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Effectiveness /Efficacy Degree of Locally Made Hand Sanitizer against Gram-Negative Bacteria (GNB) using Ultraviolet-Visible (UV-VIS) Spectroscopic Signatures

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Abstract: Hand hygiene, particularly hand sanitizing, is essential in reducing infectious disease transmission. The recent outbreak of different diseases such as Ebola and corona virus in Nigeria and other countries around the world both increased public awareness of the practice of hand sanitizing and resulted in the introduction of new products to the market. This study aimed to evaluate the degree of antibacterial effectiveness and efficacy of ten locally made hand sanitizers sold in Akungba Akoko area using agar well diffusion and dilution methods. This study was carried out from April to July, 2022. In this study, 10 alcohol-based hand sanitizers were purchased from the main stores in Akungba Akoko. Among them, 8 liquid based hand sanitizers and among 3gel based hand sanitizers. Efficacy of hand sanitizers were evaluated against some Gram negative bacterial isolates including; *Pseudomonas aeruginosa, Enterobacter cloacae, Haemophilia alvei*, *Serratia odorifera*, *Proteus* sp, *Escherichia coli*, *Salmonella multocida*, *Salmonella multocida*, *Klebsiella ornithinolytica* and *Klebsiella ornithinolytica* by agar well diffusion method. Results in this study showed higher inhibitory activity of 70% of the products to the test isolates. In general however, the sanitizers showed good activities, with inhibition of bacteria noted at concentrations as low as 25%. Products tested in this study showed higher zones of inhibition than previously reported, indicating their overall effectiveness. The variations in diffusion and dilution results highlight the effect of texture of the sanitizing product on testing methods and point at a need to properly assess if this could perhaps have any effect in real time on inhibitory activities. The hand sanitizing products tested in this study are suitable in disease prevention. However, regulatory bodies may need to focus on product texture until the effect of this on activity is determined.

Keywords: Locally Made hand Sanitizer, Gram-Negative Bacteria (GNB) Ultraviolet-Visible (UV-VIS) Spectroscopic.

Research Paper

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INTRODUCTION

A hand sanitizer or hand antiseptic is a supplement or alternative to hand washing with soap and water. Keeping hand clean is one of the most essential actions for the reduction of transmission of infectious diseases in the community and hospitals environment (Zapka *et al.,* 2017). Cold viruses, flu viruses, and pathogenic bacteria are easily spread through public meeting places such as hospital, school, bus, office etc.

One gram of human feces which is about the weight of a paper clip can comprise one trillion of microorganisms (Haque *et al.,* 2018).

Hands are primary mode of transmission of microbes and infections. Hand hygiene is therefore the most important measure to avoid the transmission of harmful germs and prevent the infections. Hand hygiene is the single most important, simplest, and least expensive means of preventing nosocomial infections. Contaminated hands can serve as vectors for the transmission of microorganisms. Pathogenic microorganisms accountable for outbreaks are spread from the hands of the food handler to others when the food handler contaminates his/her hands and then passes these microorganisms to consumers via hand contact with food or drinks. The consumer is exposed following the ingestion of these microorganisms, which may cause gastrointestinal illness.

Alcohol-Based Hand Rubs (ABHRs) are the most widely used hand sanitizers. They may contain additional active ingredients such as quaternary ammonium compounds (QAC), povidone-iodine, triclosan or chlorhexidine that mainly serve to contribute to the efficacy of formulations (Gupta *et al.*, 2019). In the use of alcohol rubs, ethanol destroys bacteria by causing damage to its cell membrane and denaturation of proteins. Ethanol also prevents the spread of microbes by interfering with cell metabolism and cell division. Although found effective, the mode of action of other antimicrobial agents is not known (Muita *et al.*, 2021).

The present study aims to determine the effective and efficacy of locally hand sanitizers against the selected Gram negative bacteria. Now, in the era of COVID-19, prevention is pricy more than ever, considering the events still taking place due to the worldwide spread of the various organisms and its ferocity, the virus lives for several hours to days depending on the environment according to the WHO (WHO, 2020). However, not all sanitizers work against all pathogens, in other words, one sanitizer is effective against one type of germs but not the other (Ochwoto *et al.*, 2017).

This effectiveness is determined by several factors including the type and concentration of alcohol, formulation and the nature of product, presence of excipients, applied volume, contact time and viral contamination load (Singh *et al.*, 2020). Hand cleanliness will be broadly perceived as a large portion vital in keeping the transmission of contamination especially in the case of disease (Donskey *et al.*, 2017).

Liquid based hand sanitizers are those type of hand sanitizers which have liquid consistency whereas gel based hand sanitizers are those type of hand sanitizers which have gel based consistency. Liquids act more rapidly (~15 s) and leave less residual substance on hands. Gels require about 30 seconds to act, and time loss can reduce compliance (WHO, 2020). Some studies reported high efficacy of cleanser in the reduction of microbial flora while others showed counter effect (Rutala *et al.*, 2016). Generally hand sanitizers are available as alcohol and non-alcohol based cleansers and their use in liquid, foam, gel and cosmetics is common.

Similar to alcohol-based hand sanitizers, benzalkonium chloride (BC), the primary ingredient of NABHS, is generally not effective against non-enveloped viruses (Rai *et al.*, 2017) though a study demonstrating its efficacy against the non-enveloped human coxsackie virus suggest exceptions exist (Rai *et al.*, 2017).

Use of waterless hand sanitizers as an alternative to conventional hand washing has long been debated. Despite some potential advantages over conventional water and soap (quicker and easier usage), instant hand products are generally considered to more effectively meet needs in hospital and health care settings rather than food preparation settings. ABHRs containing 60 to 95% alcohol are recommended as an alternative to hand washing in hospital and health care settings when hands are not visibly soiled. In contrast, use of these alternatives has not been recommended in food establishments because of the inability of these products to remove fat and food debris from soiled hands (Samuel *et al.*, 2020).

Dentists are exposed to different types of infectious microorganisms daily. A large number of pathogens are localized in the oral cavity, which can be transmitted in different ways during dental procedures (Singh *et al.*, 2020) usually by means of air/water syringe and high-speed instruments.

Figure 1: Hand Sanitizer
Source: Adekunle Ajasin University, Akungba-Akoko, (2022).

MATERIALS AND METHODS

Sample collection and size

Ten hand sanitizers used in the study were purchased and collected in their sealed form available in the local markets of Akungba. Hand sanitizers sold in markets were included or sealed or expired, and labeled hand sanitizers were excluded from the study. All the hand sanitizers were represented by their names Corysan, Tetmosol, 2-Sure, Sangel, Nice, Calvary Royal, Cussons Carex, Dettol, Demia, and Passion.

Description of the research site

The study was carried out within Akungba Akoko area, Ondo state, southwestern Nigeria, which has a border with Owo and other Akoko communities in Ondo State. It is the host community to Adekunle Ajasin University, having the location coordination of ($7^0$28'11" N, $5^0$44'10" E) (GPS). Ten hand sanitizers were purchased and collected from the local markets of Akungba. Their efficacy and effectiveness were tested against the isolated organisms in the laboratory of the Department of Microbiology, Akungba.

Research design

This study has a descriptive and quantitative type of research design. It mainly focuses on obtaining information about the effectiveness and efficacy of locally made hand sanitizers against the isolated organisms

Source of test organism

A total of ten isolated organisms i.e.; *Psudomonasaeruginosa, Enterobacter cloacae, haemophila alvei, Serratia odorifera, Protius spp, Escherichia coli, Salmonella multocida, Klebsiella ornithinolytica, Escherichia coli, Aeromonas hydrophilia* were obtained from Central Laboratory Department of Microbiology, Adekunle Ajasin University, Akungba-Akoko.

Identification of isolates

Isolates were identified based on their cellular morphological appearances and a series of biochemical tests.

Gram staining and morphological identification
Biochemical identification tests for Isolates

The isolates were identified by conventional methods. Briefly, for the identification of test isolates, using a sterile wire loop a drop of normal saline was put on the center of grease-free slide and a portion of the colony was picked and emulsified into the center of a glass slide and allowed to air dry before fixing. To gram stain, crystal violet was then applied after 3min. It was then replaced with a gram's iodine for one (1) minute, prior to rinsing with water and application of 95% alcohol until no color appeared on the flow. Slides were then rinsed with water and safranin was applied for 1-2min. This was followed by rinsing and air-drying before being observed microscopically under ×100 oil immersion lens. Growth was interpreted as described by (Ayeni *et al.,* 2019) which where interpreted that purple and blue color indicated the presence of Gram-positive bacteria and pink or red color identify the presence of gram-negative bacteria. Fungal isolates were identified based on cultural and morphological characteristics with reference to the standard atlas (Ayeni *et al.,* 2019). All slants of test organisms were kept at -4ºC prior to the bioassay of the extracts. Extensive series of biochemical tests were carried out to further confirm all the test bacterial strains. Biochemical tests done includes; Indole test, Catalase, Citrate, Methyl Red-Voges Proskaeur (MR-VP), Triple Sugar Iron (TSI), Urease, Motility Test, Oxidase Test, (Kamaliyah *et al*., 2019).

Agar Well Diffusion Test

The antibacterial effect of the isolated organisms on the hand sanitizers was performed using the well variant of the agar diffusion method described by (Valgas *et al.,* 2007). Sterile Mueller-Hinton agar was inoculated in the Petri plates. A sterile cotton swab was dipped into the test tube containing inoculum. The excess inoculum was removed by firmly pressing the cotton swab against the wall of the test tube. The cotton swab was streaked all over the agar surface by rotating the plate at an angle of 60°. Then, it was left to dry at room temperature with the lid closed. With the help of a cork borer, three (3) equally spaced holes were bored in the agar plates. The agar plugs were discarded with the help of a sterile needle. Fifty microliters of hand sanitizers were inoculated in the three wells with sterile water of equal volume in the central well. The plates were incubated at 37°C for 24hrs in an upright position. After 24hrs the zone of inhibition was observed which shows the degree of susceptibility and resistance of the standard ATCC culture. Similarly, the test was also carried out in hundred microliters and one hundred fifty microliters respectively. The zone of inhibition was measured in mm with the help of a ruler (Osuntokun, 2019).

MIC and MBC Determination of test isolates

Minimum inhibitory concentration (MIC) was also measured. Dilution law was used to determine the concentration per plate. The working concentration was 100mg/ml, 50mg/ml, 25mg/ml, 12.5mg/ml, and 6.25 mg/ml. The dilution law; C1 * V1 = C2 * V2. While C l is 100mg/ml and V2 is 20mls was employed to determine the quantity of the sanitizer needed to be added to the agar. The mixture of the agar and hand sanitizer was allowed to solidify and the standardized bacterial isolates were inoculated on the plates each. For this procedure, hand sanitizers were tested on the test bacterial isolates to determine the minimum concentration of inhibition and Minimum Bactericidal concentration. The Petri dish containing the agar and hand sanitizer with the organism was incubated at 37^0C and examined after 24 hours respectively. The lowest concentration of the hand sanitizer at which there is inhibition and a clear zone of elimination of the organism growth is taken as the minimum inhibitory concentration (MIC) and Minimum Bactericidal concentration respectively (Osuntokun *et al*., 2020).

Measurement of Growth dynamic and Death rate of the isolates using Ultra violet spectrophotometer

Growth dynamic refers to the rate at which cells of microorganisms grow at a given time. This test was done to determine the rate of growth of the isolates as well as their killing time in due time. Colony was picked from the stocked culture slant and inoculated into a nutrient broth which was incubated for 24 hours at 37^0C.

A loopful of organisms was picked from the broth culture into nutrient broth in three sets which are set A (sanitizer), B (isolate growth rate), and C (control) respectively. The ultraviolet spectrophotometer was set at 620λ wavelength, warmed up for 15 minutes and then the control was first read, the first reading was taken at zero hours and it continues after every 8 hours for 8 times. At the 8th reading, which is the 56th hour of set A, the sanitizer was added to evaluate the rate of kill. At the reading (Eni, 2019).

RESULTS

Table 1 shows the type of sample purchased, the number of samples purchased, place and time of purchase. In this table, the type of sample purchased was hand sanitizers, the number of samples purchased was 10 and the samples were purchased at the local market Akungba Akoko.

Table 1: Sampling of Hand Sanitizsr

Hand Sanitizer Company Name	Manufacturer Date	Expiry Date
Dettol	10/06/2020	10/06/2022
Evree	17/05/2020	16/06/2022
Corysan	13/03/2020	15/05/2022
Wind	02/02/2020	03/11/2022
Nice	01/03/2021	02/02/2025
Cusson carex	02/08/2022	03/07/2023
2-sure	01/01/2021	02/01/2023
Sangel	01/06/2021	01/06/2023
Tetmosol	06/02/2020	05/02/2023
Calvary Royal	02/03/2023	02/04/2024

Table 2 shows the test isolate's codes, location, time and date of collection. the isolate codes are as follow; OK3, OK22, IK35, IK4, Ok15, T4, T20, Ik16, T10 and 0k21 which were obtained from Adekunle Ajasin University Akungba Akoko, University Health Center on the 14th of September, 2022 at 10:00 AM.

Table 2: Test isolates codes, location and date of collection

Isolate codes	Location of collection	Date of collection
OK3	AAUHC	22/10/2022
OK22	AAUHC	22/10/2022
IK35	AAUHC	22/10/2022
IK4	AAUHC	22/10/2022
Ok15	AAUHC	22/10/2022
T4	AAUHC	22/10/2022
T20	AAUHC	22/10/2022
Ik16	AAUHC	22/10/2022
T10	AAUHC	22/10/2022
0k21	AAUHC	22/10/2022

Key: AAUHC = Adekunle Ajasin University's Health Center

Table 2a shows the cultural characteristic of the test isolates. It was observed in this table that isolate OK22, IK35, and T4 give a grayish color, where, have Greenish, Bluish-white, Pale-colorless, Red with a black center, and Pink to red respectively. OK3, OK22 and T20 are flat in elevation where IK4, IK35, Ok15, T4, T10 and OK3 are convex and Ik16 is raised. All isolates are smooth on the surface. It was observed in this table that all isolates; OK3, OK22, IK35, T20were irregular in shape where IK4, Ok15, T4, Ik16, and T10, OK3 are circular. Shows the characteristics of test isolates under a microscope. It was observed that all isolates; OK3, OK22, IK35, IK4, Ok15, T4, T20, Ik16 and T10were all Gram negative. Isolates OK3, OK22, IK35, IK4, Ok15, T4, T20, Ik16, and T10 were all in pair and rod in arrangement and shape respectively.

Table 2a: Cultural characteristics of test isolates

Isolate codes	Color	Elevation	Surface	Shape	Probable organism
OK3	Greenish	Flat	Smooth	Irregular	*Pseudomonas aeruginosa*
OK22	Greyish-white	Flat	Smooth	Irregular	Greyish to white
IK35	Greyish-White	Convex	Smooth	Irregular	*Haemophiliaalvei*
IK4	Bluish-white	Convex	Smooth	Circular	*Serratiaodorifera*
Ok15	Pale-colorless	Convex	Smooth	Circular	*Protiussp*

Isolate codes	Color	Elevation	Surface	Shape	Probable organism
T4	Grayish-white	Convex	Smooth	Circular	*Escherichia coli*
T20	Red with black center	Flat	Smooth	Irregular	*Salmonella multocida*
Ik16	Pink to red	Raised	Smooth	Circular	*Klebsiellaornithinolytica*
T10	Grayish to white	Convex	Smooth	Circular	*Escherichia coli*
OK3	Yellow-brown	Convex	Smooth	Circular	Yellow-brown

Table 3: Gram staining and cellular characteristics of test isolates

Isolate code	Gram stain	Arrangements	Shapes
OK3	-ve	In pairs	Rods
OK22	-ve	In pairs	Rods
IK35	-ve	In pairs	Rods
IK4	-ve	In pairs	Rod
Ok15	-ve	In pairs	Rod
T4	-ve	In pairs	Rod
T20	-ve	In pairs	Rod
Ik16	-ve	In pairs	Rod
T10	-ve	In pairs	Rod

Key: +ve = positive, -ve = negative

Table 4 shows the confirmatory biochemical tests for the test isolate. It was observed in this table that isolates OK3, OK22, IK35, IK4, Ok15, T20, and Ik16were indole negative where T4, T10 and 0k21 are negative. All isolates; OK3, OK22, IK35, IK4, Ok15, T4, T20, Ik16 and T10were catalase positive where OK3, OK22, IK35, IK4, Ok15 and Ik16 were citrate positive while T4, T20 and T10 were negative. It was observed that isolates OK3, OK22, IK4, Ik16 were Methyl red positive and T10 and OK21, IK35, Ok15, T4, T20 were negative respectively. Isolate OK22, IK35, IK4, Ik16 and 0k21 were V.P positive where OK3, Ok15, T4, T20 and T10 were negative. Isolate OK3, OK22, IK35, T4, T20, T10 and 0k21 were urease negative and IK4, Ok15 and Ik16 were negative. All isolates were motility positive except Ik16. Isolate OK22, IK35, IK4, Ok15, T20, Ik16 were oxidase negative where OK3, T4, T10 were negative.

It the table, isolate, OK3, IK35, Ok15, T20, and 0k21were observed to ferment lactose sugar while OK22, T4, Ik16 and 0k21 are lactose negative. Isolate OK3, IK35, Ok15, T4, T20 and T10 were sucrose negative while OK22, IK4, Ik16 and 0k21 were sucrose positive. It was observed that all isolates were Dextrose, maltose, and fructose positive. However, isolates were identified as *Pseudomonas aeruginosa, Enterobacter cloacae, Haemophilia alvei, Serratia odorifera, Proteus* sp*, Escherichia coli, Salmonella multocida, Salmonella multocida, Klebsiella ornithinolytica* and *Klebsiella ornithinolytica*

Table 4: Re-confirmatory Biochemical tests of Test isolates

Isolate code	**Biochemical test**														
	Indole	**Catalase**	**Citrate**	**Methyl Red-**	**V.P**	**Urease**	**Motility**	**Oxidase**	**Sugar fermentation**						**Presumptive isolate**
									Lactose	**Sucrose**	**Dextrose**	**Glucose**	**Maltose**	**Fructose**	
OK3	-	+	+	-	-	-	+	+	-	-	+	-	+	+	*Pseudomonas aeruginosa*

OK22	-	+	+	-	+	-	+	-	+	+	+	+	+	+	*Enterobacter cloacae*
IK35	-	+	+	+	+	-	+	-	-	-	+	+	+	+	*Haemophiliaalvei*
IK4	-	+	+	-	+	+	+	-	-	+	+	+	+	+	*Serratiaodorifera*
Ok15	-	+	+	+	-	+	+	-	-	-	+	+	+	+	*Proteus* sp
T4	+	+	-	+	-	-	+	+	+	-	+	+	+	+	*Escherichia coli*
T20	-	+	-	+	-	-	+	-	-	-	+	+	+	+	*Salmonella multocida*
Ik16	-	+	+	-	+	+	-	-	+	+	+	+	+	+	*Klebsiellaornithinolytica*
T10	+	+	-	+	-	-	+	+	+	-	+	+	+	+	*Escherichia coli*
0k21	+	+	+	+	+	-	+	+	-	+	+	+	+	+	*Aeromonashydrophila*

Key: + = positive, - = negative, V.P =Voges-Proskauer

The following figures 1-10 shows the Measurement of zones of inhibition of different hand sanitizer at 100%, 50%, 25%, and 12.5% concentration. Amoxicillin as control 0

Fig 1; Measurement of zones of inhibition of Corysan hand sanitizer at 100%, 50%, 25%, and 12.5% concentration. Amoxicillin as control. shows the measurements of zones of inhibition of gram-negative bacterial growth to different concentrations (100%, 50%, 25% and 12.5%) of Corysan hand sanitizer product. In this Fig all the test isolates show significant zones of inhibition to Corysan sanitizer at 100%, 50% and 25% the diameter of the zone of inhibition reduced while at 12.5% there was a less visible zone of inhibition. It was observed that *Klebsiella ornithinolytic has* the highest zones of inhibition to Corysan hand sanitizer (35mm) at 100%, while *Haemophilia alvei* has the lowest zone of inhibition having a diameter of 21mm. At 50%, *Pseudomonas aeruginosa* has the highest zone of inhibition of 28mm and *Protius sp*. has the lowest zone of inhibition of 11mm. at 25%

concentration, *Enterobacter cloacae* have the highest zone of inhibition of 22mm while *Proteus* sp has the lowest zone of inhibition of 4mm. however, at a 12.5% concentration of the hand sanitizer (Corysan) product, there was no zone of inhibition on most of the test isolates.

Fig 2; Measurement of zones of inhibition of Tetmosol hand sanitizer at 100%, 50%, 25%, and 12.5% concentration. Amoxicillin as control. shows the measurements of zones of inhibition of gram-negative bacterial growth to different concentrations (100%, 50%, 25%, and 12.5%) of Tetmosol hand sanitizer products. In this fig, all the test isolates show less significant zones of inhibition to Tetmosol sanitizer at 100% 50% and at 25% and 12.5% there was no visible zone of inhibition. It was observed that *Salmonella multocida* has the highest zones of inhibition to hand sanitizer (26mm) at 100%, while *Pseudomonas aeruginosa* has the lowest zone of inhibition having a diameter of 0mm. At 50%, *Protius spp* has the highest zone of inhibition of 10mm and *Serratiaodorifera* has the lowest zone of inhibition of 0mm. at 25% and 12.5% concentration, there was no zone of inhibition on most of the test isolates.

Fig 3; Measurement of zones of inhibition, Wind hand sanitizer at 100%, 50%, 25%, and 12.5% concentration. Amoxicillin as control. shows the measurements of zones of inhibition of gram-negative bacterial growth to different concentrations (100%, 50%, 25%, and 12.5%) of Wind hand sanitizer product. In this Fig, the entire test isolates show significant zones of inhibition to Wind sanitizer at 100%, and at 50% concentration, the diameter of the zone of inhibition reduced while at 25% and 12.5% concentration. There was a less visible zone of inhibition. It was observed that *Salmonella multocida* has the highest zones of inhibition to wind hand sanitizer (26mm) at 100%, while *Escherichia coli* has the lowest zone of inhibition having a diameter of 12mm. At 50%, *Salmonella multocida* shows the highest zone of inhibition of 23.5mm followed by *Pseudomonas aeruginosa* having a zone of inhibition of 16mm. at 25% and 12.5% concentrations, there was no visible zone of inhibition on most of the test isolates.

Fig 4; Measurement of zones of inhibition, 2Sure hand sanitizer at 100%, 50%, 25% and 12.5% concentration. Amoxicillin as control. shows the measurements of zones of inhibition of gram-negative bacterial growth to different concentrations (100%, 50%, 25%, and 12.5%) of 2Sure hand sanitizer products. In this fig, all the test isolates show significant zones of inhibition to 2Sure sanitizer at 100%, 50%, and at 25% concentration, the diameter of the zone of inhibition reduced while at 12.5% concentration. There was no visible zone of inhibition. It was observed that *Escherichia coli* has the highest zones of inhibition to 2Sure hand sanitizer (35.8mm) at 100%, while *Serratia odorifera* nation d *Protiusspp* has the lowest zone of inhibition having a diameter of 5mm. At 50%, *Escherichia coli* show the highest zone of inhibition of 22mm, and *Aeromonashydrophilia* has the lowest zone of inhibition of 1mm. At 25% and 12.5% concentration, there was no visible zone of inhibition on most of the test isolates.

Fig 5; Measurement of zones of inhibition, Sangel hand sanitizer at 100%, 50%, 25% & 12.5% concentration. Amoxicillin as control. shows the measurements of zones of inhibition of gram-negative bacterial growth to different concentrations (100%, 50%, 25% and 12.5%) of Sangel hand sanitizer product. In this Fig all the test isolates do not show significant zones of inhibition to Sangel sanitizer at 100%, 50%, and 25% concentration. It was observed that only *Escherichia coli* shows the highest zones of inhibition to Sangel hand sanitizer (30mm) at 100% and 50% (25%). At 25% and 12.5% concentrations, there was no visible zone of inhibition on most of the test isolates.

Fig 6; Measurement of Zones of Inhibition, Nice Hand Sanitizer At 100%, 50%, 25%, and 12.5% Concentration. Amoxicillin As Control: shows the measurements of zones of inhibition of gram-negative bacterial growth to different concentrations (100%, 50%, 25%, and 12.5%) of Nice hand sanitizer product. In this Fig, all the test isolates do not show significant zones of inhibition to Nice sanitizer at 100%, 50%, and at 25% concentration. It was observed that only *Salmonellamultocida* shows the highest zones of inhibition to Nice hand sanitizer (22.5mm) at 100%. At 50%, 25% and 12.5% concentration, there was no visible zone of inhibition on most of the test isolates.

Fig 7; Measurement of zones of inhibition, Calvary hand sanitizer at 100%, 50%, 25% & 12.5% concentration. Amoxicillin as control shows the measurements of zones of inhibition of gram-negative bacterial growth to different concentrations (100%, 50%, 25% and 12.5%) of Calvary hand sanitizer product. In this fig, most of the test isolates do not show significant zones of inhibition to Calvary sanitizer at 100%, 50%, and at 25% concentration. It was observed that only *Escherichia coli* shows the highest zones of inhibition to calvery hand sanitizer (21.5mm) at 100%. At 50%, 25%, and 12.5% concentration, there was no visible zone of inhibition on most of the test isolates.

Fig 8; Measurement of zones of inhibition of Cussons hand sanitizer at 100%, 50%, 25%, and 12.5% concentration. Amoxicillin as control shows the measurements of zones of inhibition of gram-negative bacterial growth to different concentrations (100%, 50%, 25%, and 12.5%) of Cussons hand sanitizer product. In this fig, all the test isolates show significant zones of inhibition to Cussons sanitizer at 100%, 50%, and at 25% concentration, the diameter of the zone of inhibition reduced while at 12.5% concentration, there was no

visible zone of inhibition. It was observed that *Escherichia coli* has the highest zones of inhibition to Cussons hand sanitizer (40mm) at 100%, while *Haemophiliaalvei* has the lowest zone of inhibition having a diameter of 17mm. At 50%, *Enterobacter cloacae* show the highest zone of inhibition of 26mm and *Haemophilia alvei* has the lowest zone of inhibition of 15mm. At 25% and 12.5% concentration, there was no visible zone of inhibition on most of the test isolates

Fig 9; Measurement of zones of inhibition of Dettol sanitizer at 100%, 50%, 25%, and 12.5% concentration. Amoxicillin as control: shows the measurements of zones of inhibition of gram-negative bacterial growth to different concentrations (100%, 50%, 25%, and 12.5%) of Dettol hand sanitizer product. In this fig, all the test isolates show significant zones of inhibition to Dettol sanitizer at 100%, 50%, and at 25% concentration, the diameter of the zone of inhibition reduced while at 12.5% concentration. there was no visible zone of inhibition. It was observed that *Haemophilia alvei* has the highest zones of inhibition to Dettol hand sanitizer (32.5mm) at 100%, while *Salmonella multocida* has the lowest zone of inhibition having a diameter of 9.85mm. At 50%, *Enterobacter cloacae* show highest zone of inhibition of 24mm and *Salmonella multocida* has the lowest zone of inhibition of 7.25mm. At 25% and 12.5% concentrations, there was no visible zone of inhibition on most of the test isolates.

Fig 10; Measurement of zones of inhibition, Passion hand sanitizer at 100%, 50%, 25% & 12.5% concentration. Amoxicillin as control shows the measurements of zones of inhibition of gram-negative bacterial growth to different concentrations (100%, 50%, 25%, and 12.5%) of Passion hand sanitizer product. In this table, most of the test isolates do not show significant zones of inhibition to Passion sanitizer at 100%, 50%, and at 25% concentration. It was observed that only *Escherichia coli* shows the highest zones of inhibition to Passion hand sanitizer (30mm) at 100%. At 50%, 25%, and 12.5% concentration, there was no visible zone of inhibition on most of the test isolates.

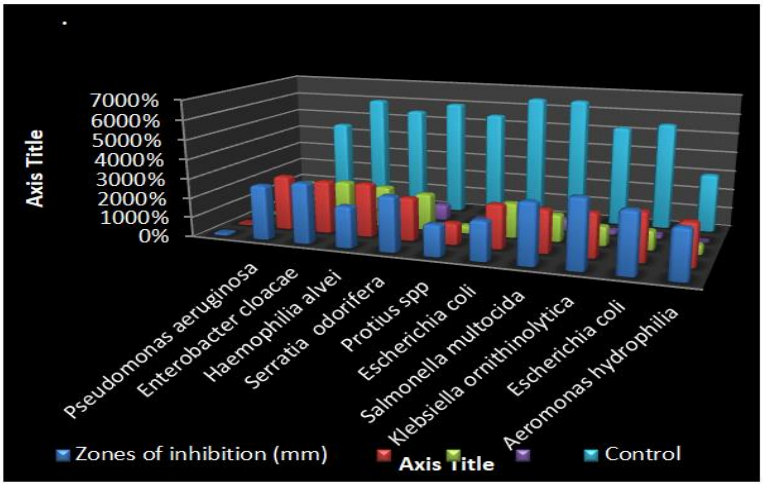

Fig 1: Measurement of Zones of Inhibition, Corysan Hand Sanitizer At 100%, 50%, 25% & 12.5% Concentration

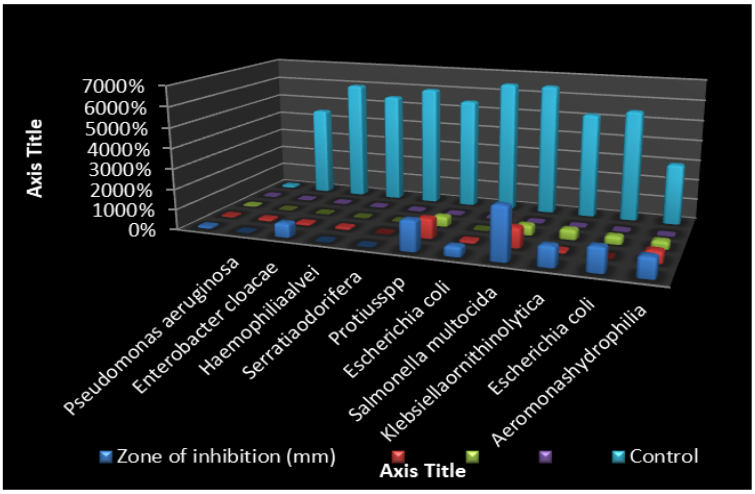

Fig 2: Measurement of Zones of Inhibition, Tetmosol Hand Sanitizer At 100%, 50%, 25% & 12.5% Concentration

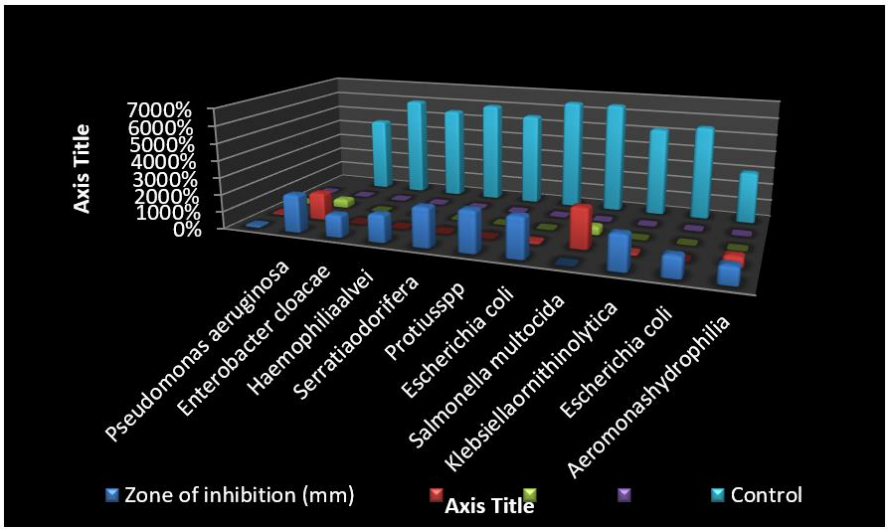

Fig 3: Measurement of Zones of Inhibition, wind Hand Sanitizer At 100%, 50%, 25% & 12.5% Concentration

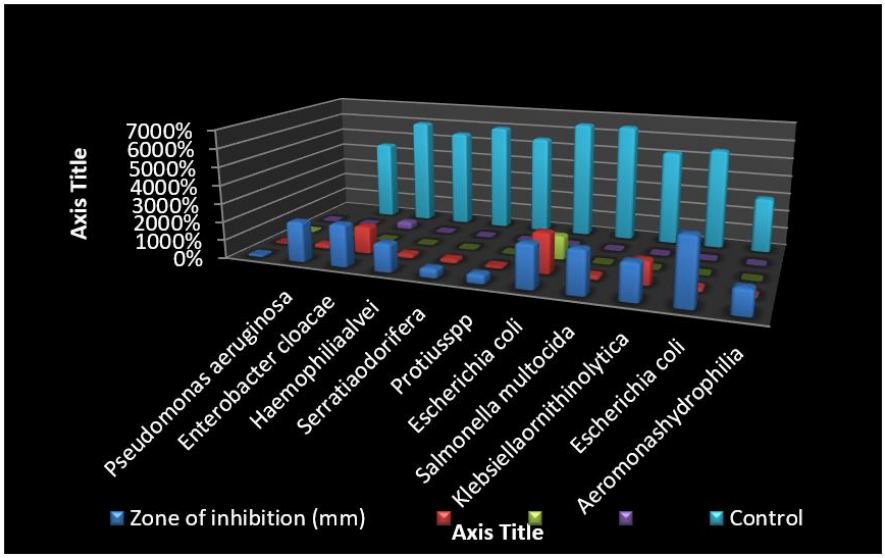

Fig 4: Measurement of Zones of Inhibition, 2sure Hand Sanitizer At 100%, 50%, 25% & 12.5% Concentration

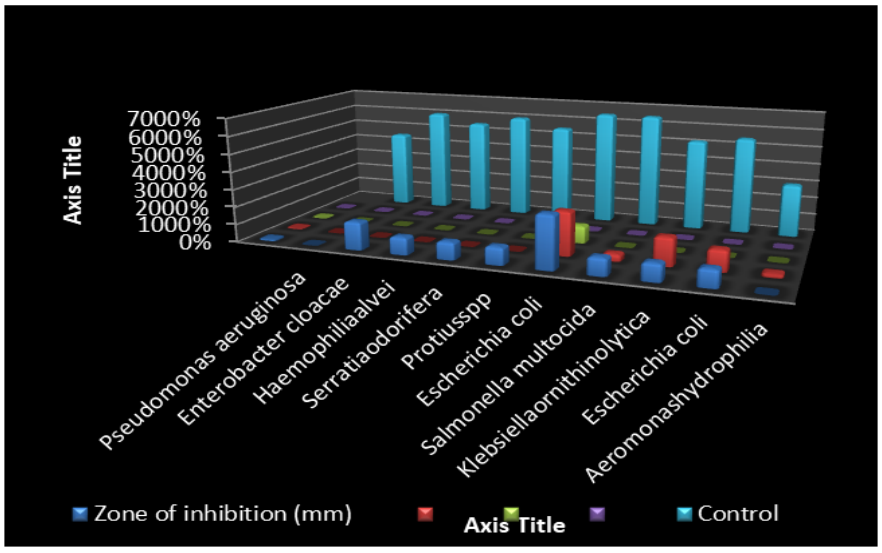

Fig 5: Measurement of Zones of Inhibition, Sangel Hand Sanitizer At 100%, 50%, 25% & 12.5% Concentration

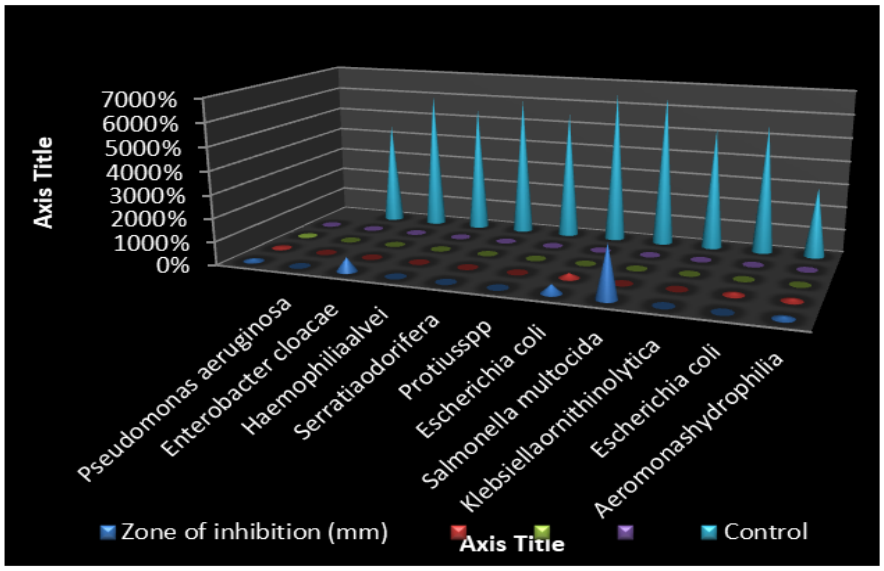

Fig 6: Measurement of Zones of Inhibition, Nice Hand Sanitizer At 100%, 50%, 25% & 12.5% Concentration

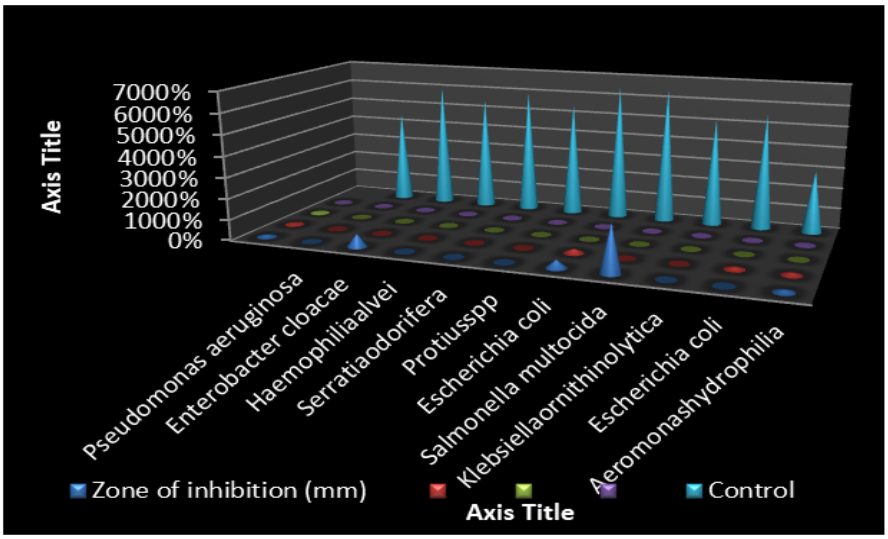

Fig 7: Measurement of Zones of Inhibition, Nice Hand Sanitizer At 100%, 50%, 25% & 12.5% Concentration

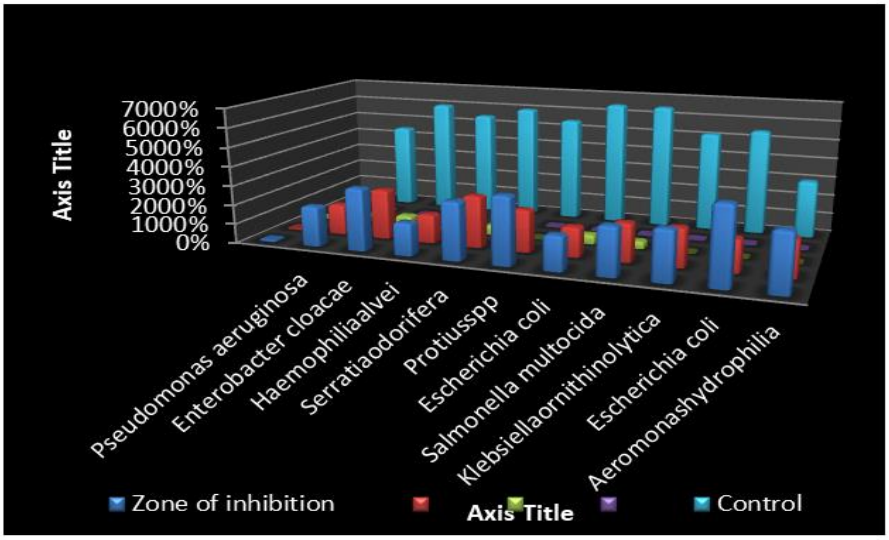

Fig 8: Measurement of Zones of Inhibition, Cussons Hand Sanitizer At 100%, 50%, 25% & 12.5% Concentration

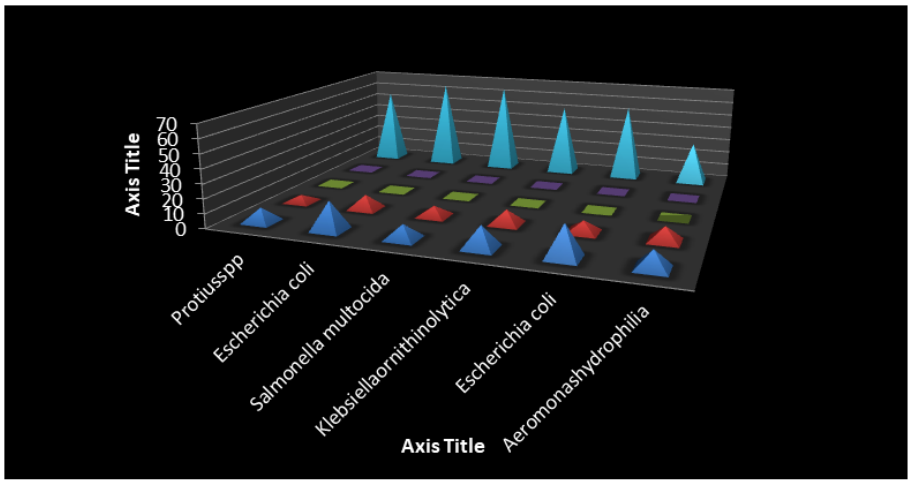

Fig 9: Measurement of Zones of Inhibition, Dettol Hand Sanitizer At 100%, 50%, 25% & 12.5% Concentration

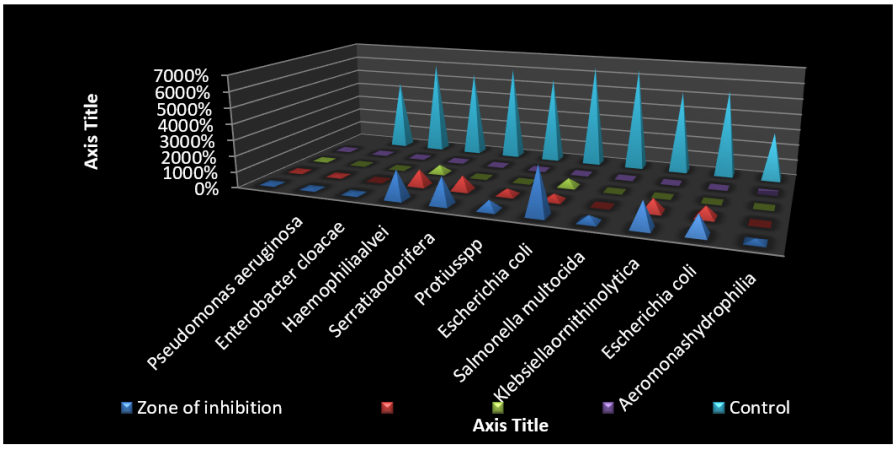

Fig 10: Measurement of zones of inhibition, Passion hand sanitizer at 100%, 50%, 25% & 12.5% concentration

Percentage zones of inhibition of hand sanitizer at 100%, 50%, 25%, and 12.5% concentration. Amoxicillin as control the following figures 11-20 shows the Measurement of percentage zones of inhibition of different hand sanitizer at 100%, 50%, 25%, and 12.5% concentration.

Fig 11; Percentage zones of inhibition of Corysan hand sanitizer at 100%, 50%, 25%, and 12.5% concentration. Amoxicillin as control

Fig 12; Percentage zones of inhibition of Tetmosol hand sanitizer at 100%, 50%, 25%, and 12.5% concentration. Amoxicillin as control

Fig 13; Percentage zones of inhibition of Wind hand sanitizer at 100%, 50%, 25%, and 12.5% concentration. Amoxicillin as control

Fig 14; Percentage zones of inhibition of Wind hand sanitizer at 100%, 50%, 25%, and 12.5% concentration. Amoxicillin as control

Fig 15: Percentage zones of inhibition of Sangel hand sanitizer at 100%, 50%, 25%, and 12.5% concentration. Amoxicillin as control

Fig 16: Percentage zones of inhibition of nice hand sanitizer at 100%, 50%, 25%, and 12.5% concentration. Amoxicillin as control Zone of inhibition (mm)

Fig 17; percentage of inhibition of Calvary hand sanitizer at 100%, 50%, 25%, and 12.5% concentration. Amoxicillin as control

Fig 18; Percentage zones of inhibition of Cusson's hand sanitizer at 100%, 50%, 25%, and 12.5% concentration. Amoxicillin as control

Fig 19; Percentage zones of inhibition of Dettol hand sanitizer at 100%, 50%, 25%, and 12.5% concentration. Amoxicillin as control

Fig 20; Percentage zones of inhibition of Corysan hand sanitizer at 100%, 50%, 25%, and 12.5% concentration. Amoxicillin as control

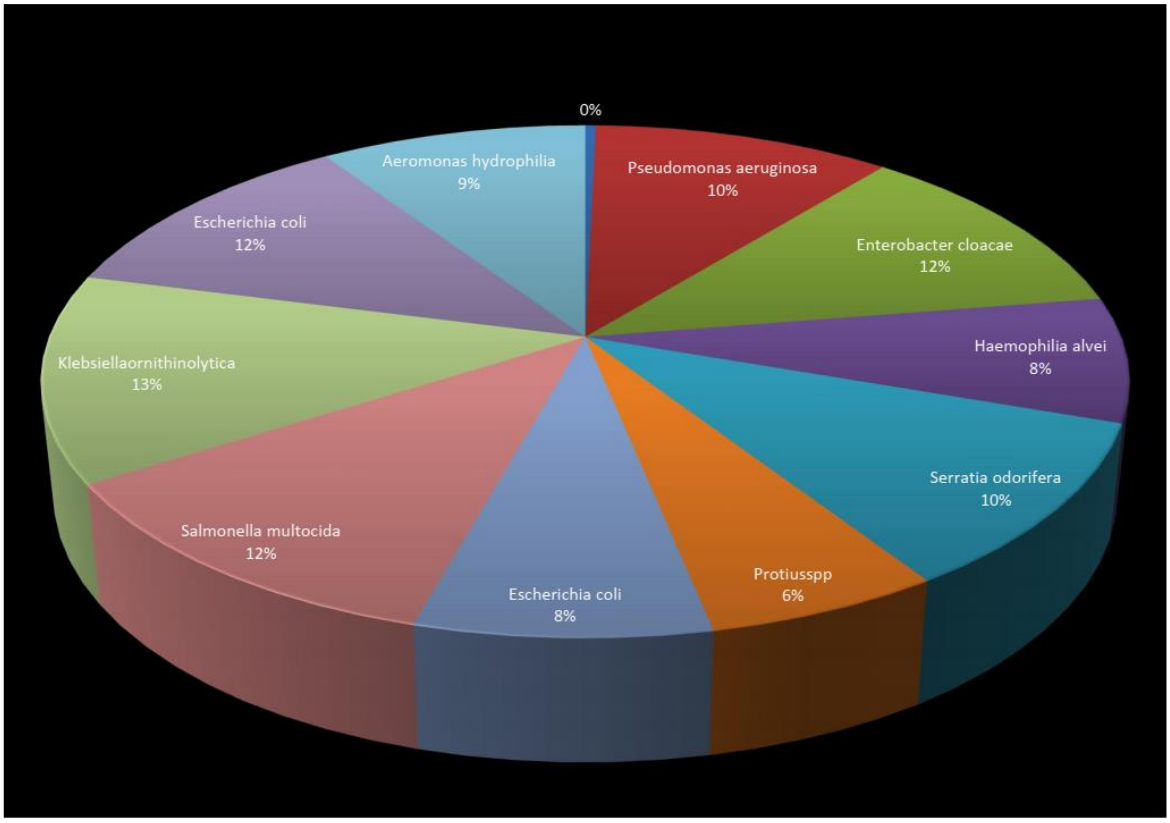

Fig 11: Percentage zones of inhibition of Corysan hand sanitizer at 100%, 50%, 25% and 12.5% concentration

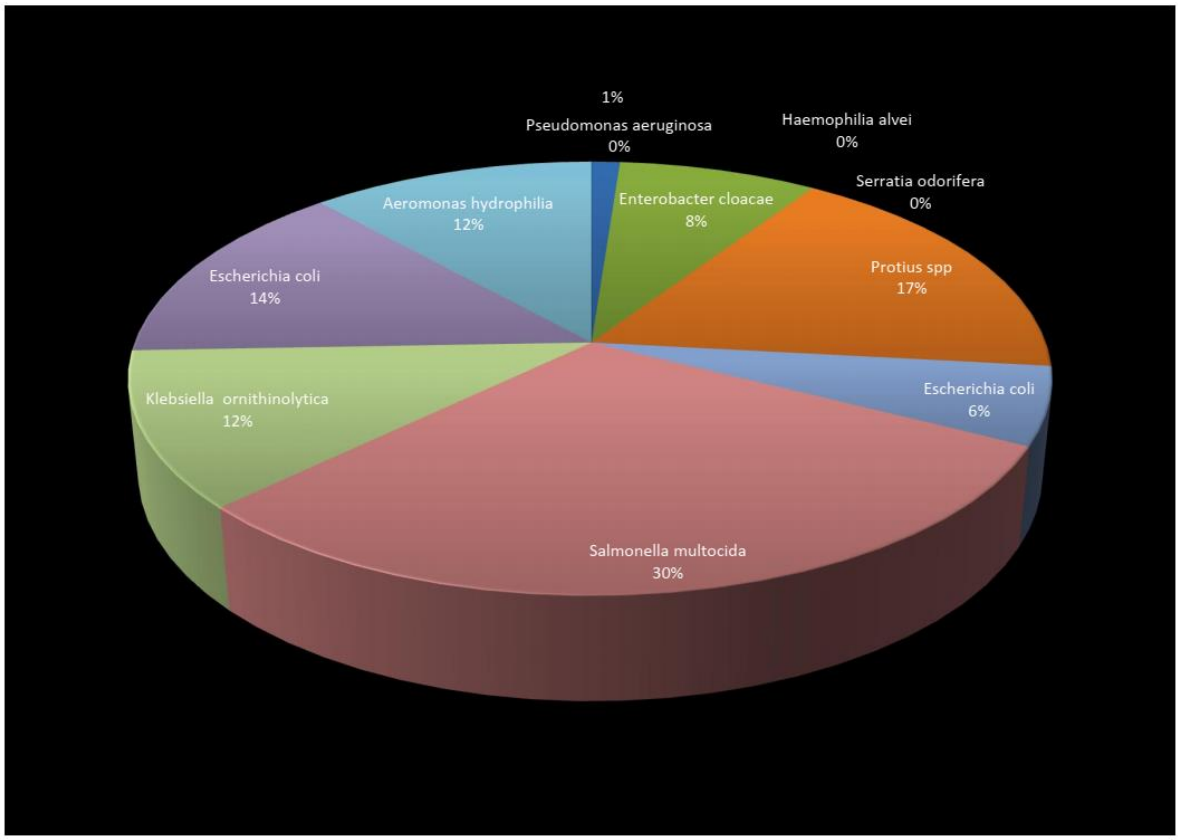

Fig 12: Percentage zones of inhibition of Tetmosol hand sanitizer at 100%, 50%, 25% and 12.5% concentration

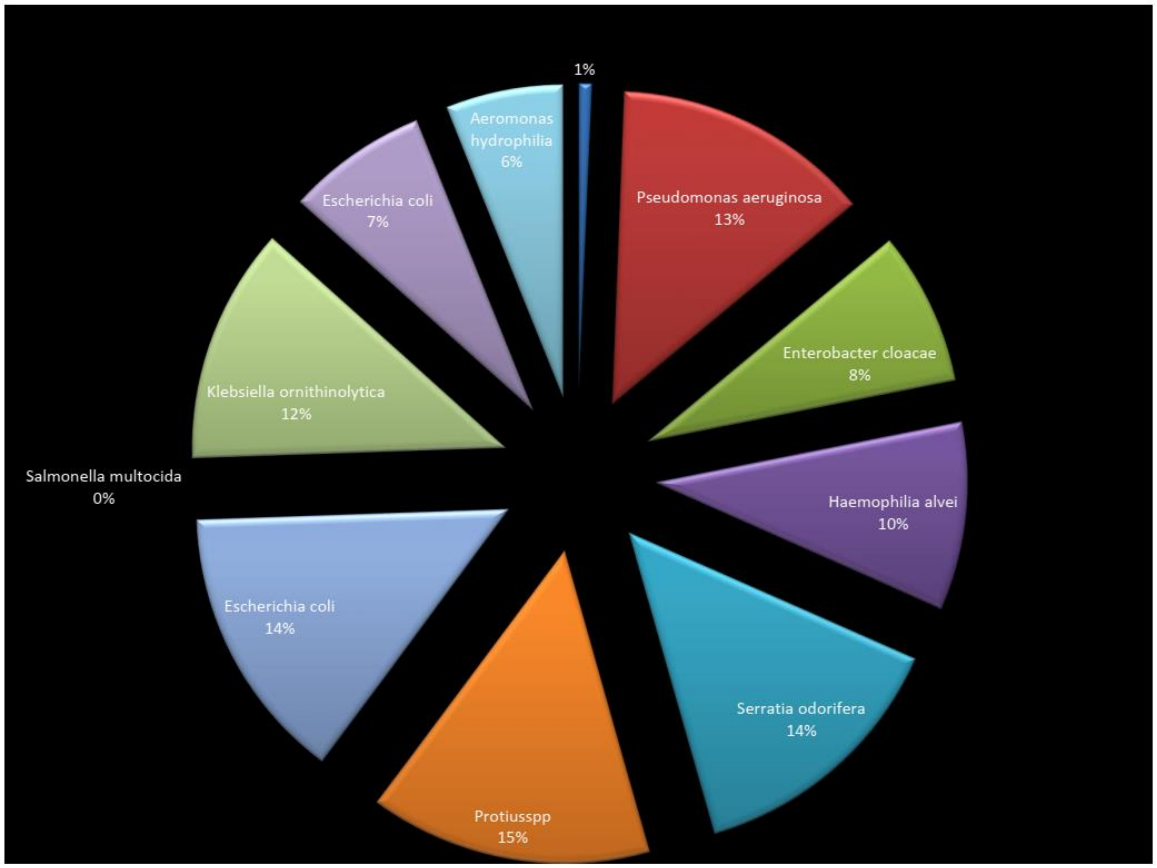

Fig 13: Percentage zones of inhibition of Wind hand sanitizer at 100%, 50%, 25% and 12.5% concentration

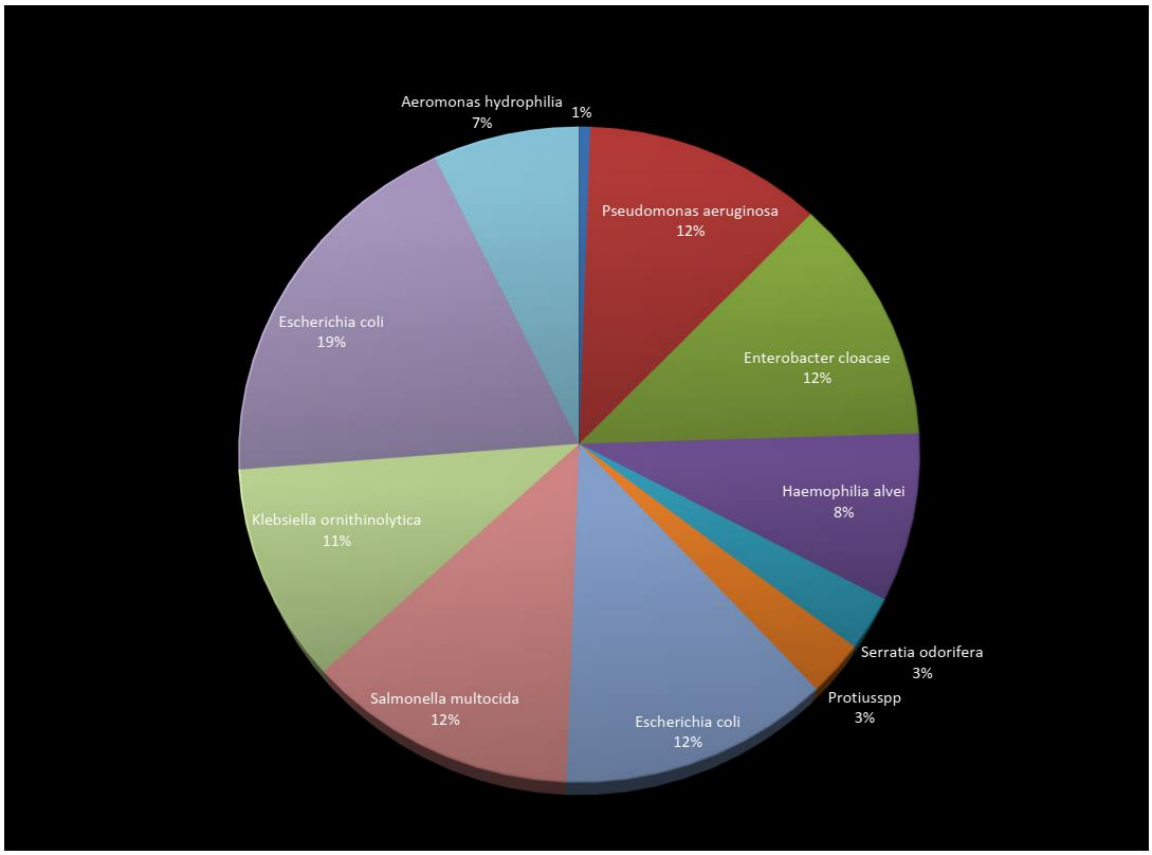

Fig 14: Percentage zones of inhibition of 2Sure hand sanitizer at 100%, 50%, 25% and 12.5% concentration

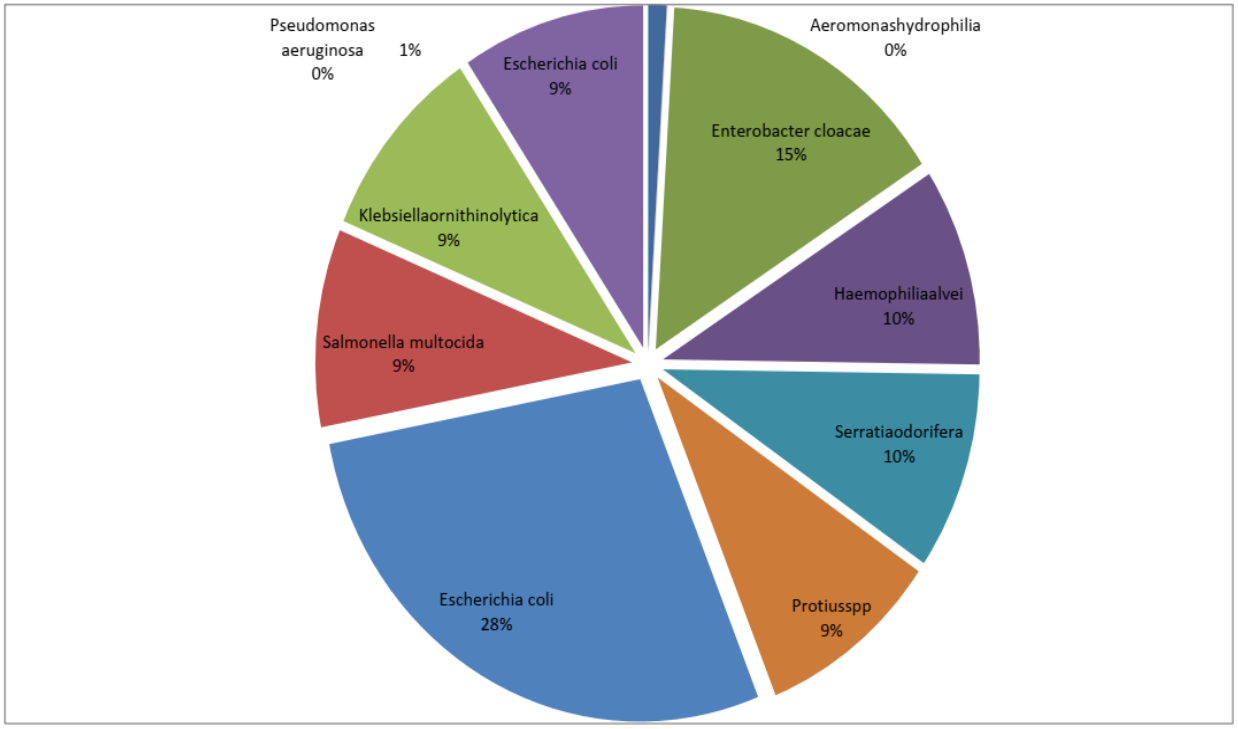

Fig 15: Percentage zones of inhibition of Sangel hand sanitizer at 100%, 50%, 25% and 12.5% concentration

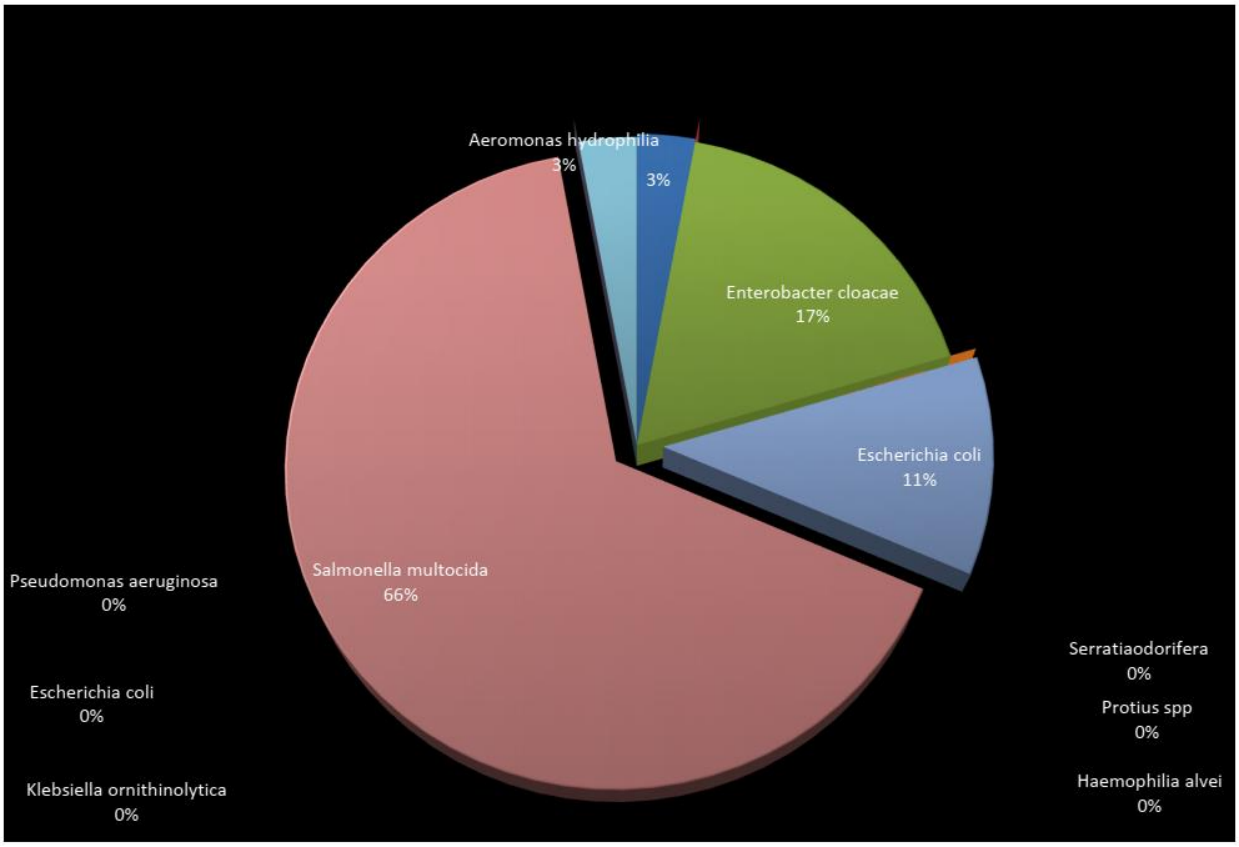

Fig 16: Percentage zones of inhibition of nice hand sanitizer at 100%, 50%, 25% and 12.5% concentration

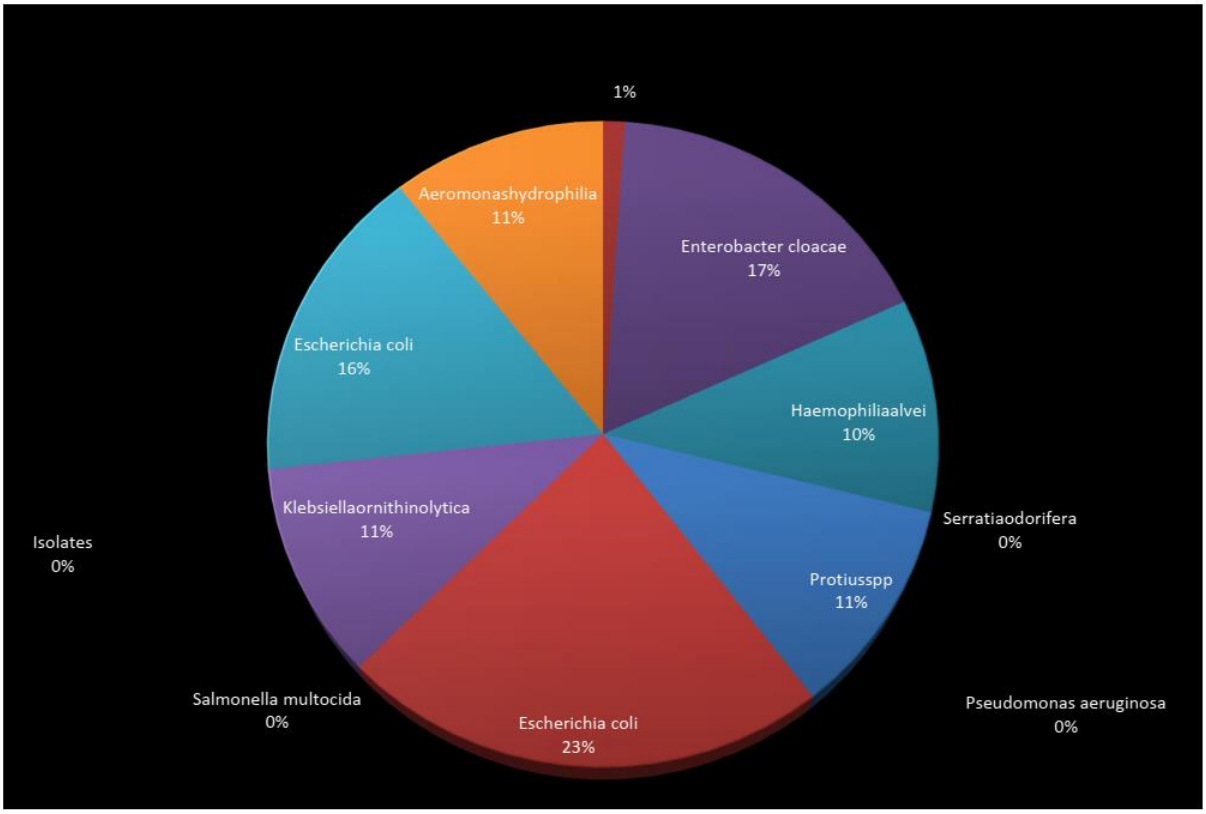

Fig 17: Percentage of inhibition of Calvary hand sanitizer at 100%, 50%, 25% and 12.5% concentration

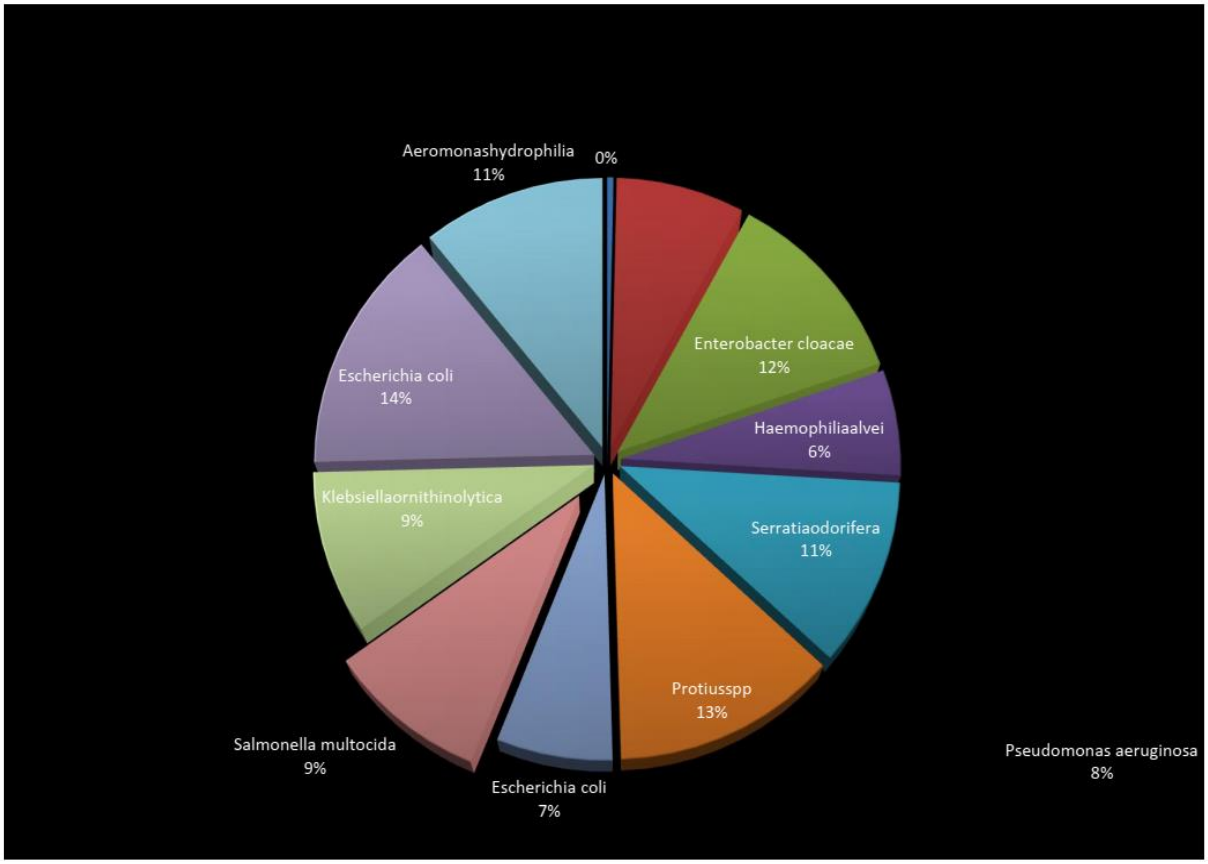

Fig 18: Percentage zones of inhibition of Cussons hand sanitizer at 100%, 50%, 25% and 12.5% concentration

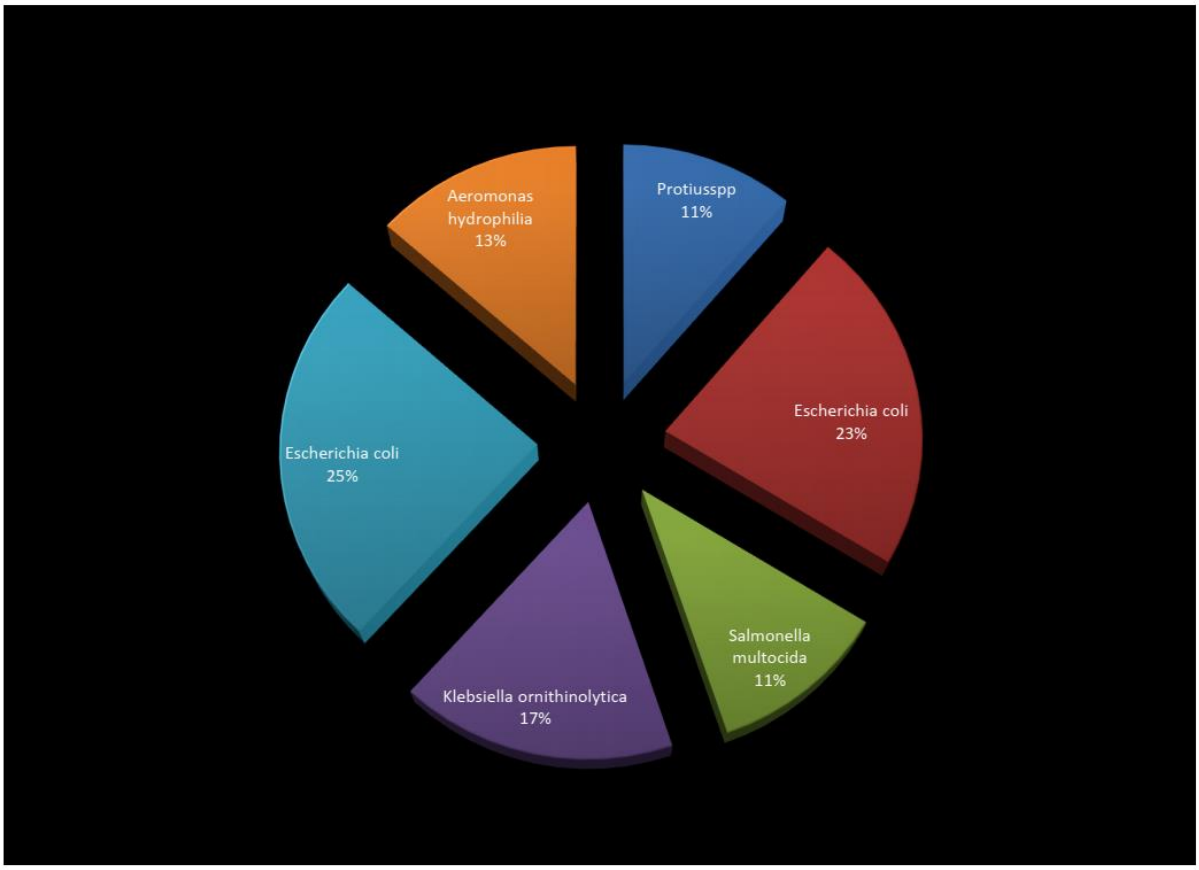

Fig 19: Percentage zones of inhibition of Dettol hand sanitizer at 100%, 50%, 25% and 12.5% concentration

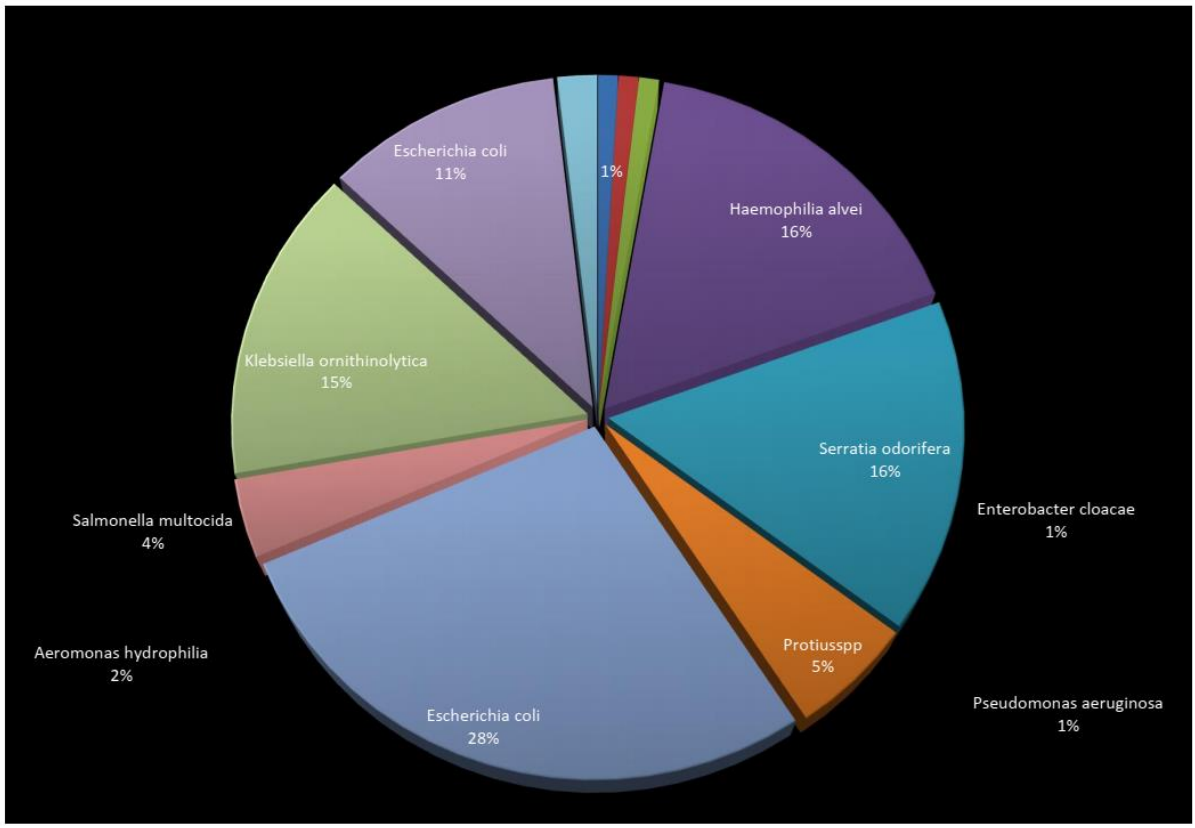

Fig 20: Percentage zones of inhibition of Corysan hand sanitizer at 100%, 50%, 25%, and 12.5% concentration

Table 5: Minimum inhibitory concentration and Minimum bactericidal concentration of hand sanitizer used

Aeromonas hydrophilia	*Escherichia coli*	*Klebsiella ornithinolytica*	*Salmonella multocida*	*Escherichia coli*	*Proteus* sp	*Serratia odorifera*	*Haemophilia alvei*	*Enterobacter cloacae*	*Pseudomonas aeruginosa*	Isolate code
100	NE	50	NE	50	100	NE	50	25	50	**DMIC**
NE	100	100	NE	100	NE	NE	100	50	100	**DMBC**
50	50	50	50	25	50	25	25	25	25	**EMIC**
100	100	100	100	50	100	50	100	50	100	**EMBC**
NE	100	100	NE	NE	NE	100	100	NE	NE	**CMIC**
NE	NE	NE	NE	100	NE	NE	NE	NE	NE	**CMBC**
100	100	NE	50	NE	NE	NE	100	100	50	**WMIC**
NE	NE	100	100	100	100	100	NE	NE	100	**WMBC**
NE	NE	NE	NE	NE	NE	NE	NE	NE	NE	**NMIC**
NE	NE	NE	NE	NE	NE	NE	NE	NE	NE	**NMBC**
50	50	50	50	50	50	50	50	50	50	**CUMIC**
100	100	100	100	100	100	100	100	100	100	**CUMBC**
100	NE	100	100	50	NE	NE	100	50	NE	**2MIC**
NE	100	NE	NE	100	NE	NE	NE	100	100	**2MBC**
NE	100	100	100	50	100	100	100	100	NE	**SMIC**
NE	NE	NE	NE	100	NE	NE	NE	NE	NE	**SAMBC**
NE	NE	NE	100	NE	NE	NE	NE	NE	NE	**TMIC**
NE	NE	NE	50	NE	NE	NE	NE	NE	NE	**TMBC**
100	100	NE	NE	NE	100	NE	100	100	NE	**CAMIC**
NE	NE	100	NE	100	NE	NE	NE	NE	NE	**CAMBC**

Key: NE = No effect

The following figures 11-21 show the growth dynamic of bacterial isolates using an ultraviolet spectrophotometer with wavelength 620λ.

Fig 21; Shows the growth dynamic of test bacteria isolates using an ultraviolet spectrophotometer. In this table, it was observed that at 0 hours, *Aeromonas hydrophilia* has the highest growth rate of 0.250λ and *Serratia odorifera* has the lowest growth rate of -0.080λ. At the 56th hour, *Enterobacter cloacae* have the lowest death rate of 0.350λ and *Escherichia coli* has the highest death rate of 0.120λ.

Fig 22; Shows the growth dynamic and killing time of bacteria isolates and the addition of Corysan hand sanitizer extract at the 56th hour using an ultraviolet spectrophotometer. In this table, it was observed that at 0 hours, *Aeromonas hydrophilia* has the highest growth rate of 0.336λ, and *Haemophilia alvei* has the lowest growth rate of 0.092λ. At 56th hour, *Escherichia coli* has the lowest death rate of 0.100λ and *Haemophilia alvei* has the highest death rate of -0.009λ

Fig 23: Shows the growth dynamic and killing time of bacteria isolates and the addition of Wind hand sanitizer at the 56th hour using an ultraviolet spectrophotometer. In this table, it was observed that at 0 hours, *Salmonella multocida* has the highest growth rate of 0.282λ and *Serratia odorifera* has the lowest growth rate of 0.127λ. At the 56th hour, *Escherichia coli* has the lowest death rate of 0.280λ and *Klebsiella ornithinolytica* has the highest death rate of 0.100λ

Fig 24: Shows the growth dynamic and killing time of bacteria isolates and the addition of Cusson hand sanitizer at the 56th hour using an ultraviolet spectrophotometer. In this table, it was observed that at 0 hours, *Enterobacter cloacae* have the highest growth rate of 0.380λ, and *Haemophilia alvei* have the lowest growth rate of 0.080λ. At the 56th hour, *Escherichia coli* has the lowest death rate of 0.150λ and *Protius spp* have the highest death rate of -0.050λ

Fig 25: Shows the growth dynamic and killing time of bacteria isolates and the addition of 2-SURE hand sanitizer at the 56th hour using an ultraviolet spectrophotometer. In this table, it was observed that at 0 hours, *Klebsiella ornithinolytica* has the highest growth rate of 0.554λ and *Pseudomonas aeruginosa* have the lowest growth rate of 0.170λ. At the 56th hour, *Serratia odorifera* has the lowest death rate of 0.500λ and *Pseudomonas aeruginosa* has the highest death rate of -0.090λ

Fig 26: Shows the growth dynamic and killing time of bacteria isolates and the addition of SANGEL hand sanitizer at the 56th hour using an ultraviolet spectrophotometer. In this table, it was observed that at 0 hours, *Enterobacter cloacae* have the highest growth rate of 0.400λ, and *Protiusspp* have has the lowest growth rate of 0.155λ. At the 56th hour, *Enterobacter cloacae* has the lowest death rate of 0.550λ and *Protius spp* has the highest death rate of 0.040λ

Fig 27: Shows the growth dynamic and killing time of bacteria isolates and the addition of NICE hand sanitizer at the 56th hour using an ultraviolet spectrophotometer. In this table, it was observed that at 0 hours, *Aeromonas hydrophilia* has the highest growth rate of 0.431λ, and *Haemophilia alvei* have the lowest growth rate of 0.135λ. At the 56th hour, *Serratia odorifera* has the lowest death rate of 0.550λ and *Haemophilia alvei* have the highest death rate of 0.030λ

Fig 28: Shows the growth dynamic and killing time of bacteria isolates and the addition of CALVARY hand sanitizer at the 56th hour using an ultraviolet spectrophotometer. In this table, it was observed that at 0 hours, *Pseudomonas aeruginosa* has the highest growth rate of 0.354λ and *Aeromonas hydrophilia* has the lowest growth rate of 0.150λ. At the 56th hour, *Enterobacter cloacae* have the lowest death rate of 0.430λ and *Protius spp* has the highest death rate of 0.030λ

Fig 29: Shows the growth dynamic and killing time of bacteria isolates and the addition of DETTOL hand sanitizer at the 56th hour using an ultraviolet spectrophotometer. In this table, it was observed that at 0 hours, *Protius spp* has the highest growth rate of 0.382λ and *Klebsiella ornithinolytica* has the lowest growth rate of 0.220λ. At the 56th hour, *Enterobacter cloacae* has the lowest death rate of 0.620λ and *Aeromonas hydrophilia* has the highest death rate of 0.180λ

Fig 30: Shows the growth dynamic and killing time of bacteria isolates and the addition of TETMOSOL hand sanitizer at the 56th hour using an ultraviolet spectrophotometer. In this table, it was observed that at 0 hour, *Aeromonas hydrophilia* has the highest growth rate of 0.512λ and *Protius spp* has the lowest growth rate of 0.067λ. At the 56th hour, *Klebsiella ornithinolytica* has the lowest death rate of 0.360λ and *Protius spp* have the highest death rate of -0.005λ

Fig 31: Shows the growth dynamic and killing time of bacteria isolates and the addition of PASSION hand sanitizer at the 56th hour using an ultraviolet spectrophotometer. In this table, it was observed that at 0 hour, *Pseudomonas aeruginosa* has the highest growth rate of 0.400λ and *Serratia odorifera* has the lowest growth rate of 0.200λ. At the 56th hour, *Aeromonas hydrophilia* has the lowest death rate of 0.550λ and *Haemophilia alvei* have the highest death rate of 0.120λ.

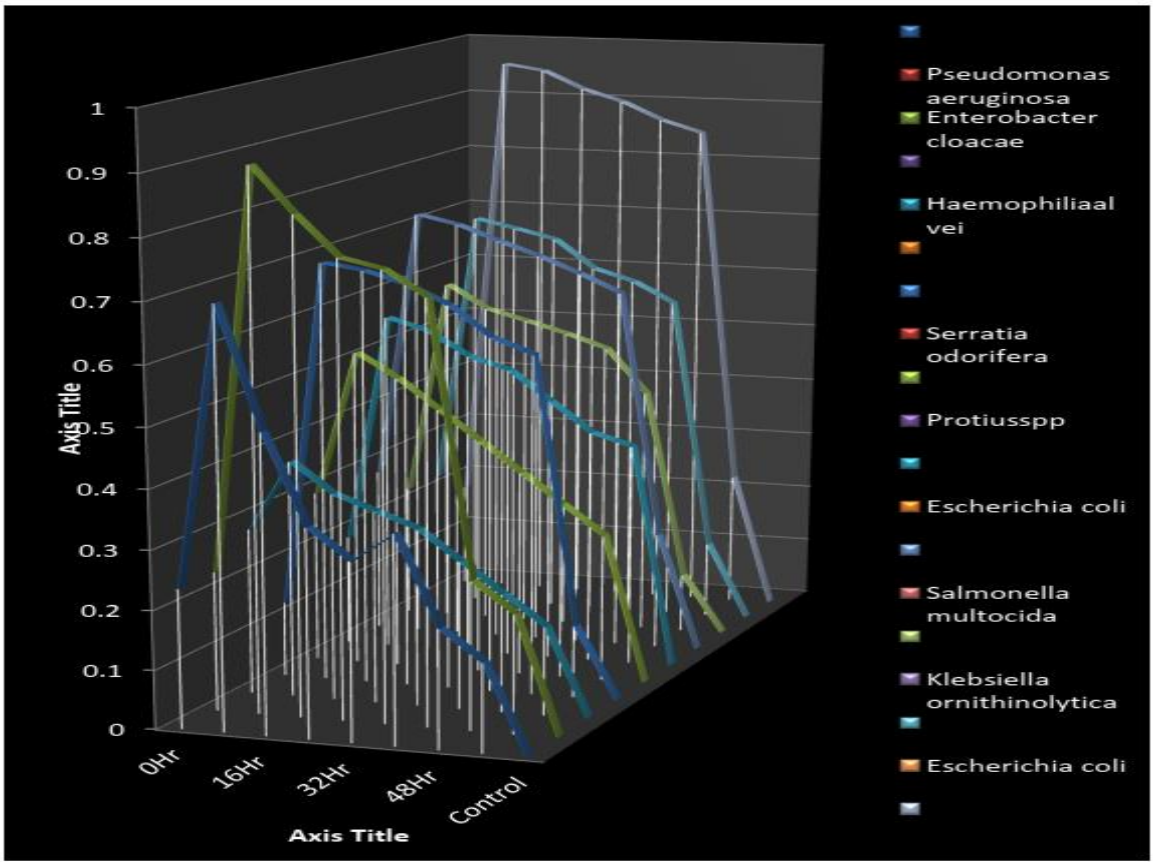

Fig 21: Growth Dynamic of Bacterial Isolates Using Ultraviolet Spectrophotometer with Wavelength 620λ

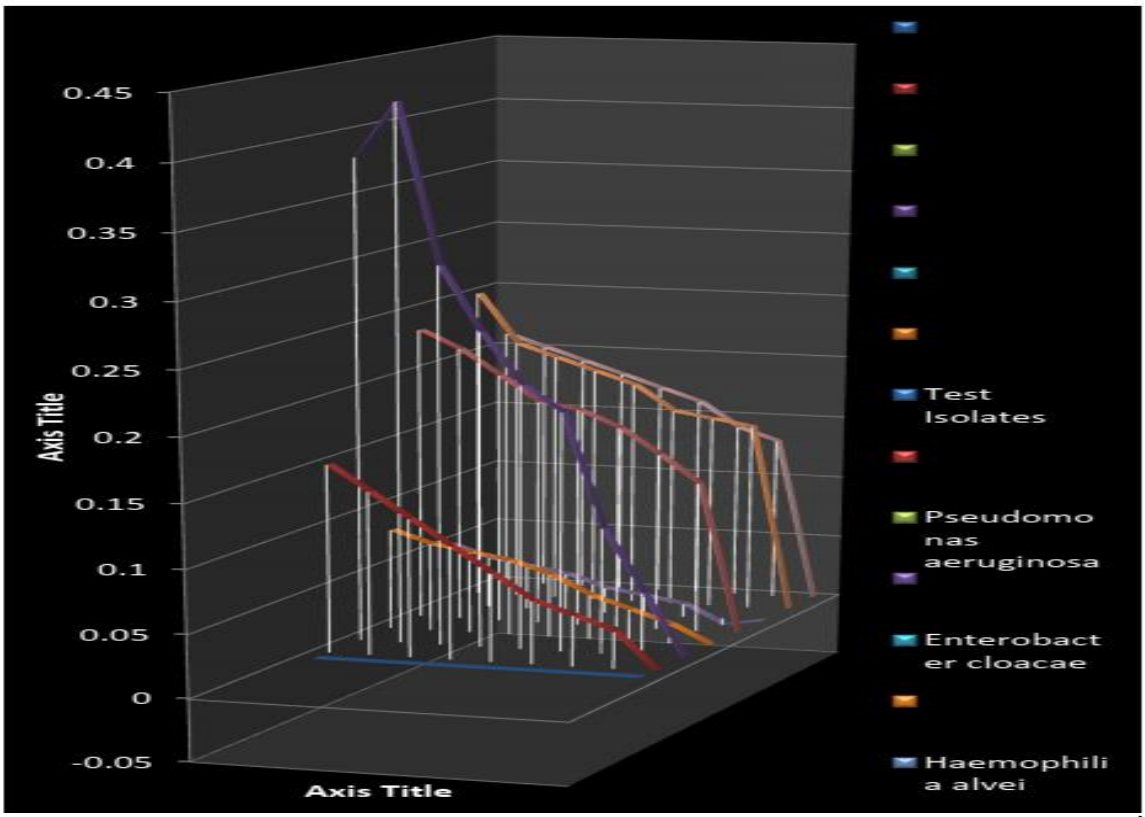

Fig 22: Growth Dynamic and killing of Bacterial Isolates with addition of Corysan hand sanitizer at the 56th using Ultraviolet Spectrophotometer with Wavelength 620λ

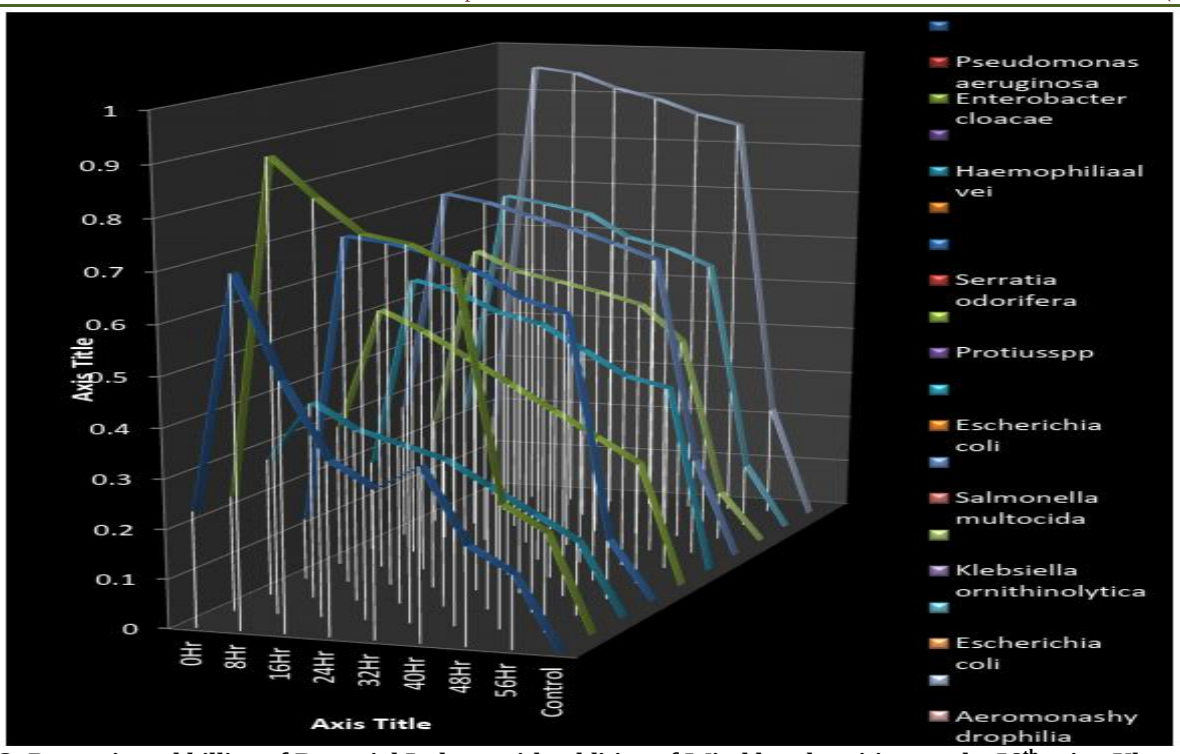

Fig 23: Dynamic and killing of Bacterial Isolates with addition of Wind hand sanitizer at the 56th using Ultraviolet Spectrophotometer with Wavelength 620λ

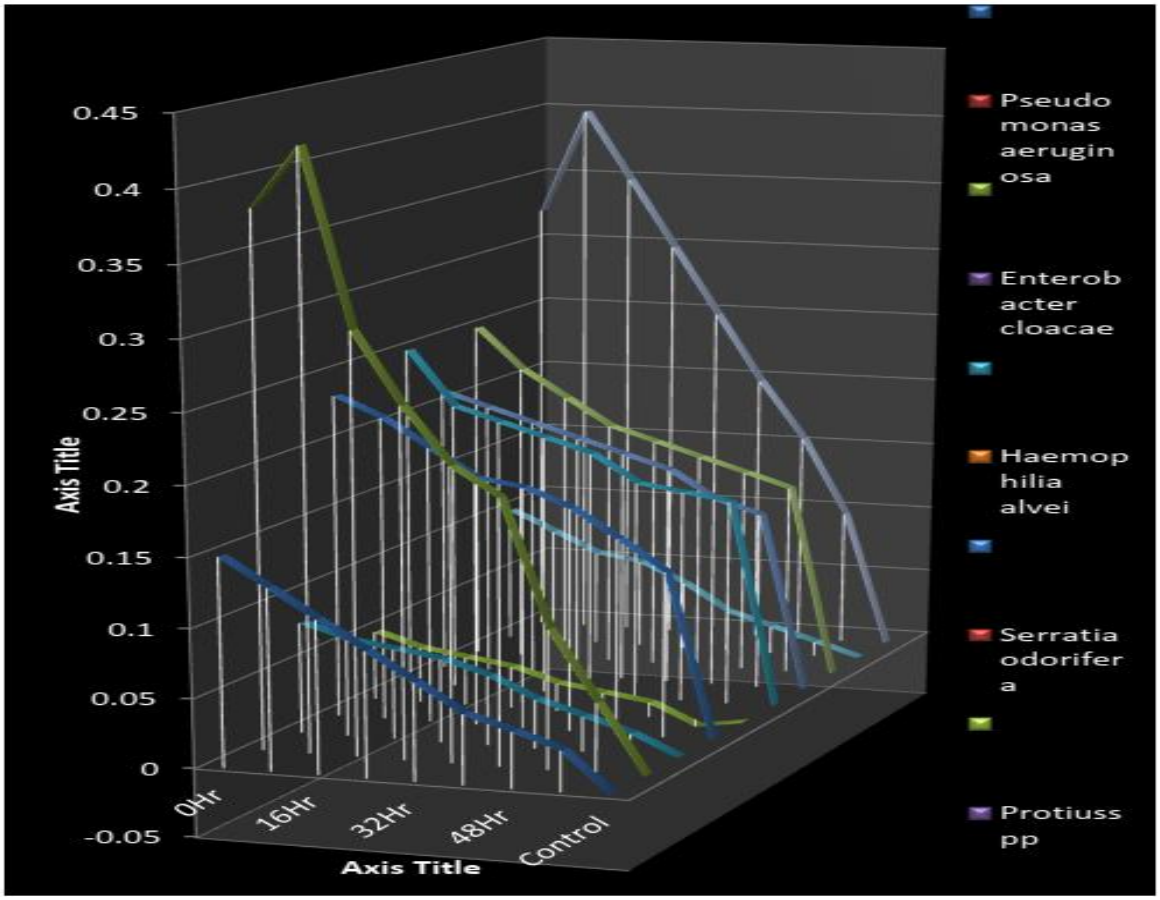

Fig 24: Growth Dynamic and killing of Bacterial Isolates with addition of Cussonhand sanitizer at the 56thhour using Ultraviolet Spectrophotometer with Wavelength 620λ

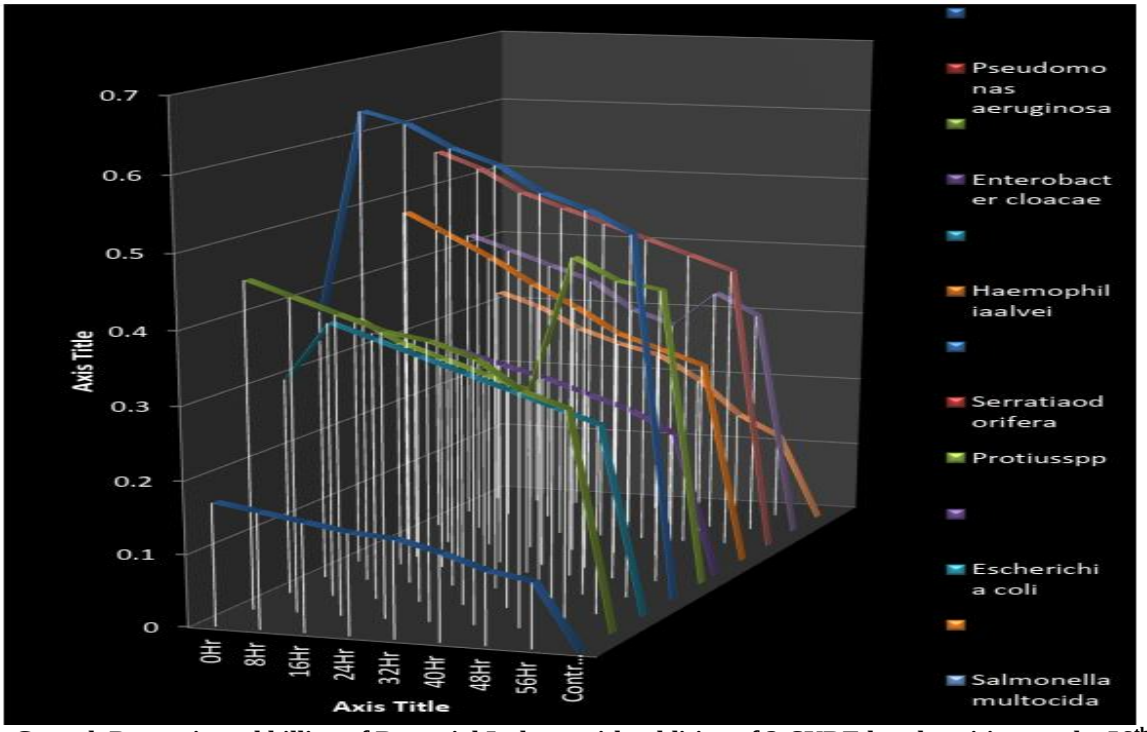

Fig 25: Growth Dynamic and killing of Bacterial Isolates with addition of 2-SURE hand sanitizer at the 56th hour using Ultraviolet Spectrophotometer with Wavelength 620λ

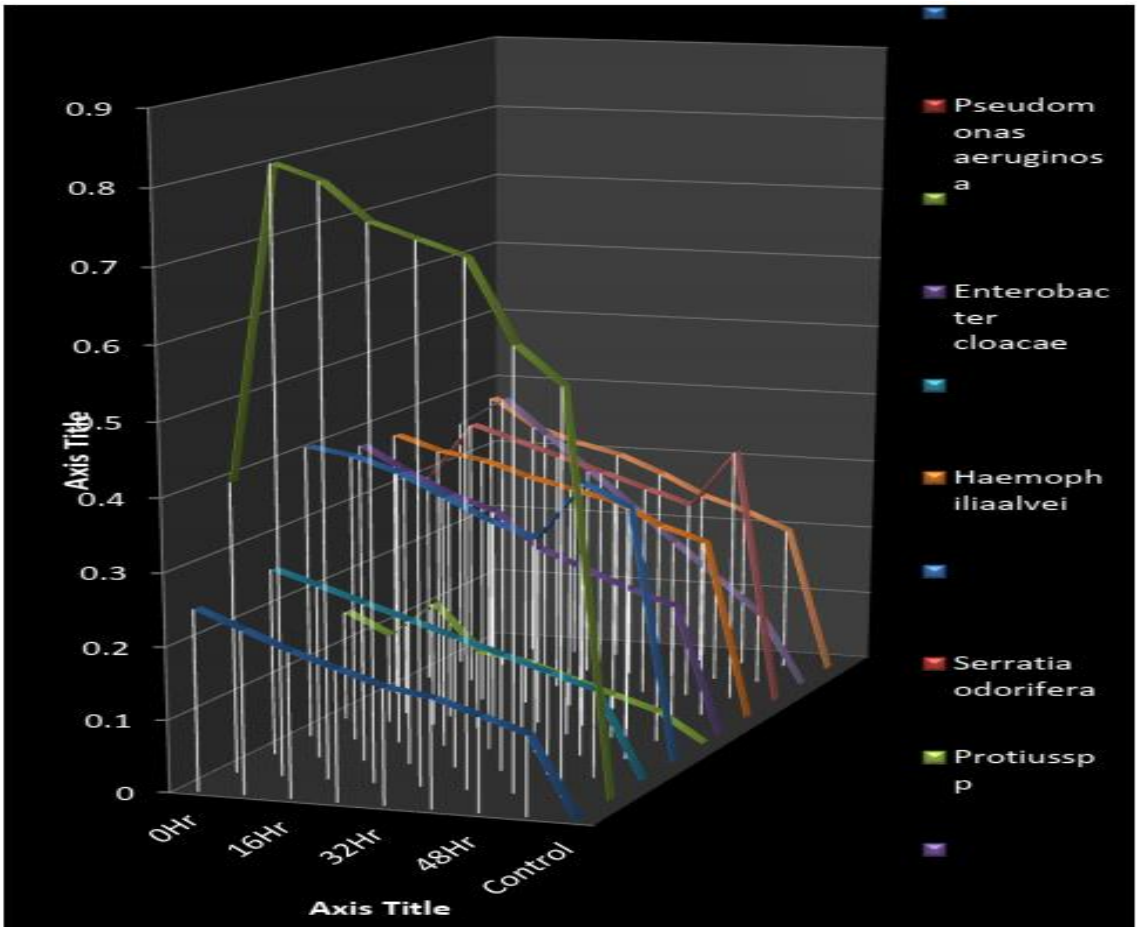

Fig 26: Growth Dynamic and killing of Bacterial Isolates with addition of SANGEL hand sanitizer at the 56th hour using Ultraviolet Spectrophotometer with Wavelength 620λ

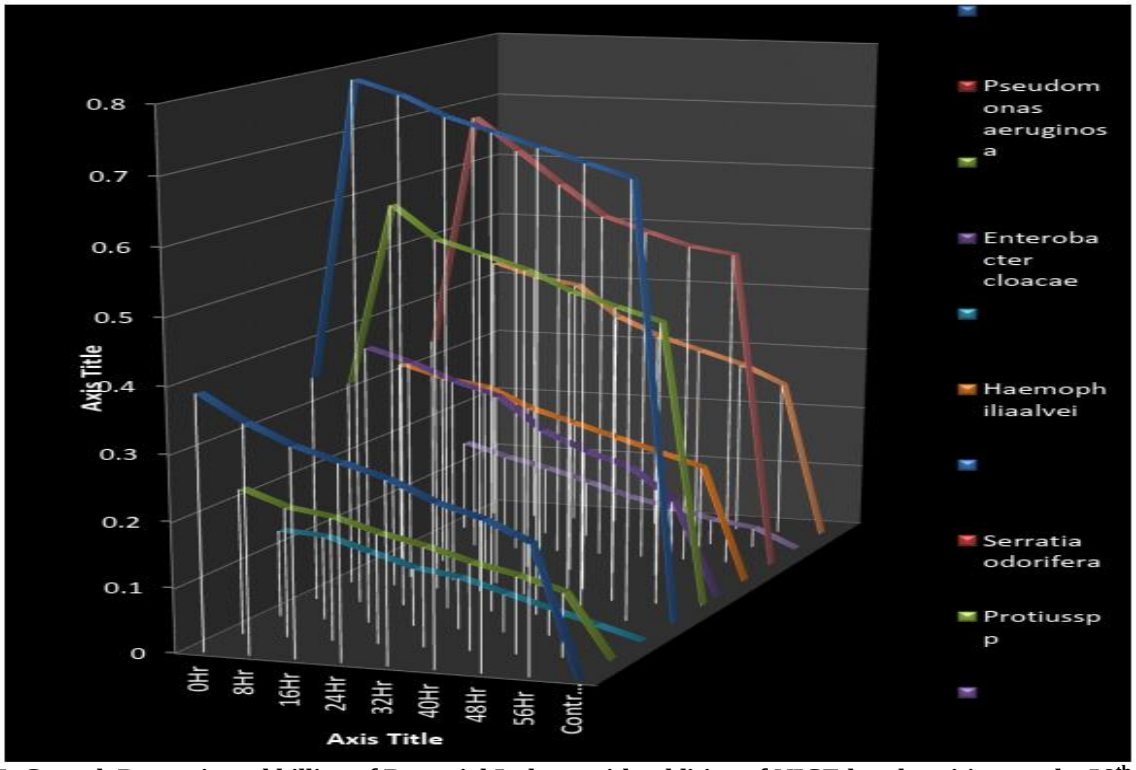

Fig 27: Growth Dynamic and killing of Bacterial Isolates with addition of NICE hand sanitizer at the 56th hour using Ultraviolet Spectrophotometer with Wavelength 620λ

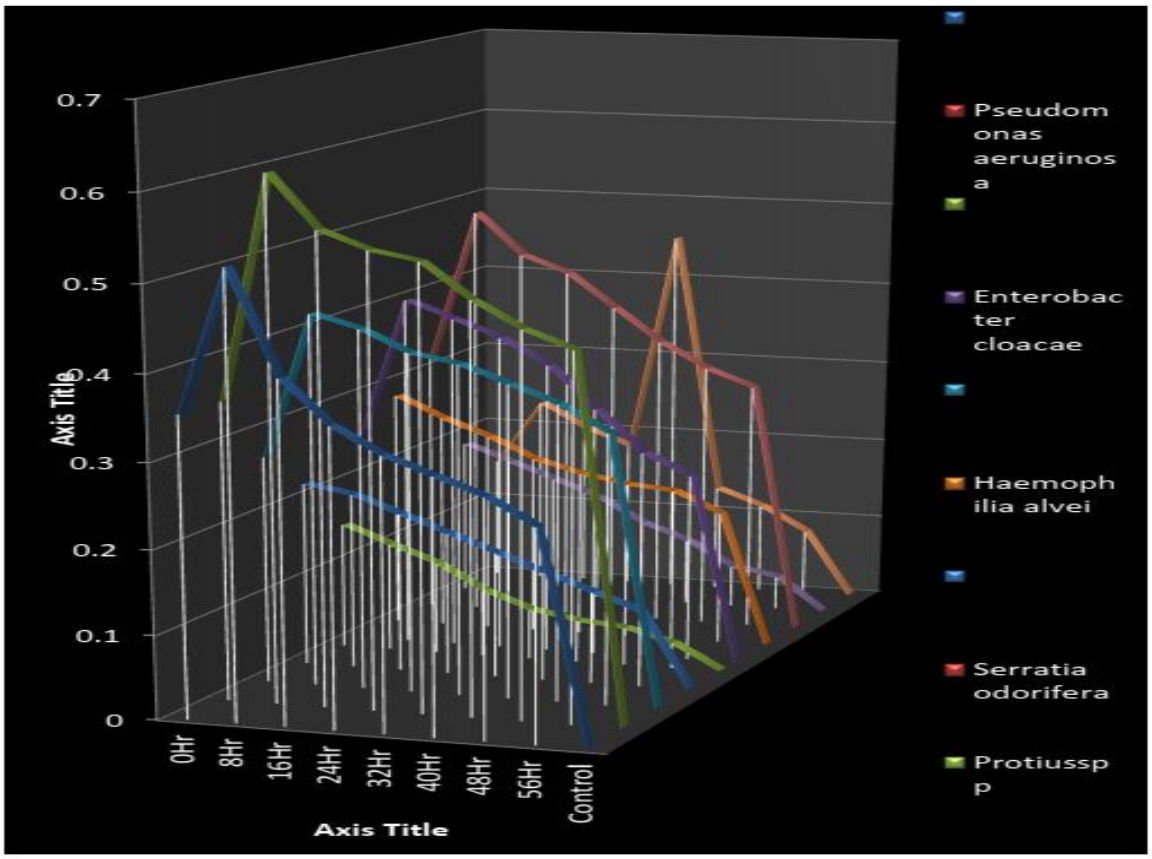

Fig 28: Growth Dynamic and killing of Bacterial Isolates with addition of CALVARY ROYALhand sanitizer at the 56th hour using Ultraviolet Spectrophotometer with Wavelength 620λ

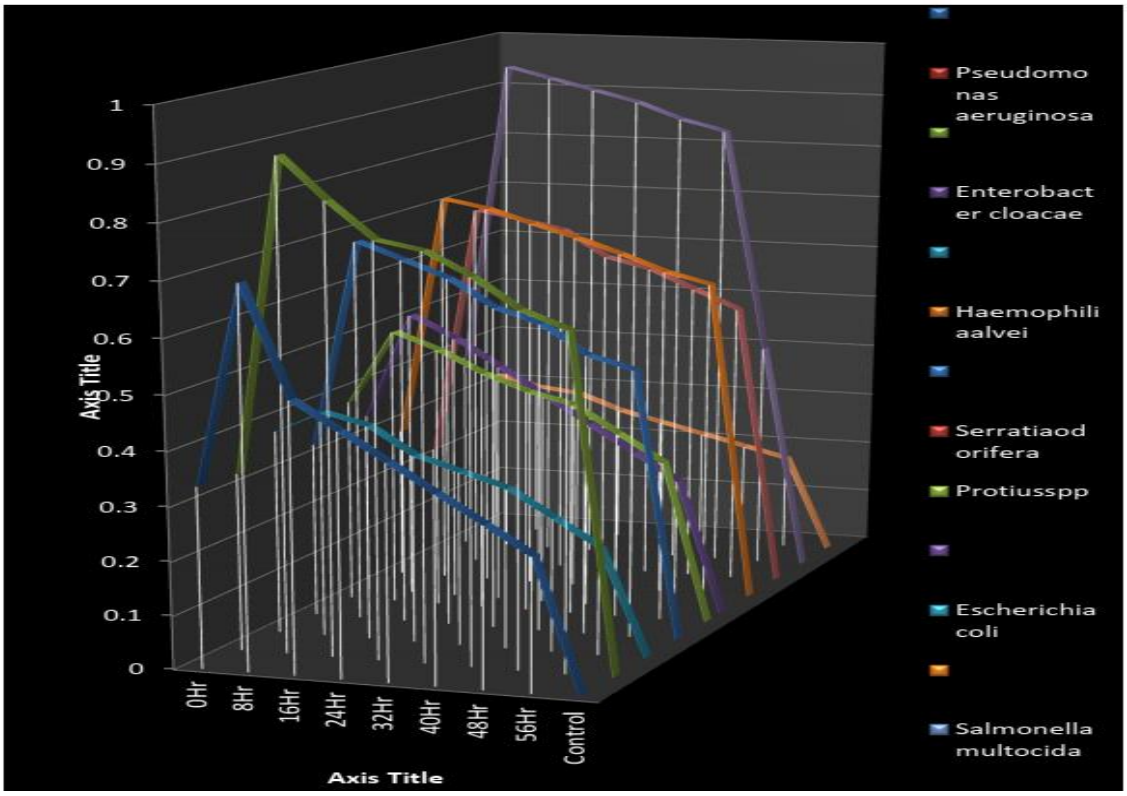

Fig 29: Growth Dynamic and killing of Bacterial Isolates with addition of DETTOL hand sanitizer at the 56th hour using Ultraviolet Spectrophotometer with Wavelength 620λ

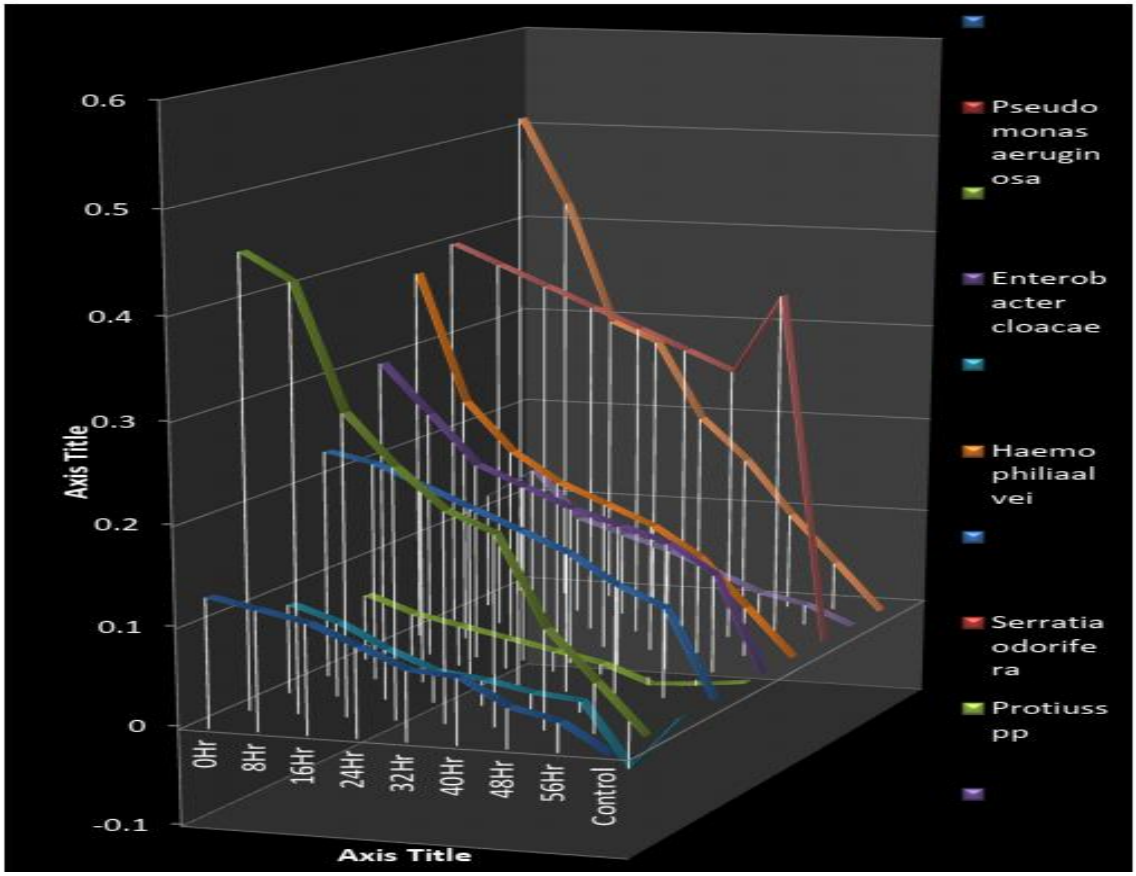

Fig 30: Growth Dynamic and killing of Bacterial Isolates with addition of TETMOSOL hand sanitizer at the 56th hour using Ultraviolet Spectrophotometer with Wavelength 620λ

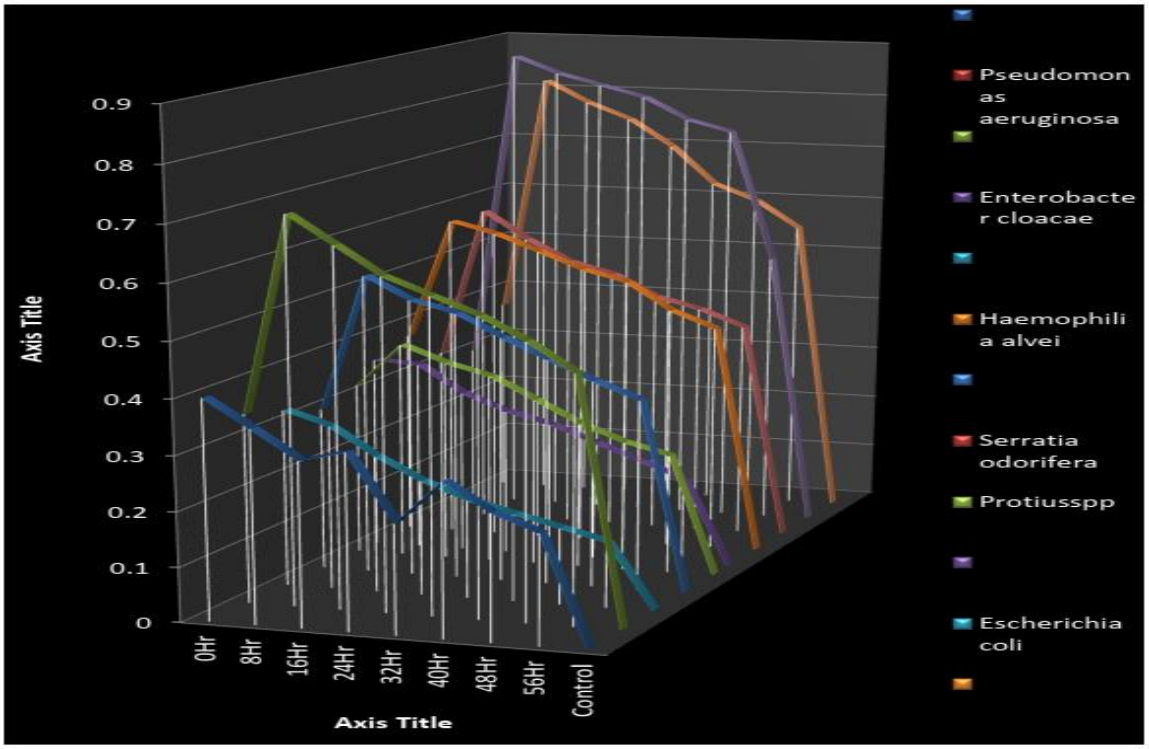

Fig 31: Growth Dynamic and killing of Bacterial Isolates with addition of PASSION hand sanitizer at the 56th hour using Ultraviolet Spectrophotometer with Wavelength 620λ

DISCUSSION

The purpose of this study is to assess the effectiveness/efficacy degree of locally made hand sanitizer against Gram-negative bacteria (GNB) using ultraviolet-visible spectroscopy signatures. Microorganisms are everywhere, and the basic reason they are called ubiquities. They can get onto our hands and items we touch during our daily activities and be infected by different microbes Cleaning our hands with soap and water or hand sanitizer is a key to perfect health and well-being. Hand sanitizer contains at least 60% alcohol and alcohol is one of the most important solvents used in disinfectants, to reduce the degree of antimicrobial proliferation and activity. but there is an unhearing shortcoming in the production of this locally-made hand sanitizer. There is research ongoing comparing the local and foreign-made hand sanitizers to compare their antimicrobial spectrum, this locally made hand sanitizer, its antimicrobial spectrum is less than 30% efficacious and 35% effective under the scope of this research work comparing the table obtained with standard used antibiotics.

Hand sanitizer acts by killing certain microbes on the skin. Although alcohol-based hand sanitizers can quickly reduce the number of germs in many situations, they should be used in the right situations. But the local way the populace uses this hand sanitizer may not reduce the microbial load it even enhance the growth of the microbe.

There are various microorganisms that are present in our body as well as the environment. Some may be harmful, and while some may be harmless, but the body harbors various degree microorganisms, which play an important role in maintaining our physical well-being. Some of the microorganisms which colonizes in our hands may lead to various infections and diseases., to prevent such infections and diseases various measures are necessary like hand washing and the use of hand sanitizers, this hand washing is recommended by W.H.O, to reduce the microbial load in our hand and even in our food before consumption, especially for the African world that prefers to use their bear hands to eat before the advent of foreign eating cutlery. Hand washing become veritable tools to reduce the microbial load in our human system

The use of hand sanitizers has recently gained popularity due to the outbreak of coronavirus in 2020. Due to the outbreak, people are aware of the spread of diseases and their consequences. hand sanitizers as an easy and convenient measure for preventing various diseases. But the efficacy and effectiveness of the locally available hand sanitizers is still unknown and raises some question that is left unanswered.

In this study, the assessment of the effectiveness/efficacy degree of locally made hand sanitizer against Gram-negative bacteria (GNB) using ultraviolet-visible spectroscopy signatures. The ten(10) locally made hand sanitizers examined include; Corysan, Tetmosol, 2-Sure, Sangel, Nice, Calvary

Royal, Cussons Carex, Dettol, Demia, and Passion, which are mainly alcohol-based hand sanitizers.

Alcohol-based hand sanitizers; the main active ingredient is alcohol. Alcohol is a natural antibacterial agent with a broad spectrum of antimicrobial potency on both Gram-positive and Negative bacteria. Some fungi were also affected but not viruses. The modus operandi of alcohol-based hand sanitizers is to exert antibacterial activity by causing protein denaturation, disruption of tissue membranes, and dissolution of several lipids in the cell wall of bacteria (Kar, 2008). Gram-negative bacteria are associated with diverse pathogenic/ infectious diseases most especially antimicrobial-resistant Gram-negative bacteria. But it was a surprising fact that some of the locally-made hand sanitizer has poor microbial activity against Gram-negative bacteria, it suggest that the potency of the so-called locally-made hand sanitizer lack merit and their production capability should be investigated

Traditionally, agar diffusion and agar dilution method are commonly employed for the assessment of the antimicrobial activity of both plant and clinical samples. The advantages of the agar disk diffusion method deplete that the chemical properties of the hand sanitizer remain unchanged, an easy and less technique-sensitive method (Aravind, 2006; Pumarola, 1992). In this study, agar well diffusion and dilution method was employed to determine the efficacy of the selected hand sanitizer.

All ten hand sanitizer products exhibited inhibitory activity against the test isolates, with zones of inhibition ranging from 6 mm to 40 mm at concentrations of 100%. This inhibitory activity varied with product concentration. A general reduction in inhibitory activity was associated with a reduction in product concentration, and inhibition was still observed at concentrations as low as 25%, in some cases compared to the known standard antibiotics.

The result of this study revealed that Curyson hand sanitizer products showed bactericidal activity against all selected test organisms, with MBC values of 100% and MBC values of 50% and 100%. Generally, the growth of selected test organisms was decreased by increasing the concentration gradient of the hand sanitizers. This bacteriostatic/bactericidal activity is more probably due to alcohol components of alcohol-based hand sanitizer which are the major active ingredients intended to exert disinfectant activity in bacteria, but due to production error, the concentration of alcohol decreases and an increase in the growth capability of Gram-negative bacterial.

Other sanitizer products used; Tetmosol, 2-Sure, Sangel, Nice, Calvary Royal, Cussons Carex, Dettol, and Passion, in this study, were selective and less active against all of the selected test organisms with a narrow degree of bactericidal activity, this rises doubt in the efficacy and effectiveness of this locally made hand sanitizer. We can probably say that they are a false sense of protection against all forms of both bacteria and viruses (Osuntokun 2020). The lack of bactericidal activity and more or less no effective inhibition zone observed in this products could be due to the relative decrement of the concentration compared with others since the efficacy of alcohol-based hand sanitizers is affected by several factors such as the type of alcohol used, the concentration of alcohol or amount of alcohol used, the possible contact time and absence of active ingredient in product (hydrogen peroxide) which may limit the bacteriocidal effect of the alcohol from attainment the bacterial cells. This variable degree of activity of hand sanitizers in the market, have previously been widely reported by some researcher, analyzing the efficacy of various hand sanitizing products, it was noted that one of their products was only effective against 6.5% of the isolates tested (Sharif and Ansari 2015).

A more recent study carried out in Kenya (Ochwoto *et al.,* 2017) noted that 25% of tested products were effective against only 33% of the test isolates. Similar to a previous report (Odebisi-Omokanye *et al.,* 2015), this study noted a lower level of susceptibility to all the tested products in Gram-negative organisms tested. In general, however, the results of this study show lower effectiveness of tested products with many mild zones of inhibition than previously published (Oke *et al*., 2013, Odebisi-Omokanye *et al.,* 2015). And unlike both studies which reported a total lack of bacteriocidal activity possibly due to improper storage, 70% of hand sanitizing products in this study exhibited lower bacteriocidal activity.

In conclusion, results obtained from this study show that the products assayed have a mild efficacy than other previously studied products in Nigeria, not all products tested were active against all the test organisms using the dilution method and UV spectrophotometry signatures. To reduce the scourge of adulterated product that lacks poor quality control, more stringent checks of products introduced into the Nigerian market is necessary to ensure that they meet set international standards both in the composition of inhibitory substance this will ensure uniformity in the bacteriological activity of hand sanitizers against human pathogens.

Therefore, it is pertinent to recommend further work on this said topic, especially to the Nigerian populace and Africa as a whole. Hand sanitizers that are prepared in the manufacturing company should be tested after preparation to maintain the proper efficacy of the hand sanitizers which are claimed by the company. Only hand sanitizers which show proper efficacy of hand sanitizers should be sold in the market to protect consumers from buying poor-quality hand sanitizers.

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