metal impurities could possibly deactivate the DOC. Increased oil consumption and oil dilution with the use of biodiesel could also contribute to increased ash in the flue gas reaching the DOC (McCormick et al. 2009; Williams et al. 2011).

[43] www.meca.org/cs/root/diesel_retrofit_subsite/what_is_retrofit/what_is_retrofit. Accessed April 15, 2011.

4.4.4 Selective catalytic reduction

Selective catalytic reduction (SCR) technology is the leading NO_X emission control system for heavy-duty and light-duty diesel applications (Johnson 2011) and it is expected to be used by the majority of new truck manufacturers to meet the US EPA 2007/2010 on-highway regulation (MECA 2009). SCR is usually applied in combination with a DOC and a DPF.

SCR was designed to reduce NO_X emissions by 70-90% and in the future could achieve 90-95% NO_X abatement efficiency (Johnson 2011). SCR can also lower a range of regulated and unregulated pollutants. Authors have not reported a significant change in the performance or efficiency of SCR systems when increased levels of biodiesel fuel were used (Rosenblatt et al. 2008; Walkowicz et al. 2009).

Winther (2009) notes that SCR-equipped engines can lead to greater differences in NO_X emissions between diesel and biodiesel-fuelled vehicles compared with older engines without SCR. This is apparently the result of the current SCR urea dosing systems not being optimized for the use of biodiesel (Winther 2009), highlighting the importance for manufacturers to consider biodiesel content when developing after-treatment devices.

4.4.5 Lean NO_X traps or NO_X adsorber catalysts

Lean NO_X trap technology is currently the leading method for reducing NO_X emissions in smaller passenger cars and applications where space is limited or the use of a diesel exhaust fluid is difficult. It is often used in combination with DPF, DOC, or SCR after-treatment devices (RWDI 2009; Johnson 2011). NO_X reductions around 70% are theoretically achievable with this type of device (Johnson 2011).

Tatur et al. (2008) tested a NO_X adsorber catalyst (NAC) in combination with other emission control systems on a light-duty engine and a LDDV fuelled with ULSD and soybean-based biodiesel blends (B5 and B20). Engine testing showed that the calibration and the parameters settings of the NAC were essentially identical for ULSD and B20, signalling that the different fuels had no impact on the emission controls. Optimal calibration of engine-out emissions with biodiesel even allowed for more stable and lower NO_X emissions with the biodiesel blends compared to ULSD (Tatur et al. 2008).

4.5 LDDV Testing at Environment Canada

The Emissions Research and Measurement Section (ERMS) at Environment Canada, in collaboration with Health Canada, tested light-duty diesel vehicles operating on different biodiesel blends (B5 and B20) and diesel fuels to evaluate their impact on exhaust emissions (Meloche et al. 2010). These tests were conducted to fill existing data gaps pertaining to the impact of biodiesel fuels on light-duty diesel vehicle exhaust emissions. Three recent vehicles equipped with a DOC or a catalyzed DPF were tested on a total of eight (8) different test fuels, which were selected to reflect fuels that may be used in LDDVs under Canadian conditions.

Testing was conducted on a chassis dynamometer over the Federal Test Procedure City Test (FTP-75) and the US06 Supplemental Federal Test Procedure (US06) at standard and cold temperatures, 24°C and -18°C, respectively. Exhaust emission data was obtained for regulated and unregulated pollutants, including CO, THC, PM, NO_X, SO_2, OC, EC, PAH, n-PAH, and metals.

The baseline fuel for this project was a seasonal commercially available ULSD (sULSD, known as No. 2), which also served as the blending stock for most of the biodiesel blends. For the cold temperature portion of the program, a winterized version of the seasonal ULSD was used, which in contrast to the standard ULSD fuel had a lower cloud point, density, and cetane number, as well as a slightly different aromatic and saturate content.

Based on a simple Student t-test at 95% confidence interval, several statistically significant changes were observed when comparing B5 and B20 biodiesel blends to ULSD. For example, PM was found to decrease in most cases with biodiesel soy or canola blends, while NO_X generally increased. However, overall, the emissions data collected as part of this study did not show many consistent results across vehicles and test cycles when comparing the various biodiesel blends with ULSD. Regarding air toxics, it was not considered relevant to report the observed changes in emissions of species like VOCs and PAHs even if results appeared statistically significant, because emission levels were close to the instrumentation detection limits, in some instances leading to high standard deviations and elevated coefficients of variance (Meloche et al. 2010).

PM size and mass measurements were obtained by means of an Engine Exhaust Particle Sizer (EEPS). The results varied according to the vehicle type, after-treatment devices and test cycle. For example, across all fuel blends tested, the DPF-equipped vehicle generally showed $PM_{2.5}$ reductions 81 times greater than non DPF-equipped vehicles and TPM was generally reduced by a factor of 5 to 10. The differences in PM mass and number emissions between standard fuels and biodiesel blends were minor and/or insignificant over both test cycles. Notably, with increasing biodiesel blend levels:

- one vehicle showed reductions in particle number and mass emission rates;
- biodiesel was observed to lower the average size of the emitted particles; and
- over the US06 cycle, a decrease in the number of ultrafine particles (UFPs) was detected.

In light of the data gathered as part of this study on LDDV exhaust emissions, the impacts due to temperature and emission control technologies were more consistently significant than the impacts related to fuel type (Meloche et al. 2010).

4.6 Summary of Impacts of Biodiesel Use on Vehicular Emissions

Biodiesel fuel components are rich in different chemical structures, resulting in complex chemical reactions during combustion. In addition, the combustion mechanisms of biodiesel fuels are still not completely understood. Compared to alcohol-type fuels (e.g., ethanol, butanol), the absolute concentration and nature of engine and exhaust emissions of biodiesel may prove harder to predict.

Nonetheless, based on the literature review, general conclusions can be drawn with regards to the impact of biodiesel fuels on vehicle exhaust emissions. The following points are considered particularly important for the current assessment. Compared to ULSD, biodiesel is expected to result in:

- significant reductions in PM, CO, and HC emissions;
- no net impact or slight increase in NO_X emissions;
- reduction in total VOC emissions with large variations between chemical species;
- downward shift in fine PM size profile, without any agreement as to the exact cause(s) (e.g., an uneven reduction of large particles or an increase in small particles);
- increase of the PM organic carbon fraction;
- reduction of PAH and n-PAH emissions; and
- reduction of ozone reactivity.

In addition:

- After-treatment devices are effective at reducing emissions of various pollutants, but any significant difference between biodiesel and petroleum fuels remains to be determined.
- Particulates from biodiesel-fuelled engines appear to be more easily oxidized, which could increase after-treatment device efficiencies.
- Engine parameters, test cycles characteristics, and the quality of the test fuels have a significant influence on emissions.

Last, although exhaust emissions from vehicles using different types of biodiesel fuels, either from animal or vegetable origin, have been observed to differ, these variations between esters remain small and secondary to general trends observed when comparing biodiesel to petroleum fuels (BIO Intelligence Service 2010).

Table 4-1 recaps the quantitative changes in heavy-duty diesel engine and vehicle emissions reported by the US EPA (2002a, 2009), the ERMS (Rosenblatt and Rideout 2007a; Rosenblatt et al. 2008), and Yanowitz and McCormick (2009).

Table 4-1 Summary of changes in regulated exhaust emissions with the use of B20 blends compared to petroleum diesel fuels according to different sources

Pollutant	US EPA (2002)	ERMS (2007a; 2008)	Yanowitz & McCormick (2009)	US EPA (2009)		
				Light load	*Soy biodiesel*	*All biodiesel*
NO_X	2.0%	B5: 0.7% B20: 3.0%	1%	-1.0%	2.2%	2.0%
PM	-10.1%	B5: -8.9% B20: -25.7%	-17%	-19.0%	-15.6%	-13.6%
HC	-21.1%	B5: -12.1% B20: -18.7%	-16%	-14.2%	-13.8%	-18.7%
CO	-11.0%	B5: -12.5% B20: -12.4%	-16%	-9.9%	-14.1%	-13.5%

The changes in exhaust emissions in Table 4-1 represent various results from key studies. In the following chapters, Canadian mobile emission impacts of biodiesel use are modelled using the MOBILE6.2C model. Background information regarding MOBILE6.2C, the basis for the development of biodiesel emissions factors within the model, and the modelling approach used are presented in Chapter 5.

4.7 Uncertainties

Understanding the impacts of biodiesel fuels on exhaust emissions is limited by multiple factors, including variable sampling methods and test procedures, as well as insufficient testing data. Sampling uncertainties are more important for VOCs, ultrafine particles and other pollutants that are emitted in minute amounts. For example, properly detecting and measuring pollutants such as UFP are challenging. Similarly, identifying sampling methods that are as relevant for petroleum and bio-based fuels remains a challenge for hydrocarbon species.

Gaining a better understanding of the effect of emission control systems and newer engine technologies on emissions is also fundamental to evaluating the general impact of using petroleum and biodiesel fuels on the emission inventory. As noted in Section 4.4, after-treatment devices such as EGR, DPFs, and catalysts are expected to have a considerable impact on vehicle emissions, notably NO_X and PM, which will outweigh any impact on emissions from the use of biodiesel fuels. It remains to be proven if the operation of after-treatment devices is significantly impacted by biodiesel fuels or if the presence of after-treatment devices in newer vehicles will render any impact from the use of biodiesel fuels insignificant. For example, Luján et al. (2009) compared emissions from petroleum fuels and biodiesel blends in a diesel engine, with and without after-treatment devices. They noted that the high efficiency of after-treatment devices like DPFs and oxidizing catalysts masked or overshadowed the noticeable advantage of biodiesel fuels in terms of particulate, CO, and HC emissions.

Similarly, the ERMS observed that the impacts of temperature and emission control technology on LDDV exhaust emissions appear more important than the combustion of the various test fuels (Meloche et al. 2010). As new diesel engines/vehicles and diesel fuels will have to meet very strict emission standards, it may be that the presence of low blends of biodiesel will not have any discernable impact on emissions and ambient air quality.

Lastly, Yanowitz and McCormick (2009) noted that the test cycle used for the certification of new engines might not be representative of actual operations. Since the impact of biodiesel fuel on pollutant emissions was found to be highly dependant on engine cycle (e.g., load, speed), current comparisons to petroleum diesel fuels might not be relevant to actual on-road use.

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Chapter 5. Impacts of Biodiesel on Canadian On-road Mobile Emissions

This Chapter outlines the methodology adopted to assess emissions from on-road mobile sources using ULSD or biodiesel blends and presents results from the MOBILE6.2C modelling tool, the Canadian version of the US EPA's MOBILE6.2 model. MOBILE6.2C is widely accepted as the best available model for the estimation of emission factors from on-road vehicles in Canada and the most recent version captures the effects of the use of renewable fuels.

As reviewed in Chapter 4, the impact of biodiesel fuels on exhaust emissions depends on many factors, such as engine technology, engine calibration, drive cycle, and ambient temperatures. MOBILE6.2C integrates the available information regarding these variables to generate pollutant emissions factors and, in combination with data of vehicle kilometres travelled (VKT), provides national emission estimates for on-road vehicles.

MOBILE6.2C is used to quantify the impacts from the use of various biodiesel fuel blends on overall exhaust emissions from the Canadian on-road vehicle fleet. Different modelling years are considered for the selected scenarios to reflect changes in vehicle technology and emissions standards.

5.1 Methodology and Inputs of MOBILE6.2C

Emission estimates from on-road vehicles are generated using a bottom-up approach, where emission factors are developed for individual vehicle classes and activity data are applied to them.

5.1.1 Pollutants of interest

This analysis focuses on emissions of the listed CACs, mobile source air toxics (i.e., benzene, 1,3-butadiene, formaldehyde, acetaldehyde, acrolein, PAHs), and metals. Greenhouse gases are not considered.

5.1.2 Emission factors

MOBILE6.2C generates emission factors in grams per mile or kilometre driven for on-road vehicles. Technical information regarding these estimates and guidance documents are available from the US EPA (US EPA 2002, 2003, 2004; US EPA and E.H. Pechan & Associates, Inc. 2008).

Estimates are provided for the exhaust, evaporative and fugitive components of all relevant pollutants. CACs, benzene, 1,3-butadiene, formaldehyde, acetaldehyde, acrolein, and methyl tertiary butyl ether are explicitly modelled. TPM is estimated from MOBILE's PM_{10} estimate,

using the US EPA's *PM Calculator* tool.[44] For evaporative and exhaust emission compounds not explicitly modelled by MOBILE6.2C, emissions factors or air toxic ratios can be used (e.g., as fractions of emitted VOCs, total organic gases, or PM) (US EPA 2002; US EPA and E.H. Pechan & Associates, Inc. 2008). For the current assessment, default MOBILE values are used and are not altered by the use of biodiesel fuel.

The full list of pollutants estimated for this analysis are presented and discussed in Section 5.2.

5.1.3 MOBILE6.2C update – renewable fuels

The Canadian version of the MOBILE model was revised by Environment Canada in 2010 to improve the modelling capabilities of renewable fuels, including biodistillates (i.e., biodiesel and renewable diesel). The data and methods for five separate biodiesel feedstocks were incorporated: canola/rapeseed, soybean, all-plant composite,[45] yellow grease (i.e., used cooking oil) and animal fat. For this assessment, canola is assumed to be the single biodiesel feedstock used by the Canadian on-road fleet.

The impacts of fuels on emissions are supported by a database used by the US EPA for its *Renewable Fuels Strategy* (RFS2)[46], in concert with test results from Environment Canada's Emissions Research and Measurement Section (ERMS), and from recent data collected by the California Air Resources Board (CARB). The final database used to develop biodiesel correction factors for MOBILE6.2C consisted solely of heavy-duty diesel engine test results.

The biodiesel fuel adjustment factors programmed directly into MOBILE6.2C are i) based on a percent reduction in diesel exhaust emissions per percent of biodiesel content of the fuel and ii) assume that biodiesel impacts are linear with biodiesel content. The method also assumes no impact of biodiesel on emissions from HDDVs meeting final 2010 model-year exhaust emission standards[47] and no impact of biodiesel on emissions from LDDVs.[48]

5.1.4 MOBILE6.2C vehicle fleet profile

MOBILE6.2C covers 28 distinct vehicle classes based on gross vehicle weight rating (GVWR) and fuel type. These 28 classes are aggregated based on GVWR for the purposes of this report:

[44] http://www.epa.gov/ttnchie1/software/pmcalc/

[45] Results for soybean and canola/rapeseed feedstocks were not statistically different in terms of the biodiesel impact on exhaust emissions, so the *all-plant composite* feedstock was created, combining data from both canola/rapeseed and soybean-based biodiesels.

[46] The US EPA dataset is outlined in Appendix A of the *Draft Regulatory Impact Analysis: Changes to Renewable Fuel Standard Program* (2009).

[47] This is due to the absence of sufficient test data on vehicles meeting 2010 model year standards. Emissions from 2010 model year and later vehicles are expected to be substantially lower and any impact of biodiesel use would be much lower in absolute values. The most recent model year diesel engine in the underlying database was 2006

[48] The biodiesel emissions factor corrections developed for HDDVs are not applicable to LDDVs. No correction was assumed for the light-duty fleet with respect to biodiesel use, in part because data on the impact of biodiesel on representative North American LDDVs are limited. This is expected to have minimal impact because the LDDV fleet represents only about 2% of vehicles on the road in Canada.

- HDDV: Heavy-duty diesel-powered vehicles (8501 lbs. and greater, GVWR)
- HDGV: Heavy-duty gasoline-powered vehicles (8501 lbs. and greater, GVWR)
- LDDT: Light-duty diesel-powered trucks (0-8500 lbs., GVWR)
- LDDV: Light-duty diesel-powered vehicles (Passenger Cars)
- LDGT: Light-duty gasoline-powered trucks (0-8500 lbs., GVWR)
- LDGV: Light-duty gasoline-powered vehicles (Passenger Cars)
- MC: Motorcycles (all motorcycles are assumed to be gasoline-powered)

Accurately defining the on-road fleet by model-year, fuel type and gross vehicle weight rating is a complicated task. While no single source comprehensively meets MOBILE's input data requirements, an effort was made to analyze and compile a trend of data from various sources (DesRosiers Automotive Consultants; R. L. Polk & Co.; Motorcycle and Moped Industry Council; NRCan; Statistics Canada; Sanford Evans Research Group; and Stewart-Brown Associates). However for the current project, due to insufficient data, the vehicle populations are left unchanged from previous Environment Canada inventories based on provincial/territorial populations for 2004 (for both GHGs and air pollutant emissions) (Environment Canada 2005). More recent and detailed information is used for Ontario and British Columbia due to the more robust data made available from their Inspection and Maintenance (I/M) programs.

Future projection of the fleet is determined based on NRCan's *Canada's Energy Outlook: The Reference Case 2006*, from which forecasts of gasoline and diesel use by province/territory to 2020 are translated into growth rates.

5.1.5 Meteorological data

Ambient temperature has an impact on emissions and consequently meteorological conditions are considered in MOBILE. Meteorological data are provided by Environment Canada's Meteorological Service of Canada (e.g., monthly and minimum temperatures). Meteorological data for 2006 were selected for emissions modelling to concur with the air quality modelling meteorology (see Chapter 6).

5.1.6 Fuel characteristics and renewable fuels scenarios

The reference fuel is assumed to be ULSD for the entire 2006 calendar year. On-road diesel and gasoline sulphur levels are defined in the report *Sulphur in Liquid Fuels* (Environment Canada 2010).[49] For this analysis, ULSD is modelled at 10 ppm sulphur content, which is assumed to be more representative of current fuels.[50] All other fuel characteristics, such as Reid Vapour Pressure (RVP) and oxygenates, are based on the report *Emissions of Air Toxics from on-Highway Sources in Canada* (SENES Consultants and Air Improvement Resources 2002).

[49] The October 1st to April 30th period was assigned winter fuel properties, while the May 1st to September 30th period was assigned summer fuel properties (or special Reid Vapour Pressures where applicable).

[50] In consultation with experts from Environment Canada's Oil, Gas and Alternative Energy Branch and from industry represented by the Canadian Petroleum Products Institute (CPPI).

MOBILE runs for the following canola biodiesel scenarios are based on year-round use: B0, B2, B5, B10, and B20. While it is not expected that higher blends of biodiesel will be used during winter months, the model is run for all these cases for every month of the year. This allows for the creation of many different scenarios.

5.1.7 Speed profiles and activity level

A speed profile allows the MOBILE model to allocate the amount of vehicle activity (distance traveled) by average speed, for each hour of the day and for roadway types. A profile is developed for each province and territory (Delcan Corporation and iTRANS Consulting Inc. 2003).

The activity level in MOBILE is expressed in vehicle miles traveled (VMT), which are converted to VKT externally to the model. Relevant VMT or VKT estimates require suitable fleet characterization data, including the number and age of vehicles in each class, and the average distance traveled for each vehicle class and age, on a yearly basis. To account for regional differences in accumulation rates of kilometres travelled, the VKT are normalized, generally using assumed fuel consumption rates and fuel availability statistics by province or territory.[51]

Table 5-1 shows the absolute values and percentages of the activity data (vehicle populations, VKT, and fuel use) used in the current study for Canada for the year 2006.

Table 5-1 Percentage of Canadian on-road fleet composition, VKT, and fuel use for different vehicle classes for 2006

Vehicle class	% of fleet	% of total VKT	% Diesel fuel use	% gasoline fuel use
Heavy-duty diesel vehicles	4	10	94	
Heavy-duty gasoline trucks	2	2		7
Light-duty diesel trucks	1	2	5	
Light-duty diesel vehicles	1	1	1	
Light-duty gasoline trucks	36	37		48
Light-duty gasoline vehicles	53	48		44
Motorcycles	2	<1		<1
% Total*	**100**	**100**	**100**	**100**
Absolute total	**20,959,517 vehicles**	**373,810,670,589 km**	**15,467 ML**	**38,520 ML**
*** Total may not add to 100 due to rounding.**				

[51] Fuel availability is taken from the *Report on Energy Supply-Demand* (Statistics Canada, annual).

5.1.8 Regions

Canada is divided into 19 distinct regions for the current analysis. Each province and territory is its own region except for British Columbia, Ontario and Québec, which are further divided by the so-called *Phases* of their inspection and maintenance programs or economic regions. Québec is divided into two regions: inside and outside the Quebec-Windsor corridor. British Columbia is divided into 3 regions according to the *Air Care* program (Phases 1, 0, and Z). Ontario is divided into four regions, aligned with the Phases of the *Drive Clean* program (Phase1, Phase2, Phase3, and Phase0).

5.2 Emission Estimates

The years 2006, 2010, 2015 and 2020 are modelled for various biodiesel scenarios. Emission impacts are assumed for HDDVs only. The only variable for scenarios within each year investigated is the volume of biodiesel content. Changes in emissions between years are also due to fleet turn-over and stricter regulatory emissions standards. Table 5-2 shows the percent change in fleet-average heavy-duty diesel emission factors estimated for B20 compared to B0 (i.e., ULSD), for 2006, 2010, and 2020 calendar years.

Overall, exhaust emissions of PM, air toxics, PAHs, CO, and total VOCs are expected to decrease, while it is estimated that NO_X emissions will increase with the use of biodiesel. Estimates indicate that the impacts of biodiesel diminish into the future, as the fleet population is comprised increasingly of newer vehicles meeting more stringent emission standards (newer vehicles are assumed not to be impacted by biodiesel use). Some pollutants, such as the fugitives (brake- and tire-wear) and metals are not sensitive to fuel characteristics and display no change. In addition, biodiesel use is not expected to impact NH_3 and SO_X emissions.

The emission factors in Table 5-2 represent national values. However, a number of region-specific variables (e.g., age profile of the fleet, meteorological data, fuel characteristics, and average vehicle speed per roadway type) influence emission factor output from MOBILE. These variables lead to region-specific emission factors (results not shown) that are used in the modelling.

Between the years investigated in this study, the absolute fleet emissions vary. All else being equal, emissions should increase as the activity level (VKT) increases. However, fleet turn-over, having more vehicles meeting newer and more stringent emission standards, coupled with the use of alternative fuels result in a decrease in emissions.

Table 5-2 Percent change in Canadian fleet-average heavy-duty diesel emission factors considered in MOBILE6.2C for B20 compared to B0 for 2006, 2010 and 2020

Pollutant	2006	2010	2020	Pollutant	2006	2010	2020
1,3-Butadiene	-18	-14	-3	NH_3	0	0	0
Acenaphthene	-13	-13	-8	n-Hexane	-18	-14	-3
Acenaphthyle	-12	-12	-9	NO_X	4	4	3
Acetaldehyde	-18	-14	-3	Naphthalene	-13	-12	-9
Acrolein	-18	-14	-3	Nickel	0	0	0
Arsenic	0	0	0	Ocarbon	-13	-12	-9
Benzene	-18	-14	-4	Phenanthrene	-13	-13	-9
Benzo[a]anthracene	-13	-12	-10	$PM_{10\ brake}$	0	0	0
Benzo[a]pyrene	-14	-12	-9	$PM_{10\ exhaust}$	-13	-12	-9
Benzo[b]fluoranthene	-12	-13	-11	$PM_{10\ tire}$	0	0	0
Benzo[ghi]perylene	-12	-13	-11	$PM_{2.5\ brake}$	0	0	0
Benzo[k]fluoranthene	-12	-13	-11	$PM_{2.5\ exhaust}$	-13	-12	-9
Chromium Cr^{3+}	0	0	0	$PM_{2.5\ tire}$	0	0	0
Chromium Cr^{6+}	0	0	0	Propionaldehyde	-18	-14	-3
Chrysene	-13	-14	-9	Pyrene	-13	-12	-9
CO	-11	-10	-7	SO_2	0	0	0
Ecarbon	-13	-12	-9	SO_4	0	0	0
Ethylbenzene	-18	-14	-3	Styrene	-18	-14	-3
Fluoranthene	-13	-13	-9	THC_{total}	-18	-14	-3
Fluorene	-13	-12	-9	Toluene	-18	-14	-3
Formaldehyde	-18	-14	-3	TPM_{brake}	0	0	0
Indeno(1,2,3-cd)pyrene	-12	-23	0	TPM_{tire}	0	0	0
Manganese	0	0	0	VOC_{total}	-18	-14	-3
Mercury	0	0	0	Xylene	-18	-14	-3

5.2.1 Effect of biodiesel on National emission results

The following sections present the results for the main pollutants of concern, including CACs and toxics. Only results for B0, B5, and B20 are displayed. Table 5-3 presents both the absolute emissions and the relative contribution of HDDVs to all on-road emissions, as a percentage.[52] Because the impact of biodiesel is only assessed for HDDVs, the MOBILE results assume that, for a given scenario year, the net impact on all vehicle emissions is solely due to the emissions change from HDDVs.

[52] As noted in Table 5-1, the heavy-duty diesel vehicles account for roughly 4% of the total on-road fleet by vehicle population, 10% of the total fleet VKT, and 94% of the diesel fuel used.

Table 5-3 PM_{10}, NO_X, VOC, and CO emissions from HDDVs, by year and biodiesel content

Year	Descriptor	B0	B5	B20*
	PM_{10}**			
2006	HDDV emissions (tonnes)	6,955	6,734	6,078
	Contribution to on-road (%)	*57*	*57*	*54*
2010	HDDV emissions (tonnes)	4,197	4,068	3,679
	Contribution to on-road (%)	*49*	*48*	*45*
2020	HDDV emissions (tonnes)	890	871	813
	Contribution to on-road (%)	*21*	*20*	*19*
	NO_X			
2006	HDDV emissions (tonnes)	278,474	280,979	288,498
	Contribution to on-road (%)	*53*	*53*	*53*
2010	HDDV emissions (tonnes)	196,172	197,929	203,201
	Contribution to on-road (%)	*49*	*49*	*50*
2020	HDDV emissions (tonnes)	54,393	54,776	55,925
	Contribution to on-road (%)	*38*	*38*	*38*
	Total VOC***			
2006	HDDV emissions (tonnes)	10,544	10,076	8,669
	Contribution to on-road (%)	*4*	*3*	*3*
2010	HDDV emissions (tonnes)	8,523	8,235	7,371
	Contribution to on-road (%)	*4*	*4*	*3*
2020	HDDV emissions (tonnes)	6,621	6,568	6,392
	Contribution to on-road (%)	*6*	*6*	*5*
	CO			
2006	HDDV emissions (tonnes)	60,578	58,951	54,072
	Contribution to on-road (%)	*1.4*	*1.4*	*1.2*
2010	HDDV emissions (tonnes)	41,095	40,044	36,893
	Contribution to on-road (%)	*1.2*	*1.2*	*1.1*
2020	HDDV emissions (tonnes)	13,114	12,900	12,259
	Contribution to on-road (%)	*0.5*	*0.5*	*0.5*

* The equivalent values in percent change are presented in Table 5-2.

**PM_{10} values and changes representative of all $PM_{exhaust}$, i.e. $PM_{2.5}$ and TPM.

*** As representative of hydrocarbons (VOC, n-Hexane, THC), benzene (ethyl benzene, styrene, toluene, xylene), and the aldehydes (acetaldehyde, acrolein, formaldehyde and propionaldehyde). [53]

[53] Note that there is some degree of variability between the representative species and the larger set of pollutants that they represent.

As shown by their contribution to on-road emissions, HDDVs influence overall on-road PM and NO_X emissions considerably. The contribution of HDDVs is quite limited for emissions of CO and VOCs, which are predominantly generated by gasoline-powered on-road vehicles.

5.2.1.1 Scenario comparisons

Results from MOBILE runs are compared within each modelling year to observe the impact of biodiesel blends on identical vehicle fleets.

Criteria air contaminants: PM, NO_X, SO_X, VOCs, CO, and NH_3

The HDDV category is generally the most important contributor to **PM** exhaust emissions (see Table 5-3), for both PM_{10} and $PM_{2.5}$. However, in 2020 light-duty gasoline vehicles are projected to dominate PM emissions, as reflected by the considerable decrease of the HDDV contribution to on-road emissions (e.g., 21% in 2020 vs. 57% in 2006 with the use of B0). The use of B20 in 2006 is estimated to reduce PM emissions by 13% or 877 tonnes, while 2020 projections show a reduction in PM emissions of 9% or 77 tonnes with B20. These changes have a minimal impact on the contribution of HDDVs to the overall on-road emissions, as reflected by a shift from 57% to 54% and 21% to 19%, respectively, in 2006 and 2020.

The impact of biodiesel on HDDV emissions is expected to decrease in 2020 compared to 2006 as newer vehicles are introduced into the vehicle fleet. For example, TPM/PM_{10} HDDV emission reductions for B5 and B20 vary from 3% and 13%, respectively, in 2006 to 2% and 9%, respectively, in 2020 (see Appendix V, Table V-1). $PM_{2.5}$ emissions are expected to vary in a similar manner to TPM or PM_{10} emissions from 2006 to 2020 with the use of biodiesel.

The HDDV category is also generally the most important contributor to on-road **NO_X** emissions. Contrary to most pollutants, NO_X emissions are expected to increase with increasing biodiesel content for all scenario years (see Appendix V, Table V-2). For HDDVs, emission increases for 2006 and 2020, respectively, are estimated to be 0.9% and 0.7% for B5 and 3.6% and 2.8% for B20.

As **CO** emissions are predominantly generated by light-duty gasoline vehicles and trucks, which account for ~95% of CO emissions, the HDDV emission reductions from the use of biodiesel blends in 2006 and 2020 translate into a decrease of less than 1% for all vehicles (see Appendix V, Table V-2).

The MOBILE output files include a variety of compounds or emission categories related to **VOC**s, notably VOC_{total}, $VOC_{exhaust}$, $VOC_{evaporative}$, and THC_{total}. Light-duty gasoline trucks and vehicles are responsible for most VOC emissions, including through evaporative losses. Because evaporative emissions for diesel and biodiesel fuels are considered negligible, the evaporative categories (e.g., $VOC_{evaporative}$ and $THC_{evaporative}$) are not considered in this analysis. For HDDVs, the relative impact of biodiesel fuel use on emissions of VOC_{total}, $VOC_{exhaust}$, and individual

VOC compounds (e.g., acetaldehyde, formaldehyde, and acrolein) compared to the B0 scenario is identical (see Appendix V, Table V-3):

- a 4.4% decrease in 2006 to a 0.8% decrease in 2020 for B5; and
- a 17.8% decrease in 2006 to a 3.5% decrease in 2020 for B20.

MOBILE6.2C does not predict any impact from the use of biodiesel fuel on **SO_2** and **NH_3** emissions.

Non-criteria contaminants

Based on the MOBILE results, gasoline-powered light-duty vehicles and trucks are responsible for most of the **aromatic VOC** emissions, including around 98% of benzene emissions and more than 99% of toluene emissions. Thus, the impact of any change in HDDV aromatic emissions from the use of biodiesel fuel is expected to have a limited impact on emissions from all vehicles (less than 1%).

MOBILE can be used to generate emission factors for **polycyclic aromatic hydrocarbons** (PAHs). The relative impact (in percent change) of biodiesel use on all mobile source emissions is similar for all PAH species in MOBILE. The impact of biodiesel use on PAH emissions is limited even for high blend scenarios. In 2006, aside from B[a]A that is reduced by 4% and B[a]P by 2% for the B20 scenario, other PAH species are limited to reductions between 0.1 and 1.3%. The impact of B20 on PAH emissions from all mobile sources projected to 2020 is limited to a reduction of less than 1% (see Appendix V, Table V-4).

Inter-year MOBILE results are compared to determine the impact of new technologies and regulatory requirements on the different biodiesel blends. The greatest impacts of newer vehicles and engines are observed under the B0 scenarios. Changes in emissions due solely to vehicle technology improvements between modelling years are considerable (Appendix V, Table V-6). Since 1998, the general approach to setting on-road vehicle emissions standards in Canada has been to harmonize them as much as possible with US EPA federal standards (detailed in the US Code of Federal Regulations). For HDDVs, recent regulations brought in more stringent standards for smog forming emissions between 2004 and 2010. These regulations result in a reduction of NO_X and PM emissions from new vehicles by 95% and 90% respectively, relative to previous requirements.[54] Compared to 2006 values, the 2020 HDDV emissions due to vehicle fleet changes under the B0 scenario are expected to change by:

- 87% for TPM, PM_{10}, and $PM_{2.5}$;
- 80% for NO_X;
- 78% for CO;
- 36% for VOCs and THCs; and

[54] http://www.ec.gc.ca/air/default.asp?lang=En&n=733706F8-1 (accessed December 29, 2011)

- 80% for PAHs.

Because of the important relative contribution of HDDVs to total mobile-source emissions, these inter-year impacts translate into considerable reductions in NO_X and PM emissions from all on-road mobile sources.

The inter-year impacts are reduced slightly for most pollutants with increasing biodiesel blends, as the impact of biodiesel on emissions mitigates a fraction of the technology-driven changes. Relative emission reductions in 2020 compared to 2006 are generally 1-2% less for biodiesel blends compared to B0 scenarios (data not shown). It does not indicate that biodiesel use lowers the efficiency of new technological improvements; rather it indicates that the potential emission benefits from the use of biodiesel are projected to be smaller with the introduction of new vehicles in the on-road mobile fleet. Regarding NO_X, the increase in emissions from biodiesel use is mitigated by the major technology-driven emission decreases.

5.2.2 *Effect of biodiesel on* provincial/territorial emission results

MOBILE emission results are analyzed on a provincial/territorial basis to assess regional variations due to factors such as heavy-duty diesel fleet turn-over, fuel characteristics, and meteorological considerations. Provincial model outputs are compared for the 2006 and 2020 modelling years.

For the 2006 scenarios, the impact of biodiesel use is estimated to have a similar impact on HDDV emissions in all provinces and estimates are generally within a few decimal points of comparable national estimates (see Appendix V, Table V-5 and Table V-6). However, inter-provincial differences are apparent in 2020, especially for Ontario. These differences in emission changes from the use of biodiesel between regions are due in part to fleet profiles and turn-over rates.[55] A more modern fleet and a more rapid turn-over will have the effect of minimizing the impact of biodiesel blends on emissions. Table 5-4 presents a summary of estimated HDDVs older than 2007 for the 2020 scenario.

These distributions show that some regions are impacted more by the introduction of biodiesel in the diesel fuel pool. Based on the expected fleet profile in 2020, the impact of biodiesel in all provinces is expected to be lowest in Ontario and greatest in Prince Edward Island.

[55] Differences in fuel characteristics (aromatics, olefins, sulphur levels), meteorological input data, and the presence of an I/M program will also influence regional emission profiles.

Table 5-4 Model-year 2007 and older on-road HDDV population by region for 2020, as assumed in MOBILE

Region	AB	BC	MB	NB	NL	NS	NT
Total HDDVs older than 2007 MY	63,315	40,270	19,337	9,319	4,350	7,125	599
Percentage of HDDV fleet older than 2007 MY (%)	30	23	38	29	40	31	44
Region	**NU**	**ON**	**PE**	**QC**	**SK**	**YT**	
Total HDDVs older than 2007 MY	85	36,014	2,590	86,629	32,620	555	
Percentage of HDDV fleet older than 2007 MY (%)	43	13	58	35	42	40	
MY: model-year; AB: Alberta; BC: British Columbia; MB: Manitoba; NB: New Brunswick; NL: Newfoundland and Labrador; NS: Nova Scotia; NT: Northwest Territories; NU: Nunavut; ON: Ontario; PE: Prince Edward Island; QC: Québec; SK: Saskatchewan; YT: Yukon Territories							

Currently, provincial requirements for the use of biodiesel exist in three provinces: 2% in Manitoba (since 2009) and Alberta (since 2010), and up to 5% in British Columbia (3% in 2010, 4% in 2011, and 5% in 2012). These are not considered in the current analysis. The impact of this modelling decision is assessed by estimating the relative weight of individual provinces compared to the national mobile inventory. Compared to the 2010 B0 scenario and considering all vehicle categories, decreases in PM, VOC_{total}, and CO, and increases in NO_X emissions from the implementation of provincial biodiesel requirements are projected to be less than 1%. The values of these changes are expected to decrease in 2020 as newer vehicles are introduced. Further, these minimal impacts will be diluted further when considered in the emissions inventory from all sectors, where transportation emissions from heavy-duty diesel vehicles contribute only in a very limited fashion. These results suggest that ignoring the provincial requirements for the purposes of the reference case (i.e., B0) does not have a considerable impact on MOBILE source emissions for all of Canada.

5.3 Conclusion

The MOBILE 6.2C model is capable of estimating vehicle emission factors for a wide range of pollutants, including criteria air contaminants and air toxics. It is the most relevant system for modelling Canadian on-road mobile source emissions (28 vehicle classes from passenger cars to heavy-duty trucks), relying on extensive US and Canadian vehicle testing data, and Canadian specific codes and inputs. It can be used to generate emissions per year for designated regions by combining pollutant-specific emission factors and vehicle kilometres traveled data.

Biodiesel use scenarios ranging from B0 to B20 in 2006, 2010, 2015, and 2020 were selected for the modelling runs. All modelled scenarios consider a national-wide use of biodiesel and are applicable year-round. Although existing and planned provincial biodiesel requirements are not

considered in the MOBILE simulations, a sensitivity analysis demonstrated they are not expected to have a significant impact on projections for the different scenarios analyzed in this assessment (see Section 5.2.2).

The MOBILE6.2C modelling quantified the changes in emissions due to biodiesel use: most HDDV exhaust emissions are expected to be reduced, with the exception of NO_X emissions, which increased slightly. The magnitude of change in HDDV exhaust emissions generally vary linearly with biodiesel content, such that B20 has roughly four times the impact of B5. For example, PM_{10} HDDV emission reductions for B5 and B20 are 3.2% and 12.6%, respectively, in 2006.

When all mobile source emissions are considered, the relative impact of biodiesel use differs significantly between chemical species. For example, pollutants predominantly originating from diesel vehicles, such as NO_X and PM, are more affected by biodiesel use than gasoline-related pollutants such as hydrocarbons and CO.

Biodiesel is projected to have less impact on exhaust emissions in 2020 due to the turn-over of the Canadian vehicle fleet, with 2010 and beyond vehicles having to meet more stringent exhaust emission standards. Baseline on-road vehicle emissions in 2020 are projected to be considerably less compared to 2006 levels due to changes in emission control technologies. This was also observed by Sauthoff et al. (2010) who assessed biodiesel use scenarios in freight trucks in the Upper Midwestern US with the *Greenhouse Gases, Regulated Emissions, and Energy Use in Transportation* (GREET) model. The modelling results led them to conclude that with the use of fuel blends containing less than 20% biodiesel, the impact on NO_X and PM emissions were outweighed by important reductions in emission rates resulting from improvements in exhaust emission control devices, vehicle efficiency, and fuel formulations.

On-road mobile source emissions generated from the MOBILE6.2C are used to populate the emissions inventory used in the AURAMS atmospheric quality model, as described in Chapter 6.

5.3.1 Limitations

All modelling activities include limitations such as modelling assumptions, inclusion/exclusion of key variables and environmental factors, and sufficient and available observational data to determine emission factors. Limitations inherent to the MOBILE6.2C model are not discussed (e.g., VOC speciation, air toxic ratios).

An important limitation in this analysis is the lack of biodiesel test data for the most recent model-year heavy-duty vehicles (i.e., 2010 model-year or later vehicles) and for vehicle classes other than HDDVs. Hence, the impacts of biodiesel fuel are limited to the impact of biodiesel fuels in HDDVs and only for HDDVs produced before 2010.

The limitation of assuming no biodiesel effect on LDDV and LDDT emissions is expected to be minor as they account for only 2% of the entire Canadian on-road vehicle fleet, 3% of the total VKT, and 6% of the diesel fuel used in 2006 (Table 5-1). Although they represent a minor portion of the diesel fleet, it would be of value to include light-duty diesel fleet effects in future scenario modelling.

Profile development for the heavy-duty diesel fleet introduces some uncertainty into this analysis. Complete access to provincial/territorial vehicle data would allow for greater confidence in the on-road vehicle fleet profile through a more transparent and robust means of dividing vehicle information into MOBILE classes. In addition, a specific set of mileage accumulation rates for each province, territory, or region would be valuable.

The previous limitations underline issues regarding the representativeness of the biodiesel emission database. Yanowitz and McCormick (2009) have noted that the biodiesel emissions database is not highly representative of the fleet currently in-use. Testing on additional engines is required to achieve a better representation. While acknowledging the analysis by Yanowitz and McCormick (2009), it is assumed for this assessment that the current biodiesel emissions database is the most relevant database available to conduct modelling of biodiesel use in Canada with MOBILE6.2C.

5.4 References

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Chapter 6. Impacts of biodiesel use on ambient air pollutant concentrations

To understand the human health implications of the emission changes associated with biodiesel use noted in Chapter 5, it is necessary to estimate the relative impact of those changes on Canadian air quality. This is a complex task that requires photochemical modelling because emissions are transported from the emission sources and chemically transformed in the atmosphere. The latter integrates the spatial and temporal distribution of air pollutant emissions over Canada with chemical reaction processes and meteorological data. This chapter presents the incremental changes in air pollutant concentrations for Canada under scenarios of national use of B5 and B20, in 2006 and 2020.

Photochemical modelling for the current project is conducted with *A Unified Regional Air quality Modelling System* (AURAMS) in collaboration with the Air Quality Modelling Application Section (AQMAS) of Environment Canada to investigate the impact of biodiesel blends on air pollution in Canada. The results of these simulations are used by the Air Quality Benefit Analysis Tool (AQBAT) to estimate the human health impacts of change in air quality resulting from biodiesel use (see Chapter 8).

6.1 AURAMS

AURAMS is a chemical dispersion model used by Environment Canada to study the formation of tropospheric ozone, particulate matter, and acid deposition in North America, in support of air quality policy and management decisions for Canada. Overall, the model resolves 157 tracers: 49 gaseous species and 9 particulate chemical components (sulphate, nitrate, ammonium, elemental carbon, primary organic matter, secondary organic matter, crustal material, sea salt, and particle-bound water) each divided into 12 size bins (logarithmic spacing between 0.01 and 41 μm). Details regarding the mechanisms behind AURAMS can be found in Samaali et al. (2009), Smyth et al. (2009), and AQMAS (2010). The AURAMS model has been validated and/or compared with observations for the following pollutants: O_3, $PM_{2.5}$, PM_{10}, NO_2, SO_2, CO, HNO_3, $PM_{2.5}$ components (e.g., sulphates, nitrates, ammonium, OC, EC, sea salt, crustal material), total VOCs, some individual VOCs, and secondary organic aerosols.[56]

AURAMS uses the meteorological fields from the Canadian Global Environmental Multiscale (GEM) model and atmospheric emissions from the Sparse Matrix Operator Kernel Emissions (SMOKE) processing system as inputs. GEM is an integrated forecasting and data assimilation system developed by Environment Canada. SMOKE[57] is an emissions processing system designed to create gridded, speciated, hourly emissions for input into a variety of air quality

[56] AQMAS, December 28, 2011. Personal communications.
[57] http://www.smoke-model.org/index.cfm

models. It supports area, biogenic, mobile, and point source emissions processing for criteria pollutants, toxics, and particulates.

Air quality modelling depends on a variety of inputs, including emissions inventories for North America and spatial allocations based on the 2006 Census of Population conducted by Statistics Canada.

6.1.1 Modelling domains

Modelling is based on a continental grid domain at 45-km resolution (143×107 grid points) and two regional grid domains at 22.5-km resolution, Eastern and Western, covering the ten Canadian provinces. These domains are represented in Figure VI-1 in Appendix VI. The Eastern domain (145×123 grid points) covers all of eastern Canada and the northeastern United States (US), and the Western domain (124×93 grid points) covers all of western Canada. Results from the two regional domains are combined to generate national annual data on a 22.5-km grid.

In addition to the continental and regional runs, a finer 3-km resolution grid domain is defined for the Montréal area (130×120 grid points) to focus on urban air pollution (see Figure VI-2 in Appendix VI). This high-resolution modelling is conducted using refined transportation emission estimates generated by the GRILLE model (see Section 6.3). High-resolution air quality modelling results are limited to a two-week episode in June.

6.1.2 Scenarios

Three basic annual scenarios are selected for modelling: a diesel fuel reference case (B0), a 5% biodiesel content scenario (B5), and a 20% biodiesel content scenario (B20), for the years 2006 and 2020. All scenarios investigated for the current assessment, described in Table 6-1, are applicable for Canada (i.e., no similar fuel substitution is assumed for the US or Mexico). Upstream emissions associated with biodiesel fuel (i.e., fuel production, transportation, and distribution) are not considered in the modelling due to a lack of data.

Table 6-1 Canadian biodiesel use scenarios for AURAMS modelling for all domains

Years	Fuel	Continental	National		Montréal
			Eastern	Western	
2006 and 2020	B0	Annual	Annual	Annual	June 12 to 23
	B5	Annual	Annual	Annual	June 12 to 23
	B20	May 1 – Sept. 30*	May 1 – Sept. 30*	May 1 – Sept. 30*	June 12 to 23
*For the B20 scenarios, B0 is used during winter months (i.e., October 1st to April 30th)					

6.1.3 Emissions inventory

The emissions inventories for the 2006 reference case and the 2020 projections are compiled by Environment Canada's Pollution Data Division (PDD). They combine emissions data for all sectors of the economy and include industrial, non-industrial, and mobile sources. Tables VI-1 to

VI-3 in Appendix VI present a list of all sectors and source categories featured in the emissions inventory. Under the different biodiesel use scenarios, only emissions from the on-road HDDV portion of the transportation sector are assumed to be impacted (see Section 5.1.3).

The US data originates from the US EPA 2005v4 inventory.[58] For Mexico, 1999 data are available (Eastern Research Group Inc. 2006).[59]

The emissions inventory for on-road mobile sources under ultra low sulphur diesel (ULSD) and biodiesel scenarios is prepared using MOBILE6.2C, and is detailed in Chapter 5. The Canadian on-road emissions are distributed across the road network as 30% highway and 70% non-highway with little variation across vehicle types, similar to the US approximations.

Some sources are not fully represented in the inventory (e.g., lightning emissions, wildfire emissions, wind blown dust) for various reasons (e.g., unpredictable nature of the events). Although some of these natural emissions may contribute significantly to pollutant levels, such as ozone, it was considered preferable to omit them from the inventory. In addition, for consistency between inventories, when corrections are required to the 2006 inventory, proportional correction factors are also applied to the 2020 projections.

Table VI-4 (Appendix VI) presents 2006 total CAC emissions (all sectors including off-road transportation) and details on-road emissions by vehicle type. Within on-road transportation emissions, the HDDV class dominates NO_X and PM emissions, while gasoline vehicles account for most CO and VOC emissions. Based on total emissions, the transportation sector is only a key driver for CO and NO_X emissions. However, this does not take into account the geographic distribution of emissions and proximity to human populations.

As biodiesel concentration increases in fuel, emissions of CO, PM_{10}, $PM_{2.5}$, and VOCs from the on-road transportation sector are expected to decrease, while NO_X emissions are expected to increase. It is assumed that there is no impact on NH_3 and SO_2 emissions from the use of biodiesel.[60]

Table VI-5 (Appendix VI) presents the 2020 inventory data for the on-road transportation sector by vehicle class and pollutant under the different modelling scenarios.[61] As for the 2006 inventory, HDDVs are the main driver of NO_X on-road transportation emissions and are also responsible for a large share of PM_{10} and $PM_{2.5}$ on-road emissions. Otherwise, the contribution

[58] See US EPA website for more information : www.epa.gov/ttn/chief/emch/index.html#2005
[59] Although the Mexican data appears somewhat outdated, their impact on air quality in Canada is very limited (AQMAS 2010).
[60] See Chapters 4 and 5 for details regarding the impact of biodiesel use on vehicle emissions,
[61] 2020 emission projections are estimated by PDD using the *Energy, Environment, Economy Model for Canada* (E3MC) modelling framework (http://www.ec.gc.ca/doc/virage-corner/2008-03/571/Annex1_eng.htm).

of HDDV emissions to the transportation sector is eclipsed by light-duty gasoline vehicles and trucks, notably for CO and VOCs.

In general, on-road transportation emissions are expected to decrease from 2006 to 2020 due to improvements in engine and vehicle technologies and the adoption of more stringent emission standards.

6.1.4 Meteorology

Meteorological fields used by the atmospheric air quality model are generated by the GEM meteorological model for the 2006 reference year. The same meteorological fields are used for 2020. The meteorological model is integrated for the full 2006 year on a global grid of variable spatial resolution with a resolution of 15 km over North America. The meteorological variables are then interpolated onto the air quality modelling grids (22.5-km and 45-km). For the high-resolution modelling around Montréal, a separate set of meteorological files is generated using a limited area version of the operational GEM on a 2.5-km grid. This dataset is then interpolated on the AURAMS 3-km grid.

6.1.5 Model output and pollutant metrics

AURAMS generates emission rates and ambient concentrations of air pollutants. Gridded metrics are then projected onto 2006 population census divisions for the estimation of health impacts (see Chapter 8). Ambient concentrations of gaseous species (O_3, NO, NO_2, NO_X, CO, VOC, formaldehyde) are reported as ppbv, while concentrations of aerosols ($PM_{2.5}$ and PM_{10} species) and PM precursors (SO_2, NH_3) are reported in $\mu g/m^3$. Changes in concentrations under the different biodiesel scenarios are expressed in comparison to the petroleum ULSD (or base case) scenarios.

The analysis focused primarily on the following metrics:

- 1-hour daily maximum for CO, NO_2, PM_{10} and $PM_{2.5}$ (annual);
- 24-hour daily average for CO, NO_2, $PM_{10,}$ and $PM_{2.5}$ (annual);
- 8-hour daily maximum (running average) and 1-hour daily maximum for O_3 (summer only).

AURAMS is not capable or considered suitable for estimating air concentrations of most mobile source air toxics (e.g., VOCs). VOCs considered in AURAMS are expressed as thirteen *mechanism species* (MS), which take into account chemical functional groups and their atmospheric reactivity with respect to the hydroxyl radical. An exception to this is formaldehyde, which is treated explicitly in AURAMS. MS are developed in relation to their ozone forming potential and are not based on toxicological relevance in humans. While the confidence level in AURAMS total VOC output is good (based on comparison with measured ozone levels), there has been no systematic validation of individual MS. Individual MS concentrations are therefore generally not used to estimate health effects. Nonetheless, a qualitative discussion of general

trends regarding formaldehyde and MS contained in mobile source air toxics is included in Chapter 8.

6.2 National Modelling

Modelling results presented in this section for the 2006 and 2020 scenarios are from the merged East and West domains, providing a national coverage with a 22.5-km horizontal resolution.

The base case scenarios are initially compared to assess the projected changes in air quality between 2006 and 2020 when biodiesel is not used. Changes in emissions are expected to result from various regulations and economic factors, such as the implementation of more stringent emission standards in vehicles, and increasing populations, vehicle traffic, and industrial activity. In general, it is observed that 8-hr average ozone concentrations are expected to increase (by less than 10 ppbv) in large urban centres and the Alberta oil sands area, while most of Canada is expected to see a decrease. Increases and decreases in $PM_{2.5}$ concentrations are also observed across Canada, but these changes are expected to be minimal (less than 3 $\mu g/m^3$).

6.2.1 Biodiesel use in 2006

Results for the B5 and B20 scenarios in 2006 are presented for O_3, $PM_{2.5}$, NO_2, CO, and VOCs.

6.2.1.1 Ozone

For the base case in 2006, modelled summer daily ozone 8-hr running average[62] maxima are above 50 ppbv in Vancouver, central Alberta, Regina, southern Ontario and Montréal. Otherwise, the annual hourly ozone average across Canada remains below 30 ppbv (data not shown; AQMAS 2010).

The use of 20% biodiesel in 2006 results in a negligible impact on ozone 8-hr daily maxima summer values (from -0.40 ppbv to +0.11 ppbv, or -1% to +0.2%) with a decrease in large urban centres, but an increase outside of these centres (see Figure VI-3 in Appendix VI)**Error! Reference source not found.**. Most of the changes in the east are located along the St. Lawrence River corridor, while in the west the southern part of the Prairies are impacted. These changes are in agreement with the variations in emissions. However, horizontal transport, which tends to disperse ozone outside of the areas where the changes in emissions occur, is noticeable, notably in the Atlantic region. The decrease in ozone concentrations over urban centres could be explained by both an increase in HDDV NO_X emissions (+3%) and a decrease in HDDV VOC emissions (-17%) due to the use of B20 (see Chapter 5). However, it is hard at this level of analysis to identify whether NO_X or VOCs contribute more or less to the reduction in ozone concentrations.

[62] The 8-hr running average maximum metric is analogous to the Canada-Wide Standard for O_3 set at 65 ppbv.

The impact of B5 is similar to B20, but proportionately scaled down. Concentration differences outside urban areas are close to model detection limits. Overall, based on the modelling results, the change observed in the number of episodes exceeding the O_3 Canada Wide Standard (CWS) of 65 ppbv is negligible (AQMAS 2010).

6.2.1.2 $PM_{2.5}$

Annual average $PM_{2.5}$ concentrations based on hourly output are examined. Typical average concentrations are 20 µg/m³ inside large cities and 5-10 µg/m³ for surrounding urban areas (AQMAS 2010).

Small changes in $PM_{2.5}$ mobile emissions from the use of B20 in 2006 are predicted across Canada. The emission reductions are greater in urban areas due to more intense transportation activity. The spatial pattern of changes in the $PM_{2.5}$ concentrations (Figure VI-4 in Appendix VI) due to biodiesel use is generally in agreement with the spatial changes in $PM_{2.5}$ emissions. Modelling results show that over the entire domain, $PM_{2.5}$ ambient levels vary only minimally, between -0.03 ug/m^3 and +0.04 ug/m^3 (or -0.3% and 0.1%, respectively) under the B20 scenario.

Examination of speciated $PM_{2.5}$ components (p-NO_3, p-SO_4, EC) indicates that while p-NO_3 and p-SO_4 may be responsible for some of the increases along the Québec-Windsor corridor, the decreases in EC levels provide a good explanation for the decrease in $PM_{2.5}$ levels in urban areas.

$PM_{2.5}$ concentrations for the B5 scenario (not shown), compared to the B20 results, do not follow a linear trend, possibly due to different particulate formation processes occurring at the same time. It is observed that the transition zone between the decreases in urban centres and the increases in the peripheral regions moves further away from urban areas under the B5 scenario. While the decrease in $PM_{2.5}$ concentrations in urban centres under the B5 scenario is approximately half the B20 value, it affects a larger area. It appears that increasing the biodiesel content from B5 to B20 has a greater impact on processes that generate secondary $PM_{2.5}$ compared to processes that decrease primary $PM_{2.5}$ emissions in urban centres.[63]

The change in the number of episodes exceeding the 30 µg/m³ CWS for fine particulate matter is negligible (AQMAS 2010).

6.2.1.3 NO_2

Average NO_2 concentrations of 20-30 ppbv and 2-10 ppbv are predicted in large cities and surrounding areas, respectively, for the base case scenario in 2006. Under the B20 scenario in 2006, NO_2 concentration changes are small, but non-negligible (AQMAS 2010). Increases of approximately 0.005 ppbv are expected on the periphery of large cities from the use of biodiesel, while increases above 0.010 ppbv are estimated in city centres. A maximum change of 0.08 ppbv is modelled in Toronto.

[63] AQMAS, June 10, 2011. Personal communications.

6.2.1.4 CO

The average concentrations of CO for the base case scenario are estimated at 100-200 ppbv, reaching a high between 500 and 1000 ppbv in large cities. The baseline spatial distribution of concentrations is in agreement with spatial allocation of emissions from the main CO sources, such as industry and transportation. Changes under the B20 scenario in 2006 are minor and limited to a decrease of about 0.3 ppbv. Similar changes are observed for the B5 scenario.

6.2.1.5 VOC

VOC concentrations under a B20 scenario in 2006 are predicted to decrease slightly in large cities (-0.030 to -0.050 ppbv) and their surroundings (-0.001 to -0.010 ppbv). This is somewhat expected as the impact of biodiesel use on VOC emissions from all sources is minimal. Reductions of approximately 0.001 to 0.005 ppbv are predicted for the whole St. Lawrence River corridor and the Atlantic region.

Total VOC and MS results are available for Montréal and Toronto. These two urban areas drive most of the VOC emission changes from the use of biodiesel in the Eastern domain. For Montréal, concentrations of all VOC mechanism species are expected to decrease, albeit by less than 1% with the exception of the higher aldehydes MS (which include acetaldehyde, propionaldehyde and benzaldehyde) at around 2%. VOC ambient concentrations in Toronto generally decrease under a B20 scenario during summer 2006, with total VOC reduction expected to reach 0.153%. Of note, alkenes, cresol, and mono-substituted aromatics MS are expected to increase slightly in Toronto.

6.2.1.6 Concentration changes by census division

Further analyses examine CO, PM_{10}, $PM_{2.5}$, O_3 and NO_2 concentration changes at the census division (CD) level. The concentration in a CD is determined by summing the product of a grid cell concentration and the area of that grid cell occupied by the CD, for all grid cells intersecting with the CD.

As a general observation, changes in air pollutant concentrations are minor under the B5 scenario, even for CDs registering the largest variations. No CD is expected to see positive or negative changes above 0.5% for any of the modelled pollutants. Results from the B20 2006 scenario are fairly similar to the B5 scenario for all pollutants (i.e., the same CDs show the most or least change), even if the direction and the amplitude of change can vary between scenarios and pollutants for individual CDs.

The number of CDs in which concentrations are expected to increase and decrease under the B5 and B20 scenarios are examined to gauge the fraction of the Canadian population affected. Generally, decreases in pollutant concentrations were predicted in fewer CDs with the use of B20 compared to B5. Decreases in $PM_{2.5}$ concentration from the use of B5 are modelled in 104 CDs, representing approximately 68% of the Canadian population. Under the B20 scenario,

decreases in $PM_{2.5}$ concentrations are expected in fewer CDs (56), representing 54% of the population. Only 19 CDs show decreases in O_3 concentrations with B5, which account for nearly 45% of the Canadian population. While O_3 reductions in large urban centres like Toronto and Montréal are beneficial (due to high population density), a larger fraction of the national population is expected to experience an increase in O_3 concentration. Results for B20 are similar. For CO and NO_2, decreases and increases in concentrations, respectively, were anticipated in most CDs (representing more than 99% of the Canadian population).

6.2.2 Biodiesel use in 2020

B5 and B20 scenarios are also available for the 2020 projected inventories. Although these projections include more inherent uncertainty than the 2006 scenarios, they incorporate the impacts of fleet turn-over and the implementation of stricter emission standards for newer vehicles. Generally, the impacts of biodiesel use are smaller in 2020 compared to the 2006 results due to these factors. In addition, B5 results are near the detection limits for most pollutants. Only the B20 scenario results are presented for the 2020 model year.

6.2.2.1 Ozone

Ozone concentration reductions are noted in most urban centres, although these reductions are very small. For example, the 8-hr average daily maximum is reduced by at most 0.1 ppbv in Calgary, no reduction is projected for Toronto, and a decrease of 0.04 ppbv is expected in Montréal. Some increases are observed in southern Quebec and Manitoba (0.02-0.05 ppbv).

6.2.2.2 $PM_{2.5}$

The B20 scenario results in minimal changes in $PM_{2.5}$ concentrations at the national scale. Only the Athabasca Oil Sand sector (northern Alberta) shows a clear increasing trend (around 0.005 $\mu g/m^3$). Slight increases are also expected around the Lower Fraser Valley, Toronto, and an area southeast of Montréal, while a small decrease is perceptible around the Calgary, Edmonton, and Montréal regions. These results are near the model detection limit.

6.2.2.3 NO_2

NO_2 is only minimally impacted by the use of biodiesel fuel in 2020. Small increases in concentration are noted in urban areas (approximately 0.01-0.03 ppbv), but changes remain close to the model detection limit (e.g., 0.006 ppbv in Toronto).

6.2.2.4 CO

Changes in CO concentrations are minor. For example, maximum decreases vary from 0.010 to 0.060 ppbv in large urban centres, where approximate average CO concentrations are expected to range between 200 and 500 ppbv.

6.2.2.5 VOC

The impact of the B20 scenario on VOC annual average concentrations is also very small in 2020. Decreases are limited to a few grid points corresponding to urban areas. Reductions observed around cities are more localized in the Western domain than in the Eastern domain.

Plumes extending from Toronto and Montréal might be due to the Québec-Windsor corridor atmospheric circulation where pollutants can be transported over long distances from their point of origin.

6.2.2.6 Concentration changes by census division

Although the most impacted CDs differ between the B5 and B20 scenarios, results are within the same order of magnitude. As was observed for 2006, O_3 concentrations are expected to decrease in a few populated CDs while increases are expected in CDs beyond urban areas. CO concentrations are reduced in most CDs. As for PM_{10} and $PM_{2.5}$, the largest decreases are estimated to occur around highly populated areas, such as Winnipeg and Edmonton. Maximum increases in PM_{10} and $PM_{2.5}$ concentration are expected in suburban and rural regions of Québec, mostly downwind of Montréal along the St. Lawrence River corridor. NO_2 concentrations are projected to increase in all CDs and the extent of the relative change does not appear to correlate with CD population.

Regarding the number of CDs where increases and decreases in air pollutant concentrations occur under the B5 and B20 scenarios in 2020, the results generally predict decreases in pollutant concentrations in fewer CDs with the use of B20 compared to B5. For both $PM_{2.5}$ and PM_{10}, decreases in concentrations are expected in approximately 50 CDs (25% of the population) under the B5 scenario, but are limited to 14 CDs (14% of the population) with B20.

Ozone results are similar for B5 and B20, with increases in concentrations expected in most CDs, affecting nearly 90% of the population.

6.2.3 Discussion of national modelling results

Important reductions in exhaust emissions are noted for the baseline ULSD scenarios in 2020 compared to 2006, due in part to the development and use of efficient emission control technologies and the implementation of new emission standards for the model year 2010 and later vehicles (see Tables VI-4 and VI-5 in Appendix VI).

In general, the proposed B5 and B20 biodiesel scenarios are associated with very minimal changes in emissions and ambient air concentrations of air pollutants, and B5 has less impact than B20. The B5 and B20 scenarios under 2006 conditions predict small, but non-negligible, changes in ambient concentrations, mostly in urban centres and surrounding areas (southern Ontario, southern Québec, central Alberta and the Lower Fraser Valley). Aside from a few CDs, ozone and $PM_{2.5}$ concentrations are reduced in urban centres, but increase slightly in surrounding areas.

Ozone concentrations are observed to be highly dependant on NO_X and VOC levels. Increases in NO_X emissions in urban centres can reduce ozone concentrations via ozone scavenging, especially if VOC levels decrease simultaneously. Regarding $PM_{2.5}$ concentrations, the

difference between primary $PM_{2.5}$ reductions and secondary $PM_{2.5}$ formation will determine the direction of change in ambient levels. Hence, biodiesel does not necessarily have a beneficial impact on $PM_{2.5}$ concentrations even if primary $PM_{2.5}$ emissions from HDDVs are expected to decrease. For example, the concentrations of $PM_{2.5}$ and PM_{10} are projected to increase in the highly populated Greater Vancouver (CD5915) and Fraser Valley (CD5909) regions under the B20 scenario in 2020, which contrasts with the B5 results. The increase in $PM_{2.5}$ is linked to the increase in NO_X emissions, which are converted to nitrate. Essentially in these two CDs, the decrease in primary $PM_{2.5}$ emissions in 2020 does not compensate for the increase in secondary nitrate production from NO_X emissions.[64]

For the 2020 projections, changes in predicted air quality are very small and often close to model detection limits. Decreases in concentrations of O_3 and PM are observed in fewer CDs in 2020 compared to 2006, under both the B5 and B20 scenarios.

Regarding model performance, summer ozone estimates are considered very good, while $PM_{2.5}$ predictions are acceptable. Better quantification of $PM_{2.5}$ is necessary to improve model performance.

6.3 High-resolution Urban Modelling

High-resolution modelling of the Montréal region is conducted using AURAMS with a 3-km horizontal resolution grid and specific input from the GRILLE traffic-demand model. GRILLE is a high-resolution emissions model combining emission rates from MOBILE6.2C with network flow data for the Greater Montréal area, based on Transport Québec's *Modèle de Transport de la Région de Montréal* (MOTREM). GRILLE provides on-road mobile emissions and displays them on a 1-km grid.

Data from GRILLE at 1-km and meteorological data generated by GEM at 2.5-km resolution are interpolated to coincide with the AURAMS 3-km grid cells. Simulations are for a two-week high pollution episode extending from June 12 to 23, 2006 and 2020. High-resolution outputs for O_3, $PM_{2.5}$, and NO_2 are discussed in this section, while other model outputs generated (e.g., CO, SO_2) are considered in Chapter 8.

The 2006 and 2020 base case scenario concentrations are compared to assess the impact of fleet composition and other projected changes on vehicle emissions. Ozone concentrations in 2020 are usually 5-10 ppbv less than 2006 values in regions surrounding the island of Montréal, while concentration levels within the island boundary in 2006 and 2020 are similar (within 5 ppbv). Notably, areas of peak ozone concentration differ because of significant reductions in on-road

[64] The results for $PM_{2.5}$ and PM_{10} for the Greater Vancouver and Fraser Valley should be interpreted with caution as the AURAMS model lacks the chemistry reactions with sea salt, which can mitigate the increase in PM concentrations.

NO_X emissions predicted for 2020. Consequently in 2020, more of the domain is NO_X-limited rather than VOC-limited with regards to ozone formation. As a result, high-traffic sectors identified in the 2006 baseline scenario (e.g., highways, bridges), which contributed to ozone destruction by increasing NO_X emissions (via the ozone scavenging effect), do not provide the same signal in 2020.

The spatial distribution and scale of $PM_{2.5}$ average concentrations in the 2006 and 2020 reference cases are similar. The greater part of the domain outside the island is defined by concentration levels between 5.0 and 10.0 µg/m^3. Maximum concentrations are modelled in the south-eastern part of the island and above the St. Lawrence River near the high-traffic bridges crossing the river (20.0-30.0 µg/m^3).

The spatial distributions of NO_2 annual average concentrations in 2006 and 2020 are similar, but lower concentration levels are projected for 2020. Maximum concentrations are attained south of the island of Montréal, mostly above water bodies in the vicinity of the bridges since traffic is a major contributing source of NO_2. Also, greater NO_2 concentration levels are distinguishable along the major roads and highways spanning from the island of Montréal.

6.3.1 Biodiesel use in 2006

Of note, projected variations in emission rates by the GRILLE model are generally small between the B20 and B0 scenarios in 2006, while they are practically imperceptible in 2020.

6.3.1.1 Ozone

The spatial distribution of ozone concentration changes under the B5 and B20 scenarios is similar, but the amplitude of change is much smaller with B5. Generally, reductions of 0.050 to 0.300 ppbv are estimated for the island of Montréal for B20, whereas an increase of less than 0.100 ppbv is predicted for surrounding areas (Figure VI-5 in Appendix VI). For the region downwind of Montréal, i.e., east of the island, reductions in O_3 concentrations up to 0.200 ppbv from the use of B20 in on-road HDDVs are modelled.

For both B5 and B20 scenarios, it is possible to distinguish the effects of highways on O_3 levels. The greatest reductions are in areas of dense traffic such as the Champlain, Jacques-Cartier and Victoria bridges, the Lafontaine tunnel, the Bonaventure expressway and Highway 15 leading to St-Jérôme.

6.3.1.2 $PM_{2.5}$

The impact of B5 on $PM_{2.5}$ concentrations is much less than that of B20 and is more homogeneous throughout the modelling domain. Only slight reductions (0.010 µg/m^3) are expected across the island of Montréal. The B20 scenario results in reductions above water bodies and along major highways (highways 20, 15, and 40 notably) and bridges (Figure VI-6 in Appendix VI). Results also suggest that urban air masses bring about a decrease in $PM_{2.5}$ concentrations eastward across the St. Lawrence River.

Because the impact of B20 on $PM_{2.5}$ HDDV emissions is four times that of B5, a proportional and scalable response on ambient levels is generally expected between scenarios. However, this proportionality is not observed, possibly due to the very small changes in $PM_{2.5}$ air concentrations under both scenarios (see section 6.2.1.2).

6.3.1.3 NO_2

Variations in NO_2 concentrations under the B5 and B20 scenarios are similarly distributed. Increases in NO_2 levels are limited to the island of Montréal and the major highways (e.g., 40, 20, 15, and 10). The area east of the island, located downwind, is also impacted by an increase in NO_2 ambient concentrations. Results from the B20 scenario are shown in Figure VI-7 (Appendix VI).

NO_2 concentrations peak along major highways and in urban areas because on-road mobile source emissions, specifically HDDVs, are responsible for a large share of NO_2 emissions, and biodiesel is assumed to increase HDDV NO_X emissions. This is important due to nitrogen dioxide's significant impact on tropospheric ozone concentrations (NO_2 being an O_3 precursor).

6.3.1.4 Concentration changes by census division

In addition to assigning area-averaged CD concentrations (see Sub-Section 6.2.1.6), population-weighted CD concentrations for $PM_{2.5}$, PM_{10}, NO_2, O_3, and CO are derived from the high-resolution results based on population data from the 2006 Canadian census at the dissemination area level (DA).[65] This method provides an estimate of the average concentration to which a population in a CD may be exposed rather than the average concentration in the CD.

Concentration changes generated on a census division basis both with and without population weighting for the 19 CDs included in the urban domain (Figure VI-2 in Appendix VI) are quite similar (i.e., within an order of magnitude). The results generally show that changes in population-weighted concentrations are slightly larger (in absolute terms) than changes in area-averaged concentrations, except for O_3. Nonetheless, changes for all pollutants are generally small. For example, the maximum recorded average (for all 19 CDs) net change was an increase of less than 3.79 ppbv for CO (daily max), equal to a 0.6% increase, under the B20 scenario.

Average O_3 and NO_2 concentrations are expected to increase in all CDs and under both scenarios. $PM_{2.5}$ and PM_{10} concentrations are expected to decrease in all CDs under both B5 and B20. As for CO, B5 use decreases concentrations, while B20 increases the average CO concentration.

[65] A DA is the smallest standard geographic area, covering all Canada, for which all census data are disseminated. DAs are composed of one or more neighbouring blocks with a population of 400 to 700 persons (see http://www12.statcan.ca/English/census01/products/reference/dict/geo021.htm).

6.3.2 Biodiesel use in 2020

6.3.2.1 Ozone

The impact of biodiesel use on ozone concentrations in 2020 is negligible under both B5 and B20 scenarios. In 2020, on-road NO_X emissions will be considerably lower than the 2006 values, possibly diminishing the area affected by ozone titration over the island of Montréal. Under the B20 scenario, concentrations throughout most of the domain remain within 0.025 ppbv (Figure VI-8 in Appendix VI).

The greatest reduction is noted near the bridges crossing the St. Lawrence River (0.200 ppbv), where maximum NO_2 concentrations are modelled (Figure VI-10 in Appendix VI). This high-traffic zone is most affected by ozone titration because the presence of water limits the deposition of ozone, enhancing the titration effect.

6.3.2.2 $PM_{2.5}$

The modelling results for $PM_{2.5}$ concentrations diverge from other pollutants analyzed in this section as the spatial distribution of changes between the B5 and B20 scenarios are quite different. The B5 scenario results in no obvious modifications in concentrations at high-resolution. With the B20 case (Figure VI-9 in Appendix VI), $PM_{2.5}$ concentrations are expected to increase by approximately 0.002 to 0.010 $\mu g/m^3$ across the island of Montréal and immediate suburbs, and regions further east. Reasons for this projected increase in $PM_{2.5}$ concentration across the Montréal area are not clear, but it could be due to the small impact that biodiesel is expected to have on overall primary $PM_{2.5}$ emissions in 2020 in comparison to secondary PM formation.

6.3.2.3 NO_2

Changes in NO_2 emissions brought on by the use of biodiesel in 2020 are relatively smaller and less heterogeneous than in 2006. The spatial distribution of variations is similar under both B5 and B20 scenarios, but the amplitude of change is approximately ten-fold less for B5. For the island of Montréal and the regions downwind (i.e., east), an increase is expected under a B20 scenario (Figure VI-10 in Appendix VI), while minimal decreases are expected for the rest of the domain.

6.3.2.4 Concentration changes by census division

As expected, changes under the B20 scenario are greater than for the B5 scenario. Also, as noted for the 2006 scenarios, the population-weighted approach generates greater concentration changes than area-weighted interpolation, except for O_3 under the B20 scenario.

The results estimate that B5 reduces the average (for all 19 CDs) CO concentrations, but B20 increases average CO levels. Average O_3 and NO_2 concentrations are expected to increase in all cases with B5 or B20. PM_{10} and $PM_{2.5}$ concentrations are expected to increase under both B5 and B20 scenarios, which differs from the 2006 results. All of these changes in concentrations are

minimal. Averaged across the 19 CDs, the maximum relative change is 0.807% (0.025 ppbv) for NO_2 under the B20 scenario.

6.3.3 Discussion of 3-km grid modelling results

In general, use of the AURAMS model at 3-km grid resolution yields greater changes in emissions and concentrations for the 2006 scenarios compared to the 2020 projections. However, all estimated changes are very minor. The high-resolution modelling runs did nonetheless allow distinguishing spatial features in the modelling domain, such as major road links, which represent areas where air pollutant concentrations are more affected by biodiesel use.

It is noted that areas of ozone reduction are correlated with areas of greater NO_2 concentrations. This feature is most probably due to the O_3 scavenging effect of NO in high-traffic areas, which is co-emitted with NO_2,[66] although other reactions involving NO_2 are possible (e.g., removal of radicals). The results suggest that highly populated areas where NO_X emissions are usually greater because of traffic related sources will see decreases in tropospheric ozone in association with biodiesel use (B5 and B20 in 2006, and B5 in 2020). Ozone increases are anticipated outside the urban core.

Increases in O_3 are nonetheless predicted for the urban domain under the B20 scenario in 2020. Unlike in 2006, the NO_X regime in 2020 will have changed due, in part, to the important reductions in NO_X emissions from model 2010 and later vehicles, the effect being that ozone scavenging will diminish or disappear. Hence the increase in NO_X emissions from the use of biodiesel will not have the same impact as in other modelled scenarios.

Results for $PM_{2.5}$ were somewhat variable between scenarios and modelling year. Notably, the use of B20 in 2020 is projected to increase $PM_{2.5}$ concentrations, while the B5 scenario led to no definite change. The relationship between primary PM emissions and secondary PM formation is possibly responsible for these results.

6.4 Conclusion

In general, the proposed B5 and B20 biodiesel scenarios are associated with very minimal changes in emissions and ambient air concentrations of the pollutants analyzed. The B5 and B20 scenarios under 2006 conditions predict small but non-negligible changes in air quality, mostly in urban centres and surrounding areas. For the 2020 projections, changes in predicted air quality are very small and often close to model detection limits. The smaller impacts observed under the 2020 scenarios are related to the significant reductions in exhaust emissions for the baseline ULSD scenarios compared to 2006. These are partly due to the development and implementation

[66] NO emissions are generally much greater than NO_2 emissions, making up approximately 90% of NO_X emissions vs. 10% for NO_2.

of more stringent emission standards for the model year 2010 and later vehicles (see Chapters 4 and 5).

The results from this study concur with previous findings from Smyth (2003) and Morris et al. (2003). Smyth (2003) evaluated the impact on vehicle emissions and air quality from the use of B20 in the Lower Fraser Valley of British Columbia. The author concluded that even marked changes in diesel vehicle emissions result in minimal variations in total emissions (less than 0.5% for the CACs) and likely no noticeable effect on air quality. Morris et al. (2003), as part of a study by the US National Renewable Energy Laboratory, assessed the impact of biodiesel fuels on air quality in various air basins across the US. It is reported that changes in air pollutants concentrations of O_3, $PM_{2.5}$, PM_{10}, and CO from the use of biodiesel would likely lead to non-measurable impacts on air quality.

The use of AURAMS with a 3-km grid resolution in a major urban area also yields greater changes in emissions and concentrations for the 2006 scenarios compared to the 2020 projections. Nonetheless, all recorded changes are minimal. High-resolution modelling does not improve the accuracy of predicted concentrations, but it provides enhanced spatial resolution of air quality impacts, bringing to light different air quality phenomena caused by smaller scale meteorological regimes and a more detailed distribution of mobile emission sources, such as the impacts of major bridges and highways.

Analysis of modelling results on a census division basis suggested that even if average pollutant concentrations are expected to increase on a national scale, average concentrations in populated areas may decrease. For example, under the B20 scenario in 2006, $PM_{2.5}$ concentration decreases were recorded in 56 CDs representing 54% of the Canadian population, while the national average $PM_{2.5}$ concentration increased. The estimation of human health impacts from changes in air quality resulting from biodiesel use will be assessed in detail in Chapter 8.

6.5 Uncertainties and limitations

Modelling capabilities can be limited by internal algorithms and process representations, in addition to scenario assumptions, available emission inventories, emission spatial and temporal allocations, boundary conditions, meteorological input, etc. The evaluation of the AURAMS model is beyond the scope of this assessment, so this analysis of uncertainties and limitations focuses on emissions data and results.

The confidence level for some pollutants represents a major limitation. For example, due to the incompleteness of CO emission sources and sinks represented in the model, there is lower confidence in model processes regarding CO concentrations. Similarly, model validation is not possible for some pollutants, notably some individual toxic VOCs, because of a lack of field monitoring data. Although there are published studies using Canadian VOC measurements (e.g.,

Jiang et al. 1997) these measurements are not relevant for the current assessment since they cover limited geographical areas and older time periods. However, total VOC values in AURAMS are considered reliable as they are linked to ozone level estimates for which the model has been evaluated.[67] Given that some VOCs are carcinogens, health assessments could greatly benefit from modelling capabilities directed at individual species such as benzene and acetaldehyde.

Validation of the high-resolution modelling results and additional tests are warranted to allow better use of current modelling capabilities. Improvements in spatial allocation and traffic-demand modelling could also lead to street level resolution, which could provide interesting insight and allow for a more accurate evaluation of air quality impacts for the populations living in the vicinity of major roads.

6.6 References

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Chapter 7. Toxicity of Biodiesel

This chapter reviews the existing health effects information for biodiesel exhaust in comparison to diesel exhaust and is followed by sections examining the potential for inhalation exposure to the Bovine Spongiform Encephalopathy (BSE) infectious agent and allergenic proteins resulting from the combustion of biodiesel derived from tallow and soy, respectively. Background and toxicity information are also presented for major fuel additive categories that are likely to be used in biodiesel fuels in Canada. The chapter begins with a review of the main health effects for air pollutants of diesel exhaust.

7.1 Health Effect Summaries

7.1.1 Health effects associated with carbon monoxide

Carbon monoxide is formed by incomplete combustion of carbon-containing fuels and by photochemical reactions in the atmosphere (US EPA 2010a). Upon inhalation, CO binds reversibly with haemoglobin (Hb) in the blood and produces carboxyhaemoglobin (COHb); reducing the oxygen (O_2) carrying capacity in blood and inhibiting the release of O_2 from oxyhaemoglobin. High levels of COHb result in tissue hypoxia and cytotoxicity. Tissues that are most dependent on O_2 such as the brain, heart, skeletal muscle and the developing foetus are particularly sensitive to CO (World Health Organization 1999; US EPA 2010a).

The health effects of CO depend on the extent of CO exposure and compensatory changes including vasodilation and increased cardiac output. As a result, individuals with coronary heart disease (CHD) are most susceptible to CO (Health Canada 2010; US EPA 2010a). Other groups with either increased probability or increased severity of health effects from CO exposure include pregnant woman, foetuses and young infants, individuals with anaemia, diabetics, the elderly, children, and persons with chronic obstructive pulmonary disease (Health Canada 1994; US EPA 2010a). Analyses of results from clinical studies with CHD patients indicate that there is a statistically significant inverse relationship between COHb concentration and time to ST segment change (a sign of myocardial ischemia) or time to exercise-induced angina, with no evidence of a measurable threshold for CO (Health Canada 2010; US EPA 2010a).

With respect to short term exposures, the following health impacts have been associated with CO (US EPA 2010a):

- Respiratory morbidity;
- Cardiovascular morbidity;
- Neurological effects; and
- Mortality.

In terms of short term exposures to CO, the US EPA (2010a) considered the results from toxicological, controlled human exposure and epidemiologic studies, as well as evidence from

atmospheric sciences, ambient air analyses, and dosimetry in order to make causal determinations for different health outcomes. It was concluded that:

- Epidemiologic and toxicological studies are suggestive of a causal relationship between short-term exposures to CO and respiratory morbidity;
- A causal relationship is likely to exist between short-term exposures to CO and cardiovascular morbidity based on evidence from epidemiologic and controlled human studies combined with CO's role in limiting O_2 availability;
- Controlled human exposure and toxicological studies are suggestive of a causal relationship between short-term exposures to CO and neurological effects; and
- Epidemiologic evidence is suggestive of a causal relationship between short-term exposures to CO and mortality.

With respect to long term exposures, the following health impacts have been associated with CO (US EPA 2010a):

- Neurological effects; and
- Birth outcomes and developmental effects.

In terms of long term exposures to CO, the US EPA (2010a) considered the results from toxicological, controlled human exposure and epidemiologic studies as well as evidence from atmospheric sciences, ambient air analyses, and dosimetry in order to make causal determinations for different health outcomes. It was concluded that:

- Controlled human exposure and toxicological studies are suggestive of a causal relationship between long-term exposures to CO and neurological effects; and
- Evidence from epidemiologic and toxicological studies is suggestive of a causal relationship between long-term exposures to CO and birth outcomes and developmental effects.

7.1.2 Health effects associated with nitrogen dioxide

Nitrogen dioxide is generated by combustion processes. The principal source of ambient NO_2 is traffic emissions while the principal indoor sources are unvented or poorly vented heating appliances.

NO_2 and nitric oxide (NO) are the most hazardous species of nitrogen oxides (NO_x) and most health studies focus on the effects of these compounds. In the atmosphere, NO is easily converted to NO_2. While not as intensively studied as PM or ozone, NO_2 has been implicated in a variety of health effects in studies conducted around the world, and has been well linked to certain respiratory outcomes.

Research shows that the groups most vulnerable to NO_2 include those with pre-existing pulmonary conditions (asthmatics), the elderly, children and people with occupations that require them to be in close proximity to traffic. In studies that examined the concentration-response relationship between NO_2 and health effects, the concentration-response appeared linear within the observed range of values indicating that there is little evidence of an effect threshold (US EPA 2008).

In terms of short term exposures to NO_2, the US EPA (2008) considered the results from toxicological, controlled human exposure and epidemiologic studies as well as evidence from atmospheric chemistry and exposure assessment studies in order to make causal determinations for different health outcomes. It was concluded that recent studies provided scientific evidence that NO_2 is associated with a range of respiratory effects (below) and provide evidence sufficient to infer a likely causal relationship between short term NO_2 exposure and adverse effects on the respiratory system:

- Impaired host-defence systems and increased risk of susceptibility to both viral and bacterial infections after NO_2 exposures;
- Evidence of airway inflammation, particularly in the more sensitive subgroups such as children or asthmatics;
- Increased airway responsiveness to specific allergen challenges;
- An association of respiratory effects with indoor and personal NO_2 exposures in children at ambient concentration levels;
- An association between short-term ambient NO_2 concentrations and increased emergency department visits and hospital admissions for respiratory causes, especially asthma.

In terms of long term exposures to NO_2, the US EPA (2008) considered the results from toxicological, controlled human exposure and epidemiologic studies as well as evidence from atmospheric chemistry and exposure assessment studies in order to make causal determinations for different health outcomes. It was concluded that epidemiologic and toxicological evidence examining the effect of long term exposure to NO_2 on respiratory morbidity (decrements in lung function, asthma, respiratory symptoms) is suggestive but not sufficient to infer a causal relationship.

7.1.3 Health effects associated with particulate matter

Respirable particles are usually categorized into various size fractions including coarse ($PM_{10-2.5}$), fine ($PM_{2.5}$) and ultrafine ($PM_{0.1}$) with approximate mass median aerodynamic diameters of 2.5 µm to 10 µm, 2.5 µm and 0.1 µm, respectively (Health Canada 2008). Many studies have been conducted to determine the health risks posed by exposure to particles. While the focus of most research has been on $PM_{2.5}$, attention has also been paid to $PM_{10-2.5}$ and $PM_{0.1}$ and some other specific size fractions and chemically oriented moieties. Overall, the scientific database

indicates a range of significant effects at the individual and population level can be attributed to PM of various sizes.

$PM_{10-2.5}$ can deposit widely within the lungs but deposits primarily in the upper portions of the respiratory system, while both $PM_{2.5}$ and $PM_{0.1}$ penetrate deep into the lungs and deposit to a greater extent in this region. Each of these size fractions can elicit a range of physiological responses (Health Canada 2008). Research results show that the groups most sensitive to PM (particularly $PM_{2.5}$, for which the evidence of health effects is strongest) are individuals with certain pre-existing diseases including individuals with asthma, chronic obstructive pulmonary disease, cardiovascular disease and diabetes. Children, especially asthmatics, older adults with cardiovascular disease and subgroups with greater exposure are more affected by PM exposure (Health Canada 2011). Most research to date has identified no threshold for PM, meaning that there is no clear evidence of a level of exposure below which health effects are not observed (Health Canada 2011).

With respect to short term exposures, the health impacts have been associated predominantly with $PM_{2.5.}$ The remainder of this section summarizes the conclusions for various categories of health effects from a recent Canadian assessment of the health effects of ambient fine PM (Health Canada 2011). These are generally similar to the conclusions of the most recent US EPA (2009) assessment.

- With respect to respiratory morbidity (including decreased lung function, increased respiratory symptoms, and pulmonary inflammation), the evidence from epidemiologic, controlled human exposure and toxicological studies exhibits strength of association, robustness, consistency, biological plausibility and coherence. It therefore provides a basis for concluding that there is a causal relationship between respiratory morbidity and acute exposure to $PM_{2.5.}$ This results in increased respiratory ERVs and hospitalizations;
- The mechanisms that appear to underlie acute PM morbidity are also applicable to acute exposure mortality. PM appears to increase all-cause and cardiorespiratory mortality, with the identification of those with respiratory infection as being particularly vulnerable; this is consistent with a progression of the inflammatory, cell damage and biochemical mechanisms observed upon acute PM exposure, and the morbidity outcomes described above. As such the evidence from epidemiologic, controlled human exposure and toxicological studies provides a basis for concluding that there is a causal relationship between acute exposure to $PM_{2.5}$ and acute respiratory mortality; and
- The consistent finding of elevated risk for hospital visits and premature mortality from cardiovascular causes in relation to PM exposure, along with the supporting work from panel and toxicological studies illustrating a range of potential mechanisms in relation to altered and impaired cardiovascular function, exhibits strength of association, robustness, consistency, biological plausibility, and coherence. The database therefore provides a basis for concluding that there is a causal relationship between acute exposure to $PM_{2.5}$ and cardiovascular morbidity and mortality.

With respect to long term exposures, the following health impacts have been associated predominantly with $PM_{2.5}$ (Health Canada 2011). These conclusions for various categories of health effects, which are taken from a recent Canadian assessment, are similar to the conclusions of the most recent U.S. EPA (2009) assessment.

- Evidence from epidemiological and animal toxicology studies is suggestive of a causal relationship between chronic PM exposure and respiratory morbidity. However, the possible role of other pollutants and the relatively small number of studies indicates that there is currently inadequate evidence to draw a more definitive conclusion;

- Many epidemiological studies of premature mortality and chronic exposure to PM have reported significant associations for cardiopulmonary mortality. However, relatively few have examined respiratory mortality in isolation, and in those that have, the findings have been somewhat inconsistent and not subject to detailed analysis. Thus, current evidence is inadequate to draw any conclusions with regard to the causal linkages between chronic exposure to PM and respiratory mortality;

- With respect to cardiovascular morbidity associated with long term exposure to PM, mechanistic findings from panel and animal toxicology studies provide biological plausibility for the effects seen in both the limited morbidity database and the more extensive chronic exposure mortality database. Because the evidence for morbidity is so limited in size and scope, the evidence for cardiovascular morbidity in relation to chronic PM exposure is suggestive of a causal relationship at this time. However, based on the consistency of the mortality database, the strong mechanistic support, and the general coherence of the database, the evidence for premature cardiovascular mortality indicates that there is a causal relationship between chronic exposure to ambient $PM_{2.5}$ and cardiovascular mortality.
- Given the importance of cardiovascular mortality in overall non-accidental mortality, and the preceding conclusion that chronic exposure to ambient PM is causally related to cardiovascular mortality, it is not surprising that exposure to PM is also consistently associated with all-cause mortality, and it is concluded that there is a causal relationship between exposure to ambient PM and total non-accidental mortality.
- There are important indications from epidemiology studies that lung cancer mortality is associated with long-term exposure to PM. While animal studies done at high exposures provide some mechanistic plausibility for the ability of PM to instigate lung cancer, the epidemiological findings are such that only lung cancer mortality has been evaluated. Though this database is limited, the findings in the strongest studies to date are indicative that there is likely a causal relationship with lung cancer mortality.
- Ambient PM has been associated with reproductive and developmental outcomes, including post-natal mortality, preterm birth, intrauterine growth retardation, and low birth weight in some epidemiological studies. However, associations with other pollutants such as CO and NO_2, as well as with roadway exposure metrics, were often stronger and/or more consistent. Overall, the evidence to date on these outcomes provides evidence that is suggestive of a causal relationship. Nevertheless, additional

understanding of the multi-pollutant issue is necessary in order to better understand the implications associated with these findings.

7.1.4 Health effects associated with ozone

Ground-level ozone (O_3) is a pollutant that is formed in the atmosphere primarily from nitrogen oxides and volatile organic compounds (Health Canada 2008). Exposure to O_3 has been demonstrated to cause various pulmonary effects in healthy individuals but is especially problematic for those with existing cardiovascular and pulmonary disease (Health Canada 2011; US EPA 2006). Research results show that the groups most sensitive to ozone pollution are the elderly, adolescents, children with asthma, and individuals who engage in outdoor activities (e.g., outdoor sports, outdoor workers) (Health Canada 2011). Studies to determine whether there is a safe level of ozone exposure suggest that there is no threshold or that, if one exists, it is much lower than the values established by the US EPA and international regulations (Bell et al. 2006).

With respect to short term exposures, the following health impacts have been associated with ozone (Health Canada 2011). These are similar to the conclusions of an earlier US EPA (2006) assessment.

- Epidemiological associations with respiratory health endpoints with support from the human clinical and animal toxicological findings exhibit strength of association, robustness, consistency, biological plausibility, and coherence. Hence, the overall evidence indicates that there is a causal relationship between acute exposure to ambient ozone and increased respiratory morbidity (including decreased lung function, as well as increases in respiratory symptoms, airway injury and inflammation, and airway hyper-responsiveness), resulting in increased asthma emergency room visits and respiratory hospitalizations.
- Animal studies indicate that exposure to ozone at relevant levels can affect the cardiovascular system, however the limited epidemiological evidence is somewhat lacking in consistency, robustness and coherence. Overall, the evidence is suggestive of a causal relationship between short-term exposure to ozone and cardiovascular morbidity, though the database is limited and more research is needed.
- Knowledge of the specific mechanisms that may underlie acute ozone-related mortality remains limited, though there are plausible pathways by which it could increase the risk of death from respiratory or cardiovascular causes. While there is uncertainty for specific causes of death, the associations with total non-accidental and cardiopulmonary mortality clearly display strength of association, robustness, and consistency. Therefore, the overall evidence indicates that there is likely a causal relationship between acute exposure to ambient ozone and non-accidental and cardiopulmonary mortality.

With respect to long term exposures, the following health impacts have been associated with ozone (Health Canada 2011). These are similar to the conclusions of an earlier US EPA (2006) assessment.

- Overall, the limited available evidence is suggestive of a causal relationship between long-term exposure to ambient ozone and each of lung function growth in children, asthma development, respiratory mortality, and morphological changes in the respiratory tract. However, for each of these the database is limited in size and scope, and more research is needed.

7.1.5 Health effects associated with acetaldehyde

Acetaldehyde is ubiquitous in the environment and is a product of numerous natural, industrial and combustion processes (including the combustion of fossil fuels). Acetaldehyde can also be produced photochemically from hydrocarbons in the atmosphere. Indoor levels of acetaldehyde can be impacted by combustion, building materials, cooking and consumer products (HEI 2007).

In the general population, acetaldehyde is an irritant of the eyes, skin, and respiratory tract at concentrations of about 45 mg/m^3 (HEI 2007; Environment Canada and Health Canada 2000a).

Susceptible populations include segments of the Asian and South American Indian populations because of a polymorphism of the $ALDH_1$ gene, which results in a non-functional enzyme. Affected individuals are less able to metabolize systemic acetaldehyde and show elevated levels in the blood after exposure to ethanol (which metabolizes to acetaldehyde). Within the $ALDH_1$-deficient population, an alcohol-induced asthmatic reaction may occur (US EPA 1999).

With respect to short term exposures, the following health impacts have been associated with inhalation of acetaldehyde in animals and/or humans (Environment Canada and Health Canada 2000a; HEI 2007):

- Irritation of respiratory tract, skin and eyes;
- Degenerative changes in the olfactory/respiratory epithelia; and
- Exacerbation of asthma.

With respect to long term exposures, the following health impacts have been associated with acetaldehyde in animals and/or humans (HEI 2007; Environment Canada and Health Canada 2000a; IARC 1999):

- Tissue damage in the respiratory tract; and
- Carcinogenesis in respiratory tract (animals).

Acetaldehyde was found to be “toxic” under The Canadian Environmental Protection Act (CEPA) (1999) based on its contribution to the formation of ground-level ozone, and the genetic component in the induction of tumours in the upper respiratory tract following acetaldehyde inhalation (Environment Canada and Health Canada 2000). In addition, acetaldehyde was classified by the International Agency for Research on Cancer (IARC) (1999) as possibly

carcinogenic to humans (Group 2B) based on evidence in animals, and as a probable human carcinogen (Group B2) based on evidence in animals by the US EPA (1991).

7.1.6 Health effects associated with formaldehyde

Formaldehyde is ubiquitous in the environment as a result of natural processes and anthropogenic sources, such as fuel combustion. In addition, formaldehyde forms secondarily in the atmosphere from the oxidation of natural and anthropogenic organic compounds present in air. Formaldehyde is also produced in the body as part of normal metabolism (Environment Canada and Health Canada 2001; HEI 2007).

Indoor air concentrations of formaldehyde are impacted by combustion sources and off-gassing of certain consumer products (Health Canada 2006). There is some evidence that exposure to formaldehyde in indoor air increases the occurrence of asthma symptoms in children (HEI 2007).

Due to its high reactivity and water solubility, formaldehyde principally impacts those tissues with which it first comes into contact (Environment Canada and Health Canada 2001). Threshold concentrations of 0.6–1.2 mg/m^3 for eye, nose, and throat irritation have been noted in studies with volunteers (HEI 2007).

With respect to short term exposures, the following health impacts have been associated with inhalation of formaldehyde in animals and/or humans (ATSDR 1999; HEI 2007):

- Irritation of respiratory tract, skin and eyes;
- Effects on lung function;
- Effects on behaviour and performance, motor activity; and
- Allergic responses and/or asthma.

With respect to long term exposures, the following health impacts have been associated with formaldehyde inhalation in animals and/or humans (ATSDR 1999; IARC 2006; Environment Canada and Health Canada 2001; Health Canada 2006; HEI 2007):

- Adverse effects on respiratory epithelium (particularly hyperplasia and metaplasia), cell death and evidence of irritation and inflammation;
- Carcinogenesis in nasal tissues (nasopharyngeal cancer); and
- Allergic responses and/or asthma.

Formaldehyde was declared “toxic” under CEPA 1999, due to its contribution to the formation of tropospheric ozone and to the carcinogenic and non-cancer hazard posed to humans (Environment Canada and Health Canada 2001). The US EPA classified formaldehyde as a probable human carcinogen (group B1) (US EPA 1991), although this is currently under review (US EPA 2010b). In addition, IARC (2006) reclassified formaldehyde as carcinogenic to humans (Group 1), based on “sufficient” evidence in humans of nasopharyngeal cancers and “strong”

human evidence of formaldehyde causing leukemia, as well as "sufficient" evidence in animals of carcinogenicity.

7.1.7 Health effects associated with 1,3-butadiene

1,3-Butadiene is formed by the incomplete combustion of olefins in gasoline and diesel fuel. Other sources include emissions from synthetic rubber and plastic factories, cigarette smoke, and forest fires. The major source of 1,3-butadiene in indoor air was identified as smoking, however wood heating and infiltration of automobile exhaust into the home are also potential sources (Environment Canada and Health Canada 2000b; HEI 2007).

There is no information to suggest that certain subpopulations have greater toxicological susceptibility to 1,3-butadiene than does the general population (US EPA 2002a).

With respect to short term exposures, the following health impacts have been associated with 1,3-butadiene in animals and/or humans (Environment Canada and Health Canada 2000b; HEI 2007):

- Developmental effects (foetal anomalies in mice).

With respect to long term exposures, the following health impacts have been associated with 1,3-butadiene in animals and/or humans (Environment Canada and Health Canada 2000b; HEI 2007; US EPA 2002a; IARC 2008):

- Carcinogenicity by inhalation (lymphohaematopoietic cancers in humans, variety of tumours in animals);
- Genotoxicity of 1,3-butadiene metabolites; and
- Reproductive effects (gonadal atrophy in mice).

In Canada, 1,3-butadiene was declared "toxic" under CEPA 1999 due to the high likelihood of it being carcinogenic in humans (Environment Canada and Health Canada 2000b). In addition, this compound is classified as carcinogenic (*Group 1*) to humans by IARC (2008) and as carcinogenic to humans by inhalation according to the US EPA (2002a).

7.1.8 Health effects associated with acrolein

Acrolein is commonly found in smoke from burning organic matter and is released into the ambient air through the combustion of petrochemical fuels and tobacco. Acrolein is also a by-product of 1,3-butadiene reactions in the atmosphere. A significant indoor source of acrolein is cigarette smoke (HEI 2007; Environment Canada and Health Canada 2000c).

Acrolein is a reactive, water soluble compound and for this reason is not expected to be distributed beyond the upper respiratory tract. Therefore impacts are mostly localized in these tissues. Acrolein is a respiratory irritant in humans at relatively low concentrations (700 $\mu g/m^3$)

(HEI 2007; Environment Canada and Health Canada 2000c). Children who are sensitive to irritants in the air (such as children with asthma and reactive airway dysfunction) may be more sensitive to lung irritation from acrolein (ATSDR 2007a).

With respect to short term exposures, the following health impacts have been associated with acrolein in animals and/or humans (Environment Canada and Health Canada 2000c; HEI 2007; ATSDR 2007a):

- Upper respiratory tract irritation; and
- Decreased body weight and pulmonary function, as well as histopathological changes in the nose, airways and lung (animals).

With respect to long term exposures, the following health impacts have been associated with acrolein in animals and/or humans (Environment Canada and Health Canada 2000c; HEI 2007):

- Upper respiratory tract irritant; and
- Histopathological effects in respiratory tract (animals).

Acrolein was found to be "toxic" under CEPA 1999 (Environment Canada and Health Canada 2000c) based on impacts on human health.

7.1.9 Health effects associated with benzene

Benzene is an organic compound found naturally in the environment in low concentrations. Most ambient benzene, however, is derived from emissions of coal and oil combustion, vehicle exhaust (gasoline and diesel), evaporative losses and refuelling emissions, evaporation of industrial solvents and hazardous waste sites. Indoor sources include tobacco smoke, building materials and furniture, household products and ambient air entering via ventilation and infiltration including air from attached garages (HEI 2007; WHO 2010; Hun et al. 2011).

Significant sources of variability in the population stem from genetic polymorphisms in key enzymes involved in the metabolism of benzene. These differences may increase or decrease an individual's susceptibility to the toxic effects of benzene (EPA 2002b; ATSDR 2007b).

With respect to short term exposures, the following health impacts have been associated with benzene in animals and/or humans (ATSDR 2007b; Environment Canada and Health and Welfare Canada 1993; Health Canada 2009):

- Blood and bone marrow toxicity;
- Immunotoxicity (animals);
- Neurotoxicity (high exposures: animals/humans); and
- Developmental and reproductive toxicity (high exposures: animals).

With respect to long term exposures, the following health impacts have been associated with benzene in animals and/or humans (ATSDR 2007b; IARC 1987; Environment Canada and Health and Welfare Canada 1993; Health Canada 2009):

- Blood and bone marrow toxicity;
- Neurotoxicity (occupational/humans);
- Carcinogenicity (acute myelogenous leukemia in humans exposed occupationally and various tumours in animals); and
- Genotoxicity (clastogen).

Benzene was found to be "toxic" under CEPA 1999, due to its classification as carcinogenic to humans. In addition, benzene is classified as a known human carcinogen (Group A) by the US EPA (2000), and as carcinogenic to humans (Group 1) by IARC (1987).

7.1.10 Health effects associated with polycyclic aromatic hydrocarbons

Polycyclic aromatic hydrocarbons (PAHs) are a component of polycyclic organic matter (POM). POM compounds with five or more benzene rings are usually associated with particulate matter, while compounds with four or fewer rings are semi-volatile and are partitioned between the particulate and gaseous phase (HEI 2007). PAHs are formed during the incomplete combustion of coal, oil, gas, wood, garbage, or other organic substances such as tobacco and charbroiled meat. PAHs normally occur as complex mixtures and enter the environment mostly as releases to air from volcanoes, forest fires, residential wood burning, exhaust from automobiles and trucks, and home-heating oil (ATSDR 1995; HEI 2007). Tobacco and wood smoke, cooking and outdoor sources may contribute to indoor PAH concentrations (HEI 2007).

There are over one hundred PAHs and the toxicological properties of most are not fully understood. The majority of toxicological information for PAHs is related to benzo[*a*]pyrene and 7,12-dimethylbenz[*a*]anthracene (ATSDR 1995; HEI 2007).

The following health impacts have been associated with PAHs in animals and/or humans:

- Carcinogenicity; and
- Genotoxicity in *in-vitro* and *in vivo* test systems.

Epidemiologic studies have reported an increase in lung cancer in humans in occupational settings when exposed to complex mixtures of POM compounds including certain PAHs, and animal studies have reported respiratory tract and upper digestive tract tumors from inhalation exposure to benzo[*a*]pyrene and PAH mixtures (ATSDR 1995; HEI 2007; US EPA 2011; IARC 2010; Environment Canada and Health and Welfare Canada 1994).

The Government of Canada (1994) classified benzo[*a*]pyrene, benzo[*b*]fluoranthene, benzo[*j*]fluoranthene, benzo[*k*]fluoranthene, and indeno[*1,2,3-cd*] pyrene as probably

carcinogenic to humans and therefore "toxic" under CEPA 1999. More recently, naphthalene was declared "CEPA toxic" on the basis of cancer and non-cancer effects (Environment Canada and Health Canada 2008). The US EPA has classified seven PAHs (benzo[*a*]pyrene, benz[a]anthracene, chrysene, benzo[b]fluoranthene, benzo[k]fluoranthene, dibenz[a,h]anthracene, and indeno[1,2,3-cd]pyrene) as probable human carcinogens (Group B2) and naphthalene as a possible human carcinogen (US EPA 1998b; US EPA 2011). In addition, IARC (2010) made the following classifications: benzo[a]pyrene as carcinogenic to humans; cyclopenta[cd]pyrene, dibenz[a,h]anthracene and dibenzo[a,l]pyrene as probably carcinogenic to humans; and benz[*j*]aceanthrylene, benz[*a*]anthracene, benzo[*b*]fluoranthene, benzo[*j*]fluoranthene, benzo[*k*]fluoranthene, benzo[*c*]phenanthrene, chrysene, dibenzo[*a,h*]pyrene, dibenzo[*a,i*]-pyrene, indeno[1,2,3-*cd*]pyrene and 5-methylchrysene as possibly carcinogenic to humans.

7.2 Toxicological Review of Biodiesel Exhaust

The following review examines the existing health effects information for biodiesel exhaust. The objectives are twofold depending on the information available: 1) to determine if biodiesel exhaust has a similar, reduced or greater impact than petroleum diesel exhaust in terms of specific health effects and outcomes; and 2) to attribute, where possible, any difference in the magnitude of effects observed to a change in the level of a specific physicochemical parameter(s).

Data relating to biodiesel toxicity was taken from the National Biodiesel Board's Tier 2 testing of biodiesel exhaust emissions (NBB 2000a/Finch et al. 2002), as well as from the broader scientific literature.

Diesel exhaust (DE) is a mixture of vapour phase chemicals and diesel exhaust particulates (DEP), formed by the combustion of diesel fuel (Hesterberg et al. 2006). DEP is characterized as elemental carbon (EC), organic mass (organic carbon, hydrogen, oxygen, and other elements associated with organic carbon), and inorganic ions (sulphate/nitrate). While the composition of the emissions may vary depending on the conditions of fuel use, DE typically contains carbon dioxide (CO_2), carbon monoxide (CO), nitrogen oxides (NO_X), sulphur dioxide (SO_2), methane and other volatile organic compounds (VOCs), as well as diesel particulate matter (Hesterberg et al. 2006).

In terms of health effects, DE exposure has been consistently associated with irritation and inflammation of the respiratory tract, increased allergic response and a compromised immune system. In addition, emerging studies suggest that DE exposure may be correlated with cardiovascular disease end points. While data on the carcinogenicity of DE are conflicting, the IARC has identified DE as *probably carcinogenic to humans* (IARC 1989). Other potential effects (e.g., neurological and reproductive) have been noted in the literature but the consistency

of these effects is not as well documented as the aforementioned respiratory and immunological effects.

Particulate matter (PM) is an important component of DE and has been extensively studied in terms of health impacts. The primary health effects associated with PM (DEP and ambient PM) in humans and animals occur in the respiratory and cardiovascular systems. Effects have also been noted on the immune and central nervous systems.

Quantitative characterization of biodiesel exhaust in terms of its individual components (vapour phase chemicals and PM) is complicated by the impacts of various factors including the type of feedstock used, the level of blending, the use of after-treatment devices and engine parameters. The information presented indicates that the use of biodiesel blends is expected to lead to reductions in emissions of CO, hydrocarbons, VOCs and PM mass. In terms of PM, some evidence suggests that biodiesel use may result in a downward shift in the particle size distribution (resulting in an increase of ultrafine particles) and an increase in the PM organic fraction. SO_2 emissions are expected to remain constant and NO_X may increase slightly.

Information for individual health effects (respiratory, immunological, cardiovascular, carcinogenicity, reproductive and developmental, neurological, and systemic) and specific outcomes are presented here for biodiesel exhaust. The focus of the assessment is exposure to biodiesel exhaust by inhalation for the general population; however, dermal exposure is also considered (see Section 7.2.7) given that potential exposure may occur during refuelling. The most informative studies examining the toxicity of biodiesel are those in which an equivalent treatment with DE was included for comparative purposes.

7.2.1 Respiratory effects

7.2.1.1 Biodiesel exhaust

The following text reviews the information on respiratory effects resulting from exposure to biodiesel exhaust and focuses on the key outcomes: inflammation; and histopathology and lung function effects.

Inflammation

The National Biodiesel Board (2000a)/Finch et al. (2002) conducted a subchronic inhalation exposure study as part of the US EPA Tier 2 testing requirements for fuel/fuel additives detailed in Title 40 of the US Code of Federal Regulations (CFR) Part 79. The study with F344 rats was conducted to determine the potential toxicity of biodiesel (soybean-derived) exhaust emissions. Twenty male and twenty female rats, per exposure group, were exposed for 6 hours/day, for 5-7 days/week for a total of 73-75 days. Rats were exposed by inhalation to diluted exhaust (from a Cummins diesel engine operated according to the transient US EPA heavy-duty engine dynamometer schedule (40 U.S. CFR, Part 86) burning 100% soybean-derived fuel) or clean air

(control). Whole emissions were diluted to targeted NO_x concentrations of 5, 25, and 50 ppm (low, intermediate, and high) levels. The highest NO_x exposure level (50 ppm) was considered to be an appropriate upper-bound to avoid conducting a study primarily reflecting high-level NO_x toxicity. The treatment levels corresponded to 0.04, 0.2, and 0.5 mg particles/m^3, respectively. The particle size distribution of biodiesel exhaust particulates expressed as a combined (calculated) Mass Median Aerodynamic Diameter (µm) was calculated at the beginning and end of the exposure period (13 weeks) for the three treatment levels: low (0.14, 0.58); intermediate (0.31, 0.40); and high (1.15, 0.06).

Rats underwent histopathology examination for the incidence and severity of chronic inflammation, as determined by the presence of inflammatory cells in the lung, after the 13-week exposure period. With the exception of one control (male), chronic inflammation was not found to be present in any of the treatment groups (NBB 2000a; Finch et al. 2002). The study did not include DE exposures.

An *in vitro* study by Swanson et al. (2009) evaluated the effect of 24-hour exposure to PM organic extracts from Standard Reference Material 1975 (SRM 1975) (an extract of diesel PM from a diesel-powered forklift), soy ethyl ester (SEE), soy methyl ester (SME), Phillips No.2 diesel (D2) (sulphur content 0.036%) and a solvent blank on BEAS-2B cells (human airway epithelial cell line). A 1997 Caterpillar 3406E heavy-duty six cylinder, four stroke, turbocharged, after-cooled 14.6 L engine was used with the EPA Heavy-Duty Engine Transient Test Cycle to generate the exhaust. Samples were extracted with dichloromethane. A cytokine assay was performed using IL-8 and IL-6 cytokines as markers of the pro-inflammatory response induced in BEAS-2B cells exposed for 24 hours to different fuel emissions particle extracts (SRM 1975, D2, SEE+SME, SEE, and SME) at doses of 10, 25, 50, 184, 369, 922, and 1,845 µg PM eq/ml for SRM 1975, and 10, 25, and 40 µg PM eq/ml for D2, SEE, and SME. Three separate experiments were carried out in order to determine the IL-8 and IL-6 response (Tables 7-1 and 7-2).

Table 7-1 Summary of IL-8 response (protein/µg PM eq) for experiments by test material

	Experiment 1 (median)	Experiment 2 (median)	Experiment 3 (median)	Average slope for IL-8
Solvent Blank	-0.021	-2.31	0.231	-0.701
SRM 1975		-1.54	0.191	-0.675
Diesel (D2)	-0.100	5.9	0.291	2.03
SEE+SME	0.516*	13.4	0.972**,+	4.97
SEE	0.525	12.7	1.67	4.96
SME	0.490	20.8	-0.416	6.97

Statistically significant difference at *p<0.05 and **p<0.01 from the solvent blank. +Statistically significant difference at p<0.05 from diesel.

Table 7-2 Summary of IL-6 response (protein/µg PM eq) for experiments by test material

	Experiment 1 (median)	Experiment 2 (median)	Experiment 3 (median)	Average. Slope for IL-6
Solvent Blank	0.043	0.028	0.017	0.029
SRM 1975		0.053	0.025	0.039
Diesel (D2)	0.000**	1.35*	0.049**	0.465
SEE+SME	0.351***	6.65**	0.128***	2.38
SEE	0.401	6.01	0.203	2.20
SME	0.211	8.13	-0.020	2.77

Statistically significant difference at *p<0.05, **p<0.01, and ***p<0.001 from the solvent blank

It was determined that biodiesel induced a dose-dependent increase in the release of pro-inflammatory cytokines (IL-6 and IL-8) by BEAS-2B cells that was higher than the cells exposed to either petroleum diesel extracts (D2 or SRM 1975). The average slope for IL-8 response for biodiesel (SME+SEE) (4.97) was approximately 2.5 times greater than D2 (2.03). The average slope for IL-6 response for biodiesel (2.38) was approximately 5 times greater than D2 (0.465). The slopes for SRM 1975, in both cases, were comparable to the solvent blank. Only in one experiment (No.3) for IL-8 response was the median value for biodiesel (SEE+SME) group statistically greater than D2 (Swanson et al. 2009). Responses were not dependent on changes in cell viability as this parameter was unchanged. Given the high variability for inflammatory response between experiments (Tables 7.1 and 7.2) the results should be interpreted with care.

The soluble organic fraction (SOF) of biodiesel extract appeared to be a more potent inflammatory stimulant in BEAS-2B cells in comparison to the SOF of two separate petroleum diesel PM fractions (diesel PM and SRM 1975). At a dose level of approximately 40 µg PM eq/ml, biodiesel SOF elicited an increased cytokine release from BEAS-2B cells, while petroleum diesel SOF required concentrations greater than 100 µg PM eq/ml to induce an increased cytokine response. The authors, however, believed that this data was too preliminary to draw any conclusions (Swanson et al. 2009). The significance of these results in terms of human exposure to biodiesel exhaust is not clear.

A study by Brito et al. (2010) examined inflammatory toxicity induced by inhalation of exhaust from 500 ppm sulphur diesel fuel, and SEE biodiesel fuel. A stationary diesel electrical generator (BD-2500 CFE; Branco) was used to generate the exhaust. Adult male BALB/c mice were exposed to B100, B50, diesel (97% diesel and 3% SEE), and a control group (filtered air) for 1 hour. Analysis of the bronchoalveolar lavage fluid (BALF) showed that there was a significant increase in the total number of cells in the BALF and pulmonary parenchyma after exposure to B100, B50, and diesel when compared to the control group.

In BALF, there was a significant increase in neutrophils (segmented cells) in the B50 and diesel groups, and a small insignificant increase in neutrophils in the B100 group, compared to the

control group. There was also a significant increase in macrophages in mice exposed to B100, B50 and diesel compared to the control group. In the pulmonary parenchyma, a significant increase in macrophages was observed for the B100 and B50 exposure groups compared to the control group (Brito et al. 2010). While the authors noted a significant increase in the total number of cells in the BALF and pulmonary parenchyma for all treatments compared to the control, there was no significant difference between the biodiesel and diesel treatments.

Tzamkiozis et al. (2010) investigated lung inflammation in female BALB/cJ mice exposed to exhaust particles. Vehicle testing was conducted in a variety of driving conditions to cover a range of real-world situations including the cold-start New European Driving Cycle followed by "Artemis" cycles which simulate urban, rural, and highway driving. Particulate samples were directly collected as waterborne suspensions using the versatile aerosol concentration enrichment system. One composite waterborne PM sample was collected per vehicle (Euro 3 gasoline (G Euro 3), diesel Euro 1 (Euro 2 car without oxidation catalyst), Euro 2 car fuelled with 100% soybean-derived biodiesel (B100), diesel Euro 4, and a diesel Euro 4 car retrofitted with a diesel particulate filter (DPF) (D Euro 4+)) over all driving cycles tested each day. Each sample was intratracheally instilled in 5 mice per concentration at 50 µL and 100 µL of PM or pure water (control). Mice were sacrificed 24 hours after exposure and analyzed for acute lung inflammation.

Bronchoalveolar lavage (BAL) fluid was assessed for the influx of polymorphonuclear neutrophils (PMNs) as a measure of the inflammatory cellular response and BAL protein concentration was measured as an indicator of lung injury (see Subsection *Histopathology and Lung Function*). Various emission characteristics were measured including the emission rates of trace elements and individual organic species.

Samples (both 50 and 100 µL) from animals exposed to B100 Euro 2 vehicle exhaust particles exhibited a small insignificant increase in BAL PMNs compared to the sham control. At the high dose level (100 µL), samples from the gasoline-powered Euro 3 vehicle and the diesel-powered Euro 1 induced a moderate but significant inflammatory response characterized by an influx of inflammatory PMNs (10^3 BAL PMN) into the airway when compared to the sham control: 73.1 ± 14; 46.9 ± 6.4 and 25.3 ± 1.8, respectively. PM from the gasoline engine at the high dose level elicited the greatest response compared to the sham control; however, none of the treatments were reported to be significantly different from each other.

The B100 Euro 2 vehicle emitted a higher content of sodium, phosphorous, sulphur, manganese, iron, cobalt, and zinc compared to the D Euro 4 and D Euro 1 vehicles, and significantly more potassium than all other vehicles. The B100 Euro 2 vehicle emitted a greater level of hopanes, steranes and organic acids compared to other vehicles, a lower level of alkanes compared to

other vehicles and higher levels of total PAHs compared to all vehicles except for the G Euro 3 car.

Tzamkiozis et al. (2010) examined the association between biological responses and different particle physicochemical characteristics. The strongest associations between inflammatory responses and PM characteristics were observed for the elements iron, manganese, copper, phosphorous, sulphur and lead as well as high molecular weight PAHs and the reactive oxygen species (ROS) induction capacity of the PM samples. In terms of the chemical species identified, the B100 Euro 2 PM sample ranked 2nd for phosphorous content, 3rd for sulphur and 2nd in terms of high molecular weight PAHs.

For the purposes of this evaluation, any comparison of the inflammation effects of biodiesel to diesel exhaust is not straight-forward given the use of various technologies. However, the information from the study indicates that in terms of the inflammatory cellular response (BAL PMNs), there was no significant difference between the B100 Euro 2 and the other treatments.

In an *in vitro* study, Jalava et al. (2010) looked at the acute inflammatory effect of exposure to particulate emissions from a non-road diesel engine operated with conventional diesel fuel (EN590) and two biofuels: hydrotreated fresh vegetable oil (HVO) and rapeseed methyl ester (RME) with and without a DOC/POC (diesel oxidation catalyst/particle oxidation catalyst) catalytic converter. A small industrial 1.123 L IDI (EPA Tier 1/EU stage II) non-road diesel engine (Kubota D1105-T) run on a dynamometer according to the international ISO standard steady state cycle C1 (8178-4:1996) was used to generate emissions. Methanol was used for high-efficiency extraction of particulate emissions from the sampling substrate in a high-volume cascade impactor. PM samples were characterized for elements, ions, and PAHs. Mouse macrophages (RAW264.7 cell line) were exposed to the PM samples at concentrations of 15, 50, 150, and 300 µg/ml for 24 hours and measured for inflammatory mediators: macrophage inflammatory protein (MIP-2) and tissue necrosis factor (TNF-α). Responses were not dependent on changes in cytotoxicity as this parameter was unchanged.

The MIP-2 and TNF-α responses increased in a dose response manner for EN590 and HVO. In the case of RME, the highest response was 150 µg/ml. The responses for HVO were slightly greater than those of EN590 and both appeared to be greater than that of RME. The authors indicated that there was no significant difference between fuels for MIP-2 and TNF-α response at the 150 µg/ml dose level. All treatments were significantly different from the controls at the 150 and 300 µg/ml dose levels. The use of a catalytic converter increased the MIP-2 and TNF-α response for EN590 and HVO samples but resulted in a slight decrease for RME samples. The authors indicated that there was no significant difference between catalyst treated and non treated samples.

Jalava et al. (2010) weighted the results for MIP-2 and TNF-α response with emission factors (mg/MJ) for each fuel. The small differences in toxicological activities between particles (described above) emitted from the engine operated with the different fuels were increased when the results were weighted with the emission factors. The strongest toxicological activities were in all cases associated with EN590 or HVO fuels. It was indicated that RME emission particles were less toxic in part because of lower particle mass emissions. For MIP-2 the relative responses without the catalyst were (mg/MJ): EN590 (4.8); HVO (5.0) and RME (3.0). TNF-α relative response without the catalyst were (mg/MJ): EN590 (4.5); HVO (4.1) and RME (2.9). All emission particles treated with the catalyst had lower relative toxicological activities.

Iron and zinc were the most abundant metals in all samples. Other metals included copper, manganese, nickel, lead and vanadium. The RME sample contained the highest level of total elements. The authors indicated that the relatively small difference in the elemental composition between fuels was possibly due to the use of lubricant oil and engine wear which was common to all treatments. Sodium (Na^+), ammonium (NH_4^+), and nitrate (NO_3^-) were the most common ions. Total PAHs were 109.7 ng/mg for conventional diesel, 50.7 ng/mg for HVO, and 37.6 ng/mg for RME. Heavy molecular weight PAHs were largely absent from the samples possibly due to the extraction method. The use of the catalytic converter resulted in significant increases in levels of Na^+ and NO_3^- and decreased emissions of metals (except Mn and Ni for EN590) and PAH levels.

Jalava et al. (2010) indicated that the absence of high-molecular-weight PAHs and the contribution of metals resulting from the use of lubricating oil may have impacted the results. Nonetheless, the results indicate that RME particles resulted in a smaller inflammatory response than conventional diesel fuel and HVO. However, the authors indicated there was no significant difference in biological responses between fuels. While this study investigated the presence of chemical constituents (elements, ions, and PAHs) in the different fuels, it did not determine specific associations or causal relationships between constituents and inflammatory responses.

In 2000, Le Prieur et al. evaluated antioxidant capacity and cytotoxicity (see Section 7.2.4), as well as the inflammatory response in rat lung slices exposed *in vitro* to whole (100%) or filtered (10, 15, 25, 60 and 85%) exhaust from DF, 100% RME or 30% RME (30% RME and 70% DF) (generated from a 5-horsepower one-cylinder Robin engine running at 3000 rpm with a load corresponding to a 1 bar water power outlet pressure) for three hours. Treatment with DE produced a statistically significant increase in TNF-α protein levels with increased concentrations, whereas supplementation with RME or filtration of each exhaust prevented an increase in TNF-α levels (Le Prieur et al. 2000).

Histopathology and Lung Function

In the 13-week subchronic inhalation study by NBB (2000a)/Finch et al. (2002), twenty male and twenty female F344 rats from each treatment group (see inflammation above for additional details) underwent an histological examination at sacrifice for the presence of certain lung lesions including the presence of dust-laden alveolar macrophages (AMs), alveolar macrophage hyperplasia, alveolar bronchiolarization, alveolar histiocytosis and centriacinar fibrosis. One female rat from the control group was moribund and sacrificed after four days of exposure. An additional ten male and ten female rats were taken from the high-level group after 13 weeks of exposure and 28 days of recovery for analysis of the incidence and severity of some lung lesions (dust-laden AMs, alveolar macrophage hyperplasia, alveolar bronchiolarization, and alveolar histiocytosis (NBB 2000a)).

A dose-dependent increase in the number of dust-laden AMs was observed in male and female rats. The authors noted that the severity of the dust-laden AMs was less in rats that had the 28-day recovery period, compared to animals with no recovery period (NBB 2000a).

A dose-dependent increase in the presence of alveolar macrophage hyperplasia was observed in both male and female rat lungs. The authors determined that the severity of the alveolar macrophage hyperplasia was less in rats that had the 28-day recovery period compared to rats with no recovery period. The authors also noted that there was a reduction in the cytoplasm of the rats with alveolar macrophage hyperplasia indicating a return toward a less active state (NBB 2000a).

Alveolar bronchiolarization (presence of ciliated and Clara cells lining the alveolar ducts and alveoli adjacent to the terminal bronchioles), a lesion indicative of tissue response to injury, was minimal in severity and found only in 4/30 high-level female rats (3/20 after 13-week exposure and 1/10 after 13-week exposure and a 28-day recovery period).

Alveolar histocytosis (aggregates of AMs associated with a reaction in the alveolar septa) was found in 1/20 control male rats, 1/19 control female rats, 1/20 intermediate-level male rats, 1/20 intermediate-level female rats, and 4/30 high-level female rats (2/20 after the final sacrifice and 2/10 after the 28-day recovery period).

Centriacinar fibrosis was seen infrequently in the study (1/20 intermediate-level male rats, 1/20 high-level male rats, and 1/19 control female rats) and could not be related with certainty to the level of exposure.

Based on the histologic findings in the lung, the authors set the no observable adverse effect level (NOAEL) at the intermediate exposure level (25 ppm NO_x or 0.2 mg particles/m^3). While dust-laden macrophages and increases in AMs were present in the intermediate-level group, and,

to a lesser extent, the low-level exposure group, the authors indicated that these findings were not accompanied by an influx of neutrophils and were judged to be normal physiological responses to particles inhaled and deposited within the lungs (NBB 2000a). The study did not include comparative DE exposures.

An *in vitro* study by Ackland et al. (2007) investigated the effects of exposure of a cultured human airway cell line (A549) to a control (ethanol) and various blends (B20, B40, B60, B80, B100) of biodiesel (feedstock not indicated) emissions particulate matter (BDEP) and petroleum diesel emissions particulate matter (PDEP) for five days. A 1979 Volkswagen Golf with a 1.6 litre engine but without an oxidation catalytic converter was run on the Economic Commission of Europe (ECE) Euro 2 drive test cycle, which consists of four urban driving cycles with a maximum speed of 50 km/h and one extra urban driving cycle with a maximum speed of 120 km/h. After a 20-minute sampling period, smoke-laden filters were removed from the sampling holder. Dichloromethane was used to extract the PM from the filters. A549 cells were treated for 5 days with particle concentrations (25 µg/ml) for both diesel and biodiesel and assayed for multinuclearity.

The authors found that B20 induced 52% of cells to be multinucleate compared to only 12% of cells treated with B100. The background multinucleate rate was 7%. There was a general reduction in the proportion of multinucleate cells as the relative proportion of BDEP in the culture medium increased, demonstrating a causal relationship between the formation of multinucleate cells and exposure to exhaust PM (petroleum) (Ackland et al. 2007).

In order to show that multinucleate cells are more likely to undergo apoptosis than cells with a single nucleus, active caspsase-3 (indicator of apoptosis) was measured in cells exposed to BDEP and PDEP. Multinucleate cells were found to have more active caspsase-3 staining compared to cells with a single nucleus. The authors concluded that PDEP was a much more effective inducer of apoptosis than BDEP. The results of the study suggest that diesel exhaust PM is more damaging to human airway cells than biodiesel exhaust PM and that exposure to diesel exhaust PM induces cell apoptosis which may contribute to the cell death observed in airway cells (Ackland et al. 2007).

While the result of this *in vitro* study indicates that diesel exhaust PM is more damaging than biodiesel to human airway cells, it is not clear how this finding translates to human exposure scenarios.

An *in vitro* study by Liu et al. (2008) examined the toxicity of exhaust emissions (more specifically the semi-volatile and particulate products in the exhaust) from the combustion of 6 blends of diesel and biodiesel (palm oil methyl ester): diesel, B10, B30, B50, B75, and B100. A 4-stroke, water-cooled, non-catalyst generator with a constant output power (13kW) was used as

the test engine to generate the samples (i.e. not a vehicular engine). The samples were extracted with dichloromethane and *n*-hexane mixture solvent (50/50, v/v).

The authors used the Microtox test to determine the TUV (toxicity unit per litre exhaust sampled) and the TUW (toxicity unit per μg soluble fraction of particulate). The Microtox test is based on the difference in light output between a control and treatment sample - exposure to a toxic test substance inhibits *V. Fischeri* (marine bacterium) and the bioluminescence of *V. Fischeri* is directly linked to respiratory activity and correlated with a toxic response. In addition, the 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) assay was used to ascertain cytotoxicity in lung epithelial cells (BEAS-2B) by determining viable cell numbers in relation to proliferation and cytotoxicity.

The results from the Microtox test revealed that diesel was the only fuel that had a higher TUV in the particulate phase than in the semi-volatile phase. For the other fuels, TUV's for the semi-volatile extracts were approximately three (B10 and B75) to five (B50) times greater than the particulate extracts suggesting that the semi-volatile phase of biodiesel exhaust is more toxic than the particulate phase. The authors determined that the TUW of particles was more meaningful than the TUV in evaluating unit toxicity. They noted that, according to the TUWs, diesel particulates were more toxic than biodiesel particulates.

The results of the MTT assay (Table 7-3) are expressed as cell viability as a percentage of the non-exposed control (dimethyl sulphoxide (DMSO)). DMSO exposure was associated with a cell viability of 93%. The results from the MTT assay indicated that the cell viability of particulate extracts exceeded 80% for the blank control and all test fuels. Only diesel and B10 exhibited particulate toxicity as demonstrated by the lower cell viability compared to the blank control. However, the particulate had minimal inhibitory effect on BEAS-2B cells (equal or 10% below the non-exposed control) and the authors concluded that the particulate extracts were not cytotoxic.

Semi-volatile extracts of B50 and B75 more strongly inhibited cell viability (Table 7-3). There was no apparent difference in cytotoxicity for semi-volatile extracts for diesel, B10, B30, and B100 when compared to the blank control. For each test fuel with the exception of diesel, the semi-volatile extract cytotoxicity (cell viability) was significantly greater than that of the particulate extract; a result that agreed with the results of the Microtox test.

The authors concluded based on the TUWs generated from the Microtox test, that soluble diesel particulates were more toxic than soluble biodiesel particulates. B50 particulates were found to be the most toxic of all biodiesel blends and diesel was associated with significant unit particulate toxicity. However, according to the results of the MTT assay, the lack of a substantial

drop in cell viability (see Table 7-3) indicated no significant difference between the cytotoxicity of diesel and biodiesel particulates.

Table 7-3 MTT Assay: cell viability % of particulate and semi-volatile extracts of test fuels

	Blank Control	Diesel	B10	B30	B50	B75	B100
Particulate Extracts	86	84 ± 1	83 ± 5	94 ± 5	95 ± 3	87 ± 7	96 ± 5
Semi-volatile Extracts	77 ± 6	82 ± 2	72 ± 2	77 ± 5	47 ± 2	66 ± 6	88 ± 3

Given the difference in results between the Microtox test and MTT assays in terms of the toxicity of diesel and biodiesel particulate extracts (i.e., significant unit toxicity for diesel versus biodiesel in Microtox test and lack of significant difference in cytotoxicity in MTT assay) the authors recommended additional studies focusing on the role of hazardous organics (such as PAHs or hydrocarbons) in inducing toxicity (Liu et al. 2008).

Swanson et al. (2009) evaluated the effect of 24-hour exposure to PM organic extracts from SRM 1975 (dichloromethane extract of diesel PM from a diesel-powered forklift), SEE, SME, petroleum diesel (D2) and a solvent blank on BEAS-2B cells (human airway epithelial cell line). Cell viability was determined using the MTT assay. Doses of 10, 25, 50, 184, 369, 922, and 1,845 µg PM eq/ml for SRM 1975, and 10, 25, and 40 µg PM eq/ml for petroleum diesel, SEE, and SME were used. The authors found that no biodiesel sample was consistently cytotoxic at the doses tested. This finding was based on the lack of changes in LDH activity, MTT metabolism, and the cells remaining attached and morphologically similar. In addition, SRM 1975 did not cause a reduction in cell viability (the article did not mention results for D2). Analysis of PAH content (13 PAH species ranging from 3-6 rings) was performed for biodiesel exhaust extracts (SEE and SME) and petroleum diesel. The authors found that PAH content for the biodiesel and petroleum diesel extracts were at, or below, the non-detectable limit for the 13 compounds tested.

Jalava et al. (2010) examined the cytotoxic effect of exposure to particulate emissions from a non-road diesel engine operated with conventional diesel fuel (EN590) and two biodiesels: HVO and RME with and without a DOC/POC catalytic converter. Exhaust gas samples were characterized for elements, ions, and PAHs (see subsection *Inflammation*).

Cell viability of macrophages was detected using the MTT assay. Mouse RAW264.7 macrophages were exposed for 24 hours, to a control and four dose levels (15, 50, 150, and 300 µg/ml) of PM samples generated from EN590, EN590+cat, HVO, HVO+cat, RME, and RME+cat. All samples for each treatment evoked a dose-dependent decrease in cell viability. And every dose, except 15 µg/ml of EN590, resulted in a statistically significant reduction in cell

viability compared to the control indicating acute cytotoxicity. The authors also determined that there were no major differences in cytotoxicity between samples from different fuels, with or without the use of the catalytic converter (Jalava et al. 2010).

Apoptotic response was detected using flow cytometric cell cycle analysis for all samples at dose levels of 15, 50, 150, 300 µg/ml and a control. At doses of 150 and 300 µg/ml for all treatments, and 50 µg/ml for HVO samples, exposure resulted in a significantly increased apoptotic response. The HVO emission particles exhibited the largest apoptotic response; however, the differences between each treatment were relatively small and not statistically significant. The use of a catalyst was shown to slightly reduce apoptosis caused by HVO and RME treatments but not for EN590 (Jalava et al. 2010).

Tzamkiozis et al. (2010) investigated the effect of exhaust PM on alveolar tissue injury in BALB/cJ mice acutely exposed to exhaust particles. Waterborne PM samples were collected from the exhaust of a gasoline Euro 3 car, a diesel Euro 1 car (Euro 2 car without oxidation catalyst), a Euro 2 car fuelled with 100% soybean-derived biodiesel (B100), a diesel Euro 4 vehicle, and a diesel Euro 4 car retrofitted with a DPF (D Euro 4+). Each sample was intratracheally instilled in 5 mice per concentration at 50 µL and 100 µL of PM or pure water (control). Mice were sacrificed 24 hours after exposure and analyzed for alveolar tissue injury by using protein concentration [µg/ml] in BAL fluid as a marker.

BAL protein concentration was significantly increased compared to the control for the high dose levels (D Euro 1, B100 Euro 2, D Euro 4, D Euro 4+) and low dose levels (D Euro 1, G Euro 3). There was no significant difference between the 50 µL B100 Euro 2 sample and the control group. PM from the D Euro 4 engine at the high dose level exhibited the greatest response compared to the control; however, none of the treatments were reported to be significantly different from each other.

For the purposes of this evaluation, any comparison of lung injury due to biodiesel to DE is not straight forward given the use of different vehicle technologies. However, the results indicate there was no significant difference in alveolar tissue injury between the Euro 2 vehicle, which used B100, and the other treatments.

A human cross-sectional study by Hasford et al. (1997) was conducted in which 763 employees were exposed to exhaust fumes from RME (n=381) or DF (n=382). Inhalation exposures were estimated by a score taking into account the characteristics of the job and the vehicle. The employees exposed to diesel served as the control group. Females were excluded from the analysis since the proportion of females to males was small in the RME and diesel groups. The study population consisted of truck drivers delivering goods over intermediate distances (10–100 km), rural road maintenance workers, drivers of heavy motor vehicles in agricultural research

and development facilities, and industrial fork lift truck drivers. The main findings of the study questionnaire suggested a dose-related association between the self-reported acute irritation of mucous membranes and the lower airways and the intensity of exposure to exhaust from RME and diesel fuel. Forty-six male volunteers were submitted to repeated pulmonary function tests before and after work periods (exposure to either RME or DE fumes) (Hasford et al. 1997).

The authors did not find any differences exceeding normal ranges between the results for RME compared to DE. There was also no difference in the reporting of the symptoms between RME and diesel emissions. Lung function tests did not reveal a risk for acute respiratory impairment by exhaust fumes from RME when compared to diesel fuel (Hasford et al. 1997). Given the lack of exposure information for this study, it is difficult to draw any conclusions with respect to biodiesel versus DE and impacts on lung function. The self-reporting of health effects and the fact that lung function tests were only administered to a limited number of volunteers were also limiting factors of this study.

Summary/conclusion

Exposure to DE and ambient PM have both been shown to cause inflammation effects in *in vitro* assays (Steerenberg et al. 1998; Dagher et al. 2005; Mitschik et al. 2008) and in experimental animals (Dye et al. 2001; Wise et al. 2006; Finnerty et al. 2007; U.S. EPA 2002c; U.S. EPA 2004). In addition, acute human experimental studies have provided evidence of mild airway inflammation resulting from DE exposure (Rudell et al. 1990; Rudell et al. 1994; Rudell et al. 1996; Nordenhäll et al. 2000). The evidence for inflammation indicates that there is an equal or reduced effect for biodiesel exhaust compared to DE. Of the three *in vivo* studies reviewed, a subchronic study (NBB 2000a/Finch et al. 2002) indicated no evidence of chronic inflammation due to biodiesel exhaust (it did not include a DE treatment) and two acute studies found no significant difference between biodiesel and diesel treatments (Brito et al. 2010; Tzamkiozis et al. 2010). Of the *in vitro* studies reviewed, Swanson et al. (2009) observed that the inflammatory response of human airway epithelial cells was higher for biodiesel (SEE and SME) compared to DE. However, it should be noted that the study was carried out with the SOF, there was considerable variation between experiments and only in one experiment out of three was the difference significant. Jalava et al. (2010) found that the inflammatory response of mouse macrophages exposed for 24 hours to particulate emissions was slightly greater for HVO compared to diesel and both were greater but not significantly different from the RME treatment. Finally, Le Prieur et al. (2000) observed that the inflammatory response in rat lung slices was significantly greater for DE compared to RME exhaust.

Chronic exposure to DE in experimental animals has been shown to cause histopathological changes and affect lung function (Pepelko et al. 1980; Vinegar et al. 1980; Vinegar et al. 1981a; Vinegar et al. 1981b; Moorman et al. 1985; Brightwell et al. 1986; Heinrich et al. 1986; McClellan et al. 1986; Mauderly et al. 1988). Exposure to ambient PM in animals has also

produced cell damage and oedema (Dye et al. 2001; Wise et al. 2006). In epidemiological studies, a reduction in lung function has been correlated with ambient PM exposure in children (Dockery et al. 1996; Raizenne et al. 1996; Kulkarni et al. 2006). Based on the limited information available comparing biodiesel and DE, it seems unlikely that biodiesel exhaust will surpass DE in terms of histopathology and lung function outcomes. Two *in vivo* studies were reviewed; a subchronic study (NBB 2000a/Finch et al. 2002) of only biodiesel showed dose dependent increases for some lung lesions in rats while an acute study found no significant difference in alveolar tissue injury between biodiesel and DE treatments (Tzamkiozis et al. 2010). The results for the *in vitro* studies reviewed were mixed. Swanson et al. (2009) found that no biodiesel or SRM 1975 diesel extract (SOF) was consistently cytotoxic for human airway epithelial cells at the doses tested. Jalava et al. (2010) found no major differences in cytotoxicity and apoptosis in mouse macrophages exposed for 24 hours to particulate emissions from HVO, diesel and RME with or without a catalyst. In contrast, Ackland et al. (2007) observed that diesel PM was more effective at inducing apoptosis in human airway cells than biodiesel PM. Finally, Liu et al. (2008) observed varying results; the Microtox test indicated that diesel particulate extracts had higher unit toxicity than biodiesel particulates while the MTT assay indicated that the particulate extracts for both fuels were similar in terms of cytotoxicity. Importantly, this study also showed that for biodiesel and biodiesel blends the semi-volatile extracts are more toxic than the particulate extracts and there was some evidence that the semi-volatile extracts from mid to high biodiesel blends elicited the greatest response. The results from a human cross-sectional study indicated there was no differences between diesel and RME exhaust in terms of irritation of mucous membranes and the lower airways as well as lung function (Hasford et al. 1997).

While some of the reviewed studies characterized emissions in terms of different physicochemical characteristics (Finch et al. 2002/NBB 2000a; Brito et al. 2010 (see 7.2.3 *Cardiovascular*); Swanson et al. 2009; Tzamkiozis et al. 2010; Jalava et al. 2010) none of the studies attributed differences in the magnitude of inflammation and histopathological outcomes between biodiesel and DE to specific chemical-physical characteristics.

7.2.2 Immunological effects

7.2.2.1 Biodiesel exhaust

No studies were located that examined immunological effects (increased allergic response and airway reactivity; reduced immune function) resulting from exposure to biodiesel exhaust, either in humans, animal or *in vitro* models. Given the lack of data for biodiesel exhaust, it was not possible to determine if biodiesel emissions result in a similar, decreased, or increased impact on immunological outcomes in comparison to DE.

7.2.3 Cardiovascular effects

7.2.3.1 Biodiesel exhaust:

The following text reviews the information on cardiovascular effects resulting from exposure to biodiesel exhaust and focuses on the key outcomes: endothelial dysfunction; prothrombosis; systemic inflammation and atherosclerosis; haematological effects; and cardiac events. It should be noted that some markers are relevant to more than one cardiovascular outcome and therefore their results are reported in more than one of the following sections.

Endothelial Dysfunction

A study by Brito et al. (2010) examined the acute cardiovascular effects induced by inhalation exposure to biodiesel exhaust from a SEE biodiesel fuel, DE (500 ppm sulphur diesel fuel), and filtered air. A stationary diesel electrical generator (BD-2500 CFE; Branco) was used to generate the exhaust. Twenty-four adult Balb/c mice per treatment group were exposed to B100, B50, diesel (97% diesel and 3% SEE), and a control (filtered air). In relation to $PM_{2.5}$ measured in the exposure chamber with a monitor, mice were exposed to 45.08 ± 36.17 μg/m^3 (control), 556.41 ± 134.69 μg/m^3 (diesel), 551.61 ± 176.07 μg/m^3 (B50), and 550.13 ± 153.55 μg/m^3 (B100) for 1 hour to simulate a concentration that would be received in 24 hours. CO concentrations were ND (control), 9.79 ± 7.32 ppm (diesel), 11.9 ± 5.61 ppm (B50), and 3.84 ± 3.17 ppm (B100). The mice were euthanized after exposure.

Mean concentrations (per filter) for $PM_{2.5}$ were 32.83 ± 12.06 μg/m^3 (control), 424.01 ± 244.03 μg/m^3 (diesel), 516.78 ± 222.82 μg/m^3 (B50), and 229.28 ± 157.69 μg/m^3 (B100). Mean concentrations of black carbon were 0.99 ± 0.62 μg/m^3 (control), 43.49 ± 29.66 μg/m^3 (diesel), 68.65 ± 19.71 μg/m^3 (B50), and 26.68 ± 17.79 μg/m^3 (B100).

Analysis of inorganic compounds on the filters indicated the total inorganics (magnesium, aluminum, phosphorous, sulphur, chlorine, potassium, calcium, titanium, vanadium, chromium, manganese, iron, nickel, copper, zinc, selenium, bromine, lead) for each treatment were: 0.65 ng/m^3 (control); 1.05 ng/m^3 (diesel); 1.43 ng/m^3 (B50); and 0.80 ng/m^3 (B100). The analysis also indicated that B50 exhaust had greater concentrations of sulphur, magnesium, potassium, zinc, copper, calcium and iron compared to diesel and B100 exhaust.

Total VOCs (aromatics, alkanes, alkenes, and alkadienes) measured were: 769.77 μg/m^3 control); 10,716.93 μg/m^3 (diesel); 2,253.08 μg/m^3 (B50); and 4,210.44 μg/m^3 (B100). Total PAHs in $PM_{2.5}$ were: 150.1 ng/m^3 (control); 8,019.55 ng/m^3 (diesel); 644.7 ng/m^3 (B50); and 95.8 ng/m^3 (B100).

Endothelin-1, a marker of endothelial dysfunction, was not measured in this study. However, blood pressure (BP), which may be associated with endothelial dysfunction, was. The authors

observed no significant differences in the mean BP at 30 minutes and 60 minutes after pre-exposure time for the control, diesel, B50, and B100 treatment groups.

Prothrombosis

NBB (2000a)/Finch et al. (2002) performed a subchronic inhalation study (see 7.2.1.1 for additional details) in F344 rats to determine the potential toxicity of biodiesel (soybean-derived) exhaust emissions. Thirty rats, 15 males and 15 females per exposure group, were used for the haematology examination (NBB 2000a).

Blood samples were obtained pre-exposure, and after 30 and 90 days of exposure. Markers of prothrombosis analyzed included platelets and leukocytes. Exposure to biodiesel exhaust did not affect either marker. Although platelet and leukocyte counts declined over time for all treatments, there was no significant difference between treatments (control and biodiesel) within each treatment period (0, 30 and 90 days).

Brito et al. (2010) analysed blood samples of mice exposed by inhalation to biodiesel exhaust, DE and filtered air 24 hours after exposure. Markers of prothrombosis included platelet count, fibrinogen concentration, leukocyte count, activated partial thromboplastin time (APTT), prothrombin time (PT), and thrombin time (TT).

Results indicated that there was a significant increase in mean platelet count in the B100 (903.73 ± 153.7 M/mm^3) group when compared to the diesel (605.60 ± 208.2 M/mm^3) and control (608.67 ± 287.9 M/mm^3) groups. There was also an increase in the mean platelet count in the B50 (682.50 ± 181.52 M/mm^3) group when compared to the control and diesel; however, it was not significant.

There was a small insignificant increase in mean leukocyte count (3.82 ± 1.22 M/mm^3) in the B50 group compared to the control group (3.49 ± 0.92 M/mm^3) and an insignificant decrease was noted in the diesel (2.66 ± 0.60 M/mm^3) and B100 (3.12 ± 0.95 M/mm^3) groups compared to the control. The difference between the B50 and B100 groups compared to the diesel treatment was insignificant.

The mean fibrinogen concentration appeared to decrease as the level of biodiesel increased; however, any differences were determined not to be significant. Mean fibrinogen concentrations were 131.54 ± 70.68 mg/dl for the control group, 125.58 ± 51.97 mg/dl for the diesel group, 113.00 ± 19.94 mg/dl for the B50 group, and 29.92 ± 59.99 mg/dl for the B100 group. The authors of the study did not comment on these levels. However, it should be noted that the numbers for all groups are low and those for the B100 group are not compatible with life.

Mean APTT, PT and TT values were not significantly different for the control, diesel, B50, and B100 treatment groups.

Brito et al. (2010) indicated that exposure to B50 and B100 exhaust and activation of bone marrow leukocytes and platelets are indicative of prothrombotic events and may parallel the series of effects associated with exposure to the organic fraction of diesel exhaust particles. This may be the case; however, the results for leukocytes in the present study do not necessarily support this. While the results for platelets indicate a significant increase for B100 compared to the diesel and control groups, there was no significant increase in leukocyte numbers as a result of exposure to biodiesel exhaust. It should be noted that haematology data from NBB (2000b) did not indicate any increase in platelet or leukocyte counts.

Systemic Inflammation and Atherosclerosis

NBB (2000a,b)/Finch et al. (2002) analyzed for markers of inflammation such as platelets, leukocytes, and lymphocytes. Blood samples were taken before exposure, after 30 days, and at the end of the exposure (90 days). Exposure to biodiesel exhaust did not affect platelet or leukocyte counts (see prothrombosis). In a similar fashion, lymphocyte numbers did not differ significantly between the control, low, intermediate and high exposure groups within each treatment period (NBB 2000b).

Brito et al. (2010) investigated the acute cardiovascular effects in mice and observed that B50 and B100 exhaust elicited a systemic inflammatory response, including leukocyte (not significant) and platelet release from the bone marrow, when compared to DE and the control. The authors indicated that such a response increases lung inflammation and changes the phenotype of atherosclerotic plaques making them more prone to rupture (Van Eeden and Hogg 2002 in Brito et al. 2010). Systemic inflammation is thought to result from a cascade of events that include the activation of cell signalling pathways and the release of pro-inflammatory mediators. According to Brito et al. (2010), this cascade was demonstrated by the activation of the bone marrow, AMs in the BALF, leukocytes, lymphocytes, and platelets in the blood (Brito et al. 2010).

Bone marrow analysis 24 hours after exposure revealed that there was a significant increase in mean number of total cells in the B100 (92.51 cells/ml x 10^4), B50 (97.9 cells/ml x 10^4), and diesel (121.9 cells/ml x 10^4) groups when compared to the control (55.54 cells/ml x 10^4) group. The mean percentage of metamyelocytes was significantly increased in the B50 (7.64%) and B100 (7.8%) groups compared to the diesel (4.55%) group. The value for the control was 5.14%. The mean percentage of monocytes was significantly decreased in the B50 (2.18%) group in comparison to the diesel (3.91%) and B100 (3.9%) groups. The value for the control was 3.42%. The mean percentage of plasmocytes was significantly decreased in the B50 (0.09%) group compared to the B100 (1.7%) and control (1.37%) groups. The value for the diesel group was

1%. The authors observed no significant differences for other bone marrow cells including myelocytes, bastonet cells, segmented cells, eosinophils, lymphocytes, macrophages and red blood cells (RBCs) (Brito et al. 2010).

There was a significant increase in mean alveolar macrophage numbers in the BALF for diesel, B50, and B100, exposure groups when compared to the control group. There was also a significant increase in neutrophils in the BALF for diesel and B50 compared to the control.

Results for leukocytes, and platelets were presented in the previous Section for prothrombosis. Blood analysis revealed that the lymphocyte count was significantly different for B50 (3.02 ± 1.10 M/mm^3) compared to the diesel treatment (1.97 ± 0.58 M/mm^3). Lymphocyte counts for the control and B100 were (2.69 ± 0.90 M/mm^3) and 2.45 ± 0.72 M/mm^3), respectively. There were no significant differences seen in the levels of neutrophils (segmented cells), eosinophils, or basophils in any of the experimental groups.

Effects on Haematology

NBB (2000a)/Finch et al. (2002) performed a subchronic inhalation study in F344 rats to determine the potential toxicity of biodiesel exhaust. Blood was collected for haematology parameters and analyzed for RBC count, Hb, haematocrit percentage (HCTPCT), platelet count, white blood cell count (segmented cells, band cells, lymphocytes, monocytes, eosinophils, and basophils), and differential white blood cell count. Blood samples were taken before exposure, after 30 days, and at the end of the exposure (90 days).

Total white blood cell counts (mainly lymphocytes and monocyte) decreased significantly in rats from all exposure groups over the 13-week-exposure period. The authors indicated that this observation (reduced lymphocyte numbers) was consistent with previous reports in aging Sprague Dawley rats (Wolford et al. 1987 *in* NBB 2000a).

The results indicated that there was no significant difference observed in platelet (see *Prothrombosis*) and lymphocyte (see *Systemic inflammation and atherosclerosis*) counts as well as for all other parameters for each treatment (control, low, intermediate and high) at pre-exposure, and after 30 and 90 days of exposure. The study authors concluded that there were no biologically significant differences observed in the haematology data for all treatment groups (NBB 2000a; Finch et al. 2002). Health Canada reviewed the data (NBB 2000b) and concurred with this conclusion.

Brito et al. (2010) investigated the cardiovascular effects induced by inhalation of DE, biodiesel exhaust, and filtered air. Haematological parameters analyzed 24 hours after exposure included: mean corpuscular volume (MCV), mean corpuscular haemoglobin concentration (MCHC), reticulocytes, platelets, leukocytes, lymphocytes, erythrocytes, Hb, haematocrit, mean

corpuscular haemoglobin (MCH), red cell distribution with variation coefficient, red cell distribution with size distribution, neutrophils (segmented cells), eosinophils, basophils, monocytes, and fibrinogen. There was a significant increase in the MCV in the B100 (45.54 ± 0.66 fl) group when compared to the diesel (44.07 ± 0.88 fl), B50 (42.90 ± 0.73 fl), and control (43.38 ± 0.91 fl) groups.

The MCHCs were: B50 (35.40 ± 1.65 g/dl); control (34.37 ± 0.60 g/dl); diesel (33.74 ± 0.75 g/dl); and B100 (33.25 ± 0.50 g/dl). The MCHC was significantly decreased in the B100 compared to the B50 and control groups but not significantly different from that of diesel. The value for the B50 group was significantly higher than the diesel and the B100 groups. Reticulocytes were significantly increased for B50 (3.87 ± 0.44%) compared to B100 (3.10 ± 0.80%), diesel (2.83 ± 0.45%), and the control (2.87 ± 0.39%) groups.

There were no differences observed between treatment groups for erythrocytes, Hb, haematocrit, MCH, red cell distribution with variation coefficient, red cell distribution with size distribution, neutrophils (segmented cells), eosinophils, basophils, and monocytes.

Results from Brito et al. (2010) for mean platelet and leukocyte counts as well as fibrinogen were presented in Subsection 7.2.3.1 *Prothrombosis*. Results for mean lymphocyte count are presented in Subsection 7.2.3.1 *Systemic inflammation and atherosclerosis*.

Cardiac Events

Brito et al. (2010) investigated the cardiovascular effects induced by inhalation of DE, biodiesel exhaust, and filtered air. Markers of cardiac events that were analyzed included heart rate (HR), heart rate variability (HRV) for the time domain (characterized by the standard deviation of normal beats (SDNN) and the root mean square of SDNN (RMSSD)), and the frequency domain (characterized by low frequency (LF), high frequency (HF), and the low frequency/high frequency (LF/HF) ratio). BP was also measured and no significant differences were observed for the different treatments (see *Endothelial dysfunction*). All parameters were measured at 30 and 60 minutes relative to pre-exposure time.

There were significant differences observed for the group HR and HRV (RMSSD and LF) parameters; however, no effect of time (30 and 60 minutes), and no interaction between time and group were observed. Mean HR was significantly increased after 30 and 60 minutes for B100 (43.84 ± 63.96 beats/min; -1.49 ± 50.58 beats/min) group in comparison to the control (-6.08 ± 52.93 beats/min, -48.42 ± 67.79 beats/min) groups. There were no significant differences after 30 and 60 minutes for the mean HR between the control (above), diesel, and B50 groups. In terms of HRV, the mean RMSSD was significantly increased after 30 and 60 minutes in the diesel (0.34 ± 19.98 ms; 11.44 ± 26.17 ms) group compared to the control (-17.07 ± 16.55 ms, -10.11 ± 26.27 ms) group. In addition, the mean LF was significantly increased after 30 and 60 minutes in

the diesel (22.88 ± 59.88 ms^2; 23.87 ± 84.41 ms^2) and B100 (1.28 ± 41.20 ms^2; 35.15 ± 130.78 ms^2) groups compared to the control (-61.75 ± 71.17 ms^2; -39.52 ± 98.33 ms^2). There were no significant differences observed for SDNN, HF, and LF/HF ratio in the control, diesel, B50, and B100 group.

Summary/conclusion

Exposure to DE, DEP and/or ambient PM by different routes have been shown to affect endothelial dysfunction (Bouthillier et al. 1998; Mills et al. 2005; Peters 2005; Tornqvist et al. 2007; Peretz et al. 2008), prothrombosis (Nemmar et al. 2004a; Nemmar et al. 2004b), systemic inflammation/atherosclerosis (Peters et al. 1997; Pekkanen et al. 2000; Peters et al. 2001a; Peters et al. 2001b; Schwartz 2001; Suwa et al. 2002; Sorensen et al. 2003; Sun et al. 2005; Van Eeden et al. 2005), haematological (Wiester et al. 1980; Brightwell et al. 1986; Peters et al. 1997; Pekkanen et al. 2000; Peters et al. 2001a; Peters et al. 2001b; Schwartz 2001) and cardiac event (Liao et al. 1999; Pope et al. 1999a; Pope et al. 1999b; Gold et al. 2000; Nadziejko et al. 2001; Holguin et al. 2003; Liao et al. 2004; Pope et al. 2004a; Pope et al. 2004b; Campen et al. 2005; Luttmann-Gibson et al. 2006; Wheeler et al. 2006; Adar et al. 2007; Mills et al. 2007) outcomes in experimental animals or humans.

With respect to biodiesel exhaust, Brito et al. (2010) observed no significant differences between biodiesel and diesel treatments for endothelial dysfunction and cardiac events. In the case of endothelial dysfunction, only one relevant endpoint (BP) was measured. Endpoints for cardiac events included: HR, HRV and BP. Given that this was the only study available which investigated the effect of biodiesel exhaust on these cardiovascular outcomes, it was not possible to draw any conclusions on how biodiesel exhaust compares to DE.

In terms of systemic inflammation/atherosclerosis and haematological outcomes, Brito et al. (2010) observed that acute exposure of mice to B100, B50 and DE resulted in a greater number of total bone marrow cells for the B50 and B100 treatments compared to the control but not compared to diesel. Significant differences between biodiesel and diesel treatments were noted for some parameters. The mean percentage of metamyelocytes (see *Systemic inflammation and atherosclerosis*) were greater for B50 and B100 compared to diesel, the mean percentage of monocytes (see *Systemic inflammation and atherosclerosis*) were decreased for B50 compared to B100 and diesel and the mean percentage of reticulocytes (see *Haematology*) were greater for the B50 compared to all other groups. A significant increase in platelet numbers (see *Prothrombosis*) for B100 compared to diesel and a greater lymphocyte count (see *Systemic inflammation*) for B50 compared to diesel also point to the inflammatory effects of biodiesel exhaust. Haematological changes included a greater MCV for B100 and a decreased MCHC for B100 compared to all other treatments. The changes in these haematological parameters may reflect the cited inflammatory effects.

Brito et al. (2010) calculated that B50 exhaust had increased emissions of $PM_{2.5}$ (21.88%), black carbon (57.85%), inorganics (37.64%), and CO (21.55%) but had lower emissions of PAHs (91.77%) and VOCs (78.98%) when compared to DE. B100 exhaust had decreased emissions of $PM_{2.5}$ (45.93%), black carbon (38.65%), inorganics (32.81%), CO (60.78%), PAHs (97.81%) and VOCs (60.71%) when compared to diesel.

It should be noted that the calculations that generated the foregoing numbers were based on averages ($PM_{2.5}$, BC, inorganics) and totals (PAHs, VOCs) and did not take into consideration standard deviations, i.e., statistical significance. As an example, the B50 $PM_{2.5}$ increase (21.88%) appears suspect given that the standard deviations for the B50 and diesel treatments indicated that they were not statistically different. Standard deviations were not provided for the inorganics, PAHs and VOCs. Therefore, it is not possible to judge the significance of the reported changes.

Although the exact reason for the increase in some inflammatory parameters, i.e., bone marrow cells, platelets and lymphocytes, as a result of the biodiesel treatments (B100 and B50) is not obvious, it may be related to the presence and/or combination of pollutants which cause inflammation including $PM_{2.5}$, black carbon, PAHs, CO, inorganics, and VOCs. In the case of B50, an increase in some inflammatory markers may be due in part to higher levels of $PM_{2.5}$, black carbon, some inorganics, CO as well as some pollutants and/or chemical-physical characteristic not measured in this study. Given that $PM_{2.5}$, black carbon, inorganics, CO, PAHs and VOCs were all lower for B100 compared to DE, other pollutants and/or chemical-physical factors not measured in the study including NO_x, specific VOCs, the SOF of PM, and specific size ranges of particles may be contributing to increases in some inflammatory markers.

The results from Brito et al. (2010) indicate that exposure to biodiesel exhaust (B100 and B50) causes increases in some inflammatory markers when compared to DE. However, given that these findings stem from only one study it is not possible to draw any final conclusions with respect to cardiovascular effects. It is also important to point out this study was based on the use of a generator and not a vehicle engine with emission controls. Brito et al. (2010) did not identify specific pollutants responsible for the increases of the inflammatory markers in question. The only other study reviewed (NBB 2000a/Finch et al. 2002) did not observe any significant effects for those cardiovascular parameters measured and it did not include a diesel treatment. Thus any comparison of cardiovascular outcomes with biodiesel was not possible.

7.2.4 Carcinogenicity

7.2.4.1 Biodiesel exhaust

The following text reviews the published literature on effects related to initiation of carcinogenesis resulting from exposure to biodiesel exhaust and focuses on the key outcomes:

clastogenic effects *in vivo* and *in vitro*; biochemical events associated with genetic instability; cytotoxic effects in cultured mammalian cells; and mutagenic effects in *Salmonella*.

Clastogenic effects in vivo and in vitro

Only one *in vivo* and one *in vitro* study in the peer-reviewed literature assessed the clastogenic effects of biodiesel exhaust (NBB 2000a/Finch et al. 2002; Eckl et al. 1997).

NBB (2000a)/Finch et al. (2002) performed a subchronic inhalation study (see Section 7.2.1.1 for additional details) with F344 rats to determine the potential toxicity of biodiesel exhaust emissions. Five male and five female rats were used per exposure group for the clastogenicity portion of the study, which included the micronucleus and sister chromatid exchange assays.

Micronuclei were examined in bone marrow collected from femurs and sister chromatid exchanges were evaluated at the end of 13 weeks (without a post-exposure recovery time) in lymphocytes in peripheral blood. Both assays were performed on rats from 7,12-dimethylbenzanthracene (DMBA) controls and the four exposure groups at terminal sacrifice. There were no statistically significant biodiesel exhaust exposure-related effects on micronuclei in bone marrow or sister chromatid exchange in peripheral blood lymphocytes (NBB 2000a; Finch et al. 2002).

The NBB (2000a)/Finch et al. (2002) study was the only available study that assessed the clastogenic effects associated with inhalation exposure to biodiesel exhaust. The study did not include DE exposure thus limiting a comparison of clastogenic outcomes between biodiesel and DE.

Eckl et al. (1997) utilized the Ames assay (TA98 and TA100) and metabolically-competent primary rat hepatocytes to examine the clastogenic effects of PM extracts and condensates of DF and RME exhaust generated from a tractor engine at five loads (e.g., 0–100% load at varying revolutions per minute). The results of the Ames assay indicated a higher mutagenic potential for the DE compared to RME. In the case of the rat hepatocyte model, no statistically significant differences were observed as measured by the induction of micronuclei and sister chromatid exchanges. The authors attributed this difference to the greater metabolic capacity of hepatocytes compared to the Salmonella strains and the greater sensitivity of the bacteria to nitro-polycyclic aromatic hydrocarbons (nitro-PAHs).

Biochemical Events Associated with Genetic Instability

The association between inflammation and the development of cancer is well established. Inflammation has been linked to the proliferation and survival of malignant cells, production of ROS, DNA damage, and an imbalance of antioxidant capacity. Furthermore, inflammation has been linked to genetic instability, which can result in the generation of cancer cells (Colotta et al.

2009). Four studies investigated the inflammatory response, DNA damage or antioxidant response to exhaust or PM extracts of exhaust from biodiesel and diesel *in vitro* (Jalava et al. 2010; Le Prieur et al. 2000; Swanson et al. 2009; Kooter et al. 2011).

In a study conducted in 2010, Jalava and colleagues evaluated ROS production, cytokine production, and genotoxicity in RAW267.4 macrophages treated with 15, 50, 150 and 300 µg/mL PM extract from exhaust of RME, DF (EN590) and HVO generated from a non-road diesel engine with indirect fuel injection (run at several modes [2.22–22.2 kW] and with or without a catalyst) for 24 hours. No statistically significant differences in ROS production were detected between the three different treatment groups involving different fuels or concentrations of PM extract. However, a statistically significant increase in ROS production was seen with increasing dose within each individual treatment (Jalava et al. 2010). The authors also noted that PM extract from RME exhaust was least potent at inducing inflammatory markers, such as TNF-α and MIP-2. However, the differences between treatments were not significant. In addition, DNA damage, as assessed by the Comet assay, was similar among PM extracts from the exhaust of all three fuels. Based on these analyses, the authors concluded that the toxicological potency of the PM extracts generated from the exhausts of RME, DF and HVO as assessed by DNA damage, inflammatory signalling and ROS production, was similar. No correlations were made between levels of individual pollutants in PM extract of the different fuel exhausts, ROS and response of inflammatory markers.

In 2000, Le Prieur et al. evaluated antioxidant capacity and the inflammatory response in rat lung slices exposed *in vitro* to whole (100%) or filtered (10, 15, 25, 60 and 85%) exhaust from DF, 100% RME or 30% RME (30% RME and 70% DF) (generated from a 5-horsepower one-cylinder Robin engine running at 3000 rpm with a load corresponding to a 1 bar water power outlet pressure) for three hours. Bosch smoke number, hydrocarbons, and CO were reduced in RME exhaust, whereas NO_x levels increased slightly with 100% RME as compared to DF exhaust. Total glutathione levels, a marker for oxidative stress or antioxidant capacity, decreased following exposure to all three exhausts, although there was no statistically significant difference between the three different fuel types. Treatment with DF exhaust produced a statistically significant increase in TNF-α protein levels with increased concentrations, whereas supplementation with RME or filtration of each exhaust prevented an increase in TNF-α levels (Le Prieur et al. 2000).

In 2009, Swanson and colleagues used an *in vitro* human bronchial epithelial cell model (BEAS-2B) to investigate the inflammatory effects of PM organic extracts from SRM 1975 (an extract of diesel PM), SEE, SME, Phillips No.2 diesel and a solvent blank (see Subsection 7.2.1.1 *Inflammation* for additional details). An analysis of the IL-8 median response revealed a statistically significant increase in IL-8 protein levels after treatment with PM extract from

exhaust of biodiesel compared to diesel in one out of three separate experiments (Swanson et al. 2009).

Kooter et al (2011) used mouse macrophage cell line RAW264.7 in the dithiothreitol (DTT) assay to test the redox activity (ROS formation) of particle extracts of diesel EN590, biodiesel EN14214, blends (B5, B10 and B20) and pure plant oil (PPO). The use of a DPF with diesel EN590 was also tested. A six cylinder Euro III truck engine with a cylinder displacement of 12 litres and 355 kW was used. The engine was tested with the European Transient Cycle on a transient engine dynamometer. The use of B100, PPO and DPF reduced PM mass and numbers by over 80%. A significant reduction in elemental carbon (90%) and oxy-PAHs (70%) were also observed. However, there was no significant reduction in nitro-PAHs. The use of B100 and PPO resulted in a NO_x increase of approximately 30%. The oxidative potential of B100, PPO, and diesel + DPF was reduced by 95% compared to diesel. The authors indicated that the reduction in redox activity was correlated with the decrease in EC and total PAH levels.

According to Jalava et al. (2010) and Le Prieur et al. (2000), the oxidative stress response and DNA damage are similarly affected after treatment with exhaust or PM extract from exhaust of biodiesel and diesel. In contrast, Kooter at al. (2011) observed a significant decrease in ROS formation for biodiesel compared to diesel. Treatment with exhaust or PM extract from biodiesel exhaust resulted in an inflammatory response that was equal or lower when compared to DE. Jalava et al. (2010) observed a slight but insignificant decrease, while Le Prieur et al (2000) detected a significant decrease with supplementation with RME. Swanson et al. (2009) observed a significant increase in the inflammatory response for the SOF of biodiesel exhaust compared to the same fraction for DE but only in one of three experiments.

It should be noted that the extraction method used in the different studies varied. Exhaust samples were extracted with methanol in the Jalava study whereas the Swanson study used dichloromethane (DCM). DCM has been shown to be a more effective solvent for the extraction of mutagenic organics from DE (Petersen et al. 1982; Montreuil et al. 1992). The equivalent studies, however, have not been performed for biodiesel exhaust emissions. In addition to the extraction methodologies, differences in engine types and running conditions used to generate exhaust samples may have also contributed to the variation in results between studies.

Cytotoxic Effects in Cultured Mammalian Cells

Several studies in the peer-reviewed literature addressed the cytotoxic and apoptotic potential of biodiesel *in vitro* (Bünger et al. 2000a; Bünger et al. 1998a; Bünger et al. 1998b; Jalava et al. 2010; Le Prieur et al. 2000; Liu et al. 2009; Kooter at al. 2011). Ackland et al. (2007), Liu et al. (2008) and Swanson et al. (2009) also investigated cytotoxicity or apoptosis in cells exposed to diesel and biodiesel extracts (see Subsection 7.2.1.1 *Histopathology and Lung Function*).

Some researchers have reported no differences in cytotoxicity or induction of apoptosis after treatment of cells or tissues with PM extract from exhaust of diesel or biodiesel (i.e., RME, SME and SEE). Bünger et al. (1998a) found no statistically significant differences in cytotoxicity between PM extracts of exhaust from DF, RME and SME (generated from a VW Vento 1.9L TDI equipped with a turbo-charged direct injecting diesel engine with a catalytic converter run using the European MVEG-A test and US Federal Test Procedure-75 test cycles) in L929 cells. Additionally, Bünger et al. (1998b) did not observe any statistically significant differences in cytotoxicity in L929 cells between PM extracts from exhaust of RME or DF (generated from a Farymann one-cylinder direct injection diesel engine with a catalytic converter running a 5-mode cycle). Lastly, Jalava et al. (2010) examined cytotoxicity of PM extract of exhaust of DF, HVO and RME (generated from a non-road indirect injection diesel engine) and found no differences in cytotoxicity between the three fuel types (Jalava et al. 2010). Other studies, however, have found that both diesel and biodiesel can be potent inducers of apoptosis and/or cytotoxicity (Bünger et al. 2000a; Le Prieur et al. 2000; Liu et al. 2009; Kooter et al. 2011).

Le Prieur et al. (2000) found a statistically significant decrease in viability (ATP assay) in rat lung slices following treatment with increasing concentrations of filtered (10, 15, 25, 60 and 85%) or whole (100%) exhaust of 30% RME (30% RME and 70% DF) and 100% RME (generated from a 5-horsepower one-cylinder Robin engine), whereas none of the DF exhaust concentrations significantly reduced viability. Although a 10% exhaust concentration of 30% RME significantly induced nucleosome release (marker of apoptosis), DF exhaust induced nucleosome release at all concentrations, and 100% RME exhaust produced minimal nucleosome release. The authors noted that although the greatest apoptotic response was produced by DF exhaust, pure RME and 30% RME exhaust had a greater cytotoxic response (cell viability) than DF exhaust. Filtered exhaust of both fuels produced an inconsistent response in apoptosis and nucleosome release (Le Prieur et al. 2000).

Liu and colleagues (2009) examined the cytotoxicity (as measured by the MTT assay) of PM extract from DF and B10 exhaust (a mix of 10% palm fatty acid methyl ester and 90% premium diesel), generated with a four cylinder diesel generator at 10, 33 and 55% loads, and found that treatment with PM extract from exhaust of B10 caused greater cytotoxicity in BEAS-2B cells as compared to PM extract from exhaust of DF (although no statistical significance was provided). Results from the Microtox test also indicated greater toxicity with exposure to PM extract from exhaust of B10 as compared to DF; however, no statistical comparisons between the two fuel types were provided. The greatest toxicity of PM extract from exhaust of DF was associated with the highest carbonyl emissions (load 10% as compared to 33 and 55% loads); however, this was not the case for PM extract from exhaust of B10, whose toxicity did not correlate with carbonyl emission levels. The authors suggested that carbonyls are not the primary pollutants associated with the cytotoxicity of PM extract from exhaust of B10. Additionally, CO_2, CO and NO_x emissions increased with engine mode when DF was used, whereas a similar trend was observed

with B10 except CO declined as engine loading increased. A correlation between emissions of these individual pollutants and toxicity was not evaluated (Liu et al. 2009).

The Bünger group used a commercially available tractor with a four-stoke direct injection diesel engine (Fendt 306 LSA, 52 kW) fuelled with RME and diesel fuel and driven in the European standard (13-mode) test cycle (ECE R49). Extracts of the particulate emissions were tested for cytotoxicity using the neutral red assay in L929 mouse fibroblast cells. It was found that PM extract from RME exhaust was more potent at inducing cytotoxicity than PM extract from DF exhaust (sulphur content 370 ppm) at idling (referring to a stationary state). This effect was attributed to higher amounts of carbonyl compounds such as the toxic aldehyde, acrolein, and unburned fuel (fatty acid methyl esters) in the extracts during idling mode. Furthermore, emissions from the engine burning RME generated less smaller-sized particles than DF at modes corresponding to idling (modes 1, 7, 13). The differences in cytotoxicity at rated power (mode 8: 100% speed and load) were noted to be small and the particle size distribution and maximum numbers of emitted particles were very similar for the two fuels (Bünger et al. 2000a).

Kooter et al (2011) used the Lactate Dehydrogenase (LDH) assay to test the cytotoxicity of particle extracts of diesel EN590, biodiesel EN14214, blends (B5, B10 and B20) and PPO. The cytotoxicity of B100 (40%) was significantly increased compared to diesel (20%).

Exhaust or PM extract from exhaust of biodiesel and biodiesel blends were shown to be equally (Bünger et al. 1998a; Bünger et al. 1998b; Jalava et al. 2010) or more potent (Bünger et al. 2000a; Le Prieur et al. 2000; Liu et al. 2009; Kooter et al. 2011) than diesel with respect to inducing apoptosis or cytotoxicity. It should be noted that in Le Prieur et al. (2000), the results indicate lower apoptotic responses but greater cytotoxicity after treatment of lung slices with RME exhaust. Investigations on the apoptotic and cytotoxic responses of exhaust or PM extract from exhaust of biodiesel as compared to diesel have demonstrated a dependence on fuel type (Le Prieur et al. 2000; Liu et al. 2009; Kooter et al. 2011) and engine load (Bünger et al. 2000a, Liu et al. 2009). However, there are not enough data to reach a conclusion on the cytotoxicity of the exhaust or PM extracts from exhaust of biodiesel. Finally, it should be noted that each of the foregoing studies used different engines and testing conditions.

Mutagenic Effects in Salmonella

Between 1997 and 2011, several studies evaluated the mutagenicity of PM extracts from biodiesel exhaust utilizing modifications of the *Salmonella typhimurium*/microsome mutagenicity bioassay or Ames test (Bagley et al. 1998; Bünger et al. 1998a; Bünger et al. 1998b; Bünger et al. 2000a; Bünger et al. 2000b; Bünger et al. 2006; Carraro et al. 1997; Eckl et al. 1997; Kado et al. 2003; Kooter et al. 2011; Krahl et al. 2001; Krahl et al. 2002a [Schroder et al. 1999; Krahl et al. 2002b; Munack et al. 2001]; Krahl et al. 2003a; Krahl et al. 2003b [Krahl et al. 2003c, Krahl et al. 2003d German]; Krahl et al. 2005; Krahl et al. 2008; Krahl et al. 2009a;

Krahl et al. 2009b;Turrio-Baldassarri et al. 2004).[68] The studies conducted the Ames test (or a modification thereof) using TA100, TA98, TA98NR, TA98DNP and/or YG1024 strains of *Salmonella typhimurium*. The majority of the studies were conducted by the Bünger group. Those conducted by other researchers are discussed first.

The majority of the studies performed by investigators outside of the Bünger group found that PM extracts from biodiesel exhaust were either less mutagenic or as mutagenic as PM extracts from DE. The investigation by Bagley et al. (1998) was primarily an evaluation of the use of biodiesel in conjunction with a diesel oxidation catalyst (DOC) as an effective strategy for Total Particulate Matter (TPM) control in a confined space, such as a mining operation. A commercially available indirect injection diesel engine typically found in mining environments was operated on SME and diesel fuel. PM samples were collected with and without the DOC in use and the mutagenicity of PM extracts was evaluated using the classic bioassay for detecting carcinogens and mutagens, the Ames Salmonella/Microsome test. The DOC is used to reduce particle-phase VOCs and hydrocarbon emissions. The use of the DOC reduced emissions for both DF and SME and reduced the mutagenicity (revertants/kW-hr) of PM extracts from exhaust of DF and SME. PM extract from exhaust of SME generated about 50% fewer mutations than DF both with and without a DOC. Condensate extract from exhaust of SME was not associated with any mutagenicity when a DOC was used. The authors noted that the presence of nitro-PAHs in the exhaust of DF contributed to its greater effect on mutagenicity compared to biodiesel. Emissions of these compounds were decreased in biodiesel, regardless of the use of a DOC (Bagley et al. 1998).

In a similar investigation, Carraro and colleagues (1997) demonstrated that PM extracts from biodiesel exhaust (type not specified) induced less mutations (revertants/kW-hr) in the Ames test than PM extract from DF exhaust. The exhaust samples were collected on the chassis dynamometer from two light-duty diesel engines (a 1930 cc direct injection engine and a 1930 indirect injection engine) running the standard driving test cycle: Economic Commission for Europe-Exhaustion Driving Cycle (ECE-EUDC). The type of engine and therefore the associated exhaust influenced the mutagenicity; PM extract from exhaust of the 1930 cc direct injection engine generated greater mutagenicity than the 1930 cc indirect injection engine using both fuels. Mutagenicity of the PM extracts from the exhaust of both fuels (DF and biodiesel) was correlated ($r>0.9$) with PAH and nitro-PAH content. Additionally, levels of both PAHs and nitro-PAHs were lower in biodiesel. No further information regarding the characteristic emissions from diesel versus biodiesel was provided (Carraro et al. 1997).

In a study by Kado and Kuzmicky (2003), the mutagenicity PM extracts from the exhaust of various types of biodiesels compared to DF (Philips No. 2 Diesel, sulphur content 300 ppm) was

[68] Articles that consist of reports completed for organisations, re-edited or unpublished data, and translations of reports originally written in German are mentioned between brackets.

compared under hot and cold engine start (using a 1991 production model Detroit Diesel 6-cylinder engine, 4-stroke, electronically-controlled, direct-injected, turbocharged engine with intercooled calibration) conditions using a microsuspension procedure, which is a modification of the Ames test. The mutagenic potency value and particle emission data collected by the authors were used to calculate an emission rate for mutagens in the engine exhaust generated from each fuel type. PM extract from DF exhaust had the highest mutagen emission rates (revertants/BHP-HR $x10^5$) as compared to canola methyl ester, pork lard methyl ester, yellow grease methyl ester and beef tallow methyl ester, but the statistical significance of the results was unclear. Additionally, the greatest particulate emissions under hot and cold start were generated by DF (Kado et al. 2003).

Turrio-Baldassarri et al. (2004) examined the mutagenicity (revertants/kW-hr) of extracts and condensates of the PM from exhaust of DF (sulphur content <300 ppm) and B20 (a mix of 80% DF and 20% RME) (generated with a turbocharged EURO 2 IVECO 8360.46R heavy duty 6-cylinder diesel engine running the steady-state European 13 mode cycle, ECE R49) and did not report significant differences between the mutagenicity of the two types of fuel exhaust extracts and condensates. It was noted that mutagenicity was generally lower after treatment with extracts of PM and condensates from exhaust of both DF and B20 in the nitropyrene resistant strain TA98/1,8 DNP_6, suggesting a role for genotoxic nitroaromatics (Turrio-Baldassarri et al. 2004).

Eckl et al. (1997) investigated mutagenicity of extract and condensate from exhaust of DF and RME generated from a Steyr WD 411/87 tractor engine at five loads (100% /2150 rpm, 50%/2150 rpm, 100%/1600 rpm, 50%/1600 rpm, and 0%/900 rpm). The testing conditions were previously described in the sub-section "Clastogenic effects *in vivo* and *in vitr*o". They found a statistically significant increase in mutagenicity (revertants per plate) in PM extract and condensate from DF exhaust compared to extract and condensate from RME exhaust, at an exhaust volume of one litre (Eckl et al. 1997).

Kooter et al (2011) used the Ames test with strains TA98 and YG1024 to assess the mutagenicity of particle extracts of diesel EN590, biodiesel EN14214, blends (B5, B10 and B20) and PPO. The results indicate increased mutagenicity (YG1024) for biodiesel (B100) and PPO compared to diesel in the absence of S9 fraction. YG1024 is a derivative of TA98 and highly sensitive to aromatic amines and nitroarenes. The authors suggested that the increased mutagenicity may have been related to the formation of nitro-PAHs.

A majority of the published studies that characterized the mutagenicity of biodiesel were performed by the same research group from Germany headed by Dr. Jürgen Bünger. Of these studies, thirteen were articles presenting original data, while the remaining papers were reports for agencies (US National Renewable Energy Laboratory), republished or unpublished data or German translations of the original documents. These remaining references are noted in the

report in square parentheses where appropriate. The investigators considered the effects of various engines (diesel, tractor, modes [loads] and fuel formulations [diesel, V-Power, 10% RME]) in their studies. Exact concentrations of PM extract or condensate from exhaust of the fuels were not provided. Instead the investigators used relative dilutions of exhaust as treatments. In the following five sections, the mutagenicity of various fuel formulations from the studies from the Bünger group are compared and summarized.

Comparison of Diesel Fuel and Rapeseed Oil Methyl Ester

Four studies comparing PM extracts and/or condensates of 100% RME and 100% DF exhaust found that PM extract and/or condensate of DF was more mutagenic than RME, which was attributed to lower polycyclic aromatic compounds in RME exhaust. The group used various engines (turbocharged direct-injection diesel engine, four-stroke direct-injection diesel engine and a one-cylinder direct-injection diesel engine), as well as various test modes in the studies. In general, DF produced higher PAH emissions and smaller diameter PM as compared to RME (Bünger et al. 1998a; Bünger et al. 1998b; Bünger et al. 2000a).

A Farymann-type 18D one-cylinder air-cooled 4.2 kW four-stroke diesel engine with direct injection running on a German agricultural five mode cycle developed by Welschof (1981), was used to investigate the mutagenic potential of RME and 370 ppm sulphur DF. PM extract from exhaust of DF generated at an engine load corresponding to approximately 60% of the maximum speed of the engine, was found to have the greatest relative mutagenic potency as compared to PM extract of RME exhaust. However, the differences in mutagenic potency between the PM extracts from exhaust of DF and RME were much smaller in magnitude under other modes (e.g., rated or partial load, idle motion) of the five-mode test cycle (Krahl et al. 2002a [Schroder et al. 1999; Krahl et al. 2002b; Munack et al. 2001]).

Comparison of Diesel Fuel with Rapeseed Oil Methyl Ester and Soybean Oil Methyl Ester

The mutagenicity of PM extracts from exhaust of 100% RME, 100% SME and 100% DF (sulphur content 0.022%) (generated with a Farymann type 18 D one-cylinder air-cooled 4.2 kW four-stroke diesel engine with direct injection using a German agricultural five mode engine test cycle developed by Welschof (1981)) was evaluated with and without the use of a DOC (Krahl et al. 2001). PM extract from exhaust of DF was shown to be dramatically more mutagenic (revertants per plate) than RME and SME; however, the statistical significance was unclear. With the use of a DOC, decreased mutations were observed for each fuel type. Additionally, without the use of a DOC, increasing RME content (several blends of RME/DF were used here as opposed to the mutagenicity portion of the study in which 100% pure DF, RME or SME were used) decreased hydrocarbon emissions. Furthermore, increased CO emissions were observed when the DOC was not used. The use of a DOC decreased hydrocarbon emissions for all fuel types, but had almost no effect on NO_x emissions (Krahl et al. 2001). Based on this experiment,

it was determined that PM extract from exhaust of DF was more mutagenic than those of RME and SME. Also, application of a DOC reduced the mutagenicity for all fuel types.

Comparison of Diesel Fuel with Rapeseed Oil Methyl Ester, Soybean Oil Methyl Ester, Low-Sulphur Diesel Fuel (LS-DF) and Other Formulations of Diesel

This section summarizes three studies which investigated the differences in mutagenicity between PM extracts from exhaust of biodiesel and different formulations of diesel fuels including LS-DF.

Mutagenicity was compared between PM extracts from RME, SME, DF (sulphur content 370 ppm) exhaust and LS-DF (generated with a Farymann type 18 D one-cylinder air-cooled 4.2 kW four-stroke diesel engine with direct injection using a German agricultural five mode engine test cycle developed by Welschof (1981) (Bünger et al. 2000b). PM extracts from exhaust of DF produced more mutations (revertants x 1,000 per h) compared to the other three fuels, especially at modes representing engines at full speed, which the authors suggested produced more mutagenic compounds. The study found that there was no statistically significant difference between the mutagenicity of the PM extract from exhaust of the other three fuels (RME, SME, and LS-DF). Notably, RME, SME and LS-DF generated less black carbon and total polynuclear aromatic compounds. Lower mutagenicity of the PM extract from exhaust of LS-DF was attributed to the lower content of sulphur and aromatic compounds.

In 2003, Krahl and colleagues evaluated the differences in mutagenic potential of PM extracts from exhaust of DF (sulphur content 0.032 mg/kg), RME and Swedish low-sulphur fuel (MK 1) generated with a four-cylinder Daimler-Chrysler engine OM 904 LA using the ECE-R 49 13–mode test cycle. In idle mode, PM extract from exhaust of DF was two to three times more mutagenic (revertants x 1,000 per h) than PM extracts from exhaust of the other fuels, although at maximum torque, the extracts of the fuels were similar (Krahl et al. 2003a). Using the same engine and test cycle, Bünger and colleagues subsequently investigated the differences in the mutagenic potential of PM extracts from exhaust of MK 1, RME, DF (sulphur content 41 mg/kg) and a low-sulphur diesel fuel with high aromatic compound content and lower boiling characteristics (DF05). PM extract from exhaust of RME produced the fewest mutations, followed by MK 1, DF05 and DF, which produced four to five times more mutations than RME. The induction of more mutations by PM extracts from exhaust of DF05, MK 1 and DF was attributed to a higher aromatic compound content. Also, RME produced the lowest hydrocarbon and CO emissions as compared to the other fuels (Krahl et al. 2003b [Krahl et al. 2003c, 2003d]).

In a similar study (see Bünger et al. 2000b) published by Bünger et al. in 2006, PM extract from exhaust of RME and SME generated with a Farymann type K 54 one-cylinder direct-injection 3.5 kW diesel engine running at 5 different load modes simulating the operating conditions of

heavy-duty vehicles was analyzed. The test was derived from the 13-mode ECE R49 test. RME and SME produced unexpectedly high numbers of mutations in TA98 with the use of a DOC. PM extract from exhaust of RME produced statistically significant increases in the numbers of mutations (revertants x 1,000 per h) in TA100 with the use of a DOC (as compared to control samples without a DOC). Despite these increases, the mutagenicity levels of RME and SME were still below those generated by PM extracts from exhaust of DF (sulphur content 370 ppm). PM extract from exhaust of LS-DF (both with and without a DOC) generally induced mutations (revertants x 1,000 per h) at levels similar to RME. PM extract from exhaust of RME, SME and LS-DF with and without a DOC induced fewer mutations than DF in all experiments; however, statistical significance was not provided. The authors concluded that use of the DOC increased the formation of direct acting mutagens in biodiesel under certain engine loads (0 and 84%), via the reaction of NO_x with PAHs resulting in the formation of nitro-PAHs. Increased PAH production with use of a DOC at loads of 0% and 84% was attributed to an increase in mutations after treatment with PM extract from exhaust of DF. Additionally, total PM emissions were higher for RME and SME as compared to DF, and these were reduced with the use of a DOC (Bünger et al. 2006).

Comparison of Diesel Fuel with Rapeseed Oil Methyl Ester, Soybean Oil Methyl Ester, Natural Gas Derived Synthetic Fuel, and various diesel Formulations and Biodiesel Blends

The mutagenicity of RME was compared to that of PM extracts from exhaust of DF (sulphur content 35 mg/kg), gas-to-liquid fuel (GTL), Aral Ultimate (Ultimate) and Shell V-Power (V-Power) (generated with a six-cylinder 205 kW Mercedes-Benz engine OM 906 using the 13-mode European Stationary Cycle test) (Krahl et al. 2005). PM extract from exhaust of DF induced the highest increase in mutations (mutations per m^3 exhaust), while RME induced the least. Notably, PM extracts from exhaust of Ultimate and V-Power fuels induced mutations at higher but comparable levels to RME. Addition of the S9 fraction slightly decreased the mutagenic response for all fuels. Rape methyl ester exhaust contained fewer hydrocarbons, CO and PM, whereas NO_x was increased in RME exhaust as compared to DF. This study indicates that lower mutagenicity can be achieved with fuels systems engineering because the reduction of mutation levels generated by the PM extract from exhaust of Ultimate and V-Power fuels approached those observed with RME (Krahl et al. 2005).

Krahl (2008) analysed PM extracts from the exhaust of different fuels generated by different engines: a six-cylinder Mercedes Euro III OM 906 LA with intercooler and turbocharger run on the 13-mode European Stationary Cycle; a Euro IV MAN D08 36 LFL51 with turbocharger, intercooler and exhaust-gas recirculation system run on the European Transient Cycle for mutagenicity and the European Stationary Cycle for determination of regulated emissions; and a one-cylinder 502.019 test engine sampled at rated power only. PM extract from exhaust of B20 (a blend of 80% diesel, Ultimate or V-Power and 20% RME) produced the highest mutations (mutations per plate) when compared to PM extract from exhaust of DF (sulphur content <1-3.0

mg/kg), Ultimate, V-Power, GTL and exhaust of several biodiesel blends. The effect was reduced with the addition of S9 fraction. PM extracts from exhaust of B10 and B30 were also more potent than those of DF at inducing mutations with the exception of DF condensate extract. The authors of the study provided no explanation for the higher mutation levels for B20 (Krahl 2008).

In another study, Krahl and colleagues (2009b) examined the mutagenicity (mutations per m^3 exhaust gas) of PM extract and condensates from exhaust of DF (sulphur content <1 mg/kg), RME, V-Power and B5Ult (5% RME and 95% Aral Ultimate Diesel) (generated with a six-cylinder Euro III Mercedes Benz OM 906 LA test engine with turbocharger and intercooler using the 13-mode European Stationary Cycle) for mutagenicity. Although PM extract from exhaust of DF was the most mutagenic, no statistically significant differences were observed between the different fuel extracts; however, the addition of a DOC reduced DF extract mutagenic levels to those of RME in the TA98 strain. No statistically significant differences were seen between mutagenic effects of the fuel condensates and use of a DOC dramatically reduced the mutagenic potency of the condensates obtained from the PM exhaust for all of the fuels. Notably, CO and hydrocarbon emissions were dramatically reduced in all the fuels with use of a DOC (Krahl et al. 2009b).

Comparison of Diesel Fuel with Rapeseed Oil Methyl Ester, Natural Gas Derived Synthetic Fuel, and Rapeseed Oil (RSO).

Krahl et al. (2009a) investigated the mutagenic effects of rapeseed oil. Extracts of PM and condensates generated from exhaust emissions of a Mercedes Benz Euro III engine OM 906 LA with turbocharger and intercooler using the 13-mode European Stationary Cycle burning DF (sulphur content met EU standard EN590), RME, GTL, and RSO were tested for mutagenicity (mutations per litre exhaust). PM extracts and condensates from exhaust of RSO were significantly more mutagenic than DF, GTL and RME, whereas little difference in mutagenicity was seen between the PM extracts and condensates from exhaust of DF, GTL and RME. Emissions of total hydrocarbons and CO for all fuels were well below the Euro 3 limits. While, RSO had the highest PM emissions they were still below the Euro 3 limit as were the other fuels. Nitrogen Oxide emissions were highest for RSO followed by RME and both exceeded the Euro 3 limit (5 g/kWh) slightly. The authors suggested that the higher induction of mutation by RSO extracts may be due to the presence of the glycerol moiety in triacylglycerols (Krahl et al. 2009a).

Summary/conclusion

Based on the review of the currently available literature of biodiesel emissions health effects related to the initiation of carcinogenesis (clastogenicity, biochemical events associated with genetic instability, cytotoxicity, and mutagenicity), several observations were made. Firstly, clastogenic effects between treatments with exhaust or extract of diesel and biodiesel exhaust *in*

vitro were similar in the one study reviewed. In the case of the *in vivo* study reviewed, no DE treatment was included therefore preventing any comparison. Secondly, biochemical events associated with genetic instability, such as oxidative stress response and DNA damage were similarly affected after treatment with exhaust or PM extract from exhaust of biodiesel and diesel. However, one study indicated a decrease in ROS formation for biodiesel compared to diesel. The inflammatory response tended to be equal or lower for biodiesel exhaust. In one study, there was a slight decrease in response for biodiesel exhaust compared to DF while in a second study there was a significant decrease with supplementation with RME. A third study indicated that the inflammatory response was greater for the SOF of biodiesel exhaust but only in one of three experiments. Thirdly, cytotoxicity was similar between biodiesel and diesel treatments in three studies while in four others, biodiesel demonstrated greater cytotoxicity. Fuel type and engine load appeared to be important factors influencing cytotoxicity; however, only a few studies considered these issues. Last, the majority of mutagenicity testing using the Ames assay demonstrated that PM extract from exhaust of biodiesel is potentially less mutagenic than PM extract from DE.

Although the balance of available literature evaluating the initiation of carcinogenesis suggests that biodiesel exhaust may be less harmful than DE, there are several shortcomings with the dataset. A lack of standardization in study design, including administered dosage, types of fuels (DF and biodiesel), and engine types and loads, makes comparisons of results difficult. The fact that the majority of studies were conducted with either the Ames Tests or short-term *in vitro* assays is also a limiting factor (i.e., a lack of *in vivo* studies). In addition, many of the existing studies did not indicate if the differences between fuel types were statistically significant.

Despite these issues, some variables seemed to be influential in terms of eliciting a mutagenic and/or cytotoxic response from biodiesel versus diesel fuel exhaust. Some biodiesel blends elicited stronger mutagenic (B20 [20% RME and 80% DF]) and cytotoxic (B10 [10% palm fatty acid methyl ester and 90% DF]) responses than others. However, due to the lack of data available, the specific blend (ratio of biodiesel to diesel) to produce a maximum or minimum effect could not be identified. Other factors that appeared to influence the cytotoxicity and/or mutagenicity were the type of biodiesel, engine load and mode, chemical constituents in fuel emissions, and the use of emission control technologies (e.g., DOC). Cold-pressed RSO (unprocessed) was consistently more mutagenic than any other fuels, including diesel. In addition, the lower emissions of hydrocarbons (including PAHs) observed in biodiesel exhaust appeared to correlate with its decreased mutagenicity compared to diesel fuel. Finally, the utilization of a DOC was shown to lower chemical emissions (e.g., particulates, hydrocarbons) and subsequently lowers the mutagenic potential; however, use of a DOC under certain engine loads was also shown to increase the formation of nitro-PAHs. In general, given the lack of information and consistency in study design, it was not possible to determine the specific effects of individual engine parameters or exhaust chemical-physical characteristics.

Although the available evidence suggests that exhaust from certain formulations of biodiesel (under some engine load/mode and emission control conditions) is less mutagenic than DF exhaust, generalizations about the carcinogenicity of biodiesel based on one type of engine mode or fuel blend should be avoided. More research is needed using standardized methods to better understand the comparative toxicity of biodiesel in relation to events potentially leading to the initiation of carcinogenicity. That being said, given the results for mutagenicity to date, it would appear that biodiesel exhaust represents a reduced carcinogenic risk compared to DE.

7.2.5 Reproductive and developmental effects

7.2.5.1 Biodiesel exhaust

The following text reviews the information on reproductive and developmental effects resulting from exposure to biodiesel exhaust and focuses on the key outcomes: teratogenic effects; decreased reproductive capacity and gonadal abnormalities; reduced birth/body weight, premature birth, intrauterine growth retardation; decreased foetal/post-natal survival; and estrogenic activity.

Teratogenic effects

NBB (2000a)/Finch et al. (2002) performed a 13-week subchronic inhalation study (see Subsection 7.2.1.1 *Inflammation* for additional details) in F344 rats to determine the potential toxicity of biodiesel exhaust emissions. Twenty-five females were used per exposure group for the reproductive toxicology portion of the study (NBB 2000a).

The reproductive tracts from animals in the reproductive toxicology (with gross lesions or failed to reproduce), the general histopathology (high-level and control groups), and the special histology (all exposure groups) portion of the study were examined. No exposure-related lesions were observed in the reproductive tracts of any rats and there were no statistically significant or biologically meaningful differences observed between control and exposed groups with respect to the number of foetal malformations. On this basis, the authors concluded that biodiesel is not teratogenic (NBB 2000a; Finch et al. 2002).

The NBB (2000a)/Finch et al. (2002) research was the only study that assessed the reproductive toxicology resulting from inhalation exposure to biodiesel exhaust. It should be noted that this study did not include animals exposed to equivalent concentrations of DE thus limiting a comparison of reproductive outcomes.

Decreased reproductive capacity and gonadal abnormalities

In the 13-week subchronic inhalation study by NBB (2000a)/Finch et al. (2002), exposure to biodiesel exhaust did not affect pregnancy rates, which were 21/22, 18/23, 22/23, and 25/25 for the control, low, intermediate, and high exposure level groups, respectively. It was also

determined that there were no statistically significant or biologically meaningful differences observed between control and exposed groups with respect to the number of corpora lutea; implantations; early, late or total resorptions; and foetal sex ratios.

The NBB (2000a)/Finch et al. (2002) study was the only research identified that investigated the potential for biodiesel exhaust to affect reproductive capacity. Although this study did not include similar exposures to DE, exposure to biodiesel exhaust did not cause any effects for the selected endpoints when compared to clean air. However, it is not clear what impact biodiesel exhaust exposure would have on the development of the foetal reproductive system.

Reduced birth weight, reduced body weight, premature birth, and intrauterine growth retardation

In the 13-week subchronic inhalation study by NBB (2000a,b)/Finch et al. (2002), mean foetal weights (grams) for the different exposure groups were: 3.14 (control); 3.14 (low); 3.07 (intermediate) and; 3.21 (high). Results indicated that there were no statistically significant or biologically meaningful differences observed between exposure and control groups in terms of foetal weights. The results contributed to the determination that biodiesel exhaust is not foetotoxic.

NBB (2000a,b)/Finch et al. (2002) was the only study that examined the effect of inhalation exposure to biodiesel exhaust on body weights. Reproductive endpoints such as pre-mature birth of foetuses were not considered in this study (i.e., rats were delivered via caesarean section).

Decreased foetal/post-natal survival

In the 13-week subchronic inhalation study by NBB (2000a,b)/Finch et al. (2002), the mean number of viable foetuses per female for each exposure group was: 7.1 (control); 7.6 (low); 8.2 (intermediate); and 8.0 (high). Results indicated that there was no statistically significant or biologically meaningful difference between exposure and control groups in terms of the viability of foetuses. The authors concluded that biodiesel exhaust is not foetotoxic. This was the only study that examined reduced foetal survival resulting from inhalation exposure to biodiesel exhaust.

Estrogenic activity

No studies were found in the literature that considered estrogenic activity as a result of exposure to biodiesel exhaust.

Summary/conclusion

The NBB (2000a)/Finch et al. (2002) study indicated that subchronic exposure to biodiesel exhaust is unlikely to result in reproductive and developmental effects based on the selected endpoints. However, it is not clear if exposure to biodiesel exhaust would affect other endpoints such as the development of the foetal reproductive system and the pre-mature birth of foetuses.

No information was located for effects on the endocrine system resulting from exposure to biodiesel exhaust.

Given the lack of data for biodiesel exhaust, it was not possible to determine if biodiesel exhaust results in a similar, decreased, or increased impact on reproductive and developmental outcomes relative to DE.

7.2.6 Neurological effects

7.2.6.1 Biodiesel exhaust

The following text reviews the information on neurological effects resulting from exposure to biodiesel exhaust and focuses on the key outcomes: effects on behaviour; and abnormal neuropathology and neurodegeneration.

Effects on behaviour

No studies were found that looked at behavioural effects resulting from exposure to biodiesel exhaust, either in humans or in animal models.

Abnormal neuropathology and neurodegeneration

NBB (2000a)/Finch et al. (2002) performed a subchronic inhalation study (see Subsection 7.2.1.1 *Inflammation* for additional details) in F344 rats to determine the potential toxicity of biodiesel exhaust emissions (NBB 2000a). The authors examined brain tissue from five males and five females per exposure group and analyzed for increases in concentrations of glial fibrillary acidic protein (GFAP) - a marker of neuronal injury (astrogliosis). The results showed a slight increase in GFAP in the high-level group compared to controls but this increase was not statistically significant. Levels of GFAP in the lowest exposure group were significantly decreased when compared to the control group, although this was not deemed to be a measure of toxicity.

During the course of this study, a number of animals were subjected to clinical observations and histopathological examinations. There were no observed changes in absolute or relative body or brain weights, or lesions in nervous tissues or tibial nerves, in the high-level exhaust exposure group compared to the control group. Given that the highest exposure level did not result in animals showing histologic evidence of nervous tissue toxicity, the intermediate- and low-level exhaust exposure groups were not evaluated. The authors concluded that, given the absence of clinical signs of neurotoxicity or brain toxicity, the observed differences between groups in terms of GFAP levels, were not biologically significant (NBB 2000a).

The NBB (2000a)/Finch et al. (2002) was the only study identified that investigated the potential for abnormal neuropathology and neurodegeneration. Exposure to biodiesel exhaust did not cause any effects for the selected endpoints.

Summary/conclusion

While the NBB (2000a)/Finch et al. (2002) study indicated that subchronic exposure to biodiesel exhaust is unlikely to result in neuropathology and neurodegeneration outcomes based on the selected endpoints, a lack of information prevents any conclusions from being drawn with respect to effects on behaviour. Given the lack of comparable data (biodiesel exhaust versus DE under similar exposure scenarios) it is not known whether biodiesel emissions result in a similar, decreased, or increased impact relative to diesel emissions.

7.2.7 Systemic effects

7.2.7.1 Biodiesel exhaust

The following text reviews the information on systemic effects resulting from exposure to biodiesel and focuses on changes in: body weight, organ weight, organ-to-body weight ratio, food consumption, and mortality. Results from dermal and oral exposure studies are also presented. The dermal studies were included because potential exposure may occur during refuelling. Results from oral exposure studies were included because information on systemic effects in general is limited.

Body Weights

NBB (2000a)/Finch et al. (2002) carried out a subchronic inhalation study (see Subsection 7.2.1.1 *Inflammation* for additional details) in F344 rats to determine the potential toxicity of biodiesel exhaust emissions.

Body weights were measured twice during the pre-study phase, biweekly throughout the study after daily exposures, and at necropsy. Rats from all exposure levels gained weight throughout the study. With the exception of 2 high dose females that weighed less than the controls, there were no significant differences in body weights between the exposure groups and the controls (NBB 2000a).

Peterson and Moller (2005) reviewed a group of studies by the University of Idaho that examined the effect of acute dermal and acute oral exposure to 100% RME, 100% rapeseed ethyl ester (REE), 100% Phillips 2-D low sulphur diesel (control), 50% RME/50% 2-D, 50% REE/50% 2-D, 20% RME/80% 2-D, and 20% REE/80% 2-D on albino rabbits (dermal) and albino rats (oral).

Ten albino rabbits, 5 males and 5 females, were administered a single dermal dose of 100% RME, 100% REE or 100% 2-D at a level of 2000 mg/kg. Body weights were obtained and recorded at the beginning of the study (day 0), day 7, and day 14. The authors concluded that dermal exposure to 100% RME, 100% REE and 100% 2-D did not cause any changes in rabbit body weights (Peterson and Moller 2005).

Ten rats, five males and five females, were administered single oral doses of 2-D diesel, 100% RME, 100% REE, 50% RME/50% 2-D, 50% REE/50% 2-D, 20% RME/80% 2-D, and 20% REE/80%2-D) at a level of 5000 mg/kg. Body weights were recorded at the beginning of the study (day 0), after 7 days, and at termination (14 days). The authors of the study determined that oral exposure to 100% RME, 100% REE, 50% RME, 50% REE, 20% RME, 20% REE, and 100% 2-D did not result in significant body weight changes (Peterson and Moller 2005).

In a 4-week oral study by Poon et al. (2007), 35 male Sprague-Dawley rats (7 animals /group) were administered one of three biodiesels – canola oil methyl ester (CaME), soy oil methyl ester (SME), or fish oil methyl ester (FiME) at 500 mg/kg, low sulphur diesel (LSD) at 500 mg/kg, or corn oil (control) at 500 mg/kg. Final body weights for CaME, SME, FiME, and LSD were not significantly different from the control (Poon et al. 2007).

A 4-week oral study by Poon et al. (2009), 77 male Sprague-Dawley rats were administered one of three biodiesels – SME, CaME, and FrAME (animal frying oil methyl ester) at 5, 50 and 500 mg/kg, ultra-low sulphur diesel (ULSD) at 500 mg/kg, or a control (corn oil) at 500 mg/kg. Growth curves and final body weight gain of all treatment groups were not significantly different from the control (Poon et al. 2009).

Organ Weights

In the NBB (2000a)/Finch et al. (2002) subchronic inhalation study organ weights were measured for the treatment (low, intermediate, and high) and control groups. The mean liver weight in the high-level group was significantly lower than the controls in both males (10.7 g vs. 11.6 g) and females (5.5 g vs. 6.2 g), respectively. The authors concluded that the liver weight difference probably had no biological significance, and was not related to the level of exposure given that no lesions were observed. A significant increase in lung weights in high-level females was observed and was consistent with histopathological changes. No difference in absolute brain weights was observed between the exposure and control groups (Finch et al. 2002).

In the 4-week oral study by Poon et al. (2007), male Sprague-Dawley rats were administered CaME, SME, FiME, LSD, or corn oil (control). The major treatment effect, produced by oral administration of CaME, SME, and LSD, was hepatomegaly. Mean liver weights were highest for SME, followed by LSD, CaME, FiME and the control; however, it was not clear if these differences were significant.

Organ-to-body weight ratio

In the 13-week subchronic inhalation study by NBB (2000a)/Finch et al. (2002) organ-to-body weight ratios were measured for the treatment (low, intermediate, and high) and control groups. Liver-to-body weight ratios were lower for female rats in the high (2.8%) and intermediate

(3.0%) treatments compared to the controls (3.1%). Lung-to-body weight ratios in high-level female rats were higher than the controls (0.52% versus 0.49% respectively). Finally, in males, testes-to-body weight ratios were greater in the high-level group compared to the control (0.91% versus 0.86% respectively). The authors concluded that since there were no lesions observed in the liver and testes, the slight weight differences were of uncertain biological importance (Finch et al. 2002).

In the 4-week oral study by Poon et al. (2007), significantly increased liver-to-body weight ratios were noted in study groups receiving CaME (0.0384 ± 0.0014 grams), SME (0.0457 ± 0.0021 grams), and LSD (0.0458 ± 0.0030 grams) compared to the control (0.0345 ± 0.0042 grams). A significant decrease in thymus to body weight ratio was also observed in animals receiving SME (0.0012 ± 0.0002 grams) compared to the control. There were no significant changes in organ-to-body weight ratios observed for other organs (Poon et al. 2007).

In the 4-week oral study by Poon et al. (2009), a significantly increased liver-to-body weight ratio (ULSD: 4.07 ± 0.18 grams; control: 3.84 ± 0.34 grams) and kidney-to-body weight ratio (ULSD: 0.834 ± 0.048 grams; control: 0.691 ± 0.053 grams) were observed in the treatment group dosed with 500 mg/kg ULSD when compared to the control. There were no significant changes in organ-to-body weight ratios for the brain, heart, spleen, or testes (Poon et al. 2009). It should be noted that this study did not present results for organ weights.

Food Consumption

NBB (2000a)/Finch et al. (2002) carried out a 13-week subchronic inhalation study in F344 rats to determine the potential toxicity of biodiesel exhaust emissions. Consumption of feed was measured throughout the course of the study for the control, low, intermediate, and high-level groups. Group mean values for feed consumption ranged from 0.062 g to 0.066 g feed consumed/gram of rat/night. The authors determined that there were no significant differences attributable to treatment levels (NBB 2000a).

Mortality

In the NBB (2000a)/Finch et al. (2002) subchronic inhalation study, the rats (120 males and 220 females) were visually inspected twice daily for mortality. With the exception of three animals, all rats survived until their scheduled sacrifice. Two rats, a control group rat and an intermediate group rat, were sacrificed due to cage-related traumas, while a third rat (from the control group) was sacrificed after marked weight loss, the cause of which was not determined. The authors of the report determined that causes of death were not associated with exposure to biodiesel exhaust emissions (NBB 2000a).

Peterson and Moller (2005) summarized a study that looked at the effect of acute oral exposure to RME and REE compared to the control diesel (2-D) in albino rats, and acute dermal exposure

to RME and REE compared to the control diesel (2-D) in albino rabbits. Both rats and rabbits were observed for mortality at 1, 3, and 4 hours post-exposure on day 0 and twice daily (morning and afternoon), every day, for 14 days. The authors of the study found that acute oral exposure (5000 mg/kg) to 100% RME, 100% REE, 50% RME, 50% REE, 20% RME, 20% REE, and 100% 2-D did not result in any deaths. Likewise, acute dermal exposure (2000 mg/kg) of white albino rabbits to 100% RME, 100% REE, and 100% 2-D did not cause any mortality (Peterson and Moller 2005).

Summary/conclusion

Subchronic and chronic inhalation exposure to DE and DEP in animals has been associated with changes in body weights (Pepelko 1982; Schreck et al. 1981; Heinrich et al. 1986; Heinrich et al. 1995; Nikula et al. 1995; US EPA 2002c; Stinn et al. 2005; US EPA 2009). Body weights were not affected by biodiesel via inhalation (NBB 2000a; Finch et al. 2002) dermal (Peterson and Moller 2005) or oral (Peterson and Moller 2005; Poon et al. 2007; Poon et al. 2009) exposures.

Inhalation exposure to DE has resulted in increases in lung weights in experimental animals (Kaplan et al. 1982; Vinegar et al. 1981a, Vinegar et al. 1981b; Brightwell et al. 1986; Henderson et al. 1988; Heinrich et al. 1995; Nikula et al. 1995; US EPA 2002c; Stinn et al. 2005; US EPA 2009). Organ weights were affected by biodiesel via inhalation and oral exposures. NBB (2000a)/Finch et al. (2002) indicated inhalation exposures of biodiesel significantly increased lung weights in female rats (high dose) compared to the control. Poon et al. (2007) indicated oral exposures of biodiesel (CaME, SME, or FiME) and LSD increased liver weights compared to the control. However, it was not clear if the differences between treatments were significant.

Subchronic and chronic inhalation exposure to DE has been shown to cause significant increases in lung-to-body weight ratio in animal studies (Kaplan et al. 1982; Wiester et al. 1980; US EPA 2002c). Different organ-to-body weight ratios were affected by biodiesel via inhalation and oral exposures. NBB (2000a)/Finch et al. (2002) indicated that inhalation exposures to biodiesel affected liver (i.e. lower for high and intermediate dose females), lung (i.e. increased for high dose females), and testes-to-body weight (i.e. increased for high dose males) ratios when compared to controls. Poon et al. (2007) indicated oral exposures of biodiesel and diesel increased liver-to-body ratios compared to the control and that there was a significant decrease in thymus-to-body weight ratio for animals treated with SME.

Chronic inhalation exposure of DE was shown to lower food consumption in rats (Stinn et al. 2005; US EPA 2009). Based on the results of the NBB 2000a/Finch et al. 2002 study, food consumption was not affected by biodiesel via inhalation exposure.

Evidence of mortality from subchronic and chronic inhalation DE studies in animals is varied (Reed et al. 2004; Kaplan et al. 1982; Heinrich et al. 1986; Nikula et al. 1995; Karagianes et al. 1981; Mauderly et al. 1984; Mauderly et al. 1987; Mauderly et al. 1996; Lewis et al. 1989; US EPA 2002c). Subchronic and chronic exposure to ambient PM is thought to be a major contributor to mortality in humans (Bunn et al. 2004; Englert 2004; Valberg 2004; Englert 2007). Mortality was not affected by biodiesel via inhalation (NBB 2000a; Finch et al. 2002), dermal or oral exposures (Peterson and Moller 2005).

Short term dermal exposure to diesel in animals and humans may result in local irritation of the skin (Beck et al. 1984; Koschier 1999; Health Protection Agency 2006). The only dermal study for biodiesel reviewed by Peterson and Moller (2005) indicated that when rabbits were administered a single dermal dose of 100% RME, 100% REE or 100% 2D at a level of 2000 mg/kg, there were no effects on body weight or mortality. No information was available on potential skin irritation resulting from exposure to biodiesel.

Given that the NBB (2000a)/Finch et al. (2002) study did not include a diesel treatment, and no other relevant information was available it was not possible to determine how biodiesel exhaust compares with DE with respect to changes in body weight, organ weights, organ-to-body weight ratios, food consumption and mortality following inhalation exposure.

7.2.8 Conclusions

First objective of evaluation: determine if biodiesel exhaust has a similar, reduced or greater impact than DE in terms of specific health effects and outcomes.

The evidence from *in vivo* and *in vitro* studies for respiratory effects resulting from biodiesel exhaust exposure is limited and varied. As such, it is not possible to make a definitive conclusion with respect to how biodiesel exhaust compares with DE. It seems that, in balance, biodiesel exhaust is unlikely to exceed DE in terms of respiratory effects.

No information was available for immunological effects resulting from exposure to biodiesel exhaust. Therefore, it is not possible to determine how biodiesel exhaust and DE compare.

Health Canada reviewed two *in vivo* studies which examined the cardiovascular effects of biodiesel exhaust. In an acute study, exposure of biodiesel exhaust resulted in increases in some systemic inflammatory markers when compared to DE. A subchronic study that included no comparative diesel treatment did not record any significant cardiovascular effects in animals exposed to biodiesel exhaust. Given the limited data set, it was not possible to draw any conclusions as to how biodiesel and DE compare with respect to cardiovascular effects.

Several outcomes (clastogenicity, biochemical events associated with genetic instability, cytotoxicity, and mutagenicity) relevant to the initiation of carcinogenesis were reviewed. With

the exception of mutagenicity, only a few studies were available for each outcome. With respect to clastogenicity effects, biodiesel and DE extracts were similar in the one *in vitro* study reviewed. In three studies that examined genetic instability, the oxidative stress response was similarly affected in two studies after treatment with exhaust or PM extract from exhaust of biodiesel and diesel. The third study indicated a decrease in ROS formation for biodiesel compared to DE. The inflammatory response was equal or lower for biodiesel exhaust except for one study that compared the SOF of biodiesel and DE. Cytotoxicity was similar for biodiesel and diesel treatments in three studies reviewed, while in four others, biodiesel demonstrated greater cytotoxicity. The limited data set indicated that biodiesel and DE are similar in terms of clastogenicity, biodiesel exhaust has a similar or lower effect for genetic instability (ROS, inflammation), and biodiesel exceeds diesel with respect to cytotoxicity. The majority of studies investigating mutagenicity demonstrated that PM extract from biodiesel exhaust is potentially less mutagenic than DE PM extract. Given that many of the studies examining clastogenicity, genetic instability, cytotoxicity and mutagenicity were conducted under different experimental conditions, generalizations about the initiation of carcinogenesis of biodiesel compared to DE based on one type of engine or fuel should be avoided.

A subchronic study was reviewed that considered reproductive and developmental effects. The results indicated that exposure to biodiesel exhaust (no diesel treatment included) in rats is unlikely to result in reproductive and developmental effects based on the selected endpoints. No information was available for other endpoints including development of the foetal reproductive system, the pre-mature birth of foetuses or endocrine effects. Given the lack of data for biodiesel and DE for reproductive and developmental effects it was not possible to determine how these two fuels compare.

A subchronic study indicated that exposure to biodiesel exhaust (no diesel treatment included) is unlikely to result in neuropathology and neurodegeneration based on the selected endpoints. No information was available for effects on behaviour. Given the lack of comparable data (biodiesel exhaust versus DE under similar exposure scenarios) for neurological effects it was not possible to determine whether biodiesel emissions result in a similar, decreased, or increased impact relative to diesel emissions.

With respect to systemic effects, a review of 4 studies indicated that body weights, food consumption and mortality were not affected by inhalation, dermal or oral exposure to biodiesel. The same studies indicated that organ weights and organ-to-body weight ratios were impacted by inhalation and oral exposures. Given that the inhalation study did not include a diesel treatment it was not possible to determine how biodiesel compares with diesel for the various systemic outcomes. A potential outcome of dermal exposure during refuelling (i.e., skin irritation) was not considered in the dermal study for biodiesel.

Second objective of evaluation: attribute any difference in the magnitude of health effects observed (between biodiesel and DE) to a change in the level of specific physicochemical parameter(s).

The literature for respiratory, cardiovascular, and outcomes associated with initiation of carcinogenesis increasingly reflect research efforts to ascribe differences in biological responses between biodiesel and DE to differences in physicochemical characteristics between the two fuels. However, in the majority of studies, differences in individual pollutant levels between biodiesel and DE have not been specifically linked to changes for a given biological response.

In the case of respiratory effects, Swanson et al. (2009) found that the biodiesel SOF was linked to the inflammatory response of BEAS-2B cells. Liu et al. (2008) observed that in the case of exhaust from biodiesel and biodiesel blends, the semi-volatile extracts tended to be more toxic than the particulate extracts whereas the opposite was observed for DE. Tzamkiozis et al. (2010) and Jalava et al. (2010) examined the association between different pollutants (inorganics, aromatics and ROS capacity) and the inflammatory response but did not establish specific causal relationships. Brito et al. (2010) characterized levels of various pollutants ($PM_{2.5}$, black carbon, inorganics, CO, PAHs, VOCs) in biodiesel and DE but did not identify which pollutants were responsible for the increases in inflammatory markers. In the mutagenicity literature, several authors have attributed lower mutagenicity rates in biodiesel extracts compared to diesel extracts, to lower PAHs levels.

7.2.9 Uncertainties

Madden et al. (2011) concluded that biodiesel can have more, less, or the same potency in inducing biological responses and health effects as petroleum DE. This conclusion parallels to some extent the findings of the present evaluation and may reflect, to a certain degree, the level of uncertainty which is inherent to the comparison of health effects for biodiesel and DE. Much of the uncertainty stems from the fact that there is a shortage of information on health effects for biodiesel. The following text outlines some important areas of uncertainty that were encountered in carrying out this evaluation.

The majority of information on health effects for biodiesel was obtained from relatively high-level or acute *in vitro* exposures. The relevancy of the high-level exposure studies to low-level chronic exposures, which may be experienced in the general population, was not always clear. The possibility in the future of more *in vitro* studies at environmentally relevant exposure levels as well as *in vivo* studies for whole body exposures in animals will hopefully contribute to a more complete data base.

The existing studies for biodiesel do not always include exposures to DE thus making any comparisons of health effects/outcomes challenging. Comparing results from biodiesel and diesel studies is also challenging given the use of different experimental designs including variation in

animal models, fuels, treatment levels, engine types and conditions, and study methodologies. In addition, many of the existing studies did not indicate if the differences between fuel types were statistically significant.

Comparing the extensive literature for diesel and the limited data set for biodiesel is another challenge. The range of methodologies (i.e., different animal models and engine systems) used in studies for conventional diesel is extensive in comparison to the limited range of permutations used in the reviewed biodiesel studies.

Madden et al. (2011) recommended improved reproducibility of design for future studies in order to assess how biodiesel compares with diesel for different health effects. Study designs should attempt to narrow down the fuel (including feedstock) type used, minimize fuel impurities, use a current and widely used engine, and standardize the run conditions, so that emissions from different studies are reasonably similar (Madden et al. 2011). Other factors to consider include animal models, extraction methodologies, and after treatment devices.

Improved reproducibility of study design would also facilitate the task of characterizing and determining the toxicology of biodiesel exhaust in comparison to DE (i.e., role of gas versus PM phase in biological responses) (Madden et al. 2011).

7.3 Biodiesel and Bovine Spongiform Encephalopathy

The objective of this section is to examine the risk that inhalation exposure of the Bovine Spongiform Encephalopathy (BSE) infectious agent will occur in the general population as a result of the combustion of biodiesel made from tallow derived from Specified Risk Material (SRM). In considering the combustion scenario, the review also examines the implications of using tallow feedstock that exceeds the regulated level for insoluble impurities and how this may impact inhalation exposure of humans to the BSE agent.

SRM includes the parts of animals that comprise the greatest potential for BSE infectivity. According to the Canadian Food Inspection Agency (CFIA), these materials include: skull, brain, trigeminal ganglia (nerves attached to the brain), eyes, tonsils, spinal cord and dorsal root ganglia (nerves attached to the spinal cord) of cattle aged 30 months or older and the distal ileum (portion of the small intestine) of cattle of all ages (CFIA 2011).

Bovine spongiform encephalopathy, commonly referred to as mad cow disease, is the disorder resulting from the accumulation of misfolded prion proteins in the brains of cattle. Alternate forms of the neurodegenerative disease (transmissible spongiform encephalopathies (TSE)) have been observed in other mammalian species, including humans. This class of disorder is unique as it is the only disease currently known that is believed to be propagated exclusively by a protein (i.e., no transmission of DNA or RNA). Prion proteins (PrP) are naturally found within brain

tissue of animals in either of two forms: PrP^{C} (C – cellular) and PrP^{Sc}(Sc – scrapie) (also known as PrP^{Res}). The PrP^{Sc}(PrP^{Res}) form is an infectious agent that is most likely responsible for transmissible spongiform encephalopathies, including BSE (ATF Can 2005).

7.3.1 Tallow and biodiesel production

7.3.1.1 Source of animal tissue

The risk of BSE presence in biodiesel can be reduced through prudent selection of starting materials. Important factors to consider in selecting starting materials include: the species, anatomical parts utilised and the country of origin of the animals (ATF Can 2005).

- Species: Regarding farmed food animals, the reported incidents of BSE have occurred primarily in cows. However, field cases have been documented in goats and experimental infections have been reported in both sheep and goats. Pigs have been infected experimentally by the intracranial, intravenous and intraperitoneal routes (CFSPH 2007). Alternate forms of TSEs have been reported in other farmed animals, zoo ruminants and captive wildcats (Sigurdson et al. 2003).

- Anatomy: The use of non-SRM tissues greatly reduces the chance of exposure to BSE agents, and thus reduces the chance of infection. Given the use of SRM, it is important to note that due to the lengthy incubation times of BSE, tissue collected from cattle aged less than 30 months has a reduced detection limit for the infectious agent. As the infection progresses, the agent becomes detectable in different tissues of the body at different concentrations. Detectable quantities may not be present in brain tissue until the later stages of the disease; however, this does not mean that the BSE agent is not present in other tissues (e.g., distal ileum) at significant quantities prior to 30 months (pers. comm. Dr. Penny Greenwood 2009; European Commission 2002; Espinosa et al. 2007).

- Geographic: The World Organisation for Animal Health (OIE) has categorized several countries based on their BSE risk and BSE risk mitigation measures. The categorization system rates a country with respect to BSE risk as: negligible, controlled or undetermined (OIE 2011).

7.3.1.2 Tallow Production

The production of biodiesel from bovine fat involves the rendering of cattle by-product tissue to release tallow and the subsequent transesterification of the tallow into biodiesel. The risk of BSE infectivity throughout the production process can be mitigated by selecting safer source tissues. However, the use of SRM as source tissue presents an economically attractive and possibly safer alternative to standard SRM disposal methods (ATF Can 2005). This practice might be considered safer since it effectively destroys the BSE agent and limits the propagation of the disease during the tallow and biodiesel production process. However, if the methods used during biodiesel production are not effective at inactivating the BSE agent, the risk of human exposure will potentially increase with biodiesel use.

Rendering refers to the process whereby animal by-products are separated through cooking. The by-product material is cooked thereby sterilizing the microbiological content, water is evaporated and the fat is extracted via pressing or centrifugation. This process generates animal fat and a protein rich mixture known as meat and bone meal (MBM). In the case of cattle rendering, the fat is known as tallow. The vast majority of proteinaceous material, including the BSE agent if present, partitions into the MBM fraction. Concentrations of insoluble material in tallow, including proteins, are expected to be minimal (≤0.15% in filtered tallow, of which 5–16% is predicted to be protein) (SSC 2001; ATF Can 2005). Rendering is carried out using either a batch or continuous processing system. A continuous system employs a constant feed stream at one end of the cooker and a constant product flow at the other. Particles in a continuous process vary in residence time. A batch system involves cooking a large mass at once, emptying the cooker, and subsequently refilling it with more material. A batch system cooks all material at once for the same residence time (CFIA 2006). Currently in North America, continuous production methods are favoured by the rendering industry (AARI 2005).

Canadian Food Inspection Agency policy considers SRM-derived tallow with a maximum of 0.15% insoluble impurities to be of negligible risk with respect to BSE agents and does not require any downstream products (i.e., biodiesel) adhering to this standard to be classified as SRM. As such, SRM-derived tallow meeting the 0.15% standard is exempted from the permit controls, record keeping and other requirements specified in the Health of Animals Regulations (pers. comm. Sergio Tolusso 2008, 2009, 2011a and 2011b; Section 6.6 of the Health of Animals Regulations; Task Specific Notes and Policy, CFIA 2007). The CFIA enforces this tallow purity standard via the Federal Health of Animals Act (Section 162(1) (e) of the Regulations) and the Feeds Act (Section 19(1) (d.3) of the Regulations).

The CFIA's tallow purity standard is based on the OIE's health standard for tallow, i.e., not more than 0.15% insoluble impurity content (OIE 2010). According to the OIE, this level represents a negligible risk for products destined for international trade (OIE 2010). The EU has also established a standard for insoluble impurity levels (0.15%) for tallow (derived from ruminant animal by-product including SRM) intended for biodiesel production (ATF Can 2005).

7.3.1.3 Biodiesel production

Fats and oils from both animal and plant sources, including tallow, are composed of triglyceride molecules. This molecule includes three long chain fatty acids (8–22 carbons) attached to a glycerol backbone. Transesterification refers to the process by which triglycerides are transformed into alkyl esters (biodiesel), glycerine, and free fatty acids through an acid or base catalyzed hydrolysis reaction with an alcohol (usually methanol) (ATF Can 2005).

Base catalyzed reactions are preferred by industry as they are faster and cheaper to run compared to acid catalyzed reactions. Acid-catalyzed reactions are slower and require higher temperatures and pressures but serve to prevent the saponification of free fatty acids which may be present in

the feedstock. The high temperatures used during the rendering of tallow increase the proportion of free fatty acids in the stock material compared to vegetable oils. The bases used during base-catalyzed reactions (usually KOH or NaOH) will react with these fatty acids to form soap, turning the stock material into a gel-phased substance and halting the conversion into biodiesel. Therefore, effective biodiesel production from tallow generally involves a combination of both processes, i.e., a combined acid-base catalyzed process (ATF Can 2005). The description that follows is of a generic process that is used by most biodiesel producers worldwide (ATF Can 2005).

Tallow is initially subjected to an acid catalyzed pre-treatment stage in order to lower free fatty acid concentrations. This reaction takes place between 60–70°C at 140–400 kPa in the presence of a strong acid (≤0.5 M H_2SO_4). The conversion of free fatty acids is usually complete within 2 hours. A molar ratio of 20–40:1 alcohol to oil is used during this step, which may result in the alcohol fixation of TSE agents, causing them to stabilise and be protected from subsequent exposure to heat. On the other hand, strong acids at elevated temperatures (>60°C) appear to have the ability to reduce the infectivity of TSE agents (ATF Can 2005).

The pre-treated mixture with less than 1 wt% free fatty acids is then subjected to transesterification via a base catalyzed reaction. Methanol is added to the triglyceride mixture at a ratio of approximately 6:1. The reaction to produce methyl esters of the fatty acids takes 1–4 hours at 60–65°C, and 140-400 kPa. Sodium hydroxide is the most common catalyst used and is added at approximately 0.5–2 wt% (0.09–0.36 M). The exposure of any TSE agents in the reaction mixture to any basic reagents is expected to be minimal and have little effect on TSE infectivity reduction. As well, the temperatures and pressures used during this phase of the process will have no significant effect on TSE infectivity reduction, especially with respect to the BSE form (ATF Can 2005).

Given the lack of empirical data, it is uncertain whether transesterification reactions result in significant inactivation of BSE agents. Research is required to determine what role the combinations of acids or bases, temperature, pressure and time play in regards to reducing BSE infectivity. Similarly, the role that alcohols and other co-solvents play needs to be investigated as well (ATF Can 2005).

The European Food Safety Authority has approved the biodiesel production process as safe for the processing of animal fat separated from Category 1 (high risk of TSE) animal by-products. However, it should be noted that this approval was based on research conducted with a biodiesel process that included one esterification step, two transesterification steps, three washing steps, and a starting material that had undergone a rendering treatment consisting of temperatures ≥ 133°C and at 300 kPa pressure for 20 minutes (EFSA 2004; Seidel et al. 2006). Mittelbach et al. (2007) calculated reduction factors for pre-esterification, transesterification and vacuum

distillation steps and concluded that prion contamination would be destroyed by the biodiesel production process. Application of these findings (EFSA 2004; Seidel et al. 2006; Mittelbach et al. 2007) to the North American situation may not be suitable given that "pressure cooking" rendering is not practiced in North America, nor is it clear (based on the absence of information) that this particular biodiesel process is routinely used in North America. With respect to the latter, it was recommended in the ATF Can report (2005) that experimental work is required to examine the deactivation capabilities of different commercially viable biodiesel production processes.

7.3.1.4 Purification of biodiesel

There are different options for biodiesel purification that include passing the biodiesel over an absorbent and then filtering it, passing the biodiesel through activated carbon to improve the colour, and using vacuum distillation to deodourise the biodiesel. Most production facilities also use a final filter of about 5 µm. Due to the adherent nature of TSE agent, the absorbent or activated charcoal steps are thought to significantly reduce any residual infectivity in biodiesel. The vacuum distillation process is used to remove methanol and water. The biodiesel itself is not distilled and therefore any protein that might be present in the biodiesel would remain after the distillation step (ATF Can 2005).

7.3.1.5 Biodiesel (tallow-derived) combustion

Tallow-derived biodiesel is expected to be used in compression ignition (CI) engines; however, there are currently no experimental data available on the inactivation of TSE agents in CI engines. Fuel in CI engines is exposed to temperatures of up to 3327°C and pressure of 4–10 MPa but only for very short periods of time, i.e., the millisecond range. As the various phases in CI engines present a non-uniform temperature gradient and the exposure times are quite short, all potential TSE agents may not be exposed to the same high temperatures required for inactivation. Generally, 2% of the fuel entering a CI engine combustion chamber is not oxidised to CO_2 and water, but is transformed into soot and other polycyclic aromatic hydrocarbons. This effect is amplified in cold start situations where emissions can contain up to 18% pyrolysis products (ATF Can 2005). In terms of new treatment technologies for CI engines, i.e., exhaust gas recirculation and exhaust after treatment, it is unclear how effective these would be in inactivating TSE agents given that these technologies can modify the engine temperature (ATF Can 2005).

In 2002, Cummins et al. developed a model to assess the human health risks associated with the combustion of SRM-derived tallow in Ireland. Routes examined included direct inhalation, the consumption of crops following deposition of airborne BSE, as well as the infiltration of BSE into surface and ground water systems. The risk of a human contracting Creutzfeldt-Jakob disease (vCJD)[69] following exposure to airborne BSE, resulting from the combustion of SRM-

[69] Variant Creutzfeldt-Jakob (vCJD) disease occurs when humans are possibly infected by animal prions, such as the infectious BSE agent (Dormant, 2002).

derived tallow, was calculated to range from $10^{-11.43}$ to $10^{-7.23}$ per year/person. Given that the spontaneous rate of vCJD occurrence in humans is approximately 10^{-6}, it was concluded that the human health risks associated with airborne BSE released from tallow combustion are negligible (Cummins et al. 2002). This being said, given a situation in which tallow with >0.15% insoluble impurities (and containing BSE agents) was used as a feedstock to produce biodiesel, and the said limitations of CI engine combustion to destroy BSE infectivity, it is not possible to rule out exposure to airborne BSE.

7.3.2 Conclusions and uncertainties

The risk of BSE infection resulting from the use of SRM-derived tallow as a feedstock for biodiesel may be divided into two scenarios based on this review. In the first, the risk is considered negligible providing that SRM and any downstream products (i.e., tallow destined for biodiesel production) are processed to achieve a tallow purity standard of not more than 0.15% insoluble impurity content. This standard is based on the World Organization of Animal Health's (OIE) health standard for tallow and is enforced by the CFIA via the Federal Health of Animals and Feeds Acts.

In a second scenario in which the insoluble content of the SRM-derived tallow exceeds 0.15% and contains BSE agents, it is expected that the various processes from rendering through combustion will contribute to a reduction in the risk of inhalation exposure to BSE agents as result of the combustion of biodiesel. However, it is difficult to establish a negligible risk of infection given the uncertainty that exists. During rendering, the vast majority of proteinaceous material, including the BSE agent (if present) partitions into the MBM fraction and not in the tallow. In terms of biodiesel production, it is expected that strong acids and elevated temperatures during an acid catalyzed pre-treatment stage, as well as biodiesel purification, will contribute to a reduction in BSE infectivity. However, there is a lack of empirical information to quantify this. In addition, little is known about the role that transesterification reactions play in the inactivation of BSE agents. Finally, limited evidence reported by Cummins et al. (2002) supports the view that BSE infectivity through inhalation exposure resulting from the combustion of SRM-derived tallow is unlikely. However, there exists some uncertainty as to whether compression ignition engine temperatures are high enough for a sufficient length of time to render BSE agents inactive.

7.4 Biodiesel and Allergenicity

In North America, the most common feedstocks for the production of biodiesel include plant oils (soy, canola), animal fat (tallow) and recycled greases. Given that soy is one of the main food categories responsible for allergic reactions, the potential for allergic reactions in the general population following inhalation exposure to exhaust from soy-based biodiesel was examined. Dermal exposure to biodiesel fuel was not considered because systemic reactions resulting from

topical exposure to food allergens are considered rare. Normally, dermal exposure to food allergens leads to isolated, local skin reactions (Metcalfe et al. 2003).

The exact prevalence of soy allergy in the general population is unknown and likely depends on local eating habits and exposure; however, a prevalence rate of 0.3–0.4% has been reported (Becker et al. 2004). Estimates of threshold levels of soy intake triggering an adverse reaction in soy-allergic individuals vary widely, ranging from 0.0013–500 mg (L'Hocine et al. 2007). Severe reactions to soy are rare compared to reactions to peanuts, tree nuts, fish and shell fish, although deaths from eating soy have been reported (L'Hocine et al. 2007).

From the majority of existing reports, it appears that the ingestion of highly refined soybean oil may not represent a risk of provoking allergic reactions in most susceptible people (L'Hocine et al. 2007). However, oils destined for biodiesel production are not necessarily refined. Available information suggests that unrefined plant oil is more likely to contain proteins and potentially allergenic proteins (Martin-Hernandez et al. 2005).

With regards to biodiesel production, purification processes, if they occur, are likely to reduce protein levels in the finished product (ATF Can 2005). In addition, denaturation (unfolding) and hydrolysis of proteins may occur during transesterification reactions depending on the conditions, i.e., temperature and pH. The extent to which these processes occur will impact potential allergenicity of the final product.

In the event that allergenic proteins were able to survive the biodiesel production process, it is highly probable that the proteins would be destroyed as a result of the combustion process given that temperatures in diesel engines range from 500^{o}C to over 2000^{o}C (Hountalas et al. 2001). While there is no information available on the effect of these temperatures on allergenic proteins, information from the food industry indicates that significant alterations in protein structure occur at significantly lower temperatures. It has been reported that loss of secondary structure of proteins (55–70^{o}C), cleavage of disulphide bonds (70–80^{o}C), formation of new intra or inter molecular interactions, rearrangements of disulphide bonds (80–90^{o}C) and the formation of aggregates (90–100^{o}C) follow destruction of the tertiary structure. These modifications eventually result in loss of organized structure and denatured protein (Wal 2003 *in* Wilson et al. 2008). Thus, any proteins existing in the fuel are not expected to survive the thermal environment of the combustion process.

In conclusion, the potential for allergic reactions in the general population following inhalation exposure to exhaust from soy-based biodiesel is unlikely given the probability that proteins present in plant oil and biodiesel would ultimately be destroyed during the combustion process.

7.5 Hazard Assessment – Biodiesel Additives

This section presents key background and toxicity information with regards to major fuel additive categories (biocides, NO_X reducers, antioxidants, and cold-flow improvers) that are likely to be used in biodiesel fuels in Canada.[70] Other existing fuel additive categories less widely used were not included in this assessment. Uncertainties and data gaps surrounding fuel additives are also discussed. Additives that are reviewed in this chapter were selected based on input from the California Air Resources Board (CARB) biodiesel multimedia assessment, collaborators at Health Canada's Environmental Health Science and Research Bureau, and industry. They represent either fuel additives currently in use or most likely to be used in a Canadian context.

Potential exposure scenarios for additives used in biodiesel include inhalation (volatilization, combustion emissions), dermal contact (e.g., fuel dispensing), and ingestion (e.g., water or soil contamination).

7.5.1 Biocides

Biocides are used to limit the growth of fungi and bacteria that can develop at the fuel-water interface. The growth of micro-organisms can alter fuel quality and impede engine performance (FTA 2007; CARB 2008). Biocides are selected based on their efficiency under a variety of conditions, their broad spectrum of action and their ability to function with minimal dose rates and treatments. It is expected that biocides, traditionally used in diesel fuels, will work equally well with biodiesel (NREL 2009). A common commercial fuel biocide is Kathon FP1.5.

Kathon FP1.5 is an industrial biocide formulation in which the active ingredients are 5-chloro-2-methyl-3(2H)-isothiazolone (CMIT) and 2-methyl-3(2H)-isothiazolone (MIT), which comprise 1–1.3% and 0.3–0.4%, respectively, of the product.[71] One of the proposed mechanisms of action of CMIT is a two-step process that begins with inhibition of cell growth and is followed by the production of free radicals leading to cell death (Williams 2007). To control biological growth in fuel storage tanks, the proposed curative dose for Kathon FP1.5 is 204–522 ppm (1.6–7.8 ppm CMIT) while the maintenance dose is 52–313 ppm (0.8–4.7 ppm CMIT) (Rohm and Haas 2007). In Canada, Kathon FP1.5 is registered with the Pesticide Management Regulatory Agency (PMRA) under the *Pest Control Products Act* (PCPA) for use in different liquid hydrocarbon fuels and oils.

7.5.1.1 CMIT/MIT

CMIT and MIT were first registered in Canada in 1978. There are 92 products registered with the PMRA that contain CMIT/MIT as an active ingredient. The PMRA conducted a re-evaluation of CMIT and MIT in 2004 and determined that, based on the US EPA re-evaluation

[70] The inclusion of any commercial additive in this assessment does not represent an endorsement of the additive by Health Canada.

[71] CMIT and MIT are commonly referred to as IST and ISL, respectively, in the PMRA re-evaluation document (2004).

of CMIT and MIT and taking into consideration Canadian use patterns and issues, CMIT and MIT are acceptable for continued registration provided that mitigation and precautionary measures are adopted (PMRA 2004). CMIT/MIT is not harmful to the environment or human health (i.e., inherently toxic and greatest potential for human exposure) according to the Canadian Environmental Protection Act (CEPA) categorization criteria for substances on the Domestic Substances List (DSL) (Environment Canada 2010).[72] The US EPA also concluded that methylisothiazolinone does not pose unreasonable risks or adverse effects to humans or the environment and that CMIT/MIT may be classified as a group D carcinogen (not classifiable as to human carcinogenicity) (US EPA 1998a).

CMIT/MIT was not found to be foetotoxic, embryotoxic, or teratogenic based on exposure in pregnant rats (Weatherholtz et al. 1980 and Thomas et al. 1992 *in* US EPA 1998a).

7.5.2 NO_X Reducers

Increasing the cetane number of a fuel is expected to result in decreased NO_X emissions.[73] Hence, so-called NO_X reducers are added to biodiesel fuels to enhance the cetane value. The majority of NO_X reducers are alkyl nitrates (CARB 2008), such as ORYXE LED+5510.[74] The active ingredient in ORYXE LED+5510 is ethyl-hexyl nitrate (2-EHN), which comprises 45% w/w of the final product (CARB 2008). ORYXE LED+5510 has not been evaluated in Canada and there is no toxicological data available for this additive. However, health and toxicological information for the active ingredient 2-EHN exist.

7.5.2.1 2-EHN

2-EHN does not meet the human health categorization criteria for substances on the DSL (Environment Canada 2010). NOAELs and LOAELs (lowest observable adverse effect levels) were determined in rats and rabbits using different test protocols (e.g., acute, sub-acute, developmental) and administration methods (e.g., oral, inhalation, dermal), but no human data is available (American Chemistry Council 2006; European Chemical Industry Council 2004; Poon 2010, 2011a, 2011b).

In a developmental toxicity study, male and female rats were exposed to 2-EHN (prior to mating, during mating and gestation, and until 5 days post-partum) by gavage at dose levels of 20, 100, or 500 mg/kg/day. The NOAEL for parental toxicity was determined to be 20 mg/kg/day and the NOAEL for toxic effect on reproductive performance of the progeny was 100 mg/kg/day (American Chemistry Council 2006).

[72] As defined in section 73 of CEPA 1999

[73] However, small increases in NO_X occur when using a biodiesel blend compared to diesel. Increases in NO_X are not determined by a single fuel property but are a result of a number of coupled mechanisms (Mueller et al. 2009). [See Chapter 4]

[74] ORYXE LED+5510 is included in CARB's Tier 1 Biodiesel Multimedia Assessment (CARB 2008). There are 12 separate ORYXE additives registered with the US EPA (Jim Caldwell, personal communication 2009).

2-EHN was not mutagenic, with or without metabolic activation in the Ames test, and was also negative for the induction of chromosome aberrations at dose levels that induced acceptable levels of toxicity in the *in vitro* chromosomal aberration assay (American Chemistry Council 2006).

7.5.3 Antioxidants

Synthetic antioxidants are added to fuels to improve their stability by slowing down the oxidation and polymerization processes and preventing the formation of insoluble compounds that can block fuel filters and nozzles (FTA 2007). Dinkov et al. (2009) suggest that antioxidants act by inhibiting the formation of secondary oxidation products in biodiesel fuels, such as insoluble compounds, which have a negative impact on density, viscosity and total acid number. t*ert*-Butylhydroquinone (TBHQ) is a synthetic antioxidant that has been shown to enhance the storage stability of biodiesel (Tang et al. 2008; NREL 2005) and is the active ingredient of BioExtend-30.[75]

BioExtend-30 is a commercial antioxidant formulated specifically for biodiesel containing 20% v/v of TBHQ. There is no information available pertaining to the use or toxicity of BioExtend-30 in Canada, but all the components of BioExtend-30 (i.e., TBHQ, butyl acetate, citric acid, and diethylene glycol monobutyl ether) are regulated under CEPA and can be found on the DSL. In the US, the components are all listed on the Toxic Substances Control Act inventory (Eastman 2008). Toxicological information for the active ingredient TBHQ is presented.

7.5.3.1 TBHQ

TBHQ does not meet the human health categorization criteria for substances on the DSL (Environment Canada 2010). Studies in rat and mouse animal models have led to mixed results with respect to carcinogenicity (Eastman 2003; Altmann et al. 1985, Hirose et al. 1993 and Kawabe et al. 1994 *in* Ghavari et al. 2007) and genotoxicity or mutagenicity (Eastman 2003; Ghavari et al. 2007). TBHQ exposure did not result in teratogenic effects when given to pregnant rats at concentrations as high as 0.5% in their diet (Eastman 2003).

7.5.4 Cold Flow Improvers

At cold temperatures biodiesel fuels can gel, resulting in equipment malfunctions (e.g., clogged filters and fuel dispensers). The behaviour of biodiesel in cold climates can vary based on the fatty acid profile and the amount of impurities in the fuel (CARB 2008; NREL 2009). Cold-flow improvers can be added to biodiesel to help prevent the formation of wax crystals at low temperatures. Most cold-flow improvers are ethylene or vinyl acetate co-polymers, but kerosene has also been used in the past to treat cold flow issues (FTA 2007; CARB 2008). There are no cold flow improvers currently available for use in Canada that can be properly defined as

[75] BioExtend-30 is included in CARB's Tier I Biodiesel Multimedia Assessment (CARB 2008) and recommended for use in the Tier II Health Risk Analysis (CARB 2009).

additives.[76] Research in the area of cold-flow improvers is currently underway and is proprietary.[77]

7.5.5 Key Uncertainties and Data Gaps

Given the evolving nature of the biodiesel industry, no definitive types of additives have been selected for use in Canada. Since fuel additives include a variety of compounds used for specific purposes, either regularly or sporadically, it is not possible to predict which fuel additives will be used in different geographical locations across Canada and according to seasonal variations. The proprietary nature of additives and on-going developments also add to the difficulty of assessing the health impacts of novel commercial fuel additive products. In addition, fuel additives currently used in conventional fuels may be effective in biodiesel fuel blends, which will likely influence the selection of fuel additives in Canada.

In general, there is a lack of data and studies regarding the health effects of whole additives in combination with biodiesel blends derived from different feedstocks. For example, the toxicological data that was available for biocides came from material safety data sheets produced by Rohm and Haas (2006, 2007) and from the U.S. EPA (1998a) for methylisothiazolinone, but the studies from which toxicity values were derived were not publically available. Also, the US EPA conclusions for CMIT/MIT were determined based on the biocide Kathon 886, which differs slightly in composition from Kathon FP1.5.

As for NO_x reducers, their use, efficiency, and the selection of a specific product are difficult to forecast. It appears that additional studies are required to identify the most effective additives under Canadian conditions (e.g., climate, feedstocks, vehicle/engine technologies) for which toxicological data can be developed.

The lack of comparative data regarding the potential health impacts resulting from inhalation, dermal or oral exposures to different additives in biodiesel blends is particularly important. Recent oral toxicity studies conducted by Health Canada have examined the health effects resulting from exposure to individual additives (e.g., CMIT, 2-EHN, TBHQ); however, the results are preliminary at this point in time.

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[76] Kerosene can be used for seasonal regional cloud point specifications (personal communication, Stuart Porter, BBI Biofuels, January 8, 2010). However, it has been tested at such high levels in biodiesel blends (21%-43% for B2) that it is not considered an additive, but more appropriately a fuel component (ARDD 2009).

[77] Ken Mitchell of Shell Canada Limited - personal communication, 2010.

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Chapter 8. Health Impacts of Biodiesel Use

The assessment of the health impacts of introducing a new or modified motor vehicle fuel requires several sequential steps: estimating changes in vehicle emissions and in the overall emissions inventory associated with the fuel's introduction; estimating the impact of these emissions changes on air quality; and estimating the health implications of the associated air quality changes. This chapter addresses the latter step.

The effects of fuel use on emissions, air quality and, subsequently, health, can vary spatially and temporally, due to the geographic distribution of populations, population growth, variability in meteorological parameters, changes to the vehicle fleet over time, and changes in overall emissions over time. The analysis of health impacts of a new or modified fuel requires an approach that accounts for the dynamic nature of these variables.

A quantitative estimate of the incremental health impacts due to biodiesel use is presented for impacts associated with exposure to the following criteria air contaminants (because robust methodologies exist to do so): $PM_{2.5}$, O_3, CO, NO_2 and SO_2. Conversely, the potential health implications of changes in emissions of mobile source air toxics and PM characteristics (e.g., particle size and number) are discussed qualitatively, due to uncertainties in the understanding of these issues and insufficient data with which to make quantitative estimates.

8.1 Quantitative Estimates of Health Impacts

Diesel-powered vehicles are a source of air pollutant emissions, particularly in urban areas where vehicle populations are high. The health impacts of $PM_{2.5}$, O_3, CO, NO_2 and SO_2, which are either emitted directly and/or formed secondarily in the atmosphere from diesel combustion and other emission sources, are well documented in the scientific literature (see Section 7.1). It is recognized that there is no exposure threshold for many of these health effects, and that these pollutants are responsible for major population health impacts in Canada and elsewhere, including cardiorespiratory mortality, hospital admissions and emergency room visits. For example, analyses have estimated that air pollutant exposure was associated with approximately 5,900 mortalities in eight Canadian urban centres in 2000 (Judek et al. 2004), and 21,000 mortalities, 11,000 hospital admissions and 92,000 emergency department visits across Canada in 2008 (CMA 2008).

Potential population health impacts associated with changes in criteria air contaminants due to biodiesel use were quantified. It should be noted that changes in air quality due to biodiesel production, transport and storage were not included due to a lack of appropriate data. For the purposes of this analysis, the incremental health risks and benefits across the Canadian population associated with the national use of either B5 (year-round) or B20 (summertime only)

as compared to the use of ultra-low sulphur diesel (ULSD) in 2006 and 2020 were evaluated using the Air Quality Benefits Assessment Tool.

8.1.1 Methodology

The Air Quality Benefits Assessment Tool (AQBAT) is a computer simulation tool developed by Health Canada to estimate the human health and welfare benefits or damages associated with changes in the ambient concentrations of criteria air contaminants (CACs) in Canada. It is a national model, with all inputs specific to the geographic areas of Canada and calculations are performed on an annual basis. The basic approach of AQBAT is to estimate changes in the average per capita risk for specific health outcomes that would occur across the population in all geographic areas of Canada due to specified incremental changes in ambient concentrations of a select list of criteria air contaminants, to translate these health risk changes into economic terms, and to then aggregate the net population health and economic impacts by region. For this analysis, the incremental changes in air pollutant concentrations assessed are equal to the differences between pollutant levels associated with ULSD and biodiesel use scenarios.

8.1.1.1 AQBAT scenario runs

National Scale Model Runs

For this analysis, AQBAT was run comparing the biodiesel use scenarios (either B5 or B20) with that of the base-case (B0), for both 2006 and 2020. The assumptions used for these scenarios are presented in detail in Chapters 5 and 6. In brief, they include:

- ultra low sulphur diesel (ULSD) as the baseline fuel;
- canola considered as the single feedstock for biodiesel production;
- biodiesel use confined to on-road heavy-duty diesel vehicles (HDDVs) across Canada;
- no effect on emissions from the use of biodiesel in light-duty diesel vehicles or in 2010 or later model year HDDVs;
- B5 used year-round and B20 use confined to summer months (May–September); and
- Emissions associated with biodiesel production, storage and transport are not included in the modelling due to insufficient data.

Each AQBAT run compares two scenarios for which the air pollutant concentrations differ, and the population health impacts associated with the difference in pollutant concentrations are estimated for individual geographic areas. For this report, four distinct runs were conducted at the national level for a one year period, as listed in Table 8-1. For each of these scenarios, air pollutant concentration data for each census division in Canada were generated by AURAMS (A Unified Regional Air-quality Modelling System) as discussed in Section 6.2. These data served as input to AQBAT. The B5 and B20 scenarios are considered distinct from one another i.e. their results are presented separately and should not be combined.

Table 8-1 AQBAT biodiesel runs

Run #	Year	Baseline scenario	Biodiesel scenario	Geographic area
1	2006	B0 (ULSD)	B5	Canada
2	2006	B0 (ULSD)	B20	Canada
3	2020	B0 (ULSD)	B5	Canada
4	2020	B0 (ULSD)	B20	Canada

Urban (Montréal) Model Runs

In addition, an analysis was undertaken of the health impacts of B5 or B20 use in Montréal in 2006 and 2020 using high-resolution emissions and atmospheric modelling output data, as described in Section 6.3 of this report. The runs specific to the urban scale modelling are listed in Table 8-2. Population-weighted concentration results from AURAMS modelling were used in the Montréal analysis (see Section 6.3.1.4). It is important to note that the high-resolution modelling was conducted for a 12-day period in June that included an episode of high air pollution levels. Because these runs were based on a short time frame, a limited population and minor air quality changes due to biodiesel use, the health impacts are expected to be very small. Moreover, the meteorological and atmospheric conditions of an air pollution episode may not be typical of a longer time frame (e.g., summer season) and therefore the results have not been extrapolated over a longer period.

In order to provide context for the interpretation of the small health impacts observed in the Montréal analysis, the percent change in health outcomes due to biodiesel use relative to all health impacts due to *above-background air pollutant concentrations* was calculated. For example, for the B5 scenario, the percent change in health impacts (%ΔHI) was calculated as follows:

$$\%\Delta HI = \frac{\text{incremental health impacts due to B5 vs. B0}}{\text{B0 health impacts due to above-background air pollution}} \times 100$$

Background air pollutant concentrations were previously determined by Judek et al. (2004). They are defined as the concentration of a pollutant that would occur if there were no North American anthropogenic sources of the pollutant and its precursors, i.e., it is assumed that only natural sources within North America and inflow from outside of North America contribute to them. Long-term monitoring data from remote locations were used in their derivation. For the Montréal analysis, the following background concentrations were assumed: $PM_{2.5}$ = 1.80 µg/m^3; O_3 =33.00 ppb; NO_2 = 0.150 ppb; SO_2 = 0.020 ppb; CO (1 hour) = 0.130 ppm; and CO (24 hour) = 0.120 ppm. These represent background concentrations specific to the month of June for O_3, SO_2 and CO, and annual average background concentrations for $PM_{2.5}$ and NO_2. AQBAT was used to estimate the B0 health impacts associated with above-background air pollution for use in these calculations.

Table 8-2 Urban AQBAT biodiesel runs for 12-day period

Run #	year	Baseline scenario	Biodiesel scenario	Geographic area
5	2006	B0 (ULSD)	B5	Montréal area
6	2006	B0 (ULSD)	B20	Montréal area
7	2020	B0 (ULSD)	B5	Montréal area
8	2020	B0 (ULSD)	B20	Montréal area

8.1.1.2 AQBAT inputs: pollutant concentration changes

National Scale Model Runs

Criteria air contaminant (CAC) concentration outputs from photochemical modelling of the six Canada-wide scenarios with AURAMS (i.e., B0, B5 and B20 in 2006 and 2020, as presented in Chapter 6) were used as data inputs for the national scale AQBAT runs. Photochemical modelling was conducted for the entire country at a resolution of 22.5 km x 22.5 km. Pollutant concentration results for individual grid cells were area-weighted for each Census Division (CD) defined by the 2006 Census of Canada, as described in Section 6.2.1.6. These CD-specific concentrations for each pollutant were used as input to AQBAT for the national scale model runs.

Urban (Montréal) Model Runs

High resolution photochemical modelling (AURAMS) results for the Montréal domain (see Section 6.3) for the six modelling scenarios (i.e., B0, B5 and B20 in 2006 and 2020, as presented in Chapter 6) were used as the CAC concentration data inputs for additional AQBAT analyses (Table 8-2). This analysis was undertaken because the high-resolution photochemical and emissions modelling upon which it is based better represent the spatial distribution of motor vehicle emissions and their proximity to human populations.

Grid cell (3 km x 3 km) pollutant concentrations were area-weighted for each CD in the domain, as described in Section 6.3.1.4, for a total of 19 CDs. In addition, each CD was assigned population-weighted concentrations of each pollutant, which reflect the average concentrations where people live. The high-resolution urban modelling was done for a 12-day period in June that included high smog conditions, and therefore is not comparable to the annual estimates from the lower-resolution modelling.

8.1.1.3 AQBAT inputs: population of Canada

AQBAT contains files of both historical and projected Canadian population data. For the 2006 biodiesel scenario, the population data were based on the 2006 Census of Canada CD level population counts (288 CDs of varying geographical and population size). The populations for the 2020 biodiesel scenario were based on population projections prepared by Statistics Canada (2005) by age and province/territory for the years 2005–2031, and applied to each of the 288 CDs.

8.1.1.4 AQBAT inputs: health outcomes and concentration response functions

The AQBAT model includes health impact information for several air pollutants in the form of concentration response functions (CRFs). Specifically, a CRF is a quantitative representation of the impact of a given air pollutant on the average per capita risk for a specific health outcome. None of the health outcomes included have a threshold for effect, i.e., the scientific evidence indicates that effects occur at all levels of exposure. Therefore, health impacts are defined as excess health risk per unit increase in ambient pollutant concentration (e.g., per 1 $\mu g/m^3$).

The CRFs employed in AQBAT are derived from published peer-reviewed scientific analyses of data pertaining to Canadian and other populations, and may be based on a single study or on pooled results from multiple studies. There is some uncertainty inherent in the CRFs, which are input as a distribution function in AQBAT. Based on these probability distributions, AQBAT generates a central "best guess" estimate of the most likely health impacts, as well as low and high end estimates. However, given the relatively small change in ambient air quality between the scenarios assessed in this analysis (see Chapter 6), health impacts are expected to be fairly small. Therefore, this report focuses on the central estimates of the results. In addition, AQBAT includes age-specific baseline incidence rates for each health outcome in the target population, in order to estimate the number of excess health outcomes associated with the increased risk due to the change in air pollutant concentration.

The pollutants and the associated health effects considered in this analysis are provided in Table 8-3. Each of the health effects is the result of either a *short-term* or *long-term exposure*. These health endpoints, their acute or chronic nature, the associated CRFs, and the population group(s) to which they apply are pre-defined within AQBAT, and represent Health Canada endorsed values drawn from the medical literature. It should be noted that although AQBAT includes a CRF for acute exposure mortality associated with NO_2 exposure, it is not assumed to reflect a causal relationship. Rather, NO_2 may be acting as a surrogate for a specific component of the air pollution mixture such as fresh combustion emissions.

Similar health outcomes are aggregated for the presentation of results. Specifically, all premature mortality, all hospital admissions, all emergency room visits and all restricted activity days are reported in aggregated form. It should be noted that although additional health endpoints have been studied in the literature and found to be associated with exposure to air pollutants, they have not been assessed quantitatively and incorporated into the framework of AQBAT.

Table 8-3 CACs and their associated health endpoints with CRFs in AQBAT

Pollutant*	Averaging period	Health Endpoint
Ozone (O_3)	1-hour maximum	Acute exposure mortality
ozone (summer May–September only)	1-hour maximum	Chronic exposure respiratory mortality
		Respiratory hospital admissions
		Respiratory emergency room visits
		Acute respiratory symptom days
		Asthma symptom days
		Minor restricted activity days
Fine particulate matter ($PM_{2.5}$)	24-hour	Chronic exposure respiratory mortality
		Chronic exposure cerebrovascular mortality
		Chronic exposure ischemic heart disease mortality
		Chronic exposure lung cancer mortality
		Respiratory hospital admissions
		Respiratory emergency room visits
		Cardiac hospital admissions
		Cardiac emergency room visits
		Adult chronic bronchitis cases
		Child acute bronchitis episodes
		Asthma symptom days
		Acute respiratory symptom days
		Restricted activity days
Carbon monoxide (CO)	1-hour maximum	Elderly cardiac hospital admissions
	24-hour	Acute exposure mortality
Nitrogen dioxide (NO_2)[#]	24-hour	Acute exposure mortality
Sulphur dioxide (SO_2)	24-hour	Acute exposure mortality

[*] Unless otherwise specified, the CRFs apply to exposure to the pollutant at any time during the year.

[#]Although a CRF for acute exposure mortality associated with NO_2 exposure is a component of AQBAT, NO_2 is generally interpreted to be acting as a surrogate for combustion, and may not be causally linked to this outcome.

8.1.1.5 AQBAT inputs: economic valuation estimates

AQBAT includes economic valuation estimates for the health outcomes assessed by the model. These estimates consider the potential social, economic, and public welfare consequences of the health outcomes, including medical costs, reduced workplace productivity, pain and suffering, and the impacts of reduced mortality risk. The sum of the valuation estimates provides an indication of the relative social benefit, or value that results from reduced risks to health. Because of uncertainty in the economic valuation estimates, they are entered as a distribution in AQBAT. Results associated with the mean estimates from the Monte Carlo simulations are reported here and all results are in 2009 dollars. Note that AQBAT valuation estimates for future years are not inflated based on projected income growth and do not account for mortality lag in valuation of premature mortality. In the case of the latter, the CRFs are assumed to represent a

steady state situation, and to be appropriate for the estimation of risks and associated welfare impacts over the longer term.

It should be noted that valuation estimates for emergency department visits associated with $PM_{2.5}$ and O_3 exposure take into account the probability and valuation of subsequent admission to hospital. For instance, if 70% of emergency department visits for cardiac disease result in a hospital admission, the valuation of emergency department visits for these conditions is weighted accordingly to reflect this probability of admission and associated increased costs. Because the model already includes the value associated with the risk of hospitalisation as part of the estimated economic impacts of emergency room visits, the model outputs a zero value for hospital admissions themselves, in order to avoid double counting of the impacts. In addition, because of the aggregation of similar health endpoints that may have different costs (e.g., respiratory and cardiac emergency room visits) in the presentation of results, the average cost per emergency room visit (or other aggregated health outcomes) may differ for the different scenarios.

8.1.2 National estimates of health impacts of biodiesel use

Results of the AQBAT analyses for the national biodiesel use scenarios for 2006 and 2020 are presented in Tables 8-4 to 8-7. Specifically, mean incremental health risks are presented for individual air pollutants modeled, representing the health costs/benefits of the use of biodiesel compared to those for petroleum diesel. Monetary values associated with these risks/benefits are largely driven by the associated mortalities. The 2.5^{th} percentile and 97.5^{th} percentile estimates of the health counts (aggregated across pollutants) are provided for comparison, reflecting the fact that the CRFs contain inherent uncertainty and are in fact distributions rather than single values.

8.1.2.1 Results for 2006

National use of B5 by the on-road heavy-duty diesel vehicle fleet is associated with a net reduction of approximately five premature mortalities annually due to decreased mortality risk associated with exposure to $PM_{2.5}$ and O_3, and despite a small increase in premature mortality associated with increased NO_2 exposure (Table 8-4). B5 use is also associated with minimal reductions in hospital admissions and emergency room visits, along with reductions in acute respiratory symptom days, asthma symptom days, restricted activity days, child acute bronchitis episodes and adult chronic bronchitis cases. The total economic value of the avoided health outcomes is approximately $33,000,000, due largely to the avoided premature mortalities.

The national use of B20 during the May–September period of 2006 by the on-road heavy-duty diesel vehicle fleet is estimated to result in a net benefit of approximately seven fewer premature mortalities (Table 8-5). Four and five avoided mortalities are associated with decreased $PM_{2.5}$ and O_3 exposures, respectively, while about three additional premature mortalities are attributed to increases in NO_2 exposure. In addition, reductions in $PM_{2.5}$ and O_3 exposure are associated with reductions in hospital admissions, emergency room visits, acute respiratory symptom days,

asthma symptom days, restricted activity days, child acute bronchitis episodes, and adult chronic bronchitis cases. The net economic value of the changes in health outcomes in 2006 is approximately $48,000,000.

8.1.2.2 Projection for 2020

The emission benefits of biodiesel fuel use are expected to diminish with time due to turnover of the on-road heavy-duty diesel vehicle fleet and the introduction of cleaner vehicles. Projection of the impacts of annual B5 use in the Canadian on-road heavy-duty vehicle fleet in 2020 reveals a reduction of benefits compared to the 2006 estimates (Table 8-6). The net impact on premature mortality is estimated to be an increase of less than one death, due to negligible increases in exposure to $PM_{2.5}$ and NO_2. There are similarly very small morbidity impacts, resulting in a net economic cost of about $4,000,000 for 2020.

National use of B20 by the on-road heavy-duty vehicle fleet in 2020 (May to September) is estimated to result in a net risk of two additional premature mortalities, due to localized minimal increases in $PM_{2.5}$, O_3 and NO_2 exposures (Table 8-7). Minor increases in morbidity outcomes related to increased $PM_{2.5}$ and O_3 exposures are also projected. The reasons for increased air pollutant concentrations in this scenario relate to secondary formation of $PM_{2.5}$ in the atmosphere and conditions of ozone formation (see Sections 6.2.3 and 6.2.4). This scenario is associated with a net cost in 2020 of approximately $15,000,000. However, the pollutant concentration changes underlying these effects are believed to be close to the AURAMS model detection limit.

Table 8-4 Mean incremental health outcomes (and cost, in 2009 dollars)[@] associated with B5 use in on-road HDDVs in 2006, Canada

Pollutant	Premature mortality	Hospital admissions	Emergency room visits	Acute respiratory symptom days	Asthma symptom days	Restricted activity days	Child acute bronchitis episodes	Adult chronic bronchitis cases	Sum of costs
$PM_{2.5}$	-4 (-$26,174,000)	-<1 (*)	-2 (-$8,000)	-13,263 (-$135,000)	-239 (-$16,000)	-5446 (-$331,000)	-34 (-$13,000)	-5 (-$1,899,000)	-$28,576,000
O_3	-2 (-$13,636,000)	-<1 (*)	-3 (-$6,000)	-5,357 (-$70,000)	-719 (-$47,000)	-1226 (-$34,000)	-	-	-$13,795,000
CO	-<1 (-$118,000)	-<1 (-$3,000)	-	-	-	-	-	-	-$121,000
NO_2	1 ($9,526,000)	-	-	-	-	-	-	-	$9,526,000
SO_2	<1 ($45,000)	-	-	-	-	-	-	-	$45,000
All pollutants combined	-5 [-2/-8][#] (-$30,357,000)	-2 [-1/-2] (-$3,000)	-5 [-3/-7] (-$15,000)	-18,620 [-5,132/-32,935] (-$205,000)	-959 [-385/-1,556] (-$63,000)	-6,672 [-3,203/-12,969] (-$365,000)	-34 [0/-73] (-$13,000)	-5 [0/-10] (-$1,899,000)	-$32,920,000

[@] Counts of health outcomes are rounded to the nearest integer and values between 0 and 1 are represented as <1. Valuation estimates are rounded to the nearest $1,000, and values between $0 and $1,000 are represented as <$1,000. As a result, column and row totals may not exactly match the values provided.

* Valuation estimates for emergency department visits associated with $PM_{2.5}$ and O_3 take into account the probability and valuation of subsequent admission to hospital, therefore no cost is presented for $PM_{2.5}$ and O_3 hospital admissions alone.

[2.5^{th}%/97.5^{th}%] provides the 2.5^{th} and 97.5^{th} percentile count estimates for the health outcome aggregated across pollutants

Table 8-5 Mean incremental health outcomes (and cost, in 2009 dollars) [@] associated with B20 use (May–September) in on-road HDDVs in 2006, Canada

Pollutant	Premature mortality	Hospital admissions	Emergency room visits	Acute respiratory symptom days	Asthma symptom days	Restricted activity days	Child acute bronchitis episodes	Adult chronic bronchitis cases	Sum of costs
$PM_{2.5}$	-4 (-$28,969,000)	-<1 (*)	-3 (-$9,000)	-14,426 (-$147,000)	-260 (-$17,000)	-5,977 (-$364,000)	-35 (-$14,000)	-5 (-$2,086,000)	-$31,607,000
O_3	-5 (-$33,137,000)	-2 (*)	-10 (-$25,000)	-20,988 (-$275,000)	-2,818 (-$184,000)	-4,803 (-$135,000)	-	-	-$33,757,000
CO	-<1 (-$171,000)	-<1 (-$4,000)	-	-	-	-	-	-	-$175,000
NO_2	3 ($17,116,000)	-	-	-	-	-	-	-	$17,116,000
SO_2	0 ($0)	-	-	-	-	-	-	-	$0
All pollutants combined	-7 [-3/ -11][#] (-$45,160,000)	-3 [-2/ -5] (-$4,000)	-13 [-4/ -22] (-$34,000)	-35,414 [-11,641/ -60,261] (-$422,000)	-3,078 [-1,043/ -5,071] (-$201,000)	-10,780 [-3,517/-20,700] (-$499,000)	-35 [0/ -77] (-$14,000)	-5 [0/ -11] (-$2,086,000)	-$48,421,000

[@] Counts of health outcomes are rounded to the nearest integer and values between 0 and 1 are represented as <1. Valuation estimates are rounded to the nearest $1,000, and values between $0 and $1,000 are represented as <$1,000. As a result, column and row totals may not exactly match the values provided.

* Valuation estimates for emergency department visits associated with $PM_{2.5}$ and O_3 take into account the probability and valuation of subsequent admission to hospital, therefore no cost is presented for $PM_{2.5}$ and O_3 hospital admissions alone.

[2.5^{th}%/97.5^{th}%] provides the 2.5^{th} and 97.5^{th} percentile count estimates for the health outcome aggregated across pollutants

Table 8-6 Mean incremental health outcomes (and cost, in 2009 dollars) [@] associated with B5 use in on-road HDDVs in 2020, Canada

Pollutant	Premature mortality	Hospital admissions	Emergency room visits	Acute respiratory symptom days	Asthma symptom days	Restricted activity days	Child acute bronchitis episodes	Adult chronic bronchitis cases	Sum of costs
$PM_{2.5}$	<1 ($1,554,000)	<1 (*)	<1 (<$1,000)	794 ($8,000)	14 (<$1,000)	334 ($20,000)	2 (<$1,000)	<1 ($119,000)	$1,703,000
O_3	<1 ($137,000)	<1 (*)	<1 ($1,000)	995 ($13,000)	134 ($9,000)	228 ($6,000)	-	-	$166,000
CO	-<1 (-$18,000)	-<1 (-<$1,000)	-	-	-	-	-	-	-$18,000
NO_2	<1 ($2,182,000)	-	-	-	-	-	-	-	$2,182,000
SO_2	-<1 (-$54,000)	-	-	-	-	-	-	-	$-54,000
All pollutants combined	<1 [<1/<1] [#] ($3,801,000)	<1 [<1/-<1] (-<$1,000)	<1 [1/<1] ($2,000)	1790 [3,029/591] ($21,000)	148 [243/52] ($10,000)	562 [1,456/197] ($27,000)	2 [4/0] (<$1,000)	<1 [<1/0] ($119,000)	$3,979,000

[@] Counts of health outcomes are rounded to the nearest integer and values between 0 and 1 are represented as <1. Valuation estimates are rounded to the nearest $1,000, and values between $0 and $1,000 are represented as <$1,000. As a result, column and row totals may not exactly match the values provided.

* Valuation estimates for emergency department visits associated with $PM_{2.5}$ and O_3 take into account the probability and valuation of subsequent admission to hospital, therefore no cost is presented for $PM_{2.5}$ and O_3 hospital admissions alone.

[2.5^{th}%/97.5^{th}%] provides the 2.5^{th} and 97.5^{th} percentile count estimates for the health outcome aggregated across pollutants

Table 8-7 Mean incremental health outcomes (cost, in 2009 dollars) [@] associated with B20 use (May–September) in on-road HDDVs in 2020, Canada

Pollutant	Premature mortality	Hospital admissions	Emergency room visits	Acute respiratory symptom days	Asthma symptom days	Restricted activity days	Child acute bronchitis episodes	Adult chronic bronchitis cases	Sum of costs
$PM_{2.5}$	<1 ($5,400,000)	<1 (*)	<1 ($2,000)	2,938 ($30,000)	53 ($3,000)	1,227 ($75,000)	7 ($3,000)	1 ($437,000)	$5,949,000
O_3	<1 ($5,819,000)	<1 (*)	2 ($5,000)	3,903 ($52,000)	524 ($34,000)	894 ($25,000)	-	-	$5,934,000
CO	-<1 (-$25,000)	-<1 (-<$1,000)	-	-	-	-	-	-	-$25,000
NO_2	<1 ($3,149,000)	-	-	-	-	-	-	-	$3,149,000
SO_2	<1 (<$1,000)	-	-	-	-	-	-	-	<$1,000
All pollutants combined	2 [3/2][#] ($14,344,000)	<1 [<1/<1] (-<$1,000)	2 [4/<1] ($6,000)	6,841 [11,600/2,247] ($81,000)	577 [955/204] ($38,000)	2,121 [5,589/722] ($99,000)	7 [15/0] ($3,000)	1 [2/0] ($437,000)	 $15,008,000

[@] Counts of health outcomes are rounded to the nearest integer and values between 0 and 1 are represented as <1. Valuation estimates are rounded to the nearest $1,000, and values between $0 and $1,000 are represented as <$1,000. As a result, column and row totals may not exactly match the values provided.

* Valuation estimates for emergency department visits associated with $PM_{2.5}$ and O_3 take into account the probability and valuation of subsequent admission to hospital, therefore no cost is presented for $PM_{2.5}$ and O_3 hospital admissions alone.

[2.5^{th}%/97.5^{th}%] provides the 2.5^{th} and 97.5^{th} percentile count estimates for the health outcome aggregated across pollutants

8.1.3 Urban estimates of health impacts of biodiesel use

High-resolution air quality modelling data were used to estimate health impacts in an urban area, specifically Montréal and its environs. Population-weighted air pollutant concentrations were used for the base-case and biodiesel scenarios (B5 and B20), the calculation of which is described in Section 6.3.1.4. It is important to note that this modelling reflects the impact of the use of biodiesel during a short (high pollution) time period (12 days) and on the population of a single urban area (3,964,938), which represented 12.5% of the Canadian population in 2006. Therefore, health impacts are expected to be accordingly small. Mean incremental health risks associated with the use of biodiesel compared to ULSD were estimated (Table 8-8). To extend the interpretation of these results, the percent change in health outcomes relative to all health impacts due to above-background air pollutant concentrations are also presented.

8.1.3.1 Results for 2006

The overall impact of B5 use by the on-road heavy-duty vehicle fleet in the Montréal area is a decrease in premature mortality and morbidity outcomes in the population, associated with a total monetary benefit of approximately $200,000. Considering all pollutants combined, B5 use would result in a 0.02%–0.04% reduction in all mortality and morbidity associated with above-background base-case air pollution levels.

Widespread use of B20 is associated with a reduction in premature mortality and all morbidity outcomes (except for hospital admissions), the total monetary value of which is about $140,000 for 2006. Relative to the impact of above-background base-case air pollution concentrations, this is equivalent to reductions of 0.01%–0.19% in mortality and morbidity, and an increase of 0.21% in hospital admissions.

8.1.3.2 Projection for 2020

Projecting forward to 2020, the use of B5 by the on-road heavy-duty vehicle fleet in the Montréal area is estimated to produce a minimal increase in premature mortality and morbidity outcomes, with a total cost of approximately $50,000 for that year, due primarily to a very small increase in mortality risk. Combining all pollutants, the percent increase in health outcomes above the base-case is less than 0.01% for all outcomes.

In 2020, the overall impact of B20 use predicted in the Montréal area is an increase in both morbidity and premature mortality, associated with an approximate additional cost of $650,000, which is due largely to mortality estimates for $PM_{2.5}$, O_3 and NO_2. As discussed in Section 6.3.2, the atmospheric modelling predicted increases in $PM_{2.5}$ for the Island of Montréal and in some of the surrounding areas and the reason for this is not clear. Considering the combined pollutants, B20 is predicted to result in a 0.07% increase in premature mortality and a 0.02%–0.42% increase in morbidity outcomes associated with above-background base-case air pollution levels.

Table 8-8 Mean incremental health impacts in Montréal from all pollutants ($PM_{2.5}$, O_3, CO, NO_2, SO_2) associated with B5 or B20 use in on-road HDDVs in 2006 or 2020 (12-day period, population-weighted concentrations), and percent change in health outcomes due to biodiesel use relative to health impacts due to above-background base-case air pollutant concentrations.

Pollutant	Parameter	Premature mortality	Hospital admissions	Emergency room visits	Acute respiratory symptom days	Asthma symptom days	Restricted activity days	Child acute bronchitis episodes	Adult chronic bronchitis cases	Sum of costs
B5 in 2006	**ΔHI BD**	-0.026	-0.014	-0.041	-132.1	-8.1	-45.8	-0.202	-0.031	
	HI ABBC	115.787	46.172	100.778	348,362.1	19,953.6	125,504.6	731.778	96.263	
	% ΔHI	-0.022	-0.030	-0.041	-0.038	-0.041	-0.036	-0.028	-0.032	
	incremental cost (2009 $)*	-$176,000	-<$1,000	-<$1,000	-$1,000	-<$1,000	-$2,000	-<$1,000	-$12,000	-$193,000
B20 in 2006	**ΔHI BD**	-0.013	0.098	-0.179	-490.2	-37.9	-157.5	-0.575	-0.09	
	HI ABBC	115.787	46.172	100.778	348,362.1	19,953.6	125,504.6	731.778	96.263	
	% ΔHI	-0.011	0.212	-0.178	-0.141	-0.190	-0.125	-0.079	-0.093	
	incremental cost (2009 $)	-$89,000	<$1,000	-<$1,000	-$6,000	-$2,000	-$8,000	-<$1,000	-$35,000	-$140,000
B5 in 2020	**ΔHI BD**	0.007	0.002	0.010	23.2	2.6	6.2	0.010	0.002	
	HI ABBC	130.286	48.107	107.073	378,291.9	19,597.8	143,917.9	782.110	117.312	
	% ΔHI	0.005	0.004	0.009	0.006	0.013	0.004	0.001	0.002	
	incremental cost (2009 $)	$47,000	-<$1,000	<$1,000	<$1,000	<$1,000	<$1,000	<$1,000	<$1,000	$49,000
B20 in 2020	**ΔHI BD**	0.094	0.201	0.082	212.0	19.7	63.2	0.172	0.029	
	HI ABBC	130.286	48.107	107.073	378,291.9	19,597.8	143,917.9	782.110	117.312	
	% ΔHI	0.072	0.418	0.077	0.056	0.101	0.044	0.022	0.025	
	incremental cost (2009 $)	$631,000	$1,000	<$1,000	$3,000	$1,000	$3,000	<$1,000	$11,000	$650,000

a: ΔHI BD = incremental health impact due to biodiesel **b**: HI ABBC = health impact due to above-background base-case concentration

c: %ΔHI = percent change in health impact due to biodiesel use relative to health impact due to above-background base-case air pollutant concentration

* Valuation estimates are rounded to the nearest $1,000, and values between $0 and $1,000 are represented as <$1,000. As a result, column and row totals may not exactly match the values provided.

8.2 Qualitative Consideration of Health Risks

In addition to the quantifiable health impacts associated with CAC emissions of biodiesel use by the on-road heavy-duty vehicle fleet detailed in Section 8.1, there are a number of aspects of biodiesel emissions for which potential health risks/benefits cannot be quantified. This may be due to several factors, such as: 1) the emissions testing database not being robust enough to draw firm conclusions regarding the impact of biodiesel use on a specific constituent; 2) MOBILE6.2C not having the capability to address a specific constituent; 3) AURAMS not providing explicit air concentration estimates for a specific constituent; or 4) AQBAT not including concentration response functions for a specific constituent. This section provides a qualitative review of the information for key components in biodiesel exhaust and what can be ascertained regarding potential health risks/benefits.

8.2.1 Mobile source air toxics

Mobile source air toxics are compounds that are emitted to air by mobile sources, including on- and off-road vehicles, and are not individually regulated through motor vehicle emission standards. In addition, they may form secondarily in the atmosphere from other mobile emissions (HEI Air Toxics Review Panel 2007). The mobile source air toxics considered here are: benzene; 1,3-butadiene; formaldehyde; acetaldehyde; acrolein; and polycyclic aromatic hydrocarbons (PAHs).

A review of emissions literature pertaining to the impact of biodiesel on air toxic emissions is provided in Chapter 4. This information is extended with modelling using MOBILE6.2C in Chapter 5 to produce national estimates of percent change in emissions for individual air toxics resulting from the use of biodiesel blends. As reviewed briefly in Section 7.1, these compounds are associated with a number of different health effects, including both threshold and non-threshold impacts. The following sections provide a brief overview of the health effects of these compounds, as well as what can be ascertained from the previous chapters regarding the potential impacts on human exposure resulting from biodiesel use.

8.2.1.1 Benzene, 1,3-butadiene, formaldehyde, acetaldehyde, acrolein

Health Effects

As summarized in Section 7.1, benzene, 1,3-butadiene, formaldehyde, acetaldehyde and acrolein are five key mobile source air toxics that are associated with significant health outcomes.

Acetaldehyde is considered toxic under CEPA 1999 due to evidence of the induction of tumours in the upper respiratory tract resulting from its inhalation. In addition, a tolerable concentration (the level to which individuals can be exposed for a lifetime without deleterious effect) of 390 $\mu g/m^3$ was derived to protect against non-neoplastic effects in the respiratory tract (Environment Canada and Health Canada 2000; Health Canada 2004). Acetaldehyde is also classified as

possibly carcinogenic to humans by IARC (International Agency for Research on Cancer) (1999) and as a probable human carcinogen by the US EPA (1991).

Inhalation exposure to formaldehyde can result in both non-cancer effects, particularly on the respiratory system, as well as carcinogenic risk to humans (Environment Canada and Health Canada 2001). Carcinogenicity is thought to result following inflammation/cytotoxicity and subsequent cell proliferation, and therefore risk is assumed to be negligible at sufficiently low exposure concentrations (Health Canada 2006). A tolerable concentration of ≤120 $\mu g/m^3$ was derived to protect against non-cancer threshold effects (Health Canada 2004). In addition, Health Canada has derived a one-hour exposure limit of 123 $\mu g/m^3$ and an eight-hour exposure limit of 50 $\mu g/m^3$ as indoor air quality guidelines for this compound (Health Canada 2006). Formaldehyde has been classified as a probable human carcinogen by the US EPA (1991), and is currently under review there. More recently, formaldehyde was reclassified as a human carcinogen (Group 1) by IARC (2006). This reclassification was based on the conclusion of "sufficient" evidence of carcinogenicity in humans and in animals, including "sufficient" epidemiological evidence for nasopharyngeal cancers in humans, and "strong" evidence for a causal association with leukemia in humans (IARC 2006).

Benzene is classified as a human carcinogen in Canada (Government of Canada 1993), in the United States (US EPA 2000) and by IARC (1987). Multiple epidemiologic studies have reported an elevated risk of leukemia in groups exposed occupationally to benzene, as well as benzene carcinogenicity in animal models (Government of Canada 1993).

Chronic exposure to 1,3-butadiene has led to international organizations concluding that it is likely a human carcinogen (Environment Canada and Health Canada 2000; IARC 2008; US EPA 2002).

Acrolein is a reactive compound whose effects from inhalation are expected to be localized in the tissues of the upper respiratory tract. Health Canada (2004) derived a tolerable concentration for acrolein of 0.4 $\mu g/m^3$.

Emissions

Chemically, the mobile source air toxics benzene, 1,3-butadiene, formaldehyde, acetaldehyde and acrolein are VOCs. In general, the VOC species present in emissions of diesel or biodiesel are similar. Although individual studies have reported varying results regarding the impact of biodiesel blends on the magnitude of emissions of individual VOCs (see Section 4.3.4), evaluation of the overall literature base for HDDVs led to the derivation of a single biodiesel VOC emission factor for use in MOBILE6.2C (see Chapter 5), due to a lack of sufficient data for individual species.

Based on MOBILE6.2C, the use of B20 would result in a reduction in VOC emissions from the Canadian on-road HDDV fleet as compared to ULSD emissions. Specifically, the HDDV fleet-average emission factors for the five key mobile source air toxics were calculated to decrease by 18% with the use of B20 in 2006. In 2010 and 2020, B20 is expected to result in decreases of 14% and 3%, respectively, in the value of ULSD emission factors for these compounds (Section 5.2). Under the ULSD base-case scenario, the HDDV fleet represents a relatively small component of the overall on-road mobile emissions inventory for VOCs: 4% in 2006 and 2010, and 6% in 2020. Therefore, although reductions in VOC emissions are anticipated with biodiesel use, it is likely that the effect on the overall emissions inventory would be very small.

Air Concentrations

It is important to note that AURAMS has not been systematically validated with respect to estimates of ambient concentrations of air toxics/VOCs, hence this added uncertainty is inherent in the following data.

AURAMS data for VOCs as a whole indicate that the use of B20 (2006) is estimated to reduce summer total VOC concentrations in major urban centres such as Toronto and Montréal by approximately 0.01–0.05 ppbv. Individual VOC species are generally not explicitly handled within AURAMS, with the exception of formaldehyde. AURAMS estimates concentration reductions for the following VOC grouped species for Toronto and Montréal, respectively (summer 2006): 0.6% and 0.8% for formaldehyde; 1.5% and 2.0% for higher aldehydes (includes acetaldehyde); 0.1% for C3H8 in both locations (includes benzene); and 0% for higher alkenes (includes 1,3-butadiene) (Section 6.2.1.5). These minor improvements in air quality due to biodiesel use are expected to be reduced in the future (2020), as a result of lessening emissions benefits from biodiesel over time.

Because human exposure to VOCs is dependent on human activity patterns and air concentrations in a number of microenvironments (e.g., indoors or in vehicles), the influence of the minor reductions in ambient VOC levels noted in this analysis on overall human exposure is likely to be very minimal.

Conclusion

The use of biodiesel fuel in Canada is expected to result in minor reductions in emissions of benzene, 1,3-butadiene, acetaldehyde, formaldehyde and acrolein, and these reductions will lessen over time. The impact of these emission changes on human exposure is expected to be very minimal.

8.2.1.2 Polycyclic aromatic hydrocarbons

Health

Five polycyclic aromatic hydrocarbons (PAHs) (benzo[*a*]pyrene, benzo[*b*]fluoranthene, benzo[*j*]fluoranthene, benzo[*k*]fluoranthene, and indeno[*1,2,3-cd*]pyrene) were classified as

“probably carcinogenic to humans" by Environment Canada and Health Canada (Government of Canada et al. 1994). More recently, naphthalene was also declared toxic on the basis of carcinogenicity (Government of Canada et al. 2008). In addition, the US EPA designated the following seven PAHs as probable human carcinogens in 1994: benzo[*a*]pyrene; benzo[*a*]anthracene; benzo[*b*]fluoranthene; benzo[*k*]fluoranthene; chrysene; dibenz[*a,h*]anthracene; and indeno[*1,2,3-cd*]pyrene (US EPA 1994a; US EPA 1994b; US EPA 1994c; US EPA 1994d; US EPA 1994e; US EPA 1994f; US EPA 1994g). Naphthalene was classified as a possible human carcinogen by the US EPA (1998).

Emissions

PAHs are generally produced through the combustion or pyrolysis of organic matter (Government of Canada et al. 1994). PAH emissions from mobile sources vary with engine and fuel type, and occur in both the particulate and gaseous phases of diesel emissions (HEI Air Toxics Review Panel 2007). Section 4.3.2 provides an overview of emission testing studies that examined the impact of biodiesel use on PAH emissions. Overall, a reduction in PAH and nitro-PAH emissions has been documented, but not in all studies.

The MOBILE6.2C model used in this analysis includes the following fleet-wide average changes in HDDV emission factors for several PAHs associated with the use of B20 compared to ULSD in 2006, 2010 and 2020, respectively: -14%, -12% and -9% for benzo[a]pyrene; -12%, -13% and -11% for benzo[b]fluoranthene or benzo[k]fluoranthene; -12%, -23% and 0% for indeno[1,2,3-cd]pyrene; -13%, -12% and -9% for naphthalene; -13%, -12% and -10% for benzo[a]anthracene; and -13%, -14% and -9% for chrysene (see Section 5.2). The MOBILE6.2C analysis used default basecase emission factors within the model for individual PAHs based on default ratios to PM_{10} emission factors (US EPA and E.H. Pechan & Ass. 2008). Thus, biodiesel was assumed to affect PM emissions only, and the relevant default PAH ratios in MOBILE6.2 were not altered due to biodiesel use. Some PAHs occur in both the particulate and gas phases, and hence calculation of the emission factor based solely on association with the particulate phase may introduce additional uncertainty in the model. However, this assumption follows the default values of the MOBILE6.2 model, which were selected to reflect the predominant phase in which the PAH is likely to be found.

These changes in emission factors represent substantial reductions within this component of the mobile fleet. The contribution of the HDD fleet under a ULSD scenario to overall on-road mobile emissions of these compounds is variable and expected to reduce over time (Table 8-12). Thus, although certain PAH emission reductions are expected to occur due to biodiesel use, this impact will be reduced by 2020.

Table 8-9 Percent contribution of on-road HDDVs to all on-road emissions of PAHs (B0 scenario)

Year	Benzo[a]pyrene (%)	Benzo[b]fluoranthene or Benzo[k]fluoranthene (%)	Indeno[1,2,3-cd]pyrene (%)
2006	14.7	10.6	1.3
2010	10.8	7.7	0.9
2020	2.8	2.0	0.1

Air Concentrations

The AURAMS model does not have the capability to calculate air quality changes in PAHs and therefore no estimates of the impact of PAH emission changes due to biodiesel use on ambient PAH concentrations in Canada have been made. Given the estimated changes in PAH emissions with B20 use, it is likely that small reductions in PAH air concentrations would occur, particularly in locations or corridors heavily impacted by HDDVs. However, little effect is likely by 2020.

Conclusion

Biodiesel use in Canada is expected to result in minor reductions in PAH emissions from mobile sources, which will lessen substantially by 2020 due to the turnover of the HDDV fleet and the introduction of cleaner vehicles. It is possible that emissions reductions would translate into reduced human exposure to individual PAHs, especially near roads with extensive HDDV traffic. However, by 2020 this impact is expected to be minimal.

8.2.2 Characteristics of particulate matter

8.2.2.1 Particle size and particle number

Particulate matter of mass median aerodynamic diameter less than 100 nm ($PM_{0.1}$) is referred to as ultrafine particles (UFP) or the transient nuclei mode. These particles are formed during the fuel combustion phase and during the gas to particle conversion phase. Road transport is a major contributor to UFP emissions, especially in urban areas (Kumar et al. 2010). Although UFPs represent a small amount of the total mass of ambient particulate matter, this fraction is characterized by very high particle number and also contributes substantially to overall surface area of PM (Harrison et al. 2000; HEI Panel on the Health Effects of Traffic-Related Air Pollution 2010). Because of their reactivity, high number and extremely small size, UFPs are short-lived and concentrations drop off rapidly at increasing distance from sources such as roadways. In general, the UFP number concentration decreases by about half within 100m of major roads (although this varies depending on the atmospheric stability of the location) and approaches background levels approximately 300m or more from a major road (CRC 2008).

The very small size of UFPs allows them to penetrate deep into the lung, and deposition in the alveolar region of the lung is higher than for larger particles. For example, it is estimated that

approximately 20–40% of $PM_{0.01-0.1}$ deposits in the alveoli of an adult male human (nose-breathing and at rest), while alveolar deposition of coarse mode particles (PM_{1-10}) is estimated at roughly 0–20% (US EPA 2009). To date, a relatively limited number of studies have investigated the relationship between UFP exposure and health outcomes. The US EPA (2009) concluded that the overall literature suggests a causal relationship between UFP exposure and both cardiovascular and respiratory outcomes, although substantial uncertainty remains.

There is a substantive body of evidence indicating a reduction of PM mass emissions associated with biodiesel use in HDD engines. A smaller number of studies have examined changes in the particle size distribution and the particle count resulting from biodiesel use. As reviewed in Section 4.2.1, results have been variable, but there is a tendency for biodiesel emissions to contain a smaller number of accumulation mode particles (i.e. $PM_{2.5-0.1}$) and an increase in nucleation mode particles (i.e. $<PM_{0.1}$) compared to conventional diesel emissions.

Because UFP concentrations are highest in close proximity to sources (Zhu et al. 2006), it is possible that widespread use of biodiesel could cause an increase in UFP concentrations in environments dominated by HDDVs, such as along specific roadways. It is estimated that approximately 6% and 32% of Canadians were living within 50m and 250m, respectively, of a major roadway in 2006 (Statistics Canada 2011). However, the MOBILE6.2C model does not address UFPs and therefore quantitative modelling of the impact was not possible, and the potential impact on air quality remains unknown at this time.

8.2.2.2 Organic carbon/elemental carbon

There is an indication in the emissions literature that the use of biodiesel results in a reduction in the elemental carbon (EC) fraction and an increase in the organic carbon fraction of PM emissions (see Section 4.3.3), due in part to a decrease in the EC mass.

Although the health effects of particulate matter as a whole have been well studied and characterized, little has been done to elucidate which PM constituents are responsible for eliciting specific health outcomes. The US EPA (2009) reports that there is some indication in the literature that EC may be linked to cardiovascular endpoints and OC to respiratory outcomes, but the data remain very limited at this point. Therefore, it is not possible to interpret the human health significance of the potential reduction in the EC/OC associated with the use of biodiesel fuels compared to conventional diesel fuel.

8.2.3 Relative toxicity of biodiesel emissions

An extensive amount of scientific literature has been published regarding the toxicological effects induced by exposure to diesel emissions. A smaller number of studies have examined the potential health impacts associated with exposure to biodiesel emissions, and their findings are reviewed in detail in Chapter 7. Generally, these studies focus on the toxicity of the biodiesel exhaust mixture, often relative to the toxicity of petroleum diesel exhaust. In summary, the

available data are insufficient to draw conclusions regarding the toxicity of biodiesel exhaust relative to that of diesel in many organ systems, such as cardiovascular, immunological and neurological systems. It was concluded that biodiesel exhaust is unlikely to be more toxic to the respiratory system than diesel exhaust although the data remain limited. With respect to outcomes relevant to the development of carcinogenesis, mutagenicity has received a fair amount of attention and the data suggest that PM extracts from biodiesel exhaust may be less mutagenic than those from diesel exhaust. More limited data suggest that biodiesel exhaust may be similar in terms of clastogenicity, have similar or less effect regarding genetic instability and be more cytotoxic than conventional diesel emissions. Thus overall the dataset pertaining to biodiesel exhaust toxicity remains limited, but the available evidence does not suggest a substantially enhanced toxicity relative to diesel exhaust.

8.3 Biodiesel Exposure Monitoring Studies

Few studies have measured changes in air pollutant exposures under specific conditions heavily influenced by biodiesel fuel emissions. These have focussed on occupational settings, and therefore the reported exposures are not necessarily reflective of general population exposure scenarios. Occupational studies in which the exposure setting would not be encountered by the general population (e.g., in a mine) have not been reviewed here.

Traviss et al. (2010) conducted limited exposure monitoring among workers at a municipal recycling facility in 2004, during which equipment (large front-end loader, small front-end loader and a skid steer loader) was operated on B20 or diesel (≤500 ppm sulphur), with the cabins open. A single day of monitoring was conducted for each fuel.

Average $PM_{2.5}$ levels were higher during the diesel operating day (95 $\mu g/m^3$) than the biodiesel day (34 $\mu g/m^3$) at the monitoring site in the plant. Similarly, PM_4 levels were more than five times higher in the cabin of the front-end loader during diesel use than during biodiesel use (5332 $\mu g/m^3$ vs. 947 $\mu g/m^3$). In addition, formaldehyde and acetaldehyde levels were substantially lower at two locations in the plant when biodiesel was in use. The authors noted the potential value of biodiesel in reducing occupational exposures in this type of working environment. However, this was a pilot study with limited sampling, and the results might be different if a similar study were undertaken with ULSD as the baseline fuel.

Vijayan and Kumar (2010) measured PM inside public transit buses operating in parallel on diesel or biodiesel (B20) in Toledo, Ohio. Sampling was carried out for a three week period in July, 2006. The diurnal concentration trends were similar between the two fuel types. However, the PM_1 concentrations in the diesel buses were two to three times higher than in the biodiesel buses throughout the day, with the relative differences being highest during the morning hours. It should be noted that ULSD was introduced in the US during 2006, and the authors state that the

buses were all running on ULSD by the end of 2006. However it is unclear if the vehicles were operating on ULSD or higher sulphur diesel during the sampling period.

8.4 Comparison to Other Analyses

To the best knowledge of the authors, very few analyses of the human health implications of biodiesel use have been undertaken.

As part of its Renewable Fuel Standard Program (RFS2) Regulatory Impact Analysis (RIA), the US EPA (2010) conducted an analysis of impacts of the overall biofuels program on non-greenhouse gas pollutants. The RIA compared the impacts of the RFS2 standards in 2022 (in the form of volumes of renewable fuels to be used in transportation as mandated by Congress in the Energy Independence and Security Act) to a base-case scenario of renewable fuel use based on a 2007 energy outlook. However, the RFS2 analysis examined the use of first and second generation ethanol and biodiesel fuels, and although at the emissions level their individual effects were reported, air quality modelling and health impact analyses include the effect of all biofuels combined, as well as of upstream and downstream emissions.

The RFS2 analysis assumed an overall annual use of 0.85 billion gallons (3.22 billion litres) of FAME biodiesel and 0.15 billion gallons (0.57 billion litres) of renewable diesel, of a total of 35.3 billion gallons (133.6 billion litres) of renewable fuels (the remainder being ethanol). Regarding the biodiesel analysis contained in the RIA, the US EPA modeled the impact of biodiesel (generally B5 or less) use in on-road HDDVs only, similar to the assumption in this Health Canada analysis.

Comparing emissions from HD vehicles operating on B20 to those operating on conventional diesel, it was estimated that NO_X emissions would increase by 2.2%, PM emissions would decrease by 15.6%, hydrocarbon (and air toxics) emissions would decrease by 13.8% and CO emissions would decrease by 14.1%. These values are similar to those derived for this report for 2006–2010 (see Table 5-10). Overall, the US EPA (2010) estimated that the RFS2 program (ethanol and biodiesel) would result in 33 to 85 additional cases of premature adult mortality due to $PM_{2.5}$ and 36 to 160 additional cases due to O_3 in 2022 (the ranges reflect variable CRFs from different studies). PM increases were expected in certain areas, and the authors stated that these are likely due to emissions from renewable fuel production plants and from the transportation of renewable fuels. In addition, reductions of 0.0001 to 0.023 $\mu g/m^3$ in annual population-weighted exposures to air toxics were predicted, which represent less than 5% reductions. These expected exposure reductions were not translated into health impacts as part of the RFS2 analysis. Overall, given that biodiesel represents a small portion of the renewable fuel volume mandate in the US RFS2 and that the US population is about 10 times larger than the Canadian population, the results support the Health Canada findings that biodiesel use is associated with very minor health impacts.

The National Renewable Energy Laboratory conducted a detailed analysis of air quality and health impacts of biodiesel (NREL 2003). For that analysis, NREL compared 100% penetration of B20 in the on-road HDDV fleet to a diesel base-case, and modeled air quality changes in O_3, CO, $PM_{2.5}$ and PM_{10} in specific locales and for specific air pollution episodes. Overall, the authors estimated that B20 use would result in an increase of 2.4% in NO_X emissions, and a decrease of 8.9% in PM_{10}, 17.9% in VOCs, 13.1% in CO and 20% in SO_2 emissions (the analysis was done prior to the implementation of ULSD use in on-road diesel vehicles). These values do not differ substantially from the Health Canada values for 2006–2010, except for SO_2 (Table 5-10). Daily maximum O_3 level changes (1- and 8-hour averaging times) in all locations varied from reductions of ≤1.2 ppb to increases of ≤0.25ppb with B20 use, and minor changes of less than 2 µg/m^3 (-1.61 to +0.62 µg/m^3) in daily maximum $PM_{2.5}$ or PM_{10} concentrations were noted. Reductions of ≤0.03 ppm were reported for CO.

In addition, the authors included a relatively simplistic estimate of change in health risk from air toxics (diesel PM, benzene, 1,3-butadiene, formaldehyde and acetaldehyde) in the south coast air basin, which suggested that annual use of B20 would result in a 5% reduction in mortality risk due to air toxics. This was due in part to the assumption that PM generated from the combustion of B20 is 5% less toxic than conventional diesel PM.

The California Environmental Protection Agency recently adopted a formal approach to guide the conduct of multimedia evaluations of fuels (University of California et al. 2008), and biodiesel is the first fuel to be evaluated under that framework. The Tier 1 report, which provides a review of the literature and identifies key elements to be addressed through research in Tier 2, was published in 2009 (University of California 2009). The report noted the importance of modelling the air quality and health impacts of changes to the lifecycle emissions inventory related to biodiesel use. New experimental research has been undertaken to address gaps relating to biodiesel emissions testing, fate and transport, biodegradation, aquatic toxicity, and toxicity. It is anticipated that detailed analyses of health impacts associated with air quality changes will be included in the Tier 3 report, but no data are available at this time.

8.5 Uncertainties

The estimation of health impacts associated with a widespread fuel change is the final step in a sequential process, and is preceded by evaluating the impact of the fuel on mobile source emissions and the impact of those emission changes on air quality. As a result, the understanding of the health implications of biodiesel is influenced by the uncertainties in each of the previous steps in the analysis (emissions impacts, mobile emissions modelling, inventory specification, forecasting and atmospheric modelling), which were discussed in Chapters 4, 5 and 6. This section focuses on uncertainties associated with the health analysis itself.

The quantitative health impact analysis presented here is based on changes in ambient concentrations of $PM_{2.5}$, O_3, NO_2, CO and SO_2 and established concentration response functions

for morbidity/mortality outcomes associated with them. Although the CRFs are derived from the peer-reviewed literature and are endorsed by Health Canada, they include some inherent uncertainty, which is reflected in the AQBAT output as the 2.5^{th} or 97.5^{th} percentile estimates for the health outcomes. Consideration of the 2.5^{th} or 97.5^{th} percentile estimates reveals that the key conclusion (based on central CRF values) that the health benefits/risks of widespread biodiesel use in Canada are very small, is not changed by the uncertainty in the CRF. This is because the largest estimates of health outcomes, i.e., the 2.5^{th} or 97.5^{th} percentile of the estimates, were less than three times those based on the mean estimates, and are still characterized as very small impacts. However, AQBAT includes a limited number of CRFs for the criteria air contaminants, although other health outcomes associated with exposure to these pollutants have been reported in the literature (e.g., reproductive and developmental outcomes) (US EPA 2009). As a result, not all health benefits/costs of air pollution changes are included in the analysis. Furthermore, some of the health endpoints included in AQBAT are based on more limited or older information (e.g., asthma symptom days associated with summer O_3 exposure) and therefore embody more uncertainty than other parameters. In addition, the health impact estimates for 2020 are based on population projections provided by Statistics Canada, which include inherent uncertainty, as well as assumptions regarding future baseline rates of the health impacts assessed.

The quantitative analysis focused solely on changes in pollutant emissions during vehicle use, and excluded upstream emissions changes such as during the growing of oilseeds, the production of biodiesel, and the storage and transport of the fuel, as well as off-road uses of the fuel. These stages of the fuel life cycle were not included due to a lack of sufficient data for quantitative analysis. It is unclear how the incorporation of these aspects would affect the net health benefits/cost estimates obtained. However, lifecycle assessment has shown that fuel use (combustion/evaporation) is a large source of emissions and associated health effects when considering all stages in a transportation fuel lifecycle (ADEME 2010; Huo et al. 2009; NRC 2003). This is likely due to the general proximity of vehicle emissions to dense human populations. Nonetheless, the lack of consideration of quantitative impacts from upstream stages represents a significant limitation in this analysis. Chapter 2 includes a qualitative overview of the biofuels production industry, which provides some general indication of the types of potential impacts of this sector on the local community population. These include chemical air emissions such as particulate matter (during seed crushing), hexane (used as a solvent in oil extraction) and methanol (used in the transesterification process), as well as combustion emissions (NO_X, CO, SO_2, $PM_{2.5}$, PM_{10}, VOCs and air toxics) from steam boilers, generators or on-site vehicles.

There is a general lack of quantitative information on the impact of biodiesel use on concentrations of mobile source air toxics, not only in ambient air but also in specific microenvironments that are more likely to be affected by transportation. Although emission

testing results can be informative, they do not include consideration of atmospheric processes and secondary production. The exposure/risk per unit of emitted pollutant can vary by orders of magnitude, depending on the spatial distribution of emissions, i.e., their proximity to human populations (McKone et al. 2011).

The AURAMS model does not include explicit treatment of individual air toxics, and does not address PAHs at all, thus limiting researchers' ability to predict changes in human exposure to these compounds. In addition, changes in ambient concentrations of air toxics do not necessarily translate into comparable changes in exposure because of other sources of these pollutants in specific microenvironments such as indoors. Far more data would be required to estimate the impact of fuel changes on total population exposure to air toxics. These compounds are associated with mortality due to chronic diseases (cancer) as well as illnesses such as asthma.

Finally, there is a general indication in the emissions testing literature that the combustion of biodiesel blends results in lower PM mass but higher numbers of very small particles. At this point in time, neither mobile sector nor atmospheric models address UFPs, and therefore changes in their spatial distribution and quantity, and hence exposure, cannot be estimated. Moreover, the understanding of the health effects of UFPs remains limited. Similarly, the significance of changes in EC/OC remains unknown with regards to emissions, air quality and human health. Overall, this represents substantial uncertainty in this analysis.

8.6 Conclusions

A detailed analysis of the population health impacts associated with widespread biodiesel use was undertaken. Mobile emissions modelling incorporating the emissions impacts of biodiesel in the on-road HDDV fleet formed the basis of this analysis. These estimates provide information on the spatial and temporal variability of expected pollutant emission changes, which formed the inputs for national scale air quality modelling of specific biodiesel scenarios.

AQBAT, Health Canada's health benefits analysis tool for air quality changes, was used to provide quantitative impact estimates for the health endpoints for which concentration-response functions have been derived for $PM_{2.5}$, O_3, NO_2, CO and SO_2. The scenarios selected for air quality and health impact modelling included a comparison of the national use of B5 or B20 to that of ULSD, in 2006 and in 2020. It was assumed that B20 use would be confined to the May–September period due to technical limitations of using B20 under low temperature conditions. It should be noted that occupational exposures to biodiesel were not considered in this analysis.

As expected due to the minimal air quality changes observed due to biodiesel use (Chapter 6), limited health benefits are estimated for the biodiesel scenarios. The majority of the mortality and morbidity benefits estimated for 2006 are associated with reductions in $PM_{2.5}$ and O_3 levels, while increases in NO_2 levels are associated with minor increases in mortality. The overall effect of annual B5 use would be a reduction of approximately five premature mortalities, and a total

monetary benefit of about $33 million. National summertime B20 use in 2006 is estimated to result in a net reduction of seven mortalities, and a total monetary benefit of approximately $48 million.

The biodiesel emission benefits from the HDDV fleet are expected to be reduced by 2020 because the incorporation of new emission control technologies will result in such low emissions that the impact of biodiesel will become less evident. As a result, the health impacts of national B5 use in 2020 are estimated to be close to neutral, with an increase of less than one death, and an associated monetary cost of about $4 million. The use of B20 is associated with approximately two mortalities, related to localized minimal increases in $PM_{2.5}$, O_3 and NO_2, and a cost of about $15 million, although the pollutant concentration changes underlying these effects are believed to be close to the AURAMS model detection limit.

High-resolution urban air quality modelling for these scenarios was undertaken for a 12-day high pollution episode for Montréal, and the health benefits calculated in terms of the overall impact on all mortality/morbidity *caused by above-background air pollutant concentrations*. The results indicate that for 2006, B5 use would result in a net decrease of 0.02% and 0.03–0.04% for the mortality and morbidity metrics, respectively. B20 would result in a net decrease of 0.01% and 0.08–0.19% in mortality and morbidity, respectively, except for an increase of 0.21% in hospital admissions. In 2020, B5 is estimated to result in net increases of ≤0.01% in mortality and morbidity, while B20 is estimated to result in net increases of 0.07% and 0.02–0.42% in mortality and morbidity, respectively. It is recognized that these represent very small changes relative to the baseline rates of these outcomes due to air pollution in the population.

Several air pollutants could not be considered quantitatively, due to limitations in MOBILE6.2C, AURAMS and/or AQBAT to model them. Qualitative consideration of the data available reveals that minimal reductions in emissions of VOCs (including benzene, 1,3-butadiene, acetaldehyde, formaldehyde and acrolein) and PAHs are expected with biodiesel use, which may translate into very minor reductions in human exposure to these compounds. The implications of emissions testing data (suggesting that biodiesel can result in an increase in particle number and a downward shift in particle size distribution) for human exposure to UFPs and toxicity are unclear at this time.

Although the scenarios examined here do not replicate specific existing Canadian biodiesel policies, they were selected in order to provide an overall picture of potential health impacts of biodiesel use in Canada. Overall, the use of B5 or B20 nationally is expected to result in very minimal air quality and health benefits/costs, and these are likely to diminish over time. Although substantial modelling and data limitations remain, the currently available evidence suggests that the incremental health impacts associated with the widespread use of low level biodiesel blends in Canada as compared to the use of ULSD, are expected to be minimal.

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Chapter 9. Overall Conclusions and Limitations

9.1 Conclusions

The health impacts analysis detailed in this report evaluates the potential human health implications of the widespread production, distribution, storage and use of biodiesel in Canada, in comparison to ULSD. A primary consideration of this analysis is the potential impact of biodiesel use on mobile sector emissions, atmospheric concentrations of air pollutants (e.g., CACs, air toxics), and consequently human health impacts.

The Government of Canada put in place a 2% renewable content requirement in diesel and heating oil on July 1st, 2011. Any fuel defined as a renewable fuel as per the *Regulations Amending the Renewable Fuels Regulations* and produced from appropriate feedstocks may be used. This national 2% renewable requirement creates an annual demand of around 650 million litres of renewable diesel fuel. Currently, more than 70% of biodiesel production originates from tallow and recycled greases. Canola and soy oil-based biodiesel fuel production remains limited. It is forecasted that the 2% requirement will not be met by Canadian producers, at least in the initial years. Imports of biodiesel and other renewable forms of diesel fuels from the US or abroad are expected to meet the demand stimulated by the Regulation.

Biodiesel: Impacts of Biodiesel Production

With respect to the upstream stages of the fuel life cycle, which can be associated with very variable emissions and impacts, this report focused on biodiesel production facilities. Preliminary analysis of emissions data from Canada's NPRI suggests that biodiesel facilities may emit more CACs than conventional petroleum refineries on a fuel unit basis, while air toxic emissions are considerably lower both in quantity and variety of pollutants. However, in terms of absolute emissions (i.e., kg or tonnes), biodiesel facilities emit a fraction of the emissions reported from petroleum refineries. In general, biodiesel production plants are not expected to emit pollutants in excess of current air quality regulations. Based on the GHGenius model, the modelling results for the whole upstream stage (i.e., feedstock and fuel production) show that, in comparison to conventional diesel, biodiesel has lower CO and SO_X emissions but higher PM, NO_X and VOC emissions, both on a g/GJ and a g/km basis. Feedstock production and handling, fertilizer production and use, and fuel processing are responsible for the greater PM, NO_X, and VOC emissions, respectively, with biodiesel fuels. SO_X emissions are not expected to be an issue with biodiesel fuels, while the generation of useful or marketable co-products from the biodiesel production process helps to lower CO emissions via displacement (i.e., CO emissions avoided from the use of the displaced product with higher lifecycle emissions become a credit for the biodiesel process).

Biodiesel: Environmental Fate and Transport

Health risks from accidental releases and spills of bulk biodiesel fuels can be expected in Canada. Total releases of up to 40,000 litres of neat biodiesel per year (based on a 2% renewable content requirement in diesel fuel and heating oil) are likely given historical spill and leak information for petroleum fuels. Physical and chemical properties of biodiesel fuel components are significantly different from those of petroleum fuels. Using screening-level modelling systems, it was found that biodiesel fuel components are expected to migrate less than ULSD fuel components following a spill to soil and groundwater, as expected based on biodiesel's physical and chemical characteristics, particularly the greater biodegradation rate of biodiesel fuel components compared to diesel fuel fractions. The limited mobility of biodiesel fuel components can be considered an environmental benefit since the contamination plumes are expected to be contained within a relatively smaller volume of soil and groundwater. Notwithstanding the modelling uncertainties, it is reasonable to conclude that biodiesel fuels would have less impact on the environment and human health than petroleum fuels following an uncontrolled release in a natural or urban environment. However, no conclusion can be drawn regarding spills of biodiesel fuel blends, as interactions between fuel components were not accounted for in the modelling.

Biodiesel Use: Impacts on Motor Vehicle Emissions

The transportation sector is responsible for emissions of several pollutants, such as CACs and air toxics. Mobile sources are the largest contributing sector to NO_X and VOC emissions, and diesel vehicles are responsible for a large share of the NO_X and PM emissions from this sector. Biodiesel fuel components are rich in different chemical structures, resulting in complex chemical reactions during combustion that are still not completely understood. In addition, results from the literature have been somewhat variable depending on test vehicles (e.g., LDDV, HDDV, model year, after-treatment devices) and procedures (e.g., test cycles, reference fuels, sampling methods). However, based on a review of the literature, general trends are observed with regards to the impact of biodiesel fuels on HDDV exhaust emissions: compared to ULSD, biodiesel is generally expected to result in reductions in emissions of PM, CO, HC, VOCs and PAHs, while no net impact or a slight increase in NO_X emissions is expected. Only limited data are available regarding the impact of biodiesel use on emissions from North American LDDVs, and thus conclusions cannot be drawn for this portion of the fleet at this point in time. After-treatment devices are expected to have a significant impact on vehicle emissions, although any additional impacts of biodiesel fuel in vehicles equipped with these devices remain to be determined.

The MOBILE6.2C model estimates emissions from on-road vehicles in Canada. The most recent version of MOBILE6.2C, which captures the effects of the use of renewable fuels, was used to quantify the changes in on-road HDDV emissions due to biodiesel use and the results are presented in Table 9-1. The results indicate that most HDDV exhaust emissions decrease, while

NO_X emissions increase slightly. The magnitude of change generally varies linearly with biodiesel content, such that B20 has roughly four times the impact of B5. Considering the impact of biodiesel use in HDDVs on emissions from all on-road mobile sources, NO_X and PM are most impacted since HDDVs are the predominant mobile source of these pollutants. Baseline on-road heavy duty vehicle emission projections for 2020 are considerably lower than 2006 levels due to the turn-over of the Canadian vehicle fleet and the introduction of new emission control technologies, with 2010 and beyond vehicles having to meet more stringent exhaust emission standards (e.g., $PM_{2.5}$ baseline emissions are estimated to be 6400 tonnes in 2006, while they are projected to be 820 tonnes in 2020). Hence, the net and relative impacts of biodiesel on exhaust emissions in 2020 are expected to be reduced compared to 2006 levels.

Table 9-1 Percent change in on-road HDDV emissions in Canada under biodiesel use scenarios compared to ULSD scenarios, estimated from MOBILE6.2C results

Scenarios		TPM, $PM_{2.5}$, PM_{10}	NO_X	SO_2	CO	VOC_{total}*	B[a]A	B[a]P
2006	**B5 vs. B0**	-3.2	0.9	0	-2.7	-4.4	-3.2	-3.3
	B20 vs. B0	-12.6	3.6	0	-10.7	-17.8	-12.6	-13.5
2020	**B5 vs. B0**	-2.2	0.7	0	-1.6	-0.8	-2.9	-2.4
	B20 vs. B0	-8.7	2.8	0	-6.5	-3.5	-9.8	-8.7
* Results for VOC_{total} are similar to: acetaldehyde, acrolein, benzene, formaldehyde, n-hexane, THC total, and 1,3-butadiene.								

Biodiesel Use: Impacts on Canadian Air Quality

Photochemical modelling for the current project was conducted with AURAMS to investigate the impact of biodiesel blend use on air quality in Canada. Overall, the proposed B5 and B20 biodiesel scenarios are associated with very minimal changes in emissions and ambient air concentrations of the pollutants analyzed. The B5 and B20 scenarios under 2006 conditions predict small (less than 1%) but non-negligible changes in air quality. In general, $PM_{2.5}$ and O_3 concentrations decrease in urban areas and increase in surrounding areas. CO concentrations are expected to decrease in all regions. For the 2020 projections, changes in predicted air quality are very small (less than 0.5%) and often close to model detection limits. Ozone and $PM_{2.5}$ concentrations are generally reduced in urban centres, but increase slightly in surrounding areas. CO concentrations are reduced in most areas. The smaller impacts observed under the 2020 scenarios are related to the significant reductions in exhaust emissions for the baseline ULSD scenarios compared to 2006, due in part to the implementation of more stringent emission standards for the model year 2010 and later vehicles. The results from this study concur with previous reports evaluating the impact of biodiesel fuels in different locations across North America, which found that impacts are likely to be minimal. High-resolution modelling provided enhanced spatial resolution of air quality impacts in the Montréal urban area.

Biodiesel Use: Impacts on Human Health due to Changes in Emissions of CACs and Air Toxics
Health Canada's Air Quality Benefits Assessment Tool (AQBAT) was used to quantify Canadian morbidity and mortality risks/benefits from criteria air pollutants associated with the use of B5 or B20 compared to ULSD in the on-road HDDV fleet, in either 2006 or 2020. In 2006, annual B5 or summertime B20 use are associated with a reduction of about five to seven premature mortalities as well as minimal reductions in hospital admissions, emergency room visits and other morbidity outcomes, due primarily to minor reductions in $PM_{2.5}$ and O_3 levels. The monetary valuation of those health benefits is estimated to be $33,000,000 for B5 and $48,000,000 for B20. The biodiesel emission benefits are expected to be reduced by 2020 due to the incorporation of new emission control technologies in the HDDV fleet: the overall health impact of B5 use in 2020 is estimated to be close to neutral, while the summertime use of B20 is associated with approximately two premature mortalities, related to localized minimal increases in $PM_{2.5}$, O_3 and NO_2, with a net health cost of about $15,000,000. However, the pollutant concentration changes underlying these effects in 2020 are believed to be close to the AURAMS model detection limit.

While HDDV emissions of mobile source air toxics are expected to be affected by the widespread use of biodiesel blends, the impact of emission changes on human exposure to these pollutants could not be quantified due to a lack of modelling capabilities. Qualitative consideration of the available VOC and PAH emissions data indicate that minimal reductions are expected for benzene, 1,3-butadiene, acetaldehyde, formaldehyde, acrolein and PAHs in association with use of biodiesel, which may translate into very minor reductions in human exposure to these pollutants, particularly near roads that are heavily trafficked with HDDVs. However, the emissions benefits and any associated reductions in human exposures are expected to diminish by 2020.

Biodiesel Use: Toxicity of Biodiesel Emissions
A detailed toxicological review of biodiesel exhaust was conducted with two objectives: to determine if biodiesel exhaust has a similar, reduced or greater impact than diesel exhaust in terms of specific health effects; and to attribute any difference in the magnitude of effects observed (between biodiesel and diesel exhaust) to a change in the level of a specific physicochemical parameter(s) in the exhaust.

A review of several studies determined that biodiesel exhaust is unlikely to exceed diesel exhaust in terms of respiratory effects (inflammation; histopathology and lung function effects). Only two studies were reviewed that examined cardiovascular effects (endothelial dysfunction, prothrombosis, systemic inflammation and atherosclerosis, haematological effects, and cardiac events) of biodiesel exhaust. Acute exposure of mice to biodiesel exhaust resulted in increases in some systemic inflammatory markers when compared to diesel exhaust. A subchronic study which included exposure of rats to biodiesel exhaust but did not include a diesel treatment,

recorded no significant cardiovascular effects. Based on this limited data set, it was not possible to draw any conclusions as to how biodiesel and diesel exhaust compare with respect to cardiovascular effects.

A review of outcomes relevant to the initiation of carcinogenesis (clastogenicity, biochemical events associated with genetic instability, cytotoxicity, and mutagenicity) indicated that biodiesel and diesel exhaust are similar in terms of clastogenicity, biodiesel exhaust has a similar or lower effect on biochemical events (ROS, inflammation) associated with genetic instability, and biodiesel is equal to or exceeds diesel with respect to cytotoxicity. The majority of studies investigating mutagenicity demonstrated that PM extract from biodiesel exhaust is potentially less mutagenic than diesel exhaust PM extract. It is important to note that with the exception of mutagenicity the data set was fairly limited for the outcomes under consideration. Also, given that many of the studies examining these outcomes were carried out under different experimental conditions, generalizations about the initiation of carcinogenesis by biodiesel exhaust compared to diesel exhaust cannot be made at this time.

Only one inhalation study considered reproductive and developmental effects, neurological effects, and systemic effects resulting from exposure to biodiesel exhaust. Given that this study did not include a diesel treatment, it was not possible to draw any comparison between biodiesel and diesel exhaust. Dermal exposure to biodiesel was also considered because of potential exposure during refuelling. However, skin irritation, a potential outcome of this type of exposure, was not considered in the study reviewed.

No information was available for immunological effects resulting from exposure to biodiesel exhaust.

Regarding the second objective of the toxicity evaluation, it was determined that toxicological studies investigating respiratory, cardiovascular, and outcomes associated with initiation of carcinogenesis increasingly reflect efforts to ascribe differences in biological responses between biodiesel and diesel exhaust to differences in physicochemical characteristics between the two fuels. However, for most studies, differences in individual pollutant levels between biodiesel and diesel exhaust have not been specifically linked to changes in a given biological response.

A review was conducted to examine the risk that inhalation exposure of the Bovine Spongiform Encephalopathy (BSE) infectious agent may occur in the general population as a result of the combustion of biodiesel made from Specified Risk Material (SRM) derived tallow. Two scenarios were considered. In the first, the risk was considered negligible provided that SRM and tallow destined for biodiesel production are processed to achieve a tallow purity standard of not more than 0.15 % insoluble impurity content, as per CFIA directives. In the second scenario, in which the insoluble content of the SRM-derived tallow would exceed 0.15% and would

contain BSE agents, it is expected that biodiesel manufacturing and combustion processes would contribute to a reduction in the risk of inhalation exposure to BSE agents. However, given the lack of information on the reduction of potential BSE infectivity during some of these processes, it is not possible to define the level of risk.

The potential for allergic reactions in the general population following inhalation exposure to exhaust from soy-based biodiesel was investigated due to the fact that soy is one the main foods responsible for allergic reactions. The review concluded that denaturation and hydrolysis of proteins during biodiesel production as well as purification processes are likely to reduce the allergenicity of biodiesel. However, if allergenic proteins were able to survive the latter processes, it is highly probable that the proteins would be destroyed during the combustion process given that temperatures in diesel engines are significantly higher than those which cause significant alterations in protein structure.

Finally, a review of the main fuel additive categories that are likely to be used in biodiesel fuels in Canada was carried out. The review included key background and toxicity information for different types of additives as well as specific products.

Overall Conclusion

Although the scenarios examined in this assessment do not replicate specific existing Canadian biodiesel use policies, they were selected in order to provide an overall picture of potential health impacts of biodiesel use in Canada. Overall, the use of B5 or B20 nationally is expected to result in very minimal air quality and health benefits/costs, and these are likely to diminish over time. Although substantial modelling and data limitations remain, the currently available evidence suggests that the incremental health impacts associated with the widespread use of low level biodiesel blends in Canada as compared to the use of ULSD, are expected to be minimal.

9.2 Limitations

In conducting this assessment of biodiesel fuel use in Canada, a number of data gaps and limitations were identified, such as: data availability, physical and chemical characterization of biodiesel fuel components, the impact of biodiesel on vehicular emissions, and the inherent limitations of the modelling tools selected.

Regarding the production stage of the biodiesel lifecycle, monitoring data of emissions from production facilities are insufficient to conduct a quantitative assessment of possible air quality impacts. Specifically, data are insufficient to include emissions associated with biodiesel production within the emissions inventory used in AURAMS modelling of national air pollutant levels, or to evaluate the local impacts of biodiesel production plants with dispersion modelling. This is due in part to the fact that this is a relatively new industry and that current processing,

use, or emission levels do not require mandatory reporting to the NPRI. It is expected that data will be collected in upcoming years when larger biodiesel facilities come on-line.

Understanding the behaviour of biodiesel fuels following accidental releases is limited by the paucity of data regarding basic chemical and physical parameters of biodiesel fuel components. Although some data gaps may be filled using available tools (e.g., EpiSuite, SPARC, carbon fraction), uncertainties remain for several key parameters. Environmental fate and transport modelling is limited to screening-level tools, which require only limited input and assume no interaction between petroleum and biodiesel fuel components. Although this type of modelling is useful, it remains highly dependent on selected values and may not be representative of all environmental conditions in Canada.

A number of key data gaps exist in the overall database of vehicle emissions testing for biodiesel. Although vehicle and engine emissions testing of biodiesel fuel blends reveal some trends with regards to the impacts on major pollutants such as PM, CO and NO_X, additional work is necessary to more precisely characterize these emissions and to expand the inventory of pollutants regularly assessed to include air toxics such as PAHs. In addition, most of the biodiesel testing has been conducted on older vehicle and engine types and there is insufficient data on newer model-year engines/vehicles developed to meet more stringent emissions regulations to quantify the impacts. Importantly, the literature suggests that LDDV and HDDV emissions may be affected differently by biodiesel, but the data for LDDVs are very limited and hence emission impacts are not quantified. Similarly, there is insufficient data to quantify the impact of biodiesel on off-road emissions. Also, determining the relative impact of biodiesel fuels on vehicles or engines equipped with after-treatment devices is important as these vehicles become more common in the on-road fleet. Lastly, emissions testing of biodiesel fuels and the characterization of some types of pollutants (e.g., VOC species, ultra-fine particles) may require the development of new standard testing methods to increase the level of precision and account for the physical and chemical aspects of biodiesel fuels.

Modelling of Canadian on-road emissions with MOBILE6.2C highlights the aforementioned data limitations regarding recent model-year heavy-duty vehicles (i.e., 2010 model-year or later vehicles) and for vehicle classes other than on-road HDDVs (LDDVs and off-road vehicles). In addition, a lack of precise provincial or territorial vehicle count and activity data introduces some uncertainty in the characterization of the Canadian vehicle fleet and its emissions. It should be noted that Environment Canada is currently adapting a new US EPA mobile source emissions model known as MOVES (Motor Vehicle Emissions Simulator) for use in Canada, which estimates significantly higher PM emissions for the diesel sector than MOBILE6.2C does. At the time of this analysis, there was no Canadianized version of MOVES capable of modelling biofuels. However, it is expected that using MOVES in this analysis would not alter the

conclusion that the impact of biodiesel on the overall Canadian emissions inventory and the resulting health impacts of that are minimal.

With regards to the air quality modelling conducted for this analysis, the confidence level for the estimated concentrations of some pollutants is a major limitation (e.g., CO, VOC species). These uncertainties are due partly to an incomplete emissions inventory, either because of unaccounted for sources (e.g., CO) and/or the absence of precise data for specific pollutants (e.g., PAHs, explicit VOC species). Validation of the high-resolution modelling results and additional tests are warranted to allow better use of current modelling capabilities. Spatial allocation and traffic-demand modelling for the Montréal area are also limited to a 1-km resolution. Although this is a vast improvement over previous modelling capabilities, street level resolution, especially along major highways and roads, would provide a more accurate evaluation of air quality impacts.
Attempting to estimate fuel impacts in future years involves projecting a number of variables into the future, all of which involve assumptions and uncertainty. For this analysis, assumptions are made about future fuel use patterns, technologies, fuel quality, vehicle populations, fleet composition, emissions inventories, human behaviour (vehicle kilometres travelled), human populations, and baseline disease and mortality rates. Although these introduce uncertainty into the analysis, the values selected represent the best estimates of various agencies and experts.

No formal uncertainty analysis amalgamating the uncertainty at the various stages of this assessment was conducted. The MOBILE6.2C and AURAMS models are deterministic, i.e., based on single values for input parameters rather than distributions, and thus uncertainty in the estimates is not formally captured. AQBAT addresses the inherent uncertainty in the CRFs and economic valuations by inputting them as distributions rather than single values, and generating a distribution for the estimates of impacts. Health impacts associated with the national modelling of biodiesel use were reported as the 2.5^{th} and 97.5^{th} percentiles as well as the mean value. Overall, the fact that the estimated effects of biodiesel use are minimal is not altered by the selection of these percentiles or the mean, and hence by the uncertainty in the estimates.

This analysis was designed to assess population health impacts of nationwide use of B5 and B20 in the on-road HDDV fleet, although the reality of biodiesel use in Canada is that it varies geographically and temporally, and in terms of the blend levels used. However, given that the potential impacts of national B20 use were estimated to be very minimal and reducing over time, it is anticipated that the use of biodiesel in Canadian urban centres will have very limited impacts on air quality and health.

Much of the uncertainty associated with the toxicological review of biodiesel exhaust is linked to the limited amount of health effects information available. Of the existing studies examining biodiesel exhaust, many were based on high-level or acute *in vitro* exposures and their significance vis-à-vis potential exposures experienced by the general population was not clear.

An important sub-chronic study in rats did not include diesel exposures thus limiting any comparison for the health effects under consideration. Significant differences in experimental designs (animal models, fuels, treatment levels, engine types and conditions, and study methodologies) in biodiesel and diesel studies also contributed to the level of uncertainty which was part of this review.

In conducting a review to examine the risk that inhalation exposure of the BSE infectious agent may occur in the general population due to combustion of biodiesel made from SRM-derived tallow containing greater than 0.15% insoluble content (and containing BSE agents), it was concluded that biodiesel manufacturing and combustion processes would contribute to a reduction in the risk of inhalation exposure. However, there is little to no empirical information on the extent to which biodiesel production will contribute to a reduction in BSE infectivity. Also, there exists some uncertainty as to whether compression ignition engine temperatures are capable of rendering BSE agents inactive.

While it was concluded that the potential for allergic reactions in the general population following inhalation exposure to exhaust from soy-based biodiesel is unlikely given that proteins would probably be destroyed during combustion, there is no explicit testing data to support this. It was also suggested that denaturation and hydrolysis of proteins during biodiesel production may affect potential allergenicity; however, once again there is no direct empirical information to support this.

There is a relatively high level of uncertainty associated with additives and their use in biodiesel in Canada. This stems from the fact that it is difficult to predict which products will be used on a consistent basis and because of the relatively limited toxicological and exposure information for these compounds.

Human Health Risk Assessment for Biodiesel Production, Distribution and Use in Canada

Appendices

May 2012

APPENDIX III. Tables and Figures

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Table III-1 Scenario specific input parameters

Parameter	Scenario	
	UST	Truck spill
NAPL volume/area release rate (m) [a]	NA	NA
Lower depth of NAPL zone (m)	NA	NA
NAPL flux (m/d)	NA	NA
Days of release (d)	NA	NA
Radius of NAPL lens source (m)	NA	NA
Simulation time (y)	5	1
Source thickness (m)	3	3
Source length/estimated plume length (m)	10	10
Depth to groundwater (m)	3.0	3.0
Source width (m)	10	10
Source concentration in groundwater (mg/L)	10	10
a- based on release of 10 000 L of NAPL over 100 m^2, no correction for density applied		

Source: Meridian 2010b, Table A-5

Table III-2 NAPL specific input values required for the HSSM model

Parameter	NAPL blend				Unit	Reference
	ULSD	B5	B20	B100		
NAPL density	0.84	0.84	0.85	0.88	kg/L	Meridian 2009a, Table 9
NAPL kinematic viscosity	2.8	2.9	3.2	5.0	cSt	Meridian 2009b, Table 1
NAPL dynamic viscosity	2.35	2.44	2.72	4.4	cP	Calculated
NAPL surface tension	25	25	26	29	mN/m	HSSM, Allen et al. 1999
K_{OC}	$7.94*10^4$	$7.81*10^4$	$7.42*10^4$	$6.31*10^4$	L/kg	Meridian 2009a, Table 11
NAPL/water interfacial tension	23.2	15.3	12.3	11.5	mN/m	Hollebone et al. 2008
NAPL solubility in water	0.00	$5.25*10^{-4}$	$2.10*10^{-3}$	$9.00*10^{-3}$	mg/L	Meridian 2009a, Table 10

Table III-3 Chemical and physical parameters of *aliphatic* and *aromatic* ULSD fuel components

Aliphatic components	**Unit**	**C>10-C12**	**C>12-C16**	**C>16-C21**	**C>21-C34**	**Reference**
Molecular weight	*g/mol*	$1.60*10^{2}$	$2.00*10^{2}$	$2.55*10^{2}$	$3.80*10^{2}$	CCME 2008b, Table B.1; Meridian 2010b, Table 4
Retardation factor (coarse soil)	-	$5.96*10^{3}$	$1.19*10^{5}$	$1.50*10^{7}$	$2.37*10^{11}$	Calculated (CCME 2008b)
Retardation factor (fine soil)	-	$3.72*10^{3}$	$7.43*10^{4}$	$9.36*10^{6}$	$1.48*10^{11}$	Calculated (CCME 2008b)
K_{OC}	*L/kg*	$2.51*10^{5}$	$5.01*10^{6}$	$6.31*10^{8}$	$1.00*10^{13}$	CCME 2008b, Table B.1; Meridian 2010b, Table 4
Half-life	*d*	$1.75*10^{3}$	$1.75*10^{3}$	$1.75*10^{3}$	$1.75*10^{3}$	CCME 2008b, Table C.4, Table C.4; Meridian 2010b, Table 4
Solubility in water	*mg/L*	$3.40*10^{-2}$	$7.60*10^{-4}$	$2.50*10^{-6}$	$1.26*10^{11}$	CCME 2008b, Table B.1; Meridian 2010b, Table 4
NAPL water partition coefficient	*L/kg*	$4.57*10^{7}$	$2.56*10^{9}$	$9.92*10^{11}$	$2.93*10^{17}$	Meridian 2010b, Table 4
Concentration in NAPL (as % of F2 or F3)[1]	*% wt*	36.0	44.0	56.0	24.0	CCME 2008b, Table C.4
Viscosity	*cSt*	2.80	2.80	2.80	2.80	Meridian 2009a, Table 1
Diffusion coefficient in water[2]	cm^2/s	$1.00*10^{-5}$	$1.00*10^{-5}$	$1.00*10^{-5}$	$1.00*10^{-5}$	CCME 2008b, Table C.4

Aromatic components	**Unit**	**C>10-C12**	**C>12-C16**	**C>16-C21**	**C>21-C34**	**Reference**
Molecular weight	*g/mol*	$1.30*10^{2}$	$1.50*10^{2}$	$1.80*10^{2}$	$2.50*10^{2}$	CCME 2008b, Table B.1
Retardation factor (coarse soil)	-	60.6	$1.20*10^{2}$	$3.76*10^{2}$	$2.99*10^{3}$	Calculated (CCME 2008b)
Retardation factor (fine soil)	-	38.2	7.53*10	$2.35*10^{2}$	$1.87*10^{3}$	Calculated (CCME 2008b)
K_{OC}	*L/kg*	$2.51*10^{3}$	$5.01*10^{3}$	$1.58E*10^{4}$	$1.26*10^{5}$	CCME 2008b, Table B.1
Half-life	*d*	$1.75*10^{3}$	$1.75*10^{3}$	$1.75*10^{3}$	$1.75*10^{3}$	CCME 2008b; Meridian 2010b, Table 4
Solubility in water	*mg/L*	25.0	5.80	0.65	$6.60*10^{-3}$	CCME 2008b, Table B.1
NAPL water partition coefficient	*L/kg*	$5.05*10^{4}$	$2.51*10^{5}$	$2.69*10^{6}$	$3.68*10^{8}$	Meridian 2010b, Table 4
Concentration in NAPL (as proportion of F2 or F3)	*% wt*	9.00	11.0	14.0	6.00	CCME 2008b, Table C.4
Viscosity	*cSt*	2.80	2.80	2.80	2.80	Meridian 2009a, Table 1
Diffusion coefficient in water	cm^2/s	$1.00*10^{-5}$	$1.00*10^{-5}$	$1.00*10^{-5}$	$1.00*10^{-5}$	CCME 2008b, Table C.4

[1] The C>10-C12 and C>12-C16 compounds are included in F2; the C>16-C21 et C>21-C34 compounds are included in F3.
[2] Based on the value for air multiplied by 10^{-5}.

Table III-4 Chemical and physical parameters of canola biodiesel methyl esters

Parameter	Unit	Methyl behenate	Methyl erucate	Methyl tetracosanate	Methyl palmitate	Methyl palmitoleate	Methyl arachidate	Methyl eicosinate	Reference
Molecular Weight	*g/mol*	$3.55*10^{2}$	$3.53*10^{2}$	$3.83*10^{2}$	$2.71*10^{2}$	$2.69*10^{2}$	$3.27*10^{2}$	$3.25*10^{2}$	a
Retardation factor (coarse soil)	-	$3.06*10^{4}$	$1.75*10^{4}$	$5.84*10^{4}$	$4.37*10^{2}$	$6.70*10^{2}$	$9.45*10^{3}$	$9.45*10^{3}$	b
Retardation factor (fine soil)	-	$1.91*10^{4}$	$1.09*10^{4}$	$3.65*10^{4}$	$2.73*10^{2}$	$4.19*10^{2}$	$5.91*10^{3}$	$5.91*10^{3}$	b
K_{OC}	*L/kg*	$1.29*10^{6}$	$7.36*10^{5}$	$2.46*10^{6}$	$1.84*10^{4}$	$2.82*10^{4}$	$3.98*10^{5}$	$3.98*10^{5}$	c
Half-life	*d*	$8.75*10^{2}$	$8.75*10^{2}$	$8.75*10^{2}$	$8.75*10^{2}$	$8.75*10^{2}$	$8.75*10^{2}$	$8.75*10^{2}$	c
Solubility in water	*mg/L*	$1.13*10^{-5}$	$6.56*10^{-6}$	$1.00*10^{-6}$	$1.10*10^{-2}$	$2.29*10^{-2}$	$1.00*10^{-4}$	$1.03*10^{-4}$	c
NAPL water partition coefficient	*L/kg*	$1.07*10^{11}$	$1.83*10^{11}$	$1.30*10^{12}$	$8.37*10^{7}$	$3.99*10^{7}$	$1.11*10^{10}$	$1.08*10^{10}$	b
Concentration in NAPL	*% wt*	0.10	0.80	0.10	4.50	0.20	0.10	0.50	d
Viscosity	*cSt*	$5.00^{\#}$	$5.00^{\#}$	$5.00^{\#}$	4.38	3.67	$5.00^{\#}$	$5.00^{\#}$	c
Diffusion coefficient in water	cm^2/s	$3.39*10^{-6}$	$3.41*10^{-6}$	$3.23*10^{-6}$	$4.02*10^{-6}$	$4.06*10^{-6}$	$3.56*10^{-6}$	$3.60*10^{-6}$	e

Parameter	Unit	Methyl stearate	Methyl oleate (cis) & elaidate (trans)	Methyl linoleate	Methyl α- & γ-linolenate	Reference
Molecular weight	*g/mol*	$2.99*10^{2}$	$2.97*10^{2}$	$2.95*10^{2}$	$2.93*10^{2}$	a
Retardation factor (coarse soil)	-	$1.48*10^{3}$	$1.48*10^{3}$	$1.48*10^{3}$	$1.48*10^{3}$	b
Retardation factor (fine soil)	-	$9.26*10^{2}$	$9.26*10^{2}$	$9.26*10^{2}$	$9.26*10^{2}$	b
K_{OC}	*L/kg*	$6.24*10^{4}$	$6.24*10^{4}$	$6.24*10^{4}$	$6.24*10^{4}$	c
Half-life	*d*	$8.75*10^{2}$	$8.75*10^{2}$	$8.75*10^{2}$	$8.75*10^{2}$	c
Solubility in water	*mg/L*	$1.00*10^{-3}$	$4.10*10^{-3}$	$1.30*10^{-2}$	$3.50*10^{-2}$	c
NAPL water partition coefficient	*L/kg*	$1.02*10^{9}$	$2.46*10^{8}$	$7.71*10^{7}$	$2.84*10^{7}$	b
Concentration in NAPL	*% wt*	1.90	60.8	22.2	8.80	d
Viscosity	*cSt*	5.85	4.51	3.65	3.14	c
Diffusion coefficient in water	cm^2/s	$3.77*10^{-6}$	$3.81*10^{-6}$	$3.85*10^{-6}$	$3.89*10^{-6}$	e

$^{\#}$ Based on equivalent carbon relationships (Meridian 2009b, Table 1); Reference: a. Meridian 2009a, Table 2A; b. Calculated (CCME 2008b); c. Meridian 2009a, Table 5A; d. Meridian 2009a, Table 9; e. US EPA 2010a

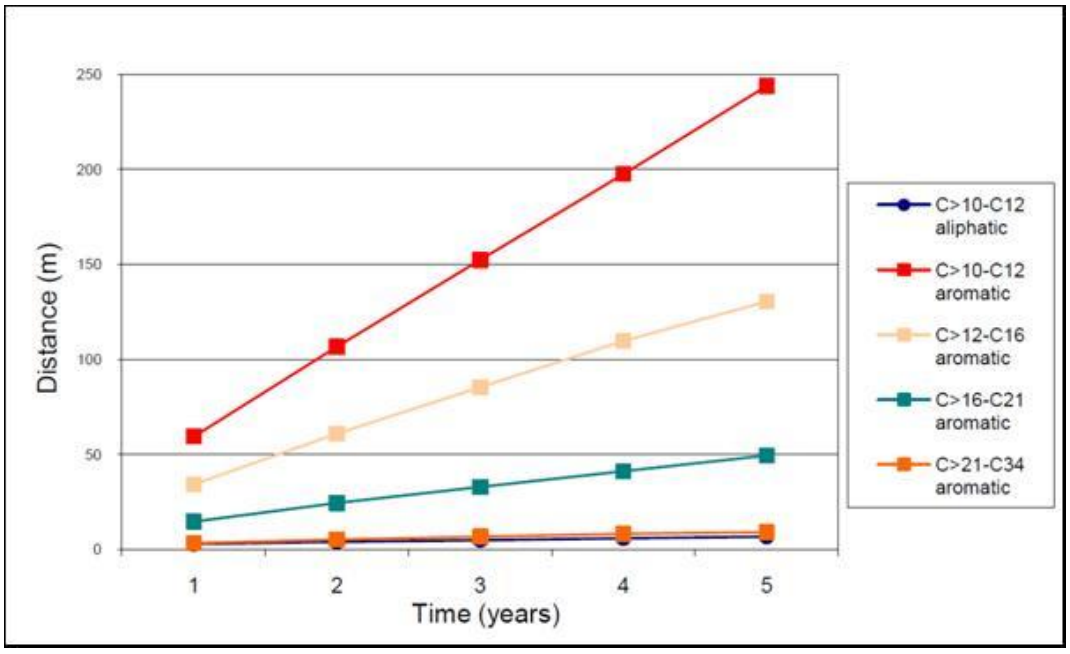

Figure III-1 UST - Maximum predicted ULSD plume length in the direction of groundwater flow for coarse-grained soil after 1 to 5 years, based on an initial source concentration of 10 mg/L with BIOSCREEN-AT[3]

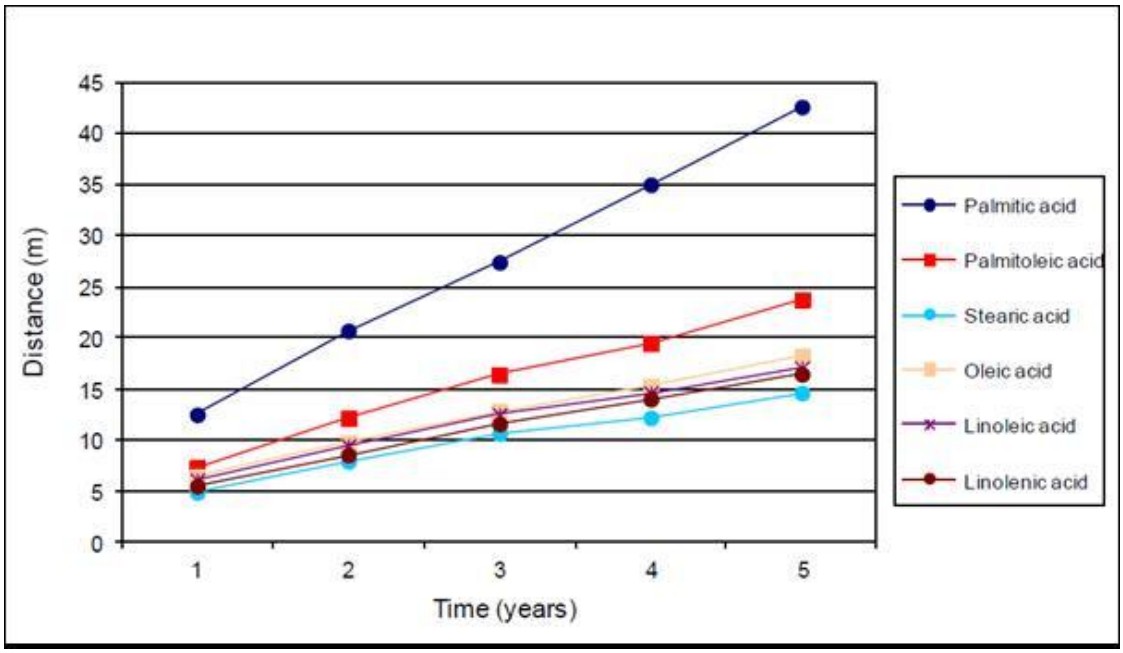

Figure III-2 UST - Maximum predicted canola B100 plume length in the direction of groundwater flow for coarse-grained soil based on an initial source concentration of 10 mg/L after 1 to 5 years with BIOSCREEN-AT

[3] *Note*: Components with predicted distances less than 3 meters are not shown. *Source*: Meridian 2010b

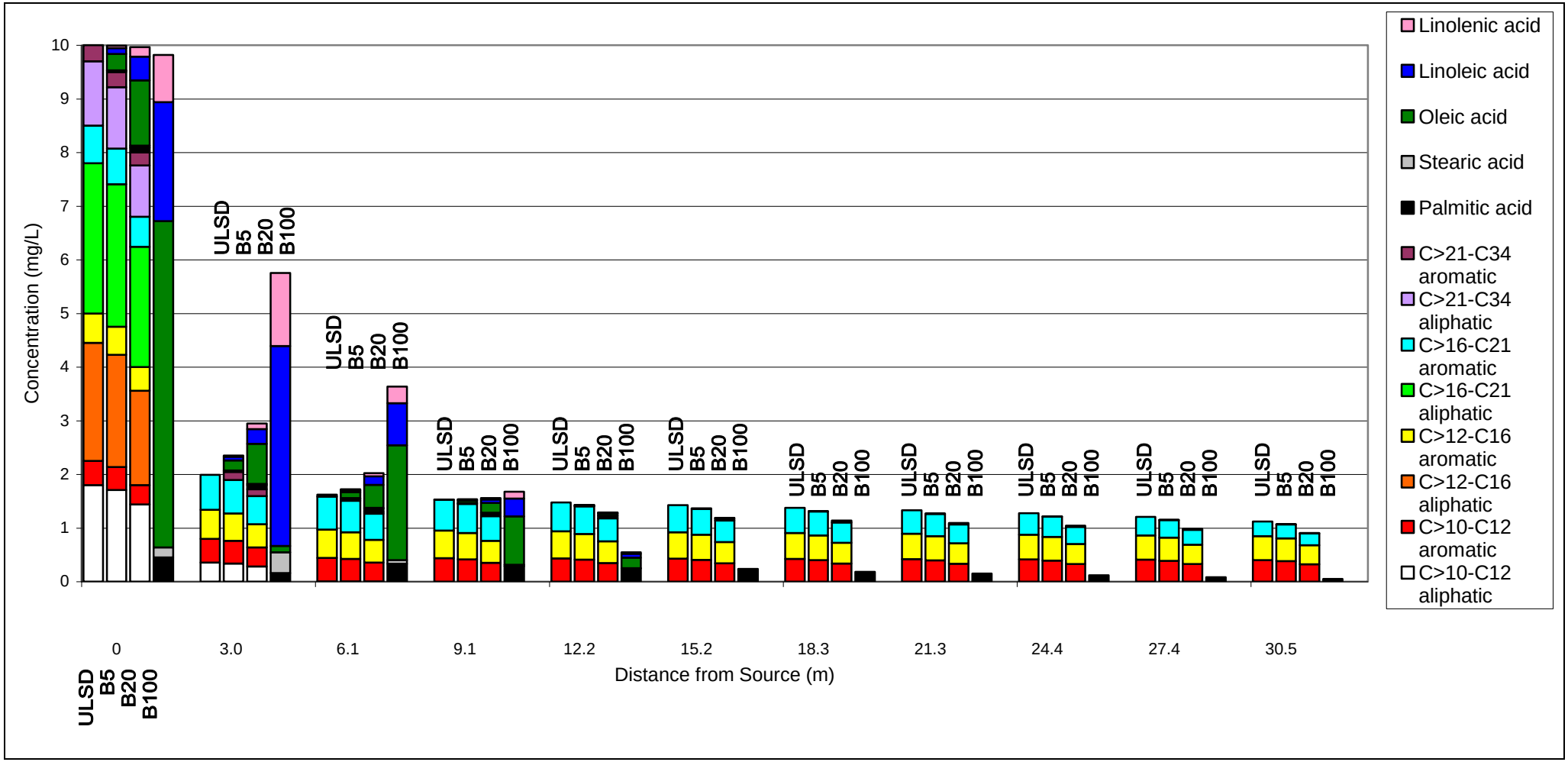

Figure III-3 UST - Predicted plume composition and concentration for coarse-grained soil after five years, for ULSD, B5, B20, and B100 with BIOSCREEN-AT

Appendix V. MOBILE6.2C results

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Table V-1 TPM, PM_{10}, and $PM_{2.5}$ emissions (in tonnes) and emission variations for biodiesel blends in comparison to a B0 scenario (in %) for HDDVs and all MOBILE6.2C sources in 2006, 2010, 2015, and 2020

		HDDV				All MOBILE6.2C					
		TPM/PM_{10} exhaust*		$PM_{2.5}$ exhaust		PM_{10} exhaust		$PM_{2.5}$ exhaust		TPM exhaust	
		tonnes	%	tonnes	%	tonnes	%	tonnes	%	tonnes	%
2006	B0	6954.60	0.00	6399.49	0.00	12135.34	0.00	10837.39	0.00	12272.08	0.00
	B1	6910.61	-0.63	6358.98	-0.63	12091.35	-0.36	10796.88	-0.37	12228.09	-0.36
	B2	6866.60	-1.27	6318.16	-1.27	12047.34	-0.73	10756.06	-0.75	12184.08	-0.72
	B5	6734.43	-3.17	6197.22	-3.16	11915.17	-1.81	10635.12	-1.87	12051.91	-1.79
	B20	6077.69	-12.61	5592.55	-12.61	11258.43	-7.23	10030.45	-7.45	11395.17	-7.15
2010	B0	4197.68	0.00	3863.55	0.00	8525.65	0.00	7666.91	0.00	8642.21	0.00
	B1	4171.56	-0.62	3839.50	-0.62	8499.53	-0.31	7642.86	-0.31	8616.09	-0.30
	B2	4146.02	-1.23	3815.68	-1.24	8473.99	-0.61	7619.04	-0.62	8590.55	-0.60
	B5	4067.57	-3.10	3743.26	-3.11	8395.54	-1.53	7546.62	-1.57	8512.10	-1.51
	B20	3678.59	-12.37	3384.69	-12.39	8006.56	-6.09	7188.05	-6.25	8123.12	-6.01
2015	B0	1836.12	0.00	1690.77	0.00	5234.97	0.00	4803.12	0.00	5329.02	0.00
	B1	1826.38	-0.53	1680.93	-0.58	5225.23	-0.19	4793.28	-0.20	5319.28	-0.18
	B2	1816.19	-1.09	1671.74	-1.13	5215.04	-0.38	4784.09	-0.40	5309.09	-0.37
	B5	1784.51	-2.81	1643.11	-2.82	5183.36	-0.99	4755.46	-0.99	5277.41	-0.97
	B20	1630.03	-11.22	1501.02	-11.22	5028.88	-3.94	4613.37	-3.95	5122.93	-3.87
2020	B0	889.92	0.00	820.26	0.00	4200.21	0.00	3859.95	0.00	4294.76	0.00
	B1	886.37	-0.40	816.36	-0.48	4196.66	-0.08	3856.05	-0.10	4291.21	-0.08
	B2	882.29	-0.86	812.73	-0.92	4192.58	-0.18	3852.42	-0.20	4287.13	-0.18
	B5	870.86	-2.14	802.34	-2.18	4181.15	-0.45	3842.03	-0.46	4275.70	-0.44
	B20	813.19	-8.62	748.48	-8.75	4123.48	-1.83	3788.17	-1.86	4218.03	-1.79
* For HDDVs, TPM and PM_{10} results are identical.											

Table V-2 NO_X and CO emissions (in tonnes) and emission variations for biodiesel blends in comparison to a B0 scenario (in %) for HDDVs and all MOBILE6.2C sources in 2006, 2010, 2015, and 2020

		HDDV				All MOBILE6.2C			
		NO_X		CO		NO_X		CO	
		tonnes	%	tonnes	%	tonnes	%	tonnes	%
2006	B0	278474.16	0.00	60577.70	0.00	529644.75	0.00	4360359.60	0.00
	B1	278973.49	0.18	60253.16	-0.54	530144.08	0.09	4360035.06	-0.01
	B2	279475.36	0.36	59927.01	-1.07	530645.95	0.19	4359708.91	-0.01
	B5	280978.71	0.90	58950.80	-2.69	532149.30	0.47	4358732.70	-0.04
	B20	288497.89	3.60	54072.18	-10.74	539668.48	1.89	4353854.08	-0.15
2010	B0	196172.03	0.00	41095.09	0.00	399516.68	0.00	3454964.59	0.00
	B1	196523.27	0.18	40886.39	-0.51	399867.92	0.09	3454755.89	-0.01
	B2	196874.80	0.36	40676.77	-1.02	400219.45	0.18	3454546.27	-0.01
	B5	197928.98	0.90	40044.21	-2.56	401273.63	0.44	3453913.71	-0.03
	B20	203200.56	3.58	36893.11	-10.23	406545.21	1.76	3450762.61	-0.12
2015	B0	100834.85	0.00	20708.39	0.00	231714.62	0.00	2772745.27	0.00
	B1	101000.51	0.16	20618.71	-0.43	231880.28	0.07	2772655.59	0.00
	B2	101170.34	0.33	20530.00	-0.86	232050.11	0.14	2772566.88	-0.01
	B5	101671.91	0.83	20260.38	-2.16	232551.68	0.36	2772297.26	-0.02
	B20	104186.70	3.32	18916.41	-8.65	235066.47	1.45	2770953.29	-0.06
2020	B0	54392.83	0.00	13114.01	0.00	144267.54	0.00	2549485.32	0.00
	B1	54470.82	0.14	13071.47	-0.32	144345.53	0.05	2549442.78	0.00
	B2	54547.70	0.28	13028.86	-0.65	144422.41	0.11	2549400.17	0.00
	B5	54776.28	0.70	12899.59	-1.64	144650.99	0.27	2549270.90	-0.01
	B20	55925.10	2.82	12258.70	-6.52	145799.81	1.06	2548630.01	-0.03

Table V-3 VOC (total, exhaust, acetaldehyde, formaldehyde, acrolein) and THC emissions and emission variations for biodiesel blends in comparison to a B0 scenario (in %) for HDDVs in 2006, 2010, 2015, and 2020

		VOC total/exhaust*		Acetaldehyde		Formaldehyde		Acrolein		THC total	
		tonnes	%	kg	%	kg	%	kg	%	kg	%
2006	B0	10544.45	0.00	318118.58	0.00	863802.07	0.00	38656.18	0.00	10690.04	0.00
	B1	10451.13	-0.89	315286.58	-0.89	856130.92	-0.89	38303.57	-0.91	10594.42	-0.89
	B2	10358.00	-1.77	312468.46	-1.78	848448.10	-1.78	37977.69	-1.76	10500.15	-1.78
	B5	10075.85	-4.44	303984.29	-4.44	825399.67	-4.45	36932.65	-4.46	10214.37	-4.45
	B20	8669.10	-17.79	261565.05	-17.78	710216.92	-17.78	31789.39	-17.76	8790.06	-17.77
2010	B0	8523.47	0.00	257273.96	0.00	698503.01	0.00	31251.59	0.00	8644.78	0.00
	B1	8464.32	-0.69	255513.12	-0.68	693774.82	-0.68	31052.61	-0.64	8585.85	-0.68
	B2	8407.56	-1.36	253776.06	-1.36	689046.85	-1.35	30838.69	-1.32	8528.22	-1.35
	B5	8235.06	-3.38	248551.63	-3.39	674878.14	-3.38	30192.25	-3.39	8351.57	-3.39
	B20	7370.53	-13.53	222439.25	-13.54	604008.67	-13.53	27030.40	-13.51	7473.32	-13.55
2015	B0	7089.39	0.00	214011.81	0.00	581103.09	0.00	26008.17	0.00	7192.86	0.00
	B1	7063.52	-0.36	213219.90	-0.37	578978.77	-0.37	25907.03	-0.39	7165.14	-0.39
	B2	7036.51	-0.75	212450.78	-0.73	576851.19	-0.73	25820.20	-0.72	7137.30	-0.77
	B5	6959.36	-1.83	210102.08	-1.83	570510.15	-1.82	25529.86	-1.84	7060.73	-1.84
	B20	6570.61	-7.32	198412.42	-7.29	538760.83	-7.29	24110.05	-7.30	6668.25	-7.29
2020	B0	6621.07	0.00	199954.30	0.00	542912.44	0.00	24294.16	0.00	6717.60	0.00
	B1	6611.37	-0.15	199598.54	-0.18	541986.14	-0.17	24252.99	-0.17	6707.92	-0.14
	B2	6599.91	-0.32	199259.48	-0.35	541031.77	-0.35	24221.45	-0.30	6695.43	-0.33
	B5	6567.91	-0.80	198212.94	-0.87	538194.77	-0.87	24097.01	-0.81	6660.01	-0.86
	B20	6392.20	-3.46	193014.43	-3.47	524061.55	-3.47	23461.29	-3.43	6485.82	-3.45
* For HDDVs, VOC total and VOC exhaust results are identical. Also, benzene and 1,3-butadiene emission variations (in %) are the same as the VOCs in this table.											

Table V-4 PAH emissions (in kg) and emission variations for biodiesel blends in comparison to a B0 scenario (in %) for all MOBILE6.2C sources in 2006, 2010, 2015, and 2020

		Benzo[a] anthracene		Benzo[a]pyrene		Chrysene		Fluoranthene		Indeno(1,2,3-cd)pyrene		Naphthalene		Pyrene	
		net	%	net	%	net	%	net	%	net	%	net	%	net	%
2006	B0	739.64	0.00	564.79	0.00	533.53	0.00	4512.93	0.00	358.58	0.00	455028.25	0.00	6197.81	0.00
	B1	738.26	-0.19	564.24	-0.10	533.35	-0.03	4512.16	-0.02	358.52	-0.02	454971.52	-0.01	6195.94	-0.03
	B2	736.72	-0.39	563.64	-0.20	532.77	-0.14	4511.06	-0.04	358.43	-0.04	454915.32	-0.02	6194.64	-0.05
	B5	731.43	-1.11	562.07	-0.48	531.98	-0.29	4508.54	-0.10	358.31	-0.08	454745.20	-0.06	6189.79	-0.13
	B20	707.47	-4.35	553.58	-1.98	527.89	-1.06	4495.18	-0.39	358.02	-0.16	453897.91	-0.25	6166.21	-0.51
2010	B0	563.71	0.00	457.27	0.00	438.94	0.00	3775.64	0.00	303.20	0.00	385160.82	0.00	5176.96	0.00
	B1	562.56	-0.20	456.98	-0.06	438.87	-0.02	3775.09	-0.01	303.17	-0.01	385127.73	-0.01	5176.17	-0.02
	B2	561.74	-0.35	456.75	-0.11	438.68	-0.06	3774.53	-0.03	303.17	-0.01	385093.89	-0.02	5175.27	-0.03
	B5	558.81	-0.87	455.94	-0.29	438.08	-0.20	3772.95	-0.07	303.05	-0.05	384993.73	-0.04	5172.58	-0.08
	B20	544.57	-3.40	451.22	-1.32	435.37	-0.81	3765.01	-0.28	302.60	-0.20	384492.23	-0.17	5158.58	-0.36
2015	B0	392.73	0.00	345.60	0.00	338.40	0.00	2967.84	0.00	241.30	0.00	303310.75	0.00	4061.10	0.00
	B1	392.42	-0.08	345.44	-0.05	338.33	-0.02	2967.70	0.00	241.30	0.00	303297.33	0.00	4060.64	-0.01
	B2	391.99	-0.19	345.30	-0.09	338.30	-0.03	2967.36	-0.02	241.30	0.00	303283.80	-0.01	4060.23	-0.02
	B5	390.92	-0.46	344.96	-0.19	338.14	-0.08	2966.72	-0.04	241.26	-0.02	303244.34	-0.02	4059.40	-0.04
	B20	385.25	-1.90	343.20	-0.69	337.16	-0.37	2963.52	-0.15	241.20	-0.04	303044.83	-0.09	4053.53	-0.19
2020	B0	354.57	0.00	331.47	0.00	328.03	0.00	2907.50	0.00	239.50	0.00	298686.03	0.00	3975.72	0.00
	B1	354.36	-0.06	331.41	-0.02	328.00	-0.01	2907.38	0.00	239.50	0.00	298681.38	0.00	3975.57	0.00
	B2	354.12	-0.13	331.32	-0.05	327.96	-0.02	2907.29	-0.01	239.50	0.00	298676.34	0.00	3975.43	-0.01
	B5	353.65	-0.26	331.25	-0.07	327.85	-0.05	2907.18	-0.01	239.50	0.00	298661.33	-0.01	3975.12	-0.02
	B20	351.43	-0.89	330.66	-0.24	327.61	-0.13	2905.94	-0.05	239.50	0.00	298586.43	-0.03	3972.98	-0.07

Table V-5 Provincial HDDV emissions of selected contaminants (in tonnes or kilograms) with biodiesel use and emission reductions (in %), in 2006

Region	Scenario	TPM/PM_{10} (tonnes)	$PM_{2.5}$ (tonnes)	NO_X (tonnes)	VOC total (tonnes)	CO (tonnes)	Benzene (kg)	1,3-butadiene (kg)	Formaldehyde (kg)	THC exhaust (kg)
AB	B0	1414.98	1302.26	53808.29	2096.94	12482.73	23068.12	13397.45	171741.9	2125.21
	B5	1371.02	1261.34	54292.39	2004.79	12147.34	22030.08	12794.4	164104	2030.56
		-3.11%	-3.14%	0.90%	-4.39%	-2.69%	-4.50%	-4.50%	-4.45%	-4.45%
	B20	1238.44	1139.47	55745.01	1724.75	11142.7	18960.67	11017.25	141204.6	1747.35
		-12.48%	-12.50%	3.60%	-17.75%	-10.74%	-17.81%	-17.77%	-17.78%	-17.78%
BC	B0	701.71	645.63	28671.18	1133.17	6211.87	12472.26	7247.34	92898.5	1149.61
	B5	679.12	625	28929.3	1082.84	6044.81	11921.5	6923.53	88769.84	1098.81
		-3.22%	-3.20%	0.90%	-4.44%	-2.69%	-4.42%	-4.47%	-4.44%	-4.42%
	B20	612.2	563.52	29703.23	931.46	5544.35	10253.15	5953.54	76384.36	945.34
		-12.76%	-12.72%	3.60%	-17.80%	-10.75%	-17.79%	-17.85%	-17.78%	-17.77%
ON	B0	1633.93	1503.54	78903.98	2927.88	15915.46	32220.8	18710.07	239955.9	2970.24
	B5	1580.61	1454.7	79613.82	2797.27	15488.53	30790.64	17886.55	229289.9	2837.66
		-3.26%	-3.25%	0.90%	-4.46%	-2.68%	-4.44%	-4.40%	-4.44%	-4.46%
	B20	1421.65	1308.41	81744.73	2406.64	14206.45	26496.24	15390.15	197288.1	2441.76
		-12.99%	-12.98%	3.60%	-17.80%	-10.74%	-17.77%	-17.74%	-17.78%	-17.79%
QC	B0	1651.67	1519.92	57508.97	2279.7	13156.85	25057.62	14562.77	186661.9	2309.71
	B5	1599.95	1472.27	58025.27	2177.54	12803.47	23945.91	13910.71	178356.1	2207.41
		-3.13%	-3.14%	0.90%	-4.48%	-2.69%	-4.44%	-4.48%	-4.45%	-4.43%
	B20	1445.8	1330.04	59578.82	1874.1	11743.59	20605.47	11973.33	153470.9	1899.3
		-12.46%	-12.49%	3.60%	-17.79%	-10.74%	-17.77%	-17.78%	-17.78%	-17.77%

Table V-6 Provincial HDDV emissions of selected contaminants (in tonnes or kilograms) with biodiesel use and emission reductions (in %), in 2020

Region	Scenario	TPM/PM_{10} (tonnes)	$PM_{2.5}$ (tonnes)	NO_X (tonnes)	VOC total (tonnes)	CO (tonnes)	Benzene (kg)	1,3-butadiene (kg)	Formaldehyde (kg)	THC exhaust (kg)
AB	B0	202.76	186.83	13476.69	1440.17	3032.86	15863.92	9221.15	118115.92	1460.94
	B5	198.10	182.28	13576.68	1427.44	2979.68	15687.34	9123.33	116920.39	1446.35
		-2.30%	-2.44%	0.74%	-0.88%	-1.75%	-1.11%	-1.06%	-1.01%	-1.00%
	B20	184.25	169.50	13874.74	1382.46	2823.77	15221.35	8845.96	113343.01	1402.42
		-9.13%	-9.28%	2.95%	-4.01%	-6.89%	-4.05%	-4.07%	-4.04%	-4.01%
BC	B0	87.71	80.76	5088.86	691.53	1262.8	7617.34	4422.09	56718.83	701.89
	B5	85.60	78.88	5123.19	686.74	1243.14	7552.76	4388.64	56261.28	696.03
		-2.41%	-2.33%	0.67%	-0.69%	-1.56%	-0.85%	-0.76%	-0.81%	-0.83%
	B20	80.25	73.83	5229.13	669.71	1184.54	7373.23	4282.65	54886.33	679.12
		-8.51%	-8.58%	2.76%	-3.16%	-6.20%	-3.20%	-3.15%	-3.23%	-3.24%
ON	B0	183.07	169.26	9379.93	1927.98	2758.51	21218.03	12326.15	158082.91	1956.19
	B5	180.42	166.63	9429.19	1921.57	2729.23	21141.66	12283.90	157466.82	1948.57
		-1.45%	-1.55%	0.53%	-0.33%	-1.06%	-0.36%	-0.34%	-0.39%	-0.39%
	B20	172.90	159.36	9576.19	1898.07	2640.93	20894.04	12138.68	155621.27	1926.18
		-5.56%	-5.85%	2.09%	-1.55%	-4.26%	-1.53%	-1.52%	-1.56%	-1.53%
QC	B0	202.67	186.95	12464.84	1296.32	2945.12	14277.79	8290.14	106284.76	1315.42
	B5	198.21	182.73	12557.58	1283.09	2892.84	14122.54	8205.02	105160.94	1301.44
		-2.20%	-2.26%	0.74%	-1.02%	-1.78%	-1.09%	-1.03%	-1.06%	-1.06%
	B20	183.77	169.24	12834.50	1241.48	2733.36	13663.60	7934.09	101797.39	1260.56
		-9.33%	-9.47%	2.97%	-4.23%	-7.19%	-4.30%	-4.29%	-4.22%	-4.17%

Appendix VI-A Tables and Figures

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Table VI-1 Industrial sources of air pollution in Canada considered in the emissions inventory for use in AURAMS

Industrial Sources		
Abrasives Manufacture	Asbestos Industry	Asphalt Paving Industry
Bakeries	Grain Industries	Pulp and Paper Industry
Cement and Concrete Industry • Cement manufacture • Lime manufacture • Concrete batching & products	Wood Industry • Sawmills • Panel board mills • Other wood products	Foundries • Die casting • Ferrous foundries • Non-ferrous foundries
Aluminum Industry • Primary aluminum smelting & refining • Primary aluminum smelting & refining • Secondary aluminum (Includes recycling)	Downstream Petroleum Industry • Other downstream Petroleum industry • Petroleum refining • Refined petroleum Products bulk storage and distribution	Iron and Steel Industries • Other (Iron and steel industries) • Primary (Blast furnace and DRI) • Secondary (Electric arc furnaces) • Steel recycling
Mining and Rock Quarrying • Coal mining industry • Rock, sand and gravel • Metal mining • Other minerals • Potash	Petroleum Product Transportation and Distribution • Natural gas distribution • Natural gas transmission • Petroleum product pipelines	Non-Ferrous Mining and Smelting Industry • Primary Ni, Cu, Zn, Pb • Secondary Pb, Cu • Other metals
Chemicals Industry • Chemical manufacture • Fertilizer production • Other (Chemical industries) • Paint & varnish manufacturing • Petrochemical industry • Plastics & synthetic resins fabrication	Other Industries • Electronics • Food preparation • Glass manufacture • Metal fabrication • Other (Other industries) • Plastics manufacture • Textiles • Vehicle manufacture (Engines, Parts, Assembly, Painting) • Paint and varnish formulation	Upstream Petroleum Industry • Oil sands in-situ extraction and processing • Oil sands mining extraction and processing • Bitumen and heavy oil upgrading • Petroleum liquids storage • Crude oil and natural gas production and processing • Other upstream petroleum industry
Mineral Products Industry • Clay products • Brick products • Other mineral products	Iron Ore Mining Industry • Pelletizing • Iron ore mining industry	

Table VI-2 Mobile sources of air pollution in Canada considered in the emissions inventory for use in AURAMS

Mobile Sources		
Air Transportation Landing / Take-off	Light-Duty Gasoline Vehicles	Off-Road Use of Gasoline/LPG/CNG
Rail Transportation	Light-Duty Gasoline Trucks	Tire Wear & Brake Lining
Heavy-Duty Diesel Vehicles	Light-Duty Diesel Vehicles	Marine Transportation
Heavy-Duty Gasoline Trucks	Motorcycles	
Light-Duty Diesel Trucks	Off-Road Use of Diesel	

Table VI-3 Non-industrial, Open, Natural, Incineration, and Miscellaneous sources of air pollution in Canada considered in the emissions inventory for use in AURAMS

Non-Industrial Sources	Miscellaneous Sources	Open Sources
Electric Power Generation (Utilities) • Coal • Natural gas • Other (EPG) Commercial Fuel Combustion Residential Fuel Combustion Residential Fuel Wood Combustion	Cigarette Smoking Dry Cleaning General Solvent Use Marine Cargo Handling Industry Meat Cooking Refined Petroleum Products Retail Printing Structural Fires Surface Coatings Human Other Miscellaneous Sources	Agriculture • Animals • Agriculture tilling and wind erosion • Fertilizer application Construction Operations Dust from Paved Roads Dust from Unpaved Roads Waste • Landfills • Water and sewage treatment • Energy from waste • Open burning Mine Tailings
Incineration Sources	**Natural Sources**	
Crematorium Industrial & Commercial Incineration Municipal Incineration Other Incineration & Utilities	Natural Sources • Biogenics (Vegetation, soils)	

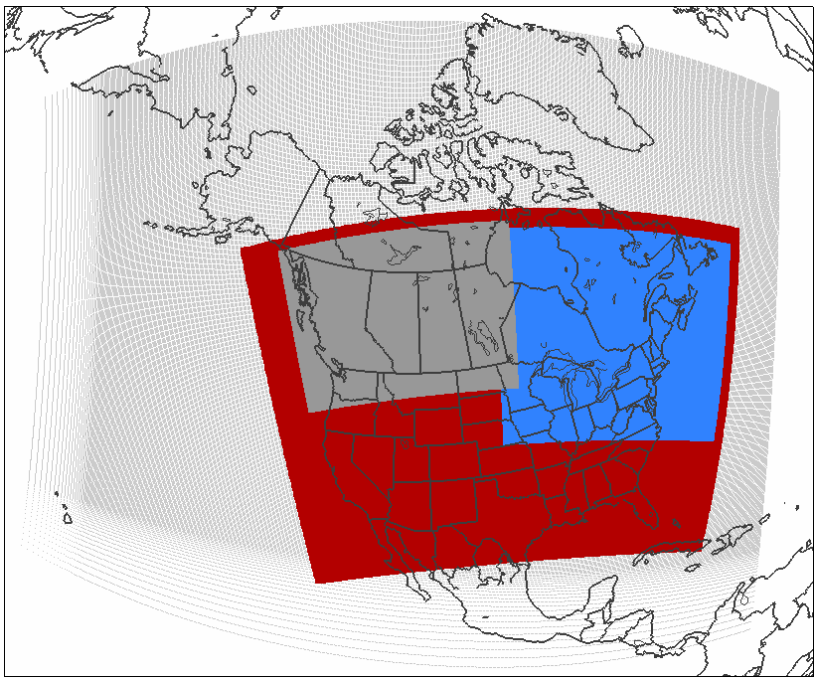
Table VI-4 Canadian inventory of CAC emissions from the on-road transportation sector under the B0, B5 and B20 scenarios in 2006, as used in AURAMS

Scenario	CLASS	POLLUTANT (in tonnes)						
		CO	NH_3	NO_X	PM_{10}	$PM_{2.5}$	SO_2	VOC
On-road transportation B0	HDDV	60,578	640	278,474	7,803	6,662	205	10,544
	HDGV	119,225	233	24,438	1,154	902	77	8,203
	LDDT	3,642	24	4,686	516	433	12	2,012
	LDDV	1,730	11	1,323	175	142	3	431
	LDGT	2,129,930	8,476	114,077	3,827	2,311	652	126,408
	LDGV	2,025,798	11,270	105,094	3,729	2,133	625	132,339
	MC	19,457	13	1,552	63	38	3	3,701
	B0 total	4,360,360	20,666	529,645	17,267	12,621	1,579	283,639
On-road transportation B5	HDDV	58,951	640	280,979	7,583	6,459	205	10,076
	HDGV	119,225	233	24,438	1,154	902	77	8,203
	LDDT	3,642	24	4,686	516	433	12	2,012
	LDDV	1,730	11	1,323	175	142	3	431
	LDGT	2,129,930	8,476	114,077	3,827	2,311	652	126,408
	LDGV	2,025,798	11,270	105,094	3,729	2,133	625	132,339
	MC	19,457	13	1,552	63	38	3	3,701
	B5 total	4,358,733	20,666	532,149	17,047	12,418	1,579	283,170
On-road transportation B20	HDDV	54,072	640	288,498	6,926	5,855	205	8,669
	HDGV	119,225	233	24,438	1,154	902	77	8,203
	LDDT	3,642	24	4,686	516	433	12	2,012
	LDDV	1,730	11	1,323	175	142	3	431
	LDGT	2,129,930	8,476	114,077	3,827	2,311	652	126,408
	LDGV	2,025,798	11,270	105,094	3,729	2,133	625	132,339
	MC	19,457	13	1,552	63	38	3	3,701
	B20 total	4,353,854	20,666	539,668	16,390	11,814	1,579	281,763
Total – other sectors[4]		**4,978,243**	**512,059**	**1,747,055**	**2,025,182**	**526,842**	**1,961,754**	**1,943,075**

[4] Represents the total emission estimates from sectors other than on-road transportation. See Tables A-1 to A-3 in Appendix VI-A for a complete listing of categories.

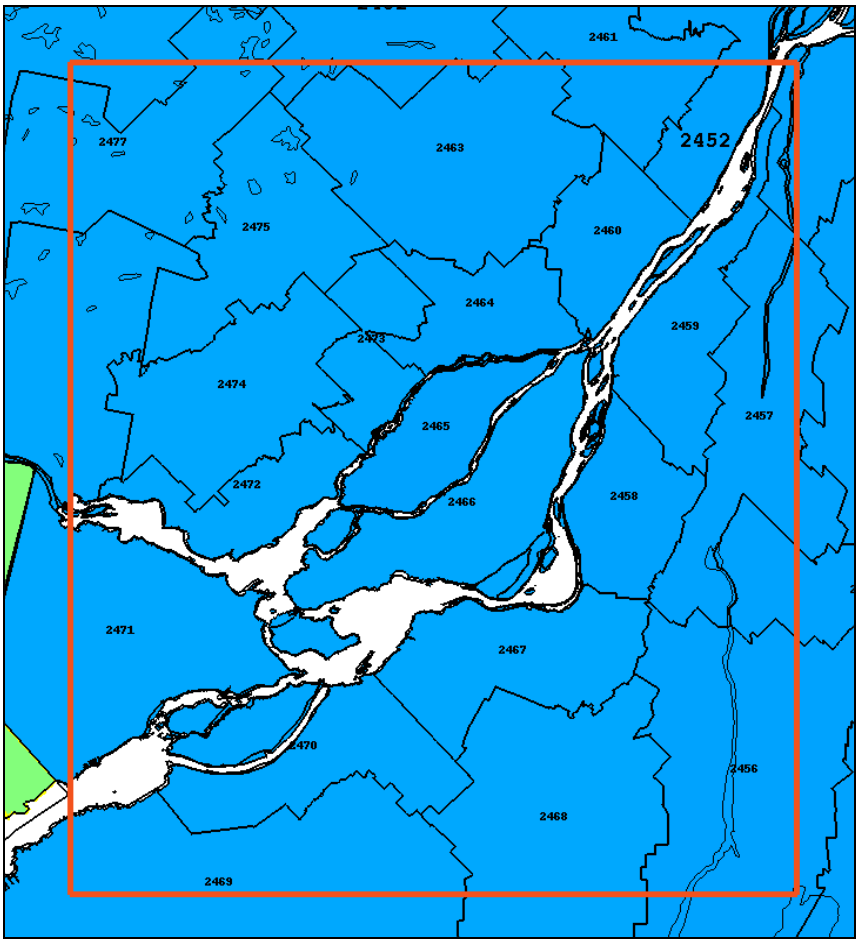
Table VI-5 Canadian inventory of CAC emissions from the on-road transportation sector under the B0, B5 and B20 scenarios in 2020, as used in AURAMS

Scenario	CLASS	POLLUTANT (in tonnes)						
		CO	NH_3	NO_X	PM_{10}	$PM_{2.5}$	SO_2	VOC
On-road transportation B0	HDDV	13,114	784	54,393	1,931	1,142	249	6,621
	HDGV	61,315	286	6,778	484	351	92	3,229
	LDDT	2,053	31	1,402	179	111	15	880
	LDDV	1,293	13	169	69	41	4	135
	LDGT	1,130,314	10,531	46,309	3,338	1,880	644	51,014
	LDGV	1,319,304	13,712	34,239	4,356	2,457	621	52,729
	MC	22,092	16	977	76	45	4	2,141
	B0 total	2,549,485	25,373	144,268	10,433	6,026	1,630	116,749
On-road transportation B5	HDDV	12,900	784	54,776	1,912	1,124	249	6,568
	HDGV	61,315	286	6,778	484	351	92	3,229
	LDDT	2,053	31	1,402	179	111	15	880
	LDDV	1,293	13	169	69	41	4	135
	LDGT	1,130,314	10,531	46,309	3,338	1,880	644	51,014
	LDGV	1,319,304	13,712	34,239	4,356	2,457	621	52,729
	MC	22,092	16	977	76	45	4	2,141
	B5 total	2,549,271	25,373	144,651	10,414	6,008	1,630	116,696
On-road transportation B20	HDDV	12,259	784	55,925	1,854	1,070	249	6,392
	HDGV	61,315	286	6,778	484	351	92	3,229
	LDDT	2,053	31	1,402	179	111	15	880
	LDDV	1,293	13	169	69	41	4	135
	LDGT	1,130,314	10,531	46,309	3,338	1,880	644	51,014
	LDGV	1,319,304	13,712	34,239	4,356	2,457	621	52,729
	MC	22,092	16	977	76	45	4	2,141
	B20 total	2,548,630	25,373	145,800	10,357	5,954	1,630	116,520
Total – other sectors		**6,306,047**	**586,642**	**1,570,612**	**2,656,112**	**653,579**	**1,652,195**	**2,117,568**



Notes: *Red*: Continental domain; *Blue*: Eastern domain; *Grey*: Western domain. The lightly shaded area covering most of North America represents the region covered the by the meteorological driver GEM.

Figure VI-1 AURAMS domains



Beauharnois-Salaberry (CD2470)
Thérèse-De Blainville (CD2473)
Pays-d'en-Haut (CD2477)
Vallée-du-Richelieu (CD2457)
Jardins-de-Napierville (CD2468)
Rivière-du-Nord (CD2475)
Laval (CD2465)
Mirabel (CD2474)
Montréal (CD2466)
Deux-Montagnes (CD2472)
Haut-Richelieu (CD2456)
L'Assomption (CD2460)
Roussillon (CD2467)
Rouville (CD2455)
Lajemmerais (CD2459)
Les Moulins (CD2464)
Vaudreuil-Soulanges (CD2471)
Montcalm (CD2463)
Longueuil (CD2458)

Figure VI-2 Census divisions included in the GRILLE domain (red outline)

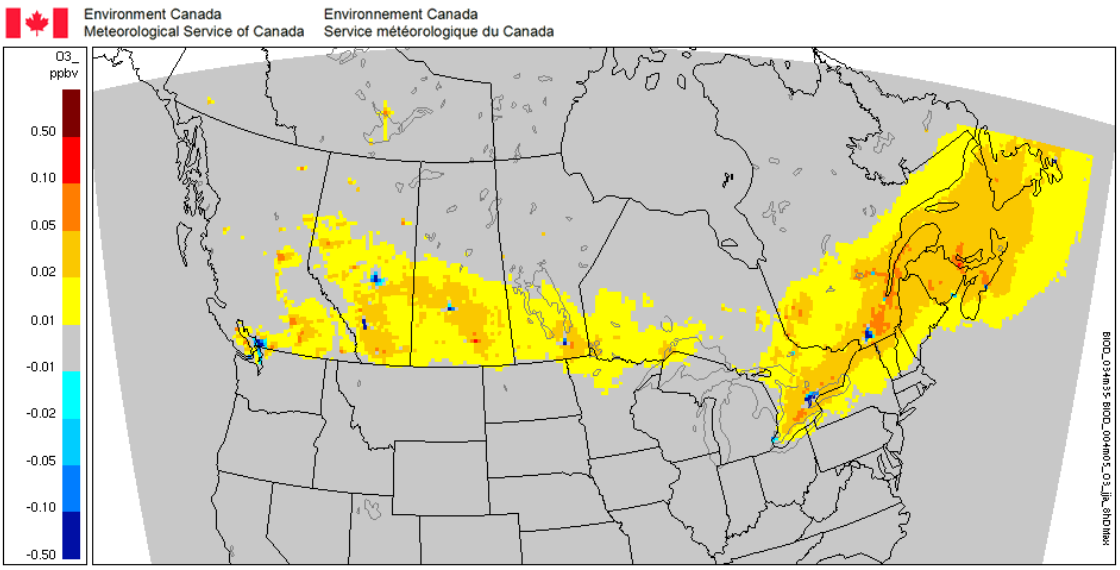


Figure VI-3 Change in ozone 8-hr average daily maxima summer (June-July-August) concentrations under a B20 scenario in 2006

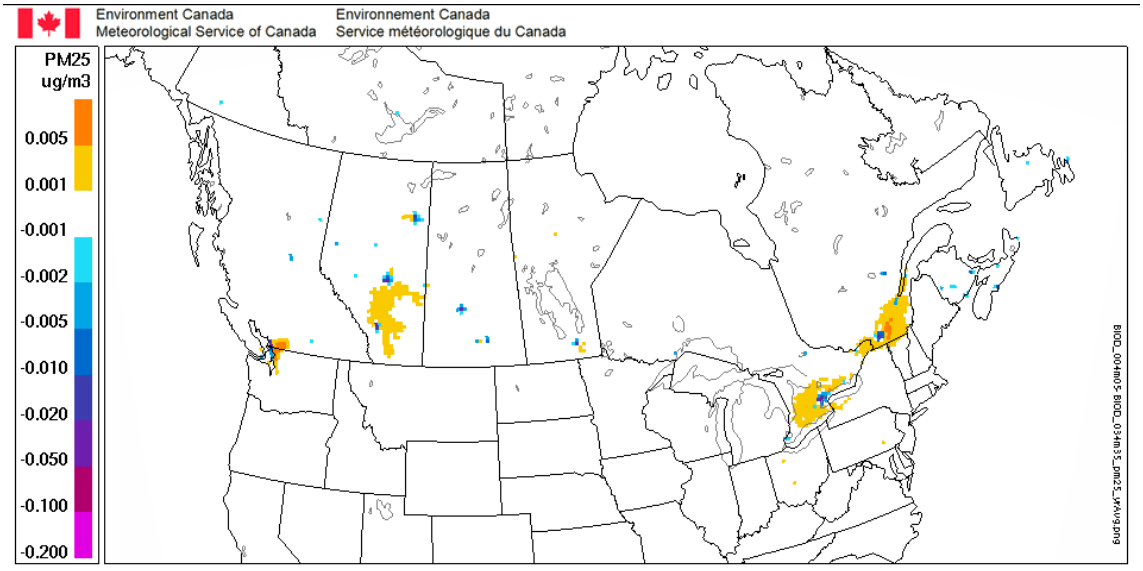


Figure VI-4 Change in $PM_{2.5}$ annual average concentrations under a B20 scenario in 2006

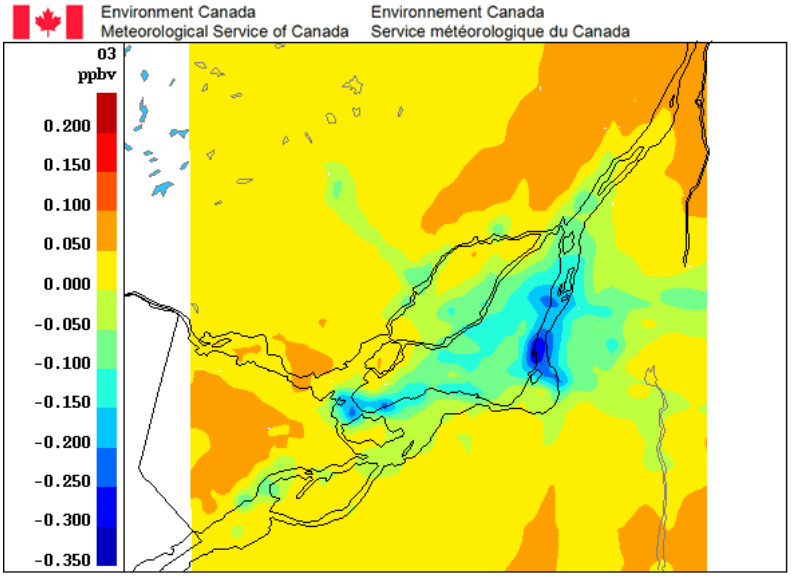


Figure VI-5 Change in O_3 8-hr average daily maxima concentrations under a B20 scenario for the June 12 to 23, 2006 episode

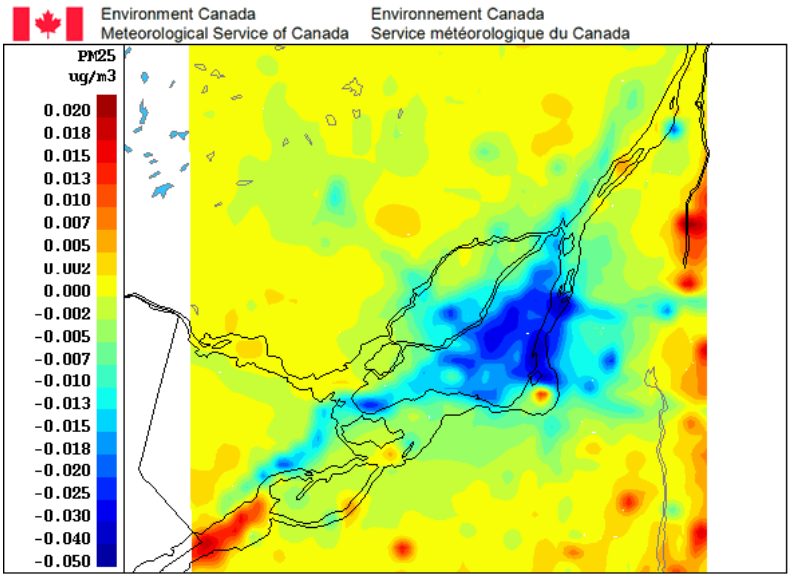


Figure VI-6 Change in $PM_{2.5}$ average concentrations under a B20 scenario for the June 12 to 23, 2006 episode

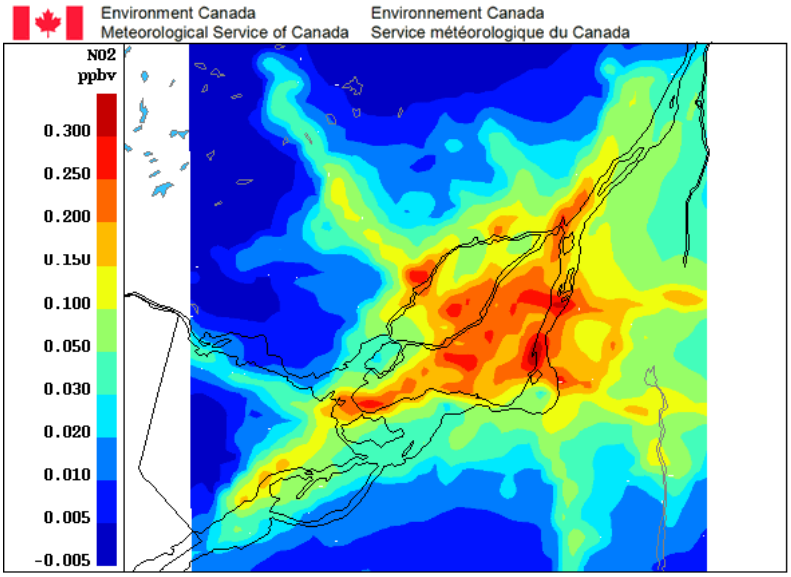


Figure VI-7 Change in NO_2 average concentrations under a B20 scenario for the June 12 to 23, 2006 episode

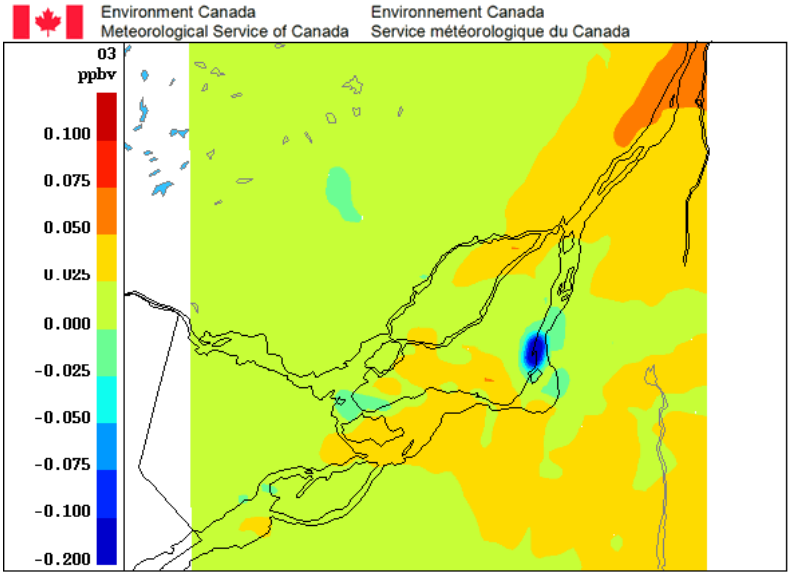


Figure VI-8 Change in O_3 8-hr average daily maxima concentrations under a B20 scenario for the June 12 to 23, 2020 episode

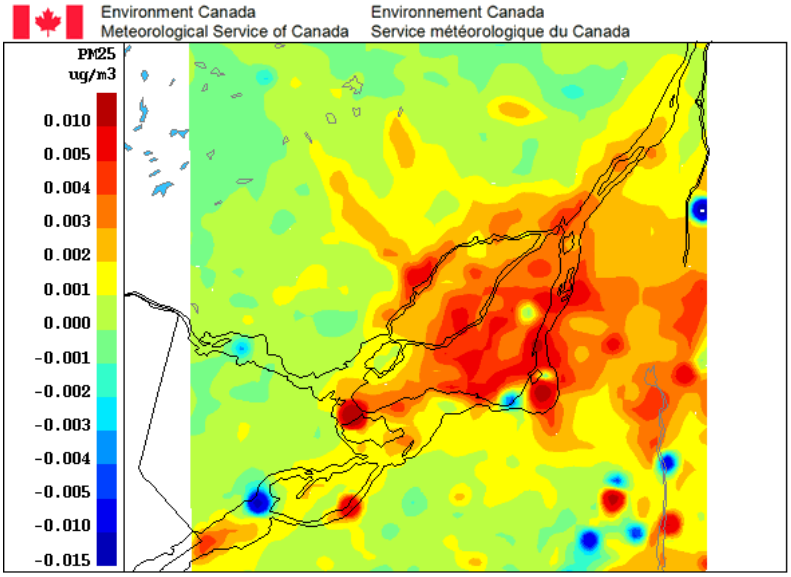


Figure VI-9 Change in $PM_{2.5}$ average concentrations under a B20 scenario for the June 12 to 23, 2020 episode

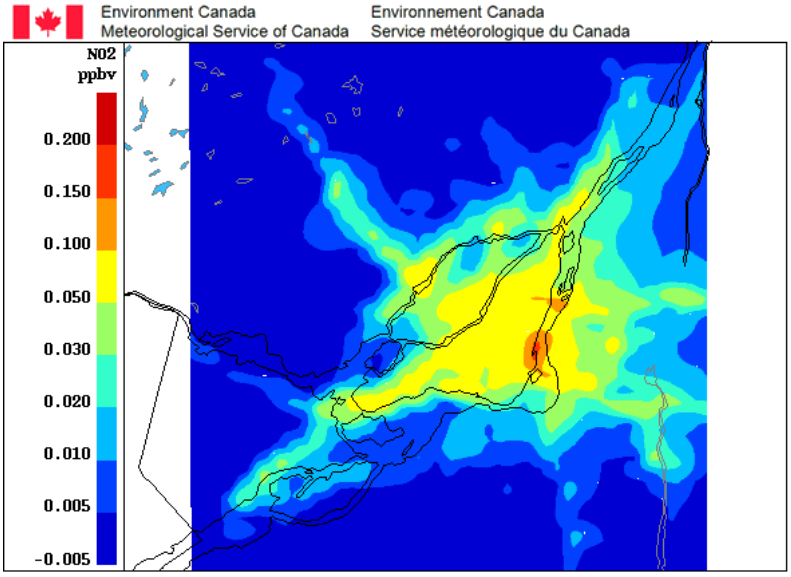


Figure VI-10 Change in NO_2 average concentrations under a B20 scenario for the June 12 to 23, 2020 episode