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In Vitro Effectiveness Of Acid And Aldehyde Base Surgical Instruments Disinfectansts Against Various Microorganismis

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Abstract

In this study, in order to define how effective aldehyde based surgical instrument disinfectant with acid based is to which concentration against Staphilococcus aureus (ATCC 29213), Escherichia coli (ATCC 25922), Pseudomonas aeruginosa (ATCC 23853), Candida albicans (ATCC 10231) strains used for Test microorganisms, final microorganisms was disinfectant into the different concentrations of the disinfectant to be tested by being prepared as 2-5x109 CFU/ml according to the McFarland 5 cloudiness of Test disinfectant. After disinfectants were activated with microorganisms at previously experimented 30 seconds 1,5,15 and 30 minute periods, colony counts at 1 ml. Levels were performed by way of cast-cultural plaque metod. Consequently, it was determined that 0.01% concentration of aldehid based surgical instrument disinfected was effective against test microorganisms in 30 seconds, 0.1% concentrations of the disinfectant containing peracetic acid was effective against Staphilococcus aureus in 30 seconds, while other test microorganisms were effective in 30 seconds at 0.5% concentrations. It was also determined that 0.1% concentrations of the disinfectant containing citric acid were effective against all test microorganisms in 30 seconds.

Key words: Acid, Aldehyde, Disinfectant, Microorganism

INTRODUCTION

The density of declarations belonging to infections which develop after inadequate/inconvenient decontamination operationsin routine invasive and non invasive disinfection applications such as medical instruments and equipments make necessary the control of disinfection procedures belonging to selection and usage or disinfectants in medical establishments [1-2].

This circumstance has an important role for avoide and controls especially the nasal infections [3]. Nowadays, the information about the mechanism of action of antiseptics and disinfectants that used widely are very limited in accordance with antibiotics. The compositions of hydrogen peroxide with 7.5% concentration and phosphoric acid with 0.85% concentration can be used for high level disinfection of endoscopes (compatible with hydrogen peroxide) [4-5]. The product which includes a mixture of 1% hydrogen peroxide, 0.08% peracetic acid is accepted by FDA and is in use epidemically in disinfection of endoscopes in other countries. This product is effective at mycobacteriums resistant to glutaraldehyde [7-8].

For sterilization of endoscopes, the mixtures of 7.35% hydrogen peroxide and 0.23% peracetic acid are accepted by FDA as a contact period of 180 minutes at 20ºC and disinfection conditions in same concentrations during 15 minutes [9-10-11].

In disinfection of hemodialysis machines the use of disinfectants with a combination of aldehyde and perasetic acid increases 10 times between years 1983 and 1997 in accordance with normal disinfectants [6]. In this study it is aimed to investigate the activity of acid and aldehyde basis disinfectants that are usually used in hospital in recent years against the surgical equipment that are contaminated with microorganisms.

MATERIALS AND METHODS

Materials

The alcohol basis disinfectant that is belonging to A Company (a combination of 3.5% perasetic acid, hydrogen peroxide, acetic acid), and to B Company (a combination of 5% organophosphate, 30-50% sodium perborate, 15-20% citric acid), aldehyde basis disinfectant that is belonging to C Company (a combination of 2% glutaraldehyde)that are used in this study are obtained from medical stores.

Methods

Test Microorganisms That Are Used In Trials

Test microorganisms that are used in this study such as Staphylococcus aureus (ATCC 29213), Escherichia coli (ATCC 25922), Pseudomonas aeruginosa (ATCC 23853), Candida albicans ATCC (10231) strains are obtained from culture collection of our laboratory. When counting colonies of these microorganisms, for Staphylococcus aureus, the Staphylococcus medium 110 (Oxoid), for Escherichia coli, Violet Red Bile Agar (Oxoid), for Pseudomonas aeruginosa, Pseudomonas selective medium (Oxoid) and for Candida albicans, Sabouraud-dextrose agar (Oxoid) are used. According to the McFarland 5 blurriness, the final concentration of each strain which is used in this trial will be 2-5x109 CFU/ml and they are prepared correspondent with this concentration [12].

Preparation of Neutralizateur That are used in trials

After the activation of microorganisms of test with disinfectants, for inactivation of disinfectant 3% Tween80 + 3% saponin + 0.1% Histidin + 0.1% Sistein combination is used as neutralizateur in the study [13, 14, 15].

Determination of Disinfectants Effects

In order to determine until which concentration the disinfectant is active, the disinfectant material with different concentration (1%, 0.5%, 0.1%, 0.05%, 0.01%, 0.005%) is distributed into tubes 9 ml. by 9 ml. in each. Then by taking 1 ml of beginning microorganism suspension for each tubes they are added to test tubes which includes disinfectants with different concentrations (1 ml + 9 ml). Microorganisms are kept waiting in test tubes that includes disinfectant materials, during designed period (1, 5, 15 and 30 minutes). At the end of these contact periods 1 ml are taken from each test tube and added on to neutralizateur materials of 9 ml which are in different test tubes. In 1-5 minutes 0.2 ml of example are taken from each tube and are placed into plaques which includes appropriate medium. After an incubation period of 48 hours at 37oC, colonies that are reproduced in appropriate mediums are counted and bacteria numbers in 1 ml. are calculated. At the end of the first minute, the concentration of the disinfectant that cause a decline 5 log and above (the reduction factor is 5 log and above) in the number of microorganism according to the number of microorganism that are treated with disinfectant materials is accepted as effective concentration. Besides, it is confirmed that the neutralizateur materials don't have a deterrent effect on the reproduction of microorganisms and don't cause decline in the number of microorganisms. And also is is confirmed that it inactivate the effect of disinfectant material by the experiments [13-14-15].

RESULTS AND DISCUSSION

The results of disinfectant A against test microorganisms are given in table 1-2-3-4

Table 1. S.aureus's number of colony in 1 ml after the time limit (CFU/ml) In different concentrations treated with A disinfectant's solutions.

Cons. (%)	Effect duration(minute)							
	1mn	RF	5mn	RF	15mn	RF	30mn	RF
10	-		-	-			-	
5	-	-	-		-	-		
2.5	-	-	-		-	-		
1	-	-	-		-	-		
0.5	$5.5x10^2$	5.65	-	-			-	
0.1	$3.3x10^3$	4.88			-	-		
0.05	$1.7x10^5$	3.16	$3x10^4$	3.92	$5.5x10^2$	5.65	$2.2x10^3$	5.05

-: microorganism did not multiply Initial suspension: 2.5 x 109 CFU/mL
RF: log reduction factor F inal con. In the disinfectant.: 2.5 x 108 CFU/mL (8.39 log CFU/ml)

Table2. E.coli's number of colony in 1 ml after the time limit (CFU/ml) In different concentrations treated with A disinfectant's solutions.

Cons. (%)	Effect duration(minute)							
	1mn	RF	5mn	RF 1	5mn	RF	30mn R	F
10	-		-		-		-	
5	-		-		-		-	
2.5	-		-		-		-	
1	-		-		-		-	
0.5	-		-		-		-	
0.1	$>10^6$	<2.2	$>10^6$	<2.2	$1.6x10^5$	3	$1.4x10^5$	3.06
0.05	$>10^6$	<2.2	$>10^6$	<2.2	$>10^6$	<2.2	$>10^6$	<2.2

-: microorganism did not multiply Initial suspension: 1.6x109 CFU/mL
RF: log reduction factor Final con. In the disinfectant: 1.6 x 108 CFU/mL (8.20 log CFU/ml)

Table3. P.aeruginosa's number of colony in 1 ml after the time limit (CFU/ml) In different concentrations treated with A disinfectant's solutions.

Cons. (%)	Effect duration(minute)							
	1mn	RF	5mn	RF	15mn	RF	30mn	RF
10	-		-		-		-	
5	-		-		-		-	
2,5	-		-		-		-	
1	-		-		-		-	
0,5	-		-		-		-	
0,1	$>10^6$	<2,17	$>10^6$	<2,17	$>10^6$	<2,17	$>10^6$	<2,17
0,05	$>10^6$	<2,17	$>10^6$	<2,17	$>10^6$	<2,17	$>10^6$	<2,17

-: microorganism did not multiply Initial suspension: 1.5x109 CFU/mL
RF: log reduction factor Final con. In the disinfectant. 1.5 x 108 CFU/mL (8.17 log CFU/ml)

Table4. C.albicans' number of colony in 1 ml after the time limit (CFU/ml) In different concentrations treated with A disinfectant's solutions.

Cons. (%)	Effect duration(minute)							
	1mn	RF	5mn	RF	15mn	RF	30mn	RF
10	-		-		-		-	
5	-		-		-		-	
2.5	-		-		-		-	
1	-		-		-		-	
0.5	-		-		-		-	
0.1	$1.1x10^5$	2.43	$2.2x10^4$	3.13	$1.1x10^5$	2.43	$4.1x10^4$	2.86
0.05	$>10^5$	<2.47	$>10^5$	<2.47	$>10^5$	<2.47	$>10^5$	<2.47

-: microorganism did not multiply Initial suspension: 3x108 CFU/mL
RF: log reduction factor Final con. In the disinfectant. 3 x 107 CFU/mL (7.47 log CFU/ml)

It is determined that the solution of disinfectant A is effective since concentration of 0.05% against microorganisms that are tested.

In preliminary tests the solutions of 10%, 5%, 1% of disinfectant B is used and after first minute, any augmentation of microorganism is determined. Hereon, in order to find lowest concentration that disinfectant D is active in subsequent experiments, solutions below concentration 1% is used. The results of disinfectant B against test microorganisms are given table 5-6-7-8.

Table 5. S.aureus's number of colony in 1ml after the time limit (CFU/ml) In different concentrations treated with B disinfectant's solutions.

Cons. (%)	Effect duration(minute)							
	30nd	RF	1mn	RF	5mn	RF	30mn	RF
1	-		-		-		-	
0.5	-		-		-		-	
0.1	-		-		-		-	
0.05	10^5	3.39	$5.5x10^2$	5.65	$5.5x10^2$	5.65	$5.5x10^2$	5.65
0.01	$1.1x10^5$	3.35	$5.5x10^2$	5.65	-		-	
0.005	$>10^6$	<2.39	$1.1x10^5$	3.35	$1.26x10^4$	4.29	$8.8x10^4$	3.45
0.001	$>10^6$	<2.39	$>10^6$	<2.39	$>10^6$	<2.39	$>10^6$	<2.39

-: microorganism did not multiply Initial suspension: 2.5 x 109 CFU/mL
RF: log reduction factor Final con. in the disinfectant.: 2.5 x 108 CFU/mL (8,39 log CFU/ml)

Table 6. E.coli's number of colony in 1 ml after the time limit (CFU/ml) In different concentrations treated with B disinfectant's solutions.

Cons. (%)	Effect duration(minute)							
	30nd	RF	1mn	RF	5mn	RF	30mn	RF
1	-		-		-		-	
0.5	-		-		-		-	
0.1	-		-		-		-	
0.05	-		$4.4x10^3$	4.66	-		-	
0.01	$>10^6$	<2.30	10^6	<2.30	-		-	
0.005	$>10^6$	<2.30	$>10^6$	<2.30	$>10^6$	<2.30	$>10^6$	<2.30

-: microorganism did not multiply Initial suspension: 2x109 CFU/mL
RF: log reduction suspension final con. in the disinfectant.: 2 x 108 CFU/mL (8.30 log CFU/ml)

Table 7. P.aeruginosa's number of colony in 1 ml after the time limit (CFU/ml) In different concentrations treated with B disinfectant's solutions.

Cons. (%)	Effect duration(minute)							
	30nd	RF	1mn	RF	5mn	RF	30mn	RF
1	-		-		-		-	
0.5	-		-		-		-	
0.1	-		-		-		-	
0.05	$>1^5$	<3.39	$>10^5$	<3.39	$6.6x10^3$	4.58	-	
0.01	$>10^6$	<2.39	$>10^6$	<2.39	$>10^6$	<2.39	$>10^6$	<2.39
0.005	$>10^6$	<2.39	$>10^6$	<2.39	$>10^6$	<2.39	$>10^6$	<2.39

-: microorganism did not multiply Initial suspension: 2.5x109 CFU/mL
RF: log reduction factor final con. İn the disinfectant: 2.5 x 108 CFU/mL (8.39 log CFU/ml)

Table 8. C.albicans' number of colony in 1 ml after the time limit (CFU/ml) In different concentrations treated with B disinfectant's solutions.

Cons. (%)	Effect duration(minute)							
	30nd	RF	1 mn	R F	5mn	RF	30mn	RF
1	-		-		-		-	
0.5	-		-		-		-	
0.1	-		-		-		-	
0.05	-		-		-		-	
0.01	$>10^5$	<2.39	$>10^5$	<2.39	$>10^5$	<2.39	$>10^5$	<2.39
0.005	$>10^6$	<1.39	$>10^6$	<1.39	$>10^6$	<1.39	$>10^6$	<1.39

-: microorganim did not multiply Initial suspension: 2.5x108 CFU/mL
RF: log reduction factor final con. İn the disinfectant: 2.5 x 107 CFU/mL (7.39 log CFU/ml)

It is determined that solutions of disinfectant B is effective in 30 second. against microorganisms that are tested.

In preliminary tests the solutions of 10%, 5%, 1% of disinfectant C is used and after first minute, any augmentation of microorganism is determined. Here on, in order to find lowest concentration that disinfectant C is active in subsequent experiments, solutions below concentration 1% is used. The results of disinfectant C against test microorganisms are given table 9-10-11-12.

Table 9. S.aureus's number of colony after the time limit (CFU/ml) In different concentrations treated with C disinfectant's solutions.

Cons. (%)	Effect duration(minute)							
	30nd	RF	1mn	RF	5mn	RF	30mn	RF
1	-		-		-		-	
0.5	-		-		-		-	
0.1	-		-		-		-	
0.05	10^5	3.39	$5.5x10^2$	5.65	$5.5x10^2$	5.65	$5.5x10^2$	5.65
0.01	$1.1x10^5$	3.35	$5.5x10^2$	5.65	-		-	
0.005	$>10^6$	<2.39	$1.1x10^5$	3.35	$1.26x10^4$	4.29	$8.8x10^4$	3.45
0.001	$>10^6$	<2.39	$>10^6$	<2.39	$>10^6$	<2.39	$>10^6$	<2.39

-: microorganism did not multiply initial suspension: 2.5 x 109 CFU/mL.
RF: log reduction factor final con. in the disinfectant.: 2.5 x 108 CFU/mL (8.39 log CFU/ml)

Table 10. E.coli's number of colony after the time limit (CFU/ml) In different concentrations treated with C disinfectant's solutions.

Cons. (%)	Effect duration(minute)							
	30nd	RF	1mn	RF	5mn	RF	30nd	RF
1	-	-			-	-		
0,5	-	-			-	-		
0,1	-	-			-	-		
0,05	-		$4,4x10^3$	4,66	-	-		
0,01	$>10^6$	<2,30	$>10^6$	<2,30	-	-		
0,005	$>10^6$	<2,30	$>10^6$	<2,30	$>10^6$	<2,30	$>10^6$	<2,30

-: microorganism did not multiply initial suspension: 2x109 CFU/mL.
RF: log reduction factor final con. in the disinfectant:
2 x 108 CFU/mL (8,30 log CFU/ml)

Table 11. P.aeruginosa's number of colony in 1 ml after the time limit (CFU/ml) In different concentrations treated with C disinfectant's solutions.

Cons. (%)	Effect duration(minute)							
	30nd	RF	1mn	RF	5mn	RF 3	0mn	RF
1	-	-			-	-		
0.5	-	-			-	-		
0.1	-	-			-	-		
0.05	$>10^5$	<3.39	$>10^5$	<3.39	$6.6x10^3$	4.58	-	
0.01	$>10^6$	<2.39	$>10^6$	<2.39	$>10^6$	<2.39	$>10^6$	<2.39
0.005	$>10^6$	<2.39	$>10^6$	<2.39	$>10^6$	<2.39	$>10^6$	<2.39

-:microorganism did not multiply initial suspension: 2.5x109 CFU/mL
RF: log reduction factor final con. in the disinfectant: 2.5 x 108 CFU/mL (8.39 log CFU/ml)

Table 12. C.albican's number of colony after the time limit (CFU/ml) In different concentrations treated with C disinfectant's solutions.

Cons. (%)	Effect duration(minute)							
	30nd	RF	1mn	RF	5mn	RF	30mn	RF
1	-	-			-	-		
0.5	-		-	-			-	
0.1	-		-	-			-	
0.05	-		-	-			-	
0.01	$>10^5$	<2.39	$>10^5$	<2.39	$>10^5$	<2.39	$>10^5$	<2.39
0.005	$>10^6$	<1.39	$>10^6$	<1.39	$>10^6$	<1.39	$>10^6$	<1.39

-:microorganism did not multiply initial suspension: 2.5x108 CFU/mL
RF:log reduction factor final con. in the disinfectant: 2.5 x 107 CFU/mL (7.39 log CFU/m)

It is determined that solutions of disinfectant C are effective in 30 second. against microorganisms that are tested.

A study shows that gluteraldehyde can kill the spores of B.subtilis in one hour and, the bleacher kills them in two hours [16]. In an other examination, it was shown that, for the decontamination of vegetative and spore forming bacteria that was contamined to dental equipments glutheraldehyde with concentration of 2% is more effective than the other disinfectants (2% sodium hypocloride and 10% polyvinyl psolydone) [17]. In a similar study, it was determined that, gluteraldehyde, bleacher and sodium hypocloryde is effective against Acinetobacter type bacteria [18].

As a result, when choosing disinfectant of equipments it will be true that the disinfectant will be effective in vegetative forms of bacteria and also they don't have to be corrosive and must have wide range of effect.

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