INHIBITION OF BACTERIA FROM MARINE SOURCES BY AUREOMYCIN*†

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INHIBITION of marine bacteria by antibiotics is engaging the attention of researchers in the fields of marine biology and fisheries during recent years. Spencer (1952) used antibiotics in the isolation of bacteria-free cultures of marine phytoplankton organisms. Shewan et al. (1954) suggested a method for rapid differentiation of asporogenous rods commonly occurring in the marine environment based on the differences in their sensitivity to certain antibiotics. Oppenheimer (1955) in his studies on the effect of marine bacteria on the development and hatching of pelagic fish eggs, employed various antibiotics including aureomycin, singly and in different combinations, for controlling bacterial growth. Tarr et al. (1950) studied the effect of several antibiotics in retarding the spoilage of fish stored in ice and reported aureomycin to be most effective in low concentrations.

The bacteria which cause spoilage in sea fish are largely of marine origin, though other bacteria might also be introduced through handling. Further, certain bacterial genera, i.e., *Achromobacter* and *Pseudomonas* appear to be more significant than the others in fish spoilage (Shewan, 1949; Wood, 1940). In view of the promise held forth by aureomycin as a possible preservative for fish it is of practical importance to know to what extent marine bacteria are affected qualitatively and quantitatively by this antibiotic. In this connection, the inhibiting action of aureomycin in different concentrations on the growth of a number of bacterial species isolated from the marine environment and from sea fish, and on the bacterial population of sea-water, was studied. The results are reported in this paper.

MATERIALS AND METHODS

The bacterial cultures used for these studies were from the stock of well-documented types isolated in this laboratory (Velankar, 1957). A freshly
prepared concentrated solution of aureomycin hydrochloride was sterilised by filtration and used for incorporating in agar medium. After melting the agar the aureomycin solution was added aseptically at 42°C. rapidly and slopes prepared. A loopful of growth on agar of the stock culture was taken up in 5 ml. of sterile saline (1% NaCl in distilled water) and a loopful of the suspension was streaked on aureomycin-treated and control agar slopes. In the case of the organisms which required sea-water or 3% NaCl for optimum growth sea-water agar was employed; for the rest of the cultures fresh-water agar was used.

The growth appearing on the inoculated slopes was observed every twelve hours.

Sea-water samples were collected in sterile 1 lb. glass bottles from about 2 miles off the shore in the Palk Bay at Mandapam.

The stability of aureomycin at the level of 2 p.p.m., i.e., the lowest concentration employed in these studies, in sea-water agar at room temperatures was determined empirically as follows: Sea-water agar slopes containing 2 p.p.m. of aureomycin hydrochloride were prepared as described above; on each successive day one of these and a control agar slope (containing no aureomycin) were inoculated with an organism which was known to be very sensitive to 2 p.p.m. of aureomycin. The growth appearing on the control and the aureomycin treated slope was observed after 24 hours. The absence of growth on the test slope and the presence of good growth on the control slope was taken as the indication of the stability of the antibiotic at room temperature in sea-water agar medium.

RESULTS

The results are shown in Tables I to V.

With the exception of two or three all the bacteria were sensitive to aureomycin. Approximately 40% were sensitive to the level of 2 p.p.m., 30% to 5 p.p.m. and 30% to 20 p.p.m. In the case of those sensitive to 2 p.p.m. the lag period (Table I) varied from ½ to 4 days; in those sensitive to 5 p.p.m. from 1 to 3 days and in those sensitive to 20 p.p.m., the period was about 4 days. Bacteria sensitive to 2 p.p.m. included spp. of Gram-negative achromic rods (motile with polar or peritrichous flagellation or non-motile), Vibrio, Flavobacterium, Bacterium and Bacillus and an agar digester resembling Cytophaga. Yellow, red and violet pigment producing polar flagellated rods and denitrifiers belonging to the Pseudomonas were sensitive to higher levels, i.e., 5 and 20 p.p.m. The cocci were more resistant than the Gram-negative rods. In the Bacillus the degree of sensitivity varied greatly; the isolate