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Acute Toxicity of Roundup® and Rodeo® Herbicides to Rainbow Trout, Chinook, and Coho Salmon

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Rodeo ® and Roundup ® herbicides are manufactured by Monsanto Company. This paper summarizes the results of aquatic toxicity testing of these two herbicides using three different fish species, different sized fish, and differences in dilution water (type/source, pH, hardness and conductivity).

MATERIALS AND METHODS

Acute lethality (96-h LC50) testing was conducted in accordance with procedures outlined by APHA (1985) for static acute bioassays. Rainbow trout (*Salmo gairdneri*), chinook salmon (*Oncorhynchus tshawytscha*) and coho salmon (*O. kisutch*) were obtained from commercial hatcheries and acclimated to laboratory conditions for a minimum period of 5d prior to bioassay initiation. Fish were fed Oregon Moist trout pellets *ad libitum*. Feeding was discontinued 24-h prior to and during testing.

Water used in the bioassays consisted of either charcoal dechlorinated municipal water (total chlorine less than 0.01 mg/L), dechlorinated water reconstituted with $NaHCO_3$, $CaCl_2$, $MgSO_4$ and KCl (U.S. EPA 1985), or natural lake water obtained from the International Pacific Salmon Fisheries Commission Laboratory, Cultis Lake, B.C., Canada.

Tests were conducted with standard commercial formulations of Roundup and Rodeo herbicides. Roundup

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was directly diluted to obtain the desired exposure concentrations. Rodeo was mixed with Ortho X-77™ surfactant prior to testing at the maximum-use dilution recommended by Monsanto. Specifically, a stock solution was prepared with 312 mL Rodeo, 699 mL water and 4 mL X-77 surfactant. Aliquots of this Rodeo/X-77 stock solution were volumetrically diluted for testing. Duplicate water samples were taken from selected Roundup test concentrations, frozen in polyethylene bottles, and used for active ingredient (glyphosate) analysis by HPLC to confirm the bioassay test concentrations.

Ten fish (weights provided in Tables 1 and 2) were exposed per test concentration. Rainbow trout testing was conducted in glass aquaria; testing of coho and chinook salmon was conducted in fiberglass tanks. Fish loading densities were maintained below 1.0 g/L.

Testing was conducted at a temperature of 11 ± 1°C and a 16 h light:8 h dark photoperiod regime. Dissolved oxygen levels were maintained above 60% saturation at all times with minimal aeration (<7.5 cc/min/L).

Median lethal values (96-h LC50s) and 95% confidence limits were calculated from survival data using probit, moving-average or binomial probability analysis, and were confirmed by graphical log-probit analysis. Acute lethality values are presented in terms of the total herbicide formulation and in terms of the isopropylamine (IPA) salt of glyphosate.

RESULTS AND DISCUSSION

Acute toxicity values derived in this and in previous studies with Roundup herbicide are presented in Table 1. Previous studies have presented such data in terms of either the total formulation (Hildebrand et al. 1982) or the isopropylamine (IPA) salt of glyphosate (Folmar et al. 1979). In order to allow data comparisons, both methods of data presentation are used herein.

There was no significant difference ($P<0.05$) in the toxicity of Roundup herbicide to 0.37 g rainbow trout, 4.6 g chinook salmon and 11.8 g coho salmon. Acute toxicity values ranged from 7.4 mg/L to 12 mg/L in

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Table 1. Roundup herbicide acute toxicity

Species	Mean fish weight (g)	Dilution Water type[b]	pH	hardness (mg/L)	conductivity (umhos/cm)	Roundup 96-h LC50[a] (95% confidence limits) Total formulation (mg/L)	IPA salt of glyphosate (mg/L)	Data source
Rainbow trout	0.37	dechl.	6.1	4.5	12	26 (12-38)	12* (5.7-18)	This study
"	0.37	recons.	7.6	85	132	22 (12-38)	11 (5.7-18)	This study
"	0.37	lake	7.7	81	132	15 (12-21)	7.4 (5.7-10)	This study
"	1.4	dechl.	6.2	n.d.	14	54.8 (50-60)	n.d.	Hildebrand et al. (1982)
"	1.6	stream	6.7	n.d.	190	52.0 (n.d.)	n.d.	Hildebrand et al. (1982)
"	2.0	recons.	7.2	40	n.d.	n.d.	8.3 (7.0-9.9)	Folmar et al. (1979)
Chinook Salmon	4.6	dechl.	6.1	4.5	12	20 (17-27)	9.6* (7.9-13)	This study
Coho Salmon	11.8	dechl.	6.2	4.5	12	22 (12-38)	11* (5.7-18)	This study

[a] Acute toxicity values given in terms of the total formulation and/or the isopropylamine (IPA) salt of the active ingredient glyphosate; * indicates test concentrations verified by glyphosate analysis; n.d. = not determined.

[b] Dilution water type: dechl. = dechlorinated municipal water; recons. = water reconstituted with hardness and pH buffering salts; lake = Cultis Lake, B.C.; stream = UBC Research Forest stream, B.C.

terms of the IPA salt of glyphosate, with 95% confidence limits ranging from 5.7 - 18 mg/L. Varying dilution water type (dechlorinated, dechlorinated and reconstituted, and natural lake water), pH (range 6.1-7.7), hardness (4.5 - 85 mg/L as CaCO3) and conductivity (12 -132 umhos/cm) did not significantly affect Roundup toxicity to 0.37 g rainbow trout.

The Roundup herbicide 96-h LC50 value of 8.3 mg/L previously determined by Folmar et al. (1979) for 2.0 g rainbow trout tested at 12°C, pH 7.2 and 40 mg/L hardness was not significantly different ($P<0.05$) from the present values (Table 1). However, in contrast to the present study, Folmar et al (1979) also presented data showing a significant increase ($P<0.05$) in Roundup toxicity when testing was done at pH 6.5 compared with pH 7.5 (96-h LC50 values, in terms of the IPA salt of glyphosate, decreased from 7.6 mg/L to 1.6 mg/L). Folmar et al. (1979) reported a similarly large difference in Roundup toxicity of 8.3 mg/L versus 1.3 mg/L for tests with rainbow trout weighing 2.0 g compared with trout weighing 1.0 g. Large differences in toxicity such as those determined by Folmar et al. (1979) are unexpected given the small changes in test pH and fish size and in particular are not supported by the values obtained in the present study.

Hildebrand et al. (1982) found that differing water type (dechlorinated versus natural stream) and conductivity (14 versus 190 umhos/cm) did not affect Roundup toxicity. However, the acute 96-h LC50 values obtained by Hildebrand et al. (1982) were over twice as great as those determined in the present study (Table 1). The reason for this difference in toxicity is unknown, but could be due to differences in bioassay testing methodology between the two studies.

Test concentrations used in the present study with Roundup were confirmed by glyphosate analysis of the bioassay tank solutions. These analyses indicated that after 24 h of testing, 70 - 87% of the original target levels remained in the chinook and coho salmon test tanks. Levels in the rainbow trout tanks were 21 - 63% of the target levels after 24 h. These differences may be largely due to higher rates of herbicide adsorption onto the glass aquaria utilized in the rainbow trout bioassay, than onto the fiberglass tanks used in the chinook and coho salmon tests. The fact that these differences are not reflected in significant differences ($P<0.05$) between the acute toxicity values of these species may indicate that the majority of the lethal herbicide dose was absorbed by the test fish prior to loss of the test material from solution.

No significant differences ($P<0.05$) were found between the Rodeo/X-77 acute toxicity values for rainbow trout, chinook salmon, and coho salmon (Table 2). The 96-h LC50 values ranged from 120 mg/L to 290 mg/L in terms of the IPA salt of glyphosate, with 95% confidence limits ranging from 68 - 380 mg/L. There was some indication that the use of waters with relatively high pH, hardness and conductivity values resulted in a decrease in Rodeo/X-77 toxicity (maximum values in the present study were pH 7.8, 77 mg/L and 130 umhos/cm, respectively). However, these toxicity differences were not significant (Table 2). Fish size also did not significantly ($P<0.05$) affect Rodeo/X-77 toxicity; test fish mean size ranged from 0.21 g for the rainbow trout to 17.9 g for the coho salmon.

Comparison of Roundup and Rodeo/X-77 acute toxicities indicates that the 96-h LC50 values, in terms of the IPA salt of glyphosate, differ by approximately 10-fold (Tables 1 and 2). This difference may be due to the difference in the type of surfactants present in these two formulations. Testing of Rodeo without prior formulation with its X-77 surfactant significantly ($P<0.05$) decreased toxicity from 130 mg/L to 580 mg/L, in terms of the IPA salt of glyphosate (Table 3). Tests of the surfactant in Roundup herbicide and of technical glyphosate [N - (phosphonomethyl) glycine] conducted by Folmar et al. (1979) also suggested that the surfactant was the primary toxic agent in the Roundup formulation. The 96-h LC50 values obtained by Folmar et al. (1979) were 2.0 mg/L for the surfactant and 140 mg/L for technical glyphosate.

While materials used to formulate Roundup and Rodeo herbicides are more toxic than the active ingredient itself, the representation of the acute toxicity of these two herbicides in terms of the IPA salt of glyphosate can be useful. Trace levels of glyphosate are quantifiable and from such data the total herbicide concentrations can be readily obtained by back-calculation.

The results of the present studies indicate that, according to a toxicity classification scheme currently in use (U.S. Fish and Wildlife Service 1984), Roundup and Rodeo herbicides would be considered to be slightly toxic and practically non-toxic, respectively, to trout and salmon species. No acute toxicity hazard to aquatic environments would be expected during the course of normal usage.

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Table 2. Rodeo /X-77 acute toxicity

Species	Mean fish weight (g)	Dilution Water type[b]	pH	hardness (mg/L)	conductivity (umhos/cm)	Rodeo /X-77 96-h LC50[a] (95% confidence limits) Total formulation (mg/L)	IPA salt of glyphosate (mg/L)
Rainbow trout	0.52	dechl.	6.0	5.0	15	680 (600-820)	130 (120-160)
"	0.21	dechl., recons.	7.8	75.	130	1070 (600-1920)	210 (120-380)
Chinook salmon	4.2	dechl.	5.8	5.0	15	750 (600-1100)	140 (120-220)
"	5.9	dechl., recons.	7.4	77.	130	1440 (1070-1920)	290 (210-380)
Coho salmon	17.9	dechl.	5.8	5.0	15	1000 (600-1900)	200 (120-370)
"	11.8	dechl.	6.2	4.5	12	600 (340-1100)	120 (68-220)

a Acute toxicity values given in terms of the total formulation and the isopropylamine (IPA) salt of the active ingredient glyphosate. Rodeo /X-77 consists of 312 mL Rodeo mixed with 699 mL water and 4 mL X-77 surfactant.

b Dilution water type: dechl. = dechlorinated municipal water; recons. = water reconstituted with buffering salts.

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Table 3. Acute toxicity of Rodeo and Rodoe X/-77 to rainbow trout[a]

Herbicide formulation	96-h LC50[b] (95% confidence limits) Total formulation (mg/L)	IPA salt of glyphosate (mg/L)
Rodeo	1100 (850-1300)	580 (460-730)
Rodeo /X-77[c]	680 (600-820)	130 (120-160)

[a] Comparison of acute toxicity values obtained for Rodeo herbicide alone, and for Rodeo herbicide with X-77 surfactant. Tests conducted with 0.52 g fish in dechlorinated municipal water (pH 6.0, hardness 5.0 mg/L, conductivity 15 umhos/cm).

[b] Acute toxicity values given in terms of the total formulation and the isopropylamine (IPA) salt of the active ingredient glyphosate.

[c] Rodeo /X-77 consists of 312 mL Rodeo mixed with 699 mL water and 4 mL X-77 surfactant.

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