



STUDYING EFFECT OF pH ON THE ANTIMYCOTIC PERFORMANCE OF SOME DISINFECTANTS BY USING QUANTITATIVE SUSPENSION TEST

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ABSTRACT :

The antimycotic effect of some disinfectants was comparatively studied under different pH gradients. Reference strains of *C.albicans*, *A.niger* and *T.mentagrophytes* were used in the current study. Quantitative suspension test was used with an initial inoculum of 1.0×10^7 CFU/mL from each organism. After different exposure time (1; 2; 5; 10; 15; 30 and 60 min), 1.0 mL was taken and added to 9.0 mL of the neutralizer to give the required concentration and left for 10 min before spreading of 0.1mL on Sabouraud Dextrose agar medium. The CFU/mL was recorded for each time point and the reduction of the viable count was recorded as \log_{10} of the count. The obtained results revealed that glutaraldehyde, formaldehyde, phenols and quaternary ammonium compounds are highly effective at alkaline pH. The acid pH gradient of these compounds either fails to give the required result or needs a longer time of exposure. On the other hand, sod. hypochlorite and standard phenol were showed high antimycotic efficiencies under acidic pH than alkaline ones. Using the alkalinizing and/or acidifying agents observed no direct toxic effect on the organism. This indicates that the pH may modify the disinfectant performance, which should be put in our mind during disinfection.

INTRODUCTION

There is no perfect disinfectant as each category has advantages and disadvantages. When a disinfectant is being evaluated, if it is not the exact formulation, their overall performance should not be accepted. Even if the active ingredients are similar in percentages in different formulations, it is unlikely that the rest of particular product contains exact chemical characteristics, pH, surfactant system and chelating agent, as the disinfectant it is being compared to (Bergan & Lystad, 1972).

When recommending any chemical agent as a disinfectant, it is essential to qualify conditions under which it will be effective. These

conditions for use are decided according to extensive "in-vitro" tests. These conditions defining the useful biological activity and determine the influence of other factors as pH, temperature, organic matter and in-use dilution on the activity and stability of the compound (Russell, 1974). The order of death of *E.coli* exposed to acid and alkaline glutaraldehyde was approximately exponential and the disinfectant efficiency of aqueous acid solution was considerably lower than activated alkaline solution (Munton & Russell, 1970). Sporicidal activity of alkaline glutaraldehyde was much higher than that of acidic glutaraldehyde (Borick *et al.*, 1964; Power & Russel, 1989 and Traore *et*

al., 2002). It was recorded that the reactivity of glutaraldehyde with the cells and spores is particularly affected by the use of sodium bicarbonate to alkalize the product (Gorman & Scott, 1977 and Gorman & Scott, 1980). Although aldehydes and quaternary ammonium compounds are more effective at a high pH, chlorines, iodophores, and phenols are more effective at a low pH (Ladd, 1993).

The purpose of this study is to examine the effect of pH on the fungicidal effectiveness of the most common commercially available germicides.

MATERIAL AND METHODS:

Disinfectants:

The following products were tested:

1- Aldehyde:

a-Glutaraldehyde: (1,5-Pentaendiol, Fair Lawn, NJ, USA), 50% active matter, pH 2.8 (Unadjusted).

b-Formaldehyde: (Sigma-Aldrich, Gillingham, Dorset, 36.5-38% aqueous solution), pH 7.0 (Unadjusted).

2- Phenolic compounds:

a-TEK-TROL: (26% Phenol, BIO-TEC Industries Inc, Atlanta, Georgia, USA), pH 8.5 (Unadjusted).

b-Standard phenols: (Saturated phenol, Sigma-Aldrich, Gillingham, Dorset)

3-Chlorine compounds: Sodium hypochlorite: (Javex 12 from Bristol-Mayer, Toronto, Ontario, Canada). About 10% w/v available chlorine; pH 12.1

4-Quaternary ammonium compounds: (from Decon Labs, Inc. Bryn Mawr, Pennsylvania, USA). *0.105 alkyl dimethyl benzyl*

ammonium chlorides and 0.105% alkyl dimethyl ethylbenzyl ammonium chloride. pH 9.5.

The pH was either alkalized by 0.3% sodium bicarbonate to be 8.2-8.4 or acidified by 0.01% HCl to be 2.4-2.6.

Prior to testing all the disinfectant solutions were freshly prepared in sterile deionized water.

Test Organisms:

Three organisms were used:

Candida albicans (SC5314), *A. niger* (ATCC 16888) and *T. mantagrophytes* (ATCC 24953). *C.albicans* was grown overnight in yeast nitrogen base (YNB) medium (Difco Laboratories, Detroit, Mich.) supplemented with 50 mM glucose. Fifty milliliters of medium (in 250-ml Erlenmeyer flasks) was inoculated with *C.albicans* from fresh Sabouraud dextrose agar plates (SDA, Difco) and incubated for 24 h at 37°C in an orbital water bath shaker at 60 rpm. Cells were harvested and washed twice with 0.15 M phosphate-buffered saline (PBS; pH 7.2, Ca^{+} and Mg^{+} -free). Cell pellet was resuspended in 10 ml of PBS, counted after serial dilution using hemacytometer, standardized (1.0×10^7 /mL), and used immediately. The number of cells in aliquots of each challenge suspension was confirmed by quantitative culturing. *A.niger* and *T.mantagrophytes* were grown on SDA for up to 7 days at 25-30 °C. The stock inocula were prepared by flooding the culture With 10 ml of 0.85% sterile saline (0.05% Tween 80). The spores were removed by scraping, washed and counted as mentioned before.

Preparation of the test suspension:

The test suspension was prepared in 10 mL sterile distilled water containing appropriate disinfectant concentration. The fungal titer in the solution at zero time was 1×10^7 CFU/mL.

Two sets of each disinfectant concentration were prepared, unadjusted pH and the second either acidified or alkalized. The test was conducted at the room temperature and the fungicidal activity was evaluated by quantitative suspension test (Griffiths *et al.*, 1999 and Fraud *et al.*, 2001). After exposure time of 1, 2, 5, 10, 15, 30 and 60 min, 0.1 mL sample withdrawn and suspended in 0.9 mL of the appropriate neutralizer for 10 min in order to inactivate any residual disinfectant (Pepper, 1981, Hosnia-Swafy, 1999, and Traore *et al.*, 2002). Serial dilutions were then made in sterile saline and 0.1 mL was directly spread out on SDA. The plates were incubated aerobically at 37 °C for 24 h (*C.albicans*) and 25-30°C for up to 7 days (*A.niger* and *T. mentagrophytes*).

The fungicidal effect (FE) expressed as \log_{10} reduction factor for each exposure time. The $FE = \log_{10} NC - \log_{10} ND$ in which NC and ND represent the CFU/mL in control and disinfectant, respectively.

Test for neutralizer:

Tests were performed to check relative efficiency of neutralizers on the disinfectant (Tuncan, 1993 and Traore *et al.*, 2002). A 1mL mixture of *C. albicans* suspension (1.0×10^7 /mL), neutralizer and the disinfectant to give the required concentration was prepared. Moreover, in order to check whether each neutralizer has a direct toxic effect on the cells, 1 mL mixture of *C. albicans* suspension (1.0×10^7 /mL) and neutralizer to give the required concentration was prepared. After 10 min, 0.1 mL of the originals as well as of the ten-fold serially diluted was spread out on SDA. The plates were incubated at 37°C for 24 h. The growing colonies were counted and CFU/mL was calculated accordingly. Percentage of inhibition was also calculated.

Effect of 0.3% sodium bicarbonate and HcL on *C.albicans* :

This test was conducted In order to exclude the direct effect of sod. bicarbonate or HcL on the organisms. Two sets of *C. albicans* (1.0×10^7 cfu/ml) were used, one was alkalized by 0.3% Sod. bicarbonate and the other was acidified by 0.01% HcL. The CFU/ml was tested before as well as every 15 min by the pour plate method.

RESULTS:

Effect of pH on antimycotic efficiency of disinfectants :

All fungal species used in the current study were sensitive to a high extent to the most disinfectants under test.

Data in table (3 & 6) indicated that, \log_{10} viable count of *C.albicans* was dropped 7 folds and no viable count could be detected after 10; 30; 60; 1; 60 and 1 min by using acidic glutaraldehyde, formaldehyde, standard phenol, Tek-Trol, quaternary ammonium compound and sod. hypochlorite, respectively. Moreover, the \log_{10} of *C.albicans* count was dropped 7 folds by alkalized disinfectants after 2; 5; 30; 2; 10 and 15 minutes by glutaraldehyde, formaldehyde, standard phenol, Tek-trol, quaternary ammonium compound and sod. hypochlorite, respectively.

It was revealed that *A.niger* and *T. mentagrophytes* exhibited a certain intrinsic resistance. Table (4 & 6) illustrated that *A.niger* was reduced 7 logs after 15; 2; 10; and 30 min by using alkaline formaldehyde; Tek-trol; quaternary ammonium compound and sod. hypochlorite, respectively. Alkaline glutaraldehyde and standard phenol failed to achieve this result. On the other hand, all acidified disinfectant failed to get 7 logs reduction in case of *A.niger* except Tek-trol and sod. hypochlorite.

Table (1): Disinfectants used and their neutralizers.

Disinfectant		Neutralizer
Name	Concentration (%)	
Glutaraldehyde	0.125	0.3% glycine Dilution with buffered saline 0.1% Tween 80 in saline 0.1% Tween 80 in saline Lethen broth 0.5% Sod. thiosulfate
Formaldehyde	1.25	
Standard phenol	1.25	
Tek-Trol	0.4	
Quaternary ammonium compound	0.005	
Sod. Hypochlorite	0.005	

Table (2): Efficiency of used neutralizers on the disinfectant and their toxicity on *C.albicans*.

Neutralizer	Efficiency					Toxicity				
	Log ₁₀ count		Log ₁₀ Reduction	Inhibition %	Neutralization efficiency	Log ₁₀ count		Log ₁₀ Reduction	Inhibition %	Recovery %
	Initial	After 10 min				Initial	After 10 min			
0.5% Sod thiosulfate	7	6.79	0.21	3.0	97.0	7	6.83	0.17	2.4	97.6
0.3% Glycine	7	6.77	0.23	3.3	96.7	7	6.81	0.19	2.7	97.3
0.1% Tween80	7	6.69	0.31	4.4	95.6	7	6.76	0.24	3.4	96.6
Lethen broth	7	6.58	0.42	6.0	94.0	7	6.85	0.15	2.1	97.9

Table (3): Efficiency disinfectant's pH on *C.albicans*.

Disinfectant		PH	Mean log ₁₀ reduction at (min)							
Name	Concentration %		1	2	5	10	15	30	60	
Glutaraldehyde	0.125	4.2*	3.6	3.7	4.6	7.0	7.0	7.0	7.0	
		8.4	4.6	7.0	7.0	7.0	7.0	7.0	7.0	
Formaldehyde	1.25	6.8*	0.8	0.8	0.9	1.4	3.4	5.0	6.2	
		8.4	0z.4	1.3	7.0	7.0	7.0	7.0	7.0	
		2.4	0.4	1.2	2.1	2.1	4.2	7.0	7.0	
Standard phenol	1.25	6.8*	2.7	7.0	7.0	7.0	7.0	7.0	7.0	
		8.4	0.2	0.2	0.4	0.4	2.6	7.0	7.0	
		2.4	0.1	0.3	1.1	1.1	3.0	5.2	7.0	
Tek-Trol	0.4%	9.8*	4.7	7.0	7.0	7.0	7.0	7.0	7.0	
		2.4	7.0	7.0	7.0	7.0	7.0	7.0	7.0	
QAC	0.005	10.5*	4.2	4.9	6.1	7.0	7.0	7.0	7.0	
		2.4	0.4	0.9	1.0	1.0	3.7	4.2	7.0	
Sod. Hypochlorite	0.005	10.2*	0.3	3.6	4.7	5.1	7.0	7.0	7.0	
		2.4	7.0	7.0	7.0	7.0	7.0	7.0	7.0	

* unadjusted pH.

Table (4): Efficiency of disinfectant's pH on *A.niger*.

Disinfectant		PH	Mean log ₁₀ reduction at (min)							
Name	Concentration %		1	2	5	10	15	30	60	
Glutaraldehyde	0.125	4.2*	0.3	0.7	1.1	1.2	1.4	1.7	2.3	
		8.4	0.9	1.5	1.7	1.7	2.1	2.7	3.4	
Formaldehyde	1.25	6.8*	0.2	0.2	0.5	1.1	2.8	4.2	5.1	
		8.4	0.2	1.9	2.2	4.5	7.0	7.0	7.0	
		2.4	0.2	0.7	1.1	20.	2.6	4.1	4.8	
Standard phenol	1.25	6.8*	4.0	5.0	6.3	7.0	7.0	7.0	7.0	
		8.4	0.0	0.1	0.3	1.6	2.1	5.3	5.8	
		2.4	0.1	0.1	0.8	1.4	1.8	3.8	5.1	
Tek-Trol	0.4%	9.8*	4.7	7.0	7.0	7.0	7.0	7.0	7.0	
		2.4	3.5	4.1	4.6	7.0	7.0	7.0	7.0	
QAC	0.005	10.5*	4.2	4.9	6.1	7.0	7.0	7.0	7.0	
		2.4	0.0	0.2	0.6	1.8	2.5	3.0	4.8	
Sod. hypochlorite	0.005	10.2*	0.1	0.8	2.6	4.7	6.3	7.0	7.0	
		2.4	4.3	5.6	7.0	7.0	7.0	7.0	7.0	

* unadjusted pH.

Table (5): Efficiency of disinfectant's pH on *T.mentagrophytes*.

Disinfectant			Mean log ₁₀ reduction at (min)						
Name	Concentration %	pH	1	2	5	10	15	30	60
Glutaraldehyde	0.125	4.2*	0.5	1.1	1.3	1.5	2.1	2.4	2.9
		8.4	1.8	2.1	3.0	4.8	5.9	6.2	7.0
Formaldehyde	1.25	6.8*	0.3	0.5	0.7	1.2	2.9	4.6	5.4
		8.4	0.2	2.2	2.8	5.0	6.8	7.0	7.0
		2.4	0.0	1.1	1.3	2.2	3.1	4.9	5.3
Standard phenol	1.25	6.8*	0.9	3.8	4.9	5.8	7.0	7.0	7.0
		8.4	0.1	0.9	1.1	2.6	2.9	4.2	6.7
		2.4	0.1	0.7	1.0	1.8	2.3	3.6	5.4
Tek-Trol	0.4%	9.8*	6.7	7.0	7.0	7.0	7.0	7.0	7.0
		2.4	4.1	4.9	5.6	7.0	7.0	7.0	7.0
QAC	0.005	10.5*	2.8	7.0	7.0	7.0	7.0	7.0	7.0
		2.4	0.0	0.1	0.7	1.3	3.2	4.0	5.3
Sod. hypochlorite	0.005	10.2*	0.4	1.2	3.1	5.8	7.0	7.0	7.0
		2.4	7.0	7.0	7.0	7.0	7.0	7.0	7.0

* unadjusted pH.

Table (6): Exposure time required for each disinfectant to achieve 7 log₁₀ reduction factor.

Disinfectant	PH	Organism		
		<i>C. albicans</i>	<i>A. niger</i>	<i>T. mentagrophytes</i>
Glutaraldehyde	Alkaline	2	NA	NA
	Acidic	10	NA	NA
Formaldehyde	Neutral	NA	NA	NA
	Alkaline	5	15	30
	Acidic	30	NA	NA
Standard phenol	Neutral	2	15	15
	Alkaline	30	NA	NA
	Acidic	30	NA	NA
Tek-Trol	Alkaline	1	2	2
	Acidic	1	10	10
Quaternary ammonium compound	Alkaline	5	10	2
	Acidic	NA	NA	NA
Sod. hypochlorite	Alkaline	15	30	15
	Acidic	1	5	1

NA, not achieved

Table (7): Effect of alkalinizing and acidifying agent on *C. albicans*.

Treatment	Initial count (log ₁₀)	Mean log ₁₀ count at (min)							
		1.0	Inhibition %	15	Inhibition %	30	Inhibition %	60	Inhibition %
0.3% Sod. bicarbonate	7	6.92	1.1	6.91	1.3	6.87	1.9	6.80	2.9
0.001% HcL	7	6.86	2.0	6.82	2.6	6.78	1.6	6.72	1.0
Control	7	6.90	1.8	6.91	1.3	6.89	1.6	6.91	1.3

Data in tables (5 & 6) showed that the viable count of *T.mentagrophytes* was dropped 7 logs after 10 and 1 min by using acidic Tek-trol and sod. hypochlorite, respectively. On the other hand, the viable count of *T.mentagrophytes* was dropped 7 folds by alkaline glutaraldehyde, formaldehyde, Tek-trol, quaternary ammonium compound, and sod. hypochrite after 60; 30; 2; 2, and 15 min,

Data in tables (3; 4; 5, and 6) revealed that, alkalinized glutaraldehyde, formaldehyde, tek-trol, and quaternary ammonium compounds are highly effective than their acidified products. The time of exposure required to reduce the viable count of *C.albicans* was shorter than that of acidified ones to drop the count 7 folds (table 3 & 6). The performance of the disinfectant against *A.niger* and *T.mentagrophytes* were

more or less the same except that these two organisms show some intrinsic resistance. Although acidified formaldehyde and quaternary ammonium compounds failed to reduce the viable count of *A.niger* and *T.mentagrophytes* 7 folds, alkalized products achieve this effect. The viable count of *A.niger* dropped 7 logs after 15 and 10 min by using formaldehyde and quaternary ammonium compounds, respectively while *T.mentagrophytes* required 30 and 2 min, respectively.

Alkalized products of most disinfectants under study were more efficient than acidified ones. Moreover, unadjusted standard phenol pH (neutral) was highly effective than alkalized and/or acidified phenol. Table (6) revealed that *C.albicans* count dropped 7 logs after 2; 30, and 30 min by using neutral, alkalized and acidified phenol, respectively. On the other hand, acidified Tek-trol and sod. hypochlorite was highly efficient than the alkalized ones. Viable count of *C.albicans*, *A.niger* and *T.mantagrophytes* dropped 7 logs by using alkalized sod. hypochlorite after 15; 30 and 15 min, respectively. On acidifying the product, the same effect was achieved after 1;5, 1 min, respectively.

Efficiencies and toxicity of the neutralizers :

Concerning neutralizers used, their efficiencies in neutralizing disinfectant were so high and in the same time they have no direct toxicity on the *C.albicans*. The viable count of *C.albicans* was normally fluctuated after 10 min exposure to neutralizers as well as in the disinfectant-neutralizer mixtures (table 2). It was recorded that the compounds used were efficient in neutralizing the disinfectants. The neutralization efficiencies were ranged from 94-97%. On the other hand, their was no direct effect on *C.albicans*. After 10 min, *C.albicans*

shown a growth ranged from 96.6-97.9% of the initial count.

Effect of alkalizing and acidifying agents:

Table (6) revealed that the viable count of *C.albicans* was fluctuated in the normal range. There was no nsignificant effect of the *C.albicans* cells through out one hour experiment. Under the effect of 0.3% sod bicarbonate, the count was dropped up to 2.9% while it was dropped up to 4% by 0.01% HcL comparing to 1.3% drop in the count of the control experiment.

DISCUSSION:

Disinfectant products were selected to represent the main categories of disinfectants used in veterinary field. The biological activity of each chemical compound is greatly influenced by some factors. Extensive tests are then required to define these conditions if the optimum performance is required.

The term high-level disinfectant is usually used for disinfectants that produce 5 log₁₀ reduction in bacterial numbers within 5 min exposure (European standard suspension test, 1997). This criterion is the minimum requirements for passing such a quantitative suspension test. Moreover, Hernandez *et al.*(2000) mentioned that the product be a disinfectant when a≥4 fold reduction on the initial inoculums. All disinfectants were used at a low concentration in order to compare their activity with Tek-Trol, which used at its recommended concentration (0.4%) by the manufacturer. However, the concentration used for each compound found to be effective on *C. albicans* during the initial study.

Aqueous solution of glutaraldehyde is mildly acidic (~4.2) while Tek-Trol, quaternary ammonium compound and Sod. hypochlorite is

alkaline (pH 9.7-10.4). Formaldehyde and standard phenol solutions are more or less neutral pH. The acidic solutions were alkalinized with 0.3% sod. bicarbonate (Gorman & Scott, 1980) and/or acidified by 0.01% HCL. The results that recorded in this study confirm that aldehydes, tek-trol, quaternary ammonium compounds are highly fungicidal at alkaline pH range. Similar results were published elsewhere indicating that alkaline glutaraldehyde is highly effective as a disinfectant than acidic one (Stonehill *et al.*, 1963; Gorman & Scott, 1980; Fraud *et al.*, 2001; Traore *et al.*, 2002). McGucken & Woodside (1973) found that a 2×10^8 cells/ml of *E.coli* was completely killed in 10 min by 100 µg/mL of alkaline glutaraldehyde compared to a 45% kill produced by the acid solution. The predominant factors governing activity of the aldehydes are, the distance between aldehyde groups as well as tendency of aldehyde to polymerize allowing free aldehyde groups to interact with amino groups of the organism cells (Boucher *et al.*, 1973). This statement essentially agrees with findings of Rubbo *et al.* (1967) as the antibacterial activity is due to free aldehyde groups present in the molecule. Alkalinizing of glutaraldehyde, increases the free aldehyde groups available to be up taken by the organism. Gorman & Scott (1977) found that *E.coli* was usually uptake more alkaline glutaraldehyde than acidic one at any particular concentration attributing this to development of fresh sites on the organism walls due to further penetration of the aldehyde and bicarbonate. Munton & Russel (1973) found that acid glutaraldehyde does not react immediately with the outer cell layer or to the same overall extent as an alkaline solution. This may explain the long time required by acidic glutaraldehyde or formaldehyde to give the same effect of alkaline glutaraldehyde (Tables 3-6). Walsh *et al.* (1999)

and Simon *et al.* (2000) stated that glutaraldehyde interact strongly with the organism cells by reacting with the primary amines present in the peptide chain. They found that the reactive amine is NH_2 and not NH_3 and this explains the higher efficiency of alkaline aldehyde where the amines are in the NH_2 .

The effect of alkalinizing is crucial in terms of fungicidal properties of formaldehyde also. It was recorded that 1.25% acidic pH was able to reduce *C.albicans* 7 logs in 30 min (table 3) comparing to 5 min in case of alkaline one (Table 3). Moreover, acidic formaldehyde fails to reduce *A.niger* and *T.mentagrophytes* count to the minimum requirements through out the experiment. On the other hand, alkaline formaldehyde reduced *A.niger* 7log_s in 15 and 30 min, respectively (table 4; 5 and 6). These results are different from those recorded by Rubbo *et al.* (1967) who found that the biocidal activity of formaldehyde was not greatly modified by changes in pH.

Data recorded in tables (3-6) revealed that acidic sod. hypochlorite is more effective against all organisms under test than alkalinized compound. *C.albicans*, *A.niger* and *T.mentagrophytes* were completely destroyed after 1; 5, and 1 min exposure by the acidified sod. hypochlorite while alkaline compound achieved this effect after longer time of exposure. These results are in agree with that reported by Ladd (1993).

Table (6) revealed that 0.3% sod. bicarbonate and/or 0.01% HCL has no germicidal effect and the count of *C.albicans* was fluctuating within the normal range through out one hour test. Power & Russel (1990) found that addition of 0.3% (w/v) NaHCO_3 to other aldehydes did not affect the antimycotic action indicating that antimycotic character of alkaline glutaraldehyde did not due

to the simple pH effect. Moreover, the same authors found that addition of NaOH increases the sporocidal activity of 2% acid glutaraldehyde not to the same extent as addition of NaHCO₃, suggesting that enhancing effect is different from one alkalizing agent from other indicating that it is not simply due to pH. Sod. bicarbonate may sensitize the outer layer of the organisms to be easily penetrated by the disinfectant. Moreover, NaHCO₃ may cause some alteration in the outer layers aiding interaction and/or penetration of disinfectant with potential substrates as proteins, enzymes and peptido-glycan (Power & Russel, 1990). Vasseur *et al.* (1999) found that decreasing the medium pH to 5.8, 5.6 or 5.4 by addition of acetic, lactic or hydrochloric acids increased lag phase and decreased the growth rate of *L. monocytogenes*. The inhibitory effect was acetic acid > lactic acid > hydrochloric acid. However, addition of NaOH to attain pH values of 9.5, 10.0, 10.5 or 11.0 in the medium produced a dramatic increase of the lag phase at pH 10.5 and 11.0. Growth rates were also decreased while the maximal population increased with high pH values. Under acidic condition (pH 4.5-5.0), *L. monocytogenes* showed resistance to niasin and NaCl while niasin was more effective at pH ranging from 5.7-9.2. This indicates that the pH itself has no harmful effect on the organism (Thomas and Wimpenny, 1996).

Concerning the efficiency and toxicity of used neutralizers, table (2) revealed that, the viable count of *C.albicans* was fluctuated within the normal range under effect of these disinfectant-neutralizer mixtures. Reduction of the viable count was ranged from 0.21-0.42 folds with an inhibition percentages of 3-6%. On the other hand, there was no direct toxicity of the neutralizers on the cells. *C.albicans* count was just dropped by 2.1-3.4% indicating that these neutralizers were perfect (table 2). The obtained

results are more or less coincided with those recorded by (Winer *et al.*, 1965; Russel *et al.*, 1979; Russel *et al.*, 1981; Gardner & Peel, 1986; Linton *et al.*, 1987 and Hosnia-Swaify, 2000).

The results indicated that some disinfectants (aldehydes, Tek-trol, and quaternary ammonium compounds are highly effective as an antimycotic under alkaline pH. Moreover, sod.hypochlorite is highly effective under acidic pH range than alkaline one. From these data, one can safely concluded that pH plays a great role in the disinfectant's performance, which should be put in mind if maximum efficiencies are required.

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استخدام اختبار المعلق الكمي في دراسة تأثير الأس الهيدروجيني علي كفاءة المطهرات

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في هذا البحث تم دراسة تأثير عدد من المطهرات واسعة الانتشار في الحقل البيطري علي ميكروبات كانديدا البيكانز والأسبرجيليس نيجر وكذلك التريكوفاييتون منتاجروفاييتس لمعرفة اقل تركيز كافي لقتل هذه الميكروبات عند أوقات مختلفة تراوحت من دقيقة واحدة وحتى ٦٠ دقيقة . تم تغيير الأس الهيدروجيني لهذه المركبات للقاعدي باستخدام ٠.٣% بيكربونات الصوديوم وكذلك للحمضي باستخدام ٠.٠١% حمض الهيدروكلوريك . ثم اختبرت هذه المركبات عند نفس التركيز علي نفس الميكروبات السابق ذكرها . قورنت النتائج عند درجات الأس الهيدروجينية المختلفة. أثبتت النتائج أن الأس الهيدروجيني يلعب دورا هاما في كفاءة المطهرات التي استخدمت في هذا البحث . في هذا الإطار ثبت أن مركبات الجلوترالديهاث ومركبات الأمونيا الرباعية ومركب أل Tek-Trol كانت أعلى كفاءة عند الأس الهيدروجيني القاعدي وأن تغيير الأس الهيدروجيني لهذه المركبات إلى الحمضي فشل في قتل الميكروبات كما أن بعض المركبات قد احتاج وقت أطول للحصول علي نفس النتائج التي تم الحصول عليها من هذه المركبات عندما تكون قاعدية، ومن ناحية أخرى فإن مركب هيبوكلوريدات الصوديوم كانت أعلى كفاءة عند شقها الحامضي. بالرغم من أن الشق القاعدي لمركبات الفورمالدهيد والفينول كانت أعلى من الشق الحمضي لها لكنها كانت أعلى كفاءة عند الأس الهيدروجيني المتعادل، من هذه النتائج ينصح بوضع الأس الهيدروجيني في الاعتبار عند استخدام هذه المطهرات وصولا لأعلى كفاءة تطهيريه .