

**Western Canada Study of Animal Health Effects  
Associated with Exposure to Emissions  
from Oil and Natural Gas Field Facilities**

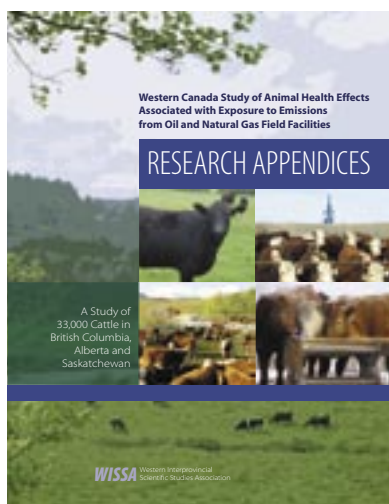
# TECHNICAL SUMMARY

A Study of  
33,000 Cattle in  
British Columbia,  
Alberta and  
Saskatchewan



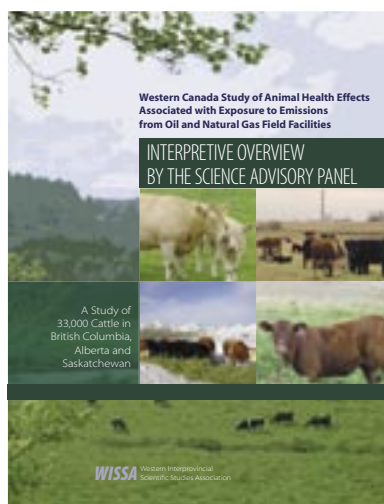
THE PUBLIC REPORT of the Western Interprovincial Scientific Studies Association (WISSA) presents the results of the Western Canada Study of Animal Health Effects Associated with Exposure to Emissions from Oil and Natural Gas Field Facilities.

This is one of four publications that comprise the public report. The others are as follows:



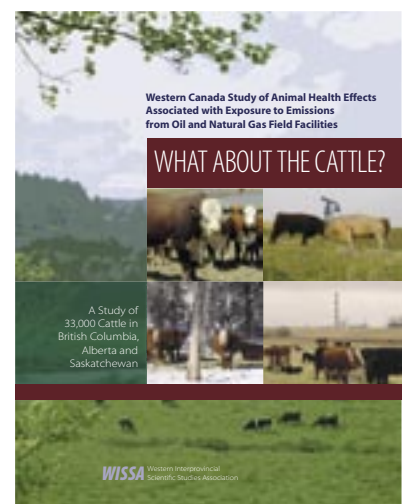
### **Research Appendices**

A collection of 17 research appendices that presents the details of all research for the study



### **The Interpretive Overview by the Science Advisory Panel**

The views of the Science Advisory Panel on the interpretation and significance of the study findings



### **What About the Cattle?**

Highlights of the study

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# Executive Summary

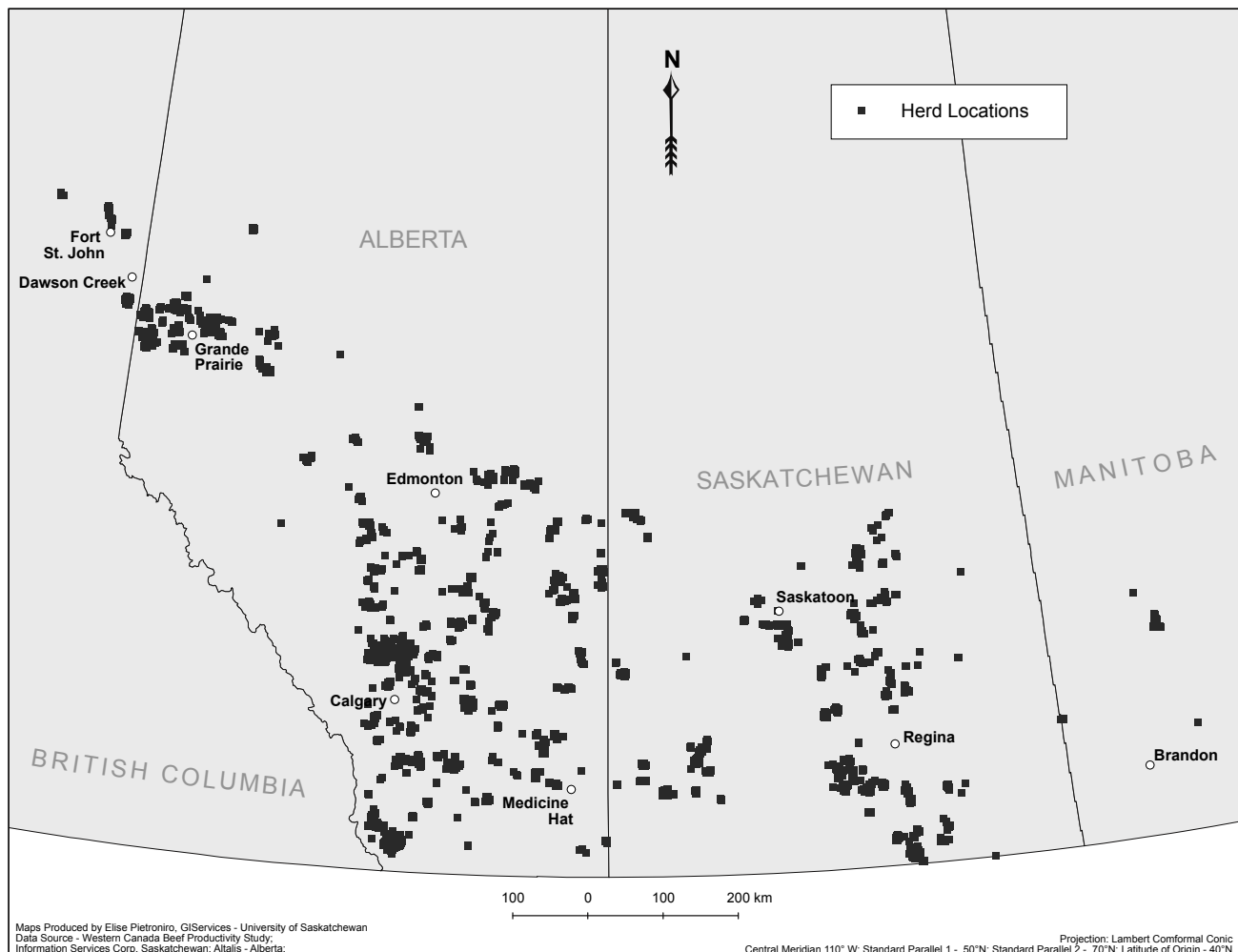
In the largest on-farm study of its kind undertaken to date, 33,000 cows in 205 beef herds from Alberta, Saskatchewan, and northeastern British Columbia were closely monitored over a 2-year period to examine the association between exposure to emissions from the oil and natural gas industry and animal health and productivity.

The study was prompted by long-standing concerns on the part of cattle producers that emissions from industry field facilities, often located on pasture, wintering, and calving areas, have contributed to reproductive failure and disease in their herds.

The study was designed to investigate the potential association between exposure to emissions and reproductive success in beef herds. Other study questions included

whether emissions were associated with effects on the immune, respiratory, and nervous systems of calves. The potential effects of emissions on the reproductive and immune system health of the European starling, a sentinel wildlife species which shared the same environment as the cattle, were also examined.

The study found no associations between exposure to emissions and whether or not the cows became pregnant, the frequency of abortions or stillbirths, as well as most of the other measures of animal health and productivity examined in either cattle or starlings. However, the highest measured concentrations of some emissions were associated with a few important indicators of animal health and productivity.



Reported location of study herds, April 2001 through November 30, 2002.

Note: Cattle were placed on the pastures in Manitoba near the end of the study after the completion of calving season in 2002. Air quality was not measured at these sites.

The most notable finding was an increase in the risk of mortality observed for calves born to cows from areas with the highest observed exposures to SO<sub>2</sub>. Some measures of exposure were also associated with a small increase in the time to calving for the cows, and also reports of calf treatment with pharmaceuticals, the occurrence of respiratory lesions in calves, and a decrease in a few types of specialized white blood cells.

The wildlife study found a small decrease in the rate of bone growth in starling nestlings from the most exposed study sites.

## Herds

The study involved close observation of herds from cow-calf operations across Western Canada to assess possible adverse impacts of industry exposure, primarily on reproduction and calf survival.

The study also examined the potential for effects on the immune system of calves and yearlings as well as on the presence of lesions in the respiratory, immune, and nervous systems of aborted, stillborn and live-born calves that died.

It provided a unique opportunity to describe other indicators of health and productivity in cow-calf herds in the region. This information will provide a valuable baseline for assessing reproductive performance and the occurrence of disease in western Canadian cow-calf herds.

The herds, which varied in size from 50 to more than 200 cows, were selected with the help of veterinarians from more than 60 private veterinary clinics in the study area such that they represented a broad range of exposures to different levels of emissions from facilities in major cattle and oil- and gas-producing areas of Western Canada. The cattle herds were monitored closely where they were pastured and wintered, to accurately determine their exposure status. Producers were selected to obtain a broad geographic coverage and to maximize the range of observed exposures. Other selection criteria included the producer's interest in the study, the availability of herd records, and a working relationship with a local veterinary clinic.

Data on the cows, collected with the help of records kept by herd owners, included the results of pregnancy testing, the number of days between calvings for each cow, whether the cow aborted, whether or not the calf died at birth or before it was 3 months old, and whether or not the producer reported treating the calf with a pharmaceutical.

The locations of land to which each cow had access during each month of the study were recorded and used in determining the extent to which each animal was exposed to industry emissions. Calf records were linked to the calving records of their dams.

## Emissions

The components of emissions selected for study included sulphur dioxide (SO<sub>2</sub>), hydrogen sulphide (H<sub>2</sub>S), volatile organic compounds (VOCs), fine particulate matter (PM<sub>1.0</sub>), and polycyclic aromatic hydrocarbons (PAHs).

These chemicals can be found in airborne emissions from facilities such as oil and gas wells, batteries, compressor stations, and gas plants, and some have been associated with adverse health effects in previous environmental and occupational studies.

During the field component of the study, concentrations of SO<sub>2</sub> and VOCs were measured more than 9,800 times at 1,200 sites; H<sub>2</sub>S was measured more than 6,300 times at 1,100 sites; and particulates and PAHs were measured 365 times at 32 sites.

This represents the largest compilation of measurements of these air quality parameters in rural western Canada to date, and is a valuable contribution to our understanding and knowledge of air quality in the region.

## Air Monitors

Two types of air-monitors were used to measure the emission components selected for study. The passive type, in which air moves across a diffusion membrane, monitored airborne concentrations of SO<sub>2</sub>, H<sub>2</sub>S, and VOCs. The active type, in which a solar-charged electric pump drew air through a filter, and a filter plus a column filled

## Animal Health Effects

### Associated with Exposure to Emissions from Oil- and Gas-Field Facilities

 association not found
  association found
  association not studied

	SO <sub>2</sub>	VOCs as benzene H <sub>2</sub> S	VOCs as toluene	PAHs	Well-Site Density PM <sub>1.0</sub>	
<b>Associations Addressing PRIMARY STUDY HYPOTHESES—CATTLE</b>						
Greater risk of non-pregnancy [RA 6]						
Longer breeding-to-calving interval [RA 6]						
Greater risk of abortion* [RA 7]						
Greater risk of stillbirth* [RA 7]						
Greater risk of mortality in calves < 3 months old (prenatal exposure - first bull exposure to calving date) [RA 8]						
Greater risk of mortality in calves < 3 months old (their dams exposed last trimester) [RA 8]						
Greater risk of mortality in calves < 3 months old (exposure during 1st month) [RA 8]						
<b>Associations Addressing SECONDARY STUDY HYPOTHESES—CATTLE</b>						
Greater risk of lesions with pre- and postnatal exposure—calf immune systems [RA 10]						
Greater risk of lesions with pre- and postnatal exposure—calf nervous systems [RA 10]						
Greater risk of lesions with postnatal exposure only—calf immune systems [RA 10]						
Greater risk of lesions with postnatal exposure only—calf nervous systems [RA 10]						
Greater risk of lesions with postnatal exposure only—calf respiratory systems [RA 10]						
Lower count of B-cells in yearling cattle immune systems [RA 11&12]						
Lower count of CD4 T-cells in yearling cattle immune systems [RA 11&12]						
Lower count of CD8 T-cells in yearling cattle immune systems [RA 11&12]						
Lower count of γδ T-cells in yearling cattle immune systems [RA 11&12]						
Lower count of WC1 T-cells in yearling cattle immune systems [RA 11&12]						
Lower response to vaccine in yearling cattle immune systems [RA 11&12]						
Lower count of B-cells—calf immune system health [RA 13]						
Lower count of CD4 T-cells—calf immune system health [RA 13]						
Lower count of CD8 T-cells—calf immune system health [RA 13]						
Lower count of γδ T-cells—calf immune system health [RA 13]						
Lower count of WC1 T-cells—calf immune system health [RA 13]						
<b>Associations Addressing OTHER STUDY HYPOTHESES</b>						
Greater risk of cow disposal (includes culling or death loss) [RA 6]						
Greater risk of calves receiving treatment < 3 months old (prenatal exposure) [RA 8]						
Greater risk of calves receiving treatment < 3 months old (dams exposed last trimester) [RA 8]						
Greater risk of calves receiving treatment after first month (postnatal exposure) [RA 8]						
Greater risk of lesions with pre- and postnatal exposure—calf skeletal or cardiac muscle [RA 10]						
Greater risk of lesions with pre- and postnatal exposure—calf thyroid glands [RA 10]						
Greater risk of lesions with postnatal exposure only—calf skeletal or cardiac muscle [RA 10]						
<b>Associations Addressing SECONDARY STUDY HYPOTHESES—AVIAN</b>						
Lower hatching success [RA 14]						
Lower fledging success [RA 14]						
Shorter tarsal length [RA 14]						
Shorter first primary feather [RA 14]						
Lower IgM antibody production [RA 15]						
Smaller spleen [RA 15]						
Smaller bursa of Fabricius [RA 15]						
Lower IgY antibody production [RA 15]						
Lower T-cell response to phytohemagglutinin [RA 15]						
Higher heterophil/lymphocyte (H/L) ratios [RA 15]						

RA refers to the applicable research appendix in the *Research Appendices*

\* H<sub>2</sub>S was not monitored before September 2001.

with absorbent material, monitored airborne concentrations of PM<sub>1.0</sub> and PAHs in a sample of study herds.

Passive air monitors were placed on or as close as possible to all pastures used by the cattle. The passive stations were relocated, or new monitoring stations set up, each month if the cattle were moved, to collect data in locations as close as possible to the summer, winter, and calving areas of the herds. More monitoring stations were needed in summer, when cattle were dispersed on pasture.

Passive samplers and the collectors from the pump-operated monitors were retrieved from the monitoring stations and replaced monthly, then shipped to the central office. After removing information identifying the location of the monitoring station, the samples were forwarded by the air-monitoring contractor to the laboratories for analysis.

At the conclusion of air monitoring, both statistical modeling of emissions and comparisons with data collected in Alberta airshed monitoring zones demonstrated that the passive monitors performed adequately for the range of concentrations examined, and that the measured concentrations of SO<sub>2</sub> at most exposed study sites were representative of data from areas of the province with a high potential for exposure.

## Relating Emissions and Exposure

In combination with data on animal locations, data on emission concentrations collected by the closest monitor or monitors were linked to each cow for each month of the study to determine her exposure for that month.

Oil and gas well sites within 1.6 km of the center of each location used by the cattle were counted to provide a value of well-site “density” that was used as an additional measure of animal exposure to industry emissions.

## Key Findings

The study was primarily designed to investigate whether emissions from oil and natural gas facilities were associated with effects on the reproductive performance of the cattle. Secondary study questions included whether emissions were associated with effects on the immune, respiratory, and nervous systems of calves.

Complementary research was conducted on the reproductive and immune system health of European starlings, a bird species that has been used in environmental research as an indicator species for studying the biological effects of contaminants.

Investigations into the potential effects of exposure on animal health are listed in the matrix, in order of primary and secondary study questions. During the course of the study additional questions arose and the results of those analyses are also listed.

The primary question in this study was whether emissions from oil and gas industry facilities were associated with reproductive losses in cattle. The study found that the frequency of non-pregnancy, abortions, or stillbirths did not differ across the measured range of exposures.

Exposure to most emissions was not associated with a longer interval between breeding and calving. The breeding-to-calving interval was, however, about 3 days longer for cows most exposed to VOCs measured as benzene, compared to the least exposed cows.

Exposure to most emissions was also not associated with a greater risk of calves dying. However, there was a small increase in the risk of dying for calves from cows in areas exposed to the highest measured concentrations of SO<sub>2</sub> during gestation and near the time of calving. For example, for a typical bull calf born to a heifer during the peak of the calving season and in good weather conditions, the risk of dying would be 1.8% higher in the most exposed calf (5.9%), compared to the least exposed calf (4.1%).

The study also found that exposure was not associated with changes to most measures of immune system health in calves and yearlings. However, statistically



significant associations were found between exposure to some VOCs and the numbers of circulating specialized white blood cells (CD4 and CD8 T-lymphocytes) in calves and yearlings.

Increasing exposure to VOCs was associated with a statistically significant increase in the risk of pathological changes in the respiratory system of older calves when examined *post mortem*. In contrast, increasing exposures to SO<sub>2</sub> and H<sub>2</sub>S were not associated with changes in the respiratory system.

Exposure was not associated with changes in the nervous system of calves when examined *post mortem*.

In a parallel investigation, exposure to emissions was not associated with the health of the immune system of European starlings, the reproductive success of the birds, or nestling development as measured by feather growth. Exposure was, however, associated with a small, but statistically significant, decrease in bone growth.

The finding of statistically significant associations does not necessarily indicate cause-and-effect relationships. Such conclusions would require other supporting evidence.

For details on the interpretation of the study findings and their significance, see the *Interpretive Overview by the Science Advisory Panel*, which forms part of WISSA's public report.

## Long-Term Benefits

Given the large number of animals studied and the wide geographic area included, the study provides valuable insights into reproductive performance and the occurrence of disease in cow-calf herds across Western Canada.

Study data provide background information, both on animal health and exposure, which should prove useful in future studies of similar issues in cattle.

# Introduction

The impact of exposure to emissions from oil- and gas-field facilities on animal as well as human health has been a long standing concern in Western Canada. Oil and gas facilities, such as well sites, batteries, compressor stations, and gas plants, are often located on land used to pasture or winter cattle. Although the direct impact of exposure to emissions from these facilities on the animals is uncertain, cattle producers have expressed concerns about lower pregnancy rates, increased numbers of abortions, and greater incidence of disease.

The Western Canada Study of Animal Health Effects Associated with Exposure to Emissions from Oil and Natural Gas Field Facilities was undertaken in response to public concern about the possible effects of petroleum industry facilities on animal health. It was initiated in 1999 by the governments of the four western provinces, which set up the Western Interprovincial Scientific Studies Association (WISSA), an arm's-length organization to manage and oversee the study.

WISSA is a federally incorporated, not-for-profit organization. One of its roles in the study was to avoid potential bias from industry, government, and interest groups, and thus ensure a high degree of public and scientific credibility.

WISSA's Board of Directors consists of appointees from each of the western provinces. It has ultimate responsibility for managing and directing the affairs of WISSA and is accountable to governments, funding partners, other stakeholders, and the public. The Board is assisted in technical matters by a Science Advisory Panel, drawn from experts in the various fields of research that apply to the study.

## Productivity and Health

The study was primarily designed to examine associations between emissions and important reproductive parameters in beef cattle, including pregnancy rates, the frequencies of abortions and stillbirths, and the risk of death in young calves. It also examined the effects of emissions on the time from one calving to the next, another measure of herd reproductive success.

A second study question was whether emissions were associated with effects on the respiratory, immune, and nervous systems of calves and yearlings. Samples collected during postmortem examinations of calves that had died were used to determine the frequency of different types of pathological changes in these systems.

An additional series of studies of yearlings and calves explored the potential for effects of emissions on the immune system using different methods. These studies compared counts of circulating specialized white blood cells among yearlings and calves with different histories of exposure to emissions, and examined the immune system's response to vaccination in yearlings.

### Study Timelines

<b>1999</b>	<ul style="list-style-type: none"><li>• Deputy Ministers of Environment from the western provinces identified the need</li><li>• Intergovernmental Ad Hoc Committee struck</li><li>• Co-chairs of the Science Advisory Panel (SAP) chosen</li></ul>
<b>2000</b>	<ul style="list-style-type: none"><li>• First study plan developed</li><li>• Western Interprovincial Scientific Studies Association (WISSA) established</li><li>• Responsibility for the study transferred from Ad Hoc Committee to WISSA Board of Directors</li><li>• SAP members appointed</li><li>• Team at University of Saskatchewan designed study</li><li>• SAP reviewed and refined detailed study design</li></ul>
<b>2001 - 2002</b>	<ul style="list-style-type: none"><li>• Cattle monitored, starting in breeding season</li><li>• Air monitoring by RWDI AIR Inc., Calgary, AB (RWDI) began</li><li>• Field work by team at University of Saskatchewan</li><li>• SAP provided oversight and quality assurance</li></ul>
<b>2002 - 2003</b>	<ul style="list-style-type: none"><li>• Calving season data collected</li><li>• Air monitoring continued</li><li>• SAP provided oversight and quality assurance</li></ul>
<b>2003 - 2004</b>	<ul style="list-style-type: none"><li>• Data analysis and evaluation, and statistical analyses, carried out by team at University of Saskatchewan</li><li>• SAP reviewed results of analyses, proposed further refinements, and provided quality assurance</li><li>• Additional analyses of exposure data, proposed by SAP, conducted by RWDI and University of Alberta</li></ul>
<b>2005</b>	<ul style="list-style-type: none"><li>• Research appendices prepared by team at University of Saskatchewan, University of Alberta and RWDI</li><li>• SAP reviewed findings with researchers and provided oversight and interpretive guidance</li></ul>
<b>2006</b>	<ul style="list-style-type: none"><li>• Preparation of research appendices continued</li><li>• WISSA public report completed and released</li></ul>

Monitoring the concentrations of various airborne contaminants from facilities provided essential information for determining the exposure of individual animals. Air-monitoring data were also used to obtain an indication of whether emissions could affect wildlife in the area, through studies of the reproduction and immune functions of a wild bird species, the European starling.

## Study Personnel

Research was carried out under the direction of two principal investigators at the University of Saskatchewan: Dr. Cheryl Waldner, Western College of Veterinary Medicine, and Dr. Mark Wickstrom, Toxicology Centre.

Dr. Waldner was the project manager and principal investigator for the beef cattle productivity study, and supervised the collection of field data for the beef cattle immunotoxicology study and the exposure assessment and statistical analysis for the immunotoxicology and avian studies. Dr. Wickstrom was the principal investigator for the beef cattle immunology and the avian studies.

The air-monitoring component of the study was undertaken by a 20-member team from RWDI Air, Inc., Calgary, AB which deployed and maintained air-sampling equipment, managed collection of sampler data, and administered the quality assurance and chain-of-custody protocol for the samples. Three contract laboratories analysed the samples. Dr. Igor Burstyn of the University of Alberta analysed WISSA's air-monitoring data.

Dr. Ted Clark, Prairie Diagnostic Services, Saskatoon, SK, a pathologist certified by the American College of Veterinary Pathologists, completed detailed histological analyses of the samples submitted from field postmortem examinations.

Dr. Henrik Stryhn, a biostatistician from University of Prince Edward Island, provided advice on aspects of the statistical analysis.

In addition to the above, the study team included three graduate students, six project veterinarians employed by the university, and private veterinarians from more than 60 clinics that provided veterinary services to study participants. Several field and office personnel

and part-time laboratory technicians also provided invaluable assistance.

WISSA provided a full-time Study Manager to administer the day-to-day operations, provide secretarial support to the Board of Directors and the Science Advisory Panel, and manage contracts, communications, and preparation of the study report.

## Confidentiality

Because of the nature of the information collected, it was essential that herd owners who took part in the study remain anonymous. To maintain the herd owners' anonymity, research personnel agreed not to release the identities of study participants to anyone not directly involved in the day-to-day work of the study, including the sponsoring agencies and associated committees. Confidentiality agreements were signed by all project participants with access to the data.

## Data Handling

All production and animal location information was entered into a database by the veterinarians who collected the data and checked them for accuracy. Pathology data were entered by the pathologist and associated staff, and air-monitoring data by the contractor. Reproductive indices, immune system outcomes, pathology results, exposure data, and information on other factors of interest were described and the data checked before linking the exposure data to animal health outcomes.

## Statistical Analyses

Mixed-effect regression models were used to investigate whether measures of reproductive, immunological, and pathology outcomes were associated with emissions from the petroleum industry.

The plan for analysis of the data included procedures to account for:

- similarities between animals in the same herd and between herds in the same geographic areas
- confounders, which refer to factors that can mask or distort the exposure-response relationship. Confounders

are related to both the outcome (e.g., breeding-to-calving interval) and the air pollutant under consideration (e.g., exposure to SO<sub>2</sub>).

- potential intermediate variables, which refer to factors that may be part of the biological causal pathway
- interactions, which refer to factors that modify or change the exposure-outcome relationship.

In addressing study questions, the researchers needed to account for other influential factors, such as the weather or the age of the cattle, that could potentially mask the primary exposure-outcome relationship. For example, in preliminary analyses, it might appear that higher levels of an airborne emission such as SO<sub>2</sub> were associated with lower numbers of stillbirths. But what if, in that situation, higher levels of SO<sub>2</sub> happened to coincide with areas less affected by drought? The researchers applied a systematic approach to determine answers to study questions that controlled for the many other factors known to affect animal health.

Appropriate statistical adjustments were made to correct for multiple comparisons, following standard statistical practice. One such adjustment, the Bonferroni correction, controls the likelihood of falsely declaring a non-existent association, usually known as a Type I error. Only associations that were statistically significant using these stringent criteria were presented as findings for discussion.

## Associations vs Effects

The study was an epidemiological investigation that drew on large blocks of data collected from privately owned cow-calf operations, laboratory analyses of biological samples and samplers from air monitors, and advanced statistical techniques to determine whether there were associations between emissions from oil and gas industry facilities and measures of beef cattle health and production.

Since this was an observational study that depended on data collected in the real world, where conditions of the experiment were not controlled by the researchers, the

researchers could conclude, for example, that more lesions in the heart muscle of calves were associated with exposure to higher levels of SO<sub>2</sub>, not necessarily that more lesions were caused by higher exposure to SO<sub>2</sub>.

Epidemiological evidence of an association can show only that a risk factor (such as an emission) is associated with a higher incidence of disease in the population most exposed to that risk factor. The stronger the association between the risk factor and the disease, the more likely the risk factor contributes to the disease and the less likely the disease may be due to some other, unmeasured factor. A consistent increase in effect with increasing exposure and supporting evidence from other studies also suggest that the association could be consistent with a cause-and-effect relationship.

On the other hand, where a carefully designed and adequately sized observational study such as this does not demonstrate an association, this “negative” finding helps rule out a cause-and-effect relationship as it relates to the conditions of exposure observed within the study. That is why the “no association” findings of this study are of such value.

## Organization of the Technical Summary

The 17 numbered sections of this Technical Summary present the highlights of each of the 17 major research appendices in the Research Appendices volume. Numbers of the sections coincide with numbers of the Research Appendices, for ease of reference.

For further details on the background, scope, and purpose of the study see the Introduction in the Research Appendices volume. Following the Introduction, there is an overview of the methods used to analyse the data. The “Overview of the statistical analysis” is then followed by “Inferring Causation from Findings in Observational Studies,” which takes a closer look at the methods of epidemiology and why they are helpful for determining a causal effect, and at the limitations of a single study in determining causation with certainty.

# 1. Selecting the Herds and Collecting Data

Herd selection was guided by three objectives: to obtain broad coverage of the major cattle and oil- and gas-producing areas of Western Canada, to achieve a wide range of exposures to different levels of emissions, and to involve herd owners most likely to comply with the study design.

## Herd Selection

Six “project” veterinarians, employed by the University of Saskatchewan, began the herd selection process by recruiting local veterinary clinics. The clinics contacted herd owners within their practice area and provided a list of potential participants to the project veterinarians, who then contacted the herd owners on the list.

For a herd to be invited to participate, the following selection criteria were to be met:

- Herd size, where possible, was to be between 50 and 250 breeding females.
- All animals were to be individually identified with at least one readily visible ear tag.
- All calf births should have been recorded during the 2000 calving season.
- The herd owner was to have access to facilities suitable for pregnancy testing, bull evaluation, and blood sample collection.
- Cows and heifers should have been pregnancy tested by a licensed veterinarian following the 2000 season.
- The herd owner needed to have an established spring-summer breeding season for the herd.
- Bulls should have been evaluated by a licensed veterinarian before use in the 2000 breeding season
- The herd owner needed to have an established, working relationship with a local veterinary clinic.
- Both the herd owners and their veterinarians had to be interested in the study and commit to completing the study protocol.

Project veterinarians were asked to classify the herds based on information about the proximity of oil- and gas-field facilities provided by the herd owner, and to use this information to maximize the range of potential exposures for the herds within their geographic regions.

For the purpose of selection, exposure potential was classified as follows:

- **higher exposure**—herds pastured within 1.6 km (1 mile) of well and battery sites or within 8 km (5 miles) of a compressor station or gas plant with a large flare stack or an incinerator stack
- **lower exposure**—herds pastured at least 3 km (2 miles) from small facilities and at least 16 km (10 miles) from large facilities.
- **no identifiable exposure**—herds with no oil or gas field facilities within 50 km (30 miles) of any pasture.

Herds with obvious exposure to emissions from significant sources other than oil- and gas-field facilities, such as very large pulp and paper operations, coal-fired power plants, and intensive livestock operations were not considered.

More than 400 herd owners were contacted, informed about the study, and asked if they wanted to participate. Those that expressed an interest in the study were interviewed to see if their herds met the selection criteria and to estimate exposure potential.

The project veterinarians selected 212 herds because of their potential for exposure, interest on the part of the herd owner, and fit of the operation with the selection criteria. Five herds changed their decision to participate before the start of the study and two more shortly after the start of data collection; 205 herds with some 33,000 animals completed the first phase of the study to pregnancy testing in fall 2001.

The resulting sample size was considered sufficient to address the primary study questions to the end of June 2002, when air monitoring of most herds was discontinued. Successful retention of participating herd owners and efforts to minimize the loss of data from each herd helped to maintain the sample size and resulting power of the study to examine the primary study questions.

Cooperation and compliance by participating producers and veterinarians were essential to the success of this project. While incentives paid to producers for each major milestone in the study likely helped with compliance,



## Overview of the time line for collecting data

2001				2002		
Spring	Summer	Fall	Winter	Spring	Summer	Fall
<b>Cattle Productivity and Herd Management</b>						
<p>Complete final selection of study herds</p> <p>Collect baseline records and establish herd inventory</p> <p>Collect calving records for spring 2001</p> <p>Score body condition of cows before breeding season and verify inventory</p> <p>Evaluate breeding soundness of herd bulls</p> <p>Avian Study—1st Field Season, April to June</p> <p>Start post-mortem examinations for breeding herd (May 1, 2001 through June 1, 2002)</p> <p>Record cow membership in breeding groups and breeding season management</p> <p>Track inventory changes in beef herds</p>	<p>Track breeding group dynamics</p> <p>Track animal location on summer pasture</p>	<p>Pregnancy test cows and heifers from study herds</p> <p>Body condition score cows and heifers and verify inventory</p> <p>Case-control study of infectious disease in breeding herd</p> <p>Record observed abortions</p>	<p>Monitor winter feeding program and pre-calving management</p> <p>Continue to track inventory changes and animal location</p> <p>Score body condition of cows before calving</p>	<p>Collect records of calving and calving management</p> <p>Record calf death losses</p> <p>Conduct post-mortem examinations of losses from 2002 calf crop</p> <p>Record treatments in calves and breeding herd</p> <p>Score body condition of cows and heifers before breeding and verify inventory</p> <p>Evaluate breeding soundness of herd bulls.</p> <p>Collect feed samples</p> <p>Avian Study—2nd Field Season, April to June</p> <p>Beef Cattle Immunotoxicology Study—Spring 02</p> <p>Record cow membership in breeding groups and breeding season management</p> <p>Track inventory changes</p>	<p>Track breeding group dynamics</p> <p>Track animal location on summer pasture</p>	<p>Pregnancy test cows and heifers from study herds</p> <p>Score body condition of cows and heifers and verify inventory</p> <p>Conduct serology study of infectious disease in calves born in spring</p> <p>Collect water samples</p>
<b>Air Monitoring</b>						
<p>Collect data on location of study herd pastures</p> <p>Set up passive monitoring network for SO<sub>2</sub> and VOCs for first bull exposure in all study herds, Apr &amp; May 01</p>	<p>Establish passive monitoring network for H<sub>2</sub>S – Sep 01.</p> <p>Continue passive monitoring for SO<sub>2</sub> and VOCs</p>	<p>Continue passive monitoring</p>	<p>Set up particulate monitors for PM<sub>1.0</sub> and PAHs—Jan/Feb 02</p> <p>Continue passive monitoring</p>	<p>Passive monitoring discontinued in all but 50 study herds Jun 30, 02</p> <p>Continue particulate monitoring</p>	<p>Continue passive monitoring in 50 herds</p> <p>Continue particulate monitoring</p>	<p>Stop remaining passive air monitoring Dec 02, particulate monitoring Jan 03</p>

the ongoing on-farm presence and personal contact with study personnel also assisted producers in keeping on track and helped ensure the timely recording of data.

## The Production Cycle

The study was designed to coordinate data collection with the production cycle, which for most cow-calf herds in Western Canada begins with the start of breeding season.

Typically, bulls are placed with the cows after calving, usually between April and June. Most cow-calf pairs are put onto pasture during the summer. At the end of the grazing season, most cows are moved back to more confined locations to facilitate fall processing and winter feeding management. The calves are weaned in the fall and either sold, retained and fed for later sale, or kept as herd replacements.

Many producers will pregnancy test their cow herd in the fall and sell any cows that are not pregnant to reduce winter feeding costs. In some herds the cows are not processed again until calving. Other herd owners will vaccinate the cows before calving to increase protective immunity passed to the calf in the colostrum and minimize the risk of diarrhea in the calf.

Most herds are moved to areas that allow close monitoring of at least some part of the herd during calving. Herds that calve in the coldest part of winter tend to be more confined during calving than herds that calve later in the spring.

Near the end of the calving season and before turnout to summer pasture, the herd is often processed again, to vaccinate the cows and calves in preparation for the start of the next breeding season.

Given this production cycle, there are only two times during the year when it is practical to collect individual animal information and samples from all herds: when the cows are tested for pregnancy status, and when they calve. The primary reproductive outcomes measured during the production cycle were records of pregnancy status and the occurrence of abortions, stillbirths, and calf deaths before 3 months of age.

## Data Collection

In addition to information on herd productivity, data were collected on a range of factors that could influence the reproductive success of individual animals, such as cow age, breed, body condition, vaccination status, and herd breeding and calving management.

The following reproductive outcomes were recorded for each cow or calf:

- **Pregnancy Status.** All cows were pregnancy-tested, typically between September and November, when most were between 2 and 6 months into gestation. The outcome of the testing was recorded as whether the cow was pregnant or not pregnant.
- **Abortion.** A known or suspected abortion was defined as either an observed premature calving judged to be at least 1 month before full term, or an assumed calf loss when a cow was diagnosed as pregnant but failed to calve.
- **Calving-to-Calving Interval.** The calving-to-calving interval was defined as the number of days from calving in 2001 to calving in 2002.
- **Breeding-to-Calving Interval.** The breeding-to-calving interval was estimated as the time from first bull exposure in 2001 to calving in 2002; or the time from 21 days after calving in 2001 to calving in 2002, if the cow calved after being placed on pasture with the bull.
- **Stillbirth.** A stillbirth was defined as a calf that was dead at or within 1 hour of birth, or a calf that was found dead, had not been observed alive, and was obviously recently born.
- **Calf Death.** Calf death was defined as a calf that died more than 1 hour after birth and before the earlier of 3 months of age or June 30.
- **Calf Treatment.** Calf treatment was defined as the producer reporting administering any pharmaceutical for therapeutic or prophylactic indications to the calf before 3 months of age or June 30.

Herd veterinarians examined animals that died to describe their physical condition, report gross postmortem examination findings, and collect a standard set of tissue samples for laboratory analysis (Sections 9 and 10).

Each cow was tracked throughout the study to determine the reasons for changes in herd inventory and to determine the extent and reasons for any missing data required to address important study questions.

Through the cooperation of local herd owners and veterinarians, on-farm collection of detailed individual animal data was successful in this group of cow-calf operations.

Complete data for the 2002 calving season were available from 96% of the herds initially selected to participate. Herd production records were rated as satisfactory or better in 94% of the herds and individual animal records were available for more than 98% of potentially eligible cows from participating herds, for each measurement period throughout the study.

## **Data Management**

Data on herd management, animal inventory, individual cow reproductive performance, and calf health were collected with the help of the herd owners and entered into a central database by the project veterinarian for each herd. Data from postmortem examinations and other diagnostic testing were stored in separate databases linked to the main database.

The location of each cow for each month during the study was also entered by the project veterinarian. Animal locations were recorded as the legal description of land to which the cow had access at the beginning and midpoint of each month. Calf information was linked to the calving record for the cow.

Data on the results of air monitoring were stored in a separate database and linked to the location record of each cow using a geographic information system (GIS).

The resulting electronic map was also used to assign each herd to an ecoregion and to link herd records and the most appropriate meteorological data from Environment Canada.

# 2. Herd Location and Industry Facilities

A key objective in herd selection was to maximize the differences in exposure to emissions from oil- and gas-field facilities across the herds. At the end of the study, data on facility locations and emissions relative to each herd were compared with the original rankings for the exposure potential of the herds to determine if that objective had been met.

Data on the locations of oil and gas wells, gas plants, gas-gathering systems, and compressor stations, and volumes of gas flared and vented from each location in 2001 and 2002, were provided by the British Columbia Oil and Gas Commission, the Alberta Energy and Utilities Board, and Saskatchewan Industry and Resources. The data were incorporated into the study GIS, along with herd location data collected for replacement heifers and cows from 205 herds on 3,355 parcels of land between April 2001 and the end of data collection for each herd in the fall of 2002.

Numbers of oil and gas well sites and batteries within 1.6 km and gas plants within 8 km of the centre of each quarter section were determined. For pastures larger than one quarter section, the numbers of facilities were averaged. Results were used to arrive at a measure of the “density” of facilities on and around the location of the cattle. Average exposure to industry operations and emissions was determined for the land available to each herd, based on the density of surrounding facilities and the volumes of gas



Flaring

Estimated exposure of study herds to oil and gas facilities based on information provided by herd owners at the time of herd selection

Exposure Category	Number of Herds
high exposure	92 (45%)
low exposure	71 (35%)
no exposure	42 (20%)

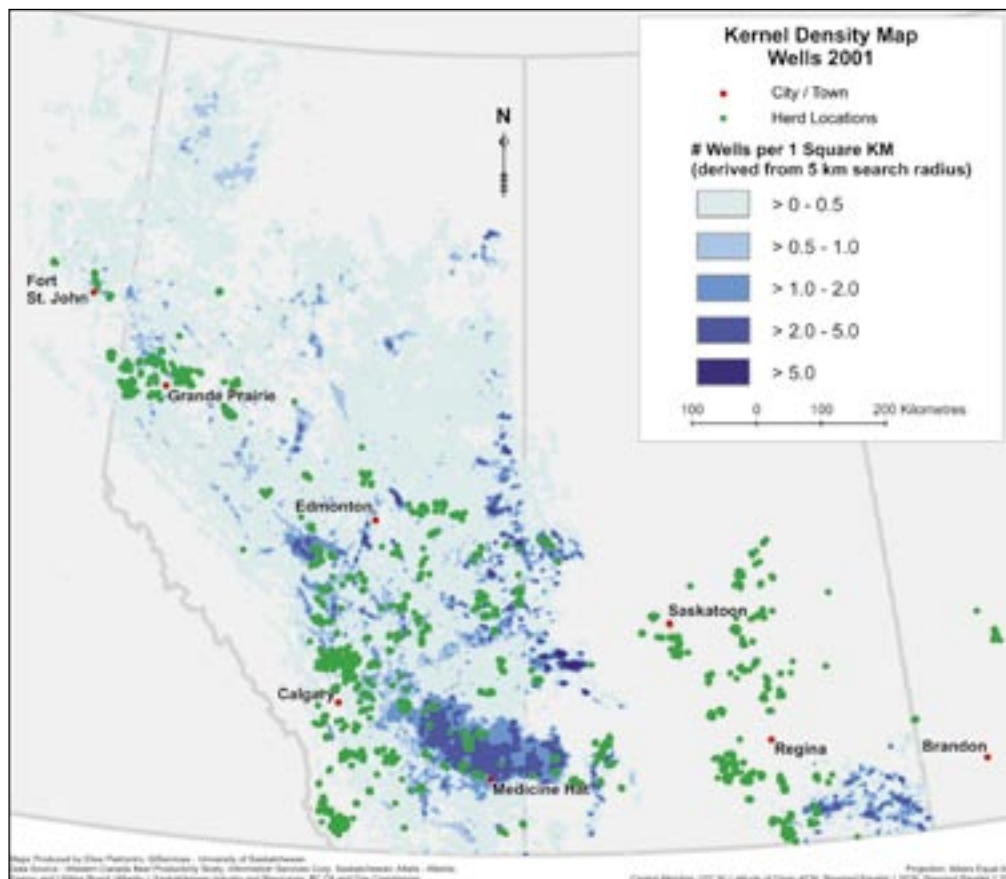
that the facilities were reported to have flared and vented in 2001 and 2002. When the average exposures were compared to the exposure rankings assigned to each herd location during the selection process, results showed that the original rankings—high exposure, low exposure, and no exposure—were successful in identifying herds with significant differences in exposure to both various facility types and associated reported emissions.

Mapping of herd locations in relation to the major oil- and gas-producing areas of Western Canada, or in relation to Environment Canada data on reported air-contaminant emissions, showed that the study herds were adequately distributed across the areas of interest, and included a range of locations with no, low, moderate, and high industry activity.

Types and concentrations of contaminants can vary, however, depending on the composition of the field, type and technology of the facility, and level of activity. The locations



Oil Well



Note: Cattle were placed on the pastures in Manitoba near the end of the study after the completion of calving season in 2002. Air quality was not measured at these sites.

Herd locations for 2001 and 2002 in relation to oil and gas well sites in 2001, showing a broad distribution of herds and wells in areas of no, low, moderate, and high petroleum industry activity.

of facilities, and the volumes of emissions they released, alone were not sufficient to serve as an adequate measure of animal exposure at specific herd locations and times.

Passive air-monitoring (Section 3) provided more accurate location- and time-specific information on exposure than proximity-based information, for examining potential associations between the health of the cattle and industry emissions.

## Oil and Gas Industry Facilities and Their Emissions

Common oil and gas industry field facilities include oil and gas wells, battery sites, gas-gathering systems, and natural gas processing plants. The following descriptions of these facilities, and their emissions, were drawn from websites maintained by the Alberta Energy and Utilities Board, the Clean Air Strategic Alliance, and the Canadian Association of Petroleum Producers.

A **facility** is an arrangement of equipment or buildings for the production, injection, or movement of petroleum products associated with the oil and gas industry.

A **battery** is a facility that controls production from an oil or gas well or group of wells, and may include equipment for separating, treating, cleaning, and storing products.

A **compressor** station comprises service equipment that maintains or increases the flowing pressure of the gas from a well, battery, or gathering system, for delivery to market or other disposition.

A **gas-gathering system** consists of gas lines that move products from one facility to another and may include compressors and line heaters.

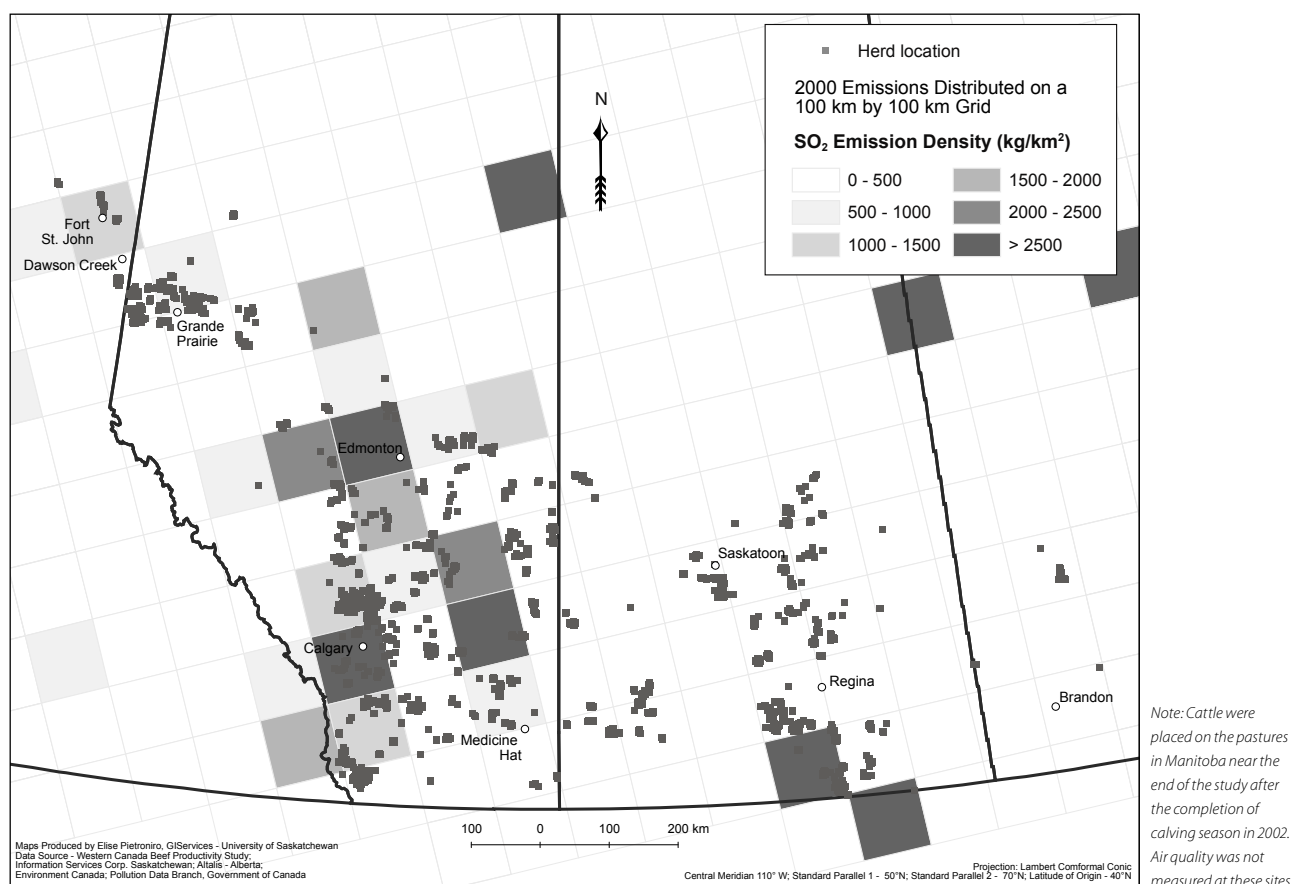
A **gas plant** is a facility for processing raw gas to meet specifications required to sell the gas and transmit it by pipeline.

Airborne contaminants can be released from these facilities through fugitive emissions, venting, flaring, and the operation of incinerator stacks.

**Fugitive emissions** are unintentional leakages of gas from connections or valves.

**Venting** is the controlled release of natural gas that is not processed for sale or use because of technical or economic reasons. It has typically been used as a low-cost





*Herds participating in the study in 2001 and 2002 in relation to reported emissions of SO<sub>2</sub> (kg/km<sup>2</sup>) in 2000, from Environment Canada's database on Criteria Air Contaminants, showing a broad distribution of herd locations relative to reported SO<sub>2</sub> emissions.*

option to manage quantities of waste natural gas too small to be conserved or to support combustion in a flare.

**Flaring** involves the combustion of natural gas and is used to prevent potential explosions, to depressurize gas-processing equipment for maintenance, or to dispose of unwanted or unusable volumes of gas. Flaring can occur at wells, dehydrators, compressors, pipelines, batteries, and gas-processing plants.

**Incineration** is used at sulphur recovery plants to dispose of tail gases remaining after all saleable components have been extracted. True incinerators have enclosed flames where the air-to-fuel ratio is controlled for optimum combustion. This process results in near-complete combustion of any petroleum hydrocarbons, the end products being carbon dioxide, water, and, in the case of sour gas, SO<sub>2</sub>.

Oil and gas containing hydrogen sulphide (H<sub>2</sub>S) is called "sour" and presents unique safety and environmental challenges.

**Sour gas** is not intentionally vented or released to the atmosphere because of the H<sub>2</sub>S hazard.

In addition to H<sub>2</sub>S, contaminants of the raw gas that must be removed during processing include water, carbon dioxide, and nitrogen. Other acid-forming emissions associated with petrochemical production include nitrogen oxides, ozone, and SO<sub>2</sub>.

Where combustion of flare gas is incomplete, emissions may contain various compounds, such as BTEX (benzene, toluene, ethylbenzene, and xylenes) and other volatile organic compounds (VOCs) as well as polycyclic aromatic hydrocarbons (PAHs).

Crudely defined, a VOC is any hydrocarbon, other than methane, that has a boiling point of 180°C or less and that has less than 10 carbon atoms in its molecule. BTEX compounds are examples of aromatic hydrocarbons or unsaturated cyclic hydrocarbons containing one or more benzene rings.

PAHs are aromatic hydrocarbons consisting of two or more benzene rings fused together.

### 3. Air Monitoring Technology, Methods, and Highlights

To determine potential associations between emissions from oil and gas production facilities and cattle health, the study used air-concentration data supplied by a network of air samplers deployed as close as possible to the herds.

The samplers measured mean monthly air concentrations of selected contaminants near cows in 205 extensively managed beef cow-calf herds. Air-concentration data were obtained from more than 1,200 locations that included the major oil- and gas-producing areas of Western Canada.

The air-monitoring program began in April 2001 and was completed in January 2003. It focused on the following types of oil and gas industry emissions:

- $\text{SO}_2$ , to represent combustion emissions associated with sour-gas flaring and the operation of sulphur recovery plants
- $\text{H}_2\text{S}$ , to represent incomplete combustion and fugitive emissions from sour-gas collection and processing facilities
- VOCs (volatile organic compounds), to represent fugitive, dehydrator, and incomplete combustion emissions
- $\text{PM}_{1.0}$  (fine particulate matter with an aerodynamic diameter of  $1.0\ \mu\text{m}$  or less), to represent incomplete combustion from sweet and sour gas combustion processes
- PAHs (polycyclic aromatic hydrocarbons), to represent incomplete combustion from sweet- and sour-gas combustion processes
- metals, as they have often been associated with combustion emission sources.

$\text{SO}_2$ ,  $\text{H}_2\text{S}$ , and VOCs were measured for all participating study herds from April 2001 to June 2002, and for a subset of 50 herds from July 2002 to December 2002.

Particulate matter ( $\text{PM}_{1.0}$ ) was measured for a subset of 32 study herds, from January 2002 to January 2003.



Top: Passive  $\text{SO}_2$  and  $\text{H}_2\text{S}$  sampler shelter, showing three passive samplers.

Bottom: Typical field installation of a passive sampler shelter.



*PM<sub>1.0</sub> sampling system, shown without surrounding protective fencing*

## Passive and Active Air Monitors

Two types of air monitors—passive and active—were used to monitor emissions. The passive type, in which air moves across a diffusion membrane, measured concentrations of SO<sub>2</sub>, H<sub>2</sub>S, and 26 VOCs. The active type, in which a solar-charged electric pump draws air through a filter, and a filter plus a column filled with absorbent material, measured concentrations of PM<sub>1.0</sub>, 30 PAHs, and 14 metals.

Passive monitors consisted of a shelter 200 mm (8 inches) in diameter, housing three samplers, one each for SO<sub>2</sub>, H<sub>2</sub>S, and VOCs. The samplers were attached under the shelters, to allow undisturbed movement of air across diffusion membranes and protect the membranes from the weather.

Passive samplers were used near all study pastures because they are simple to operate, reliable, and relatively inexpensive. They are also effective in sampling low concentrations of air contaminants.

Active monitors require more maintenance and are relatively more expensive; they were used only at a subset of study sites for part of the study.

For optimum placement, both types of monitors were located on or as close as possible to the pastures where the cattle were reported to be spending most of their time. They were placed so that they would not be affected by local sources such as roadways or homesteads, nor by nearby obstacles that could interfere with airflow.

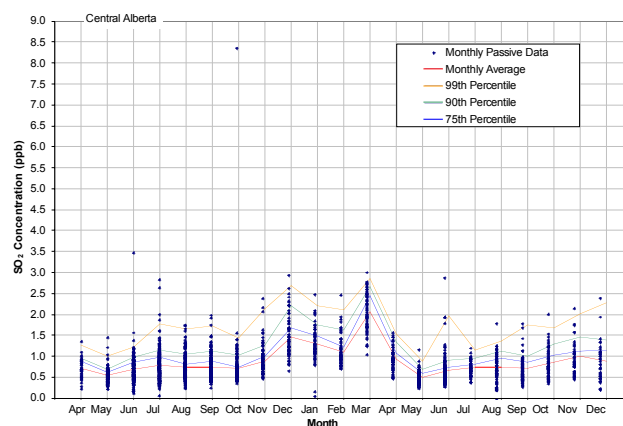
## Monthly Replacement of Media

Passive samplers and the collectors from the pump-operated monitors were installed, then retrieved and replaced each month, by a team of 15 technicians from RWDI Air Inc., Calgary, Alberta. Exposed samples were shipped to RWDI's offices in Calgary, where quality assurance/quality control documentation was prepared. The samples were then shipped to contract laboratories for analysis after coding, so that the lab would not know where the samples had been collected.

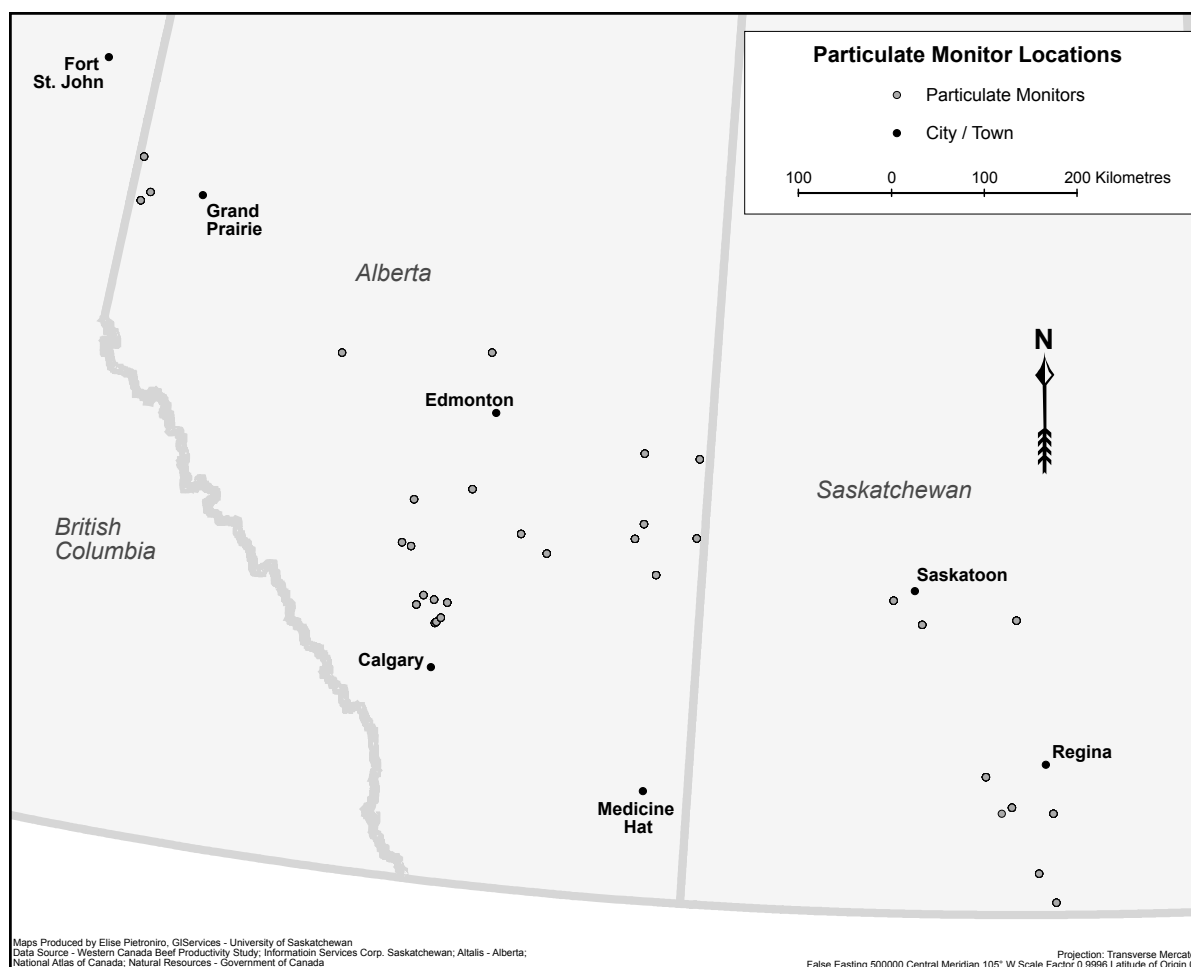
A thorough chain-of-custody protocol was followed in sample collection and handling. Duplicate samplers (10% of the total sampler numbers), field blanks, and a series of laboratory and field audits were used in quality assurance and control.

Passive monitors were moved as necessary at the end of each calendar month to ensure that there was a monitor at or near each location with cattle at all times during the study. During the year, each herd might be moved between several pastures and holding areas, depending on herd management, the available land base, the time of year, and pasture conditions.

The team from RWDI worked with the project veterinarians from the University of Saskatchewan, to ensure that the monitors were deployed in accordance with herd movements.



*Monthly concentrations of SO<sub>2</sub> (ppb) measured from April 2001 to December 2002 in Central Alberta. Each blue dot represents one reading from a passive air monitor; 3,006 readings were recorded.*



Locations of  $PM_{1.0}$  monitoring stations in 2002

Active monitors were usually set up on the “home quarter” to measure exposure near the calving area and the winter feeding pens for yearling animals in a subset of 32 study herds (Sections 5 and 12). These monitors were not moved during the study.

## Overview of Results

Air-concentration data were broadly grouped into six geographic regions: Northern Alberta (which for the study included part of northeastern British Columbia), Eastern Alberta, Central Alberta, Southern Alberta, Central Saskatchewan and Southern Saskatchewan. The data were reviewed by the air-monitoring contractor to identify obvious spatial and temporal trends.

### $SO_2$

Monthly average  $SO_2$  concentrations were determined from 13,991 samples collected from 1,211 pasture locations over 21 months. Regions with higher concentrations of  $SO_2$  experienced those concentrations in winter.

Average and maximum monthly concentrations were lowest in Central Saskatchewan, at 0.37 ppb and 2.32 ppb; and highest in Central Alberta, at 0.79 ppb and 8.35 ppb.

### $H_2S$

Monthly average  $H_2S$  concentrations were determined from 7,502 samples collected from 1,143 pasture locations over 16 months. There did not appear to be a consistent temporal pattern for  $H_2S$  concentrations.

Average and maximum monthly concentrations were lowest in Southern Alberta, at 0.14 ppb and 2.00 ppb; and highest in Eastern Alberta, at 0.27 ppb and 8.32 ppb.

### VOCs

Monthly average VOC concentrations were determined from 11,447 samples collected from 1,209 pasture locations over 21 months. Higher benzene concentrations tended to occur in winter.

The lowest VOC concentrations were in Southern Saskatchewan, Central Saskatchewan, and Southern Alberta.

Average benzene concentrations ranged from 200 to 210 ng/m<sup>3</sup>; and 95th percentile benzene concentrations from 490 to 560 ng/m<sup>3</sup> range.

The highest VOC concentrations were in Northern Alberta, Eastern Alberta, and Central Alberta. Average benzene concentrations ranged from 320 to 364 ng/m<sup>3</sup> range; and 95th percentile benzene concentrations from 810 to 880 ng/m<sup>3</sup>.

### **PM<sub>1.0</sub>**

Monthly average PM<sub>1.0</sub> concentrations were obtained from 365 sample pairs collected from 32 locations over 12 months. For all regions, the highest concentrations were measured in May.

Average and maximum monthly concentrations were lowest in Southern Saskatchewan, at 7.1 µg/m<sup>3</sup> and 23.8 µg/m<sup>3</sup>; and highest in Eastern Alberta, at 9.3 µg/m<sup>3</sup> and 45.4 µg/m<sup>3</sup>.

### **PAHs**

Monthly average particle and vapour phase PAH concentrations were obtained from 365 samples collected from 32 locations over 12 months. Seasonal trends tended to vary with the compound.

Regions with lower and higher PAH concentrations varied with the compound. The lowest naphthalene concentrations were measured in Southern Saskatchewan, with average and 95th percentile values of 36 and 140 ng/m<sup>3</sup>. The highest naphthalene concentrations were measured in Central Saskatchewan, with average and 95th percentile values of 57 and 290 ng/m<sup>3</sup>.

### **Metals**

Monthly average concentrations of metals in PM<sub>1.0</sub> samples were determined from 365 samples collected from 32 locations over 12 months. The highest concentrations tended to occur in May and were associated with the high concentrations of PM<sub>1.0</sub> that occurred at the same time.

Most metal air-concentration measurements were less than the minimum detection limits. Only aluminum and calcium were consistently above the minimum limits, with respective averages of 22 and 130 ng/m<sup>3</sup>.

## **Comparing the Data**

Where possible, ambient air-monitoring data obtained from the study were compared to ambient air-monitoring data collected in Alberta airshed monitoring zones; these data were obtained from the Alberta Clean Air Strategic Alliance (CASA) or from the respective airshed zones.

For SO<sub>2</sub> and H<sub>2</sub>S, monthly average and period average concentrations measured at study locations were found to be similar to those measured concurrently by other monitoring programs in similar areas.

For VOCs, study concentrations were compared to non-concurrent VOC data collected within and downwind of an industrial region airshed. For some compounds, values for the two datasets were similar; for others, study values were higher or lower, depending on the compound.

For PM<sub>1.0</sub>, study concentrations were compared to PM<sub>2.5</sub> concentrations measured and summarized by others in Alberta. Average study values were similar to those for “rural, source-influenced sites,” which tend to be in the 4.5 to 11.4 µg/m<sup>3</sup> range.

For ambient PAH concentrations, which are not routinely measured in Western Canada, comparable data were not from periods concurrent with the study. In general, the study values of PAHs tended to be similar to those reported by Environment Canada. There was good agreement between measurements from Grande Prairie and the study for some compounds, but concentrations of others were either higher or lower, depending on the compound.

For metals, study concentrations were compared to non-concurrent measurements derived from PM<sub>2.5</sub> provided by Alberta Environment. In some cases, concentrations were similar (e.g., aluminum) and in other cases, study results were higher (e.g., calcium). Comparisons of information on metals were limited by the lack of concurrent data from similar regions.

Comparisons indicate that the results of the sampling program provided data representative of the range of exposures to the emissions of interest expected in other similar rural areas across Western Canada.



## 4. Modeling Pollutant Concentrations

Statistical models were developed to predict concentrations of air pollutants at locations where the study herds were pastured in British Columbia, Alberta, and Saskatchewan.

The models described the distribution of air concentrations for all measured substances, except metals, and identified factors that would predict their concentrations in the air. They were developed to determine whether proximity to oil and gas facilities explained variability in airborne concentrations of  $\text{SO}_2$ ,  $\text{H}_2\text{S}$ ,  $\text{PM}_{1.0}$  (fine particulate matter), VOCs (volatile organic compounds), and PAHs (polycyclic aromatic hydrocarbons).

The models drew on data from the air-monitoring stations set up to measure monthly average concentrations of the pollutants at pasture, wintering, and calving areas (Section 3).

The coordinates of the monitoring stations were combined with the coordinates of oil and gas facilities to calculate distances between the sources and the stations. Effects of the sources were considered for several distance classes: 0-2 km, 2-50 km, 0-50 km, 50-1000 km, and 0-1000 km. The number of facilities was determined for each distance class around a monitoring station, and a weighted sum was used to express the proximity of the facilities to the stations.

The following sources or groups of sources were considered separately: all wells, oil wells, gas wells, bitumen wells, batteries, and large facilities (gas plants and gas-gathering plants). Only active facilities were considered.

The effects of  $\text{H}_2\text{S}$  concentrations in the pool from which oil and gas were extracted, and the volume of gas flared during well tests of measured  $\text{H}_2\text{S}$  concentrations, were studied in a subset of data from Alberta.

There was some indication of seasonal trends, but most of the variability in concentrations was due to differences between the locations of monitoring stations.

The exposure models developed revealed an association between the proximity of oil and gas facilities and airborne concentrations of  $\text{H}_2\text{S}$ ,  $\text{SO}_2$ , and some VOCs and PAHs. The exposure models also indicated that increasing  $\text{H}_2\text{S}$  content of oil and bitumen wells, and sour-gas flaring, were associated with higher recorded airborne

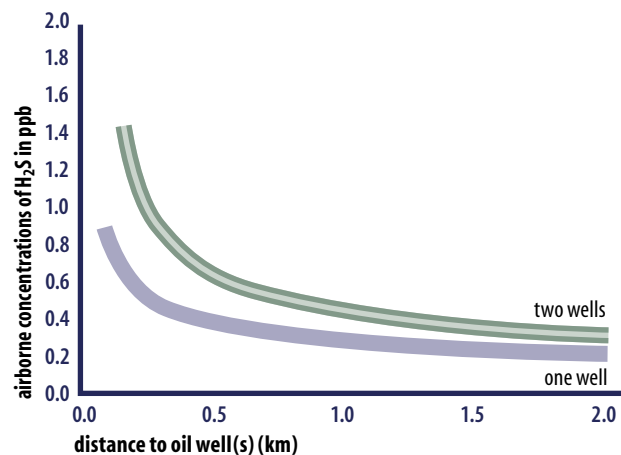


Chart comparing proximity to oil wells and airborne concentrations of  $\text{H}_2\text{S}$ , showing a steep decline in concentrations with increasing distance from the source of the pollution.

concentrations of  $\text{H}_2\text{S}$ . This suggests that oil and gas facilities contribute to environmental concentrations of the studied pollutants. On the other hand, the models did not reveal any clearly interpretable associations between the proximity of various oil and gas facilities and concentrations of  $\text{PM}_{1.0}$ . This finding was not surprising, given that a great many sources emit fine particles.

The pattern of modeled association between distance to oil wells, and number of oil wells, and concentrations of  $\text{H}_2\text{S}$  in the air is representative of the pattern for  $\text{SO}_2$  and some VOCs and PAHs. As shown in the chart, there is a steep decline in concentrations of  $\text{H}_2\text{S}$  when oil wells are farther away, even within a 2-km range. Further, as the number of wells within a given radius increases, so do expected concentrations of  $\text{H}_2\text{S}$ .

Concentrations of air pollutants predicted by the models agreed very well with concentrations of pollutants measured by the monitoring stations.

Overall, it is reassuring that concentrations of  $\text{H}_2\text{S}$ ,  $\text{SO}_2$ , and some VOCs and PAHs measured by the monitoring stations were associated with the distance of the stations to oil and gas facilities, indicating that the monitoring stations can discriminate between locations with different air quality. This finding increased confidence in the dependability of air pollutant concentrations measured by the air-monitoring stations.

## 5. Determining Animal Exposure to Contaminants

Air-monitoring stations were used to measure emissions from oil- and gas-field facilities for 205 cow-calf herds across the major cattle and oil- and gas-producing areas of Western Canada.

Stations were set up as close as possible to the summer, winter, and calving areas of the herds to measure concentrations of airborne  $\text{SO}_2$ ,  $\text{H}_2\text{S}$ , and VOCs.

Other types of stations were also set up to measure amounts of  $\text{PM}_{1.0}$ , which in turn yielded data on volatile and particulate-bound PAHs, and selected metals, for 32 study herds (Section 3).

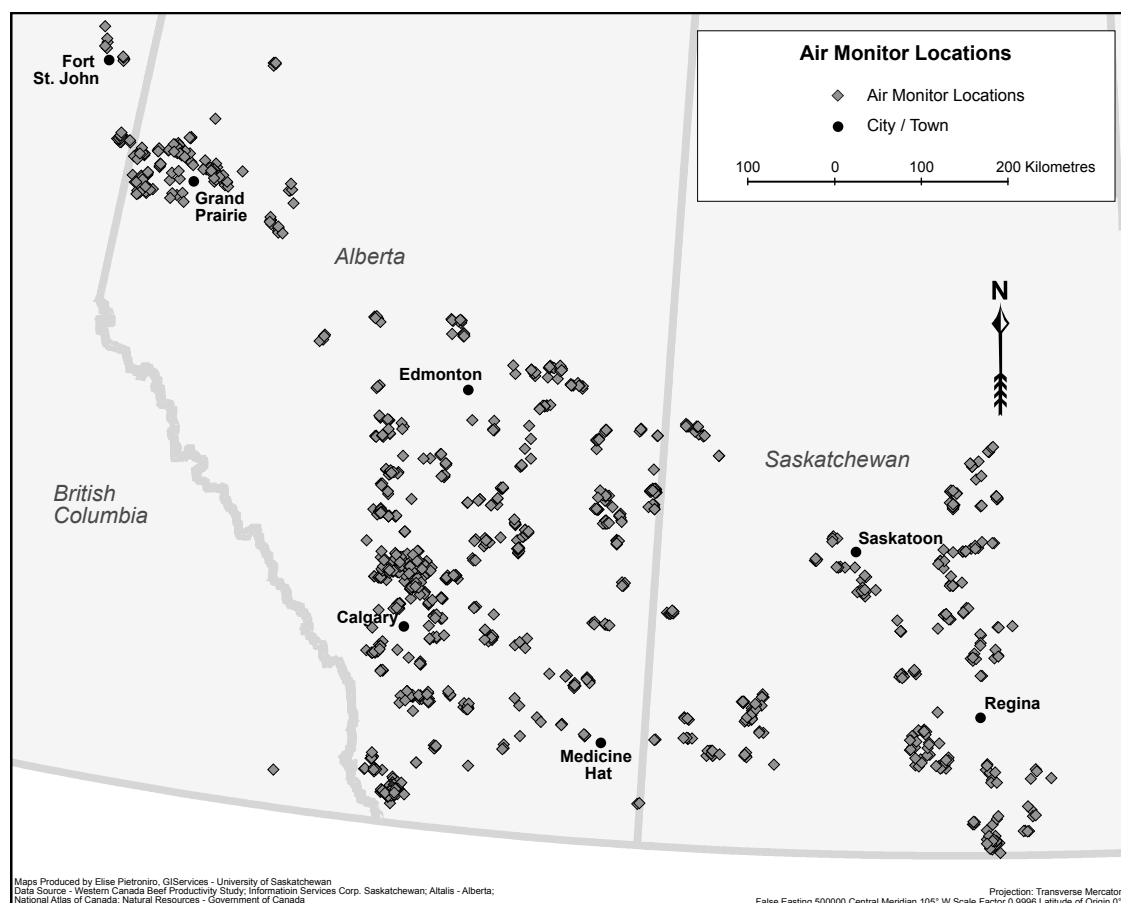
In addition to the monitoring stations, data on the “density” of surrounding oil and gas well sites, determined from information supplied by provincial regulatory authorities, served as a second measure of exposure.

### Distances of Monitors from Pastures

The objective of the air-monitoring program was to place a monitor within 1.6 km (1 mile) of as many quarter sections as possible, used by the cattle during each month of the study.

**$\text{SO}_2$ ,  $\text{H}_2\text{S}$ , and VOCs.**  $\text{SO}_2$  and VOCs were monitored starting in April 2001;  $\text{H}_2\text{S}$  was not monitored until September 2001. There was at least one passive air monitor within 1.6 km of the centre of 75% of all quarter sections used to pasture cattle, and at least one passive air monitor within 5.3 km (3.3 miles) of 90% of all quarter sections.

Most of the larger distances from each quarter section to a passive air monitor were reported on large pastures where terrain limited access to potential monitoring sites; the largest distances were reported on very large community pastures in southern Saskatchewan, in an area where



Locations of passive air monitors, April 1, 2001 through June 30, 2002

there were no oil and gas facilities. The most extreme situations had very low potential for industry contamination; therefore, there was less concern about unmonitored variations in air quality from one point in the pasture to another in those locations.

**PAHs and PM<sub>1.0</sub>.** An active air-monitoring station was placed at a central location within the wintering and calving grounds of each of 32 study herds.

## Determining Exposure

Mean concentrations of SO<sub>2</sub>, H<sub>2</sub>S, and selected VOCs (benzene, toluene, ethylbenzene, m&p-xylene, o-xylene, and hexane) were assigned to each quarter section of land containing cattle for each month of the study. Concentrations of these pollutants were determined from data collected by all air monitors within 1.6 km of the centre of each quarter section, or, when necessary, from data collected by the nearest available air monitor.

Where cattle had access to more than one quarter section during a month, the values of the concentrations read by the stations for each quarter section were averaged for that month.

Information on the locations of 39,269 animals from 205 herds on 3,355 different parcels of land was recorded for each half-month interval from April 2001 to December 2002. The result was a total of 3,792,925 animal locations.

A GIS was used to link the location of each animal in each month and data on concentrations of pollutants for the pasture for that month, to determine the animal's exposure.

To determine the total exposure of each animal over any particular "at-risk" period (i.e., a length of time biologically relevant to the studied parameter, such as abortion, for example), the monthly values of mean concentrations were averaged over the appropriate at risk period to arrive at the cumulative exposure for each study question.

## Density of Surrounding Facilities

Numbers of oil and gas well sites and batteries within 1.6 km of each quarter section, and large field facilities (gas plants and gas-gathering systems) within 8 km of the centre of each quarter section, were recorded and averaged for pastures with more than one quarter section.

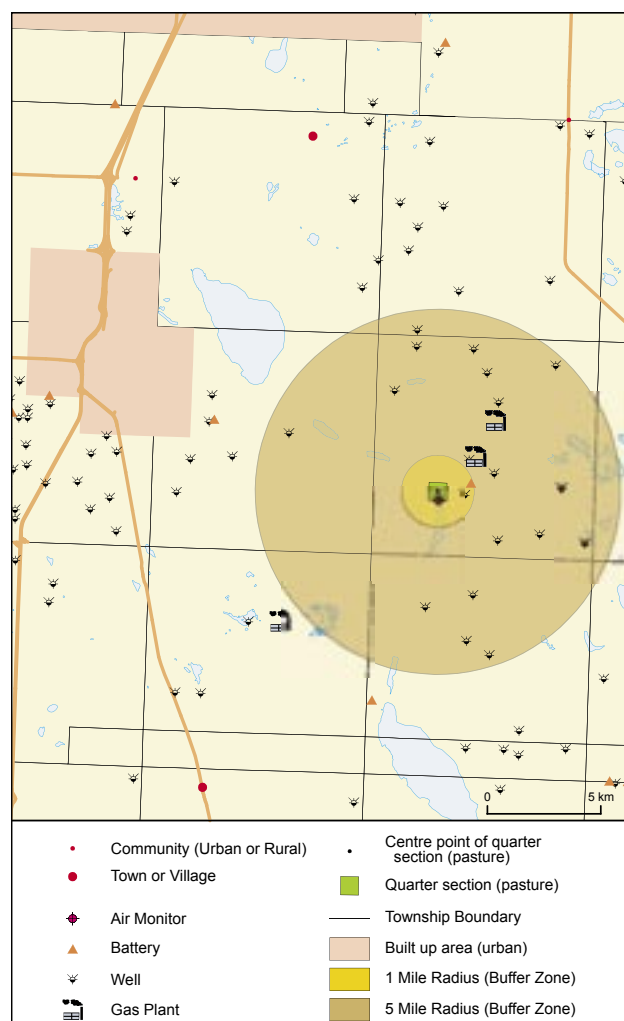
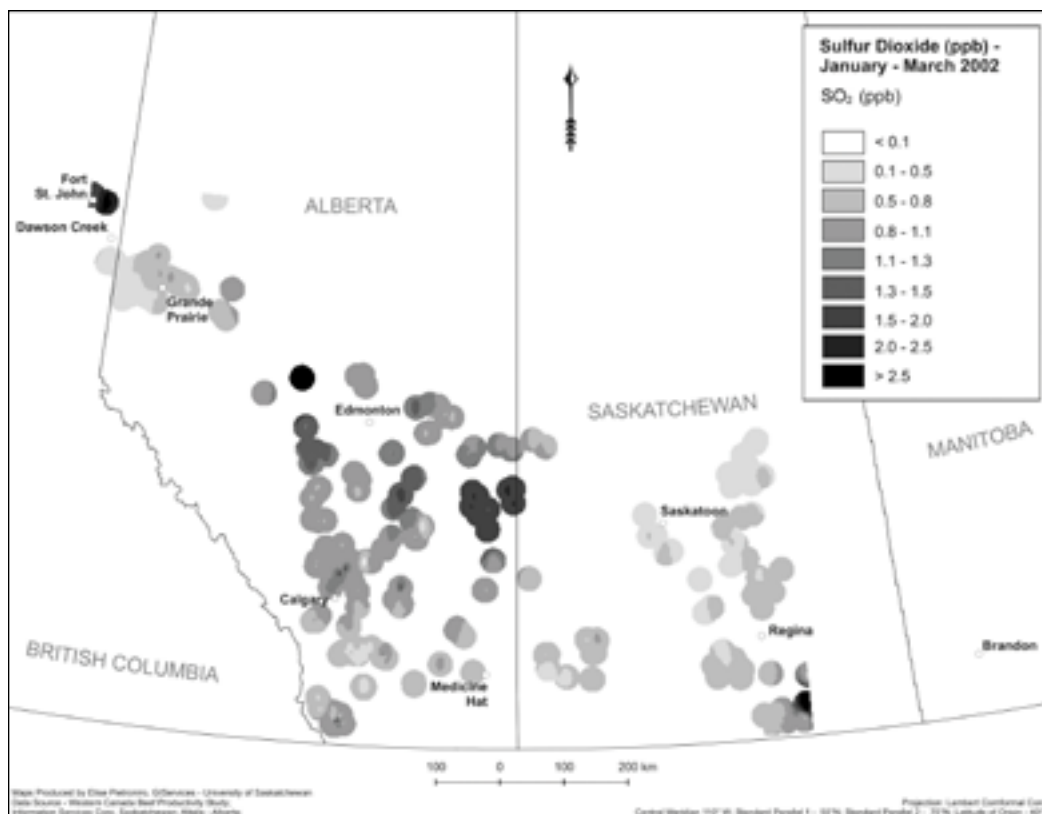


Diagram showing a 1 mile zone from the centre of a quarter section containing one air monitor, one well site, and one battery and a 5 mile zone containing 2 gas plants. The zones were used to determine the "density" of facilities in relation to pastures.

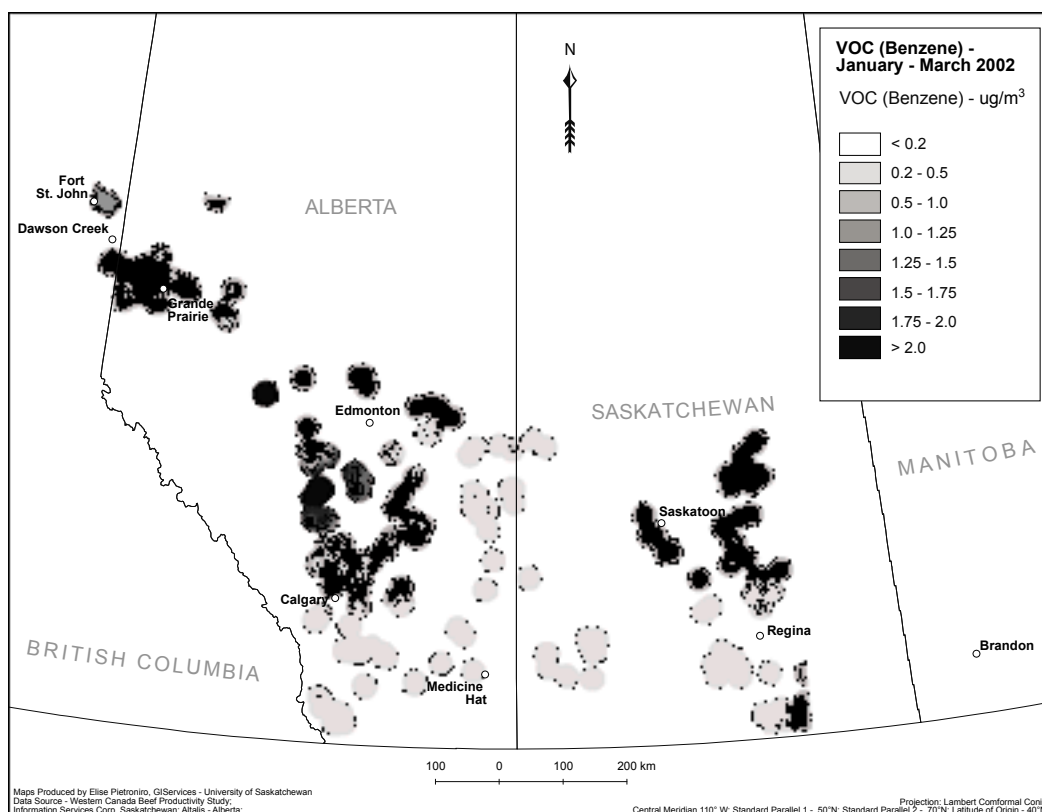
The resulting measures of surrounding facility density, combined with reported volumes of gas flared and vented from the facilities for each location, were also linked to the individual animals on that location for each month of the study.

The number of oil and gas well sites was chosen to represent the "density" of all facilities, because they are the most common types of facilities and data on them appeared to be the most verifiable and consistently reported for all facility types across the provinces.

Because well sites were only a surrogate for industry activity in an area, and because there is substantial variation in the amount and types of emissions from different field facilities, data on concentrations of airborne contaminants collected by the air monitors were the primary method of measuring exposure for the study.



Mean  $\text{SO}_2$  concentrations from passive monitors, January 1, 2002, to March 31, 2002



Mean benzene concentrations from passive monitors, January 1, 2002, to March 31, 2002

## 6. Nonpregnancy, Calving Interval and Disposal

The risk of nonpregnancy and the length of the calving interval are key measures of fertility, the most important factor affecting the profitability of cow-calf operations. Potential associations between emissions from oil- and gas-field facilities and reproductive success were examined for more than 32,000 beef cows across Western Canada.

The exposure of each cow to SO<sub>2</sub> and VOCs was measured using air monitors located near each pasture for 205 herds during the 2001 breeding season. SO<sub>2</sub>, VOCs, and H<sub>2</sub>S were measured for a subset of 49 herds during the 2002 breeding season for which there was also pregnancy testing information. The density of facilities surrounding each pasture was also used as a secondary measure of exposure.

Pregnancy status was determined by herd veterinarians in fall 2001 and 2002, typically between September and November, when most cows were between 2 and 6 months into gestation. The calving interval for each cow was determined by comparing the records of calving dates for 2001 and 2002.

### Risk of Nonpregnancy

The risk of nonpregnancy was calculated by dividing the number of nonpregnant females by the number of females that had been examined for pregnancy.

Animal- and herd-management factors known or suspected to affect these parameters were measured. These factors included the age and breed type of the cow, whether the cow was purchased or born on farm, vaccination status, details of bull management on pasture, and the month of pregnancy testing. Cow body condition score was also measured.

The mean risk of nonpregnancy was 7.0% in 2001 and 8.2% in 2002. These findings were only slightly higher than the 6.5% reported from a mail-out survey by Alberta Agriculture in 1986/87; or the 6.6% reported from a smaller, on-farm, multi-year study of cow-calf operations in Central Alberta.

Reproductive success varied among the study herds: the 5% of herds reporting the lowest nonpregnancy rates

had less than 2.0% nonpregnant or open cows in 2001 and less than 2.1% in 2002. The 5% of herds with the highest nonpregnancy rates reported that more than 13.0% of cows were open at pregnancy testing in 2001 and 16.9% in 2002.

**Percentage of cows exposed to any oil- and gas-field facilities during each at-risk period used for examining associations between exposure and reproductive success**

Distance from facility	Non-pregnancy		Breeding-to-Calving	Disposal after pregnancy testing
	2001	2002	2001-2002	2001
within 1 mile of a well site	72%	70%	68%	72%*
within 1 mile of a battery site	34%	34%	27%	34%
within 5 miles of a gas plant or gas-gathering facility	68%	67%	62%	68%

\* For example, for the at-risk period for pregnancy status in 2001, 72% of cows were pastured within 1 mile of at least one well site during the period from first bull contact to the time the bulls were removed or to the date of pregnancy testing

**Exposure to contaminants, as measured by passive air monitors; median (5th to 95th percentile range)**

	SO <sub>2</sub> (ppb)	H <sub>2</sub> S (ppb)	VOCs (µg/m <sup>3</sup> )	
			as benzene	as toluene
Risk of non-pregnancy 2001	0.5 (0.2-1.1)*	--	0.18 (0.10-0.44)	0.31 (0.15-0.78)
Risk of non-pregnancy 2002	0.6 (0.1-1.4)	0.16 (0.07-0.51)	0.22 (0.13-0.50)	0.24 (0.07-0.56)
Breeding-to-calving interval	0.4 (0.2-1.0)	--	0.15 (0.02-0.39)	0.24 (0.07-0.56)

\*For example, for the at-risk period for pregnancy status in 2001, the typical cow was exposed to an average of 0.5 ppb during the period from first bull contact in 2001 to the time the bulls were removed or to the date of pregnancy testing. The reported average for the 5% of cows that were least exposed to SO<sub>2</sub> was less than 0.2 ppb, and the exposure reported for the 5% of cows that were most exposed was greater than 1.1 ppb.

The at-risk period for examining associations between exposure to emissions and the risk of nonpregnancy was defined as the period from first bull contact with the cows in spring to last bull contact or to pregnancy testing—whichever was shorter.

**2001 Breeding Season.** For 32,871 cows in 2001, neither exposure to increasing concentrations of SO<sub>2</sub> and VOCs measured as benzene and toluene nor well-site density was associated with an increase in the risk of nonpregnancy.

**2002 Breeding Season.** Passive air monitoring was scaled back at the end of June 2002, with the result that exposures during the at-risk period for nonpregnancy were available only for a subset of 49 herds in 2002.

For 9,038 cows from the 49 herds with extended air-monitoring data in 2002, exposure to increasing concentrations of SO<sub>2</sub>, H<sub>2</sub>S, and VOCs measured as benzene and toluene were not associated with an increase in the risk of non-pregnancy. However, the small sample size limited the power of the analysis to detect only relatively large exposure effects.

The study also found that for 31,052 cows from 197 herds in 2002, well-site density was not associated with an increase in the risk of non-pregnancy.

### Calving Interval

The calving-to-calving interval was defined as the number of days from the calving date in 2001 to the calving date in 2002. Calving interval is another indicator of cow fertility.

Calving dates were available for 22,402 cows with exposure data from 202 herds that calved in 2001 and that gave birth to a full-term live calf in 2002. The average calving-to-calving interval was 369 days. The finding is comparable to the 365 days reported in a 1986 study of 180 beef cow-calf herds in Ontario and to the 367 to 369 days for a smaller, but multi-year study of cow-calf herds in central Alberta.

The breeding-to-calving interval was used when examining the potential effect of exposure in the study, because the calving-to-calving interval includes the time from the previous calving to the time the bull is

**Summary of the risk of nonpregnancy and calving interval for the 2001 – 2002 cow-calf production cycles for all cows with complete exposure records**

Productivity Outcome	Study Population
Risk of nonpregnancy in 2001 (205 herds)	7.0% (2296/32871)
Breeding-to-calving interval 2001-2002 (202 herds)	305 days (standard deviation, 23)
Risk of nonpregnancy in 2002 for herds with complete air monitoring (49 herds) where air monitoring was continued after June 30, 2002.	8.2% (743/9038)

turned out with the cows. The length of the period from calving to the start of breeding is influenced by herd management decisions and can change from one year to the next affecting the length of the calving interval. The breeding-to-calving interval includes only the time from first bull contact to conception and then the gestation period.

For the study, the breeding-to-calving interval was defined as the difference between the date of first bull contact with the cows in spring 2001 and calving in 2002, or the difference between 21 days after calving in 2001 and calving in 2002, for cows that had not yet calved at the time of first bull contact. The 21-day period was intended as an adjustment factor that might represent the shortest possible time required for a typical cow to come into heat and to potentially be at risk of becoming pregnant again after calving.

The primary at-risk period considered for examining the association between exposure and the breeding-to-calving interval was the 30-day period after the bull was first turned out with the cows in spring 2001 .

The average breeding-to-calving interval was 305 days. There was no evidence of an association between the duration of the breeding-to-calving interval and exposure to SO<sub>2</sub>, VOCs measured as toluene, or well-site density. There was, however, an association between exposure to VOCs measured as benzene and a small increase in the duration of the breeding-to-calving interval—approximately 3 days for cows exposed to the highest 25% of VOC



concentrations measured as benzene, compared to cows exposed to the lowest 25% of measured concentrations.

There will be some error in estimating any potential effect of exposure on the time to conception, because the breeding-to-calving interval includes both the time to conception and the gestation period. The importance of the 3-day difference found in the study should be evaluated against the substantial variation in length of gestation among breeds and among animals of the same breed.

For example, average gestation for Angus cattle varies from 273 to 282 days and for Limousin cattle, 287 days to 290 days. Although general information on breed type was accounted for in the analysis, exact breed was not available for every animal enrolled in the study, so the potential to account for genetic differences in length of gestation was limited.

Cows are expected to produce one viable calf every year. Given that a typical gestation period for the common breeds used in Western Canada can range from 273 days to 290 days, this leaves between 75 and 92 days for the cow to come into heat after calving and successfully conceive.

## Risk of Disposal

The study also considered possible associations between emissions and the risk of pregnant cows being culled from the herds (or disposed of), after pregnancy testing in 2001 and before calving in 2002.

Although the calving interval was a good and practical means of estimating any potential effects of exposure on time to conception, this index would miss cows that were culled after pregnancy testing because they had become pregnant too late in the breeding season and were sold before calving. By examining the association between exposure and the risk of pregnant cows being culled, the potential association between exposure and late pregnancies that were eliminated from the herd by management decisions was determined. A limitation of this analysis was that the reason for culling was not consistently recorded.

The analysis included all cows removed or lost from inventory including those removed due to the sales of breeding stock, for other problems not related to reproductive failure, and cows that died.

The at-risk period for examining the association between exposure and the risk of culling was defined as the period from first bull contact with the cows in spring to last bull contact or pregnancy testing—whichever was shorter.

Of the 32,600 cows with follow-up data, 4,247 (13.0%) were recorded as removed from inventory before calving in 2002; and of the 4,247 cows, 50.8% (2,158) were reported as pregnant at the time of removal.

There was no evidence of an association between exposure to emissions from the oil and gas industry and an increased risk of pregnant cows being culled from the herds after pregnancy testing and before calving.

## 7. Abortion and Stillbirth

Potential associations between emissions from oil- and gas-field facilities and the risks of abortion and stillbirth were determined for more than 28,000 beef cows in 203 herds followed from the beginning of the breeding season in 2001 through calving in 2002.

The exposures of each cow to SO<sub>2</sub>, H<sub>2</sub>S, and VOCs over appropriate, biologically relevant, at-risk periods for abortion and stillbirth were calculated from air-monitoring data collected at each pasture, wintering, and calving area. The density of facilities surrounding each quarter section was also used as a second measure of exposure.

The abortion analyses also examined the effects of other potential risk factors, including: the cow's age, breed, vaccination status, and body condition; whether the cows were purchased or born on farm; and twin pregnancies. The stillbirth analyses considered the effects of: the cow's age, breed, vaccination status, and body condition; whether the cows were purchased or born on farm; the month of calving; the gender of the calf and whether the calf was a twin; and the temperature on the day of birth. Precipitation in the 2001 growing season was examined to account for the possible effects of drought on both outcomes.

### Risk of Abortion

An abortion was defined as a premature calving, judged to be at least 1 month before full term, or an assumed calf loss when a cow was diagnosed as pregnant but failed to calve. All pregnant cows with exposure data and for which a calving outcome could be determined were considered at risk of abortion in this analysis.

To determine the frequency or risk of abortion, 28,144 pregnant cows were followed from pregnancy testing in fall 2001 to the end of calving in 2002. The risk of abortion for the cows averaged 1.6% and for the herds, 1.6%. The frequency of abortions ranged from none in 20% of the herds to more than 4.0% in 5% of the herds.

Exposure of the cows to SO<sub>2</sub>, VOCs, and well-site density was measured for three at-risk periods for abortion. The first period ran from the first reported bull contact with the cows to the date of pregnancy testing. A second

exposure period, November and December, was examined for all cows that were still pregnant at the end of December to examine potential effects of exposure in late gestation. A third analysis determined the risk of abortion in any month, from the time of pregnancy testing to the end of May 2002, and examined the association with exposure in the preceding 3 months. Data on H<sub>2</sub>S were available for the latter two exposure periods.

No significant associations were found between the risk of abortion and exposure to SO<sub>2</sub>, H<sub>2</sub>S, VOCs measured as benzene and toluene, or between the risk of abortion and the density of surrounding well sites.

The study did, however, identify some other factors related to the risk of abortion in these herds. There was a

**Percentage of cows exposed to any oil- and gas-field facilities during each at-risk period used for examining associations between exposure and abortion and stillbirth**

Distance from facility	Abortion 2001-2002	Stillbirth 2001-2002	
	Full- prenatal	Full- prenatal	Last trimester
within 1 mile of a well site	72%	73%	63%
within 1 mile of a battery site	34%	37%	26%
within 5 miles of a gas plant or gas-gathering facility	68%	69%	64%

**Exposure to contaminants, as measured by passive air monitors; median (5th to 95th percentile)**

	SO <sub>2</sub> (ppb)	H <sub>2</sub> S (ppb)	VOCs (µg/m <sup>3</sup> )	
			as benzene	as toluene
Risk of Abortion (to pregnancy testing)	0.5 (0.2-1.1)	--	0.18 (0.10-0.47)	0.31 (0.15-0.79)
Risk of Stillbirth (entire prenatal)	0.7 (0.3-1.5)	--	0.30 (0.20-0.71)	0.39 (0.16-1.61)
Risk of Stillbirth (last trimester)	0.9 (0.5-1.9)	0.16 (0.11-0.37)	0.50 (0.32-1.17)	0.47 (0.11-4.44)

small but significant decrease in the risk of abortion associated with the use of bovine viral diarrhea and infectious bovine rhinotracheitis vaccines in these herds. For example, cows that were vaccinated with a modified live vaccine were on average 1.5 times less likely to abort than cows that were not vaccinated. There were some small differences in the effectiveness of modified live and inactivated products, depending on the time period examined. In addition, cows that were thin at pregnancy testing, and those that were pregnant with twins, were more likely to abort than other cows.

## Risk of Stillbirth

A stillbirth was defined as a calf that was dead at or within 1 hour of birth; or a calf that was found dead, had not been observed alive, and was obviously recently born. All calves born (dead or alive) to cows for which there was exposure data and that were born within one month of full term were considered in this analysis.

*In utero* exposure of the calves to emissions throughout gestation was measured from the time of first reported bull contact with the cows to the time of calving. Exposure data for this interval were available for 28,402 calves born to cows in 203 herds.

The potential effects of exposure were also examined during the last trimester of gestation—the last 3 months before calving.

Of the 28,402 full-term calves, 2.6% were born dead or died within 1 hour of birth. The risk of stillbirth varied between herds, with 5% of herds reporting no stillbirths and 5% reporting more than 6.4% stillbirths. Half of the herds reported that at least 2.5% of calves were stillborn.

There were no associations between the frequency of stillbirth and exposure to SO<sub>2</sub>, VOCs measured as benzene or toluene, or the density of well sites, regardless of whether the exposure was considered for the entire prenatal period or just the 3 months before calving. There was also no association between the risk of stillbirth and exposure to H<sub>2</sub>S in the last trimester of gestation.

Several other factors were found to be associated with the risk of stillbirth. Cows that were thin before calving,

## Summary of the risks of abortion and stillbirth for the 2001–2002 production cycle.

Productivity outcome	Study population <sup>a</sup>	Individual herds <sup>b</sup>	
		Mean	Median
Risk of abortion <sup>c</sup> (2001–2002)	1.6%	1.6%	1.3%
Risk of stillbirth <sup>d</sup> (2002)	2.6%	2.7%	2.5%

<sup>a</sup> Risk calculated across the entire study population (28,144 cows for abortion assessment and 28,402 calves for stillbirth assessment).  
<sup>b</sup> Risk calculated for each herd and then summarized across all 203 study herds.  
<sup>c</sup> Risk calculated for reported and assumed abortions from breeding season in 2001 through calving season in 2002.  
<sup>d</sup> Risk calculated for calves reported as having died full-term (> 8 months gestation) and when they were less than 1 hour old during the 2002 calving season.

first-calf heifers, and very old cows were more likely to have a stillborn calf. Twins, bull calves, calves born very early in the calving season, and calves that required assistance at birth were also more likely to be born dead or die very shortly after they were born.

The drought in 2001 also appeared to affect calf survival. Cows in areas with the lowest precipitation were more likely to give birth to a stillborn calf than cows in areas with the highest precipitation. This effect may have been related to the quality of pasture during early gestation or to the amount and types of stored feeds available during the winter feeding period.

The overall risks of abortion (1.6%) and stillbirth (2.6%) reported for the study herds were reasonably consistent with those reported for other beef cattle herds in Western Canada.

In conclusion, although the large sample size and detailed individual animal data provided sufficient statistical power to detect important associations, the study found no evidence of a difference in the risk of abortion or stillbirth across the observed range of exposures to emissions from the oil and gas industry, as measured by airborne contaminants or the density of surrounding facilities.

## 8. Calf Mortality and Treatment

Potential associations between emissions from oil- and gas-field facilities and the risk of calves dying or receiving treatment with pharmaceuticals were determined for 27,511 calves born to cows in 203 herds followed from the beginning of the breeding season in 2001 through calving in 2002.

Exposures of the calf, both *in utero* (during gestation) and in the early postnatal period, to SO<sub>2</sub>, H<sub>2</sub>S, and VOCs measured as benzene and toluene were calculated from data collected by air monitors located at the pastures and winter holding areas. The density of facilities surrounding each pasture and wintering area was used as a second measure of exposure.

Animal- and herd-management factors known or suspected to affect the risk of calves dying or receiving treatment were also considered. Factors examined included the cow's age, breed, vaccination status, and body condition; whether the cow was purchased or born on the farm; sex of the calf; whether twins were born; the month of calving; and average temperature on the day of birth. Additional factors examined included precipitation in the 2001 growing season, to account for the possible effects of drought, and the ecoregions where the herds were pastured, to account for differences in climate, nutrition, or vegetation that could affect calf survival.

### Risk of Calf Mortality

Calf mortality was defined as a calf that died more than 1 hour after birth and before it was 3 months old or before June 30, whichever happened first. Herd risk of calf mortality was the percentage of calves that were born alive but died before 3 months of age or June 30.

Two at-risk periods for exposure were considered when examining whether there was an association between *in utero* exposure of the calf and the risk of the calf dying:

- exposure of the developing calf measured from the time of first bull contact with the cow to the calving date, to study associations for the period just before conception to the end of gestation
- exposure of the developing calf measured from 3 months before calving to the calving date, to study

associations for the period of *in utero* exposure in the last trimester

All calves born alive between November 1, 2001, and June 30, 2002, from dams with exposure data, were included in analyses. Of these calves, 3.6% died within 3 months (or before June 30). The frequency of calf death varied among herds, with 5% of herds reporting no calf mortalities, 50% reporting at least 2.9%, and 5% reporting more than 8.2%.

Exposure to VOCs measured as benzene or toluene, and the density of surrounding well sites, was not associated with a significant increase in calf mortality for any of the at-risk periods. In addition, exposure to H<sub>2</sub>S during the last three months of gestation was not associated with increased calf mortality.

Exposure to SO<sub>2</sub> during the last trimester (as well as during the full prenatal period, which included the last trimester) was associated with a small increase in the risk of calf mortality. Calves exposed to the highest concentrations of SO<sub>2</sub> during the last trimester were about 1.4 times more likely to die before 3 months of age than calves

**Exposure to contaminants, as measured by passive air monitors, for determining the risks of calves dying or receiving treatment; median (5th to 95th percentile)**

At-risk period	SO <sub>2</sub> (ppb)	H <sub>2</sub> S (ppb)	VOCs as benzene (µg/m <sup>3</sup> )	VOCs as toluene (µg/m <sup>3</sup> )
Full prenatal	0.7 (0.3-1.5)	--	0.30 (0.20-0.71)	0.39 (0.16-1.65)
Last trimester	0.9 (0.5-1.9)	0.16 (0.11-0.37)	0.51 (0.32-1.17)	0.48 (0.11-4.47)
Early neonatal	0.8 (0.2-1.9)	0.16 (0.09-0.33)	0.46 (0.21-0.86)	0.18 (0.02-0.90)

**Percentage of calves exposed to facilities, calf mortality and treatment studies**

Distance from facility	Full prenatal	Last trimester
within 1 mile of a well site	72%	64%
within 1 mile of a battery site	37%	25%
within 5 miles of a gas plant or gas-gathering facility	69%	63%

exposed to the lowest concentrations. For example, the risk of dying before 3 months of age was estimated at 4.1% for a male calf born to a replacement heifer on a day in March where the temperature was 0°C and where the calf was exposed to the lowest measured concentrations of SO<sub>2</sub>. For the same calf from an area exposed to the highest measured concentrations of SO<sub>2</sub>, the risk of dying was estimated at 5.9%—an increase of 1.8% in this example.

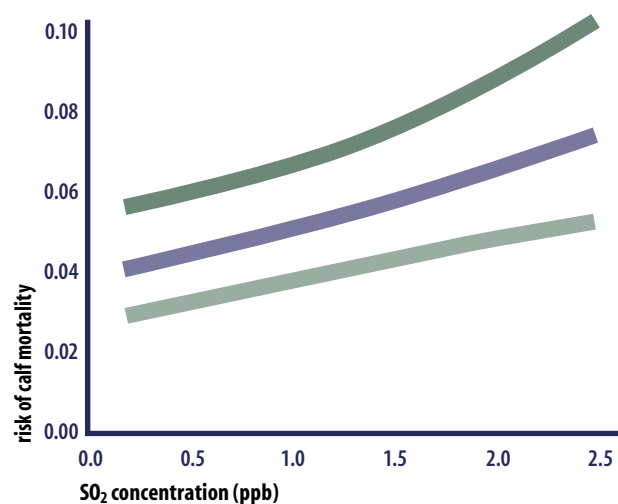
The potential effects on the calf of exposure during the first month of life were also determined. Associations were examined between exposure during the first month of life and the risk of dying after 1 month of age, and the association between exposure in the month of calving and the risk of a calf dying in any month during the calving season. Although the associations were not significant, the risk of calf mortality did tend to be higher in those calves most exposed to SO<sub>2</sub> during the postnatal period in both analyses, by an amount similar to that associated with *in utero* exposure.

### Risk of Calves Receiving Treatment

Treatment was defined as any pharmaceutical given to a calf for therapeutic or prophylactic indications. Risk of treatment was defined as the percentage of calves that were reported to have received treatment with a pharmaceutical.

The at-risk periods for determining possible associations between exposure of the cows during gestation and of the calves after birth and the risk of calves receiving treatment were defined as for calf mortality.

Of the calves born alive between November 1 and June 30, 12.9% were reported as having been treated in their first 3 months of life. The frequency of calf treatment varied among herds, with 5% of herds reporting that no calves received treatment, 50% reporting that at least 6.5% of calves received treatment, and 5% reporting that more than 48% of calves received treatment.



The average predicted risk of mortality in a male calf from a heifer born in March at 0°C increases with increasing exposure to SO<sub>2</sub> (centre line). The upper and lower lines are the upper and lower 95% confidence intervals for the predicted risk for each level of exposure.

Exposure during gestation to SO<sub>2</sub>, VOCs measured as benzene or toluene, and density of surrounding well sites was not associated with frequency of calf treatment.

As for the analysis of calf mortality, associations were also examined between exposure of the calf during the first month of life and the risk of treatment after 1 month of age, and between exposure in the month of calving and the risk of a calf being treated for the first time in any month during the calving season.

Exposure to H<sub>2</sub>S, VOCs measured as benzene or toluene, and SO<sub>2</sub> in the first month after calving was associated with a small increase in the risk of a calf receiving treatment after the first month of life. However, the risk of treatment increased in a manner consistent with a causal association only for exposure to H<sub>2</sub>S.

In conclusion, the study found evidence that exposure to some emissions from oil and gas industry facilities was associated with a small, adverse effect on calf survival and risk of treatment.

### Summary of associations between exposures to the oil and gas industry and the risks of calves dying or receiving treatment before 90 days of age or June 30, 2002.

Risks	SO <sub>2</sub>	H <sub>2</sub> S	Benzene	Toluene	Well-Site Density
death, when cows were exposed from first bull contact to calving	↑	N/A	—	—	—
treatment, when cows were exposed from first bull contact to calving	—	N/A	—	—	—
death, when cows were exposed for 3 months before calving	↑	—	—	—	—
treatment, when cows were exposed for 3 months before calving	—	—	—	—	—
death after 30 days, when calves were exposed from calving to 30 days	—	—	—	—	—
treatment after 30 days, when calves were exposed from calving to 30 days	↑	↑	↑	↑	—
— increasing exposure caused no change    ↑ increasing exposure increased the risk    N/A H <sub>2</sub> S not measured during prenatal period					



## 9. Diagnostic Pathology Findings

Gross postmortem and histological examinations were completed on aborted, stillborn, and live-born calves from the 2002 calving season that died before May 31, 2002, to determine the immediate cause of death and describe pathological lesions or disease processes.

This was the largest and most complete cross-sectional study of calf loss ever conducted in North America. The success of the study was due to the cooperation by herd owners in submitting calves for postmortem examination and to the quality of necropsies performed by participating veterinarians and their compliance in submitting targeted tissues for histopathological evaluation.

Findings, which provide unique insights into beef herd losses in Western Canada, supported work on assessing potential associations between exposure and the risk of lesions, as described in Section 10.

Diagnostic techniques included gross examination of carcasses (necropsy), microscopic examination of tissues (histopathology) from each calf, and specialized techniques to confirm diagnoses and identify infectious agents (histochemistry and immunohistochemistry).

Tissue samples were collected by herd veterinarians and submitted to the study office together with postmortem examination forms that included data on the age of the cow, date of abortion or calf death, duration of illness,

history of treatments if any, and condition of the carcass. All identifying information was removed before forwarding the coded samples to the diagnostic laboratory.

Herd veterinarians examined 1,689 carcasses and submitted tissues from 1,678. Eleven carcasses (3 aborted fetuses, 3 stillborn calves, and 5 live born calves) were considered too scavenged or decomposed to warrant submission.

A standard set of tissues was requested from all postmortem examinations. Herd veterinarians submitted tissues from 180 aborted fetuses, 557 stillbirths, and 941 full-term calves that died when they were more than one hour old.

One pathologist, certified by the American College of Veterinary Pathologists, completed the histological examinations of all submissions. This ensured consistency in histological examinations, which is critical for ensuring the quality of data for subsequent studies.

Success in establishing the cause of death increased with the age of the calf. Causes of death, or potentially significant histological lesions, were reported for 52% of aborted fetuses, 68% of stillborn calves, 89% of calves 1-3 days old, and 94% of older calves.

**Most common causes of the death of calves in each age category (%)**

Aborted (n=183)		Stillborn (n=560)		Newborn (n=388)		>3days old (n=558)	
Unknown	48.1	Unknown	32.3	Heart Failure	12.6	Starvation (primary and secondary)	19.7
Placental infection	13.1	Dystocia	30.2	Unknown	10.8	Abomasal ulcer/torsion/perforation	15.4
Cow Death Prior to Birth	9.3	Thyroid Gland Malfunction	12.7	Dystocia	8.2	Enteritis/colitis	9.5
Heart Failure	8.2	Heart Failure	10.7	Thyroid Gland Malfunction	7.7	Heart Failure	5.9
Thyroid Gland Malfunction	8.2	Congenital Anomaly	2.9	Generalized bacterial infection	7.5	Unknown	5.7
Congenital Anomaly	3.8	Hemosiderosis	2.5	Trauma	7.2	Generalized bacterial infection	5.6
Disseminated Intravascular Coagulation	1.6	Placental infection	1.6	Pneumonia	5.4	Intestinal accident	5.2
		Skeletal myopathy	1.4	Skeletal Myopathy	4.6	Pneumonia	4.1

In addition to these common causes, there were numerous other causes of death that affected only a small number of animals, hence, the percentages in the table do not total 100

**Key findings included the following:**

- Noninfectious disease processes were considered the most likely cause of most fetal and calf deaths.
- Cardiac lesions, thyroid abnormalities, and congenital anomalies were the most frequent pathological findings associated with noninfectious causes of abortions.
- Infectious or inflammatory disease associated with placentitis was present in 13% of all abortions.
- Problems associated with parturition (dystocia) were the most common reason for stillborn calves (30%). Dystocia (8%) was also an important cause of neonatal calf loss, second only to cardiac failure (13%).
- Infectious disease, uncommon in stillborn calves (2%), was more frequently associated with mortality in neonatal calves (20%) and calves older than 3 days (33%).
- Starvation, which was often secondary to or found in association with some other disease process, was the immediate cause of loss in another 20% of older calves.
- Abomasal lesions, including inflammation, ulcers, torsions and perforation, accounted for 15% of older calf losses.
- Significant histopathological findings in the respiratory tract included pneumonia and pulmonary vascular lesions. The relative frequency of different types of pneumonia varied by age class.
- Hypomyelination of the sciatic nerve was the most common lesion of the nervous system.
- The most frequent lesions in the immune system were atrophy or depletion of the thymus and Peyer's patches of the ileum.

Findings included previously undocumented pathological changes and unique prevalence estimates for other conditions of interest in beef calves. Examples included degenerative and hypoplastic thyroid lesions, hemosiderosis of the liver, skeletal muscle myopathy, and myocardial necrosis with features atypical of selenium deficiency. Cardiac muscle necrosis, hypomyelination, and pulmonary vascular lesions were strongly associated with the occurrence of skeletal myopathy.

**Most frequent histological lesions in each age category (%)**

	Aborted	Stillborn	Newborn	>3days old
Pneumonia	22	6	24	18
Sciatic Nerve Hypomyelination	2	5	6	6
Thymus Atrophy	15	6	20	35
Any Thyroid Lesion	30	39	29	19
Skeletal Muscle Myopathy	6	19	42	39
Cardiac myopathy or necrosis	9	14	24	19
Hemosiderosis	8	28	30	13

## 10. Assessing the Risk of Lesions in Calves

Potential associations between emissions from oil- and gas-field facilities and the health of beef calves were investigated by assessing the risk of lesions in tissue samples collected from 1,531 aborted fetuses, stillbirths, and calf mortalities from the 2002 calf crop.

Tissues from the immune, nervous, and respiratory systems, as well as skeletal and cardiac muscle and the thyroid gland, were among the samples collected by herd veterinarians during postmortem examinations and evaluated by a pathologist certified by the American College of Veterinary Pathologists as described in Section 9.

Each tissue was classified by the presence or absence of a series of specified lesions, as follows:

- **Immune System.** Lymph nodes, spleen, thymus, and Peyer's patches from the ileum were examined for lympholysis, necrosis, inflammation, degeneration, presence of anomalous development, and signs of infection.
- **Nervous System.** Brain, spinal cord, and sciatic nerve tissues were examined for necrosis, inflammation, degeneration, presence of anomalous development, and signs of infection.
- **Respiratory System.** Lung and tracheal tissues were examined for necrosis, inflammation, degeneration, presence of anomalous development, signs of infection, and cell proliferation.

The results of histological examinations of tissues from skeletal and cardiac muscle, and the thyroid gland, were also included in this analysis, after a review of the diagnostic reports. These tissues were examined for necrosis, inflammation, degeneration, presence of anomalous development, signs of infection, autolysis, and, for the thyroid gland, cell proliferation.

Exposures of the calves *in utero* and in the early postnatal period to SO<sub>2</sub>, H<sub>2</sub>S, and benzene and toluene (as surrogates for all VOCs) were measured using data from passive air monitors installed at pastures and wintering and calving areas. The density of oil and gas well sites near each pasture was used as a second measure of exposure.

Other potentially important animal- and herd-management risk factors were examined as potential risk factors for the occurrence of lesions of interest. They included the cow's age, breed, and vaccination status; whether the cows were purchased or born on the farm; the age of the calf at death, gender of the calf, and whether twins were born; and the temperature on the day of birth.

Potential associations between emissions and the risk of lesions developing in tissues were examined for two at-risk exposure periods.

The first period covered only postnatal exposure and was determined only for calves that were at least 4 days

**Summary of associations between exposures to the oil and gas industry and the risk of lesions in the immune, nervous and respiratory systems, skeletal and cardiac muscle, and the thyroid glands of aborted fetuses, stillbirths, and calf mortalities from the 2002 calf crop.**

Risk of lesions in:	SO <sub>2</sub>	H <sub>2</sub> S	Benzene	Toluene	Wells
immune system with postnatal exposure	—	—	—	—	—
immune system with exposure from first bull contact to death	—	NA	—	—	—
nervous system with postnatal exposure	—	—	—	—	—
nervous system with exposure from first bull contact to death	—	N/A	—	—	—
respiratory system with postnatal exposure	—	—	↑	↑	—
skeletal & cardiac muscle with postnatal exposure	—	—	—	—	—
skeletal & cardiac muscle with exposure from first bull contact to death	↑	N/A	—	—	—
thyroid gland with exposure from first bull contact to time of death	—	N/A	↑	—	—
— increasing exposure caused no change    ↑ increasing exposure increased the risk    N/A H <sub>2</sub> S not measured during prenatal period					

old when they died. It started at the calving date, and ended at the earlier of the reported date of death or 1 month of age.

The second at-risk period covered prenatal and postnatal exposure. It started with the time of first bull contact with the cow and ended with the death of the calf.

### Risk of Lesions in the Immune and Nervous Systems

Potential associations were examined between prenatal and postnatal exposure and lesions in the immune or nervous systems of the calves.

Lesions of degeneration, lympholysis, and necrosis were most common in calves from 4 days to about 6 weeks old. Lympholysis was identified in at least one tissue from 49% of calves examined, and was the most commonly recorded lesion in the immune system.

Lesions in the nervous system were rare compared with those identified in the immune and respiratory systems.

Exposures to SO<sub>2</sub>, H<sub>2</sub>S, and VOCs measured as benzene and toluene were not associated with an increased risk of lesions in tissues of either the immune or nervous systems of the calves.

### Risk of Lesions in the Respiratory System

Potential associations between postnatal exposure and lesions in the respiratory system were examined in calves that died after they were more than 3 days old. The *in utero* period was not considered because the initial exposure of interest in this analysis was direct inhalation.

Increasing exposure, after birth, to VOCs measured as benzene and toluene was associated with an increased risk of lesions in the respiratory tissues of calves that died after they were more than 3 weeks old. Exposure to SO<sub>2</sub> and H<sub>2</sub>S was not associated with the occurrence of respiratory pathology.

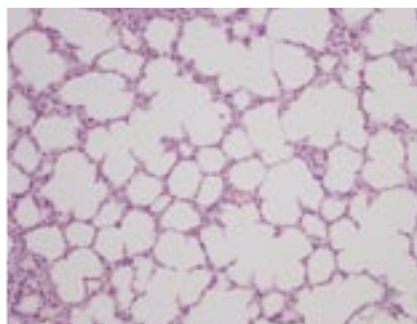
### Risk of Lesions in the Thyroid Gland and Skeletal or Cardiac Muscle

Potential associations between exposure and the presence or absence of lesions in the thyroid gland and either skeletal or cardiac muscle were examined in response to interest generated by histopathological findings.

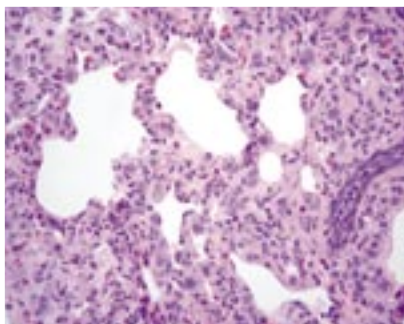
Thyroid lesions occurred in more than 18% of all calves with exposure data, most frequently in stillborn calves. Three types of thyroid lesions were identified: follicular hypoplasia, follicular degeneration with or without necrosis, and goiter or follicular hyperplasia. The thyroid glands affected were often characterized by a lack of colloid within the follicular cells.

The most common lesions in skeletal and cardiac muscle were degeneration and necrosis. Muscle lesions were most common in calves that died before 3 weeks of age; these lesions were identified in more than 25% of tissues examined.

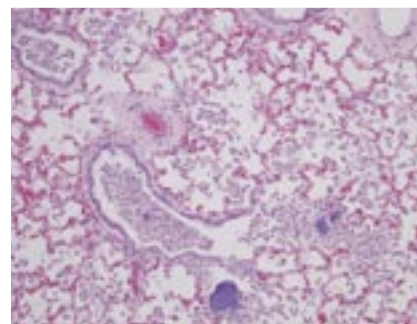
Increased exposure to SO<sub>2</sub> during gestation was associated with a small increase in the risk of lesions in skeletal and cardiac muscle, and increased exposure during gestation to VOCs measured as benzene was associated with a small increase in the risk of lesions in the thyroid gland.



Normal lung tissue, live newborn calf



Interstitial pneumonia, 40-hour calf



Acute suppurative bacterial bronchopneumonia, 28-day-old calf

## 11. Yearling Immune System Health and SO<sub>2</sub>, H<sub>2</sub>S, and VOCs

Potential associations between exposure to airborne emissions of SO<sub>2</sub>, H<sub>2</sub>S, and VOCs from oil-and gas-field facilities and the structure and function of the immune systems of yearling beef cattle were investigated using blood samples collected from 27 mixed-breed herds in Alberta and Saskatchewan in spring 2002.

A parallel study that considered emissions of fine particulate matter, polycyclic aromatic hydrocarbons, and selected metals in 22 of the herds is summarized in Section 12.

The yearling cattle, primarily replacement heifers, were exposed to a range of emissions from oil- and gas-field facilities. Yearling animals were chosen for study to minimize the potential for age-related differences in immune system outcomes and to take advantage of the exposure history available for these animals.

Exposure of the cattle to SO<sub>2</sub>, H<sub>2</sub>S, and VOCs (measured as benzene and toluene) was determined from data collected by monitors installed on and near pastures and wintering areas. The mean exposure of each animal was calculated for the 6-month period before the start of immune system assessment. Exposure was further evaluated by determining the proximity of study animals to emission sources such as well sites.

Potential associations between airborne contaminants and the structure and function of the immune systems of the animals were investigated in two ways.

The first was to determine concentrations of specific types of white blood cells in peripheral blood. These blood cells—B-lymphocytes and populations of four T-lymphocyte subtypes (CD4, CD8,  $\gamma\delta$ , and WC1)—play critical roles in the immune response to infection.

The second method of assessing the immune system was to measure antibody production in response to vaccination.

The project veterinarians visited the study herds three times during this investigation. On the first visit, they collected blood samples from a select group of yearling cattle, then inoculated the animals with a 2-ml dose of a commercial, inactivated rabies vaccine licensed for use in cattle. On the second visit, 3 weeks later, they collected a

second blood sample and inoculated the animals with a second 2-ml dose (booster) of the vaccine. On the third visit, 6 weeks after the first, they collected a final blood sample.

Concentrations of specific lymphocyte subtypes were determined in blood samples from 574 yearlings in the 27 herds sampled during the first visit. Rabies antibody concentrations were measured in blood samples from all three visits. Increases in the concentrations of rabies antibody between the first and third visits (i.e., the response to vaccination) were determined for 537 yearlings from the 21 herds available for testing at that time.

Increasing exposure to SO<sub>2</sub> and proximity to well sites were not significantly associated with concentrations of specific lymphocyte subtypes in circulating blood, nor with the production of antibodies in response to the rabies vaccination.

Increasing exposure to VOCs measured as toluene was associated with decreasing numbers of CD4 T-lymphocytes in peripheral blood. The number of CD4 T-cells was 30% lower in cattle exposed to VOCs measured as toluene at concentrations > 0.82  $\mu\text{g}/\text{m}^3$  (highest quartile) compared with cattle exposed to concentrations < 0.41  $\mu\text{g}/\text{m}^3$  (lowest quartile).

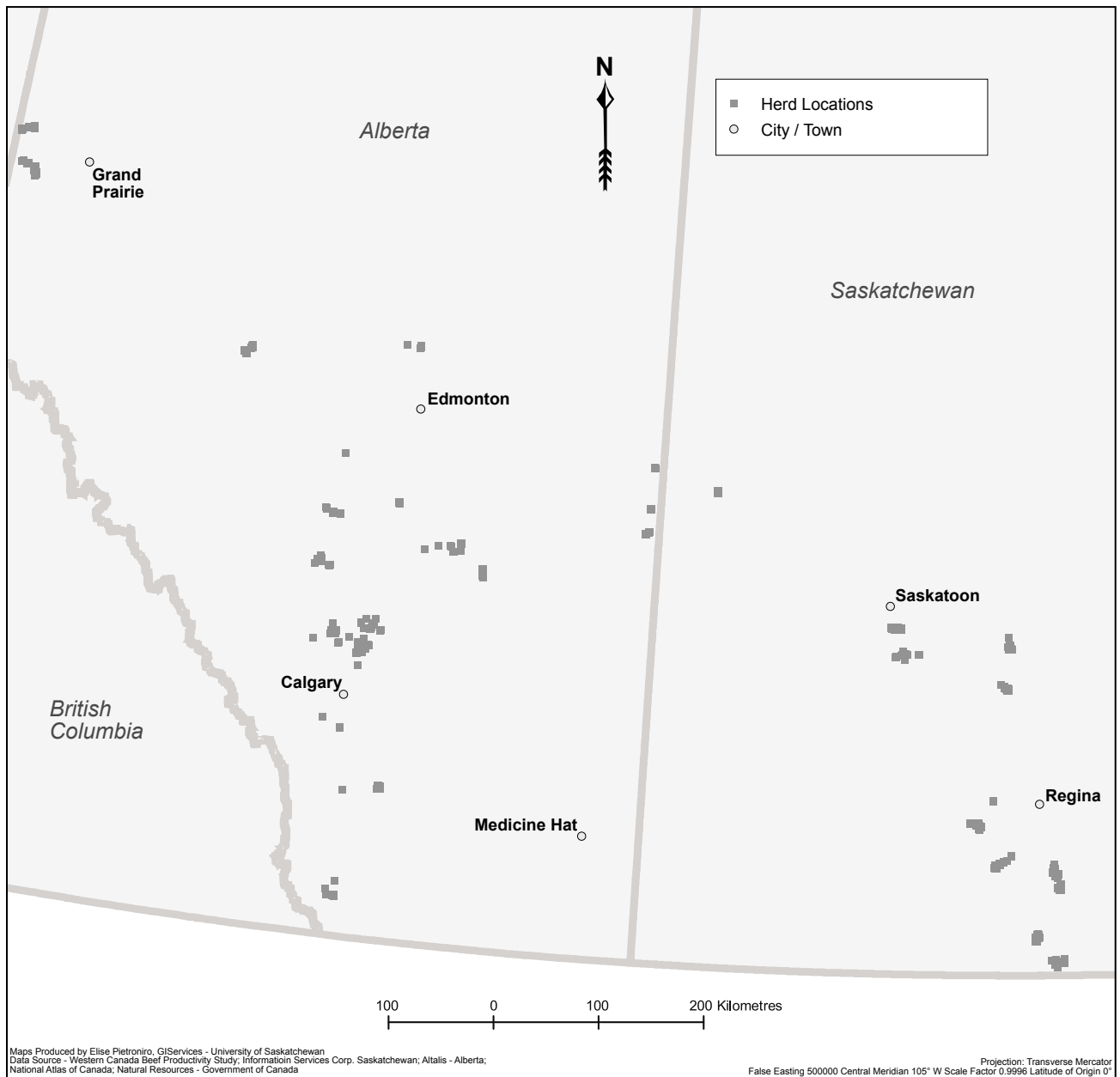
Exposure to VOCs measured as toluene was also associated with  $\gamma\delta$  and WC1 T-lymphocyte numbers, but the response did not consistently increase with exposure.

Increasing exposure to H<sub>2</sub>S measured as a continuous variable was associated with a small increase in antibody production, but the association was not significant when exposure was categorized into quartiles.

No other significant associations were observed between measures of exposure and the immune system outcomes examined in the study.

The statistical power of this aspect of the study was limited by the sample size. A larger sample size with more groups, permitting the detection of smaller differences across groups, would have been ideal, but practical considerations limited the number of herds that could be examined and the number of samples that could be processed.





*Locations used by the 27 herds taking part in the immunology study*

Results suggest that exposure to emissions from oil- and gas-field facilities may alter the immune systems of beef cattle; however, the biological significance of these alterations is unclear.

## 12. Yearling Immune System Health and PM<sub>1.0</sub>, PAHs, and Metals

Potential associations between exposure to airborne emissions of fine particulate matter (PM<sub>1.0</sub>), polycyclic aromatic hydrocarbons (PAHs), and metals from oil- and gas-field facilities and the structure and function of the immune system of beef cattle were investigated using blood samples collected from yearling cattle in 22 mixed-breed herds in Alberta and Saskatchewan in spring 2002.

A parallel study of SO<sub>2</sub>, H<sub>2</sub>S, and VOCs and the immune system health of yearling cattle in 27 herds, including the 22 discussed here, is summarized in Section 11.

The herds were selected from areas with a range of potential exposures to emissions from oil- and gas-field facilities. Particulate air monitors were placed near each wintering area in January and February 2002.

Exposure of the cattle to mean monthly airborne concentrations of PM<sub>1.0</sub>, PAHs, and metals was determined from two sets of air-sampling data. The first set included all exposure data available for each herd until the completion of blood sample collection in that herd. The second included only data collected in the 30 days before the first blood samples were taken.

As in Section 11, potential associations between airborne contaminants and the structure and function of the immune systems of the animals were investigated in two ways. The first was to determine concentrations of white blood cells in samples of peripheral blood. These blood cells—B-lymphocytes and populations of four T-lymphocyte subtypes (CD4, CD8, γδ, and WC1)—play critical roles in the immune response to infection.

The second method was to measure antibody production in response to vaccination. Project veterinarians visited the study herds three times. On the first visit, they collected blood samples from a select group of yearling cattle, then inoculated the animals with a 2-ml dose of a commercial, inactivated rabies vaccine licensed for use in cattle. On the second visit, 3 weeks later, they collected a second blood sample and inoculated the animals with a second 2-ml dose (booster) of the vaccine. On the third visit, 6 weeks after the first, they collected a final blood sample.

Concentrations of specific lymphocyte subtypes were determined in blood samples from 469 yearlings from the

22 herds sampled during the first visit. Increases in the concentration of rabies antibody between the first and third visits (i.e., the response to vaccination) were determined for 446 heifers from 18 herds available for testing.

### PM<sub>1.0</sub>

The geometric mean monthly concentration of PM<sub>1.0</sub> in the ambient air to which study animals were exposed from January 1, 2002 to June 30, 2002, was 7.1µg/m<sup>3</sup>. There was no association between exposure to PM<sub>1.0</sub> and immune system outcomes.

### PAHs

Concentrations of 24 PAH were measured during the same period. Naphthalene and 1-methylnaphthalene were found in the highest concentrations, with geometric means of 5.6 ng/m<sup>3</sup> and 2.2ng/m<sup>3</sup>, respectively.

It was impractical to examine potential associations between immune system outcomes and exposure to all 24 PAHs. To make the best use of the data, PAH concentrations were summarized for groups of PAHs with similar chemical properties and distribution patterns in the environment, using factor analysis (Research Appendix 4). The resulting factor scores provided a summary value for each of four groups of PAHs, for use in determining potential associations with immune system outcomes.

No associations were found, for either of the two exposure periods examined, between measured airborne concentrations of PM<sub>1.0</sub> or PAHs and lymphocyte counts or response to rabies vaccination.

### Metals

Mean metal concentrations, measured in the particulate matter samples, were very low (Section 3). Of the six metals with known potential to affect immune function, cadmium, lead, and selenium were below the level of quantification at all 22 herds. Airborne chromium concentrations were slightly above the level of quantification at 12 herds; copper, at 3 herds; and zinc, at 2 herds.

Given the small number of samples in which metals were detected, potential associations between immune system outcomes and concentrations of metals were not examined.

## 13. Calf Immune System Health

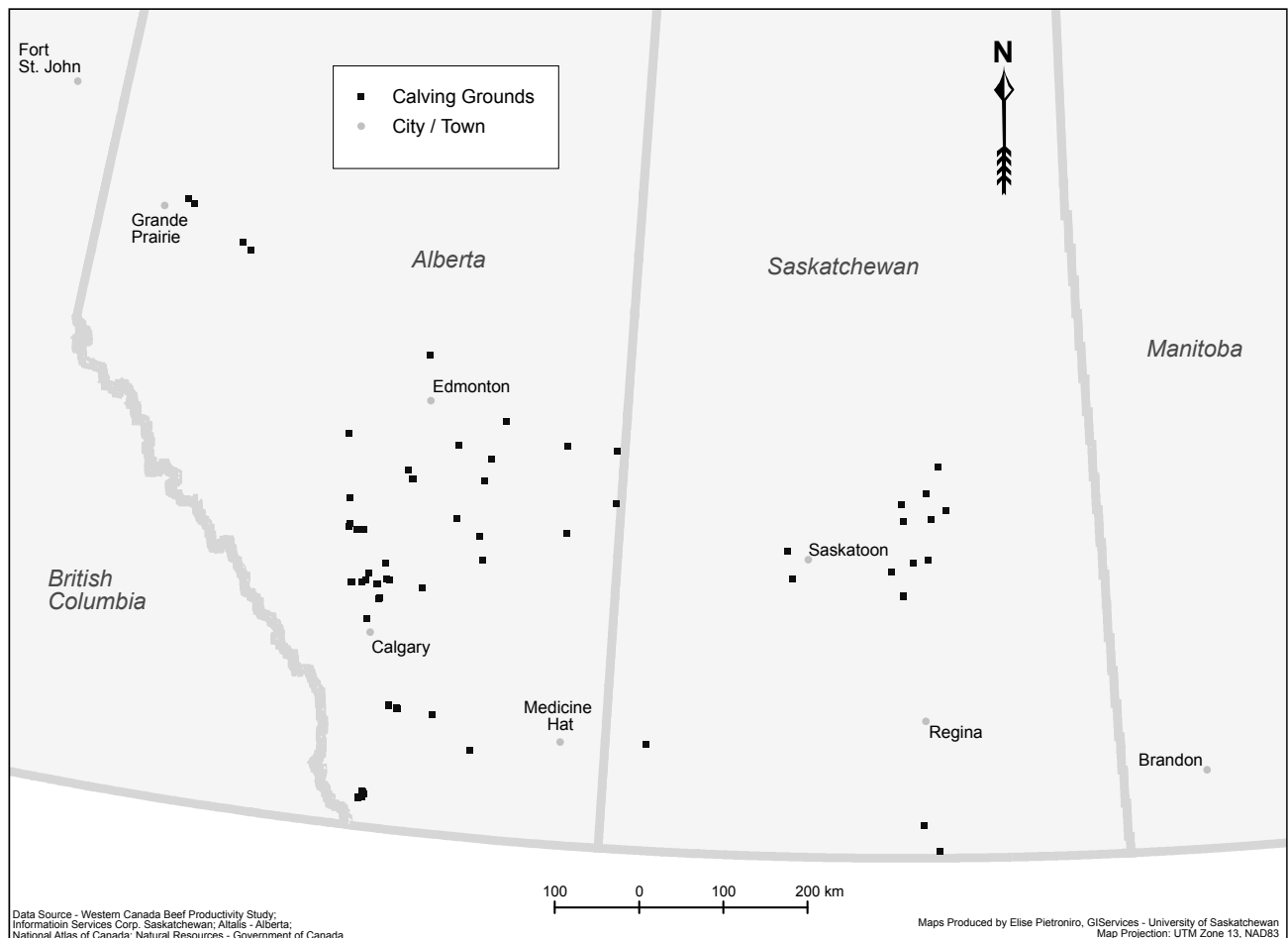
Potential associations between exposure to airborne emissions of SO<sub>2</sub> and VOCs from oil-and gas-field facilities and specific measures of the immune status of newborn calves were investigated using blood samples collected from 325 calves from 60 study herds in Alberta and Saskatchewan in spring 2002.

The immune system of young calves continues to develop after birth, leaving neonates at a relatively increased risk of contracting an infectious disease during the first weeks of life. This investigation was particularly relevant because much of the historical concern among livestock producers living near oil and gas facilities has centred on the occurrence of infectious diseases in young calves.

For this part of the study, the project veterinarians collected samples from calves available during regularly scheduled herd visits. Most of the calves were between 3 and 8 days old.

Mean *in utero* exposures to emissions were estimated for each calf, for the period from the earliest possible breeding date for the cow to the calving date. Levels of exposure to SO<sub>2</sub> and VOCs were determined from passive monitors installed on and near pastures, wintering areas, and calving grounds. The density of facilities surrounding each pasture was used as second measure of exposure.

More than 70% of the calves were exposed—*in utero*—to either a well site or a large field facility. The range of measured airborne concentrations of SO<sub>2</sub> and VOCs was



Locations of the 60 herds where blood samples were collected from 325 newborn calves

similar to the concentrations reported in Section 8 for exposure during gestation.

Potential effects of exposure on the immune systems of newborn calves were assessed by determining concentrations of specific types of white blood cells in peripheral circulation using flow cytometry. Cells populations evaluated included B-lymphocytes and four T-lymphocyte subtypes (CD4, CD8,  $\gamma\delta$ , and WC1), all of which play critical roles in the immune response to infection.

Exposure to SO<sub>2</sub> and the density of surrounding facilities were not associated with numbers of B-lymphocytes and the CD4, CD8,  $\gamma\delta$ , and WC1 T-lymphocyte subtypes. However, in calves exposed to VOCs measured as benzene at concentrations  $\geq 0.38 \mu\text{g}/\text{m}^3$  (highest quartile) compared with calves exposed to concentrations  $< 0.28 \mu\text{g}/\text{m}^3$  (lowest quartile), the numbers of CD4 T-lymphocytes averaged 42% lower and the numbers of CD8 T-lymphocytes averaged 43% lower.

Similarly, in calves exposed to VOCs measured as toluene at concentrations  $\geq 0.71 \mu\text{g}/\text{m}^3$  (highest quartile) compared with calves exposed to concentrations  $< 0.35 \mu\text{g}/\text{m}^3$  (lowest quartile), the number of CD4 T-lymphocytes was 40% lower.

Because of the small number of calves in this part of the study, there was no conclusive way to examine the relationship between the numbers of CD4 or CD8 cells and health status; however, the findings may indicate that exposure could affect a crucial component of the immune system.

In conclusion, results suggest that exposure to emissions from oil- and gas-field facilities may alter the immune systems of neonatal beef cattle; however, the biological significance of these alterations is unclear.

## 14. Reproductive Performance of European Starlings

Potential associations between exposure to emissions of  $\text{SO}_2$ ,  $\text{H}_2\text{S}$ , and VOCs from oil- and gas-field facilities and the reproductive performance of European starlings (*Sturnus vulgaris*), as measured by hatching and fledging success, and nestling growth, were examined in breeding starlings that used nest boxes that were placed on a subset of cattle ranches taking part in the beef productivity study in 2001 and 2002.

European starlings were chosen as a sentinel wildlife species to complement research on beef cattle productivity. Starlings have been used as indicators of environmental contamination in previous studies.

The widespread distribution of this species across the study area, its tolerance of human activity, and its willingness to use nest boxes made it a suitable model for the investigation. In addition, the use of an avian model was appropriate, because the contaminants of concern in this study were airborne, and passerine birds are more sensitive to most inhaled toxicants than mammals, as a result of the unique anatomy and physiology of their respiratory systems.

To attract breeding starlings, 300 plywood nest boxes were installed in suitable locations on 30 of the 205 participating cattle ranches during April 2001. Sites were selected based on proximity to emission sources reported by herd owners. The sites represented a wide range of exposure potentials, from those with a high density of surrounding oil and gas facilities and observed flaring, to those with little or no exposure.

The ranches were located in a band across central Alberta (24) and Saskatchewan (6) in geographically and ecologically homogeneous zones to reduce the potential effects of variations in habitat and weather.

The six sites in Saskatchewan were dropped from the study in 2002 because few or no starlings nested in the boxes at those sites.

Data were collected on the eggs, hatchlings, and fledglings in occupied boxes, starting in May of each year. Starlings occupied 136 of the 300 nest boxes in 2001, and 128 of the 240 boxes in 2002.

Monthly mean concentrations of airborne  $\text{SO}_2$ ,  $\text{H}_2\text{S}$  (2002 only), and VOCs at the nest boxes were obtained

using data from air monitors located near the nest sites, for use in the cattle study. If there was more than one monitor within 1.6 km, an average concentration was used. If there were no monitors within 1.6 km, concentration data from the nearest monitor were used.

Numbers of oil and gas well sites within 1.6 km, and numbers of gas plants and gas-gathering system facilities within 8 km, as well as data on flaring and venting from each facility, were linked to the location of each nest box using a GIS system. The density of surrounding facilities and reported emissions from these facilities were examined as a second method of exposure measurement.

Typical nest box





Time-weighted averages for all measurements of exposure were calculated for each nest box for three at-risk periods.

The first at-risk period began 7 days before laying, and ended on the day when the next-to-last egg was laid (which is also when incubation generally starts). The second period began 7 days before laying, and ended on the day that hatching started. The third period began 7 days before laying, and ended with fledging of the brood 19 days after hatching. Each of the three risk periods started 7 days before laying began, because of the uncertainty about the locations and feeding ranges of the birds any earlier than that.

Associations were assessed between exposure and egg weight and egg volume during the first period; between exposure and hatching failures during the second period; and between exposure and fledging failures, as well as chick weight and size, during the third period.

Data on mean daily temperatures and mean daily precipitation during each risk period were recorded as factors potentially affecting reproductive success. The percentage of nests that contained eggs was also recorded for each farm and for each ecoregion in which the nests were located.

In 2001, starting May 1, nest boxes were checked for signs of nest building every 2 or 3 days. After the first egg was laid, the nest was monitored until the clutch was complete. Once an average clutch of five eggs was complete (no additional egg after two consecutive visits), each egg in the nest was weighed and measured. If there were fewer than five eggs, then the eggs were weighed and measured after one visit with no new eggs. If more than five eggs were laid, the additional eggs were weighed and measured when discovered.

Once the eggs were weighed and measured, the box was not revisited until 10 days after laying had begun. Then the box was checked every 2 days, to determine the hatching date. At 19 days after hatching, the chicks were weighed and the length of their 10th primary feather was measured as an indication of development.

The same sequence was followed in 2002, except that the tarsal length of the chicks was also measured as an index of body size and growth. Tarsal length is the length of the tarsometatarsal bone, from the level of the hock joint to the bottom of the foot.

Hatching and fledging failures were not significantly associated with exposure, which included airborne SO<sub>2</sub>, H<sub>2</sub>S, VOCs measured as benzene and toluene, and the density of surrounding oil and gas-well sites.

In addition, nestling weight, egg weight, and egg volume were not associated with any measurement of contaminant exposure.

However, nestling size, based on tarsometatarsal bone length, decreased significantly with increasing exposure to VOCs measured as benzene and toluene in 2002. Tarsometatarsal bones were 2.0 and 2.3 mm shorter in nestlings exposed to VOCs measured as benzene and toluene, respectively, in the highest quartile ( $> 0.37 \mu\text{g}/\text{m}^3$  for benzene and  $> 0.39 \mu\text{g}/\text{m}^3$  for toluene) compared with nestlings exposed to concentrations in the lowest quartile ( $\leq 0.19 \mu\text{g}/\text{m}^3$  for benzene and  $\leq 0.20 \mu\text{g}/\text{m}^3$  for toluene). For birds exposed to the upper range of VOCs measured as benzene and toluene concentrations, respectively, this represents a reduction in bone length of 7.1 and 8.1%, relative to average bone length.

The significance of this finding on fledgling fitness and ability to survive is uncertain. Tarsometatarsal length was not measured in 2001, and there was no association between exposure and fledgling development as determined by length of the 10th primary feather in either 2001 or 2002.

In conclusion, apart from an association between decreased nestling size and exposure to VOCs measured as benzene or toluene, there were no indications that starlings living near oil and gas facilities and exposed to greater levels of contaminants were at increased risk of reproductive failure.

## 15. Starling Immune System Health

Potential associations between exposure to airborne emissions of SO<sub>2</sub>, H<sub>2</sub>S, and VOCs from oil- and gas-field facilities and the structure and function of the immune system of European starlings (*Sturnus vulgaris*) were examined in nestling birds that used nest boxes on 30 study sites in Alberta and Saskatchewan in 2001, and 24 study sites in Alberta in 2002.

This investigation was completed in parallel with the investigations of potential associations between starling reproductive success and levels of emissions, as described in Section 14.

Immunological endpoints evaluated during the first field season (2001) included:

- primary humoral immune response, measured by IgM production following immunization with sheep red blood cells (2001 only)
- T-cell response to intradermal injection of a mitogen (phytohemagglutinin)
- somatic indices of lymphoid organs, including spleen and bursa of Fabricius weights as a function of total body weight

- heterophil/lymphocyte (H/L) ratios, as determined by differential cell counts on blood smears.

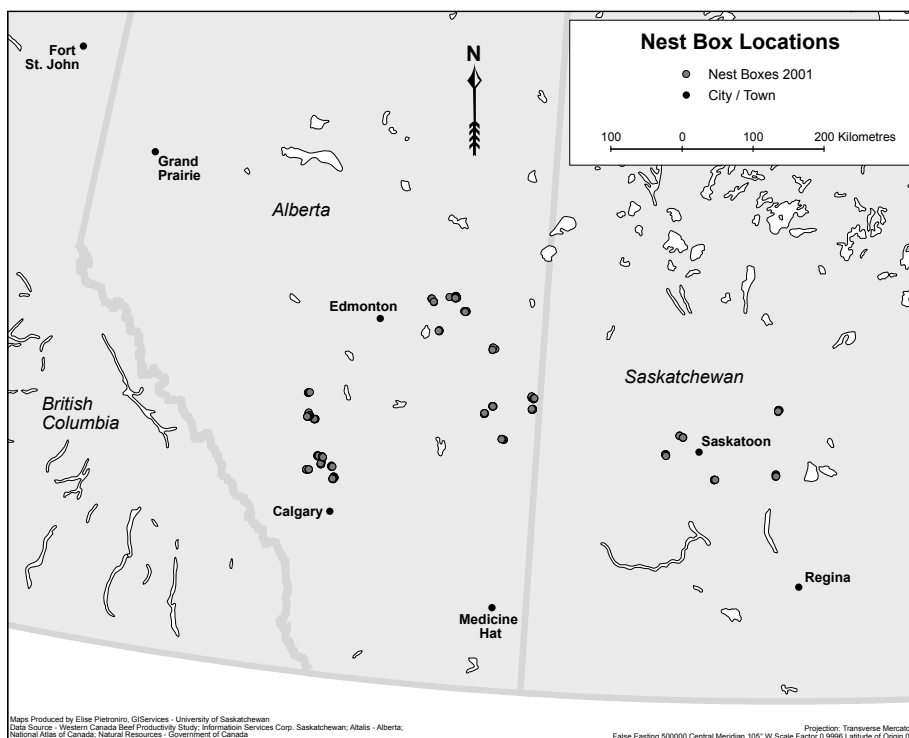
To further characterize immune system effects in the second field season (2002), the following were added:

- evaluation of the secondary humoral immune response, by measuring IgY production in response to immunization with bovine serum albumin
- histopathological evaluations of immune organs.

No significant associations were found between exposures to airborne emissions and any of the immunological endpoints evaluated, except for the H/L ratios.

Increasing concentrations of airborne SO<sub>2</sub> and H<sub>2</sub>S were associated with increased H/L ratios in the peripheral blood of nestling starlings in 2002. An increased H/L ratio is indicative of stress in some birds. However, the association was not observed in 2001.

The significance of this finding to nestling immune competence is likely to be minimal.



Nest boxes in Alberta and Saskatchewan in 2001. Each open circle represents one of 30 farms, each with 10 nest boxes, for a total of 300 boxes.

## 16. Water Quality Testing

In fall 2002, herd veterinarians collected samples from sources of drinking water, primarily from wells, on the home quarters of a subset of 53 study herds.

The water samples were collected from water supplies of all herds where air-monitoring data were collected after June 30, 2002, and included those herds that were part of the immunotoxicology studies and particulate monitoring studies (Sections 11 and 12).

The samples reflected what the cattle had been drinking during winter 2001/02. Samples were analysed for a range of materials, including pesticides, soil sterilants, insecticides, volatile organic compounds, phenolics, and base/neutral extractables.

Concentrations of volatile organic compounds, soil sterilants, and herbicides were undetectable in all samples except as noted below. Inorganic substances were either undetectable or within water quality guidelines for most samples.

In two herds, three organic substances (trihalomethanes) were detected with concentrations above the laboratory's level of detection. These compounds are produced during water chlorination.

There was no indication that the water sources tested had been directly contaminated by oil or gas industry activity. Concentrations of metals in a few samples exceeded established water quality guidelines for community aesthetic values or drinking water standards for livestock.

## 17. Feeding Management and Forage Testing

Because of the possibility that nutritional management and forage quality could have had an effect on the health of calves in the study, herd feeding practices and forage quality were examined in spring 2002.

### Nutritional Data

Nutritional management data collected in spring 2002 indicate that 98% of the producers provided mineral and vitamin supplements to their cattle during the winter feeding period.

The supplements were provided in various formulations, including loose vitamin and mineral mixes, commercial liquid and solid formulations provided free choice, and pre-mixes formulated to be included in predetermined measured amounts in total mixed rations.

Before calving in 2002, 61% of the producers fed cereal grains or commercial concentrates to mature cows and 69% fed a concentrate to their heifer groups.

### Forages

In spring 2002, project veterinarians and herd owners collected 501 samples from the remaining winter supply of forage from 159 study herds. Samples were analysed for crude protein, acid detergent fibre, neutral detergent fibre, nitrates, calcium, phosphorus, magnesium, manganese, sulphur, zinc, copper, selenium, and molybdenum. Values for total digestible nutrient, digestible energy, net energy (metabolism, gain and lactation), and relative feed value were calculated based on laboratory results.

Results indicated that much of the forage would require supplementation to meet the requirements of an average beef cow in early lactation. For example, 95% of the forages were deficient in zinc, 93% in copper, 64% in phosphorus, and 60% in selenium. Some samples contained higher than recommended concentrations of molybdenum, sulphur or nitrates.

These results of feed analyses were expected, based on earlier studies in Western Canada. Feeding practices reported during herd visits, as indicated above, suggested that these deficiencies are recognized by producers, and that some type of supplementation is used in most herds.

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