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The Impact of Cobalt and Strontium Radiation on *Pseudomonas aeruginosa*

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ABSTRACT

The research aims to study the effect of radiation emitted from different clinical sources, including cobalt and strontium. The study was carried out using isolates of Pseudomonas bacteria isolated from Baghdad hospitals during the year 2021, about 20 isolates from the blood of patients infected with the bacteria. The results showed a lethal effect on Pseudomonas bacteria when exposed to radiation at different times during one hour, two hours and three hours. Different doses using cobalt with effective 1 mci and 10 mci in the presence and absence of aluminium, and the use of strontium in the presence and absence of aluminium. Co^{60} (10Mci) without Aluminium, with dose 12.48472106(1hr.), 24.96944212 (2hr.), 37.45416318 (3hr.) that emit Beta and Gamma; Co^{60} (10Mci) with aluminium with dose 0.023315312×10^{-7} (1hr.), 0.046630625×10^{-7} (2hr.), 0.069945936×10^{-7} (3hr.) that emit Beta ray; Co^{60} (1Mci) without Aluminium with dose 3.4178394 (1hr.), 6.835678887 (2hr.), 10.2535182 (3hr.) that emit Beta and Gamma; Co^{60} (1Mci) with aluminium with dose 0.155992856*10-4 (1hr.), 0.311985712*10-4 (2hr.), 0.467978568×10^{-4} (3hr.) that emit Beta ray and Sr90 with dose 0.73251×10^{-2} (1hr.) 1.4650×10^{-2} (2hr.), 2.19753×10^{-2} (3hr.) The results showed that the killing rate of *P. aeruginosa* bacteria using cobalt 10 mCi without aluminium was 78%, and in the presence of aluminium 100%. In comparison, the killing rate of Pseudomonas bacteria with effective effectiveness of 1 mC without aluminium was 100% and in the presence of aluminium 98%, and the killing rate of Pseudomonas bacteria without aluminium was 83% and in the presence of aluminium 96% compared with control.

KEYWORDS: Irradiation, radiation, bacteria, cobalt, strontium

INTRODUCTION

Pseudomonas aeruginosa is a Gram-negative, aerobic rod-shaped bacterium belonging to the Pseudomonadaceae bacterial family, a gamma-proteobacteria member. *P. aeruginosa* is one of 12 subtypes that make up the group. The pathogen is a free-living organism that thrives in various planktonic environments. These bacteria can also build biofilm and accounts for 10 to 20% of hospital infections [1]. One of the most therapeutically and epidemiologically important microorganisms is Pseudomonas aeruginosa. It's the most common source of nosocomial infections in non-fermenting patients. Gram-negative bacteria are the most common cause of opportunistic infections in immunocompromised individuals (Killed *et al*.,2013). It is one of the most common etiological agents of nosocomial infections [2] and continues to be under selection pressure in hospital settings [3]. Radiation is often classified as ionizing or non-ionizing based on the particle energy emitted. Ionizing radiation with a wavelength energy greater than 10 eV is sufficient to ionize atoms and molecules and break chemical bonds. This distinction is critical because of the considerable disparity in the harmfulness to living beings. Ionizing radiation is commonly produced by radioactive elements emitting radiation made up alpha particle, electrons or positrons, and photons. Other sources include medical X-rays positrons, neutrons, muons, mesons, and other particles that make up secondary cosmic rays. The ionizing part of the electromagnetic spectrum includes X-rays, gamma rays, and the higher energy range of ultraviolet light. The term "ionize" alludes to breaking one or more electrons from an atom, necessitating the comparatively high energy these

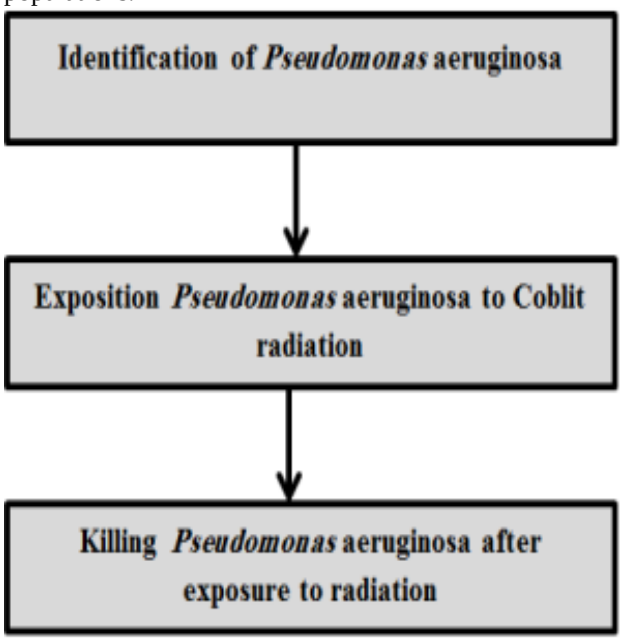
electromagnetic waves provide. For example, burn is an example of a chemical reaction in which molecules are broken down by lower UV wavelengths, which are not capable of ionizing them but can disrupt interatomic bonds that hold them together. Visible light, infrared, and microwave waves with longer wavelengths are incapable of breaking bonds, although they can cause vibrations in bonds that are viewed as heat. Biological systems are generally not considered to be harmed by radio frequencies and below. There is a lot of overlap regarding frequency-specific effects [4]. Thus, they are no clear delineations of energy. The nuclei of helium-4 are known as alpha particles (two protons and two neutrons). Due to their charges and total mass, they interact aggressively with the matter. Their speed is limited to a few millimeters of air or a few centimeters of low-density material (such as the thin mica material specially placed in some Geiger counter tubes to allow alpha particles in). A typical alpha decay process doesn't penetrate the outer layers of dead skin cells, enabling alpha particles to reach the living tissues below. 10% of cosmic radiation is high-energy alpha particles, which may penetrate the body and even thin metal plates. However, they only pose a danger to astronauts due to the Earth's magnetic field deflecting and blocking them and the atmosphere [5]. Beta-minus radiation is more penetrating than alpha but less than gamma. Several millimeters of plastic or metal can block radioactive beta rays. A beta particle and an antineutrino are formed when a neutron decays into a proton in a nucleus. Natural beta radiation is significantly less powerful and penetrating than the beta radiation produced by linac accelerators. It is sometimes employed as a therapeutic tool in treating superficial tumours. The release of positrons, which are electrons created by antimatter, is known as beta-plus (+) radiation. When a positron slows down to the same speed as electrons in a material, it will annihilate an electron, generating two 511 keV gamma-rays. Two gamma rays travel in opposite directions. High-energy photons from positron annihilation produce ionizing gamma radiation [6].

Cobalt-60 is made by putting Cobalt-59, a naturally occurring metal, into a nuclear reactor, where it absorbs neutrons and transforms into the radioactive Cobalt-60. The Cobalt-60 is extracted from the nuclear reactor and sent to a facility encased in stainless steel pencils. These Cobalt-60 pencils, also known as sources, have been tested and verified to satisfy international design standards. The Cobalt-60 sources are placed in gamma irradiators, which treat products and materials by exposing them to the gamma radiation released by the sources. In a gamma irradiator, cobalt-60 sources were mounted [7]. The sources are kept in a secure undersea location [8, 9].

METHODOLOGY

The clinical investigation comprises study populations from various sources, VITECK2-GN *P. aeruginosa* screening, radiation impact, and statistical analysis [10]. Figure 1 shows the methodology of this study. The study design is a strategy or protocol for a direct study that allows the researcher to translate the conceptual hypothesis into an operational one. It is also the formulation of trials, experiments, and observational studies in medical, clinical, or other human research (e.g., epidemiological) [11].

Figure 1. Methodology of research project Study populations.



In 2021, obtained blood samples from 20 *P. aeruginosa* isolates from Baghdad hospitals. According to [12], these isolates were identified by conventional biochemical tests. It was found that the results after 24 hours of incubation at 37°C with Nutrient agar inoculation remained stable for the isolates. It was identifying blood *P. aeruginosa*. Examine morphology Diagnosis of bacterial

isolates based on colony shape, texture, color, and edges in culture media. [13] MacConky agar was used. Examination Under a Microscope To compare the results with Holt, 1998, to do the Gram stain reaction on an isolate, one colony was chosen and put on a microscope slide. Vitek2-GN was used to identify *P. aeruginosa* from blood samples. The Vitek2 GN system for *P. aeruginosa* determined the bacterial isolates by the manufacturer's instructions. To determine carbon source use, enzyme activity, and antibiotic resistance, scientists use 64 different biochemical tests. This system performs 64 biochemical assays from a single pure isolate colony. Turbidity was adjusted to 0.5 McFarland for the Vitek2 GN suspension solution. Gram-Negative (GN) cards are used to identify Gram-Negative bacteria automatically. The biological procedures used to create the GN identification card are well-established. Biomerieux (2013) inoculated French and newly designed substrates with a sterile Pasteur pipette according to each test's recommendations and compatibility. After 24 hours of incubation at 37°C, an automatic analytical, quick identification at the species level was used to determine the isolate's identity. The GN card and suspension tubes were inserted with a single negative control well in a cassette. Biochemical patterns are subjectively assessed and stored by the device. After snuggling, the device software evaluated the patterns and gave diagnostic data for each Reader/Incubator card, per Biomerieux [14, 15, 16, 17].

Impact of Coblit and Strontium radiation on *P. aeruginosa*

Centrifuged at 5000 rpm, the *P. aeruginosa* was cultured for 24 hours in Nutrient stock at 37°C, similar to [18] with some modifications. Using a Coblit radio source and the control configuration (outward radiation exposure), 1 ml of the solution was injected into Trypton soy agar and analyzed in triplicate. Using sterile normal saline, the residue was compared to a control of MacCfrland 0.5. The equation percentage of killing:

$$Percentage\ of\ killing = \frac{Control - treated}{Control} \times 100$$

Statistical analysis

The Statistical Analysis System (SAS) program was utilized to alter various study parameters. This comparison was made using a Chi-square test[19].

RESULTS AND DISCUSSIONS

P. aeruginosa [20] was the subject of a descriptive cross-sectional investigation in which blood samples were taken. Study populations *P. aeruginosa* isolated from *P. aeruginosa* from Baghdad hospitals through 2021. Identification by using vitek2- GN system.

Identification of *P. aeruginosa*

Morphological and Microscopic examination of *P. aeruginosa*

The collected isolates were initially diagnosed in hospitals as *P. aeruginosa*. All isolates were grown on MacConkey agar to confirm this diagnosis, where gram-positive and gram-negative bacteria grew [13].

Impact *P. aeruginosa* to Coblit radiation

Results listed in Tables 1 and 2 show the results of this study, Co^{60} (10Mci) without Aluminum, with dose 12.48472106(1hr.), 24.96944212 (2hr.), 37.45416318 (3hr.) that emit Beta and Gamma; Co^{60} (10Mci) with aluminum with dose $0.023315312*10^{-7}$ (1hr.), $0.046630625*10^{-7}$ (2hr.), $0.069945936*10^{-7}$ (3hr.) that emit Beta ray; Co^{60} (1Mci) without Aluminum with dose 3.4178394 (1hr.), 6.835678887 (2hr.), 10.2535182 (3hr.) that emit Beta and Gamma; Co^{60} (1Mci) with aluminum with dose 0.155992856×10^{-4} (1hr.), 0.311985712×10^{-4} (2hr.), 0.467978568×10^{-4} (3hr.) that emit Beta ray and Sr^{90} with dose 0.73251×10^{-2} (1hr.) 1.4650×10^{-2} (2hr.), 2.19753×10^{-2} (3hr.).

Table 1. Radiation dose, energy and activity of Isotope are all factors in determining the percentage of *P. aeruginosa* colony annihilation.

Sources isotope	Dose (msV)	Energy (Kev)	Type of radiation	Activity (Mci)
Co ^{60} (10Mci)	12.48472106(1hr.), 24.96944212 (2hr.), 37.45416318 (3hr.)	1173.1332	Beta and Gamma	10
Co ^{60} (10Mci)	0.023315312×10^{-7} (1hr.), 0.046630625×10^{-7} (2hr.), 0.069945936×10^{-7} (3hr.)	1173.1332	Beta ray	10
Co ^{60} (1Mci)	3.4178394 (1hr.), 6.835678887 (2hr.), 10.2535182 (3hr.)	1173.1332	Beta and Gamma	1
Co ^{60} (1Mci)	0.155992856×10^{-4} (1hr.), 0.311985712×10^{-4} (2hr.), 0.467978568×10^{-4} (3hr.)	1173.1332	Beta ray	1
Sr^{90}	0.73251×10^{-2} (1hr.), 1.4650×10^{-2} (2hr.), 2.19753×10^{-2} (3hr.)	0.198	Beta ray	10

Table 2. Cobalt radiation kill and viability percentages for *P. aeruginosa* at various dosages and energies.

No.		Viable cell			Percentage of killing		
		1h.	2h.	3h.	1h.	2h.	3h.
1 CO (10Mci)	P1	86	16	66	71%	94%	78%
	P2	0	2	0	100%	99%	100%
2 CO (1Mci)	P1	11	19	0	96%	93%	100%
	P2	21	70	4	93%	76%	98%
3 Sr^{90}	P1	80	10	50	73%	96%	83%
	P2	70	30	10	76%	90%	96%
Control = 300							

P1: mean without aluminum, P2: mean with aluminum.

The results showed that the killing rate of *P. aeruginosa* bacteria using cobalt 10 mCi without aluminum was 78%, and in the presence of aluminum, 100%. In comparison, the killing rate of Pseudomonas bacteria with effective effectiveness of 1 mC without aluminum was 100% and in the presence of aluminum 98%. The killing rate of Pseudomonas bacteria using ^{90}Sr without aluminum was 83% in the presence of aluminum 96% compared with control.

Studies have shown that the Nd: YAG laser has an effect on *Klebsiella* pneumoniae while also having an effect on beta and gamma rays, using ^{137}Cs, ^{60}Co, ^{90}Sr, and ^{22}Na Tl as well as Cs 137 [20]. The exposure was in dose 0.3863×10^{-8}, Co^{60} in dose 1.826×10^{-8}, ^{90}Sr in dose 1.973×10^{-8}, Na in Gamma-ray 0.31993×10^{-8} and 1.4157×10^{-8} in Beta ray in 3hr with done control. Additionally, K.pneumoniae was exposed to Nd: YAG laser pulses at 1064° with a 6-second interval between each pulse for 500, 1000, and 1500 pulses in triplicate. Laser and radiation exposure to K. pneumonia-resistant colistin decreased the bacteria's vitality, with a large percentage of the bacteria being killed. For destroying K. pneumoniae resistant colistin, Nd: YAG laser and Beta, Gamma radiation proved highly effective. Researchers at Imam Ali Hospital in Babil Governorate and Hilla Teaching Hospital in Baghdad examined the effects of radiation on various Staphylococcus aureus bacterial isolates obtained from those same hospitals and patients with various types of burns and wounds of the urinary tract and infected skin from those same hospitals. A variety of radionuclides, including ^{137}Cs and ^{60}Co, with activity levels of 1-10 c and varying radiation doses, were used to identify *S. aureus*, which was then implanted in Nutrient broth and the nutrient agar was first placed in a test tube containing 5 ml of distilled water before being exposed to varying radiation dosages for times ranging from 1-2-3 hours. The bacteria were then placed on Petridishes plates of Nuterinat. The effects of being exposed to beta and gamma rays from ^{137}CS and ^{60}Co radioactive sources with 10 µci and 1 µci. for 1hrs.,2hrs. 3hrs, with various doses includin Cs (10 µci) Beta 3.160×10^{-1} (1hrs.), 6.32×10^{-1} (2hrs.), 9.48×10^{-1} (3hrs.) 98.3%, 99.5%, 99.5% respectively; Cs (10 µci) Gamma5.815×10^{-1} (1hrs.), 11.63×10^{-1} (2hrs.), 17.445×10^{-1} (3hrs.) 98.9%, 99.6%, 99.5% respectively; Cs (1 µci) Beta 3.417×10^{-1} (1hrs.) , 6.835×10^{-1} (2hrs.), 9.253×10^{-1} (3hrs.), 99.3%, 99.5%, 99.9% respectively ; Cs (1 µci) Gamma 6.289×10-1 (1hrs.), 12.678×10^{-1} (2hrs.), 18.867×10-1 (3hrs.) 99.2%, 99.6%, 99.8% respectively; Coblit with Co(1 µci) Beta 1.2×10^{-1} (1hrs.), 2.4×10^{-1} (2hrs.), 3.6×10^{-1} (3hrs.) 99.4%, 99.2%, 99.8%; Co(10 µci) Beta, 1.86×10^{-1} (1hrs.), 3.7×10^{-1} (2hrs.), 5.5×10^{-1} (3hrs.) 99.6%, 99.8%, 99.9% respectively ;

Co(10 µci) Gamma, 2.33×10^{-1} (1hrs.), 4.66×10^{-1} (2hrs.), 6.99×10^{-1} (3hrs.), 95.2%, 99.8%, 99.8% respectively. While, ^{90}Sr with Beta(1µci), 0.155992856×10^{-4} (1hr), 0.311985712×10^{-4} (2hr) 0.467978568×10^{-4} (3hr), 76%, 90%, 96%, respectively. Calculate the number of colonies with the percentage of killing. Calculate the number of colonies with the percentage of killing.

Radiation therapy is a complex process that requires a team of experts from a wide range of fields, including doctors, nurses, and radiation therapists. Each of the 20 to 40 fractions or sessions of external beam radiation calls for a plethora of machine and patient parameters that are typically the same yet unique to each patient. The International Basic Safety Standards for the Protection against Ionizing Radiation and the Safety of Radiation Sources (the BSS), which establish requirements for investigating accidental medical exposure and implementing corrective measures to prevent a recurrence, have given special radiotherapy consideration in safety standards [9].

CONCLUSIONS

There are a high number of *Pseudomonas aeruginosa* isolated from blood specimens. There are Pseudomonas aeruginosa isolated from blood killed by radiation with a high dose of Co60 and Sr90 with different doses through exposure to different hours.

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