

Rapapport's Broth, A Better Enrichment Medium in the Identification of *Salmonella* from Processed Frog Legs

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Salmonella serotypes recovered from processed frog legs were compared on three enrichment media, Selenite cystine broth (SC), Tetrathionate broth (TT) and Rapapports broth (RB) 25/37°C after pre-enrichment in lactose broth. The studies indicate that enrichment media RB 25/37°C is superior to SC broth as well as TT broth. TT broth facilitated more of *Salmonella* serotypes to grow, where as SC broth enabled different groups of salmonella serotypes to grow.

The WHO/World Association of veterinary Food Hygienists Round Table Conference on the present status on the *Salmonella* problem, held in Netherlands in 1980 noted that the *Salmonella* from food of animal origin is the most common food borne diseases in the world and causes great morbidity and economic loss (WHO, 1981). Frog legs as mode of spread of *Salmonella* contamination was reported in 1967 in Belgium (Desmet Paix *et al.*, 1968). Andrews *et al.* (1977) and Shrivastava (1978) reported the incidence of *Salmonella* from processed frog legs to the extent of 60% and 30% by modifying the isolation procedures.

National Academy of Sciences and Association of Official Analytical Chemists (Baily *et al.*, 1981) have recommended the use of TT broth and SC broth for recovery of *Salmonella* from foods. Many modification of RB broth enrichment are in use for better recovery of *Salmonella* (Vassiliadis *et al.*, 1974; 1978, 1979). International Standards Organisation (ISO) has recommended pre-enrichment in buffered peptone water followed by enrichment in Muller Kauffman tetrathionate broth (Vassiliadis, 1983). In the present study growth obtained from SC, TT and RB 25/37°C were compared. Number of *Salmonella* serotypes isolated by all three methods were taken as 100%.

Materials and Methods

A total of 160 frozen frog leg samples were analysed. Frog legs wrapped in clean

sterile polyethylene bags and frozen individually at -40°C are termed as individually quick frozen (IQF) products. Sampling scale was as recommended by Foster (1971). The samples were analysed according to AOAC (1970). One ml of 1/9 dilution from pre-enriched lactose broth was inoculated into 10 ml of SC broth, TT broth and 0.1 ml RB broth. After scheduled incubation period of 24 h at 37°C the growth from the broth was streaked on Brilliant green agar BGA (Difco) and bismuth sulphite agar BSA (Difco). The characteristic colonies from BGA and BSA were stab inoculated into triple sugar iron agar TSI (Difco) and lysin iron agar LIA. Further identification and confirmation of the colonies were carried out as per Rajagopalan *et al.* (1984).

Results

Table 1 shows the percentage isolation of *Salmonella* from different enrichment media namely, RB 25/37°C 88.8 % TT broth 79.6% and SC broth 74.07%.

Table 2 shows serotypes of *Salmonella* on different enrichment media. Maximum number (9 groups) were recovered from RB 25/37°C followed by SC broth (7 groups) and TT broth (3 groups).

Discussion

It was observed that RB 25/37°C enabled different groups of *Salmonella* to grow freely. Vassiliadis (1983) has reported superiority

Table 1. *Comparative efficiency of different enrichment media in the isolation of salmonella*

Enrichment medium used	Percentage of <i>Salmonella</i> isolated
Rapapport's broth	88.8
Tetrathionate broth	79.6
Selenite cystine broth	74.07
Rapapport's broth plus tetrathionate broth	94.5
Rapapport's broth plus selenite cystine broth	91.01
Selenite cystine broth plus tetrathionate broth	83.0

of RB 25/37°C over other enrichment media for the isolation of *Salmonella* from contaminated meat products, sewage and from healthy pigs. Efficiency in detecting the same number of *Salmonella* organisms when inoculated into RB 25/37°C and RV 10/43°C has been accepted (Harvey *et al.*, 1979, 1981; Vassiliadis, 1983). Even though TT broth showed a recovery rate of 79.60% it could not isolate different serotypes. Tayler & Silliker (1962) demonstrated that when egg albumen in samples were pre-enriched and inoculated into selective enrichment media recovery of *Salmonella* from TT broth was

superior than recovery from SC broth. In the present study maximum number of *Salmonella* in TT broth was from group C, which shows that TT broth is more selective. RB broth which showed a greater recovery rate of 88.8% could be used as single efficient media.

Salmonellae surviving under freezing condition are likely to be missed if the choice of isolation are not improved. The study enables to detect more *Salmonella* in routine analysis of foods.

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Table 2. *Salmonellae serotypes isolated from different enriched broth*

Rapapport's broth	Selenite cystine broth	Tetrathionate broth
<i>S. barielly</i> C1	<i>S. barielly</i> C1	<i>S. barielly</i> C1
<i>S. hvitting-foss</i> I	<i>S. paratyphi</i> C C1	<i>S. hvitting-foss</i> 1
<i>S. tennessees</i> C1	<i>S. typhimurium</i> B	<i>S. paratyphi</i> C C1
<i>S. paratyphi</i> C C1	<i>S. salford</i> I	<i>S. choleraesuis</i> C1
<i>S. berlin</i> J	<i>S. virchow</i> C1	<i>S. stendal</i> F
<i>S. typhimurium</i> B	<i>S. concord</i> C1	<i>S. oranienburg</i> C1
<i>S. virchow</i> C1	<i>S. telhashomer</i> F	<i>S. tennessee</i> C1
<i>S. newport</i> C2	<i>S. saint-paul</i> B	<i>S. arizonae</i>
<i>S. saint-paul</i> B	<i>S. enteritidis</i> D	Rough strain
<i>S. paratyphi</i> A A	<i>S. kentucky</i> C2	
<i>S. stendal</i> F	<i>S. anatum</i> E1	
<i>S. richmond</i> C1	Rough strain	
<i>S. anatum</i> E		
<i>S. roan</i> P		
Rough strain		
Nine <i>Salmonella</i> groups	Seven <i>Salmonella</i> groups	Three <i>Salmonella</i> groups

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