

THE ISOLATION OF SALMONELLA ORGANISMS FROM FRESH FISH SOLD IN A COLOMBO FISH MARKET

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Reports on the isolation of human intestinal pathogenic bacteria from fresh fish are relatively few (Trawinska, 1949; see also Venketaraman & Sreenivasan, 1952, 1953, and Tanner, 1944). Hompesch (1953) traced an outbreak of typhoid fever to direct contact with contaminated fresh fish. More recently, Floyd & Jones (1954) isolated several shigella and salmonella organisms from the intestinal contents of fresh fish sold in Cairo.

The present investigation was undertaken to study the possibility of Salmonella organisms being present in fresh fish intended for human consumption in Ceylon.

MATERIALS AND METHODS

Fresh fish were purchased during the period July to November 1955 from fish-stalls in a market in Colombo typical of fish-markets in the city. The fish had been caught in the nearby open sea and lagoons and fresh-water sources of various coastal towns all round the island, packed in ice and transported by train or lorry to Colombo. The consignments generally arrived at the Market within about 24 hr., were immediately unpacked and the fish displayed for sale on open-tiled fish slabs.

The types taken for examination depended on availability; usually nine to ten fish were randomly selected from several stalls on each sampling day. They were all less than 18 in. in length, and consisted of species typically small or the young of larger varieties. Each fish was wrapped separately in sterile kraft paper, brought to the laboratory and examined within 30 min. of collection.

Samples of water which had been used for washing fish were collected from buckets in the market, brought to the laboratory in sterile bottles and examined within 30 min. of collection.

All procedures in the laboratory, including dissection of the fish, were carried out with sterile precautions. One group (series A) was examined by culturing the pooled gills and intestines; another (series B), intestines only; and a third group (series C) gills only. The relevant organs from each fish were carefully dissected out, ground in a mortar with a few millilitres of tetrathionate broth, and 1 ml. of this suspension transferred to a tube containing 10 ml. of tetrathionate broth.

The water (fish-washings) was examined by (1) direct plating on S-S agar (Difco), and (2) enrichment in tetrathionate broth (1 ml. sample + 10 ml. tetrathionate broth).

The tubes of inoculated tetrathionate broth were subcultured after 24 hr. on to S-S agar (Difco) plates. These were incubated at 37°C. for 24 hr. Suspicious

colonies were picked into combined Kligler-urea-soft agar tubes (Schmid & Velaudapillai, 1954). Cultures showing reactions on this medium suggestive of *Salmonella* or *Shigella* were examined serologically.

RESULTS

Six hundred and twenty-nine fresh fish consisting of sixty-five species were studied. The species were identified according to Mendis (1954), and *Salmonella* organisms were isolated from the following: *Alectis ciliaris*, *Caranx* sp., *Chirocentrus dorab*, *Chorinemus lysan*, *Clupea fimbriata*, *C. kanagurta*, *C. leiogaster*, *Engraulis baelama*, *E. grayi*, *Exocoetus volitans*, *Gazza minuta*, *Gerres* sp., *Lates calcarifer*, *Leiognathus splendens*, *Mene maculata*, *Mugil cephalus*, *Myripristis murdjan*, *Nemipterus tolu*, *Sardenella* sp., *Sparus berda*, *S. spinifer*, *Sphyaena jello*, *Syngnathoides biaculeatus* and *Tachysurus thalassinus*.

Thirty-nine fish belonging to the twenty-four species listed above were found to harbour salmonella organisms. In each of the series A, B and C, described earlier under Methods, the number of fish examined, the number positive and the types of salmonella isolated are given in the table. In all, thirteen types of the organism were recovered.

Salmonella organisms in fish

Salmonella types	No. of fish positive for <i>Salmonella</i>			All series
	Series A (pooled gills and intestines)	Series B (intestines only)	Series C (gills only)	
<i>Salm. paratyphi</i> B	1	—	—	1
<i>Salm. stanley</i>	1	1	—	2
<i>Salm. saint paul</i>	—	1	1	2
<i>Salm. typhimurium</i>	—	1	—	1
<i>Salm. virchow</i>	—	—	1	1
<i>Salm. oranienberg</i>	—	2	—	2
<i>Salm. bovis morbificans</i>	1	1	—	2
<i>Salm. give</i>	1	—	—	1
<i>Salm. weltevreden</i>	10	2	5	17
<i>Salm. chittagong</i>	—	—	1	1
<i>Salm. newbrunswick</i>	1	2	—	3
<i>Salm. huttingfoss</i>	1	—	—	1
<i>Salm. waycross</i>	—	—	5	5
All salmonella types	16	10	13	39
No. of fish examined	278	217	134	629

Samples from some forty-one other species were examined, but salmonella organisms were not identified in any of this material.

Of the forty-two samples of fish-washings examined, five were positive for salmonella organisms of four serotypes, two of which, *Salm. weltevreden* and *Salm. newbrunswick*, had also been recovered from the fish, but the other two, *Salm. javiana* and *Salm. adelaide*, were found only in the fish-washing water.

DISCUSSION

Attention has already been drawn to the fact that in published records fresh fish have only rarely been incriminated as a source or vehicle in the spread of enteric infections, so much so that Hobbs (1954) considered that fresh fish could be regarded as one of the safest articles of our dietary. This opinion was based on experience in the United Kingdom where the majority of the fish offered for sale come from unpolluted river water or from deep-sea fishing, but this is not true for all countries. The fish examined by Floyd & Jones (1954) and the present authors were caught in shallow inshore waters or in rivers where pollution with faecal material of human or animal origin was either known or suspected.

Of the salmonella types isolated from the fish, *Salm. weltevreden*, which was recovered on seventeen occasions, had previously been isolated in this country from cases of infantile diarrhoea (Schmid, de Silva & Soysa, 1953), rats (Schmid, Lieb & Velaudapillai, 1953), two fish from a fresh-water canal into which several houses discharged polluted material, swans and a wild elephant (McGaughey, Schmid, George & Velaudapillai, 1954). *Salm. waycross*, *Salm. give*, *Salm. new-brunswick* and *Salm. hvittingfoss* had also been isolated in routine examinations of human faeces in the laboratory, though rarely, while *Salm. oranienberg* and *Salm. chittagong* were isolated for the first time in Ceylon during the present study. No island-wide study of the distribution of salmonella types in man and animals has yet been undertaken, and there is, therefore, no accurate information with regard to the types most prevalent in Ceylon. Nevertheless, it is of interest to record that *Salm. typhimurium*, generally regarded as the most common salmonella type in nature, was in this survey found only once.

All the species of fish found to be harbouring salmonella organisms could be caught in shallow seas and lagoons, that is, waters that are prone to human and animal pollution. The presence of the organisms in the fish intestines suggests that they must have entered live fish inhabiting polluted waters. Several workers have found that fish living in such waters could carry human and animal faecal organisms in their intestines (see Tanner, 1944; Griffiths, 1937; Guelin, 1952; and Floyd & Jones, 1954).

The isolation of salmonella organisms from the gills and fish-washings may well have been due to post-mortem contamination at any period from the netting of the fish to the time they were taken at the fish market for examination. However this contamination may have occurred it would appear to constitute as great a health hazard as the presence of the bacteria in the gut itself.

It is probable that the majority of the pathogenic organisms found in the raw fish would be destroyed during the normal cooking processes, but it is possible that a significant number might survive other forms of preparation for human consumption. The greater danger, however, would appear to lie in the handling of the raw fish and the 'discards' during their cleaning and preparation in the kitchen. There the fingers of the cooks or utensils could readily be contaminated and the organisms transferred to other 'dishes' already prepared. Many of these may act as excellent culture media and allow great multiplication of salmonella organisms—sufficient to give rise to manifest outbreaks of food-poisoning.

SUMMARY

Of 629 fresh fish (sixty-five species) and forty-two samples of fish-washings obtained from a fish-market in Colombo, and investigated bacteriologically for the presence of *Salmonella*, thirty-nine fish (twenty-four species) and five samples of washings were found to be harbouring the organisms.

Of fifteen salmonella types found, all except *Salm. chittagong* and *Salm. oranienberg* had been previously isolated from human and animal sources in Ceylon.

The presence of *Salmonella* in the intestines of fresh fish examined suggested that the organisms had entered the live fish, whilst their isolation from the gills and fish-washings was evidence that the fish could have been contaminated post-mortem.

The significance of these findings in relation to hazard to health is discussed.

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