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COLONIZATION RATES AND PROCESSES AS AN INDEX OF POLLUTION SEVERITY

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Rockville, Md. April 1983



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ABSTRACT

A comparative survey of colonization rates and events for the biological communities developing on identical hard substrates was conducted at several Puget Sound localities. These sites differed in the extent to which chemical toxicants were present in the water column and in the sediments. Colonization rates and patterns were compared at sites which differed with respect to pollution severity.

Initial patterns of colonization suggest trends that may be related to degree of chemical contamination of the sites. This survey indicates that the reference station, Manchester, tended to accumulate more species than any of the experimental sites at Commencement Bay and Duwamish, during the first months of colonization. After the initial colonization period, fluctuation in species number was greatest at the reference site in this experiment and at Elliott Bay in an earlier experiment. At the Commencement Bay sites, where the potential for pollution was greater, there was less fluctuation in species richness. Least fluctuation, as well as lowest species richness, was observed at the Duwamish site, another locality where pollution was potentially high. Between six months and 1 year after experiments began, the reference site had a statistically greater mean number of species present than any of the experimental localities. In the second year of the survey, colonization curves tended to converge at all but the Duwamish site, suggesting that the initial differences in colonization were obscured. In total the Duwamish site attracted fewest species, the Commencement Bay Waterways considerably more, although less than the reference site, Manchester.

No correlation was found between dominance of particular functional morphological groups and sites when attached organisms were classified as either solitary or colonial. Reference sites and experimental sites in potentially polluted areas showed similar patterns with solitary organisms predominating towards the end of the study. Nor did plots of the log-normal distribution of individuals per species indicate differences between sites. All showed a break in the log-normal distribution, possibly reflecting the increased larval settlement which occurred.

1. INTRODUCTION

Field studies documenting the responses of communities to different types of pollutants (c.f., Davis et al., 1977; Cory and Nauman, 1970; Rastetter and Cooke, 1979) are necessary to assess man's impact on the marine environment and to determine the resiliency of natural communities to man-induced perturbations. The present study attempts to compare fouling community development at several localities around Puget Sound, Washington, which differ with respect to their ambient concentrations of heavy metals, synthetic organic compounds and aromatic hydrocarbons. Several of these sites have received municipal and industrial wastes for a considerable period of time. Three of the sites located on Commencement Bay are part of an area designated by the U.S. Environmental Protection Agency as a hazardous waste site in need of remedial action. To date, several surveys sponsored by various government agencies have monitored particular chemical and biological aspects of the overall problem (Malins et al., 1982). The high frequency of lesions observed in bottom fishes has been shown to be correlated with increased pollutant levels (Malins et al., 1982). In addition, Swartz et al. (in press) have shown a strong negative correlation between amphipod species richness and abundance and increasing sediment toxicity. In the present study, the fouling community dynamics over a two year period at these pollutant stressed sites are compared with a relatively unpolluted reference site in Manchester, Washington and with some previous experimental data from other localities in Puget Sound. Differences in rates of colonization, number of species accumulating, and types of colonists might be expected to occur under different pollution regimes. To test this hypothesis identical colonization experiments were begun at reference and polluted sites.

2. MATERIALS AND METHODS

2.1 Field Sites

Experiments were conducted from stationary piers at all sites. The choice of sites was dependent upon the availability of chemical data from sediments and/or water columns (Riley et al., 1980, 1981). At some localities, seasonal collections of biota and sediments had been made by the National Marine Fisheries Service for the NOAA/MESA Puget Sound Project (Malins et al., 1982). Final choice of sites was made by mutual agreement with the MESA Puget Sound Office of NOAA. A brief description of the sites follows.

a. Commencement Bay Sites: Commencement Bay, located in the southern portion of Puget Sound (Figure 1), is a major industrial harbor adjacent to Tacoma, Washington. The port area is northeast of Tacoma. Three localities at the Port of Tacoma were chosen for experimental work: (1) Milwaukee Waterway, (2) Blair Waterway and (3) Hylebos



Figure 1. Map of Puget Sound indicating study sites: Clam Bay, Manchester (A), Duwamish River (B) and Port of Tacoma Waterways (C).

Waterway. These waterways are periodically dredged to maintain their depth.

(1) Milwaukee Waterway: This waterway extends towards East lith Avenue. Experiments were conducted from the piers adjacent to the now demolished warehouses along the southwestern side of the waterway. Little activity occurred on this waterway during monitoring periods.

(2) Blair Waterway: The experimental site was located at the easternmost sector of Pier 4, a cargo transfer pier. Experiments were conducted just west of the East 11th Avenue Bridge. During monitoring periods ships were often transfering cargo and this resulted in some damage to our experiments.

(3) Hylebos Waterway: Hylebos Creek empties into the upper regions of this Waterway. The dock of the Naval Reserve grounds served as the experimental site. The permanent walkway leading from shore to the minesweepers was utilized. Although periodic activity was noted at this site, little damage to experiments was observed.

b. Clam Bay, Manchester: This, the reference site for the experiments, is the northernmost of the study sites. It is located west of Seattle, on the Kitsap Peninsula. The pier at Clam Bay faces eastward, extending towards Rich Passage between central Puget Sound and Bremerton's Sinclair Inlet. Depth below the pier increased rapidly in an easterly direction. Experiments occupied the easternmost sector of the pier on its northern side.

c. Duwamish River: Experiments were conducted below the 16th Avenue South bridge, from the eastern catwalk. The Duwamish is a stratified estuary, with the salt wedge typically extending to this locality from Elliott Bay, thus the salinity regime was expected to be similar to that of the other sites.

2.2 Field Methods

At each site, 12 identical experimental surfaces were suspended independently approximately 0.5 m apart from each other along the length of each dock. Anticipating that some losses would occur, this number of panels was determined sufficient by a previous study of variability among fouling panels in Puget Sound (Schoener and Greene, 1980). The experimental surfaces were constructed from pieces of textured white formica, cut to measure 20.3 x 20.3 cm and glued to similarly sized backing. Each square panel was suspended by a 0.64 cm polypropylene hollowbraid rope which passed from a cleat on the dock through the panel's center, to a weight suspended approximately 1 m below the panel. Weights did not touch the substrate bottom. All panels in a series were submerged within an hour's time, and floated 3.1 m below mean tide level. Panels were spaced along the dock to avoid putting them too close to pilings where their colonization might be influenced by organisms already attached, yet sufficiently adjacent to each other, so that similar events might be expected to occur in each series.

2.2.1 Sampling Schedule

The experiments began in mid-summer, 1980, panels being submerged on July 27, 1980, at Clam Bay, Manchester, and on July 28, 1980, at the Commencement Bay sites. Approximately two weeks later, on August 8, 1980, identical panels were submerged at the Duwamish site. Panels were observed biweekly at first and subsequently at monthly intervals during the two-year period with only one hiatus. A schedule of sampling dates is given in Table 1.

No panel losses occurred at the Manchester site during the course of the study. Only one panel was disturbed at the Hylebos site, and this at a late observation date. Two panels were lost at the Duwamish Waterway, while both Milwaukee Waterway and Blair Waterway lost four panels apiece. By the end of the experiment eight panels remained at Blair and Milwaukee Waterways, 10 at the Duwamish site, 11 at Hylebos and 12 at Manchester.

2.2.2 Biological Analyses

Panels submerged horizontally accumulate least sediment and most macrofouling growth on their undersurface. Therefore, the undersurface of each panel was studied. Observations were made while panels were temporarily retained in trays of seawater. After examination panels were returned to their original positions dockside. Measurements included an enumeration of the attached species and the coverage on each panel. The latter measure was estimated utilizing a point-sampling technique. This method involved generating anew at each sampling date a number of random points on an area equal to that of the panel. These points were then transferred to a clear plexiglass sheet which was suspended over the panel. By sighting which species was under each point, an estimate of percent cover could be made to within 5% accuracy compared to camera lucida determinations (Sutherland, 1974). All observations were made in a nondestructive manner with little disturbance to the growth on the panel.

RESULTS

3.1 Community Richness

A total of 74 species, primarily macrofaunal invertebrates, plus several species of urochordates and algae were observed on panels submerged at the combined study sites (Table 2). The total number of species was 66 at the Manchester reference site. This site had accumulated considerably more species than had any of the Commencement Bay sites. Milwaukee Waterway had 47 species, Blair Waterway had 48 species and Hylebos Waterway had 50 species. In total the Commencement

	· · · · · · · · · · · · · · · · · · ·	DATE OF	LOCALITY SUF	RVEY	· · · · · · · · · · · · · · · · · · ·
MONTH, YEAR	MANCHESTER	DUWAMISH	HYLEBOS	BLAIR	MILWAUKEE
August, 1980	23		20	20	20
September, 1980	5,17	6,21	3,16	3,16	3,16
October, 1980	10	12	9	9	9
November, 1980	8	11	10	10	10
December, 1980	17	25	15	15	15
January, 1981	29	25	24	24	23
February, 1981	21	22	26	25	23
March, 1981	17	19	18	18	18
April, 1981	27	30	29	29	29
May, 1981	24	29	24	22	22
June, 1981	23	30	25	24	24
July, 1981	30*	*(8)11	28	31	28
August, 1981	25	*(9)16	27	28	27
September, 1981	24	*(10)27	26,30	30	30
January, 1982	23	21	18	18	15
February, 1982	27	*(3)4	23	24	24
March, 1982	19	21	18	22	18
April, 1982	25	24	21	21	21
May, 1982	25	24	21	21	21
June, 1982	28	27	*(7)2,7	*(7)7	*(7)10

Table 1. Schedule of sampling periods.

* month shown in parentheses

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Algae	Mollusca
Antithamnion sp.	Pelecypoda
Callophyllis sp.	Chlamys sp.
Ceramium sp.	Hiatella sp. 1
Desmerestia sp.	Hiatella sp. 2
Laminaria sp.	Mytilus edulis
Microcladia sp.	unk, bivalve sw
Polyneura sp.	Gastropoda
Polysiphonia sp.	Calvotraea fastigiata
Ulva lactuca	Corvphella sp. eggs
Protozoa	Hermissenda crassicornis eg
Foliculina en	Onchidoris bilamellata eggs
Foliculina sp.	
Porifera	Arthropoda
Halichondria or Haliclona sp.	ampnipod tube
Leucosolenia sp.	Balanus crenatus
Scypha raphanus	Bryozoa
Cnidaria	Bowerbankia gracilis
Hydrozoa	Bugula pacifica
Clytia sp.	Callopora horrida
Obelia longissima	Cribrilina annulata
Obelia sp.	Cryptosula pallasiana
Tubularia marina	Dendrobeania lichenoides
hydroid sp. 1	Hippothoa hyalina
Scyphozoa	Membranipora membranacea
scyphistomae	Reginella nitida
Anthozoa	Scrupocellaria sp.
Metridium senile	Tubulipora tuba
Tealia crassicornis	unk. bryozoan l
	unk. bryozoan 2
Nemertea	unk. bryozoan 3
Emplectonema sp.	unk. bryozoan 4
Nemertina sp.	Chardete
Annelida	Urochordata
Polychaeta	
Eudistylia vancouveri	Ascidia paratropa
Myxicola infundibulum	Roltonia villago
Pseudopotamilla occelata	Bottenla Villosa Potrullus or Botrulloidos
Sabellaría cementarium	Cholycopp productum
Schizobranchia insignis	
Serpula vermicularis	Diplocome meedopoldi
Spionid sp.	Dipiosoma macuonaldi Distanlia ossidentalia
Spirorbis sp.	stuala dibbeii
Nereis sp.	unk colonial an #1
Terebellid sp.	Unt. COLUMIAL Sp. #1 Vertebrata
Svllid sp. 1	vertebrata vellow fish soos
Sv1lid sp. 2	AGTTOM ITSU GSS2
-,	Misc.
	unk. green egg mass
	unk. eggs

Table 2.	Combined	list of	sessile	species	colonizing	panels
	from all	sites.				

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Bay sites had 56 species. Fewest total species, 19, were present in the Duwamish. Algal species were noticeably lacking at this site.

During the course of the experiments various species immigrated onto panels, while others disappeared. Appendix A summarizes these changes at each of the five sites. Characteristic species assemblages, designated alphabetically, were determined based on cluster diagrams for each site. Only those species which occupied greater than 10% of the space in a given assemblage were included in these calculations, a criterion consistent with Sutherland and Karlson's (1977) definition of the foundation species of a fouling community. By following the transitional sequences of species assemblages observed for each of the 12 panels at each site through time, and then inspecting the composition of these assemblages, a fairly detailed picture of the community's development can be portrayed.

Panels at Manchester were primarily covered by hydroid and bryozoan growth during the first few weeks. Panels submerged in each of the three Tacoma Waterways developed different dominants during this time, even though all panels were submerged within hours of one another. Initial coverage in Hylebos Waterway was by a colonial tunicate, <u>Botryllus or Botrylloides</u> sp., while Blair achieved extensive coverage by an arborescent bryozoan <u>Bugula pacifica</u>. Milwaukee Waterway during this time had a heavy barnacle set (<u>Balanus crenatus</u>). The Duwamish River accumulated high initial coverage of the encrusting bryozoan Membranipora membranacea.

By the end of the two-year observation period, panels had considerable coverage by barnacles (<u>Balanus crenatus</u>) at the Duwamish. Manchester had well-developed calcareous polychaete tubes (<u>Serpula</u> <u>vermicularis</u>) accounting for extensive coverage, along with additional coverage by the anemone, <u>Metridium senile</u>, and the bryozoan <u>Scrupocellaria</u>. Milwaukee Waterway had an abundance of <u>Serpula</u> and <u>Balanus predominating</u>. Blair Waterway had <u>Balanus</u> along with <u>Metridium</u> and Serpula. Hylebos was dominated by Balanus and Bugula.

3.2 Rates of Colonization

To compare colonization patterns among sites with different biotas, taxonomic and environmental differences should be minimized. At each monitoring period, physical parameters including water temperature and salinity were measured at each site with a portable Yellow Springs Water Quality Instrument (Table 3). In order to minimize taxonomic differences an estimate of species-richness was made by summing the total number of sessile species per panel. Mean number of species and standard deviations were determined for each series (Table 4).

Initially all substrates were empty. Therefore, all colonization curves plotting the number of sessile species accumulating with time started from the origin (Figure 2). For visual clarity the zero points are not shown in the figure. Although most curves were monotonically

	MILWAUKEE	WATERWAY	BLAIR I	ATERWAY	HYLEBOS	6 WATERWAY	MANC	HESTER	DUW	AMISH
DATE	T(°C)	S(o/oo)	T(°C)	S(o/oo)	T(°C)	S(o/oo)	T(°C)	S(o/oo)	T(°C)	S(0/00)
August, 1980	15.6	25.3	15.2	20.9	15.9	20.9	14.1	29.9	15.2	27.2
September 3, 1980 16, 1980	14.9 15.6	28.5 27.8	15.0 14.0	29.1 28.7	16.0 16.5	28.8 27.9	14.9 13.9	30.1 30.2	15.0 14.7	22.2 21.8
October, 1980	12.3	27.5	13.6	28.7	13.2	27.9	13.1	28.9	12.8	28.0
November, 1980	11.6	28.2	11.5	28.1	11.8	28.6	11.8		11.3	25.8
December, 1980	10.2	28.0	10.5	27.7	11.5	27.2	10.1	27.2	9.9	23.8
January, 1981	9.8	26.2	9.7	26.3	9.8	26.0	9.4	26.6	9.3	25.0
February, 1981	9.0	26.3	9.2	26.5	9.9	26-4	9.7	26.1	7.6	22.5
March, 1981	9.2	25.8	10.9	26.0	10.1	25.8	9.5	27.1	10.2	20.5
April, 1981	10.5	28.9	13.0	25.1	10.8	29.2	10.9	22.5	10.9	22.5
May, 1981	10.9	25.2	13.0	24.0	10.9	25.2	11.8	26.3	11.8	23.0
June, 1981	13.9	23.0	11.9	24.2	14.5	23.9	12.5	25.5	16.0	2.0
July, 1981	15.1	23.6	13.8	24.1	14.9	23.8	14.0	25.6	19.0	11.5
August, 1981	13.6	24.8	13.8	24.8	14.5	25.9	15.1	25.8	14.2	23.8
September, 1981	12.8	22.2	13.2	25.9	13.5	24.5	14.1	25.6	11.8	23.8
January, 1982	9.5	24.4	8.8	23.8	9.5	25.8	9.1	21.0	11.3	22.6
February, 1982	8.3	23.2	8.5	23.8	8.3	21.9	8.3	24.0	8.3	22.2
March, 1982	8.3	22.1	8.3	23.7	9.2	23.8	9.0	20.1	8.0	20.8
April, 1982	9.0	22.6	10.2	22.5	11.6	21.8	9.8	19.2	9.9	18.3
May, 1982	11.1	22.1	12.0	22.4	12.8	22.4	11.9	23.2	12.6	2.1
June, 1982	14.5	23.0	16.0	21.2	13.3	22.5	12.8	23.2	13.5	22.2

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Table 3. Temperature and salinity data for field sites.

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TIME		DUWAMISH	MANCH	ESTER	BL	AIR	MILW	AUKEE	HYI	LEBOS
(weeks)	Meàn (x)	Standard Deviation (s	x)	S	x	S	x	S	x	S
3	8.50	1.51	7.33	1.30	6.67	1.30	1.83	0.58	7.25	1.30
5	8.00	1.41	10.75	2.63	9.17	0.94	3.33	0.78	8.58	2.02
7	5.00	1.83	12.75	2.45	10.00	1.54	3.42	0.79	9.58	2.35
10	3.70	0.82	14.75	2.53	11.92	1.62	4.42	1.00	9.58	2.02
14	3.33	0.50	14.08	2.11	13.58	2.23	8.17	1.75	9.58	1.44
18	5.44	1.24	15.83	3.01	12.45	2.73	8.00	2.34	11.92	2.54
23	5.70	1.06	18.00	1.95	12.55	1.69	9.00	1.48	12.33	2.02
27	5.30	1.06	18.42	1.88	13.20	1.99	9.42	1.44	14.42	1.88
31	4.80	0.92	19.58	2.43	15.20	2.25	11.58	2.23	16.08	1.98
36	4.10	0.88	20.33	2.61	14.90	1.70	14.58	1.98	16.50	1.98
40	4.70	0.82	19.50	3.42	15.40	2.32	14.45	1.13	15.33	2.35
45	4.90	1.20	20.00	3.10	14.13	2.53	14.64	1.69	14.36	1.43
50	4.50	0.97	18.17	3.04	16.00	2.67	15.00	1.84	15.83	1.64
54	4.30	0.68	16.50	3.32	15.75	2.43	14.82	0.98	15.17	2.17
58			17.83	2.98	15.75	3.11	15.64	1.75	15.92	1.93
76	4.10	0.57	13.08	2.43	13.12	1.64	14.27	1.90	14.83	1.11
80	3.67	0.71	13.83	2.82	13.88	1.81	14.09	1.70	14.25	1.36
84	3.70	0.48	15.25	3.39	15.63	3.20	15.56	1.88	17.09	0.94
89	3.70	0.48	14.33	2.23	16.38	2.77	17.50	1.77	16.91	1.87
93	4.00	0.67	15.83	3.54	17.37	2.45	15.88	2.10	15.91	1.92
98	4.50	0.53	15.25	2.83	15.88	2.90	14.25	2.31	16.27	2.72

Table 4. Colonization parameters for species richness at survey sites.

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Figure 2. Colonization curves for panels at several Puget Sound localities. E=Elliott Bay, A= Manchester, B=Blair Waterway, H=Hylebos Waterway, M=Milwaukee Waterway and D= Duwamish River.

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increasing throughout the initial weeks, not all did. The Duwamish colonization curve initially rose as did the curves at the Commencement Bay and Manchester sites, but it soon fell. Visual observation of the colonization curves suggested that the most rapid increases took place during these first six months after submergence. During this time there was a tendency, although not a significant one, for the reference site to accumulate a greater number of species than any of the experimental localities did during the same time period. A linear regression model fitted through the origin was used to determine the slopes of the individual colonization curves during this period. At all experimental localities the early portion of these colonization curves had rapidly rising and significantly positive (P < 0.05) slopes which were not significantly different from each other. Differences in total number of species accumulated could have been due to the fact that panels at the reference site continued to add species for a longer time than did any of the experimental localties.

3.3 Community Stability

Comparing the later months of colonization with the first six months of the study indicated that the rate of colonization was leveling off. The colonization curves from six months through the remainder of the study had more gradual slopes, none of which was statistically different from zero. The probability that the latter 1-1/2 years of the colonization curve rose less rapidly than the early portion of the curve was calculated using t-tests comparing these slopes. Significant differences (P < 0.05) between these two portions of the curve were observed at all localities except the Duwamish. These results indicated that species were being added at a slower rate as time progressed, and suggested that the communities were approaching an equilibrial number of species. At the Duwamish site the number of species was low and differences in rate might have been difficult to establish because of this. Between the first six months of colonization and the first year, the reference site, Manchester, had a significantly greater number of species than did the experimental localities (P < 0.05). After the first year of colonization substrates from all but the Duwamish site had approximately similar numbers of species present. The Manchester site even decreased its number of species, a phenomenon which also occurred in an earlier study in Elliott Bay (Schoener and Schoener, 1981).

3.4 Community Organization

When colonists on panels at survey sites were classified with respect to their functional morphology as either solitary or colonial organisms (Jackson, 1977) it was evident that colonial organisms dominated initially at almost all study sites (Figure 3, 4). Only at Milwaukee Waterway were there slightly more solitary individuals during the initial year of colonization. However, at this site total coverage was considerably lower during this time period than at the other sites (Table 5). A brief period of colonial dominance was recorded thereafter, and by the end of the experiments solitary organisms prevailed both at Milwaukee Waterway as well as at all the other sites. Two



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TIME	DUWAMISH		MANCHE	MANCHESTER		IR	MILWA	MILWAUKEE		HYLEBOS	
(weeks)	Mean (x)	Standard Deviation	x (s)	S	x	S	x	S	x	S	
3	28.00	9.73	6.58	6.99	2.17	1.34	1.92	1.31	2.25	1.14	
5	70.20	7.69	34.50	22.28	22.08	8.17	19.33	12.56	11.17	6.63	
7	91.00	6.88	43.58	19.68	36.25	6.62	25.75	8.85	27.00	12.00	
10	97.90	3.93	33.33	10.02	39.67	9.58	16.67	11.37	51.42	12.41	
14	92.00	5.27	35.00	17.88	30.17	7.22	17.83	12.43	71.08	15.84	
18	51.22	11.11	37.83	12.26	21.00	6.75	22.83	14.45	39.08	15.31	
23	44.10	13.90	32.50	11.67	15.18	3.43	19.92	13.11	21.67	9.99	
27	26.50	15.78	36.75	13.61	24.10	4.89	19.08	13.57	19.33	6.79	
31	76.10	6.31	45.33	11.37	19.00	8.03	30.58	10.36	20.08	5.84	
36	88.20	5.77	62.75	9.52	52.30	11.81	57.25	7.15	46.92	7.12	
40	88.80	5.77	80.75	7.12	70.10	13.25	74.91	6.61	79.75	8.92	
45	98.60	2.17	81.83	9.17	74.00	16.56	80.36	9.05	83.36	12.67	
50	98.20	1.87	84.50	6.71	80.00	11.99	87.91	8.75	85.00	12.98	
54	94.70	3.56	82.75	.9.58	76.75	11.71	87.00	4.12	83.33	8.25	
58 ्			82.33	8.14	76.75	6.52	84.10	4.66	82.42	6.63	
76	83.70	6.45	73.58	13.18	59.63	12.65	65.09	6.25	68.92	4.10	
80	80.44	7.32	77.50	13.25	62.63	13.10	64.91	6.96	65.08	7.63	
84	77.20	8.43	76.75	13.08	60.25	13.37	69.33	7.28	63.82	10.05	
89	74.70	11.82	75.00	11.56	60.63	12.43	67.13	6.17	66.18	10.44	
93	80.60	8.63	80.67	12.40	68.50	12.46	83.63	5.53	79.64	8.61	
98	89.80	5.67	82.58	11.59	75.13	9.70	88.13	4.79	83.55	6.15	

Table 5. Colonization parameters for percent cover at survey sites.

previous studies elsewhere in Puget Sound are shown for comparative purposes (Greene and Schoener, 1982; Schoener and Schoener, 1981), and similar patterns were found at these localities, too.

3.5 Panel Similarities Between Localities

The similarity between panels at different localities can be evaluated by utilizing a measure of the similarity of all species which takes into account the proportions of individual species on panels. Similarity was computed for each of the 60 panels at various times throughout the study. Panels numbered 1-12 were from Blair Waterway, 13-24 from the Duwamish, 25-36 from Manchester, 37-48 from Milwaukee Waterway and 49-60 from Hylebos Waterway. The similarity of these panels was computed using the Bray-Curtis similarity index. Similarity values were then clustered to produce the dendrograms shown in Figures 5-12. The longer the bar under each locality name the longer the sequence of similar panels. The height of the cluster on the ordinate scale indicates the degree of dissimilarity of the grouping with other groupings. The higher the value at which they cluster, the more dissimilar the group.

Several trends are apparent from these data. First, throughout the course of the experiment all the Duwamish panels clustered together and were more dissimilar from other sites than was the case for any other series. Panels from the Clam Bay, Manchester, reference site clustered together and formed a distinctive group throughout most of the study, and their clustering was more apparent during the later portion of the survey. Commencement Bay panels at each site at first grouped separately, but as time progressed, considerable overlap between sites occurred. Panels which were lost from the study formed an artificial grouping observed in the dendrogram. These trends are examined in more detail below.

Fall, 1980: (Figure 5) The cluster diagram from this date shows that the Duwamish panels clustered together. These panels were dominated by the encrusting bryozoan <u>Membranipora</u>. This series was least similar to the other sites. The Commencement Bay panels were distinctive at each waterway, with considerable similarity between panels in each series. Blair and Manchester shared certain similarities in that both were characterized by moderate hydroid growth and considerable empty space. Milwaukee panels also lacked extensive coverage, but barnacles were present. Hylebos panels clustered together with a colonial tunicate occurring interspersed with arborescent bryozoans and hydroids.

Winter, 1980: (Figure 6) The Duwamish series was again distinctive from the remaining clusters of panels with <u>Membranipora</u> and barnacles equally abundant. Manchester panels all clustered together, characterized by the encrusting bryozoans <u>Reginella</u> and <u>Membranipora</u>, and the calcareous tubes of the polychaete <u>Serpula</u>. Hylebos panels formed two distinctive groups, the more extensive group clustering more closely with the Manchester panels. The colonial tunicate Botryllus/



DISTANCE

Figure 5. Cluster diagram for panels at all study sites in September, 1980, indicating their similarity. Underlined localities show the extent of similar panels.



Figure 6. Cluster diagram for panels at all study sites in December, 1980, indicating their similarity. Underlined localities show the extent of similar panels.

Botrylloides was present at this time, along with a few hydroids and Serpula tubes. Blair panels clustered together and were similar to the remaining Hylebos panels and Milwaukee series. The Serpula polychaete tubes and encrusting bryozoans were present.

Spring, 1981: (Figure 7) The Duwamish series clustered together but had some similarities with a considerable number of the Commencement Bay series. At the Duwamish the encrusting bryozoan <u>Membranipora</u> as well as barnacles were present. Barnacles were also predominant at Milwaukee Waterway along with some coverage by <u>Serpula</u> tubes. These polychaete tubes were also present at the other waterways although at Hylebos the colonial tunicate <u>Botryllus/Botrylloides</u> was important, and at Blair an encrusting bryozoan took up considerable space. Manchester panels were fairly distinctive as a group but clustered with the rest of the Milwaukee panels; both were distinct from the Duwamish and remaining Commencement Bay panels.

Summer, 1981: (Figure 8) The Duwamish series was most distinctive again, characterized by extensive barnacle growth. All Manchester panels clustered together and were distinguished by a low to moderate coverage of the polychaete <u>Serpula</u>, as well as some encrusting and arborescent bryozoans. The Commencement Bay Waterways no longer exhibited distinctive clusters. Instead, the dendrogram was characterized by considerable overlap and short runs of similar panels. A few of the longer runs are shown by underlining.

Fall, 1981: (Figure 9) The Duwamish series clustered together well apart from any of the other sites and barnacles prevailed. The Manchester series was also distinctive and clustered together, although at a lower similarity value. Manchester panels had persistent and moderate polychaete coverage, along with some encrusting bryozoan growth. Hylebos panels were similar among themselves and characterized by solitary tunicates (Corella), barnacles, polychaetes, and encrusting and arborescent bryozoans. Blair Waterway had several similar panels, with moderate barnacle coverage and some encrusting bryozoans. Very few panels at Milwaukee Waterway clustered together.

Winter, 1981-82: (Figure 10) The Duwamish panels continued to be covered by extensive barnacle growth and these panels clustered well apart. The entire Manchester series clustered together, having a moderately high coverage by <u>Serpula</u>. Where some polychaete tubes had dropped off, empty space was renewed for further recruitment. Hylebos panels clustered together and had very apparent similarities with some of the Milwaukee and Blair series. These all had low to moderate barnacle coverage, low <u>Serpula</u> coverage and limited encrusting bryozoan coverage.

Early Spring, 1982: (Figure 11) While the Duwamish panels were covered primarily by barnacles and the Manchester panels by extensive tubeworm growth, there was so much variation in the remaining series as to make their characterization difficult. Considerable overlap occurred between the Port of Tacoma Waterways at this time.



Figure 7. Cluster diagram for panels at all study sites in March, 1981, indicating their similarity. Underlined localities show the extent of similar panels.



DISTANCE

Figure 8. Cluster diagram for panels at all study sites in June, 1981, indicating their similarity. Underlined localities indicate extent of similar panels.



Figure 9. Cluster diagram for panels at all study sites in September, 1981, indicating their similarity. Underlined localities show the extent of similar panels.



Figure 10. Cluster diagram for panels at all study sites in January, 1982, indicating their similarity. Underlined localities show the extent of similar panels.



Figure 11. Cluster diagram for panels at all study sites in March, 1982, indicating their similarity. Underlined localities show the extent of similar panels.

Late Spring, 1982: (Figure 12) Duwamish panels continued to be dominated by barnacles quite distinct from the remaining series. Manchester panels formed the next most distinctive grouping. The Tacoma Waterway panels had some similar series but there was considerable overlap between them, and the clusters were not as extensive as at the other localities.

3.6 Species Abundances

It has been suggested that the log-normal distribution of individuals per species can indicate the pollution stress to which benthic communities are exposed (Gray and Mirza, 1979). Plotting the cumulative percentage of species against the geometric class of individuals, i.e., the number of individuals per species in increasingly large arithmetic classes, at each of the five sampling sites indicated that the experimental and reference sites all showed a distinct break in the curve at the last observation period (Figure 13). The number of geometric classes spanned was 10 at Blair and Hylebos Waterways, 9 at Milwaukee, 8 at the Duwamish and 10 at Manchester. The Duwamish may exhibit a second break in the curve.

3.7 Relationships Between Types of Colonists and Chemical Toxicity

The slope of the colonization curves and the total species richness of the sites are community parameters that showed correlation with the potential toxicity of the sites. Chemical measurements by other investigators of the sediments from the survey localities indicated that the Tacoma Waterways and the Duwamish were considerably higher in their concentration of toxic chemicals than was the reference site at Manchester.

Chemical analyses of the sediments indicated that the Tacoma Waterways had extremely high concentrations of aromatic hydrocarbons (Malins et al., 1982). Hylebos Waterway, slightly seaward of the survey site, had 50,000 ppb dry weight of aromatic hydrocarbons and landward, 21,000 ppb (Malins et al., 1982). Blair and Milwaukee Waterways had considerably lower amounts, but 7,000 ppb (Malins et al., 1982) and 6,500 ppb, respectively, have been measured there. Concentrations of polychlorinated biphenyls (PCBs) were extremely high at Hylebos (150-553,000 ppb dry weight) seaward from the experimental site and 1,200 ppb landward of it. Both Blair and Milwaukee Waterways had considerably lower amounts of PCBs (Malins et al., 1982; Riley et al., 1981).

The concentration of heavy metals in sediments or in suspended particulate matter suggested that the potential for chemical pollution was considerable at the Port of Tacoma and Duwamish Waterways and considerably less so for Manchester. Arsenic in sediments was present at less than 1 ppm dry weight at Manchester (Cummins et al., 1976) and in suspended particulate matter varied from 36-95 ppm in the Duwamish and 42-106 ppm at the Port of Tacoma Waterways (Riley et al., 1980). Less



Figure 12. Cluster diagram for panels at all study sites in May, 1982, indicating their similarity. Underlined localities show the extent of similar panels.



Figure 13. Plots of log-normal distribution of individuals per species at Blair (A), Milwaukee (B). Manchester (C), Hylebos (D) and Duwamish (E) at the termination of the experiments in June, 1982. Geometric class I= 1 individual per species, class II= 2-3, class III=4-7, class IV=8-15, class V=16-31, etc.

than 21 ppm dry weight of copper in sediments was found at Manchester (Cummins et al., 1976) but between 100-500 ppm was present in suspended particulate matter in the Duwamish (Riley et al., 1980) and Tacoma Waterways. Similar figures apply to concentrations of lead at these sites (Cummins et al., 1976; Malins et al., 1982). Less than 1 ppm of cadmium was recorded in sediments at Manchester (Cummins et al., 1976) and nearly an order of magnitude greater concentration at Hylebos. Milwaukee Waterway had twice the Manchester level of cadmium and five times this amount has been recorded at Blair in sediments (Malins et al., 1982).

Water column data showed high concentrations of copper at Hylebos with values reaching 1,637 μ g/l upstream from our Hylebos site (unpublished Environmental Protection Agency (EPA) region 10 data). Purgeable halocarbons were high landward (values to 7,873 μ g/l). Values of 1,423 μ g/l were present seaward of the study site (unpublished EPA region 10 data). Chloroform values of as high as 6,740 μ g/l were present landward and ca. 800 μ g/l seaward from the study site (unpublished EPA region 10 data). Arsenic has also been measured at 12,000 μ g/l landward from this site.

4. DISCUSSION

Several of the study sites in Puget Sound, Washington, differed in the degree and type of toxic chemical concentrations present in the water column or in the benthic sediments. Since studies on individual toxicants have been shown to affect individual species, community properties might also be expected to reflect differences in these parameters. Surveys of comparative colonization on identical substrates were made at these sites. Colonization studies recorded the accumulation of species under varying environmental conditions. During at least the first six months of settlement and growth of attached biotas, species accumulation was greatest at the reference site in this survey and at a relatively uncontaminated site in Elliott Bay, Puget Sound, during an earlier study (Schoener and Schoener, 1981). After the initial period of colonization, the sites with the greatest species richness declined and approached the fairly constant values of panels from the Port of Tacoma Waterways. This decline in species richness may reflect an oscillation of seasonal duration or longer. Osman (1977) has empirically and theoretically shown that in a relatively unpolluted environment near Woods Hole, Massachusetts, epifaunal communities on artificial substrates undergo an oscillatory equilibrium where species richness varies between winter and summer values. A comparably longterm oscillation may have occurred at the Manchester and previously studied Elliott Bay sites (Schoener and Schoener, 1981). This pattern, however, was not as evident at any of the Commencement Bay sites. These results correspond to those reported by Malins et al. (1982) for the recolonization of sediments which were collected, frozen and then repositioned at the Port of Tacoma and Duwamish. Low species richness of the infauna was found in the Tacoma Waterways. Their relatively uncontaminated reference area, Port Susan, had the greatest number of species present. The total species richness at the Port of Tacoma sites may not be as great as at the reference site. Swartz et al. (1982) discovered species of amphipods that responded lethally in bioassays were absent from samples from the Tacoma Waterways. The low number of species at the Duwamish Waterway may have been attributable to either physical or chemical parameters or both. Although panels were positioned in the salt wedge through most of the year, observations made at extreme low tides in June of each year indicated that salinity was considerably reduced at this upriver site. Although the low number of species did not vary seasonally, the decrease in salinity may have reduced species richness at this site.

Other trends that were examined for their potential correlation with chemical conditions in the study sites included functional group analysis of colonists. Solitary organisms because of their hard calcareous skeletons may undergo secondary colonization by other organisms which attach to them and form a secondary layer of organisms that could reduce contact with pollutants for the basal species. Furthermore, solitary colonists can retreat into their skeletons when not actively feeding, thereby avoiding contact with pollutants. Many colonial organisms, however, do not accumulate secondary fouling because of their soft-bodied surfaces, and might be more exposed to pollutants than solitary organisms would be. In the only analysis of its kind, Jackson (1977) determined that in pristine coral reef areas attached organisms on hard substrates showed a continual prevalence of colonial organisms after an initial dominance by solitary organisms. The present survey provided data from contrasting areas in terms of potential pollution differences but which also differed in a wide variety of environmental features. Although it might have been expected that differences between experimental localities would be observed, colonial organisms prevailed early in the initial colonization period at almost all sites in Puget Sound in both this and in previous studies in Elliott Bay (Schoener and Schoener, 1981) and Phinney Bay (Greene and Schoener, 1982). After this initial period, however, solitary organisms tended to dominate panels with different species often dominating at particular sites. There is no obvious difference between relatively contaminated and uncontaminated sites with respect to the prevalence of particular functional groups.

Gray and Mirza (1979) suggested that when plotted on probability paper, the cumulative percentage of species in geometric classes of individual per species gives a straight line under nonpolluted conditions but shows a distinct break in the straight log-normal plot and also covers more geometric classes, where populations were exposed to slight pollution. All of the five sites in this study showed a break in the line. It was later found that a variety of conditions, including season of year and increased pollution load, altered the shape of the curve (Gray, 1980). Larval settlement in spring and summer may also lead to a break in the log-normal similar to that found under slightly polluted conditions. This may best explain the experimental results. The reference site covered the same number of geometric classes as two of the Port of Tacoma sites. Panels at the reference site, Manchester, were typically most similar to each other and distinct from those at the other sites. Panels at the Duwamish were also similar to one another and clustered together. These differences may be related to the location of the sites. Panels at the Port of Tacoma Waterways were at first quite distinctive, but as time progressed, similarities between panels from the different Waterways became more apparent and each series no longer clustered separately.

In conclusion, the differences in colonization patterns which appeared between localities differing in potential pollution severity were patterns which were most evident during the first year of colonization. There was a tendency for the reference site to accumulate a greater number of species than any of the experimental localities during the first six months of colonization. Although colonization rates were not statistically significant between localities at this time the mean number of species accumulating was significantly higher at the reference site than at any experimental locality from six months to a year after the survey was initiated. As the experiment progressed most colonization curves tended to converge, suggesting that initial differences in colonization were obscured by the second year of colonization. In this survey comparative colonization processes best reflected differences in pollution severity during the first year of colonization and their usefulness as an index of pollution severity decreased as community development proceeded into the second year.

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APPENDIX

Community composition and development in terms of the relative abundances of solitary and colonial species on panels are shown for each of the experimental localities (Appendix A-1 through A-5). The upper portion of each figure illustrates the composition of species assemblages determined from cluster analysis of panels. Only the foundation species are included. The coded key corresponds to five relative abundance categories based on mean percent cover. Mean percent cover for a given species was determined from all the panels represented in each species assemblage. The lower portion of each figure lists the species assemblage corresponding with each panel on a given sampling date.

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SOLITARY ANIMALS	ABCDEFG
Balanus crenatus	
Membranipora sp.	
EMPTY SPACE	

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	Date							
Panel	9/80	12/80	3/81	6/81	9/81	1/82	3782	5/82
1	•	В	D	E	E	F	F	F
2	A	В	C	Е	E	Έ	F	F
- 3	A	*	с	F	E	F	C	F
4	-	-	-	-	-	-	-	-
5	-	-	-	-	-	-	-	-
. 6	۸	A	с	F	E	F	C	G
7	*	в	c	F	E	F	G	G
8	A	В	с	F	Е	F	G	C
9	A	в	с	F	E	F	f	F
10	٨	B	D	E	E	P	F	C
11	*	٨	с	P	E	P	F	F
12	٨	В	D	E	E	F	F	P
					- pa * pa	nel lo nel no	st t cens	used

Appendix A-1. Community development at the Duwamish River site .

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SOLITARY ANIMALS	A	8	С	D	Ε	F	G	н	1	J	к	L	M
Balanus crenatus	:.												
Metridium senile		.:::			ſ		· :.	;				. i.	
<u>Mytilus</u> edulis													
Pseudopotamilla occelata						njii							
Serpula vermicularis			·				·	86					*
COLONIAL ANIMALS													
Bowerbankia gracilis				000 1									
Hydroid sp.1					<u> </u>								Ц
Leucosolenia sp.													
Membranipora membranacea		·	888) 1										
Obelia sp.				;: : :									
Reginella nitida													
Scrupocellaria sp.	•		·:.•.		:. •	÷.,		*			:".		
Tunicate sp.1													
EMPTY SPACE	្រា			-		8 2			*	8			
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1-25	26-50	51-75	76-100
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Date

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Panel	9/80	12/80	3/81	6/81	9/81	1/82	3/82	5/82
1	٨	с	٨	G	н	м	м	м
2	D	с	E	F	H	F	E	Ξ
3	D	٨	Ē	C	к	L	L	к
4	٨	c	A	F	۶	м	м	м
5	D	Е	F	ĸ	L	L	L	L
6	D	A	E	H	н	ĸ	м	к
7	D	с	E	G	ĸ	м	к	к
8	D	A	в	н	F	м	м	м
9	٨	I	ĩ	L	t	J	J	J
10	D	A	E	ห	ห	м	м	н
11	D	A	E	મ	์ห	м	н	L
12	В	A	E	G	н	L	L.	L

Appendix A-2. Community development at the Manchester site.

SOLITARY ANIMALS	A	8	<u>C</u>	D	Ε	F	G	н	1	J	ĸ	L	м	N
Balanus crenatus														
Corella willmeriana	÷:::													
Metridium senile														
Serpula vermicularis														
COLONIAL ANIMALS						_								
Bugula pacifica														
Bryozoan sp.1				_	÷									
Callopora horrida					::::.					÷				
Dendrobeania lichenoides												чн 1	:	
Hippothoa hvalina				ŧ:										
Hydroid sp.l		L.,												
Scyphistomae										ille;				
EMPTY SPACE				- (-	- 							98 5.	
						<u>.</u>	-							
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nel	9/80	12/80	3/81	6/81	9/81	1/82	3/82	5/82
1	*	С	В	E	G	C	G	G
2	A	D	B	I	J	J	J	I
3	A	С	с	н	L	м	м	N
4	A	D	c	н	F	к	ĸ	N
5	A	D	D	F	F	к	N	N
6	A	с	-	-	-	-	-	-
7	A	D	с	E	F	к	ĸ	ĸ
8	A	D	D	-	-	-	-	-
9	A	A	В	в	L	в	в	в
10	A	D	с	-	-	-	-	-
11	A	-	-	-	-	-	-	-
12	A	с	D	Ε	C	к	ĸ	н

Appendix A-3. Community development at the Blair Waterway site.

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SOLITARY ANIMALS	A	8	C	D	E	F	G	н	1	J	К	L	M	N
Balanus crenatus					1		:				1			
Corella villmeriana		- :: ^		<u> </u>		Ì								
Metridium senile													-	
Pseudopotamilla occelata				. :	;								A	
Serpula vermicularis							••••	•						
COLONIAL ANIMALS Botryllus sp.		::: :	¢.							:::;`				
Bryozoan sp.1										•	::;:			
Bryozoan sp.2														
Callopora horrida					.:									::.
Cryptosula pallasiana					•								·:	92
Dendrobeania lichenoides				11 -				••••			111			
Hydroid sp.1												ŀ		
<u>Reginella nitida</u>							11.1		••••		[*]		; *	
EMPTY SPACE	2		1. j. f.	1	:	i.	•			8		88		

				Ser.
0	1-25	26-50	51-75	76-100
	A>10			

X.

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	Date							
Panel	9/80	12/80	3/81	6/81	9/81	1/82	3/82	5/82
1	. A	A	٨	A	G	J	J	N
2	A	3	A	Ø	н	н	м	м
3	A	с	A	D	к	J	J	N
4	в	A	A	I	н	I	I	I
5		B	A	D	ĸ	I	к	G
6	в	в	A	D	н	I	ĸ	G
7	A	A	A	D	D	I	r	ท
8	٨	A	A	З	F	н	м	r
9	A	8	A	Е	З	н	м	м
10	в	в	٨	D	۶-	J	J	C
11	A	8	A	L	N	м	L	N
12	в	в	A	D	ĸ	I	ĸ	-
					- pa	inel lo	ost	

Appendix A-4. Community development at the Hylebos Waterway site.

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SOLITARY ANIMALS	A	8	С	D	Ε	F	G	Н	1	J	K	L	Μ	N	0	P	Q	R
Balanus crenatus	: A				*													
Corella villmeriana																-		
Foliculina sp.			<u></u>	::::							L			L				
Pseudopotamilla occelata																		
Serpula vermicularis																: err		
COLONIAL ANIMALS Boverbankia gracilis										<u> </u>								
Bryozoan sp.1																:		
Bugula pacifica				<u></u>									:::··					
Dendrobeania lichenoides															88 S	×		
Bydroid sp.l					<u> </u>			,										
Reginella nitida		<u> </u>		199			- <u> </u> -				анц. С				.:::	•••••		:: : ::.
Yellow sponge spp.*	L																· ••.	
EMPTY SPACE	.	iller,		-14				***	*	*					*			
^e <u>Hallsbondera</u> panista <u>Halislana</u> permatis			E		E	-25	26	<u>-</u> 50	51		76	-100	2					

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	Date							
Panel	9/80	12/80	3/81	6/81	9/81	1/82	3/82	5/82
	٨	F	A	G	Q	ĸ	ĸ	L
2	E	F	F	N	R	I	I	Q
3	A	A	A	o	P	I	I	R
4	A	D	D	N	R	I	I	L
5	с	D	A	0	P	I	I	н
6	B	٨	٨	н	P	I	I	н
7	В	с	D	G	Q	L	-	-
8	B	D	D	C	Q	L	-	-
9	В	с	D	G	P	н -	ห	н
10	В	D	D	c	R	I	L	ĸ
11	с	с	D	-	-	-	-	-
12	с	D		G	Q	I	-	-
•••					— pa	snel la	ost	

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