Effects of Roundup® Herbicide on Diatom Populations in the Aquatic Environment of a Coastal Forest

Druscilla S. Sullivan¹, Thomas P. Sullivan¹ and Thana Bisalputra²

¹Northwest Ecological Animal Research Ltd., #1205-2233 Allison Road, Vancouver, B.C., V6T 1K9, Canada; ²Department of Botany, University of British Columbia, Vancouver, B.C. V6T 1W5, Canada

Roundup herbicide³ is a non-selective postemergence herbicide. It can be used to control perennial weeds in vegetable crops, orchards, and industrial areas and in conifer plantations and reforestation areas. As with any pesticide application, consideration must be given to the possibility of contamination of nontarget environments such as streams and ponds.

Similarities between the physiology of higher plants, such as those directly affected by Roundup herbicide, and phytoplankton in the aquatic environment make the latter an obvious place to study the potential effects of Roundup on nontarget primary producers. Concern over the presence of unnatural chemicals and other environmental alterations to streams and ponds is based on the results of many ecological studies describing food chains. From this concept it is obvious that changes in any one trophic level will affect all others. Phytoplankton in streams and ponds form an integral part of the aquatic food web providing food for larger organisms such as zooplankton and ultimately fish.

The purpose of this study was to determine the effects of Roundup herbicide on algae (diatoms) in two streams and one pond under two different experimental conditions.

METHODS AND MATERIALS

This research was conducted at the University of British Columbia Research Forest, Maple Ridge, B.C.

Experiment A - Aerial Application of Roundup herbicide

The experimental study area was a small, rocky stream running through a 20-year old Douglas fir plantation containing a variety of deciduous growth such as alder (Alnus rubra), salmonberry (Rubus spectabilis), and vine maple (Acer circinatum). The stream flows all year, though portions of it may be subterranean during drier seasons, and leads to the small experimental pool (about 2 m in diameter). The benthic sediment of this pool is approximately 5 cm deep and rests on a bed of small rocks and pebbles. The pond is just outside the edge of the plantation and therefore has a reduced coniferous canopy. This resulted in a direct application of Roundup herbicide to the pond at the time of spraying.

a commercial formulation by Monsanto containing 356 g/L glyphosate as isopropylamine salt

R Trademark of Monsanto Company

The control study area was logged in the fall of 1973 and planted with Douglas fir in the spring of 1975. The steam runs through the center of this area and feeds a pool of standing water approximately 3 m in diameter. The bottom of the pond is composed of a thick layer (greater than 0.5 m) of sediment. There is a heavy cover of deciduous shrubs and trees along both sides of the stream.

The experimental area (4.9 ha) was sprayed with Roundup herbicide at a rate of 2.2 kg a.i./ha (2 lb/acre) on Sept. 12, 1979. A spray volume of 90 L/ha was produced from a Simplex spray boom mounted on an Okanagan helicopter. The weather was sunny and warm (22°C). The foliage was dry at application time 1130 to 1200 h) and there was no senescence of plant tissues observed at this date. The first recorded rainfall occurred 14 days after spraying.

Sediment samples and two glass slides (placed in the water on Aug. 30) were removed from the experimental and control streams and ponds immediately preceding the spraying. Slides were held in plastic boxes with no top and all but a 0.3 cm rim of the bottom removed. Sample slides were replaced with new clean slides. Sediment samples were taken at 1600 h on Sept. 13 and sediment and slide samples were taken on Sept. 17, Oct. 11 and 29.

Ten ml of sediment and associated water was collected from each of the control and experimental streams and ponds. These samples were fixed in 2% glutaraldehyde with the natural water acting as buffer. At the time of counting, the sediment samples were agitated and a single drop removed and placed on a Hemacytometer. Two such samples were analysed. Identification of the diatom genera was determined using PRESCOTT's key (1972) in conjunction with STEIN's (1975) work in the Lower Fraser Valley, B.C.

Slide samples were allowed to air-dry in a dust free environment. Twelve subsamples were uniformly distributed as follows: four (1.5 cm apart) per each of 3 rows (1.5 cm apart). All subsamples were at least 0.25 cm from the edge of the slide, and were photographed at 63x and then processed into 4 x 5 prints. On these prints, genera were identified and counted for each slide.

Duplicate slide and sediment samples were then enumerated. A 2-way analysis of variance and Duncan's multiple range test were used to analyse the effects of time and treatment on dominant genera that were consistently present in the samples.

Experiment B - Manual Application of Roundup herbicide

A third stream (3-5 m in width) with a mainly solid rock base was chosen for this experiment. Deciduous shrubs and trees covered both edges of the stream. This study stream was divided into three regions: upstream was the control, midstream was misted with a manually operated spray boom at the field dose (2.2 kg/ha) of Roundup herbicide, while further downstream was sprayed manually with 10x field dose. Each region had an aluminum slide tray containing four upright slides (placed in the water on Aug. 10), upstream from, and braced against a rock. Two slides from each area were removed and replaced just prior to spraying on Sept. 12. Two more slides were removed on Sept. 17. The two slides that were replaced on Sept. 12, were removed on Oct. 11.

Preparation of samples, enumeration of genera, and statistical analysis were conducted in the same manner as described for experiment A.

RESULTS AND DISCUSSION

Experiment A - Aerial Application of Roundup herbicide

Pond sediments

Figure 1 illustrates the data for the dominant diatoms. genera with significant differences over time or between experimental and control areas will be discussed. Low densities of stream sediment diatoms precluded analysis.

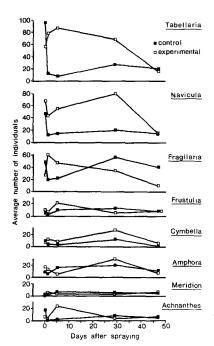


Fig. 1. Effects of an aerial application of Roundup herbin the sediments of a forest pond. Data points represent the average number of individuals per 0.1 ml sedimentwater mixture.

Surirella, Enotia and Diatoma.

Tabellaria numbers were not significantly different over time or between the experimental and control areas. However, overall mean values of control (32.8) versus experimental (59.8) areas suggest a biologically significant increase in the number of Tabellaria individuals on the treated area. Differences in the overall mean values for the 5 sampling periods (75.8, 41.3, 47.8, 48.5 and 18.2) suggest a biologically significant decline in total diatom numbers over time.

Significant differences were found between Navicula abundance in the control versus experimental areas (p < 0.01). The overall mean for the experimental (51.6) was more than double that of the control (21.6). The first and fourth sampling times have significantly more Navicula diatoms than the census on Oct. 29 (day 47) (p=0.03).

Cymbella also showed a significantly higher density in the experimental area when compared with the control (p=0.02). The abunicide on dominant diatom genera dance of this diatom was significantly greater (p=0.02), at Oct. 11 compared to all other sampling times.

Other genera found in the sediments in very low or inconsistent numbers were: Gomphonema, Cocconeis, Ceratoneis, Peronia, Synedra, Any variation in algal densities appears to be due to habitat or seasonal variation rather than due to the treatment itself. In general, the experimental area supported a greater density of diatoms than the control.

Pond slides

The colonization of slides by diatoms in the streams was

extremely low, such that the analysis was restricted to pond slide data. The dominant genera are presented in Figure 2. Although no significant difference was found between the control and treatment areas, there was a significant change in <u>Achnanthes</u> numbers over time (analysis of variance p=0.02). Comparison of means showed the Oct. 11 density to be significantly higher than either Sept. 17 or Oct. 29 (days 5 and 47). Figure 2 indicates that this is mainly a result of a peak in <u>Achnanthes</u> abundance in the experimental pond at this time.

There was no significant differences in treatment or over time for <u>Meridion</u>. However, overall mean values for the four sampling times (9.9, 4.7, 4.0, and 0.9) indicate a significant decline by Oct. 29 (day 47).

Tabellaria showed a significant initial increase and then decrease over time (p=0.04). The density of <u>Tabellaria</u> was significantly (p=0.01) higher on the control area when compared with the experimental.

Gomphonema showed no significant differences over time but had a significantly greater density on the control versus the experimental area (p=0.02).

Other genera identified but inconsistently present were: Cocconeis, Cymbella, Navicula, Pinnularia and Amphora.

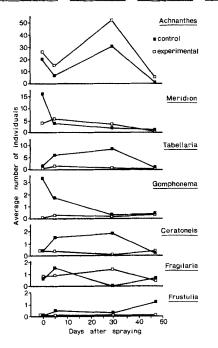


Fig. 2. Effect of an aerial application of Roundup herbicide on diatom genera colonizing glass slides in the forest pond. Data points represent the average number of individuals found in each of 24 subsamples.

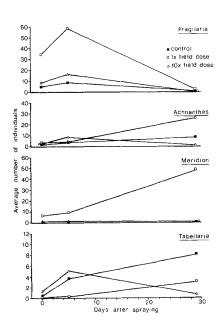


Fig. 3. Effect of a manual application of Roundup herbicide (1x and 10x field doses) on diatom genera colonizing glass slides in a forest stream. Each data point represents the average number of individuals in each of 24 subsamples.

This study stream represents a considerably different habitat compared with the streams in Experiment A. However, similar genera of diatoms were encountered (see Figure 3).

<u>Fragilaria</u> and <u>Meridion</u> densities were found to be significantly greater at the field dose treatment (p=0.01). However, pre-treatment (Sept. 12) numbers indicate that this is a result of environmental differences at each of the three study areas. <u>Fragilaria</u> numbers increased by Sept. 17 (day 5) and then showed a significant decline by Oct. 11 (day 29) (p= 0.01).

Other genera observed inconsistently were: $\underline{\text{Navicula}}$, $\underline{\text{Gomphonema}}$, Cymbella, Ceratoneis, and Cocconeis.

Results from the colonization of glass slides in Experiments A and B suggest that the explanation for differences in algal density between the control and experimental pond is a result of habitat and seasonal variation rather than treatment effects. This is also verified if one takes into account the differences in colonizing density before spraying as seen on Sept. 12 (day 0) for both ponds (Figure 2) and the manually sprayed experimental stream (Figure 3). BLUM (1960) states that only large old aquatic environments can be considered so stable that they will not undergo week to week composition changes.

While laboratory studies may determine conclusive effects of pesticides on unicultures of algae, such results may not represent the effects of such a pesticide in the field. Synergistic effects between the environment and the pesticide and interspecific responses to disturbance cannot be predicted from laboratory studies alone. According to LIVINGSTON (1977), there is almost no evidence concerning the activity of a given pesticide at the population or community level. The purpose of this study was to gain such evidence for an important basic portion of the aquatic food web.

Diatoms appeared to be the most ubiquitous algae in the study areas and have been used as indicators in other pesticide studies. A larvicide (Dursban) was shown to totally prevent the growth of fresh-water diatoms (BROWN et al. 1976) while an insecticide (DDT) is apparently detoxified by diatoms (MIYAZAKI and THORSTEINSON 1972). Achnanthes lanceolata (Breb) is considered to be a fairly resistant diatom to certain poisons and sewage pollutants (SCHROEDER 1939). Cocconeis placentula (Ehr.) is somewhat sensitive to pollutants (BLUM 1960). However, this genus was not consistently present in the ponds and streams in our study.

More interesting effects of Roundup herbicide on the aquatic community may occur in the future due to delayed, indirect, effects of the herbicide on the surrounding environment. Changes in algal species composition may result from changes in stream temperature, chemistry, illumination and precipitation protection that are likely to occur in a defoliated environment (WHITFORD 1960, McINTIRE 1968, LIKENS et al. 1970, HANSMANN and PHINNEY 1973). The chemistry of the stream may change due to a lack of new vegetative growth taking up nutrients from the soil. It has been shown that glyphosate enhances cellulase activity (ABU-IRMAILEH et al. 1979) and this may explain the stimulated cellulose decay observed when glyphosate is applied to soil or substrate (GOSSBARD and WINGFIELD 1978). This

increased cellulose break-down may result in a pulse of carbon to the aquatic environment via leaching of the degraded products into the streams.

The work of SPRANKLE et al. (1975a and b) and RUEPPEL et al. (1977) suggest very little glyphosate or its break-down products will be leached into run-off leading to streams and ponds. The chance of glyphosate occurring in run-off is also decreased by the method of application which requires that spraying is followed by at least six hours of dry weather. GOTTRUP et al. (1976) found that the type of leaf and its relative size determines the absorption rate and therefore the sensitivity of a plant to glyphosate. In their study, the larger-leaved Canadian thistle was more sensitive to glyphosate and therefore possibly absorbs more glyphosate than a leafy spurge. Thus, the microscopic sizes of diatoms may slow down their absorption of glyphosate to an even greater degree and thus increase the chances of it being degraded and absorbed by some other means.

It is concluded from this study that variations in abundance of diatoms observed in the ponds and streams were mainly determined by habitat and seasonal factors. The variation in algae in the pond water might be attributed to the aerial application of Roundup herbicide. However, the inconsistency of the results from the other techniques used makes it much more probable that density changes were due to environmental causes.

Acknowledgments We thank I.W. Taylor I. Walters and his staff and

Acknowledgments. We thank L.W. Taylor, J. Walters and his staff and are grateful for a NSERC grant to T. Bisalputra.

REFERENCES

ABU-IRMAILEH, B.E., L.S. JORDAN and J. KUMAMOTO: Weed Sci. 27, 103 (1979).

BLUM, J.L.: The ecology of algae, C.A. Tryon and R.T. Hartman, eds. Edwards Brothers, Inc. Ann Arbor, Michigan. (1960).

BROWN, J.R., L.Y. CHOW and C.B. DENG: Bull Environ. Contam. Toxicol. 15, 437 (1976).

GOTTRUP, O., P.A., O'SULLIVAN, R.J. SCHRAA and W.H. VANDEN BORN: Weed Res. 16, 197 (1976).

GOSSBARD, E. and G.I. WING-FIELD: Weed Res. 18, 347 (1978).

HANSMANN, E.W. and H.K. PHINNEY: Ecology 54, 194 (1973).

LIKENS, G.E., F.H. BORMAN, N.M. JOHNSON, D.W. FISHER and R.S. PIERCE: Ecol. Monogr. 40, 23 (1970).

LIVINGSTON, R.J.: CRC Critical Rev. Environ. Cont. 7, 325 (1977). McINTIRE, C.D.: Ecology 49, 520 (1968).

MIYAZAKI, S. and A.J. THORSTEINSON: Bull. Environ. Contam. Toxicol. 8, 81 (1972).

PRESCOTT, G.W.: How to know the freshwater algae. Wm. C. Brown Co. publishers, Bubuque, Iowa 1961.

RUEPPEL, M.L., B.B. BRIGHTWELL, J. SCHAEFER and J.T. MARVEL: J. Agric. Food Chem. 25, 517 (1977).

SCHROEDER, H.: R. Kolkwitz, Pflanzen forschung 21, vi, 88 pp. (1939).

SPRANKLE, P., W.F. MEGGITT and D. PENNER: Weed Sci. 23, 224 (1975a). SPRANKLE, P., W.F. MEGGITT and D. PENNER: Weed Sci. 23, 229 (1975b).

STEIN, J.R.: Syesis 8, 119 (1975).

WHITFORD, L.A.: The ecology of algae. C.A. Tryon and R.T. Hartman, eds. Edwards Brothers, Inc. Ann Arbor, Michigan 1960.