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MICROBIOLOGICAL INVESTIGATIONS IN INDIAN COASTAL WATERS AND THE INDIAN OCEAN*

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ABSTRACT

The past two decades have witnessed several marine microbiological investigations being carried out by different workers on the east and west coasts of India. The enumeration of the heterotrophic bacteria of the surface and bottom waters, muds and plankton tows and of certain physiological groups had been carried out off the coast of Madras, in the Palk Bay and the Gulf of Mannar. The nature of the bacterial flora associated with sea water and fishes off Malabar Coast had also been worked out though less exhaustively. The quantitative as well as the generic distribution of bacteria in sea water off Cochin and in fresh fishes caught therein has recently been studied. Marine bacteria producing B12 and certain antibiotics had been isolated from the sea water and marine muds off Bombay Coast. Investigation on marine yeasts was first done in Bombay waters and subsequently studies were made in Vellar Estuary and the near-shore waters of Porto Novo. The distribution of yeasts in the Indian Ocean has been studied recently during the International Indian Ocean Expedition. The occurrence of marine bacteriophages had been reported earlier at Bombay and the recent International Indian Ocean Expedition has met with bacteriophages in the bottom and off East African Coast. Valuable information on the geographical distribution, movement and the biochemical activities of bacteria inhabiting the Indian Ocean has been provided by some of the earlier Soviet Oceanic Expeditions.

INTRODUCTION

IT is well known that marine environment harbours bacteria of diversified character which influence the chemical and biological condition of the sea. In contrast to other major habitats, the marine environment is characterised by its vast area and volume, relatively high salinity, its low content of organic matter, low temperature and high pressure at great depths. Under these conditions too, bacteria exist and bring about certain transformations of organic and inorganic constituents of the sea. The breakdown of complex compounds like cellulose and chitin, the transformation of phosphorus and sulphur compounds and the interconversion of salts of nitrogen are but a few of the processes which determine the fertility of the sea.

Investigations so far carried out by bacteriologists all over the world have established the existence of a bacterial flora autochthonous to the marine environment. The review on the subject by Zo Bell (1946), the Symposium in Marine Microbiology held in 1961 (Oppenheimer, 1961) and the monography by Kriss (1963) give an account of the advance made in this field.

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It is pertinent to mention here that a fairly good proportion of the available data on marine bacteria has resulted from studies on spoilage of sea fish. A large volume of literature is available on the nature of the commensal flora of fishes and other marine animals habitating the Northern temperate waters (Stewart, 1932 ; Reay and Shewan, 1941 ; Liston, 1956, 1957 ; Georgala, 1958 ; Reed and Spence, 1929 ; Hunter, 1920 ; Liston, 1959). These studies have substantially contributed to the knowledge of the bacteria of marine origin.

The microbiological investigations of the open seas and oceans have been carried out by Soviet scientists. Starting in early forties, Soviet vessels have explored World Oceans for this purpose. The expedition on the OB conducted by Soviet scientists have collected a large mass of microbiological data on World Oceans including the Indian Ocean. During the more recent International Indian Ocean Expedition, vessles METEOR and ANTON BRUNN had collected interesting data on micro-organisms in the Indian Ocean.

The present review purports to give an account of the microbiological investigations of the coastal waters of India and of the Indian Ocean in its entirety.

OCCURRENCE OF HETEROTROPHIC BACTERIA IN INDIAN COASTAL WATERS

East coast : The earlier studies in India on marine bacteria have been carried out incidental to investigations on spoilage of fish. Studies were confined to specific areas at distances not far from the coast. The earliest studies were started in 1948 on the bacteriology of sea water off the coast of Madras (Velankar, 1950). These studies revealed following features. The average bacterial count per ml of sea water ranges from a few hundred to about 1000 in surface sea water, greater numbers were being associated with preponderence of phytoplankters ; lowering the nitrate concentration resulted higher bacterial counts and during part of the year, the environment was subjected to the influence of land drainage.

The next scene of activity was the Gulf of Mannar and the Palk Bay areas at Mandapam. These areas were practically free from terrestrial influences because of scarcity of human population and absence of land drainage. Studies here included enumeration of bacteria associated with plankton low samples, the aerobic and anaerobic counts of the bottom mud and the determination of the physiological groups of bacteria. Many bacterial types isolated during these investigations have been described in detail (Velankar, 1957). The studies revealed that surface sea water contained a bacterial load of the order of few hundreds per ml ; water from near bottom had a lower bacterial population. The plankton tows and mud samples contained very high numbers of bacteria. Magnitude of the bacterial population in the Palk Bay was found to be related to the phytoplankton content. The mud contained more aerobes than anaerobes. Physiological groups like nitrifiers and denitrifiers, sulphate reducers and agar digesters were isolated from sea water, plankton and mud. A high prevalence of gram negative asporogenous motile rods with a low count of gram positive types and absence of bacteria producing gas from sugars were the salient features of the bacteria of these waters. Studies were also conducted by Venkataraman and Sreenivasan (1956) in the marine environs off Tuticorin. Surface and bottom sea water samples were analysed for total bacterial count and for certain physiological groups such as denitrifiers, agar, alginic acid, cellulose and chitin digesters.

Some preliminary studies on the distribution of yeasts on the east coast were carried out in Vellar Estuary (Seshadri, 1966). Subsequently the seasonal variation in the distribution of yeasts in the near-shore waters off Porto Novo at 10 fathom line was studied (Ramamurthy and Seshadri, 1967). These workers noted the occurrence of black yeasts in the waters. Non ascosporogenous yeasts were found to be more abundant than ascosporogenous yeasts.

The bacteria associated with fresh fish were very largely gram negative rods. It was also found that fishes taken from shore seine catches showed the presence of gram positive sporeformers. For fishes like shark taken by hook and line, the spoilage flora were found to contain entirely of gram negative rods. The absence of *Coryne bacterium* was noted in the Mandapam area unlike the Australian environments investigated by Wood (1953). The influence of environmental flora on the fish flora was thus thought to be different in the two cases. Further the spoilage rates of fishes at refrigeration temperature were lower than those recorded for fishes of temperate waters. This has been attributed to the greater susceptibility of bacteria of the tropical waters to the effect of cold as compared with the bacteria of temperate waters. The bacterial flora of pearl oysters (*Pinctada vulgaris*) and chanks (*Xancus pyrum*) were found to contain chitin digesters (Venkataraman and Sreenivasan, 1956). The occurrence of bacteria tolerating 750 to 1000 ppm of copper sulphate was also noted on the chanks and oysters.

West coast : The surface inshore waters off Tellicherry Coast and the flora of the Indian Mackerel (*Rastrelliger kanagurta*) were examined (Venkataraman and Sreenivasan, 1954). *Micrococci* was dominant (about 50%) in the sea water. The microflora of mackerel consisted of large numbers of *Bacillus* and *Micrococci*.

The bacterial flora associated with Bombay-duck (*Harpodon nehereus*) caught off Bombay Coast consisted of a high incidence of gram negative rods with a smaller number of cocci, gram positive rods and sporeformers (Bhat and Albuquerque, 1953) Gandhi (1962) examined the production of vitamin B_{12} by marine bacteria. One hundred and three out of 130 cultures isolated from sea water samples collected 5 miles off Bombay were found to produce vitamin B_{12}. Production of antibiotics by bacteria isolated from sea water off Bombay has also been reported. The occurrence of marine bacteriophages was reported by Chaina (1963) at Bombay. The incidence of yeasts in the waters off Bombay has been reported (Bhat and Kachwalla, 1955). Asporogenous yeasts predominated. The most common genus was found to be Candida.

Detailed work on the quantitative and qualitative aspects of the bacterial flora of fresh sardines (*Sardinella longiceps*) has been carried out in the Cochin area (Karthiayani and Mahadeva Iyer, 1967). Plate counts at room temperature showed a seasonal variation when regular samplings were carried out over a period of 12 months. The bacterial counts of the guts ran parallel with those of the skin but at a higher level of magnitude. Qualitatively, the analysis of 360 cultures of bacteria isolated from the skin and muscle and 100 cultures from the guts during a year's study showed a high preponderance of gram negative rods, mainly of *Achromobactor*, *Vibrio* and *Pseudomonas* groups. The percentage of gram positive organisms was very low or nil at times in ocean fresh sardines. Seasonal variations in the luminiscent bacteria in guts and in the biochemical characteristics of the microflora in general associated with sardines were observed ; biochemically less active groups being present during the month of December and January (Karthiayani and Mahadeva Iyer, under publication).

Sea water and bottom and samples collected off Cochin Coast were subjected to bacteriological analysis. These samples showed a predominance of gram negative rods. More gram positive organisms were found associated with bottom mud. Occurrence of gram positive organisms in prawns was more frequent than in sardines. The generic distribution of the microflora of the sea water bottom mud, fishes and crustaceans showed wide variation. However the floral composition of bottom mud showed more or less a regular pattern with the paucity of *Vibrio* and a constant presence of gram positive organisms.

The Indian Ocean

During the oceanographical expedition of OB in 1956-1957, extensive investigations were carried out on the distribution of heterotrophic micro-organisms in the Indian Ocean. Microbiological stations included all geographic zones from Antarctic Coast to the northern tropical regions in the Bay of Bengal. The area from west to east covered from the southern tip of Africa to the 98°E.

A total of 1117 water samples from various depths were collected and analysed. Starting from the Antarctic and subantarctic regions, the count of heterotrophic increased considerably as one approached the tropical zones. This increase was particularly rapid along the section 93-98°E than along that of 20°E meridian. It was observed that there was a lack of variety in cultural characteristics of the colonies growing on the membrane filters, apparently indicating a scarcity of species in the waters rich in heterotrophs. In the subantarctic and Antarctic regions the water samples, though containing a lower count, produced a greater variety of colonies, indicating richness of species.

Out of 1058 cultures isolated from the water samples more than 700 cultures consisted of asporogenous motile and non-motile rods. Thus in the Indian Ocean also, just as in other oceanic areas, the asporogenous rods appear to constitute the predominant part of the microbial flora. The large numbers of such heterotrophic bacteria on the surface and in the deep waters of the trophical Indian Ocean are suggestive of the waters having a very high proportion of easily assimilable organic substances. Regarding the biochemical capabilities of these heterotrophs, the tropical Indian Ocean suffers from a scarcity of biochemically active microbial forms whereas the waters of the high latitudes of the ocean abound with biochemically active ones. A possible explanation of this phenomenon is suggested (Kriss, 1962). The same author has suggested the use of microbiological indicators in the study of movement of water masses in the depths of the Indian Ocean from tropical areas to high latitudes and vice versa (Kriss, 1963).

During the Meteor Expedition in the Indian Ocean during 1964-1965, data of the bacterial distribution of four sections, each about 200 miles long and perpendicular to the west coast of India were collected (Anon, 1960). The bacterial numbers increased from west to east and from south to north. Bacterial counts from a station in the Gulf of Aden were compared with values of organic bound carbon (particulate and total). In addition, data of the distribution of bacteria in a deep sea core are presented.

Distribution of Yeasts and Molds

Yeasts and molds are collected from 16 stations during the cruise 3 of the R.V. Anton Brunn for the U.S. programme in Biology, International Indian Ocean Expedition (Fell, 1967). The section investigated extended along the 60°E meridian from 11°N to 40°S at a depth of 0-2000 metres. The yeast counts ranged from 0 to 513 cells/liter. The greatest of concentrations were noticed at the Somali current and Antarctic Intermediate waters. A total of 25 species of yeast was isolated. The commonly isolated species were *Rhodotorula rubra* and *Candida atmospherica*. The relationship of yeast distribution to hydrographic and biological condition is discussed. The molds occurred in the proximity of terrigenous material.

Occurrence of Marine Bacteriophage

15 deep bottom mud samples collected, during the 8th cruise of the R.V. Anton Brunn taking part in the International Indian Ocean Expedition, were screened for bacteriophages using bacteria from these muds as host. Only one mud sample collected off the coast of East Africa from a depth of 300 metres showed plaque formation against bacteria isolated from the same mud (Johnson, 1968).

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