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## EFFECTS OF FEEDLOT RUNOFF ON FREE-LIVING AQUATIC CILIATED PROTOZOA

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### FINAL REPORT

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Water samples and free-living and sessite ciliated protozoa were collected at various distances above and below a stream that received runoff from a feedlot. No correlation was found between the species of protozoa recovered, water chemistry, location in the stream, or time of collection.

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### INTRODUCTION

The current trend for feeding livestock in the United States is toward large confinement types of operation. Most of these large commercial feedlots have some means of manure disposal and programs to prevent runoff from feedlots from reaching streams. However, there are still large numbers of smaller feedlots, many of which do not have adequate facilities for disposal of manure or preventing runoff from reaching waterways. The production of wastes by domestic animals was often not considered in the past, but management of wastes is currently one of the largest problems facing the livestock industry. It has been estimated that 3 pounds of manure are produced for each quart of milk produced and 6 to 25 pounds of manure are produced for each pound of livestock weight gain (Hart and McGanhey, 1963). In Illinois the equivalent population of animal wastes is considered to be greater than the wastes produced by the human population (Loehr, 1968).

That ciliate protozoa are common in streams polluted by biological wastes has been well documented (Lackey, 1925; Liebmann, 1951), and the numbers of ciliated protozoa were reported to decrease with increased biological pollution (Cairns, 1965). However, a different study reported that highest population levels of ciliated protozoa were found nearest the discharge of a waste treatment plant (Small, 1973).

Aquatic ciliated protozoa have a cosmopolitan distribution, but few recent studies have been concerned with their role as indicators of pollution or their possible role in removing bacteria from polluted streams. Many species of ciliated protozoa are bacterivores and are an important source for removing bacteria from water. That the distribution of ciliated protozoa depends on their food supply has been well described (Fauré-Fremiet, 1950).

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#### OBJECTIVES

The objectives of the study were: (1) to identify the ciliated protozoan profile in a stream which receives direct runoff from a cattle feedlot and (2) to correlate the ciliated protozoan profile with biological and chemical indicators of pollution.

### MATERIALS AND METHODS

### Study Area:

The study area was near DuQuoin, Perry County, Illinois. The feedlot used may contain as many as 4,000 cattle at any one time and has an annual turnover of approximately 10,000 animals. Runoff from the feedlot reaches Reese Creek, a tributary of the Little Muddy River (Appendix I). There is no runoff into the stream from other feedlots. Farmers in the area below the feedlot were concerned with pollution of the stream and were cooperative in establishing collection sites on their farms.

Although personnel at the feedlot stated that no runoff from the feedlot reached Reese Creek, there was visual evidence of runoff at one point. Settling basins were constructed during the summer of 1974 to receive. Two primary settling basins received liquid wastes from the feedlot. These basins were connected to canals which fed liquids into three large lagoons, which were completed on November 17, 1974, and after that controlled feedlot runoff.

Collection sites were located above and at various distances below the feedlot runoff. During the first year of the study, one collecting site was used above the stream flow through the feedlot to determine baseline chemical and biological properties of the stream. Two sites were established in each of the following zones below the feedlot--"septic, oxygen sag, and clean water." The above zones were determined by collecting invertebrate indicator organisms

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such as physid snails, tubificid oligochaetes, and midge larvae. The invertebrates were collected by using standard quantitative limnological methods. The collecting sites (SI-S6) are illustrated in Appendix 1. SI was located above a bridge approximately 1 mile above the feedlot. The stream at this point was approximately 30 feet wide and 3 feet deep. The stream was slow, contained much wooded debris, and had little visual evidence of pollution. No runoff from any feedlot entered the stream above this point. Site S2 was located about 40 feet downstream from the area which received periodic runoff from the feedlot. At this point the stream was 10-12 feet wide and 1-3 feet deep and was rapidly flowing. Site S3 was located approximately 15 feet below an area which received raw municipal sewage from the city of DuQuoin during heavy rainstorms. No raw sewage was allowed to run into the stream after May 30, 1975. Site S4 was located approximately 100 yards below S3. The stream was about 10 feet wide and 3 feet deep with slow running water. Site 55 was about two miles below site S4. At this point the stream was about 15 feet wide and 3 feet deep. There was sluggish water flow. Studies during 1974 indicated that there were few differences in ciliated protozoa at all sites. Because of widespread pollution the normal bactivore/carnivore protozoan profile could not be established. For this reason two additional collection sites were established. Site S0 was approximately 1.5 miles upstream from S1, and S6 was approximately 1.5 miles below S5. It was anticipated that these sites would provide additional data to establish the ciliate profile of the stream.

### Collection of Samples:

Ciliated protozoa and water samples were collected at weekly to 10-day intervals from July 3, 1974, to November 5, 1975. Thereafter collections were made at biweekly intervals until June 5, 1975. Samples were then collected at weekly intervals until August 7, 1975, when biweekly collection began. These were continued until termination of the study on April 16, 1976.

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Samples collected at each site were (1) slide traps for sessile protozoa, (2) bulk water samples for motile protozoa, and (3) bulk samples to determine ammonia, nitrate, nitrite, dissolved oxygen, and biological oxidation demand.

The coverslip traps have been previously described (Small, 1973). Traps containing 10 coverslips were placed at each site one week prior to collection. The traps were attached to a string which was anchored to the stream bank. After collection a new coverslip trap was placed at each site. As soon as the coverslip trap was removed from the water it was placed in a jar containing 15 cc of Holland's fixative. Approximately 1 liter of water was collected in bottles to study free-living protozoa and an additional 250 cc was collected for chemical analyses. The water samples and coverslip traps were placed in insulated styrofoam containers packed with artificial freezer containers and transported to the laboratory.

Coverslips were retained in the fixative until staining with protargol.

The water samples containing the free-living protozoa were concentrated with a continuous flow centrifuge. The samples were then affixed to coverslips with albumin and stained as described above.

Chemical analyses were performed by the Illinois Environmental Protection Laboratory, Urbana, Illinois.

#### RESULTS

## First Year of Study:

The first site chosen, about 1 mile above the feedlot, had approximately the same types of bactivorous ciliates as were found at the point of visual feedlot runoff into the stream. (A rise in bactivorous ciliates parallels a rise in bacterial growth associated with increased biological pollution.)

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This pattern was constant far downstream although the diversity of species decreased from that area accepting the greatest direct runoff (sites 2, 3, and 4). At site 1, 11 different genera were found. These were all bactivores. At site 2, one of the two sites of visual runoff, 15 species were found. These were all bactivores except for 1 carnivore. There were 14 genera found at site 3, the second area of visual pollution. Again, there was only one carnivore. At site 4, where the stream widens, flows slowly and where it receives bright sun most of the day, 20 genera were found. Of these, only 2 genera were carnivores. Finally at site 5, about 2 miles downstream from site 4, the number of genera dropped to 13. Two of these were carnivores. The genera and the number of times each was found are given on Table 1.

It is clear from the results of this study thus far that the ciliate profile presents a general picture of all sites. Because of the widespread pollution, the normal bactivore/carnivore profile of a "clean" stream has not been established. In further studies, sites will be established much further downstream and upstream to see how or whether the protozoan picture varies much from that found at the sites near the feedlot.

The chemical indices of pollution at each site confirmed that the stream was polluted at all sites except site 1. According to standards set for Illinois waterways<sup>1</sup> the dissolved oxygen should not be less than 5.0 mg/liter at any time. All sites except site 1 had readings at or below this level at least once during the study period. Also according to the same publication, the concentration of ammonia nitrogen (N) should not exceed 1.5 mg/liter. Site 1 again was the only sample site that did not exceed this reading at least once in the year. The only index for which more than one site remained continuously in the safe range was biological oxygen demand (BOD). Here both

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sites 1 and 2 remained well below the established 30 mg/liter throughout the entire year.

The food sources of the various protozoa found are presented in Table 3.

No correlation was found between the species of protozoa recovered, water chemistry, collection site, and time of collection.

		No. of times
<u>Site No</u>	Genera	found
1	Vorticella	17
-	Paramecium	9
	Tetrahymena	8
	Blepharisma	5
	Epistvlis	5
	Glaucoma	2
	Zoothamnium	2
	Carchesium	-
	Colpidium	-
	Glenodinium	1
	Euplotes	1
2	Vout	16
2		16
	Tetranymena	. 0
	Epistylis	8
		4
	Glaucoma	с С
	Cotpidium	2
		2
	Hotophrya	
	<u>TINCINNOPSIS</u>	2
	Biepharisma	1
	<u>Dileptus</u>	1
	Gienoainium	· · · · · · · · · · · · · · · · · · ·
		· <u> </u>
	Platycola	1
	Zoothamnium	1
3	Vorticella	8
	Tetrahymena	3
	Colipidium	5
	Glaucoma	2
	Paramecium	2
	<u>Tintinnopsis</u>	2
	Blepharisma	1
	Chilodonella	1
	Didinium	1
	Epidinium	1
	Epistylis	1
	Frontonia	1
	Holophrya	1
	Tetrahymena	1

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TABLE 1. Genera of protozoa and the numbers of times each was found at each site.

Table 1 (continued)

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		No. of times
Site No.	Genera	found
4	Vorticella	13
	Tetrahymena	10
	Blepharisma	3
	Colpidium	3
	Glaucoma	3
	Paramecium	3
	Epistylis	2
	Tintinnopsis	2
	Chilodonella	1
	Climacostomum	1
	Coleps	1
	Colpoda	1
	Cymatocyclis	1 ~
	Didinium	1
	Epidinium	1
	Holophrya	1
	Leucophrys	1
	Malacophrys	1
	Opercularia	1
	Zoothamnium	1
5	Vorticella	17
	Tetrahymena	9
	Paramecium	7
	Didinium	3
	Blepharisma	2
	Chilodonella	2
	Colpidium	2
	Glaucoma	2
	Coleps	1
	Cothurnia	1
	Epistvlis	1
	Euplotes	1
	Holophyra	1
	<u></u>	

		No. of times
Site No.	Protozoa	found
S0	Vorticalla	21
		21
	Tetranymena	19
	Cothurnia	11
	Platycola	5
	Tintinnidae	4
	ZOOThamnium	3
	Blepharisma	3
	Colpidium	3
	Epidinium	
	Glaucoma	
	Paramecium	1
	Podophrya	1 -
Sl	Vorticella	29
	Holophrya	11
	Tetrahymena	9
	Cothurnia	7
	Coleps	6
	Colpidium	6
	Cymatocyclis	5
	Epistylis	4
	Glaucoma	3
	Platycola	l
	Spirostomum	1
	Tintinnidae	1
	Zoothamnium	· 1
C 2	Vorticalla	О
52		5
	Banama sium	5
	Tetrahecium	5 1
	Chiledenelle	±. 1
		1
	GLAUCOMA	T
S3	Vorticella	11
	Holophrya	4
	Tetrahymena	3
	Chilodonella	3
	Zoothamnium	2
	Blepharisma	2
	Colpidium	1
	Didinium	1
	Frontonia	1
	Glaucoma	1
	Leucophrys	1
	Prorodon	1
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TABLE 2. Genera of protozoa and the numbers of times each was found at each site.

TABLE 2 (continued)

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		No. of times
Site No.	Protozoa	found
S4	Vorticella	18
	Holophrya	±0 6
	Tetrahymena	5
	Zoothampium	5
	Chilodonella	4
	Colpidium	1
	Cumatocyclis	1
	Paramogium	1
	Taramecium	1
	Didinium	1
	Erontonia	1
	Loncophrus	1
	Leucopintys	I .
S5	Vorticella	29
	Holophrya	18
	Tetrahymena	12
	Glaucoma	7
	Paramecium	4
	Zoothamnium	1
	Frontonia	1
	Hypostomata	1
	Oxytricha	1
	Tokophyra	1
<b>S</b> 6	Vorticella	11
	Cothurnia	6
	Didinium	6
	Tetrahymena	5
	Glaucoma	5
	Holophrya	4
	Chilodonella	3
	Epidinium	1
	Euplotes	1
	Heliophra	1
	Nassula	1
	Paramecium	1
	Zoothamnium	1

Protozoan	Food Source*
Blepharisma	B
Carachesium	B
Chilodinella	
Climacostomum	R,D,D
Coleps	
Colpeda	
Colpidium	ם,ר סיד
Cothurnia	D,I
Cuamatoguelis	D D
Didinium	В
Dileptus	· C
Dileptus	C
	G
Epistylis	в
Euplotes	А, В, С, Р
Frontonia	A,C,D
Glaucoma	В
Glenodinium	В
Heliophyra	С
Holophyra	E.
Hypostomata	A
Leucophrys	В
Malacophyrys	В
Nassula	В
Opercularia	В
Oxytricha	A,B,F
Paramecium	B,F
Platycola	А,В
Podophyra	С
Prorodon	В,Н
Spirostomum	A,B,F
Tetrahymena	В
Titinidae	B,D,F
Tintinnopsis	B,D,F
Tintinus	B,D,F
Tokophyra	С
Vorticella	A,B
Zoothanium	В

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TABLE 3. Food source of protozoa found during

A - algae

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B - bacteria

C - ciliates

D - diatoms

F - flagellates

G - rumen ciliates

H - nematodes

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