# Estimation of $D_{10}$ Value for Salmonella Serotypes Isolated from Processed Frozen Frog Legs

Salmonella has been regarded as major aetiological agent of foodborne infection even in advanced countries despite high standard of hygiene. These organisms can contaminate food supply to such an extent that it has been practically difficult to eliminate them completely. Preservation by irradiation has recently been accepted as one of the best and cleanest methods. A knowledge of the D value of particular organisms and of the extent of contamination of the raw materials is necessary to determine the irradiation dose. The process required to eliminate salmonella in frozen frog legs by irradiation in Indonesia has been reported (Anon,1981). Nerkar & Lewis (1982) has made significant contribution in this direction. The present study reports the D value of rare and unreported serotypes of salmonella isolated from frozen frog legs. Knowledge gained from this invitro studies on the serotypes of salmonella will provide sufficient material for future research, since the choice of dose for radicidation depends upon the individual serotypes of salmonella.

Twentyfour serotypes of salmonella isolated from frozen frog legs were studied. The serotypes studied are given in Table 1. Lyophinsed strains mentioned were used for the purpose. Twentyfour hour old cultures of samonella strains grown in trytipic soy broth (TSB, Difco) was diluted to the litre of $10^8$ cell/ml. These cell suspension was exposed to gamma radiation in the range of 50,100, 150, 200, 250, 300 rads using a gamma cell 220 (AECL, Canada) with $Co^{60}$ source at the rate of 3kilo rad per minute. The test was performed at Bhabha Atomic Research Centre, Trombay. Sufficient ice was used during irradiation in the sampled beaker ($2^oC$) throughout the experiment. The viable cell count was determined by plating on Difco tryptic soy yeast extract suppremented with 0.3 per cent yeast extract. Total number of bacteria was counted from the incubated plates after 48 h. $D_{10}$ value (dose required for 90 percent inactivation) was determined from the survival curve. A graph of log of the surival microbial population on the ordinate and irradiation dose on the absissa was plotted. The line of the best fit is drawn through these points. D value was calculated from the straight line plotted.

D value of different serotypes of salmonella are shown in Table 1. The lowest value was shown by S.concord with 35 K rad and higher

**Table 1.** *Sensitivity of salmonella serotypes suspended in tryptic soy broth to gamma radiation*

| Salmonella serotypes | D10 value in K red |
|---|---|
| *S. barielly* | 50 |
| *S. paratyphi C* | 60 |
| *S. hvittfoss* | 70 |
| *S. typhimurium* | 42 |
| *S. tennessee* | 50 |
| *S. virchow* | 60 |
| *S. concord* | 35 |
| *S. stendal* | 40 |
| S. salford | 40 |
| S. newport | 45 |
| S. choleraesuis | 40 |
| S. paratyphi A | 50 |
| S. oranienburg | 60 |
| S. richmond | 45 |
| S. telehashomer | 50 |
| S. berlin | 40 |
| S. saintpaul | 58 |
| S. arizonae | 50 |
| S. rough strain | 50 |
| S. enteritidis | 62 |
| S. kentukey | 48 |
| S. anatumm | 50 |
| S. roan | 50 |
| S. 41 z 10 | 40 |

value of 70 K rad and higher value of 70 K rad was shown by S.hvittingfoss. Six serotypes of salmonella showed values between 40 and 50 K rad, 5 serotypes between 50 and 60 K rad and 2 serotypes of salmonella between 60 and 70 K rad in tryptic soy broth in iced conditions. Variation of sensitivity among different serotypes of salmonella was carried out for comparative purpose in the present study. D value of salmonella in Hartsell's broth was reported to be between 0.025 and 0.55 M rad. *S.pvllorum, S.enteritidis and S.paratypht* A showed 0.025 Miad while *S.choleaesuns and S.paratyphi B*.0.055 M rad (Dorethy *et al.*, 1970). In brain heart infussion broth at 4° C resistance of salmonella serotypes ranged from 0.042 M rad for *S. entcritidis* and for *S. thompson* 0.084 M rad value (Previte *et al.*, 1982). Nerkar & Lewis (1982) reported $D_{10}$ value of *S. typhimurium, S. enteritidis* and *S. newport* to be between 18 and 30 K rad, when the cells were irradiated at 2°C in phosphate buffer. D value obtained in the present study (35 K rad in the case of *S. Concord* and 70 K rad for *S. typhimurium* in tryptic soy broth) are identical to that of other investigators. Nerkar & Lewis (1982) reported that salmonella species normally encountered in frog legs were almost equally sensitive to gamma irradiation whether irradiated in buffer or in frog lengs homogenite. Temubunan (1983) has conducted a similar study from frog from Indonesia and identified several species of salmonella from frogs.

The authors are thankful to Dr. G.B. Parulkar, Dean, Seth G.S. Medical College and K.E.M. Hospital, Parel for permission to undertake this study and to Department of Biochemistry and Food Technology Division of BARC (Bombay) for providing facility for irradiation.

## References

Anon (1982) *Food Irradiation Newsletter*, No. 6 (1), 11 RC 2711/RB joint FAO/IAEA Division of Isotope and Radiation Application of Atomic Energy for Food and Agricultural Development, International Atomic Energy Agency, Vienna.

Dorothy, J., Anderson, A.N. & Dyer, J.F. (1970) Technical Report Series 114, IAEA Vienna, 1-14.

Nerkar, D.P. & Lewis, N.F. (1982) *J. Good Prot.* **45**, 820.

Previte, J.J., Chung, Y. & Bisi. H.M.E. (1971) *Can. J. Microbiol.* **17**, 385.

Temabunan, P.R. (1983) *Food Irradiation Newsletter*, **17** (8), 28 Agricultural Development, International Atomic Energy Agency, Vienna.

Department of Molecular Biology,
Seth G.S. Medical College,
Bombay - 400 012.

D. RAJAGOPALAN
C.K. DESHPANDE &
LEELA JOSHI.