Bacteriological Evaluation of Antiseptic Solutions Used to Keep Cheattle's Forceps

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Received: 20 February 2014 / Accepted: 26 May 2014 / Published online: 11 June 2014
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Abstract There is little information available on the bacteriological contamination of Cheattle's forceps during routine use and the effects of antiseptic solutions. This study was conducted to detect infection in various antiseptics and disinfections in-use used for keeping the Cheattle's forceps in surgical wards. An aqueous solution of 0.5 and 1 % cetrimide/chlorhexidine, 2.4 % glutaraldehyde, 5 % povidone iodine, and 10 % chloroxylenol were compared using the normal saline as control. The samples from each bottle were sent for culture on the 3rd, 5th, and 7th day. All the culture reports were analyzed and statistically compared. A total of 2,160 samples were sent for culture from solutions including control over a period of 7 days. A total of 360 samples were sent for culture from each antiseptic solution. Out of 2,160 samples, 240 (11.11 %) were positive over a period of 7 days for contamination with microbial organisms. The most common organism isolated was *Pseudomonas* present in 140 samples followed by *E. coli* in 71 samples. *S. aureus* was present in seven samples, bacillus in ten samples, and Coagulase -ve *Streptococcus* and *Klebsiella* in six samples each. Number of positive culture consistently increased in normal saline and cetrimide/chlorhexidine, but povidone iodine, chloroxylenol, and glutaraldehyde did not show any positive culture on the 3rd day. Povidone iodine and glutaraldehyde showed positive culture on the 5th and 7th day, respectively, but they were statistically insignificant. On the 7th day, glutaraldehyde was the only solution which did not show any positive culture. We strongly recommend that the Cheattle's forceps should be kept in a bottle containing glutaraldehyde or chloroxylenol.

Keywords Disinfectants · Bacterial flora · Contamination · *Pseudomonas*

Introduction

Cheattle's forceps are commonly used in surgical wards and operation theaters to transfer sterilized instruments and materials to the doctors' hands. This forceps is kept in a variety of antiseptic/disinfectant solutions, often in open bottles which are unchanged for many days. Contamination of these solutions is a likely possibility, thus conveying infection, via the use of Cheattle's forceps. Surprisingly, there is no information about this important issue. Hence, this study was conducted to test the presence of bacteriological contamination in various antiseptics and disinfections used for keeping the Cheattle's forceps in surgical wards.

Methods

This study was conducted in the Department of Surgery, NSCB Government Medical College, Jabalpur (MP) India, a tertiary teaching institution in Central India over a period of 6 months from January 2012 to June 2012. Five solutions were assessed for contamination and were compared with normal saline as control. Cheattle's forceps were kept in the dressing trolley in the surgical wards in various solutions for this study. These five solutions were as follows:

1. 0.5 % cetrimide and chlorhexidine combination (Savlon, Manufactured by Johnson & Johnson, India)
2. 1 % cetrimide and chlorhexidine combination (Savlon, Manufactured by Johnson & Johnson, India)
3. 5 % povidone iodine (Betadine, manufactured by Win-Medicare Pvt. Ltd. India)

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4. 10 % chloroxylenol (Dettol, manufactured by Reckitt Benckiser, India)
5. 2.4 % glutaraldehyde (Cidex, manufactured by Johnson & Johnson, India).

Sterile glass bottles 500 ml were used to keep Cheattle's forceps, and these bottles were filled two-thirds with different solutions. These bottles were not capped. The regular dressings were performed using Cheattle's forceps to transfer the sterile swabs, cotton bandages, and instrument from autoclaved drums to the surgeon's hand. The samples from each bottle were sent for bacteriological culture on the 3rd, 5th, and 7th day. The solution was not changed for 1 week. On the 7th day, sterile bottles filled with different antiseptic solutions were replaced with the same fresh solution. All the culture reports were analyzed and statistically compared using SPSS 18 (SPSS Inc. Released 2009. PASW Statistics for Windows, Version 18.0. Chicago: SPSS Inc.) Statistical analysis was done using the Fisher's exact test and proportion (Z test) with critical level at 0.05.

Swabs from different antiseptic solutions were inoculated in the MacConkey Agar and blood agar plate and incubated for 18–24 h at 37 °C aerobically. These agar plates were observed for the growth of bacteria and confirmed using standard protocol like gram staining, motility testing, and biochemical reaction. The presence of colony was taken as an indicator of contamination. Only aerobic cultures were performed, no fungal or anaerobic cultures were done.

Results

A total of 2,160 samples were sent for culture from solutions including control over a period of 7 days. A total of 360 samples were sent for culture from each solution (120 samples on the 3rd, 5th, and 7th day,

respectively, from each antiseptic). Out of 2,160 samples, 240 (11.11 %) were positive over a period of 7 days for contamination with microbial organisms.

Out of 240 positive samples, the most common organism isolated was *Pseudomonas* present in 140 samples followed by *Escherichia coli* in 71 samples. *S. aureus* was present in seven, *Bacillus* in ten, and Coagulase -ve *Streptococcus* and *Klebsiella* in six samples each. There was no mixed infection present (Table 1).

The results indicated that on the 3rd day, the control group (normal saline) had the highest incidence of positive culture (20 %) followed by cetrimide/chlorhexidine 0.5 % (15 %) and cetrimide/chlorhexidine 1.0 % (11.6 %) while povidone iodine, glutaraldehyde, and chloroxylenol did not show any positive culture. On the 3rd day, povidone iodine, glutaraldehyde, and chloroxylenol were significantly better than cetrimide/chlorhexidine (0.5 and 1.0 %) and normal saline for the prevention of contamination ($P<0.0001$, Tables 2 and 3).

On the 5th day, normal saline (NS) produced 35.0 %, cetrimide/chlorhexidine (0.5 %) 18.33 %, cetrimide/chlorhexidine (1.0 %) 17.5 %, and povidone iodine 3.33 % positive cultures while glutaraldehyde and chloroxylenol did not show any positive culture. On the 5th day, povidone iodine, glutaraldehyde, and chloroxylenol were significantly better than NS and cetrimide/chlorhexidine in preventing contamination ($P<0.001$), and povidone iodine, glutaraldehyde and chloroxylenol were having an insignificant difference to each other ($P>0.05$, Tables 2 and 3).

On the 7th day, the positive cultures were present 40 % in NS, 22.5 % in cetrimide/chlorhexidine (0.5 %), 20.8 % in cetrimide/chlorhexidine (1 %), 5 % in povidone iodine, and 0.83 % in chloroxylenol. Glutaraldehyde did not show positive culture even on the 7th day. On the 7th day, glutaraldehyde and chloroxylenol were significantly better than normal saline and cetrimide/chlorhexidine ($P<0.0001$, Tables 2 and 3). Glutaraldehyde 2.4 % did not produce

Table 1 Organism isolated from positive antiseptic solutions including control

Organisms	NS 0.45 %			Savlon 0.5			Savlon 1 %			Betadine 5 %			Dettol 10 %			Cidex 2.4 %		
	Days			Days			Days			Days			Days			Days		
	3	5	7	3	5	7	3	5	7	3	5	7	3	5	7	3	5	7
<i>Pseudomonas</i> (140)	16	20	30	10	12	14	10	12	13	0	0	2	0	0	1	0	0	0
<i>E. coli</i> (71)	8	10	18	0	0	2	4	9	12	0	4	4	0	0	0	0	0	0
<i>Bacillus</i> (10)	0	0	0	2	4	4	0	0	0	0	0	0	0	0	0	0	0	0
<i>S. aureus</i> (7)	0	0	0	2	2	3	0	0	0	0	0	0	0	0	0	0	0	0
<i>Klebsiella</i> (6)	0	0	0	2	2	2	0	0	0	0	0	0	0	0	0	0	0	0
Coagulase -ve <i>Streptococcus</i> (6)	0	0	0	2	2	2	0	0	0	0	0	0	0	0	0	0	0	0
Total (240)	24	30	48	18	22	27	14	21	25	0	4	6	0	0	1	0	0	0

Table 2 Rate of contamination of different antiseptic solutions including control

Antiseptic	No. of sample	Samples					
		3rd day		5th day		7th day	
		Positive	Negative	Positive	Negative	Positive	Negative
Normal saline (0.45 %)	360	24 (20 %)	96	30 (25 %)	90	48 (40 %)	72
Cetrimide/chlorhexidine (0.5 %)	360	18 (15 %)	102	22 (18.33 %)	98	27 (22.5 %)	93
Cetrimide/chlorhexidine (1 %)	360	14 (11.6 %)	106	21 (17.5 %)	99	25 (20.8 %)	95
Povidone iodine (5 %)	360	0	120	4 (3.33 %)	116	6 (5.0 %)	114
Chloroxylenol	360	0	120	0	120	1 (0.83)	119
Glutaraldehyde (2.4 %)	360	0	120	0	120	0	120
Total	2,160	56 (2.59 %)	664	77 (3.56 %)	643	107 (4.95 %)	613

any positive culture even after the span of 7 days while chloroxylenol showed only (0.83 %) a positive bacterial culture; therefore, both the solutions were good in the prevention of bacterial contamination (Table 2).

Discussion

There are two important issues when using antiseptics and disinfectants for keeping the Cheattle's forceps in surgical

Table 3 Total number of positive samples with their statistical correlation

Sample	3rd day		5th day		7th day	
	Positive	Negative	Positive	Negative	Positive	Negative
S 0.5 %	16	104	22	98	33	87
S 1.0 %	4	116	6	114	13	107
B 5.0 %	0	120	7	113	10	110
C 2.4 %	0	120	4	116	4	116
D	0	120	6	114	8	112
NS	24	96	36	84	46	74
t-1/2	2.85	$P<0.05$	3.29	$P<0.001$	3.36	$P<0.001$
t-1/3	4.30	$P<0.0001$	3.03	$P<0.05$	4.00	$P<0.0001$
t-1/4	4.30	$P<0.0001$	3.85	$P<0.001$	5.50	$P<0.0001$
t-1/5	4.30	$P<0.0001$	3.29	$P<0.001$	4.46	$P<0.0001$
t-1/6	1.39	$P>0.05$	2.13	$P<0.05$	1.80	$P>0.05$
t-2/3	2.03	$P<0.05$	0.29	$P>0.05$	0.66	$P>0.05$
t-2/4	2.03	$P<0.05$	0.65	$P>0.05$	2.29	$P<0.05$
t-2/5	2.03	$P<0.05$	0.00	$P>0.05$	1.15	$P>0.05$
t-2/6	1.72	$P>0.05$	2.54	$P<0.05$	2.84	$P<0.05$
t-3/4	N/A	N/A	0.93	$P>0.05$	1.66	$P>0.05$
t-3/5	N/A	N/A	0.29	$P>0.05$	0.49	$P>0.05$
t-3/6	5.48	$P<0.0001$	5.14	$P<0.0001$	5.88	$P<0.0001$
t-4/5	N/A	N/A	0.65	$P>0.05$	1.19	$P>0.05$
t-4/6	5.48	$P<0.0001$	5.94	$P<0.0001$	7.40	$P<0.0001$
t-5/6	5.48	$P<0.0001$	5.40	$P<0.0001$	6.35	$P<0.0001$

S Savlon 0.5 %, S Savlon 1 %, B Betadine 5 %, C Cidex, D Dettol, NS normal saline

$P<0.05$, $P<0.001$, and $P<0.0001$ are considered statistically significant

$P>0.05$ is considered as non significant

wards. These are bacteriological contamination and loss of their anti-microbial activity over a period of time, thus necessitating its change to a fresh solution. There are many reports using bacterial culture to determine contamination of in-use (not from bottles keeping the Cheattle's forceps) antiseptics and disinfections [1–7]. These reported the following:

1. Contamination incidence varied from 1.8 to 63.1 % [1, 4, 6, 7].
2. The rates of contamination correlated with the duration of use [1, 8].
3. *Pseudomonas* was the commonest bacteria to be cultured [2, 3, 6, 7]. Its resistance to microbial agents was a concern [2].
4. Risk factors for contamination were improper preparation, storage, and prolonged use [1, 3, 7, 8].
5. Periodic quality testing and strongly adhering to standard disinfection procedures were advised to prevent this problem [3–7].

While it is expected that the continuous use of a solution to sterilize instruments for a long period would inevitably result in the dilution of the sterilizing solution; exact time frame for the loss of potency of a chemical is specifically mentioned only for glutaraldehyde. It is recommended that this solution should be discarded 14 days after activation because of the reported significant decrease in activity and free aldehyde concentration [9]. For povidone iodine, it has been mentioned that previously opened solutions may lose their anti-microbial effectiveness because of a partitioning of the iodine between the micelle structure of the surface active agent and the water phase [10, 11]. However, time frame, after which solution should be changed, is not specified.

Practicing and maintaining asepsis remains the Holy Grail for every hospital. It is well known that asepsis, like any other chain, is only as strong as its weakest link. Many diverse aspects of this chain have received scientific attention: contamination from opening glass ampoules [12], risk of multiple-use vials/bottles [8, 13], contamination of pagers/mobiles [14, 15], and even deliberate contamination of in-use antiseptic solutions to establish the minimum care necessary [16]. However, it is extremely surprising that a very important issue of contamination of various antiseptics and disinfections used for keeping the Cheattle's forceps has not received any scientific scrutiny, a classic example of missing the wood for the trees.

Our study's prevalence rate of contamination was 11.11 %, a finding similar to other studies which found 15.14 and 16.4 % of their samples contaminated with bacteria [4, 6]. As expected, positive rates of contamination increased with passage of time, a finding again consistent with other reports [1, 8]. Gram-negative bacteria, especially *Pseudomonas*,

followed by *E. coli* were the commonest to be cultured, as has been found in other studies [2, 3, 6, 7].

Our study addresses an important issue for “day-to-day” practice of medicine and patient health and safety. To summarize our findings, the growth of bacteria was found after 1 week, in 22.5 % of samples at 0.5 % concentration of cetrimide/chlorhexidine combination and in 20.8 % of samples at 1 % concentration of cetrimide/chlorhexidine combination, and it cannot be recommended as a safe solution for keeping Cheattle's forceps. Povidone iodine samples of 5 % were positive for the growth of bacteria after 1 week. After 1 week, bacterial culture was positive in 0.83 % (1/120 samples) and 0 % (0/120 samples) for chloroxylenol and 2.4 % for glutaraldehyde, respectively. Both chloroxylenol and 2.4 % glutaraldehyde were consistently shown to have better anti-microbial activity than cetrimide/chlorhexidine combinations and 5 % povidone iodine. We can safely conclude that the ideal solution for keeping Cheattle's forceps is 2.4 % glutaraldehyde followed by chloroxylenol.

Conflict of Interest None.

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