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HETEROPHIC BACTERIA DECOMPOSING POLYSACCHARIDES IN FRESH-WATER LAKES

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Aerobic heterotrophic bacteria decomposing various polysaccharides such as cellulose, chitin, starch, pectin and lingnin have been isolated from two fresnwater lakes namely, Keoladev National Park, Bharatpur and Ramgarh Lake, Jaipur. The former is an eutrophic and the latter an oligotrophic lake. The bacteria isolated from the lakes were pure cultured and were identified to generic and specific levels whereever possible. Out of the eighteen forms isolated five belonged to the genus *Micrococcus*, three *Flavobacterium*, two *Pseudomonas*, two *Corynebacterium*, two *Vibrio* and one each of *Achromobacter*, *Zoogloea Bacillus* and *Cellulomonas*. It was found that cellulose was decomposed by thirteen forms, pectin by seventeen forms, chitin by thirteen forms, starch by five forms and lignin by none.

Introduction

Polysaccharides such as cellulose, chitin, pectin, starch and lignin constitute one of the basic structural materials of animal and plant cell walls. These materials are released into the water bodies such as lakes and ponds by the death and decay of the plant and animal residues or brought in from various other sources along with the rain water. The decomposition of these polysaccharides is necessary for the normal functioning of the ecosystem as the decomposition leads to energy flow, mineralization and nutrient recycling. Work on decomposition of various polysaccharides in the water bodies has been attempted in the past (Kormonday, 1968; Laurent, 1969; Hofsten and Edberg, 1972; Zdanowskii, 1977; Patrica and Tracey, 1956; Fjerdingstad, and Fjerdingstad 1978 and Parvateesam and Narayana, 1984). In the present investigation heterotrophic bacteria decomposing various polysaccharides from two freshwater lakes of Rajasthan is being reported for the first time.

Materials and Methods

Water and lake bottom mud samples from Keoladev National Park, Bharatpur and water samples from Ramgarh lake, Jaipur were collected in sterile polythene bottles and were brought to the laboratory for the isolation of various bacterial forms. These bacteria were further purified and were identified using Bergey's (1957) determinative tests. These bacteria

were tested using various media (pH-6.8) as given below to know their ability to decompose various polysaccharides. All the tests were made on prepoured plates by spread plate technique.

Cellulolytic activity—To detect cellulolytic activity the medium described by Mc Intyre and Hankin (1978) was used. After a period of 6 to 8 days incubation the plates were flooded with 1% aqueous solution of hexadecyltrimethyl ammonium bromide. This reagent precipitates intact cellulose and thus a clear zone around the colony in an otherwise opaque medium indicated degradation of cellulose.

Chitinolytic activity—Chitinolytic activity was determined with the medium described by Hankin and Anagnostakis (1975). After 5 to 8 days of incubation clear zones were seen in the opaque ager around colonies able to degrade chitin.

Pectolytic activity—The medium described by Hankin et al. (1971) was used to detect pectolytic activity. The plates were incubated for 3 to 5 days and then flooded with 1% aqueous solution of hexadecyltrimethyl ammonium bromide which precipitates intact pectin in the medium and thus clear zones around a colony in an otherwise opaque medium indicated degraded pectin.

Amylolytic activity—The ability to degrade starch was used as the criterion for the determination of ability to produce amylolytic enzymes. The medium used contained Nutrient Agar 0.2% soluble starch. After 3 to 5 days of incubation the plates were flooded with an iodine solution. A yellow zone around colony in an otherwise blue medium indicated amylolytic activity.

Lignolytic activity—Mineral salt solution medium with Agar Hankin *et al.* (1971) and lignin powder incorporated in to the medium was used to detect lignolytic activity. The plates after 10 to 15 days of incubation were flooded with phloroglucinol and HCl mixture. This solution gives typical pink coloration when comes in contact with lignin. The areas where lignin is degraded does not produce this colour.

Results and Discussion

The ability of the bacterial species isolated from the two water bodies to produce enzymes on solid media is shown in table 1. The word enzyme production is here intended to mean both synthesis of the enzyme by the bacteria and the ability of the enzyme in the medium after its production. From the data in the table it is clear that some bacteria *Pseudomonas putida*, *Vibrio* sp. 2 and *Cellulomonas* sp. produced cellulase, pectinase, chitinase and amylase. While the

Table-1 : Bacterial isolates from the freshwater lakes producing extracellular hydrolytic enzymes on solid media.

Bacterial forms	Enzymes produced				
	Cellulase	Chitinase	Lignase	Pectinase	Amylase
Micrococcus Conglomeratus	□	*	*	*	□
M. Varians	*	□	*	□	*
M. agilis	□	□	*	□	*
Micrococcus sp. 1	□	□	*	□	*
Micrococcus sp. 2	*	□	*	□	*
Achromobacter sp.	*	□	*	□	*
Zoogloea sp.	*	□	*	□	*
Flavobacterium sp.	□	□	*	□	*
Flavobacterium aquatile	□	□	*	□	*
F. difusum	□	*	*	□	*
Pseudomonas putida	□	□	*	□	□
Pseudomonas sp.	□	□	*	□	*
Corynebacterium sp. 1	□	□	*	□	*
Corynebacterium sp. 2	□	*	*	□	*
Bacillus megaterium	□	*	*	□	□
Vibrio sp. 1	□	*	*	□	*
Vibrio sp. 2	□	□	*	□	□
Cellulomonas sp.	□	□	*	□	□

□—Indicates production

*—Indicates non-production.

others *Flavobacterium aquatile*, *Pseudomonas* sp., *Corynebacterium* sp. 1 *Flavobacterium* sp., *Micrococcus* sp. 1, and *Micrococcus agilis* produced cellulase, pectinase and chitinase. *Bacillus megaterium* produced cellulase, pectinase and amylase. *Flavobacterium diffusum*, *Corynebacterium* sp. 2 and *Vibrio* sp. 1 produced only cellulase and pectinase, *Zoogloea* sp., *Achromobacter* sp., *Micrococcus* sp. and *Micrococcus varians* produced pectinase and chitinase. *Micrococcus conglomeratus* produced cellulase and amylase.

In the earlier studies on cellulose decomposition Rodina (1967) reported cellulolytic bacteria from humified lakes of Russia. Koromondy (1968) found that degradation of cellulose increased oxygen consumption. Berg *et al.* (1968) isolated Vibrios that degrade cellulose. In the present study it was found that as many as 12 bacteria from Bharatpur and 5 bacteria from Ramgarh lake out of 18 forms isolated decomposed cellulose. It was further found that out of 18 bacterial forms only one from *Micrococcus conglomeratus* isolated from the lake water of Bharatpur did not utilise pectin.

There are several reports of chitin degrading bacteria from marine environments. However from the freshwater bodies the reports are few. Out of the total 18 bacterial forms isolated as many as 17 forms readily decomposed this polysaccharide. None of bacteria under study could use lignin even after prolonged incubation periods.

Rodina (1967) and Jones (1979) reported bacteria degrading starch from lakes. In the present study it was found that only 5 forms out of 18 forms isolated degraded starch.

Such type of studies on hetrotrophic aquatic bacteria producing various enzymes that decompose the polysaccharides will be very useful in identifying the lakes with the type of polysaccharide input and also in the classification of lakes on the basis of the predominant bacterial types which decompose a specific or a group of polysaccharides. It will also help in the better management of the water bodies which have recently become health hazards due to increased human activities.

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