# ASSESSMENT OF MATERIALS FOR MICROBIOLOGICAL SAMPLING OF CARCASSES

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**Objectives**

To compare six materials for their potential use as carcass swabs by investigating recovery of pure cultures of a spoilage organism, *Pseudomonas fluorescens*, and three pathogens; *Listeria monocytogenes*, *Staphylococcus aureus* and *Salmonella mbandaka* from blended samples. A cheap, effective swab was sought for use on beef carcasses.

**Materials and methods**

Six materials selected were and four of these were made into sampling swabs in the laboratory. The latter were cotton pads (CP), using Propax™ bandage gauze (Smith & Nephew Medical Ltd., England), Co-Op sponge wipes (SW) , Co-Op sponge cloths (SC), (Belfast Co-Operative Society Ltd) and Spontex Sponge Wipes (SP) (Spontex Ltd., Swansea, Wales). Two commercial sampling sponges were evaluated; sterile foam sampling sponges (TSC) (Technical Service Consultants Ltd., Lancs., England), and Speci-Sponge™ (NASCO) ( NASCO, Fort Atkinson, WI, USA).

*Preparation of swabs in the laboratory*

CP - 7.5cm fabric folded to eight layers thick and 6cm width then stapled at one side to form a pocket for three fingers. Sterilised by autoclaving at 121°C for 15 minutes. SP, SW and SC were cut into 8 by 9 cm sections. Following any required treatments samples were sterilised by gamma irradiation at 25 kGy. Neutralising buffer (NB) consisting of 0.1% (w/v) sodium thiosulphate; 0.1% (v/v) Tween 80 in deionised water was prepared for use with these samples.

The following bacterial cultures were utilised: *Listeria monocytogenes* NCTC 04885; *Pseudomonas fluorescens* (wild type, local isolate from spoiled beef); *Staphylococcus aureus* NCTC 1803 and *Salmonella mbandaka* NCTC 7892.
All cultures were stored at -80°C in bacterial preservers. All media used were obtained from Oxoid, Basingstoke, England. For each test, cultures were transferred to tubes containing nutrient broth (5ml) (NB) and incubated at 37°C overnight, except *P. fluorescens* which was incubated at $25^0$C for 2 days. The cultures were diluted to give approximately $10^3$ colony forming units per millilitre (cfu $ml^{-1}$) in maximum recovery diluent (MRD) before being inoculated onto the test materials.

*Recovery of bacterial cultures from test materials*

After dilution, 4 ml was pipetted onto each material in stomacher bags and held refrigerated ($4^0$C) for three and a half hours (to simulate transit time from abattoir to laboratory). Buffered peptone water (90ml) (BPW) was then added to each bag and the sample homogenised for two minutes (Stomacher Model 400, Seward Laboratories, London) then manually squeezed to express all of the liquid. Controls were prepared by adding 4 ml of inoculum to an empty stomacher bag and treating as for the samples. Each sample was then plated, in duplicate, onto Baird-Parker agar (BP); Oxford agar (OX); Pseudomonas selective agar (PSA) and modified brilliant green agar (BGA). Plates were incubated at 37°C for 48 hours (PSA $25^0$C) then characteristic colonies counted.

All experiments were conducted in triplicate.

**Results and discussion**

Swabs are available for surface sampling (Shilliker and Gabis 1975, Baldock 1974) but for the purpose of swabbing carcasses robust, cheap swabs were sought and the swab was required to leave no inappropriate residue on the carcass after sampling (Dorsa et al. 1996, Lasta et al. 1992). To evaluate the swabbing materials the number of organisms in the control were determined then compared with those recovered from the swabs. Since some organisms would be expected to bind to the swabs a slight reduction in counts is to be expected but any large difference implies excessive binding or some anti-microbial activity (Llabrés and Rose, 1989). Initial experiments with SW showed very low recoveries of all of the challenge organisms hence some samples were hand washed in two changes of drinking water prior to use. This increased recoveries implying the sponges contained anti-microbial activity. Therefore both SW and SC swabs were placed (separately) in a domestic washing machine and the rinse cycle run. This further increased recoveries, but this recovery was still poor for all bacterial strains in comparison with the TSC commercially produced sampling swabs, Fig 1. The use of NB was then investigated as a means of raising recoveries but *S. aureus* proved sensitive to the buffer as tested. The acceptability of the residue left by such a buffer was also open to question. Thus SW and SC were regarded as unsuitable as potential swab material and cotton swabs (CP) and Spontex Sponge Wipes (SP) evaluated.

The CP swabs showed good recovery whilst the SP swabs required a wash treatment. However once washed the SP swabs gave results very similar to the NASCO sample swabs, Fig 2. The treatment required by the SP swabs was simple and hence swabs could be produced with less effort than for the cotton swabs. The cost of the SP swabs was also considerably less than the commercial

products, being about £0.09 per swab of size 8 by 9 cm whilst the NASCO swabs cost £0.68 per 4 by 8 cm swab and those from TSC were £0.28 per swab of size 6 by 8 cm.

Since the swabs were intended for use in a major sampling excercise requiring about 1 000 swabs the benefits from preparing swabs in the lab were considerable. Locally produced swabs also have the advantage of being larger than commercial swabs allowing easier handling, and also permit the option producing of very large swabs for use on large areas of carcasses or processing equipment.

**Conclusions**

Commercial swabs showed high recoveries of the challenge organisms but were rather awkward to use due to their small size. However swabs prepared from some retail sponges gave very low recoveries even after washing, or treatment with neutralising buffer. Cotton gauze was effective in recovering the challenge micro-organisms but the production of swabs required time to assemble the swabs and the staples used to form the swab could be a hazard if they were to break and become embedded in the carcass. Spontex sponge wipes proved cheap, and once washed, effective in recovering the challenge micro-organisms and hence were selected for use in a major carcass sampling programme.

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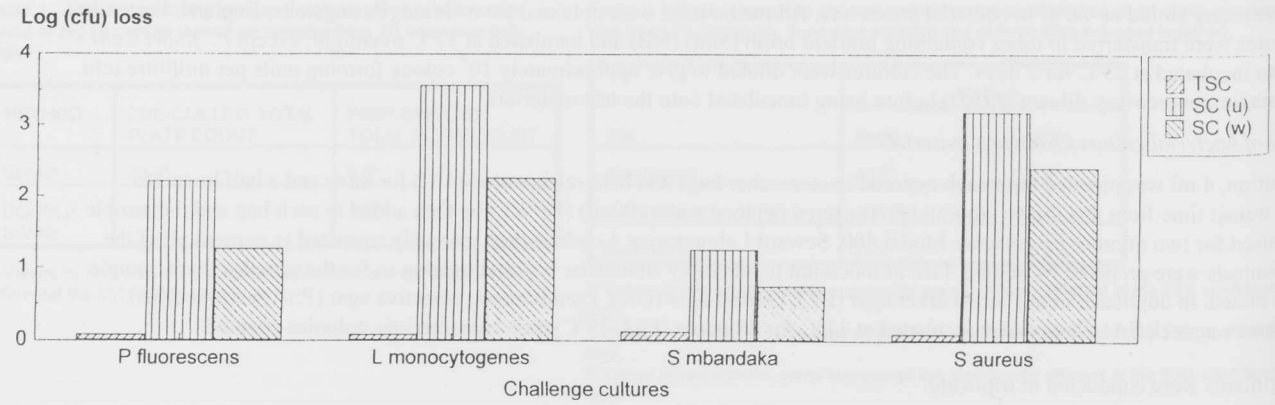


Fig1 Loss of viability with washed (w) and unwashed (u) retail sponge and commercial swab

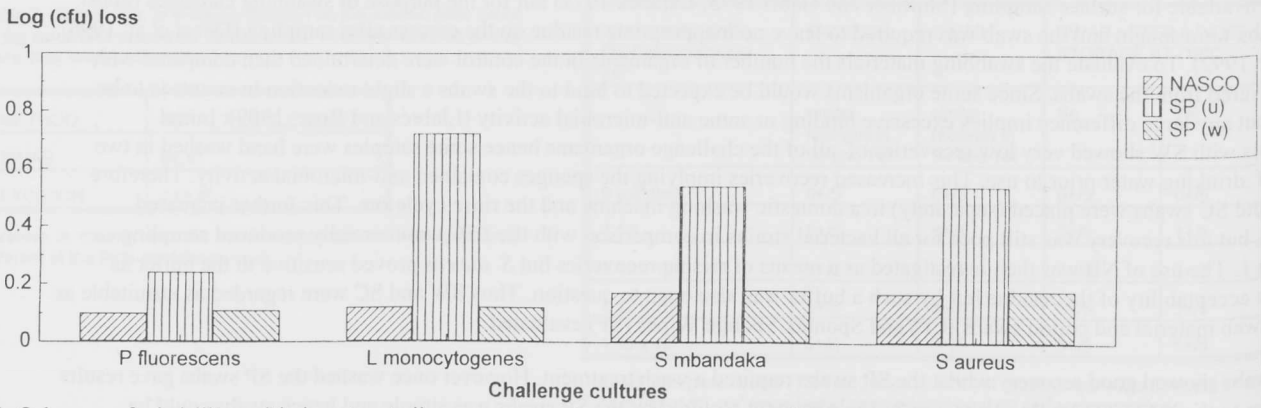


Fig2 Loss of viability with best retail sponge and commercial swab