

Comparative efficacy evaluation of disinfectants routinely used in hospital practice: India

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Abstract

Aim: The aim of this study was to evaluate and compare practically achieved disinfection efficacy of some locally available disinfectants on surfaces and infectious microbiological hospital waste. **Materials and Methods:** Seven disinfectants were tested at concentrations recommended by manufacturers on rough and smooth surfaces that were contaminated experimentally by locally circulating isolates of methicillin-resistant *Staphylococcus aureus*, multidrug-resistant *Acinetobacter baumannii*, *Klebsiella pneumoniae*, *Enterobacter aerogenes*, *Pseudomonas aeruginosa* strains, standard isolate of *Salmonella typhi* and *Candida albicans*. Reduction in microbial counts before and after surface disinfection was expressed as log reduction. A very heavy microbial waste load was simulated by immersing culture plates with heavy microbial growth in disinfectants. Daily, a sample of disinfectant was taken and subjected to in-use test. **Results:** The highest average log reduction of test microbes on the rough surface was given by DesNet (5.05) and Bacillocid special (5.02). A comparable average log reduction of test microbes on a smooth steel surface was noted (5.68, 5.67, 5.50) for Lysol, Bacillocid sp. and DesNet, respectively. In the discard jars, Bacillocid special worked satisfactorily for 4 days, DesNet for 3 days and Hi-giene Germitol for 1 day. The remainder of the disinfectants failed in the in-use test on Day 1. Phenolics, although widely used in our settings, may not be as good surface disinfectants as newer formulations like DesNet and Bacillocid special. **Conclusions:** Newer quaternary ammonium compounds and aldehyde formulations were found to be the best disinfectants for disinfection of heavy contamination.

Keywords: Disinfectant, evaluation, hospital practice

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Introduction

Appropriate disinfection and sterilization procedures are a must for control of hospital-acquired infection, as failure can result in many hospital-acquired infections thus leading to increased cost, morbidity and mortality. Disinfection in hospital practice is mainly achieved either by surface disinfection (e.g., disinfection of surfaces of the tables, trolleys, instruments, walls and floors, etc.) or immersing the contaminated objects in the disinfectant solution. Disinfectants may also be used to chemically

treat infectious hospital waste, especially the disposable plastic and microbiological wastes. Different disinfectant formulations have different applications. The process of disinfection may be affected by many variables like temperature, contact period, pH and concentration of the disinfectant, bioburden, organic soil and hardness of water used for dilution. Therefore, the disinfectant ought to be tested in the field for the specified application to ensure its effectiveness. There is limited awareness among health care workers about choosing an appropriate disinfectant, especially in small health care settings. Usually, an agent with broad-spectrum antimicrobial activity is chosen based on the literature provided by manufacturers. Many hospitals are still using phenolic disinfectants, while their use is being discouraged throughout advanced countries. Toxicity issues have led to discontinued use of glutaraldehydes in some developed countries^[1] but, in developing countries,

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they are used very frequently. The standard tests to check disinfection efficiency include Rideal-Walker phenol coefficient (R.W.C) test,^[2] Chick-Martin and Garrod's test,^[3] Kelsey and Maurer's in-use tests and surface disinfection tests capacity use dilution test (Kelsey and Sykes, 1969),^[4] modified by Kelsey and Maurer, 1974,^[5] various other microbial time kill assays^[6] and standard carrier tests such as EN 13697,^[7] ASTM E2197,^[8] etc.

Because these standard tests cannot be performed by the laboratories belonging to small hospitals, one has to solely rely upon the literature provided by the manufacturer regarding the efficiency of the disinfectants. Almost all the manufacturers claim their disinfectant as a broad-spectrum antimicrobial agent suitable for diverse applications.

Keeping in view the above, the following study was planned with an aim to evaluate and compare the practically achieved disinfection efficacy of some locally available disinfectants for disinfection of surfaces and infectious microbiological and other hospital waste keeping their cost-effectiveness in mind. The efficacy was tested against locally isolated highly drug-resistant isolates of *Klebsiella pneumoniae*, *Enterobacter aerogenes*, *Acinetobacter calcoabmannii*, *Pseudomonas aeruginosa*, methicillin-resistant *Staphylococcus aureus* (MRSA), *Candida albicans* and standard strain of *Salmonella typhi*.

Materials and Methods

Seven disinfectants were purchased from the market and taken up for the study: phenol (carbolic acid 99.5%) manufactured by Qualigens Fine Chemicals, a division of Glaxo Smith Kline Pharmaceutical Ltd., Mumbai, India; phenyle (Rideal-Walker coefficient 5-7) manufactured by Bengal Chemicals and Pharmaceuticals Ltd. (a Government of India Enterprise); Lysol (50% cresol solution in soap) manufactured by Loba Chemie Pvt. Ltd., Mumbai, India); Hi-gene Germitol, manufactured by HiMedia Laboratories (P) Ltd., Mumbai, India (combination of Benzalkonium chloride 20% W/v, Cetrimide, isopropyl alcohol; 5% v/v, Emulsifier); Clea-N-Sept tablets (an effervescent disinfectant tablet containing the active ingredient sodium Dichloroisocyanurates 50%); DesNet (a combination of quaternary ammoniums didicyldimethylammonium chloride, sodium carbonate and surfactants) imported, processed and marketed in India by Quincon Enterprises, New Delhi, India, under the licence of ALPRO GMBH, St. Georgen/Schwaizwald, Germany; and Bacillocid special (combination of 1, 6 Dihydroxy-2,5-dioxyhexane 11.2%, Glutaraldehyde 5%, Benzalkonium chloride 5% and Alkyl Urea Derivative 3%) manufactured by Raman and Weil Pvt. Ltd., Daman,

India, in collaboration with Bode Chemie, Hamburg, Germany. All the disinfectants were used at the following working dilutions as per the manufacturers' instructions: phenol (5%), phenyl (3%), Lysol (3%), Hi-Giene Germitol (0.5%), Clean-N-Sept (one tablet per 2 L of water), DesNet (2%) and Bacillocid special (2%).

Seven microbial strains were included in this study: multiple drug-resistant (MDR) *Klebsiella pneumoniae* and *Enterobacter aerogenes* (resistant to amikacin, cefotaxime, ciprofloxacin, imipenem, piperacillin-tazobactam, cefoperazone-sulbactam), MDR *Acinetobacter anitratus* (resistance same as *Klebsiella pneumoniae* and additionally resistant to tigecycline and colistin), *Pseudomonas aeruginosa* (resistant to imipenem, ceftriaxone, amikacin, piperacillin-tazobactam, cefoperazone-sulbactam), *Salmonella typhi* NCTC 786, methicillin-resistant *Staphylococcus aureus* (MRSA) and *Candida albicans*. All the microbial strains used in this study were clinical isolates, except *Salmonella typhi* standard strain NCTC 786 obtained from Colindale London and being maintained in our laboratory.

Surface disinfection activity was tested by in-house standardized procedures, which is a modified quantitative surface disinfection test.^[7] Two types of surfaces were chosen: (i) rough surface (representative of floors, walls, etc.) and (ii) stainless steel surface, representing the instrument tables and trolleys, etc. made up of steel and other shining surfaces. All disinfectants were diluted to working concentration and microbial strains were suspended in normal saline to optical density equal to 0.5 McFarland opacity tube. Six ceramic plaster tiles (representing rough surface) and shiny stainless steel plates (representing smooth surface) of dimensions 10 cm × 10 cm were autoclaved for each disinfectant. One milliliter of microbial suspension of various organisms was evenly spread over each of the labeled surfaces with a micropipette tip and was allowed to dry for 1 h. To one labeled square, the test disinfectant was applied by a sterile cotton gauge soaked in 5 mL of disinfectant and the other was left without disinfectant. After a contact period of 10 min, both templates were swabbed and labeled as follows (nondisinfected area A and the disinfected B). Each swab was vortexed in a tube containing 10 mL of Bacto D/E neutralizing broth. The following serial dilutions were prepared: 1:10, 1:100 and 1:1000. Five drops of 100 mL of each of the dilutions were dropped on Mueller Hinton Agar, (Make: Difo Laboratories, BD, 1 Becton Drive, Franklin Lakes, NJ, USA, 07417, 201 847 6800, USA) from a height of 2.5 cm. The plates were incubated for 48 h for bacterial growth at 37°C and 7 days at 22°C. After incubation, the number of

the microorganisms were counted, and total counts were calculated by multiplying with the dilution factor. Each test was carried out in triplicate. All procedures were performed by a single person using the same technique.

For checking the ability to decontaminate infected hospital waste, 1 L of each disinfectant was prepared at the recommended working dilution and was poured in a glass container. Five culture media plates having heavy growth of different organisms (*Pseudomonas aeruginosa*, MRSA, *Salmonella typhi*, NCTC 786, *Acinetobacter anitratus* and *Candida albicans*) were discarded in each of the disinfectant-containing jar and immersed for 24 h. Each day, the in-use test was performed, old culture plates were taken out and then fresh culture plates were added. A period of 24 h was chosen to simulate the practical condition in which the microbiological discard is being treated chemically. The process was continued till the time disinfectants failed the in-use test. Briefly, the in-use test was performed by diluting 0.5 mL of the test sample into 4.5 mL nutrient broth (Difco Laboratories) in a sterile test tube. Ten drops (20 μ L volume) of this mixture were placed on 10 different areas marked on each of the two well-dried nutrient agar plates. One plate was incubated for 3 days at 37°C and the other for 7 days at room temperature. The test was considered failed if there was growth in five or more than five inoculated areas on either plate.

For statistical analysis, an average of six observations (three at 37° and 22°C each) was taken for calculating log reduction achieved by the disinfectant for each organism. Log reduction was calculated by using the following formula:

$$\text{Log}_{10} \text{ Reduction Factor (RF)} = \text{Log}_{10} \text{ Prevalue} - \text{Log}_{10} \text{ Postvalue}$$

Our aim was to find out the statistically significant difference in overall efficacy of the disinfectants. Box plots were drawn [Figures 1 and 2] for determining the overall efficacy of a disinfectant using R-Gui software version 2.11.1 to determine whether there was any statistically significant difference. The independent t test was applied for all possible comparisons using SPSS software version 16. Only statistically significant difference comparisons have been shown in Tables 1 and 2.

Results

Maximum average log reduction of test microbes on rough surface was achieved by DesNet (5.05) and Bacillocid special (5.02). Clea-N-Sept and lysol have

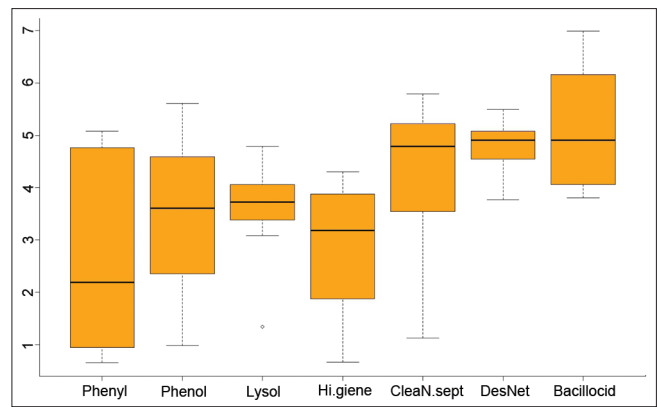


Figure 1: Box plot showing overall disinfection efficacy of various disinfectants on rough surface. Y-axis shows log reduction values

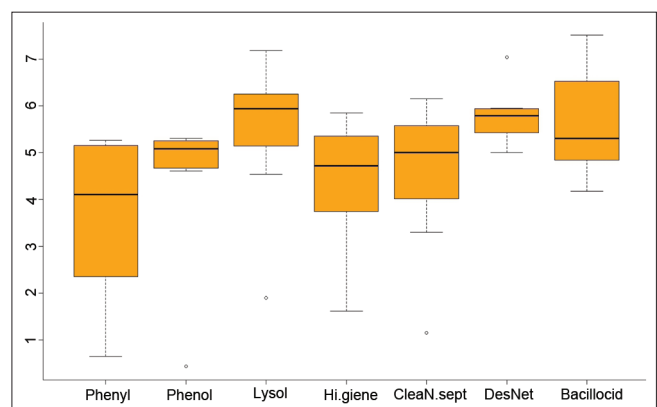


Figure 2: Box plot showing overall disinfection efficacy of various disinfectants on smooth surface. Y-axis shows log reduction values

almost equal surface disinfection activity on the rough surface, and showed an average log reduction of 4.17 and 4.10, respectively. Comparable log reduction values (3.33, 3.29 and 3.09) were achieved by phenyl, phenol and Hi-giene, respectively. [Table 3 and Figure 2]. The log reduction against *Salmonella typhoon* NCTC 786 was shown to be maximum by Bacillocid special and DesNet (log reduction 6.99 and 6.92, respectively). DesNet showed a statistically significant more activity as compared with phenyl and Hi-gene (*P*-value 0.013 and 0.015, respectively). Bacillocid was significantly more active as compared with phenyl (*P*-values 0.004, 0.032, 0.042 and 0.004, respectively) [Table 1]. Bacillocid special and DesNet could achieve a 100% kill for all microorganisms, whereas all other disinfectants showed a variable kill of different microorganisms except *Candida albicans*, on which each disinfectant achieved a 100% kill.

Surface disinfection on smooth steel surface showed that average log reduction of test microbes was found to be comparable (5.68, 5.76 and 5.50) for lysol, Bacillocid special and DesNet, respectively. Clean-N-Sept, phenol, Hi-giene and phenyl showed an average log reduction

Table 1: Comparative efficacy of different disinfectants on rough surface based on log reduction

		Mean difference	Std. error	P value τ	95% confidence interval	
					Lower bound	Upper bound
DesNet	Phenyle	2.0128571	0.7771677	0.013*	0.444469	3.581245
	Hi-giene	1.9700000	0.7771677	0.015*	0.401612	3.538388
Bacillocid	Phenyle	2.4014286	0.7771677	0.004*	0.833041	3.969817
	Phenol	1.7228571	0.7771677	0.032*	0.154469	3.291245
	Lysol	1.6300000	0.7771677	0.042*	0.061612	3.198388
	Hi-giene	2.3585714	0.7771677	0.004*	0.790183	3.926959

τ independent t-test; *Significant at P value < 0.05

Table 2: Comparative efficacy of different disinfectants on smooth surface based on log reduction

		Mean diff.	Std. error	P value τ	95% confidence interval	
					Lower bound	Upper bound
Lysol	Phenyl	1.8257143	0.8255783	0.033*	0.159630	3.491799
DesNet	Phenyl	2.2185714	0.8255783	0.010*	0.552487	3.884656
Bacillocid	Phenyl	2.1000000	0.8255783	0.015*	0.433915	3.766085

τ independent t-test; *Significant at P value < 0.05

Table 3: Results of surface disinfection activity by different disinfectants on rough surfaces contaminated with different organisms

Name of disinfectant (average log reduction)		<i>Pseudomonas aeruginosa</i>	<i>Acinetobacter anitratus</i>	MRSA	<i>Salmonella typhi</i>	<i>Klebsiella pneumoniae</i>	<i>Enterobacter aergens</i>	<i>Candida albicans</i>
Phenyle (3.33)	A	6.3×10^3	1.4×10^5	1.2×10^5	7.2×10^4	4.5×10^3	5.4×10^4	6.3×10^4
	B	1×10^3	9.0×10^2	Nil	5.8×10^3	1×10^3	Nil	Nil
	C	2.79	2.19	5.07	1.09	2.65	4.73	4.79
	D	84.12%	99.35%	100%	91.94%	77.77	100%	100%
Phenol (3.29)	A	3.7×10^4	4.1×10^5	1.5×10^5	7.6×10^4	4.1×10^4	8.2×10^4	1×10^4
	B	2×10^2	Nil	Nil	8×10^3	1×10^1	3×10^2	Nil
	C	2.26	4.61	5.17	1.97	2.61	2.43	4.00
	D	99.45%	100%	100%	89.45%	99.97%	99.63%	100%
Lysol (4.10)	A	5.2×10^3	4.9×10^3	1.6×10^4	1.3×10^7	1.2×10^3	8.3×10^3	6.2×10^4
	B	Nil	Nil	Nil	6×10^5	Nil	Nil	Nil
	C	>3.71	3.69	6.20	1.33	5.07	3.91	4.79
	D	100%	100%	100%	95.38%	100%	100%	100%
Hi-giene (3.09)	A	9.2×10^3	2.7×10^3	3×10^5	2×10^5	6.8×10^4	1.3×10^6	8.4×10^3
	B	2×10^3	1×10^2	2×10^2	1×10^1	1×10^1	6.2×10^3	Nil
	C	2.66	3.43	3.17	3.30	2.83	2.32	3.92
	D	78.26%	96.29%	99.93%	99.99%	99.98%	99.52%	100%
Clea-N-Sept (4.17)	A	7.7×10^4	6.1×10^4	6.1×10^5	5.7×10^5	4.7×10^4	3.6×10^5	2.6×10^4
	B	Nil	Nil	Nil	4.3×10^4	1×10^2	Nil	Nil
	C	>4.88	4.78	5.78	1.12	2.67	5.55	4.41
	D	100%	100%	100%	92.45%	99.78%	100%	100%
DesNet (5.05)	A	3.1×10^5	5.9×10^3	2.1×10^4	8.5×10^4	8.2×10^4	1.7×10^5	6×10^4
	B	Nil	Nil	Nil	Nil	Nil	Nil	Nil
	C	>5.49	3.77	4.32	6.92	4.91	5.23	4.77
	D	100%	100%	100%	100%	100%	100%	100%
Bacillocid (5.02)	A	6.5×10^3	1.7×10^4	7.7×10^3	9.8×10^6	1×10^6	2.1×10^6	8.2×10^4
	B	Nil	Nil	Nil	Nil	Nil	Nil	Nil
	C	>3.81	4.23	3.88	6.99	5.00	6.32	4.91
	D	100%	100%	100%	100%	100%	100%	100%

A: Number of colony forming units (cfu) present on the surface before disinfection; B: Number of cfu present on the surface after disinfection; C: Log reduction in cfu after disinfection; D: Percent kill after disinfection

of 4.77, 4.69, 4.49 and 3.89, respectively. The maximum log reduction against *Salmonella typhi* NCTC 786 was shown by Bacillocid special and DesNet (log reduction 7.07 and 7.04, respectively) [Table 4]. Table 2 shows the significant comparative differences. Lysol, DesNet and

Bacillocid showed significant differences (P-values 0.033, 0.01 and 0.015, respectively) as compared with phenyl by the independent t test [Table 2]. All disinfectants could achieve a 100% kill on MRSA and *Candida albicans*. *Salmonella typhi* appeared to be the most difficult target

Table 4: Results of surface disinfection activity by different disinfectants, on smooth (Steel) surface contaminated with different organisms

Name of disinfectant (average log reduction)		<i>Pseudomonas aeruginosa</i>	<i>Acinetobacter anitratus</i>	MRSA	<i>Salmonella typhi</i>	<i>Klebsiella pneumoniae</i>	<i>Enterobacter aerogens</i>	<i>Candida albicans</i>
Phenyl (3.89)	A	1.3×10^4	1×10^5	1.8×10^5	1.2×10^7	4.5×10^3	1.2×10^5	1.7×10^5
	B	Nil	3×10^2	Nil	7.8×10^4	1×10^3	Nil	Nil
	C	4.11	2.75	5.25	2.18	2.65	5.07	5.23
	D	100%	99.7%	100%	99.35%	77.77%	100%	100%
Phenol (4.69)	A	2×10^5	1.2×10^5	1.6×10^5	6.4×10^3	4.1×10^4	2×10^5	5.2×10^4
	B	Nil	Nil	Nil	2.4×10^3	Nil	Nil	Nil
	C	5.30	5.07	5.20	2.42	4.85	5.30	4.71
	D	100%	100%	100%	62.5%	100%	100%	100%
Lysol (5.68)	A	9.6×10^5	3.4×10^6	3.5×10^4	1.5×10^7	8.8×10^5	1.5×10^7	5.6×10^5
	B	Nil	Nil	Nil	1.9×10^5	Nil	Nil	Nil
	C	5.98	6.53	6.54	1.89	5.94	7.17	5.74
	D	100%	100%	100%	98.73%	100%	100%	100%
Hi-giene (4.49)	A	1.1×10^6	5.3×10^4	2.8×10^5	1.8×10^5	3×10^6	1.8×10^7	7×10^5
	B	1.1×10^3	Nil	Nil	Nil	1×10^2	4.4×10^5	Nil
	C	3.0	4.72	5.44	5.25	4.47	3.61	5.00
	D	99%	100%	100%	100%	99.99%	97.55%	100%
Clea-N-Sept (4.77)	A	2×10^3	2.4×10^5	1.4×10^6	7.2×10^5	1.0×10^5	5.9×10^5	5.2×10^4
	B	Nil	Nil	Nil	5.1×10^4	Nil	Nil	Nil
	C	5.30	5.38	6.14	1.14	5.00	5.77	4.71
	D	100%	100%	100%	92.91%	100%	100%	100%
DesNet (5.50)	A	3.6×10^5	8.5×10^5	8.9×10^5	1.1×10^7	1.0×10^5	2×10^5	6.2×10^5
	B	Nil	Nil	Nil	Nil	Nil	Nil	Nil
	C	5.55	3.92	5.94	7.04	5.00	5.30	5.79
	D	100%	100%	100%	100%	100%	100%	100%
Bacillocid (5.76)	A	1.5×10^4	4.1×10^4	2×10^5	1.2×10^7	1.2×10^5	3.2×10^7	9.5×10^5
	B	Nil	Nil	Nil	Nil	Nil	Nil	Nil
	C	4.17	4.61	5.30	7.7	5.07	7.50	5.97
	D	100%	100%	100%	100%	100%	100%	100%

A: Number of colony forming units (cfu) present on the surface before disinfection; B: Number of cfu present on the surface after disinfection; C: Log reduction in cfu after disinfection; D: Percent kill after disinfection

organism to be killed. Only Bacillocid special, DesNet and Hi-giene could achieve a 100% kill of this organism.

For ability to decontaminate infected hospital waste by the in-use test, Bacillocid special was found to be active till 4 days. DesNet worked satisfactorily for 3 days. Hi-giene worked satisfactory for only 1 day. All the three phenolic disinfectants and Clean-N-Sept could not tolerate the microbial load even for 1 day.

Discussion

This study evaluated and compared the practically achieved disinfection efficacy of some locally available disinfectants on surfaces and very heavy microbial waste load. In routine hospital practice, disinfection of surfaces like floors and walls may not be required except where they have been contaminated by infectious materials or agents. Thorough cleaning of these surfaces may be sufficient in noncritical areas, although some studies advocate the use of a disinfectant in floor cleaning.^[9-11] Although cleaning may remove a large number of bacteria, the microorganisms that have been left behind soon begin to grow and accumulate, and may

cross-contaminate the clean areas. It is more important to disinfect the "near patient" hand touch areas that are most implicated in transmission via the contaminated hands of the health care workers.^[9,11] Therefore, use of disinfectants in critical and high-risk areas like burn units and Intensive Care Units (ICUs) is justified,^[9-13] where the environment may be heavily contaminated with drug-resistant pathogens like MRSA, *Klebsiella pneumoniae*, *Acinetobacter* species and *Pseudomonas aeruginosa*.^[14] On many occasions, while investigating outbreaks caused by MRSA, we have noted gross contamination of the articles and the surfaces like medicine trolleys, the patients' cabinets, railing of the beds, the nurses' lockers, electric switches and door handles, etc. (unpublished data). In various ICUs and emergency wards, 27.3% of the environmental surfaces showed contamination with *Staphylococcus aureus*, and 30% of these were MRSA.^[14] Proper disinfection of the surfaces is also important in operation theaters^[15] and other areas to disinfect blood spillages and other grossly infected surfaces. Many studies have emphasized that routine cleaning, hand washing and barrier nursing alone were not sufficient to control protracted outbreaks of MRSA, but required proper disinfection of the environment.^[16,17]

A wide range of disinfectants are available commercially that undergo extensive testing in controlled environments before market release. However, often, the products and procedures as described in the literature may not be able to adequately disinfect or decontaminate items when the surfaces have been contaminated with highly resistant or unusual organisms, or if the bioload of microorganisms is very heavy. The matter may be further complicated by the quality of environmental hygiene because of dust and other organic matter. When choosing a disinfectant for specific hospital use, it may be necessary to know the expected number and the types of organisms likely to be present on the surface. It is critical that the disinfectant be selected based on its ability to be effective against the prevalent pathogenic microorganisms that can be transmitted by direct or indirect contact with the environment.^[18] Therefore, we chose to test the disinfectants on multidrug-resistant isolates that are circulating in our hospital environment. The type of surface to be disinfected and applications for the product must always be considered. An ideal disinfectant should have a broad antimicrobial spectrum, should be nonirritating, less toxic, noncorrosive and inexpensive.^[9]

As expected, this study showed that all disinfectants were highly effective in killing microbes on the smooth steel surface as compared with the rough surface. The decrease in efficacy of the test disinfectants on rough surface may be attributed to presence of organic matter, protection provided to the organisms and increased adherence.^[9] On both surfaces, different disinfectants showed a variable effect on different microorganisms. Only Bacillocid special and DesNet could achieve a 100% kill for all microorganisms. All others showed a variable kill of different microorganisms except *Candida albicans*, on which each disinfectant achieved a 100% kill. Similarly, on smooth surface, all disinfectants could achieve a 100% kill on MRSA. *Salmonella typhi* appeared to be the most difficult target organism to be killed; only Bacillocid special and DesNet could achieve a 100% kill of this organism on both surfaces. Hi-gene could also achieve a 100% kill on smooth surfaces. Phenolics and Clea-N-Sept were least effective against *Salmonella typhi* NCTC 786, which is a standard stain used in the R.W. phenol coefficient test.^[2] The statistical analysis also supported the overall best efficacy achieved by DesNet and Bacillocid special on both surfaces. Clea-N-Sept could achieve a comparable efficacy on smooth surface.

Although phenolic agents exhibit high toxicity and low biodegradability, they are still in use in developing countries because of their low cost. They are considered

a health risk by the EPA and NIOSH,^[19] and cannot be used in neonatal, pediatric ICU or on any infant contact surface. Eye irritation, contact dermatitis/urticaria and depigmentation of the skin have been linked to phenol residue contact.^[12,19] In this study, phenolics showed poor activity on rough surfaces that represent cracks and grooves on the floors and walls, very commonly seen in developing country health care settings. Therefore, better and safer disinfectants are required to replace them.

Chlorine, although being rapidly active on bacteria, viruses and most fungi,^[12,20] has some disadvantages,^[20] which include effect of pH on stability, reduced efficacy in the presence of organic matter, unpleasant smell, irritation to skin, eyes and mucous membranes and corrosiveness to metals. Various chlorine preparations are commercially available, including sodium and calcium hypochlorite, sodium dichloroisocyanurate, etc. In the present study, the antimicrobial activity of Clean-N-Sept was in the acceptable range. It could achieve a 100% kill on five and six of the seven target organisms on rough and smooth surfaces, respectively. However, its use on metallic surfaces cannot be recommended.

Aldehyde-containing disinfectant, "Bacillocid special" was found to be a very effective antimicrobial agent on almost all the tested organisms on both types of surfaces. Although widely used for the chemical sterilization of heat-sensitive equipments, there are safety concerns as aldehydes cause irritation to skin, eyes and air ways, allergic asthma and contact exema.^[21,22] Therefore, their use is now discouraged throughout the world. A popular brand of gluteraldehyde has been withdrawn from sale in the UK^[1] and replaced with ortho-phthalaldehyde.

Two quaternary ammonium compound (QAC)-containing products, DesNet and Hi-gene Germitol, were tested in this study. Newer QACs are active against a wide range of microorganisms including yeasts and moulds, are odorless, colorless, noncorrosive and highly stable compounds over a wide range of pH (3–10.5) and temperature. They are relatively stable in the presence of organic matter, have bacteriostatic residual effect on treated surfaces and cause low irritation and low toxicity.^[12,19] Our study has shown high antimicrobial activity of one of the third-generation QAC formulations (DesNet), but lesser activity was shown by the other disinfectant (Hi-giene) containing benzalkonium chloride.

In this study, we also wanted to test the disinfectants against heavy microbial waste load, which was simulated by immersing culture plates in disinfectants

and monitored by in-use test. The microbiological waste is best treated by autoclaving.^[23] However, it is a common practice for microbiologists/technicians working on the bench to discard many infectious items into jars containing disinfectants. Chemical disinfection is also required for the disinfection of sharps, which are dipped in disinfectant solutions. Under such conditions, disinfection is required in the presence of organic matter, a variable pH and temperature conditions, and should be free of toxic and irritant fumes. The best disinfection activity was shown by Bacillocid special, followed by DesNet and Hi-giene. Clea-N-Sept and phenolics could not tolerate the tested bacterial load even for 1 day.

There are some limitations of this study as we did not test for antimycobacterial and antiviral effect of any of the test disinfectants. It can be concluded from the present study that newer QACs with surfactants and aldehyde formulations can be used as general purpose disinfectants. Both the disinfectants are noncorrosive to rubber and metals. However, for safety reasons, QAC formulations may be preferred over potentially toxic aldehydes.

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