The Effect of Various Ultra High Frequency Radiation on the Evolution of the Staphylococcus aureus

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Abstract—The World Health Organization (WHO) has declared the outbreak of Coronavirus disease 19 (COVID-19) on March 2020. Severe acute respiratory syndrome-coronavirus 2 (SARS-CoV2) is the cause of the COVID-19 disease. Staphylococcus aureus, like SARS-CoV2, is a harmful microorganism. Once the skin is breached, S. aureus can cause infections like pneumonia The coinfections between these two and skin infections. microorganisms present high risk on human health. This study deals with investigating the effect of electromagnetic radiation on Staphylococcus aureus. It focuses on exposing the S. aureus bacteria samples to different Ultra High Frequencies (UHF). The number of bacterial colonies that grew from the exposed sample and unexposed samples were assessed. The average bacterial growth was 90.51%. This means that some of the bacteria did not survive after exposure to radiation. This result showed that some of the UHFs had affected the growth of Staphylococcus aureus. The correlation between the frequency and the growth of the bacteria was also calculated by means of computing the Pearson Correlation coefficient. The result is 0.4344 which signifies a small positive linear relation between the two parameters mentioned. These results can be preliminary stage on exploring other possible application of electromagnetic radiation in prevention of diseases.

Keywords— Ultra High Frequency, Electromagnetic radiation, Staphylococcus aureus bacteria

I. INTRODUCTION

In 2019, the spread of the virus called Severe acute respiratory syndrome-coronavirus 2 (SARS-CoV2) had been very prominent worldwide that it was declared by the World Health Organization (WHO) as a pandemic [1]. There were about 170,000 confirmed cases of coronavirus disease. It included 7,000 deaths in almost 150 countries [2]. Staphylococcus aureus is also a harmful microorganism. It causes pneumonia [3]. The risks of coinfection between these microorganisms post high risk on human health [4]. WHO recommended the community to maintain social distancing, avoid congested places, wear masks, avoid touching the eyes, nose and mouth and thoroughly disinfect and rinse the hands [5]. All of these precautions aim to reduce the chances of being infected or spreading the virus. Some studies explored

on electronic means. The study of Ali et al. focused on experimenting THz frequencies to perceive the existence of bacteria and high power electromagnetic wave can be used to make some bacteria indolent without terminating others after their discovery [6].

In relation to the study mentioned, it can be seen that electromagnetic waves can affect the growth of a microorganism. And that it can further be exploited in different frequency ranges. The researcher intends to explore on the Ultra High Frequency (UHF) range and its effect on the bacteria, Staphylococcus aureus.

The general objective of this research is to evaluate the effect of electromagnetic radiation on the growth of Staphylococcus aureus. Specifically, the objectives are as follows: 1) To design a machine that radiates Ultra High Frequency, 2) To expose Staphylococcus aureus to Ultra High Frequency radiation, 3) To identify the number of bacterial colonies in the experimental sample and control sample, and 4) To evaluate the relation between the frequency and bacterial growth.

Staphylococci are common bacteria that thrive on skin. Staphylococcus is one of its types. Once it breached the skin, it is capable of causing infections like pneumonia, dermatitis, boils and other skin infections. Investigating the effect of UHF radiation on the evolution of S. aureus can be a preliminary stage on further studies of alternative solutions in removing harmful microorganisms and the prevention of diseases that they cause.

This study focuses on using UHF frequencies 750MHz, 760MHz, 770MHz, 780MHz, 870MHz, 885MHz, 890MHz and 900MHz. It investigates the effect of the UHF radiation to Staphylococcus aureus. The study did not use other type of microorganism. The bacteria samples were exposed to each frequency once due to financial constraints.

II. RELATED LITERATURE

A. Staphylococcus aureus and COVID-19

Staphylococcus aureus is a commensal microorganism and a human pathogen that causes community acquired pneumonia [3]. It also causes health care-associated infections like infective endocarditis, prosthetic device infections and skin infections [7]. Coronavirus disease 19 (COVID-19), because of severe acute respiratory syndrome-coronavirus 2 (SARS-CoV2), consists a wide range of clinical management ranging from flu-like condition to organ failure [4].



Fig. 1. Staphylococcus aureus skin and soft tissue infections

A patient had a high risk of mortality due to viral-bacterial (SARS-CoV2 - S.aureus) coinfection and advance cancer status. Viral infection contributes in the spread of bacteria which provides more adhesion sites causing impairment on the response of the immune system. Coinfection due to bacteria is responsible for a worse diagnosis characterized by higher disease severity also influenced by viral and bacterial strains, density of bacterial colonization, and the time between viral infection and bacterial coinfection [4].

B. Ultrahigh frequencies (UHFs)

The Electromagnetic Spectrum Electromagnetic waves are signals which the amplitudes of the electric and magnetic fields vary at a specific rate. The electromagnetic waves differ sinusoidal. The frequency is measured in cycles per second (cps) or hertz (Hz). Ultrahigh frequencies (UHFs) include the 300- to 3000-MHz series. It is used in TV channels 14 to 51, and it is used for land mobile communication, cellular telephones and military communication. Some radar, navigation services and radio amateurs also have bands in this range [8].

C. Effect of Electromagnetic Waves on Saccharomyces cervisiae.

The effect of electromagnetic waves on Saccharomyces cervisiae was investigated in a study. A system was designed to expose Saccharomyces cerevisiae to radiation. The experiment used 1200 kHz and magnetic flux density of 1.57x10^-3 T. Evaluation was conducted be means of comparing visual images after graphical editing between the unexposed and exposed samples. The ratio between the experimental and control sample was 3/4. The result shows inhibit effect after 72 hours exposures [9].

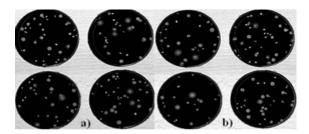


Fig. 2. Reference sample with yeast culture after 72 hours

D. Effect of Mobile Phone to Staphylococcus aureus

Buniyamin et al. used Staphylococcus aureus as their bacterial sample in knowing the effect of electromagnetic radiation from mobile phone. Users are prone to the electromagnetic waves coming from mobile phones. The radiation can cause increase in hotness in sense of hearing and portion of the head. In this research, S. aureus were exposed to mobile phone radiation in different durations for both call and standby mode. S. aureus decreased in numbers when the phone is in on-call mode. Fig. 4 shows the setup where mobile device was located in the middle of two dishes that hold the bacteria. The distance is approximately 1 centimeter [10].



Fig. 3. Exposing the S. aureus Bacteria to the EMF

E. Effects of Low Frequency EMF to Deinococcus radiodurans and Enterococcus faecium

Deinococcus radiodurans and Enterococcus faecium are microorganisms as well. These samples were used to know the effects of Low Frequency EMF. The exposed and unexposed samples were evaluated via bacterial growth. As it is shown in Fig. 5, experimental subjects are in the petri dish and unexposed ones are in a shielded container. The samples were exposed for four hours. Results seen affected the growth after exposure. It was observed that when the concentration is high, the inhibition of cell spread is lesser. In the case of Deinococcus radiodurans, it can be seen that it is unaffected to ionizing radiation. In the case of Enterococcus faecium, LF EMF prevents its spread [11].

F. Electromagnetic radiation affects the Staphylococcus aureus

Samortin et al. have investigated how electromagnetic radiation affects the Staphylococcus aureus. According to their study, upon using the 1GHz frequency, there were 62% of the bacteria that were eliminated [12].

G. ANSYS Simulation of Multi-band compact Microstrip

There are researches that used ANSYS software in simulating their concepts and designs. Kumar et al. used it in designing and simulating multi-band compact microstrip patch. They assessed the measurements of their element and simulated it on ANSYS software. The resulted indicated that antenna can operate in 11.25GigaHertz - 12.05GigaHertz, 13.05GigaHertz - 14.85GigaHertz and 16.7GigaHertz - 17.3GigaHertz [13].

H. Bowtie Antenna

In designing an antenna, good return loss, flat input impedance, and constant radiation pattern are important factors. Conical antennas are known for its wide bandwidth. The bowtie antenna is a variation of cone antenna. A two-dimensional cone is triangle hence the flat version of cone antenna looks like two triangles. Bowtie antennas maintain a constant impedance and gain over a 4:1 range of frequency [8].

I. Matlab

Matlab is mathematical software used in numerical calculation. It has several types of modeling and disciplines. Its use is becoming an emerging technology for many. [14] In terms of numerical calculations, it is one of the best equipment used for mathematical application software. It provides the representation of different datum, operating in matrix, formation of user interface, and algorithm implementation. It is also extensively applied in image processing, engineering calculations, financial modeling designs, communications and signal processing, and control design. [15]

III. METHODOLOGY

A. Conceptual Framework

The research framework provides the process of developing a good quality project. It begins with the input which is the Staphylococcus aureus bacteria. The preparation of the culture of Staphylococcus aureus was done by licensed medical personnel. Next is the designing of the system that will radiate the electromagnetic waves. When the device is properly working, the bacteria are exposed to different Ultra High Frequency electromagnetic radiation. Lastly, the control samples and experimental samples were incubated and the bacterial colonies formed were evaluated.

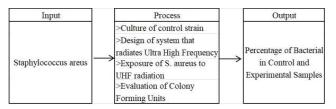


Fig. 4. Conceptual Framework

B. System Block Diagram

The system is consisting of various parts to produce Ultra High Frequency. The Wide Band Synthesizer generates a range of frequencies from the single reference frequency, 25MHz. It has an integrated oscillator which is voltage controlled with a fundamental output frequency ranging from 2.200 GHz to 4.4GHz. The loop filter filters undesired higher frequency which might permit out of the phase detector and act in the VCO tune line. The matching circuit is a mirror LC network needed to match 50 ohm output impedance of the synthesizer. Attenuator reduces the power of a signal without distorting its

waveform. Low frequency cannot be used due to amplifier. To compensate for this, the signal is attenuated. The signal will be decreased in such a way that it will not exceed the input limit. DC Filter is used to produce pure DC from an unfiltered pulsating DC. Power amplifier increases the magnitude of power of a given input signal. It has RF Gain Block with high open loop gain and differential inputs which is used for adaptability; gain, bandwidth and other attributes can be manipulated by feedback through an external circuit. Low Pass Filter permits signals with a frequency lesser than a designated cutoff frequency and weakens signals with frequencies greater than the cutoff frequency. Matching circuit in the last stage is used to match the power amplifier and the antenna. Lastly, the antenna radiates the electromagnetic signals.

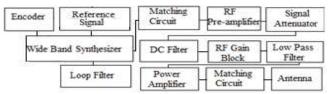


Fig. 5. System Block Diagram

C. Antenna Simulation

The element between the transmitter and receiver is the antenna. [6] This research used bowtie antenna. Several studies have used ANSYS software in simulating their antenna designs. For this research, the design simulated using the software. The first antenna effectively operates from 713 MHz to 836MHz with the center frequency of 760 MHz. The return loss at the 760MHz center frequency is -22.89dB.

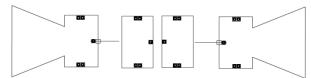


Fig 6. Bowtie Antenna Layout

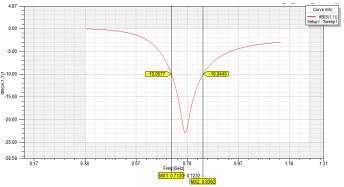


Fig. 7. 760MHz Bow tie Antenna Return Loss

The second antenna has center frequency of 886MHz. It effectively operates from 820 MHz to 964MHz with the return loss of -23.02 dB.

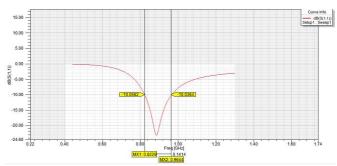


Fig. 8. 870MHz Bow tie Antenna Return Loss

D. Experimental Procedure

The experimentation starts with the preparation of the Staphylococcus aureus sample. The sample is diluted to achieve the quantity of a Colony Forming Unit: 10⁸ and 10⁶ bacterial concentrations. Microorganisms are processed in Tryptic Soy Broth solution as shown in Fig. 9. 5ml of the solution is pipetted on a Petri and then exposed to radiation.



Fig. 9. Processing of S. aureus in Trypticase Soy Broth Suspension Solution

A portion of the diluted solution, 5ml, was used and placed in Petri dish.



Fig. 10. S. aureus processed in Trypticase Soy Broth (TSB)

The exposure system is shown in Figure 10. It is consisting of a signal generator that produces Ultra High Frequency; the antenna that radiates the electromagnetic waves; and a small compartment that holds the Petri Dish.

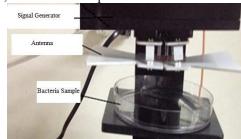


Fig. 10. Exposure System

During exposure, samples are covered with a shielding box. After exposure, a volume of samples are streaked to Blood Agar Plate (BAP). The samples are placed in an incubator with constant temperature from 35°C to 37°C for one day. After incubation, plates are removed from the incubator and the quantity of colonies in the plates is assessed.



Fig. 11. S. aureus being streaked on Blood Agar Plate

E. Data Gathering and Processing

After the 24-hour incubation, the bacterial colonies were analyzed by a professional Medical Technologist. The first batch of plates evaluated used a bacterial concentration of 10⁶. These plates were exposed to the following frequencies: 770 MHz, 780MHz, 885MHz, 890 MHz and 900MHz. The second batch of plates evaluated had a bacterial concentration of 10⁸. The frequencies used were 750MHz, 760MHz, 870MHz and 890MHz.

All the results were consolidated and the bacterial growths were calculated in terms of percentage. The percentage of bacterial growth was computed using the formula:

$$\%BG = \frac{CE}{CR} \times 100 \quad (1)$$

Where BG is Bacterial growth, CE is the number of colonies in Exposed sample and CR is the number of colonies in the Reference sample.

Using Machine Learning tool which is Matlab, the average bacterial growth and standard deviation achieved on the exposed and unexposed samples were calculated using the preceding formula:

$$\bar{x} = \frac{1}{n} \sum_{i=1}^{n} \mathbf{x}_{i} \qquad (2)$$

Where \bar{x} is the mean, n is the number of samples and x is the sample value.

Standard deviation =
$$\sqrt{\frac{1}{n-1}}\sum_{i=1}^{n}(xi-\bar{x})^{2}$$
 (3)

Where xi is the value at ith point in the samples, x is the mean value of the growth of bacterial colonies and n is the number of frequencies used.

Matlab was also used to calculate the relation between the parameters frequency and the percentage growth of bacteria was calculation using linear regression. This technique analyzes the possibility of manifestation of a binary response variable (y) with respect to predictor variable (x) [16].

$$Y = b_o + b_1 X_1 + b_2 X_2 + b_3 X_3 + b_4 X_1 X_2 + b_5 X_1 X_3 + b_6 X_2 X_3 + b_7 X_1 X_2 X_3$$
 (4)

The Pearson correlation coefficient was simulated using Matlab. It quantified the correlation between frequency and bacterial growth.

$$r = \sqrt{\frac{n(\sum xy) - \sum x \sum y}{\sqrt{[n \sum x^2 - (\sum x)^2][n \sum y^2 - (\sum y)^2]}}}^2$$
(5)

IV. RESULTS AND DISCUSSION

In the experimentation, different UHF frequencies were used. The volume of the sample was 5 milliliters. The samples were diluted to achieve a number of desired colonies. TABLE I shows the result for using 770 MHz, 780MHz, 885MHz, 890 MHz and 900MHz.

TABLE I. BACTERIAL COLONIES (REFERENCE= 10^6 concentration, 1,927 colonies)

Number	Frequency	Colonies in Experimental Sample (approximate)
1	770MHz	1,927
2	780MHz	1,927
3	885MHz	1,927
4	890MHz	802
5	900MHz	1,927

When 890MHz was used, the number of colonies after exposure was 802. On the other hand, when frequencies 770MHz, 780MHz, 885MHz and 900MHz were used, bacteria grew were 1,927 colonies.

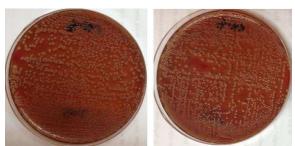


Fig. 14. Unexposed (left) and Exposed (right) Incubated Samples

TABLE II shows the results for using 750MHz, 760MHz, 870MHz and 890MHz. When 750 MHz was used, the bacteria that grew were 9,416 colonies. Using 760MHz, 9,322 colonies grew. When 870MHz and 890MHz were used, 9,132 and 7,610 colonies of the bacteria grew.

TABLE II. BACTERIAL COLONIES (REFERENCE=10^8 CONCENTRATION, 9,512 COLONIES)

Number	Frequency	Colonies in Experimental Sample (approximate)
1	750MHz	9,416
2	760MHz	9,322
3	870MHz	9,132
4	890MHz	7,610

TABLE III shows the percentage of the bacteria that grew after exposure to Ultra High Frequencies.

TABLE III. BACTERIAL GROWTH AFTER EXPOSURE

Number	Frequency	Bacterial Growth
1	750MHz	98.99%
2	760MHz	98.00%
3	770MHz	100.00%
4	780MHz	100.00%
5	870MHz	96.01%
6	885MHz	100.00%
7	890MHz	41.62%
8	890MHz	80.00%
9	900MHz	100.00%

100% means that the bacterial growth is not affected by the radiation. On the other hand, the bacterial growth that is less than 100% means that not all the bacteria survived after the radiation. Numbers 8 and 9 are both 890MHz because this frequency was used in both tests, that is, in using 10^8 and 10^6 bacterial concentrations.

The plot of the corresponding bacterial growth at a certain frequency is shown in Fig. 15. Looking at the data, the frequencies that affected the growth of Staphylococcus aureus are 750MHz, 760MHz, 870MHz, and 890MHz. At these frequencies, the bacteria that grew were only 98.99%, 98%, 96.01%, 41.62% and 80% respectively.

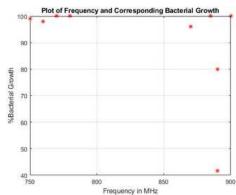


Fig. 15. Scatter Plot of Frequency and Corresponding Bacterial Growth

The plot for the average bacterial growth and standard deviation is shown in Fig. 16.

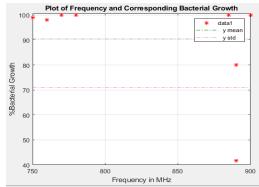


Fig. 16. Plot of Matlab Simulation for Mean

Matlab simulation result is shown in Table IV. The average bacterial growth was 90.51% and the Standard deviation is 19.43%.

TABLE IV. BASIC STATISTICS DATA FROM MATLAB SIMULATION	TABLE IV.	BASIC STATISTICS	S DATA FROM MAT	TLAB SIMULATION
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	Frequency in MHz (x)	%Bacterial Growth (y)
min	750	41.62
max	900	100
mean	832.8	90.51
median	870	98.99
mode	890	100
std	65.25	19.43
range	150	58.38

Using the basic linear fitting, the equation derived is y=-0.1293x+198.2. The correlation coefficient is 0.4344. There is a small positive linear relation between the frequency and the growth. This means that for the tests conducted, increasing the value of the frequency does not necessarily result to a lesser percentage of bacteria that would survive.

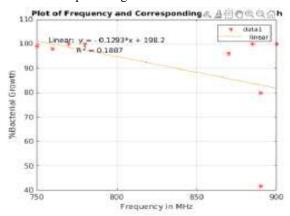


Fig. 18. Basic Linear Fitting using MATLAB

V. CONCLUSION

This research focuses on investigating the effect of Ultra High Frequencies when radiated to the bacteria, Staphylococcus aureus. Consolidating the data, the frequencies 750MHz, 760MHz, 870MHz, and 890MHz caused a change in the bacterial growth with colonies survival of 98.99%, 98%, 96.01%, 41.62% and 80% respectively. The average bacterial growth was 90.51%. This means that some of the bacteria did not survive after exposure to the radiation. The Pearson Correlation coefficient between the frequency and the growth of the bacteria is 0.4344. This means that for the tests conducted, increasing the value of the frequency does not necessarily mean that there will be lesser percentage of bacteria that would survive. From the results, it can be concluded that the radiation of Ultra High Frequencies, 750MHz, 760MHz, 870MHz, and 890MHz, affected the growth of the bacteria, Staphylococcus aureus. Conducting further repetitive tests will help gain a more comprehensive outcome. Other variables can also be explored including a different type of bacteria, antenna and bacterial concentrations. The result of this study can be an initial stage in discovering other possible applications of electromagnetic radiation in biomedical sciences.

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REFERENCES

- [1] World Health Organization. Coronavirus disease 2019 (COVID-19) situation report–51. Geneva, Switzerland: World Health Organization; 2020
- [2] World Health Organization. Coronavirus disease 2019 (COVID-19) situation report–57. Geneva, Switzerland: World Health Organization; 2020.
- [3] Self, W., Wunderink, R., Williams, D., Zhu Y., Anderson, E., Balk, R., Fakhran, S., Chappell, J., Casimir, G., Courney, M., Trabue, C., Watere G., Bramley, A., Magill, S., jain, S., Edwars, K, Grijalva., "Staphylococcus aureus Community-acquired Pneumonia: Prevalence, Clinical Characteristics, and Outcomes", US National Library of Medicine, National Institutes of Health, 2016 May 8
- [4] Spoto S, Valeriani E, Riva E, De Cesaris M, Tonini G, Vincenzi B, Locorriere L, Beretta Anguissola G, Lauria Pantano A, Brando E, Costantino S, Ciccozzi M, Angeletti S., "A Staphylococcus aureus Coinfection on a COVID-19 Pneumonia in a Breast Cancer Patient", Dovepress, International Journal of General Medicine, September 2020 Volume 2020:13 Pages 729-733
- [5] World Health Organization. Health topics. Corona Virus, 2020
- [6] Ali, F., Ray, S., "Detection and Selective destruction of Bacteria Colony at THz Frequencies", Conference: Communications (NCC), 2012 National Conference on, 2012
- [7] Tong S,. Davis J., Eichenberger E., Holland T., Fowler V., "Staphylococcus aureus Infections: Epidemiology, Pathophysiology, Clinical Manifestations, and Management" Jul; 28(3): 603–661. Published online 2015 May 27.
- [8] Frenzel, L Jr., "Principles of Electronic Communication Systems", McGraw Hill, 2014
- [9] Malikova, I., Janousek, L. "Non thermal Effects of Low-frequency Electromagnetic Field on Biological Cells", 2014, IEEE,
- [10] Buniyamin, N., Rosman, M., Ismail, M., Mohd-Zain, Z., "The effect of Mobile Phone Electromagnetic Radiation n Staphylococcus aureus: A Preliminary Investigation", IEEE, 2012
- [11] Malikova I., Janousek ,L., Fantova, V., Jira, J., and Kriha, V., "Impact of Low Frequency Electromagnetic Field Exposure on Selected Microorganisms", IEEE, 2015
- [12] Samortin A., Garcia R., Dela Cruz J., "The Utilization of Electromagnetic Radiation from a Bowtie Antenna for Eradicating Staphylococcus Aureus Bacteria", ICBET, 2020
- [13] Kumar, R., Kartikeyan, M.,"ANSYS Design and Simulation of Multi Band Compact Microstrip Patch" IEEE, 2019
- [14] R. C. Compton, R. C. Mophedran, Z. Popovic, G.M. Rebeiz and P.P. Tong, "Bow-Tie Antennas on a Dielectric Half-space: Theory and Experiment", IEEE Transactions on Antennas and Propagation, Vol. Ap-35, No. 6, June 1987
- [15] Yu, L., "Matlab Programming Environment Based on Web", IEEE 2018
- [16] Imamou D., "Application of MATLAB in Solving Function and Derivation", Mathematics Learning and Research, no. 15, pp. 11-12, 2012.
- [17] A. Urso, A. Fiannaca, M. La Rosa, V. Ravì, and R. Rizzo, "Data mining: Prediction methods," Encycl. Bioinforma. Comput. Biol. ABC Bioinforma., vol. 1–3, pp. 413–430, 2018, doi: 10.1016/B978-0-12-809633-8.20462-7.