1982

Effect of Herbicides on Performance of Aerobic Dairy Lagoons

John D. Bottom
University of Kentucky

Joseph L. Taraba
University of Kentucky, joseph.taraba@uky.edu

Effect of Herbicides on Performance of Aerobic Dairy Lagoons

John D. Bottom, Joseph L. Taraba
ASSOC. MEMBER
ASAE

ABSTRACT

THE herbicides glyphosate, bromacil, and dalapon were assessed for their affect on the oxygen uptake of microorganisms from an aerated dairy lagoon at 20 °C. The maximum recommended addition of herbicide active ingerdient to a lagoon is: glyphosate (isopropyl amino salt)—0.42 mg/L; bromacil—3.70 mg/L; and dalapon (85.8 percent Na salt and 14.2 percent Mg salt)—1.95 mg/L. In addition, microorganism isolates from the aerobic dairy lagoon were incubated under aerobic and anerobic conditions at room temperature to assess the effects of the herbicides. Under aerobic incubation, it was found that neither herbicide type nor herbicide concentration affected isolate growth. Under anaerobic incubation, both inhibition and stimulation effects were found with gram-positive rod organisms being least affected.

INTRODUCTION

As the farmer increases the use of herbicides, the effect thay have on the environment must be examined. The effect these chemicals have on the microorganisms in a system, such as a diary lagoon, is an important consideration in assessing their use and disposal characteristics.

REVIEW OF LITERATURE

Several researchers have considered the effect of microbial action on various herbicides. Jensen (1957, 1959) found that soil fungi and a species of *Agrobacterium,* as well as certain pseudomonads, would decompose the herbicide dalapon in soil. Hirsch (1960) and Hirsch and Stillmach-Hellwig (1961) found that *Pseudomonas dehalogenans* removed the chlorine from dalapon and metabolized the remaining residues. Along with bacteria, they observed that fungal growth can decompose herbicides and that *Nocardia sp.* would dechlorinate dalapon. Torgenson and Mee (1967) found *Penicillium paraherquei* Abe. active in degrading bromacil. Terbacil (closely related to bromacil) was also found to be degradable by *Penicillium paraherquei* Abe. (Torgeson, 1971).

Microbial growth can be stimulated by specific herbicides. Van Schreven et al. (1970) reported that the herbicides ioxynil, dalapon, mecoprop, dichlorprop, MCPA + dichlorprop, picloram, and amitrole—T increased microbial growth. In another study, Fletcher (1960) observed that dalapon had a broad stimulating effect on soil microflora. In work completed by Magee and Colmer (1959), many bacteria, actinomycetes and molds were found to grow readily in the presence of high concentrations of dalapon. Torgeson (1971) reported on bacteria grown under anaerobic conditions and found that gram-positive cocci and gram-positive rods were not affected by the herbicide DCPA, while gram-negative rods did not grow. In lake water treated with the herbicides diuron, dichlobenil and diquat, a stimulating effect on bacterial growth was observed by Yazar et al. (1975). The bacterial populations in the treated samples indicated an extended lag period and a higher population decrease than the control samples. After the initial adaptation period, bacterial populations of the treated samples exceeded the control samples by approximately 70 percent.

Other factors can stimulate microbial growth. Holstun and Loomis (1956) found that microbial growth in the soil evironment is related to soil moisture, pH level, addition of organic matter and temperature. According to Norman and Newman (1950), the addition of organic matter accelerated the disappearance of herbicides from soil. As the metabolic activity of the microbial population is stimulated, the length of persistence for most herbicides is decreased rapidly by increased microbial degradation.

The literature does not address the effects of herbicides on the microorganisms found in animal manure treatment lagoons or the effect on the dynamics of the waste lagoon. Two situations can be posed for the entrance of herbicides into dairy lagoons: first, the washing-off of applied herbicide, used to rid the inside embankment of the lagoon of tall weeds, into the lagoon during rainfall events; and second, the use of the lagoon as the disposal site of unused herbicides or for the wash water from cleaning equipment or containers.

The objective of the experiments, reported here, was to assess the effect of herbicides with three different persistence levels and five concentrations on microorganisms from a mechanically aerated dairy lagoon by considering: (a) aerobic degradation using lagoon water as an inoculum, and (b) the growth of isolated microorganisms from the lagoon incubated under aerobic and anaerobic conditions.

METHODS AND MATERIALS

The procedures utilized in this rsearch were not anticipated to simulate lagoons, but were only a first order assessment of the effects of herbicides on the microbial environment of a dairy lagoon to see if these effects are adverse.

Article was submitted for publication in April 1981; reviewed and approved for publication by the Structures and Environment Division of ASAE in September 1981. Presented as ASAE Paper No. SER-81-003.

Common names for herbicides have been used in the paper. Specific information can be found in WSSA, 1979.

This paper is published with the approval of the Director of the Kentucky Experiment Station and designated Paper No. 81-2-57.

The authors are: JOHN D. BOTTOM, Agricultural Engineer, and JOSEPH L. TARABA, Assistant Extension Professor, Agricultural Engineering Dept., University of Kentucky, Lexington.

  0001-2351/82/2504-1026$02.00

The herbicides dalapon, glyphosate, and bromacil* were used in this study. These can be considered to have short-, medium- and long-term persistence levels, respectively. The active ingredient concentration levels used were 0.01, 0.05, 0.1, 1, and 10 times a base concentration. The base concentration of the herbicide in the lagoon water was calculated from a field situation in which it was assumed that all the herbicide applied to the inside embankment of an aerated dairy lagoon (volume equal to 353,796 L) was washed into the lagoon. The herbicide was assumed to be applied at recommended levels. The base concentration levels of active ingredient were 1.95 mg/L for dalapon (85.8 percent Na salt and 14.2 percent Mg salt), 0.42 mg/L for the isopropylamine salt of glyphosate, and 3.70 mg/L for bromacil.

The effect of the herbicides on the aerobic activity of microorganisms was assessed using a modified 5-day biological oxygen demand (BOD) method to measure oxygen uptake. The inoculum, used as seed, was obtained from a mechanically aerated lagoon receiving manure from diary heifers located on the University of Kentucky experimental dairy farm. The inoculum was added in a concentration equal to 2 mL lagoon water per liter of dilution water. The lagoon water, used as the seed, was obtained within a few hours before initiation of the experiment and stored at 5 °C until used. A sufficient amount of simulated organic waste (150 mg glucose and 150 mg glutamic acid per liter or 312 mg BOD/L) was added to obtain a dissolved oxygen reduction of approximately 50 percent for the control after 5 days at 20 °C in the BOD bottle. It was found that 14 mL/L of dilution water was needed. The experiment was then run in triplicate with a control containing no herbicide. The dissolved oxygen reduction data was determined at the end of 5 days and was used to assess the effect of the herbicide treatment on the aerobic activity of the lagoon microorganisms. An analysis of variance (ANOVA) for a 5 × 3 factorial experiment was used to determine the significance of the treatments.

The effect of the herbicides on specific microorganisms isolated from the aerobic dairy waste lagoon was followed under aerobic and anaerobic conditions. Forty microorganisms were isolated from the lagoon on trypticase agar (TSA) pour plates. The isolates were purified and the type and the gram stain of each was determined.

TSA streak plates were used to assess the effect of each herbicide and herbicide concentration treatment on the bacterial isolates. The agar was sterilized and tempered to 50 °C. The herbicides were filter sterilized and aseptically added to the tempered agar to give the required final herbicide concentration. The agar was then poured into petri plates and cooled. If no microbial growth was found on the plates after 24 hours, the plates were considered free from contamination. Each isolate was streaked on duplicate plates for each herbicide-concentration treatment plus a control with no herbicide. One series of the duplicates was incubated aerobically and another was incubated anaerobically in an anaerobic glove box. Both series of plates were incubated at room temperature (~20 °C). At 24, 72, and 168 hours each of the plates was read to determine if there was growth or no growth for each isolate for all of the herbicide and

*Herbicides manufactured by Dow Chemical, Monsanto, and Du-Pont, respectively.

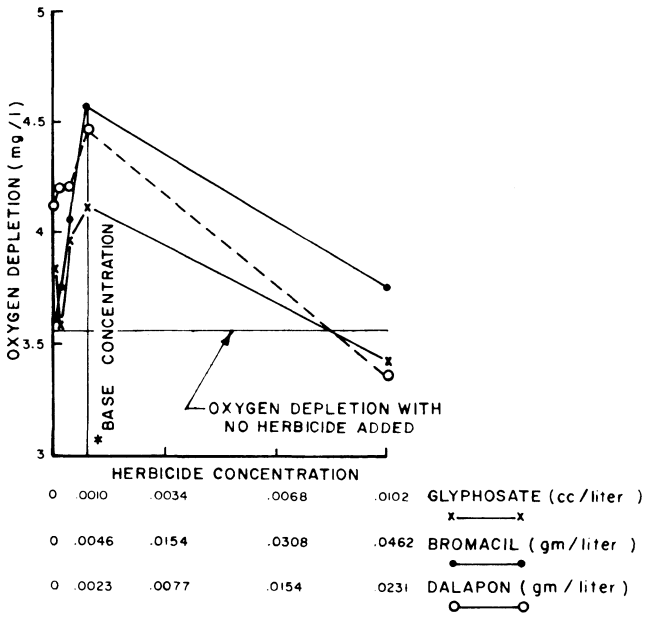

FIG. 1 Oxygen depletion with and without herbicide added using dairy lagoon water as inoculum.
***Base Concentration = Maximum recommended concentration of herbicide.**

concentration treatments. The anaerobic plates were always kept in an anaerobic glove box during incubation.

RESULTS AND DISCUSSION

The oxygen depletion after 5 days at 20 °C in the BOD bottles for all treatments is shown in Fig. 1. The ANOVA for this experiment indicates that the herbicide and the concentration treatments were significantly different at the 99 percent confidence level. It was also found that the interaction between the herbicide species and herbicide concentration at the 99 percent confidence level had affected the oxygen depletion significantly. The oxygen depletion for herbicides is greater than the control and increases as the concentration of the herbicides increases up to the base concentration. At ten times the base concentration, the oxygen depletion associated with each herbicide decreases with only bromacil remaining above the control oxygen depletion. The herbicide that is added does contribute to oxygen depletion if metabolized by the bacteria. The estimated BOD for each herbicide, as sold, are 0.566 m O_2/mg dalapon, 0.956 mg O_2/mg bromacil, and 341 mg O_2/mL glyphosate. The medium persistence herbicide, glyphosate, yielded oxygen depletion above the estimated BOD depletion for all concentrations except at ten times the base concentration. The long-persistence herbicide, bramacil, and the short-persistence herbicide, dalapon, show a higher than estimated oxygen depletion only at the concentration of 0.01 times the base concentration, but are always below the estimated oxygen depletion at all the other concentrations. Based on the concentrations of herbicides tested, the maximum loading of the active ingredient of the herbicides that an aerobic dairy lagoon can handle without showing inhibition of oxygen uptake after 5 days is: glyphosate (isopropylamine salt)—0.42 mg/L; bromacil—3.70 mg/L; and dalapon (85.8 percent Na salt and 14.2 percent Mg salt)—1.95 mg/L. Since the data show such a large degree of difference in the affects that the herbicides have on the oxygen depletion between the base concentration and ten times this value, it is sug-

TABLE 1. RESULTS OF ISOLATES GROWN UNDER ANAEROBIC CONDITIONS

Organism type	Gram$^+$ rod	Gram$^-$ rod	Gram$^+$ coccus
Total number streaked	17	16	7
Control positive, no growth by 168 hours on herbicide	3	4	0,3*
Control negative, positive growth by 168 hours on herbicide	1,1*	3,2*	2,0*
Organisms not stimulated or inhibited	12	5	2

*First number indicates that an isolate was observed more than two times. Second number indicates isolate observed only once.

gested that further studies be completed to assess the herbicides' effect in this intermediate range. These results are similar to those found by Van Schreven et al. (1970) and Yazar et al. (1975) who found a stimulation of bacterial growth was associated with the addition of a herbicide.

The 40 microorganisms isolated from the aerated dairy lagoon were found to be grouped as 17 gram-positive rods, 16 gram-negative rods, 7 gram-positive cocci, and no gram-negative coccus. These organisms were streaked onto TSA plates that were prepared with all the herbicide-concentration treatments. All 40 of the isolates subjected to aerobic incubation grew for all herbicide and concentration treatments. There were 38 isolates that indicated growth at 72 h and the other two indicated growth by 168 h. There was no indication of an effect of herbicide type or herbicide concentration on the growth results.

The isolates incubated under anaerobic conditions gave far different results. A summary of these results is shown in Table 1. Herbicide concentration did not indicate any positive or negative effect on whether an isolate was stimulated to grow or inhibited on the agar plates. As shown in Table 1, the gram-positive rods were the least affected by the presence of herbicide while the gram-negative rods and gram-positive cocci show equivalent proportions of isolates that show inhibited growth or stimulated growth in the presence of herbicides. These results are slightly different from those found by Torgeson (1971). There were nine isolates that did not indicate growth within 168 hours on the control plates but were found to show growth on plates that had a herbicide present. There was no particular preference of a herbicide to stimulate the growth and no indication that an organism type was favored. There were 10 isolates that indicated growth within 168 h on the control plates but did not indicate growth within 168 h on herbicide-treated plates. Two gram-positive rods did not show growth in the presence of any of the herbicides while the other eight isolates were inhibited by at least two of the herbicides. There was no specific indication of organism type or herbicide preference. As indicated by these results, further studies should be continued on the effect of herbicides on microbial isolates from anaerobic lagoons when incubated anaerobically.

CONCLUSIONS

The following conclusions were derived from oxygen depletion experiments and the dairy lagoon microorganism isolates tested with three different herbicides at five concentrations. The primary conclusion is that the herbicides tested can be added to an aerobic dairy lagoon without inhibiting the microbial activity if the final concentration does not exceed the base concentration of active ingredient: glyphosate (isopropylamine salt)—0.42 mg/L, bromacil—3.70 mg/L, and dalapon (85.5 percent Na salt and 14.2 percent Mg salt)—1.95 mg/L.

The other conclusions are:

1 Herbicide treatment of lagoon water with dalapon, glyphosate, and bromacil enhanced oxygen depletion, as compared to a control containing no herbicide.

2 Glyphosate and dalapon inhibit oxygen depletion at ten times the base concentration, whereas bromacil stimulates the oxygen depletion.

3 Lagoon water microorganism isolates grew without preference to the type of herbicide used or the concentrations of the herbicide under aerobic incubation at room temperature.

4 Lagoon water microorganism isolates showed no signs of preference toward specific herbicide or herbicide concentration under anaerobic incubation at room temperature.

5 Gram-positive rods incubated anaerobically are least affected by herbicides while gram-negative rods and gram-positive cocci show equivalent proportions of isolates that have inhibited growth or stimulated growth in the presence of herbicides.

References

1 Fletcher, W. W. 1960. The effect of herbicides on soil microorganisms. Herbicides and the Soils: pp. 20-62.

2 Hirsch, P. 1960. Die Entgiftung von unkrautbekampfungsmittln durch boden mikroorganismen. Kalibriefe. (Weed Abs. 11:547, 1962).

3 Hirsch, P. and R. Stellmach-Hellwig. 1961. Zur Frage des Abbaus von α- Dichloropropionsaure (Dalapon) und Trichloroessigsaure (TES) durch propionsaureverwendende Pansen und Bodenbakterien. Zentrallel. fur Bakt. Parasitenk., Infektionskr .u Hyg. 2:113:683-686.

4 Holstun, J. T. and W. E. Loomis. 1956. Leaching and decomposition of 2, 2- dichloropropionic acid in several Iowa soils. Weeds 4:205-217.

5 Jensen, H. L. 1957. Decomposition of chloro-organic acids by fungi. Nature 180:14-16.

6 Jensen, J. L. 1959. Biologisk sonderdeling of ukrudlsmidler i jordbunden. Fidsskr. Planteaul 63:470-499.

7 Magee, L. A. and A. R. Colmer. 1959. Decomposition of 2, 2-dichloropropionic acid by soil bacteria. Can. J. Microbiol. 5:255-260.

8 Norman, A. G. and A. S. Newman. 1950. The presistence of herbicides in soils. Proc. Northeastern Weed Control Conf.: pp. 7-12.

9 Torgeson, D. C. and H. Mee. 1967. Microbial degradation of bromacil. Proc. Northeast. Weed Contro. Conf. 21:584.

10 Torgeson, D. C. 1971. Interaction of herbicides and soil microorganisms. U.S. Environmental Protection Agency, Water Pollution Control Research Series 16060 DMR: 72 pp. (NTIS PB 211 566).

11 Van Schreven, D. A., D. J. Lindenbergh and A. Koridon. 1970. Effect of several herbicides on bacterial populations and activity and the persistence of these herbicides in soil. Plant and Soil 33:513-532.

12 Weed Science Society of America. 1979. Herbicide handbook. 4th ed. Weed Science Society of America. Champaign, IL.

13 Yazar, Oya, J. M. Shively and N. D. Camper. 1975. Effects of selected herbicides on bacterial populations in nontreated and treated water. Water Resources Bulletin, Paper No. 75001, Vol. II, No. 2. 2:294-299.