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Sodium dodecyl sulphate - polyacrylamide gel typing system for characterization of *Staphylococcus aureus* strains of bovine mastitis origin

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The present study was to compare different strains of *Staphylococcus aureus* of bovine mastitis origin by SDS-PAGE of whole-cell proteins, and to know about the suitability and practicality of the method in differentiation of the strains in epidemiological situation.

Staph. aureus isolates (50) were identified from 354 milk samples of cows and buffaloes with a history of clinical and sub-clinical cases of mastitis in and around Madras city on the basis of morphological and biochemical characteristics (Buchanan and Gibbons 1974). The whole-cell protein samples of 50 isolates of *Staph. aureus* for SDS-PAGE analysis were prepared (Krikler *et al.* 1986) and stored at –70°C. The whole - cell sonicates were analysed by a modified version of the SDS-PAGE technique (Laemmli 1970, Carter and Pennington 1989 a). The average similarity between 2 *Staph. aureus* isolates analysed by Coomassie-brilliant blue stained gels was assessed using the coefficient of Dice (1945), whereby

$$\text{average percentage similarity (\%S)} = \frac{\text{Number of matching bands} \times 2}{\text{Total number of bands in both isolates}}$$

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Reproducible gel profiles were obtained after staining with Coomassie blue (Fig. 1). Each whole - cell polypeptide profile comprised 52 to 60 bands, with an average of 56 as ascertained per gel scanned peaks (LKB Ultroscan XL). Visual inspection of gels showed several differences between isolates. However, these were almost entirely due to variations in band intensity. The 50 isolates were differentiated into 5 groups (10%) based on the number of bands in each isolate (Table 1). Calculation of Dice coefficients showed high degrees of similarity between different isolates of *Staph.aureus*. The least degree of similarity (91.0%) occurred between isolates 13 and 14. Isolate 14 possessed 8 more bands than No. 13.

All other pairs differed by only 2 to 6 bands. The average similarity value was 98.8%. Out of the 50 isolates only 5 types (10%) could be distinguished on the basis of the number of bands in each of the isolate (Table 1). Due to the high degree of similarity observed among the banding patterns analysis of whole - cell extracts by SDS-PAGE was unable to distinguish themselves between the isolates of *Staph.aureus*, but it could be concluded that all the isolates were closely related. In comparison where similarity values approach 100% different isolates will be indistinguishable from each other. However, in conjunction with other typing

Table 1. Grouping of 50 isolates of *Staphylococcus aureus* (bovine mastitis origin) based on SDS-PAGE whole-cell polypeptide profile

Group	Number of polypeptide bands in each isolate	*Staphylococcus aureus* isolate (total number in each group)	Per cent
1	52	Staph 13 (1)	2
2	54	Staph 1, Staph 2, Staph 3, Staph 4, Staph 18, Staph 20, Staph 21, Staph 28, Staph 33, Staph 34, Staph 36, Staph 41, Staph 42, Staph 43, Staph 47, Staph 49 and Staph 50 (17)	34
3	56	Staph 5, Staph 7, Staph 19, Staph 23, Staph 24, Staph 29, Staph 32, Staph 35, Staph 37 and *Staph 40 (10)*	20
4	58	Staph 6, Staph 8, Staph 11, Staph 15, Staph 17 Staph 22, Staph 25, Staph 27, Staph 31 and Staph 38 (10)	20
5	60	Staph 9, Staph 10, Staph 12, Staph 14, Staph 16, Staph 26, Staph 30, Staph 39, Staph 44, Staph 45, Staph 46 and Staph 48 (12)	24

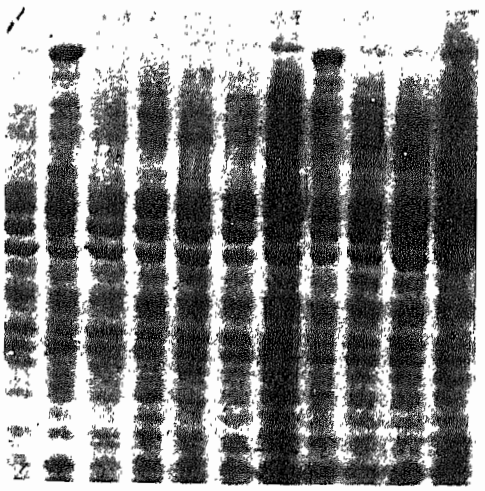

Fig. 1. Wide-cell polypeptide profile of 50 isolates of *Staph. aureus* (bovine mastitis origin).

methods such as resistogram typing, antibiogram typing and plasmid profile analysis, more number of isolates could be differentiated. Hence, SDS-PAGE analysis of whole-cell extracts alone could not be used to distinguish between *Staph. aureus* isolates. In this study, the results concur with the findings of Kirkler *et al.* (1986), Stephenson *et al.* (1986) and Carter and Pennington (1989 a, b).

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