

**CAPP Freshwater Salinity Working Group and  
the Salt Technical Advisory Sub-committee of the  
British Columbia Upstream Petroleum Committee**

***A Review of the Toxicological Literature for Salt –  
2002 to 2007***

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## EXECUTIVE SUMMARY

As a follow up to the Derivation of Soil Quality Matrix Soil Standards for Salt under the British Columbia Contaminated Sites Regulation Draft Document, prepared by Doug Bright and Jan Addison (2002), a literature review was conducted to assess any new scientific research into the toxicity potential of salt ions to aquatic life and plants. In particular, there is an interest in whether recent and emerging scientific knowledge would alter conclusions in Bright and Addison (2002) regarding thresholds of toxicity for sodium or chloride in soil and water. The literature review, conducted by UMA Engineering Ltd. (UMA) using online searchable databases of scientific journals, was limited to new research published since 2002.

Seventeen relevant research papers were identified, although very few of these were directly related to sodium chloride (NaCl) toxicity to terrestrial and freshwater life. The purpose of this report is to provide a summary of recent research examining salt toxicity to wildlife and plants and to evaluate this new data in the context of the draft soil matrix standards for salts and the BC Working and Approved Water Quality Guidelines for the protection of aquatic life.

Table S-1 summarizes the toxicity data applicable to plant and animal species that might be surrogates for those found in BC, and compares toxicity values to the draft soil matrix salinity standards (Bright *et al*, 2002) and the provincial Working and Approved Water Quality Guidelines (MoE, 2006). Exposures involving simultaneous exposure to another substance are not summarized in Table 1. Similarly, studies involving hydroponic or soil solution exposures are also not included Table 1, since the basis of expression of exposure concentration is not conducive to comparison with BC draft soil matrix standards for salt ions.

While limited new data were located for freshwater organisms and plants, no new data were found for soil invertebrates.

Table S-1: Summary of Toxicity Test Results for BC Species				
Study	Toxicity Test	Organism	Result	Guideline
Davies and Hall (2006)	48 hour acute Cl <sup>-</sup> toxicity test in water with varying Ca:Mg ratios.	<i>Daphnia magna</i>	LC50 results: 3,136 mg/L Cl <sup>-</sup> (0.7 Ca:Mg); 3,222 mg/L Cl <sup>-</sup> (1.8 Ca:Mg); 3,137 mg/L Cl <sup>-</sup> (7.0 Ca:Mg);	BC Approved Water Quality Guideline for chloride: 150 mg/L
Diamond <i>et al</i> (2005)	Pulsed exposure of NaCl in water at varying concentrations and durations for 7 days	Fathead minnows ( <i>Pimephales promelas</i> )	Decreased survival with 4 g/L NaCl for 96 hours, 8 g/L for 24 hours and 12 g/L for 3 hours	BC Approved Water Quality Guideline for chloride: 150 mg/L
Sanzo and Hecnar (2005)	96 hour acute NaCl toxicity test	Wood frog tadpoles ( <i>Rana sylvatica</i> )	LC50 results: 2,636 mg/L by Spearman-Kaber and 5,109 mg/L by probit analysis	BC Approved Water Quality Guideline for chloride: 150 mg/L
Sanzo and Hecnar (2005)	90 day chronic NaCl toxicity test using concentrations of 0.0, 0.39, 77.8, and 1030 mg/L NaCl	Wood frog tadpoles ( <i>Rana sylvatica</i> )	Lower survivorship, decreased time to metamorphosis at 1030 mg/L NaCl, as well as reduced weight and activity, and increased physical abnormalities with increasing salt concentration.	BC Approved Water Quality Guideline for chloride: 150 mg/L

Based on a review of the most recent published scientific information, it is concluded that toxicological data for salt which became available post-2002 would not substantively change the draft matrix soil standards derived in 2002 for either sodium (Na<sup>+</sup>) or chloride (Cl<sup>-</sup>).

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## 1. Introduction

As a follow up to the Derivation of Soil Quality Matrix Soil Standards for Salt under the British Columbia Contaminated Sites Regulation Draft Document, prepared by Doug Bright and Jan Addison (2002), a literature review was conducted to assess any new scientific research into the toxicity potential of salt ions to aquatic life and plants. In particular, there is an interest in whether recent and emerging scientific knowledge would alter conclusions in Bright and Addison (2002) regarding thresholds of toxicity for sodium or chloride in soil and water. The purpose of this report is to provide a summary of scientific research published between 2002 and August 2007 regarding the ecotoxicity of chloride or sodium ion.

The literature review was conducted by UMA Engineering Ltd. (UMA) and utilized online searchable databases of scientific journals. The literature review was limited to new research published since 2002. Online databases searched included –

Springerlink,	ScienceDirect,	Wiley Interscience,
Royal Society of Chemistry,	Oxford University Press,	JSTOR,
American Chemical Society,	SETAC online journals	

A summary is provided below of recent knowledge on –

- Salt Toxicity to Aquatic Life (Section 2), and
- Salt Toxicity to Plants (Section 3).

No new studies on soil invertebrates and salt exposures were found.

Section 4 provides major conclusions. Sections 2 and 3 include a review of studies where the test organism(s) were exposed to multiple substances including salt ions, as well as hydroponic exposures of plants. Such studies, however were deemed to be not relevant for assessing salt toxicity thresholds.

## 2. Salt Toxicity to Freshwater Life

Recent research on salt toxicity to wildlife is limited. Few studies were found that pertain directly to NaCl toxicity. Several studies found pertained to the chemical toxicity of metals, organic and inorganic pollutants in saline solutions and whether or not salts enhanced or inhibited a toxicological response in test organisms.

**Dethloff et al (2007)** examined the chronic toxic effects of silver on the early life stages of rainbow trout (*Oncorhynchus mykiss*) in the presence and absence of 49 mg/L NaCl and assessed possible protective effects of sodium chloride to silver toxicity. Chronic toxicity did not appear to be greatly modified by the presence of sodium chloride in this experiment although a reduction in silver uptake was observed based on whole body analyses.

A similar study was conducted by **Naddy et al (2007)** that looked at chronic exposure of an early life stage and short term chronic effects of silver toxicity on fathead minnows (*Pimephales promelas*) in water amended with or without 60 mg/L NaCl. The researchers found that chronic toxicity of silver was mitigated to some extent by NaCl addition based on increases in the maximum acceptable tolerable concentration (MATC) and inhibition concentration of 20% of the population (IC<sub>20</sub>) compared to those found in unamended waters. Sodium chloride also appeared to provide a level of protection from silver accumulation in the early life stage study but not in the short term chronic study. The results of this research suggest a protective chloride and/or sodium effect in short-term and chronic exposures from ionic Ag<sup>+</sup> using fathead minnows.

Protective effects of chloride from sulfate toxicity were assessed by **Soucek and Kennedy (2005)** in acute toxicity tests of sulfate to *Ceriodaphnia dubia*, *Chironomus tentans*, *Hyalella azteca*, and *Sphaerium simile*. The toxicity tests were used to determine the LC<sub>50</sub> concentrations for sulphate in solutions of varying water compositions including high TDS solutions. Protective effects of chloride on sulfate toxicity to *H. azteca* was assessed by exposing *H. azteca* to 2800 mg/L sulphate in six different concentrations of chloride for 96 hours. The results showed that sulfate toxicity to *H. azteca* decreased with increased levels of chloride. At the lowest measured chloride concentration of 5 mg/L, only 20% of the test organisms exposed to 2846 mg/L sulfate were alive after 96 hours. At 13 mg/L Cl<sup>-</sup> survival increased nominally, but not significantly; however significant increases in survival were observed at and above 18 mg/L Cl<sup>-</sup>. Survival was 85% and 100% in the 36 and 67 mg/L Cl<sup>-</sup> treatments respectively. The results of the *H. azteca* experiment support the hypothesis that chloride has a protective effect against sulphate toxicity, because incremental increases in chloride were associated with incremental increases in survival.

In another experiment, **Soucek (2007)** examined the influence of both chloride and water hardness on acute toxicity of sodium sulfate to *Hyalella azteca* and *Ceriodaphnia dubia*. Results from the study indicated that chloride had varying effects on sodium sulfate toxicity to *C. dubia* and *H. azteca* over the range of 5 to 500 mg/L Cl<sup>-</sup>. For *H. azteca*, increasing chloride

concentration from 5 to 25 mg/L resulted in increased sulfate LC<sub>50</sub>s. For *C. dubia* the slope was not significantly different from zero over this chloride concentration range. In addition LC<sub>50</sub>s for *C. dubia* were higher than those for *H. azteca* for each chloride concentration over this range. Although a positive relationship between chloride concentration and sulphate LC<sub>50</sub> was observed for *H. azteca* over the range of 5 to 25 mg/L Cl<sup>-</sup>, a significant negative trend was observed over the range of 25 to 500 mg/L Cl<sup>-</sup>. An even stronger negative relationship was observed for *C. dubia* over the same chloride range.

These studies did not assess the toxic effects of chloride directly on test organisms; therefore, a comparison to the BC Approved Water Quality Guidelines (BC AWQG) for chloride could not be made.

Only one study was found that investigated chloride toxicity to *Daphnia magna* in exposure waters of varying Ca:Mg ratios and hardness.

**Davies and Hall (2006)** conducted acute chloride toxicity tests using reformulated waters with Ca:Mg ratios of 0.7, 1.8, and 7.0. *D. magna* was exposed to saline waters (NaCl) at 100 mg/L hardness for all Ca:Mg ratios. Mortality was determined at the end of the 48 hour exposure period. The mean median lethal concentration for *D. magna* in NaCl toxicity tests at 100 mg/L hardness expressed as Cl<sup>-</sup> anion concentration was 3,136 mg/L (0.7 Ca:Mg), 3,222 mg/L (1.8 Ca:Mg), and 3,137 mg/L (7.0 Ca:Mg). No significant difference was found between exposure waters at different Ca:Mg ratios and chloride toxicity. The chloride concentrations found to elicit mortality in this study were far higher than the BC AWQG for chloride, which is 150 mg/L chloride. This indicates that the BC water quality guideline for chloride would be protective of chloride toxicity to freshwater aquatic invertebrates.

In the above-described studies by Dethloff *et al* (2007) and Naddy *et al* (2007), protective effects of sodium chloride to silver toxicity on two freshwater fish species was observed. Similarly a protective effect of chloride on sulfate toxicity to *H. azteca* was also found by Soucek and Kennedy. However, a study conducted by Barwardi (2007) found sodium chloride to enhance the toxic effects of the agricultural pesticide fethion on three freshwater fish species.

**Barwardi *et al* (2007)** examined the impacts of hypersaline water on the biotransformation and toxicity of fethion, an agricultural pesticide, on rainbow trout (*Oncorhynchus mykiss*), striped bass (*Morone saxatilis* x *Morone chrysops*) and tilapia (*Oreochromis mossambicus*). Results from the

96 hour toxicity test indicated that rainbow trout exposed to fenthion in a hypersaline environment experience significantly greater toxicity (LC<sub>50</sub> of 0.18 mg/L fenthion) than those fish in freshwater treatments with no additional NaCl (LC<sub>50</sub> 1.12 mg/L fenthion). The LC<sub>50</sub> for freshwater treated tilapia was 6.59 mg/L fenthion, compared to 4.56 mg/L fenthion in hypersaline conditions. Although a trend toward salinity-enhanced toxicity was observed, a significant difference between the two treatments was not observed in experiments with rainbow trout and tilapia. In contrast, toxicity testing with striped bass indicated a significant difference between freshwater and hypersaline environments, with LC<sub>50</sub> values being 13.2 mg/L fenthion and 2.8 mg/L fenthion, respectively. A comparison of the three freshwater fish species indicated that trout were approximately 10 times more sensitive to fenthion toxicity than striped bass or tilapia in freshwater treatments. In hypersaline conditions, trout were 36 and 16 times more sensitive than tilapia or striped bass respectively.

Although these studies are not directly related to NaCl toxicity on aquatic life they do indicate that hypersaline solutions may enhance or inhibit the chemical toxicity of pollutants within the aqueous environment on aquatic organisms.

Two studies were found that included toxicity tests of NaCl on fish species.

**Arezon et al (2003)** evaluated the feasibility of use of a Brazilian fish, *Cynopoecilus melanotaenia*, as a test organism in toxicity tests. Three reference substances were used in a 96 hour acute toxicity test, one of which was sodium chloride (NaCl). Fry of *C. melanotaenia* were placed in five concentrations of NaCl plus one control (0.6, 0.9, 1.4, 1.7 and 2.0 g/L NaCl). Results of the 96 hour toxicity test for sodium chloride showed a mean EC<sub>50</sub> of 1.7 g/L NaCl. Although this fish species is not found in British Columbia, a comparison of the EC<sub>50</sub> result for sodium chloride in this study with the BC AWQG for salinity in freshwater environments shows that the BC guideline of 150 mg/L (0.15 g/L) NaCl would provide protection from NaCl toxicity for this fish species.

The second, more relevant study of NaCl toxicity to fish was conducted by **Diamond et al (2005)** who examined the toxic effects of pulsed or fluctuating contaminant exposures to fathead minnows (*Pimephales promelas*) using concentrations of NaCl. Six day old fathead minnows were exposed to pulses of NaCl at varying concentrations and durations for 7 days (4 g/L for 96 hours, 6 g/L for 24 hours, 8 g/L for 24 hours, and 12 g/L for 3 hours). Results showed significant decreases in survival with the 4 g/L 96 hour, 8 g/L 24 hour and 12 g/L 3 hour pulses compared to



the control. Fish lethality generally occurred within 24 hours of the pulse, however, continued mortality over time was observed in the highest magnitude pulse (12 g/L) when exposure duration was relatively short (3 hours). The lack of change in survival after 72 hours of testing in most treatments indicated that mortality effects of NaCl had stabilized.

**Gomez-Mestre and Tejedo (2003)** conducted several experiments to determine the effect of salinity on embryonic and early larval stages of *Bufo calamita*, a species of frog that breeds in both brackish and freshwater environments in Spain. Through a combination of field transplant and common garden experiments this study showed that water salinity decreased survival probability of individuals in all populations, prolonged their larval period and reduced their mass at metamorphosis. Acute toxicity experiments on embryonic and early larval phases found that for three freshwater populations of *B. calamita* mortality within the first 48 hours remained low until a concentration of 10 g/L total dissolved solids (TDS) was reached and increased steeply afterward. Embryos from the brackish populations of *B. calamita* suffered no mortality at all during the first 48 hours of the experiment at all salinity concentrations (2, 4, 6, 8, 10, 12 g/L total dissolved solids). Differences in tolerance among populations were highly significant during the first 48 hours of exposure. After 72 hours, differences among populations had disappeared. LC<sub>50</sub> values decreased as time went on, suggesting that mortality was not just restricted to the impact of the initial exposure to the osmotic stress but that water salinity had a chronic effect on continuous exposure. Embryos from the brackish water population showed an initial higher tolerance to osmotic stress, but after three days of exposure the acute levels of salinity had the same effect on it as on the freshwater populations and all ended up with LC<sub>50</sub> values ranging from 8 to 10 g/L. Higher osmotolerance of brackish water populations was further supported by the results of the embryonic common garden experiment. Survival of the embryos decreased at the highest salinity on average 62% in the freshwater populations but only 20% in the brackish populations. In BC, there are no approved guidelines for TDS for the protection of freshwater aquatic life; however, there exists a working water quality guidelines for TDS for livestock watering (1 g/L for sensitive species, and 3.0 g/L for other species), and for irrigation (0.5 to 3.5 g/L max, crop and soil dependent). A concentration of 500 mg/L is the BC AWQG for TDS in drinking water.

**Sanzo and Hecnar (2005)** examined the effects of roads salts (NaCl) on wood frog (*Rana sylvatica*) tadpoles exposed for 96 hours. Tests revealed 96 hour LC<sub>50</sub> values of 2636 mg/L by the Spearman-Kaber method and 5109 mg/L by probit analysis. Linear regression revealed a significant decrease in tadpole weight at 96 hours as NaCl concentrations increased. Physical

and behavioral effects in all salt exposure levels were observed with the most pronounced effects seen at higher NaCl concentrations. A 90 day chronic experiment revealed significantly lower survivorship, decreased time to metamorphosis, reduced weight and activity, and increased physical abnormalities with increasing salt concentration (0.00, 0.39, 77.9 and 1030 mg/L). Time to metamorphosis was significantly different from the control at an exposure concentration of 1030 mg/L. Similarly, number of successfully metamorphosed tadpoles per treatment group was only significantly lower than the control in the 1030 mg/L treatment. The BC WWQG for salinity is set at 150 mg/L chloride, which is equivalent to 247 mg/L NaCl assuming both anion and cation is present on an equimolar basis. BC AWQG for chloride in freshwater environments would provide protection from NaCl toxicity for this amphibian species.

**Valenti et al (2006)** conducted a study on the toxicity of chlorine as hypochlorite to freshwater mussels. This study, therefore, is not relevant for assessing toxicity of chloride ion. The objective of the study was to assess the level of risk that chlorine toxicity poses to early life stages of unionids. A series of experiments were conducted with glochidia from various species of freshwater mussels to determine their tolerance of total residual chlorine (TRC). Chronic tests were conducted using juvenile mussels for 21 days. Bioassays with 3, 6 and 12 month old juveniles of *Villosa iris* were conducted to examine the relationship between age and toxicity. In addition a 21 day test with 2 month old juveniles of *Epioblasma capsaeformis*, a United States federally endangered species was conducted to compare sensitivities between species. For the acute toxicity test glochidia were exposed to varying concentrations of calcium hypochlorite (high test hypochlorite, (HTH) in moderately hard reconstituted water as the TRC toxicant. Survivorship was assessed every 24 hours until 72 hours of exposure. Juvenile mussels were exposed to HTH at varying concentrations for 21 days in the chronic toxicity tests. After 21 days the juveniles were removed from test chambers and assess for growth and survivorship. The results of the acute glochidia tests showed that the three endangered species *E. brevidens*, *E. capsaeformis* and *Alasmidonta. heterodon* were slightly more sensitive to chlorine than *Lampsilis fasciola* and far more sensitive than *V. iris* after 24 hours of exposure. At 250 µg TRC/L average survivorship for the more sensitive species (<20%) was nearly half that of the respective value for *L. fasciola* (35%) and less than a third for *V. iris* (66%). In concentrations of 30 µg TRC/L and lower, survivorship remained greater than 90% for all species after 24 hour except *E. brevidens* (79-87%). All exposed glochidia died at 500 µg TRC/L. After 48 hours of exposure, survivorship in chlorinated treatments differed only slightly for *V. iris* (mean LC<sub>50</sub>=260 µg/L) and *A. heterodon* (mean LC<sub>50</sub>=95 µg/L), although it decreased substantially for *L. fasciola* (mean LC<sub>50</sub>=80 µg/L). Significant declines were observed in experiments with three and six

month old *V. iris* juveniles in the 21 day chronic toxicity tests. Adverse effects were observed at lower concentrations in experiments with three month old juveniles as survivorship declined to 50% at 30 µg TRC/L. Survivorship for six month old juveniles remained =90% in concentrations as high as 120 µg/L and was significantly lower than the control only at concentrations =250 µg/L TRC/L. No concentration in the tests caused significant declines in survivorship for 12 month old juveniles and survivorship remained 80% even at 500 µg TRC/L. In terms of growth all three age classes grew significantly less at concentrations =60 µg TRC/L than those in controls. Average growth was reduced relative to controls by 37 to 80% in exposures with TRC concentration of 30 to 120 µg/L and by 90% in exposures of 250 µg/L and greater. The lowest observed adverse effect concentrations for three-month old *V. iris* was determined to be 30 µg/L and 60 µg/L for 6 and 12 month old *V. iris* juveniles. Two month old *E. capsaeformis* juveniles were more sensitive than any age class of *V. iris*. Growth was significantly reduced at concentrations of 20 µg TRC/L and higher, as exposed individuals grew less than 20% relative to those in the control. Observed mortalities was considerably high in the tests, as 50% or more of the individuals died at concentrations of 30 µg/L and higher. The lowest observed adverse effect concentration for *E. capsaeformis* approached the US EPA 4-day maximum freshwater water quality criteria of 11 µg/L TRC. All individuals in the 120 µg/L exposure died after 21 days of exposure, whereas those in the control and 5 µg/L had average survivorship of 80 and 100%, respectively. The results of this study show that glochidia are more tolerant of TRC than many aquatic species. In particular, researchers have reported toxicological endpoints for cladocerans that were much lower than those found in this study. This study also showed that juvenile mussels may be able to survive high dose acute exposures; the impact of long-term exposure to low doses may result in sublethal impairment that could lower their chances of surviving the multi-year juveniles stage and being recruited to the reproducing population. Since this study looked at toxicity of total residual chlorine in water no direct comparison can be made to the chloride concentration set to protect freshwater aquatic life in British Columbia.

### 3. Salt Toxicity to Plants

Research found on toxicity of salt to plants focused mainly on agricultural crops (soybeans, barley, etc) and its effect on nutrient uptake, root and shoot growth. Two studies were found to pertain to forest tree species and NaCl toxicity. A significant limitation of many of these studies relative to define acceptable soil-based sodium or chloride thresholds is that the plants were exposed hydroponically (Cramer, 2002; Bayuelo-Jimeniz *et al.*, 2003; Luo *et al.*, 2004) or the concentration was expressed as a concentration in irrigation water (Franklin *et al.*, 2002; Wilson *et al.*, 2006; Apostol *et al.*, 2002).

**Cramer (2002)** evaluated the inhibitory effect of salinity on leaf extension of three different grass species: *Hordeum jubatum* L. (Foxtail Barley), *Hordeum vulgare* L. (Common Barley) and *Zea mays* L. (Sweet Corn). Plants were exposed to NaCl added to a hydroponic solution (0, 40, 80 and 120 mM NaCl) and changes in leaf elongation rate (LER) were measured over time with a displacement transducer. Leaf elongation of plants was reduced immediately by the addition of salinity to the nutrient solution. Initially, LER declined rapidly in response to salinity, but then entered a recovery phase, reaching a steady state rate after 5 hours for all species and salt treatments. The steady-state response was lower than the controls and was proportional to the level of salinity. At lower salt concentrations (40 and 80 mM), distinctions between species was small. However, clear distinctions were found between species responses at 120 mM NaCl. In general *H. jubatum* was more tolerant than *Z. mays*, which was more salt tolerant than *H. vulgare* to these short-term salinity stresses. In contrast, barley was more salt tolerant than maize over the long term. The mechanism of inhibition of LER by salinity as tested by the applied tension technique varied with the species examined affecting either the apparent yield threshold, the hydraulic conductance of the whole plant or both.

**Wilson et al (2006)** studied the physiological processes involved in cowpea differential growth response of four major USA cowpea cultivars to increasing salinity. The effect of salinity on leaf gas exchange of net photosynthetic rate per unit leaf mass (P<sub>nm</sub>), and per unit leaf area (P<sub>na</sub>) and stomatal conductance (g<sub>s</sub>) were examined. Seven salinities ranging from 2.6 to 20.5 dSm<sup>-1</sup> were constructed using NaCl, CaCl<sub>2</sub> and MgSO<sub>4</sub> as the salinization salts. A highly significant salt effect on P<sub>na</sub>, P<sub>nm</sub>, g<sub>s</sub> and specific leaf weight (SLW) was found for all four cowpea cultivars. The salinity level resulting in a maximum leaf gas exchange (C<sub>max</sub>) was determined to be 6.1 dSm<sup>-1</sup> compared to 6.0 dSm<sup>-1</sup> for the vegetative stage and the flowering stage respectively, and C<sub>50</sub> values (salinity level resulting in a 50% reduction of the maximum leaf exchange) was 17.5 dSm<sup>-1</sup> versus 17.8 dSm<sup>-1</sup> for the vegetative stage and the flowering stage respectively.

**Bayuelo-Jimeniz et al (2003)** studied the effects of salinity on four wild (*Phaseolus angustissimus*, *P. filiformis*, *P. microcarpus*, and *P. vulgaris*) and two cultivated (*P. acutifolius* and *P. vulgaris* L.) *Phaseolus* species. Relative growth rate (RGR), unit leaf rate (ULR), leaf area ratio (LAR), specific leaf area (SLA) leaf weight ratio (LWR) and rate of ion uptake were calculated for the period between 10 and 20 days after planting. Salinity stress had a significant effect on root, shoot and total dry weight and root:shoot ratio, and differences among species for all characters were highly significant. In both cultivated and wild accessions, salinity inhibited

shoot growth more than root growth. Shoot dry weight was significantly reduced in all taxa at all salinity levels, whereas root dry weight was significantly reduced at 40 mM NaCl but no further reduction was observed at 80 mM NaCl. At day 20 shoot and root dry weights of *P. filiformis* were reduced to 50 and 25% of control plants by 80 mM NaCl. For all other species, shoot and root dry weights were reduced by 60 -75% and 29-56% respectively of the control values in the 80 mM NaCl treatment. Salinity increased the root:shoot ratio, but these ratios significantly decreased in *P. acutifolius* after 20 days of growth. The effect of salt stress on the number of leaves and leaf area were similar to that on shoot growth in all accessions. Number of leaves at day 20 significantly decreased with increasing salinity and duration of salt stress, resulting in decreased leaf area. After 20 days of salt stress total leaf area was reduced by 44-66% in accessions grown in 40 mM NaCl and 77-91% in those grown in 80 mM NaCl. Salinity stress decreased RGR. Differences in RGR between unsalinized plants and plants treated with 40 mM NaCl were evident after 20 days. In all species RGR decreased with increasing salinity and with the period of exposure. ULR decreased over time in all species, particularly in salinized plants. Salinity reduced LAR in all species. The lowest LAR was found in one of the slowest growing species, *P. angustissimus*. Salinity significantly affected leaf water, osmotic, and turgor potentials as well as stomatal conductance and CO<sub>2</sub> assimilation rate. Tissue concentrations of Cl<sup>-</sup> and Na<sup>+</sup> ions increased significantly in response to salt treatments. In all taxa the concentration of Na<sup>+</sup> increased almost in parallel in stems and roots with increasing salinity, whereas the concentration of Cl<sup>-</sup> increased more in stems and leaves than in roots. Uptake rate of K<sup>+</sup>, Ca<sup>2+</sup>, and Mg<sup>2+</sup> were significantly affected by increasing NaCl. On day 20 salinity had significantly increased the absorption rates of Cl and Na.

**Luo et al (2004)** conducted experiments to determine ion-specific stress effects of Na<sup>+</sup> and Cl<sup>-</sup> on seedlings of cultivated (*Glycine max* L. Merr) and wild soybean (*Glycine soja* Sieb & Zucc.). Results showed that under NaCl stress Cl<sup>-</sup> was more toxic than Na<sup>+</sup> to seedlings of *G. max*. A positive correlated was found with the content of Cl<sup>-</sup> in the leaves and ionic stress resulting in injury in *G. max*. A negative correlation was found between Cl<sup>-</sup> content in roots and plant injury. Seedlings of *G. max* cultivars (salt-tolerant Nannong 1138-2 and salt sensitive Zhongzihuangdou-yi) and two *G. soja* populations were exposed to 150 mM Na<sup>+</sup>, Cl<sup>-</sup> and NaCl respectively. *G. max* Nannong 1138-2 and Zhongzihuangdou-yi were damaged much more heavily in the solution of Cl<sup>-</sup> than in that of Na<sup>+</sup>. The leaves were found to be more sensitive to Cl<sup>-</sup> than to Na<sup>+</sup>, and salt tolerance of these two *G. max* cultivars was mainly due to successful withholding of Cl<sup>-</sup> in the roots and stems to decrease its content in the leaves. A reverse response was observed to isoosmotic stress of 150 mM Na<sup>+</sup> and Cl<sup>-</sup> was shown in *G. soja* populations, their leaves were not

as susceptible to toxicity of  $\text{Cl}^-$  as that of  $\text{Na}^+$ . In this case salt tolerance was mainly due to successful withholding of  $\text{Na}^+$  in the roots and stems to decrease its content in the leaves. The results of this experiment indicate that *G. soja* have advantages over *G. max* in those traits associated with the mechanism of  $\text{Cl}^-$  tolerance, such as its withholding in roots and vauoles of leaves.

**Franklin et al (2002)** compared the effects of  $\text{NaCl}$  and  $\text{Na}_2\text{SO}_4$  on the nutrient status of jack pine (*Pinus banksiana*). Twenty-eight week old germinated seedlings of *P. banksiana* were exposed to 60 mM  $\text{NaCl}$  and 60 mM  $\text{Na}_2\text{SO}_4$  and maintained for 10 weeks. Salt treatments decreased shoot dry weights and shoot elongation rates. Shoot dry weight was reduced by 36% in 60 mM  $\text{Na}_2\text{SO}_4$  treated seedlings and by 40% in 60 mM  $\text{NaCl}$  treated seedlings. The percentage of seedlings exhibiting terminal bud flush decreased slightly from 98.4% in the control seedlings to 91.9% and 90.2% in  $\text{Na}_2\text{SO}_4$  and  $\text{NaCl}$  treatments, respectively. Growth and injury of seedlings were found to be more affected by sodium chloride than sodium sulfate. While a small but significant amount of needle necrosis (8%) occurred in  $\text{Na}_2\text{SO}_4$  treated seedlings,  $\text{NaCl}$  treatment resulted in a significantly greater necrosis of approximately 21% of the needle dry weight. Chlorophyll *a* and total carotenoid content were reduced in  $\text{NaCl}$  treated plants. Sodium chloride treated plants exhibited a delay in flushing of the terminal buds, reduced carotenoid content, extensive needle necrosis, and elevated levels of K, Mg, Mn, N and P in the shoot.  $\text{Na}_2\text{SO}_4$  treated seedlings resulted in reduced shoot calcium and potassium concentrations, while those of nitrogen and phosphorus increased. Needle necrosis was correlated with tissue  $\text{Na}^+$  only in sodium chloride treated plants, and no relationship was found between growth or necrosis, and tissue levels of  $\text{Cl}^-$  or nutrition elements. This study showed that greater toxicity of sodium chloride in jack pines was not due to nutrient deficiency.

**Apostol et al (2002)** examined the response of 6 month old jack pine seedlings to boron and salinity ( $\text{NaCl}$  and  $\text{Na}_2\text{SO}_4$ ) treatments. During 4 weeks of exposure 60 mM  $\text{NaCl}$  and 60 mM  $\text{Na}_2\text{SO}_4$  significantly decreased survival, new shoot length, number of new roots, shoot to root dry weight ratio and transpiration rates. When applied in absence of the salts Boron had little effect on the measured variables. However, when applied together with salts, Boron decreased seedling survival, increased needle injury and altered tissue elemental concentrations in jack pine seedlings. In 2 mM Boron treatment Boron concentration was higher in shoots than in the roots. However, when 2 mM Boron was present in  $\text{NaCl}$  and  $\text{Na}_2\text{SO}_4$  treatments, shoot Boron concentration declined and greater proportion of Boron accumulated mostly in the roots. Based on the electrolyte leakage and needle necrosis data,  $\text{Cl}^-$  appears to be the major factor

contributing to seedling injury and Boron aggravates the injurious effects of NaCl. The authors suggest that Cl<sup>-</sup> may contribute to Na and B toxicity in jack pine by altering cell membrane permeability leading to increased Na concentration in the shoots.

#### 4. Conclusions

Very limited research has been conducted since 2002 on the toxicity of sodium chloride to plants and wildlife. Seventeen relevant research papers were identified, although very few of these were directly related to sodium chloride (NaCl) toxicity to terrestrial and freshwater life.

Table 1 summarizes the toxicity data applicable to plant and animal species that might be surrogates for those found in BC, and compares toxicity values to the draft soil matrix salinity standards (Bright *et al*, 2002) and the provincial Working and Approved Water Quality Guidelines (MoE, 2006). Studies involving simultaneous exposure to one or more additional substance are not summarized in Table 1, since these are not deemed to provide comparable data for assessing salt toxicity in isolation.

Table 1: Summary of Toxicity Test Results for BC Species				
Study	Toxicity Test	Organism	Result	Guideline
Davies and Hall (2006)	48 hour acute Cl <sup>-</sup> toxicity test in water with varying Ca:Mg ratios.	<i>Daphnia magna</i>	LC50 results: 3,136 mg/L Cl <sup>-</sup> (0.7 Ca:Mg); 3,222 mg/L Cl <sup>-</sup> (1.8 Ca:Mg); 3,137 mg/L Cl <sup>-</sup> (7.0 Ca:Mg);	BC Approved Water Quality Guideline for chloride: 150 mg/L
Diamond <i>et al</i> (2005)	Pulsed exposure of NaCl in water at varying concentrations and durations for 7 days	Fathead minnows ( <i>Pimephales promelas</i> )	Decreased survival with 4 g/L NaCl for 96 hours, 8 g/L for 24 hours and 12 g/L for 3 hours	BC Approved Water Quality Guideline for chloride: 150 mg/L
Sanzo and Hecnar (2005)	96 hour acute NaCl toxicity test	Wood frog tadpoles ( <i>Rana sylvatica</i> )	LC50 results: 2,636 mg/L by Spearman-Kaber and 5,109 mg/L by probit analysis	BC Approved Water Quality Guideline for chloride: 150 mg/L
Sanzo and Hecnar (2005)	90 day chronic NaCl toxicity test using concentrations of 0.0, 0.39, 77.8, and 1030 mg/L NaCl	Wood frog tadpoles ( <i>Rana sylvatica</i> )	Lower survivorship, decreased time to metamorphosis at 1030 mg/L NaCl, as well as reduced weight and activity, and increased physical abnormalities with	BC Approved Water Quality Guideline for chloride: 150 mg/L

			increasing salt concentration.	
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Similarly, experiments involving soil solution or hydroponic exposures are excluded from the Table 1 summary, since the data are of limited relevance in the context of this exercise.

Additional research not directly related to BC species or sodium chloride toxicity was included in this review to provide complimentary information for risk assessors on salt interactions with other toxicants, and on altered bioavailability of environmental contaminants to terrestrial and aquatic life.

Based on a review of the most recent published scientific information, it is concluded that toxicological data for salt which became available post-2002 would not substantively change the draft matrix soil standards derived in 2002 for either sodium (Na+) or chloride (Cl-).

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