Laboratory evaluation of disposable and reusable disinfectant cloths for cleaning food contact surfaces

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SUMMARY

A comparison of five methods of cleaning Formica surfaces contaminated with bacteria dried in milk has been carried out. A standardized procedure was developed, and impression plates were found to be at least as sensitive as a swab-rinse method for detecting bacteria on the surfaces. The most satisfactory results were obtained with one type of disposable alcohol-impregnated wipe and with a detergent/hypochlorite solution applied with paper. A reusable cloth impregnated with disinfectant initially performed well against all test organisms, but was less reliable against *Staphylococcus aureus* and *Streptococcus faecalis*, after the cloth had been used and rinsed several times. The importance of introducing methods to reduce the high risk of cross-contamination presently associated with the use of wiping cloths in catering premises is stressed.

INTRODUCTION

Although the advantages of paper for cleaning in catering premises have been stressed (Davis, Blake & Woodall, 1968; Gilbert, 1969; Tebbutt, 1984), reusable wiping cloths are still used in the vast majority of premises. Such cloths are frequently heavily contaminated with bacteria. A recent study in shops selling raw and cooked meats found that 19% of cloth samples contained more than 10^5 c.f.u./cloth of *Escherichia coli* (Tebbutt, 1986).

In most small premises a single cloth is used for cleaning in raw and cooked food areas. Generally these cloths are rinsed after each use, but proper cleansing and disinfection is inpracticable. Some advocate the use of colour-coded cloths for different food areas. Unless rigidly supervised, however, these cloths are unlikely to remain in their designated areas. Continuous soaking of cloths in dilute disinfectant solution has been suggested. In practice these solutions are rarely prepared accurately, and are changed infrequently, giving rise to inactivation of the disinfectant and contamination by bacteria.

Recently, disposable alcohol-impregnated hand wipes have been introduced to supplement existing hand-washing facilities. In certain circumstances these have been used successfully to disinfect surfaces and equipment. However they have limitations, and are only suitable for light to medium soil loads, and cannot be

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used to mop up spillages. These wipes are potentially useful in situations where soiling is light or where disinfection after preliminary cleaning is desirable.

Because field-tests designed to measure bacterial contamination of surfaces and equipment cannot be easily standardized, a laboratory model has been developed to compare different cleaning procedures. In this investigation the performance of two types of disposable alcohol wipes, a non-ionic detergent applied with paper, a combined detergent/disinfectant solution applied with paper, and a reusable cloth impregnated with disinfectant has been studied.

MATERIALS AND METHODS

Organisms

Ten strains of *Escherichia coli* and three isolates of each of *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Streptococcus faecalis* were studied. These were isolated from wiping cloths obtained from catering premises. Also three *Klebsiella aerogenes* strains, which were isolated from urines from hospital patients, were studied. All organisms were suspended in nutrient broth containing 10% (v/v) glycerol and stored frozen at -70 °C.

Surfaces

Formica boards were used. Sixteen squares each measuring 25 cm^2 were drawn on each board. Before use each board was wiped with a paper towel moistened with a combined detergent/hypochlorite solution. The board was allowed to dry, rinsed with sterile distilled water, and allowed to dry thoroughly.

Cloths

Cloth A (Levertex) was supplied by Lever Industrial Limited. It was a heavyduty paper wipe incorporating a solution containing 30% ethyl alcohol, surfactant, humectant and emollient skin-care agents. Cloth B was obtained from Southern Hygiene Chemicals Limited. It was made of non-woven rayon, and incorporated a solution containing 10% ethyl alcohol and cetrimide.

The reusable cloth (Difco Disinfectant Cloth) was a non-woven fabric sheet impregnated with biocides (a blend of quaternary ammonium compounds and biguanides). The cloth also contained a blue indicator dye to indicate the presence of active biocides. The cloth was rinsed in clean water after each use as recommended by the manufacturer.

Diluents

In most experiments pasteurized milk submitted for routine examination was used. All samples were screened by a methylene blue dye reduction test, a phosphatase test, and for the presence of any test organisms in a 20 μ l sample. A sample giving a positive result in any of these tests was not used. Sometimes bacteria were suspended in $\frac{1}{4}$ -strength Ringer solution or in an extract prepared from sliced cooked meats. For this, meat samples were homogenized in $\frac{1}{4}$ -strength Ringer solution, and large food particles were allowed to settle. The fluid was poured off and heated at 80 °C for 15 min. Samples of cooked-meat fluid were stored at -20 °C.

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Preparation of inoculum and contamination of test surfaces

The organisms were grown in nutrient broth (Oxoid CM67) at 37 °C for 24 h. Each culture was agitated on a vortex mixer for 30 s, and diluted a hundredfold in pasteurized milk or occasionally in either $\frac{1}{4}$ -strength Ringer solution or cooked-meat fluid. Sometimes serial tenfold dilutions were prepared to determine the number of organisms inoculated onto test surfaces. After mixing, 20 μ l of the suspension were inoculated onto the centre of a 25 cm² area marked on the Formica board. The inoculum was spread with an L-shaped glass rod (made from a Pasteur pipette), and was allowed to dry at 25 °C.

Drying experiments

A 10^{-4} dilution of an overnight broth culture was used. Test surfaces were contaminated with bacteria suspended in either pasteurized milk or in $\frac{1}{4}$ -strength Ringer solution as previously described. Using contact plates surfaces were sampled immediately after contamination, after the inoculum had dried, and after, 5, 10, 30 and 60 min. Two surfaces were examined for each sampling time.

Surface cleaning and sampling technique

Four cleaning tests were studied: (1) non-ionic detergent (0.5% v/v) applied with paper, (2) a solution containing 0.5% detergent and hypochlorite (final concentration 200 ppm available chlorine) also applied with paper, (3) a disposable alcohol wipe (cloth A), and (4) another alcohol wipe (cloth B). Each contaminated surface was wiped in two directions, and, as far as possible, a uniform downwards pressure was applied to the cloth or paper. Each surface was allowed to dry. Generally surfaces were sampled using contact plates containing Columbia Agar (Oxoid CM331) with 1% Tween 80. Preliminary tests confirmed that the addition of Tween 80 did not reduce the plating efficiency of the medium. In some experiments Columbia Agar was replaced with Blood Agar Base No. 2 (Oxoid CM271) or with Buffered Peptone Water (Oxoid CM509) solidified with 1.2% agar. All contact plates were incubated overnight at 37 °C.

Because the recovery of organisms from surfaces may be greater by a direct swabbing method than by an agar-impression technique (see Gilbert, 1970), some surfaces were sampled with alginate swabs (Medical Wire and Co. Ltd) using two swabs/area. The first swab was moistened in saline before it was rubbed on the surface. The swabs were dissolved in $\frac{1}{4}$ -strength Ringer solution containing 1% sodium hexametaphosphate, 1% Tween 80, 0.1% peptone, and where needed 0.5% sodium thiosulphate. After incubation in this fluid for 1 h to try to resuscitate any sublethally-damaged bacteria (Mossel & Corry, 1977), serial dilutions were cultured onto Columbia Agar.

Tests with reusable cloths impregnated with disinfectant

Test organisms were suspended in pasteurized milk as previously described. At intervals, 20 μ l of the suspension were spread onto the test surface and allowed to dry. Before use the cloth was rinsed in tap water, and between each use the cloth was rinsed three times in tap water at 5 min intervals. The technique used to clean

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surfaces was as described for disposable cloths. After use the cloth was wiped on a clean test surface to determine whether or not the cloth transfered bacteria from one surface to another. This procedure was repeated after 5 min to determine whether any organisms remaining on the cloth had been killed. In most experiments contact plates were used to sample cleaned surfaces, but sometimes an alginate swab-rinse method was used as previously described.

RESULTS

Drying experiments

Many organisms were killed during drying on the Formica surface. The survival of $E.\ coli,\ Staph.\ aureus$ and $Str.\ faecalis$ suspended in either pasteurized milk or in Ringer solution are shown in Fig. 1. In preliminary experiments the recovery of $P.\ aeruginosa$ or $K.\ aerogenes$ was similar to that of $E.\ coli$. Bacteria suspended in milk or in cooked-meat fluid survived better than those suspended in an inorganic salts solution. Whatever the suspending fluid Staph. aureus and Str. faecalis survived considerably better than did $E.\ coli$. After drying the mean survival rates for bacteria suspended in milk were 27 % for $E.\ coli,\ 88\%$ for Staph. aureus and 91 % for Str. faecalis.

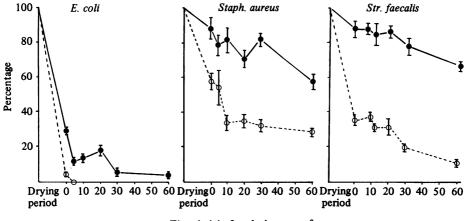
Cleaning experiments

The results of preliminary experiments in which test organisms were suspended in either pasteurized milk or in cooked-meat fluid did not differ significantly, and all subsequent tests were done with bacteria suspended in milk. Although the size of colonies was smaller on buffered-peptone agar, viable counts were similar on each of the three test media (see Methods), and contact plates containing Columbia Agar were used throughout the study. Counts showed that approximately 10^5 c.f.u. were spread on the 25 cm² test surface. Although some bacteria, in particular those belonging to Gram-negative genera, were killed during drying, a confluent growth was always obtained on contact plates applied to the surface at this stage of the experiment.

Fig. 2 shows the results of cleaning with a detergent alone and with a detergent and hypochlorite solution. In simultaneous experiments the combination consistently performed better against each of the test organisms than did the detergent alone. Some surfaces contaminated with *E. coli* or with *Staph. aureus* were cleaned with detergent and hypochlorite solution and sampled with alginate swabs. Of *E. coli* none was recovered, and the counts for *Staph. aureus* were similar on contact plates and in swab fluids (mean counts 28 for plates and 20 for swabs).

Fig. 3 shows the results of cleaning with two types of alcohol impregnated wipes. Although both wipes removed *Staph. aureus* and *Str. faecalis* from the surfaces to similar extents, wipe A performed consistently better than wipe B against *E. coli*, *P. aeruginosa*, and *K. aerogenes*. Experiments in which alginate swabs were used to sample surfaces contaminated with either *E. coli* or *Staph. aureus* failed to detect either organism on surfaces after cleaning with wipe A.

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Time (min) after drying on surface

Fig. 1. Survival of *E. coli, Staph. aureus* and *Str. faecalis* suspended in either pasteurized milk ($\bigcirc - \bigcirc$) or in $\frac{1}{4}$ -strength Ringer solution ($\bigcirc ---\bigcirc$). Each count is expressed as a percentage of the number of organisms recovered from the contaminated surface before drying (\pm s.E.M.).

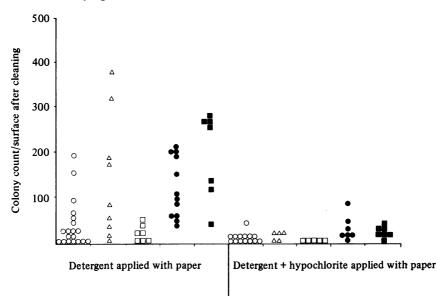


Fig. 2. Recovery of *E. coli* (\bigcirc). *P. aeruginosa* (\triangle), *K. aerogenses* (\square), *Staph. aureus* (\bigcirc) and *Str. faecalis* (\blacksquare) from Formica surfaces after cleaning with detergent or with a combined detergent/hypochlorite solution.

Experiments with reusable disinfectant cloths

Fig. 4 shows the plate counts obtained from surfaces after cleaning with a cloth impregnated with disinfectant. Initially these cloths performed well, and none of the test organisms was detected on cleaned surfaces. However after the cloth had been rinsed several times, some viable bacteria, in particular *Staph. aureus* and *Str. faecalis*, remained on the surfaces. To determine whether or not these cloths could transfer bacteria from one surface to another, a used cloth was immediately wiped on a clean surface. Initially no bacteria were transfered, but after several

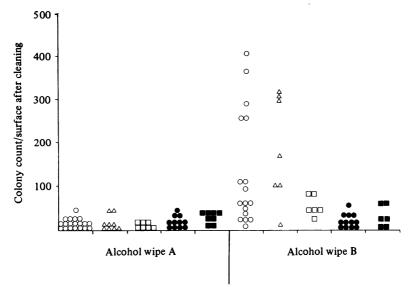
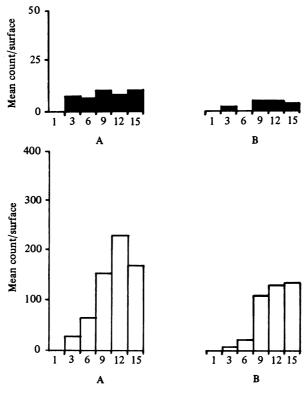


Fig. 3. Recovery of *E. coli* (\bigcirc) , *P. aeruginosa* (\triangle) , *K. aerogenes* (\Box) , *Staph. aureus* (\bigcirc) and *Str. faecalis* (\blacksquare) from Formica surfaces after cleaning with two types of disposable alcohol-impregnated wipes (A and B).



Number of times cloth rinsed

Fig. 4. Recovery of *E. coli/P. aeruginosa* (\blacksquare) and *Staph. aureus/Str. faecalis* (\square) from Formica surfaces after cleaning with a reusable cloth impregnated with disinfectant. Column A, colony count from contaminated surface after cleaning and the surface had been allowed to dry; column B, colony count from a surface which had been wiped with a cloth immediately after the cloth had been used to clean a contaminated surface.

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uses and rinses some organisms, mostly Staph. aureus or Str. faecalis, were detected on the clean surface (Fig. 4 column B). Cloths left for 5 min after cleaning a contaminated surface were much less likely to transfer bacteria. None of the E. coli and P. aeruginosa was recovered, and small numbers of Staph. aureus and Str. faecalis were transfered only after a cloth had been used to clean six surfaces and had been rinsed a total of 15 times (mean count 13 c.f.u./surface).

DISCUSSION

The importance of effective cleaning in catering premises is not always understood. Although reusable wiping cloths remain popular, many are disinfected infrequently and are heavily contaminated with bacteria. Daily cleaning is not sufficient, and more reliable disinfection is necessary. Paper or disposable wipes could be considered, but these are more expensive than cloths, in some cases are less absorbent, and the accumulation and disposal of waste can present problems. Their use, however, would considerably reduce the risk of cross-contamination.

As far as possible a standardized cleaning technique was used in this study. Some variation between experiments occurred and may be associated with differences in drying times, in the amount of pressure applied to paper or cloths when cleaning surfaces, and in plating efficiency. A surface material commonly found in catering premises was chosen, but in practice other surfaces such as wooden chopping boards damaged by repeated use are probably more difficult to clean and disinfect. The presence of grease or dried food debris on surfaces could also influence cleaning results. To compare the effectiveness of different cleaning procedures, a range of marker organisms was examined. The presence of these organisms in anything more than minimal numbers suggests poor hygiene practices, and contamination of foods by *E. coli* suggests a potential risk of enteric pathogens (Mossel, 1982). Although relatively large numbers of bacteria were spread on surfaces (about 10^5 c.f.u.) previous work showed similar levels of contamination in wiping cloths obtained from food premises (Tebbutt, 1986).

Although field-tests have showed that direct swabbing techniques are superior to agar-impression methods for counting bacteria on surfaces (see Gilbert, 1970), both techniques produced similar results in the laboratory model described here. Several reasons could explain why this difference occurred. First, clumps of bacteria on food surfaces are more likely to be broken up when the rinse solution is shaken to dissolve the swab, whereas impression techniques do not distinguish between single cells and clumped organisms. In this study bacterial suspensions were agitated thoroughly before inoculation onto surfaces to break up large clumps or chains of bacteria. Secondly, the swab-rinse method is more likely to detect bacteria trapped in surface irregularities, e.g. as might be expected with wooden cutting boards in frequent use. In this study new Formica boards were used, and surface damage was probably minimal. Thirdly, in some studies selective or less nutritious media have been used in contact plates which might have contributed to the lower recovery rates using this technique. In this investigation a rich non-selective medium was used in both contact plates and for culturing swab-rinse solutions.

Several studies have suggested that the frequency and efficiency of cleaning

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procedures are more important than the inclusion of a disinfectant (Gilbert & Maurer, 1968; Tebbutt, 1984). It seems that a combination of detergent activity and physical removal of bacteria are important. In this study more satisfactory results were always obtained when hypochlorite was added to the detergent solution. One explanation for this might be that only light pressure was applied to paper during surface cleaning, and more vigorous wiping might have removed more bacteria in tests with detergent alone. The results, however, support the observation that a disinfectant used properly can provide an extra margin of safety.

The higher concentration of alcohol in wipe A than in wipe B (30% compared with 10%) probably explained its better performance in these laboratory tests. Both cloths, however, contained considerably less than the optimal concentration of alcohol for killing organisms. The presence of cetrimide in wipe B might explain why it worked well against Gram-positive organisms. One disadvantage of alcohol is its poor penetrative powers such that it cannot be used to disinfect dirty surfaces. A field-study is needed to determine the value of alcohol wipes for routine use in catering premises.

It is impossible to determine accurately how much active biocide remains in a cloth impregnated with disinfectant. Blue indicator dye provides only an approximate guide. This study showed that breakthrough by some bacteria occurred relatively easily, and it was not possible to predict disinfection failures from the appearance of the cloth. Because these cloths are relatively expensive, and catering staff may be reluctant to replace them frequently, such cloths cannot be relied upon for surface disinfection unless their use is carefully supervised.

As a follow up to this study a field-trial is planned to evaluate some methods of cleaning surfaces in local food-manufacturing premises. The performance and acceptibility of a disposable alcohol wipe, a combined detergent/hypochlorite solution applied with paper, and of a reusable cloth which is kept in detergent/ hypochlorite solution between each use will be compared. In this way it is hoped to suggest a practical method of reducing the risk of cross-contamination which is presently associated with reusable cloths used in a large number of commercial food premises.

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