

# Bacterial Number, Heterotrophy and Extracellular Enzyme Activity in the Bransfield Strait, Antarctica

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**Abstract :** To study the structure and function of bacterial populations in the Bransfield Strait, Antarctica, which is located between 62°~64° S and 56°~62° W, twenty nine sampling stations were chosen. Samples were collected from sea water and sediment during austral summer (Dec. 1989~Jan. 1990) and analyzed for total bacterial number, total saprophytic bacterial number, heterotrophic activity and extracellular enzyme activity.

The number of total bacteria in sea water was between  $1.0 \times 10^4$  cells/ml and  $1.6 \times 10^5$  cells/ml, and total saprophytic bacteria were between  $0.5 \times 10^2$  CFU/l and  $8.0 \times 10^4$  CFU/l in their numbers. The population density of saprophytic bacteria was significantly low, giving less than  $10^{-4}$  of the total bacterial number, in this region. Turnover times of glucose and leucine in sea water were in the ranges of 41 and 2094 hrs, and 56 and 980 hrs, respectively. Turnover times of these organic matters were extremely variable depending on the sampling station and water depth. In the sediments, the enzyme activities of  $\alpha$ -glucosidase, N-acetyl- $\beta$ -glucosaminidase and aminopeptidase of the fast growing bacterial population were higher than those of the slow growing bacterial population, but  $\beta$ -glucosidase activities were higher in the slow growing bacterial population.

**Key words :** bacterial number, bacterial activity, Antarctic Ocean

## Introduction

The Antarctic is an area of outstanding scientific interest. Bacterial heterotrophy, once considered negligible in antarctic water is now emerging as an important pathway of secondary production, and detrital material particularly derived from krill appears to enter the food web at many points (Vincent, 1988). Bacteria in the antarctic environment exhibit a number of interesting properties and additionally play a crucial role in the cycling of nutrients, and many scientists regard marine bacterial metabolism as a principal feature in the regeneration of primary nutrients (Tanner, 1985). From this view, we carried out this study to understand the structure and function

of bacterial populations in this specialized environment.

## Materials and Methods

The locations of sampling stations are shown in Figure. 1. The sampling period was between Dec. 29, 1989 and Jan. 7, 1990, during austral summer. To estimate total bacterial cell number, the epifluorescent microscopic method was used (Zimmermann, 1977). The basic procedures were followed by the recommendations of Cassell (1965), Zimmermann and Meyer-Reil (1974), Daley and Hobbie (1975) and Pomroy (1984). To enumerate total saprophytic bacterial number, the membrane filter method

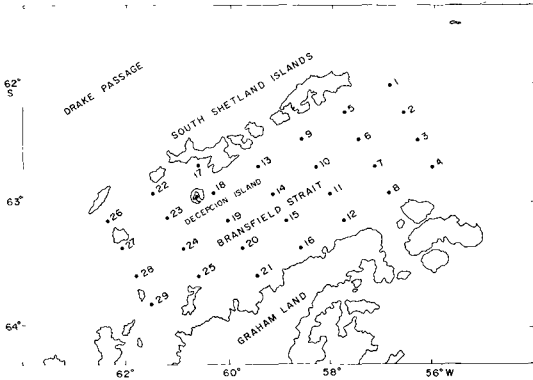


Fig. 1. Map of the Bransfield Strait with Stations 1–29.

(pore size :  $0.45 \mu\text{m}$ ) and plate count method were used for sea water and sediment samples, respectively, using ZoBell 2216E agar medium.

Plates were incubated at  $8^\circ\text{C}$  for 15 days. The extracellular enzyme activities of  $\alpha$ - and  $\beta$ -glucosidase, N-acetyl- $\beta$ -glucosaminidase and aminopeptidase were determined by the method of Kim and Hoppe (1986). For the assessment of microbial activity, turnover time was measured using  $^{14}\text{C}$ -glucose and  $^{14}\text{C}$ -leucine by the method of Williams and Askew (1968).

## Results and Discussion

### 1. Total bacteria and saprophytes

Table 1 shows the numbers of total bacteria and total saprophytes in each station of the Bransfield Strait at different water depths. The number of total bacterial cells varied from  $1.0 \times 10^4$  cells/ml to  $1.6 \times 10^5$  cells/ml. In most

Table 1. Number of total bacteria and saprophytes in each station of the Bransfield Strait.

Station	Sampling Depth (m)	Total bacterial number <sup>1</sup>	Total number of saprophytes <sup>2</sup>	Station	Sampling Depth (m)	Total bacterial number <sup>1</sup>	Total number of saprophytes <sup>2</sup>
1	0	13.6	31	16	0	4.8	17
	30	8.3	11		30	3.9	16
	700	2.2	16		150	4.3	4.9
2	0	7.5	24	17	0	12.9	—
	30	11.3	29		30	6.7	—
	750	6.5	2.5		200	3.0	—
3	0	2.1	15	18	0	8.1	—
	30	2.3	0.5		30	—	—
	200	3.0	3.5		750	2.1	28
4	0	8.0	24	19	0	5.3	—
	30	5.8	8.5		30	7.3	—
	200	4.5	15		700	2.3	95
5	0	5.6	29	20	0	8.9	—
	30	6.6	21		30	4.4	10
	1500	1.3	8.0		100	3.8	31
6	0	9.8	11	21	0	6.0	800
	30	8.2	1.5		30	6.4	11
	1000	1.3	2.5		150	3.4	12

Bacterial activity in the Bransfield Strait

Table 1. Continued.

Station	Sampling Depth (m)	Total bacterial number <sup>1</sup>	Total number of saprophytes <sup>2</sup>	Station	Sampling Depth (m)	Total bacterial number <sup>1</sup>	Total number of saprophytes <sup>2</sup>
7	0	7.0	30	22	0	11.4	—
	30	4.9	3.5		30	6.9	110
	400	2.3	5.5		150	8.6	69
8	0	10.5	25	23	0	4.0	—
	30	8.1	30		20	3.7	140
	100	9.3	18		500	7.0	45
9	0	4.2	62	24	0	8.3	—
	30	1.9	4		30	5.5	—
	1000	4.7	4.5		300	2.4	19
10	0	10.8	36	25	0	5.7	—
	30	9.1	56		30	7.1	—
	750	1.6	3.5		200	4.2	—
11	0	6.5	20	26	0	15.6	—
	30	3.3	1		30	10.6	120
	500	2.3	12		150	7.9	60
12	0	4.5	13	27	0	15.1	100
	30	3.9	11		10	10.1	20
	320	2.8	8		20	7.5	23
13	0	8.1	120	28	0	2.6	—
	30	6.2	10		30	6.3	—
	1000	1.0	7.1		800	2.5	140
14	0	4.4	32	29	0	5.4	—
	30	6.5	39		30	9.2	640
	700	2.1	12.2		750	3.2	67
15	0	7.9	33				
	30	9.6	14				
	75	6.0	3.6				

1 :  $\times 10^4$  cells/ml  
2 :  $\times 10^2$  CFU/l

stations, the total cell number in the upper layer of the water column was higher than that in the deeper layer. The total cell number obtained in the present investigation was found to be similar to that reported by Hanson et al. (1983) for the Drake Passage, in January, with values ranging between  $1 \times 10^4$  and  $2 \times 10^5$  cells/ml but less than 1/10 of that in the Scotia Sea and

coastal antarctic waters (Hanson et al., 1983).

As shown in Table 1, the total saprophyte number was between  $5 \times 10^1$  and  $8 \times 10^4$  CFU/l during the sampling period which is similar to that reported in the same sampling area in 1981 by Zdanowski (1985). Compared with the total bacterial number, the saprophyte number was extremely low, giving less than  $10^{-4}$  of the total

bacterial number, in this region. The ratio of saprophytes to total bacteria in this cold environment is significantly low compared with the ratios in the temperate climatic zone. This difference might be due to the saprophyte counting method and the composition of media as pointed out by Simidu et al. (1986). It was postulated that a large proportion of the bacterial population in the Antarctic waters was in an actively growing state and almost all recovered in the artificial medium.

## 2. Heterotrophic activity

The turnover times for glucose and leucine in seawater samples from the Bransfield Strait are

given in Table 2. These values were in the range of 41–2094 hours and 56–980 hours, respectively and varied depending on the sampling station and depth. The turnover times of these two compounds from 0 m, 30 m and deeper layer were increasing in this order. However, in the case of the station numbers 23 and 27, the opposite pattern was observed. The turnover times of leucine were significantly shorter than those of glucose indicating that leucine could be uptaken and remineralized faster than glucose in the water column of the Bransfield Strait.

A high percentage (>50~70%) of the planktonic bacteria in this marine environment

**Table 2.** Turnover time of glucose and leucine in sea water samples from the Bransfield Strait.

Station	Sampling Depth(m)	Glucose(h)	Leucine(h)
1	0	112.1	92.3
	30	170.1	235.5
	700	835.0	743.3
5	0	88.9	62.3
	30	664.6	499.1
	1500	1703.4	725.9
9	0	90.7	62.3
	30	212.1	192.8
	1000	1628.9	780.9
13	0	214.5	82.6
	30	167.6	110.2
	1000	1663.8	912.8
18	0	178.2	84.2
	30	—	—
	750	530.9	294.6
23	0	1291.8	816.6
	20	1168.2	979.9
	500	515.8	604.8
27	0	2093.7	687.7
	10	161.5	124.8
	20	41.3	55.6

### Bacterial activity in the Bransfield Strait

appears to be metabolically inactive or dormant. In antarctic water, as elsewhere, there is little or no correlation between bacterial DNA synthesis and bacterial cell concentrations (Vincent, 1988). In the present study a correlation between the turnover times of two dissolved organic compounds and the number of total bacterial cells was found, but no correlation of the turnover times with the total number of saprophytes.

The data summarized in Table 3 imply the great variability of heterotrophic activity in different marine areas. Compared with other results obtained from the different regions, the turnover time of glucose in the Bransfield Strait was shorter than the mean values obtained from the Subarctic Pacific (Seki et al., 1972) and the western side of McMurdo Sound

(Hodson et al., 1981). In contrast, much shorter turnover time was reported from the samples of the eastern side of McMurdo Sound and Bossiere Fjord, Antarctica.

Water temperature is not likely to play a important role for the spatial distribution of bacterial heterotrophic potential as long as it remains unfluctuated in the Bransfield Strait. Under this circumstance, the effect of total bacterial cell number and its metabolic activity (active or dormant) together with the concentration of organic nutrients on heterotrophic potential may become dominant.

### 3. Bacterial extracellular enzyme activity

Table 4 shows the percentage of positive colonies with enzyme activities to total colonies during different incubation periods from sedi-

Table 3. Turnover time of glucose at various locations.

Location	Tt (h)	References
Western north Pacific	6,000	Seki et al.(1972)
Kuroshio current	2,700	Seki et al. (1972)
Subarctic Pacific	1,300	Seki et al. (1972)
McMurdo Sound eastern side	116	Hodson et al. (1981)
western side	20,454	
Bossiere Fjord Kerguelen Island (Mussel bed)	0.5~23	Delille and Cahet (1985)
Kiel Bight (Brackish water)	5.1~523	Gocke(1977)
Kiel Fjord (Brackish water)	2.8~63.4	Gocke(1977)
Shimoda Bay (Estuary)	31	Seki et al. (1975)
Tokyo Bay	8.7	Seki et al. (1975)

Table 4. Percentage of positive bacterial colonies showing enzyme activities to total colonies during different incubation periods from sediment samples in the Bransfield Strait.

Station	$\alpha$ -glucosidase			$\beta$ -glucosidase			N-acetyl- $\beta$ -glucosaminidase			Aminopeptidase		
	Within 0-5 days	Within 6-15days	Total period	Within 0-5 days	Within 6-15days	Total period	Within 0-5 days	Within 6-15days	Total period	Within 0-5 days	Within 6-15days	Total period
2	59	64	62	0	47	25	17	5	11	96	79	86
3	96	50	78	0	25	7	0	0	0	100	63	90
6	86	70	82	0	33	16	5	0	4	100	87	94
9	71	63	69	0	30	8	0	0	0	100	77	93
10	96	78	91	2	5	3	0	0	0	62	64	63
11	100	0	98	0	0	0	5	0	4	75	0	67
12	21	10	17	2	7	3	68	32	56	76	50	66
14	100	63	83	0	14	5	55	17	49	63	33	56
15	24	25	24	0	25	3	100	0	85	87	33	85
16	71	8	53	0	0	0	0	0	0	75	27	52
17	54	25	46	0	18	7	15	0	8	85	56	74
18	82	23	57	0	33	11	22	0	8	93	92	92
19	92	92	92	0	76	53	50	2	13	100	94	95
21	97	40	83	0	4	2	0	0	0	84	0	59
22	85	69	80	0	39	14	38	23	35	65	25	54
23	88	100	89	0	14	3	5	0	4	88	29	69
24	88	20	69	0	9	3	6	7	7	100	75	94
25	39	21	36	0	52	15	16	0	11	96	62	86
29	31	18	26	0	20	9	26	0	15	93	56	80
$\bar{X}$	72.6	44.2	54.6	0.2	23.7	9.8	22.5	4.5	16.3	86.2	52.7	76.6
SD	26.8	30.2	29.4	0.9	19.9	12.3	27.9	9.2	23.3	13.3	28.7	15.3

$\bar{X}$  : average value  
SD : standard deviation

ment samples in the Bransfield Strait. The bacteria growing within 5 days were classified as the fast growing population and those growing after 6 days of incubation as the slow growing population. The enzyme activities of  $\alpha$ -glucosidase, N-acetyl- $\beta$ -glucosaminidase and aminopeptidase in the fast growing population were higher compared with those in the slow growing population. In case of  $\beta$ -glucosidase activities, however, the opposite result was obtained implying that the slow growing bacterial population can play a major role for cellulose decomposition in the sediment environment of the Bransfield Strait.

The average values of each enzyme activity increased in the order of  $\beta$ -glucosidase, N-acetyl- $\beta$ -glucosaminidase,  $\alpha$ -glucosidase and aminopeptidase. Among the 4 different enzymes, aminopeptidase showed the highest activity supporting the phenomenon described by Zobell (1946) that most marine bacteria possess high proteolytic activity.

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