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# Contamination of chlorhexidine cream used to prevent ascending urinary tract infections

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## SUMMARY

Chlorhexidine-containing cream is often used as an antimicrobial barrier to ascending urinary tract infection in patients with indwelling urethral catheters. The cream is dispensed in small tubes for personal use but repeated use of a tube still entails a potential infection hazard. The extent of cream contamination was analysed by emulsifying it in 1% peptone broth with 1% Tween-80 added as a wetting agent, and culturing quantitatively for bacteria and fungi by membrane filtration. Twenty-three per cent of cream samples and 35% of swabs taken from outside the tube beneath the screw cap demonstrated microbial contamination. Isolates included potential pathogens such as enterococci, staphylococci, *Proteus mirabilis*, *Pseudomonas aeruginosa*, opportunists like *Moraxella* spp. and diphtheroids, and contaminants such as *Bacillus* spp., micrococci, and a mould of the genus *Cladosporium*. Contamination of cream with a particular bacterial strain was found to precede urinary tract infection with the same microbe. We recommend that chlorhexidine cream for this use be dispensed in single dose units to ensure sterility.

## INTRODUCTION

The calamities that may arise when pharmaceutical preparations for intravenous use are contaminated have been well documented (Editorial, 1971; Goldman, Martin & Worthington, 1973; Maki, Anderson & Shulman, 1974). The Public Health Laboratory Service reported on the contamination of 1220 samples of preparations for oral administration or topical use collected in 19 British hospitals (Working Party, 1971). Eighteen per cent of the specimens contained more than $10^4$ viable organisms per ml or g. The ubiquitous pathogen *Pseudomonas aeruginosa* was identified in 3% of samples and other Gram-negative pathogens in another 9%.

The consequences of contaminated parenteral solutions are dramatic but contamination of topical preparations may also cause serious infections (Baird, Awad & Shooter, 1980). The working party of the Public Health Laboratory Service (1971) and various pharmacopoeias (Pharmacopoea Nordica (1966),

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United States Pharmacopeia (1975)) recommend conditions for production and sterility testing to ensure contamination free topical products. However, lack of microbes after preparation does not ensure sterility during use (unless the product is in single dose containers). We report on how sterile chlorhexidine cream may become contaminated during use and cause the urinary tract infections that the preparation is intended to prevent.

## MATERIALS AND METHODS

### *Cream*

The preparation studied was 'Kateterkrem' from Norges Apotekerforening (NAF). The formulation consists of 1% chlorhexidine acetate in a base of 25 g hypromellose-4000, 200 mg propyleneglycol, and 1000 g of aqua purificata. The water is distilled and autoclaved before use. The product is sterilized by autoclaving and dispensed in tubes of 6 g. Its sterility is controlled as specified by Pharmacopoea Nordica (1966).

### *Test bacteria*

To test the recovery methods samples of cream contaminated with known numbers of standard bacterial strains were used. The strains used were those proposed as test microbes in the European Pharmacopoeia, i.e. *Escherichia coli* ATCC 8739, *Bacillus subtilis* ATCC 6633, *Staphylococcus aureus* ATCC 6538, and a strain of *Pseudomonas aeruginosa*.

### *Microbial recovery*

The methods previously used to recover bacteria from ointments and creams have produced low yields; so various alternatives were tried. As the cream is water soluble, one would expect that mixing in any fluid would have been satisfactory. However, the recovery from using peptone broth with saline was not acceptable and so a wetting agent (1% Tween-80) was added. The final recovery medium contained 1% peptone broth, 1% Tween-80 and 0·5% sodium chloride (to stabilize the osmotic conditions and improve microbial recovery).

Samples (0·1 g) of cream were added to 10 ml of recovery medium and mixed in a whirlymixer; the liquid was then filtered through a 0·04 $\mu$m membrane filter (Millipore) which was then washed with 200 ml of broth (in 4 aliquots) to remove any residual chlorhexidine. Afterwards the filters were gently transferred to the surface of HS-T Agar (Oxoid, London, U.K.) (Clausen, Aasgaard & Solberg, 1973; Pharmacopoea Nordica, 1966). This medium is used to recover microbes after exposure to disinfectants and may neutralize any small residual quantities of chlorhexidine that have remained on the filters despite washing.

Microbial growth was determined after 48 h incubation at 30 °C. Subcultures were made on blood agar (Tryptone Blood Agar Base (Oxoid) with 5% human blood) and lactose bromothymol blue agar. Identification was made by standard determinative methods (Lassen, 1975) and API-E, API-S, and API-A (Analab,

Inc., New York, N.Y., U.S.A.) for the species identification of Gram-negative aerobic bacteria, staphylococci, and anaerobic bacterial strains respectively.

*Use of cream before sampling*

The tubes of chlorhexidine cream were intended for use twice daily. A fresh tube was used in the morning and then placed in the drawer of the patient's bedside table where it lay without protection until reuse. The tubes were used by removing the screw cap and squeezing out some cream directly onto the urethral orifice. The tip of the tube (4 mm wide and 5 mm long) invariably touched the urethral catheter or surrounding mucosa and skin.

*Sampling*

In normal use around 10 h passed between the first and second application of the cream. Therefore, after 10 h, instead of reuse the sample tubes were removed from the drawers for microbial examination. The results should thus have reflected exactly the flora to which the patients would have become exposed by use of contaminated cream.

Each tube was sampled three times. The first sample was 0·1 g of cream which was transferred to the recovery broth and homogenized for 2 min in the whirlymixer. The second sample was a swab from the external surface of the tip of the tube normally covered by the screw cap. Cotton swabs moistened with 1% peptone broth were used and any cream that was present within the grooves of the screw thread was collected; however, areas not normally covered by the screw cap were not touched. The last sample consisted of the outer 3 mm of the application tip which was cut off with sterile scissors and transferred to the peptone recovery medium for whirlymixing. Ten-fold dilutions of the recovery broth were used for membrane filtration and counts were made from the filters that had yielded 100–400 colonies.

## RESULTS

*Recovery of test bacteria*

The difficulty of quantitative bacterial recovery from ointments is well documented (Allwood & Hambleton, 1973; Sokolski & Chidester, 1964; Tsuji & Robertson, 1970; Walsh & Allwood, 1973). Although preparations containing paraffin have a low water miscibility and represent particular problems, our cream was water soluble and so should have been easier to sample. However, previous experience has amply demonstrated that the test procedure must be controlled carefully for each different type of cream base as different technical problems are involved. The method we have described was the most efficient of several we studied. Parallel examinations were made of the cream base and the cream containing 1% chlorhexidine acetate. The contact time between adding known numbers of test bacteria to the cream and recovery was 5 min, and the results of recovery of standard strains are shown in Table 1. There was complete recovery

Table 1. *Recovery of bacteria of known quantity from cream base and cream with 1 % chlorhexidine acetate*

| Test strain | Number of organisms added per g $\times 10^{-8}$ | Recovery from cream base (%) | Recovery from cream with chlorhexidine (%) |
|---|---|---|---|
| *Bacillus subtilis* | 2·0 | 79 | 35 |
| *Escherichia coli* | 3·3 | 86 | 55 |
| *Pseudomonas aeruginosa* | 1·8 | 116 | 107 |
| *Staphylococcus aureus* | 3·6 | 104 | 85 |

Table 2. *Number of contaminated cream and tube tip samples*

| Microbial strains per sample | Cream | Tip of tube |
|---|---|---|
| 0 | 66 | 61 |
| 1 | 12 | 18 |
| 2 | 8 | 7 |
| 3 | 3 | 3 |
| Total samples | 89 | 89 |
| Total positive | 23 | 28 |

from the cream base. The range of 79 % (*Bacillus subtilis*) to 116 % (*Pseudomonas aeruginosa*) suggests random variations consistent with the technical method. The lower recovery from medium containing chlorhexidine demonstrates the antibacterial effect of this agent, except against Pseudomonas which is well known to be resistant to chlorhexidine (Bergan, 1977).

*Microbial recovery from patients' cream*

The extent of microbial contamination of the tubes of chlorhexidine cream at the time of their intended use is shown in Table 2. Twenty-three of 89 samples of the cream and 28 of 89 samples of the tube tips were contaminated and 39 strains of bacteria were isolated. Thus, use of cream contaminated from previous use may occur frequently. The patients' urines were always examined at the time when their chlorhexidine creams were examined for a complete microbial picture. In one case a strain of *Proteus mirabilis* was isolated from cream 7 days before the same strain caused urinary tract infection. The urine of this patient did not contain bacteria when the cream was examined. After a week, the patient had acquired clinical symptoms of urinary tract infection and the urine contained more than $10^5$ bacteria per ml. Consequently, a causal relationship between the use of contaminated cream and a subsequent urinary tract infection is possible.

## DISCUSSION

Table 3 shows the types of bacteria isolated. The Gram positive strains represent mostly normal skin flora. Enterococci were the only likely pathogens and no strains

Table 3. *Microbial species isolated*

| Species | Cream | Tip of tube |
|---|---|---|
| Gram-positive bacteria | | |
| *Bacillus* sp. | 2 (2)* | 6 (4) |
| *Corynebacterium* sp. | 7 (1) | 8 (2) |
| Enterococci | 8 (6) | 6 (6) |
| *Micrococcus* sp. | 8 (2) | 11 (3) |
| *Staphylococcus aureus* | 0 | 3 (1) |
| *S. epidermidis* | 19 (3) | 14 (2) |
| Gram-negative bacteria | | |
| *Moraxella* sp. | 2 (2) | 0 |
| *Proteus mirabilis* | 7 (1) | 5 (1) |
| *Pseudomonas aeruginosa* | 1 (1) | 2 (0) |
| Fungi | | |
| *Chladosporium* | 0 | 1 (1) |
| Unidentified mould | 0 | 2 (1) |

* Numbers in parenthesis indicate strains isolated in monoculture.

of *Staphylococcus saprophyticus* were isolated. The Gram negative bacteria found included both *Proteus mirabilis* and *Paeudomonas aeruginosa* which are well established as problem microbes of the urinary tract of patients with indwelling catheters. Moulds were contaminants. Tubes of the chlorhexidine cream used as a barrier against ascending urinary tract infections in patients with indwelling catheters have been provided for repeated use by the individual patient. By restricting one tube to one patient cross-infection is avoided. But the question was, would repeated use of individual tubes be undesirable? Our study clearly shows that the cream and the screw thread of the tubes become contaminated during use so that repeated use of the same tube represents an infection hazard. Contamination of the cream is unavoidable during use.

The method used to elute bacteria from the cream was effective, 79–116% recovery being found with standard bacterial test strains. This range compares favourably with previously reported recovery rates, which ranged from 50 to 82% (Allwood & Hambleton, 1973); Sokolski & Chidester, 1964; Tsuji & Robertson, 1970). One reason for our better success may be that we examined water miscible cream, whereas many of the lower recovery rates apply to paraffin based ointments.

Our recovery method combined previously suggested improvements. Thus, Sokolski & Chidester (1964) demonstrated enhanced recovery with membrane filtration, which allows residual antibacterial substances to be washed away before cultivation. Mixing cream with isopropyl myristate gave bacterial recoveries of 30–73%. Allwood & Hambleton (1973) and Walsh & Allwood (1973) reported that improved recovery could be obtained by combining membrane filtration with a medium containing 1% peptone and 1% Tween-80. This raised recovery rates for *Escherichia coli* to 74–82% compared with 5–11% with isopropyl myristate. Our tests also showed less recovery in the absence of Tween-80. The bacterial species recovered from the chlorhexidine cream 10 h after it had been used was

relevant to their intended use. As Table 1 shows that chlorhexidine reduces the count of the test organisms after only 5 min exposure the bacterial numbers in the medium immediately after its use must have been high. The bacteria isolated reflect the flora of the mucosa and the skin around the urethral orifice. Contaminants, such as the *Bacillus* sp. and the *Cladosporium* sp., may have been acquired during the time in the patient's bedroom table drawer since these microbes were more frequently isolated from the screw thread caps of the tubes. We must stress that surfaces not covered by the screw caps were carefully avoided. Accordingly, the microbes derived from the external surface and tube tips can be presumed to be available for contamination of the cream, the catheter and the mucosa around the urethra. The surface contamination might be reduced if the tubes were kept in plastic covers or other protection between uses.

The potential hazard of contaminated cream is well illustrated by the patient who developed a urinary tract infection with a strain of *Proteus mirabilis* which had previously been isolated from the cream. If the contaminated cream is applied to the space between the catheter and the urethral mucosa ascending infection with chlorhexidine resistant bacteria is likely.

The Pharmacopoea Nordica and other pharmacopoeias require that cream applied on mucosal surfaces must be sterile. This in itself makes it intolerable that a preparation is contaminated during use and still not discarded. One might, perhaps, argue that a certain level of contamination could be tolerable compared with the hygienic and clinical risks involved. Non-pathogenic species were isolated and these would appear to represent a minor problem. A rather different situation applies to the potential pathogens which constitute a significant portion of the isolates.

Contamination might be lower if the tubes were equipped with narrow, long tips of the kind used for eye ointments, instead of the wide necked tubes presently used. However, contamination would occur even with such modifications. Since the number of specimens with contaminated cream was so high (one quarter of the samples), repeated use even to the same patient clearly represents an intolerable hygienic hazard counteracting the very purpose of the preparation, i.e. prevention of ascending urinary tract infections in patients with indwelling catheters. It is consequently recommended that the cream be dispensed in smaller single-dose tubes. The pharmaceutical requirements of sterility for preparations with intended mucosal surface contacts are there, but the purpose must be followed up also by the way in which the products are used and handled in the clinical departments.

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