

BACTERIAL FLORA IN THE ALIMENTARY TRACT OF *PLEUROPLOCA TRAPEZIUM* (LINNAEUS)

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ABSTRACT

Total aerobic heterotrophic bacterial flora (THB), amylolytic, proteolytic, and lipolytic activities were estimated monthly in the gut of *Pleuroploca trapezium*. The bacterial populations increased from the foregut to hind gut. Maximum number of THB was recorded in May (41×10^9 CFU/g) and the minimum in November (11×10^3 CFU/g). In general, bacterial populations were more abundant in summer months (April-June) and less so in monsoon months (October-December). The bacterial strains *Micrococcus*, *Bacillus*, *Pseudomonas*, *Vibrio*, *Corynebacterium*, Enterobacteriaceae, *Flavobacterium/Cytophaga*, *Aeromonas* and *Achromobacter* were commonly encountered. *Micrococcus* was the dominant group. The role of symbiotic bacteria in the digestion is discussed.

INTRODUCTION

The alimentary tract of marine invertebrates may be intermittently or permanently populated by micro-organisms from the environment. If the bacterial flora of molluscs is pathogenic, then diseases can be transmitted (WHO 1968). The horse conch, *P. trapezium* is a commercially important snail marketed as seafood. Comprehensive knowledge of the microbes in the alimentary tract is quite essential to keep the seafood uncontaminated with pathogenic micro-organisms. Watkins & Simkiss (1990) estimated the bacterial populations of various snails and recorded colonies similar to those found in soil. Since *P. trapezium* occurs in marine environments, which are influenced by river flow and man-made pollution, any fluctuation in the water quality may be reflected in the bacterial flora of this taxon.

The purpose of this study is to provide basic information on quantitative and qualitative occurrence of gut microflora in *P. trapezium* for a period of one year.

MATERIALS AND METHODS

Monthly collections of live *P. trapezium* were made from August 1993 to July 1994 on the south-eastern coast of India. Snails were brought to the laboratory in sterile polythene bags, washed with sterile 50 % sea water, the shells removed, and the soft body dissected aseptically. The alimentary tract was divided into foregut (oesophagus), mid gut (stomach and digestive gland) and hindgut (intestine and rectum). One

g of each gut region was homogenized in 99 ml 50 % sterile sea water. Eight serial dilutions from 10^{-3} to 10^{-10} were made (9 ml sterile blanks). Pour plate technique was employed using Zobell's 2216e marine agar medium, Starch agar medium, Gelatine agar medium and Tween '80 agar medium to enumerate the total viable heterotrophic (THB), amylolytic, proteolytic and lipolytic bacterial forms respectively. Plates were incubated at room temperature (28-30 °C) in triplicate. Representative cultures were collected at random from all incubated plates. Isolated pure strains were obtained after streaking and were stored in nutrient agar slants. Cultures were identified after 24 hrs according to Simidu & Aiso (1962).

RESULTS

There was a remarkable difference in the number and composition of gut microflora associated with the *P. trapezium* during various seasons. The highest bacterial count was observed during summer season (April-June) compared to the pre-monsoon (July-September), post-monsoon (January-March) and monsoon seasons (October-December). Table 1 shows the monthly THB, amylolytic, proteolytic and lipolytic bacterial flora. The gram-positive bacteria, *Micrococcus*, *Corynebacterium*, *Bacillus*, and gram-negative *Vibrio*, *Flavobacterium/Cytophaga*, *Pseudomonas*, *Aeromonas*, *Achromobacter* and Enterobacteriaceae were encountered. *Micrococcus* was the dominant strain (Table 2).

Table 1. Monthly counts (Log₁₀) (August 1993 - June 1994) of total heterotrophic (THB), amylolytic, proteolytic and lipolytic bacterial populations in three regions of the alimentary tract of *P. trapezium* (CFU/g tissue). FG: foregut; MG: midgut; HG: hindgut.

Months	Heterotrophs			Amylolytic			Proteolytic			Lipolytic		
	FG	MG	HG	FG	MG	HG	FG	MG	HG	FG	MG	HG
Aug.	7.66	7.00	7.94	3.08	4.04	4.90	4.30	5.15	6.11	2.30	2.30	3.70
Sep.	6.49	7.45	7.80	2.60	2.60	3.60	3.30	5.00	5.95	2.23	2.48	3.78
Oct.	5.36	6.30	7.38	1.30	1.85	3.00	2.30	3.70	5.30	1.00	1.30	2.30
Nov.	4.04	4.59	5.32	1.30	1.30	1.30	2.11	3.34	2.90	-	-	-
Dec.	7.89	8.17	8.04	5.66	5.41	7.51	6.01	6.25	8.24	5.52	5.88	6.81
Jan.	8.02	8.04	8.73	5.79	6.01	7.48	5.81	5.89	7.67	5.85	6.07	6.05
Feb.	7.52	8.36	8.65	6.00	6.11	6.82	6.07	7.03	7.96	4.60	5.86	6.20
Mar.	8.07	9.16	9.56	6.08	6.89	7.75	7.23	8.10	7.57	5.77	6.32	6.23
Apr.	9.59	10.34	10.48	7.64	9.96	10.15	8.52	8.81	9.23	7.11	8.74	8.30
May	10.13	10.42	10.61	7.25	9.17	9.11	8.49	8.86	10.31	7.26	9.11	9.63
Jun.	9.74	9.80	9.48	6.85	8.56	8.76	8.05	8.82	9.11	6.89	8.48	9.04
Jul.	8.01	8.38	8.53	6.38	7.38	7.85	5.30	7.85	5.26	4.48	4.60	6.20

Table 2. THB, amylolytic, proteolytic & lipolytic bacterial strains isolated (Si) from three gut regions of *P. trapezium*. 1: *Vibrio*, 2: *Flavobacterium/Cytophaga*, 3: *Pseudomonas*, 4: *Aeromonas*, 5: *Achromobacter*, 6: Enterobacteriaceae, 7: *Bacillus*, 8: *Corynebacterium*, 9: *Micrococcus*, 10: Unidentified, T: total. The percentage of strains with indicated activity is shown in brackets.

Si	THB	Amylolytic	Proteolytic	Lipolytic No.
1	3 (4.0)	2 (8.3)	3 (5.8)	5 (16.1)
2	2 (2.7)	1 (4.2)	1 (1.9)	-
3	7 (9.3)	1 (4.2)	4 (7.7)	4 (12.9)
4	2 (2.7)	1 (4.2)	2 (3.9)	1 (3.2)
5	2 (2.6)	1 (4.2)	2 (3.9)	1 (3.2)
6	3 (4.0)	6 (25.0)	2 (3.9)	-
7	1 (1.3)	2 (8.3)	11 (21.2)	7 (22.6)
8	10 (3.3)	2 (8.3)	7 (13.5)	2 (6.5)
9	39 (52.0)	6 (25.0)	18 (34.6)	9 (29.0)
10	6 (8.0)	2 (8.3)	2 (3.85)	2 (6.5)
T	75 (100)	24 (100)	52 (100)	31 (100)

DISCUSSION

Low bacterial counts during the monsoon (Oct.-Dec.) coincided with heavy rain and river run off. The resul-

ting rapid decrease of temperature and salinity could adversely influence the feeding rate of this gastropod. The highest bacterial numbers were recorded in the hindgut followed by midgut and foregut. All digestive processes are completed in the midgut region. The low bacterial counts in the foregut might be caused by the digestive secretions in this region (Kanakasabai, 1985). Heterotrophic bacteria have an active role in the digestion of food and their numbers in various regions of the gut are inversely related to enzyme activity in that region.

This study slightly differs from Kanakasabai (1985), who reported dominance of the Enterobacteriaceae in the guts of 5 herbivore neritids from the Vellar-Coleeroon estuarine complex. In the present investigation of a carnivore, more beneficial forms than pathogenic forms were noted. *Vibrio*, *Pseudomonas*, *Aeromonas* and *Bacillus* are the likely pathogenic. *Micrococcus* was the dominant form, which could contribute to hydrolysis of protein, carbohydrates and fat in all three regions of the alimentary tract.

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