Bull. Environ. Contam. Toxicol. (1990) 44:650–656

Bulletin of Environmental Contamination and Toxicology

Acute Lethal Toxicity of Ammonia and Suspended Sediment Mixtures to Chinook Salmon (*Oncorhynchus tshawytscha*)

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Ammonia poses a potential hazard to aquatic life in the vicinity of many municipal and industrial discharges. Criteria were established for ammonia to protect aquatic life and were revised to take account of the lower tolerance of salmon and trout and influence of pH and temperature. (Anon 1985). Suspended sediments can be directly lethal to salmonids and have been shown to cause sub-lethal histopathological, physiological and behaviourial responses (Servizi and Martens 1987; Redding and Schreck 1987).

Ammonia and suspended sediments occur together in many aquatic environments but there is no published information on their combined toxicity. The purpose of the work reported herein was to examine the acute lethal toxicities of mixtures of Fraser River sediments and ammonia to juvenile chinook salmon (*Oncorhynchus tshawytscha*).

MATERIALS AND METHODS

Sediments were obtained from a sand bar in the lower Fraser River near Ft. Langley and consisted primarily of particles 5 µm and smaller (geometric mean diameter). For further details of the sediments and their similarity to suspended sediments in the upper Fraser River, refer to Servizi and Martens (1987).

Static bioassays of underyearling chinook salmon were conducted in inverted cone shaped vessels (42 cm x 52 cm high x 35 l) fitted with recirculation pumps to maintain sediments in suspension (Servizi and Martens 1987). Temperature was maintained with a waterbath. Test fish were confined to cylindrical cages (30 cm x 8 cm high) placed 18 cm below the surface. Confinement at a pre-determined depth assured fish were exposed to the measured values of suspended solids.

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During bioassays, samples for suspended solids were taken twice daily at the top of the cages using a 23 mL cylinder (2.7 cm x 3.8 cm high), which was opened and re-stoppered under water. Samples were vacuum filtered through tared glass fiber Whatman GF/C filters, dried at 105°C for 1 hr, desiccated and weighed. Mean suspended solids were calculated for each bioassay concentration.

Total ammonia (NH_3) was measured during bioassays using the improved phenol-hypochlorite method (Harwood and Kuhn 1970). Prior to reagent addition, water and ammonia sediment mixtures were filtered through Whatman GF/C filters to remove suspended material. Ammonia was measured once daily and values averaged for each bioassay. Results are expressed as mg/L un-ionized ammonia (NH_3) which were calculated using Thurston et al., (1979) and the mean total ammonia, pH and temperature.

Temperature, pH and dissolved oxygen (D.O.) were measured daily during bioassays and the results averaged for each test. The mean pH, obtained with a Radiometer Model PHM64 pH meter (Radiometer Co. Copenhagen, Denmark), ranged from 7.81 to 8.11. Mean temperatures ranged between 5 and 8°C with the season. Dissolved oxygen, measured with a YSI Model 54 oxygen meter (Yellow Springs Instrument Co., Ohio, USA), exceeded 90% of saturation in all bioassays.

Natural water from Cultus Lake was used for rearing fish and for dilution water. This water has been tested regularly for 25 yr and is stable with respect to pH (7.8), conductivity (168 umhos/cm), alkalinity (60 mg/L), hardness (84 mg/L) and turbidity (2 NTU).

The mean wet weights of fish in test vessels ranged from 1.0 to 7g. Fish densities were below 0.8 g/L (American Public Health Association 1985) whenever possible but on a few occasions were about 1 g/L. Six single test solutions, each containing 10 fish were usually used for bioassay tests. When large fish (>3.5g) were used the test consisted of three different concentrations in duplicate, each containing five fish. Dead fish were summed for concentration pairs when calculating percent mortalities. Mortalities were removed daily. Buccal cavities and guts were examined for sediment.

Acute lethality of ammonia-sediment mixtures is expressed as percent mortality in 96h. Acute lethalities of ammonia and suspended sediment were quantified as 96-h LC50 values with 95% confidence limits (Litchfield and Wilcoxon, 1949).

Eyed chinook salmon eggs (upper Fraser River) were transferred from Quesnel River Hatchery to Cultus Lake Laboratory for incubation and rearing. Fry were fed Biodiet starter and later progressed to increasing sizes of Oregon Moist Pellets. Feed rate was 1.5% of body wet weight. Fish were not fed for 1d prior to or during bioassays.

RESULTS AND DISCUSSION

The 96-h LC50 for juvenile chinook exposed to ammonia at 7.0°C was 0.45 mg/L un-ionized NH_3 with 95% confidence limits of 0.43 to 0.47 mg/L. Buckley (1978) reported a value of 0.45 mg un-ionized NH_3-N/L (0.55 mg/L NH_3) for juvenile coho (*O. kisutch*) in the temperature range 14 to 19.5°C. For rainbow trout (*O. mykiss*) the mean 96-h LC50 was 0.61 mg/L un-ionized NH_3 in the temperature range 11.9 - 14.5°C (Thurston and Russo 1983). These authors also reported tolerance to ammonia was greatest for rainbow trout in the size range 0.7 to 10 g. While these comparisons suggest that juvenile chinook may be less tolerant of ammonia than rainbow trout or coho, there is also evidence that fish in general are less tolerant of un-ionized ammonia as temperatures decrease (Erickson 1985). Thus the apparent lower tolerance to ammonia by chinook may be associated with the lower bioassay temperatures.

The 96-h LC50 for juvenile chinook exposed to suspended sediment was 31 g/L at 7.0°C with 95% confidence limits from 29 to 33 g/L. The buccal cavities and gastro-intestinal tracts of 149 fish exposed to only suspended sediments were examined. Buccal cavities of the 49 fish which died were filled with sediment but none was observed in the digestive tract beyond this point. On the other hand, the buccal cavities and digestive tracts of the 100 survivors examined were free of sediment. We postulate that buccal cavities filled with sediments when fish became too fatigued to continue clearing buccal cavities via the cough reflex. For fingerling sockeye (*O. nerka*), the 96-h LC50 was 17.6 g/L for the identical suspended sediment at temperatures in the range 7.8 to 8.3°C (Servizi and Martens, 1987). Thus juvenile Chinook tolerated nearly twice as much suspended sediment as sockeye.

The 96-h toxicities of ammonia-suspended sediment mixtures increase with increasing content of these two constituents (Fig. 1). For exposure times of 24 and 48h, the mean mortalities for all mixtures tested were 20 and 57%, respectively, of the mortalities at 96h. The nature of joint toxic action is not immediately evident in Figure 1 but this characteristic of the mixtures can be examined using toxic units (Sprague 1970). Acute lethal toxic units (TU) are defined by:

$$TU = \frac{\text{Concentration}}{\text{96hLC50}}$$

The possibility that acute toxicities are additive can be examined by calculating the sums of toxic units for mixtures causing 50% mortality. If toxicities are additive, the sum of toxic units will equal unity for combinations of ammonia and suspended sediment which cause 50% mortality. When making this analysis, allowance was made for the possibility that there is a lower limit at which a toxic constituent is unlikely to contribute to the observed acute toxicity of a mixture. The "no effect" level

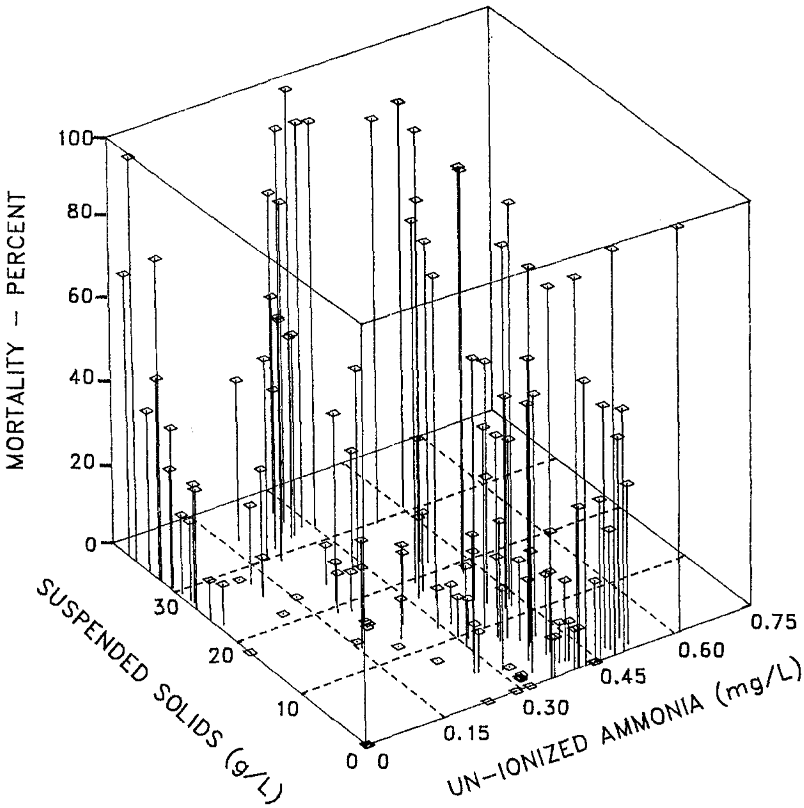

Figure 1. Mortalities of juvenile chinook salmon exposed to suspended sediment - ammonia mixtures for 96h.

reported for un-ionized ammonia is 0.12 TU (Lloyd and Orr, 1969). For Fraser River suspended sediment the "no effect" limit was 0.2 TU (Servizi and Martens, 1987). Six mixtures of ammonia and suspended sediments yielded 50% mortality when both constituents were present in excess of their "no effect" levels. The mean toxic unit total for these six mixtures was 1.34 TU (S.D. = 0.22). According to the terminology recommended by Sprague (1970), this result indicates that the combined toxicity of ammonia and suspended sediment is less-than-additive.

Summation of toxic units (Sprague 1970) has been used successfully to compute the joint toxic action of some mixtures of soluble toxicants; copper and zinc (Sprague and Ramsay 1965). Although the lethal mechanisms may differ for these toxicants, the results agreed with an empirical model represented by summation of toxic units. On the other hand, two components of a herbicide mixture were less-than-additive (Servizi et al. 1987). In the case of ammonia and suspended sediment reported herein, the less-than-additive result may be related in part to the capacity of colloidal clay fractions to adsorb ammonium ions. Ammonia adsorbed to suspended colloidal clay particles is unlikely to contribute to toxicity but would pass through glass fiber filters and be subjected to analysis for ammonia. Montmorillonoid and kaolin have been cited as clay components of Fraser River suspended sediments (Mackintosh and Gardner 1966).

Water quality and fisheries habitat managers have a need for readily interpreted joint toxicity data. To assist this need, the data in Figure 1 have been summarized into acute toxicity zones (Fig. 2). The zones were established using fit-by-eye and computer generated contour plotting based on quadratic smoothing software. The boundary between the "greater than 0% mortality" and "greater than 50% mortality" zones approximates a 50% mortality line. The sum of toxic units along this line exceeds unity and confirms that the lethalities of ammonia and suspended solids are less-than-additive. For example, 0.6 TU ammonia and 0.8 TU suspended solids intersect on the 50% mortality line and their sum is 1.4 TU. Ideally, mixtures which are additive fall along a straight line connecting 1.0 TU on the abscissa and ordinate (Sprague 1970).

Using Figure 2, it can be seen which non-lethal concentrations of suspended sediment and ammonia occurring together are likely to create lethal or sub-lethal mixtures. For example, 0.55 TU un-ionized ammonia plus 0.55 TU suspended sediment would be expected to cause mortality. On the other hand a mixture of 0.33 TU un-ionized ammonia and 0.31 TU suspended sediments is likely to cause only sub-lethal toxic effects.

Sublethal effects can be expected for some mixtures within the non-lethal zone. Gill pathology has been reported among juvenile chinook chronically exposed to 6-8 ug/L un-ionized ammonia but no effect was observed on similar fish exposed to 15 ug/L for 1 hr/d (Burrows 1964). Gill hypertrophy and necrosis were reported for juvenile sockeye exposed to 3.1 g/L Fraser River suspended sediments for 96-h while adult sockeye displayed elevated plasma glucose following chronic exposure to 500 mg/L (Servizi and Martens 1987). Gill pathology may provide an entry for infectious organisms. Redding and Schreck (1987) reported reduced tolerance among yearling steelhead (*O. mykiss*) to a bacterial pathogen following exposure to 2.5 g/L topsoil for 2d. Since both ammonia and suspended solids cause sublethal gill pathology there is a distinct possibility that these may combine to reduce tolerance to infectious diseases, especially if environmental factors such as temperature are adverse. Fraser River salmon may be especially susceptible since high pre-spawning mortalities among sockeye were attributed to *C. Columnaris* brought on by high temperatures (Colgrove and Wood 1966). Both suspended sediments and temperatures are high in the Fraser River during summer (Servizi and Gordon 1989). In addition, the Fraser River received about 1.5×10^6 m^3/d of municipal and industrial wastewaters containing ammonia in 1985 (Servizi 1989).

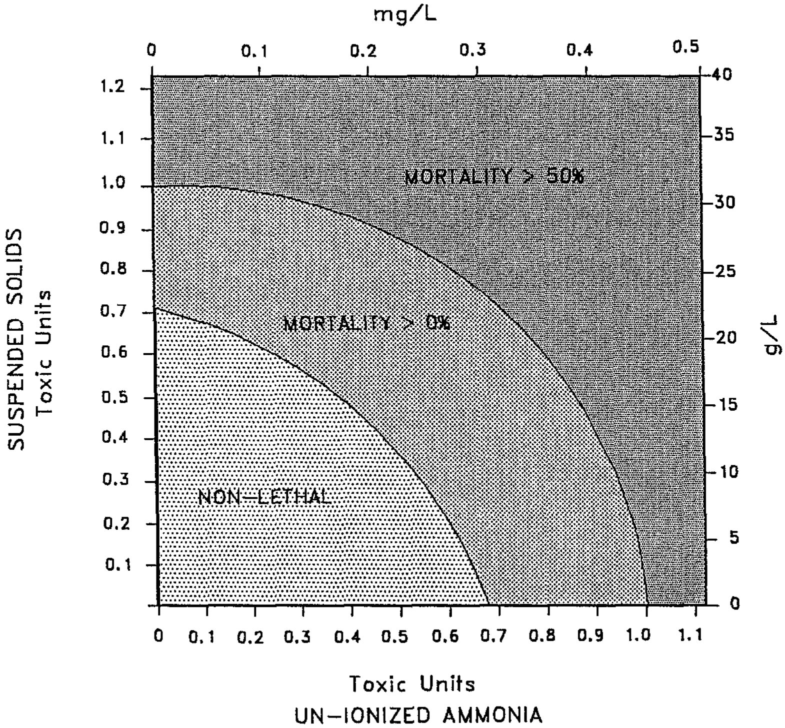

Figure 2. Acute toxicity zones for juvenile chinook salmon exposed to mixtures of ammonia and Fraser River suspended sediment for 96h at 5 to 8°C.

A feature of the toxic unit model is its potential to broaden application of toxicity data. Figure 2 provides such an opportunity when LC50 data are available, or may be estimated, for fish species and sediments other than juvenile chinook and Fraser River suspended sediment, respectively. Although based on juvenile Chinook salmon, Figure 2 could be used to estimate toxicity for other species assuming the mechanisms of toxicity for ammonia and suspended sediments are similar for juvenile chinook and the species in question. Application of Figure 2 to suspended solids besides those of Fraser River origin should be based on comparison of the sediments with those from the Fraser River. Fraser River suspended sediments are described as to particle size, shape (including photomicrographs) and mineralogy (Servizi and Martens 1987). This reference also includes measurements of 96-h LC50 for four particle size ranges.

Acknowledgments. The authors gratefully acknowledge the efforts of Jeremy Hume and Vivian Ogilvie who produced the figures.

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Received July 23, 1989; accepted October 21, 1989.