# **EVALUATION OF DISINFECTANT: ALGARD**

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# **Abstract**

Algard HWS-256 is a one step disinfectant manufactured by Alliance World. Testing algard under laboratory conditions is undoubtedly useful in assessing the activity of disinfectants. However, such testing must be regarded as no more than a preliminary to field trials. The aim behind my study is to record the end-point of a disinfection procedure. In this study few resistant and ATCC strains were used to check the efficacy of the disinfectant by suspension method. To establish the test concentration and the contact time a suspension method is generally applied because the bacteria are uniformly exposed to the disinfectant. As per recommendation, 3.9ml of algard is effective per liter of water with the contact time of 10 minutes. And this study was able to evaluate that the concentration of algard recommended is effective against organisms to kill them at a rapid rate for MDR and ATCC strains.

Keywords: Algard, Disinfectant, Efficacy.

# **INTRODUCTION:**

Disinfectant is a chemical agent which is used to destroy and eradicate variety of microorganisms and their spores. Disinfectants are used within hospitals, dental surgeries, clinics, laboratories, kitchens and bathrooms regularly to kill infectious organisms and their spores. The process of killing the microbes is called disinfection. Evaluation of disinfectant is very useful in order to control hospital acquired infections, as failure can result in many hospital acquired infections thus leading to increased cost, morbidity and mortality by using the lowest concentration available. It can also help in preventing the spread of harmful bacteria and viruses, so once in a while their efficacy must be tested. Disinfectants are different from other agents which are used to resist the growth of organisms, such as antibiotics, which destroy micro-organisms within the body, and antiseptics, which destroy micro-organisms on living tissue. They work by damaging the cell wall of microbes or interfering with their metabolism very rapidly so that microbes cannot produce any response in return.<sup>2</sup>

The discussion below focuses on the action of particular disinfectant Algard HWS-256 by Alliance Formulations on some targeted micro organisms suspended in a media by suspension method.<sup>3</sup> Algard is a one step disinfectant, bactericidal, fungicidal and virucidal and having contact time of 10 minutes. Each 100ml contains active ingredients like didecyl dimethyl ammonium chloride and n-alkyl dimethyl benzyl ammonium chloride<sup>4</sup>. The solutions should be prepared on the day of test. Specified strains are recommended. A synthetic broth is used for preparing a series of subcultures to be used in the test, incubated and CFU is counted. Due to effective contact time, it kills bacteria at a rapid rate. Disinfectants generally kill bacteria on non-living surfaces like floors and inanimate objects. <sup>5</sup> Over the

last few decades there had been a huge increase in the emergence and spread of antimicrobial resistant enzymes of antibiotic resistant bacteria and contributing to morbidity, mortality, and increased hospital expenditure. So it becomes very important to use specific concentration of certain disinfectant in hospital premises to avoid cross infections and hospital acquired infections.<sup>6</sup> To evaluate the efficacy of HWS- 256, we will use suspension method since it is easy to perform; material is easily available and offers quick results. The premise is that after introducing a microbe into a broth medium containing a specific concentration (.39%) of disinfectant, will not allow them to grow<sup>7</sup>. The type of disinfectant to be used in the hospitals depends on various factors like the contact time, the spectrum, irritability rate etc. Some possess a wide spectrum (which kills as many as different types of microorganisms) while others kill a smaller range of disease-causing organisms but are preferred for having other properties like being non-corrosive, non-toxic, less irritable to humans, longer shelf life or inexpensive).<sup>8</sup>

Correct solution strength for 660 ppm quat 0.39%					
Product	Water				
3.9ml algard	1 litre				
10ml algard	2.56 litres				

Table 1: Dilution chart

The aim behind this study is to record the end-point of a disinfection procedure. In this study we tried to check the efficacy of algard disinfectant by suspension method. As per recommendation, 3.9ml of algard is effective per liter of water with the exposure time of 10 minutes. So we made the dense suspension of organisms which are resistant to antibiotics and few sensitive strains to check their survival in the presence of algard disinfectant with different exposure times. We also checked the efficacy of disinfectant by using separate organism's suspended in a media containing algard. So objective behind this study is to evaluate the use of algard in hospitals. If algard is working effictively or not. The suspension test estimates in-vitro bactericidal activity of the disinfectant

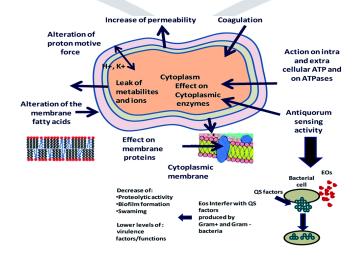


Figure: 1 Action of disinfectant

# **MATERIAL AND METHOD**

# **Test group**

A total of 11 clinical isolates of different multi drug resistant organisms and we also included the ATCC strains from different samples like pus, urine, blood received from patients admitted in Fortis Hospital, Mohali formed the test group of this study.

Table 2: Micro-organisms used

ATCC STRAINS	RESISTANT STRAINS		
ESCHERICHIA COLI	ACINETOBACTER BAUMANNII- MDR		
ENTEROCOCCUS FECALIS	KLEBSIELLA PNEUMONIAE-MDR		
HEMOPHILUS INFLUENZA	METHICILLIN-RESISTANT		
	STAPHYLOCOCCUS AUREUS		
STAPHYLOCOCCUS AUREUS			
PSEUDOMONAS AERUGINOSA			
SALMONELLA			
CANDIDA ALBICANS			
ENTEROCOCCUS HORMAECHEI			

# Materials used

The materials used in the present study are given in the following table. All the glassware, media, equipment used in this project was provided by the Microbiology Department of Fortis hospital.

Table 3: List of materials used in project.

S.No.	Material used	Type Purpose	
1.	Media	MaCconkey agar Culture/Isolation	
		Peptone broth	Isolation
		Blood agar	Culture/Isolation
2.	Glassware	Conical flasks	Media preparation
		Beaker	Media preparation
		Measuring cylinder	Media preparation
		Petri plates	Isolation
		Test tubes	Isolation
		Stirring rods	Mixing of solution

3.	Instruments and	Weighing balance	Media preparation	
	Equipments			
		Bacteriological	Incubation	
		Incubator		
		Bio-safety cabinet	Inoculation into	
			various culture media	
			and biochemical test	
			media, Subculture	
		Density meter	Measure density of	
			colonies	
4.	Others	Scissors, wooden		
		sticks, stop watch,		
		inoculating loops,		
	JUL	face mask, gloves		
	146	etc.		

This study is carried out during training period from 1st January to 30th April, 2019 in the microbiology laboratory of Fortis hospital Mohali, in order to check the efficacy of algard disinfectant in the laboratory by covering a period of 4 months.<sup>2</sup>

#### Sterilization

Glassware used in the project was washed with 2% soap solution followed by washing with distilled water. They were then dried in hot air oven and wrapped in paper and plugged using cotton plug and sterilized in autoclave for 15 minutes at 15 per square inch (psi) pressure at 121 degree Celsius (°C).

# Methodology:

So, in this study a suspension method was used by using ATCC strains like E.coli, E.fecalis, H.influezae, S.aureus, P.aeruginosa, Salmonella, C.albicans, E. hormaechei and few resistant strains like Klebsiella, A.baumannii, MRSA and mixed with disinfectant for testing algard under laboratory conditions. A loopful of bacterial suspension was brought in contact with the disinfectant.<sup>11</sup>

We also performed the experiment by using organisms such as E.coli, MRSA, P.aeruginosa, suspended in a separate broth mixed with disinfectant to check its efficacy.

**Suspension Test:** A sample of bacterial culture was suspended into the peptone water and density of the solution was checked i.e. density should be more than 5. Then 3ml of .3% amount of disinfectant was added into it and after

that suspension was exposed to different time durations i.e. 5, 10, 15 and 20 minutes . It was verified by subculture on to the solid media and incubated for 24-48 hours to check whether the organisms are killed or not. 12

**Processing:** After the proper denser solution was made, then was crossed checked by following guidelines in the laboratory:-

- 1. Density of solution was checked (more than 5).
- 2. Labeling of the test tubes was checked.
- 3. Volume of peptone water (3ml).

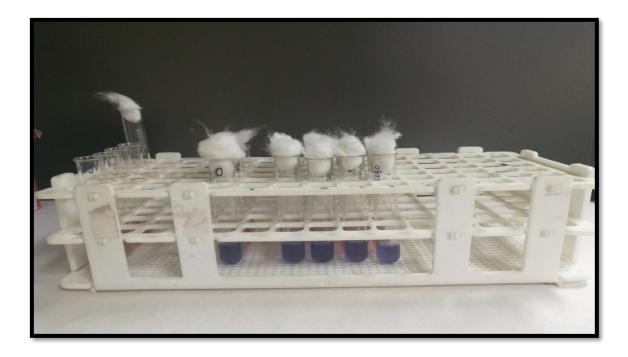


Figure 2: Preparation of microbial suspension mixed with Algard.

A master dilution was prepared by mixing 3ml of peptone water with few colonies of different ATCC and resistant strains into it. Check density of the solution on density meter. The required density should be more than 5. Add 10 ml of algard disinfectant into it. Mix well and then pipette 1ml of dense solution from master dilution to 4 new test tubes and give different exposure time to each tube of 5, 10, 15 and 20 minutes respectively.<sup>13</sup>

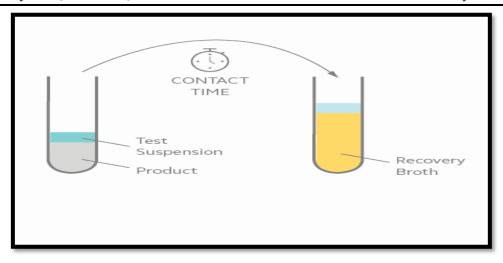


Figure 3: Preparation of suspension

**Media inoculation:** When the processing was done for preparing the suspension and exposure time was complete, then the solution was inoculated on Mac-Conkey and blood agar plates by surface streak plate technique. <sup>9</sup>

**Incubation:** After the culturing is done on solid media. The plates were incubated at  $37^{\circ}$  C  $\pm 2^{\circ}$ C f or 24-48 hours.



Figure 4: Culture plates ready for incubation.

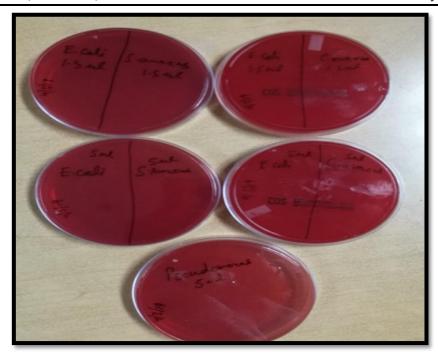


Figure 5: Shows sub culturing of suspension containing separate organisms on MacConkey and blood agar plates

**Results:** A total of 11 microbial isolates from hospitalized patients were used and culture was sterile after 24-48 hours of incubation. None of the organism was able to survive separately as well within the suspension in the presence of the disinfectant which means the test was effective to resist the growth of organisms. Control plate showed growth of mixed bacterial flora in 5, 10, 15 and 20 minutes of exposure.

**Table 4:** shows results of all the incubated plates with different exposure time.

Exposure	Organism 1	Organism 2	Organism 3	Suspension	Control
Time(mins)	(E.coli)	(MRSA)	(P.aeruginosa)		
5	NG	NG	NG	NG	+
10	NG	NG	NG	NG	+
15	NG	NG	NG	NG	+
20	NG	NG	NG	NG	+

NG indicates: No growth

+ indicates: Growth



Figure 6: Culture plates are sterile after 24 hours of incubation.

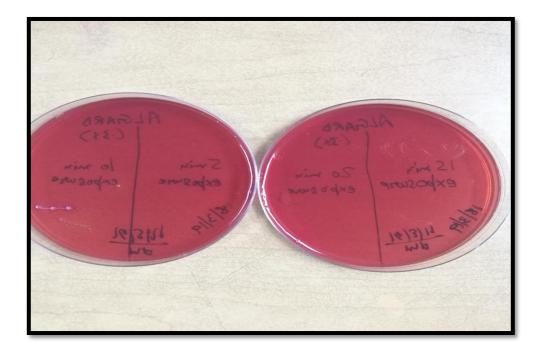


Figure 7: Culture plates are sterile after 48 hours of incubation.

#### **Discussion:**

Under laboratory conditions- Algard turned out to be very useful for variety of organisms which contains bacteria (ATCC & MDR) and Fungus (C.albicans). Control was used to check the sensitivity of the procedure. Algard was effective to kill the entire bacterial load. Manufacturer recommends the time exposure of 10 minutes to kill the micro-organisms and their spores but our study found that even after the shortest exposure of 5 minutes with this disinfectant can kill the organisms which is really effective to prevent the spreading of pathogenic organisms in the hospital premises. And it is also helpful for the safety of doctors, technicians, patients, attendants and other staff of the hospital. The continue evolution of antimicrobial resistance of the organisms in the hospital threatens to evaluate disinfectant after every 6 months to ensure quality results and safety of the hospital workers.

## **Limitations:**

We used qualitative suspension method and prevention of organic matter. Organic matter in the form of serum, blood, pus, egg, or lubricant material can interfere with the antimicrobial activity of disinfectants. My study includes bacteria and fungi, but not viruses or yeast species.

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