

Comparative Efficacy of Four different Culture Media for Isolation of *Salmonella gallinarum*

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We evaluated the effectiveness of Xylose Desoxycholate agar (XLD) and Salmonella Shigella agar (SSA), MacConkey's agar (MCA) and Brilliant Green Agar (BGA) for isolation of *Salmonella Gallinarum*. Among 500 dead broiler birds, 69 were positive for *Salmonella Gallinarum* using Selenite broth F and all plating media combined. XLD and SSA gave better results than BGA and Mac Conkey agar with less false positive colonies. The most effective isolation media was XLD agar, wherein *Salmonella* was identified in 39 (56.5%) of the 69 positive samples followed by 29 (42.0%) for SSA, 15 (21%) BGA and 7(10.0%) for MCA.

Key words: *Salmonella Gallinarum*, isolation, Birds.

Successful isolation of *Salmonella* is a complex process which involves many factors including various media and cultural conditions. There is bewildering variety of methods available for isolation of *Salmonella*, which makes it difficult to compare results obtained by different workers and optimize the technique for isolation. The present investigation was undertaken to standardize the protocol for isolation of *Salmonella gallinarum* which is particularly desirable because of wide variation in techniques adopted by different laboratories (Geissler & Koster, 1970; Gosh & Pande, 1988) and wide distribution of this among the poultry flocks (Prakesh *et al.* 2005) Selective enrichment broths lead to increased *Salmonella* numbers because they contain inhibitory compounds that limit non-*Salmonella* microorganisms. The objective of this study was to evaluate the effectiveness of four plating agars for isolation of *Salmonella gallinarum* from field outbreaks of fowl typhoid.

MATERIAL AND METHODS

Samples

500 broiler chickens suspected to have died of fowl typhoid collected from 140 outbreaks (April 2008 to Oct. 2009) in Srinagar district and its adjoining areas, were necropsied following standard procedures. From each of the broiler bird organs (heart, liver and spleen) were collected and processed for isolation of *Salmonella*.

Procedures

Samples were collected from liver following sterile precautions and *Salmonella* was isolated according to standard methods (ISO 6579, 1993). Briefly sample was aseptically added to buffered peptone water (Pre-enrichment media) in the ratio of 1:10. The samples were incubated for 18hr at 37°C. 0.1ml of pre-enriched liver cultures were then transferred to Selenite broth F, and incubated at 42°C. After incubation at 42°C, a loop full of each broth was plated onto plates of Xylose Lysine deoxycholate (XLD) agar, Salmonella-Shigella agar(SSA), Brilliant green agar (BGA) and MacConkey's agar (MCA) incubated at 37°C for 24 h. After incubation, five typical colonies from each agar plate were submitted to biochemical

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tests (Edward and Ewing, 1972). The isolates were then sent for serotyping at "National Salmonella and *E. coli*, Central Research Institute", Kasauli, HP. The efficacy of different culture media used in isolation of *Salmonella* was assessed by comparing the results of isolation.

RESULTS AND DISCUSSION

In all 69 *Salmonella gallinarum* (antigenic structure of 9, 12) could be isolated from 500 samples of poultry origin (Table 1). Of these 50 were isolated from liver, 13 from Spleen and 6 from heart. By using Selenite broth F as enrichment media and XLD as selective media about 56.5 % of the isolates were recovered. The results obtained in the present study indicated that MCA could disclose more number of *Salmonella gallinarum* than BGA. Ghosh and Pande (1988) indicated BGA as an ideal selective media for isolation of *Salmonella gallinarum*. However they reported

that MCA could not disclose *S. Gallinarum*, whereas in the present study MCA was found superior than BGA as more number of isolations was possible on MCA. XLD was found to be best plating media among these for isolation of *S. Gallinarum* (56.5%) followed by SSA (42.0%), MCA (21.7%) and BGA (10%). Taylor and Schelhart (1968) reported XLD (94%) as preferred media for isolation of *Salmonella* from stool specimens when compared to MCA. Rall *et al.* (2005) also reported XLD as best classic media (34.5%) followed by SSA (27.6%) and BGA (13.8%) for isolation of *S. Gallinarum* from Poultry carcasses. Shah *et al.* (2000) observed MCA as more effective media for the isolation of *S. gallinarum* when compared to BGA.

It can thus be concluded that maximum isolation of *Salmonella gallinarum* could be made by selective media such as XLD and SSA. Selenite broth F when used as enrichment media may enhance the growth of *S. gallinarum*.

Table 1. Comparative efficacy of different culture media used for isolation of *Salmonella gallinarum*

Organs	Number of Samples	Number of <i>Salmonella</i> Strains isolated after direct plating on media				Total number of positive <i>Salmonella</i> isolated
		XLD	SSA	BGA	MCA	
Liver	500	28	22	6	11	50
Spleen	500	8	5	1	3	13
Heart	500	3	2	-	1	06
Total		39	29	7	15	69

REFERENCES

- Edward, P.R., Ewing, W.H., Identification of enterobacteriaceae 3rd ed. Burgess publication Company, Minneapolis, Minnesota 1972.
- Geissler, H., Kusters, J. Zur Diagnose de Salmonellose des Geflugels mit Hilfe der Anreicherung bei 43 C. Zentralblat fur Bakteriologie, Parasitenkunde, Infektionskrankheiten und Hygiene. *Abt. I Originale*, 1970; **214**: 207-215.
- Ghosh, N. N., Pande, S N. Isolation of *Salmonella gallinarum* from Poultry in Orissa. *Indian Vet. J.*, 1988; **65**: 1049-1051.
- ISO 6579. 1993. Microbiology - General guidance on methods for the detection of *Salmonella*.
- Prakesh, B., Krihnappa, G., Munlyappa, L., Kumar, B.S. Epidemiological characterization of Avian *Salmonella enterica serovar* infections in India. *Intern. J. Poult. Sci.*, 2005; **4**: 388-395.
- Rall, V.L.M., Rall, R., Aragon, L.C., Silva, M.G.D. Evaluation of three enrichment broths and five plating media for *Salmonella* detection in Poultry. *Braz. J. Microb.*, 2005; **36**: 147-150.
- Shah, D.H., Roy, A., Purohit, J.H. Comparative efficacy of different culture media for isolation of *Salmonella gallinarum*. *Indian J. Comp. Microbiol. Immunol. Infect. Dis.*, 2000; **21**: 79-80.
- Taylor, W.I., Schelhart, D. Isolation of Shigellae VI Performance of media with stool specimens. *App. Microbiol*, 1968; **16**: 1387-1393.