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ALBERTA OIL SANDS ENVIRONMENTAL RESEARCH PROGRAM

The Impact of Saline Waters upon Freshwater Biota
(A Literature Review and Bibliography)

AF 1.2.1

January 1977

Sponsored jointly by

Alberta ENVIRONMENT

Environment Canada Environnement Canada

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ALBERTA OIL SANDS ENVIRONMENTAL RESEARCH PROGRAM RESEARCH REPORTS

These research reports describe the results of investigations funded under the Alberta Oil Sands Environmental Research Program, which was established by agreement between the governments of Alberta and Canada in February 1975.

Enquiries pertaining to the Alberta-Canada Agreement or requests for information about the research reports should be directed to:

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The Impact of Saline Waters Upon Freshwater Biota
(A Literature Review and Bibliography)

Sub-project AF 1.2.1

AOSERP Report 8

LETTER OF TRANSMITTAL

The Hon. D. J. Russell
Minister of the Environment
228 Legislative Building
Edmonton, Alberta

and

The Hon. R. Le Blanc
Minister of Fisheries and Environment
Parliament Buildings
Ottawa, Ontario.

Sirs:

Enclosed herein is the report on "The Impact of Saline Waters Upon Freshwater Biota", which is a literature review and bibliography.

This report was prepared for the Aquatic Fauna Technical Research Committee of the Alberta Oil Sands Environmental Research Program, under the Alberta-Canada Agreement of 28 February 1975.

Respectfully,

W. Solodzuk, P.Eng.
Chairman, Steering Committee, AOSERP
Deputy Minister, Alberta Environment

J. S. Tener, Ph.D.
Member, Steering Committee, AOSERP
Assistant Deputy Minister
Environmental Management Services
Environment Canada.

The Impact of Saline Waters Upon Freshwater Biota

(A Literature Review and Bibliography)

DESCRIPTIVE SUMMARY

ABSTRACT

The impact of saline waters upon freshwater biota, having special reference to the AOSERP study area, is reviewed. Available information on water quality in the AOSERP study area indicates that: the natural regional surface water quality does not appear to have any toxic effects, and chloride concentrations of groundwaters are quite variable although those associated with mine depressurization are apparently low to moderate. The effects of saline waters in general on major groups of organisms including aquatic plants, aquatic invertebrates, fish, birds and mammals are discussed. Toxicity summaries for individual ions are presented. The report also contains an extensive bibliography of scientific references pertaining to the impact of saline waters upon freshwater biota.

BACKGROUND AND PERSPECTIVE

Test pits excavated in the AOSERP study area indicate that a significant inflow of groundwater can be expected in surface mining operations in the oil sands area. In order to prevent the accumulation of groundwater during surface mining, the local water table must be lowered by pumping. Subsequently, all groundwater seepage to the mine pit area is controlled and removed. The disposal of this groundwater may present a problem because of its salinity.

The purpose of this literature review and bibliography on saline waters was to provide the Aquatic Fauna Technical Research Committee and researchers with a review of the literature on the effects of saline waters on biota. The bibliography was deemed an important part of the research effort since it would form a basic reference source on the current literature for future researchers as well as those now working on the problem.

ASSESSMENT

This study was designed to serve as an initial phase of aquatic toxicological investigations begun by the Aquatic Fauna Technical Research Committee. It will serve as a basic reference source for future researchers.

The report was reviewed by the Aquatic Fauna Technical Research Committee and has been recommended for publication. The content of this report does not necessarily reflect the views of Alberta Environment, Fisheries - Environment Canada, or the Oil Sands Environmental Study Group. The mention of trade names for commercial products does not constitute an endorsement or recommendation for use.

The Aquatic Fauna Technical Research Committee of the Alberta Oil Sands Environmental Research Program accepts "The Impact of Saline Waters Upon Freshwater Biota (A Literature Review and Bibliography)" and thanks the author, Kazimierz Machniak and Aquatic Environments Ltd., Calgary, for their contributions.

Ron R. Wallace, PhD.
Chairman,
Aquatic Fauna Technical
Research Committee

ALBERTA OIL SANDS ENVIRONMENTAL RESEARCH PROGRAM STUDY AREA

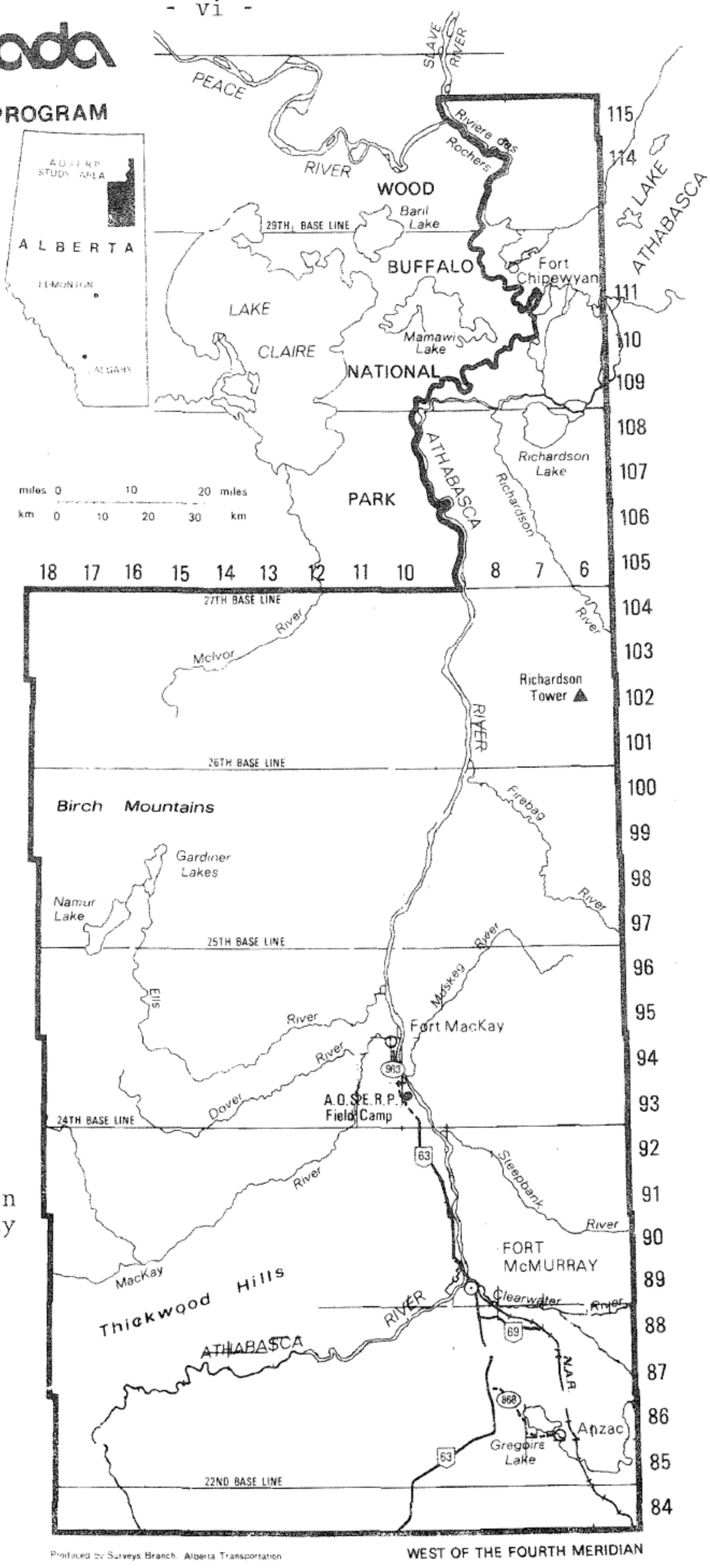

FIGURE 1. Location of the AOSERP study area.

THE IMPACT OF SALINE WATERS UPON FRESHWATER BIOTA
(A Literature Review and Bibliography)

by

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ALBERTA OIL SANDS ENVIRONMENTAL RESEARCH PROGRAM

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1. INTRODUCTION

The Aquatic Fauna Technical Research Committee of the Alberta Oil Sands Environmental Research Program (AOSERP) requested that Aquatic Environments Limited review the literature with respect to the impact of saline waters upon freshwater biota, having special reference to the AOSERP study area. The effects of saline waters in general on major groups of organisms is discussed. Toxicity summaries for individual ions are presented but the values given need not necessarily be applicable as "standards" for saline discharges and receiving waters in the AOSERP area. The lack of available data particularly on the chemistry of saline discharges limits somewhat the assessment of the potential effects of saline waters in the study area.

2. WATER QUALITY IN THE AOSERP STUDY AREA

SURFACE WATER

The available surface water quality data as well as biological data for the AOSERP study area have been compiled and summarized by Renewable Resources Consulting Services Ltd. (RRCSL, 1975a; b; c) and Griffiths (1973). Some current surface water data (primarily on the Athabasca and Clearwater rivers) are also available from the Water Quality Branch of Environment Canada (H. Block, District Resource Officer, Inland Waters Directorate, pers. comm.). The following discussion is limited to those natural waters with reported high conductivities and chloride levels.

Specific conductance values of lakes in the area are generally less than 250 micromhos/cm @ 25°C indicating low dissolved salt contents. However, Saline Lake has been reported to have conductivities ranging from 3,000 to 4,800 micromhos and chloride levels of 600 to 1,570 mg/1 (Aquatic Environments Limited, unpublished data). A spring located at Saline Lake had conductivities of 141,900 to 179,480 micromhos (salinity of 75-90‰) and chloride levels of

40,280-60,000 mg/1 with sulphate levels of 3,950 to 4,500 mg/1.

The water quality parameters of rivers and streams in the AOSERP study area show greater ranges than those found in most lakes. Griffiths (1973) reported conductivities of streams and rivers in the tar sands area generally ranged from 27 to 550 micromhos. He did, however, sample two locations on the Christina River (Sec. 33, Twp. 88, Rge. 7, and Sec. 26, Twp. 87, Rge. 6) where conductivities were 870 and 1,050 micromhos and chlorides of 100 and 241 mg/1, respectively.

Ellis's (1937) studies of inland fresh waters indicated that the specific conductance of streams and rivers supporting a good mixed fish fauna lay, in general, between 150 and 500 micromhos/cm @ 25°C, except in streams of the western plains and desert areas, particularly those carrying more alkaline natural waters. On the basis of his studies, Ellis concluded that conductances in excess of 1,000 micromhos in most types of streams, or in excess of 2,000 micromhos in the alkaline western streams,"are probably indicative of the presence of acid or salt pollution of various kinds". Ellis (1943) also found that a specific conductance of 4,000 micromhos is approximately the upper limit of ionizable salts tolerated by fish in mixtures of Na, Mg, and Ca compounds in gas well waters.

The level of dissolved NaCl in the Christina River appears to be about 3-4 times greater than in any other river.

However, regional water quality of AOSERP rivers appears good with no toxic or harmful effects from any of the parameters measured (RRCSL, 1975b).

Syncrude Canada Ltd. (1975) reports levels of total dissolved solids in Poplar Creek surpassing 1,000 mg/l (1,778 mg/l) on one occasion and levels of the chloride ion to regularly surpass 200 mg/l and on one occasion greater than 700 mg/l. The chloride levels are similar to chloride levels found in Beaver Creek (RRCSL, 1974b). The relatively high chloride level in Poplar Creek appears to be natural.

GROUNDWATER

At present, only limited data are available on the chemistry of groundwater in the AOSERP area. The Groundwater Division of the Alberta Research Council (ARC) is, or has analysed the water quality of 35 wells in the area (the results on 7 wells are presented in Table 1 and Appendix I). Total dissolved solids (TDS) range from 196-3,182 mg/l in these wells. The U.S. Geological Survey defines saline water as containing more than 1,000 mg of dissolved solids per litre of solution (Krieger *et al.*, 1957). Conductivities of these well waters range from 280-6,000 micromhos/cm at 25°C with chlorides of 12-338 mg/l. West of the Athabasca River, chloride concentrations in groundwater as high as 118,636 mg/l have been reported (RRCSL, 1975c). The formation containing this high chloride groundwater is not, however, involved in mine depressurization schemes. Groundwater data for the area,

TABLE 1. Ranges of some chemical parameters of groundwater determined for several ARC wells[1] in the AOSERP study area (from Groundwater Division, Alberta Research Council) compared with seawater (Smith, 1974) and oil field brines (Reid *et al.*, 1974).

Groundwater

Parameter	Range
TDS (mg/l)	196-3,182
Hardness (as $CaCO_3$)	33-599
Alkalinity (as $CaCO_3$)	110-1,432
Conductivity (micromhos/cm @25°C)	280-6,000
Lab pH	7.1-12.3
Minor Constituents	
Iron (mg/l)	4-7,100

Major Constituents (in mg/l)

	Groundwater	Seawater	Oil field brine
Calcium (Ca)	2-240	409-417	1,000-120,000
Magnesium (Mg)	0-46	1,292-1,301	500-25,000
Sodium (Na)	18-1,000	10,720-10,800	12,000-150,000
Potassium (K)	2-533	393-405	30-4,000
Carbonate (CO_3)	0-67	-	-
Bicarbonate (HCO_3)	0-1,693	137-153	0-1,200
Sulphate (SO_4)	0-1,496	2,701-2,724	0-3,600
Chloride (Cl)	12-388 (118,636)[2]	19,353	20,000-250,000
Nitrate (NO_3)	0-2		
Hydroxide (OH)	160-460		
Silica (SiO_2)	0-6		
Calcium (Acid)	3-700		
Magnesium (Acid)	0-251		

[1]The available data (Appendix I) limits the high range characteristics of groundwater.

[2]RRSCL (1975c).

including chemical analyses, are available in the Groundwater Division (ARC) data files (R. Green, Chief of the Earth Sciences Branch, ARC, pers. comm.). Many leaseholders in the oil sands area have conducted groundwater analysis programs. Syncrude Canada Ltd. has also conducted groundwater toxicity studies (these unpublished studies as yet are unavailable).

Hydrogeological mapping in the AOSERP area has been carried out by Dr. Georgi Ozoray of the Alberta Research Council (Groundwater Division). Dr. Ozoray (pers. comm.) states that the results of his work (field mapping and groundwater flow patterns) are not all published as of yet, and hence are not available for this report.

2.1 THE GROUNDWATER PROBLEM IN OIL SANDS DEVELOPMENT

A test pit excavated on the Syncrude Lease in the late summer of 1972 illustrated that a significant inflow of groundwater could be expected in commercial mining operations in the tar sands area (Syncrude Canada Ltd., 1973a). In order to prevent the accumulation of groundwater in the mining operation, the local water table must be lowered by pumping. Subsequently, all groundwater seepage to the mine pit area is controlled and removed (dewatering). The disposal of this groundwater may present a problem because of its salinity.

Northwest Hydraulic Consultants Ltd. (1975) discussed the hydrologic impacts of oil sands development. Some of the potential impacts indicated with regards to saline waters were:

"1. Dewatering operations would draw down the water table, resulting in groundwater flow into the area increasing. Lower hydrostatic pressures may allow saline groundwater to enter fresh water aquifer zones.

"2. Bitumen extraction would result in increased porosity and permeability of reservoir. Groundwater flow rates could increase; connections with deeper saline water bearing formations could result.

"3. Deep well injection (disposal) could exert such pressures that the flow rate from existing saline springs would increase, tending to pollute surface and groundwater systems."

Renewable Resources Consulting Services Limited (1975c) summarized what little information is available regarding mine pit dewatering operations:

"The dewatering problem associated with an open pit mining operation has resulted in test drilling and aquifer testing on many leases. The bulk of this data as well as information on the methods and technology of dewatering has remained confidential. The composition and quantity of groundwater removed during dewatering and mine pit depressurization operations is expected to vary considerably from lease to lease, reflecting variations in local permeability of the geologic formations of each lease. The formation permeability is dependent on the lithology and structure of the particular formation involved. In general, saline waters can be expected from Devonian sediments, brackish to saline to moderately fresh water from the McMurray and Wabiskaw Formation and fresh water from the Grand Rapids Formation."

Syncrude's operation at the Mildred Lake mine site discharges saline groundwater into Beaver Creek. Since Syncrude Canada Ltd. is required to monitor its discharge of saline groundwater, more detailed information may be obtained from the company in future. Other surface mining and in-situ operations in the oil sands area also have to cope with this type of groundwater.

2.2 DISPOSAL OF SALINE WATERS

2.2.1 Present License Requirements for Disposal

Approval for disposal of saline groundwater is required by the Standards and Approvals Division of the Alberta Department of the Environment. The Department has, at present, no specific requirements for the disposal of saline waters except for those specified in Syncrude Canada Ltd.'s letter of permission allowing the discharge of saline water to Beaver Creek (J. Defir, Director of Standards and Approvals, pers. comm.).

The discharge is to be controlled for chloride concentration, and oil and grease in Beaver Creek. Other monitoring requirements are also stipulated for both the effluent and the receiving stream. These include: pH, suspended solids, C.O.D., phenolics, anion concentration (sulfate, bicarbonate, etc.), metals (vanadium, iron, copper, nickel, arsenic, etc.), and toxicity evaluation.

A copy of the letter of permission issued to Syncrude Canada Ltd. in 1976 is presented in Appendix II.

2.2.2 Disposal Practices in Other Areas

Deep-well injection is used in oil-producing states for underground disposal of the saline water brought to the surface with the oil (Warner, 1965). Deep-well injection is not actually a disposal method, but rather a storage method. Subsurface brine (saline) disposal can be categorized as confinement or containment; confinement is the placement of brines in a horizon where any movement can be controlled or monitored, while containment is the placement that precludes the movement of brines out of a formation or zone.

Formations into which brines are often pumped for disposal are called salaquifers, and these zones consist of permeable sedimentary rock. Some information needed before such a zone can be used for disposal operations is as follows (Collins, 1971):

1) The size of the zone.

2) The migration of the brine in the zone (whether it might reappear in another zone or perhaps migrate to the surface).

3) The mechanisms that control movement in a given salaquifer or out of it.

4) The steps necessary to assure containment or confinement of the brine within the salaquifer.

Disposal into salaquifers is limited and cannot continue indefinitely; therefore, other methods of disposal need development.

Various methods of disposal of saline water in East Texas oil fields included (Morris, 1960):

1) evaporation in open pits,

2) evaporation in open pits with the aid of heat (gas flares),

3) continuous discharge of saline water into surface drainage and streams,

4) storage of saline water in earthen pits within the field, with controlled discharge into streams during periods of heavy rainfall,

5) transportation of saline water from the field by pipeline or open ditches to a body of salt water,

6) the injection of saline water into subsurface formations.

The discharge of oil treater wastes (with a TDS of between 3,000 and 4,500 mg/1) is not permitted into any of the flowing waters in Wyoming which support a fisheries. In these situations, the oil treater water has to be reinjected (R. Millis, Supervisor Water Quality Lab, Wyoming Game and Fish Dept., pers. comm.).

2.3 SOURCES OF SALINE WATERS

Oil field brine (saline water) is one of the major sources of surface fresh water and groundwater pollution in oil producing states. The high salinity of most oil field brines is caused by soluble salts. The major salts contributing to brine salinity are the sulfates (SO_4^{-2}), bicarbonates (HCO_3^-), and chlorides (Cl^-) of the cations, sodium ($Na+$), calcium ($Ca+^2$), and magnesium ($Mg+^2$)(Reid and Streebin, 1972; Reid *et al.*, 1974). Chlorides in oil field brines generally range from 20,000 to 250,000 mg/l while chlorides in AOSERP groundwaters range from a low of 12 mg/l (ARC, Appendix I) to a high of 118,636 mg/l (RRCSL, 1975c).

In addition to salinity, certain nonionic materials are common to oil field brines, such as oil, dissolved organics, and dissolved gases. The groundwaters of the tar sands area are similar in this respect because they too contain hydrocarbons and other residues which have leached out from the oil bearing strata. Oil field waste waters contain as much as 0.1 to 0.33% oil by volume (McKee, 1956). Although the dissolved gases and components of the oil are potentially hazardous, these substances tend to dissipate before they accumulate and reach toxic levels. Should oil accumulate to a concentration of 3-5 mg/1, freshwater fish which are especially sensitive to oil components may begin to experience toxic effects (Water Quality Criteria, 1968).

Some other sources of saline waters include the following:

Runoff from agricultural irrigation (Skogerboe *et al.*, 1974a, b).

Strip mining waters (Lewis and Peters, 1955).

Potash mine wastes (Albrecht, 1954; Schmitz, 1956; 1957a; b; 1958; Hammer *et al.*, 1975).

Runoff from street deicing salt (Judd, 1969; Hanes *et al.*, 1970).

Sea water intrusion (Butler and Popham, 1958).

Metal mine effluents (Clarke, 1974a).

Dewatering operations (Todd, 1974).

Industrial wastes (Gregory, 1974; Thompson, 1974; Clarke, 1974b).

Drilling fluid sump (Falk and Lawrence, 1973; Land, 1974).

Springs (Hager and De Geer, 1965; Rueffel, 1966; Royer, 1966).

There are natural saline waterbodies (including springs) in the AOSERP area which, from a scientific point of view, offer a broad spectrum of salinities where the impact of salinity on the flora and fauna may be studied.

2.4 SUMMARY

The available information on water quality in the AOSERP study area is reviewed:

1) Regional surface water quality of the AOSERP area appears good with no toxic or harmful effects from measured parameters.

2) Chloride concentrations of groundwaters in the area are quite variable although those associated with mine depressurization are apparently low to moderate.

3) At present, there are no specific regulations for the disposal of saline waters in the province.

3. TOXICITY OF SALINE WATERS

Chlorides as a Toxic Component of Saline Waters

"There is no convincing evidence that chloride ions have any specific toxicity. The toxicity of physiologically unbalanced solutions of various chlorides (salts), including sodium chloride (NaCl), is apparently attributable to the specific toxicity of the cations present and not to any toxicity of the chloride ions (anions). The specific toxicity of the different cations varies greatly. Sodium, calcium, strontium, and magnesium are among the least toxic, while the chlorides of such metals as copper, mercury, cadmium, zinc, and lead are among the most toxic. Because the toxicity of the chloride ion has been found to be so much less significant than that of the cations associated with it, an expression of chloride ion concentration in any water has no meaning as far as toxicity is concerned" (Ohio River Valley Water Sanitation Commission--ORSANCO, 1956).

Physiologically balanced salt solutions, such as sea water, may be harmful to freshwater organisms because of their excessive overall salt content and osmotic pressure rather than the specific toxicity of any particular ions present. However, when consideration is given to mixed salt solutions of unknown and varying composition such as oil field waters and other industrial wastes, their chloride ion content is not a reliable index of osmotic strength. For example, a waste water containing a large amount of sodium sulfate (Na_2SO_4) can be much more active osmotically than another water with a much higher chloride ion content but containing only sodium and calcium chlorides (ORSANCO, 1956).

Each particular mixture of chlorides and other salts found in polluted waters should be considered individually, with attention being given to the specific toxicity of all the components, their synergistic and antagonistic relationships or interactions, and the osmotic pressure of the waters under consideration.

Tolerance to Salinity

Many freshwater animals are unable to tolerate much salinity and are thus eliminated by salts (Beadle, 1957). On the other hand, many aquatic invertebrates, particularly among Oligochaeta, prosobranch Gastropoda, Crustacea, Odonata, Hemiptera, Coleoptera, Diptera, and Trichoptera can tolerate quite high salinities (Hynes, 1970).

Different organisms vary in their ability to tolerate, let alone maintain, normal activity in high concentrations of dissolved substances. For example, some fishes are equally at home in sea water and in fresh water, but others are reported to die in water where the total dissolved materials are present in no higher concentrations than in fish blood. The widely prevalent notion that the limit of salinity tolerance of freshwater fishes is, in general, the equivalent of 7,000 mg NaCl is misleading (ORSANCO, 1956). While some salts can often be tolerated in relatively high concentrations, others are toxic at much lower concentrations.

Many authors today stress that study of acute toxicity is not sufficient, that there must be more concern with sublethal effects (e.g., Alderdice, 1967; Cairns, 1966; Fujiya, 1965). In order to predict any sublethal effects (i.e., stress) of saline waters, it is important to understand the modes of action of individual ions. Since the range of capacity of different species to regulate their internal environment tends to bear a relation to the stress imposed by their external environment, osmotic and ionic regulation should be considered. These subjects are of fundamental interest in the understanding of salinity tolerance and toxicity, and therefore, much of the literature has been included in the extensive bibliography of this review. If the modes of action of saline waters were better understood, we could more accurately predict their effects as pollutants.

Osmoregulation also demonstrates some of the forms of adjustment (or adaptation) that organisms often make in order to tolerate salinity. In many instances, however, the limit of salinity tolerance of a given organism may be modified by other factors (i.e., temperature, oxygen, season, size, water quality, etc.).

3.1 THE IMPACT OF SALINE WATERS UPON FRESHWATER BIOTA

The effects of saline waters including the toxicity of individual ions to freshwater biota is reviewed under separate headings of organisms. Those particular species or genera known to occur within the AOSERP study area are denoted with an asterisk (*).

3.1.1 Aquatic Plants

It has long been known that high concentrations of chlorides are related to plant distribution. This may well be an osmotic effect for it has been demonstrated there are optimum osmotic concentrations for plants (Moyle, 1956). Several salts are necessary for plants, for example potassium salts, but others are detrimental not to say toxic, for instance sodium chloride (NaCl).

3.1.1.1 Algae

Chlorides do not appear to limit algal production directly in nature but, in the form of NaCl, play a major part in the determination of the kinds of algae which can grow in the water. "The importance of chloride alone may be over-emphasized because of the relative ease of estimating its concentration and so using it as an index of salinity" (Lund, 1964).

The effect of various salinities on the photosynthetic rate of freshwater alga (*Scenedesmus obliquus*)* was studied by Vosjan and Siezen (1968). It was observed that with increasing salinity, photosynthesis decreases gradually until photosynthesis and respiration are about equal near 30‰ salinity.

The range of total salt concentration and ionic ratios over which alga can live is often considerable but as the conditions become more extreme such algae grow less well and meet with competition from species adapted to these conditions. Wood and Straughan (1953) observed reduction in size, as well as sexual retardation in *Lemanea fucina* growing in a stream where it was exposed to brackish water twice daily. However, a few freshwater algae, such as *Cladophora glomeruta*, may grow very successfully in estuarial waters in apparent response to the increased concentrations of limiting salts or other nutrients (Hamel, 1924). *Chlamydomonas** seem to adjust readily to gradual changes in salt concentrations (Flowers and Evans, 1966).

The ratio of mono- to divalent ions can affect the constitution of the flora. "When the amount of NaCl increases, Na^+ may have more effect than Cl^-, although most accounts stress the effect of the latter" (Lund, 1964). Sodium chloride is one of the most important salts which limits the distribution of diatoms. Kolbe (1927, 1932) divides diatoms on the basis of salinity tolerance into four groups--"polyhalobiens", "euhalobiens", "mesohalobiens", and "oligohalobiens".

The polyhalobiens are species which can stand a salt concentration greater than that of the sea. To this group belongs *Navicula longirostris*.

The euhalobien species develop best in water with total salt concentration of 3-4% (NaCl 1.7-2%). To this group belong the marine and brine water species.

The mesohalobien species have their optimum in a total salt concentration of 0.5-2% (NaCl 0.2-1.5%). To this group belong such brackish-water species as *Achnanthes brevipes* var. *intermedia**, *Amphora coffeaeformis*, *Nitzschia hungarica**, *Stauroneis salina* var. *latior*, *Navicula salinorum**, *N. integra*, *N. pygmaea*, and *Diploneis interrupta*.

The oligohalobien species have their optimum condition in water with a very low salt concentration. This group is subdivided into three subgroups. First, those species which have their best development in water with a small amount of salt, such as *Navicula cincta**, *Anomoeoneis sphaerophora**, *Caloneis amphisbaena*, *Cyclotella meneghiniana*, *Diatoma elongatum**, and *Navicula hungarica*. These are known as halophilic species. Second, those freshwater species, such as *Diploneis elliptica*, *Cymbella lacrustris**, *Gyrosigma attenuatum*, *Melosira arenaria*, and *Hantzschia elongata*, which are not sensitive to a little salt, are known as indifferent species. Third, those which live in very pure water and dislike salt, are halophobs. To this group belong most species of *Eunotia**, *Actinella*, *Stenopterobia*, *Tabellaria flocculosa**, and *Asterionella ralfsii*, and some of the species of *Pinnularia** and *Frustulia*.

Diatoms are particularly suitable for the investigation of saline waters, because they abound in all waters and are regarded as good indicators of salinity (Ziemann, 1968). The quantitative relationship between biological conditions of diatom communities and salinity in several inland saline waters were examined by Ziemann (1967, 1968). The observations were made in the rivers Werra and Wipper which had received salt for 50 years from potash mine wastes.

In the course of the rivers Werra and Wipper, differences of salinity exist within the range from fresh water up to 45‰ salinity in the River Werra and up to 30‰ salinity in the River Wipper. At different salinities, diatom communities which represent distinct indications of different salt concentrations in stretches of the rivers were found (h:halobiont diatoms)(Table 2).

TABLE 2a. Diatom communities representative of different salt conditions (from Ziemann, 1968).

Freshwater stretches	Stretches with Moderate Salt Concentration	Stretches with High Salt Concentration
Navicula avenacea	*Navicula avenacea*	*Navicula salinarum* (h)*
N. gregaria	*N. salinarum* (h)*	*N. pygmaea* (h)
*N. cryptocephala**	*N. gregaria*	*N. gregaria*
N. gracilis	*N. cryptocephala**	*Amphora coffaeiformis* (h)
Surirella ovata	*Surirella ovata*	*Nitzschia apiculata* (h)
Synedra ulna	*Nitzschia apiculata* (h)	*N. hungarica*
Cymbella ventricosa	*N. hungarica* (h)	*N. frustulum* var. *subsalina* (h)
Rhoicosphenia curvata	*N. frustulum* var. *subsalina* (h)	*Gyrosigma peisonis* (h)
*Nitzschia palea**	*Rhoicosphenia curvata*	*Synedra pulchella* (h)
*Diatoma vulgare**	*Cyclotella meneghiniana* (h)	*Surirella ovata*
Gomphonema olivaceum		*Cydotella meneghiniana* (h)
Melosira varians		

TABLE 2b. Toxicity of various salts to the diatom, *Nitzschia linearis* (from Patrick *et al.*, 1968).

Salt	Toxicity (120 hr TLm mg/1)	Comments
$CaCl_2$	3,130	Soft dilution water
KCl	1,337	50% reduction in number of
NaCl	2,430	cells produced TLm
		for diatoms.

In the freshwater reaches of rivers, halobiont diatom species are not found. With increase of salinity, the frequency of halobiont species increases, whereas the freshwater species decrease with almost any halobiont species remaining when the salinity is very high. The relationship between diatom association and salinity may be expressed as the quotient of the frequency of halobiont diatom species to the frequency of all diatom species. This relationship between salinity and diatom flora is only valid for salinities higher than 1.5-2‰.

Patrick *et al.* (1968) determined median tolerance limits for several chloride salts on the diatom, *Nitzschia linearis*. The 120 hr TL_m's were based on a 50% reduction in numbers of cells produced. They found that the TL_m's for $CaCl_2$, KCl, and NaCl were 3,130 mg/l, 1,337 mg/l, and 2,430 mg/l, respectively (Table 2b). It was also observed that diatoms appear to be more sensitive to chloride salts than fish.

3.1.1.2 Higher Plants (in particular Macrophytes)

Rawson and Moore (1944) found that higher plants first appeared at a salinity of 20,000 mg/l total dissolved solids (TDS) and increased in number of species with decreasing salinity below that concentration. *Brachionus plicatilis, B.p. spatuosus, B. satanicus, Conochilus unicornis,* and *Pedalia* sp. were taken in lakes with salinities (TDS) ranging from 10,000 to 30,000 mg/l. They presented a table showing how the number of species increases with decreasing salinity.

Ungar (1965) presents a qualitative and quantitative description of the vegetation of the Big Salt Marsh, Stafford County, Kansas. Groundwaters in the vicinity of the Big Salt Marsh are highly mineralized and contain high concentrations of chloride (13,050 - 24,440 mg/l), principally as NaCl. There is a discussion of the salt tolerance of marsh species. The species which were found to have the greatest salt tolerance were *Suaeda depressa, Sesuvium verrucosum, Salicornia rubra,* and *Distichlis stricta.*

In southwestern Manitoba, plants indicative of higher salinities were: *Salicornia rubra, Sueda depressa, Scirpus paludosus, Scirpus americanus, Atriplex hastata, Distichlis stricta, Potamogeton pectinatus**, *Ruppia americana,* and *Zannichellia palustris. Ruppia* is probably the most tolerant aquatic and *Distichlis* the most tolerant riparian species (Dr. G.D. Adams, Canadian Wildlife Service, pers. comm.).

Seddon (1972) correlated floristic assemblages of aquatic macrophytes with water quality (conductivity and total dissolved solids).

Haller *et al.* (1974) examined the effects of salinity on the growth rates of 10 aquatic macrophytes. The aquatic plants studied can be roughly divided into three groups according to salinity tolerance. The larger floating species *Eichornia* and *Pistia* were most susceptible to low levels of salinity (2.5‰). With the exception of *Myriophyllum (brasiliense* and *spicatum)* species, the submersed plants were the next most susceptible to increasing salinities. Concentrations toxic to this group were between 5‰ and 10‰. Salinities toxic to smaller floating species and *Myriophyllum** species ranged between 10.0‰ and 16.6‰. This grouping indicates that morphology of aquatic plants may be a factor in determining salt tolerances.

Symptoms of salinity stress on the larger floating plants *(Eichornia crassipes* and *Pistia stratiotes)* and emergent *Myriophyllum* were similar to symptoms described for terrestrial plants (Gauch, 1972). Leaf margins became chlorotic; the leaves twisted and finally became necrotic in toxic concentrations. Toxicity symptoms first appeared on the older leaves and progressed to younger, more immature foliage. Frequency of occurrence and severity of symptoms were dependent on salt concentrations. The smaller floating species *(Lemna minor, Azolla caroliniana,* and *Salvinia rotundifolia)* became chlorotic and, in toxic solutions, sank to the bottom of the containers. Submersed species *(Hydrilla verticillata, Myriophyllum spicatum, Nujas guadalupensis,* and *Vallisneria americana*) at toxic salinity levels did not exhibit any of these characteristic symptoms but rather remained dark green as if preserved, and after 2-3 weeks decayed and sank.

Weirich (1974) observed marked stunting in *Myriophyllum spicatum* in habitats of high salinity. Examination of specimens collected from the habitat of high salinity had less wall-thickness in the epidermis, the sub-epidermal cortical layers, and in the outer tangential endodermal wall. Smaller cells and fewer cells for each tissue accounted for the stunting. The author concludes that the toxic environment may limit the plants' ability to produce cell walls sufficiently thick to limit salt intake.

3.1.2 Protozoa

Freshwater Protozoa are hyperosmotic to their medium, and the contractile vacuola eliminates excess water load. A survey of many ciliates shows that few freshwater species can survive in more than 5‰ (ppt)[1] salinity (Ax and Ax, 1960).

Water can enter protozoans by exchange diffusion, by osmotic movement, and by food vacuoles. Contractile vacuoles are present in all freshwater Protozoa. From volume and rate of pulsation measurements, an osmotic function is indicated.

When the freshwater *Amoeba verrucosa* was cultured in increasing concentrations of sea water, the vacuolar pulsations slowed and no vacuole was seen when the animals were in 50% sea water (Prosser, 1973).

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[1]parts per thousand

When *Paramecium* is first put into a hyperosmotic solution, the body becomes flattened but later recovers its normal shape, and vacuolar output is markedly reduced. In one saline tolerant species of *Paramecium*, the average intervals between vacuolar pulsations increased from 13 seconds in fresh water to 32 seconds in 50% sea water and to 65 seconds in 100% sea water (Frisch, 1944).

3.1.3 Annelida

3.1.3.1 Oligochaeta (Aquatic Earthworms)

Brackish water species (such as *Paranais litoralis*) may be found in inland waters affected by saline discharges from salt deposits utilized by chemical industries, such as those along the St. Clair River and the River Werra in Germany (Wachs, 1963).

Kamemoto and Goodnight (1956) investigated the effects of various salt concentrations of the inorganic ions of calcium, potassium, and magnessium on asexual reproduction in *Aeolosoma hemprichi*. They concluded that the inorganic environment affected the asexual reproduction of *Aeolosoma* by means of a complex interaction of various ions together with the temperature. Whereas calcium in certain concentrations had a stimulating effect and high concentrations of potassium were toxic, no one ion could, in general, be singled out from the complex in its effects.

3.1.3.2 Hirudinea (Leeches)

In Europe, no leeches occur in a salinity higher than 6‰, except *Piscicola geometra* which may occur up to 8‰ (Herter, 1937).

In certain Gulf coast rivers the freshwater species, *Helobdella stagnalis**, *H. elongata*, *Placobdella parasitica*, *Illinobdella moorei*, *Erpobdella punctata*, and *Mooreobdella microstoma*, are confined to water with a total salinity of less than 0.68‰ (Wurtz and Roback, 1955).

Not all leeches have the same tolerance to salinity changes: the erpobdellids and certain glossiphoniids (e.g., *H. stagnalis** and *H. lineata*) are especially sensitive to salinity, whereas some glossiphoniids (e.g., *P. parasitica*), all piscicolids, and perhaps the hirudinids are remarkably tolerant. For example, the average survival time (in days) of the following European and American leeches placed in water of 9.7‰ at 10°C are: *H. stagnalis** (1), *H. lineata* (2), *Theromyzon tessulatum* (3.6), *Glossiphonia complanata* (8), *P. parasitica* (23), *Erpobdella octuculata* (3.8), and *Macrobdella ditetra* (3.8)(unpublished data in Sawyer, 1974).

The piscicolids, *Piscicola geometra*, *I. moorei*, and

Myzobdella lugubris, will live indefinitely under the conditions described above.

The role that water conductivity (salinity) plays in the distribution of leeches in a series of saline lakes in the Southern Interior Plateau region of British Columbia has been investigated (Scudder and Mann, 1968). With the exception of *Theromyzon rude* (3200 micromhos/cm), no leeches were encountered in water with a conductivity above 1650 micromhos/cm. In laboratory tests, *Nephelopsis obscura* died in two hours at 3770 micromhos/cm.

Similarly, the role that total residue content of the water plays in the distribution of leeches in Colorado has been investigated (Herrmann, 1970). The maximum value of total residue (in ‰) at which each species occurs is: *Helobdella stagnalis**, 2.99; *Glossiphonia complanata*, 0.95; *Theromyzon rude*, 2.09; *Illinobdella moorei*, 2.05; *Erpobdella punctata*, 1.86; *Dina dubia*, 1.69; *D. parva*, 1.10; *Nephelopsis obscura*, 0.65; and *Haemopsis (Percymoorensis) marmorata*, 3.80.

3.1.4 Mollusca

3.1.4.1 Bivalvia (Clams and Mussels)

Destruction of mussels by chloride-laden oil field brine was first reported by Shira (1913). Williams (1969) reviewed oil brine damage in the Green River of Kentucky. In 1958, the chloride concentration rose from 10 to more than 1,000 mg/l. Mussel bed recruitment became possible in the affected area only where high flow and dissolved oxygen levels prevailed (i.e., just below dams). *Amblema plicata* and *Megalonaias gigantea* withstood brines best among mussels. Reporting on the mussels of Turtle River, North Dakota, Cvancara and Harrison (1965) recorded none where the river flows through a belt of naturally saline soils and chloride concentration rises to 87 mg/l and more.

The sensitivity of the freshwater mussel *Anodonta cygnea* to various salts was studied by Lukacsvoics and Salanki (1968). They found *A. cygnea* more sensitive to potassium (K+) than to any other cation. The effective order of cations, which caused a closure of the inflow siphon, was $K>Cd>Ca>Mg>NH_4>Na$. Imlay (1971) added several observations on the relation of potassium to mussels.

Nagabhushanam and Lomte (1971) investigated the effect of salinity on the survival of the freshwater mussel *Parreysia corrugata*. Between 0.5% and 0.7% NaCl, all freshwater mussels tightly closed their shells till they died. In 0.3%, NaCl, out of 10 animals tested only 3 died during the experimental period. In the higher salinities, *P. corrugata* could not survive probably partly due to dehydration and partly due to the influx of ions following an osmotic gradient between the body fluid and the medium.

Lomte and Nagabhushanan (1971) also investigated the respiration of *Parreysia* in relation to salinity. The media, with salinities ranging from 0.1 to 0.7% NaCl were made up from NaCl and distilled water. They observed that oxygen consumption decreased with an increase in salinity. Their results are in agreement with those of Hiscock (1953) on *Hyridella* where the oxygen consumption decreased considerably with an increase of external chlorinity. Thus it would appear that salinity has a significant effect on respiration.

3.1.4.2 Gastropoda (Snails)

"There is no physiological reason, at least from the point of view of water balance, why freshwater species should not survive well in water with a higher salt content. As long as the body fluids remain iso-osmotic or hyper-osmotic to their surroundings, no fundamental change in water balance physiology is required" (Machin, 1975).

Under experimental conditions, it has been shown that *Lymnaea stagnalis* readily survive saline solutions and respond to them by decreasing urine production in proportion to the reduced osmotic gradient across the body wall. There are also a number of recorded instances of freshwater snails inhabiting natural brackish water environments.

However, chloride wastes from Oil Chemical Corporation's soda ash operation in the North Fork of the Hoston River over the last 75 years has apparently eliminated *Io fluvialis* (Goodrich, 1940), *Oxytrema unicale* and *Leptoxis subglobsa* (Stansbery, 1972). All of these species occur above the pollution source.

For the freshwater snail *Physa heterostropha*, the acute dose 96 hr LC50 for Na (as NaCl) was 6,200 mg/l (Wurtz and Bridges, 1961). Other industrial wastes have been checked for toxicity to *P. heterostropha*; for example, KCl, a 96 hr LC50 of 2,010 mg/l. Dowden and Bennett (1965) give an acute (one day) dose of 1,941 mg/l KCl to *Lymaea* sp.

3.1.5 Arthropoda

3.1.5.1 Insecta (Aquatic Insects)

The regulatory abilities of several aquatic insects kept in saline media are known in some detail. In particular, salt and water regulation in mosquito *Aedes aegypti* larvae has been studied intensively by Wigglesworth (1933, 1938) and Ramsay (1950, 1951, 1953). Larvae of *Sialis lutaria* were also thoroughly investigated by Beadle and Shaw (1950) and Shaw (1955).

In both *A. aegypti* and *Sialis* larvae, the haemolymph osmotic pressure rises when the external salt concentration is increased, so that the haemolymph remains hyper-osmotic to the medium. Regulation of the haemolymph salt concentration begins to break down when the external salt concentration is increased to a level roughly equivalent to the normal total concentration of the haemolymph, i.e., at about 170 mM/l[1] (9,937 mg/l) NaCl. Larvae begin to die when the external concentration is increased above this level. External concentrations greater than about 170 mM/l are also fatal to larvae of *Corethra* (Schaller, 1949) and *Helodes* (Treherne, 1954). Thus, to all of these freshwater insects, external salinities greater than 30-40%

[1]mM/l--millimoles per litre

sea water are rapidly fatal. Butler and Popham (1958) considered a salinity of about 8.5‰ as being critical to many freshwater insects.

Shaw (1955) noted that the fact that *Sialis* larvae are unable to live in saline more concentrated than the blood could be explained from a knowledge of their power of ion and water regulation. Osmotic and ionic regulation in aquatic insects is reviewed by Hoar (1966) and Prosser (1973), among others.

Roback (1974) lists those aquatic insect species or genera which are tolerant of high Cl levels (Tables 3a and 3b).

Ephemeroptera (Mayflies)

Forbes and Allanson (1970) undertook an investigation of the osmoregulatory abilities of three species of mayfly nymph of the family Baetidae. The tolerance of nymphs was investigated by exposing them to dilutions of sea water or to an artificial river water of equivalent total dissolved solids (TDS) content. In order to compare the effects of different media, the time at which 50% of the experimental animals survived was referred to as the Median Time of Survival (MTS).

The laboratory experiments described in their paper

TABLE 3a. Aquatic insect species found at chloride greater than 1,000 mg/l (from Roback, 1974).

Odonata

Hetaerina americana (Fabr.)
Argia moesta (Hagen)
Argia sedula (Hagen)
Argia violacea(Hagen)
Enallagma civile (Hagen)
Enallagma signatum (Hagen)
Enallagma traviatum Selys
Ischnura posita (Hagen)
Erpetogomphus designatus Hagen
Dromogomphus spinosus Selys
Gomphus vastus Walsh
Gomphus lividus Selys
Basiaeschna janata (Say)
Boyeria vinosa (Say)
Erythemis simplicicollis (Say)

Ephemeroptera

Baetis spp.*
Callibaetis spp.
Neocloeon alamance? Trav.

Plecoptera

Paragnetina sp.

Hemiptera

Gerris marginatus Say
Rheumatobates tenuipes Meinert
Trepobates inermis Esaki
Microvelia americana Uhl.
Rhagovelia obesa (Uhl.)

Coleoptera

Laccophilus spp.
Thermonectes spp.
Gyrinus spp.
Berosus spp.
Tropisternus spp.
Helichus lithophilus (Germ.)

Lepidoptera

Parargyractis sp.
Paraponynx sp.

(Continued)

TABLE 3a. Continued.

Trichoptera
- *Nyctiophylax* sp. A Flint
- *Hydropsyche betteni* Ross*
- *Hydropsyche* (*bifida* gp.) sp.
- *Leptocella candida* Hagen

Diptera
- *Procladius bellus* (Loew)
- *Coelotanypus concinnus* (Coq.)
- *Clinotanypus pinguis* (Loew)
- *Cricotopus bicinctus* (Meig.)*
- *Cricotopus* nr. *exilis* Joh.
- *Cryptochironomus* nr. *fulvus* Joh.*
- *Dicrotendipes modestus* (Say)
- *Dicrotendipes nervosus* (Staeger)
- *Endochironomus nigricans* (Joh.)
- *Glyptotendipes* sp.
- *Polypedilum illinoense* (Mall.)*
- *Palpomyia* gp. spp.

TABLE 3b. Aquatic insect species found at chloride greater than 2,000 mg/l plus magnesium greater than 150 mg/l (from Roback, 1974).

Odonata
- *Ischnura posita* (Hagen)
- *Erythemis simplicicollis* (Say)

Ephemeroptera
- *Baetis* spp.*
- *Callibaetis* spp.
- *Neocloeon alamance?* Trav.

Hemiptera
- *Rheumatobates tenuipes* Meinert
- *Trephobates inermis* Esaki

Coleoptera
- *Gyrinus* spp.

Trichoptera
- *Nyctiophylax* sp. A Flint

Diperta
- *Procladius bellus* (Loew)
- *Coelotanypus concinnus* (Coq.)
- *Clinotanypus pinguis* (Loew)
- *Endochironomus nigricans* (Joh.)
- *Glyptotendipes* sp.
- *Polypedilum illinoense* (Mall.)*
- *Palpomyia* gp. spp.

*Denotes species or genera collected from Poplar Creek (Syncrude Canada Ltd., 1975).

showed that all nymphal stages could survive TDS levels up to 7,000 mg/1, but the smaller nymphs were adversely affected once the level reached about 9,000-10,000 mg/1. The TDS tolerance increased markedly with age, i.e., juvenile nymphs are more sensitive than adults to high concentrations of dissolved solids. Macan (1961) stated that tolerance to environmental factors may vary with age in freshwater animals.

The tolerance tests showed that the ionic variation of the water was unimportant and that the important restricting factor was the TDS level. This supports Beadle's (1943) contention that overall TDS levels, and hence osmotic pressures, are of more importance to aquatic insects than the nature of the substances contributing to the osmotic pressures.

Sutcliffe's (1962a) work on larvae of Trichoptera provides a parallel to the above study in that he also found species within the same genus with markedly different TDS tolerances.

Mayfly larvae and other aquatic insects possess chloride cells in the integument. The number of the chloride cells is correlated with the osmoregulatory situation and the different salinities of the surrounding water. Wichard (1974) suggested the use of chloride cells in the tracheal gills of mayfly larvae as indicators of salinity. An increase in the external salinity decreases the need for salt absorption in osmoregulation and hence the number of chloride cells.

Larvae of *Callibaetis* (Baetidae) are apparently very sensitive to major changes in salinity. When directly transferred into concentrated fresh water with 100 or 150 mM NaCl/1, which is approximately iso-osmotic (Sutcliffe, 1962a; Forbes and Allanson, 1970), all larvae died within a few days (Wichard *et al.*, 1973).

Plecoptera (Stoneflies)

Roback (1974) reports no plecopterans occurring at a chloride concentration greater than 2,000 mg/1 plus magnesium greater than 150 mg/1. At chloride greater than 1,000 mg/1, only a *Paragnetina* species was present.

Hemiptera (Corixidae and Notonectidae)

Previous studies of osmoregulation in Corixidae inhabiting saline waters (Claus, 1937; Scudder, 1969; Scudder *et al.*, 1972; Knowles and Williams, 1973; Frick and Sauer, 1974) have shown that corixids, with the exception of the brackish water species, *Sigara stagnalis*, are essentially freshwater species although some have developed limited salinity tolerance.

The osmotic regulation and salinity tolerance of *T. verticalis interiores* (Corixidae) were investigated by Tones and Hammer (1975). *T. verticales interiores* was apparently tolerant to changes in ionic, particularly anionic, composition. The tolerance limit (TL50) of the adults at 13°C occurred when the freezing point depression of the medium was 1.64°C, which corresponded to a conductivity of 45,000 μmhos/cm at 25°C.

The common European backswimmer, *Notonecta glauca*, is sometimes found in brackish waters. However, field data indicates that the adults avoid or are unable to endure salinities higher than about 7‰, that is, higher than about a fifth of the salinity of normal sea water (35‰) (Poisson, 1924; Lindberg, 1948; Butler and Popham, 1958). Adults are unable to osmoregulate their haemolymph in media more concentrated than normal haemolymph (Staddon, 1973a). However, the adults of *N. glauca* seem better able to regulate the total concentration of the haemolymph than adults of the freshwater corixids *Sigara distincta* and *S. fossorum* (Claus, 1937).

Trichoptera (Caddisflies)

Sutcliffe (1961a, b) compared salinity tolerances of brackish and freshwater species of caddis larvae (*Limnophilus*). He found that the brackish water species, *L. affinis*, could tolerate external salt concentrations up to at least 410 mM/l NaCl (23,965 mg/l) and survive for short periods in 470 mM/l NaCl (27,472 mg/l) whereas the two freshwater species, *L. stigma* and *Anabolia nervosa*, died rapidly in 30-40%

sea water. The majority of larvae of both freshwater species did not survive for more than a few days at external salt concentrations greater than about 60 mM/l NaCl (3,507 mg/l). In 120 mM/l NaCl (7,014 mg/l) about 50% of the larvae died within three days while in 170 mM/l NaCl (9,937 mg/l) about 75% died within three days and in 220 mM/l NaCl (12,859 mg/l) only a few individuals survived for more than two days.

Unlike *L. affinis*, larvae of the freshwater species are unable to survive in salt water hyper-osmotic to the normal haemolymph level. Drinking occurs but is not controlled and very large amounts of salt water are swallowed. Moreover, salt water has a decidedly adverse effect on the alimentary canal, manifested by massive regurgitation of fluid, eversion of the rectal wall, and expulsion of the peritrophic membrane. This behaviour is accompanied by a rapid decrease in weight which must be due largely to exosmosis across the gut wall. In this respect, the effect of salt water appears to be very similar to that observed by Wigglesworth (1933) in freshwater mosquito larvae placed in salt water. Clearly, successful adaptation to salt water must involve an increased tolerance of the gut wall to high concentrations of salt (Sutcliffe, 1962).

Roback (1965) gives a table showing 8 out of 20 genera of Trichoptera being tolerant to a chloride concentration of over 2,500 mg/l. This is equivalent to a salinity concentration of 4,500 mg/l. The net makers appear to be slightly more common at these higher salinities. Under brackish situations (chloride greater than 2,000 mg/l, magnesium greater than 150 mg/l), only *Nyctiophylax* is found (Roback, 1974).

He (1965) also reported that caddisfly larvae *(Hydropsyche)** were far more tolerant to NaCl than mayflies *(Stenonema rubrum)**. The 48 hr TL_m for *Hydropsyche* was 9,000 mg/l while *Stenonema* was 2,500 mg/l NaCl.

Haage (1968) studied the salinity tolerance of eggs and recently hatched larvae of *Phryganea grandis* from brackish water in the Baltic Sea. He found that mature eggs had a higher tolerance than freshly deposited eggs or larvae. The experiments indicated that the limit of the salinity tolerance of larvae of *Phryganea grandis* in their first stages of development was about 7‰--freshly deposited eggs would not develop to hatching in salinities above 7‰.

The reason why the differentiated (mature) eggs show a higher tolerance than newly deposited ones may be that the properties of the egg membranes or the jelly mass change during development. The difference in volume change indicates this. The results show that the eggs close to hatching have no difficulties in tolerating an occasional increase of salinity.

Haage also found that case building becomes abnormal in salinities above 10‰. "In salinities of 12-20‰ the cases were very badly built, the algal fragments were not of the same size and they were loosely and unevenly joined so that the cases showed holes and easily fell to pieces."

Diptera

When the mosquito larvae *(A. aegypti)* are placed in artificial sea water of various dilutions, it is found that both the osmotic pressure and the chloride content of the haemolymph remain constant until the concentration of the external medium reaches 0.65-0.75% NaCl. In more concentrated media, the chloride content of the haemolymph increases and the total osmotic pressure is thereby maintained slightly in excess of that of the medium up to an external concentration of 1.6% NaCl, at which the larvae die rapidly (Wigglesworth, 1938).

Curry (1965) gives a table listing the ranges of environmental factors for freshwater tendipedid (midge) species. Maximum chloride concentrations tolerated ranged from 6.0 to 12.0 mg/l to 6% sea water. Roback (1974) gives a similar table for dipterans. The most tolerant of chemical extremes are Chironomidae; for example, in brackish situations, 6 of the 7 species present are Chironomidae and where chloride exceeds 1,000 mg/l, only Chironomidae are present.

Thornton and Wilhm (1974) measured the effects of pH, phenol, and NaCl on the survival of the life stages of *Chironomus attenuatus* as well as the caloric lipid and protein-nitrogen content of larvae and adults. They found that NaCl affected the lipid content of the fourth instar larvae. The lipid content was higher with NaCl present than without it. Survival was also affected by the interaction between phenol and NaCl. Survival was higher when phenol and NaCl were present than when either was present without the other.

3.1.5.2 Crustacea

The blood concentrations maintained by most freshwater crustaceans are considerably lower than those of species normally inhabiting dilute brackish water. These blood concentrations are rather accurately regulated when the animals are in their normal environment, but an increment of the salinity of the environment is accompanied by an increase in the blood concentration. Freshwater species are not able to tolerate wide changes in the blood concentration, and any marked rise results in death. The inability of *Gammarus pulex* to tolerate high salinities is associated with its inability to prevent chloride from entering the tissues when the blood concentration is raised above the normal level (Shaw and Sutcliffe, 1961). In general, the higher the initial blood concentration, the greater is the range of salinity tolerated (Lockwood, 1962).

A few crustaceans from fresh water exhibit wider tolerance of saline media. For example, *Telphusa* (Duval, 1925), fresh water *Saduria (=Mesidotea) entomon* (Lockwood and Croghan, 1957), and *Caecosphaeroma burgundum* (Dresco-Derouet, 1959) can be acclimatized to normal sea water.

Adaptation to high salinities necessitates other physiological adaptations besides those of osmoregulation. The decrease in oxygen levels with increase in salinity requires an effective mode of oxygen uptake. Gilchrist (1954) has shown that in *A. salina* there is an increase in blood haemoglobin in response to a decrease of oxygen level as salinity increases.

Salinity has an important influence on the duration of the juvenile phase. This was pointed out by Engel (1961), who caused female *Daphnia magna* to mature after four, five, or six instars by using different concentrations of diluted sea water. In these experiments, the juvenile phase was prolonged by the higher concentrations.

Toxicity of Saline Waters to Crustaceans

Holm-Jensen (1944) discussed the survival time of *Daphnia magna* when kept in several concentrations of diluted sea water. If *D. magna* specimens are allowed to starve in the laboratory, their brackish water tolerance diminishes

(Lagerspetz, 1955).

Gresens (1928) studied the survival time of an isopod, *Asellus aquaticus*, in different salinities. His test animals were obtained from fresh water (salinity $\leq 0.6‰$) in the estuary of the River Ryck. He concluded, that after a sudden increase of salinity, *Asellus* is able to survive permanently in 5.25 (some individuals in 7‰) which is the upper limit for breeding. After a gradual acclimation to higher salinities, these isopods were able to survive in a salinity of 15‰. Similarly, Lagerspetz (1958) found that *Asellus* does quite well in a salinity of 4.9‰.

Lagerspetz (1958) also conducted brackish water tolerance experiments on three species of Daphnias: *Daphnia magna*, *D. pulex* and *D. longispina*. He found that *D. magna* could withstand brackish water with a salinity of 5.8‰ whereas the other two species, *D. pulex* and *D. longispina* showed a high mortality even at a salinity of 2.9‰.

There are data given in the literature about the influence of specific ions on Daphnidae. Anderson (1948) presented threshold concentrations of toxicity to *Daphnia magna* for 25 cations when added to Lake Erie water. His results and comparisons with others are presented below:

Calcium Chloride ($CaCl_2$)

The threshold of toxicity for *D. magna* was 0.0083 molar (920 mg/l). This value was about two-thirds that reported by Anderson (1944) on the basis of an exposure period of 16 hours.

Naumann (1934) stated that 0.0066 molar (approximately 730 mg/l) $CaCl_2$ in a soft water was without effect on *D. magna* in 24 hours, but that 0.0165 molar (1,830 mg/l) produced a weakening. In a hard water, 0.0165 molar was without effect but 0.033 molar (3,670 mg/l) immobilized *D. magna*.

Ramult (1925) found that 0.0167 molar (1,860 mg/l) in a pond water caused a weakening of *Daphnia pulex* adults by the second day and inhibited development of the eggs.

Fowler (1931) reported that 0.05 molar (5,560 mg/l) $CaCl_2$ killed *D. longispina* in 41 hours when he used a well water as the diluent.

Anderson *et al.* (1948) gave 0.0156 molar (1,730 mg/l) and 0.013 molar (11,440 mg/l) as the thresholds to *Cyclops vernalis* and *Megacyclops leuckarti*, respectively.

Magnesium Chloride ($MgCl_2$)

Magnesium chloride was somewhat more toxic than $CaCl_2$

to *D. magna*, its threshold was 0.0078 molar (740 mg/1).

Hutchinson (1933) gave the following tolerance limits for survival and reproduction of Cladocera: *D. magna*, 0.01 molar (950 mg/1); *D. thomsoni*, 0.00125 molar (120 mg/1); *D. pulex*, 0.005 molar (480 mg/1); *D. longispina* (clone 1), 0.0025 molar (240 mg/1); *D. longispina* (clone 2), 0.00125 molar (120 mg/1); *Ceriodaphnia reticulata*, 0.0025 molar (240 mg/1); and *Moina macrocopa*, 0.0075 molar (710 mg/1), when he used pond water as the diluent.

Potassium Chloride (KCl)

Its toxicity threshold was 0.0058 molar (432 mg/1).

Ramult (1925) stated that 0.0167 molar (1,246 mg/1) in pond water killed *Daphnia pulex* adults and prevented the eggs from developing but that 0.0083 molar (619 mg/1) was without effect.

Fowler (1931) reported that *D. longispina* survived less than 7 hours in 0.025 molar (1,865 mg/1) in well water.

Naumann (1934) found that 0.01 molar (746 mg/1) immobilized *D. magna* in soft water and irritated those in hard water.

Baylor (1942) gave 0.005 molar (373 mg/1) in aerated, non-chlorinated tap water as "sub-threshold" for effect on the heart beat of *D. magna*. He found that 0.01 molar inhibited the heart to a slight extent but his exposure times were less than 2 hours.

Sodium Chloride (NaCl)

Sodium chloride was the least toxic of all chlorides tested. Its toxicity threshold was estimated at 0.063 molar (3,680 mg/1). This value is considerably lower than that reported by the same author (1944) when the thresholds were calculated on the basis of a 16 hour exposure period and is somewhat lower than that based on a 48 hour exposure period (Anderson, 1946).

Warren (1899) found that 0.55 per cent (0.094 molar, 5,491 mg/1) NaCl was lethal to *D. magna*.

Ramult (1925) reported that *D. pulex* failed to develop in 0.1 molar (584 mg/1) NaCl but that some developed in 0.05 molar (2,921 mg/1) in pond water. He also stated that *Ceriodaphnia laticaudata* adults were weakened by 0.05 molar but that *Pleuroxus aduncus* tolerated this concentration.

Fowler (1931) determined that *D. longispina* survived 66 hours in 0.05 molar in a well water.

Naumann (1934) found that *D. magna* were immobilized by 0.05 molar solutions of NaCl in both soft and hard waters.

Anderson *et al.* (1948) carried out a series of experiments on plankton Crustacea and reported that *Leptodora kindtii*, *Cyclops vernalis*, and *Diaptomus oregonensis** were immobilized by 0.063 molar (3,680 mg/l), 0.104 molar (6,075 mg/l), and 0.052 molar (3,037 mg/l), respectively, in Lake Erie water.

On the basis of the above evidence, one may conclude that concentrations of NaCl above 0.05 molar (approximately 3,000 mg/l) are deleterious to Crustacea.

Dowden (1960) tested the toxicity of 13 inorganic salts to *D. magna*. The results of the 24 hour and 48 hour Median Tolerance Limits (TL_m's) for the chloride salts are summarized in Table 4. TL_m's for each salt for 24 and 48 hours were compared to show cumulative toxicity which was then compared to NaCl to show the variation(s) in ionic structures that cause the cumulative toxicity. It was found that sodium nitrate, sodium sulfate, sodium bisulfite, sodium dichromate, chromic sulfate, and calcium chloride were cumulatively toxic, and that sodium chloride, sodium silicate, sodium pyrophosphate, potassium chloride, and ammonium chloride were not cumulatively toxic.

Table 4. Summary of some toxicity data for calcium, magnesium, potassium, and sodium salts to *Daphnia magna* (from Gregory, 1974).

Salt	n	Temperature (°C)	Exposure Time	LC50 (mg/l) Salt	LC50 (mg/l) Cation		Authority
$CaCl_2$	10	23±1	24 hrs	1,838	-		Dowden (1960)
	10	23±1	48 hrs	759	-		
	-	-	48 hrs	759	276		Dowden and Bennett (1965)
	-	-	100 hrs	649	236		
$CaCl_2.2H_2O$	10	18±1	48 hrs		52	(without food)	Biesinger and Christensen (1972)
	10	18±1	48 hrs		464	(with food)	
	5	18±1	3 wks		330	(308-335)	
	5	18±1	3 wks		116	(16% reproduction impairment)	
	5	18±1	3 wks		220	(50% reproduction impairment)	
$MgCl_2.6H_2O$	10	18±1	48 hrs		140	(without food)	Biesinger and Christensen (1972)
	10	18±1	48 hrs		322	(with food)	
	5	18±1	3 wks		190	(167-217)	
	5	18±1	3 wks		82	(16% reproduction impairment)	
	5	18±1	3 wks		125	(50% reproduction impairment)	

(Continued)

Table 4. Continued.

Salt	n	Temperature (°C)	Exposure Time	LC50 (mg/l) Salt	LC50 (mg/l) Cation	Authority
KCl	10	23±1	24 hrs	343		Dowden (1960)
	10	23±1	48 hrs	357		
	10	18±1	48 hrs		93 (without food)	Biesinger and Christensen (1972)
	10	18±1	48 hrs		166 (with food)	
	5	18±1	3 wks		97 (87-108)	
	5	18±1	3 wks		53 (16% reproduction impairment)	
	5	18±1	3 wks		68 (50% reproduction impairment)	
NaCl	10	23±1	24 hrs	3412		Dowden (1960)
	10	23±1	48 hrs	3318		
	10	18±1	48 hrs		1,640 (without food)	Biesinger and Christensen (1972)
	10	18±1	48 hrs		1,820 (with food)	
	5	18±1	3 wks		1,480 (1180-1840)	
	5	18±1	3 wks		680 (16% reproduction impairment)	
	5	18±1	3 wks		1,020 (50% reproduction impairment)	

In addition to examining salts individually, Freeman and Fowler (1953) and Dowden and Bennett (1965) studied salts in combinations of two's and three's. In each case, the LC50 values for the individual salts in combination were less than for the same salts individually. Dowden and Bennett's data for $CaCl_2$ are presented in Table 4.

Biesinger and Christensen (1972) demonstrated that daphnids are five to six times more sensitive to calcium than to sodium ions (Table 4). An accumulation of toxicity with time for potassium is indicated from their data. The three week LC50 for KCl is lower than the 48 hour value, while the results for NaCl and $CaCl_2$ are similar for these two times. The 16% reproduction impairment (and "safe" concentration) of 53 mg/l potassium for daphnids, together with the lethal concentration limit of 50 mg/l potassium for stickleback (Jones, 1939a), indicate that KCl concentrations down to 100 mg/l may be harmful in the aquatic environment.

Land (1974) gives a comparison of the toxicity of three drilling fluid chlorides to crustaceans (Table 5) which shows that KCl is approximately 10 times more toxic than NaCl. Calcium ions are eight times as toxic to daphnids as sodium ions on a molar basis (Anderson, 1948).

Table 5. Comparative toxicity of chlorides to crustaceans (from Land, 1974).

Test Animal	Time and Type of Exposure	mg/l Chloride KCl	mg/l Chloride $CaCl_2$	mg/l Chloride NaCl	Authority
Cyclops vernalis	64 hour	640	1,730	6,100	Anderson *et al.*
Mesocyclops leukarti	threshold	566	1,440	-	(1948)
Diaptomus oregonensis	immobilization	134	-	3,030	
Leptodora kindtii		127	-	3,700	
Daphnia magna		432	920	3,680	
Daphnia magna	48 hr LC^{50}	317	1,288	4,625	Biesinger and
	3 wk LC^{50}	185	916	3,761	Christensen (1972)
	50% reproduction impairment	130	611	2,592	
	16% reproduction impairment	101	322	1,728	

Decapoda (Crayfishes)

Among the Crustacea, there are a number of freshwater forms, of which the crayfishes are the best known examples, which are typical freshwater animals. "The features which characterize an animal as such are: 1) the fact that it cannot survive in sea water solutions much stronger than its normal blood concentration, 2) a body surface relatively impermeable to salts, 3) a relatively low blood concentration, and 4) the production of a dilute urine" (Shaw, 1959b).

Salinity Tolerance of Crayfish

Herrmann (1931) found *Astacus astacus (=Potamobius astacus)* would survive for three months in 50% sea water while Bogucki (1934) found that the same species, which he designated *Astacus fluviatilis*, would tolerate 66% sea water for one month.

Steeg (1942), in studying the resistance of young (16 to 25 mm carapace length) *Procambarus clarkii* to NaCl solutions, discovered that 100% mortality results in 23‰ (ppt), and, while able to tolerate concentrations of 15 ppt, as the salinity increases there is a corresponding increase in death rate. He suggested the possibility that individuals occurring naturally in low concentrations of salt might be able to withstand higher salinities than those individuals frequenting salt-free water.

Clemens and Jones (1954) found an Oklahoma crayfish (*Cambarus* sp.) tolerant of oil well brine solutions which were diluted to 8.7% by volume of the original brine.

Kendall and Schwartz (1954) found that some individuals of *O. virilis* (80 to 110 mm total length) were able to tolerate salinities of approximately 0, 6, 14, and 30 ppt for a maximum of 218, 216, 218, and 240 hours, in one test and 696, 696, 528, and 120 hours in a second one. In the second experiment, 50% of the animals subjected to each of the four concentrations died in approximately 416, 96, 108, and 40 hours, respectively.

In similar experiments with *Cambarus b. bartonii*, this crayfish was found to tolerate salinities of 0, 5.2, 13.6, and 27.5 ppt for 240, 252, 108, and 192 hours, respectively, but 50% of the test population died within 72 to 96 hours. Salinities of 6 to 15 ppt were readily tolerated for a period of 27 days, while higher concentrations were detrimental.

Penn (1956) reported that *P. clarkii* has been collected in littoral areas of Lake Pontchartrain where salinities were as high as 6‰.

Loyacano (1967) compared the salinity tolerance of individuals of an inland population of *P. clarkii* with those of a coastal marsh population. He found that newly hatched young are unable to withstand salinities of 15 to 30 ppt for as much as a week; those with a total length of 30 mm are able to tolerate concentrations up to 20 ppt but die within 2 or 3 days in 30 ppt. In contrast, individuals from 40 to 120 mm live at least a week in the latter concentration. It was discovered that growth varies inversely with salinity--individuals having a total length of 40 to 50 mm subjected to salinities of 0, 10, and 20 ppt for 4 weeks exhibited a weight increase of 4.4, 13.5, and 4.9% respectively. He concluded that adult *P. clarkii* are able to withstand salinities almost equivalent to that of sea water for a short time and that their tolerance to salinity is directly proportional to the size of the crayfish.

Toxicity to Individual Salts

Helff (1931), in testing the antagonistic properties of four metallic ions (Ca, Mg, K, and Na) on *P. clarkii* (Table 6), found the sequence of toxic effects to be K, Ca, Mg, Na. The toxicity of all four in general, with the possible exception of potassium, was reduced when introduced to the crayfish in combination with one or more of the others.

Table 6. Toxicity of various salt solutions to the crayfish, *Procambarus clarkii* (from Helff, 1931).

Concentration of Solutions (M)	Duration of Life (hours)			
	$CaCl_2$	$MgCl_2$	KCl	NaCl
0.5[a]	17.0	12.6	2.5	20.0
0.45	25.2	13.6	2.9	36.2
0.4	25.3	17.8	3.0	65.9
0.3	32.4	24.0	3.9	157.3 (7[b])
0.2	43.1	65.0	4.9	160.2 (7[b])
0.1	78.8 (2[b])	99.5	6.5	(10[b])
0.05	95.2 (1[b])	(10[b])	17.2	(10[b])
Distilled water	(10[b])	(10[b])	(10[b])	(10[b])

[a]The $MgCl_2$ solutions used were twice the molecular strength of corresponding $CaCl_2$, KCl, and NaCl solutions indicated above.

[b]Denotes number living for 7 days when test was discontinued.

3.1.6 Chordata

3.1.6.1 Pisces (Fishes)

The effects of saline waters on fish may be: 1) increase in osmotic pressure (i.e., stress), 2) a direct toxic effect, or 3) an indirect effect (i.e., toxic to fish-food organisms).

Osmotic Responses to Salinity Changes

Since fish in fresh water maintain their body fluids at a concentration higher than that of the surrounding medium, they must be continually taking on water through the tissues by osmosis. The skin is relatively impermeable and little water is taken in by the mouth, but some water enters osmotically through the gills and oral membrane. The inward stream provides a water load, and the kidneys have well developed glomeruli that filter a considerable volume. As the filtrate passes down the tubules, most of the solutes are reabsorbed, leaving a dilute urine. However, the urine is not as dilute as the freshwater medium, and some salt is lost by diffusion and a small amount by feces. The salt loss is compensated partly by food and partly by active absorption from the medium by special cells of the gills.

In summary, osmotic regulation in freshwater fishes is accomplished by the excretion of a hypotonic urine and the acquisition of salts from food and from the surrounding water (Krogh, 1939; Black, 1951; Hoar, 1966; Prosser, 1973).

Inorganic ions which appear to be necessary for a normal functioning of freshwater fish include Na^+, K^+, Mg^{2+}, Ca^{2+}, NH_4^+, Cl^-, HCO_3^-, PO_4^{3-}, and SO_4^{2-}. The ions Na^+, Cl^-, HCO_3^-, Ca^{2+}, K^+, PO_4^{3-}, and Mg^{2+} are involved in osmotic balance. Sodium and potassium regulate the active transport mechanism. The ions K^+, Mg^{2+}, Ca^{2+}, and Cl^- are necessary for activation of various enzymes of the body. All of these ions in solution in natural waters collectively exert osmotic pressure on the aquatic organisms living in the water, and many of these ions are physiologically active, so that freshwater fishes and other animals inhabiting these waters have become adapted to the physical and physiological actions of this salt complex. An influx of additional ions and/or salts could upset this equilibrium between organism and environment. For this reason, and if one is to understand the individual actions (toxic) of saline waters, it is important to consider the physiological response to changes in the osmotic and ionic environment.

Generally, mechanisms of osmoregulation in fishes have been studied by experimental variations in the external media, by the ecological observation of migrating or salt-tolerant fishes, and by evaluating the effects of other extrinsic and intrinsic factors.

The physiological responses of fish to changing environmental osmotic stresses are most easily measured by following osmotic or ionic changes in the blood or other body fluids, or in the composition of the tissues. The principal electrolytes in fish blood are sodium and chloride. When fish are transferred from one salinity to another, any osmotic change is largely accounted for by changes in these ions; however, other electrolytes in the blood change in a similar fashion. A vast body of literature records such changes and many of the major papers have been included in the Bibliography section of this report.

Basically, fish may be grouped by their response to salinity change in the following ways (Parry, 1966):

"1. They may survive only in conditions of isosmotic and isotonic constancy and quickly die if transferred to another environment. They may die as a result of concentration changes in the body fluids following movement of either water or salts. The marine hagfishes are stenohaline in this way, and many fish eggs and embryos also fall into this category.

"2. They may tolerate some degree of change in osmotic or ionic levels in their environment by changing and tolerating in the extracellular and intracellular fluid concentration changes which follow external changes. Some euryhaline fishes are tolerant in this way, but to a limited degree.

"3. They may be 'semi-permeable' and behave as 'perfect osmometers' allowing free water movements in or out of the animal or cells in response to external changes. These animals may tolerate change in body volume or cell volume. Some fish eggs can tolerate changes of this sort to a limited degree or for short periods during development.

"4. In contrast, they may be impermeable to both water and salts and thus isolated from external changes. Among the teleosts, the eel most nearly approaches this sort of behaviour, although it is far from being impermeable.

"5. Some may be able to regulate, that is to compensate for movements of water or solutes caused by external changes, by employing active mechanisms. Many fish use a combination of ways to do this and of responding to a changing environment and many have the potentiality to regulate their body fluids in a changing environment by such physiological regulation."

In summary, resistance to high salinities will depend on a tolerance to internal changes and on active regulatory processes. However, in a number of species, skin permeability is also very much reduced; for example, scaly lateral plates in stickleback (Heuts, 1947).

Toxicity of Saline Waters to Fish

The physiological and toxic effects of saline water, including dilute sea water, on freshwater fish have been studied by many workers, and the earlier literature has been well summarized by Black (1951) and Doudoroff and Katz (1953). The literature, however, has very little that applies specifically to this particular problem (saline groundwater). A great deal of toxicity threshold work has been done but primarily on warm water fish and for short periods with extreme concentrations. Nonetheless, the reviews of the above authors supplemented by more recent literature are discussed. In addition, data abstracted from Water Quality Criteria (McKee and Wolf, 1963) concerning the individual ions of calcium, magnesium, potassium, and sodium and their chlorides are cited in the following sections.

Tolerance to Natural Saline

The total salinity of inland waters usually is dominated completely by four major cations, calcium (Ca), magnesium (Mg), sodium (Na), and potassium (K), and the major anions, carbonate (CO_3), sulfate (SO_4), and chlorides (Cl)(Wetzel, 1975). The terms saline and alkaline have been used interchangeably by some investigators in the past when referring to inland mineral waters. The Nebraska, Minnesota, Saskatchewan, and North Dakota mineral waters may be classified as alkaline whereas the salterns and inland brine lakes of Utah, Nevada, and California are saline types (McCarraher, 1962).

Physiological effects of various salt combinations on pike* reproduction have been recorded from Devils Lake, North Dakota (Young, 1924). The disappearance of pike from the lake primarily resulted from increased salinity and secondarily from the loss of suitable spawning habitat (salinity effects on vegetation).

Rawson and Moore (1944), who investigated relationships between salinity and biological characteristics of 60 lakes in Saskatchewan, found walleye* and perch* in waters with a total salinity (TDS) of 8,000 mg/1. Most of this salinity was of the sulphate salts. Many other fish species were found in waters with a salinity greater than 1,000 mg/1. They stated "...The decrease in the number of species beyond 7,000 mg/1 suggests that higher salinities are detrimental to most species..." Pike apparently disappeared from water having more than 6,034 mg/1 TDS. In a later paper Rawson (1946) recorded the successful introduction and growth of whitefish* and walleye* in a lake with a salinity of 15,000 mg/1.

It appeared that water in saline lakes in North Dakota with an early summer TDS concentration of 7,000 mg/l was not detrimental to reproduction and growth of fathead minnows* (Held, 1971). However, no fish were present in North George Lake which had a mean of 23,489 mg/l TDS. In Nebraska lakes of the sodium potassium bicarbonate type, spawning was not successful at TDS greater than 2,000 mg/l (McCarraher and Thomas, 1968), whereas in Saskatchewan lakes of the sodium sulfate types, populations of fatheads were found in TDS of nearly 15,000 mg/l (Rawson and Moore, 1944).

Tolerance to Salt Water

Most workers seem to be of the opinion that in a physiologically balanced salt solution (such a sea water) lethal effects are the result of osmotic pressure rather than the specific toxicity of individual components of the solution. Most freshwater fishes are unable to maintain nearly normal composition (salt content) of their body fluids in a strongly hypertonic (that is, osmotically much more concentrated) medium, such as sea water. The freezing point (or its depression below that of pure water) for the normal blood of freshwater fishes, which is a measure of its osmotic pressure, has been reported to be in the neighbourhood of -0.5°C.

Garrey (1916) found that in diluted sea water with approximately corresponding osmotic pressure (or freezing point lowering), minnows *(Notropis blennius)* were able to live indefinitely (more than 6 weeks). However, in pure NaCl solutions with freezing points near -0.5°C (0.76-0.9% NaCl), the survival time averaged only 2 to 4 days.

Gueylard (1924a) reported that the survival time of three kinds of minnow *(Cyprinopsis, Leuciscus, Gobio)* in diluted sea water with a freezing point depression of -0.6°C averaged only 6 to 8 days. Perch* *(Perca fluviatilis)* lived as long at a slightly higher salinity. Tests of lower salinities with minnows and perch were not continued for longer periods, so that salinities tolerated indefinitely by these fish were not determined.

Duval (1925) acclimatized the carp *(Cyprinus carpio)* to salt water by addition of small amounts of sea salt once or twice daily from 5 to 50 days until the desired external concentration was reached, whereupon he determined the freezing point depression of the blood. He succeeded in raising the external medium to a salinity above 19‰ and his data show that this species appears to regulate a salinity of concentration until the external concentration reaches a salinity of around 11‰ after which the blood concentration follows rather closely that of the external medium.

The available data indicate that the concentrations of NaCl solutions corresponding to the normal bloods of freshwater fishes with respect to osmotic pressure are generally between 8,000 and 9,000 mg/l NaCl. It has been reported that some hardy freshwater fishes, such as goldfish *(Carassius auratus)*, can withstand indefinitely, or for long periods, salinities as high as 14,000 to 15,000 mg/l (Pora, 1939; Veselov, 1949; Black, 1951).

There is, however, considerable species variation. Pora (1937) found that freshwater fish representing five different genera could not survive in sea water having an osmotic pressure considerably lower than the blood. He reported that *Cottus*, *Cobitis*, *Gobius*, *Leuciscus*, and *Barbus* could not tolerate for 12 days a balanced solution of salts from artificial sea water with a salinity of only 4,000 mg/l. Larger individuals of each species were decidedly less resistant than small ones.

Black (1951) reviewed the literature on the osmotic regulation of teleost fishes and the effects on freshwater and euryhaline fishes of saline waters, with special reference to sea water. Black (1957) also discussed the characteristics which appear to regulate the ability of various species to tolerate increased osmotic pressures.

Renfro's (1959) data indicates that many freshwater fishes have the physiological ability to tolerate brackish waters for short periods of time (Table 7).

TABLE 7. A summary of some toxicity (lethal and non-lethal) data for various species of fishes.

A. Seawater

Species	Toxicity	Comments	Authority
Carassius auratus (goldfish)	SW[1] > 15 ppt[2] (lethal)		Black (1951)
Lampetra planeri (freshwater lamprey)	6 hr ST[3] at 270 mM[4] 2-3 hr. ST pure SW	NaCl sol'ns <120 mM non-lethal. Adults less resistant than ammocoetes.	Hardistry (1956)
Notropis venustus (Spottail shiner)	SW > 12 ppt (lethal)	pH 7.0-7.5 Sol'ns prepared from sea salt crystals.	Renfro (1959)
Notropis lutrensis (Redhorse shiner)	11.5 ppt (non-lethal)	Exp. > 9 days.	
Gambusia affinis (Mosquito fish)	15-24 ppt (lethal)	Exp. > 29 days.	
Gambusia hurtadoi (Mosquito fish)	17 ppt (critical)	Exp. > 21 days SW ≤ 15 ppt no deaths.	
Micropterus salmoides (Largemouth bass)	13 days ST at 14 ppt 9-15 ppt (slow lethal effect)	Exp. > 7 days Finglerings (standard length SL 50-155 mm).	
Lepomis punctatus (Spotted sunfish)	≥ 15 ppt (lethal)	Exp. 10 days SL of 78-90 mm.	
Etheostoma fonticola (Fountain darters)	>13 ppt (lethal)	Exp. > 6 days.	
*Culea inconstans** *(Brook stickleback)	60% SW (lethal within 43 hr)	Temp. 22-27°C Acclimated in step-up stages.	Armitage and Olund (1962)

(Continued)

TABLE 7. Continued.

A. Seawater (cont).

Species	Toxicity	Comments	Authority
Umbra pygmaea (Eastern mudminnow)	15 ppt (lethal) ST < 15 hr for 100% mortality in 15 ppt	Exp. 7 days Smaller fish always died first.	Hoese (1963)
Micropterus salmoides	96 hr TLm[5] 38% SW	Finglerings (350-420 mm)	Tebo and McCoy (1964)
Lepomis macrochirus (Bluegill)	96 hr TLm 29% SW (fingerlings) 11 day TLm 13% SW	Fry 14-21 mm pH 7.9-8.0	
Ictalurus punctatus (Channel catfish)	Eggs tolerant to 16 ppt At hatching 8 ppt	"Instant Ocean" Tolerance 9-10 ppt after yolk absorption. 12 ppt for 6 mon.-1 yr. fish.	Allen and Avault (1969)
Carassius auratus	ST 2-3 hr at 50% SW	Dies within 1/2 hr in 100% SW	Lahlou *et al.* (1969)
Cyprinus carpio (Carp)	SW ≥ 13% (lethal to juveniles)	Exp. 10 wks Egg hatchability affected in SW ≥ 7%	Al-Hamed (1971)
Ictalurus punctatus and *I. furcatus* (Blue catfish) hybrids	SW > 33% (lethal in 12 hr) Hybrids tolerated 14-15 ppt for 96 hr	Temp. 25-28°C Fish SL 40-71 mm	Stickney and Simco (1971)

(Continued)

TABLE 7. Continued.

A. Seawater (cont).

Species	Toxicity	Comments	Authority
Salvelinus alpinus (Arctic char)	ST 72 hrs in 100% SW 100% survived in 33,50 and 66% SW	100% SW has composition: Na,420 mM; Cl, 535 mM; K, 10 mM.	Roberts (1971)
Perca fluviatilis (Perch)*	Died in 50% SW	Perch survived 33% SW for 3 mon. test period.	Lutz (1972)
*Culea inconstans**	96 hr TLm ≅ 21 ppt	Temp. 20±1°C 96 hr acclimation in 15 ppt did not increase salinity tolerance.	Ahokas (1973)
I. cyprinellus (Bigmouth buffalo)	ST 1 hr in 20 ppt 4 hr in 15 ppt (lethal to yearlings)	D.O. 9 mg/l Temp.19-21°C pH 8.5 Eggs tolerated 15 ppt.	Hollander and Avault (1975)

B. Calcium chloride ($CaCl_2$)

Species	Toxicity (conc.in mg/l)	Comments	Authority
Notemigonus crysoleucas (golden shiner)	ST 143.5 hrs at 5,000 6.4 hrs at 20,000	Number (n) of fish 4-5 Size 90-115 mm	Wiebe *et al.* (1934)
Bream	48.8 hrs. at 10,000 19.5 hrs at 20,000	n 10-11 Size 20-50 mm	

(Continued)

TABLE 7. Continued.

B. Calcium chloride (cont).

Species	Toxicity (conc. in mg/1)	Comments	Authority
Lepomis macrochirus	96 hr TLm 10,650	18 ± 2°C D.O. 5-9mg/1.	Trams (1954) Patrict *et al.* (1968)
	24 hr TLm 8,350	3,036 for Ca.	Dowden and Bennett (1965)
Gambusia affinis	96 hr TLm 13,400	Turbid water 24 hr TLm and 48 hr TLm 13,400.	Wallen *et al.* (1957)

C. Potassium chloride (KCl)

Species	Toxicity (conc. in mg/1)	Comments	Authority
Notropis blennius (Straw-colored minnow)	ST 12-29 hr at 373	Distilled water.	Garrey (1916)
Carassius auratus	4 2/3 - 15 hr at 74.6	Distilled Water.	Powers (1917)
Lepomis macrochirus	96 hr TLm 2,010	Synthetic.	Trama (1954) Patrick *et al.* (1968)
	24 hr TLm 5,500	Standard reference water.	Dowden and Bennett (1965)
Gambusia affinis	96 hr TLm 920	24 hr TLm 10,000 48 hr TLm 4,200	Wallen *et. al.* (1957)

(Continued)

TABLE 7. Continued.

D. Magnesium chloride ($MgCl_2$)

Species	Toxicity (Conc. in mg/l)	Comments	Authority
Notropis blennius	ST 4-6 days at 476	Distilled water.	Garrey (1916)
Carassius auratus	ST 78 hr - 21 days at 6,757	Distilled water.	Powers (1917)
Notemigonus crysoleucas	96.5 hrs at 5,000	Distilled water.	Wiebe *et.al.* (1934)
	0.5 hrs at 20,000	n 3-4 Size 95-115 mm $MgCl_2$ kills fish more rapidly than NaC or $CaCl_2$ at all conc.	
Lepomis macrochirus	96 hr T_{Lm} 16,500	Turbid water.	Wallen *et al.* (1957)

E. Sodium chloride (NaCl)

Species	Toxicity (Conc. in mg/l)	Comments	Authority
Notropis blennius	ST 9-24 days at 2,500	Distilled water.	Garrey (1916)
Carassius auratus	17 hrs at 11,765	Distilled water.	Powers (1917)
Notemigonus crysoleucas	148 hrs at 5,000	Distilled and tap water n 5-6	Wiebe *et al.* (1934)
	1.33 hrs at 20,000	Size 85-115 mm	
Micropterus salmoides	200-250 hrs at 5,000	pH 7.8-7.9 D.O. 7-8 mg/l	
Carassius auratus	4-10 days at 10,000 30-40 min. at 50,000 ≤ 5,000 (non-lethal)	Mississippi R. water pH 7.7 Exp. > 25 days Specific conductance (245-62,699 micromhos/cm) at NaCl conc. of 10-50,000 mg/l.	Ellis (1937)

(Continued)

TABLE 7. Continued.

E. Sodium chorlide (cont.)

Species	Toxicity (Conc. in (mg/l)	Comments	Authority
Gambusia affinis	NaCl MTT[6] 10,670 Brine MTT 12,690	Pure NACl sol'n (tap water) Oil field brine (calculated hypothetical conc. NaCl)	Clemens and Jones (1955)
Notropis lutrensis (Red shiner)	9,513 (NaCl) 8,803 (brine)	n 10 Temp. 23°C	Clemens and Jones (1955)
Lepomis cyanellus (Green sunfish)	10,713 (NaCl) 9,349 (brine)	The thresholds for the green sunfish, red shiner, and fathead minnow were lower in the brine waste then in NaCl. For these species the ions may have been acting synergistically or perhaps the greater toxicity of the brine might have resulted from the presence of toxic soluble oil products.	
*Pimephales promelas** (Fathead minnow)	8,718 (NaCl) 7,212 (brine)		
Ictalurus melas (Black bullhead)	7,994 (NaCl) 8,569 (brine)		
Fundulus kansae (Plains killifish)	16,000 (NaCl) 18,783 (brine)	n 10 Temp. 15°C	
Lepomis macrochinus	96 hr TLm 12,946	18 ± 2°C D.O. 5-9 mg/l.	Trama (1954) Patrick *et al.* (1968)
	24 hr TLm 14,125		Dowden and Bennett (1965)
Gambusia affinis	96 hr TLm 17,550	24 hr and 48 hr TLm 18,100	Wallen *et al.* (1957)

(Continued)

TABLE 7. Continued.

E. Sodium chorlide (cont.)

Species	(Conc. in Toxicity (mg/l)	Comments	Authority
Poecilia reticulatus (Guppy)	NaCl > 40 g/l (lethal to all ages)	5-10 g/l (non-lethal)	Amouriq (1970)
Carassius auratus	96 hr LC 50[7] 7,341	Threshold LC 50 (6 days) 7,322 ± 224 Fish age 5 mon - 1 1/2 yr Mean weight 1.37 - 2.70 g	Adelman *et al.* (1976)
*Pimephales promelas**	96 hr LC 50 7,650	Threshold LC 50 (6 days) 7,650 ± 234 Age 11 wk old Mean weight 0.12 - 0.38 g.	

[1]SW - seawater (diluted).

[2]ppt - parts per thousand.

[3]ST - survival time.

[4]mM - millemoles per litre.

[5]TLm - Median Tolerance Limit. (equivalent to LC 50).

[6]MTT - Median Toxicity Threshold (96 hr).

[7]LC 50 - Median Lethal Concentration (concentration which results in 50% mortality, usually within a specified period of time, e.g. 96 hr LC 50).

Armitage and Olund (1962) exposed brook stickleback* *(Culaea inconstans)* to dilutions of sea water (Table 7). At each concentration, the level of activity, measured as the response of tapping on the side of the aquarium, of the fish was noted. The fish were fed once during each 24 hours and the feeding activity was also recorded.

The fish appeared normal in 10% and 20% sea water. They were less active in 30% sea water, but still fed and responded to tapping on the aquarium. All responses became more sluggish in 40% sea water. Less than half of the fish fed in 50% sea water. There was no feeding activity in 60% sea water and all fish were dead at the end of 43 hours. Also, all the fish turned black in 60% sea water--a colouration identical to the breeding colour.

Armitage and Olund (1962) also measured oxygen consumption in different salinities. On the basis of oxygen consumption, 10% sea water was readily tolerated by the fish. However, 20% sea water induced a marked stress; oxygen consumption increased markedly, but normal activity was maintained. At all concentrations of sea water above 20%, the rate of oxygen consumption decreased, indicating that the fish could not satisfy their metabolic requirements. This decrease in oxygen consumption was paralleled by a decrease in activity. That the decrease in metabolism resulted from an inability to obtain sufficient oxygen was supported by counts of opercular movements. These movements increased from about 100 per minute in fresh water to about 170-180 per minute at 10%, 20%, and 30% sea water.

Ahokas (1973), using *Culaea inconstans**, investigated salinity tolerance, and changes in blood and muscle osmotic properties in relation to changing environmental salinity. At 20±1°C, the 96 hour TL 50 value for brook stickleback was about 21 ppt sea water (Table 7). Salinity tolerance did not increase for *C. inconstans* with 96 hour acclimation in 15 ppt sea water. Blood osmolality increased to nearly isosmotic proportions with the environment at salinities above the tolerance level, followed by death.

Stenohaline fishes (either freshwater or marine) maintain the constancy of their internal fluids only when the salinity of the environment remains within a limited range. Their osmoregulatory mechanisms cannot cope with rapid or wide fluctuations in external salinity. Lahlou *et al.* (1969) attempted to find out how a stenohaline freshwater fish, the goldfish, adapts to a hyperosmotic environment (diluted sea water) and how its means of adaptation differ from those of a truly euryhaline fish.

Pronounced melanization occurred in some goldfish placed in diluted artificial sea water (Na=190 meq/1). This indicates that high salinity produces some endocrine stimulation.

Although goldfish survive in a hyperosmotic environment, they do not maintain the osmotic concentration of their plasma at levels found in normal freshwater fish. Fish in this medium (190 meq/l of Na) increase Na outflux about 20 times. They also increase their Na influx and drinking rates and decrease free water (renal) excretion.

The goldfish appears to be stenohaline chiefly because it lacks the ability to excrete enough Na actively across the gills to balance the increased Na intake from the hyperosmotic environment.

Allen and Avault (1969) found that channel catfish *(Ictalurus punctatus)* were unable to maintain themselves at salinities greater than 12 mg/l (brackish water). A salinity of 5 mg/l had no long-term effects on survival or growth of 1 to 4 month old fish while 11 to 14 month old fish were not affected by 10 mg/l.

Allen and Avault (1971) also compared the salinity tolerance of the channel catfish to the blue catfish *(Ictalurus furcatus)* and found the blue catfish more tolerant. Both species took food at 14 mg/l but lost weight, and mortalities were high over the 37 day test.

Roberts (1971) determined the salinity tolerance and regulatory ability of a "land-locked" freshwater population of Arctic char *(Salvelinus alpinus)*. He found that fish transferred directly from fresh water to sea water usually died after 24 to 48 hours and none survived after 72 hours. Survival in sea water was improved by prior acclimatization to dilutions of sea water. Survival was 100% in salinities of 33, 50, and 66% sea water, but in more hypertonic salines mortality increased. Only 50% of the fish transferred to 80% sea water were alive after 7 days, and of these only a half survived the transfer to full-strength sea water.

Total body-sodium analyses of char which died in 80 and 100% sea water showed a Na content much higher than survivors in full-strength sea water, indicating that mortality was probably the result of a failure of the osmoregulatory system and not due to extraneous factors.

Al-Hamed (1971) exposed carp *(Cyprinus carpio)* directly to various levels of salinity for 10 weeks and found that levels of 13% and greater were lethal to juvenile fish. With a gradual 4 day acclimation period, fish could live in salinities as high as 17%. Egg hatchability was affected at concentrations of 7% or greater.

Lutz (1972) measured the ionic and osmotic stress on perch* *(Perca fluviatilis)* placed in various dilutions of sea water. In 33% sea water, perch survived for the three month test period with K, Mg, and Cl in the plasma, and K and

Ca in the muscle showing some increase above the normal. However, perch failed to survive in 50% sea water evidently due to a breakdown of ionic control, as all ions increased greatly in the body. Sodium and chloride invaded the muscle cells in large amounts and all cells lost water.

Tolerance to Individual Salts and Ions

Many workers have studied the effects of single salts or unbalanced salt solutions upon freshwater fishes.

Hirsch (1914) determined the highest concentrations of several salts, dissolved in tap water, which could be tolerated by eels (more than 6 days). These threshold concentrations were reported to be N/2 NaCl; N/4 $MgCl_2$, $MgSO_4$ and $CaCl_2$; and less than N/100 KCl.

Garrey (1916) exposed minnows *(Notropis blennius)* for prolonged periods to solutions of Na, Ca, Mg, and K chlorides prepared with distilled water. In 0.025 M and 0.0075 M $CaCl_2$ solutions, fish died in 2 to 4 days and 14 to 21 days, respectively. At the next lower concentration of $CaCl_2$ (0.0025 M), they lived for 5 to 7 weeks (end of experiment). In 0.025 M $MgCl_2$, 0.005 M $MgCl_2$, and 0.005 M KCl solutions, the fished lived only for about 2 days, 4 to 6 days, and 12 to 29 hours (average 24 hours) respectively. In a 0.25 per cent

(0.043 M) NaCl solution, the minnows died in 9 to 24 days.

Garrey's data indicate that potassium was decidedly more toxic than magnesium, which was more toxic than calcium; and that sodium was the least toxic of the metals tested (as chlorides).

Powers (1917) studied the toxicity of a number of metal salts to goldfish. The experimental results were presented in tables and graphs, and certain indices of the "relative toxicity" of the salts were computed. Powers noted that his measure of relative toxicity which he described as "natural criterion" is not universally applicable. When applicable, it still can be very misleading, as it involves an unusual concept of relative toxicities.

Powers (1921a) also reported results of some similar experiments on the toxicity of Na, Ca, Mg, and Ba chlorides to bluntnose minnows. The most dilute solution of each salt tested was 0.025 M. This concentration sooner or later proved fatal to minnows in every case.

Krüger (1928) presented results of a detailed study of the cause of death of sticklebacks in pure (unbalanced) NaCl solutions.

Wiebe *et al.* (1934) determined the average survival time of 3 to 6 golden shiners *(Notemigonus crysoleucas)* (Table 7) to lethal concentrations of Na, Ca, and Mg chlorides. The water used was a mixture of distilled and tap waters with pH 7.8 to 7.9. Their results show that $MgCl_2$ kills fish more rapidly than NaCl or $CaCl_2$ at all concentrations used. In 5,000 mg/l, $MgCl_2$, $CaCl_2$, and NaCl solutions, the survival time averaged 96.5, 143.5, and 148 hours respectively, and in 10,000 mg/l solutions it averaged 4.6, 27.6, and 97 hours respectively. NaCl however, killed fish more rapidly at 20,000 and 15,000 mg/l than did $CaCl_2$ of the same concentration. The difference between $CaCl_2$ and NaCl at 20,000 and 15,000 mg/l was believed to be due to the difference in the rate of absorption of the two salts by the blood.

Edmister and Gray (1948) reported the "toxicity thresholds" of $CaCl_2$, NaCl, and KCl for 24 hour old fry of lake whitefish* *(Coregonus clupearformis)* in Lake Erie water to be 22,080, 16,500, and 10,368 mg/l respectively. Corresponding values for fry of yellow walleye* *(Stizostedion vitreum vitreum)* of the same age were reported to be 12,060, 3,859, and 751 mg/l, respectively.

Abegg (1949, 1950) reported the 24 hour Median Tolerance Limits (TL_m) of NaCl, $CaCl_2$, and KCl for bluegill *(Lepomis macrochirus)* in a synthetic river water to be 14,100, 8,400, and 5,500 mg/l, respectively.

House (1958, 1959) examined the toxicity of major cations (Ca, Mg, and Na) as the chloride salts on rainbow trout *(Salmo gairdneri)*. He showed that the toxicity of $CaCl_2$ was significantly higher than the toxicity of either Na or $MgCl_2$. At 5,000 mg/l, mortality in $CaCl_2$ was 50%; NaCl, 45%; $MgCl_2$, 15%, while at 7,500 mg/l, it was 95%, 20%, and 10%, respectively.

The low toxicity of mixed, physiologically balanced solutions of chlorides (e.g., NaCl and $CaCl_2$) is evidence that chloride ions are relatively harmless, even when compared with ions of sodium, the least toxic of the metals (Doudoroff and Katz, 1953). The specific toxicity of NaCl alone is evidently attributable to Na ions.

The Anion Chloride

Hart *et al.* (1945) cite data indicating that among U.S. waters supporting a good fish fauna, ordinarily the concentration of chlorides is below 3 mg/l in 5%; below 9 mg/l in 50%, and below 170 mg/l in 95% of such waters.

The following concentrations of chloride have been reported to be harmful to fish (McKee and Wolf, 1963):

Concentration in mg/l	Type of Fish	Authority
400	trout	Adams (1940)
2,000	some fish	Brenneke (1945)
4,000	bass, pike*, perch*	Adams (1940)
4,500-6,000	carp eggs	Nakamura (1948)
8,100-10,500	small bluegills	Wood (1957)

The salinity of brine water from oil fields, expressed as total chloride ion concentration, has a range of toxic threshold levels from 4,900 to 12,700 mg/l chloride for several freshwater fish species (Clemens and Jones, 1954; Wood, 1957). In the River Werra, freshwater fish died mainly at above 6,000 mg Cl/l (Schmitz, 1956) and in the River Wipper, at values exceeding 3,000 mg Cl/l.

A personal communication from Shaw (Falk and Lawrence, 1973) stated that drilling fluid sump with a chloride concentration greater than 500 mg/l were usually lethal to trout.

"Metal chloride toxicity would be the result of metal cations or the osmotic pressure from all cations and chloride ions, not specifically chloride ions alone. The above value of 500 mg/l chloride does not appear to be valid" (Land, 1974).

ORSANCO (1956) states that "there is no convincing evidence that chloride ions have any specific toxicity" (also discussed in a previous section). The toxicity of various chlorides is apparently due to the specific toxicities of the cations (e.g., Ca, Mg, K, and Na) present and not to any toxicity of the chloride ion.

The Cations

Calcium. The data of Garrey (1916), Powers (1921a), and Jones (1939a) indicate that calcium chloride and nitrate, when added to distilled or soft waters, can be toxic to fish at concentrations between 300 and 1,000 mg/l as Ca.

A concentration of 277 mg/l of $CaCl_2$ in distilled water has not been harmful to minnows over a period of 5 to 7 weeks. Other available data concerning lethality of higher concentrations of calcium salts (generally $CaCl_2$) in various waters indicate that fish have survived from 1 to 3 days at concentrations of 2,500 to 4,000 mg/l of calcium (e.g., Wells, 1915; Oshima, 1931; Wiebe *et al.*, 1934; Abegg, 1950).

Wiebe *et al.* (1934) found golden shiners *(Notemigonus crysoleucas)* were killed by 5,000 mg/l $CaCl_2$ in 143 hours.

Of the waters of the United States supporting a good mixed fish fauna, ordinarily 5% have less than 15 mg/l of calcium; 50% have less than 28 mg/l; and 95% have less than 52 mg/l (Hart *et al.*, 1945).

Edmister and Gray (1948) reported that fry of yellow walleye* and lake whitefish* were immobilized by 12,060 mg/l and 22,080 mg/l $CaCl_2$, respectively, but survived lower concentrations in Lake Erie water.

The following concentrations of $CaCl_2$ have been reported to have killed or injured fish (McKee and Wolf, 1963):

Concentration (mg/l)	Exposure Time	Type of Fish
555	1 week	rock bass
2,775	2-4 days	minnows
5,000	143 hours	shiners
7,752	22-27 hours	goldfish
10,000	3 hours	tench
10,000	-	sunfish
10,000	27.6 hours	shiners
10,000	48.8 hours	bream
12,060 (threshold)	-	walleye*
22,080 (threshold)	-	whitefish*

The 24 hour TL_m for bluegills in a synthetic river water was reported to be 8,350 mg/l for the salt and 3,036 mg/l for Ca (Dowden and Bennett, 1965). A 96 hour TL_m of 10,650 mg/l $CaCl_2$ has been reported by Trama (1954) and Patrick *et al.* (1968)(Table 7).

Magnesium. Magnesium chloride and nitrate can be toxic to fish in distilled water or tap water at concentrations between 100 and 400 mg/l as magnesium (Garrey, 1916; Powers, 1921a; Jones, 1939a). However, magnesium chloride, nitrate, and sulfate at concentrations between 1,000 to 3,000 mg/l as Mg have been tolerated for 2 to 11 days (e.g., Hirsch, 1914;

Oshima, 1931; Wiebe *et al.*, 1934; Jones, 1939a). Some freshwater fish have been found in saline lake waters containing over 1,000 mg/l of magnesium, as well as much sodium and calcium (Huntsman, 1922).

Among U.S. waters supporting a good fish fauna, ordinarily 5% have less than 3.5 mg/l of magnesium; 50% have less than 7 mg/l; and 95% have less than 14 mg/l (Hart *et al.*, 1945).

The following concentrations of $MgCl_2$ have been reported to have killed freshwater fish (McKee and Wolf, 1963):

Concentration (mg/l)	Exposure Time	Type of Fish
476	4-6 days	minnows
5,000	96.5 hours	golden shiners
6,757	3-21 days	goldfish
8,132	-	carp
10,000	4.6 hours	shiners
15,000	0.8 hours	shiners
16,500	96 hr TL_m	mosquito fish
20,000	0.5 hours	shiners
23,800	24 hours	*Orizias*

Potassium. The survival time of Garrey's (1916) minnows averaged about one day in a distilled-water solution of KCl with a potassium concentration of only 200 mg/l. "Much lower concentrations (perhaps well below 50 mg/l) probably would have proved fatal after more prolonged exposure" (Doudoroff and Katz, 1953).

Threshold concentrations of KCl in tap water and moderately hard lake water near 400 mg/l as potassium have been reported by Hirsch (1914) and Edmister and Gray (1948).

Other investigators have demonstrated the toxicity of higher concentrations, which proved fatal to fish in various waters within two days or less.

The following concentrations of potassium have been reported to have killed fish (McKee and Wolf, 1963):

Concentration (mg/l)	Type of Water	Exposure Time	Type of Fish
74.6	Distilled	4.5-15 hrs	Goldfish
373	Distilled	12-29 hrs	Minnows
751	Lake Erie	-	Walleye*
920	Turbid	96 hr TL_m	Mosquito fish
1,360	Well	3 days	Some perch*
2,010	-	96 hr TL_m	Bluegills
2,300	-	24 hrs	Small freshwater fish
5,500	Synthetic	24 hr TL_m	Bluegills
10,368	Lake Erie	-	Whitefish* fry
12,060	-	24 hr TL_m	Walleye*

Doudoroff and Katz (1953) found it noteworthy that sticklebacks *(Gasterosteus aculeatus)* (Jones, 1939a) could survive one day at 500 mg/l potassium, yet were ultimately killed in less than 7 days by 70 mg/l. This accumulation of toxicity with time for potassium can also be seen in data from Wallen *et al.* (1957). Mosquito fish *(Gambusia)* in pond water demonstrate a TL_m for KCl which decreases in order of magnitude from the 24 hour value (10,000 mg/l) to only 920 mg/l

KCl after 96 hours exposure. The maximum concentration at which all fish appeared normal was 5,600 mg/l at 24 hours, 1,800 mg/l at 48 hours, and 560 mg/l after 96 hours.

Sodium. Krüger (1928) reported that his sticklebacks died a little sooner in 0.05 N NaCl solutions (1,150 mg/l Na) than in solutions which were 2 to 4 times as concentrated (0.1 to 0.2 N). The median survival time averaged about 8 to 9 days in these solutions, but decreased sharply at higher NaCl concentrations.

Gueylard's (1923, 1924) data indicate much greater resistance of the same species. Concentrations of NaCl up to 12,000 mg/l (4,720 mg/l Na) in distilled water and 20,000 mg/l (7,870 mg/l Na) in tap water were found to be harmless. The survival time of three species of the minnow family and of perch (which were somewhat more resistant) averaged 4 to 8 days at NaCl concentrations of 8,500 or 10,000 mg/l (3,340 to 3,930 mg/l Na) in distilled and tap waters (Gueylard, 1924).

In harder, alkaline waters, NaCl has been reported to be toxic to some freshwater fish at concentrations between 1,500 and 2,000 mg/l as Na (Wiebe *et al.*, 1934; Edmister and Gray, 1948). Edmister and Gray reported that fry of yellow walleye* and lake whitefish* were immobilized by 3,859 mg/l and 16,500 mg/l NaCl, respectively.

However, Ellis (1937) found that 5,000 mg/l NaCl (1,970 mg/l Na) in Mississippi River water had no effect on goldfish in 25 days (Table 7); and Ramult (1927a, b) found that salmon eggs and larvae, and also young stickleback, did well in 0.1 N NaCl solutions (about 2,300 mg/l Na) prepared with tap water. Doubling these concentrations resulted in mortality of the experimental animals, but even higher tolerable concentration limits have been reported for eels (Hirsch, 1914) and whitefish* fry (Edmister and Gray, 1948). Trout eggs resumed development after 24 hours exposure to 21,000 mg/l NaCl (Devillers, 1950). Also, higher concentrations of NaCl have been found to decrease the toxicity of some metallic compounds toward fish.

Brook trout have survived and recovered from immersion for 30 to 60 minutes in a 30,000 mg/l solution of NaCl, but 50,000 mg/l caused deaths of 50% of the fish within 15 minutes (Phillips, 1944).

Of the United States waters supporting a good fish fauna, ordinarily the concentration of sodium (Na) plus potassium (K) is less than 6 mg/l in about 5%; less than 10 mg/l in about 50%; and less than 85 mg/l in about 95% (Hart *et al.*, 1945).

The following concentrations of NaCl have killed or immobilized freshwater fish (McKee and Wolf, 1963):

Concentration (mg/1)	Type of Water	Exposure Time	Type of Fish
2,540	soft, distilled	-	fish
	-	120 hrs	shiners
	-	-	fish
	distilled	9-24 days	minnows
5,100	hard, alkaline	-	fish
	natural	-	whitefish* and walleye
	-	-	newly hatched trout
	distilled	148 hrs	shiners
	distilled	200-250 hrs	bass
	distilled	4-8 days	minnows, perch
	-	97-148 hrs	shiners, bass
	tap	4-8 days	minnows, perch
	river	4-10 days	goldfish
	distilled	6 hrs	minnows
10,200	hard	6 hrs	minnows
11,680-14,600	-	-	stickleback
11,700	tap	-	salmon eggs and fry
11,765	distilled	17 hrs	goldfish
12,946	aerated	96 hr TL_m	bluegills
14,000	synthetic	24 hrs	bluegills
	-	-	goldfish
15,000	-	4.7 hrs	bass
15,000-30,000	-	24 hrs	freshwater fish
16,500	natural	-	whitefish*
17,550	turbid	96 hr TL_m	mosquito fish
20,000	-	1.3 hrs	shiners
50,000	-	15 min	brook trout

The following concentrations of NaCl have been reported as harmless to fish in the time specific (McKee and Wolf, 1963):

Concentration (mg/1)	Type of Water	Survival Time	Type of Fish
5,000	river	25 days	goldfish
5,850	tap	-	salmon eggs and larvae, and young stickleback
9,100-17,500	-	30 days	perch
11,700	-	50 hrs	young eels
12,000	distilled	-	stickleback
20,000	tap	-	stickleback
25,000-30,000	-	-	older trout
30,000	-	30-60 mins	brook trout

It is indicated that perch* are able to acclimatize somewhat to gradually increasing concentrations of NaCl. Some perch have survived for as long as a month in gradually increasing concentrations to end point concentrations of 9,100 to 17,500 mg/1 (Doudoroff and Katz, 1953).

Anderson (1948) considered NaCl concentrations above 3,000 mg/1 deleterious to both fish-food organisms and fish fry. He recommended a permissible limit of 2,000 mg/1 in fresh waters.

Gordon (1959a) tentatively proposed that euryhalinity in the brown trout *(Salmo trutta)* was correlated with the ability to regulate intracellular Na concentration. Brown trout tolerating the full transfer from fresh water to sea water, via one-half sea water, showed an almost constant intracellular (muscle) Na concentration in spite of an elevated plasma concentration whereas fish dying showed invasion of the cells by Na.

Dowden and Bennett (1965) determined the 24 hour TL_m for bluegill as 14,125 mg/1 NaCl; the 96 hour TL_m was 12,946 mg/1 (Table 7). Wallen *et al.* (1957) reported higher TL_m's of 18,100 mg/1 NaCl and 17,550 mg/1, respectively.

The effect of NaCl at concentrations of 5, 10, 15, 30, 40, 41, and 45 g/1 was studied on the motor activity of the guppy *(Poecilia reticulatus)* by Amouriq (1970). Concentrations above 40 g/1 were lethal to all ages, while 5 to 10 g/1 had no toxic effects on any age group. However, the motor activity was affected at all salt levels tested.

Sharp (1971) reported bluegills will tolerate 10,000 mg/1 NaCl while rainbow trout sustained 20% mortality at 12,000 mg/1.

The lethal concentrations (LC50's) of four toxicants on fathead minnows* *(Pimephales promelas)* and goldfish were determined and the resistance of the two species were compared by means of toxicity curves (Adelman *et al.*, 1976). It was found that most mortality from NaCl occurred within the first 48 hours, after which the toxicity curves paralleled the abscissa. Goldfish were initially more resistant than fathead minnows but after 48 hours became significantly less resistant with a threshold LC50 of 7,322±224 mg/1 for goldfish and 7,650±234 mg/1 for fathead minnows (Table 7).

In summary, potassium chloride (KCl) appears to be the most toxic of the chlorides to fish, with sodium chloride (NaCl) being the less toxic.

Antagonism and Synergism of Cations

Antagonism is reduced toxicity due to the presence of ions which counteract each other. Synergism is increased toxicity due to the presence of ions which act together.

Many investigators have noted that mixed (physiologically balanced) salt solutions are less harmful than, for example, simple NaCl solutions with corresponding osmotic pressures. A physiologically "balanced" salt solution is one in which different salts, and particularly Ca, Mg, K, and Na ions (the principal cations found in natural waters) occur in such proportions that the specific toxicity of each is neutralized or reduced through the antagonistic action of the other salt or ions.

Loeb and Wasteneys (1911, 1915) reported some of the earliest studies of the antagonism of the principal cations of sea water to fish.

Garrey (1916) determined the average survival times of minnows *(Notropis)* in various simple and mixed solutions of

Ca, Mg, K, and Na chlorides in distilled water. The following data, showing salt concentrations and corresponding survival times, illustrates the antagonism of some of the cations (from Doudoroff and Katz, 1953):

Salt Concentrations	Survival Time
9,000 mg/1 NaCl (about 0.15 M)	2 days
9,000 mg/1 NaCl and 0.001 M $CaCl_2$ (additional $CaCl_2$ produced no more favourable results)	8-10 days
0.025 M $CaCl_2$ (about 2,775 mg/1)	2-4 days
0.025 M $CaCl_2$ and 6,750 mg/1 NaCl	8-10 days
0.025 M $MgCl_2$ (2,380 mg/1)	2 days
0.025 M $MgCl_2$ and 0.01 M $CaCl_2$	6 days
0.005 M KCl (373 mg/1)	1 day
0.005 M KCl and 0.0005 M $CaCl_2$	6+ days
0.005 M KCl and 5,175 mg/1 NaCl	2-3 days
0.015 M KCl (1,118 mg/1)	20 hours
0.015 M KCl and 0.0075 M $CaCl_2$	13 days
0.015 M KCl and 0.005 M $MgCl_2$	3-5 days

It should be noted that relatively small amounts of $CaCl_2$ added to toxic solutions of Na, Mg, and K chlorides markedly reduced their toxicity, as little as 20 to 40 mg/1 of Ca being highly effective against NaCl and KCl. High concentrations of NaCl counteracted the toxicity of $CaCl_2$ and KCl; and $MgCl_2$ also was antagonistic to KCl.

Powers (1921) also studied the reciprocal antagonism of NaCl and $CaCl_2$, using minnows. Reduction of the lethality of NaCl solutions by the addition of small amounts of $CaCl_2$ was clearly shown.

Krüger (1928) found that 0.35 M and 0.4 M NaCl were rapidly fatal to sticklebacks *(Gasterosteus aculeatus)*, the median survival time averaging about 2 hours. In 0.4 M mixed solutions prepared by adding KCl, $MgCl_2$, $CaCl_2$, or combinations of two or more of these salts to 0.35 M NaCl, the fish invariably lived longer. However, all the mixtures containing no Ca proved more rapidly fatal than a mixture of NaCl and $CaCl_2$ only. Median survival times of fish in the NaCl-$CaCl_2$ solution ranged from 13 to more than 30 days.

House (1958, 1959) determined the concentrations of combinations of salts that rainbow trout will tolerate over an extended period. His experiments indicated that calcium reduces the toxicity of potassium, a finding similar to Garry (1916). He also showed that $MgCl_2$ reduced the toxicity of $CaCl_2$ if present in a large enough quantity. The same antagonistic effect on $CaCl_2$ appeared to be true of NaCl. Beatty (1959) and Hepworth (1965) also discussed antagonistic and synergistic actions of certain ions on rainbow trout. For example, the presence of NaCl reduced the toxic effects of sodium sulfate (Na_2SO_4) when the two salts were combined (Beatty, 1959). The 8,500 mg/l level of the two salts which contained about 8,000 mg/l Na_2SO_4 had a total mortality of 50% in six weeks, whereas Na_2SO_4 tested singly at 7,000 mg/l had a total mortality of 85% in six weeks. The two salts combined at the 7,500 mg/l level, of which nearly 7,000 mg/l was Na_2SO_4, had a total mortality of 50% in six weeks. This was 35% less mortality than for Na_2SO_4 tested singly at a comparable concentration for the same time period. The synergistic effects of combinations of sodium salts were also studied by Dowden and Bennett (1965).

Hepworth (1965) speculated "that since fish are known to be able to physiologically adapt to chemical changes in their environment, perhaps certain ion combinations or concentrations may trigger this adaptability. Maybe low concentrations institute subtle effects that are lethal where, on the other hand, high concentrations stimulate strong physiological reactions which enable individuals to respond in a way to overcome the toxic ions in their environment."

In summary, sodium chloride is antagonistic to the toxicity of calcium and potassium chlorides. Calcium chloride is, however, the most effective chloride in antagonistic ability, as a relatively small amount reduces the toxicity of sodium or potassium chloride.

Factors Affecting Salinity Tolerance

Seasonal Effects

Seasonal changes, especially the onset of the spawning season, may or may not alter the salinity tolerance of fish. For example, during the summer spawning season, osmotic regulation by sticklebacks in varying salinities is greatly diminished (Gueylard, 1924; Huets, 1943; Black, 1948).

This fact is also supported by Smith's (1956) study on seasonal changes in tolerance of brown trout *(Salmo trutta)* to artificially increased salinities. Salinity tolerance was lowest during the summer, went through a peak, was low again from October to December (the spawning season), then went through a broad peak during the spring.

Gordon (1959a) found that both survival and regulatory ability of the brown trout were lower during the summer. However, they were seemingly constant over the entire period from autumn to spring. Gordon thought these changes in regulatory abilities may be influenced by photoperiod and/or the thermal history of the fish.

Size and Age

Survival and the ability to osmoregulate in different salinities depends upon the size and age of fish (Black, 1951) as well as on the species (Parry, 1960). Parry in her study of three salmonid species concluded that size, rather than age, appears to be the more important factor in their tolerance of higher salinities.

Busnel *et al.* (1946) reported that recently hatched rainbow trout could not withstand a salinity of 5,000 mg/l as NaCl whereas older trout withstood salinities 5 to 6 times as great. Similarly, Hoese (1963), using the eastern mudminnow *(Umbra pygmaea)*, found that the smaller (presumably younger) individuals always died first in salinity tolerance experiments. Speshilov and Agrba (1970) reported that yearling steelhead trout *(Salmo gairdneri)* easily tolerated salinities of 0.6, 11.6, and 17%, while trout under a year had a limited capacity and were quickly killed at a salinity of 6%. Channel catfish fry had a salinity tolerance of about 9-10 ppt but by age 11 to 14 months had increased their upper tolerance to 12.5 ppt (Allen, 1971).

Pora (1937) on the other hand, found that large individuals of some freshwater species were less resistant to salt water than small ones. Kinne (1960b) observed that the ability of the desert pupfish *(Cyprinodon macularius)* to tolerate salinity decreases with increasing age. One month old fish could tolerate a sudden salinity change of 20-35 ppt whereas one year old fish were unable to survive a change of more than 10-15 ppt. "This decreasing plasticity seems to be related to the growth rate: fast-growing fish readjust and adapt faster. Not only the rate of adaptation but also its intensity decreases with age--witness the intense, partially irreversible adaptation to salinity in eggs and freshly hatched fry" (Kinne, 1960a).

Hardisty (1956) noted that ammocoetes of the freshwater lamprey *(Lampetra planeri)* were more resistant to higher concentrations of sea water than the adult. Drilhon (1943) also noted that small eels survived better than large ones. This might be correlated with Key's (1931) work on *Fundulus* which indicated that the higher the ratio of head length (gill surface) to body length, the greater the ability of the fish to survive salinity changes.

Acclimation

The degree of resistance of freshwater fish to different salinities may be increased by acclimation (previous history). Clemens and Jones (1954) demonstrated that the plains killifish could withstand 8,649 mg/l more NaCl when it had been living in waters containing 6,329 mg/l Cl^- than when it had been in waters containing less than 100 mg/l Cl^-. Mitchum (1961) reported that offspring of brook trout *(Salvelinus fontinalis)* living in saline water are more tolerant of the toxic effects of major ions (SO_4, CO_3, Cl, Ca, Mg, Na, K) than offspring of brook trout living in soft water.

Temperature

The temperature may have an effect on the salinity tolerance of fish (Bishai, 1961). Strawn and Dunn (1967) and Hill and Carlson (1970) reported that as temperatures decreased, optimum salinities for survival decreased for freshwater fish.

Stanley and Colby (1971) have shown a detrimental effect of low temperature on salt water osmoregulation in the alewife *(Alosa pseudoharengus)*. The experiments of Davis and Simco (1976) suggest that low temperatures (e.g., winter months) may limit the salt tolerance of channel catfish.

On the other hand, salinity has been found to increase resistance to high temperatures. Loeb and Wasteneys (1912) showed that dilutions of sea water or Ringer's solution increased the resistance of *Fundulus heteroclitus* to high temperatures above that in fresh water. Arai *et al.* (1963) speculated that the protective action of sea water is possibly due to increased internal salt concentration which increases the resistance to higher temperatures. Strawn and Dunn (1967) also indicated that the presence of some dissolved solids (salts) in the water benefited both freshwater and salt marsh fishes during exposure to high temperatures.

Water Hardness

Hard water not only has more calcium than soft water but nearly always more of other ions, e.g., chloride, sulfate, and sodium, and therefore a higher osmotic pressure. It is important to consider the effect water hardness may have on the salinity tolerance of fish. Calcium not only affects the efficiency of osmoregulation, but it as well as magnesium affects the rate of respiration (Schlieper *et al.*, 1952).

Effects of Salinity on Reproductive Success and Early Life

Bogucki (1930) found that perivitelline membrane formation in trout eggs was impeded in hypotonic salt solutions. Similarly, Fisher and Warren (1948; in Black, 1951) showed that hardening of speckled trout eggs did not occur in or above a salinity of 6‰.

Busnel (1943) stated that the Service des Eaux et Forêts (France) found better and more rapid hatching of rainbow trout eggs in brackish water and that the adults thrived in a saline environment because their parasites were killed by the salt water. However, Busnel *et al.* (1946) found that alevins after hatching sustained mortality as follows:

‰ NaCl	% Mortality	No. of Days
0	10	50
5	80	45
10	100	31
15	100	13

The question of oxygen consumption on fresh water and salt water was also investigated. Alevins and fry showed an oxygen consumption about 8% lower in 5‰ NaCl than in fresh water. In 10‰ NaCl oxygen consumption decreased 35%.

Tebo and McCoy (1964), using bioassay methods, determined the sea water concentrations which were lethal to the eggs, fry, and fingerlings of the largemouth bass *(Micropterus salmoides)* and the bluegill *(Lepomis macrochirus)*. On the basis of their bioassay studies, it appears that approximately 10 to 12% sea water is the maximum concentration at which bass and bluegills can successfully reproduce. Swingle (1956) stated that bass could reproduce at higher salinities than could bluegills. Neither could reproduce at a salinity of 5 to 5.5‰. Fingerlings of these species can survive, at least for short periods, in concentrations of 29 to 38% sea water (Tebo and McCoy, 1964).

Increasing levels of salinity generally result in retardation of the rate of embryogenesis in fishes (Kinne and Kinne, 1962). Higher numbers of structures are also generally associated with such conditions (Hubbs, 1926; Taning, 1952). When delay is caused by unusual levels of salinity, a greater amount of energy may be needed for osmoregulatory work in the developing embryo, and hence, growth of the embryo is retarded (Tay and Garside, 1975).

Allen and Avault (1969) and Allen (1971) determined the effect of salinity on survival of channel catfish *(Ictalurus punctatus)* eggs, sac fry, and fry. Eggs obtained at either one or two days of age were found to increase in tolerance at first, presumedly due to the formation and hardening of the vitelline membrane. At the end of the second day, the upper tolerance was about 10 ppt salinity. By the third or fourth day, the upper tolerance had increased to about 16 ppt where it remained until hatching. At hatching, there was an immediate drop in tolerance to about 8 ppt. This occurred as the vitelline membrane ruptured, permitting the embryo to come in contact with the saline water.

There did not appear to be a change in tolerance during the sac fry stage. However, following yolk absorption, the upper tolerance increased slightly to about 9 or 10 ppt during the fry stage.

Mossier (1971) exposed the eggs and sac fry of the fathead minnow* *(Pimephales promelas)*, northern pike* *(Esox lucius)*, and walleye* *(Stizostedion vitreum vitreum)* to highly saline lake water to learn about hatching success and sac fry survival as influenced by: 1) salinity at fertilization, 2) the salinity of water to which eggs and sac fry were then exposed, and 3) the age of eggs when first exposed. Eggs fertilized in water of 500, 1,300, and 4,000 micromhos, were removed about four hours after fertilization and placed in water of 500, 1,300, 4,000, 6,000, and 12,000 micromhos.

Hatching success was highest ranging from 92.2 to 94.1% for fathead minnow eggs fertilized in water of 1,300 micromhos and held in water of 500, 1,300, 4,000, and 6,000 micromhos, dropping to 54% for eggs held in water of 12,000 micromhos.

Fathead minnow sac fry survival generally decreased as salinity increased, which followed the hatching success trends.

Hatching success ranged from 0.0 to 4.2% for northern pike eggs fertilized in water of 500, 1,300, and 4,000 micromhos and held in water of 4,000, 6,000, and 12,000 micromhos; those fertilized and held in water of 500 and 1,300 micromhos had the highest hatching success ranging from 22.1 to 24.7%. Survival of northern pike sac fry was similar to hatching success trends. Generally, no sac fry from eggs fertilized in water of 500, 1,300, and 4,000 micromhos and held in water of 4,000, 6,000, and 12,000 micromhos survived.

Hatching success of walleye eggs fertilized in water of 4,000 micromhos and treated in water of 500, 1,300, 4,000, and 6,000 micromhos was only 0.7%. Hatching success was highest ranging from 14.8% to 34.4% for eggs fertilized and held in water of 500 and 1,300 micromhos. Survival of walleye sac fry was similar to hatching success trends. No sac fry from eggs fertilized in water of 500, 1,300, and 4,000 micromhos and held in water of 4,000 and 6,000 micromhos survived.

Peterka (1971) found that in water of 1,300 micromhos, eggs of fathead minnow, northern pike, and walleye hatched well, but when salinity reached 4,000 micromhos, there was no hatching of walleye, a very poor hatch of northern pike, but a good hatch of fathead minnow eggs. No northern pike sac fry survived in water of 6,000 micromhos and all fathead minnow sac fry (about 1%) surviving in 12,000 micromhos had physical abnormalities.

Eggs and sac fry of the fathead minnow were subjected by Burnham and Peterka (1975) to various dilutions of water obtained from four saline North Dakota lakes; three were sodium-sulfate-, one a sodium-chloride-type lake. Salinity tolerance tests were run in diluted waters with conductivities of 500, 1,300, 4,000, 6,000, 12,000, and 18,000 micromhos at water temperatures of 25 ± 1°C. In water from the sodium chloride lake (Kelly's Slough), the per cent of fertilized eggs surviving as sac fry for 5 days remained the same as the per cent hatched: about 90% at treatment levels of 500-12,000 micromhos and 68% in undiluted lake water of 18,000 micromhos.

A difference in response between sodium sulfate and sodium chloride water of $\geq$12,000 micromhos was observed in regard to spinal curvature of hatched fry. In dilutions of water from Kelly's Slough of 12,000 micromhos, mean per cent normal sac fry hatched was 96% but only 21-24% for the sodium sulfate type lakes. Burnham and Peterka (1975) also observed that in lake waters of conductivity 8,000 micromhos, spawned fathead minnow eggs lost their adhesiveness.

Hollander and Avault (1975) conducted salinity tolerance tests with eggs through yearlings on buffalo fish (*Ictiobus* spp.). They found the same general pattern of salinity tolerance as observed by Allen and Avault (1969) and Allen (1971) for channel catfish. They felt that the abnormalities in the spinal column, low per cent of normal fry, and low survival of newly hatched fry in 12 and 15 ppt salinities were due to lack of gas exchange since gases diffuse slower in high salinities than in fresh water.

Effects of Salinity on Fish Food Organisms

Anderson (1948) considered NaCl concentrations above 3,000 mg/l deleterious to fish food organisms. In Lake Erie water at 20-25°C, threshold limits for several fish food organisms were reported (Anderson *et al.*, 1948) as follows:

Organism	Threshold Concentration (mg/l)
Daphnia magna (young)	3,700
Daphnia magna (adult)	4,600
Leptodora kindtii	3,700
Cyclops vernalis	6,100
*Diaptomus oregonensis**	3,030

Anderson *et al.* reported the susceptibility of *D. magna* to toxic substances was representative of the susceptibility of predominant zooplanters to toxic substances.

Effects of Salinity on Fish Growth

There is some evidence that fish which can withstand a wide range of salinity will grow better in a more saline environment. For example, Gibson and Hirst (1955) found that guppies *(Lebistes reticulatus)* reared in dilute sea water had a faster rate of growth than those reared in fresh water. However, there was no experimental evidence that the addition of sea water enhanced the size of adult fish. Canagaratnam (1959), however, reported that the goldfish did not show any significant increase in weight when reared in saline water.

Physiological Effects of Salinity

The manner in which salts cause toxicity to fish is not well understood. Jones (1939b) stated that the alkaline metals enter the body and act as internal poisons and that other metal salts bring about death by precipitation of the gill secretions thus causing asphyxiation. He also pointed out that since, in most cases, toxic substances have to penetrate cell membranes and tissues before they can bring about the disturbances of metabolism that result in death, their degree of toxicity is largely determined by their penetrating power.

House (1958) examined tissues from rainbow trout which had been exposed to various salts ($CaCl_2$, $MgCl_2$, $NaCl$). He observed some striking differences between normal fish tissue and that taken from fish exposed to salts:

1) gill tissues showed edema when compared to normal gill tissue,

2) the mucous layer of a fish in $MgCl_2$ had been almost completely eroded. The scale receptacles existed only as disturbed remanents. Tissue from fish subjected to $MgCl_2$ showed the early stages of necrosis with the areas nearest the blood vessels being affected most,

3) kidney tissue from fish in the $CaCl_2$ solution showed contraction and darkening of the nuclei (pyknosis). There was some shifting of the connective tissue in the Bowman's capsules and some degeneration in the tubules, especially the inner walls.

Some of the pathological changes noted, however, may have been due in part to post-mortem changes.

Tiernan (1962) also subjected rainbow trout to various saline solutions. The seven major ions of natural waters, i.e., the cations calcium, magnesium, potassium, and sodium, and the anions carbonate, sulfate, and chloride, were tested in combination, at concentration levels ranging from 3,200 to 8,100 mg/l. In solutions containing the seven major ions, fish responded by a loss of equilibrium and impairment of normal swimming movements; this did not occur in solutions of single salts, nor in solutions of two, three, or four salts together. In addition to the symptoms mentioned above, a paralysis or muscular tetany was often observed in the fish. Hemorrhaging was sometimes seen in the superficial blood vessels of the pectoral fin region and occasionally in the gill area.

Osmotic dehydration of the tissues of freshwater fish dying in saline media has been reported often. Excessive salt levels apparently produce adverse effects by creating high osmotic concentrations which may dehydrate the gills of fish (Jones, 1964). However, death may be due to any of several possible causes (Doudoroff and Katz, 1953). The toxic action of potassium chloride to fish at concentrations below isotonicity may be due to internal effects on muscular and neuronal systems (Jones, 1964).

Adelman *et al.* (1976) observed that upon introduction of NaCl, goldfish and fathead minnow* were affected even at concentrations that caused no mortality. For about the first five minutes of exposure there was increased swimming activity, particularly toward the water surface. "Within one to two hours prior to death (those fish exposed to toxic concentrations of NaCl) began increased respiratory movements at the surface accompanied by occasional bursts of frenzied swimming. Just prior to death swimming ceased and opercular movements slowed considerably." Death was presumed to be due to massive osmoregulatory failure.

Effects of Salinity on Toxicity of Pollutants

Tests have been conducted to determine what effects salinity has upon toxicity of certain compounds to fish. Experiments show that as salinity increases to the isotonic point, zinc becomes less toxic (Herbert and Wakeford, 1964); ammonia, somewhat less toxic (Herbert and Shurben, 1965); and phenol, more toxic (Brown *et al.*, 1967).

The ammonium ion is three times less toxic to yearling rainbow trout in water with 30‰ salinity than in fresh water. Above 30‰ salinity, trout become less resistant to NH_4^+.

Yearling rainbow trout and Atlantic salmon can withstand 15 and 13 times, respectively, more $ZnSO_4$ in 30-40‰ sea water than they can in fresh water. It is significant that these species are more resistant to zinc poisoning in solutions that result in a minimal inward diffusion of water since it is postulated that zinc causes deterioration of gill epithelium (Lloyd, 1960).

The sensitivity of rainbow trout to poisoning by phenol increases as salinity increases. At 15°C the 48 hour median lethal concentration (48 hr LC50) decreases from 9.3 mg phenol/l in fresh water to 5.2 mg phenol/l in 60% sea water. In the phenol test, no explanation is given for the increased toxic effects in saline waters.

3.1.6.2 Amphibia (Amphibians)

Most amphibians spend at least part of their life in a freshwater environment. Among the lower vertebrates, the amphibians are probably the group in which mechanisms of osmotic and ionic regulation have been most carefully studied and hence are best understood (e.g., Krogh *et al.*, 1938; Gordon *et al.*, 1961; Deyrup, 1964; Gordon and Tacker, 1965; Shoffeniels and Tercafs, 1966; Greenwald, 1972; Prosser, 1973). Despite the general belief that amphibians cannot survive for long periods in solutions with a salinity exceeding 1%, investigations on osmotic regulation in species that normally inhabit brackish water, e.g., *Rana cancrivora* (Gordon *et al.*, 1961; Schmidt-Nielsen and Lee, 1962) and *Bufo viridis* (Gordon, 1962), have shown prolonged tolerance to salinities as high as 2.8%. It is also known that freshwater species can tolerate hypertonic saline for several days (e.g., Adolph, 1933), and isotonic saline for many weeks (Maetz *et al.*, 1958; Crabbé, 1961).

The experiments of Ackrill *et al.* (1969) show that, as observed previously in experiments over shorter periods (Adolph, 1933) frogs immersed in saline solutions respond to maintain body fluid hypertonicity; that changes in water, salt, and urea metabolism all contribute to the maintenance of hypertonicity; and that some of these changes are similar to those enabling brackish water species, such as

Rana cancrivora, to withstand a high salt environment (Gordon *et al.*, 1961; Schmidt-Nielsen and Lee, 1962). The physiological adaptations necessitated by changes to environmental salinity must include marked changes in the passive and active transport characteristics of skin, kidney, and bladder of anurans (toads and frogs).

Ferreira and Jesus (1973) assessed the relative importance of skin, kidney, and urinary bladder in controlling the balance of water and salt in *Bufo bufo* living in environments of high salinity. The toads were kept in NaCl solutions of 20, 50, 110, 150, and 220 mM (1169, 2923, 6430, 8768, and 12,859 mg/l) and studied in their fourth week of adaptation. The survival rates of toads kept in saline concentrations up to 150 mM were identical to those of control animals, but half of the toads kept in 220 mM died within four weeks.

Strahan (1957) studied the effects of salinity on the survival of larvae of *Bufo melanostictus*. He found that the ability to survive in brackish water was a function of age. For example, individuals placed into water of 0.5% NaCl at an age of 1.5 or 2.5 days became stunted and died 5.5 days later, whereas at an age of 5.5 or 8.5 days, larvae were only slightly retarded in their otherwise normal development.

Every structure in the body of an embryo or tadpole probably suffers to a certain extent from dehydration when an individual is exposed to a medium of higher salinity. Strahan's observations indicate that two of these structures are particularly sensitive, the tail and the external gills, each of which presents a large surface area, is thin-walled, and possesses a rich blood supply.

Exposure to high salinity (0.5-1.0% NaCl) at the neurola stage inhibits the formation of a tail bud or, of formed, may cause it to regress. Such deformed tadpoles are seriously impeded in their locomotion and this interferes with their feeding. The reduction of the external gills upon exposure to saline medium is very rapid, being noticeable in 1-2 hours (Strahan, 1957). The great majority of tadpole deaths occurs at the time of resorption of the external gills.

Ruibal (1959) studied a population of frogs of the species *Rana pipiens* which inhabits San Felipe Creek, a saline desert stream. The tolerance of the eggs of *R. pipiens* to various salinities was determined and compared with that of frogs from non-brackish habitats (i.e., Vermont and "Wisconsin" frogs). The minimum lethal concentration was 5‰ while salinities ranging from 3.8‰ to 4.5‰ were semi-lethal. No difference was detected in the tolerance of eggs from non-brackish habitats as compared to San Felipe eggs. The tolerance limits of the eggs of San Felipe frogs indicate that these eggs are not adapted to tolerate higher salinities than the eggs of freshwater populations.

Adult frogs from San Felipe and "Wisconsin" were found to successfully tolerate a salinity of 6‰ for three months. A concentration of 13‰ rapidly killed the frogs.

Peabody and Brodie (1975) undertook a study to determine whether temperature, salinity, and photoperiod influence the number of trunk vertebrae which develop in the spotted salamander, *Ambystoma maculatum*. A highly significant ($p<0.01$) effect of salinity on vertebral number occurred throughout the range of treatments (1.5‰ [control] to 6‰). The correlation between salinity and vertebral number was positive. Also, the percentage of eggs that hatched was highly dependent ($p<0.01$) on temperature and salinity, ranging from 81% at 10°C (1.5‰) to 28% at 10°C (6‰).

3.1.6.3 Reptilia (Reptiles)

The osmotic concentration of reptile blood tends to be higher than that in amphibians but similar to birds and mammals (Prosser, 1973). Freshwater reptiles may actively absorb ions; when submerged, uptake of water may be considerable.

A freshwater race of the snake *Natrix sipedon* is killed in sea water because it drinks the water, but a race which lives in salt marshes, although still preferring fresh waters, tolerates salinities up to 75% sea water because it avoids drinking (Pettus, 1958).

3.1.6.4 Aves (Birds)

The presence of supraorbital nasal glands in birds has been known for many years but their functional significance was not elucidated until 1958 when Schmidt-Nielsen and his coworkers demonstrated their role as extrarenal excretory organs. These workers have shown that when osmotically active loads of NaCl are administered to birds, the nasal glands secreted an extremely hypertonic solution of Na^+, K^+, and Cl^-. Since the birds nephron has only a limited capacity to produce hypertonic urine (Holmes *et al.*, 1968), those species in which the nasal glands have not developed into extrarenal excretory organs cannot survive the prolonged ingestion of hypertonic saline drinking water (Holmes *et al.*, 1961). Hughes (1969) and Hughes and Ruch's (1969) studies suggested that tears have the potential to eliminate significant quantities of cations, particularly potassium, in salt-stressed birds.

Dr. W. Cowan (Canadian Wildlife Service, pers. comm.) feels that a change in salinity in the AOSERP study area will probably not affect bird physiology but it may be detrimental to food resources (i.e., vegetation and invertebrates). The discharge of saline waters could also affect the quality of drinking water.

Saline Waters as Bird Habitat

Anderson (1970) evaluated salt evaporation ponds as wildlife habitat for birds. Salinity ranged from 9 Sal. (per cent of saturation) as the minimal in Pond 1, to a maximum of 93 Sal. in Pond 6. Fifty-five bird species were recorded in the ponds. Among the birds making heavy use of salt ponds were shorebirds, ducks, grebes, and Bonaparte's gulls *(Larus philadelphia)*. Dabbling ducks*, coots*, and fish-eating birds exhibited a marked preference for Pond 1. Diving ducks*, grebes, phaloropes, and Bonaparte's gulls disclosed a high degree of salinity tolerance and a predilection for food items in ponds of high salinity.

Syncrude (1973b) undertook a study of waterfowl utilization in the area of Syncrude Tar Sands Lease No. 17. It was found that Saline Lake was the waterbody most intensively used by birds in the area. Apparently the abundance of duck foods in Saline Lake tended to concentrate ducks (especially scaups, *Aythya* spp.) and also provided an attractive habitat for both diving and dabbling ducks. High productivity of invertebrates in saline waters has been mentioned by Rawson and Moore (1944), McCarraher and Thomas (1968) and Held and Peterka (1972).

Waterfowl populations appear to use saline waters for staging purposes, and occasionally for feeding in the spring and fall (Dr. G.D. Adams, Canadian Wildlife Service, pers. comm.).

Effects of Salinity on Vegetation

Metcalf (1931) relates the distribution of aquatic duck food plants in North Dakota lakes to the total dissolved mineral content of the water.

Neely (1962) gives a figure showing maximum salinity tolerances of some plants. A number of duck food plants will tolerate salinities up to 3,000-4,000 mg/1 with widgeon grass *(Ruppia maritima)* tolerating up to 35,000 mg/1.

Additional data on salinity effects on vegetation is presented in tha aquatic plants section.

Saline Waters as a Drinking Source

When freshwater-maintained ducks *(Anas platyrhynchos)* were given hypertonic saline (284 ml NaCl and 6.0 ml KCl; 16,600 mg/1 NaCl and 447 mg/1 KCl) as their sole source of drinking water, a rapid increase in the total osmolality and in the plasma concentrations of Na and Cl ions was apparent (Fletcher and Holmes, 1968). After 10 hours of exposure to the saline drinking water, both plasma concentrations showed a steady decline, and by 50 hr, they were not significantly different from the levels observed before transfer to the saline water.

These observations suggested that the duck regulated its plasma electrolyte composition within a few hours after being transferred to the hypertonic saline water.

However, these results were in marked contrast to those obtained from ducks which had been transferred from fresh water to saline drinking water of a somewhat higher concentration (472 mM/1 NaCl and 10.0 mM/1 KCl; 27,588 mg/1 NaCl and 746 mg/1 KCl). In this case, the plasma levels of total osmolals and of sodium and chloride ions again rose rapidly within the first few hours after exposure but the elevated concentrations did not decline during the experimental period. Instead, they continued to increase for a period of 14 days when the birds either died or were in a seriously deteriorated physiological state. Clearly, the ducks were unable to maintain homeostasis when exposed to this concentration of hypertonic saline drinking water. This study also indicated that the prior adaptation of the birds to dilute saline did not increase their survival upon transfer to the more concentrated saline.

It is apparent from the above observations that the ducks living on saline water containing 284 mM/1 NaCl and 6.0 mM/1 KCl were maintaining a steady rate in respect of the intake and the excretion of water and electrolytes. However, upon transfer to saline drinking water, each duck began to lose weight, and after three weeks, the body weight of each individual was significantly lower than it was before transfer to the saline regimen. The drop in body weight was accompanied by a simultaneous reduction in the food and water intake.

Apparently some birds select drinking water which is most suited to their survival. Studies on the herring gull *(Larus argentatus smithsonianus)* and laughing gull *(Larus atricilla)* indicated that when these birds are simultaneously presented with distilled water and hypertonic saline, they drink more of the former (Harriman and Kare, 1966; Harriman, 1967).

Suppression of salt gland function could be detrimental to survival in habitats of high salinity. Recent studies have suggested that DDT and/or its metabolites might produce such alterations (Friend *et al.*, 1973). They determined the effects of DDE (Dichlorodiphenyldichloroethylene) on salt gland function in the mallard, *Anas platyrhynchos**. It appears from their data that sublethal levels of DDE have no effect on extra-renal elimination of salt in mallards whose salt glands have been previously stimulated by low-level (1%) salt exposure. However, DDE can apparently suppress salt gland secretion in mature mallards not previously exposed to salt.

The significance of these data lies in the fact that the salt gland is the major route of salt elimination for birds living in saline habitats. Interference with its function in juveniles or other birds not previously (or perhaps not recently) exposed to salt could result in inability to eliminate toxic levels of salt taken in while feeding (invertebrates [crustaceans] for example, have salt concentrations equal to their surroundings).

Crustaceans are an important food item of waterfowl. Food habit studies of some waterfowl in North Dakota revealed that crustaceans, by volume of the total diet of breeding males and females, comprised 77% of the shoveler diet *(Spatula clypeata*)*, 32% of the gadwall diet *(Anas strepera*)*, and 8% of the blue-winged teal *(Anas discors*)* diet (Swanson and Nelson, 1971). These investigators also found that dipteran larvae comprised 34% by volume of the invertebrate diet of breeding teal.

3.1.6.5 Mammalia (Mammals)

Many mammals drink salt water solutions up to a limit. Mammals also tend by behaviour to regulate salt intake according to blood levels (Prosser, 1973). The impact of saline water, if any, to aquatic mammals will probably be to food resources although osmotic stress through ingestion is also possible.

Aquatic mammals such as the American water shrew

*(Sorex palustris palustris)**, mink *(Mustela vison lacustris)**, muskrat *(Ondatra zibethicus spatulatus)**, otter *(Lutra canadensis preblei)**, and beaver *(Castor c. canadensis)** consume a wide variety of aquatic foods (i.e., aquatic plants, insects, fish, mollusks, frogs, etc.)(Soper, 1964). In addition, aquatic vegetation is important as summer feed to the larger land mammals such as the moose *(Alces alces andersoni)** and to the prey (e.g., waterfowl) of smaller mammals.

3.1.7 Effects of Saline Waters on Lake and Pond Biology

Keeton (1959) experimentally introduced oil field brine into two turbid farm ponds in Oklahoma to reduce their turbidity. Brine was introduced a) intermittently into the First Experimental Pond during periods of circulation, and b) in a large single application into the Second Experimental Pond when the water was thermally stratified. Specific conductance was an efficient method for determining the dispersal of the brine when introduced. During the introduction, the various ions contained in the brine (Na, Ca, K, Cl) became more concentrated in the ponds. This concentration subsequently decreased as a result of absorption by the bottom sediments. As the brine was introduced, turbidity decreased and light penetration increased.

Several species of net plankton (cladocerans and copepods) and Tendepedidae were limited in the First Experimental Pond by the increased brine concentration; however, other species increased. The production of net plankton, phytoplankton, *Physa**, *Hyalella azteca**, fish, and aquatic plants was greater in the clear First Experimental Pond than in a turbid Control Pond. The greater production was directly attributed to increased light penetration as a result of decreased turbidity.

After 8,400 gallons of brine were introduced into the Second Experimental Pond in a single, large application, the pond cleared. Brine concentrated at the deepest point in the pond immediately after introduction, dispersing uniformly throughout the pond in three months. The brine apparently was not harmful to the zooplankton population or the fish of the Second Experimental Pond. The benthos and net plankton were, however, almost certainly killed in the region of concentrated brine. As a whole, the biota of the two ponds were not seriously affected.

Judd (1969, 1970) studied the effects of deicing salt runoff on stratification within a small lake in Michigan. Salt entering First Sister Lake increased the density of the water in the lower lake strata so that during 2 or the 3 years studied, the increased density prevented complete spring overturn. The lake would probably have become meromictic if the salts had remained in the lake. The salt, however, leached into the bottom muds. Sufficient salt was lost during the summer of 1965 that by fall, density due to salt had been reduced and the lake mixed completely.

Salt water runoff also caused changes in the benthic community. In samples taken in June 1966, three dipteran larva and seven oligochaete species were found. A large amount of salts entered the lake during the following winter. No dipteran larva were found in samples taken below 3 m in February, 1967. Only four oligochaete species were found. Changes in species composition of the benthic community were confirmed by laboratory experiments.

The major physical effect of salt water on a freshwater lake is due to the differences in density between the two types of water. The more dense saline water, in most instances, will move into the deeper part of the lake basin where it may remain separate from the lower density fresh water. The development of meromixis affects the distribution of many chemical parameters within a lake, of which the most important is oxygen (Smith and Frey, 1971; German, 1972). After the oxygen has been depleted in the high density strata, the lack of circulation means that the oxygen will not be replenished, so oxygen levels will remain approximately zero throughout the year. This is reflected in the very poor benthic communities of meromictic lakes.

3.1.8 Effects of Saline Discharge on Stream Biology (in particular, Benthic Macroinvertebrates)

The effects of brine (saline) discharge on an Oklahoma stream (Salt Creek) were studied by Clemens and Finnell (1955). Concentrations of chlorides in Salt Creek ranged from 20,000 mg/l at the headwaters to 100 mg/l at the confluence of the brine-polluted stream and a larger stream. The community composition of Salt Creek was directly correlated with the concentration of the chloride ions. The 34 species collected in the stream throughout the year were arranged according to their chloride tolerance. These animals included fish, crustaceans, nematodes, insects, rotifers, and protozoans.

The average number of organisms found in chloride concentrations ranging from 13,000 to 20,000 mg/l was 4. This number increased to 6 for the 10,000 to 13,000 range, to 7 for the 8,000 to 10,000 range, to 8 for the 4,000 to 8,000 range, to 10 for 1,000 to 4,000 range, and to 13 where the chlorides were always below 1,000 mg/l.

The only organisms found in the 13,000 to 20,000 mg/l range were: *Euglena*, nematodes, chironomids, *Fundulus kansae*, and on one occasion, *Cyclops*.*

In the 10,000 to 13,000 mg/l group, three rotifers, *Notommata*, *Notholca*, and *Monostyla*, appeared. *Notommata* and *Notholca* were most abundant during the cold water period (November-April) when chloride concentrations were not above 13,183 mg/1.

Difflugia, *Notropis lutrensis*, *Ceratium*,* dragonfly nymphs, *Alona*,* and ostracods were collected in waters of 8,000 to 10,000 mg/1 chloride. Only single specimens of *Alona*, *Notropis lutrensis*, and *Ceratium* were sampled, which suggest that these organisms were by no means well adapted to such chloride concentrations.

Five additional species appeared in water containing 4,000 to 8,000 mg/1 chloride: *Pimephales promelas*,* *Gambusia affinis*, *Cambarus*, and two rotifers, *Brachionus* and *Lecane*. The latter two, however, were so few that they may lack tolerance to this chloride concentration.

At stations where the chloride concentration was as high as 4,000 mg/1, the collections included *Cephalodella*, *Keratella*, *Platyias*, *Lepadella*, *Epiphanes*, *Pedalia*, and *Daphnia*.* The presence of only one specimen of the last four organisms probably denotes their sensitivity to the brine wastes. In fact, the specimens of *Pedalia*, *Keratella*, and *Daphnia* all occurred in samples where the chloride concentration was less than 2,000 mg/1.

The toxicity of brine waters was discussed by Clemens and Jones (1954) who determined median toxicity thresholds (MTT's) on 10 species of invertebrates. Thresholds were expressed as percentage volume of the original waste since the brine contained large amounts of calcium, magnesium, potassium, and sodium chlorides as well as soluble oil products. Invertebrates employed as test animals included four crustaceans--water fleas *(Daphnia pulex)*, copepods *(Diatoms clavipes)*, sideswimmers* *(Hyallela azteca)*, and crayfish *(Cambarus* sp.)-- as well as a snail *(Physa)**, an annelid (Tubificidae), and four kinds of aquatic insects, namely: a dragonfly (Libellulidae), a damselfly (Coenagrionidae), a burrowing mayfly (*Hexagenia*) and another species of mayfly (Baetidae).

Daphnia pulex was the most susceptible, having a threshold of 1.8%. *Diaptomus clavipes, Physa,* Baetidae, and *Hyallela azteca* were next with respective thresholds of 3.1, 3.2, 3.6, and 3.8%. Tubifex worms, mayflies *(Hexagenia)*, damselflies, and dragonflies followed with respective thresholds of 4.9, 5.1, 7.2, and 7.2%. Crayfish, the most tolerant of the invertebrates, had a threshold of 8.7%. However, computations of hypothetical combinations of chlorides in the brine indicated NaCl to be present in such proportions as to be most toxic.

Determinations of the MTT's for NaCl were made for five species of invertebrates. Crayfish and dragonflies were the most tolerant, having thresholds of 17,403 and 15,943 mg/l NaCl, respectively. The common pouch snail (*Physa*),* the copepod (*Diaptomus*), and the water flea (*Daphnia*) possessed much lower thresholds--5,353, 4,329, and 2,932 mg/l NaCl, respectively.

Harrel and Dorris (1968) correlated morphometry, physico-chemical conditions, and community structure of benthic macroinvertebrates of the Otter Creek drainage basin, Oklahoma, with stream order. Oil field brine was one of the factors which upset the stream order-community structure relationship. Marked increases in NaCl and sulfates were observed at stations 9(4) and 22(5), both of which received oil field brines by seepage through the soil. Conductivities were consistently higher at station 22(5) ($\bar{X}$=2,462 micromhos/cm) than at station 9(4) ($\bar{X}$=607 micromhos/cm). Total alkalinity and dissolved oxygen were also higher at these stations.

Harrel and Dorris calculated total community species diversity, diversity per individual, and redundancy for each season and for the year. "Total community species diversity (H) reflects the manner in which individuals are distributed among species in the community. Maximum diversity (H_{max}) exists if all individuals belong to different species. Minimum diversity (H_{min}) occurs if nearly all individuals are of one species. Total diversity of natural communities usually lies between H_{max} and H_{min}. Diversity per individual ($\bar{H}$) becomes smaller as the probability of selecting a particular species becomes a certainty, and larger as the choice becomes more uncertain. Redundancy (R) is maximal when no choice exists and minimal when there is more choice" (Patten, 1962; in Harrel and Dorris, 1968).

Algae were more abundant at stations which received brines than at other stations, probably because of decreased turbidity. Keeton (1959) used oil field brines to reduce turbidity and increase light penetration in turbid ponds. *Spirogyra* sp. was dominant at station 9(4) and *Vaucheria* sp. at station 22(5). Similarly, benthic invertebrates were more numerous at these stations than at other stations. More species (62) were collected at station 9(4) than at any other station. Among the species most numerous were: *Dero digitata*, *Nais variabilis*, *Tubifex* sp., *Hyallela azteca*,* *Caenis* sp.*, *Nehalennia* sp., *Sialis* sp., *Oecetis inconspicua*, *Tropisternus lateralis*, *Hydroporus* sp., *Dubiraphia vittala*, several dipterans, *Physa anatina*, and *Spaerium transversum*.* *Polypedilum illinoense* (Diptera) reached its highest densities at stations which received oil field brines. Harrel and Dorris stated that Paine and Gaufin (1956) considered *P. illinoense* to be a positive indicator of an unpolluted habitat.

Generally, total diversity (H) was higher at stations 9(4) and 22(5) than at any other station. However, moderate oil field brines at station 9(4) seemed to improve the stream by reducing turbidity, thus permitting higher primary productivity. The increase in diversity per individual ($\bar{H}$) and decrease in redundancy (R) indicated a more random distribution of species and improved stream conditions. More concentrated brines at station 22(5) did not seem to exclude species, but did allow certain species (*Limnodrilus* sp. [Oligochaeta] and several tendipedids) to become superabundant. Diversity per individual decreased and redundancy increased at station 22(5).

In summary, influx of low concentrations of brines caused diversity per individual to increase and redundancy to decrease, indicating improved stream conditions and more random distribution of individuals into species. More concentrated brines caused diversity per individual to decrease and redundancy to increase, denoting less random distribution and greater repetition of selected species.

In a similar study, the effects of oil field brines discharged to an intermittent watercourse on the diversity of macroinvertebrates was investigated in Oklahoma (Mathis, 1965; also in Mathis and Dorris, 1968). These data were analysed according to information theory methods (Patten, 1962) and generally a more diverse community existed as the brines were diluted with either increasing tributary water additions or increasing runoff.

Mathis used specific conductance as a means of detecting brine pollution produced by water from oil wells (see Ellis, 1937). Chloride concentrations were not measured in Black Bear Creek (they were assumed to vary with specific conductance). Tarzwell (1956) and also ORSANCO (1956) indicated that the chloride ion is not a reliable index of osmotic strength when dealing with a mixed salt solution such as oil field brines. They concluded that the cations are of greater importance as a toxic agent to aquatic life and that the chloride ion is of little significance. Specific conductance below the brine influx ranged from 231 micromhos at station 125 to 16,000 micromhos at station 43.

Seventy-nine species in 74 genera and 10 orders were collected during the study. The majority of benthic organisms in Black Bear Creek belonged to three families of insects: Hydropsychidae, Tendipedidae, and Simulidae. Distinct differences in species composition were found above and below the source of brines. During the year, 55 species were collected from station 125, where brine concentrations were relatively low. Above the outfall, 47 species were collected. The smallest number of species, 31, was found at station 43 located just below the outfall.

Maximum numbers of individuals occurred in the middle reaches of Black Bear Creek where turbidity was lowest and brines more concentrated. The large numbers of bottom organisms in these reaches may have resulted from increased primary productivity associated with decreased turbidity and increased light penetration. Algal blooms were noted on several occasions, especially in less turbid reaches of the stream.

Evidence that oil field brines may be limiting certain species comes from a comparison of the faunal composition of Black Bear Creek and one of its brine free tributaries, Camp Creek. At some stations in Camp Creek, *Sphaerium transversum* reached a density of $1300/m^2$. This animal was absent from the middle reaches of Black Bear Creek where brine concentrations were high. Other organisms such as mosquito larvae, phatom midge, alderflies, and leeches were abundant in Camp Creek but were rare or absent in Black Bear Creek.

However, oil field brines in Black Bear Creek apparently are not as harmful to benthic organisms as are domestic and oil refinery effluents. Only 42 species of benthic macro-invertebrates were found in Skeleton Creek, a stream comparable to Black Bear Creek, but which differs from it in that Skeleton Creek receives domestic and oil refinery effluents near its origin (Wilhm and Dorris, 1966).

Numerical indices derived from information theory make possible quantitative comparisons of benthic community structure in different streams under different conditions. Comparative mean annual diversity indices show that oil field brines in Black Bear Creek were less restrictive to bottom organisms than effluents in Skeleton Creek. With the exception of R (redundancy), parameters for Black Bear Creek generally exceeded those in Skeleton Creek. Other parameters such as H, $\bar{H}$, H_{max}, and H_{min} indicate faunal diversity to be much greater in Black Bear Creek than in Skeleton Creek.

Olive and Dambach (1973) evaluated the quality of water in Whetstone Creek, Ohio, in relation to certain characteristics of six benthic invertebrate communities. The community characteristics included the kinds of organisms present, the diversity of the organisms, the number of taxa present, and the Shannon-Wiener index of biological diversity. Prior to and during their period of study, Whetstone Creek received moderate quantities of pollutants from two major sources: 1) organic effluents from a sewage treatment plant, and 2) brines from oil field operations.

Chlorides ranged from 8-12 mg/l at headwater stations but increased to 105-276 mg/l at stations located within the oil field region. High chloride concentrations in Whetstone Creek resulted from seepage of brines into the stream and from direct discharge into the stream channel. Although no chloride concentration greater than 276 mg/l was recorded during the study, Pettyjohn (1971) reported that, during peak drilling activity in the region, chloride levels as high as 1,350 mg/l were noted in the stream.

Olive and Dambach conclude "that because of the large diversity of pollution-sensitive and facultative organisms in brine-polluted areas of Whetstone Creek, it is doubtful that brines have damaged the benthic macroinvertebrate communities as much as have organic effluents." At station 31, for example, where organic and brine pollution are most severe, *Goniobasis livescens* (snail), *Acroneuria* sp. (stonefly), *Libellula* sp.* (dragonfly), and *Pycnopsyche gentilis* (caddisfly) were absent. But the presence of these organisms at stations farther downstream, where organic pollution was less severe but where chlorides remained high, suggests that brines were not as damaging to these populations as were the low dissolved oxygen concentrations or the sludge deposits associated with decaying organic wastes at station 31.

Harrel *et al.* (1967) studied the relation between stream order and species diversity of fishes in the Otter Creek drainage basin, Oklahoma.

They found that stations which received oil-field brines by seepage through the soil had higher conductivities, lower turbidities, and more algal growth than other stations. Species diversities at stations that received brines were similar to other stations of the same order above and below the influx with one exception. During June, species diversity decreased from 1.18 at an upstream station 21(5), to 0.73 at station 22(5), then increased to 2.57 at downstream station 23(5). Numbers of species exhibited a similar pattern.

RRCSL (1974b) conducted a study on Beaver Creek with respect to the discharge of saline water from dragline operations on the Syncrude mining lease. The diversity and abundance of benthic macroinvertebrates was the major subject of investigation. Benthic macroinvertebrates were sampled monthly at suitable riffle sites along the creek from September, 1973 to August, 1974. However, no suitable riffles could be found upstream from the discharge to serve as controls so sampling sites furthest downstream from the discharge were chosen as partial controls.

The discharge of saline water commenced in early September, 1973, at about 700 imperial gallons per minute (IGPM) containing about 5,000 mg/l NaCl and stabilized in the early summer of 1974 at about 150 IGPM. Chlorides ranged from control background levels of 3-46 mg/l to highs of 512-522 mg/l near the point of discharge. Generally, the highest chloride levels occurred during the winter months when stream flow was low.

Changes were observed in the abundance of Ephemeroptera (mayflies) and Plecoptera (stoneflies), the dominance of

Chironomidae (midges) and the diversity of benthos. Mayfly and stonefly nymphs largely disappeared from collections (numbers per sample decreasing from 10-46 in September, 1973, to 0-7 in November-March, to 0-1 during May to August, 1974). Chironomids thus rose in dominance from about 50% (percentage of total individuals) in September, 1973, to around 80% during November, 1973, to August, 1974. Mean diversity of benthos decreased from 1.45-2.56 in September, 1973, to 0.34-1.80 during November-March, and 0.55-1.52 during May-August, 1974. There was, however, no evidence of adverse effects of saline discharges on aquatic plants or fish.

Apparently, some combination of chloride discharge, increased silt loads, oil/tar input, and insecticides is the cause of these observed changes in benthos. However, the lack of good baseline data on Beaver Creek benthos precludes any meaningful interpretation on the impact of the above pollutants on such changes.

"In general, discharges which result in conditions of high salinity in freshwater streams cause the normal community to be replaced by one characteristic of brackish waters. It is probable that such a community can only persist in fairly constant conditions of salinity. Alternating conditions of high and low salinity could result in neither the normal nor the replacement community being present" (Hawkes, 1962).

4. SUMMARY

The effects of saline waters in general on major groups of organisms is discussed and the major findings were as follows:

4.1 TOXICITY OF SALINE WATERS

1. The toxicity of physiologically unbalanced solutions of various chlorides (salts), including sodium chloride (NaCl), is apparently attributable to the specific toxicity of the cations present and not to any toxicity of the chloride ions (anions).

2. Sodium chloride is antagonistic to the toxicity of calcium and potassium chlorides. Calcium chloride ($CaCl_2$) is however the most effective chloride in antagonistic ability.

4.2 AQUATIC PLANTS

1. Chlorides do not appear to limit algal production directly in nature but, in the form of NaCl, play a major role in the determination of the kinds of algae which can grow in the water.

2. Diatoms are regarded as good indicators of salinity.

3. Morphology of aquatic plants may be a factor in determining salt tolerances.

4.3 INVERTEBRATES

1. Many aquatic invertebrates, particularly among the Oligochaeta, Gastropoda, Crustacea, Odonata, Hemiptera, Diptera, and Trichoptera can tolerate quite high salinities.

2. The few studies on the effects of saline discharges on benthic invertebrates indicate that changes in species composition may result and that certain (less tolerant) species are limited.

4.4 VERTEBRATES

4.4.1 Fish

1. A great deal of toxicity threshold work has been done but primarily on warm water fish and for short periods with extreme concentrations.

2. Many freshwater fishes have the physiological ability to tolerate saline water for short periods of time.

3. However, many factors affect salinity tolerances, e.g., season, life history stage, size and age, acclimation, temperature, etc.

4. Of the fish species in the AOSERP region, the yellow walleye *(Stizostedion vitreum vitreum)* is probably the most

sensitive to saline waters.

5. Potassium chloride (KCl) appears to be the most toxic of the chlorides to fish with sodium chloride (NaCl) being the less toxic.

6. NaCl concentrations above 3,000 mg/l are considered deleterious to fish food organisms.

4.4.2 Amphibians and Reptiles

1. Although it is known that amphibians have well developed mechanisms which enable them to live in very dilute environments, apparently some species can also survive in media of high salinity.

4.4.3 Birds

1. Saline waters are probably not directly toxic to birds but may be detrimental to food resources (i.e., aquatic plants and invertebrates).

4.4.4 Mammals

1. The effects of saline water, if any, to aquatic mammals will probably be to food resources although osmotic stress through ingestion is also possible.

APPENDIX I. Chemical analyses of seven ARC wells in the AOSERP study area.

ALBERTA RESEARCH COUNCIL
WATER ANALYSIS REPORT

Mer 4 Tp 94 Rge 6 Sec 4 Lsd 16

			D	M	Yr.
Lab No.	75 756	Date sampled	12	8	75
Index No.	DH 57	Date submitted	18	8	75
well depth (ft)	43.0	Date analysed (major)	20	8	75
water Level (ft)	-	Date analysed (minor)	-		
Top open interval (ft)	31.0	Sampled by	ARC		
Bottom open interval (ft)	36.0	Sample Source	BAILED		
Altitude (ft)	2020.0	TDS (mg/1)	874.0		
Bedrock elevation (ft)	-	Hardness (as $CaCO_3$)	598.8		
Owners name	ARC 9-43	Alkalinity (as $CaCO_3$)	581.6		
Field Cond (micromhos/cm)	-	Cond. (micromhos/cm@25C)	2700.0		
Field pH	11.0	Lab pH	12.0		

MAJOR CONSTITUENTS

	mg/1	meq/1	% of total anion or cation
Calcium (Ca)	240.0	11.98	84.1
Magnesium (Mg)	0.0	0.00	0.0
Sodium (Na)	36.2	1.57	11.1
Potassium (K)	26.7	0.68	4.8
Carbonate (CO_3)	67.0	2.23	12.8
Bicarbonate (HCO_3)	0.0	0.00	0.0
Sulphate (SO_4)	118.0	2.46	14.1
Chloride (Cl)	118.0	3.33	19.1
Nitrate(NO_3)	1.0	0.02	0.1
Hydroxide (OH)	160.0	9.41	53.9
Silica (SiO_2)	4.3	Total anions (epm)	17,440
Calcium(Acid)	265.0	Total cations (epm)	14,234
Magnesium (Acid)	0.4	Ion balance error (%)	10
		TDS balance error (%)	-11

MINOR CONSTITUENTS

Iron 4.20 mg/1

OTHER MEASUREMENTS

Field Temp (C)	9.00	Flouride (F)	0.30 mg/1
Field Hydroxide (OH)	38.10 mg/1)		
Field Carbonate (CO_3)	4.90 mg/1) wrong		
Field Bicarbonate (HCO_3)	0.00 mg/1)		

ALBERTA RESEARCH COUNCIL
WATER ANALYSIS REPORT

Mer 4 Tp 94 Rge 6 Sec 9 Lsd 1

			D M Yr.
Lab No.	75 757	Date sampled	12 8 75
Index No.	DH 58	Date submitted	18 8 75
well depth (ft)	161.0	Date analysed (major)	20 8 75
water Level (ft)	-	Date analysed (minor)	-
Top open interval (ft)	146.0	Sampled by	ARC
Bottom open interval (ft)	156.0	Sample Source	BAILED
Altitude (ft)	2018.0	TDS (mg/1)	1942.0
Bedrock elevation (ft)	-	Hardness (as $CaCO_3$)	384.2
Owners name	ARC 9-161	Alkalinity (as $CaCO_3$)	1432.1
Field Cond (micromhos/cm)	-	Cond. (micromhos/cm@25C)	6000.0
Field pH	11.4	Lab pH	12.3

	MAJOR CONSTITUENTS mg/1	meq/1	% of total anion or cation
Calcium (Ca)	154.0	7.68	24.9
Magnesium (Mg)	0.0	0.00	00.0
Sodium (Na)	218.0	9.48	30.8
Potassium (K)	533.0	13.64	44.3
Carbonate (CO_3)	48.0	1.60	4.2
Bicarbonate (HCO_3)	0.0	0.00	0.0
Sulphate (SO_4)	6.6	0.14	0.4
Chloride (Cl)	338.0	9.53	24.9
Nitrate(NO_3)	1.0	0.02	0.0
Hydroxide (OH)	460.0	27.04	70.6
Silica (SiO_2)	2.6	Total anions (epm)	38,328
Calcium(Acid)	198.0	Total cations (epm)	30,802
Magnesium (Acid)	35.0	Ion balance error (%)	10
		TDS balance error (%)	-9

MINOR CONSTITUENTS

Iron 13.00 mg/1

OTHER MEASUREMENTS

Field Temp (C)	4.00	Flouride (F)	0.20 mg/1
Field Hydroxide (OH)	93.80 mg/1)		
Field Carbonate (CO_3)	4.94 mg/1) wrong		
Field Bicarbonate (HCO_3)	0.00 mg/1)		

ALBERTA RESEARCH COUNCIL
WATER ANALYSIS REPORT

Mer 4 Tp 94 Rge. 7 Sec 30 Lsd 5

			D	M	Yr.
Lab No.	751185	Date sampled	10	9	75
Index No.	DH 201	Date submitted	12	9	75
well depth (ft)	34.0	Date analysed (major)	3	10	75
water Level (ft)	-	Date analysed (minor)	-		
Top open interval (ft)	24.0	Sampled by	-		
Bottom open interval (ft)	29.0	Sample Source	Well		
Altitude (ft)	1413.0	TDS (mg/l)	474.0		
Bedrock elevation (ft)	-	Hardness (as $CaCO_3$)	115.8		
Owners name	ARC 8-34	Alkalinity (as $CaCO_3$)	355.2		
Field Con.d (micromhos/cm)	-	Cond. (micromhos/cm@25C)	820.0		
Field pH	8.7	Lab pH	7.1		

MAJOR CONSTITUENTS

	mg/l	meq/l	% of total anion or cation
Calcium (Ca)	23.0	1.15	13.9
Magnesium (Mg)	14.2	1.17	14.1
Sodium (Na)	133.0	5.79	70.0
Potassium (K)	6.3	0.16	2.0
Carbonate (CO_3)	0.0	0.00	0.0
Bicarbonate (HCO_3)	444.0	7.28	92.0
Sulphate (SO_4)	0.0	0.00	0.0
Chloride (Cl)	22.0	0.62	7.8
Nitrate (NO_3)	1.0	0.02	0.2
Hydroxide (OH)	0.0	0.00	0.0
Silica (SiO_2)	0.6	Total anions (epm)	7,914
Calcium(Acid)	29.0	Total cations (epm)	8,262
Magnesium (Acid)	14.8	Ion balance error (%)	2
Calculated TDS	418.4	TDS balance error (%)	-11

MINOR CONSTITUENTS

Iron 45.80 mg/l

OTHER MEASUREMENTS

Field Temp (C)	6.50	Flouride (F)	0.40 mg/l
Field Hydroxide (OH)	—		
Field Carbonate (CO_3)	24.00 mg/l		
Field Bicarbonate (HCO_3)	415.00 mg/l		

ALBERTA RESEARCH COUNCIL
WATER ANALYSIS REPORT

Mer 4 Tp 95 Rge. 8 Sec 20 Lsd 13

			D	M	Yr.
Lab No.	751205	Date sampled	10	9	75
Index No.	DH 221	Date submitted	12	9	75
well depth (ft)	32.0	Date analysed (major)	7	10	75
water Level (ft)	-	Date analysed (minor)	-		
Top open interval (ft)	22.0	Sampled by	-		
Bottom open interval (ft)	27.0	Sample Source	Well		
Altitude (ft)	1072.0	TDS (mg/1)	348.0		
Bedrock elevation (ft)	-	Hardness (as $CaCO_3$)	60.5		
Owners name	ARC 7-32	Alkalinity (as $CaCO_3$)	257.6		
Field Cond (micromhos/cm)	-	Cond. (micromhos/cm@25C)	610.0		
Field pH	9.5	Lab pH	8.3		

	MAJOR CONSTITUENTS mg/1	meq/1	% of total anion or cation
Calcium (Ca)	6.3	0.31	5.1
Magnesium (Mg)	10.9	0.90	14.5
Sodium (Na)	108.0	4.70	75.8
Potassium (K)	11.3	0.29	4.7
Carbonate (CO_3)	0.0	0.00	0.0
Bicarbonate (HCO_3)	322.0	5.28	84.7
Sulphate (SO_4)	26.3	0.55	8.8
Chloride (Cl)	14.0	0.39	6.3
Nitrate (NO_3)	0.5	0.01	0.1
Hydroxide (OH)	0.0	0.00	0.0
Silica (SiO_2)	0.4	Total anions (epm)	6,228
Calcium(Acid)	15.1	Total cations (epm)	6,198
Magnesium (Acid)	10.0	Ion balance error (%)	0
Calculated TDS	336.0	TDS balance error (%)	-3

MINOR CONSTITUENTS

Iron 12.00 mg/1

OTHER MEASUREMENTS

Field Temp (C)	7.50	Flouride (F)	0.80 mg/1
Field Hydroxide (OH)	--		
Field Carbonate (CO_3)	72.00 mg/1		
Field Bicarbonate (HCO_3)	220.00 mg/1		

ALBERTA RESEARCH COUNCIL
WATER ANALYSIS REPORT

Mer 4 Tp 95 Rge. 9 Sec 18 Lsd 2

			D M Yr.
Lab No.	751210	Date sampled	11 9 75
Index No.	DH 266	Date submitted	11 9 75
well depth (ft)	21.0	Date analysed (major)	9 10 75
water Level (ft)	-	Date analysed (minor)	-
Top open interval (ft)	14.0	Sampled by	-
Bottom open interval (ft)	18.0	Sample Source	Well
Altitude (ft)	986.0	TDS (mg/1)	196.0
Bedrock elevation (ft)	-	Hardness (as $CaCO_3$)	98.7
Owners name	ARC 6-21	Alkalinity (as $CaCO_3$)	109.6
Field Cond (micromhos/cm)	-	Cond. (micromhos/cm@25C)	280.0
Field pH	6.4	Lab pH	7.3

	MAJOR CONSTITUENTS mg/1	meq/1	% of total anion or cation
Calcium (Ca)	31.0	1.55	55.7
Magnesium (Mg)	5.2	0.43	15.4
Sodium (Na)	17.5	0.76	27.4
Potassium (K)	1.7	0.04	1.6
Carbonate (CO_3)	0.0	0.00	0.0
Bicarbonate (HCO_3)	137.8	2.25	84.0
Sulphate (SO_4)	3.3	0.07	2.6
Chloride (Cl)	12.0	0.34	12.7
Nitrate (NO_3)	1.2	0.02	0.7
Hydroxide (OH)	0.0	0.00	0.0
Silica (SiO_2)	2.0	Total anions (epm)	2,672
Calcium(Acid)	31.0	Total cations (epm)	2,779
Magnesium (Acid)	6.2	Ion balance error (%)	1
Calculated TDS	141.3	TDS balance error (%)	-27

MINOR CONSTITUENTS

Iron 18.00 mg/1

OTHER MEASUREMENTS

Field Temp (C)	11.00	Flouride (F)	0.20 mg/1
Field Hydroxide (OH)	--		
Field Carbonate (CO_3)	0.00 mg/1		
Field Bicarbonate (HCO_3)	171.00 mg/1		

ALBERTA RESEARCH COUNCIL
WATER ANALYSIS REPORT

Mer 4 Tp 90 Rge. 11 Sec 15 Lsd 8

			D M YR.
Lab No.	751196	Date sampled	10 9 75
Index No.	DH 212	Date submitted	10 9 75
well depth (ft)	32.0	Date analysed (major)	7 10 75
water Level (ft)	-	Date analysed (minor)	-
Top open interval (ft)	22.0	Sampled by	-
Bottom open interval (ft)	27.0	Sample Source	Well
Altitude (ft)	850.0	TDS (mg/l)	2220.0
Bedrock elevation (ft)	-	Hardness (as $CaCO_3$)	33.2
Owners name	ARC 1-32	Alkalinity (as $CaCO_3$)	1354.4
Field Cond (micromhos/cm)	-	Cond. (micromhos/cm@25C)	4000.0
Field pH	9.0	Lab pH	8.3

	MAJOR CONSTITUENTS mg/l	meq/l	% of total anion or cation
Calcium (Ca)	2.1	0.10	0.3
Magnesium (Mg)	6.8	0.56	1.5
Sodium (Na)	838.0	36.45	97.8
Potassium (K)	6.7	0.17	0.5
Carbonate (CO_3)	0.0	0.00	0.0
Bicarbonate (HCO_3)	1693.0	27.75	72.3
Sulphate (SO_4)	260.0	5.41	14.1
Chloride (Cl)	184.0	5.19	13.5
Nitrate(NO_3)	1.8	0.03	0.1
Hydroxide (OH)	0.0	0.00	0.0
Silica (SiO_2)	3.6	Total anions (epm)	38,381
Calcium(Acid)	3.4	Total cations (epm)	37,286
Magnesium (Acid)	6.6	Ion balance error (%)	1
Calculated TDS	2135.4	TDS balance error (%)	-3

MINOR CONSTITUENTS

Iron 8.00 mg/l

OTHER MEASUREMENTS

Field Temp (C)	8.00	Flouride (F)	0.90 mg/l
Field Hydroxide (OH)	--		
Field Carbonate (CO_3)	144.00 mg/l		
Field Bicarbonate (HCO_3)	1440.00 mg/l		

ALBERTA RESEARCH COUNCIL
WATER ANALYSIS REPORT

Mer 4 Tp 97 Rge. 12 Sec 11 Lsd 1

			D M Yr.
Lab No.	751194	Date sampled	9 9 75
Index No.	DH 210	Date submitted	10 10 75
well depth (ft)	46.0	Date analysed (major)	7 10 75
water Level (ft)	-	Date analysed (minor)	-
Top open interval (ft)	36.0	Sampled by	-
Bottom open interval (ft)	41.0	Sample Source	Well
Altitude (ft)	1060.0	TDS (mg/1)	3182.0
Bedrock elevation (ft)	-	Hardness (as $CaCO_3$)	313.9
Owners name ARC	2-46	Alkalinity (as $CaCO_3$)	1015.2
Field Cond (micromhos/cm)	-	Cond. (micromhos/cm@25C)	5750.0
Field pH	7.4	Lab pH	8.0

MAJOR CONSTITUENTS

	mg/1	meq/1	% of total anion or cation
Calcium (Ca)	50.0	2.50	5.0
Magnesium (Mg)	46.0	3.78	7.6
Sodium (Na)	1000.0	43.50	87.0
Potassium (K)	8.8	0.23	0.5
Carbonate (CO_3)	0.0	0.00	0.0
Bicarbonate (HCO_3)	1269.0	20.80	40.5
Sulphate (SO_4)	1085.0	22.59	44.0
Chloride (Cl)	284.0	8.01	15.6
Nitrate(NO_3)	0.0	0.00	0.0
Hydroxide (OH)	0.0	0.00	0.0
Silica (SiO_2)	5.8	Total anions (epm)	51,399
Calcium(Acid)	700.0	Total cations (epm)	50,000
Magnesium (Acid)	251.0	Ion balance error (%)	1
Calculated	3103.6	TDS balance error (%)	-2

MINOR CONSTITUENTS

Iron 7100.00 mg/1

OTHER MEASUREMENTS

Field Temp (C)	4.00	Flouride (F)	0.80 mg/1
Field Hydroxide (OH)	--		
Field Carbonate (CO_3)	60.00 mg/1		
Field Bicarbonate (HCO_3)	1220.00 mg/1		

ALBERTA RESEARCH COUNCIL
WATER ANALYSIS REPORT

Mer 4 Tp 97 R.ge. 13 Sec 26 Lsd 9

			D M Yr.
Lab No.	751209	Date sampled	9 9 75
Index No.	DH 225	Date submitted	10 9 75
well depth (ft)	44.0	Date analysed (major)	9 10 75
water Level (ft)	-	Date analysed (minor)	-
Top open interval (ft)	34.0	Sampled by	-
Bottom open interval (ft)	39.0	Sample Source	Well
Altitude (ft)	2014.0	TDS (mg/1)	2596.0
Bedrock elevation (ft)	-	Hardness (as $CaCO_3$)	384.0
Owners name	ARC 3-44	Alkalinity (as $CaCO_3$)	339.2
Field Cond (micromhos/cm)	-	Cond. (micromhos/cm@25C)	4200.0
Field pH	8.2	Lab pH	7.7

	MAJOR CONSTITUENTS mg/1	meq/1	% of total anion or cation
Calcium (Ca)	117.0	5.84	14.4
Magnesium (Mg)	22.4	1.84	4.6
Sodium (Na)	750.0	32.62	80.6
Potassium (K)	6.7	0.17	0.4
Carbonate (CO_3)	0.0	0.00	0.0
Bicarbonate (HCO_3)	424.0	6.95	18.0
Sulphate (SO_4)	1496.0	31.15	80.9
Chloride (Cl)	14.0	0.39	1.0
Nitrate(NO_3)	1.8	0.03	0.1
Hydroxide (OH)	0.0	0.00	0.0
Silica (SiO_2)	3.2	Total anions (epm)	38,521
Calcium(Acid)	148.0	Total cations (epm)	40,475
Magnesium (Acid)	50.0	Ion balance error (%)	2
Calculated TDS	2619.6	TDS balance error (%)	0

MINOR CONSTITUENTS

Iron 100.00 mg/1

OTHER MEASUREMENTS

Field Temp (C)	2.50	Flouride (F)	1.40 mg/1
Field Hydroxide (OH)	--		
Field Carbonate (CO_3)	0.00 mg/1		
Field Bicarbonate (HCO_3)	451.00 mg/1		

Appendix II

Syncrude Canada Ltd. letter of permission to discharge saline water.

Alberta
ENVIRONMENT

LETTER OF PERMISSION

PERMISSION NO. 76-LP-003

FILE NO. 75-SA-1081

WHEREAS

Syncrude Canada Ltd.
c/o Mr. V. P. Kaminsky
9915 - 103 Street
Edmonton, Alberta
T5K 2G8

has, pursuant to section 11 of the Clean Water (General) Regulations applied to the Director of Standards and Approvals for permission to

discharge saline water from the mine dewatering program to the Beaver Creek reservoir

NOW THEREFORE

this permission is issued subject to the terms and conditions and requirements attached hereto.

Edmonton February 26 1976

Director of Standards and Approvals
for

PERMISSION NO.
76-LP-003

TERMS, CONDITIONS AND REQUIREMENTS ATTACHED TO LETTER OF PERMISSION

1. The saline water discharge from the mine dewatering program to Beaver Creek shall be located as shown by Drawing No. 1-E-A-130, submitted by the company on November 25, 1975.

2. The discharge of saline water to Beaver Creek reservoir shall be controlled such that the chloride concentration in Poplar Creek in the vicinity of Poplar Creek Bridge does not exceed the ambient concentration by 400 milligrams per litre.

3. The saline water discharged to the Beaver Creek Reservoir shall be:

   a) monitored for volume flow rate from all wells being pumped in terms of Canadian gallons per day;

   b) analyzed for water contaminants according to the following schedule:

      i) daily, by means of representative grab samples: chloride ion concentration, pH, total suspended solids, chemical oxygen demand and phenolics; and

      ii) weekly, by means of 24-hour composite samples: anion concentration (chloride, sulphate, fluoride, carbonate and bicarbonate), oil and grease, pH, total suspended solids, total dissolved solids, chemical oxygen demand and phenolics.

4. A 24-hour composite sample of water collected from Poplar Creek in the vicinity of Poplar Creek Bridge shall be analyzed once per month for the following parameters: oil and grease, chloride, chemical oxygen demand, phenolics and pH.

5. The results obtained in Clause 3 and the number of wells being pumped shall be submitted to the Director of Pollution Control in a monthly report which shall be submitted within 30 days of the end of the month for which the observations were made.

6. The following tests shall be conducted once every two months and submitted along with every second monthly report specified in Clause 5:

   a) TL_m (median tolerance limit); and

   b) metals analysis (on one 24-hour composite sample). This analysis shall include but not be limited to the following metals: vanadium, iron, copper, nickel, arsenic, titanium, zirconium, potassium, sodium, mercury, lead, zinc and silver.

PERMISSION NO.
76-LP-003

TERMS, CONDITIONS AND REQUIREMENTS ATTACHED TO LETTER OF PERMISSION

7. This letter of permission is granted on the basis of current knowledge, technology and circumstances and, if deemed necessary by the Director of Standards and Approvals, the terms, conditions and requirements may be revised, amended or revoked at any time. In any event, this Letter of Permission shall expire after a period of one year commencing the first day of the month of issue of this Letter of Permission.

Date: February 26, 1976

for J. DEFIR, DIRECTOR

Review Engineer: J. Nagendran
Section Review:

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