# Generation time of heterotrophic bacteria in fish ponds of West Bengal.

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Generation of heterotrophic bacteria in water and sediment of fish ponds was measured. The average generation time of heterotrophic bacteria of sediments was relatively shorter than that of water bacteria in these ponds. The generation time of bacteria was variable in different months but no significant difference in generation time was found among various culture systems of fish ponds.

Key Words: Generation time, heterotrophic bacteria; fish culture ponds, trophic status

### INTRODUCTION

Heterotrophic bacterial community prevails in aquatic ecosystems and plays a major role in nutrient releasing from organic matter in water. Godlewska-Lipowa (1972 a, b, and c) has observed a correlation between the generation time and the density of bacterial population and also between the intensity of increase in number of bacterial community and the organic matter present in water. The present investigation has been aimed to study the range of variation of generation time of heterotrophic bacterial population in fish ponds of West Bengal, India, under the organic loading in water.

# MATERIALS AND METHODS

The investigation was carried out in six fish ponds which were used as polyculture, monoculture and traditional systems of fish culture. Three ponds are located in the district of Nadia and other three are located in the district of North 24-Parganas.

The water area of these ponds ranged from 0.04 ha to 6.4 ha and their depth ranged from 1.0 m to 3.6 m respectively. The polyculture ponds were stocked with three species of Indian major carps and three exotic fishes. In monoculture system of ponds *Cirrhinus mrigala* were stocked. The traditional ponds were stocked with three Indian major carps without maintaining any ratio and these ponds naturally

harboured a number of stray fish species like *Punctius ticto*, *Channa* sp., *Anabas* sp. *Clarias battrachus*, *Heteropneustes fossilis* etc. All these fish ponds were properly managed for fish culture excepting two traditional ponds.

Both water and bottom sediment from six ponds were collected twice in month in sterilised glass bottles (125 ml capacity) for bacteriological culture.

The generation time of heterotrophic bacteria was determined by culturing them in nutrient agar medium. The initial populations of heterotrophic bacteria of water samples were obtained. Filtered water samples avoiding phyto- and zooplankton populations were incubated in the temperature of pond water for 24 h after which the final populations were estimated. Mud water suspension was prepared by mixing 1 g bottom sediment in 99 ml sterile water. Bacterial cultures were incubated at 37° C for 72 h.

The generation time of heterotrophic bacteria was determined according to the method described by Rodina (1972). The generation time (g), generation number (n) and percent increase were calculated according to Iwanoff (1955) as follows:

$$g = \frac{0.301 \text{ t}}{\log b - \log a}$$
  $n = \frac{\log b - \log a}{\log 1.8}$ 

% increase = 
$$\frac{b-a}{a} \times 100$$

where t = time interval in hour; a = number of bacteria at the beginning of experiment; b = number of bacteria at final hour; 1.8 = a coefficient taking into account 10% bacterial mortality in the population.

#### RESULTS

The generation time of viable heterotrophic bacteria of these fish ponds ranged from 6 to 433 hrs. The generation time of water bacteria, on an average, was relatively higher (39.8 hrs) than the sediment bacteria (22.8 hrs). The generation time, however, did not differ from one farm to another ( $F_{1,2} = 5.80$ ; P > 0.05). Likewise, no significant difference was observed between the ponds (Table 2) of three cultural types ( $F_{2,2} = 1.08$ ; P > 0.05) although the generation observed in traditional ponds (48.4 hrs) was higher than the monoculture (29.5 hrs) and polyculture ponds (41.7 hrs).

The seasonal variations of generation time of heterotrophic bacteria were less pronounced in these fish ponds ( $F_{27.54} = 1.0$ ; P > 0.05).

The magnitude of seasonal differences was maximum in Kalyani ponds (7.2 fold) than in Naihati ponds (3.64 fold). This suggests that the ecological condition were more stable in the latter ponds than in former.

The seasonal trends in three cultural ponds of Naihati

were represented by using the model of harmonic functions ( $r \ge 0.664$ ; P < 0.001) (Table-1), while the running average was good fit to the data in fish ponds of Kalyani.

In general, the generation time was relatively shorter in sediment bacteria. There was no difference in the generation time of heterotrophic bacteria between two farm sites ( $F_{12} = 4.24$ ; P > 0.05). The sum total of generation time was 1794.4 and 2039.6 hrs in the farms of Kalyani and Naihati, respectively. Comparison of mean generation time revealed no significant differences (P > 0.05) among three cultural systems ( $F_{22} = 5.8$ ). The generation time (20/hrs) observed in the polyculture system was about 23% lower than the values (26 hrs) found in traditional ponds. The generation time found in monoculture ponds was 22.2 hrs. The statistical treatment of data values (F<sub>22</sub> = 5.8; P > 0.05).

The generation time of heterotrophic bacteria was variable in different months of the year ( $F_{27.54}$ =1.79; P < 0.05). The development of seasonal peak of generation time in heterotrophic bacteria was obviously related to the abundance of viable heterotrophic bacteria.

The running average frequency (Table 1) was the good fit to the data of seasonal changes of generation time of heterotrophic bacteria in all the fish ponds except Monoculture-1 in which the seasonal trend was fitted with sine and cosine waves of periodic functions.

Table 1: A. Coefficients of harmonic functions generation time of heterotrophic bacteria: water and sediments

N				Pond Water			Pond Sedime	Pond Sediments	
Months		Coefficients	Poly-2	Mono-2	-Tradi-2		Mono-I		
July		a	24.680	28.625	30.194		20.819		
August		a,	1.447	-6.516	-6.051		-5.901		
September		a,	-2.069	6.180	12.777		-3.888		
October		a,	0.500	5.722	-0.583		0.805		
November		$a_4$	-3.597	5.708	5.138		0.388		
December	1 10	a,	0.052	-2.955	-1.868		-().32()		
January		a <sub>6</sub>	1.986	-().347	-1.611		-0.236		
February		b	0.464	1.388	-3.584		2.914		
March		Ь,	-1.756	-9.117	-2.453		-2.742		
April		b,	1.916	-2.916	2.694		1.666		
May		b <sub>4</sub>	2.574	0.360	-4.041		-0.817		
Júne	Ÿ	b,	1.619	-4.721	-().553		1.085		
Spearman's ranks			0.769	0.713	0.860		0.846		
Correlation									
Coefficient (y s)			p<0.01	p<().()1	p=0.01		p=0.01		

B. Moving points of the moning averages: water & sediments

	Pond Water			Pond Sediments					
	Poly-1	Mono-1	Tradi-1	Poly-1	Poly-2	Mono-2	Tradi-1	Tradi-2	
Moving									
points :	4	3	3	3	3	3	5	3	

Table 2: Analysis of variance: Generation time of heterotorphic bacteria

Source	df	MS		Variance Ratio (F)		Significance	
		Water	Sediment	Water	Sediment	Water	Sediment
Form Site (R)	1	27591.70	357.88	5.80	4.249	P>0.05	P>().()5
Culture (C)	2	5175.07	492.34	1.00	5.846	P>0.05	P>().()5
Error - A	2	4756.46	84.21				
Session (T)	27	3458.59	166.37	1.009	1.79	P>0.05	P>0.05
Interaction (C X T)	54	3263.28	125.18	0.952	1.35	P>().()5	P>(),()5
Interaction (R X T)	27	2980.37	209.15	0.869	2.25	P>0.05	P<0.01
Error - B	54	3426.02	92.71				
Total	167						

The generation time of heterotrophic bacteria in these sediment samples were found to be independent of the effect of interaction between culture practice and season ( $F_{54.54} = 1.35$ ; P > 0.05), whereas the interaction between farm site and season ( $F_{27.54} = 2.25$ ; P < 0.05) had a strong effect on the generation time of heterotrophic bacteria.

# **DISCUSSIONS**

The generation number and generation time of bacteria are often considered as index of population reproduction intensively (Godlewaska-Lipowa, 1976). The range of variation of generation time of heterotrophic bacteria in these experimental fish ponds (sediments; 7 to 83 hrs, water; 6 to 433 hrs) was longer than generation time (2.8 to 37.9 hrs) for heterotrophic saprophytic bacteria of water in some mono- and polyculture ponds in the U.S.S.R. (Antipchuk and Jana. 1978) because of trophic status differences. However, the generation time of bacterioplankton was considerably higher (134' to 755 hrs) in a small eutrophic lake in Sweden (Coveney et. al., 1977). According to Kaplan and Bott (1983), the mean generation time of bacteria in sediments ranged from 12.5 to 46.2 hrs at 15°C. It has been shown that the generation time of bacterial population was strongly dependent upon organic loading in the water (Godlewaska-Lipowa, 1970, 1972b, 1976; Jana et. al. 1980) and water temperature (Godlewaska-Lipowa, 1972 c). Further, the generation time of bacteria was inversely correlated with the initial density of bacteria (Godlewaska-Lipowa, 1972 a).

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