

Determination of the Lethal Dose of Ultraviolet Rays on The Growth of Some Human on Pathogenic Bacteria

Russul Arkan Hassan¹, Sajjad Hamid Khazaal¹

¹ Department of Medical Laboratory Techniques, College of Medical Techniques, Uruk University, Baghdad, Iraq
Email: russularkan.1995@gmail.com

Abstract

Aim: The important time to stop the growth of some types of bacteria that cause human diseases by using UV- light (500nm), which reduces the effort and danger to health care workers. **Background:** In this study, we included the effect of ultraviolet UV- light (500nm) on some types of bacteria such as Staphylococcus aureus, Klebsiella pneumonia, Acinetobacteria that cause human diseases. Staphylococcus aureus is a Gram-positive bacteria and causative agent of wide range of infectious diseases such as skin infections, bacteremia, endocarditis, pneumonia and food poisoning. K. pneumoniae now commonly occurs in healthcare facilities, such as hospitals, and is responsible for a range of serious infections involving the urinary tract, lungs, abdominal cavity, intra-vascular devices, soft tissues surgical sites and causing bacteraemia, Acinetobacter baumannii can cause infections in the blood, urinary tract, and lungs (pneumonia), or in wounds in other parts of the body. It can also "colonize" or live in a patient without causing infections or symptoms, especially in respiratory secretions (sputum) or open wounds. **Method:** The study included 50 samples (swab) of S. aureus taken from people suffering from burns and wounds. And 50 samples (intestinal swab) were taken from patients infected with Acinetobacter and 50 samples (sputum) from patients infected with K. pneumonia. Samples were collected from Al-private International Hospital in Baghdad _ during the period 11/15/2021 to 11/20/2022. All of these samples were .diagnosed by a medical staff using a (fine) device and agricultural media. The samples were preserved with glycerin to be preserved for a longer _ period and used for conducting experiments, and at the same time the samples .were used as a (control).From the same tube containing the sample, a percentage (0.5) was taken) _ to conduct experiments, and the rest was used to conduct other experiments _ Using a dose of (UV and shedding it on the growth of some types of pathogenic bacteria). **Result:** After preparing plankton, some types of pathogenic bacteria are: S. aureus, Acinetobacter, K. pneumonia. A working method is prepared for applying UV doses to bacteria at different intervals of time: (10, 30, 60) minutes. It was concluded that time is important to know the time period in which the growth of each of the S. aureus bacteria whose growth stopped by a period (30 minutes, 60 minutes), K. pneumonia, which stopped growing by a period (60 minutes), and the Acinetobacter by a time period (60 minutes) is stopped. **Conclusion:** Time periods were determined to stop the growth of each of the bacteria (Staphylococcus aureus - Klebsiella pneumoniae - Acinetobacter) after exposure to ultraviolet rays with a wavelength of (500nm). As the S. aureus bacteria stop its growth for a period (30-60 min), the - Klebsiella pneumoniae bacteria stop its growth for a period of time (60 min), and the Acinetobacter) bacteria stop its growth for a period of time (60 min).

Keywords: Ultraviolet rays, Staphylococcus aureus, Klebsiella. pneumonia, Acinetobacter.

1. Introduction

Staphylococcus aureus is a Gram-positive round-shaped bacterium, a member of the Firmicutes, and is a usual member of the microbiota of the body, frequently found in the upper respiratory tract and on the skin. It is often positive for catalase and nitrate reduction and is a facultative anaerobe that can grow without the need for oxygen. Klebsiella is a type of Gram-negative bacteria that can cause different types of healthcare-associated infections, including pneumonia, bloodstream infections, wound or surgical site infections, and meningitis. Increasingly, Klebsiella bacteria have developed antimicrobial resistance, most recently to the class of antibiotics known as carbapenems. Acinetobacter spp. are

Gram-negative coccoid rods that are sometimes difficult to stain. They grow aerobically and are oxidase negative using Kovac's reagent, O-F negative, nonmotile in hanging drop preparations, catalase positive, mostly nitrate negative, and mostly positive for Tween hydrolysis. DNA from an isolate that is Acinetobacter will transform auxotrophic Acinetobacter test strains to prototrophy and so confirm the isolate as Acinetobacter. **Aim of study:** The important time to stop the growth of some types of bacteria that cause human diseases by using UV-light (), which reduces the effort and danger to health care workers.

2. Material and Method

Collection of specimens: The study included 50

samples (swab) of *Staphylococcus aureus* taken from people suffering from burns and wounds. And 50 samples (intestinal swab) were taken from patients infected with *Acinetobacter*. And 50 samples (sputum) from patients infected with *Klebsiella pneumoniae*. Samples were collected from Al-Ahli International Hospital in Baghdad during the period 11/15/2021 to 11/20/2022. All these samples were diagnosed by a medical staff using a (fine) device and agricultural media the samples were preserved with glycerin to be preserved for a longer period and used for conducting experiments, and at the same time the samples were used as a (control). From the same tube containing the sample, a percentage (0.5) was taken to conduct experiments, and the rest was used to conduct other experiments. Using a dose of (UV and shedding it on the growth of some types of pathogenic bacteria). After preparing plankton, some types of pathogenic bacteria are: 1) *Staphylococcus aureus* 2) *Acinetobacter* 3) *Klebsiella pneumoniae*. A working method is prepared for applying UV doses to bacteria at different intervals of time: (10, 30, 60) minutes.

Procedure: Using a dose of ultraviolet rays and applying them to the growth of some types of pathogenic bacteria (*Staphylococcus aureus*, *Klebsiella pneumoniae*, *Acinetobacter*). Before starting work, we prepare the culture media. Method for preparing the culture media: 1-Suspend 28.0 grams in 1 liter of purified/distilled or deionized water. 2-Heat to a boil to completely dissolve the medium. 3-Sterilize by autoclaving at a pressure of 15 lb. (121°C) for 15 minutes. 4-After autoclaving, cool to 45-50°C. 5-Pour nutritious agar into petri dishes (until the agar is solidified). 6-Keep boxes in the refrigerator at 2-8°C. After preparing of some types of pathogenic bacteria (*Staphylococcus aureus*, *Klebsiella pneumoniae*, *Acinetobacter*) A special method is prepared for exposing doses of ultraviolet rays to bacteria at different period which are as follows (10minutes, 30 minutes,60 minutes).

-*Staphylococcus aureus* (1-3 Tubes) was numbered with different time periods to expose each tube according to the time period (Tube 1, 10 minutes - Tube 2, 30 minutes - Tube 3, 60 minutes). 2) (0.5) ml of the suspended *Staphylococcus aureus* was taken using a pipette and placed in 3 numbered tubes (with different time periods). After that, (Tube1) was placed in a hood for the purpose of exposing it to UV rays of wavelength (500nm) for a period of 10 minutes. After that, (Tube2) was placed for a period of 30 minutes in a hood device for the purpose of being exposed to UV rays of a wavelength, and (Tube3) was placed in a hood device for a period of 60 minutes for the purpose of being exposed to UV rays. 4) Bacteria exposed to UV rays were cultured for different periods of time (10, 30, 60) in a nutrient medium.

- *Klebsiella pneumoniae* (1-3 Tubes) were numbered with different time periods to expose each tube according to the time period (Tube 1, 10 minutes -

Tube 2, 30 minutes - Tube 3,60 minutes). 2) (0.5) ml of the suspended *Klebsiella pneumoniae* was taken using a pipette and placed in 3 numbered tubes (with different time periods). After that, (Tube1) was placed in a hood for the purpose of exposing it to UV rays of wavelength for a period of 10 minutes. After that, (Tube2) was placed for a period of 30 minutes in a hood device for the purpose of being exposed to UV rays of a wavelength (500nm), and (Tube3) was placed in a hood device for a period of 60 minutes for the purpose of being exposed to UV rays. 4) Bacteria exposed to UV rays were cultured for different periods of time (10, 30, 60) in a nutrient medium.

-*Acinetobacter* (1-3 Tubes) were numbered with different time periods to expose each tube according to the time period (Tube 1, 10 minutes - Tube 2, 30 minutes - Tube 3,60 minutes). 2) (0.5) ml of the suspended *Acinetobacter* was taken using a pipette and placed in 3 numbered tubes (with different time periods). After that, (Tube1) was placed in a hood for the purpose of exposing it to UV rays of wavelength for a period of 10 minutes. After that, (Tube2) was placed for a period of 30 minutes in a hood device for the purpose of being exposed to UV rays of a wavelength (500nm), and (Tube3) was placed in a hood device for a period of 60 minutes for the purpose of being exposed to UV rays. 4) Bacteria exposed to UV rays were cultured for different periods of time (10, 30, 60) in a nutrient medium.

3. Result

Determination of the lethal dose of ultraviolet rays on bacteria growth (*Staphylococcus aureus*, *Klebsiella pneumoniae*, *Acinetobacter*) that cause human diseases. After preparing the suspension bacteria that cause human diseases as mentioned in chapter three and exposing them to ultraviolet rays, the suspension exposed to ultraviolet rays is cultivated on culture media (nutrient agar), To note the period during which the growth of bacteria is stopped.

Table (1) shows the growth stop of each of the bacteria (*S. aureus*, *Acinetobacter*, *K. pneumoniae*) for different periods of time after exposure to ultraviolet rays at a wavelength (500nm)

Growth time	<i>S. aureus</i>	<i>Acinetobacter</i>	<i>K. pneumoniae</i>
	picture (1)	picture (2)	picture (3)
0	50	50	50
10 min	15	30	50
30min	0	2	25
60min	0	0	0
Growth	<i>S. aureus</i>	<i>Acinetobacter</i>	<i>K. pneumoniae</i>

1- After exposing the suspension to ultraviolet rays at intervals of (10 minutes, 30 minutes, 60 minutes) and cultivating it on culture media as explained in the third chapter, it was observed that the bacteria *S. aureus* stopped its growth in periods of time (30 minutes, 60 minutes) Table (1) illustrates this. The lethal dose of

bacteria was determined *S. aureus* at intervals (60 minutes, 30 minutes), as shown in Figure (1)

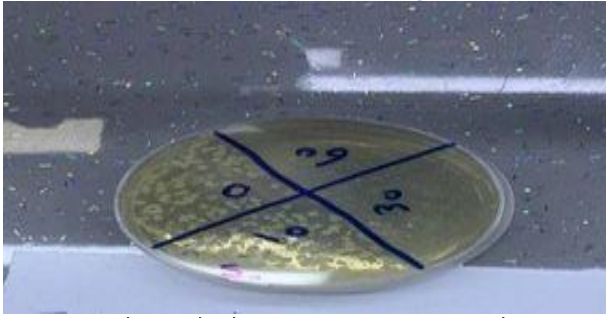


Figure (1) shows the bacteria *S. aureus* growth stopped in period (30 min-60min)

2-After exposing the suspension to ultraviolet rays at intervals of (10 minutes, 30 minutes, 60 minutes) and cultivating it on culture media as explained in the third chapter, it was observed that the bacteria *Acinetobacter* stopped its growth in periods of time (60 minutes) Table (1) illustrates this. the lethal dose of bacteria was determined *Acinetobacter* at intervals (60 minutes), as shown in Figure (2).



Figure (2) shows the bacteria *Acinetobacter* growth stopped in period (60 min)

3-After exposing the suspension to ultraviolet rays at intervals of (10 minutes, 30 minutes, 60 minutes) and cultivating it on culture media as explained in the third chapter, It was observed that the bacteria *Klebsiella pneumoniae* stopped its growth in periods of time (60 minutes) Table (1) illustrates this. the lethal dose of bacteria was determined *Klebsiella pneumoniae* at intervals (60 minutes), as shown in Figure (3).

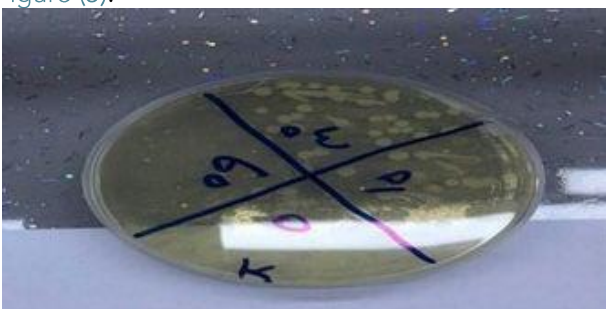


Figure (3) shows the bacteria *Klebsiella pneumoniae* growth stopped in period (60 min)

4. Discussion

The results show that After preparing a suspension of *S. aureus* and exposing it to ultraviolet rays at different periods of time, which are as follows (10,30,60minutes) with a wavelength (500nm), as mentioned in the three-chapter, and cultivating it on

the nutrient media (Nutrient agar) as shown in the figure(1), the results were that the *S. aureus* bacteria stopped its growth in a period of time (30,60) minutes, and this study agrees with the study Hardjawinata, K., Setiawati, R., and et al. (2005).[12] After preparing a suspension of *Acinetobacter* and exposing it to ultraviolet rays at different periods of time, which are as follows (10,30,60minutes) with a wavelength (500nm), as mentioned in the three chapter, and cultivating it on the nutrient media (Nutrient agar) as shown in the image (2), the results were that the *Acinetobacter* stopped its growth in a period of time (60 minutes) minutes , and this study agrees with the study Dai, T., Murray, C. K., Vrahas, M. S.,et al . (2012). Hare, J. M., Bradley, et al (2012).[14]

After preparing a suspension of *K. pneumoniae* and exposing it to ultraviolet rays at different periods of time, which are as follows (10,30,60minutes) with a wavelength (500nm), as mentioned in the three chapter, and cultivating it on the nutrient media (Nutrient agar) as shown in figure (3), the results were that the *K. pneumoniae* bacteria stopped its growth in a period (60) minutes, and this study agrees with the study Behzadi, E., Behzadi, P., et al. (2014).[13]

It was concluded that time is important to know the time period in which the growth of each of the *S. aureus* bacteria whose growth stopped by a period (60 minutes, 30 minutes), *K. pneumoniae*, which stopped growing by a period (60 minutes), and the *Acinetobacter* by a time period (60 minutes) is stopped.[15]

5. Conclusion

Time periods were determined to stop the growth of each of the bacteria (*Staphylococcus aureus* - *Klebsiella pneumoniae* - *Acinetobacter*) after exposure to ultraviolet rays with a wavelength of (500nm). As the *S. aureus* bacteria stop its growth for a period (30-60 min), the - *Klebsiella pneumoniae* bacteria stop its growth for a period (60 min), and the *Acinetobacter* bacteria stop its growth for a period (60 min).

References

- 1- Shon AS, Bajwa RP, Russo TA. Hypervirulent (hypermucoviscous) *Klebsiella pneumoniae* a new and dangerous breed. *Virulence* 2013; 4:1–12.
- 2- Kocsis B, Szabó D. Antibiotic resistance mechanisms in *Enterobacteriaceae*. In: A Méndez-Vilas, ed. *Microbial pathogens and strategies for combating them: science, technology and education*. Spain: Formatex Research Center; 2013. pp. 251–257.
- 3- Euzéby JP. List of Bacterial Names with Standing in Nomenclature: a folder available on the Internet. *Int J Syst Bacteriol*1997; 47:590–2.
- 4- Brisse S, Verhoef J. Phylogenetic diversity of *Klebsiella pneumoniae* and *Klebsiella oxytoca* clinical isolates revealed by randomly amplified polymorphic DNA, *gyrA* and *parC* genes sequencing

and automated ribotyping. *Int J Syst Evolut Microbiol* 2001; 51:915–24.

5- Podschun R, Ullmann U. *Klebsiella* spp. as nosocomial pathogens: epidemiology, taxonomy, typing methods and pathogenicity factors. *Clinic Microbiol Rev* 1998; 11:589–603.

6- Source: 2019 AR Ttreats Report CDC: Centers for Disease Control and Prevention Report.

7- Greenwood D, O'Grady F. Scanning electron microscopy of *Staphylococcus aureus* exposed to some common anti-staphylococcal agents. *J Gen Microbiol.* 1972; 70:263–270. DOI: 10.1099/00221287-70-2-263.

8- Foster T. Chapter 12: *Staphylococcus*. *Medical Microbiology*. 4th edition. Galveston (TX): University of Texas Medical Branch at Galveston, Galveston, Texas; 1996.

9- Blair JE. Factors determining the pathogenicity of staphylococci. *Annu Rev Microbiol.* 1958; 12:491–506. DOI: 10.1146/annurev.mi.12.100158.002423

10- Brown DF, Edwards DI, Hawkey PM, Morrison D, Ridgway GL, Towner KJ, Wren MW; Joint Working Party of the British Society for Antimicrobial Chemotherapy; Hospital Infection Society; Infection Control Nurses Association. Guidelines for the laboratory diagnosis and susceptibility testing of *ursingii*. *Microbiology*, 158(Pt 3), 601.

methicillin-resistant *Staphylococcus aureus* (MRSA). *J Antimicrob Chemother.* 2005; 56:1000–1018. DOI: 10.1093/jac/dki372

11- Archer GL. *Staphylococcus aureus*: a well-armed pathogen. *Clin Infect Dis.* 1998; 26: 1179–1181. PMID: 9597249.

12- Hardjainata, K., Setiawati, R., & Dewi, W. (2005). Bactericidal efficacy of ultraviolet irradiation on *Staphylococcus aureus*. *Asian Journal of Oral and Maxillofacial Surgery*, 17(3), 157-161.

13- Behzadi, E., Behzadi, P., & Ranjbar, R. (2014). In vitro apoptotic activity of UVB light in *Klebsiella Pneumoniae*. *Alban Med J*, 2, 18-22.14 - Dai, T., Murray, C. K., Vrahas, M. S., Baer, D. G., Tegos, G. P., & Hamblin, M. R. (2012). Ultraviolet C light for *Acinetobacter baumannii* wound infections in mice: potential use for battlefield wound decontamination? *The journal of trauma and acute care surgery*, 73(3), 661.

15- Hare, J. M., Bradley, J. A., Lin, C. L., & Elam, T. J. (2012). Diverse responses to UV light exposure in *Acinetobacter* include the capacity for DNA damage-induced mutagenesis in the opportunistic pathogens *Acinetobacter baumannii* and *Acinetobacter*