NOTES

Response of Extract Release Volume to the Variations in pH and Microbial Load During Spoilage of Fish at Refrigeration Temperature

Little work has been done using livestock sewage in fish culture. Present report describes the microbial status and storage property of fishes raised under composite fish culture reared in livestock sewage fed pond.

During low temperature storage, microbial growth occurs resulting in physico-chemical changes and ultimate spoilage. The phenomenon of extract release volume (ERV) has been reported as a rapid test for detecting incipient spoilage of meat and shrimp (Jay & Kontou, 1964; Shelef & Jay, 1971). How this phenomenon responds to the change in microbial growth and pH of fishes during spoilage at refrigeration temperature is reported.

Fishes were obtained from composite fish culture pond of the institute. They were brought to the point of sale in gunny bags from where rohu fishes of 1 to 1.5 kg were collected and refrigerated for 10 days. Fresh fishes were analysed for microbial load, pH and ERV on 1st, 4th, 7th and 10th days. Measured area of skin surface (Jay, 1970) was swabbed using sterile template under aseptic precautions and the swab placed in 10 ml sterile diluent of 0.1 % peptone water (IS: 5887-1976). It was agitated and mixed well to give 10^{-1} dilution. This was utilised to prepare further serial ten fold dilutions. Pour plates in quadruplicate were prepared from each of the consecutive three dilutions using standard plate count agar (IS: 5402–1969). Two plates of each dilution were incubated at 37°C for 24h to obtain total mesophilic count. Remaining plates were incubated at 5°C for 5 to 7 days for psychrophilic count. Colonies were counted with an electronic colony counter and reported as count/cm² of the examined surface. Microbial counts reported and discussed in this note are in logarithmic units.

Shelef (1974) was followed for determining ERV with following modifications. 20 g muscle tissue was scrapped from the region behind head. It was homogenised with glass distilled water for one min in a meat blender, filtered through Whatman filter paper no. 1 of 16 cm² size. The volume of extract released during the first 15 min was reported as ERV in ml (Murthy & Bachhil, 1980). pH was recorded with single electrode pH meter by directly inserting the pointed electrode into the muscle tissue.

Anterior half was utilized for ERV and pH and the posterior half for swabbing the skin surface for microbial estimations.

The range and mean values of microbial counts, pH and ERV are presented in Table 1. On an average, mesophilic aerobes

Table 1. *Changes in ERV, pH and microbial counts during refrigeration storage of whole fish*

	1st day		4th day		7th day		10th day	
	R	A	R	A	R	A	R	A
pH	6.40– 6.50	6.46	6.30– 6.40	6.36	6.20– 6.60	6.46	6.60– 6.80	6.72
ERV	25.00–45.00	34.10	17.50– 26.0	21.56	12.0– 18.0	14.65	5.00– 8.50	7.40
Mesophiles	4.83– 6.20	5.34	4.99– 6.93	5.73	5.82– 7.82	6.86	7.00– 8.25	7.76
Psychrophiles	3.78– 5.77	4.48	4.50– 5.55	4.79	4.94– 6.88	5.98	7.00– 8.80	7.45

R = Range, A = Average observation from five fishes for each parameter

were observed to be more than the psychrophiles. This can be attributed to the warmer climate. Shewan (1961) mentioned that the marine fishes show microbial load upto 7.0/cm² of the skin. In warmer seas larger number of these mesophiles are to be expected. The observed higher limit in the present study is less than that of marine fishes. Nair *et al.*, (1971) reported microbial load of 4.14/cm² from fresh water fishes. Skin surface has been stated to harbour heavy bacterial load ranging from 2 log to 5 logs/cm² (Nair & Lahiry, 1968). The microbial load of fishes raised under present system utilizing livestock sewage did not reveal counts higher than the general pattern (Nair *et. al.* 1971).

Increase in microbial numbers were observed both in mesophiles and psychrophilic groups during refrigerated storage at 8°C. Increase in counts upto 4th day is slow, but it is comparatively rapid thereafter (Fig. 1). On 7th day the level of increase was recorded to be almost the same for both the groups. Though the initial psychrophilic count was less than the mesophilic, it almost equalled on the 10th day. Obviously, this is because of the storage temperature. Organoleptically fishes were acceptable on 4th day, but exhibited signs of spoilage on 7th day. Considerable bacterial multiplication did not occur during first four days, probably due to the low pH of 6.3 to 6.4 (Fig. 1). Low pH during rigour is unfavourable to bacteria which explains the less increase in counts. The rigour pH values have been reported in the range of 6.2 to 6.5 (Amlacher, 1961). Nair & Lahiry (1968) mentioned that the growth of organisms responsible for spoilage is effectively checked even at this level. Fig. 1 shows that the pH and microbial counts started rising after 4th day reaching 6.72 and 7.76 on 10th day respectively and the fishes exhibited clear cut spoilage. Nair & Lahiry (1968) observed that the bacterial spoilage produces more undesirable changes in the flavour, odour and appearance in fishes, though oxidative rancidity may precede bacterial deterioration in fatty fishes.

Fig. 1 explains the response of ERV to microbial counts and pH of fishes stored at 8°C. They exhibited symptoms of spoilage in six days. As the pH and microbial

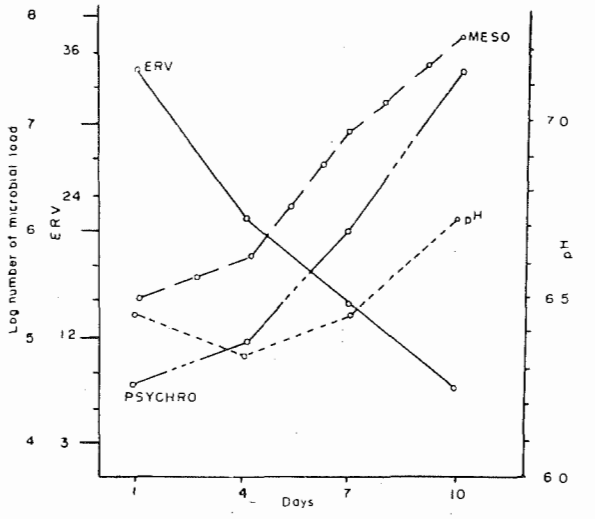

Fig. 1. Response of ERV to changes in pH and microbial load of fishes during spoilage at 8°C

counts increased, ERV decreased. On an average, it reduced to 14.6 on 7th day and at clear spoilage, it came down to 7.4 (Table 1). Raw meat of good organoleptic quality releases large volumes of extract while beef of poor microbial quality releases smaller volumes or none (Jay, 1970). Murthy & Bachhil (1980) also reported reduction in ERV of spoiling pork at refrigeration temperature. Jay (1970) further mentioned that when meats undergo spoilage, ERV is decreased rather than increased. Ingram & Dainty (1971) observed that the physicochemical basis of increase in hydration during storage is not properly understood, but it is postulated that the spoilage flora damage the sarcolemma membrane which controls the permeability of muscle fibre. Murthy & Bachhil (1980) considered pH as an important factor for increase in meat hydration besides alteration in metal ion balance and production of amino sugar complexes by spoilage flora. An interesting phenomenon revealed by ERV is that break down of primary proteins, alteast complete break down, does not occur in meats (Jay, 1970). Lerke *et al.* (1963) showed raw fish press juice to display all apparent fish spoilage as may be determined by whole fish. This indicates a general lack of attack upon insoluble proteins by fish spoilage flora since these proteins are absent from filtered press juice.

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