EFFECT OF PRE-OPERATIVE HAND SCRUBBING AND INFLUENCE OF PINHOLES APPEARING IN SURGICAL RUBBER GLOVES DURING OPERATION

BY

Masayoshi Furuhashi and Takuyuki Miyamae*1

ABSTRACT

Evaluation of a handwash can be made by various methods. The effect of handwashing and disinfectants was evaluated by scrubbing of the hands and forearms with soap and a brush before and after the application of a disinfectant, and by quantitatively determining the viable counts of microorganisms which fell off into the water in the basin. The disinfectants studied were 4% chlorhexidine detergent, 10% benzethonium chloride, and 7.5% povidone-iodine, as well as 70% and 90% ethanol and 50% isopropanol. Some other preparations were also examined. With chlorhexidine detergent, benzethonium, or povidone-iodine, no significant bacterial reduction was obtained by only a single 2-min brushing, but by two 3-min brushings, reduction of 87.8% to 94.5% was obtained. When a further treatment was tested in which the hands and forearms were thoroughly rubbed with 0.5% chlorhexidine in 90% ethanol or 0.5% benzethonium chloride in ethanol solution, which was taken on the palms, following the above 6-min brushing, significant bacterial reduction of 99% or more was obtained in a short period of time.

Small pinholes develop in surgical gloves during the course of surgical operation, at the finger tip of the gloves in most cases. Experimental pinholes were made in gloves, and the degree of transfer of bacteria from the skin of the hand was examined. After brushing of the hands and forearms with an effective disinfectant, the bacterial count transferring from the hand was markedly small.

INTRODUCTION

Hand washing before surgical operation is very important for the prevention of cross-infection. Usually, pre-operative hand treatment is performed in hospitals under stricter hand washing standards than the hand disinfection in wards and out-patient clinics. Disinfectants for such uses have been developed, and many effective antiseptic handwashes have recently been introduced in Japan. We have performed various in vitro studies on anti-bacterial action of these newly introduced disinfectants and, this time, we performed in vivo study on several hand disinfectants, results of which are reported here.

Our main test method is a modification of Price’s serial basin hand-washing test method.4) The quantity of specimen solutions taken from a basin was relatively large so that error in the estimation of bacterial counts would be minor. Bacteria were collected by the filter method for quantitative determination of the bacterial counts.

The antiseptic detergent preparations used in our tests were as follows:

(1) 4% Chlorhexidine digluconate detergent solution (Hibiscrub)
(2) 10% Benzethonium chloride detergent

*1 吉橋正吉・宮前卓之： Department of Operating Center (Chief: Prof. K. Asano), Hospital attached to the Faculty of Medicine, Tokyo Medical and Dental University (Tokyo Ika Shika Daigaku).
Received for publication, March, 12, 1979.
solution (Hyamine-T)
(3) 7.5% Povidone-iodine (0.75% available iodine; Isodine Surgical Scrub)
(4) 70% Ethanol
(5) 50% Isopropanol
(6) 0.5% Chlorhexidine in 70% ethanol
(7) 90% Ethanol
(8) 0.5% Chlorhexidine in 90% ethanol
(9) 0.5% Benzethonium chloride in 90% ethanol
(10) Unmedicated bar soap, Pluronic F-87, and Triton X-100 (control application)

The subjects participating in the study were limited to only two persons (a surgeon and an assistant) to avoid, as far as possible, variation in scrubbing techniques. The studies were repeated during the past three or more years, once a week, ensuring a 1-week interval for the skin bacterial flora to recover to the normal number. We also studied the effect of pinholes appearing in surgical rubber gloves during the operation.

MATERIALS AND METHODS

1. Comparison of the effect of various disinfectants by the serial basin hand-washing test

Eight sterile basins were each filled with 2 liters of sterile distilled water. The hands and forearms up to the elbows were scrubbed with a Nylon brush impregnated with about 200 mg of soap, Pluronic F-87, or Triton X-100, each time for a period of 2 min. After scrubbing, the hands and forearms were rinsed thoroughly in water in another basin.

At the completion of one cycle, the same maneuver was repeated at the next basin. This way of hand washing was repeated eight times, and the whole serial hand washing was regarded as the 'control'.

Antiseptic handwashes were applied between 4th and 5th hand washings in the serial basin hand washing test, to evaluate their antimicrobial effect. After the 4th scrubbing with soap and a brush, the remaining soap was rinsed off under running water, and either one of the following disinfectants was applied to the hands and forearms in the following manner.

In the case of chlorhexidine detergent (Hibiscrub), benzethonium chloride (Hyamine-T), or povidone-iodine (Isodine), 5 ml was taken on a brush, and the hands and forearms up to the elbows were scrubbed with the brush for 2 min, followed by a rinse under running water.

In the case of 70% ethanol, 50% isopropanol, or 0.02% chlorhexidine in 70% ethanol solution, the hands and forearms were immersed in the alcoholic solution for 2 min, in one test, without any rubbing; and in another test, together with concurrent gentle rubbing with a urethan sponge.

In both of above cases, further serial four hand-washings with a brush and soap were performed after the application of a test disinfectant.

Appropriate neutralizers were added when necessary to the basins to collect the fallen-off bacteria just after the application of the antiseptic handwashes. The neutralizer used was composed of lecithin (0.5%), Tween-80 (1%), and Lubrol-W (1%). For Isodine, sodium thiosulfate (2%) was used.

Sampling from the respective eight basins of the control series and the test series was made by taking exactly 0.1 and 5 ml of the solutions from each basin just after each rinsing was finished. Specimen solutions were filtered through a membrane filter (Nuclepore Filter; pore size: 0.2 μm; diameter, 47 mm), and the membrane filter taken from the filter body was aseptically placed on a Trypticase soy agar medium (diameter, 90 mm) for the aerobic incubation at 32°C for 24 and 48 hr. The number of colonies
grown on the filter was counted, and by multiplying this number by 400 (5 ml sample) and 20,000 (0.1 ml sample), the viable count of bacteria in 2 liters of water was obtained.

II. Effect on reduction of bacteria by the combined use of a disinfectant and an alcoholic solution

As shown in experimental method I, it was clear that an alcoholic solution had a strong bactericidal effect with quick action. Rubbing of the hands and forearms with a urethan sponge in an alcoholic solution in a basin was especially effective. This method, however, is not so practical since a large quantity of alcohol is necessary. Lowbury\(^3\) presented a method in which a small quantity of an alcoholic solution was taken on the palms and applied over the hands and forearms. This method is simpler and more practical than the rubbing method. The application of this method into practice, however, revealed that by this method it was hard to disinfect the nail area where a rather large number of bacteria was present. We therefore designed a combination hand washing with brushing with a disinfectant and rubbing with an alcoholic solution, and examined its effect. This method is practiced in the following way:

Hand brushing with unmedicated bar soap (1 min) hand brushing with either Hibiscrub or Hyamine-T (3 min) hands and forearms dried on a towel (1 min). An alcoholic solution of a disinfectant is applied on hands and forearms, and allowed to become dry (about 2 min) the same alcoholic solution is applied on hands below the wrist, especially on the nail area and hands are rubbed until they become dry (about 2 min).

In order to determine the bacterial counts on the skin before the application of disinfectants, the hands and forearms were brushed with soap for 2 min and washed in 2 liters of sterile distilled water in a basin. The hands and forearms were then rinsed thoroughly under running water so that no soap remained on the skin. They were then scrubbed with 5 ml of either Hibiscrub or Hyamine-T contained in a sterile brush, for 3 min. The hands and forearms were rinsed under running water and dried on a sterile towel. Ten milliliters of 0.5% chlorhexidine in 90% ethanol and 0.5% benzethonium chloride in 90% ethanol were respectively taken on the palm, applied evenly over the hands and forearms, and rubbed in until they became dry. Then, 5 ml of the same alcoholic solutions was applied well on the hands and wrists, especially on the nail area, and was rubbed in until the hands became dry. It took about 2 min for the alcohol to dry up.

After completion of the above procedure, the hands and forearms were scrubbed with soap and a brush, and washed in 2 liters of sterile distilled water in a basin, to evaluate the bactericidal effect of the disinfectant.

Further, to determine the bactericidal effect maintained on the skin after a certain lapse of time, sterile surgical gloves and a surgical gown were worn just after the hands dried after the second application of an alcoholic solution; and the subjects carried out their routine work for 3 hr. After the rubber gloves were taken off, the hands and forearms were scrubbed with soap and a brush for 2 min and rinsed in water.

Bacterial counts were taken on three occasions; before, just after, and 3 hr after the application of a disinfectant, by brushing the hands and forearms with soap for 2 min and washing them in water in a basin, and the in results were compared.

The use of neutralizers and bacterial in-
cubation methods were the same as those used in Experiment I.

As the number of bacteria which fell off into the basins was small in both cases, i.e., just after the hand washing with a disinfectant and 3 hr after the hand washing, 20 and 40 ml of specimen solutions were taken from each basin.

As the control, the hands and forearms were scrubbed twice for 3 min with a brush with Hyamine-T and then immersed in 0.02% chlorhexidine in distilled water in a basin for 1 min.

III. Pinholes in surgical rubber gloves developing during the operation and skin bacterial counts transferring from the hand

The surgical gloves examined totalled 2,284 gloves (Thoma) used in 220 operations in our theater, plus others in which pinholes were made artificially.

The presence of pinholes in rubber gloves was detected in the following way: Each of the rubber gloves collected just after each operation was filled with tap water and the presence of pinholes was checked by the leakage of water from the glove after closing the wrist of the glove by the left hand and pressing the water in the body of the glove by the right hand. The number of holes and their places were recorded on each glove.

For the investigation of the skin bacterial counts escaping from the hands through pinholes, surgical gloves were punctured artificially with one pinhole in each finger tip and in the middle of the palm with an 18.5-gauge needle. After being sterilized, these gloves were worn, and one hand was immersed up to the wrist in 2 liters of sterile distilled water in a basin and the fingers were folded and extended 120 times over the period of 5 min. From this basin, 50 ml of the specimen solution was sampled from 2 liters of water, filtered through a membrane filter, and incubated on a Trypticase soy agar medium at 32°C for 48 hr. The bacterial count escaping from one hand into distilled water was then calculated from the number of colonies grown on the medium.

When a neutralizer was necessary, an appropriate neutralizer for the disinfectant tested was added to the sterile distilled water.

Results
I. Comparison of the bacterial reduction of unmedicated detergents and various disinfectants

(A) Bacteria reduction after the application of unmedicated detergents

The bacterial reduction rates in the course of the serial 2-min hand washings were 39.4% with bar soap, after 4 min and 57.7%, even after a total of 16 min. With Pluronic F-87, 79.3% reduction was obtained after a 16-min brushing, the skin bacterial counts being $10^4$ to $10^5$ (Table 1).

(B) Bacteria reduction after the application of disinfectants

The bacteria reduction effect of various disinfectants was compared. The bacteria reduction rate of a disinfectant was obtained from the pre-hand washing bacterial count (bacteria falling off into 2 liters of water in the first basin in the serial basin scrubbing, after the first 2-min brushing with bar soap and a brush) and counts of bacterial falling off into basins after 2, 3, and 6 min of scrubbing with the disinfectant, by calculating the latter as a percentage of the former.

After a 2-min brushing, 10% benzethonium chloride showed the highest bacteria reduction rate of 87.6%. After a 3-min scrubbing, 4% chlorhexidine detergent was best, showing a reduction rate of 89.2%. When scrubbing for a two 3-min periods, both disinfectants produced high bacteria
reduction rates of 98.8\% for the former and 94.5\% for the latter (Table 2).

(C) Bacteria reduction after the application of alcoholic solutions

With 70\% and 90\% ethanol, and 50\% isopropanol, the soaking method (in an alcoholic solution for 2 min) and the rubbing method (in an alcoholic solution for 2 min; the hands and forearms being gently rubbed with a urethan sponge while being soaked) were compared. The results revealed that the rubbing method produced a better reduction rate than mere soaking of the hands. When using the rubbing method with a sponge, 70\% ethanol showed higher bacteria reduction rate than 50\% isopropanol (Table 3).

II. Bacteria reduction effect of a combined use of a disinfectant and an alcoholic solution

Although skin bacteria counts before dis-
Table 3. Disinfection of Hands by Various Preparations: Reduction in Bacterial Counts from Hand Washing (2-min), Expressed as Percentage of Initial Counts

<table>
<thead>
<tr>
<th>Antiseptic preparations</th>
<th>Method of application</th>
<th>No. of experiments</th>
<th>Mean reduction in viable counts (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>70% ethanol</td>
<td>Hands immersed in solution and dried on towel</td>
<td>6</td>
<td>(A) 77.0 ± 9.5</td>
</tr>
<tr>
<td></td>
<td>Hands washed with urethan sponge and dried on towel</td>
<td>11</td>
<td>(B) 97.0 ± 2.1</td>
</tr>
<tr>
<td>50% isopropanol</td>
<td>Hands immersed in solution and dried on towel</td>
<td>5</td>
<td>(C) 67.9 ± 10.1</td>
</tr>
<tr>
<td></td>
<td>Hands washed with urethan sponge and dried on towel</td>
<td>6</td>
<td>(D) 90.3 ± 7.8</td>
</tr>
<tr>
<td>0.5% chlorhexidine in 70% ethanol</td>
<td>Rubbed in till hands dried</td>
<td>4</td>
<td>(E) 90.5 ± 2.8</td>
</tr>
<tr>
<td>0.5% chlorhexidine in 90% ethanol</td>
<td>Rubbed in till hands dried</td>
<td>4</td>
<td>(F) 91.8 ± 9.1</td>
</tr>
</tbody>
</table>

Comparison of treatment:
(A) vs. (B): t = 6.88, P < 0.01
(B) vs. (E): t = 4.89, P < 0.01
(B) vs. (D): t = 2.74, P < 0.01

Infection were over 10⁶, the application of a disinfectant in our said method resulted in the reduction of bacterial counts to 10³ or less just after disinfection, producing a bacteria reduction rate of 99.7%. The bacterial counts were maintained also in a level of 10³ or less, even 3 hr after disinfection, showing a reduction rate of 99.9% (Table 4).

III. Pinholes in surgical rubber gloves appearing during operation and the skin bacterial counts transferring from the hand

(A) The ratio of the number of rubber gloves in which pinholes appeared during operation to the total number of gloves used in operation was obtained for each medical department involved. The mean ratio was 14.8% among all the departments. The means for surgeons and nurses were 12.3% and 22.8%, respectively, showing a higher rate for the nurses. The number of pinholes appearing in a glove was one to two in most of gloves. It was rare that one glove had six or more pinholes.

The total of 722 pinholes appearing in 339 rubber gloves consisted of 297 in the gloves worn on the right hand and 425 in the gloves worn on the left hand. Thus pinholes appeared more in left hand gloves. A breakdown of the number by palm and dorsum revealed that the pinholes in the palm (559) were 3.4 times more than those in the dorsum (163), while 92% (664) of the total pinholes appeared in the fingers of the gloves, especially within the areas from the first joint to the finger tip. As to the distribution of these pinholes by fingers, such punctures appeared most in the index finger of the left hand (203), followed by the
Table 4. Effect of Wearing Surgical Gloves on Skin Bacteria after Disinfection of Hands: Reduction in Bacterial Counts from Hand Samplings, Expressed as Percentage of Initial Counts

<table>
<thead>
<tr>
<th>Disinfectants</th>
<th>Method of application</th>
<th>Mean reduction in viable counts (%)</th>
<th>Immediately after disinfection</th>
<th>After gloves worn for 3 hr after disinfection</th>
</tr>
</thead>
<tbody>
<tr>
<td>4% chlorhexidine detergent and chlorhexidine (0.5%) in 90% ethanol</td>
<td>Washed with brush for 3 min and rubbed in till hands dried (2 times)</td>
<td>(A) 99.7±0.48 (n=17)</td>
<td>(D) 99.9±0.04 (n=4)</td>
<td></td>
</tr>
<tr>
<td>10% benzethonium chloride detergent and benzethonium chloride (0.5%) in 90% ethanol</td>
<td>Washed with brush for 3 min and rubbed in till hands dried (2 times)</td>
<td>(B) 99.7±0.45 (n=8)</td>
<td>(E) 99.9±0.66 (n=4)</td>
<td></td>
</tr>
<tr>
<td>10% benzethonium chloride detergent and chlorhexidine (0.02%) in water</td>
<td>Washed with brush for 3 min (2 times) and immersed in solution (1 min)</td>
<td>(C) 99.2±0.68 (n=8)</td>
<td>(F) 99.9±0.05 (n=8)</td>
<td></td>
</tr>
</tbody>
</table>

Comparison of treatments:
(A) vs. (B): Not significant
(B) vs. (C): Not significant
(D) vs. (E): Not significant
(E) vs. (F): Not significant

thump of the left hand, index finger of the right hand, and thumb of the right hand, in this order.

(B) The skin bacterial counts transferring from a hand through pinholes experimentally punctured were $10^3$ to $10^4$ when the hands and forearms were scrubbed without a disinfectant. In contrast, when the hands and forearms were brushed with a disinfectant, bacterial counts were not more than 100, even 3 to 4 hr after hand washing.

**Discussion**

There are many methods to evaluate hand disinfection procedures. We have used a method in which the post-disinfection hands and forearms were scrubbed with soap and a brush, and the number of living bacteria which fell off into the water in a basin was qualitatively determined. This is a well-known method developed by Price, and is the most strict method for evaluating hand washing.4-6

The membrane filter method used for collection and incubation of bacteria is better than the mix-dilute method because, by this method, it is possible to detect viable counts even from specimens containing a very small amount of bacteria, by using increased amount of the specimen solution, and because this method produces almost the same size colonies on the medium, which provides simplicity in counting. It has also been confirmed that there is no significant difference in the colony counts after incubation between the mix-dilute method and the filter method.

When evaluated by Price’s method, 2-min brushing of the hands and forearms with Hibiscrub, Hyamine-T, or Isodine did not provide marked bacteria reduction. However, 6-min brushing (two 3-min washings)
with Hibiscrub or Hyamine-T produced a remarkable bacteria reduction effect. These disinfectants are more quick-acting than Phisohex. However, we think that these do not produce their full effect when applied only for single 2 min.\textsuperscript{1)}

Alcoholic solutions are more quick-acting than these disinfectants. Especially, application of 0.5\% chlorhexidine gluconate in 90\% ethanol or 0.5\% benzethonium chloride in 90\% ethanol by rubbing onto the hands and forearms was simple and could produce a remarkable bacteria reduction effect of 99\% or more in a short period of time. This method gives a bacteria reduction rate of around 90\% with only one application. Therefore, this is recommended for hand disinfection for emergency surgical treatment or hand disinfection in wards.

Lowbury reported a reduction rate of 97.9\% after a single application of 0.5\% chlorhexidine gluconate in 95\% ethanol.\textsuperscript{2)}

We tried to prepare the alcoholic solution of the same formulation but at this concentration, complete dissolution did not occur and white turbidity was produced. Therefore, we reduced the concentration of ethanol to 90\%. Further, although they added 1\% glycerol to the above alcoholic solution for the reason that Lowbury’s participants felt discomfort when alcohol vaporized, we did not add it because ours did not feel special discomfort.\textsuperscript{2)}

It is better to prepare this alcoholic chlorhexidine solution from Hibitane Concentrate solution (5\% chlorhexidine and 3.75\% surfactant solution) than from Hibitane Gluconate solution (20\% chlorhexidine solution), as the former preparation did not leave stickiness on the skin after vaporization of alcohol and it was thus easier to wear rubber gloves.

Most pinholes appearing during operation are in the finger tips. When performing pre-operative hand washing, therefore, we should concentrate on deterging and disinfecting the hands below the wrists, especially on the finger tips, as well as on the use of an effective disinfectant.

Acknowledgement

The authors wish to thank Prof. R. Nakaya of the Department of Microbiology, Tokyo Medical and Dental University, for many helpful discussions and suggestions during this work.

References


