

Evaluating the Efficacy of Some Disinfectants, Sterilizers and Detergents Against *Streptococcus Pyogenes* Isolates from Tonsillitis Patient in Kirkuk City

Aylan Arslan Ali¹, Shymaa Hassan Ali²

¹Assist. Lecturer, Central Public Health Kirkuk, ²Assist. Lecturer, Education Directorate Kirkuk

Abstract

The aim of the study was to evaluate the inhibitory effectiveness of some disinfectants, sterilizers and detergents commonly used, including: Dettol (Sabtol), Bleach, ethanol alcohol, liquid soap, dishwashing liquid, ActiveX and Sterile Gel 85 on bacterial growth *Streptococcus pyogenes*. The study included collecting (150) swabs from Patients referred to the hospital and from private medical laboratories (age groups were 16-40 years), the swabs were taken by (Transport media) containing the prepared agricultural medium and transported to the laboratory for the purpose of isolation and diagnosis.

The results of the bacterial sensitivity test under study for these disinfectants, sterilizers and detergents according to the type and concentrations of the preparation (50,25,12.5)%, and that alcohol ethanol, hypochlorite sodium and sterile gel (Gel 85) have a high effect on the growth of bacteria *Streptococcus pyogenes* and with all the concentrations prepared, and while detergents Liquid and liquid soap, dishwashing, had less effect on the bacteria under study, according to the prepared concentrations.

Key words: *Streptococcus pyogenes*, disinfectants, sterilizers, chemical detergent

Introduction

Inflammation of the upper respiratory tract is one of the most common diseases, especially the tonsils, and its treatment depends mainly on taking antibiotics. Hand contamination and respiratory secretions such as cold and mucus are among the most important ways for infection to spread between patients or health care workers and social and economic institutions to a healthy person¹.

The discovery of antibiotics had a great impact on reducing the rate of bacterial infections, which encouraged the production of these antibiotics industrially, but nowadays we find a large number of antibiotics used in treating diseases and bacterial infections, but the effectiveness of these antibiotics is constantly decreasing with the ability of germs to develop Means of self-defense and resistance to the action of antibiotics in several ways, As this resistance has the ability to transmit from one bacterial genus to another that was previously sensitive to a specific antibiotic, and this resistance spreads in direct proportion to the increase in the randomized use of antibiotics^{1,2}.

Sterilization is defined as the process by which all microorganisms and their forms (germs, their spores, filtrates, fungi and their spores) are killed or removed, while disinfection is the process in which part or not all living organisms are removed or killed and it does not affect the spores, germs vary in their sensitivity to sterilizers and disinfectants, the germs negative for the dye of honor are less sensitive to disinfectants than the germs that are positive for the dye of generosity, perhaps due to the nature of their composition and their possession of the outer coating, which may hinder the action of disinfectants, and Mycobacteria is relatively resistant, while spores are highly resistant³, and the random and increasing use has led to the emergence of resistant strains and made the disinfectant lose its effectiveness⁵ and a few of them have the ability to kill spores, and from the need to touch the disinfectant to the area to be disinfected, because Most of these disinfectants do not have the ability to penetrate into the materials surrounding the micro-organism, such as (blood, pus, wounds), Chemical disinfectants are also characterized by instability as it sometimes decomposes into other compounds that help the growth of the microorganism, and some microorganisms have the ability to transform

and change to become resistant to disinfectants ².

Antiseptic term refers to non-toxic chemical sterilizers when used externally to disinfect skin or wounds and do not affect living tissues ³, as for detergents is an important and inexpensive way to prevent the transmission of infection in microorganisms because they are effective in removing contaminants that include bacteria, viruses or parasites. As Antibacterial or Antimicrobial, which is characterized by containing the compound Ingredients with an active anti-microbial activity, the current industrial cleaners are made either from petrochemicals (derived from fats and oils) or other chemical materials such as (sulfur trioxide, sulfuric acid, ethylene oxide and alkalis) and all have microbial effectiveness. Bactericidal ^{4,16}

Studies indicate that this efficacy increases with an increase in temperature, so washing hands with soap and hot water eliminates a large part of the pathological bacteria, and the vital effectiveness of these compounds may be due to their entry into the cell's cytoplasm and its interference in its own cell interactions to form metabolites that lead to bacterial cell death or The similarity of fatty acids to compounds with those that enter into the formation of the cell wall, which leads to the failure to complete its construction and consequently the death of the cell, or the effect may extend to itself, which leads to an increase in its clarification of these cells and consequently their death⁶.

The bacteria *Streptococcus pyogenes* is one of the common causes of disease events in humans and is

responsible for at least 616 million cases of tonsillitis and pharyngitis per year in the world and 111 million cases of infections in children in developing countries as well as causing many diseases, including tissue inflammation. Cellular and cutaneous trauma and toxic shock syndrome, *Streptococcus pyogenes* susceptibility to infection is due to its possession of many virulence factors, including capsule, surface proteins of the cell wall ^{17,1}.

Given the importance of evaluating the effectiveness of sterilizers, disinfectants and detergents against *Streptococcus pyogenes*, we decided to conduct this study, which aims to measure the effectiveness of disinfectants and sterilizers, in inhibiting or killing bacteria

Materials and working Methods

Collecting (150) swabs from the nasopharynx area from patients (male and female) using ready-made swabs containing the culture medium, after they were transferred to the laboratory, they were incubated for 24 hours, after which they were implanted on a blood agar medium and incubated on 37c for 24 hours , (20) of the bacteria *Streptococcus pyogenes* were isolated , and was diagnosed based on the dorsal characteristics that included the size, shape, color, texture of the colonies, and the type of hemolysis, and then microscopically diagnosed for the purpose of describing the shape of the cells by staining them with the Cram statin, Catalase test, sensitivity to bacitracin test, APE20, ^{7,8,9}.

Table (1) Types of disinfectants, chemical sterilizers and detergents used:

	Trade Name	The scientific name	Origin
1	Dettol (Sabtol)	Chloroxymenol	Iraq
2	Bleach	Hypochlorite Sodium	Iraq
3	Ethanol alcohol	Ethyl alcohol	Iraq
4	Liquid Soap	Bright	Iraq
5	Dishwashing liquid	O2	Iraq
6	Sterile gel	Activex	Turkey
7	Sterile gel	Aniosgel 85 NPC	France

The test to detect contamination of disinfectants before performing an allergy test:

For the purpose of ensuring that the disinfectants used in this study are free from bacterial contamination, in order to conduct a sensitivity test for bacteria, the following test was performed according to the method 2,11

1- Inoculating the blood agar with a spreading method with (0.1) ml of disinfectant and incubating at 37 ° C for 7 days.

2 - Take (1) ml of antiseptic and add (1) ml of the (heart and brain broth), the tubes were incubated at 37 ° C for 7 days. The appearance of more than 5 colonies on the plate and the appearance of turbidity in tubes in the middle of the infusion of the heart and brain broth (BHI) indicates Contamination of the disinfectant and its unfit for testing.

Preparation of disinfectant concentrations:

According to the method 2, The stock solution was prepared for seven sterilizers and disinfectants (Dettol (sabot), Bleach, liquid soap, sterile gel, dishwashing liquid, ethanol alcohol) by taking 10 ml of commercially prepared concentrations of these chemical disinfectants and disinfectants and adding it to 90 ml of Sterile distilled water ,The final concentration becomes 100%, and under sterilization conditions the following concentrations were prepared (12.5, 25, and 50%(10,12.

The sensitivity of bacterial isolation testing to disinfectants, sterilizers and detergents using the diffusion drill method:

1- Transfer (2-4) of pure colonies to test tubes each containing 5 ml of Muller-Hinton liquid medium and incubated at 37 ° C for 24 hours.

2 - Reducing bacterial growth Use physiological saline solution until the turbidity occurring is similar to McFarland(0.5) tube turbidity.

3- The sterile cotton swab was inserted into the tubes containing the bacterial growth and the increase was removed by applying pressure on the inner walls of the test tube. Then the swab was spread on the surface of Muller-Hunton medium and blood agar medium, and in different directions to ensure that the bacteria were spread evenly 13.

4- Pits with a diameter of 5 mm were made in the inoculated medium with a cork drill,0.1 ml of previously prepared disinfectants were added to each hole using a micro pipette. The dishes were incubated at 37 ° C for 24 hours.

5 - Measure the diameter of the inhibition zone in millimeters 14.

Results and discussion:

20 samples of *Streptococcus pyogenes* were isolated out of (150) swabs, and were diagnosed based on Phenotypic, Microscopic, Chemical and APE20 tests.

Sensitivity testing of bacterial isolation to disinfectants, sterilizers, and detergents:

Table (2) Results of the different concentrations effect of disinfectants, sterilizers and detergents used on *Streptococcus pyogenes*:

Types	Average inhibition zone diameters measured in (mm) according to the concentrations used		
	%12.5	%25	%50
Dettol	R	(25) S	(30) S
Bleach	(30)S	30 >S	30 >S
Ethanol alcohol	30 >S	30 >S	30 >S
Liquid Soap	R	R	(10) S
Dishwashing liquid	R	(8) S	(8) S
Activex sterile gel	R	(16) S	(18) S
Gel 85 sterile gel	30 >S	30 >S	30 >S

Table (2) shows the bacterial isolation under study showed different sensitivity to the disinfectants, sterilizers and detergents used, and this discrepancy was evident depending on the type and concentration of the disinfectant used. The results showed that all the disinfectants used at a concentration of 100% have a great effect on the growth of bacteria compared to other concentrations, and the disinfectants (Bleach, ethanol alcohol and Gel85) are among the most sterilizers and disinfectants that affect the growth of *Streptococcus pyogenes* and in all the prepared concentrations, as shown in picture (1) This is consistent with the study 5 which indicated that *Streptococcus spp.* Kill with specified time and concentration when exposed to the Bleach, The Bleach is one of the disinfectants with a wide range of effectiveness against many microbes by destroying bacteria cells through the production of hypochlorous acid, which is a strong oxidizing agent that binds directly with cellular membrane proteins and enzymes as well as its effect on DNA by inhibiting its synthesis in the bacterial cell, Likewise, ethyl alcohol has the advantage of its ability to affect bacteria by drawing water from the cell and draining it or depleting it inside the cell, working on coagulation, denaturing protein and depositing fats ¹⁵.

As for the antiseptic Dettol, the bacteria under study showed resistance to it at the concentration (12.5), while the sensitivity to it was shown at the concentration (25)% in the inhibition area (25) mm and at the concentration (50)% in the inhibition area (30) mm, as in the picture (2) , And Al-Khalidi indicated in her study that Dettol disinfectant was the least effective on its bacterial

isolates, and this is not in agreement with the current results that showed that the dettol has a high efficiency in inhibiting the bacterial isolation under study at a concentration of (25 and 50(% 5.

Activex sterile gel had an effect on the growth of the bacteria under study at the concentration (25)% in the inhibition area (16) mm and at the concentration (50)% in the inhibition area (18) mm, while the bacteria did not show any sensitivity at the concentration (12.5%), as shown in Image (2).

As for detergents, liquid soap and dishwashing liquid are among the least disinfectants affecting the growth of bacteria under study, especially at concentrations (12.5 and 25)% compared to a concentration of (50)%, as in picture (3), The present study agrees with Rama when it was mentioned that the detergent inhibitory concentration increases with the increase in the detergent concentrations used in the study, due to a number of components included in the composition of the detergents that work to inhibit the metabolic activities and kill the bacteria ^{18,4}.

The resistance of bacteria under study to sterilizers, disinfectants and chemical cleaners may be due to the bacteria acquiring the characteristic of resistance resulting from mutations that lead to mutations in cellular metabolism or the bacteria acquiring resistance genes from plasmids or transposone genes, or the occurrence of a change in the target sites or a change in the permeability of the outer membranes of the bacterial cell walls ^{4,2}.

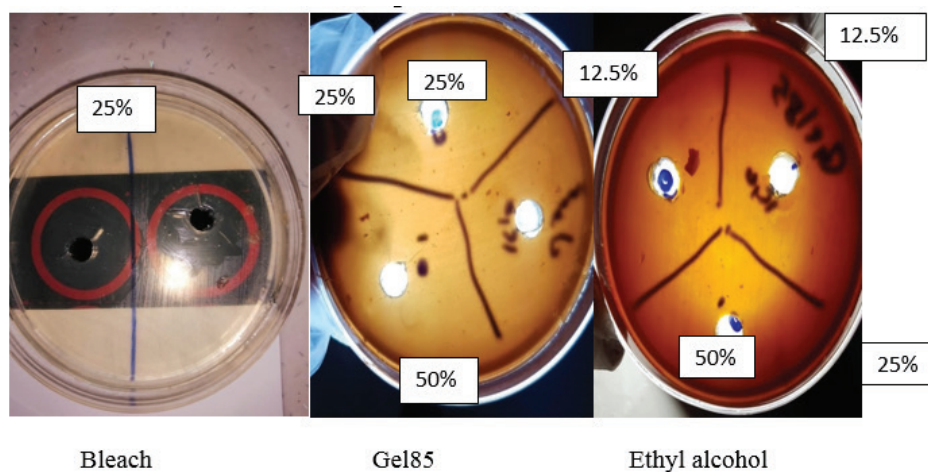


Image (1) The effect of ethyl alcohol, Gel85 and Bleach on the growth of *Streptococcus pyogenes* and at all the prepared concentrations.

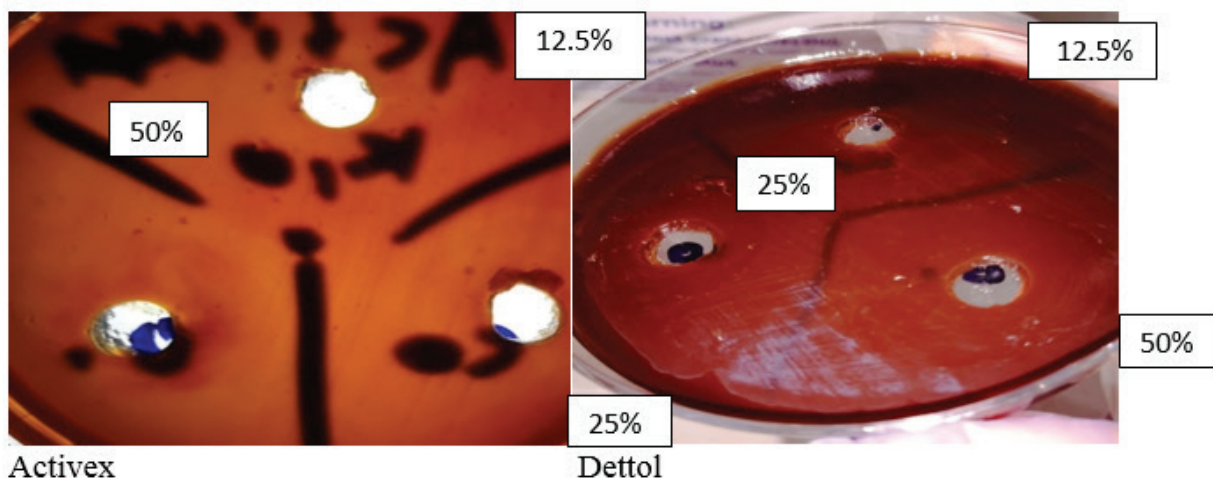


Image (2) The effect of Dettol and ActiveX Gel on the growth of *Streptococcus pyogenes*

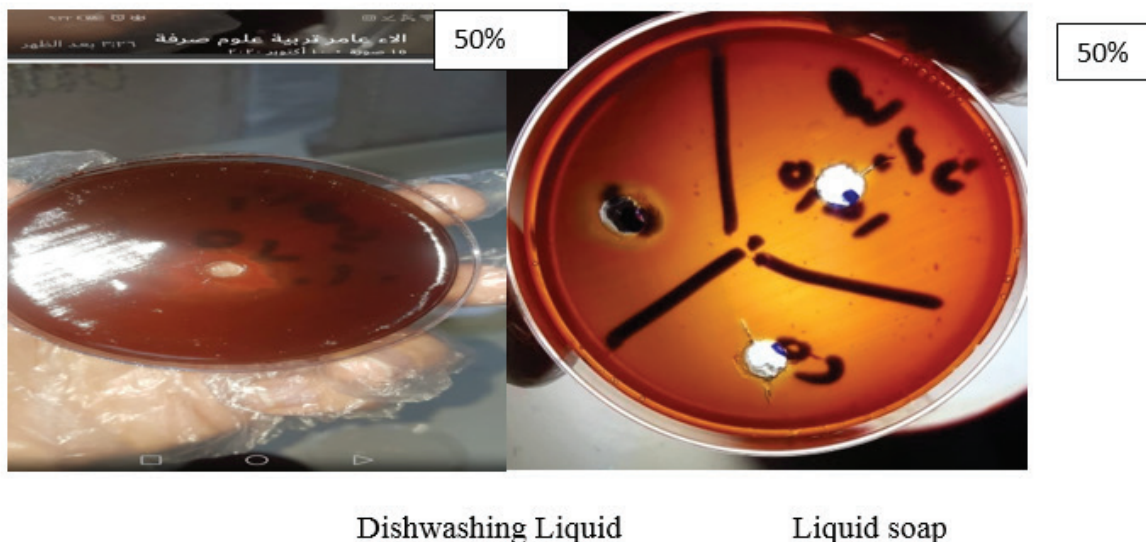


Image (3) The effect of liquid and dishwashing liquid soap on the growth of *Streptococcus pyogenes*.

Conclusions

The results of this study showed that the most disinfectants and sterilizers affected the growth of *Streptococcus pyogenes*, with all prepared concentrations being alcohol Ethanol, Bleach and Sterile gel Gel 85, and while detergents, Liquid soap and Dishwashing liquid, had less effect on the bacteria under study, according to the concentrations used.

I recommend that chemical disinfectants and sterilizers be used randomly and without referring to the manufacturer’s instructions and installed on the packaging of the chemical disinfectant, which makes the disinfectants lose their effectiveness due to inappropriate

use and in the wrong place or because they are used in inappropriate concentrations, as this leads to the emergence of many resistant bacterial strains For many disinfectants, sterilizers and antibiotics alike, which results in a serious problem that lies in the difficulty of controlling infectious diseases, and conducting a molecular and genetic scientific study to find out the relationship between bacterial resistance to antibiotics and their resistance to disinfectants and chemical sterilizers.

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