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EFFECTIVITY OF ANTISEPTICS AGAINST SOME PATHOGENS

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ABSTRACT

INTRODUCTION. The efficacy of antiseptics against bacteria and fungi is different. The choice of optimal antiseptic solution is very important in prophylaxis of hospital infections.
MATERIAL AND METHODS. In this study the efficacy of different antiseptics against some pathogens (*Klebsiella pneumoniae* ESBL (+), *Acinetobacter baumannii*, *Pseudomonas aeruginosa*, *Staphylococcus aureus* MRSA, *Candida dublinensis*) was analyzed. The disc diffusion, similar to the method used in antibiotic sensitivity testing was applied. We assumed that the size of inhibition zone of bacterial growth corresponds with the efficacy of antiseptic.
RESULTS AND CONCLUSION. The 2% alcoholic solution of chlorhexidine was the most effective antiseptic in our study.

Key words: *antiseptics, bacteria, fungi*

INTRODUCTION

More than 150 years ago Ignaz Philipp Semmelweis have proven the efficacy of antiseptics against puerperal infections. From that time many chemical compounds with disinfectant properties was synthetized. Those substances are used in skin, objects and surfaces decontamination and disinfection. For all those antiseptics their bacteriostatic and bactericidal properties was confirmed. However, clinical observations concerning their properties and efficacy are not clear (1-4).

The objective of the study was to analyze the efficacy of several antiseptics against some pathogens. The choice of bacterial and fungal species in our study corresponded with the high incidence of hospital infections, including infections of vascular catheters and bacteremia, caused by these pathogens. We assumed that the proper choice of optimal antiseptic might decrease the incidence of infections in hospitalized patients.

MATERIAL AND METHODS

In the study species of *Pseudomonas aeruginosa, Acinetobacter baumannii, Klebsiella pneumoniae* ESBL (+), *Staphylococcus aureus* MRSA, and *Candida dublinensis* isolated from biological materials collected from patients hospitalized in Military Institute of Medicine in Warsaw, Poland, in September 2014 were used.

The following antiseptics were analyzed: (1) Braunol (7,5% Providonum iodinatum; B/Braun, Melsungen, *Germany)* – sample A, (2) Braunoderm (50% Alcohol isopropylicus + 1% Povidonum iodinatum; B/Braun, Melsungen, *Germany)* – sample B, and (3) 0,5% solution of chlorhexidine in 70% ethanol) – sample C, (4) 2% solution of chlohexidine in 70% ethanol) – sample D, (5) 4% solution of chlorhexidine in water) – sample E, and (6) Caphosol (supersaturated solution of calcium and phosphates ions; EUSA Pharma Oxford, UK) – sample F. Samples C, D and E were prepared by hospital pharmacy of Military Institute of Medicine in Warsaw.

At the beginning of the study a suspension of analyzed pathogens with the density of McFarland Standard No. 0.5 was prepared. Each suspension was distributed on the Mueller-Hinton agar (bioMérieux, France). Then the blotting-paper discs with diameter of 5 mm were placed on the microbial growth media. After that, on every disc a 10 µL of analyzed antiseptics was transferred. Then the growth media were incubated for 24 hour in 37°C. After the incubation period the sizes of growth inhibition zones were noted (Fig. 1-5).

The analyses were repeated three times.

RESULTS

We assumed that the biggest diameter of growth inhibition zone around the disc with antiseptic corresponds with the biggest antibacterial / antifungal efficacy of the antiseptic. The results of analyzes were presented in Table I. The percentage comparison of growth inhibition zones was shown in Table II. The sizes of all zones were compared to the smallest one, which was get as 100%. The size of Caphosol (sample F) inhibition zone was excluded from the comparison because of its antiseptic properties concerning mouth mucosa only

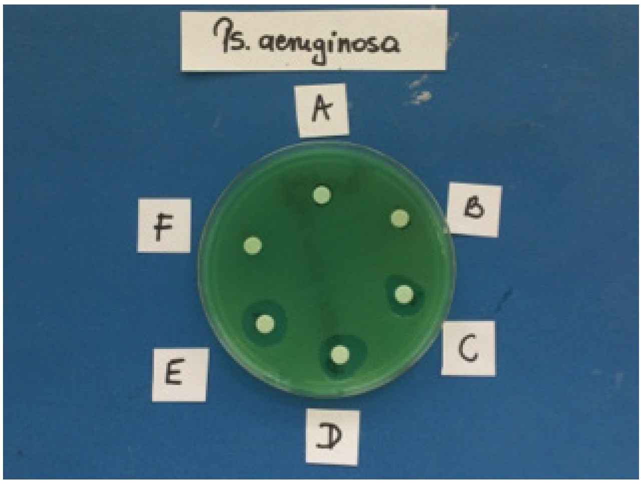

Fig.3. The inhibition zones of growth of *Pseudomonas aeruginosa*

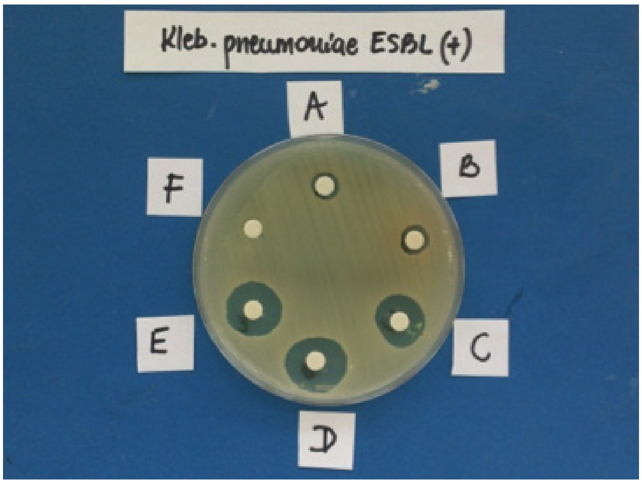

Fig.1. The inhibition zones of growth of *Klebsiella pneumoniae* ESBL (+)

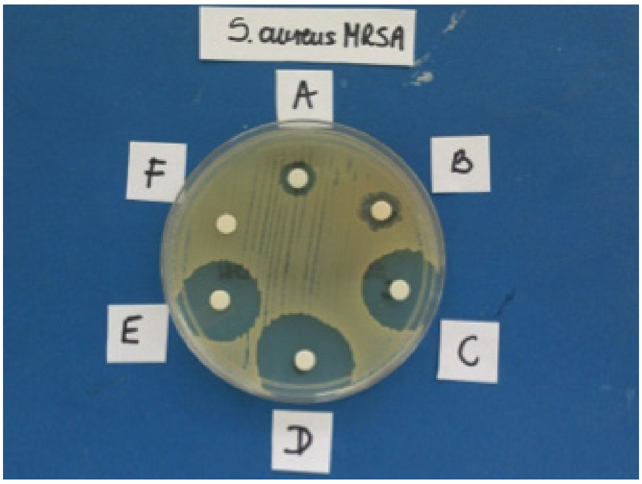

Fig.4. The inhibition zones of growth of *Staphylococcus aureus MRSA*

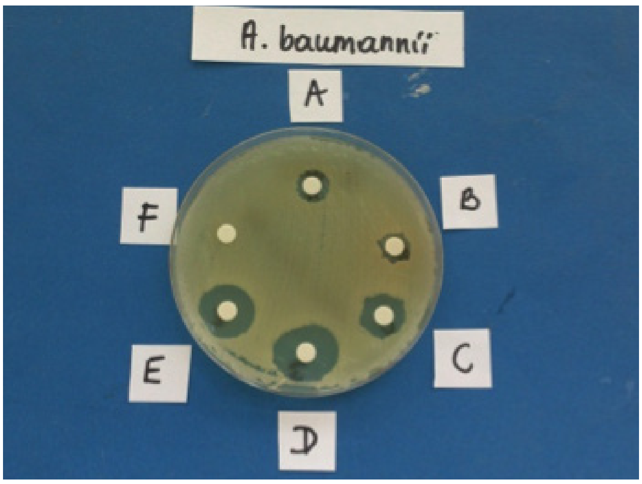

Fig.2. The inhibition zones of growth of *Acinetobacter baumannii*

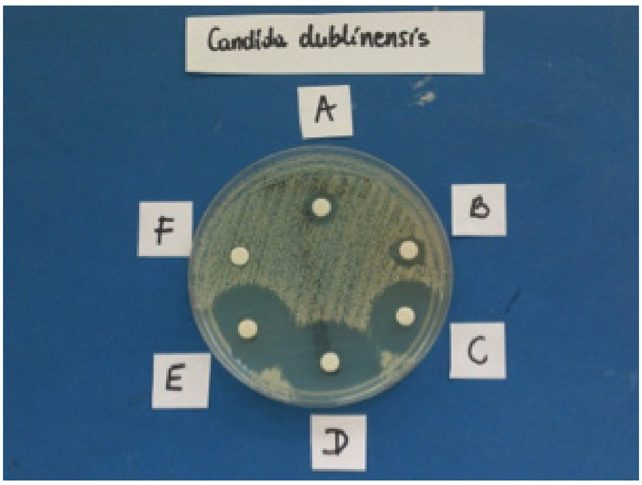

Fig.5. The inhibition zones of growth of *Candida dublinensis*

Table I. The growth inhibition zones of pathogens for analyzed antiseptics

pathogen	sample					
	A	B	C	D	E	F
K. pneumoniae ESBL +	8 ± 1 mm	9 ± 0 mm	16 ± 0 mm	18 ± 1 mm	17,3 ± 1,5 mm	6 ± 0 mm
A. baumannii	10 ± 0 mm	9 ± 0 mm	13 ± 1 mm	19 ± 1 mm	17 ± 0 mm	6 ± 0 mm
Ps. aeruginosa	8 ± 1 mm	7 ± 0 mm	14 ± 0 mm	15 ± 0 mm	15 ± 0 mm	6 ± 0 mm
S. aureus MRSA	10 ± 0 mm	10 ± 0 mm	24 ± 0 mm	29 ± 1 mm	24 ± 1 mm	6 ± 0 mm
Candida dublinensis	11 ± 0 mm	11 ± 1 mm	25 ± 0 mm	31 ± 1 mm	28 ± 1 mm	6 ± 0 mm

Sample A: Braunol (7.5% Providonum iodinatum; B/Braun, Melsungen, *Germany)*. Sample B: Braunoderm (50% Alcohol isopropylicus + 1% Povidonum iodinatum; B/Braun, Melsungen, *Germany)*. Sample C: 0.5% solution of chlorhexidine in 70% ethanol. Sample D: 2% solution of chlohexidine in 70% ethanol. Sample E: 4% solution of chlorhexidine in water. Sample F: Caphosol (supersaturated solution of calcium and phosphates ions; EUSA Pharma Oxford, UK).
Data were presented as mean ± standard deviation.

Table II. The comparison of growth inhibition zones of pathogens for analyzed antiseptics, showed as a percentage.

pathogen	sample				
	A	B	C	D	E
K. pneumoniae ESBL +	100%	112.5%	200%	225%	216.25%
A. baumannii	111.11%	100%	144.44%	211.11%	188.89%
Ps. aeruginosa	114.29%	100%	200%	241.29%	214.29%
S. aureus MRSA	100%	100%	240%	290%	240%
Candida dublinensis	100%	100%	227.27%	281.82%	254.55%

Sample A: Braunol (7.5% Providonum iodinatum; B/Braun, Melsungen, *Germany)*. Sample B: Braunoderm (50% Alcohol isopropylicus + 1% Povidonum iodinatum; B/Braun, Melsungen, *Germany)*. Sample C: 0.5% solution of chlorhexidine in 70% ethanol. Sample D: 2% solution of chlohexidine in 70% ethanol. Sample E: 4% solution of chlorhexidine in water. Sample F: Caphosol (supersaturated solution of calcium and phosphates ions; EUSA Pharma Oxford, UK).
Data were presented in comparison to the size of the smallest diameter of growth inhibition zone, showed as 100%.

In our study the 2% solution of chlorhexidine in 70% ethanol was the most effective antiseptic.

DISCUSSION

The antiseptics play an important role in decreasing the number of bacterial species colonizing human skin. The vascular catheterization is the most common procedure in critical care units with skin decontamination. The usefulness and effectiveness of antiseptics in the decreasing of incidence of catheter infections with bacteremia was analyzed in the literature (1-6).

In clinical practice, skin is decontaminated with 60% - 90% solutions of ethanol or isopropanol, or mixture of both. Alcohol is a potent bactericidal agent causing denaturation (7). Iodine with synthetic polymer (povidone-iodine) has anti-bacterial activity and is less toxic than tincture of iodine. Povidone-iodine is a broad-spectrum antiseptic that destroys microbial protein and DNA.

Very popular antiseptic is chlorhexidine. It is well tolerated by the skin and easily absorbed by the epidermis. Due to it chlorhexidine protects against bacterial growth, especially Gram-positive species, including *Staphylococcus epidermidis*, frequent etiologic factor of catheter-related infections. Chlorhexidine is considered more effective antiseptic in comparison to povidone-iodine (8,9). In our study both aqueous and alcohol solutions of chlorhexidine were more effective than povidone-iodine. It is confirmed by the literature: the lower incidence of catheter-related infections were observed when skin was decontaminated with chlorhexidine solutions in comparison to povidone-iodine (1,2,10).

Parienti et al. (5) found the lower incidence of vascular catheter colonization when skin was decontaminated with alcohol solution of powidone-iodine in comparison to aqueous one. In our study we did not observed differences between both forms of povidone-iodine. We found only small disparities in sizes of growth inhibition zones caused by 4% solution of chlorhexidine in water (E sample) in comparison to 0.5% (sample C) and 2% (sample D) solutions of chlorhexidine in 70% ethanol. It corresponds with observations of other authors (1) that efficacy of aqueous and an alcohol solution of chlorhexidine is similar.

The growth inhibition zones of 2% solution of chlorhexidine in 70% ethanol were larger in comparison to 0.5% solution of chlorhexidine in 70% ethanol. It confirms some guidelines (11) suggesting that skin should be decontaminated with alcohol solutions of chlorhexidine with concentration above 0.5%.

We found no antibacterial properties of Caphosol, supersaturated solution of calcium and phosphate ions. Only its anti-inflammatory and disinfectant properties concerning mouth mucosa can be useful.

The methodology of our study was similar to those used in laboratory testing for sensitivity of an isolated bacterial strain to different antibiotics. However, in current literature we did not found a reference model.

CONCLUSION

The solutions of chlorhexidine were more potent against some pathogens in comparison to povidone-iodine. One should consider to use 2% solution of chlorhexidine in ethanol rather than 0.5% one in skin decontamination.

REFERENCES

1. Maki DG, Ringer M, Alvarado CJ. Prospective randomised trial of povidone-iodine, alcohol, and chlorhexidine for prevention of infection associated with central venous and arterial catheters. Lancet 1991; 338 (8763): 339-343.
2. Mimoz O, Pieroni L, Lawrence C, et al. Prospective, randomized trial of two antiseptic solutions for prevention of central venous or arterial catheter colonization and infection in intensive care unit patients. Crit Care Med 1996; 24(11): 1818-1823
3. Calfee DP, Farr BM. Comparison of four antiseptic preparations for skin in the prevention of contamination of percutaneously drawn blood cultures: a randomized trial. J Clin Microbiol 2002; 40(5): 1660-1665.
4. Seguin P, Laviolle B, Dahyot-Fizelier C, et al. Effect of oropharyngeal povidone-iodine preventive oral care on ventilator-associated pneumonia in severely brain-injured or cerebral hemorrhage patients: a multicenter, randomized controlled trial. Crit Care Med 2014; 42(1): 1-8.
5. Parienti JJ, du Cheyron D, Remakers M, et al. Alcoholic povidone-iodine to prevent central venous catheter colonization: a randomized unit-crossover study. Crit Care Med 2004; 32(3): 708–713.
6. Langgartner J, Linde HJ, Lehn N, et al. Combined skin disinfection with chlorhexidine/propanol and aqueous povidone-iodine reduces bacterial colonisation of central venous catheters. Intensive Care Med 2004; 30(6): 1081-1088.
7. Sykes G. The influence of germicides on the dehydrogenases of Bact. coli: I. The succinic acid dehydrogenase of Bact. coli. J Hyg (Lond) 1939; 39(4): 463–469.
8. Goldblum SE, Ulrich JA, Goldman RS, et al. Comparison of 4% chlorhexidine gluconate in a detergent base (Hibiclens) and povidone-iodine (Betadine) for the skin preparation of hemodialysis patients and personnel. Am J Kidney Dis 1983; 2(5): 548-552.
9. May SR, Roberts DP, DeClement FA, et al. Reduced bacteria on transplantable allograft skin after preparation with chlorhexidine gluconate, povidone-iodine, and isopropanol. J Burn Care Rehabil 1991; 12(3): 224-228.
10. Garland JS, Buck RK, Maloney P, et al. Comparison of 10% povidone-iodine and 0.5% chlorhexidine gluconate for the prevention of peripheral intravenous catheter colonization in neonates: a prospective trial. Pediatr Infect Dis J 1995; 14(6): 510-516.
11. O'Grady NP, Alexander M, Burns LA, et al. Guidelines for the prevention of intravascular catheter-related infections. Am J Infect Control 2011; 39(Suppl 1): S1-34.

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