

# Southern Alberta Resource Economics Centre

Department of Economics, University of Lethbridge  
4401 University Drive, Lethbridge, Alberta, Canada, T1K 3M4

**SAREC Report 2010-10**

**Non-point Agricultural Pollutants in Alberta**

**2010**

**Chelsea Matisz and Ruth Grant-Kalischuk**



## **Southern Alberta Resource Economics Centre Publications**

The mission of the Southern Alberta Resource Economics Centre (SAREC) is to study resource issues that affect growth and development of Southern Alberta. Multi-disciplinary research is required on the impacts of human activity on water quality, water quantity, climate change, impacts of biotechnology, growth of the bio-economy, manure disposal from intensive livestock production, and other agricultural and resource issues. These resource-based issues provide the motivation for socio-economic research in this dynamic region of Canada.

While SAREC is centred in the Department of Economics at the University of Lethbridge, SAREC is informally organized and includes researchers in several disciplines, including, but not limited to, economics, geography, management, and public health. The purpose of SAREC publications is to provide a forum for the exchange of information on all aspects of resource issues in Southern Alberta and related areas. The research reports published in this series may be completed research, works in progress or thought-provoking information pieces submitted by academic researchers, graduate students or others with an interest in resource issues in Southern Alberta. SAREC researchers have been supported financially by the Social Science and Humanities Research Centre, the Alberta Water Research Institute (formerly the Alberta Ingenuity Water Research Centre), the Canadian Water Network, SouthGrow Regional Initiative, and other funding agencies.

The opinions expressed in this series of research reports represent those of the authors and not of University of Lethbridge officials or of funding agencies. Prospective authors for this series are invited to submit completed manuscripts or to contact the research leaders regarding proposed topics.

K. K. Klein and Henning Bjornlund, SAREC Research Leaders

Department of Economics, University of Lethbridge

Lethbridge, Alberta T1K 3M4

([klein@uleth.ca](mailto:klein@uleth.ca); [henning.bjornlund@uleth.ca](mailto:henning.bjornlund@uleth.ca))

## **ABSTRACT**

This report aimed to identify the primary threats of agriculture to source waters in Southern Alberta. Many of the agricultural pollutants identified in the present report have been long known to pose threats to the health of both aquatic ecosystems and the public. The aquatic and public health consequences of nutrients, including nitrogen and phosphorus, pesticides, sediments, and fecal coliform bacteria in source waters are well described, and risk assessment and monitoring protocols have been developed accordingly. Other agricultural contaminants demand a closer examination of recent scientific research to adequately assess the threats to public health and ecosystems. Recent molecular advances will undoubtedly play an enormous role in assessing the threat that livestock wastes play in dispersing zoonotic strains of the infective stages of *Giardia* and *Cryptosporidium* into source waters. Emerging threats, such as those posed by endocrine disrupting compounds, warrant additional research, monitoring, and risk assessment protocols. Endocrine disrupting chemicals have the potential to contaminate source waters through livestock wastes, which contain some quantity of excreted estrogens. The intensive agriculture that exists in Southern Alberta, combined with extensive irrigation development, requires extreme vigilance and continued research of both well recognized, and emerging threats to source waters.

## Contents

1. Introduction .....	5
2. Nutrients .....	6
2.1. Nitrogen .....	6
2.1.1. Threats to Public Health and Wildlife .....	6
2.1.2. Risk of Nitrogen Water Contamination.....	8
2.1.3. Residual Soil Nitrogen (RSN) .....	9
2.1.4. Indicator of the Risk of Water Contamination by Nitrogen (IROWC-N) .....	10
2.1.5. Nitrogen Levels in Alberta.....	11
2.2. Phosphorus .....	11
2.2.1. Threats to Public Health and Wildlife .....	12
2.2.2. Contamination of Water by Phosphorus .....	13
2.3. Conclusions .....	13
3. Sediment Pollution.....	14
3.1. Sediments as Threats to Aquatic Ecosystems.....	14
3.2. Ecological Significance of Sedimentation.....	15
3.3. Conclusions .....	16
4. Pesticides .....	17
4.1. Threats to Public Health and Wildlife .....	18
4.2. Factors of Pesticide Toxicity.....	20
4.3. Environmental Distribution and Fate of Pesticides .....	21
4.4. Monitoring, Surveillance and Prevalence of Pesticides in Alberta .....	21
4.5. Conclusions .....	23
5. Bacterial Pathogens: Fecal Coliforms.....	24
5.1. Sources of Fecal Coliforms .....	25

5.2. Fecal Coliform Prevalence in Alberta .....	26
5.3. Conclusions .....	29
6. Protozoan Pathogens: Giardia and Cryptosporidium .....	30
6.1. Giardiasis and Giardia Genotypes .....	30
6.2. Cryptosporidiosis and Cryptosporidium Species and Genotypes .....	31
6.3. Agricultural Sources of Giardia and Cryptosporidium .....	32
6.4. Giardia in Livestock .....	34
6.4.1. Giardia in Cattle .....	34
6.4.2. Prevalence of Cattle with Zoonotic Giardia Assemblages .....	35
6.5. Prevalence and Distribution of <i>Cryptosporidium parvum</i> Among Humans.....	36
6.6. Cryptosporidium Prevalence in Livestock.....	37
6.6.1. Cryptosporidium Prevalence in Cattle .....	37
6.7. Giardia and Cryptosporidium in Raw, Treated, and Drinking Water .....	39
6.8. Giardia and Cryptosporidium in Raw and Treated Drinking Water in Alberta .....	40
6.9. Prevalence of Giardia and Cryptosporidium in Alberta .....	41
6.10. Conclusions .....	41
7. Endocrine Disrupting Compounds .....	43
7.1. Hormone Mimics.....	43
7.2. Natural & Synthetic Estrogens: Pharmaceuticals .....	44
7.3. Agricultural Pharmaceuticals .....	45
7.4. Binding Affinities and Potency .....	46
7.5. Estrogenic Effects on Humans .....	47
7.6. Estrogenic Effects on Wildlife .....	48
7.7. Estrogens and Agriculture.....	49
7.7.1. Manure Application as a Source of Estrogen.....	50

7.7.2. Estrogens in Soils.....	51
7.7.3. Estrogens in Agricultural Runoff .....	52
7.7.4. Androgen and Progesterone in Agriculture .....	54
7.8. Municipal Sources of Estrogenic Compounds.....	56
7.9. Conclusions .....	57
8. Emerging threats to Alberta source waters .....	58
8.1. Methylmercury .....	58
8.2. Oil Sands Development.....	59
8.3. Fire Retardant Additives: PBDEs .....	61
9. General Conclusions.....	62
References .....	66

# 1. Introduction

Agricultural non-point sources of pollution are recognized as a leading threat to water quality. The dominance of the agricultural industry in Alberta and the density of confined livestock operations in parts of southern Alberta, in concert with extensive irrigation networks, indicates an enormous potential for non-point sources of agricultural pollution to contaminate source waters. A variety of agricultural activities, including feedlot operations, grazing, tilling, pesticide application, irrigation, and fertilizing, have the potential to impact water quality. Understanding the nature of specific threats to source water is a fundamental component to Alberta's Water for Life Strategy, as it is integral to establishing safe, secure drinking water supplies, promoting and protecting healthy aquatic ecosystems, and ensuring reliable water quantities to sustain Alberta's agricultural industry. The presence of excess nutrients, pesticides and waterborne pathogens represent some of the dominant forms of agricultural-based water pollution, though emerging threats, such as endocrine disrupting compounds, are receiving increasing amounts of attention. The objectives of this report are to: 1) identify the primary agricultural pollutants in Southern Alberta's source waters, 2) describe how these pollutants impact environmental and public health, 3) describe how these pollutants infiltrate/access source water, 4) describe the monitoring and surveillance techniques employed, and 5) assess the prevalence of identified agricultural pollutants in Alberta. This literature review will focus on nutrients, pesticides, pathogens, sediments, and endocrine-disrupting compound pollutants, and their non-point sources. The threats posed by organic mercury, oil sands development, and fire retardant additives to source waters are also briefly addressed.

## **2. Nutrients**

### **2.1. Nitrogen**

Nutrient loading, a common practice within the agricultural industry, poses a threat to water quality, with nitrogen and phosphorus being of primary concern (McRae et al., 2000). Nitrogen is an essential crop nutrient, and exists naturally in soils through decomposition of organic matter, and nitrogen-fixing legumes. Nitrogen is readily oxidized by bacteria in the soil to nitrate ( $\text{NO}_3^-$ ), a water-soluble form of the nutrient that is accessible for uptake by plants. Nitrogen-laden fertilizers and manures are applied to agricultural plots to promote high crop yields. Through inefficient fertilizer and manure application practices, or environmental factors that result in less-than expected nitrogen uptake by crops, excess nitrates accumulate in the soils. Nitrates are very mobile, and thus are capable of leaching into groundwater below the root zone. Nitrate is the most abundant pollutant of aquifers, with crop and livestock practices representing the dominant anthropogenic source of contamination (Burkart & Stoner, 2001). Once in groundwater, nitrates are discharged into surface waters, contributing to water pollution.

#### **2.1.1. Threats to Public Health and Wildlife**

An excess of nitrates in water poses a significant threat to both the health of aquatic ecosystems, and humans. Excess nitrogen in surface waters increases the number of photosynthetic organisms within an ecosystem, a process called eutrophication. Altering the community structure in aquatic ecosystems can have dramatic ramifications, such as declines in aquatic biodiversity, changes in species composition, and increased toxicity to aquatic organisms. The increased quantity in aquatic plant life in turn depletes oxygen supplies, and reduces water flow in streams and canals. While

nitrate themselves are non-toxic to humans, they are reduced to highly reactive nitrites ( $\text{NO}^{2-}$ ) by bacteria in the digestive tract (Camargo & Alonso, 2006). Nitrites are known potent mammalian carcinogens, and have been implicated in stomach, liver, bladder, and ovarian cancers (Camargo & Alonso, 2006). Nitrites reduce the capacity of hemoglobin to carry oxygen, resulting in a condition called methemoglobinemia (Wolfe & Patz, 2002; Camargo & Alonso, 2006). Infants are especially prone to this disease, with potentially fatal consequences (Greer & Shannon, 2005). However, the role that nitrate-polluted drinking water plays in the development of this condition is unknown, and disputed (Fewtrell, 2004).

The eutrophication caused by nitrogen inputs poses an additional concern to public health. Treatment of source waters with chlorine or bromine, in concert with an overabundance of organic material in the source waters, results in the formation trihalomethanes as a by-product. These compounds possess a public health risk, as trihalomethanes are believed to be carcinogenic in humans (Chambers et al., 2001). Given that the majority of water treatment facilities in Canada rely on the disinfectant properties of chlorine, the Canadian Drinking Water Quality Guidelines have included trihalomethanes as a parameter (Health Canada, 2009).

Nitrogen can be converted into several gaseous forms, including nitrous oxide ( $\text{N}_2\text{O}$ ), that are found in the atmosphere. Thus, inefficient nutrient management and environmental factors which indirectly result in excess nitrogen in the soils can lead to an increase in these nitrogenous gases. Nitrous oxide is the 4<sup>th</sup> greatest contributing gas to the greenhouse effect, with a warming potential 200 times that of carbon dioxide (Wolf & Patz, 2002). Ultimately, climate change threatens the availability and reliability of safe clean water sources.

### **2.1.2. Risk of Nitrogen Water Contamination**

Many Canadian rural wells are at high risk of nitrate contamination, with 20-40% bearing nitrate concentrations exceeding those recommended by Canadian Water Quality Guidelines (van der Kamp & Grove, 2001). As 60% of rural wells possessed unsafe nitrate levels in areas of crops with high nutrient requirements, or animal farming operations, Alberta's rural well water quality is suspect (van der Kamp & Grove, 2001). The leaching of nitrates into groundwater occurs via over-application of manure (Komor & Anderson, 1993; Wassenaar, 1995; Chang & Entz, 1996), commercial fertilizers (Exner & Spalding, 1994; Herbel & Spalding 1993), storage lagoons (Arnold & Meister, 1999), and from feedlot pens (Coote & Hore, 1979).

As shallow aquifers are commonly recharged beneath agricultural landscapes, they are especially susceptible to nitrate contamination (Burkart & Stoner, 2001). Based on studies conducted in the United States, unconsolidated aquifers, followed by alluvial and carbonate aquifers, have the highest levels of nitrate contamination (Burkart & Stoner, 2001). The denitrification processes that are capable of reducing nitrate levels in groundwaters below agricultural landscapes can take decades, even in concert with environmental restrictions (Burkart & Stoner, 2001). As shallow aquifers represent the primary source of drinking water for many rural areas in Alberta, nitrate contamination is a definite concern for rural communities (Alberta Agriculture and Rural Development: AARD, 2009b). Once contaminated groundwater discharges into surface waters, contamination spreads throughout the watershed. A study by Rodvang et al. reported an increase in both nitrate and chloride concentrations in the unconsolidated aquifers underlying manure-applied and commercially-fertilized fields in the Lethbridge North Irrigation District between 1995 and 2001 (2004). Nitrate and chloride concentrations in shallow aquifers underlying

pasture or native ranges in the Lethbridge Northern Irrigation District were significantly lower by comparison, and did not change over time (Rodvang et al., 2004). Models estimate that shallow water discharges of nitrate and chloride into the Oldman River could increase by 4.3 and 1.3 times, respectively (Rodvang et al., 2004). Irrigated cropland augments the risk of nitrogen contamination of waters, as runoff can access irrigation canals and contaminate streams and rivers via return flow channels. With 65% of irrigated land (Statistics Canada, 2009) and 40% of the cattle population in Canada located in Alberta (AARD, 2009a), nitrate contamination of surface and groundwater is a significant concern to source water quality in Alberta.

### **2.1.3. Residual Soil Nitrogen (RSN)**

Excessive nitrogen in soils provides the means for contamination of water sources. Indicators have been developed in Canada to assess and report the level of nitrogen present in soils, and the risk of nitrogen movement into water supplies. Residual soil nitrogen (RSN) reflects the annual amount of nitrogen present in the soil profile after harvest. It is calculated as the disparity between total nitrogen inputs from chemical fertilizers, manure application, nitrogen fixation and crop residues, minus the amount of nitrogen present in harvested crops (Yang et al., 2007). While there are many approximations and assumptions in RSN calculations (see Yang et al., 2007 for details), the results are useful for identifying areas where risk of nitrate contamination of water sources is high, and where inefficient nutrient application practices may occur.

Studies conducted by De Jong et al. (2005) document the temporal changes of Alberta's residual nitrogen in the soils from 1981 to 2001. While dry climates like Alberta's possess a reduced risk of soil nitrates infiltrating ground and surface water supplies, the observed general trend of increasing RSN may pose a risk to human health via

degradation of drinking water quality (Chambers et al., 2001). The proportion of farmland with very low and low amounts of RSN decreased from 84% to 56% between 1996 and 2001, concomitant with an 18% increase in the proportion of farmland with moderate levels of RSN; there was an additional 11% increase in the proportion of farmland with high, and very high RSN levels (De Jong et al., 2005).

In Canada overall, 63% of RSN could be explained by increased nitrogen inputs, via increased farming of nitrogen-fixing legumes, and decreased farming of wheat, barley, and cereals. The remaining RSN was explained by a reduced nitrogen output due to lower-than-expected crop yields (De Jong et al., 2005). Over the last 20 years Alberta has seen an increase in the sale of fertilizers, and cattle and pig manures; in combination with declines in crop yields, this is believed to account for an increased proportion of moderate levels of RSN in Alberta's farmlands (De Jong et al., 2005).

#### **2.1.4. Indicator of the Risk of Water Contamination by Nitrogen (IROWC-N)**

Under certain climactic conditions, such as spring thaw, or storm events, residual soil nitrogen is transferred from agricultural lands to surface waters. The Indicator of the Risk of Water Contamination by Nitrogen (IROWC-N) evaluates the risk of residual soil nitrogen infiltrating ground and surface waters. The indicator links the residual soil nitrogen at harvest with the following winter climactic conditions, and places regions in 5 IROWC-N classes. The indicator has evolved to include a more in-depth analysis of surface runoff, soil-water balances, crop cover, and evapotranspiration, among other things (DeJong et al., 2009). Over the last 10 years, Alberta has seen a decline in the proportion of farmlands in the very low and low IROWC-N classes, from 96% to 87%, concomitant to a 9% increase in the proportion of farmlands in the moderate IROWC-N class (De Jong et al., 2005).

### **2.1.5. Nitrogen Levels in Alberta**

A study conducted by the Canada-Alberta Environmentally Sustainable Agriculture Agreement (CAESA) indicated that nitrogen levels sometimes exceed human drinking water guidelines, and often exceed guidelines recommended for healthy aquatic life (1998). Of the 376 shallow wells examined across Alberta, 13% exceeded the recommended human drinking water guidelines; only 0.6% of 448 deep wells exceeded recommended levels of nitrate-nitrite (CAESA 1998). Although it was believed that poor well design and maintenance were the major cause of contamination, shallow aquifers (which feed shallow wells) are considered the most susceptible to contamination by agricultural runoff (Burkart and Stoner, 2001). The 1998 study also found a relationship between total nitrogen levels in streams, and the level of agricultural intensity surrounding streams (CAESA). Not surprisingly, samples of streams in areas of high intensity most frequently exceeded guidelines for aquatic life (87%); 65% and 32% of samples exceeded these guidelines in areas of moderate and low agricultural intensity, respectively.

## **2.2. Phosphorus**

As with nitrogen, fertilizers and manures are applied to farmland to provide crops with phosphorus, an essential crop nutrient. Phosphorus can dissolve in water and enter ground and surface waters via runoff from agricultural lands, or it can attach to fine soil particles and access waters via soil erosion. For more information on the ecological consequences of sediment-bound contaminants, see section 3. The primary concern of excess phosphorus in surface water is eutrophication; this nutrient is limiting in aquatic ecosystems, and excessive amounts cause increased algae and plant growth. This drastically disrupts the delicate equilibrium in aquatic environments, as increased plant growth leads to oxygen depletion, changes in water pH, and a reduction in aquatic plant

and animal diversity. In turn, water flows in canals can be reduced by the influx of plant growth (Correll, 1998).

The application of manures to meet nitrogen requirements often results in the excess application of phosphorus, resulting in phosphorus loading in the soils (Wortmann & Walters, 2006). Risks of phosphorus losses are dependent upon runoff volume, and sediment concentration in runoff (Wortmann & Walters, 2006). Overall, the concentration of phosphorus in runoff is linearly correlated with soil test levels of phosphorus (Sauer et al., 2000; McDowell & Sharpley, 2001; Andraski & Bundy, 2003; Daverede et al., 2003). However, even when soil test phosphorus levels are moderate, high risks of phosphorus transport can result in phosphorus losses from soils (Wortmann & Walters, 2006). Thus, those factors that increase the risk of phosphorus transport may be more important in assessing risks of phosphorus loading than levels of soil phosphorus tests.

### **2.2.1. Threats to Public Health and Wildlife**

Increases in phosphorus levels usually exceed increases in nitrogen. Cyanobacteria are capable of nitrogen fixation; thus low nitrogen: phosphorus ratios produce an environment conducive to cyanobacterial blooms (Schindler & Donahue, 2006). Toxins produced by these organisms make their population explosions a severe risk to public health (Bell & Codd, 1994). Human exposure to these toxins can cause skin irritation, stomach cramps, vomiting, nausea, diarrhea, fever, sore throat, headache, muscle and joint pain, blisters of the mouth and liver damage (Bell & Codd, 1994).

### **2.2.2. Contamination of Water by Phosphorus**

The amount of naturally occurring phosphorus which reaches surface waters is unknown, making estimates of phosphorus runoff from agriculturally treated lands difficult to assess. An indicator for the risk of Water Contamination by Phosphorus (IROWC-P) has been developed, and levels have been assessed for Quebec (Bochove et al., 2006). However, the applicability of current IROWC-P models for use in the prairie provinces has been challenged, as phosphorus contamination in these regions is driven more by snowmelt runoff on flat landscapes, rather than rainfall-induced soil erosion on sloped landscapes (Salvano et al., 2009). Further, the standard preliminary phosphorus indicators were not correlated with average phosphorus concentrations in the water, or levels of phosphorus exported from agricultural lands. Thus models that incorporate climactic and regional differences are necessary to assess phosphorus-loading risks. Salvano *et al.* (2009) did determine that soil test phosphorus concentrations, livestock density, and crop cover were some features that are strongly correlated with phosphorus concentrations in the waters. Total phosphorus levels in lakes in areas of Alberta under high agricultural activity exceeded maximum accepted concentrations (MAC) for the protection of aquatic life in 96% of the samples taken. In areas of moderate and low agricultural intensity, 65% and 38% (respectively) of the samples exceeded MAC for total phosphorus levels (CAESA, 1998).

### **2.3. Conclusions**

It is well established that nutrient loading has the potential to significantly impact source waters. There are public health risks associated with nitrite toxicity, cyanobacterial blooms, and trihalomethanes, and risks to aquatic ecosystems through eutrophication. Still, the challenges surrounding nutrient loading will only become exacerbated by climate

change; warmer waters and longer ice-free periods contribute to increased growth of aquatic organisms that choke out native aquatic plants, altering the food webs, and contributing to ecosystem decline. (Schindler & Donahue, 2006). An assessment of nutrient loading into Canadian surface and groundwaters revealed that in 1996, agricultural activities contributed by far the largest source of nitrogen (293 thousand tones) and phosphorus (55 thousand tones) into the environment (Chambers et al., 2001). While effective nutrient management is integral to protecting source waters, it should be complemented by science-based solutions and the implementation of new technologies. Chambers et al. (2001) outline multiple approaches to reduce anthropogenic loading of nutrients, including the development of watershed management plans that assess current levels of eutrophication, and establishing nutrient guidelines specific to rivers, lakes, and streams. Improved nutrient monitoring plans, expanded research on environmental indicators and technologies used to recover and recycle nutrients, increased research on management practices, and increase public education are also believed to be integral in promoting source water protection and environmental sustainability (Chambers et al., 2001).

### **3. Sediment Pollution**

#### **3.1. Sediments as Threats to Aquatic Ecosystems**

Sediments are a vital component in aquatic ecosystems. Sediments provide substrate for a variety of organisms, and play an integral role in nutrient cycling in aquatic ecosystems (Forstner & Owens, 2007). Both sediment quantity, and quality have the capacity to significantly influence water quality. Sedimentation is the movement and

deposition of soil particles into bodies of water, occurring primarily via soil erosion. While sedimentation is a naturally occurring process, land-use practices such as agriculture have greatly impacted rates of sedimentation. Sediments can function as both a pollutant, and a carrier of pollutants (Robbins, 1979). Thus, excessive sedimentation poses a risk to aquatic environments. Sedimentation can result in the suspension of sediment particles in bodies of water. As a result, sunlight penetration through the water column is reduced, which can significantly alter the production dynamics of aquatic ecosystems. High levels of sedimentation also can impact spawning grounds of fish, and impact food supplies of both aquatic vertebrates and invertebrates. Turbid water also has the capacity to impair water treatment processes, and represents a public health concern (Cheremisinoff, 2002).

A wide variety of contaminants can adhere to sediment particles through the process of adsorption, compromising sediment quality. Sedimentation processes can result in loading of pesticides (Akerblom, 2007), estrogens (Colucci & Topp, 2001), phosphorus (House et al., 1998), and a wide variety of heavy metals (Bryan & Langston, 1992), as these contaminants can have an affinity for sediment particles. Thus the diversity of sediments sources, which contribute a variety of contaminated sediments that accumulate in river basins, can produce a convoluted assortment of pollutants in a given river basin (Forstner & Owens, 2007).

### **3.2. Ecological Significance of Sedimentation**

The ecological significance of potentially toxic sediments is huge. Through leeching and resuspension processes, toxic components can become available to biological systems (bioavailable) (Giesy et al., 1990), and thus contaminated sediment beds can serve as long-term sources of pollutants to aquatic ecosystems (Larsson, 1985;

Salomons et al., 1987; Loring & Rantala, 1992). As benthic (sediment-dwelling) communities directly interact with sediment profiles, these organisms are particularly vulnerable to the bioavailable fraction of toxic sediments. Benthic communities play a vital role in the cycling of energy and nutrients (Forstner & Owens, 2007). Thus, through potential chronic and acute toxic effects, and bioaccumulation processes, toxic sediment profiles have the capacity to cause dramatic impacts on higher trophic levels, and shifts in community composition and function (Akerblom, 2007.).

A study conducted by Gannon et al. (2005) suggested that reduced concentrations of bacteria in certain water samples obtained from the Lethbridge Northern Irrigation District was due to bacterial sedimentation in reservoirs. Thus, reservoirs, like constructed wetlands, have the capacity to remove microbial pollutants through sedimentation, biological, physical, and chemical processes (Gearheart, 1999; Goulet et al., 2001; Shutes, 2001; Rumes et al., 2003). However, this presents a public health concern; bacterial concentrations in sediments have been reported to be up to 1000x higher than concentrations reported in the overlying water columns (Karim et al., 2004). Additionally, bacteria display a prolonged survival rate in sediments (Karim et al., 2004). Environmental events, such as spring flows, or heavy rainfall, could promote sediment suspension, causing the resuspension of bacteria in the water, and thus creating a public health risk (Craig et al., 2002; Alm et al., 2003).

### **3.3. Conclusions**

Water samples are not the sole indicator of the health of aquatic systems, as levels of contaminants in open waters are a poor proxy of the bioavailability of sediment-bound toxins. For example, studies have demonstrated that the concentration of heavy metals in

sediments is between 3-5 orders of magnitude greater than concentrations in the overlying waters (Bryan & Langston, 1992). Further, some metals such as lead and mercury, can undergo transformative processes in sediments that result in increased bioavailability and toxicity to aquatic biota (Bryan & Langston, 1992). As described previously, the concentration of bacteria is also several orders of magnitude higher in sediments, than in the open, overlying waters (Gannon et al., 2005). Thus assessing sediment quality, in addition to the benthic communities that exist on the front lines of sediment contamination, is an integral part of environmental risk assessment. A report published by Alberta Environment in 2007 highlighted the extreme lack of knowledge related to sediment quality in Alberta's aquatic ecosystems. Additionally, biological criteria, used to assess the condition of non-fish biota, are not established for Alberta (AENV, 2007). A lack of appropriate ecological indicators, and more broadly, the lack of data on sediment quality, is a significant obstacle in assessing the health of Alberta's aquatic ecosystems.

#### **4. Pesticides**

Pesticide is an umbrella term that denotes all chemicals used to control pests. These include insecticides, herbicides, fungicides, and rodenticides. The risk of pesticide use is that these chemicals will adversely affect non-target species. The ultimate sink for many of these chemicals is the aquatic environment, due to point source discharges, and non-point hydrologic and atmospheric sources (Stegeman & Hahn, 1994). Thus, it is not surprising that given the wide use of pesticides in the agriculture industry in Alberta, these chemicals represent a threat to source waters. Over 60 pesticides are licensed for use in Alberta (AENV, 2005).

#### **4.1. Threats to Public Health and Wildlife**

The neurotoxic and carcinogenic effects of pesticides have been well studied, and represent the primary public health concern of human exposure to pesticides (Alavanja et al., 2004). Pesticide exposure has been linked to the development of leukemia, non-Hodgkin's lymphoma, tumors, skin and neurological diseases, and various reproductive defects (Ontario College of Physicians and Surgeons, 2004). Endocrine disrupting abilities have been implicated for a wide variety of herbicides (2,4-D, 2,4,5-T, alachor, amitrole, atrazine, metribuzin, nitrofen, trifluralin), fungicides (benomyl, mancozeb maneb, tributyltin, zineb, and ziram), and insecticides (aldicarb, carbaryl, chlordane, dicofol, dieldrin, DDT, endosulfan, heptachlor, lindane, methomyl, arathion, synthetic pyrethroids, and toxaphene) (Matthews, 2006). The herbicide atrazine, for example, is widely used across Alberta (Colburn et al., 1993). For more a more detailed discussion on the public health risks of endocrine disrupting compounds, see section 7.

Pesticides can produce lethal, and a wide variety of sublethal effects on aquatic biota, including impaired behavioural, physiological, reproductive function through the disruption of enzyme and hormone processes (Khan & Law, 2005). Additionally, through the processes of bioconcentration and biomagnification, pesticides have the capacity to affect organisms at numerous trophic levels within an aquatic ecosystem. Thus, the effects of pesticides do not necessarily stop at those organisms to which pesticides are directly exposed. Bioaccumulation is the process by which the concentrations of pollutants accumulate within an organism at the base of the food chain; biomagnification is the increase in pollution concentration as it moves up the food chain through predator interactions. Pesticide compounds frequently accumulate in the fatty tissues of animals, posing health risks for both aquatic and terrestrial animals.

Generally, a large period of time is required for the negative effects caused by various pollutants to be manifested by populations of aquatic biota; by the time observable effects are noticed, the process may be irreversible or unstoppable (van der Oost et al., 2003). Research related to assessing the effects of pollutants on the health of non-target species relies heavily on the use of biomarkers. Biomarkers are sensitive indicators that examine changes in a biological state due to toxin exposure, and can thus serve as early warning signals of potential negative effects of toxins on individual organisms (van der Oost et al., 2003). For example, pesticides are known to inhibit the activity of acetylcholinesterase (AChE), and decrease the activity of this neurotransmitter enzyme (Sturm et al., 1999). Numerous studies have been conducted to ascertain the toxic effects of pesticides using AChE as a biomarker (Hansson et al., 2006; Li et al., 2008; Linde-Arias et al., 2008; Miller et al., 2009)

It is vitally important to consider the conditions under which aquatic organisms will be exposed to pesticides. Simple lab-based limits of toxicity are inadequate indicators of the potential effects of pesticide exposure. This is because while levels of identified pesticides may be well below recommended surface water quality guidelines, these levels do not take into account the potentially additive and synergistic effects that numerous low, environmentally relevant concentrations of pesticides might have on the aquatic biota (AENV, 2001). Pesticide mixtures have been demonstrated to frequently occur in streams, lake, and rivers (Gillion, 2007). Research has shown that mixtures of commonly employed pesticides produce additive and synergistic deleterious effects on both fish and aquatic invertebrates (Bailey et al., 1997; Pape-Lindstrom & Lydy, 1997; Belden & Lydy, 2000; Anderson et al., 2002; Laetz et al., 2009). Recently, evidence has demonstrated that atrazine, a herbicide widely used in Alberta, invokes both chronic and acute effects of other environmental pollutants (Hayes et al., 2002; Schuler et al., 2005). Currently, risk

assessment of levels of pesticides in source waters and aquatic environments does not take into account potential additive or synergistic effects.

## **4.2. Factors of Pesticide Toxicity**

The severities of toxic effects, which can be either acute or chronic, are a function of both toxicity and exposure time. The toxicity of pesticides are frequently denoted as  $LD_{50}$ , which translates to the concentration of pesticide required to produce a lethal dose (LD) in half (50%) of the population of test organisms, due to exposure over a certain period of time. Thus, the lower the  $LD_{50}$ , the greater the pesticide toxicity is. To assess the sub-lethal effects of pesticides, an  $EC_{50}$  index is employed;  $EC_{50}$  indicates the concentration of pesticide required for 50% of test organisms to display a specific response. Typically, the measured response is a particular behaviour, though other endpoints may be used. Pesticides also vary in their persistence in the environment, which is measured as half-life, or the time required for the concentration of pesticide to be decreased by 50%. A variety of biotic (ex. biodegradation and metabolism) and abiotic factors (ex. hydrolysis, oxidation) affect pesticide persistence (Calamari & Barg, 1993). In the process of degradation, through biotic or abiotic mechanisms, pesticide metabolites may be formed. These transformed pesticide products may possess similar, greater, or lesser toxicity than the original compound (Akerblom, 2004). The environmental fate and toxicity of pesticides and their transformed products should be considered, although few studies have been conducted on the latter in this regard (Akerblom, 2004). Pesticide affinity for solids, liquids, and gases can additionally impact both toxicity and persistence.

### **4.3. Environmental Distribution and Fate of Pesticides**

The environmental fate of pesticides, whether they are dissolved in river water, bio-concentrated in the tissues of living organisms, or accumulated in sediments, is largely dependant on the molecular characteristics of the specific pesticide. The degree to which pesticides bind to substances, such as lipids or soils, is measured by “k”, a binding coefficient. Studies have determined that the organic component in soils is the primary adsorbent of pesticides to sediments; thus  $K_{oc}$  represents the binding affinity of a pesticide to soils (Spark & Swift, 2002). Pesticides with a  $K_{oc} > 50$  are weakly adsorbed by soils, and thus have the potential to access groundwater through leaching, or surface waters via runoff (Comfort et al., 1996). Those pesticides with a  $K_{oc}$  between 50 and 5000 are moderately adsorbed by soils, and possess a high potential for runoff, while pesticides of a  $K_{oc}$  greater than 5000 are likely to access surface waters via soil erosion (Comfort et al., 1996). The  $K_{ow}$  is an index used to assess the affinity for lipids, thus determining the ability of a substance to pass through biological membranes, and bioaccumulate in organisms. Additional characteristics of pesticides include the water solubility coefficient (S), which expresses how likely pesticides will be dissolved by water, and their volatility, which is measured by vapour pressure. Volatile substances are more likely to be incorporated into the atmosphere, where they can be carried for great distances. For more information regarding the ecological implications of sediment-pesticide interactions, see sediments in section 3.

### **4.4. Monitoring, Surveillance and Prevalence of Pesticides in Alberta**

For the pesticide component of the Alberta River Water Quality Index (ARWQI), water samples are taken 4 times during the open-water season and tested for 17 pesticide variables (2,4-D, MCP, MCPA, Diazinon, Lindane, Picloram, Dicamba, Triallate,

Altrazine, Bromoxynel, Cyanazine, Malathion, ethoxychlor, Chlorpyrifos, Imazamethabenz, Diuron, Dichlorprop) (AENV). Based on the presence and occurrence of these variables, monitoring sites are indexed as excellent, good, fair, marginal, or poor. Several studies have provided more detailed information regarding the current state of Alberta's water quality in regards to pesticide contamination. A study conducted in 2005 for Alberta Environment determined that pesticides are generally widespread in Alberta's surface waters. Additionally, research indicates that levels of pesticide contamination of Alberta's surface waters are positively correlated to agricultural intensity (Anderson et al., 1997). Further, the frequency, concentration, and likelihood of non-compliance with surface water quality guidelines was greater where pesticide use was higher (Anderson et al., 1997). Overall, pesticide concentrations exceeded water quality guidelines in 30% of water samples (Anderson, 2005). The most frequently detected pesticide was 2,4-D, which is believed to possess endocrine disrupting abilities. Irrigation return flow channels displayed high frequency, number, and concentration of pesticides, with peaks occurring in June and July. These peaks coincided with seasonal hydrology (storm runoff, snow melt), and agricultural activities (pesticide application) (Anderson, 2005). Over half of the pesticides detected in this reports are not mentioned in the surface water quality guidelines.

In 2001, AENV conducted a laboratory-based study to evaluate the effects that mixtures composed of the most frequently detected pesticides in Alberta have on a micro-crustacean, *Ceriodaphnia*. Results indicated that various herbicide mixtures at environmentally relevant levels did not significantly alter survival or reproduction of the crustacean (AENV, 2001). However, the authors do suggest that water quality conditions in the field may play a role in the toxicity of herbicide mixtures. Clearly, field studies are needed in this regard. It would have been interesting to assess changes in specific

biomarkers of fish, in addition to using survival and reproduction of micro-crustaceans as endpoints. Another criticism was that the study did not test the toxicity of mixtures of both herbicides and insecticides, which possess unique modes of action (Aberklom, 2004); therefore, these mixtures may possess particularly potent effects. Previous studies have indeed demonstrated the synergistic effects of herbicide and insecticide mixtures (Trimble & Lydy, 2006). Generally, it is clear that an effective risk assessment of the surface water quality guidelines cannot be achieved given the current limitations of surface water quality guidelines, which are non-existent for many frequently detected pesticides in Alberta's surface waters, and do not account for synergistic effects of pesticides (Anderson, 2005)

In 2008, a proposal was published to develop a pesticide toxicity index (PTI), which would account for the additive effects of pesticide mixtures (Anderson). Briefly, the PTI would serve as a tool that would rank and compare the toxicity of various water river samples over time, thus identifying those pesticides of greatest concern. To assess its potential usefulness, Anderson applied the pesticide toxicity index to data compiled by the long-term river network (2008). Results indicate that the Alberta PTI is a useful complement to the Alberta River Water Quality Index, as it provides valuable information on the potential toxicity of pesticide mixtures. Additionally the PTI exhibits potential as a screening tool that can assess risks of specific pesticides. While this index does not account for synergistic effects of pesticide mixtures, it certainly contributes to a more holistic assessment of pesticide risk.

#### **4.5. Conclusions**

Pesticides are unique from other agricultural pollutants, in that because they are manufactured, there are legislative measures that are used to control those pesticides

licensed for use in Canada, and in turn control which pesticides are likely to be impacting Alberta's source waters. Through the Federal Canada Pest Control Act, the Pest Management Regulatory Agency regulates pesticides used in Canada. Scientific examination of the toxicity, environmental fate, potential health and environmental hazards of pesticides are required; only pesticides registered for use by the Canadian government may be used in Alberta. Overall, there appears to be a strong scientific base of the risks associated with exposure and effects of pesticides in general. Although the present monitoring techniques do not yet include assessing the risks of additive and synergistic effects of pesticide mixtures, it would appear that attempts are being made to modify and enhance monitoring practices to address these issues. Although pesticides are commonly detected in Alberta's source waters, concentrations are generally below recommended guidelines. However, it is obvious that the parameters outlined in Alberta's surface water quality guidelines are insufficient, given the number of pesticides detected in source waters that were not mentioned in these guidelines.

## **5. Bacterial Pathogens: Fecal Coliforms**

Fecal coliforms represent a sub-group of coliform bacteria that include a variety of bacteria that are significant to human health. Fecal coliform bacteria are a widely used indicator of fecal contamination in water supplies, although some species of fecal coliform bacteria exist naturally in the soils and on vegetation. Thus, the presence of *E. coli*, a fecal coliform, is used as a definitive indicator of fecal contamination from warm-blooded animals (CAESA, 1998). There are a wide variety of strains or subgroups of *E. coli*. While most strains do not pose a threat to human health, some serotypes are capable of producing highly pathogenic toxins that can cause severe illness or death in humans.

Serotypes of enteric *E. coli* can be grouped together according to similar virulence and serological characteristics. The serotype *E. coli* O157:H7 is of particular significance to public health, as this common Enterohemorrhagic strain can cause severe illness or death in humans (Hudault et al., 2001).

## 5.1. Sources of Fecal Coliforms

Livestock manure is recognized as a source of fecal coliform bacteria, including *E. coli* (Johnson et al., 2003). According to Statistics Canada, livestock produce about 10% of total fecal coliform population in Canada; approximately 71% of these fecal coliforms are produced by beef cattle (1996). Dairy cows and hogs contributed to 10% of livestock fecal coliform production, with remaining contributions from calves (7%), poultry (1%), sheep (1%), and horses (<1%) (Statistics Canada, 1996). Alberta houses nearly 40% of Canada's cattle population (AARD, 2009a), so it is unsurprising that Alberta has the greatest number of sub-basins with highest levels of fecal coliform bacteria in Canada (Statistics Canada, 1996)

Cattle are known reservoirs of *E. coli* O157:H7 (Dean-Nystrom et al., 1999) and high cattle density was found to be an effective predictor of the incidence of *E. coli* infections in humans (Johnson 1999; Michel et al., 1999; Valcour et al., 2002). The source of *E. coli* infections in the outbreak in Walkerton, ON, in 2000, which resulted in 5 deaths and the illness of thousands, was attributed to cattle manure (Kondro et al., 2000; Hruday et al., 2003). However, studies have failed to find a direct correlation between confined feedlot operations, and the prevalence of fecal coliform bacteria (Johnson et al., 2003). Nonetheless, Alberta reports the second highest incidence of *E. coli* isolates in Canada (8.8 per 100,000 people). (Public Health Agency of Canada, 2007).

## **5.2. Fecal Coliform Prevalence in Alberta**

Research indicates that levels of fecal coliform bacteria often exceed maximum acceptable concentrations (MAC) for human consumption in surface, ground, and irrigation waters in Alberta, as outlined by the Canadian Water Quality Guidelines. Agricultural practices are the most likely source of fecal contamination of these water supplies. A comprehensive study conducted by Alberta Agriculture Food and Rural Development evaluated fecal coliform levels in 27 streams, at locations with varying levels of agricultural intensity, and hundreds of shallow and deep wells across Alberta (CAESA, 1998). Additionally a pilot study was included in the report, which examined fecal coliform levels in dugouts around Lethbridge, AB. All streams exceeded maximum acceptable concentrations for human consumption, regardless of agricultural intensity of the land surrounding the sample stream (CAESA, 1998).

Interestingly, the level of fecal coliform contamination was greatest in areas of moderate agricultural intensity, with 100% of all samples acquired from streams exceeding human consumption guidelines (CAESA, 1998). These results suggest that moderate-intensity agricultural operations may benefit from increased education and technical training regarding best management practices. In areas of high and low agricultural intensity, 94% and 90% (respectively) of samples acquired from streams exceeded maximum acceptable concentrations for human drinking water. Fecal coliform levels in these samples were high enough to exceed irrigation guidelines in 25% of the samples acquired in areas of high agricultural intensity, 68% of the samples acquired in areas of moderate intensity, and 16% of the samples acquired in areas of low agricultural intensity. Fecal coliform levels obtained from samples in streams exceeded recreation guidelines in

areas of high, moderate, and low agricultural intensity in 9%, 44%, and 6% of samples, respectively (CAESA, 1998).

Shallow wells were more vulnerable to fecal coliform contamination than deep wells, with 5% of 376 tested shallow wells exceeding MAC for human drinking levels, while only 2% (11/448) of deep wells exceeded guidelines. Generally wells are not ideal locales for fecal coliform bacteria, as they are devoid of the nutrients and organic processes these organisms require to survive. Thus, the presence of fecal coliforms in shallow and deep wells could indicate septic contamination, damaged well seals, vermin entry into the well, or livestock too close to the well head (CAESA, 1998).

Irrigation canals in Alberta were in frequent violation of water quality guidelines. Samples obtained from both supply flows (96%) and return flows (95%) exceeded levels acceptable for human consumption. Additionally, 14% of samples obtained from supply sources and 33% of samples obtained from return flows exceeded levels of fecal coliform bacteria deemed acceptable for irrigation use. Fecal coliform and *E. coli* concentrations exceeded maximum acceptable concentrations for recreation in 18% and 25% of samples, respectively, at return flow channels (CAESA, 1998).

High concentrations of fecal coliforms and *E. coli* in Alberta watersheds are further demonstrated by peer-reviewed studies. Hyland *et al.* evaluated fecal coliform and *E. coli* concentrations in the Oldman River Basin, between 1998 and 2000 (2003). Waters bearing highest fecal coliform and *E. coli* counts were downstream of intensive agricultural operations, with levels peaking during the summer months (Hyland *et al.*, 2003). The trend of peaking fecal coliform counts in the spring and summer months is usually indicative of agricultural sources, and is believed to be directly related to increased livestock

distribution, increased livestock numbers, and manure application practices (Hunter et al., 2002).

*E. coli* O157:H7 isolates were first detected in the Oldman River Watershed in 1999 and 2000, by Johnson *et al.* (2003). Both *E. coli* and *Salmonella* spp. were detected in waters from surface drainage canals and storm drains; an overall prevalence of 0.9% and 6.2% of *E. coli* and *Salmonella*, respectively, was reported between 1999 and 2000 (Johnson et al., 2003). Another study tested raw, untreated and irrigation water in two irrigation districts in Southern Alberta for the presence of *E. coli* O157:H7 and *Salmonella* spp. between 2000 and 2001 (Gannon *et al.* 2004). The number of these bacterial pathogens increased during the summer months, which corresponds to the increased prevalence of the bacteria in cattle feces (Elder et al., 2000), and an increased incidence in human-related illness due to *E. coli* (Griffin & Tauxe, 1991). Although *E. coli* was isolated from only 1.3% of raw water samples between 2000-2001, the site incidence of *E. coli* was much higher, with detection in 28.5% of samples in 2000, and 32.5% of samples in 2001 (Gannon et al., 2004). The overall isolation rate of *Salmonella* spp. was much higher, at 10.3%; *Salmonella* spp. was isolated in approximately 63% and 90% of sampling sites in 2000, and 2001, respectively (Gannon et al., 2004).

The low levels of sensitivity in some isolation methods, in addition to the transitory nature of *E. coli* pathogens in waters make it difficult to isolate *E. coli* O157:H7 from waterborne outbreaks or in routine monitoring (Gannon et al., 2004). Establishing an effective standard isolation protocol for the identification of *E. coli* O157:H7 could improve monitoring methods and aid in the identification of sources of outbreaks, which in turn can help target sources of pollution.

### 5.3. Conclusions

Overall, fecal contamination of surface waters in Alberta is widespread and persistent. Evidence suggests that agricultural sources represent a significant threat to water supplies as a dominant source of fecal coliform bacteria. The widespread presence of other bacteria, such as *Salmonella* spp., additionally indicates the need to treat source waters before consumption. The relationship between high cattle density and incidence of enteric pathogens translates to an increased need for vigilance in Southern Alberta. Luckily, these bacteria are easily and economically eliminated with standard treatments, such as chlorination and UV disinfection. Indeed, the addition of a UV disinfection system to the Lethbridge water treatment plant in 1999 has resulted in a significant decrease in fecal coliform and *E. coli* counts downstream (Hyalnd et al., 2003). However, if water treatment failure occurs, wells become contaminated, or the accidental ingestion of recreational waters with high levels of pathogens occurs, public health is compromised. While fecal coliforms are easily treated, they may potentially serve as proxies for other types of contaminants that are more difficult to treat. When fecal coliform counts are high, it follows that other sources of pollution that accompany livestock wastes, such as nutrients, less-easily treated waterborne pathogens, or environmental estrogens, are similarly high, although studies are required to evaluate the utility and reliance of fecal coliform levels as proxies. Thus, continued monitoring programs to evaluate the quality of source waters, which in turn can be used to assess the effectiveness of current agricultural practices, legislation, and water quality guidelines, are instrumental in protecting source waters.

## 6. Protozoan Pathogens: Giardia and Cryptosporidium

*Giardia duodenalis* (syn. *G. lamblia*, *G. intestinalis*) and *Cryptosporidium parvum* are protozoan parasites that infect a wide range of vertebrates, including humans and their domestic stock. Both diseases are considered to be zoonoses, with livestock and wildlife representing potential sources of infection for humans. The transmissive stages of these parasites, cysts (*Giardia*) and oocysts (*Cryptosporidium*), are shed along with host feces. Infection occurs directly via a faecal-oral route, and indirectly if transmissive stages become waterborne, or contaminate food. The extremely low doses required to produce infection (Pereira, 2002; Caccio et al., 2005), in combination with the highly resistant nature of the transmissive stages make these diseases difficult to control. Both *Giardia* cysts and *Cryptosporidium* oocysts are resistant to environmental desiccation, high and low temperatures, and traditional chemical disinfectants, such as chlorine (Campbell et al., 1982). Additionally, these parasites are capable of passing through water filtration treatment processes (Carey et al., 2004). *Cryptosporidium* oocysts possess innate microbial resistance (Ramirez et al., 2004), and remain viable up to 140 days in water (Hooda et al., 2000). *Giardia* cysts are viable for up to 84 days in lakes and streams under winter conditions (deRegnier et al., 1989). Both protozoans are known to inhabit Canadian (Le Chavallier et al., 1991a; Wallis et al., 1996) and Albertan watersheds (Heitman et al., 2002). Thus, the presence of these parasites in source waters represents a valid public health concern in Alberta.

### 6.1. Giardiasis and Giardia Genotypes

Giardiasis is the most common enteric waterborne disease in Canada (Public Health Agency of Canada, 2008). It is characterized by persistent diarrhea, abdominal cramps, and loss of appetite (Thompson et al., 1993). Some cases are asymptomatic, and

these hosts represent a significant reservoir of infection (Percival et al., 2004). Untreated giardiasis lasts for a minimum of 10 days, up to three months, or even years (Percival et al., 2004), although a variety of antibiotic agents can be used to treat the disease (Gardner & Hill, 2001). Multiple host-adapted assemblages of *G. duodenalis* exist, of which two (assemblages A and B) are of significance to human public health (Thompson, 2003). As these two genotypes (primarily assemblage A) are also capable of infecting livestock, there is risk of zoonotic transmission. The complexities in identifying specific *Giardia* strains involved in an infection outbreak has made it difficult to assess the relative importance of animals and humans in waterborne transmission (see Erlandsen, 1994; Thompson and Boreham, 1994; Thompson, 1998; Thompson et al., 1990; 2000). Although the world health organization has deemed *Giardia* to be a zoonose, direct evidence documenting the role of livestock in direct (faecal-oral) and indirect (waterborne paths) transmission is limited (Thompson, 1998, Thompson et al., 2000).

## **6.2. Cryptosporidiosis and *Cryptosporidium* Species and Genotypes**

The first cases of Cryptosporidiosis were not reported until 1976, although infections are now reported worldwide (Ungar, 1990). Like *Giardia*, this disease presents itself as a typical flu-like illness, with symptoms including nausea, vomiting, and abdominal cramps. While the disease lasts between 9 and 15 days in healthy adults (Ramirez et al., 2004), Cryptosporidiosis can persist for years in immuno-incompetent hosts. Children under two are especially susceptible to acquiring the disease (Ungar, 1990), with potentially permanent growth and developmental problems occurring as a result of oocyst exposure (Molbak et al., 1997). Given the absence of effective adult drug treatments, and the potentially fatal nature of the disease, particularly for the immune-compromised, Cryptosporidiosis is generally considered to be a greater threat than *Giardia*. The

Milwaukee waterborne outbreak in 1993, in which 400,000 people were infected and 100 died, created an increased need of awareness for the modes of transmission and sources of the disease. Mathematical modeling based on data from this outbreak determined that ingestion of a single oocyst by some individuals was sufficient to invoke the disease (Haas & Rose, 1994).

Currently, 16 species and over 40 genotypes of *Cryptosporidium* are recognized (Fayer et al., 2006). Although 8 species have been identified in humans (Xiao & Ryan, 2004), nearly all (97%) cases of human Cryptosporidiosis are attributed to 2 species: *C. parvum* and *C. hominis* (Caccio, et al., 2005). *C. hominus* is characterized by an anthropogenic pattern of transmission. It was not until recently that species designation was given to *C. hominis*, which had previously been described as a human-adapted genotype of *C. parvum* (Xu et al., 2004). *C. parvum* infects bovine, sheep, goats, swine, and humans, consequently possessing zoonotic potential (Ramirez et al., 2004). Thus, molecular methods recently applied to *Cryptosporidium* studies have greatly advanced the epidemiological understanding of this disease.

### **6.3. Agricultural Sources of Giardia and Cryptosporidium**

The zoonotic potential of *C. parvum* and *G. duodenalis* from livestock sources is well established by molecular studies. Specifically, cattle are believed to be the most dominant reservoir host of zoonotic infection for these diseases, although other species of livestock and wildlife should not be ignored as potential sources. The transmission of *Giardia* (Majewska 1994) and *Cryptosporidium* (Miron et al., 1991; Lengerich et al., 1993; Prieser et al., 2003; Roberson et al., 2006) from cattle to humans has been confirmed.

There exists concern that zoonotic assemblages of *Giardia* and *Cryptosporidium* can be transported from fecally-contaminated soils to surface and groundwater. Despite the complex pattern of transmission such a route entails, in addition to the absence of knowledge on the role of livestock on waterborne outbreaks of *Giardia* and *Cryptosporidium*, there is a definite potential for agricultural sources of (oo)cysts to contaminate source water (Atwill et al., 2002; Thompson et al., 2003). Agricultural sources are often cited as the source of waterborne outbreaks, though confirmation by molecular evidence is lacking (Ramirez et al., 2004). Indeed, pasture runoff has been implicated with outbreaks of both Giardiasis and Cryptosporidiosis in humans, though it is evident that additional molecular studies on waterborne outbreaks are integral to our understanding of the epidemiology of these diseases (LeChevallier et al., 1991a; Olson, 2004). Thus, the livestock industry represents a valid public health concern as a potential source of waterborne outbreaks of Cryptosporidiosis and Giardiasis, especially given Alberta's dominance in the livestock industry in Canada. Alberta boasts 2400 confined feedlot operations, and over 5.5 million head of cattle (Stats Canada, 2009). High densities of livestock ensure high rates of transmission (Ramirez et al., 2004), and thus produce regions of continuous environmental contamination with high prevalence of disease (Jager et al., 2005). There is ample molecular evidence that *C. parvum* (Weilinga et al., 2008) and specific *Giardia* assemblages have anthropogenic potential (Thompson, 2003). Evaluating the prevalence of zoonotic genotypes of parasite infections in both humans and livestock represents the first step in producing an accurate risk assessment of livestock as sources of *Giardia* and *Cryptosporidium*.

## 6.4. Giardia in Livestock

Although *Giardia* has been detected in sheep, horses, and swine across Canada, the focus of *Giardia* in livestock has been on cattle (Olson et al., 1997). Cattle livestock operations are considered the greatest source of potential *Giardia* contamination, due to prevalence and intensity of *Giardia* infection in cattle (Olson et al., 1997b), high faecal outputs associated with cattle farming, and the high densities of cattle involved in these operations (Appelbee et al., 2003), especially in Alberta. Generally, the prevalence of *Giardia* in cattle is believed to be greatly underestimated, as cysts are shed intermittently and studies often only collect a single faecal sample per individual (O'Handley et al. 2000; Ralston et al., 2002; Appelbee et al. 2003). Additionally, methods to test for *Giardia* are not always sensitive enough for positive detection of cysts. Thus, infected hosts are capable of producing faecal samples void of *Giardia* cysts, or below levels of detection.

### 6.4.1. Giardia in Cattle

Overall, *Giardia* infection is higher among calves than cows, with greatest prevalence in dairy calf-cow operations (Olson, et al., 1997a). Generally, this trend is primarily attributed to features of the industry. As transmission is particularly high among calves (Xiao, 1994), a relatively short 2 month beef calving season reduces transmission among this population, compared to year round introduction of calves in dairy farms (Mohammed et al. 1999; Jenkins, 2001). Several studies estimate the prevalence of *Giardia* in young beef calves (<6 months) at approximately 30% (Buret et al., 1990; Olson et al., 1997b), with a prevalence of 6-11% in adult cows (Olson et al., 1997b; Hoar et al., 2001; Ralston et al., 2002). A study conducted in the North Saskatchewan River Basin in Alberta found a similar prevalence of *Giardia* in cow-calf operations. Of the cow-calf fecal samples collected from 7 operations in 1998, 20% were found to be positive for *Giardia*

*duodenalis*; in 1999, 46% of fecal samples across 11 operations tested positive (Heitmen et al., 2002). Only one study to date has examined prevalence and genotype of *Giardia* in beef calves in Alberta. Results of this study indicate similar prevalence to those previously reported for beef calves, around 34%, with the vast majority of infections (97.4%) belonging to a non-zoonotic assemblage (Appelbee et al., 2003). Thus, this study suggests that beef cattle operations pose a minimal threat as a source of *Giardia* assemblages known to infect humans, though additional genotyping studies are needed.

The prevalence of *Giardia* in dairy farms is reported to be very high, ranging from 50-100% in North America (Xiao, 1994; Xiao & Herd, 1994; Olson et al., 1997b; O’Handley et al., 1999). Approximately 25% and 5% of samples collected from two individual dairy farms in the North Saskatchewan River Basin in Alberta were positive for *Giardia* (Heitmen et al., 2002). One Alberta-based study reported a *Giardia* prevalence of 57% in dairy calves, with 17% of these calves bearing the assemblage with zoonotic potential (O’Handley et al., 2000).

#### **6.4.2. Prevalence of Cattle with Zoonotic *Giardia* Assemblages**

Generally, *G. duodenalis* genotyping studies report a higher prevalence of the host-adapted assemblage E than zoonotic assemblages A and B among cattle. This is not surprising, as host-adapted genotypes are often capable of out-competing those assemblages with a wide range of hosts (O’Handley et al., 2000). However, a series of studies conducted by Trout *et al.* report prevalence of zoonotic assemblages of *Giardia duodenalis* in cattle that should warrant concern (2004; 2005; 2006; 2007). Zoonotic assemblages of *Giardia* were reported in nearly 25% of dairy cattle from farms in Ontario, and some calves were found to harbour mixed *G. duodenalis* assemblages A and E (Coklin et al., 2007). Studies conducted in Belgium and New Zealand also provide

evidence that dairy calves in particular represent a significant source of the zoonotic Assemblage A (Geurden et al., 2008; Winkworth et al., 2008). Interestingly, it has been suggested that humans may serve as a source of assemblage A infection in cattle (O'Handley & Olson, 2006).

## **6.5. Prevalence and Distribution of *Cryptosporidium parvum* Among Humans**

Evaluating the relative prevalence of *C. parvum* and *C. hominus* infections in humans is key in assessing transmission dynamics and risk assessments of livestock sources of *Cryptosporidium*. However, it should be noted that the presence of *C. parvum* is not indicative of zoonotic transmission, as *C. parvum* can be transmitted between humans (McLauchlin et al. 2000). An extensive review by Xiao (2010) evaluated the geographic distribution of *Cryptosporidium* spp. in humans. Infection by *C. hominus* and *C. parvum* were common in European countries; *C. parvum* infections dominated in the Middle East, and infections in developing countries were primarily due to *C. hominus* (Xiao, 2010). Western and Southern Australia reported 83% and 72% of *Cryptosporidium* cases, respectively, due to *C. hominus*, with remaining proportions due to *C. parvum* (Morgan et al., 1998; Chalmers et al., 2005; O'Brien et al., 2008). Closer associations to agriculture have been suggested to result in higher rates of infection by *C. parvum* (Ramirez et al., 2004). For example, estimates by several studies indicate that over 75% of *Cryptosporidiosis* cases in the U.S are caused by *C. hominus* (Peng et al. 1997; Sulaiman et al., 1998). In contrast, a study conducted in the U.K. determined only 38% of *Cryptosporidium* cases are caused by the human genotype, with the remaining 62% due to *C. parvum* infection (McLauchlin et al., 2000).

The species of *Cryptosporidium* may impact the transmission dynamics of the disease. It has been suggested that zoonotic transmission of *C. parvum* results in sporadic cases, rather than outbreaks of the disease (Feltus et al., 2006). As sporadic cases of *Cryptosporidium* are less frequently reported than waterborne outbreaks, it follows that *C. parvum* infection may be underestimated. Indeed, the number of cases reported is believed to be only a fraction of those that occur (Fayer et al., 2000)

## **6.6. Cryptosporidium Prevalence in Livestock**

Cattle are widely regarded as the primary source of *C. parvum*; the contribution by other livestock species is relatively insignificant. Studies conducted in Australia, Denmark, Norway, Northern Ireland, and Spain have reported a marked absence of *C. parvum* alongside the presence of pig-adapted genotypes/species in swine (see Xiao 2010 for review). Overall prevalence of *C. parvum* in sheep is low. However, geographic differences in the distribution of *C. parvum* in sheep indicate the potential for human transmission to occur in some areas; studies conducted in Australia, U.S., Belgium, the U.K., and Tunisia determined sheep were primarily infected with *C. bovis* and cervine *Cryptosporidium* genotypes, although recent studies indicated *C. parvum* dominated infections of lambs in the U.K. and Spain (Xiao, 2010).

### **6.6.1. Cryptosporidium Prevalence in Cattle**

Overall, prevalence of *Cryptosporidium* spp. in Canada appears highly variable within the literature. The most comprehensive Canadian study reported 20% of cattle across 15 geographical locations to be infected with *Cryptosporidium* spp. (Olson et al., 1997b). Gow and Waldner (2006) recorded relatively low levels of *Cryptosporidium* spp. in beef calves (3.1%) across 100 Western Canadian farms. *Cryptosporidium* prevalence of

beef calves on a ranch near Calgary, Alberta, was also reported to be relatively low, with only one (5%) beef calf shedding *C. parvum* oocysts (Ralston et al., 2003). Although *Cryptosporidium* prevalence on two calf-cow dairy operations in the North Saskatchewan River basin was only around 4%, oocyst concentration from these agricultural sources far exceeded concentrations from wildlife and sewage influent (Heitmen et al., 2002).

The dominant species of *Cryptosporidium* that infect cattle are *C. parvum*, *C. andersoni*, *C. bovis*, and *C. ryanae* (Xiao, 2010). Extensive literature describes an age-related pattern of prevalence of these genotypes in cattle. Studies of dairy cattle indicate *C. parvum* dominates in pre-weaned calves, *C. bovis* and *C. ryanae* in weaned calves, and *C. andersoni* in yearling and adult cattle (see review by Xiao, 2010). It is suggested that these patterns are similar in beef cattle, though more study is needed (Xiao, 2010).

The primary factors affecting *Cryptosporidium* spp. prevalence among cow-calf operations are age (calf versus cattle), and livestock operation type (beef versus dairy farms). Generally, calves display a higher prevalence of *Cryptosporidium* spp. infection than cattle. Estimates of dairy calf infection with *C. parvum* in the first months of life are virtually 100% (Xiao & Herd, 1994; O'Handley et al., 1999). Adult cattle have been implicated as potential parasite reservoirs to calves (Upton, 2003), though the likelihood of this statement has been disputed (O'Handley, 2007). Evidence suggests that exposure to *C. parvum* in calves provides lasting immunity to subsequent infection (Harp et al., 1990). A recent review by O'Handley suggests that high rates of infection among dairy calves, combined with immunological developments, translate to the low rates of infection observed among adult dairy cattle (2007). Indeed, studies have reported very low prevalence of *C. parvum* among post-weaned calves and adult dairy cows in the Eastern

U.S. (Santin et al., 2004; Fayer et al., 2006), and Eastern Canada (Uehlinger et al., 2006). Studies on prevalence of *C. parvum* in beef calves and cattle, however, are lacking.

Cryptosporidium prevalence on dairy cow-calf operations generally exceeds that of beef cow-calf operations (Kvac et al. 2006). This trend is also observed with *Giardia*, and is similarly suggested to be the result of specific calving practices employed by dairy and beef operations (Olson et al., 2004). Additionally, beef cows are reported to have higher rates of passive immunity than their dairy counterparts (Guy et al., 1994), which are also believed to play a role in Cryptosporidium host distribution (O’Handley 2007). Furthermore, the higher quantities of oocysts excreted by dairy calves relative to beef calves should translate to increased transmission among dairy farms (Gow & Waldner, 2006).

## **6.7. Giardia and Cryptosporidium in Raw, Treated, and Drinking Water**

Clearly, the consumption of unfiltered and untreated drinking water represents a significant risk for both Giardiasis (Hoque et al., 2002; Jakubowski & Craun, 2002) and Cryptosporidiosis. However, evidence suggests that even treated drinking water can pose a risk of these diseases. While conventional UV and chlorine treatment is generally effective in reducing numbers of *Giardia* and *Cryptosporidium* (oo)cysts to acceptable levels (Rose et al., 1997), these protozans have been found to persist past tertiary water treatments such as coagulation, flocculation, sedimentation, and filtration (Gennaccaro et al., 2003; Quintero-Betancourt et al., 2003; Carey et al., 2004). Studies conducted on waterborne outbreaks of both *Giardia* and *Cryptosporidiosis* confirm that (oo)cysts are capable of passing through drinking water treatment processes (Wallis et al., 1996; Fayer

et al., 2000). However, there does appear to be a reduced viability and prevalence of these (oo)cysts in treated drinking water compared to raw, untreated water.

## **6.8. Giardia and Cryptosporidium in Raw and Treated Drinking Water in Alberta**

A study by LeChavellier *et al.* reported high levels of *Giardia* spp. and *Cryptosporidium* spp. in raw water samples collected from surface water treatment plants in 14 Central-Eastern and Eastern states, and Alberta (1991a). *Giardia* cysts and *Cryptosporidium* oocysts were detected in 81% and 87% of these raw water samples, respectively (LeChavellier, et al. 1991a). An extension of this study reported that upon treatment by the surface water treatment plants, *Giardia* cysts and *Cryptosporidium* oocysts were detected in 17% and 27% of drinking water samples, respectively, with low levels of (oo)cyst viability reported (LeChavellier, 1991b).

Using a slight modification of water analysis methods employed by LeChavellier et al. (1991a,b), Wallis *et al.* (1996) conducted a comprehensive study of *Giardia* and *Cryptosporidium* prevalence in Canada. A total of 1760 raw water, treated water, and raw sewage samples across 72 Canadian municipalities were tested. Water treatment plants in approximately 80% of these municipalities relied solely on chlorination disinfection methods (Walls et al., 1996). *Giardia* cysts were prevalent in 73% of raw sewage, 21% of raw water, and 18.2% of treated water samples. The prevalence of *Cryptosporidium* was much lower, with oocysts detected in 6.1% of raw sewage, 4.5% of raw water, and 3.5% of treated water samples. The combined prevalence of *Giardia* and *Cryptosporidium* in 39 samples of raw and treated drinking water in Alberta was approximately 20% and 5%, respectively (Wallis et al., 1996). Similar to reports by LeChavallier *et al.*, (1991b) the viability of (oo)cysts was determined to be frequently low.

## **6.9. Prevalence of Giardia and Cryptosporidium in Alberta**

In 2004, 491 cases of Giardiasis were reported in Alberta (15.4/100,000 people), with the highest rates of infection occurring in Southern Alberta, and along the western border of the province (Alberta Health and Wellness, 2008b). Cryptosporidiosis is generally low in Alberta, with only 104 reported cases in 2004 (3.2 per 100,000 people) (Alberta Health and Wellness, 2008a). Risk models have been developed to assess the maximum acceptable concentration of Giardia and Cryptosporidium (oo)cysts in drinking water. Estimates of the maximum acceptable concentration (MAC) of (oo)cysts are made difficult by theoretical calculations that suggest acceptable concentrations far below current detection limits. Presently, Canadian Water Quality Guidelines do not propose a numerical guideline for Giardia or Cryptosporidium. If (oo)cysts are suspected to be present in source waters, a treatment regime, or watershed/wellhead protection plan should be executed (Health Canada).

## **6.10. Conclusions**

It is important to recognize the recent advancements in molecular and genetic identification of host-specific strains of both *G. duodenalis* and *C. parvum*. Molecular studies are integral in assessing the risk of livestock transmission and confirming specific sources of waterborne Giardia and Cryptosporidium outbreaks. A greater understanding of the potential sources of zoonotic transmission, and those hosts which have the capacity to act as reservoirs of disease, in addition incorporating genotyping data via molecular techniques, and viability assays are necessary to produce a more effective risk assessment framework.

The livestock industry is of significance to public health as a source of both *Giardia* and *Cryptosporidium* infection to humans. Both parasites are widespread, with *Giardia* and *Cryptosporidium* present in as many as 29% and 20% of cattle at various locations across Canada (Olson et al., 1997b). The large number of livestock operations, in addition to high density of operations in Southern Alberta result in potential elevated risks of Giardiasis and Cryptosporidiosis. High rates of cyst and oocyst shedding can produce elevated levels of environmental contamination, and pose a potential threat to drinking water supplies.

Assessing the likelihood that livestock play a role in waterborne outbreaks of *Giardia* and *Cryptosporidium* is no easy task. First, the prevalence of the disease in livestock animals must be known. Additionally, the proportion of those infections with zoonotic potential must be determined, using molecular techniques. The potential for excreted (oo)cysts to be transferred from fecally-contaminated soils to surface and ground waters needs to be evaluated. Those parasites in groundwater may go on to contaminate wells, or become discharged in surface waters. To contaminate drinking waters, the transmissive stages must reach and pass through drinking water treatment plants, and still retain viability. Although this appears to be an extraordinarily complex path of transmission, the sheer densities of livestock in Southern Alberta, coupled with enormous levels of (oo)cyst excretion by livestock, in addition to manure application practices and the capacity for agricultural runoff to permeate our watersheds, the potential for the spread of waterborne infection are definite. The severity of these infections, particularly *Cryptosporidium*, warrants extreme caution.

## **7. Endocrine Disrupting Compounds**

Endocrine Disrupting Compounds (EDC's) are those compounds that disrupt the endocrine system through mimicking, antagonizing, or interfering with the biosynthesis or biodegradation of endogenous hormones. The most common endocrine disruptors are those that possess estrogenic and androgenic effects. EDC are an emerging contaminant, as research has only recently begun to establish the effects these compounds have on aquatic environments. The presence of these compounds in source waters also represents a threat to public health. The scope of this document will be to focus on those estrogenic and, to a lesser extent, androgenic compounds that are present in aquatic environments, at effective concentrations that pose a threat to the quality of surface and groundwaters.

### **7.1. Hormone Mimics**

Environmental estrogens, also referred to as xenoestrogens, are an umbrella term used to describe all those anthropogenic compounds that mimic estrogens. Common environmental estrogens include pesticides (ex. DDT, methoxychlor, and chlordane), surfactants (Alkylphenol polyethoxylates: APE's), and coolants such as polychlorinated biphenyls (PCB's). Natural plant and fungi products are known to function as estrogens (Arcand-Hoy et al., 1998). Recently bisphenol A (BSA), a component used in a wide variety of plastics, has received significant media attention due to its estrogenic capacities. As a result, the Canadian government is taking steps to limit the exposure of infants to BSA via banning the sale and import of polycarbonate baby bottles (Webster, 2008).

## 7.2. Natural & Synthetic Estrogens: Pharmaceuticals

Natural estrogens refer to the group of steroid compounds that include estrone (E1), estradiol (E2) and estriol (E3). Estradiol (also known as  $17\beta$ -estradiol) is the primary female sex hormone responsible for the development of secondary sexual characteristics; estriol and estrone represent its derivatives. Estradiol is also naturally present, at lower concentrations, in males. Both naturally occurring and synthetic or environmental estrogens bind to specific receptors in the body, eliciting an estrogenic response. Thus at higher than normal concentrations, even naturally-occurring estrogenic hormones have the capacity to act as endocrine disruptors.

Unlike environmental estrogens, which possess estrogenic qualities as an unintended side effect, a variety of compounds have been synthetically developed with the intention of producing estrogenic results. Consequently, these compounds are designed to produce physiological results at low concentrations. The synthetic analogue of estradiol,  $17\alpha$ -ethynylestradiol or EE2, is used extensively in oral contraception and hormone replacement therapy (Arcand-Hoy et al., 1998). Approximately 1.3 million women between 15-49 years of age reported using oral contraceptives in Canada (Wilkins et al., 2000), while approximately 648 thousand women aged 45-64 use some form of hormone replacement therapy (Beaudet et al., 1997). Endogenous estrogens are excreted by both men and women; estrogen excretion is highest among pregnant women, followed by women in the follicular phase of the menstrual cycle. Oral contraceptives have been shown to cause increases in the rates of estrogen excretion of women, to levels comparable to that of pregnant women (Fahl and Rose, 1970). Though no studies have been conducted on estrogen excretion rates of women on hormone replacement therapy,

it follows that the oral ingestion of low levels of synthetic estrogens should cause some increase in estrogen excretion.

### **7.3. Agricultural Pharmaceuticals**

Both natural and synthetic estrogens are also used in veterinary medicine as growth enhancement products in the beef industry. There are 6 hormone growth products (HGPs), both naturally occurring and synthetic, that are approved for use in Canada (and the United States). Naturally occurring hormones licensed for use include 17- $\beta$  Estradiol (E2), testosterone, and progesterone. Synthetic products include zeranol (an estrogenic), melengestrol acetate (MGA; a progesterone mimic), and trenbolone acetate (TbA; an androgenic) (Health Canada, 2005). HGP's are used to promote muscle growth, which in turn reduces fat deposition, and improves feed conversion efficiency. Thus, a greater proportion of lean meat is produced at a lower cost. All hormonal growth products are administered as implants injected behind the ear, with the exception of MGA, which is administered as a food additive (Health Canada, 2005)

Extensive studies conducted by Health Canada and the US Food and Drug Administration have determined that in concert with proper veterinary practices, HGP pose no risk to consumers. However, there exists concern about the fate of excreted estrogens and androgens in the environment. Livestock manure naturally contains significant quantities of steroid hormones; the addition of HGP exacerbates these quantities. Cattle that have been administered growth hormone products have been shown to have urine estrogen concentrations 5 to 6 times greater than their relative controls (Callentine et al., 1961). Studies suggest that cattle administered exogenous hormones excrete higher quantities of estrogens in their feces as well. Liquid fecal slurry from steers administered

HGPs had detectable levels of E2 and E3, whereas steers without HGP did not (Sellin et al., 2009). Additionally, the estrogenic activity of feces excreted by HGP implanted steers was found to be greater than that of relative controls (Sellin et al., 2009).

Regardless of the source, endogenous and exogenous compounds function as endocrine disrupting chemicals in the environment. There is increasing anxiety regarding the levels of estrogenic compounds in the environment. In addition to the wide variety of domestic and industrial compounds that act as estrogen mimics, there is concern regarding the abundance of natural and synthetic estrogens excreted by humans into wastewater. Evidence suggests that the processes employed by some sewage treatment plants do not effectively remove estrogenic compounds from wastewater before it is released into the environment for use downstream. Additionally, there is concern that estrogenic and androgenic compounds excreted in the waste products of livestock could accumulate in the soils and potentially contaminate aquatic ecosystems.

#### **7.4. Binding Affinities and Potency**

Both naturally occurring and synthetic estrogens and androgens and their mimics (ligands) bind to their respective receptors in target cells, eliciting estrogenic or androgenic responses. The ligand-receptor binding affinity is variable; those compounds with a higher binding affinity generally elicit a more pronounced effect (Arcand-Hoy et al., 1998). Thus, it is important to characterize the relative potency of estrogens, androgens, and their metabolic constituents, in order to develop an effective risk assessment of these compounds.

Industrial pollutants and pesticides generally have low levels of potency compared to endogenous hormones. Thus, exposure to pharmaceutical estrogens is of particular

concern, as estradiol (E2) is anywhere from 50 to 10, 000 times as estrogenic as pollutants known to have estrogenic effects such as APE's and organochlorides (Arnold et al., 1996). The synthetic analogue of estradiol (EE2) and estrone (E1) are reported to have the most potent estrogenic effects (Harries et al., 1996, 1997; Metcalfe et al., 2001). Given that laboratory studies, using in vivo mammalian assays, have determined E2 and E3 to be about 10, 000 times more potent than BSA (Milligan et al., 1998), it is not surprising that there is mounting concern over the presence of natural estrogens and their synthetic analogues in the aquatic environment.

## **7.5. Estrogenic Effects on Humans**

It is widely accepted that EDC play a role in many adverse health conditions in both humans and wildlife. A comprehensive review conducted by Choi et al. reported that of 48 EDC reported in the literature 79% were carcinogens, 52% were immunotoxic, and 50% functioned as neurotoxins (2004). As those organs which possess receptors for steroidal hormones are at a high risk of estrogenic effects, reproductive disorders represent a primary effect of exposure to environmental estrogens, though non-reproductive related health problems are by no means uncommon. Exposure to EDC's are believed to play a role in the increased prevalence of testicular cancer, prostate cancer, breast cancer, reductions in sperm counts, negative impacts on fetal development, and reduced mental and physical development in children (Sharpe & Skakkebaek, 1993; Harrison et al., 1997; Toppari et al. 1996; Sonnenschein and Soto, 1998; Colburn et al., 1993; Matozzo et al., 2008).

## 7.6. Estrogenic Effects on Wildlife

All vertebrates excrete estrogens, and respond to them (Safar-Hermann et al., 1987; Shore & Shemesh, 2003). The effects of estrogenic exposure on aquatic vertebrates have been well documented. The production of vitellogenin in male fish and amphibians, an egg precursor in oviparous vertebrates, is widely regarded as a biomarker for estrogenic contamination in the aquatic environment (Sumpter & Jobling, 1995). Extensive studies have documented a significant production of vitellogenin in freshwater and marine male fish (Segner et al., 2003; Jobling & Tyler, 2003; Sumpter, 2005; Metthiesen, 2003; Folmar et al., 1996; Bjerregaard et al., 2008) and amphibians (Kloas et al., 1999) due to the presence of estrogenic pollutants in aquatic environments. E2 (estradiol), E1 (estrone), and ethynylestradiol (EE2-synthetic analogue of E2) are believed to be the primary causative agents in the feminization of male fish, although industrial chemicals, such as APE's are known to play a role in some cases (Sumpter, 2005). Segner et al. (2003) provide evidence that suggests exogenous estrogens have the capacity to impact the reproductive and developmental capacity of both aquatic invertebrates as well. Effects of environmental estrogens have additionally been reported in panthers (Facemire et al., 1995), alligators (Guillette et al., 1994), and birds (Shugart, 1980; Fry, 1995). The contamination of surface waters by estrogenic compounds has been recently shown to be detrimental to entire fish populations (Kidd et al., 2008)

Estrogens are capable of producing estrogenic effects at remarkably low concentrations in the aquatic environment. Levels ranging from 10-100ng/L are regarded as biologically significant (Hanselman, 2003), although estrogenic effects have been observed at concentrations below 10 ng/L. Studies have reported that concentrations as low as 1ng/L of E2 are capable of inducing vitellenogen production in male fathead

minnows (Purdom et al., 1994) and male trout (Hansen et al., 1998), while concentrations as low as 0.1ng/L of 17 $\alpha$ -ethynylestradiol (the synthetic analogue of E2) caused the formation of ova in the testes of male Japanese medaka (Metcalf et al., 2001). Several studies have observed greater estrogenic effects using bioassays than otherwise predicted by chemical assays (Soto et al., 2004; Sarmah et al., 2006). Thus, combinations of some estrogenic compounds in the environment have been found to have synergistic effects that augment estrogenicity (Sumpter & Jobling, 1995).

## **7.7. Estrogens and Agriculture**

Estrogens are excreted into the environment through the feces and urine of all livestock animals. The amount of excreted estrogen is dependent upon a wide variety of factors, including animal age, sex, species, reproductive status, diet, and treatment with HGP (Lorenzen et al., 2006; Sarmah et al., 2006). The highest levels of estrogens appear to be from swine waste, followed by dairy cattle waste and chicken litter (a mixture of chicken wastes, bedding, and feathers) (Lee et al., 2007). Although beef cattle have been reported to possess relatively low levels of estrogen excretion compared to dairy cattle (Lee et al., 2007), the number of confined beef feedlot operations in Southern Alberta raises legitimate concerns over potential EDC contamination from beef cattle wastes. Although it might follow that the risk of estrogen contamination by agricultural runoff via manure application compared to direct discharge of wastes into waterways via sewage treatment plants should be relatively low, studies suggest that livestock excretions represent a significant source of estrogen contamination in the aquatic environment. The estimated emissions of E1 and E2 from swine and dairy farms in the U.S are greater than the total estrogen flow from sewage treatment plants (Raman et al., 2004).

The route of excretion (feces versus urine), and the resulting residues following the metabolism of estrogens is also species-specific. For example, cattle excrete the majority (58%) of estrogens via a fecal route, with over 90% of estrogens excreted as both 17 $\alpha$ -estradiol, and E2, with estrone excreted in both conjugated and free forms (Erb et al., 1977, as cited in Hanselman et al., 2003). Conversely swine and poultry excrete most (96% and 69% respectively) of their estrogen via urine, and rarely as 17 $\alpha$ -estradiol. Generally fecal-excreted estrogens are more often in free, un-conjugated forms, while urinary estrogens tend to be conjugated; the environmental significance of un-conjugated versus conjugated estrogen is not yet known, though conjugated forms are believed to be comparatively short-lived (Belfroid et al., 1999; Baronti et al., 2000).

#### **7.7.1. Manure Application as a Source of Estrogen**

The agricultural practice of manure application to fertilize fields presents a potentially significant means for estrogens and their metabolites to access the aquatic environment via run-off. Thus, evaluating the persistence and viability of excreted estrogenic compounds in animal manure, soil profiles, and surface water are important parameters in assessing the risks of estrogenic contamination of watersheds. Generally, literature on the persistence of estrogens and their metabolites in soils or the aquatic environment is limited. The physical structure of estrogenic compounds, in addition to environmental factors such as moisture, sunlight, oxygen availability, temperature, and soil types, affect the half-life of estrogens. Assessing the environmental fate of these compounds can be a complex and laborious process.

### **7.7.2. Estrogens in Soils**

Estrogens are slightly hydrophilic, nonvolatile compounds, which are readily adsorbed (bound) into soils and aquatic sediments, and are thus believed to have limited mobility (Colucci et al., 2001; Halthaus et al., 2002; Casey et al. 2003). Research indicates that estrogen leaching is limited to top soil profiles (Lee et al., 2003; Casey et al., 2005). Laboratory studies conducted by Colucci and Topp (2001) indicate that estrogens rapidly dissipate in agricultural soils under conditions present during the growing season, and thus contribute environmentally insignificant levels of estrogens to aquatic environments. Similar conclusions were drawn by Lorenzen et al. (2006) when examining the persistence of a variety of natural and synthetic estrogenic compounds in animal manure and municipal bio-solids. These conclusions are supported by a field study that reported the half-lives of estradiol and estrone in English streams to be between 0.2-9 days and 0.1-11 days, respectively (Jürgens et al., 2002). Therefore, it is believed that the rapid biodegradation of these compounds would contribute insignificant estrogenicity to aquatic environments. Additionally, studies suggest that estrogens are prone to degradation by a wide variety of microorganisms (review by Hanselman et al., 2003), by exposure to oxygen/aerobic conditions (Lorenzen et al., 2006), and UV radiation/sunlight (Liu & Liu, 2004).

Although bound estrogen residues are believed to pose a reduced risk of contaminating the water supplies (Colucci & Topp, 2001), laboratory-based experiments on the sorption of estrogenic compounds to soil particles do not include soils that have been treated with manure. The ways in which chemical, physical, and microbiological changes that accompany manure applications to soil alter the sorption of estrogens is unknown (Hanselmann et al., 2003). It should be noted that natural estrogens estradiol,

estrone and estriol, are generally considered more biodegradable than synthetic estrogens such as ethynylestradiol (EE2) (Fine et al., 2003). Understanding the behaviour and transportation of steroidal hormones through soil profiles would provide a better understanding of the risks manure and its application as fertilizer plays as a non-point source of EDC's (Arnon et al., 2008).

### **7.7.3. Estrogens in Agricultural Runoff**

While laboratory-based studies generally conclude that estrogens biodegrade too quickly to contaminate soil and waters, evidence provided by field studies suggest otherwise. First, several studies have demonstrated that appreciable levels of steroid hormones are capable of leaching into soil, thereby potentially contaminating groundwater. Low levels of estrogens derived from chicken wastes were detected in well water samples (Shore et al., 2003). Kjaer et al. (2007) reported the leaching of E1 and E2 from the root zone of soils treated with manure, into tile drainage systems. Three months after manure application, biologically significant levels of E1 were still detected. Another study detected estrogens in deep soil profiles 32m below a dairy farm manure storage lagoon (Arnon et al., 2008). Additionally, these dairy farm wastes were found to impact groundwater quality. The depths at which estrogens were detected are unexplained by hormone advection, dispersion, and sorption transport models. Thus, the authors suggest additional mechanisms must influence the transport of hormones in soil profiles, such as preferential flow rates (Arnon et al., 2008).

Peterson et al. (2000) found evidence to suggest that E2 can be transported to aquifers in a manner similar to fecal coliforms. This study examined groundwater springs in Northwestern Arkansas; the study area boasts a strong poultry production industry, in addition to 500,000 head of cattle. Results indicate that levels of E2 in spring groundwater

were highest following recharge events, and lowest when water levels were at base-flow conditions. The source of E2 is believed to be animal waste, as peak fecal coliforms, *E. coli*, and E2 levels coincided with each other. Thus, it appears that E2 displays similar movement patterns as fecal coliforms, as E2 concentration curves follow that of other parameters indicative of animal waste products.

Additional evidence suggests that manure application associated with the agriculture industry represents a potential non-point source for estrogen contamination of surface water. Livestock wastes from swine, cattle, and poultry represent major sources of estrogen loading in the environment (Hanselman, 2003; Raman et al., 2004; Furuichi et al., 2006). Multiple studies have determined significant concentrations of estrogens in streams receiving runoff from fields fertilized with chicken litter (Nichols et al, 1997; Finlay Moore et al., 2000, Shore, 2009), dairy (Dyer et al., 2001; Matthiesen et al., 2006), and cattle farms (Soto et al., 2004). Estrogenic effects on aquatic vertebrates in waters downstream from agricultural runoff should serve as further proof of the capacity for estrogen contamination to occur, although research has focused almost exclusively on the estrogenic effects of vertebrates downstream of municipal effluents. The few studies that have examined the estrogenicity of agricultural effluent indicate that, although generally displaying lesser estrogenic potential than municipal sources, agriculture does contribute to estrogen loading in aquatic environments. While beef cattle feedlot effluents were insufficient to feminize juvenile and male painted turtles, females had higher levels of circulating vitellogenin in plasma, which could have negative impacts on fitness (Irwin et al., 2001). Orlando et al. (2004) demonstrated that while feedlot effluents possessed androgenic properties, no indications of exposure to environmental estrogens were detected.

#### **7.7.4. Androgen and Progesterone in Agriculture**

Although the UN has reported the use of MGA and TbA as hormone growth products to be safe for both animals and consumers (Lauderdale et al., 1977; Lauderdale et al., 1983), as with estrogens, it is the presence of TbA, MGA, and their residual compounds which are excreted into the environment that raise concern among researchers. The metabolites of synthetic androgens possess longer half-lives, in manure and open water, than their naturally occurring counterparts, which possess half-lives on the order of hours or days (Meyer, 2001). A detailed study conducted by Schiffer et al. (2001) sought to determine the half-lives of hormonal growth products TbA (including both 17 $\alpha$ - and 17 $\beta$ - trenbolone residues) in cattle dung, liquid cattle manure and soil, and MGA in solid cattle dung and soil. The half-life of TbA residues in liquid manure under anaerobic conditions was approximately 260 days, while TbA metabolites were still detectable in solid cattle dung after 4.5 months of storage (Schiffer et al., 2001). Soils fertilized with cattle manure bore TbA metabolites 58 days after application (Schiffer et al., 2001). Like TbA, metabolites of the progesterone MGA were still detectable in solid dung for 4.5 months later. Soils fertilized with cattle manure still possessed MGA residues after 195 days (Schiffer et al., 2001). Not surprisingly, both TbA and MGA levels in the soils were lower than concentrations found in solid dung, as manure application effectively dilutes concentrations.

The extent to which TbA, MGA and their metabolites are susceptible to photochemical reactions caused by exposure to UV radiation, or biodegradability by microorganisms is unknown. The manner in which precipitation events affect the leaching of TbA and MGA residues into lower soil profiles, or promote run-off into surface waters is similarly unknown. A study detected the presence of testosterone 42 meters below a

dairy-farm manure storage lagoon, suggesting that hormonal transport through certain soil profiles occurs (Arnon et al., 2008). It is highly likely that the binding of TbA and MGA residues to various molecules impacts the mobility and solubility of these compounds. Schiffer et al. (2001) speculate on the behaviours of TbA and MGA based on well-studied industrial and agricultural contaminants. The authors suggest that those TbA and MGA residues that are not readily adsorbed could readily contaminate surface and groundwaters (Schiffer et al., 2001). Conversely, those TbA and MGA metabolites, which are readily sorbed by various compounds, could form stable associations, which allow them to persist and accumulate in the environment (Schiffer et al., 2001).

Like estrogens, remarkably low levels of androgens are required to product an effect on target cells. A concentration of approximately 0.027 µg/L resulted in the development of male secondary sexual characteristics, and reduced fecundity of female fathead minnows (Ankley et al., 2003); the range of 1-10 ng/L was sufficient enough to alter the reproductive biology of female mosquitofish (Sone et al., 2005). Jensen et al. (2006) and Seki et al. (2006), reported similar androgenic effects on fish exposed to low levels of androgenic compounds (cited by Sellin et al., 2009). Given the relatively long half-lives of TbA and MGA residues, in concert with the low concentrations required to produce biological effects, it is not surprising that effluent from cattle feedlots has been found to possess androgenic activity. Bioassays conducted by Soto et al. (2004), and in vitro studies by Durhan et al. (2006) detected androgenic activity in cattle feedlot effluent; additional studies determined that cattle feedlot effluents produced androgenic effects in fathead minnows as evidenced by reduced fecundity, and masculinization of female fish (Ankley et al., 2003; Orlando et al., 2004).

## 7.8. Municipal Sources of Estrogenic Compounds

While this report is focused on literature related to agricultural pollutants, there has been much important research conducted on the estrogenic effects of municipal effluent that should be mentioned. The evidence that human populations represent a major source of endocrine disrupting compounds, particularly estrogens, is undeniable. Municipal wastewaters are a complex mixture of natural estrogens, synthetic pharmaceutical estrogens used for oral contraception and hormone replacement therapy, estrogen mimics such as detergents, plastics, and cosmetics, etc., and their associated metabolites (Norris, 2007). These municipal wastewaters must pass through sewage treatment plants (STP) before being released into the environment, although several studies have demonstrated the inability of STP to filter out estrogens and their metabolites, resulting in estrogenic effluents (Daughton et al., 1999). Natural and synthetic estrogens, for example, were found to persist through processes in Ontario STP (Ternes et al., 1999). Studies conducted in the U.K. have determined that the active residues of E1, E2 and EE2 are released from STP into the environment (Desbrow et al, 1998). Thus, municipal sources contribute to a persistent discharge of estrogens into the aquatic environment.

The estrogenic effects of municipal effluent on aquatic vertebrates have been extensively studied for over a decade (Sumpter, 2005). Vitellogenin production due to estrogen exposure via STP effluent has been demonstrated in male carp (Folmar et al., 1996) roach (Jobling et al., 1998) fathead minnows (Purdom et al., 1994), and rainbow trout (Knudsen et al., 1997). Furthermore, these estrogenic effects are observable for several kilometers downstream of the effluent discharge (Harries et al., 1996; 1997). Studies have additionally demonstrated population level effects on aquatic vertebrates due to low-level chronic exposure of estrogenic compounds in surface waters. Populations

of fathead minnows in lakes experimentally exposed to levels of EE2 collapsed after 2 years (Kidd et al., 2007). The levels of EE2 employed in the study were comparable to those found in untreated and treated municipal wastewater (Kidd et al., 2007).

## **7.9. Conclusions**

Current concerns regarding the presence of endocrine disrupting compounds in biological organisms has resulted in the Canadian Environmental Protection Act (CEPA)'s mandate that Environment Canada and Health Canada conduct research on hormone-disrupting compounds (Servos et al., 2008). It is suggested that current ecosystem and human health monitoring should be modified to incorporate more sensitive endpoints related to growth, reproduction, and development (Sevros et al., 2001). Further, monitoring programs should be adapted to integrate and complement mechanistic and toxicology studies (Sevros et al., 2001). Intensive agriculture has been deemed an important site for consideration of the impacts of endocrine disruption (Sevros et al., 2001). Through discussion fostered by a multi-departmental workshop arranged by the Five Natural Resources Departments Working Group on Endocrine Disrupting Substances, the priorities of research needs and risk assessment, were determined (Servos et al., 2000a); further details on the workshop are available in Servos et al., 2000a. Overall, four components of a National Agenda for EDC in the Canadian Environment have been proposed; increased national leadership and communication on the issues of EDC in Canada, establishment of a stronger foundation of knowledge related to exposure and effects of EDC in the environment, the development of standardized national and international screening and testing protocols, and focused scientific assessment and action on priority substances (Servos et al., 2000b). Presently, despite

the intensity of livestock operations in Alberta, there does not appear to be any initiative from Alberta Environment to address the issues surrounding EDC in source waters.

## **8. Emerging threats to Alberta source waters**

While this report focused on the dominant agricultural pollutants in Alberta's source waters, there are numerous emerging threats to source waters that warrant mention here. Increasingly, there are concerns being raised about mercury levels in fish in Southern Alberta, toxic contamination of the Athabasca watershed as a result of oil sands development, and the presence of fire retardant additives in Canadian waters. Although this certainly doesn't exhaust the emerging threats to source waters, these represent some of the more pressing concerns highlighted by media coverage.

### **8.1. Methylmercury**

Mercury is widespread throughout the environment. The dominant anthropogenic source of mercury is via combustion of wastes and fossil fuels, such as coal, that contain inorganic mercury (National Research Council, 2000), although mercury is also naturally present in sediment, soils, rocks and waters. Through the process of methylation by anaerobic aquatic organisms, an organic carbon bonds to a mercury atom; the resulting compound is an organic compound, methylmercury, a well-known, persistent environmental toxicant. This compound is biomagnified throughout the food chain; top fish predators have been documented to contain levels of mercury one million times greater than in the surrounding waters (Weiner et al., 2003). The consumption of fish bearing high levels of methylmercury is a significant public health concern, as the gastrointestinal tract readily absorbs this compound, and can lead to impairments of the immune system,

reproductive system, cardiovascular system, nervous system, and the liver (National Research Council, 2000).

Through the creation of water storage reservoirs, naturally occurring mercury is released from disturbed sediments, soils, and rocks into the surrounding waters. Peaks in mercury concentrations in the associated biota has been reported approximately 2-5 years after initial reservoir impoundment (Abernathy & Cumbie, 1977; Tremblay et al., 1991, Hall et al., 1998), following a slow 15-30 year decline to background levels (Bodaly et al., 2007). A recent study conducted on mercury biomagnification in the Twin Valley Reservoir in Southern Alberta determined that the majority of fish sampled, predominantly northern pike and white sucker, contained levels of mercury exceeding Health Canada consumption guidelines (Brinkmann & Rasmussen, 2010). The levels of total mercury in northern pike were approximately three times greater than levels observed of the same species of similar size in the Oldman River (Brinkmann & Rasmussen, 2010). Presently, 17 consumption advisories in Alberta recommend that women between the ages of 15 and 49, and children under 11 years old, should not consume fish, due to potentially toxic levels of mercury (Water Matters, 2010). Alberta's Chief Medical Officer of health, Andre Corriveau, recently advised people to limit the consumption of certain fish from the Twin Valley Reservoir, the Red Deer River near the mouth of the Blindman River, and the South Saskatchewan River at the Blindloss Ferry and at Medicine Hat (Nationtalk, 2009).

## **8.2. Oil Sands Development**

Polycyclic aromatic compounds (PAC) are chemical compounds that occur in tar, coal, and oil deposits, and are produced by the combustion of fuels. PACs are recognized pollutants that possess carcinogenic and mutagenic properties, thus representing a cause

for public health concern. A recent study by Kelly et al. (2010) reported that oil sands upgrading facilities in the Athabasca oil sands deposited approximately 391kg of airborne PAC, and 168kg of dissolved PAC into the surrounding snowpack within 50km. The concentration of PAC in source waters downstream from oil sands development facilities increased 2.5 times in the winter, and 22 times in the summer months (Kelly et al., 2010). The authors believe that PAC concentrations are high enough to pose a toxic threat to fish embryos within the Athabasca river, and its tributaries (Kelly et al., 2010). Landscape disturbance, a well-known component of oil sands development, is further believed to represent a source of PAC (Kelly et al., 2010). The study further criticized the Regional Aquatic Monitoring Program for failing to recognize sources of PAC in their monitoring approaches, and for the lack of complete transparency regarding the acquisition, analyses, and reporting of surveillance data.

In addition, there is considerable concern that oil sands tailing ponds, which serve as containment units for the water that becomes toxic through the bitumen extraction process, pose a threat to groundwater through leaching. These ponds contain PACs, naphthenic acids (known to be both persistent and acutely toxic), and metals in concentrations exceeding those outlined by Canadian guidelines for aquatic life (Environmental Defense, 2008). Currently, there are over 720 billion liters of toxic tailings in the Athabasca oil sands (Alberta Energy Conservation Board, 2008). Seepage has been reported from one of the first constructed tailings ponds, at a rate of 67 liters per second (Alberta Energy Utilities Board, 2008, cited by the Pembina Institute). While both the Alberta Premier and Alberta minister of the Environment claim that tailings seepage has only occurred in old tailings ponds, and that leaked tailings are captured (Alberta Hansard, 2008, cited by Environmental Defense, 2008), the possibility poses a severe threat to source waters.

### **8.3. Fire Retardant Additives: PBDEs**

Increasingly, there are concerns being raised over the presence of fire retardant additives, called polybrominated diphenyl ethers (PBDE) in surface waters, sediments, and soils. Since the 1970s, these compounds have been added to a wide variety of products, such as plastics, foam padding, and textiles (Environment Canada, 2009). Globally, North America is by far the greatest consumer of PBDE products (Hale et al., 2003). Research has demonstrated that these products are persistent, bioaccumulative, and possess toxic effects, specifically effects related to thyroid hormones (Health Canada, 2007). Indeed, rising concentrations of PBDE in human breast milk (Betts, 2002), has led to the ban of one particularly lipophilic commercial PBDE mixture by the European Union (Hale et al., 2003). Despite the concerns surrounding PBDEs, and their high consumption in North America, research on the impacts of these chemicals in the North America environment is limited. The release of PBDEs into the environment can occur during their production, incorporation into various products, or during the disposal and recycling of PBDE products (Danish Environmental Protection Agency, 1999). The physical properties of these compounds suggest their ultimate residence in sediment and soils (Palm et al., 2002), through transport via air and water (Hale et al., 2003). Data on the prevalence of these compounds in Canadian surface waters, soils, and sediments are generally quite limited. PBDEs have been detected in the effluent of Montreal City, in the surface waters of the St. Lawrence River, and the sediments in Lake Sainte-Pierre. Although the concentrations observed in these locations are not believed high enough to pose a risk to human health, the primary concern is the threat of bioaccumulation within the food chain (Environment Canada, 2009). PBDE concentrations have increased 10 times in Arctic mammals, 10-100 times in aquatic birds, 100-1000 times in lake trout in the Great Lakes, and 1000 times in Beluga Wales in the St. Lawrence Estuary, since the 1980s (Health

Canada, 2007). Generally, these compounds are believed to be ubiquitous in the Canadian Environment, and data suggests that their presence is on the rise across Canada (Health Canada, 2007).

## **9. General Conclusions**

Generally, the consensus is that nutrients are the dominant agricultural pollutant of source waters. The ramifications of nutrient loading to aquatic ecosystems and public health are well documented. Excess nitrogen and phosphorus contribute to overall eutrophication, which has an enormous impact on the health of aquatic ecosystems. Nitrite toxicity, through the leaching and runoff of excessive nitrates, and cyanobacterial blooms, generally caused by an excess of phosphorus, pose a definite public health risk. More novel threats, such as the presence of drinking water treatment by-products, which are formed in the presence of excessive plant growth caused by eutrophication, are emerging as well. The development of residual soil nitrogen and phosphorus soil tests, in addition to the index for the risk of phosphorus and nitrogen water contamination, are instrumental in monitoring, surveillance, and risk assessment practices.

Pesticides, like nutrients, are also an endemic agricultural threat to source waters. However, because pesticides and commercial fertilizers are manufactured, there exists the capacity for licensing, thus providing a regulatory measure to help manage and control the introduction of these pollutants into the environment. The toxicity, solubility, and persistence of pesticides are assessed before being licensed for use. Despite the government's ability to control the use of specific pesticides in Alberta, pesticides are designed to disrupt biological process in pests, and thus can pose significant threats to non-target species and public health. Further, licensing a pesticide does not guarantee the

development of the appropriate water quality guidelines, as indicated by the presence of numerous pesticides frequently found in Alberta's source waters, without guidelines.

Sediment quality is intertwined with the overlying water quality, as sediments have the capacity to serve as a transport vehicle for a wide variety of contaminants including pesticides, phosphorus, and heavy metals. Additionally sediments themselves can impair source waters through increasing the costs of drinking water treatment, and affecting ecosystem dynamics through affecting sunlight penetration via sediment suspension in the water column. While sedimentation is a natural process, it can be accelerated through certain land-use practices, including agriculture.

The dominance of the livestock industry in southern Alberta, which boasts the highest density of confined feedlot operations in Canada, warrants extreme vigilance on the potential for livestock wastes to contaminate source waters. In addition to possessing nutrients that contribute to ecosystem degradation and cause concern for public health, livestock wastes are a source of fecal coliform bacteria, including strains of *E.coli* that pose severe risks to human health. Indeed, Alberta is frequently cited as having one of the highest rates of *E.coli* infection in Canada (Public Health Agency of Canada, 2007). Livestock wastes also may bear infective stages of protozoan parasites like Giardia and Cryptosporidium. The infective stages of these parasites can produce severe and lasting health consequences upon ingestion by humans. Their ability to resist the traditional disinfection practices employed by drinking water treatment plants, the low doses required for infection, and capacity to remain viable in extreme conditions and in open waters for long periods of time makes these parasites a significant public health concern. The challenge of protozoan parasite disinfection in particular emphasizes the importance of the preventative aspects of source water protection. The severity of these diseases warrants

close examination of molecular techniques that can be used to assess the sources of these infective stages of parasites. Recent molecular advances have determined that only certain genotypes of *Giardia*, and certain species of *Cryptosporidium* can be transmitted from cattle to humans. Evidence based on a limited number of prevalence and genotyping studies indicate that in Alberta, cattle may not be as significant a source of human strains of *Giardia* and *Cryptosporidium* as previously believed. Incorporating molecular techniques into standard monitoring and surveillance practices will play a significant role in developing accurate risk assessment programs.

Recently, media attention has focused on the wide variety of manufactured and naturally produced compounds that can disrupt the endocrine system of animals. Thus, the presence of these compounds in source waters represents an emerging public health concern, as these compounds are known to possess carcinogenic, immunotoxic, and neurotoxic properties. The relative potency of natural estrogens and synthetic estrogens, compared to those manufactured compounds with endocrine disrupting capabilities, warrants concern. As it relates to agriculture, livestock wastes represent a significant source of estrogen and testosterone. Synthetic hormones that are injected into cattle as growth products, in addition to naturally produced hormones, are excreted in the feces. Through soil erosion processes, these hormones can reach source waters. Research has demonstrated that agricultural effluent has the capacity to impair reproductive abilities of aquatic biota.

Generally, agricultural pollutants are a significant impairment of source waters, as nonpoint sources of pollution are notoriously difficult to monitor, manage, and control. Many agricultural pollutants, including those incorporated in livestock wastes such as nutrients, pathogens, and steroidal hormones, are best controlled through controlling soil

erosion. The challenge then lies in finding ways to encourage the adoption of practices that are known to reduce soil erosion, and additional practices that will lessen the likelihood of pollutants accessing source waters. Additional threats to source water are constantly emerging. Recently, Alberta Health and Wellness recommended restrictions on fish consumption in various locations in Southern Alberta, due to the bioaccumulation of organic mercury in the muscle tissues of certain fish. The Athabasca oil sands are under constant media scrutiny due to the potential of by-product contaminants, namely naphthenic acids and polycyclic aromatic compounds, to infiltrate ground and surface waters, which poses an enormous threat to the Athabasca watershed. Finally, certain additives used in fire retardants for a wide variety of consumer products, of which North America is the largest global consumer, are being detected in surface waters across Canada.

Thus, the proactive measure of controlling and minimizing the contamination of source water by pollutants through source water protection is vital to both public health and healthy ecosystems. Alberta Environment employs a “source-to-tap, multi-barrier approach” to drinking water. This policy engages a variety of facets, including legislation, drinking water systems, knowledge and awareness, performance assurance, and protection, to create a safe secure drinking water source for Albertans. It is clear that a significant challenge to source water protection is the ability to be readily adaptable to acknowledge and incorporate the continuously developing scientific research related to both “traditionally recognized” water pollutants, and emerging threats to aquatic ecosystems and public health.

## References

- Abernathy, A.R. & Cumbie, P.M. (1977). Mercury accumulation by largemouth bass (*Micropterus salmoides*) in recently impounded reservoirs. *Bulletin of Environmental Contamination and Toxicology*, 17, 595–602.
- Akerblom, N. (2004). Agricultural pesticide toxicity to aquatic organisms: a literature review. Uppsala, Department of Swedish Environmental Assessment, Swedish University of Agricultural Sciences.
- Akerblom, N. (2007). Bioavailability of Pesticides in Freshwater Sediments: the importance of sorption and uptake routes (Doctoral dissertation). Swedish University of Agricultural Sciences, Uppsala, Sweden.
- Alavanja, M.C.R., Hoppin, J.A., & Kamel, F. (2004). Health effects of chronic pesticide exposure: cancer and neurotoxicity. *Annual Review of Public Health*, 25, 155-197.
- Alberta Agriculture and Rural Development (2009a). Agriculture Statistics Factsheet 2009. Retrieved on October 12<sup>th</sup>, 2009, from [www1.agric.gov.ab.ca/\\$department/deptdocs.nsf/all/sdd12807](http://www1.agric.gov.ab.ca/$department/deptdocs.nsf/all/sdd12807)
- Alberta Agriculture and Rural Development. (2009b). A primer on Water Quality: impact of Crop production practices on water quality. Retrieved on October 13<sup>th</sup>, 2009, from [www1.agric.gov.ab.ca/\\$department/deptdocs.nsf/all/wat3348](http://www1.agric.gov.ab.ca/$department/deptdocs.nsf/all/wat3348)
- Alberta Energy Resources Conservation Board. (2008, June 26<sup>th</sup>). ERCB releases draft directive on oil sands tailings management and enforcement criteria. Retrieved on February 20<sup>th</sup>, 2009, from [www.ercb.ca/docs/new/newsrel/2008/nr2008-14.pdf](http://www.ercb.ca/docs/new/newsrel/2008/nr2008-14.pdf)
- Alberta Environment (2007). Water for Life. Healthy Aquatic Ecosystems. Summary Report on the initial assessment of ecological health of aquatic ecosystems in Alberta: Water Quality, Sediment Quality, and Non-fish biota. Retrieved on November 18<sup>th</sup>, 2009, from <http://environment.gov.ab.ca/info/posting.asp?assetid=7860&categoryid=5>
- Alberta Environment (2001). Ecological relevance of pesticide residues in Alberta Surface Waters: an evaluation based on toxicity testing. Retrieved on January 28<sup>th</sup>, 2010, from [www.environment.gov.ab.ca/info/library/6785.pdf](http://www.environment.gov.ab.ca/info/library/6785.pdf)
- Alberta Environment. Alberta River Water Quality Index. Retrieved on November 2<sup>nd</sup> 2009, from [www.environment.alberta.ca/1777.html](http://www.environment.alberta.ca/1777.html)
- Alberta Environment (2005). Pesticides in Surface Waters: Facts at your fingertips. Retrieved on January 28<sup>th</sup>, 2010, from [http://www3.gov.ab.ca/env/water/swq/assets/Pesticides\\_SurfaceWaterFactSheet.pdf](http://www3.gov.ab.ca/env/water/swq/assets/Pesticides_SurfaceWaterFactSheet.pdf).
- Alberta Hansard, May 27, May 28, and June 4, 2008.

- Alberta Health and Wellness (2008a). Public Health Notifiable Disease Management Guidelines – Cryptosporidiosis. Retrieved on September 22<sup>nd</sup>, 2009, from [www.health.alberta.ca/documents/ND-Cryptosporidiosis.pdf](http://www.health.alberta.ca/documents/ND-Cryptosporidiosis.pdf)
- Alberta Health and Wellness. (2008b). Public Health Notifiable Disease Management Guidelines – Giardiasis. Retrieved on September 22<sup>nd</sup>, 2009, from [www.health.alberta.ca/documents/ND-Giardiasis.pdf](http://www.health.alberta.ca/documents/ND-Giardiasis.pdf)
- Alm E.W., Burke J., & Spain A. (2003). Fecal indicator bacteria are abundant in wet sand at freshwater beaches. *Water Research*, 37, 3978–3982.
- Andersen, H., Siegrist, H., Halling-Sørensen, & B., Ternes, T.A. (2003). Fate of estrogens in a municipal sewage treatment plant. *Environmental Science Technology*, 37, 4021–4026.
- Anderson, A.M. (2005). Review of pesticide data in Alberta surface waters since 1995. Environmental Monitoring and Evaluation Branch, Alberta Environment.
- Anderson, A.M., Saffran, K.A., & Byrtus, G. (1997). Pesticides in Alberta surface waters. Prepared for CAESA, Alberta Environment.
- Anderson, A.M. (2008). Development of an aquatic pesticide toxicity index for use in Alberta. Alberta Environment, Environmental insurance.
- Anderson, T.D., & Lydy, M.J. (2002). Increased toxicity to invertebrates associated with a mixture of atrazine and organophosphate insecticides. *Environmental Toxicology And Chemistry*, 21, 1507-1514.
- Andraski, T.W., & Bundy, L.G. (2003). Relationships between phosphorus levels in soil and in runoff from corn production systems. *Journal of Environmental Quality*, 32, 310–316.
- Ankley, G.T., Jensen, K.M., Makynen, E.A., Kahl, M.D., Korte, J.J., Hornung, M.W., Henry, T.R., Denny, J.S., Leino, R.L., Wilson, V.S., Cardon, M.D., Hartig, P.C., & Gray, L.E. (2003). Effects of the androgenic growth promoter 17 $\beta$ -trenbolone on fecundity and reproductive endocrinology of the fathead minnow. *Environmental Toxicology and Chemistry*, 22, 1350–1360.
- Appelbee A.J., Frederick L.M., Heitman T.L., & Olson, M.E. (2003). Prevalence and genotyping of *Giardia duodenalis* from beef calves in Alberta, Canada. *Veterinary Parasitology*, 112, 289–94.
- Arcand-Hoy, L.D., Minrod, A.C., & Benson, W.H. (1998). Endocrine-modulating substances in the Environment; Estrogenic effects of pharmaceutical products. *International Journal of Toxicology*, 17, 139-158.

- Arnold, S.D., & Meister, E.A. (1999). Dairy feedlot contributions to groundwater contamination: A preliminary study in New Mexico. *Journal of Environmental Health*, 62, 16–19.
- Arnold, S. F., Klotz, D. M., Collins, B. M., Vonier, P. M., Guillette, L. J., Jr., & McLachlan, J. A. (1996). Synergistic activation of estrogen receptor with combinations of environmental chemicals. *Science*, 272, 1489-1491.
- Arnon, S., Ofer, D., Elhananni, S., Cohen, K., Pankratov, I., Gross, A. (2008). Transport of testosterone and estrogen from dairy farms waste lagoons to groundwater. *Environmental Science Technology*, 42, 5521-5526.
- Atwill, E.R., Hou, L., Karle, B.M., Harter, T., Tate, K.W., & Dahlgren, R.A. (2002). Transport of *Cryptosporidium parvum* oocysts through vegetated buffer strips and estimated filtration efficiency. *Applied Environmental Microbiology*, 68, 5517-5527.
- Bailey H.C., Miller J.L., Miller M.J., Wiborg L.C., Deanovic L., & Shed T. (1997). Joint acute toxicity of diazinon and chlorpyrifos to *Ceriodaphnia dubia*. *Environmental Toxicology and Chemistry*, 16, 2304-2308.
- Baronti, C., Curini, R., D'Ascenzo, G., Di Corcia, A., Gentili, A., & Samperi, R. (2000). Monitoring natural and synthetic estrogens at activated sludge sewage treatment plants and its receiving river water. *Environmental Science and Technology*, 34, 5059–5066.
- Beaudet, M.P., Walop, W., Le Petit, C. (1997). Women on Hormone Replacement Therapy. *Health Reports*, 9, 9-18.
- Belden J.B., & Lydy M.J. (2000). Impact of atrazine on organophosphate insecticide toxicity. *Environmental Toxicology and Chemistry*, 19, 2266-2274.
- Belfroid, A., Van der Horst, A., Vethaak, A., Schafer, A., Rijs, G., Wegener J., & Cofino, W. (1999). Analysis and occurrence of estrogenic hormones and their glucuronides in surface water and waste water in The Netherlands. *Science of the Total Environment*, 225, 101-108.
- Bell, S.G. & Codd, G.A. (1994). Cyanobacterial toxins and human health. *Reviews in Medical Microbiology*, 5, 256-264.
- Betts K.S. (2002). Science news: rapidly rising PBDE levels in North America. *Environmental Science and Technology*, 36, 50A–52A.
- Bjerrgaard, P., Hansen, P.R., Larsen, K.J., Erratico, C., Korsgaard, B., Holbech, H. (2008). Vitellogenin as a biomarker for estrogenic effects in brown trout, *Salmo trutta*: laboratory and field investigations. *Environmental Toxicology and Chemistry*, 27, 2387-96.

- Bochove, E., Thériault, G., Dechmi, F., Rousseau, A.N., Quilbé, R., Leclerc, M. L., & Goussard, N. (2006). Indicator of risk of water contamination by phosphorus from Canadian agricultural land. *Water Science & Technology* 53, 303–310.
- Bodaly, R. A., Jansen, W.A., Majewski, A.R., Fudge, R.J.P., Strange, N.E., Derksen, A.J., & Green, D.J. (2007). Post-impoundment time course of increased mercury concentrations in fish in hydroelectric reservoirs of northern Manitoba, Canada. *Archives of Environmental Contamination and Toxicology*, 53, 379–389.
- Brinkmann, L., & Rasmussen, J.B. (2010). High levels of mercury in biota of a new Prairie irrigation reservoir with a simplified food web in Southern Alberta, Canada. *Hydrobiologia*, 641, 11-21.
- Bryan, G.W., & Langston, W.J. (1992). Bioavailability, accumulation and effects of heavy metals in sediments with special reference to United Kingdom estuaries: a review. *Environmental Pollution*, 76, 89-131.
- Burkart, M.R. & J.D. Stoner. (2001). Nitrogen in groundwater associated with agricultural systems. In J. L. Hatfield & R. F. Follett (Eds.), *Nitrogen in the Environment: Sources, Problems, and Management*, Second edition (123-145). Elsevier: New York.
- Caccio, S.M. Thompson, R.C.A., McLauchlin, J., & Smith, H.V. (2005) Unravelling *Cryptosporidium* and *Giardia* epidemiology. *Trends in Parasitology*. 21, 430–437.
- Calamari, D., & Barg, U. (1993). Hazard assessment of agricultural chemicals by simple simulation models. In *Prevention of Water Pollution by Agriculture and Related Activities. Proceedings of the FAO expert consultation*, Santiago, Chile, 20-23 Oct. 1992. Water report. FAO, Rome.
- Callantine MR, Stob M, & Andrews FN. (1961). Faecal elimination of estrogens by cattle treated with diethylstilbestrol and hexestrol. *American Journal of Veterinary Research*, 22, 462–465.
- Camargo, J.A., & Alonso, A. (2006). Ecological and toxicological effects of inorganic nitrogen pollution in aquatic ecosystems: A global assessment. *Environment International* 32, 831–849.
- Campbell, I., Tzipori, A.S., Hutchison, G., & Angus, K.W. (1982). Effect of disinfectants on survival of *Cryptosporidium* oocysts. *The Veterinary Record*, 111, 414–415.
- Canada Alberta Environmentally Sustainable Agriculture Agreement. (1998). Agricultural impacts on water quality in Alberta. An Initial Assessment. Canada – Alberta Environmentally Sustainable Water Quality Committee, Alberta Agriculture Food and Rural Development.
- Carey, C.M., Lee, H., & Trevors, J.T. (2004). Biology, persistence and detection of *Cryptosporidium arum* and *Cryptosporidium* oocyst. *Water research*, 38, 818-862.

Casey, F.X.M., Larsen, G.L. Hakk, H., & Šimunek. J. (2003). Fate and Transport of 17 $\beta$ -Estradiol in Soil-Water Systems. *Environmental Science and Technology*, 37, 2400–2409.

Casey, F. X. M. Simunek, J. Lee, J. Larsen, G. L. & Hakk, H. (2005). Sorption, mobility, and transformation of estrogenic hormones in natural soil. *Journal of Environmental Quality*, 34, 1372–1379.

Chalmers, R.M. Ferguson,C., Caccio, S., Gasser, R.B., Abs El-Osta, Y.G., Hijnen, L., Xiao, L.,Elwin, K., Hadfield, S., Sinclair, M., Steens, M. (2005). Direct comparison of selected methods for genetic categorization of *Cryptosporidium parvum* and *Cryptosporidium hominis* species. *International Journal of Parasitology*, 35, 397-410.

Chambers, P.A., Guy, M., Roberts, E., Charlton, M.N., Kent, R., Gagnon, C. Grove, G., Foster, N., DeKimpe, C., and Giddings, M. (2008). National Water Research Institute. Threats to Sources of Drinking Water and Aquatic Ecosystem Health in Canada: Nutrients-Nitrogen and Phosphorus. Prepared for Environment Canada. Retrieved on January 6<sup>th</sup>, 2010, from <http://www.ec.gc.ca/inre-nwri/default.asp?lang=En&n=235D11EB-1&offset=7&toc=show>

Chang, C., and Entz, T. (1996). Nitrate leaching losses under repeated cattle feedlot manure applications in southern Alberta. *Journal of Environmental Quality*, 25, 145–153.

Cheremisinoff, N.P. 2002. An Overview of Water and Wastewater Treatment. In Handbook of Water and Wastewater Treatment Technologies. Butterworth-Heinemann, Woburn, MA.

Choi S.M, Yoo, S.D., & Lee B.M. (2004). Toxicological characteristics of endocrine-disrupting chemicals: developmental toxicity, carcinogenicity, and mutagenicity. *Journal of Toxicology and Environmental Health B Critical Reveiws* 1, 1-32.

Coklin, T., Farber, J., Parrington, L., & Dixon, B. (2007). Prevalence and molecular characterization of *Giardia duodenalis* and *Cryptosporidium* spp. in dairy cattle in Ontario, Canada. *Veterinary Parasitology*, 150, 297-305.

Colborn, T., F.S. vom Saal, & A.M. Soto. (1993). Developmental effects of endocrine-disrupting chemicals in wildlife and humans. *Environmental Health Perspectives*, 101, 378-384.

Colucci, M. S., Bork, H. & Topp, E. (2001). Persistence of estrogenic hormones in agricultural soils: I. 17 $\beta$ -Estradiol and estrone. *Journal of Environmental Quality* 30, 2070–2076.

Colucci, M.S., & Topp, E. (2001). Persistence of Estrogenic Hormones in Agricultural Soils. *Journal of Environmental Quality*, 30, 2077-2080.

- Comfort, S.D., Franti, T.G., & Smith, S.K. (1996). Pesticide Runoff and Water Quality in Nebraska. Nebraska Cooperative Extension, U.S. department of Agriculture. Retrieved on January 8<sup>th</sup>, 2010, from [www.stopaptrail.org/images/summaryofadsorption.pdf](http://www.stopaptrail.org/images/summaryofadsorption.pdf)
- Coote, D.R., & F.R. Hore. (1979). Contamination of shallow ground- water by an unpaved feedlot. *Canadian Journal of Soil Science*, 59, 401–412.
- Correll, D. (1998). The role of Phosphorus in the eutrophication of receiving waters: a review. *Journal of Environmental Quality*, 27, 261-26.
- Craig, D.L., Fallowfield, H.J., Cromar, N.J. (2002). Enumeration of faecal coliforms from recreational coastal sites: evaluation of techniques for the separation of bacteria from sediments. *Journal of Applied Microbiology*, 93, 557-565.
- Danish Environmental Protection Agency. (1999). Brominated flame retardants: substance flow analysis and assessment of alternatives.
- Daughton, C.G., & Ternes, T.A. (1999). Pharmaceuticals and personal care products in the environment: Agents of subtle change? *Environmental Health Perspectives*, 107, 907–938.
- Daverede, I.C., Kravchenko, A.N., Hoeft, R.G., Nafziger, E.D., Bullock, D.G., Warren, J.J., & Gonzini, L.C. (2003). Phosphorus runoff: Effect of tillage and soil phosphorus levels. *Journal of Environmental Quality*, 32, 1436–1444.
- Dean-Nystrom, E.A., Bosworth, B.T., O'Brien, A.D., & Moon, H.W. (1999). Bovine infection with *Escherichia coli* O157:H7. In C.S. Stewart & H.J. Flint. (Eds.) *Escherichia coli* O157:H7 in farm animals (pp. 51-58) New York: CABI Publishing.
- De Jong, R., Drury, C.F., Yang, J.Y., & Campbell, C.A. (2009). *Journal of Environmental Management* 90, 3169-3181.
- De Jong, R., Yang, J.Y., Drury, C.F., Huffman, E.C., Kirkwood, V., & Yang, X.M. (2005). Indicator of Water Contamination by Nitrogen (IROWC-N). In A. Lefebvre, W. Weilers, & B. Chunn, B. (Eds.) *Environmental Sustainability of Canadian Agriculture: Agri-Environmental Indicator Report Series - Report #2*. (pp. 124-130), Agriculture and Agri-Food Canada/Agriculture et Agroalimentaire Canada, Ottawa, ON.
- deRegnier, D.P., Cole, L., Schupp, D.G., & Erlandsen, S.L. (1989). Viability of *Giardia* cysts suspended in lake, river, and tap water. *Applied Environmental Microbiology*, 55, 1223–1229.
- Desbrow C., Routledge E.J., Brighty G.C., Sumpter J.P., & Waldock M. (1998). Identification of estrogenic chemicals in STW effluent: 1. Chemical fractionation and in vitro biological screening. *Environmental Science and Technology*, 32, 1549–58.

Durhan, E. J., Lambright, C. S., Makynen, E. A., Lazorchak, J., Hartig, P. C., Wilson, V. S., Gray, L. E., & Ankley, G. T. (2006). Identification of metabolites of trenbolone acetate in androgenic runoff from a beef feedlot. *Environmental Health Perspectives*, 114, 65-68.

Dyer, A. R., Raman, D. R., Mullen, M. D., Burns, R. T., Moody, L. B., Layton, A. C., & Sayler, G. S. (2001). ASAE Meeting Paper No. 01- 2107; ASAE: St. Joseph, MI.

Dyer, S. (2009, Sept 22). Environmental Impacts of Oil Sands Development in Alberta. The Oil Drum. Retrieved from <http://www.energybulletin.net/node/50186>

Elder, R.O., Keen, J.E., Siragusa, G.R., Barkocy-Gallagher, G.A., Koohmaraie, M., & Laegreid, W.W. (2000). Correlation of enterohemorrhagic *Escherichia coli* O157 prevalence in feces, hides, and carcasses of beef cattle during processing. *Proceedings of the National Academy of Sciences*, 97, 2999–3003.

Environmental Defense. (2008). The Tar Sands leaking legacy. Retrieved on February 20<sup>th</sup>, 2010, from [www.environmentaldefence.ca/reports/pdf/TailingsReport\\_FinalDec8.pdf](http://www.environmentaldefence.ca/reports/pdf/TailingsReport_FinalDec8.pdf)

Environment Canada. (2009). Tracking Polybrominated Diphenyl Ethers (PBDE): New Chemical Contaminants in the Environment. Retrieved on February 20<sup>th</sup>, from [www.ec.gc.ca/eau-water/default.asp?lang=En&n=7BBC611F-1](http://www.ec.gc.ca/eau-water/default.asp?lang=En&n=7BBC611F-1)

Erb, R.E., Chew, B.P., & Keller, H.F. (1977). Relative concentrations of estrogen and progesterone in milk and bold, and excretion of estrogen in urine. *Journal of Animal Sciences*, 46, 617–626.

Erlandsen, S.L. (1994) Biotic transmission -- Is giardiasis a zoonosis? In R.C.A. Thompson, J.A. Reynoldson, & A.J. Lymbery, (Eds.), *Giardia: From Molecules to Disease*, (pp. 83–97). Cambridge: CAB International, University Press.

Exner, M.E., and R.F. Spalding. (1994). N-15 identification of nonpoint sources of nitrate contamination beneath cropland in the Nebraska Panhandle: Two case studies. *Applied Geochemistry*, 9, 73–81.

Facemire, C.F., T.S. Gross, & Guillette, L.J. Jr. (1995). Reproductive impairment in the Florida Panther: Nature or nurture? *Environmental Health Perspectives*, 103, 79-86.

Fahl, W.E., & Rose, D.P. (1975). Effect of estrogen-containing oral contraceptives on urinary corticosteroid sulfate extraction. *Clinica Chimica Acta*, 63, 189-192.

Fayer, R., Morgan, U., Upton, S.J. (2000). Epidemiology of *Cryptosporidium*: transmission, detection, and identification. *International Journal for Parasitology*, 30, 1305-1322.

Fayer, R. Santin, M., Trout, J.M., & Ellis, G. (2006). Prevalence of species and genotypes of *Cryptosporidium* found in 1-2-year-old dairy cattle in the eastern United States. *Veterinary Parasitology*, 135, 105–112.

Feltus, D.C., Giddings, C.W., Schneck, B.L., Monson, T., Warshauer, D., & McEvoy, J.M. (2006). Evidence Supporting Zoonotic Transmission of *Cryptosporidium* spp. in Wisconsin. *Journal of Clinical Microbiology*, 44, 4303-4308.

Fewtrell L. (2004). Drinking-water nitrate, methemoglobinemia, and global burden of disease: a discussion. *Environmental Health Perspectives*, 112, 1371–1374.

Fine, D.D., Breidenbach, P.G., Price, T.L., & Hutchins, S.R. (2003). Quantitation of estrogens in ground water and swine lagoon samples using solid-phase extraction, pentafluorobenzyl/trimethylsilyl derivatizations and gas chromatography–negative ion chemical ionization tandem mass spectrometry. *Journal of Chromatography A*, 1017, 167-185.

Finlay-Moore, O., Hartel, P. G., & Cabrera, M. L. (2000). 17 $\beta$ -estradiol and testosterone in soil and runoff from grasslands amended with broiler litter. *Journal of Environmental Quality*, 29, 1604-1611.

Folmar, L. C., Denslow, N. C., Rao, V., Chow, M., Crain, D. A., Enblom, J., Marcino, J., & Guilette, Jr., L. J. (1996). Vitellogenin induction and reduced serum testosterone concentrations in feral male carp (*Cyprinus carpio*) captured near a major metropolitan sewage treatment plant. *Environmental Health Perspectives*, 104, 1096-1101.

Forstner, U. & Owens, P.N. (2007). Sediment Quantity and Quality Issues in River Basins. In B. Westrich & U. Forstner (Eds.) *Sediment Dynamics and Pollutant Mobility in Rivers: An Interdisciplinary Approach*. New York: Springer.

Fry, D.M. (1995). Reproductive effects in birds exposed to pesticides and industrial chemicals. *Environmental Health Perspectives*, 103, 165–171.

Furuichi, T., Kannan, K., Suzuki, K., Tanaka, S., Giesy, J.P., & Masunaga, S. (2006). Occurrence of Estrogenic Compounds in and Removal by a Swine Farm Waste treatment Plant. *Environmental Science and Technology*, 40, 7896–7902.

Gannon, V.P.J., Duke, G.D., Thomas J.E., VanLeeuwen, J., Byrne, J., Johnson, D., Kienzle, S.W., Little, J., Graham, T., & Selinger, B. (2005). Use of in-stream reservoirs to reduce bacterial contamination of rural watersheds. *Science of the Total Environment*, 348, 19-31.

Gannon, V.P.J., Graham, T.A., Read, S., Ziebell, K., Muckle, A., Mori, J., Thomas, J. Selinger, B., Townshend, I., & Byrne, J. (2004). Bacterial Pathogens in Rural Water Supplies in Southern Alberta, Canada. *Journal of Toxicology and Environmental Health, Part A*, 67, 1643–1653.

Gardner, T.B., & Hill, D.R. (2001). Treatment of Giardiasis. *Clinical Microbiological Review*, 14, 114-128.

Gearheart RA. (1999). The use of free surface constructed wetland as an alternative

process treatment to meet unrestricted water reclamation standards. *Water Science Technology*, 40, 375–382.

Gennaccaro, A.L., McLaughlin, M.R., Quintero-Betancourt, W., Huffman, D.E. & Rose, J.B. (2003). Infectious *Cryptosporidium parvum* oocysts in final reclaimed effluent. *Applied Environmental Microbiology*, 69, 4983–4984.

Geurden T., Geldhof P., Levecke B., Martens C. Berkvens D., Casaert S., Vercruyse J., & Claerebout, E. (2008). Mixed *Giardia duodenalis* assemblage A and E infections in calves. *International Journal for Parasitology*, 38, 259-264.

Giesy, J.P., Rosiu, C.J., Graney, R.L., & Henry, M.G. (1990). Benthic invertebrate bioassays with toxic sediment and pore water. *Environmental Toxicology and Chemistry*, 9, 233-248.

Gilliom, R.J. (2007). Pesticides in U.S. streams and groundwater. *Environmental Science and Technology*, 41, 3407-3413.

Giullette, L.J. Jr., Gross, T.S., Masson, G.R., Matter, J.M., Percival, H.F., & Woodward, A.R. (1994). Developmental abnormalities of the gonad and abnormal sex hormone concentrations in juvenile alligators from contaminated and control lakes in Florida. *Environmental Health Perspectives*, 102, 680-688.

Goulet R.R., Pick F.R., & Droste R.L. (2005). Test of the first-order removal model for metal retention in a young constructed wetland. *Ecological Engineering*, 17, 357–371.

Greer, F.R., & Shannon, M. (2005). Infant Methemoglobinemia: The Role of Dietary Nitrate in Food and Water. *Guidance for the Clinician in Rendering Pediatric Care Pediatrics*, 116, 784-786.

Griffin, P. M., & Tauxe, R. V. (1991). The epidemiology of infections caused by *Escherichia coli* O157:H7 and other enterohemorrhagic *E. coli*, and the associated hemolytic uremic syndrome. *Epidemiology Reviews*, 13, 60–98.

Gow S., & Waldner C. (2006). An examination of the prevalence of and risk factors for shedding of *Cryptosporidium* spp. and *Giardia* spp. in cows and calves from western Canadian cow calf herds. *Veterinary Parasitology*, 137, 50–61.

Guy, M.A., McFadden, T.B., Cockrell, D.C., & Besser, T. E. (1994). Regulation of colostrum formation in beef and dairy cows. *Journal of Dairy Science*, 77, 3002–3007.

Haas, C.N. & Rose, J.B. (1994) Reconciliation of microbial risk models and outbreak epidemiology: The case of the Milwaukee outbreak. In: *Proceedings of the Annual American Water Works Association Conference, Denver, CO*. (pp. 517-523).

- Hale, R.C., Alaei, M., Manchester-Neesvig, J.B., Stapleton, H.M., & Ikonou, M.G. (2003). Polybrominated diphenyl ether flame retardants in the North American Environment. *Environment International*, 29, 771-779.
- Hall, B.D., Rosenberg, D.M., & Wiens, A.P. (1998). Methyl mercury in aquatic insect from an experimental reservoir. *Canadian Journal of Fisheries and Aquatic Sciences*, 55, 2036–2047.
- Hanselman, T.A., Graetz, D.A., & Wilkie, A.C. (2003). Manure-borne estrogens as potential environmental contaminants: a review. *Environmental Science and Technology*, 37, 5471-5478
- Hansen P.D., Dizer H., Hock B., Marx A., Sherry J., McMaster M., & Blaise, C. (1998). Vitellogenin—a biomarker for endocrine disruptors. *Trends in Analytical Chemistry*, 17, 448–451.
- Hansson, T. Schiedek, D., Lehtonen, K.K., Vuorinen, P.J., Liewenborg, B., Noaksson, E., Tjarnlund, U., Hanson, M., & Balk, L. (2006). Biochemical biomarkers in adult female perch (*Perca fluviatilis*) in a chronically polluted gradient in the Stockholm recipient (Sweden). *Marine Pollution Bulletin*, 53, 451-468.
- Harp, J.A. & Goff, J.P. (1995). Protection of calves with a vaccine against *Cryptosporidium parvum*. *Journal of Parasitology*, 81, 54-57.
- Harp, J.A., Woodmansee, D.B., & Moon, H.W. (1990). Resistance of calves to *Cryptosporidium parvum*: effects of age and previous exposure. *Infection and Immunity*, 58, 2237–2240.
- Harries, J.E., Sheahan, D.A., Jobling, S., Matthiessen, P., Neall, P., Routledge, E.J., Rycroft, R., Sumpter, J.P., & Taylor, T. (1996). A survey of estrogenic activity in United Kingdom inland waters. *Environmental Toxicology and Chemistry*, 15, 1993-2002.
- Harries J.E., Sheahan D.A., Jobling S., Matthiessen P., Neall P., Sumpter J.P., Taylor, T., & Zaman, N. (1997). Estrogenic activity in five United Kingdom rivers detected by measurement of vitellogenesis in caged male trout. *Environmental Toxicology and Chemistry*, 16, 534–542.
- Harrison, P.T.C., Holmes, P., Humfrey, C.D.N. (1997). Reproductive health in humans and wildlife: are adverse trends associated with environmental chemical exposure? *Science of the Total Environment*, 205, 97-106.
- Hayes T., Haston K., Tsui M., Hoang A., Haeffele C., & Vonk A. (2002) Atrazine-induced hermaphroditism at 0.1 ppb in American frogs (*Rana pipiens*): Laboratory and field evidence. *Environmental Health Perspectives*, 111, 568–575.

Health Canada. (2005). Drugs and Health Products, Veterinary Drugs: Hormonal Growth Promoters. Retrieved on December 6<sup>th</sup>, 2009, from [http://www.hc-sc.gc.ca/dhp-mpps/vet/faq/growth\\_hormones\\_promoters\\_croissance\\_hormonaux\\_stimulateurs-eng.php](http://www.hc-sc.gc.ca/dhp-mpps/vet/faq/growth_hormones_promoters_croissance_hormonaux_stimulateurs-eng.php)

Health Canada (2009). Environmental and Workplace Health. Trihalomethanes. Retrieved on January 12<sup>th</sup>, 2010, from [www.hc-sc.gc.ca/ewh-semt/pubs/water-eau/trihalomethanes/guide-eng.php](http://www.hc-sc.gc.ca/ewh-semt/pubs/water-eau/trihalomethanes/guide-eng.php)

Health Canada. Guidelines for Canadian Drinking Water Quality: Appendix 4.1 Retrieved on November 5<sup>th</sup>, 2009, from [www.env.gov.nl.ca/env/sourcetotap/.../FMT-Appendix4-1.PDF](http://www.env.gov.nl.ca/env/sourcetotap/.../FMT-Appendix4-1.PDF)

Health Canada (2007). Science and Research. Impact of polybrominated diphenyl ethers on the Canadian environment and the health of Canadians. Retrieved on February 20<sup>th</sup>, from <http://www.hc-sc.gc.ca/sr-sr/finance/tsri-irst/proj/persist-org/tsri-237-eng.php>

Heitman, T.L., Frederick, L.M., Viste, J.R., Guselle, N.J., Morgan, U.M., Thompson, R.C.A., & Olson, M.E. (2002). Prevalence of *Giardia* and *Cryptosporidium* and characterization of *Cryptosporidium* spp. isolated from wildlife, human, and agricultural sources in the North Saskatchewan River Basin in Alberta, Canada. *Canadian Journal of Microbiology*, 48, 530-541.

Herbel, M.J., & Spalding, R.F. (1993). Vadose zone fertilizer-derived nitrate and 15N extracts. *Ground Water*, 31, 376–382.

Hoar, B.R., Atwill, E.R., Elmi, C., & Farver, T.B. (2001). An examination of risk factors associated with beef cattle shedding pathogens of potential zoonotic concern. *Epidemiology and Infection*, 127, 147–155.

Holthaus, K.I.E., Johnson, A.C., Jürgens, M.D., Williams, R.J. & Carter, J.E. (2002). The potential for estradiol and ethinylestradiol to adsorb to suspended and bed-sediments in some English rivers. *Environmental Toxicology and Chemistry*, 21, 2526-2535

Hooda, P.S., Edwards, A.C., Anderson, H.A., & Miller, A. (2000). A review of water quality concerns in livestock farming areas. *Science of the Total Environment*, 250, 143–167.

House, W.A., Jickells, T.D., Edwards, A.C., Praska, K.E., & Denison, F.H. (1998). Reactions of phosphorus with sediment in fresh and marine waters. *Soil Use and Management*, 14, 139-146.

Hoque, M.E., Hope, V.T., Kjellstrom, T., Scragg, R., & Lay-Yee, R. (2002). Risk of Giardiasis in Aucklanders: a case-control study. *International Journal of Infectious Diseases*, 6, 191–197.

Hrudey, S.E., Payment, P., P.M. Huck, P.M., R.W. Gillham, R.W., & Hrudey, E.J. (2003). A fatal waterborne disease epidemic in Walkerton, Ontario: comparison with other waterborne outbreaks in the developed world. *Water and Science Technology*, 47, 7-14.

- Hudault S., Guignot J., Servin A.L. (2001). *Escherichia coli* strains colonising the gastrointestinal tract protect germfree mice against *Salmonella typhimurium* infection. *Gut*, 49, 47–55.
- Hunter, P.R., Colford, J.M., Lechevallier, M.W., Binder, S., & Berger PS. (2002). Waterborne diseases. *Emerging Infectious Diseases*, 7S, 544-545.
- Hyland, R., Byrne, J., Selinger, B., Graham, T., Thomas, J., Twnshed, I., & Gannon, V. (2003). Spatial and Temporal distribution of fecal indicator bacteria within the Oldman River Basin of Southern Alberta, Canada. *Water Quality Research Journal of Canada* 38,15-32.
- Irwin, L.K., Gray, S., & Oberdorster, E. (2001). Vitellogenin induction in painted turtle, *Chrysemys picta*, as a biomarker of exposure to environmental levels of estradiol. *Aquatic Toxicology*, 55, 49-60.
- Jager, M., Gauly, M., Bauer, C., Failing, K., Erhardt, G., & Zahner, H. (2005). Endoparasites in calves of beef cattle herds: Management systems dependent and genetic influences. *Veterinary Parasitology*, 131, 173-191.
- Jakubowski, W. & Craun, G.F. (2002). Update on the control of Giardia in water supplies. In B. E. Olson, M. E. Olson & P. M. Wallis. (Eds.), *Giardia, the Cosmopolitan Parasite* (pp. 217–238). Wallingford: CAB International.
- Jenkins, M.C. (2001). Advances and prospects for subunit vaccines against protozoa of veterinary importance. *Veterinary Parasitology*, 101, 291–310.
- Jensen, K.M., Makynen, E.A., Kahl, M.D., Ankley, G.T. (2006). Effects of the feedlot contaminant 17 $\alpha$ -trenbolone on reproductive endocrinology of the fathead minnow. *Environmental Science and Technology*, 40, 3112–3117.
- Jobling S., Nolan M., Tyler C.R., Brighty G., & Sumpter J.P. (1998). Widespread sexual disruption in wild fish. *Environmental Science and Technology*, 32, 2498–2506.
- Johnson, J.Y.M., Thomas, J.E., Graham, T.A., Townshend, I., Byrne, J., Selinger, B.L., Gannon, V.P.G. (2003). Prevalence of *Escherichia coli* O157:H7 and *Salmonella* spp in surface waters of southern Alberta and its relation to manure sources. *Canadian Journal of Microbiology*, 49, 326–335.
- Johnson, R.P., Wilson, J.B., Michel, P., Rahn, K., Renwick, S.A., Gyles, C.L., & Spika, J.S. (1999). Human infection with verocytotoxigenic *Escherichia coli* associated with exposure to farms and rural environments. In C.S. Stewart & H.J. Flint (Eds.) *Escherichia coli O157:H7 in farm animals* (pp. 147-168). New York: CABI Publishing.
- Jürgens M.D., Holthaus K.I.E., Johnson A.C., Smith J.J.L., Hetheridge M., & Williams R.J. (2002). The potential for estradiol and ethinylestradiol degradation in English rivers. *Environmental Toxicology and Chemistry*, 21, 480–488.

- Karim M.R., Manshadi F.D., Karpiscak M.M., Gerba C.P. (2004). The persistence and removal of enteric pathogens in constructed wetlands. *Water Research*, 38,1831–1837.
- Kelly, E.N., Short, J.W., Schindler, D.W., Hodsom, P.V., Ma, M., Kwan, A.K. & Fortin, B.L. (2010). Oilsands development contributes polycyclic aromatic compounds to the Athabasca River and its tributaries. *Proceedings of the National Academy of Sciences*, 107, 951-952.
- Khan, M.Z., & Law, F.C.P. (2005). Adverse effects of pesticides and related chemical on enzyme and hormone systems of fish, amphibians, and reptiles: a review. *Proceedings of the Pakistan Academy of Sciences*, 42, 315-323.
- Kidd, K.A., Blanchfield, P.J., Mills, K.H., Palace, V.P., Evans, R.E., Lazorchak, J.M., & Flick, R.W. (2007). Collapse of a fish population after exposure to a synthetic estrogen. *Proceedings of the National Academy of Sciences*, 104, 8897-8901.
- Kjaer, J., Olsen, P., Bach, K., Barlebo, H.C., Ingerselv, F., Hansen, M., & Sørensen, B.H. (2007). Leaching of estrogenic hormones from manure-treated structured soils. *Environmental Science and Technology* 41, 3911–3917.
- Kloas, W., Lutz, I., & Einspanier, R. (1999). Amphibians as models to study endocrine disruptors: II. Estrogenic activity of environmental chemicals *in vitro* and *in vivo*. *Science of the Total Environment*, 225, 59–68.
- Knudsen F.R., Schou A.E., Wiborg M.L., Mona E., Tollefsen K.E., Stenersen J., & Sumpter J.P. (1997). Increase of plasma vitellogenin concentration in rainbow trout (*Oncorhynchus mykiss*) exposed to effluents from oil refinery treatment works and municipal sewage. *Bulletin of Environmental Contamination and Toxicology*, 59, 802–806.
- Komor, S.C., & Anderson, H.W. (1993). Nitrogen isotopes as indicators of nitrate sources in Minnesota sand-plain aquifers. *Ground Water*, 31, 260–270.
- Kondro, W. (2000). *E. coli* outbreak deaths spark judicial inquiry in Canada. *Lancet*, 355, 2058.
- Kvac, M., Kouba, M., & Vitovec, J. (2006) Age-related and housing-dependence of Cryptosporidium infection of calves from dairy and beef herds in South Bohemia, Czech Republic. *Veterinary Parasitology*, 137, 202–209.
- Laetz, C.A., Baldwin, D.H., Collier, T.K., Hebert, V., Stark, J.D., & Scholz, N.L. (2009). The synergistic toxicity of pesticide mixtures: implications for risk assessment and the conservation of endangered Pacific Salmon. *Environmental Health Perspectives*, 117, 348-353.
- Larsson, P. (1985). Contaminated sediments of lakes and oceans act as sources of chlorinated hydrocarbons for release to water and atmosphere. *Nature*, 317, 347–349.

- Lauderdale JW. (1983). Use of MGA® (melengestrol acetate) in animal production. In Meissonnier E, Mitchell-Vigneron J, (Eds.), *Proceedings of the Symposium on Anabolics in Animal Production: Public Health Aspects, Analytical Methods, and Regulation* (pp. 15–17) Paris: Office International des Epizooties.
- Lauderdale J.W., Goyings L.S., Krzeminski L.F., Zimbelman R.G. (1977). Studies of a progestogen (MGA) as related to residues and human consumption. *Journal of Toxicology and Environmental Health*, 3, 5–33.
- LeChevallier, M.W., Norton, W.D., & Lee, R.G. (1991a). Occurrence of *Giardia* and *Cryptosporidium* spp. in Surface Water Supplies. *Applied and Environmental Microbiology*, 57, 2610-2616.
- LeChevallier, M.W., Norton, W.D., & Lee, R.G. (1991b). *Giardia* and *Cryptosporidium* spp. in filtered drinking water supplies. *Applied and Environmental Microbiology*, 57, 2617-21.
- Lee, L.S., Carmosini, N., Sassman, S.A., Dion, H.M., & Sepulveda, M.S. (2007). Agricultural Contributions of Antimicrobials and Hormones on Soil and Water Quality. (pp. 2-69). In *Advances in Agronomy*, Volume 93. San Diego: Elsevier.
- Lengerich, E.J., Addiss, D.G., Marx, J.J., Ungar, B.L., & Juranek, D.D. (1993). Increased exposure to Cryptosporidia among dairy farmers in Wisconsin, *Journal of Infectious Diseases*, 167, 1252–1255.
- Li, H., Jiang, H., Gao, Z., Wang, X., Qu, W. Lin, R., & Chen, J. (2008). Acute toxicity of the pesticide methomyl on the topmouth gudgeon (*Pseudorasbora parva*): mortality and effects on four biomarkers. *Fish Physiology and Biochemistry*, 34, 209-216.
- Linde-Arias, A., Inacio, A.F., de Albuquerque, C., Freire, M.M., & Moreira, J.C. (2008). Biomarkers in an invasive fish species, *Oreochromis niloticus*, to assess the effects of pollution in a highly degraded Brazil River. *Science of the Total Environment*, 399, 186-192.
- Liu, B. and Liu, X. (2004). Direct Photolysis of estrogens in aqueous solutions. *Science of the Total Environment*, 320, 269-274.
- Lorenzen, A., Burnson, K., Servos, M., & Topp, E. (2006). Persistence of endocrine disrupting chemicals in agricultural soils. *Journal of Environmental Engineering Science*, 5, 211-219.
- Loring, D.H., & Rantala, R.T.T. (1992). Manual for the geochemical analysis of marine sediments and suspended particulate matter. *Earth-Science Reviews*, 32, 235.
- Majewska, A.C. (1994). Successful experimental infections of a human volunteer and Mongolian gerbils with *Giardia* of animal origin. *Transactions of the Royal Society of*

*Tropical Medicine and Hygiene*, 88, 360–362.

Matozzo, V., Gagne, F., Marin, M.G., Ricciardi, F., & Blaise, C. (2008). Vitellogenin as a biomarker of exposure to estrogenic compounds in aquatic invertebrates: a review. *Environment International*, 34, 531-545.

Matthews, G.A. (2006). Approval of Pesticides. In *Pesticides: Health, safety, and the environment*. Oxford: Blackwell.

Matthiessen P. (2003). Endocrine disruption in marine fish. *Pure Applied Chemistry*, 75, 2249–2261.

Matthiessen, P., Arnold, D., Johnson, A. C., Pepper, T.J., Pottinger, T.G., & Pulman, K.G.T. (2006). Contamination of headwater streams in the United Kingdom by oestrogenic hormones from livestock farms. *Science of the total environment*, 367, 616-630.

McDowell, R.W., & Sharpley, A.N. (2001). Approximating phosphorus release from soils to surface runoff and subsurface drainage. *Journal of Environmental Quality*, 30, 508–520.

McLauchlin, J., Amar, C., Pedraza-Diaz, S., & Nichols, G.L. (2000). Molecular epidemiological analysis of *Cryptosporidium* spp. in the United Kingdom: results of genotyping *Cryptosporidium* spp. in 1705 fecal samples from humans and 105 fecal samples from livestock animals. *Journal of Clinical Microbiology*, 38, 3984–3990.

McRae, T., Smith, C.A.S., & Gregorich, L.J. (2000). *Environmental Sustainability of Canadian Agriculture: Report of the Agri-Environmental Indicator Project. A Summary*. Agriculture and Agri-Food Canada, Ottawa, Ont.

Metcalfe C., Metcalfe, T., Kiparissis, Y., & Koenig, B. (2001). Estrogenic potency of chemicals detected in sewage treatment plant effluents as determined by in vivo assays with Japanese medaka (*Oryzias latipes*). *Environmental Toxicology and Chemistry*, 20, 297–308.

Meyer H.H.D. (2001). Biochemistry and physiology of anabolic hormones used for improvement of meat production. *Acta Pathologica, Microbiologica et Immunologica*, 109, 1–8.

Michel P, Wilson J.B., Martin S.W., Clarke R.C., McEwen S.A., & Gyles C.L. (1999). Temporal and geographical distribution of reported cases of *Escherichia coli* O157:H7 infection in Ontario. *Epidemiology and Infection*, 122, 193–200.

Miller, L.L., Rasmussen, J.B., Palace, V.P., & Hontela, A. (2009). Physiological stress response in white suckers from agricultural drain waters containing pesticides and selenium. *Ecotoxicology and Environmental Safety*, 72, 1249-1256.

- Milligan, S.R., Balasubramanian, A.V., & Kalita, J.C. (1998). Relative potency of xenobiotic estrogens in an acute in vivo mammalian assay. *Environmental Health Perspectives*, 106, 23-26.
- Miron, D., Kenes, J., & Dagan, R. (1991). Calves as a source of an outbreak of cryptosporidiosis among young children in an agricultural closed community. *Pediatric Infectious Disease Journal*, 10, 438–441.
- Mohammed, H.O., Wade, S.E., & Schaaf, S. (1999). Risk factors associated with *Cryptosporidium parvum* infection in dairy cattle in Southeastern New York State. *Veterinary Parasitology*, 83, 1–13.
- Molbak, K., Andersen, M., Aaby, P., Hojlyng, N., Jakobsen, M., Sodemann, M., & da Silva, A.P. (1997). *Cryptosporidium* infection in infancy as a cause of malnutrition: a community study from Guinea-Bissau, West Africa. *American Journal of Clinical Nutrition*, 65, 149–152.
- Morgan, U.M., Sargent, K. D., Deplazes, P., Forbes, D.A., Spano, F., Hertzberg, H., Elliot, A., & Thompson, R. C. A. (1998). Molecular characterization of *Cryptosporidium* from various hosts. *Parasitology*, 117, 31-37.
- National Research Council, (2000). Chapter 5: Health Effects of Methylmercury. In *Toxicological effects of Methylmercury*, (pp.147-249). Washington D.C: National Academy Press.
- Nationtalk. (2009, October 23<sup>rd</sup>). Health: Albertans urged to limit consumption of some species of fish. Retrieved on February 22<sup>nd</sup>, 2010, from <http://www.nationtalk.ca/modules/news/article.php?storyid=24874>
- Nichols, D. J., Daniel, T. C., Moore, P. A., Edwards, D. R., & Pote, D. H. (1997). *Journal of Environmental Quality*, 26, 1002-1006.
- Norris, D. O. (2007). Xenoestrogen Actions on Reproduction: Implications for Health of Wildlife And Humans. American Water Resources Association, Emerging Contaminants of Concern in the Environment: Issues, Investigations and Solutions.
- O'Brien, E., McInnes, L., Ryan, U. (2008). *Cryptosporidium* Gp60 genotypes from humans and domesticated animals in Australia, North America, and Europe. *Experimental Parasitology*, 118, 118-121.
- O'Handley, R. M. (2007). *Cryptosporidium parvum* infection in cattle: are current perceptions accurate? *Trends in Parasitology*, 23, 477-480.
- O'Handley, R.M., Cockwill, C., McAllister, T.A., Jelinski, M., Morck, D.W., and Olson, M.E. (1999). Duration of naturally acquired Giardiasis and Cryptosporidiosis in dairy calves and their association with diarrhea. *Journal of the American Veterinary Association*, 214, 391–396.

- O'Handley, R.M., & Olson, M.E. (2006). Giardiasis and cryptosporidiosis in ruminants. *Veterinary Clinics of North America: Food Animal Practices*, 22, 623–643.
- O'Handley, R.M., Olson, M.E., Fraser, D., Adams, P., & Thompson, R.C.A. (2000). Prevalence and genotypic characterisation of *Giardia* in dairy calves from Western Australia and Western Canada. *Veterinary Parasitology*, 90, 193-200.
- Olson, M.E., Guselle, N.J., O'Handley, R.M., Swift, M.L., McAllister, T.A., Jelinski, M.D., & Morck, D.W. (1997a) *Giardia* and *Cryptosporidium* in dairy calves in British Columbia. *Canadian Veterinary Journal*, 38, 703–706.
- Olson, M.E., O'Handley, R.M., Ralston, B.J., McAllister, T.A. Thompson, R.C.A. (2004). Update on *Cryptosporidium* and *Giardia* infections in cattle. *Trends in Parasitology*, 185-191.
- Olson, M.E., Thorlakson, C.L., Deselliers, L., Morck, D.W., & McAllister, T.A. (1997b). *Giardia* and *Cryptosporidium* in Canadian Farm Animals. *Veterinary Parasitology*, 68, 375-381.
- Ontario College of Physicians and Surgeons (2004). Pesticides Literature Review. Retrieved on January 15<sup>th</sup>, 2010, from [www.ocfp.on.ca/English/OCFP/Communications/CurrentIssues/Pesticides/](http://www.ocfp.on.ca/English/OCFP/Communications/CurrentIssues/Pesticides/)
- Orlando, E.F., Kolok, A.S., Binzick, G.A., Gates, J.L., Horton, M.K., Lambright, C.S., Gray, L.E. Jr., Soto, A.M., & Guillette, L.J. Jr. (2004). Endocrine-disrupting effects of cattle feedlot effluent on an aquatic sentinel species, the fathead minnow. *Environmental Health Perspectives*, 112, 353-358.
- Palm, A., Cousins, I.T., Mackay, D., Tysklind, M., Metcalfe, C., Alaei, M. (2002). Assessing the environmental fate of chemicals of emerging concern: a case of the polybrominated diphenyl ethers. *Environmental Pollution*, 117, 195–213.
- Pape-Lindström P.A., & Lydy M.J. (1997) Synergistic toxicity of atrazine and organophosphate insecticides contravenes the response addition mixture model. *Environmental Toxicology and Chemistry*, 16, 2415-2420.
- The Pembina Institute. Sustainable Energy Solutions. Written Submission: federal Parliamentary Committee Hearing on Water and the Oil Sands. Retrieved on February 20<sup>th</sup>, 2010, from [pubs.pembina.org/reports/oil-sands-and-water-submission.pdf](http://pubs.pembina.org/reports/oil-sands-and-water-submission.pdf)
- Peng, M.M., Xiao, L., Freeman, A., Arrowood, M.J., Escalante, A.A., Weltman, A.C., Ong, C.S., MacKenzie, W.R., Lal, A.A., & Beard, C.B. (1997). Genetic polymorphism among *C. parvum* isolates: Evidence of 2 distinct human transmission cycles. *Emerging Infectious Diseases*, 3, 567-73.
- Percival, S., Chalmers R., Embrey, M., Hunter, P., Sellwood, J., Wyn-Jones, P. (2004). *Microbiology of waterborne diseases*. Amsterdam: Elsevier.

- Pereira, S.J., Ramirez, N.E., Xiao, L., & Ward, L.A. (2002). Pathogenesis of human and bovine *Cryptosporidium parvum* in gnotobiotic pigs. *Journal of Infectious Diseases*, 186, 715–718.
- Peterson, E.W., Davis R.K., & Orndorff, H.A. (2000). 17-beta estradiol as an indicator of animal waste contamination in mantled karst aquifers. *Journal of Environmental Quality*, 29, 826-834.
- Preiser, G., Preiser, L., & Madeo, L. (2003). An outbreak of cryptosporidiosis among veterinary science students who work with calves. *Journal of American College Health*, 51, 213–215.
- Public Health Agency of Canada. (2007). Laboratory surveillance data for enteric pathogens in Canada: Annual Summary 2006. Retrieved on October 6<sup>th</sup>, 2009, from <http://www.nml-lnm.gc.ca/NESP-PNSME/assets/pdf/2006AnnualReport.pdf>
- Public Health Agency of Canada. (2008). Public Health Notifiable Disease Management Guidelines – Cryptosporidiosis. Retrieved on September 25<sup>th</sup>, 2009, from [www.health.alberta.ca/documents/ND-Cryptosporidiosis.pdf](http://www.health.alberta.ca/documents/ND-Cryptosporidiosis.pdf).
- Public Health Agency of Canada. (2008). Public Health Notifiable Disease Management Guidelines – Giardiasis. Retrieved on September 24<sup>th</sup>, 2009, from [www.health.alberta.ca/documents/ND-Giardiasis.pdf](http://www.health.alberta.ca/documents/ND-Giardiasis.pdf).
- Purdom, C.E., Hardiman, P.A., Bye, V.J., Eno, N.C., Tyler, C.R., & Sumpter J.P. (1994). Estrogenic effects of effluents from sewage treatment works. *Chemical Ecology*, 8, 275–285.
- Runes, H.B., Jenkins, J.J., Moore, J.A., Bottomley, P.J., Wilson, B.D. (2003). Treatment of atrazine in nursery irrigation runoff by a constructed wetland. *Water Research*, 37, 539–550.
- Robbins, J.W.D. (1979). Impact of unconfined livestock activities on water quality. *Transaction of the American Society of Agricultural Engineers*, 22, 1317-1323.
- Rodvang, S.J., Mikalson, D.M., & Ryan, M.C. (2004). Changes in Ground Water Quality in an Irrigated Area of Southern Alberta. *Journal of Environmental Quality*, 33, 476-487.
- Ralston, B.J., McAllister, T.A., & Olson, M.E. (2003). Prevalence and infection pattern of naturally acquired Giardiasis and Cryptosporidiosis in range beef calves and their dams. *Veterinary Parasitology*, 114, 113–122.
- Ramirez, N.E., Ward, L.A., & Sreevatsan, S. (2004). A review of the biology and epidemiology of Cryptosporidiosis in humans and animals. *Microbes and infection*, 6, 773-785.

- Robertson, L., Gjerde, B., Forberg, T., Haugejorden, G., & Kielland, C. (2006). A small outbreak of human Cryptosporidiosis associated with calves at a dairy farm in Norway. *Scandinavian Journal of Infectious Diseases*, 38, 810-813.
- Rose L.B., Lisle J.T., & LeChavallier, M. (1997). Waterborne Cryptosporidiosis: incidence, outbreaks, and treatment strategies. In R. Fayer, (Ed.). *Cryptosporidium and Cryptosporidiosis*, (pp. 93-110). Boca Raton: CRC Press.
- Raman, D. R., Williams, E. L., Layton, A. C., Burns, R. T., Easter, J. P., Daugherty, A. J., Mullen, M.D., & Sayler, G. S. (2004). Estrogen content of dairy and swine wastes. *Environmental Science and Technology*, 38, 3567-3573.
- Safar-Hermann, N., Ismail, M.N., Choi, H.S., Mostl, E., & Bamberg, E. (1987). Pregnancy diagnosis in zoo animals by estrogen determination in feces. *Zoo Biology*, 6, 189–193.
- Salomons, W., N.M. de Rooij, H. Kerdijk, & J. Bril. (1987). Sediments as a source for contaminants. *Hydrobiologia*, 149, 13–30.
- Salvano, E., Flaten, D.N., Rousseau, A.N., & Quilbe, R. (2009). Are current phosphorus risk indicators useful to predict the quality of surface waters in southern Manitoba, Canada? *Journal of Environmental Quality*, 38, 2096-2105.
- Santín, M., Trout, J.M., Xiao, L., Zhou, L., Greiner, E., & Fayer, R. (2004). *Veterinary Parasitology*, 122, 103-117.
- Sarmah, A.K., Northcott, G.L., Leusch, F.D.L., & Tremblay, L.A. (2006). A survey of endocrine disrupting chemicals (EDCs) in municipal sewage and animal waste effluents in the Waikato region of New Zealand. *Science of the Total Environment*, 355, 135-144.
- Sauer, T.J., Daniel, T.C., Nichols, D.J., West, C.P., Moore, P.A. Jr., & Wheeler, G.L. (2000). Runoff water quality from poultry litter treated pasture and forest sites. *Journal of Environmental Quality*, 29, 515–521.
- Schiffer B., Daxenberger A., Meyer K., & Meyer, H.H.D. (2001). The fate of trenbolone acetate and melengestrol acetate after application as growth promoters in cattle: environmental studies. *Environmental Health Perspectives*, 109, 1145-1151.
- Schindler, D.W., & Donahue, W.F. (2006). An impending water crisis in Canada's western prairie provinces. *Proceedings of the National Academy of Sciences*, 103, 7210-7216.
- Schuler, L.J., Trimble, A.J., Belden, J.B., Lydy, M.J. (2005) Joint toxicity of triazine herbicides and organophosphorous insecticides to the midge, *Chironomus tentans*. *Archives of Environmental Contamination and Toxicology*, 49, 173-177.
- Segner, H., Carroll, K., Fenske, M., Janssen, C.R., Maack, G., Pascoe, D., Schäfers, C., Vandenberg, G.F., Watts M., & Wenzel, A. (2003). Identification of endocrine-disrupting

effects in aquatic vertebrates and invertebrates: report from the European IDEA project. *Ecotoxicology and Environmental Safety*, 54, 302-314

Seki, M., Fujishima, S., Nozaka, T., Maeda, M., & Kobayashi, K., (2006). Comparison of response to 17 beta-estradiol and 17 beta-trenbolone among three small fish species. *Environmental Toxicology and Chemistry*, 25, 2742–2752.

Sellin, M.K., Snow, D.D., Gustafson, S.T., Erickson, G.E., & Kolok, A.S. (2009). The endocrine activity of beef cattle wastes: Do growth-promoting steroids make a difference? *Aquatic Toxicology*, 92, 221-227.

Servos, M., Delorme, P., Fox, G., Sutcliffe, R. & Wade, M. (2000a). Proceedings of the 5-NR Workshop: Establishing a national agenda for the scientific assessment of endocrine disrupting substances.

Servos, M., Delorme, P., Fox, G., Sutcliffe, R., & Wade, M. 2000b. Establishing a national agenda for the scientific assessment of endocrine disrupting substances: workshop executive summary.

Sevros, M., Delorme, P., Fox, G., Sutcliffe, R., Wade, M. (2008). Threat to sources of drinking water and aquatic ecosystem health in Canada. Prepared for Environment Canada. Retrieved on January 27<sup>th</sup>, 2010, from [www.ec.gc.ca/inre-nwri/default.asp?lang=En&n=235D11EB-1&offset=6&toc=show](http://www.ec.gc.ca/inre-nwri/default.asp?lang=En&n=235D11EB-1&offset=6&toc=show)

Servos, M., Delorme, P., Fox, G., Sutcliffe, R., & Wade, M. (2001). Endocrine Disrupting Substances: Water Quality Issues in Canada. pp. 17-21. In *Threats to Sources of Drinking Water and Aquatic Ecosystem Health in Canada*. Prepared for Environment Canada. NWRI Scientific Assessment Report Series, ISSN 1499-5905.

Sharpe, R.M., & Skakkebaek N.E. (1993). Are oestrogens involved in falling sperm counts and disorders of the male reproductive tract? *Lancet*, 341, 1392-1395.

Shore, L. (2009). Chapter 4: transport of steroids in surface waters. In L.S. Shore, & A. Pruden (Eds.) *Hormones and pharmaceutical generated by concentrated animal feeding operations. Emerging topics in ecotoxicology, Principles, approaches, and perspectives, Volume , 1* (p. 23-27). New York: Springer.

Shore L.S., & Shemesh M. (2003). Naturally produced steroid hormones and their release into the Environment. *Pure and Applied Chemistry*, 75, 1859–1871.

Shugart, G. (1980). Frequency and distribution of polygony in Great Lake herring gulls in 1978. *Condor*, 82, 426-429.

Shutes, R.B.E. (2001). Artificial wetlands and water quality improvement. *Environment International*, 26, 441–447.

- Sone, K., Hinago, M., Itamoto, M., Katsu, Y., Watanabe, H., Urushitani, H., Tooi, O., Guillette, L. J., Jr., & Iguchi, T. (2005). Effects of an androgenic growth promoter 17alpha-trenbolone on masculinization of mosquitofish (*Gambusia affinis affinis*). *General and Comparative Endocrinology*, 143, 151-160.
- Sonnenschein, C. & Soto, A. M. (1998). An updated review of environmental estrogen and androgen mimics and antagonists. *Journal of Steroid Biochemistry and Molecular Biology*, 65, 143–150.
- Soto, A.M., Calabro, J.M., Prechtel, N.V., Yau, A.Y., Orlando, E.F., Daxenberger, A., Kolok, A.S., Guillette, L.J., le Bizec, B., Lange, I.G. & Sonnenschein, A. (2004). Androgenic and estrogenic activity in water bodies receiving cattle feedlot effluent in eastern Nebraska. *Environmental Health Perspectives*, 112, 346-352.
- Spark, K.M., & Swift, R.S. (2002). Effect of soil composition and dissolved organic matter on pesticide sorption. *The Science of the Total Environment*, 298, 147-161.
- Stegeman, J.J. ,& Hahn, M.E. (1994). Biochemistry and molecular biology of monooxygenase: current perspective on forms, functions, and regulation of cytochrome P450 in aquatic species. In D.C. Malins, & G.K. Ostrander, (Eds.), *Aquatic Toxicology; Molecular, Biochemical and Cellular Perspectives*. (pp. 87-207). Boca Raton: Lewis Publishers, CRC press.
- Statistics Canada. (2009). Agricultural Water Use Survey 2007, Methodology Report. Environment Accounts and Statistics Analytical and Technical Paper Series, Catalogue no. 16-001-M2009008.
- Statistics Canada (2009). Cattle inventories, by province.
- Statistics Canada. (1996). Environment Accounts and Statistics Division and Agriculture Division.
- Sturm, A., Wogram, J., Hansen, P.D., & Liess, M. (1999). Potential use of cholinesterase in monitoring low levels of organophosphates in small streams: natural variability in three-spined stickleback (*Gasterosteus aculeatus*) and relation to pollution. *Environmental Toxicology and Chemistry*, 18, 194–200.
- Sulaiman, I.M., Xiao, L., Yang, C., Escalante, L., Moore, A., Beard, C.B., Arrowood, M.J., & Lal, A.A. (1998). Differentiating human from animal isolates of *Cryptosporidium parvum*. *Emerging Infectious Diseases*, 4, 681–685.
- Sumpter J.P. (2005). Endocrine disrupters in the aquatic environment: An overview. *Acta Hydrochimica et Hydrobiologica*, 33, 9–16.
- Sumpter, J.P., & Jobling, S. (1995). Vitellogenesis as a biomarker for estrogenic contamination of the aquatic environment. *Environment Health Perspectives*, 103, 174-178.

- Ternes, T.A., Stupf, M., Mueller, J., Haberer, K., Wilken R.D., & Servos, M. (1999). Behaviours and the occurrence of estrogens in municipal sewage treatment plants 1. Investigations in Germany, Canada, and Brazil. *Science of the Total Environment*, 225, 81-90.
- Thompson, R.C.A. (1998). Giardia infections. In S.R. Palmer, Lord Soulsby, D.I.H. Simpson, (Eds.), *Zoonoses: Biology, Clinical Practice, and Public Health Control* (pp. 545–561). Oxford: Oxford University Press.
- Thompson, R.C.A. (2003). Molecular epidemiology of Giardia and Cryptosporidium infections. *Journal of Parasitology*, 89, S134–S140.
- Thompson, R.C.A., & Boreham, P.L.F. (1994). Biotic and abiotic transmission. In R.C.A Thompson, J.A. Reynoldson, & A.J. Lymbery (Eds.), *Giardia from molecules to disease* (pp. 83-97). Cambridge, UK: CAB International.
- Thompson, R.C., Hopkins R.M., & Homan, W.L. (2000). Nomenclature and genetic groupings of Giardia infecting mammals, *Parasitology Today*, 16, 210–213.
- Thompson, R.C.A., Lymbery, A.J., Meloni, B.P., & Binz, N. (1990). The zoonotic transmission of Giardia species. *Veterinary Record*, 126, 513–514.
- Thompson, R.C.A., Reynoldson, J.A. & Mendis, A.H.W. (1993). Giardia and Giardiasis. *Advances in Parasitology* 32, 71-160.
- Toppari, J., Larsen, J.C., Christiansen, P., Giwereman, A., Grandjean, P., Guillette, I.J. Jr., Jegou, B., Jensen, T.K., Jounnet, P., Keiding, K., Leffers, H., Mclachlan, J.A., Meyer, O., Muller, J., Rajpert De Meyts, F., Seheike, T., Sharpe, R., Sumpter, J., & Skakkebaek, N.E. (1996). Male reproductive health and environmental xenoestrogens. *Environmental Health Perspectives*, 104, 741-803.
- Tremblay, A., M. Lucotte & R. Schetagne. (1998). Total mercury and methylmercury accumulation in zooplankton of hydroelectric reservoir in northern Quebec (Canada). *Science of the Total Environment*, 213, 307–315.
- Trimble, A.J., & Lydy, M.J. (2006). Effeectis of Triazine herbicides on organophosphate insecticide toxicity in *Hyalella asteca*. *Archives of Environmental Contamination and Toxicology*, 51, 29-34.
- Trout, J.M., Santín, M. & Fayer, R. (2007). Prevalence of *Giardia duodenalis* genotypes in adult dairy cows. *Veterinary Parasitology*, 147, 205-209.
- Trout, J.M., Santin, M., Greiner, E., & Fayer, R. (2004). Prevalence of *Giardia duodenalis* genotypes in pre-weaned dairy calves. *Veterinary Parasitology*, 124, 179–186.
- Trout J.M., Santin M., Greiner E., & Fayer, R. (2005). Prevalence and genotypes of *Giardia duodenalis* in post-weaned dairy calves. *Veterinary Parasitology* 130, 177–83.

- Trout, J.M., Santín, M., Greiner, E., & Fayer, R. (2006). Prevalence and genotypes of *Giardia duodenalis* in 1–2 year old dairy cattle. *Veterinary Parasitology*, 140, 217-222.
- Uehlinger, F.D., Barkema, H.W., Dixon, B.R., Coklin, T., & O’Handley, R.M. (2006). *Giardia duodenalis* and *Cryptosporidium* spp. in a veterinary college bovine teaching herd. *Veterinary Parasitology* 142, 231-237.
- Ungar, B.L.P. (1990). Cryptosporidiosis in Humans (*Homo sapiens*). In J.P. Dubey, C.A. Speer, & R. Fayer, (Eds.), *Cryptosporidiosis of man and animals*, (pp. 59-82). Boca Raton, FL: CRC press.
- Upton, S.J. (2003). Cryptosporidium: they probably taste like chicken. In R.C.A. Thompson, A. Armson, & U.M. Ryan, (Eds.), *Cryptosporidium: from Molecules to Disease* (pp. 38-50). Amsterdam: Elsevier Science B.V.
- Van der Oost R., Beyer J., & Vermeulen N.P.E. (2003). Fish bioaccumulation and biomarkers in environmental risk assessment: a review. *Environmental Toxicology and Pharmacology*, 13, 57–149.
- Valcour, J.E., Michel, P., McEwen, S.A., & Wilson, J.B. (2002). Associations between indicators of livestock farming intensity and incidence of human Shiga toxin-producing *Escherichia coli* infection. *Emerging Infectious Diseases*, 8, 252–257.
- Van der Kamp, G., & Grove, G. (2001). Well water quality in Canada: An overview. p. 39–42. In Proc. 2nd Joint Conf. of the Int. Assoc. of Hydrogeol. and the Canadian Geol. Soc., Calgary, AB. 16–19 Sept. 2001. Canadian Geotech. Soc., Calgary.
- Vighi, M., & Di Guardo, A. (1995). Environmental distribution and fate and exposure prediction: Predictive approaches for the evaluation of pesticide exposure. In M. Vighi, and E. Funari (Eds.) *Pesticide Risk in Groundwater*, CRC Press, Boca Raton, Florida.
- Wallis, P.M., Erlandsen, S.L., Issac-Renton, J.L., Olson, M.E., Robertson, W.J., & Van Keulen, H. (1996). Prevalence of *Giardia* cysts and *Cryptosporidium* oocysts and characterization of *Giardia* spp isolated from the drinking water in Canada. *Applied Environmental Microbiology*, 62, 2789-2797.
- Wassenaar, L. (1995). Evaluation of the origin and fate of nitrate in the Abbotsford Aquifer using the isotopes of <sup>15</sup>N and <sup>18</sup>O in NO<sub>3</sub><sup>-</sup>. *Applied Geochemistry*, 10, 391–405.
- Water Matters, (2010). What does Kennedy know about mercury in Alberta’s fish? Retrieved on February 27<sup>th</sup>, 2010, from <http://www.water-matters.org/story/355>
- Webster, P. (2008). Canada moves to protect babies from chemical. *The Lancet*, 371, 2074.

- Wielinga P. R., de Vries A., van der Goot T.H., Mank T., Mars, M. H., Kortbeek L. M., & van der Giessen, J. W. B. (2008). Molecular epidemiology of *Cryptosporidium* in humans and cattle in The Netherlands. *International Journal for Parasitology*, 38, 809-817.
- Wiener, J.G., Krabbenhoft, D.P., Heinz, G.H., and Scheuhammer, A.M. (2003). Chapter 16: Ecotoxicology of mercury. In D.J. Hoffman, B.A. Rattner, G.A. Burton, Jr., & J. Cairns, Jr. (Eds.), *Handbook of Ecotoxicology, 2nd edition*, (pp. 409-463). Boca Raton, Florida: CRC Press.
- Wilkins, K., Johansen, H., Beaudet, M.P., & Ineke Neutel, C. (2000). Oral Contraceptive Use. *Health Reports*, 11, 25-37.
- Winkworth, C.L., Learmonth, J.J., Matthaei C.D., & Townsend, C.R. (2008). Molecular characterization of *Giardia* isolates from calves and humans in a region in which dairy farming has recently intensified. *Applied Environmental Microbiology*, 74, 5100–5105.
- Wolf, A.H., & Patz, J. A. (2002). Reactive Nitrogen and Human Health: Acute and Long-term Implications. *Royal Swedish Academy of Sciences*, 31, 120-125.
- Wortmann, C.S. & Walters, D. (2006). Phosphorus runoff during four years following composted manure application. *Journal of Environmental Quality*, 35, 651-657.
- Xiao, L. (1994). *Giardia* infection in farm animals. *Parasitology Today*, 10, 436–438.
- Xiao, L. (2010). Molecular Epidemiology of Cryptosporidiosis: an update. *Experimental Parasitology*, 124, 80-89.
- Xiao, L. & Herd, R.P. (1994). Infection pattern of *Cryptosporidium* and *Giardia* in calves. *Veterinary Parasitology*, 55, 257–262.
- Xiao, L., & Ryan, U.M. (2004). Cryptosporidiosis: an update in molecular epidemiology. *Current Opinion in Infectious Diseases*, 17, 483-490.
- Xu, P., Widmer, G., Wang, Y., Ozaki, L.S., Alves, J.M., Serrano, M.G., Puiu, D., Manque, P., Akiyoshi, D., Mackey, A.J., Pearson, W.R., Dear, P.H., Bankier, A.T., Peterson, D.L., Abrahamsen, M.S., Kapur, V., Tzipori, S., & Buck, G.A. (2004). The genome of *Cryptosporidium hominis*. *Nature*, 432, 415.
- Yang, J.Y., Huffman, E.C., De Jong, R., Kirkwood, V., MacDonald, K.B., & Drury, C.F. (2007). Residual soil nitrogen in soil landscapes of Canada as affected by land use practices and agricultural policy scenarios. *Land Use Policy*, 24, 89–99.