Effect of Storage on Enterotoxigenic *Staphylococcus aureus* in Cured Fish

S.Sanjeev and P.K.Surendran
Central Institute of Fisheries Technology, Cochin - 682029, India

On a global basis about 14% of the marine fish landings are processed by curing and in India consumption by this sector is of a higher order of around 32% (Francis & Balachandran, 1989). The study of pathogenic bacteria in cured fish is very important since many of these cured products are not hygienically prepared. Among these *Staphylococcus aureus* is of particular significance, since this bacteria is tolerant to high concentrations of salt and reduced water activity (Marcy *et al.*, 1985). Also, the organism is the cause of many food-borne diseases (Rajalakshmi & Rajyalakshmi, 1982; Todd, 1983; Holmberg & Blake, 1984). Sanjeev *et al.* (1985) studied the occurrence of enterotoxigenic staphylococcus in cured fishery products collected from Cochin and found that 23 out of 112 cured fishery products (20.53%) contained enterotoxigenic staphylococcus and they produced enterotoxins A,B,C,D and E either singly or in combinations. No information is available on the effect of storage on enterotoxigenic *S. aureus* in cured fish. Against this background the present study was undertaken.

Five enterotoxigenic strains of *S. aureus* A-100, BS-6, C-361, D-472 and E-326 which produced enterotoxin A, B, C, D and E respectively obtained from Dr. M.S.Bergdoll, Food Research Institute, University of Wisconsin, U.S.A were used for this study. Individual strains were grown in brain heart infusion broth (Difco) at 37°C for 48 h. Two ml of the broth culture from each of the tubes were pipetted into another sterile tube, centrifuged, washed twice with sterile normal saline and 0.2 ml of the undiluted cell suspension was inoculated into a bottle containing 750 ml sterile normal saline.

Two kg of cured fish *Lactarius lactarius* was purchased from the market. Individual sample was then dipped in the enterotoxigenic *S. aureus* cell suspension for one second, and after removing the excess inoculum, was removed and arranged in a container. The container was then kept at ambient temperature with good ventilation.

The samples were analysed for total bacterial count and *S. aureus* load immediately after inoculation and after 2, 4, 6, 10 and 13 days' storage. From each set, 10 g of the sample was withdrawn and analysed for total bacterial count and *S. aureus* load as per ICMSF (1978). The moisture and salt content of the samples were determined as per AOAC methods (AOAC, 1980).

Total bacterial count and *S. aureus* load of the samples immediately after inoculation and during the period of storage are given in Table 1. Two log reduction in total bacterial count by 13th day and 3 log reduction in enterotoxigenic *S. aureus* load by 10th day of storage were observed. The samples were found to be free from the enterotoxigenic *S. aureus* at the 13th day of storage. The moisture content of the samples decreased from 35.4 to 33.2% after 13 days of storage and the salt content was 20.6%. Adesiyun (1984) observed that over 28 days storage of dried fish at ambient

temperature, the mean staphylococcal count was reduced from 4.6×10^5 to 2.2×10^4 CFU/g. Staphylococci, especially enterotoxigenic staphylococci do not form part of the normal bacterial flora of marine fish. Sanjeev *et al.* (1987) reported that out

Table 1. *Changes in total bacterial count (TBC)* and *S. aureus load during storage*

Period of storage, days	TBC. g^{-1}	*S. aureus*. g^{-1}
0	3.6×10^5	2.7×10^5
2	8.5×10^4	4.6×10^4
4	1.3×10^4	1.8×10^3
6	1.0×10^4	1.4×10^3
10	1.2×10^4	1.8×10^2
13	7.1×10^3	0

of 128 strains of *S. aureus* isolated from fish processing factory workers, 85 strains produced enterotoxin A, B, C, D and E either singly or in combinations.

The present studies have shown that enterotoxigenic *S. aureus* strains could not survive in cured fishes for more than 13 days even if the initial load was 2.7×10^5/g.

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