ALBERTA WATER QUALITY GUIDELINE FOR THE PROTECTION OF FRESHWATER AQUATIC LIFE

DISSOLVED OXYGEN

Alberta
ENVIRONMENTAL PROTECTION

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The water quality guideline derived in this document protects freshwater aquatic life against the adverse effects of low concentrations of dissolved oxygen. This guideline replaces the 1993 Alberta Ambient Surface Water Quality Interim Guideline of 5 mg/L.

Because the Alberta guideline in this document is based on the dissolved oxygen guidelines of other jurisdictions (Canadian Water Quality Guideline, United States Environmental Protection Agency Criteria), the dissolved oxygen guideline is considered final. This guideline has already been reviewed in a public forum, the Northern River Basins Study. Any public comments will therefore be incorporated when new information becomes available that indicates that the Alberta guideline requires review.

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PREFACE

Alberta Environmental Protection prepared this *Alberta Water Quality Guideline for the Protection of Freshwater Aquatic Life, Dissolved Oxygen*. Dissolved oxygen (DO) is necessary to support all forms of life in water. DO levels in water vary as a result of natural processes and human activity. Problems occur when there is too little DO for aquatic life forms to survive and thrive. A DO guideline can be used to assess conditions in a water body, to identify when human activity has the potential to cause problems for the aquatic ecosystem, and to prevent these potential problems.

DO is formed in water primarily through photosynthesis by plants; it can also be transferred to water from the air, especially through wave action or other turbulence. DO is removed from water mainly through chemical and biological reduction. Reduction occurs when organic matter, from natural sources or from industrial or municipal wastes, breaks down and consumes DO. Reduction also occurs when animals in water respire (breathe) by taking in DO and releasing carbon dioxide.

This Guideline identifies the dissolved oxygen levels that are necessary to protect aquatic life in Alberta. Alberta Environmental Protection (AEP), industries and municipalities will use the guideline to assess the potential for impacts of oxygen consuming discharges from industrial and municipal wastewater treatment plants. The public can use the guideline as a reference when assessing the environmental health of their water.

After a brief introduction (Section 1), Section 2 of the document provides background information on the physical and chemical characteristics of DO and analytical methods. Sections 3 through 5 describe the acute (severe effects, such as mortality, caused by exposure to very low levels of DO over short periods) and chronic effects (less severe effects, such as reduced rates of growth, caused by exposure to long term low levels of DO) of DO levels on fish, amphibians and invertebrates. The importance of natural daily (diurnal) fluctations and seasonal variations in DO levels in water is considered in Sections 6 and 7. Section 8 presents the process used to determine the Guideline levels and provides advice on how to apply the numbers in Alberta. Other jurisdictions have adopted their own DO guidelines, Section 9 describes these and explains how and why they differ from the Alberta Guideline. Section 10 recommends additional studies that will assist in the refinement of our understanding of the effects of fluctuations in DO on aquatic biota and Section 11 presents a list of references cited in this guideline document.

EXECUTIVE SUMMARY

Oxygen requirements of freshwater aquatic life have been reviewed numerous times (Doudoroff and Shumway 1970; Davis 1975; Hughes 1981; Alabaster and Lloyd 1982; Chapman 1986; Barton and Taylor 1994; Truelson 1997). Rather than providing another broad literature review, this document uses these reviews with more recent literature to rationalize the dissolved oxygen water quality guideline estalished for the protection of freshwater aquatic life in Alberta. Reference to other documents is provided in case the reader requires further information on physical and chemical characteristics or on analytical methods.

The dissolved oxygen guidelines for surface water in Alberta are:

		Guideline (mg/L)
Acute	1-day minimum	5
Chronic	7-day mean	6.5[1,2]

1 The chronic guideline should be increased to 8.3 from mid May to the end of June to protect emergence of mayfly species into adults.

2 The chronic guideline should be increased to 9.5 mg/L for those areas and times where embryonic and larval stages (from spawning to 30 days after hatching) develop within gravel beds (some salmonids). The chronic guideline is increased by 3 mg/L to account for the depletion of dissolved oxygen within the gravel.

For those situations where natural conditions do not meet the guideline for dissolved oxygen, the following guidance should be applied:

> *Where natural conditions alone create dissolved oxygen concentrations less than 110 percent of the applicable criteria means or minima or both, the minimum acceptable concentration is 90 percent of the natural concentrations.*

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1. INTRODUCTION

Toxicological methods for measuring the effects of chemicals on aquatic life have been standardized for many years. These methods involve the addition of a chemical in different amounts to determine the concentrations at which the chemical has an effect or no effect.

In contrast, dissolved oxygen is an integral component of water. It is the lack of oxygen (rather than an excess of oxygen) that causes adverse effects. Nitrogen stripping or vacuum degassing is therefore required to lower dissolved oxygen levels in water for the toxicity tests. The exposure chamber (whether it is open to the atmosphere or sealed) influences the maintenance of dissolved oxygen levels. Removal of dissolved oxygen rather than the simple addition of a toxicant may have influenced the number of different dissolved oxygen concentrations tested. In several instances, the effects of only two different dissolved oxygen concentrations were tested.

Further complications arise when effects of low dissolved oxygen concentrations are determined on fish eggs and invertebrates. The delivery of dissolved oxygen to fish eggs and invertebrates depends on flow rates. For instance, some invertebrates are found on rocks in extremely swift currents. These areas experience turbulent mixing and reaeration and would least likely suffer significant dissolved oxygen depletion. Laboratory tests of these species at much slower flow rates may indicate a greater need for high concentrations of dissolved oxygen in the laboratory than what they would require in their natural environment.

The literature often does not provide all relevant information on test conditions. Toxicity endpoints for dissolved oxygen tests can be expressed in many different units: e.g., mg/L, Torr, mm Hg, kPa, percent saturation. These units can be converted into a common unit provided all the necessary information on temperature and pressure is provided. This however is not the case for some studies. Throughout the text different units of dissolved oxygen concentrations are used, consistent with the original publication. In the tables however, units other than mg/L have been converted to mg/L. In these cases, the toxicity tests were assumed to have taken place at standard air pressure at sea level (760 mm Hg). Dissolved oxygen saturation decreases with altitude and decreased air pressure: i.e., saturation is achieved at lower DO concentrations (see Section 2). If the test was carried out at non-standard conditions, assuming standard conditions would result in a higher DO concentration (mg/L) and thus a more protective endpoint.

The type of toxicity endpoints also varies. For acute exposure, the endpoints include actual mortality, loss of equilibrium, lack of opercular movement. The exposure time for these short-term tests also varies widely (Chapman 1986). For chronic exposure, endpoints include the traditional growth and reproduction parameters. Some studies however measure other parameters such as oxygen uptake/consumption and swimming capacity. The variability in toxicity endpoints and exposure times challenges the derivation of a guidelines for dissolved oxygen.

Due to these different challenges outlined above, guidelines for dissolved oxygen cannot follow the procedures outlined in the Protocol to Develop Alberta Water Quality Guidelines for Protection of Freshwater Aquatic Life (AEP 1996). Instead, the latest USEPA guideline for dissolved oxygen (Chapman 1986) was used as a starting point for the Alberta guideline. These guidelines are the basis for the Canadian water quality guideline for dissolved oxygen (CCREM 1987) and are considered the most advanced general criteria (Barton and Taylor 1994, 1996). The recent literature was reviewed to investigate whether these dissolved oxygen guidelines should be adjusted.

The literature regarding dissolved oxygen effects on aquatic biota is immense. Effects on biota include many different aspects ranging from blood characteristics, respiration, reproduction to survival. The literature review presented in this document focusses on those endpoints most relevant to setting dissolved oxygen guidelines. These endpoints include: survival/mortality over relatively short periods for acute guidelines and endpoints such as growth and reproduction over longer time periods for chronic guidelines. Other effects (such as blood characteristics) will be discussed to provide a general overview, but this description will be brief and may not be extensive. References are provided in case the reader requires further information.

Many excellent reviews of the literature regarding effects of low dissolved oxygen levels on aquatic biota have been prepared (e.g., Doudoroff and Shumway 1970; Davis 1975; Chapman 1986; Barton and Taylor 1994; Truelson 1997). The review of Doudoroff and Shumway (1970) was used as a starting point for this document. Doudoroff and Shumway (1970) found little agreement of the reported findings and approached this problem by a careful reevaluation of evidence. Their findings are presented here and supplemented with more recent literature. The more recent findings are compiled to evaluate whether adjustments to the 1986 USEPA guidelines are justified and to illustrate the level of protection provided by the new dissolved guideline for Alberta.

2. BACKGROUND INFORMATION ON DISSOLVED OXYGEN

Truelson (1997) provided a very extensive review of the physical and chemical characteristics of oxygen, and of the analytical methods to determine oxygen concentrations in water. Rather than repeating this information, the most relevant information on chemical and physical characteristics is summarized below. For more information, the British Columbia criteria document should be consulted (Truelson 1997).

Physical and Chemical Characteristics

Oxygen is the most abundant element in soils and plants (weight content is 49 % and 70 %, respectively: Bohn et al. 1979). In addition, oxygen is the most abundant element in water by weight (Truelson 1997). Divalent oxygen combines with two single valent hydrogen atoms to form the extremely stable water molecule. Under normal pH for surface water (4 to 10), water is stable in the redox intensity range of -10 to +17: beyond this range, water is reduced to H_2 or oxidized to O_2, respectively (Stumm and Morgan 1981).

The double-bonded, two-atom molecule in water is the form of interest here, described as dissolved oxygen in the remainder of this document. Dissolved oxygen is the most fundamental parameter in water: it is essential to the metabolism of all aerobic, aquatic organisms (Wetzel 1975). Low levels of dissolved oxygen result in unbalanced ecosystems, fish mortality, odours and other aesthetic nuisances (Thomann and Mueller 1987).

Fate and Behaviour

The main reactions that form or remove dissolved oxygen from water are oxidation and reduction, respectively. Photosynthesis is the only natural reaction that oxidizes water to oxygen:

$$6CO_2 + 6H_2O \rightarrow C_6H_{12}O_6 + 6O_2$$

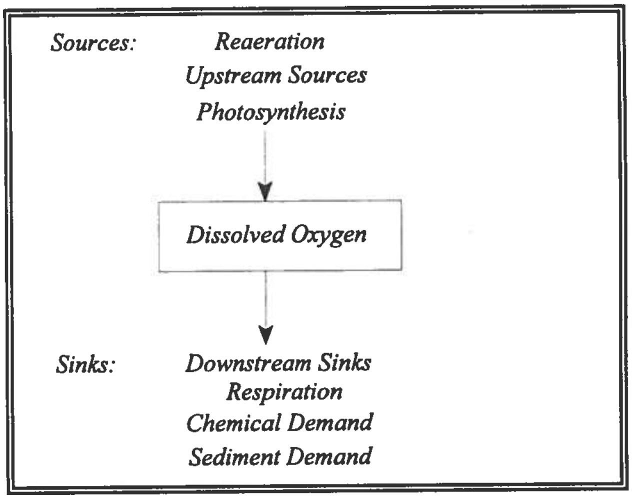
The reverse reaction reduces oxygen in water:

$$CH_2O + O_2 \rightarrow CO_2 + H_2O$$

The organic compound (CH_2O) can either be an anthropogenic organic compound (reduced carbon and nitrogen compounds in municipal and industrial wastes discharged into the water) resulting in chemical oxygen demand or a biological compound resulting in respiration by aquatic biota. Apart from the oxidation and reduction reactions, external sources and sinks may affect dissolved oxygen levels in water.

The following figure illustrates the major sources and sinks of dissolved oxygen in water. In addition to the internal oxygen source of photosynthesis, external sources such as reaeration and upstream sources can increase the dissolved oxygen content in water. Reaeration involves the transfer of oxygen from air to water. Transfer into water is increased with an increase in turbulence (both in water and air). However, transfer of oxygen into water is severely reduced in winter when ice-cover is present. Upstream sources such as tributaries, groundwater and possibly effluents can also add oxygen to water.

Respiration and chemical demand as well as downstream sinks and sediment oxygen demand can remove oxygen from water. Downstream sinks may include outlet streams and groundwater. Sediment oxygen demand may include both respiration by benthic biota (including the biolfilm) and chemical demand. Chemical demand may include the oxidation of iron, manganese, sulphur, carbon, and nitrogen compounds. Sediment oxygen demand can either be caused by anthropogenic sources (e.g., sewage or industrial effluents) or natural sources (e.g., runoff, decaying aquatic plants).



The dissolved oxygen content in water decreases as temperature and salinity increase (APHA 1992). Dissolved oxygen further decreases with an increase in altitude or decrease in atmospheric pressure (Barton and Taylor 1994).

In Alberta, two seasons are significant with regard to potential concern about low dissolved oxygen concentrations: winter and summer. In winter, ice-cover to a large degree prevents reaeration. Due to the oxygen removal processes, the oxygen content of water decreases over time in standing waters or decreases with distance downstream in flowing waters. In summer, oxygen is produced by photosynthesis during sunlight hours while oxygen is removed by respiration during day and night. In extreme cases, this can result in dissolved oxygen fluctuating from high levels during the afternoon to low levels just before dawn.

Analytical Methods

Dissolved oxygen (DO) can be analyzed by two main methods: the Winkler or iodometric method and the electrometric method using membrane electrodes. The choice of procedure depends on the interferences present, the accuracy required, and convenience or expedience (APHA 1992).

The iodometric method is the most precise and reliable titrimetric procedure for DO analysis. The method is based on the addition of several chemicals resulting in the liberation of iodine equivalent to the original DO content in the sample. The iodine is then titrated to determine the original DO content. Experienced analysts can maintain a precision of ± 50 μg/L with visual endpoint detection and a precision of ± 5 μg/L with electrometric detection (APHA 1992). Various modifications of the basic iodometric method exist (APHA 1992) to minimize the effect of interfering materials (such as oxidizing agents, reducing agents, or organic matter):

Azide modification	most wastewater, effluent, stream samples, especially if samples contain more than 50 μg NO_2^--N/L and not more than 1 mg ferrous iron /L
Permanganate modification	samples containing ferrous iron
Alum Flocculation modification	samples high in suspended solids
Copper Sulfate-Sulfamic Acid Flocculation modification	biological flocs

The major disadvantage of the iodometric method is that it is not appropriate for *in situ* or continuous measurement of DO.

Membrane electrodes provide an excellent method for DO in polluted waters, highly colored waters, and strong waste effluent. Oxygen sensitive membrane electrodes are composed of two solid metal electrodes in contact with supporting electrolyte separated from the test solution by a selective membrane. Polyethylene and fluorocarbon membranes are commonly used because they are permeable to molecular oxygen and are relatively rugged. The "diffusion current" is linearly proportional to the concentration of molecular oxygen present in the water (APHA 1992). The membrane electrodes however are susceptible to various physical conditions, which affect the diffusion rate of oxygen through the membrane. These influences are (roughly in order of decreasing importance): temperature, water flow, membrane fouling, salinity and barometric pressure (Truelson 1997). The main advantage of the electrometric method is that DO measurements are done *in situ* and the measurements can be continuous.

Both iodometric and electrometric methods are used by Alberta Environmental Protection for individual samples. The electrometric method with Automatic dataloggers (Hydrolab Datasondes) is used situations when a continuous record of DO concentrations is required: for instance, under-ice conditions or fluctuations of DO within a day.

3. EFFECTS OF DISSOLVED OXYGEN LEVELS ON FISH

3.1 Acute Effects

In most toxicity studies, acute toxicity data are expressed in concentrations that are lethal to 50% of the organisms in 96 hours (or LC_{50}). However, few toxicity studies concerning dissolved oxygen reported these LC_{50} values as was previously indicated by Chapman (1986). The data summarized by Doudoroff and Shumway (1970) were derived from highly variable test procedures, differing in duration, exposure regime and reported endpoints (Doudoroff and Shumway 1970; Chapman 1986). More recent reports have not addressed this variability. No attempt to classify the information into primary or secondary quality as outlined in the Protocol to Develop Alberta Water Quality Guidelines for Protection of Freshwater Aquatic Life (AEP 1996) was therefore made in this document. Instead, the available information on acute effects to fish is summarized in Table 1.

Younger fish tended to be more sensitive than older fish (Doudoroff and Shumway 1970; Alabaster and Lloyd 1982). A more recent study on yellow perch supported this general finding (Johnson and Evans 1991). In contrast, adult grayling were more sensitive to low dissolved oxygen than fry: at the same water temperature, adults lost equilibrium at dissolved oxygen concentrations 0.4 mg/L higher than fry (Feldmeth and Eriksen 1978). Furthermore, weight and size had no significant effect on the critical dissolved oxygen concentration of several species (Smale and Rabeni 1995a). Acclimation to low dissolved oxygen concentration decreased the lethal concentration (Doudoroff and Shumway 1970).

The lower incipient lethal concentration was determined *in situ* in three lakes in Ontario. For northern pike the incipient lethal limit was below 0.75 mg/L dissolved oxygen (DO) for all three lakes. Northern pike were extremely tolerant to low winter dissolved oxygen concentrations: some were found alive at 0.04 mg/L DO (Casselman 1978).

Oxygen consumption of juvenile paddlefish did not change significantly when dissolved oxygen pressure was reduced from 150 to 90 mm Hg. A further decrease to 5-10 mm Hg resulted in cessation of swimming, exaggerated buccal movements. When fish were returned to water saturated with dissolved oxygen, four of the 10 fish died (Burggren and Bemis 1992).

Table 1. Dissolved oxygen concentrations resulting in short-term toxic effects to fish.

Species	Acute Effects (in mg/L dissolved oxygen unless otherwise indicated)	Temperature (°C)	Reference
91 species	3		Doudoroff and Shumway 1970
bluegill	0.25	2.5 - 4	Petrosky and Magnuson 1973
4 species	1.2 [1]	25	Matthews and Maness 1979
grayling cutthroat trout	<2.2 <1.7	18 18	Feldmeth and Eriksen 1978
rainbow trout	<2.6	12.4 - 13.3	Thurston et al. 1981
rainbow trout	1.5, 1.6	15	Wirosoebroto-Hartadi 1986, as referenced in Truelson 1997
salmonids non-salmonids	3.4 3.0		Alabaster and Lloyd 1982; Slavonic literature
brown trout	3.2	16.3 - 16.6	Garric et al. 1990
central stoneroller minnow	2.33, 1.91	7.5, 23	Hlohowskyj and Chagnon 1991
6 species	< 8.4 Torr[2]	20	Castleberry and Cech 1992
paddlefish	5-10 mmHg[4]	24	Burggren and Bemis 1992
35 species	< 1.6	26	Smale and Rabeni 1995a

1 survival time varied from 60 min for emerald shiner to 118 min for plains minnow

2 1 Torr=1 mmHg, <1.07 mg/L DO calculation according to Davis (1975) at 20 °C (test condition) and assuming 760 mmHg air pressure

3 <0.1 mg/L DO calculation according to Davis (1975) at 24 °C (test condition) and assuming 760 mmHg air pressure

3.2 Chronic Effects

Growth

The efficiency of yolk conversion to larval body tissue was not materially affected at dissolved oxygen concentrations of 5 mg/L. However, yolk conversion was significantly impaired at 3 mg/L DO when water velocity was low (Doudoroff and Shumway 1970). Doudoroff and Shumway's (1970) review further indicated that low dissolved oxygen during embryonic development resulted in a reduction of alevin size. Doudoroff and Shumway (1970) questioned the significance of juvenile growth endpoints. Test conditions with an abundant supply of food to the test organisms may be irrelevant: in nature, food is never plentiful and fish have to expend energy to eat (Doudoroff and Shumway 1970).

Alabaster and Lloyd (1982) concluded from reviewing the information available at the time that a minimum value of 5 mg/L would be satisfactory for most stages and activities in the lifecycle of fishes. Processes such as juvenile growth, fecundity, hatch of eggs, larval morphology and survival, upstream movement of migratory salmon and schooling behaviour of some species were not particularly susceptible to dissolved oxygen (DO) levels above 5 mg/L. However, Alabaster and Lloyd state further that the 5 mg/L value may be unnecessarily high merely to ensure satisfactory survival of fish and adequate growth of juveniles.

At low dissolved oxygen concentrations, growth of coho salmon (Mason 1969), mountain whitefish (Siefert et al. 1974), smallmouth bass (Siefert et al. 1974), lake trout (Carlson and Siefert 1974), and lake herring (Brooke and Colby 1980) was reduced. The same effect was described for pike although no supporting data were presented (Casselman 1978). No clear "effect" concentrations and "no effect" concentrations were identified in these studies to include them in Table 2.

Keesen et al. (1981) determine weight gain of rainbow trout at 4 and 8 mg/L dissolved oxygen over a 6-week period. The experiment also determined the effect of feeding intensity and initial weight of the fish on weight gain. On average, weight gain was reduced 20% at 4 mg/L DO compared to the weight gain at 8 mg/L DO. No information was provided on water temperature during the test to evaluate these findings against a control (fully saturated conditions): the values were therefore not incorporated in Table 2.

Juvenile catfish experienced a decrease in body condition at 64% dissolved oxygen saturation compared to 100% saturation (Peterson and Brown-Peterson 1992). Length and weight did not change or increased slightly over the 20-day exposure period. The juveniles in 64% and 40% saturation treatment were stressed by a bacterial infection later in the experiment. This additional stress influenced growth: therefore results of this experiment were not included in Table 2.

Table 2. Dissolved oxygen concentrations (in mg/L unless otherwise noted) resulting in chronic effects on fish growth or feeding.

Species	Effect Level	No Effect Level	Temperature (°C)	Reference
literature review	3	5		Doudoroff and Shumway 1970
literature review: at moderately high temperature		5		Alabaster and Lloyd 1982
northern pike juveniles	2.6 fed ad libidum	≥5.4	18.6 - 18.7	Adelman and Smith 1970
fathead minnow fry	5	7.3	17.5 - 24	Brungs 1971
channel catfish	below 36% air sat (3 mg/L) @ 3% BW (body weight) feeding, at 60% air sat. (5 mg/L) ad libidum feeding	5 mg/L @ 3% BW feeding 8 mg/L ad libidum feeding	26.6	Andrews et al. 1973
channel catfish	3.5, 5.1	4.9, 6.3	25	Carlson et al. 1980
walleye embryo	3.4	4.8	17.2	Siefert and Spoor 1974
coho salmon	5.8	11	7 - 10	Siefert and Spoor 1974
coho salmon	4	5	15	Brett and Blackburn 1981
white sucker	2.5	4.9	18	Siefert and Spoor 1974
white bass		1.8 to 9.2	16	Siefert et al. 1974
smallmouth bass	6.3 6	8.6 8.3	20 23	Carlson and Siefert 1974
arctic char larvae	25% air saturation (<3.4)	normoxic (<12.3)	6.5	McDonald and McMahon 1977
yellow perch	growth 2.1 food consumption 3.5	growth 3.4 food consumption 4.8	20	Carlson et al. 1980
sockeye salmon	4	5	15	Brett and Blackburn 1981
rainbow trout		7, 10 and 14	9	Smart 1981
rainbow trout	6	7	15	Pedersen 1987

Smaller surviving yellow perch experienced less weight loss than the larger surviving fish (Johnson and Evans 1991). Channel catfish adjusted or missed their daily feeding in outdoor ponds when dissolved oxygen concentrations dropped below 5 mg/L (Randolph and Clemens 1976).

Reproduction

Doudoroff and Shumway's (1970) review indicated that low dissolved oxygen during embryonic development resulted in delayed development, and increased mortality as embryos aged. In fish species other than salmonids low dissolved oxygen in some instances resulted in mortality and delay in development. Alabaster and Lloyd (1982) concluded than any reduction of DO from air saturation value can slow development and embryonic growth, or delay hatching in salmonids. Most salmonid embryos will hatch successfully between 2 and 3 mg/L DO to produce relatively small and underdeveloped larvae that are viable and not deformed. Reduction in hatching success however can occur at higher DO concentrations.

Effects of low dissolved oxygen concentrations on hatching and survival of fish embryos are presented in Table 3. At low dissolved oxygen concentrations, hatching of fathead minnow (Brungs 1971), walleye (Oseid and Smith 1971), mountain whitefish (Siefert et al. 1974), white sucker (Siefert and Spoor 1974), coho salmon (Mason 1969; Siefert and Spoor 1974), brook trout (Siefert and Spoor 1974), lake trout (Carlson and Siefert 1974), scale carp (Kaur and Toor 1978), lake herring (Brooke and Colby 1980), and burbot (Giles et al. 1996) was delayed (Table 3).

Spawning behaviour was also affected at lower dissolved oxygen concentrations. Spawning behaviour of black crappie was affected at 2.5 mg/L dissolved oxygen, when fish became territorial. At this level, spawning fish started and finished spawning earlier than fish at higher concentrations of dissolved oxygen. No difference in success of spawning, number of embryos, viability of embryos, and hatching success of survival through swimup was observed between dissolved oxygen levels ranging from 2.5 to 6.5 mg/L (Siefert and Herman 1977). In contrast, the spawning period of burbot was extended under hypoxic (DO concentrations less than fully saturated) conditions (Giles et al. 1996).

Table 3. Dissolved oxygen concentrations resulting in chronic effects on hatching and survival of fish embryos.

Species	Effect Level	No Effect Level	Temperature (°C)	Reference
fathead minnow	eggs/female 2; survival 4	eggs/female 3; survival 5	17.5 - 24	Brungs 1971
northern pike	20-d survival 2.6	20-d survival 4.9	15	Siefert et al. 1973
northern pike	8-hr 2.2	8-hr 4	19	Peterka and Kent 1976, as referenced in Truelson 1997
white sucker	22-d survival 1.2; hatch time 2.5	22-d survival 2.5; hatch time 4.9	18	Siefert and Spoor 1974
coho salmon	119-d survival, hatch time 5.7	119-d survival, hatch time 11	7 - 10	Siefert and Spoor 1974
brook trout	133-d survival, hatch time 2.3	133-d survival, hatch time 2.9	8	Siefert and Spoor 1974
white bass	11-d survival 1.8	11-d survival 3.4	16	Siefert et al. 1974
mountain whitefish	193-d survival 4.6; 158-d survival 3.1	193-d survival 6.5; 158-d survival 6.0	4 7	Siefert et al. 1974
mountain whitefish	yolk area 5	mortality same 3 to 13.5 mg/L; yolk area 7	2-10.5	Giles and Van der Zweep 1996
smallmouth bass	14-d survival 4.4	14-d 8.7	20	Siefert et al. 1974
smallmouth bass	20-d survival 3.1 20-d survival 1.7; hatch time 4.2	20-d survival 4.5 20-d survival 3.0; hatch time 6	20 23	Carlson and Siefert 1974
lake trout	131-d survival, hatch time 4.3 108-d survival, hatch time 5.6	131-d survival, hatch time 6.0 108-d survival, hatch time 10.5	7 10	Carlson and Siefert 1974
walleye	20-d survival 3.4	20-d survival 4.8	17.2	Siefert and Spoor 1974
black crappie		survival, # embryos, hatch 2.5	20	Siefert and Herman 1977
scale carp (India)	% hatch 6	% hatch 9	25	Kaur and Toor 1978
lake herring	size of fry 2	size of fry 3,4	2, 4, 6, 8	Brooke and Colby 1980
bull trout	yolk area 5; hatch time 3	mortality same 3 to 13.5; yolk area 7; hatch time 5	2 - 10.5	Giles and Van der Zweep 1996
burbot		survival eggs same at 6 and 13	3	Giles et al. 1996
nase	0.7	7.5	15.9	Keckeis et al. 1996

Behaviour and Oxygen Demand

Fish compensate for hypoxia by several behavioural responses: increased use of air breathing, increased use of aquatic surface respiration (ASR), habitat changes, or changes in activity level (Kramer 1987).

Aquatic surface respiration and bubble exchange prevented loss of equilibrium in several east African cichlids. Tolerance to low dissolved oxygen concentrations of these cichlids may prevent predation by Nile perch in Lake Victoria (Chapman et al. 1995). Five electroid species (freshwater gobioid fishes) performed aquatic surface respiration when dissolved oxygen levels fell below 2 mg/L (Gee and Gee 1991). The west African elephant nose fish *Gnathonemus petersii* has an exceptionally large brain, which requires 60% of the total oxygen consumption of the fish. Gulping of air however was only identified below a dissolved oxygen concentration of 0.8 mg/L. Upright body posture was lost below a dissolved oxygen concentration of 0.3 mg/L (Nilsson 1996).

Fish frequently show a preference for areas with higher dissolved oxygen levels (Kramer 1987). The location of fish under ice was observed in response to progressive hypoxia (Magnuson and Karlen 1970). All fish species moved closer to the bottom of the ice as winter progressed. Pike survived the longest due to its positioning below the ice and lack of much motor activity. In contrast, perch swam rapidly and bluegill remained deeper in the water (Magnuson and Karlen 1970). During winter conditions, the depth of pike and perch capture was related to the concentration of dissolved oxygen present in the water. Pike and perch both moved up in the water column as dissolved oxygen concentrations decreased (Casselman 1978). In an aquarium with 0.5 mg/L DO, pike and perch had a tendency to move up in the aquarium, whereas bluegills tended to stay further down (Petrosky and Magnuson 1973). All species increased ventilation rates as dissolved oxygen concentrations dropped and the activity of the fish increased. Bluegill died at 0.25 mg/L DO (Petrosky and Magnuson 1973).

A combination of field and laboratory experiments indicated that central mudminnows entered hypoxic water (<0.4 mg/L dissolved oxygen; Rahel and Nutzman 1994). This severe hypoxic level was lethal within 4 hours based on an *in situ* bioassay. This behaviour coincided with feeding on *Chaoborus* larvae: in the field, these larvae were only present in severe hypoxic conditions. With access to the water surface, mudminnows engulfed airbubbles, which enabled them to spend more time in the hypoxic zone. Lack of access to the water surface in laboratory bioassays resulted in shorter times spent in hypoxic zones and consumption of fewer prey (Rahel and Nutzman 1994).

Some roach were homed to one half of a laboratory channel, whereas others were not intensively homed (Stott and Cross 1973). When dissolved oxygen was reduced to 0.7-1.1 mg/L within the home range, roach moved but stayed close to the home range. Homed roach moved back quicker into the home range when dissolved oxygen levels increased compared to the roach that were less succesfully homed (Stott and Cross 1973). A similar experiment with minnows (*Phoxinus phoxinus*) was performed. The minnows did not move outside of the home range until dissolved oxygen concentrations dropped to 1.2 mg/L, but they quickly returned within the home range once oxygen levels rose (Stott and Buckley 1979).

Walleye in their second summer remained in the shade at dissolved oxygen levels greater than 5.5 mg/L (Scherer 1971). At dissolved oxygen levels below 5.5 mg/L, walleye increased mobility into higher light intensities. Below 2 mg/L, walleye remained longer in higher light conditions.

Avoidance behaviour due to low dissolved oxygen levels can have significant effects: migrating salmonids avoided dissolved oxygen levels of 3.5 to 5 mg/L (Birtwell and Kruzynski 1989 as referenced in Barton and Taylor 1996).

Oxygen demand can also be reduced by selecting a lower water temperature (behavioural hypothermia, Jensen et al. 1993). For example, plains minnows (Bryan et al. 1984) and rainbow trout (Schurmann et al. 1991) preferred lower water temperatures at lower DO levels.

Exposure of fish to lower DO levels resulted in both increases and decreases in activity. A rapid increase in swimming may improve irrigation of gills and partly offset the effects of low dissolved oxygen levels. However, an increase in activity is usually temporary (Doudoroff and Shumway 1970). Changes in activity may include changes in ventilation frequency, reduced growth rates and feeding, reproduction, predatory avoidance (Kramer 1987). Remaining inactive however is not possible over the long term (Jensen et al. 1993).

Swimming capacity of many different species, at different temperatures and CO_2 levels were studied (Doudoroff and Shumway 1970). However, they questioned the ecological significance of the capability to swim at maximum sustainable speeds for prolonged periods. Burst swimming capability might be much more relevant as it relates to the capability to capture prey and to avoid predators (Doudoroff and Shumway 1970).

In small tanks or containers with very sharp gradients, fish usually appeared to avoid unfavourable conditions. In one case however activity increased at low dissolved oxygen concentrations but the direction was random (Doudoroff and Shumway 1970). In large tanks or natural conditions, fish moved to areas with higher dissolved oxygen levels. Different fish species are present along different zones in a gradient of dissolved oxygen levels; however fish are present at low levels even if higher levels are nearby (Doudoroff and Shumway 1970). The response of many fish to hypoxia (condition with low dissolved oxygen levels) is to escape (Jensen et al. 1993).

Pairing of two Nile tilapia alevins resulted in agonistic behaviour with frequent median and tail nippings (Alvarenga and Volpato 1995). The mean survival time under increasing hypoxia was similar for dominants and subordinates of the pairs, although the subordinates died faster than the dominants.

Physiology and Oxygen Demand

Raising the conductance of oxygen into the blood of fish is another possible mechanism to compensate for low DO levels. Oxygen conductance can be increased through increasing the flow of water over the gills (increased ventilation frequency, Jensen et al. 1993). The frequency and amplitude of opercular movements increased with a decrease in dissolved oxygen levels (Doudoroff and Shumway 1970). Arctic char larvae were exposed to normoxic and hypoxic (25% air saturation) conditions. The hypoxic larvae had coordinated buccal and opercular movements in contrast to the normoxic larvae (McDonald and McMahon 1977). Oxygen conductance can also be increased through an increase in gill oxygen diffusion conductance by: increasing the diffusion area (lamellar recruitment: Booth 1979 as referenced in Jensen et al. 1993), reducing diffusion distance across the gill (Soivio and Tuursala 1981 as referenced in Jensen et al. 1993), or increasing the diffusion gradient across the gill by reducing arterial blood oxygen pressure (Jensen et al. 1993). Hypoxic arctic char larvae had fewer filaments and lamella than the normoxic larvae. However, stimulated growth after 38 days resulted in the same lamellar surface area (McDonald and McMahon 1977).

Although reduced arterial blood oxygen pressure results in increased oxygen diffusion into blood, it requires compensation to provide more oxygen to cells. Compensation for reduced arterial blood oxygen tension can occur either through increased cardiac output (which is not typical) or an increase in the blood capacitance coefficient (Jensen et al. 1993). The blood capacitance coefficient increases through elevated hemoglobin concentration (which happens in some species, less in others) or an increase in the affinity of hemoglobin for dissolved oxygen. Affinity of hemoglobin for dissolved oxygen can also be adjusted by changing concentrations of intracellular co-factors (H^+, organic phosphates ATP, GTP). Hemoglobin affinity for dissolved oxygen increases with an increase in pH (decrease CO_2, Bohr effect). Adregenic stimulation of Na^+/H^+ exchange across membrane results in swelling and an increase in pH (Jensen 1993). Swelling of erythrocytes in response to hypoxic conditions appears to occur more readily at higher water temperature than at lower temperature (Soivio and Nikinmaa 1981). However, a decrease in hemoglobin due to a reduction in oxidative phosphorylation and an increase in intracellular pH both increase the blood oxygen affinity at lower water temperatures (Soivio and Nikinmaa 1981).

Blood characteristics may change in fish exposed to hypoxic conditions: increases in erythrocyte count, hemoglobin content, hematocrit (packed blood vell volume), lactate, and serum proteins and cellular swelling have been observed (Doudoroff and Shumway 1970). Hemoglobin levels in rainbow trout were the same at two saturation levels (>80% sat, 40% sat) with only a transient difference in hemocrit levels. However, there was a difference in presence and abundance of isomorphs (Marinsky et al. 1990). In contrast, no difference in hematocrit, hemoglobin levels in channel catfish were observed at different DO saturation levels (Andrews et al. 1973). Blood parameters and steroids in burbot also did not differ when they were exposed to 6 and 13 mg/L DO (Giles et al. 1996).

Many studies determined critical dissolved oxygen concentrations: sustainable rates of oxygen uptake below these concentrations cannot be maintained. Critical dissolved oxygen concentrations determined at the same temperature vary for the same species, and are dependent on flow, vessel, nutritional status fish, handling, illumination, and acclimation (Doudoroff and Shumway 1970). However, respiratory compensation is a normal adaptive response to environmental conditions. If there is not evidence that other functions are impaired (growth, etc.) an increase in gill irrigation is no evidence of injury (Doudoroff and Shumway 1970). Hughes (1981) also does not consider adaptive responses to hypoxia stressfull unless it represents an extreme hypoxic condition or persists for a very lengthy period. Adaptive changes within the respiratory chain usually compensate and lead to the restoration of new equilibria (Hughes 1981). Because the ecological and practical significance of critical dissolved oxygen levels based on oxygen consumption is questionable, these endpoints are not considered for deriving Alberta guidelines.

Teratogenic Effects

Low dissolved oxygen during embryonic development could result in structural deformities (Doudoroff and Shumway 1970, Table 4). Exposure of chum salmon embryos to low DO levels resulted in shortening of the vertebral column and abnormal alevins (Alderdice et al. 1958). Low dissolved oxygen levels resulted in an irreversibly locked lower jaw of largemouth bass larvae. These fish were unable to swim up and could not feed (Spoor 1977). Exposure of steelhead trout eggs to low DO concentrations resulted in significant abnormal development: twisted deformed tails or backs, abnormal structure nervous system/brain. Most of these embyos survived the hatching stage (Silver et al. 1963). Most chinook salmon embryos exposed to 1.6 mg/L DO were abnormal and did not survive the hatching stage (Silver et al. 1963). High numbers of abnormal fry of lake herring were produced at very low dissolved oxygen concentrations. The abnormal fry had deformed heads, jaws did not articulate, and the eyes were irregularly shaped (Brooke and Colby 1980). Exposure to 0.7 mg/L DO during the gastrula or eyed embryo stage did not affect mortality of nase up to hatching (Keckeis et al. 1996). However, all survivors were deformed and did not survive three days after hatching (Table 4).

Table 4. Dissolved oxygen concentrations resulting in teratogenic effects on fish embryos.

Species	Effect Level	No Effect Level	Temperature (°C)	Reference
chum salmon	below 0.3 mg/L DO		10	Alderdice et al. 1958
steelhead trout	2.6 mg/L		9.5	Silver et al. 1973
largemouth bass	1 mg/L DO for 3 hr on 6th day after fertilization		20	Spoor 1977
lake herring	1 mg/L DO at temperature of 2 and 4 °C and 2 mg/L at temperature of 6 and 8 °C		2, 4, 6, 8	Brooke and Colby 1980
bull trout		3 to 13.5 mg/L	2-10.5	Giles and Van der Zweep 1996
mountain whitefish		3 to 13.5 mg/L	2-10.5	Giles and Van der Zweep 1996
nase	0.7 mg/L DO during gastrula or eyed embryo stage		15.9	Keckeis et al. 1996

Effect of Combined Stressors

Combined hypoxia and hypercapnia (high CO_2 levels) result in respiratory acidosis (lower pH). Hyperventilation in this situation will not result in an increase in affinity of blood for dissolved oxygen (Jensen et al. 1993). However, high CO_2 levels can be avoided by fish and fish can acclimate to gradually increasing CO_2 levels (Doudoroff and Shumway 1970).

Hydrogen sulfide (H_2S) liberates oxygen from oxygenated hemoglobin and forms sulphaemoglobin. More importantly, H_2S inhibits cytochrome(c) oxidase in mitochondria of tissue cells, which blocks the last step in the respiratory chain where oxygen is the final electron acceptor (Jensen et al. 1993). The toxicity of hydrogen sulfide to goldfish increased at reduced dissolved oxygen concentration, both with and without acclimation of fish (Adelman and Smith 1972). Nitrite oxidises hemoglobin into metaemoglobin which does not transport oxygen; arterial dissolved oxygen content and blood capacitance were reduced (Jensen et al. 1987).

Increase in respiratory volume due to lower dissolved oxygen levels can result in more uptake of other toxic substances. For instance, uptake of mercury by carp increased when exposed to lower dissolved oxygen concentrations (Yediler and Jacobs 1995).

Some metals interfere with respiration: zinc and nickel reduce the diffusing capacity of the gills, resulting in a fall of oxygen supply to fish tissue (Hughes 1981). Nickel also increased the diffusion distance. The ventilatory frequency and amplitude increased during hypoxia following zinc treatment (Hughes 1981). An increase in H^+ and aluminum obstructs gill function: aluminum precipitates on the gill surface. Mucus production increased and gill lamellae fused and thickened (e.g. McDonald and Wood 1993 as referenced in Jensen et al. 1993). The toxicity of Zn, Pb, Cu, and phenols to rainbow trout increased at lower dissolved oxygen concentrations (Lloyd and Herbert 1962). The toxicity of zinc, naphthenic acids, and potassium cyanide to bluegills also increased at lower dissolved oxygen levels (Cairns and Schreier 1957 as referenced in Chapman 1986).

Ammonia was more toxic to rainbow trout at lower dissolved oxygen concentrations (Downing and Merkens 1955; Thurston et al. 1981). There was a strong positive relationship between the LC_{50} values (concentration lethal to 50 percent of the organisms) for ammonia and dissolved oxygen concentrations for rainbow trout: i.e., the toxicity of ammonia increased at lower dissolved oxygen levels.

Low dissolved oxygen was more toxic to brown trout at higher concentrations of suspended solids: the time lethal to 10 percent of the organisms (LT_{10}) decreased with decreasing dissolved oxygen concentrations and increasing concentrations of suspended solids (Garric et al. 1990).

The toxicity of linear alkylate sulfonate (LAS) to bluegill fingerlings increased with decreasing dissolved oxygen concentrations (Hokanson and Smith 1971). Fathead minnows were more susceptible to 1,2,4-trichlorobenzene at low dissolved oxygen concentrations than at higher concentrations (Carlson 1987). The central stoneroller minnow experienced loss of equilibrium at progressively higher dissolved oxygen levels as phenol concentrations increased (Hlohowskyj and Chagnon 1991). Phenols (phenol, dinitrophenol and pentachlorophenol) were also more toxic to the freshwater teleost *Notopterus notopterus* at lower dissolved oxygen levels (Gupta et al. 1983). The toxicity of 3-trifluoromethyl-4-nitrophenol (TFM) did not change at different dissolved oxygen levels for lamprey or rainbow trout at 20 °C. However, toxicity of TFM to rainbow trout increased at lower dissolved oxygen levels and 13 °C (Seelye and Scholefield 1990).

Survival time of coho salmon in a dilution of pulp mill effluent increased with increased dissolved oxygen concentrations (Hicks and DeWitt 1971). The principal toxicants of pulp mill effluent from softwood were resin acids. Resin acids and its principal component dehydroabietic acid were more toxic to fish at lower dissolved oxygen concentrations (Taylor et al. 1988 and Kruzynski 1979 as referenced in Barton and Taylor 1994).

Aquatic surface respiration increased at higher dissolved oxygen concentrations for parasitized three-spined stickleback compared to non-parasitized fish (Giles 1987).

The photo-induced toxicity of anthracene to bluegill sunfish (24-hr and 96-hr LC_{50}) was greatest at 6.9 mg/L DO and less at 5.0 mg/L and 8.1 mg/L DO (McCloskey and Oris 1991)

Despite the previous discussion, dissolved oxygen did not seem to greatly affect lethality of toxicants to fish (Sprague 1985). The increase in LC_{50} at levels of 20-30% dissolved oxygen saturation increased only about 1.5-fold. Sublethal effects are also not greatly modified by low dissolved oxygen (Sprague 1985).

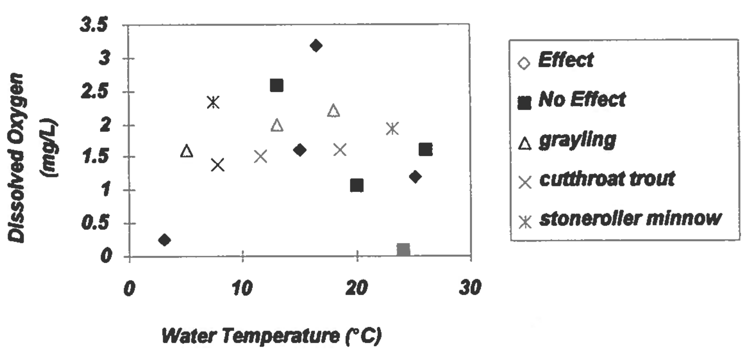
3.3 Water Temperature

The metabolic rate increases 2 to 3 times for every 10 °C increase in temperature. Heart rate and cardiac output increases. Changes in hemoglobin play a minor role and blood volume also appears to be relatively insensitive to temperature change (Jensen et al. 1993).

The minimum dissolved oxygen level that fish are able to tolerate increases with a rise in temperature (Alabaster and Lloyd 1982). At higher temperature, requirements for dissolved oxygen are generally greater for bluegill, largemouth bass, channel catfish (Moss and Scott 1961), chinook and coho salmon (Warren et al. 1973 as referenced in Chapman 1986) and largemouth bass (Brake 1972 as referenced by Chapman 1986). Survival of perch, roach chub, rainbow trout at low dissolved oxygen levels did not change from 16 to 20 °C, but decreased significantly from 10 °C to 16 °C (Downing and Merkens 1957). In a salmonid culture, the requirement for dissolved oxygen increased with an increase in water temperature (Forster et al. 1977 as cited in Smart 1981). Grayling and cutthroat trout lost equilibrium at higher dissolved oxygen concentrations when water temperature was increased form 5 to 19 °C (Feldmeth and Eriksen 1978: Figure 1). The only exception was that equilibrium of the central stoneroller minnow at increased temperature was lost at lower dissolved oxygen concentrations (Hlohowskyj and Chagnon 1991: Figure 1).

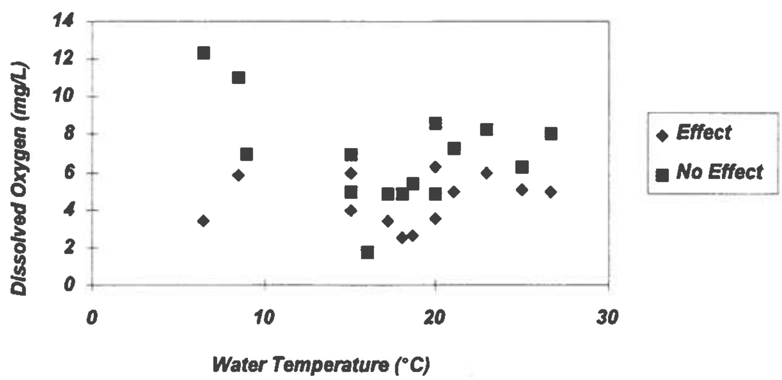
The acute toxicity data from Table 1 are plotted against water temperature in Figure 1. The effect and no effect data are single data points from different species. The acute effects data from the same species at different water temperature are presented separately. Figure 1 does not indicate a clear correlation between water temperature and acute toxicity data for the individual species. In addition, the effect levels and no effect levels of the various species clearly overlap over a wide temperature range.

Figure 1. Correlation between temperature and acute toxicity to fish.



The chronic toxicity data related to fish growth and feeding from Table 2 are graphed against water temperature in Figure 2. Very few data at different water temperature are available for the same species. The no effect level for coho salmon at higher temperature was lower than the effect level at lower temperature (Siefert and Spoor 1974; Brett and Blackburn 1981). The no effect level for rainbow trout was the same at different temperatures (Smart 1981; Pederson 1987). These limited data were not presented separately. Effect levels and no effect levels for the various species vary over the wide temperature range. However, Figure 2 does not indicate a clear correlation between water temperature and growth or feeding data.

Figure 2. Correlation between temperature and growth or feeding effects on fish.



Temperature has several effects on hatching and survival of fish embryos. Mountain whitefish, smallmouth bass and white bass embryos hatched quicker at higher temperatures (Siefert et al. 1974). Mountain whitefish fry were smaller and smallmouth bass fry were bigger at higher temperatures. Survival of white bass at low dissolved oxygen levels was much poorer at high temperature than at low temperature (Siefert et al. 1974). Lake trout developed faster at 10 °C compared to 7 °C, but did not survive as well at comparable oxygen levels (Carlson and Siefert 1974). The difference in survival and development in largemouth bass was the same at 20 and at 23 °C (Carlson and Siefert 1974). Development of walleye embryos was faster at 20 °C than at 17 °C at comparable oxygen levels, but few fish survived to the end of the test at 20 °C even at fully saturated conditions (Siefert and Spoor 1974). Low temperature appeared to favor the hatching of a higher percentage of normal fry (Brooke and Colby 1980). The effect data from the same species at different temperatures (previously presented in Table 3) are presented in Figure 3. There does not appear to be a significant trend in increased sensitivity to low DO concentrations with increased temperatures for the individual species.

Figure 3. Correlation between temperature and hatching or embryo survival of individual fish species.

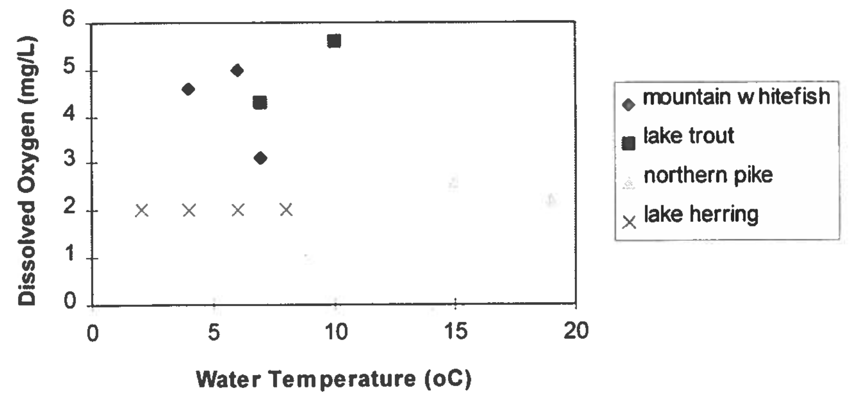
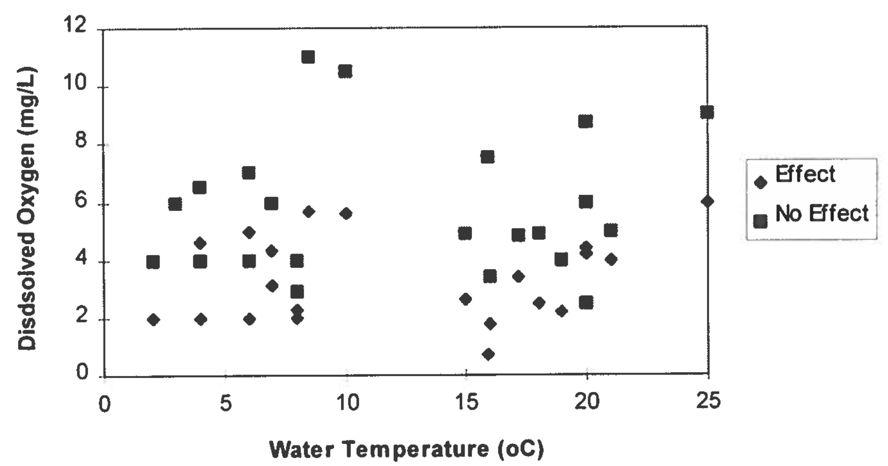


Figure 4. Correlation between temperature and hatching or survival of fish embryos.



The chronic toxicity data related to fish hatching and embryo survival from Table 3 are graphed against water temperature in Figure 4. Effect levels and no effect levels for the various species vary over the wide temperature range. However, Figure 4 does not indicate a clear correlation between water temperature and embryo hatching or survival data for all species.

3.4 Field Studies

Heuer and Sewell (1987) performed a 2-month experiment with large outdoor channels (43 m long). The channels were stocked with smallmouth bass, channel catfish, golden shiner, bluegill and grass carp. Reproduction of bluegills only occurred in the channel saturated with dissolved oxygen. Lack of spawning in other channels could be due to lower dissolved oxygen concentrations or to stress. Large losses of bluegills occurred during the experiment. In addition, competition occurred between bluegills and channel catfish. Biomass of golden shiners was high at saturated conditions, 5 and 4 mg/L DO, but was less at levels of dissolved oxygen below 4 mg/L. Embryos readily passed through downstream screen. Shiners were also subject to predation: biomass of shiners was lowest when the largest number of smallmouth bass were present. Survival of smallmouth bass was lower at dissolved oxygen concentrations below 5 mg/L compared to survival at higher DO concentrations. Bass were able to escape the enclosure and were also subject to cannabalism and predation.

Survival of steelhead trout embyos in Oregon streams was greater at sites with higher water velocity and dissolved oxygen concentrations (Coble 1961).

Survival of rainbow trout embryos was investigated in Young Creek, Ontario (Sowden and Power 1985). Survival was negligable in redds (salmonid spawning area within a creekbed) where dissolved oxygen concentrations were less than 5.2 mg/L. At DO concentrations greater than 5.2 mg/L, survival of embryos improved. However, the variability in survival between the redds was very large. This variability correlated with velocity above the threshold level of 5 cm/hr. Removal of metabolites was indicated as a potential cause for improved survival.

During a field study, Muller (1992) collected whitefish eggs from several Swiss lakes. The proportion of viable eggs was not related to dissolved oxygen content of water overlying the sediment. However, mortality of eggs was high in eutrophic lakes. Muller (1992) suggests that eutrophic lakes may have a very thin boundary layer in the sediment and that anoxic metabolites from below this boundary layer (H_2S and other toxic compounds) could diffuse into the eggs and prevent further development.

Turnpenny and Williams (1980) determined the survival of rainbow trout eggs in a river system influenced by industrial discharges. The survival of eyed eggs in artificial redds was greatest when the interstital dissolved oxygen concentrations was within 1-2 mg/L of that in the overlying water. No eyed eggs in artificial redds survived when DO in interstitial water was 6-8 mg/L less than DO in the overlying water; these sites corresponded to high siltation, low permeability, low apparent velocity and low supply rate of dissolved oxygen. Some survival occurred at DO concentrations greater than 4.9 mg/L: the LC_{50} was 6.5 mg/L DO. Alevin size was greater at higher velocities (increase in dissolved oxygen supply).

Matthews and Maness (1979) subjected four fish species to dissolved oxygen levels lowered to 1.2 mg/L at a temperature of 25 °C. Loss of equilibrium occurred at 118.5 min for plain minnow, 94 min for Arkansa river shiner, 75 min for red shiner and 60 min for the emerald shiner. However, temperature maxima of these fish species were more important as a selection pressure in the field than dissolved oxygen minima.

The headwater regions in Missouri were monitored for water quality and assemblages of fish during 1987 to 1990. Eighteen sites were monitored to determine minimum dissolved oxygen concentrations and maximum temperature and were sampled for fish during spring and fall. The observed minimum dissolved oxygen concentration observed in the field were significantly correlated with the hypoxia index (a calculated estimate of the critical DO for an "average fish" present at a site) (Smale and Rabeni 1995b).

4. EFFECTS OF DISSOLVED OXYGEN LEVELS ON AMPHIBIANS

The only information regarding low dissolved oxygen levels effects on amphibians relates to oxygen consumption and behaviour.

Metabolic oxygen regulation was tested for three species of salamanders while submerged. Two amphiuma species (3-toed and 2-toed amphiuma) were metabolic conformers over a 10 to 240 mm Hg range: that is, oxygen consumption of amphiuma increased with oxygen tension. Half of the greater siren were conformers. The other half of the sirens were regulators with a constant oxygen consumption at oxygen tensions (pO_2) greater than 92 mm Hg (Duke and Ultsch 1990).

Two aquatic salamanders, hellbenders and mudpuppies, were able to survive in severely hypoxic water (9-10 mm Hg) for 5 to 41 days by breathing from an air pocket. The critical oxygen tension for mudpuppies (*Necturus maculosus*) was 40 mm Hg and for hellbenders (*Cryptobranchus alleganiensis*) was 90 mm Hg (Ultsch and Duke 1990).

Tiger salamander selected a lower temperature when exposed to lower oxygen tension (Dupre and Wood 1988).

Most larvae of the brown-striped frog remained at the bottom of aquaria during normoxic conditions. Air breathing increased moderately with reduced dissolved oxygen concentrations at 20 °C. At 30 °C, air breathing increased below 15 kPa and increased more rapidly below 10 kPa. At 7 kPa, larvae continuously swam to the surface (Wong and Booth 1994).

Surfacing of the larvae of the South African clawed frog increased in response to lowered dissolved oxygen concentrations only below the critical tension for oxygen consumption (Hastings and Burggren 1995).

5. EFFECTS OF DISSOLVED OXYGEN LEVELS ON INVERTEBRATES

Doudoroff and Shumway (1970) stated in their literature review that in nature dissolved oxygen is depleted through respiration of organisms or through organic matter degradation. Although some sensitive invertebrate may disappear at chronic low DO concentrations, the increased productivity of other invertebrates can lead to more productive fisheries (Doudoroff and Shumway 1970). According to Davis (1975), dissolved oxygen requirements of invertebrates appear to reflect the environment they live in. Some may experience physiological and behavioural adaptations: chironomid larvae and leeches have the ability to acclimate by regulating oxygen uptake, *Daphnia* can produce hemoglobin and turn red, *Gammarus* can increase activity and leave the low oxygen environment, and freshwater snails can go to the surface. Davis (1975) concludes that due to the lack of knowledge, it was impossible to set a guideline to protect invertebrates. Literature reviewed by Alabaster and Lloyd (1982) indicated that many invertebrates survived DO levels substantially lower than 5 mg/L.

5.1 Acute Effects

The acute toxicity data for invertebrates are presented in Table 5.

Mortality was determined for several invertebrate species during short term tests (Jacob and Walther 1981; Jacob et al. 1984). A simple method was used where dissolved oxygen concentrations were lowered and the 2-5hr LC_{50} was determined (dissolved oxygen concentration lethal to 50% of the organisms). For the seven species tested, LC_{50} values (expressed as percentage saturation) varied from 5.2% for the mayfly *Siphlonurus aestivalis* to 49.5% for the mayfly *Rhithrogena iridina* (0.5 to 5.3 mg/L at an assumed atmospheric pressure of 101.3 kPa; Jacob and Walther 1981). Jacob et al. (1984) reported lethal oxygen conditions for 22 species: LC_{50} values (expressed as percent saturation) varied from 0.3% for the mayfly *Ephemera vulgata* to 96.5% for the mayfly *Epeorus sylvicola* (0.03 to 8.77 mg/L at an assumed atmospheric pressure of 101.3 kPa).

Males and females of the isopod *Asellus intermedius* and the amphipod *Hyalella azteca* had similar tolerance to low dissolved oxygen concentrations. Females of the amphipod *Gammarus fasciatus* however were more resistant to low dissolved oxygen than males (Sprague 1963). Juveniles of *Asellus aquaticus* and the amphipod *Gammarus pulex* were more sensitive (lower 24-hr LC_{50}) than male adults (Maltby 1995).

Four species of invertebrates (2 daphnids, 2 amphipods) stayed near the surface when exposed to lethal oxygen levels (Nebeker et al. 1992). When access to the surface was prevented, animals were unable to survive at the same dissolved oxygen concentration as when they had surface access (Nebeker et al. 1992).

Table 5. Dissolved oxygen concentrations resulting in short-term toxic effects to invertebrates.

Species	Acute Effects (in mg/L dissolved oxygen unless otherwise indicated)	Temperature (°C)	Reference
Daphnia magna	48-hr LC_{50} 0.6-0.7	-0.7	Nebeker et al. 1992
Daphnia magna	72-hr LC_{50} 1.27-3.19 male 72-hr LC_{50} 1.77-3.26 female 72-hr LC_{50} 0.94-1.91 juvenile	10, 20, 25	Hoback and Barnhart 1996
Daphnia pulex	96-hr LC_{50} 0.4-0.7	-0.5	Nebeker et al. 1992
Hyalella azteca	96-hr LC_{50} <0.3	16.8	Nebeker et al. 1992
Hyalella azteca	24-hr LC_{50} 0.7	20	Sprague 1963
Gammarus lacustris	7-d LC_{50} <0.2 (access to surface), 7-d LC_{50} 0.6-0.4 (no access to surface)	12.9	Nebeker et al. 1992
Gammarus pulex	24-hr LC_{50} 1.63 adult 24-hr LC_{50} 1.26 juvenile	15	Maltby 1995
Gammmarus fasciatus	24-hr LC_{50} 4.3	20	Sprague 1963
Gammarus pseudolimnaeus	24-hr LC_{50} 2.2	20	Sprague 1963
Gammarus limnaeus	96-hr LC_{20} 3	6.4	Gaufin 1973
Pteronarcys dorsata	96-hr LC_{50} 2.2	18.5	Nebeker 1972
Pteronarcys californica	96-hr LC_{50} 3.2-3.9	6.4	Gaufin 1973
Acroneuria lycorias	96-hr LC_{50} 3.6	14	Nebeker 1972
Acroneuria pacifica	96-hr LC_{50} 1.6	6.4	Gaufin 1973
Hexagenia limbata	96-hr LC_{50} 1.4	18.5	Nebeker 1972
Hexagenia limbata	96-hr LC_{50} 1.8	6.4	Gaufin 1973
Ephemera simulans	96-hr LC_{50} <1.5	18.5	Nebeker 1972
Ephemeralla subvaria	96-hr LC_{50} 3.9	18.5	Nebeker 1972
Ephemerella doddsi	96-hr LC_{50} 5.2	6.4	Gaufin 1973
Ephemerella grandis	96-hr LC_{50} 3.0	6.4	Gaufin 1973
Leptophlebia nebulosa	96-hr LC_{50} 2.2	18.5	Nebeker 1972
Baetisca laurentina	96-hr LC_{50} 3.5	18.5	Nebeker 1972
Hydropsyche betteni	96-hr LC_{50} 1.0-2.9	10-21	Nebeker 1972
Hydropsyche sp.	96-hr LC_{50} 3.6	6.4	Gaufin 1973
Tanytarsus dissimilis	96-hr LC_{50} <0.6	18.5	Nebeker 1972

Species	Acute Effects (in mg/L dissolved oxygen unless otherwise indicated)	Temperature (°C)	Reference
Asellus aquaticus	24-hr LC_{50} 0.32 male 24-hr LC_{50} <0.25 juvenile	15	Maltby 1995
Asellus intermedius	24-hr LC_{50} 0.03	20	Sprague 1963
Dinocras cephalotes	45-hr LOEC (LC_{60}) 80% sat (9.6 mg/L)* 45-hr NOEC (LC_0) 90% sat (10.8 mg/L)*	7.5	Benedetto 1970
Diura knowltoni	96-hr LC_{50} 3.6	6.4	Gaufin 1973
Diura bicaudata	50-hr LOEC (LC_{100}) 40% sat (4.8 mg/L)* 50-hr NOEC (LC_0) 60% sat (7.2 mg/L)*	7.5	Benedetto 1970
Nemoura cinerea	50-hr LOEC (LC_{40}) 40% sat (4.8 mg/L)* 50-hr NOEC (LC_0) 60% sat (7.2 mg/L)*	7.5	Benedetto 1970
Nemoura cinctipens	96-hr LC_{50} 3.3	6.4	Gaufin 1973
Arcynopteryx aurea	96-hr LC_{50} 3.3	6.4	Gaufin 1973
Arcynopteryx parallela	96-hr NOEC (LC_0) 2-5	6.4	Gaufin 1973
Brachyptera nigripennis	96-hr LC_{40} 2.3	6.4	Gaufin 1973
Pteronarcella badia	96-hr LC_{50} 2.4	6.4	Gaufin 1973
Callibaetis montanus	96-hr LC_{50} 4.4	6.4	Gaufin 1973
Rhithrogena robusta	96-hr LC_{50} 3.3	6.4	Gaufin 1973
Baetis bicaudata	72-hr LC_{90} 3.8	6.4	Gaufin 1973
Brachycentrus occidentalis	96-hr LC_{10} 2-4	6.4	Gaufin 1973
Drusinus sp.	96-hr LC_{50} 1.8	6.4	Gaufin 1973
Lepidostoma sp.	96-hr LC_{20} 3-4	6.4	Gaufin 1973
Limnephilus ornatus	96-hr LC_{50} 3.4	6.4	Gaufin 1973
Neophylax sp.	96-hr LC_{50} 3.8	6.4	Gaufin 1973
Neothremma alicia	96-hr LC_{50} 1.7	6.4	Gaufin 1973
Simulium vittatum	96-hr LC_{60} 3.2	6.4	Gaufin 1973

* assuming 760 mm Hg air pressure at 7.5 °C (test temperature)

5.2 Chronic Effects

The chronic effects of low dissolved oxygen levels on invertebrates are summarized in Table 6. Long-term survival (up to 120 days) was measured in several insect species (Gaufin 1973). The chronic survival data are not quite as well documented as the acute toxicity data, so only the information on the most sensitive species is listed in Table 6. The highest DO level affecting chronic survival of invertebrates was 5.8 mg/L: at this level, 50 percent of the stonefly *Acroneuria pacifica* survived 111 days. The least sensitive invertebrate was the fly *Atherix variegata*: 70% of the larvae survived 1.7 mg/L DO for 90 days (Gaufin 1973).

Respiration of the stonefly *Phasganophora capitata* nymph was independent of its size. However, respiration of the stonefly decreased with decreasing DO concentrations (Kapoor and Griffiths 1975).

Contrary to what Sprague (1963) found for acute toxicity, adult female *Gammarus* were more sensitive to chronic hypoxia than either adult males or juveniles. Adult males were least sensitive. Mate guarding behaviour was affected at 5 mg/L DO (no-observed effect concentration [NOEC] was 7 mg/L) during a 15-min duration test. During a longer term test (3 days) the lowest observed effect concentration (LOEC) for mate guarding behaviour was 2 mg/L DO with an NOEC of 2.7 mg/L DO (Hoback and Barnhart 1996).

Survival and growth of the mayfly *Hexagenia limbata* was determined at numerous combinations of temperature and dissolved oxygen (Winter et al. 1996). Based on multiple regression models survival and growth were related to dissolved oxygen concentrations and water temperature. However, some conditions were supersaturated with respect to dissolved oxygen and results from these conditions appeared to influence the regression relationships. The raw data were grouped to investigate the possible impact of dissolved oxygen on survival and growth by eliminating the variability due to water temperature. The data were grouped in four temperature ranges: 6.5-6.65 °C, 10.05-10.33 °C, 11.25-11.75 °C, and 13.95-15 °C. Corresponding ranges in DO were 5.1-11.47 mg/L, 2.7-12.58 mg/L, 3.86-7.64 mg/L and 0.82-8.9 mg/L, repectively. Results from some trials were combined when DO concentrations were virtually the same. Significant differences were only noted within the highest temperature group (13.95-15 °C): the NOEC was 6.29 mg/L DO and the LOEC was 2.44 mg/L DO (ANOVA: F=31.509, df=1,28, $P<5.3\times10^{-6}$). A comparison of mean growth (based on head width) indicated similar results: average growth at temperature between 13.95 and 15 °C was 5.58% at 8.9 mg/L DO, 4.49-6.87% at 6.17-6.41 mg/L DO, and 2.44% at 2.44 mg/L DO. Within the temperature range of 14 to 15 °C, dissolved oxygen affected the survival and growth of the mayfly. At his temperature, the LOEC and NOEC values for survival and growth were the same (6.29 mg/L and 2.44 mg/L, respectively): these values were entered in Table 6.

Table 6. Dissolved oxygen concentrations (in mg/L) resulting in chronic effects on invertebrates.

Species	Effect Level	No Effect Level	Temperature (°C)	Reference
Daphnia magna	21-d survival LOEC 0.5 5-d reproduction LOEC <0.9 19-d reproduction LOEC 0.6	21-d survival NOEC 0.9 5-d reproduction NOEC 0.9 19-d reproduction NOEC 0.9	17 - 19.2	Nebeker et al. 1992
Daphnia magna	26-d reproduction, time to 1st brood LOEC 1.8; final weight LOEC 2.7	26-d reproduction, time to 1st brood NOEC 2.7; final weight LOEC 3.7	21	Homer and Waller 1983
Daphnia pulex	10-d survival LOEC 0.8-1.1; reproduction LOEC 1.6-2.2	10-d survival NOEC 1.1-1.6; reproduction LOEC 2.1->2.2	16.4-16.9	Nebeker et al. 1992
Hyalella azteca	30-d LC_{50} <0.3; survival LOEC <0.3; growth and reproduction LOEC 1.2	survival NOEC 0.3; growth and reproduction NOEC >1.2	16.8	Nebeker et al. 1992
Gammarus lacustris	7-d survival LOEC <0.2	7-d survival NOEC 0.2	12.9	Nebeker et al. 1992
Pteronarcys dorsata	30-d LC_{50} 4.8-4.4		18.5	Nebeker 1972
Ephemera simulans	30-d LC_{50} 4.5		18.5	Nebeker 1972
Baetisca laurentina	30-d LC_{50} 5.0		18.5	Nebeker 1972
Tanytarsus dissimilis	30-d LC_{50} <0.6		18.5	Nebeker 1972
most sensitive of several species	111-d LC_{50} 5.8		10	Gaufin 1973
Diura nanseni		1-mo prior to emergence LC_5 5.1; close to emergence LC_5 7.1	8	Nagell and Larshammer 1981
Taeniopteryx nebulosa		1-mo prior to emergence LC_5 3.1; close to emergence LC_5 5.3	8	Nagell and Larshammer 1981
Cloeon dipterum		>200-hr LC_5 2.2	8	Nagell and Larshammer 1981
Baetis tricaudata	14-d survival, food removal lower at 5	14-d survival, food removal higher at 11	4.5	Lowell and Culp 1996
Hexagenia limbata	21-d survival and growth LOEC 2.44	21-d survival and growth NOEC 6.17	14 - 15	Winter et al. 1996; analysis raw data

The requirements for dissolved oxygen increased closer to the time of emergence of the stoneflies *Diura* and *Taeniopteryx* (Nagell and Larshammer 1981). However, dissolved oxygen requirements of benthic invertebrates emerging into adults have only been investigated for four species (Table 7). The DO requirements for emergence of three mayflies (*E. simulans*, *L. nebulosa*, *B. laurentina*) are among the highest chronic requirements found among fish and invertebrates.

Table 7. Dissolved oxygen concentrations affecting hatching of invertebrates into adults.

Species	Effect Level	No Effect Level	Temperature (°C)	Reference
Ephemera simulans	30-d LC_{50} 4.5; emergence 20% at 7.6, 15% at 6.1, 0% at 4.1	emergence 40% at 9	18.5	Nebeker 1972
Leptophlebia nebulosa	emergence 30% at 7.6, 20% at 6, 10% at 4, 0% at 2.4	emergence 70% at 9	18.5	Nebeker 1972
Baetisca laurentina	emergence 30% at 6, 0% at 4	emergence 70% at 7	18.5	Nebeker 1972
Tanytarsus dissimilis	emergence 0% at 0	emergence >80% at <0.6	18.5	Nebeker 1972

The effect of brooding behaviour and dissolved oxygen levels on the glossiphoniid leech *Glossiphonia complanata* was evaluated. The hatching rate was generally independent of oxygen saturation and brooding. Hatching of the leeches in conditions without parents present and at low dissolved oxygen concentrations (25% saturation) took significantly longer. Brooding enhanced survival of hatchlings at 25% saturation; no difference in survival was apparent at 50% dissolved oxygen saturation or higher. The weight of juveniles without parents was reduced at dissolved oxygen saturation of 50% or less compared with juvenile weight with parents (Milne and Calow 1990).

Behavioral hypothermia may also occur in invertebrates: they may select a lower water temperature to reduce oxygen demand during hypoxia. Crayfish selected a lower temperature when exposed to lower oxygen tension (Dupre and Wood 1988). The mayfly larvae *Cloeon dipterum* moved towards the dark end and the warm end in a light and temperature gradient, respectively (Nagell 1977). The reaction reversed under anoxic conditions (no oxygen present). In a combined light and temperature gradient, larvae moved to the dark and cold end under normoxic conditions, but moved to the warm and light end under anoxic conditions. Observations under field conditions indicated that larvae moved just beneath the ice surface.

Mayflies were put in stratified tanks consisting of bottom refugia with very low dissolved oxygen levels and higher dissolved oxygen levels at the surface (Rahel and Kolar 1990). The effect of fish (brook trout or brown trout) on the behaviour of the mayflies was investigated. At dissolved oxygen levels below 1.5 mg/L mayfly larvae left the bottom of tanks (bottom refugia) regardless of whether fish were present. Increased mayfly mortality occurred due to predation. However, mortality was lower than if the mayflies remained in the low-oxygen environment. A similar experiment was performed with four different species together (mayfly, amphipod, caddisfly and beetle; Kolar and Rahel 1993). All four taxa moved off the bottom when dissolved oxygen was lowered to 0.1 mg/L in the absence of fish. Mayflies and amphipods endured the low oxygen environment in the presence of fish. The behaviour of caddisflies and beetles was not affected by the presence of fish. As dissolved oxygen levels declined, all taxa except for mayflies decreased activity in the absence of fish. In the presence of fish, mayflies, amphipods and caddisflies were less active. This is not an uncommon reaction as outlined in the literature summarized by Kolar and Rahel (1993).

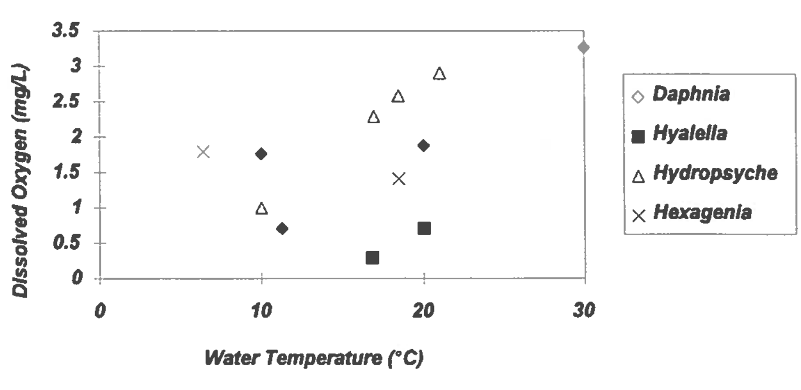
Mayflies were exposed to two different dissolved oxygen concentrations (5 and 11 mg/L - nominal) in recirculating streams (Lowell and Culp 1996). Mayflies at the low dissolved oxygen concentrations moved into areas with higher velocity. The survival of mayflies at low dissolved oxygen levels was improved when 1% effluent containing municipal and pulp mill effluent was added. The authors indicated that the increased survival with the addition of effluent may have stimulated the feeding rate of mayflies. This increased feeding rate may have partially offset the reduction in feeding rate caused by low DO concentrations. Lowell et al. (1995) indicated that pulp mill effluent had a stimulatory effect on mayfly growth. They proposed three possible mechanisms: increased nutritive value of the food, enhanced palatability of periphyton (attached algae), or increased growth via hormonal or other growth-stimulation effects (Lowell et al. 1995).

5.3 Water Temperature

The requirements of invertebrates for oxygen generally increased as temperature increased. LC_{50}s for the caddisfly *Hydropsyche betteni* were determined at different water temperatures (ranging from 10 to 21 °C). The LC_{50} increased from 1 mg/L DO at 10 °C to 2.9 mg/L DO at 21 °C (Nebeker 1972). Jacob and Walther (1981) tested four invertebrate species at different temperatures. In some cases, LC_{50} values were dependent on temperature: LC_{50} expressed as percent saturation increased with increasing temperature. In other cases, LC_{50}s did not change with temperature: for instance, mortality of the mayfly *Ephemera vulgata* only occurred when anoxic conditions were reached regardless of exposure time and test temperature (Jacob and Walther 1981). Jacob et al. (1984) tested 14 invertebrate species at different temperatures. At higher temperatures, lethal oxygen levels generally increased when expressed as percent saturation; expressed as concentration (mg/L), lethal dissolved oxygen levels either increased or were virtually the same at higher temperature (Jacob et al. 1984). Survival time of the midge *Chironomus anthracinus* in bottles starting with a dissolved oxygen concentrations of 0.15 mg/L was inversely correlated with temperature (Hamburger et al. 1995). The 72-hr LC_{50}s of female and juvenile cladoceran *Daphnia magna* increased as temperature increased. The change in LC_{50}s for males with increased temperature was variable (Hoback and Barnhart 1996: Figure 5).

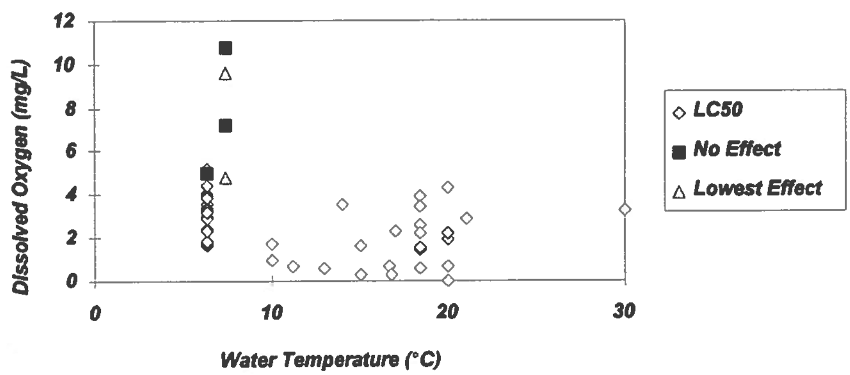
Figure 5 represents the acute toxicity data for individual invertebrate species. Although the data from some species indicate a decreased tolerance to low DO concentrations with increasing temperature (e.g., *Hydropsyche*), other data are not as clear (increasing tolerance with temperature for *Hexagenia*, small temperature range for *Hyalella*, relevance of *Daphnia* data point at extremely high temperture for Alberta).

Figure 5. Correlation between temperature and acute toxicity to individual invertebrate species.



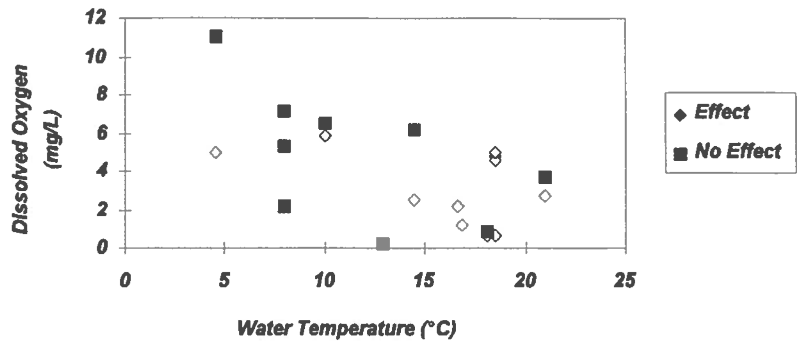
All the acute toxicity data (mostly LC_{50} values) from the various invertebrate species listed in Table 5 are presented in Figure 6. No clear correlation between water temperature and acute toxicity of the various invertebrate species is apparent.

Figure 6. Correlation between temperature and acute toxicity to invertebrates.



The chronic toxicity data are generally from different invertebrate species. The only data at different temperatures for the same species were for *Daphnia magna*. At higher temperature, tolerance to low DO appeared to decrease although the exposure to low DO at higher temperature was longer (Nebeker et al. 1992; Homer and Waller 1983, Table 6). The data are plotted against water temperature in Figure 7. There is significant variability among the "effect" data and among "no effect" data. In addition, the overlap between the "effect" and "no effect" data is substantial. No clear correlation between temperature and the chronic effects of low DO concentrations on invertebrate species is apparent.

Figure 7. Correlation between temperature and chronic toxicity to invertebrates.



6. DIURNAL FLUCTUATIONS OF DISSOLVED OXYGEN

Natural fluctuations of dissolved oxygen (DO) occur due to photosynthesis by plants and algae during daylight hours and respiration of all organisms throughout the day. The largest fluctuations of DO occur when the plant biomass is largest and most active (July and August). During these periods, river flows are generally low and water temperatures high. At lower flows, water temperature equilibrates faster to air temperature and oxygen saturation concentrations in water decrease at higher temperature. These conditions lower dissolved oxygen concentrations while the oxygen consumption of fish and invertebrates increases at higher temperature. Minimum levels of DO occur in early morning and coincide with increasing water temperature and light. These coinciding factors complicate interpretation of behavioral studies in the field (e.g., Suthers and Gee 1986). One attempt to simulate the effects of these factors identified light as a main trigger for increased activity of roach (Alabaster and Robertson 1961). In addition, very few laboratory studies have been carried out to determine the effect of fluctuating DO levels on fish and invertebrates. Thus, information regarding the effects of dissolved oxygen fluctuations on aquatic species or communities is sketchy (Alabaster and Lloyd 1982, Bagenal 1978, Chapman 1986).

Diurnal fluctuations are an important consideration when establishing DO guidelines that are intended to protect aquatic organisms and communities. Guidelines for DO (e.g., Chapman 1986, Truelson 1997) provide both an instantaneous minimum and multi-day mean DO level that is to be attained or surpassed. Several problems exist when applying these guidelines to diurnally fluctuating systems. The instantaneous minimum would be derived from a constant DO exposure in a laboratory, which would not represent a realistic aquatic system where low dissolved oxygen conditions would exist for brief periods. Dissolved oxygen levels below 5 mg/L have been reported during the early morning in reaches of several southern Alberta rivers. These reaches do not represent pristine, natural conditions, but generally represent areas affected by water withdrawal and nutrient enrichment. Although it is unclear whether DO conditions have changed due to these man-induced changes, fish and benthic invertebrate communities exist in these areas and are subjected to diurnal changes in DO. Problems also exist in calculating a multi-day mean. For example, it has been recommended that a daily average be used to evaluate a 30-day mean (Chapman 1986). The inherent assumption is that the daily average accurately reflects the trade-off between periods of higher and lower DO. A summary of studies examining the effects of diurnal fluctuations on aquatic organisms is presented in the following sections.

<u>Fish</u>

Serum Proteins- An increase in certain serum proteins in fish can be an indicator of biological stress (Bouck 1972). Bouck and Ball (1965) found that diurnal DO fluctuations altered the serum protein composition in bluegill and largemouth bass, but not in yellow bullhead. Similar effects in protein fraction were observed in rock bass subjected to diurnal DO fluctuations. However, the biological significance of changes in serum proteins was questioned (Bouck 1972).

Growth- Fluctuating DO reduced the growth rate of rock bass (Bouck 1972), juvenile largemouth bass (Stewart et al. 1967), yearling brook trout (Whitworth 1968), brook trout in some (but not all) experiments (Dorfman and Whitworth 1969), and coho salmon in one of two experiments (Fisher 1963). In seawater, diurnal fluctuations of dissolved oxygen reduced the growth rate of winter flounder (Bejda et al. 1992). Growth rates under fluctuating dissolved oxygen conditions were intermediate between growth rates at constant high DO levels and constant low DO levels (assumed Stewart et al. 1967; measured Fisher 1963 and Bejda et al. 1992). A comparison of growth rates of coho salmon under fluctuating and constant (mean or median of fluctuation) DO levels indicated that growth under fluctuating conditions was less than under constant levels (Fisher 1963). The same finding was reported for steelhead trout alevins (unpublished information Doudoroff and Shumway 1967 and Shumway and Putnam cited by Doudoroff and Shumway 1970). Growth rates of channel catfish were decreased at the lowest fluctuating DO levels (1 mg/L) compared to control fish exposed at uniform high DO levels during the same experiment (Carlson et al. 1980). However, growth rates of yellow perch exposed to diurnal fluctuation of DO were not affected compared to control fish exposed at uniform high DO levels during the same experiment (Carlson et al. 1980).

Feeding/Activity- Food consumption changed with growth of juvenile largemouth bass; fluctuating DO levels reduced feeding (Stewart et al. 1967). Reduced feeding under fluctuating DO levels was also observed in brook trout (Whitworth 1968) and rock bass (Bouck 1972). Feeding under fluctuating DO levels by black crappie was normal up to 20 °C (Carlson and Herman 1978). Non-lethal changes in DO caused more activity in roach, bream and perch (Alabaster and Robertson 1961). Largemouth bass larvae were also more active under fluctuating DO levels, which may result in higher mortality/predation (Spoor 1977).

Spawning- Black crappie did not spawn when DO fluctuated between 1.8 and 4.1 mg/L or 2.6 and 5.6 mg/L in one of the two replicate tanks. Spawning occurred under all other fluctuating DO conditions with a minimum DO levelof 3.6 mg/L or greater (Carlson and Herman 1978).

Invertebrates

Only one study was located dealing with the effects of diurnal DO fluctuations on aquatic invertebrates. Grant and Hawkes (1982) examined the effects of fluctuating DO under different conditions (temperature, exposure duration and amplitude) on the amphipod *Gammarus pulex*. Minimum dissolved oxygen levels were 0.5, 1 and 1.5 mg/L with maximum levels of 6 or 10 mg/L (at 10 °C) and 6 or 8 mg/L (at 20 °C). Minimum levels were maintained for 4, 8 or 12 hours during a 24-hr period. Amphipods were generally exposed to these conditions until 100% mortality occurred. Higher temperatures and longer exposure durations to low DO decreased survival of the amphipod. The maximum DO concentration did not significantly affect survival. Grant and Hawkes (1982) concluded that temperature, exposure duration and minimum DO level were critical factors determining survival of *Gammarus pulex* under the conditions of their experiment.

7. ENVIRONMENTAL CONCENTRATIONS

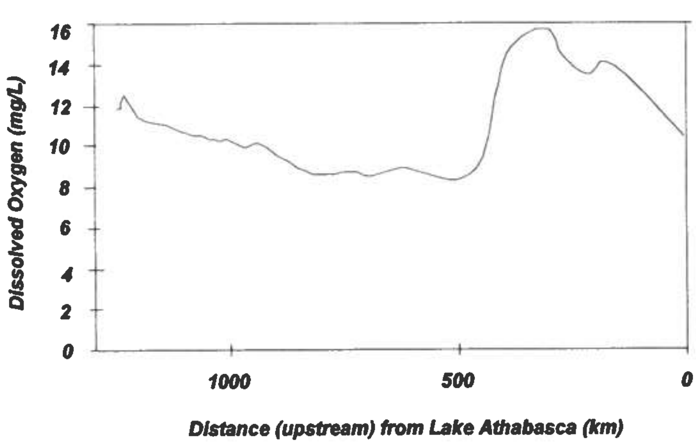
This section discusses the environmental concentrations of dissolved oxygen in rivers. In Alberta, dissolved oxygen concentrations in lakes are generally not influenced by point source discharges such as oxygen consuming industrial or municipal discharges. Dissolved oxygen concentrations in lakes are influenced by climatic conditions (e.g., ice cover), morphometry (e.g., depth), productivity, and watershed characteristics (e.g., oxygen sources, non-point source loading of organic material). These characteristics vary tremendously between Alberta lakes and between years. A general discussion of dissolved oxygen concentrations in lakes is provided in Wetzel (1975).

7.1 Winter Conditions

Most winter monitoring of dissolved oxygen levels in rivers has been carried out in northern Alberta.

In the Peace River, dissolved oxygen concentrations were highest in the winter due to greater solubility of oxygen at lower temperature. Dissolved oxygen depletion during the winter is minimal (Shaw et al. 1990).

Figure 8. Dissolved oxygen in the Athabasca River (Feb-Mar. 1993).

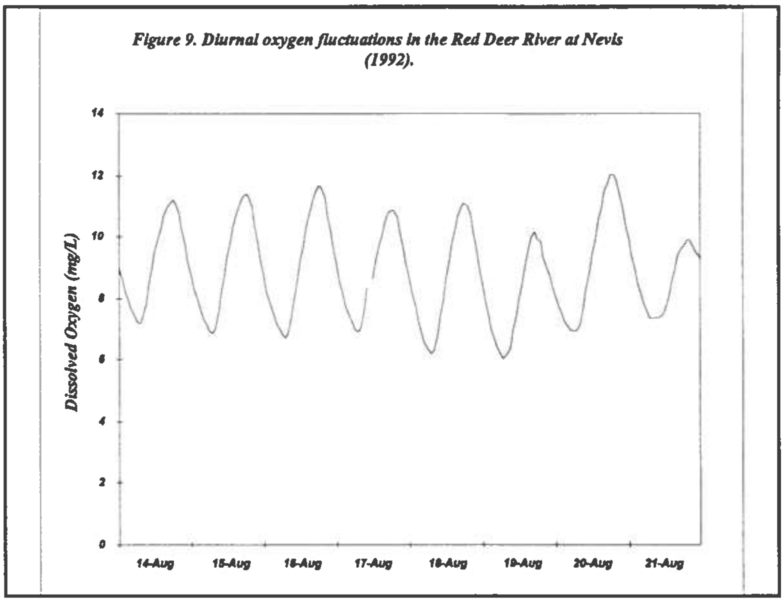


Dissolved oxygen concentrations in the Athabasca River during the winters of 1990 to 1993 exceeded 6.5 mg/L (Noton and Saffran 1995). Concentrations from the upstream open water reaches near Hinton declined due to ice cover, a number of oxygen demanding discharges from industries and municipalities, some tributaries with extremely low dissolved oxygen concentrations, and sediment oxygen demand (Figure 8). Dissolved oxygen concentrations increase when the Lesser Slave River joins the Athabasca River and due to reoxygenation at the Grand Rapids (Noton and Shaw 1989; Noton and Saffran 1995).

Dissolved oxygen concentration in the North Saskatchewan River declined from about 12 mg/L upstream of Edmonton to 9 mg/L at the Alberta-Saskatchewan border during the winters of 1986 to 1989 (Shaw et al. 1994).

7.2 Summer Conditions

Dissolved oxygen concentrations in the North Saskatchewan River downstream of Edmonton fluctuated during summer and early autumn. Concentrations varied from being supersaturated conditions during the day and decrease by as much as 4 mg/L during the night (Shaw et al. 1994).



Dissolved oxygen concentrations fluctuate in several rivers in southern Alberta. Fluctuations of dissolved oxygen concentrations in the Red Deer River are illustrated in Figure 9 (AEP monitoring records). Dissolved oxygen concentrations in the Bow River fluctuated over 6 mg/L in 1994 at several sites (AEP monitoring records). Fluctuations in the Highwood River, a tributary to the Bow River, were generally less than 1 mg/l during the summer of 1995 (AEP monitoring records). During 1990, dissolved oxygen fluctuations were greatest at a downstream site on Willow Creek compared to an upstream site. The fluctuations at an upstream site were less than 2 mg/L, whereas the fluctuations at a site further downstream were greater than 3 mg/l on some days (Shaw 1992). The fluctuations in the Oldman River during 1993 were generally less than 1 mg/L at several sites; however, the site at Pavan Park in Lethbridge indicated fluctuations less than 3 mg/L (AEP monitoring records). The maximum daily fluctuation of dissolved oxygen during the summer of 1990 was 4.34 mg/L for the Waterton River, 5.55 mg/L in the Belly River, and 5.86 mg/L in the St. Mary River (AEP monitoring records).

7.3 Interstitial Conditions

Very little information is available on dissolved oxygen concentrations in the interstitial water of bottom sediments in Alberta rivers. The following is therefore a general description of dissolved oxygen concentrations in rivers in North America. Interstitial concentrations of dissolved oxygen vary greatly temporally and spatially.

Changes in intergravel dissolved oxygen concentrations in four spawning streams in Alaska were greater over season and year than over a day. Many sites were subject to tidal changes. Spatial differences were greatest during periods of low discharge and warm weather (McNeil 1962).

In a small rural stream in east Texas, dissolved oxygen levels in interstitial water were generally lower during summer months compared to fall and winter months. Interstitial dissolved oxygen concentrations were generally less than in surface water. During cooler periods, the dissolved oxygen concentrations at 5 cm depth in the sediment approached surface water values. Dissolved oxygen concentrations generally decreased with depth in the sediment (Whitman and Clark 1982).

In June, interstitial dissolved oxygen concentrations in an Ontario stream decreased from 82% saturation at the surface to 10% saturation at 55-65 cm deep in a riffle. In October, saturation of DO in the interstitial water was less: 60% at the surface to 10% at a depth of 15-25 cm (Godbout and Hynes 1982).

In Sycamore Creek, Arizona, interstitial dissolved oxygen levels were determined in August when the water temperature was 24 to 30 °C. Dissolved oxygen concentrations in interstitial water generally were only slightly lower than concentrations in surface water. At 15 cm depth concentrations were within 1.5 mg/L of the dissolved oxygen concentration in surface water (Grimm and Fisher 1984).

Temporal and spatial variability of infaunal distribution and chemistry in a Canadian Shield stream was investigated during a spring flood. The difference in dissolved oxygen content between surface water and interstitial water varied from 2.2 to 6.4 mg/L in a riffle and from 0.2 to 10 mg/L in a pool (Giberson and Hall 1988)

The biology and chemistry of two Canadian rivers were investigated by Williams (1989). In August, the interstitial water beneath Duffin Creek was 20-30% saturated with dissolved oxygen and beneath Rouge River was 4-70% saturated. Surface water at that time was fully saturated with dissolved oxygen in both rivers (Williams 1989)

Interstitial dissolved oxygen concentrations were determined in a northern Michigan river at different sites along a riffle and at different times. At the downwelling (where river water moves into groundwater) upstream end of a riffle, interstitial dissolved oxygen concentrations reflected surface water characteristics. At the upwelling (where groundwater moves into the river) downstream end of the riffle, interstitial dissolved oxygen concentrations were "depleted" compared to surface water concentrations. These differences changed with the season. In summer (August), interstitial depletion of dissolved oxygen is more pronounced due to higher sediment oxygen demand and higher water temperature. In February, the difference is not nearly as pronounced: the maximum difference between surface water and interstitial water (top 5 cm) was 2 mg/L. For large areas in the riffle the difference between surface water and interstitial water was only 1 mg/L dissolved oxygen (Hendricks and White 1991).

Based on these studies, interstitial DO concentrations vary with discharge in a stream, discharge or recharge of groundwater, temperature, sediment oxygen demand (including biological activity) and depth in the sediment.

8. DERIVATION OF ACUTE AND CHRONIC ALBERTA GUIDELINES

As outlined in Chapter 1, the most recent USEPA guideline (Chapman 1986) was used as a starting point for the new Alberta guideline. The following sections discuss the available toxicity information in relation to the USEPA guideline.

8.1 Acute Guideline

Figures 10 and 11 graphically present the acute toxicity information for fish and invertebrates, respectively. The data were previously presented in Tables 1 and 5. The line in Figure 11 represent 5 mg/L dissolved oxygen, the recommended USEPA acute guideline (Chapman 1986).

Figure 10. Acute Effects on Fish.

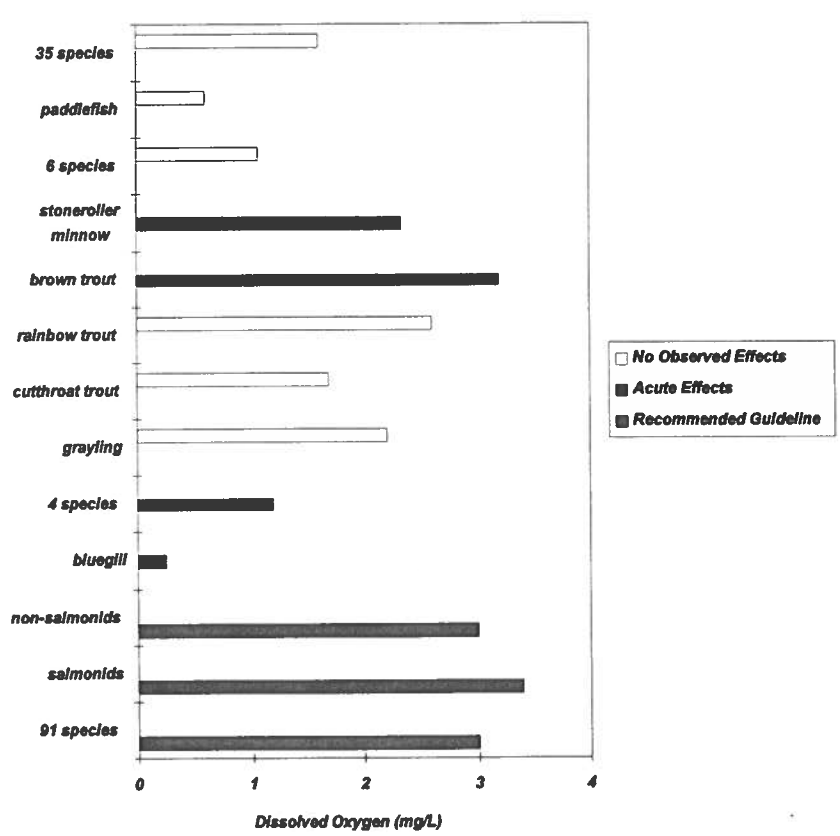
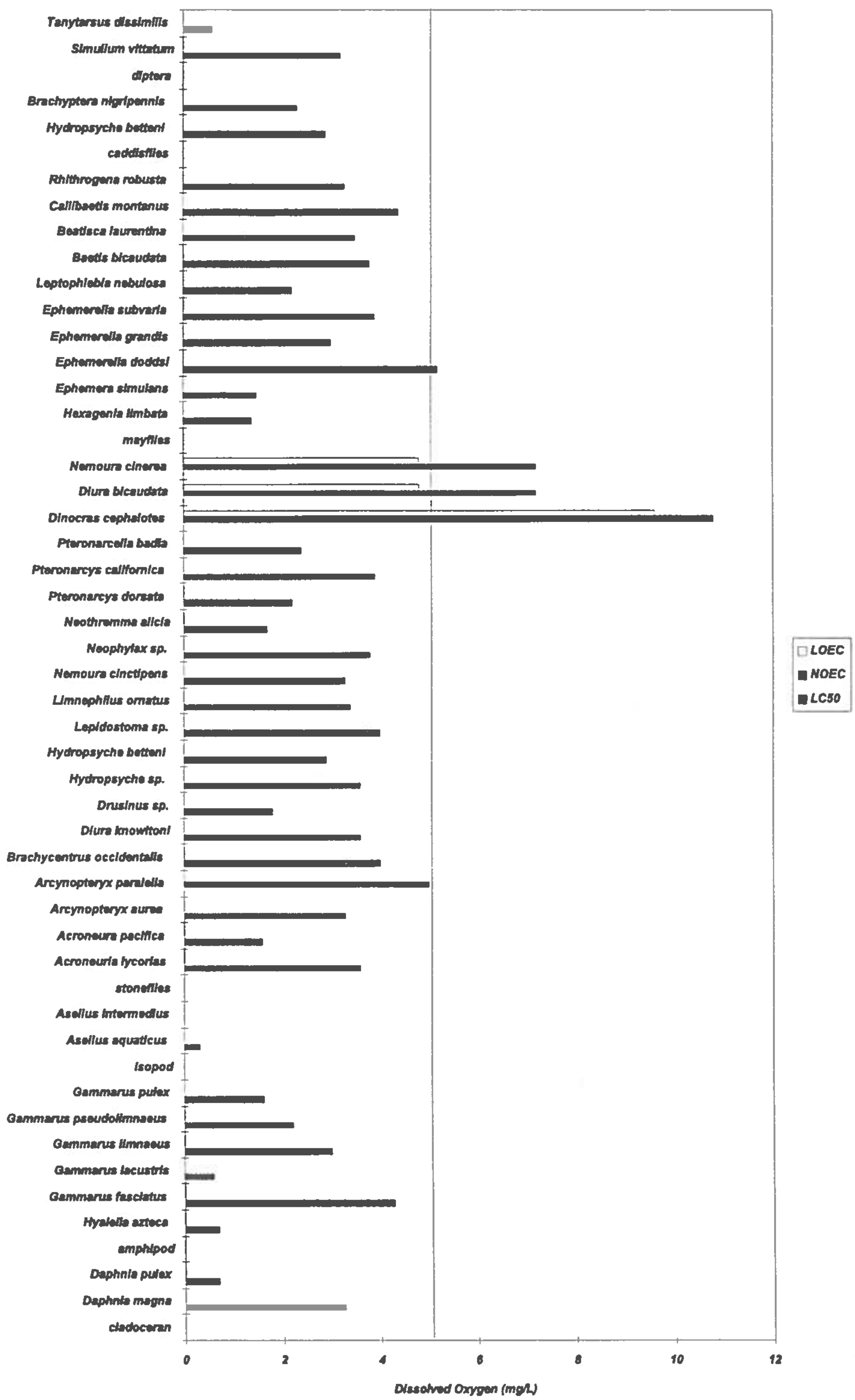


Figure 11. Acute effects on invertebrates.



The 5 mg/L DO recommended by Chapman (1986) is protective for most species represented in these figures. With respect to fish, the recommended 5 mg/L guideline is more protective than several recommended guidelines, several NOECs (no observed effect concentrations), and any acute toxicity effects by at least 1.8 mg/L. With respect to invertebrates, the recommended 5 mg/L guideline is more protective than one NOEC, some LOECs (lowest observed effect concentrations) and all but one short-term LC_{50}s (concentration lethal to 50% of the organisms). However, the LOEC for *Dinocras cephalotes* and the LC_{50} for *Ephemerella doddsi* was greater than the recommended 5 mg/L acute guideline. However, *Dinocras* is a European stonefly, not present in North America. The 96-hour LC_{50} for *Ephemerella* was only slightly greater than 5 mg/L DO (5.2 mg/L DO). Instead of adjusting the acute guideline value for this one species, the averaging period was adjusted to a period much shorter than the 96-hour exposure *Ephemeralla* was subjected to (see Section 8.3). In addition, short-term survival is compared to longer-term survival. Nebeker (1972) provided 96-hr and 30-day LC_{50} values for four species: the most sensitive 30-day LC_{50} value was 5 mg/L. Nebeker et al. (1992) determined that two species of daphnids and two species of amphipods had NOECs less than 2.2 mg/L in tests of 7 to 30-day duration. Gaufin's (1973) work indicated that different orders of invertebrates were able to survive concentrations less than 4.9 mg/L as follows:

	Survival > 50% at DO (mg/L)	days
Plecoptera	4.9	62
Ephemeroptera	4.6	30
Trichoptera	4	85
Diptera	2.4	40
Odonata	2.2	39
Amphipoda	2.8	20

These studies indicate that a wide variety of invertebrate species survive at 5 mg/L DO for periods much longer than those associated with an acute guideline. The 5 mg/L dissolved oxygen concentration is therefore adopted as the acute Alberta guideline. This acute guideline of 5 mg/L DO provides a high level of protection to both fish and invertebrate species over short-term exposures (see also Section 8.3).

Table 4 indicated that short-term exposure of early life stages of fish to concentrations less than 3 mg/L could result in teratogenic effects. Therefore the acute Alberta guideline provides ample protection against teratogenic effects.

The effect of temperature on acute toxicity of low DO concentrations was described previously in Figure 1 for fish and in Figures 5 and 6 for invertebrates. These figures indicated a lack of correlation between temperature and acute toxicity to low DO concentrations. Therefore, temperature was not incorporated as a factor to modify the acute guideline of 5 mg/L DO.

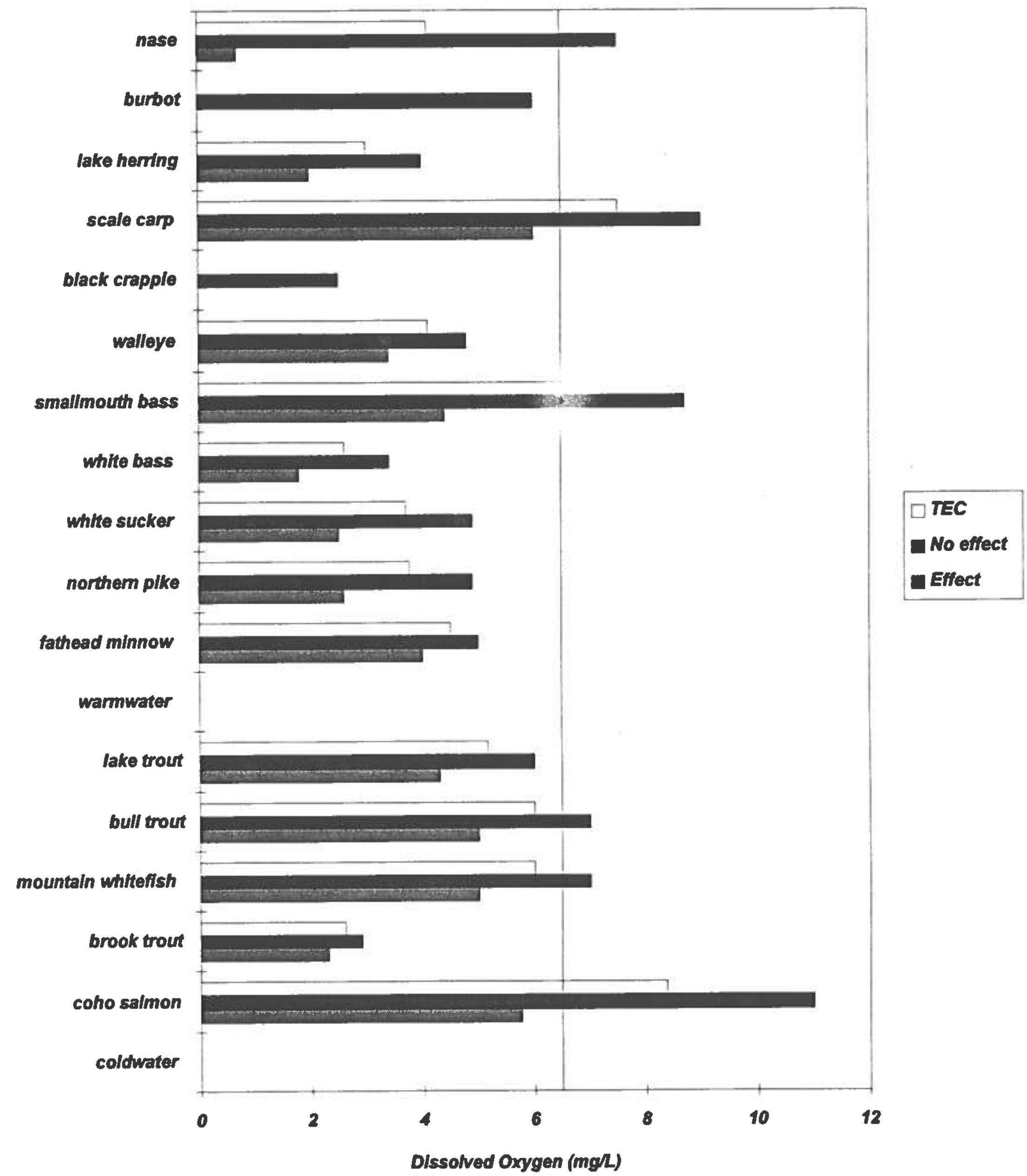
8.2 Chronic Guideline

Figures 12, 13, 14, and 15 graphically present the chronic toxicity information for fish (hatching and survival of early life stages and growth) and invertebrates (survival and growth, and hatching into adults). The data were previously presented in Tables 2, 3, 6 and 7. Only the most sensitive endpoints from each species are depicted. The lines in Figures 12, 13, 14, and 15 represent 6.5 mg/L dissolved oxygen, the recommended USEPA chronic guideline for coldwater criteria (Chapman 1986).

A threshold effect level (TEC) is generally calculated as the geometric mean of the highest no observed effect concentration (NOEC) and the lowest observed effect concentration (LOEC). This approach is similar to that outlined in the Alberta Protocol to Develop Alberta Water Quality Guidelines for Protection of Freshwater Aquatic Life (AEP 1996). However, a geometric mean provides a value closer to the lower of the two values: for most compounds this means closer to the NOEC and therefore more protective than an arithmetic mean. In the case of dissolved oxygen, a value closer to the higher concentration (NOEC) would be more protective. Therefore an arithmetic mean rather than a geometric mean is used to calculate TEC to compare the chronic toxicity data to the chronic recommended USEPA guideline.

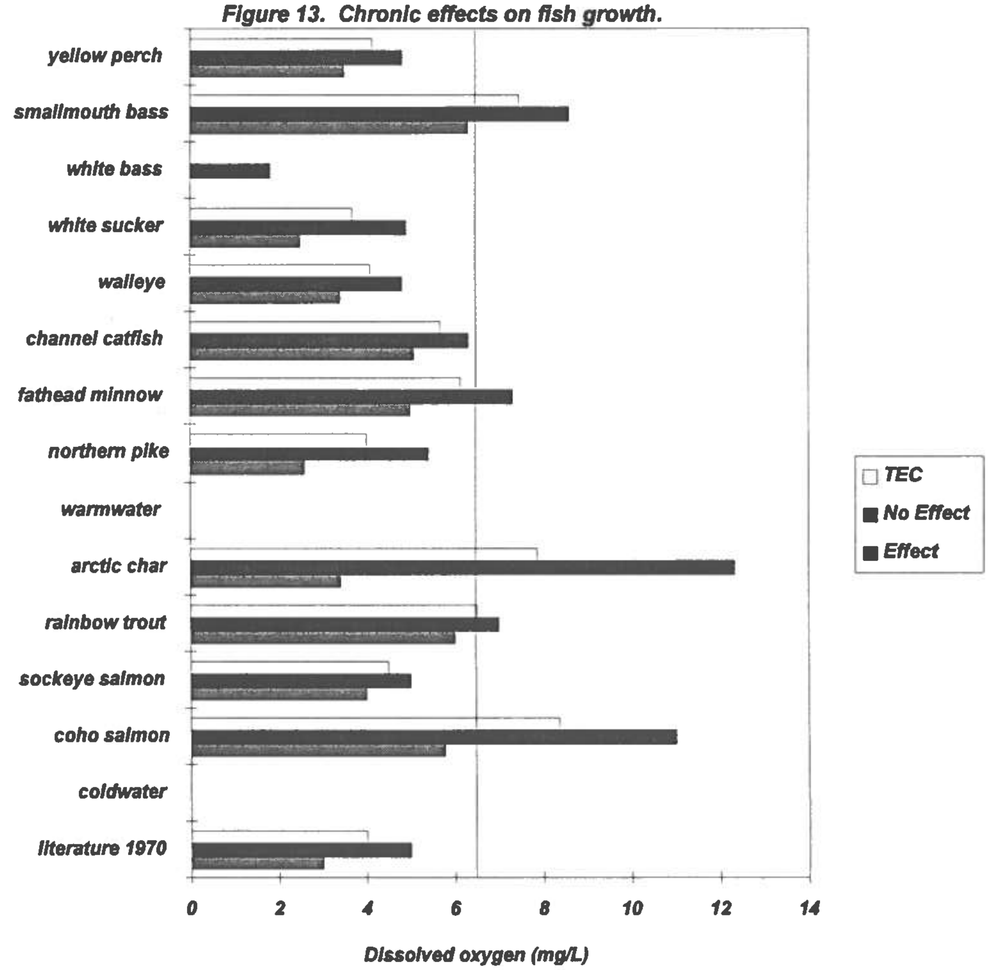
The TEC for hatching and survival of early life stages exceeded 6.5 mg/L DO for three species (Figure 12). The TEC for scale carp (Kaur and Toor 1978) was 7.5 mg/L. However, this is a species present in India and not in Alberta. The TEC for coho salmon (Siefert and Spoor 1974) was 8.38 mg/L DO. Survival of hatchlings was slightly lower at 5.75 mg/L DO compared to survival at 11 mg/L DO (73.1 % versus 79.6 %, respectively). However, the difference in survival was slight over a huge range in DO concentrations. The TEC for smallmouth bass (Siefert et al. 1974) was 6.55 mg/L DO. At 20 °C, survival of smallmouth bass was reduced 10% at 4.4 mg/L DO compared to 8.7 mg/L DO. Again this is a reduction over a large range in DO concentrations. A similar study on smallmouth bass with more DO concentrations tested in replicates and similar test conditions indicated that survival was not affected at 4.5 mg/L (Carlson and Siefert 1974).

Figure 12. Chronic effects on hatching and survival of early lifestages of fish.



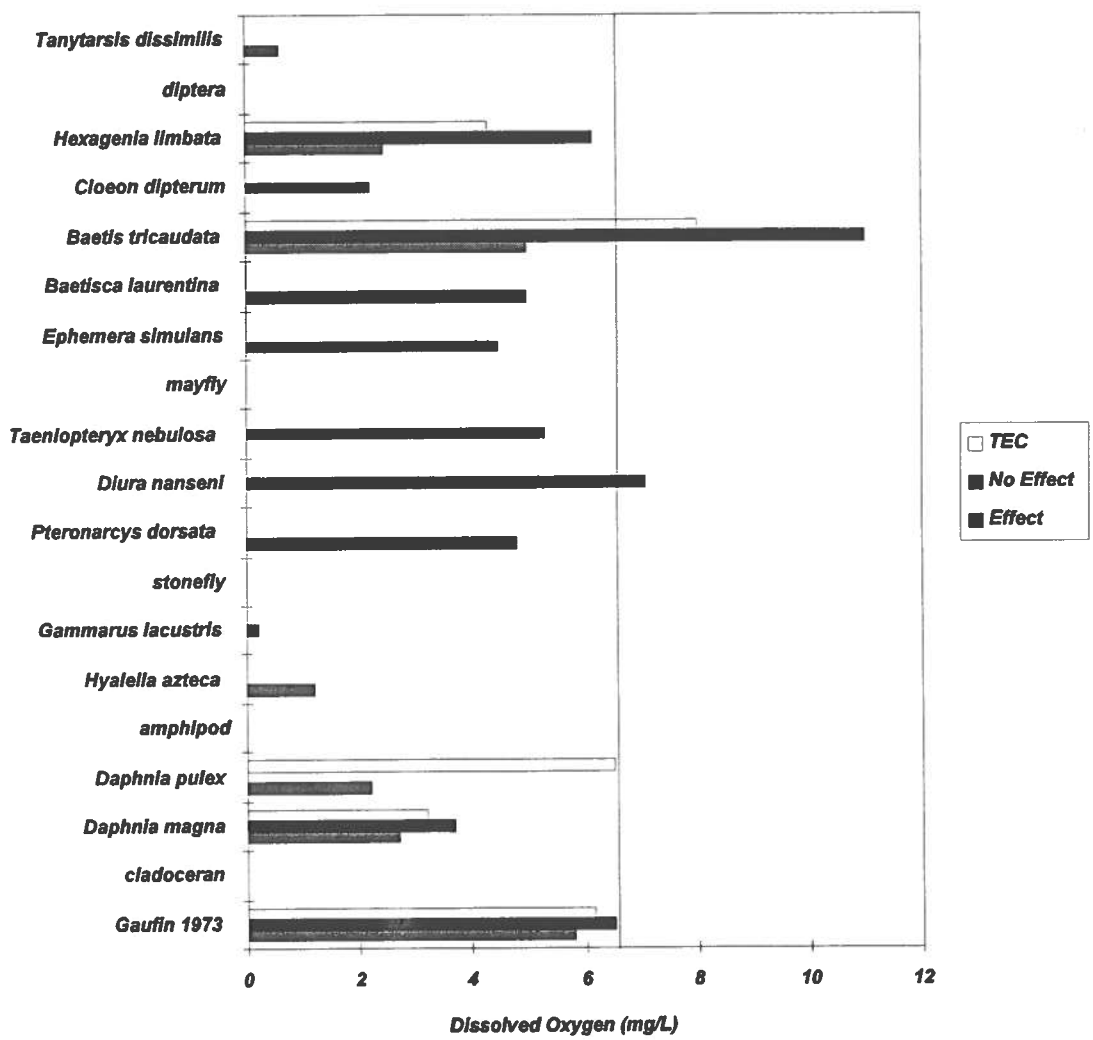
The TEC for fish growth exceeded 6.5 mg/L DO for three species (Figure 13). The TEC for coho salmon (Siefert and Spoor 1974) was 8.38 mg/L DO. The growth of hatchlings was slightly lower at 5.75 mg/L DO compared to survival at 11 mg/L DO (31.6 mm versus 33.8 mm, respectively). However, the range in length overlapped: 30-40 mm at 11 mg/L DO and 30-34 mm at 5.75 mg/L DO. The reduction in size was probably not significant and occurred over a huge range in DO concentrations. The TEC for arctic char (McDonald and McMahon 1977) was 7.85 mg/L DO, based on an LOEC of 3.4 mg/L DO and an NOEC of 12.3 mg/L DO. The actual value may be lower: the assumption of 760 mm Hg pressure had to be made to convert 33% and 100% air saturation at 6.5 °C into a mg/L concentration level. The hypoxic larvae weighed 20% less than the normoxic larvae 47 days after hatching. However, the range in DO concentrations is extremely large. The TEC for smallmouth bass (Carlson and Siefert 1974) was 7.45 mg/L DO. Length of the larvae was reduced by 0.9 mm at 6.3 mg/L compared to the length at 8.6 mg/L DO. However, the range in length of the larvae at the two DO concentrations was large and overlapped to a significant degree (9.4-13 mm at 8.6 mg/L DO versus 9-12.1 mm at 6.3 mg/L DO). The difference in length was not statistically evaluated to determine whether the reduction was significant.

Figure 13. Chronic effects on fish growth.



The TEC for survival and growth of invertebrates exceeded 6.5 mg/L DO for one species (Figure 14). Survival of and food removal by the mayfly *Baetis tricaudata* was lower at 5 mg/L compared to 11 mg/L (Lowell and Culp 1996). However, the range in dissolved oxygen concentrations was extremely large.

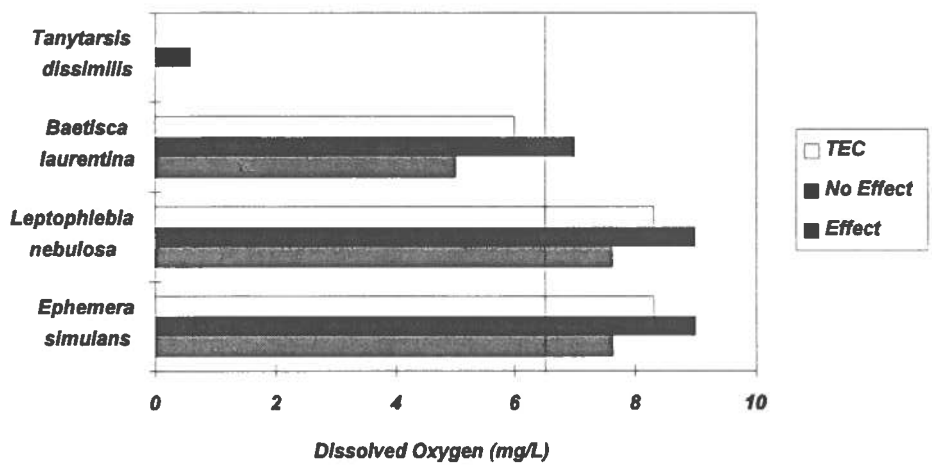
Figure 14. Chronic effects on invertebrate survival and growth.



The TEC and Effect Levels for emergence of two mayflies exceeded 6.5 mg/L DO (Figure 15). Emergence of *Ephemera simulans* at 7.6 mg/L DO was reduced to half compared to 9 mg/L

(Nebeker 1972). Emergence of *Leptophlebia nebulosa* at 7.6 mg/L DO was reduced more than 50% compared to 9 mg/L (Nebeker 1972). The TEC for both mayfly species was 8.3 mg/L.

Figure 15. Chronic effects on invertebrate emergence.



The recommended 6.5 mg/L chronic USEPA guideline exceeded several threshold effect levels (TECs). In total, TECs exceeded 6.5 mg/L for three studies on hatching and survival of early lifestages of fish, three studies on fish growth, one study on invertebrate survival and growth, and two studies on invertebrate emergence. However, several of the TECs were based on studies with a huge range between the "No Effect" and "Effect" concentrations (2 fish hatching/survival, 1 fish growth, 1 invertebrate survival). The lack of information within the wide range would disqualify these endpoints for guideline derivation for most compounds (AEP 1996). One TEC (fish hatching/survival) was on a species not present on this continent. The endpoint of two studies (fish growth) was likely not statistically significant. The endpoints in these studies were therefore not used to modify the recommended 6.5 mg/L chronic DO guideline.

The remaining endpoints concerned invertebrate emergence into adults of two mayfly species. The period of mayfly emergence is very short. Rather than modifying the guideline itself (which would extend over the entire year), the recommended chronic guideline was modified for the period of emergence for these species. Emergence of *Ephemera simulans* occurs between June 2 to June 14 in Michigan streams (Leonard and Leonard 1962) and end of May to first few days in June in various parts of North America (Edmunds et al. 1976). Emergence of *Leptophlebia nebulosa* occurs between May 11 to June June 15 in Michigan streams (Leonard and Leonard

1962) and late May and June in southern Canada (Edmunds et al. 1976). According to Clifford et al. (1979), *L. nebulosa* is very closely related to *L. cupida* and could possibly be a variant of the same species. An extensive study on *L. cupida* in the Bigoray River (Alberta) including field notes of nine years indicated that emergence of *L. cupida* starts mid May and extends to early July (Clifford et al. 1979). The TEC for both species (*E. simulans* and *L. nebulosa*) was 8.3 mg/L DO. Emergence of both species occurred between mid May and the end of June. For this period, the chronic guideline is adjusted to the TEC of 8.3 mg/L DO. The chronic Alberta dissolved oxygen guideline is therefore 8.3 mg/L between mid May to the end of June and 6.5 mg/L for the remaining period.

Concentrations of 6.5 mg/L also did not affect fish behaviour (Section 3.2).

The effect of temperature on chronic toxicity of low DO concentrations was described previously in Figures 2, 3 and 4 for fish and in Figure 7 for invertebrates. These figures indicated a lack of correlation between temperature and chronic toxicity to low DO concentrations. Therefore, temperature was not incorporated as a factor to modify the chronic guideline of 6.5 mg/L DO.

8.3 Application

Early Life Stages

As outlined in Section 7.3, dissolved oxygen concentrations in salmonid spawning habitats (redds) are less than those in overlying water. The intergravel dissolved oxygen concentrations are reduced due to sediment oxygen demand and respiration of fish embryos. To properly protect the early lifestages of these buried embryos, the guideline value (6.5 mg/L dissolved oxygen) should apply to conditions in the redds. The available information on dissolved oxygen levels in redds and overlying water has been described in Truelson 1997 and is summarized below. Based on some of this information, the USEPA adopted a differential of 3 mg/L between the overlying and interstitial water. The same differential is used by CCREM (1987) and Truelson (1997).

Dissolved Oxygen Differential: SW vs IW[1] (mg/L unless otherwise indicated)	Species	Reference
100% vs 75% saturation	natural sockeye salmon redds	Pyper and Vernon 1955
3.2, 3.5, 1.4	natural coho salmon redds	Koski 1965
2.1, 2.8, 3.7	natural brook trout redds	Hollender 1981
5	artificial steelhead trout redds	Coble 1961
100% vs 80-103% saturation, 2, 3	artificial rainbow trout redds	Turnpenny and Williams 1980
3.4	artificial salmon redds	Chevalier and Murphy 1985

1 surface water versus interstitial water

Rather than applying the differential of 3 mg/L dissolved oxygen at all times and to all places, the differential should be applied during those times and at those places where it is required. The differential should be applied from the period of spawning to 30 days after hatching, when early life stages are present. In addition, the differential should be applied to those areas where salmonids are known (or are likely) to bury their eggs. During these periods and for these areas, the chronic guideline for DO in surface water becomes 9.5 mg/L.

The application of the correct dissolved oxygen guideline value therefore requires information on the biota present at a site. For instance, are salmonid species present or likely to be present at the site, are these salmonid species spawning at the site or likely to spawn at the site, are these species fall or spring spawners, when are the embryos likely to hatch? Some of the general information on spawning and hatching of the various species in Alberta is outlined in Taylor and Barton (1992). Where specific information (species present at the site and use of the site) is not available, it is recommended that the appropriate studies be performed prior to granting any industrial or municipal license limits that may affect dissolved oxygen levels in rivers.

Natural Conditions

The dissolved oxygen guideline for Alberta does not apply to those situations which do not meet the guideline naturally.

The degree to which dissolved oxygen declines over the winter or fluctuates over the summer depends on watershed-specific and site-specific conditions. During the winter, the lack of reaeration can cause significant depletion of dissolved oxygen even under natural conditions

(Section 7.1). For instance, dissolved oxygen concentrations below 2 mg/L were observed in the Pembina River (Noton and Saffran 1995). Runoff from the watershed may contribute organic material to rivers, which under ice-cover could result in significant oxygen depletion. During the summer, the presence of aquatic macrophytes or benthic algae can result in significant oxygen fluctuations (Section 7.2). The dissolved oxygen fluctuations observed during the summer in Alberta depend on the degree of macrophyte or benthic algae biomass. Macrophyte or algal biomass depends on a great many influences: water velocity, light penetration, substrate characteristics, water temperature and nutrients.

In these situations, the following guidance provided by Doudoroff and Shumway (1970) and Chapman (1986) should be applied:

> *Where natural conditions alone create dissolved oxygen concentrations less than 110 percent of the applicable criteria means or minima or both, the minimum acceptable concentration is 90 percent of the natural concentrations (Chapman 1986).*

The Alberta DO guideline protects freshwater aquatic life: the guideline values should therefore be used for rivers and lakes. However, natural DO concentrations in lakes can vary due to climatic conditions, watershed characteristics, morphometry (depth and size of lake), etc. The Alberta guideline should therefore be used as a screening tool to evaluate the monitoring data in lakes. A more in-depth evaluation may be required to determine whether low dissolved oxygen concentrations in lakes are natural or due to other factors.

Temperature, Altitude and Oxygen Pressure

As depicted in Figures 1 to 7, temperature seemed to affect the DO requirements of a few species but not others. In addition, Chapman (1986) indicated that the USEPA guideline values are intended to be protective at typically high seasonal environmental temperatures for the warm and cold water species and the various lifestages. No temperature component is therefore incorporated in the Alberta guideline at this time, which is consistent with Truelson (1997).

Altitude has no known effect on the amount (concentration) of DO needed by aquatic organisms (Chapman unpublished as cited in Truelson 1997). Altitude is therefore not incorporated in the Alberta guideline for dissolved oxygen.

The Alberta guideline for DO is expressed in concentration values rather than as a percent saturation for the following reasons:

- Oxygen transfer to fish blood is dependent on the diffusion of DO down a concentration gradient across the gill. As outlined in Section 3.2, many factors can influence oxygen demand and the minimal tissue oxygen pressure necessary for metabolic activity. Because this minimal pressure has not been established, the necessarry diffusion gradient cannot be used to calculate the oxygen pressure (percent saturation) needed in water. Therefore, the percent saturation in water would not provide a true indication of the pressure differences across the gill or the needs of a fish.
- An expression of the guideline in percent saturation would result in lower DO concentration needs with increasing altitude. This contradicts the finding by Chapman that altitude has no known effect on the amount of DO needed by aquatic organisms (as cited in Truelson 1997).
- An expression of the guideline in percent saturation would result in lower DO concentration needs at higher temperature and higher DO needs at lower temperature. The available information does not support this concept. Instead, some information indicates that the DO needs of a few species may actually increase with an increase in temperature.

Averaging Period and Monitoring Frequency

The acute guideline of 5 mg/L should be applied as an instantaneous minimum. Using this guideline as a minimum protects aquatic biota (in particular early life stages and invertebrates) against the possible negative effects of fluctuating dissolved oxygen levels. As outlined in Section 7.2, dissolved oxygen concentrations can fluctuate diurnally during summer months. The available research seems to indicate that effects of diurnally fluctuating dissolved oxygen levels could be similar to constant levels equal to the minimum of the fluctuating dissolved oxygen levels (Section 7.2). If this limited research is correct, subjecting aquatic biota for any length of time to fluctuating conditions could potentially cause unacceptable effects. Therefore, application of the acute guideline as an instantaneous minimum would be environmentally protective. This "averaging" period is the same as the averaging period used for the acute USEPA and British Columbia guidelines (Chapman 1986; Truelson 1997).

The chronic guideline should be applied as a 7-day average value. To be protective, the averaging period should be less than the duration of the tests used to derive the guideline. The 7-day period is less than the exposure period of the fish and invertebrate studies listed in Tables 2, 3, and 6 (many of the invertebrate studies lasted 10 to 30 days). The 7-day averaging period is the same as the averaging period used for the early life stages in the chronic USEPA guideline (Chapman 1986), and the averaging period for chronic criteria (Taylor and Barton 1992, see next section).

Toxicity depends on the magnitude and duration of exposure. The magnitude of exposure is captured in the different acute and chronic guideline values. The duration of exposure is captured in the averaging period (AEP 1996). The use of a guideline with an averaging period is two-fold: to determine effluent limits and to evaluate field monitoring data. The use of the averaging period in setting effluent limits is described in the Water Quality Based Limits Procedure Manual (AEP 1995).

The use of an averaging period in evaluating monitoring data requires careful monitoring. The frequency of sampling has an impact on evaluating the seriousness of a DO value that does not meet the guideline. When monitoring is continuous, a single value not meeting the DO guideline may not be serious, whereas a single grab sample not meeting the guideline could be serious. Although continuous monitoring of DO would provide the best data to evaluate field conditions against the DO guideline, this may not always be possible. When continuous monitoring is not possible, it is important to monitor DO during periods that are most likely to be of concern. For instance, in the case of fluctuating DO concentrations, more than one sample per day needs to be collected. If the fluctuation is due to photosynthesis by plants, two samples collected early morning (7 or 8 am) and late afternoon would be sufficient to determine the range and average of the DO concentrations. If the fluctuation is due to anthropogenic causes (flow or effluent discharge fluctuations), more than two samples may need to be collected to determine the daily variability. These collections may need to be repeated for several days to establish a daily pattern. The values collected within a day are then averaged to determine the daily mean value. No values exceeding the air saturation value should be used to calculate a daily average: values greater than air saturation should be substituted by the air saturation value. The individual data should be time-averaged if the daily pattern is not sinusoidal.

Existing Alberta Guidelines and Objectives

Although the new Alberta guideline for dissolved oxygen appears more protective than the 1993 ambient surface water quality interim guideline (5 mg/L DO), the new guideline is consistent with current practices in setting license limits and determining instream flow needs (IFN). For example, Alberta Environmental Protection has applied stringent end-of-pipe limitations for oxygen-consuming organic material (biological oxygen demand or BOD) to pulp mills on the Athabasca River with very conservative assumptions. The result has been that over the last seven years DO concentrations remained above 6.5 mg/L during intensive monitoring of the river.

Also, Fish and Wildlife Division has been using the following DO criteria (in mg/L) for salmonid and other fish species in IFN studies (Taylor and Barton 1992):

	Life Stage	Averaging Period	Salmonids		Non-Salmonids
			Trouts	Whitefishes	
Acute	early life	daily minimum	8*	5	5
	fry	daily minimum	9.5*	5	5
	adult	daily minimum	4	4	3
Chronic	early life	7-day mean	9.5	6.5	6
	fry	7-day mean minimum	5.5	5.5	5
		7-day mean	6.5	6.5	6
	adult	7-day mean minimum	5	5	5
		7-day mean	6	6.5	5

* values are for the water column to achieve an intergravel DO concentration 3 mg/l lower.

The Prairie Provinces Water Board established water quality objectives for those sites where rivers cross provincial boundaries. The Water Quality Agreement (Schedule E of the Master Agreement of Apportionment) established the following objectives for dissolved oxygen:

PPWB sites at Alberta-Saskatchewan Border	Dissolved Oxygen Objective (mg/L)
Beaver River	6 during open water
North Saskatchewan River	6.5
Battle River	6 during open water

8.4 Alberta Dissolved Oxygen Guideline

In summary, the Alberta guideline for dissolved oxygen is:

		Guideline (mg/L)
Acute	1-day minimum	5
Chronic	7-day mean	6.5[1,2]

1 The chronic guideline should be increased to 8.3 from mid May to the end of June to protect emergence of mayfly species into adults.

2 The chronic guideline should be increased to 9.5 mg/L for those areas and times where embryonic and larval stages (from spawning to 30 days after hatching) develop within gravel beds (some salmonids). The chronic guideline is increased by 3 mg/L to account for the depletion of dissolved oxygen within the gravel.

For those situations where natural conditions do not meet the guideline for dissolved oxygen, the following guidance should be applied:

> *Where natural conditions alone create dissolved oxygen concentrations less than 110 percent of the applicable criteria means or minima or both, the minimum acceptable concentration is 90 percent of the natural concentrations.*

If natural natural conditions alone result in a dissolved oxygen concentration of 5 mg/L (which is below any of the guideline values), the minimum acceptable concentration would be 90% of 5 mg/L or 4.5 mg/L.

This guideline replaces the Alberta Ambient Surface Water Quality Interim Guideline of 5 mg/L (1994).

9. GUIDELINES IN OTHER JURISDICTIONS

In a document for the Food and Agriculture Organization of the United Nations, Doudoroff and Shumway (1970) proposed a set of different curves as the dissolved oxygen requirements for fish. These curves represent different levels of protection and their application is based on the natural seasonal DO minimum at a site.

Davis (1975) proposed a guideline to the National Research Council of Canada based on percent saturation levels and concentration values. At lower temperatures, guidelines were derived expressed as percent saturation and at higher temperatures, guidelines were derived expressed as concentration values. According to Davis, this would provide sufficient dissolved oxygen to maintain proper gas exchange across fish gills.

Alabaster and Lloyd (1982) proposed a dissolved oxygen guideline for the European Inland Fisheries Advisory Committee. A minimum constant value of 5 mg/L dissolved oxygen would be satisfactory for most stages and activities in the lifecycle of fish, provided that other environmental factors are favourable. The only identified attributes of the life cycle of fish that could be affected were slight reduction in larval weight, and sheltering behaviour of walleye. Tentative criteria are proposed because the effect of fluctuating DO levels on fish are not well understood. Annual 50-percentile and 5-percentile DO values should be greater than 5 mg/L and 2 mg/L for moderately tolerant freshwater fish and 9 and 5 mg/L for salmonids. These levels may not apply when young stages of fish are present or when temperatures are high.

The Canadian water quality guideline for the protection of freshwater aquatic life (CCREM 1987) is a variation of the USEPA guideline (Chapman 1986):

	Early Life Stage	Other Life Stage
Warmwater	6	5
Coldwater	9.5 (6.5[1])	6.5

[1] Interstitial water of the gravel

The most recent dissolved oxygen guideline in North America was prepared by British Columbia (Truelson 1997). The BC guideline is similar to those values proposed by the Science Advisory Board (USEPA 1986), which reviewed an earlier draft of the Chapman (1986) guideline. The following criteria were proposed:

	All Life Stages other than Buried	Buried Embryo/Alevin Life Stages	
	Embryo/Alevin	Water Column	Interstitial
Instantaneous Minimum	5	9	6
30-day Mean	8	11	8

Saskatchewan adopted the CCME guideline in their Surface Water Quality Objectives (Saskatchewan Environment and Public Safety 1988).

The DO guideline in Manitoba is 60% saturation (instantaneous minimum) for waters inhabited by cold water salmonids, and 47% saturation (instantaneous minimum) for waters inhabited by warm and cool water species (Williamson 1988).

The provincial water quality objective in Ontario and the Quebec guideline for dissolved oxygen are the same: they vary with water temperature and biota present (Ontario 1994; Quebec 1992). The values are the same as Davis proposed for the freshwater mixed fish population, including some salmonids, and for the freshwater mixed fish population with no salmonids, both with a harm commitment level B (some degree of risk to a portion of the fish population):

Temperature	Cold Water Biota		Warm Water Biota	
°C	% Saturation	mg/L	% Saturation	mg/L
0	54	8	47	7
5	54	7	47	6
10	54	6	47	5
15	54	6	47	5
20	57	5	47	4
25	63	5	48	4

Both jurisdictions added the following footnotes to the above table:

- In waters inhabited by sensitive biological communities, or in situations where additional physical or chemical stressors are operating, more stringent criteria may be required.
- In some hypolimnetic water, dissolved oxygen is naturally lower than the concentrations specified in the above table. Such a condition should not be alterred by adding oxygen

demanding materials causing a depletion of oxygen.

Quebec (1992) further specified that these values were the chronic guideline.

The most recent United States Environmental Protection Agency (USEPA) guidelines for dissolved oxygen are outlined in Chapman (1986). Different values are proposed for coldwater and warmwater species and early life stages and other life stages of fish as follows:

Averaging period	Coldwater Criteria		Warmwater Criteria	
	Early Life Stages[1,2]	Other Life Stages	Early Life Stages[2]	Other Life Stages
30-day Mean	NA[3]	6.5	NA	5.5
7-day Mean	9.5 (6.5)	NA	6	NA
7-day Mean Minimum	NA	5	NA	4
1-day Minimum[4,5]	8.0 (5.0)	4	5	3

1 These are water column concentrations recommended to achieve the required intergravel dissolved oxygen concentrations shown in parentheses. For species that have early lifestages exposed directly to the water column, the figures in parentheses apply.

2 Includes all embryonic and larval stages and all juvenile forms to 30-days following hatching.

3 Not Applicable.

4 For highly maniputable discharges, further restrictions apply.

5 All minima should be achieved as instantaneous concentrations to be achieved at all times.

The new Alberta guideline for dissolved oxygen is similar to the CCME guideline for coldwater species. The Alberta guideline does not distinguish between coldwater and warmwater species because both co-exist in Alberta rivers. The difference between the two guidelines is: (1) the addition of an Alberta acute guideline, which allows Alberta Environmental Protection to evaluate water quality and manage water quality by preventing short-term lethal and long-term sublethal effects, and (2) the refinement of the Alberta chronic guideline to account for the sensitive endpoint of mayfly emergence.

The BC guideline may seem to be more protective than the new Alberta guideline. This is the case for the acute guideline: the BC guideline for buried lifestages is 1 mg/L higher (more protective) than the Alberta guideline. The rationale for the additional 1 mg/L DO is to provide additional protection. However, a scientific basis is not provided by Truelson (1997) nor by the Science Advisory Board (USEPA 1986). The BC chronic guideline is difficult to compare with the Alberta chronic guideline because the averaging period is different (30 versus 7 days, respectively). A shorter averaging period results in a more protective guideline. Therefore the chronic guidelines cannot be compared.

The difference between the Ontario and Quebec guideline and the Alberta guideline are based on a different concept: oxygen saturation versus oxygen concentration. The fundamental differences are explained in Section 8.3.

The Alberta guideline is similar to the EPA coldwater criteria (Chapman 1986) as explained in the comparison with CCME guidelines. The Alberta acute guideline value is higher (more protective) than the USEPA acute value for other life stages. The reason is that some invertebrates experienced toxicity to DO concentrations close to 5 mg/L. The acute 1-day minimum Alberta value is the same as the USEPA 7-day mean minimum value for coldwater fish. The 7-day mean minimum value was therefore not incorporated in the Alberta guideline. The chronic guideline was the same as the chronic USEPA coldwater guideline. The only difference was the averaging period for other lifestages: the averaging period for Alberta and the USEPA is 7 days and 30 days, respectively. As explained in Section 8.3, the invertebrate studies used to derive the Alberta guideline generally lasted 10 to 30 days. A 30-day averaging period would not be sufficiently protective against the chronic effects of low DO concentrations on invertebrates.

10. RECOMMENDED RESEARCH

From the available data, it appears that dissolved oxygen requirements are highest for mayflies emerging into adults and for buried early lifestages of fish. The information on emerging insects is limited. In addition, information regarding the effects of fluctuating DO concentrations on aquatic biota is limited. The following studies are therefore recommended:

- validate the recommended DO guideline from May 15 to the end of June
  perform chronic toxicity tests on sensitive invertebrates through to emergence as adults

- determine how DO fluctuations affect sensitive aquatic biota
  1. determine egg viability, growth and activity for early life stages of rainbow trout (spring spawner);
  2. determine growth and activity levels for early life stages of mountain whitefish (fall spawners);
  3. determine growth and survival for sensitive aquatic invertebrates present in situations where DO concentrations fluctuate.

  The experiments should be performed to represent field conditions (water temperature, hardness, pH and light regime). The chronic toxicity endpoints should be determined under stable high dissolved oxygen conditions (afternoon maximum observed in the field), stable low dissolved oxygen conditions (early morning minimum observed in the field) and fluctuating dissolved oxygen conditions (between the two stable levels similar to field conditions).

No acute toxicity data are available on invertebrates at temperatures below 6.4 °C and only one chronic toxicity datapoint on invertebrates is available below 8 °C. In comparison, several datapoints on different species of fish at low temperatures (below 3 °C)are available. Therefore, research on invertebrates at low temperatures is recommended:

- only those species and lifestages present in rivers during winter conditions should be tested under controlled laboratory conditions at temperatures representing field conditions.

The results from these studies can be used to refine the Alberta guideline for dissolved oxygen. The results also will help refine the application of the guideline in situations where DO concentrations fluctuate.

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