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Algal epilithon and water quality of a stream receiving oil refinery effluent

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Abstract

Changes in epilithic algal communities colonizing introduced substrata were determined in a stream polluted with oil refinery effluent at Digboi (Assam, India). The number of algal taxa was reduced but the growth of blue-green algae, particularly two species of *Oscillatoria*, was encouraged. Epilithic biomass (as chlorophyll *a*) also declined at polluted stations. The algal community of the upstream station was markedly different from the community occurring just after the confluence of effluent; however, the differences were gradually reduced downstream, indicating improvement in water quality. Of the various criteria tested for possible relationships with the level of pollutants, species richness, Shannon diversity and biomass showed significant relationships. The study demonstrates the usefulness of algal criteria for monitoring oil pollution in running waters.

Introduction

Oil pollution effect studies on algae have so far been confined largely to laboratory-based investigations (O'Brien & Dixon, 1976; Vandermeulen & Ahern, 1976). These studies have demonstrated harmful effects of the oil, though gradual recovery has been seen due to loss of toxic volatile fractions (Dunstan *et al.*, 1975; Soto *et al.*, 1977). Therefore, laboratory data cannot be of sole use for predicting effects in nature, because of interactive effects of environmental and other factors.

Studies in the aftermath of oil spills have revealed contradictory observations: in some cases no effect could be seen and in others, marked toxic effects (see O'Brien & Dixon, 1976). Studies on algal communities in waters contaminated with oil refinery effluents are rare, though phytoplankton communities have been studied by Minter (1964) and McCauley (1966). Cooper & Wilhm (1975) studied colonization of periphytic algae on plexiglass slides in a stream receiving refinery and domestic wastes, but did not include measurements of the pollutants. Gaur & Kumar (1985) described algal community structure in effluent holding and treatment ponds of five oil refineries.

The purposes of this study were to determine the extent of oil and phenol contamination in the refinery effluent, and the receiving stream and to correlate their concentrations with changes in structure of epilithic algal community for biomonitoring of water quality.

Site description

The study was conducted in a small stream receiving 380 m^3 h^{-1} effluent from Digboi refinery (27°64'E long. and 96°18'N lat.) in the state of Assam. This is one of the oldest refineries in Asia and does not have adequate facilities for the treatment of wastewater. The liquid wastes generated in the refinery are merely given physical treatment in API seperators, where free-floating oil is removed by skimming. The effluent is eventually carried in an open channel (about 1 km long) and discharged into the receiving stream.

Methods

Sampling was conducted on 6 April and 10 September 1986 and 6 January 1987, representing summer, rainy and winter seasons, respectively. Five stations were marked for sampling, one in the effluent channel before its mixing with the stream (Station B) and the remaining four in the receiving stream: A, upstream prior to confluence with the effluent channel; C, immediately downstream of the confluence; D and E situated downstream 1 and 2 km, respectively, from the confluence.

The pH of water was measured in the field with a combination electrode (Ingold). For other estimations, samples were transferred quickly to the laboratory and analysed as early as possible following the methods described in APHA (1985). NH_4-N was estimated by the phenate method within a few hours after sample collection. The samples for NO_3-N and total reactive phosphorus were stored in freezer and analyzed within 24 h after collection by Brucine-sulfanilic acid, and stannous chloride methods, respectively. For phenol estimation, the samples were first acidified to pH 4.0 with phosphoric acid and stored at 4 °C. Phenol content was determined by 4-aminoantipyrine method. Samples for oil estimation were extracted with carbon tetrachloride and analysed by gravimetric method.

In order to study the periphyton community at the various sampling stations, rock substrata of uniform size and shape (ca. 20 cm diameter) were selected from the area close to the receiving stream and brushed clean before placing them in the stream. Eight-week exposure was found to be sufficient for colonization. Exactly eight weeks before sampling, the substrata were secured at a depth of one metre in a similar flow condition. Substrata were withdrawn from all sampling locations and a 5-cm^2 area was scraped from them for estimation of biomass (chlorophyll *a*) and relative abundance of algae. Chlorophyll *a* was estimated (using four replicates) on substrata by the method of Mackinney (1941). Six replicates of substrata were utilised for determining algal community structure. Algal growths were scraped carefully and diluted with a known amount of water. Approximately 500 individuals were counted in several microscope fields (McIntire & Overton, 1971). Diatom samples were boiled in concentrated HNO_3 for 30 min; empty frustules were mounted in euparaol and observed at 1000 × magnification. Algal identification was based on keys by several workers.

The data pertaining to relative abundance of algal taxa were used to calculate evenness or equitability and Shannon-Weiner index, and index of dissimilarity between various stations. Diversity (H′) was calculated by the following formula as given by Shannon (1948):

$$H' = \sum_{i=1}^{k} P_i \ln P_i ,$$

where k is the number of categories and P_i is the proportion of observations found in category i.

Species evenness was calculated using the expression of Pielou (1966):

$$J = \frac{H'}{\ln S} ,$$

where J is the evenness, H′ is the observed diversity and S is the number of categories (taxa).

Dissimilarity index or DI (Levandowsky, 1972) was computed using the following formula:

$$DI = 1 - \sum_{i=1}^{S} (\min X_{1i}, X_{2i}) / \sum_{i=1}^{S} (\max X_{1i}, X_{2i}),$$

where X_{1i} and X_{2i} are values based on relative abundance of ith taxon in communities 1 and 2, respectively, and S is the total number of species. DI value of zero indicates identical communities whereas communities totally different from each other have DI value of 1.

Results

Chemical features of the refinery effluent and the receiving stream are given in Table 1. The pH of the refinery effluent was always alkaline, whereas the receiving stream was slightly acidic. Entry of the effluent to the stream caused a slight increase in pH. NH_4-N concentration in the effluent was much higher than in the receiving stream.

There was not much variation in concentration of NO_3-N and total reactive phosphate at various sampling stations. Maximum concentration of oil and phenol occurred in the effluent channel, but downstream decline was observed.

The largest number of algal taxa were recorded from Station A (Table 2): Chlorophyta (7 species) was the most abundant algal phylum followed by Bacillariophyta (6 species) and Cyanophyta (3 species). Disposal of refinery effluent into the stream caused an immediate decline in the number of algal species, but, further downstream, many of these species reappeared. The distribution of individuals among various species was more or less uniform at Station A leading to high evenness. However, evenness was lowest at Station B due to the largest share of two species of *Oscillatoria* (Fig. 1). Regular increase in evenness occurred downstream due to gradual reduction in relative abundance of two species of *Oscillatoria* and concomitant increase in abundance of other species. Following a similar trend, S, H′ and J declined immediately after the input of effluent, but established a regular increasing trend downstream. The changes in algal community along the gradient of decreasing pollution load became more conspicuous when DI was taken into consideration (Table 3).

Temporal variation in the amount of chlorophyll *a* at various stations is shown in Table 4. The amount of chlorophyll *a* was maximum at Station A and minimum at Station B, but, downstream from Station B, gradual increase occurred.

Table 1. Chemistry of refinery effluent and the receiving stream at various sampling stations on three dates. All values in mg l^{-1} except for pH. See Materials and Methods for details sampling stations. ND = concentration below the detection limit.

Date	Sampling station	pH	NH_4-N	NO_3-N	PO_4-P	Oil	Phenol
6 Apr. 1986	A	5.9	1.17	1.20	0.17	ND	ND
	B	8.1	4.37	1.23	0.24	13.16	1.05
	C	7.9	3.48	1.08	0.16	12.55	0.79
	D	8.0	2.23	0.94	0.14	10.19	0.63
	E	7.8	2.12	1.05	0.11	7.45	0.56
10 Sept 1986	A	6.2	0.69	1.20	0.14	ND	ND
	B	8.3	2.24	0.81	0.10	11.72	0.95
	C	8.0	2.17	0.64	0.07	9.45	0.45
	D	7.6	1.87	0.73	0.13	7.12	0.45
	E	7.6	1.18	0.78	0.10	6.87	0.36
6 Jan 1987	A	6.1	0.80	0.93	0.17	ND	ND
	B	8.0	2.23	0.41	0.12	14.04	1.26
	C	7.6	2.74	0.63	0.12	10.43	0.78
	D	7.8	2.02	0.52	0.10	8.45	0.71
	E	7.6	1.90	0.41	0.07	7.17	0.69

Table 2. Relative abundance (expressed in percentage) of algal taxa in effluent channel and the receiving stream at various sampling dates (see Materials and Methods for the location of Sampling Stations).

	6 Apr 1986 Station					10 Sep 1986 Station					6 Jan 1987 Station				
	A	B	C	D	E	A	B	C	D	E	A	B	C	D	E
Chlorophyta															
Chlorella vulgaris Beijerinck	6	–	–	–	–	4	–	–	2	4	10	–	–	–	1
Cosmarium contractum Kirch. var. *papillatum* W. & G. S. West	13	–	–	4	6	10	4	6	4	8	16	2	3	4	5
C. bioculatum Bréb.	3	–	–	2	4	8	–	–	3	–	2	–	1	–	1
Closterium moniliforme (Bory) Ehr.	5	–	2	4	3	–	–	–	–	–	1	–	2	4	2
Pediastrum boryanum (Turp.) Menegh.	2	–	–	–	–	9	–	–	–	–	5	–	–	–	–
Scenedesmus dimorphus (Turp.) Kütz.	3	2	4	2	6	–	–	3	–	4	1	–	–	–	–
Spirogyra parvula (Trans.) Czurda	16	–	–	–	–	8	–	–	–	–	12	–	–	–	–
Cyanophyta															
Gloeocapsa magma (Bréb) Kütz.	2	–	–	–	1	–	–	–	–	–	3	–	–	2	2
Oscillatoria formosa Bory ex Gom.	14	24	33	36	24	11	23	28	30	32	14	25	29	28	30
O. princeps Vauch. ex Gom.	22	58	46	39	33	16	59	47	41	36	18	60	49	47	41
Bacillariophyta															
Cymbella turgida Greg.	2	3	1	2	4	7	–	–	4	3	4	4	3	4	3
C. lanceolata (Ehr.) Grun.	–	–	–	–	3	4	6	5	5	4	–	–	–	2	–
Fragilaria crotenensis Kitt. var. *praelonga* Grun.	6	3	4	2	5	10	–	2	6	–	3	–	–	–	3
Navicula gracilis Ehr.	–	–	1	3	2	–	–	3	–	4	6	2	6	5	5
Nitzschia gracilis Hantzch.	2	4	5	2	4	6	–	–	–	2	1	4	3	2	5
Synedra ulna (Nitzsch.) Ehr.	4	6	4	3	6	7	8	6	5	4	4	3	4	2	2

This trend was seen during three samplings, but chlorophyll *a* content was maximum in April followed by September and January.

Discussion

The study showed detrimental effects of oil refinery effluent on the receiving stream. Disposal of refinery wastes elevated ammonia level and introduced high concentration of oil and phenol in the receiving stream. Release of refinery effluent also caused an immediate loss of several algal species. On the other hand, two species of *Oscillatoria* greatly increased in abundance. Such differences resemble those found in other studies (see O'Brien & Dixon, 1976). Greater abundances of *Oscillatoria* spp. in habitats exposed to oil spills and refinery effluents has been observed by a number of authors (Hanna *et al.*, 1975; Schultz & Tebo, 1975; Snow & Scott, 1975; Gaur & Kumar, 1985). Palmer (1969) placed *Oscillatoria* at the top of his list of algae most tolerant to organic pollution, but it is unclear whether the remarkable tolerance to oil is related to this or other factors.

The refinery effluent had a small number of algal taxa, probably due to high pollution load. Occurrence of more algal species at the upstream station is presumably due to lack of pollution. Although the disposal of effluent caused an abrupt decline in number of algal species, the majority reappeared downstream where the level of pollution was well within their tolerance range. Due to this, there was a highly significant negative relationship between the level of pollution and species number (Table 5). Marked changes in evenness also occurred downstream. Many workers emphasize that the relationship of even-

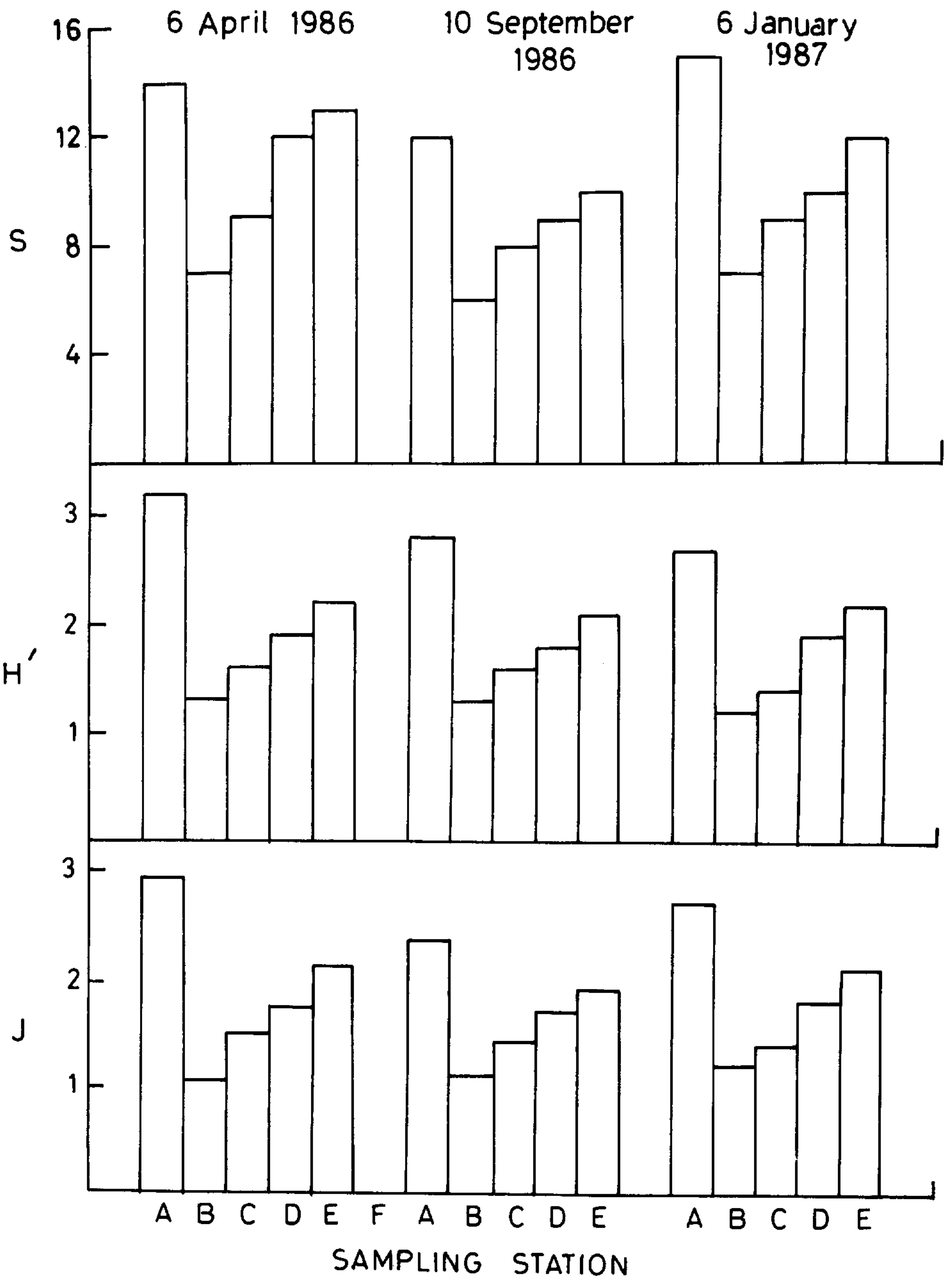

Fig. 1. Species richness (S), Shannon-Weiner index (H′) and evenness (J) of algal communities growing at various stations.

ness with stress is not clear (see Sheehan, 1984) and conflicting composition and evenness data have led them to question the value of evenness-based indices, including the Shannon index, in assessment and prediction of community response (Gray, 1979). Absence of a statistically significant relation of evenness with the concentration of phenol, and a slightly significant relation with the amount of oil in the medium, indicate that evenness is not a very effective criterion in the present case. Alteration in community structure may be the most serious consequence of oil pollution, because it could have profound impact on food webs and the entire ecosystem.

Table 3. Dissimilarity index of algal communities between pairs of sampling stations.

Date	Stations						
	A & B	A & C	A & D	A & E	B & C	C & D	D & E
6 Apr 1986	0.72	0.68	0.62	0.58	0.16	0.19	0.23
10 Sep 1986	0.81	0.78	0.68	0.52	0.21	0.24	0.30
6 Jan 1987	0.86	0.73	0.70	0.65	0.19	0.18	0.26

Table 4. Epilithic algal biomass (chlorophyll *a*) at stations A, B, C, D and E on three dates.

Station	Chlorophyll *a* (mg m^{-2})					
	6 Apr 1986		9 Sep 1986		6 Jan 1987	
	$\bar{X}$	SE	$\bar{X}$	SE	$\bar{X}$	SE
A	110.8	3.7	72.5	5.1	100.8	5.5
B	17.1	1.4	11.5	1.5	19.0	1.9
C	21.2	3.0	15.1	1.3	26.4	2.4
D	50.4	4.0	28.0	2.3	40.2	3.7
E	65.9	4.9	45.8	3.1	60.3	3.4

Table 5. Correlation coefficients between community indices (S = species richness, H′ = Shannon-Weiner diversity, J = evenness, and chlorophyll *a*) and related chemical parameters in refinery effluent channel or receiving water (*P < 0.05, **P < 0.01, ***P < 0.001).

	NH_4-N	Oil	Phenol
S	- 0.62**	- 0.82***	- 0.76***
H′	- 0.72**	- 0.95***	- 0.86***
J	- 0.73**	- 0.48*	- 0.40
Chlorophyll *a*	- 0.68**	- 0.89***	- 0.79***

Changes in algal community along the gradient of decreasing pollution load became more prominent when the Shannon-Weiner diversity index (H′) is taken into consideration. A highly significant relationship between diversity and the level of pollutants in the medium was apparent (Table 5). This observation fits with several other studies reporting decline in diversity following the release of pollutants (Cooper & Wilhm, 1975; Rai *et al.*, 1981; Gaur & Kumar, 1985). Though Shannon diversity and other non-parametric indices have met with considerable criticism (Hurlbert, 1971), they can be applied carefully in specific situations where the main aim is to detect gross changes (Sheehan, 1984).

The epilithic algal communities colonizing various stations were compared with each other using the dissilimarity index: this showed a clear relationship with the level of pollutants. The epilithon of the upstream station and the effluent channel were largely dissimilar, but the communities developing downstream were not much different from those at upstream station; the degree of similarity increased with increase in distance from the confluence.

There was a statistically significant inverse relationship between the concentration of pollutants (oil, phenol, ammonia) and algal biomass (Table 5), a result similar to that made by Bott *et al.* (1978) in an experimental stream exposed to three petroleum oils. Our findings differ however with those of Federle *et al.* (1979), who observed increased algal biomass after a simulated oil spill, and ascribed it to reduced grazing pressure due to elimination of herbivores. In spite of their results we suggest that the response of epilithic algal communities to oil stress indicates their use for monitoring purposes. Industries, Pollution Control Boards and other organizations should make use of algal criteria, besides the physico-chemical parameters which have long been employed.

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