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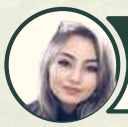

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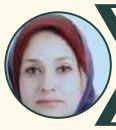

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PREFACE

The 11th International Conference on Applied Sciences and Technology Research "MINAR CONGRESS", was organized by Igdir University in collaboration with Rimar Academy. The primary objective of this event was to compile and disseminate valuable scientific knowledge and make a meaningful contribution to the future.

Remarkably, a substantial number of researchers, both from local and international backgrounds, demonstrated their interest in this conference. The scientific committee meticulously reviewed the submissions and ultimately accepted a select group of individuals, totaling 55 applicants, 30 of them were accepted by the scientific committee.

This conference was truly a global endeavor, as it drew participation from 5 attendees who joined in person, alongside 25 who engaged in the event remotely. These participants collectively enriched the conference with their expertise and insights.

The core of this conference was the presentation of 16 complete research papers, while the remaining articles and research findings are set to be featured in forthcoming issues of the MINAR Journal.

I would like to extend my sincere appreciation to all the contributors and scholars who played an essential role in making this conference a resounding success. Your dedication and valuable contributions are deeply respected and acknowledged.

Editor-in-Chief

Prof. Dr. Ghuson H. MOHAMMED

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The Effect of an Argon Plasma Jet and Transient Spark Discharge On Bacteria, Cells, And Aquatic Biomolecules in Fish Ponds

Ghaith H. Jihad [1], Zahraa A. Abdul Muhsin [2]

Abstract

Self-pulsed transient spark discharge (TS) operating at atmospheric pressure in DBD and plasma jetting for pulse-driven dielectric (PJ) dielectric discharge operating in argon and air at atmospheric pressure Chemical reactions were induced in water solutions using water-based solutions. The study aimed to treat Escherichia coli bacteria taken from a local aquarium and analyze them with emission and absorption spectroscopy. The study compared the results of TS and PJ systems, direct exposure to the vacuum solution, and ancillary contact with vacuum-activated gas flow. The aqueous solutions were analyzed for chemical composition using UV-VIS absorption spectrophotometry. The study aimed to determine the concentration of the primary bactericidal component in fish pond water using infrared and ultraviolet absorption spectroscopy in the air plasma jet. This was done to increase the reliability of measurements. The study demonstrated that some cells can be targeted by cold plasma, which also inactivates germs. This has significant implications for developing new plasma therapeutic strategies in biomedicine. Specifically, plasma jets' effects on S. aureus bacteria were studied, and results suggest that this technique could hold great promise for developing new treatments.

Key Words

Production And Absorption Spectroscopy; Transient Spark Discharge; Plasma Jet; Deactivation Of Bacteria.

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Introduction

Recent research on the wide range of biological applications of nonthermal (cold) atmospheric pressure plasmas demonstrates the rapid development and great promise of this novel, interdisciplinary field [1]. Over the past decade and a half, research in the field of Biocontamination and sterilizing using cold plasma has shown that ionizations, dissociations, excitations, and chemical low-gas-temperature reactions that take place can effectively clean and sterilize various surfaces, equipment, water, air, food, and even live tissue [2,3]. Cold plasmas are particularly useful for destroying a wide range of microorganisms, including extremely resilient structures like biofilms and bacterial spores. This technology offers a promising solution for maintaining cleanliness and sterility in various settings [4,5].

When properly calibrated and dosed, several cold plasma sources have been shown to cause intriguing events in the cells of higher species, which has led to promising therapeutic results [6]. Although plasma has been shown to have many beneficial benefits in therapy and disinfection, little is known about how plasma interacts with live cells and microbes. There is ongoing discussion over the plasma therapy of biomolecules, cells, and tissues [7,8]. The earliest known plasma treatments of mammalian cells were most likely non-destructive manipulations that resulted in the reversible separation of the cells from the substrate and from one another [9]. Later, it was reported that mammalian cells may undergo plasma-induced apoptosis. In contrast to necrosis, which is an early form of cell death brought on by severe external stimuli, apoptosis is a type of physiologic cell death that higher organisms normally employ to get rid of damaged or non-functioning cells [10-12].

Lipid peroxidation, which is often caused by plasma treatment of lipids, might impact the fluidity, integrity, and transport of cell membranes [13]. This has been thoroughly investigated and recorded in studies. Analogously [14], oxidations [15], structural alterations in amino acids [16], and denaturation of proteins are commonly observed upon plasma treatment of amino acids and proteins. There has also been a lot of focus on this field of study [17].

Arcing can help generate radiation and reactive species, but it can also overheat the gas. Electrical circuits or dielectric barriers that limit the current or discharged energy can be utilized to stop or manage arcing [18]. An illustration of this is the transient spark (TS), a self-pulsing discharge powered by direct current (dc) that generates high current pulses with a typical repetition frequency of 1–10 kHz and a very brief duration (about 0-100 ns) [19-21]. The TS exploits the reactive pulsating streamer and transition conditions in atmospheric air that are initiated by a streamer-to-spark [22]. To prevent Overheated Gases and Plasma Thermalization,

the TS sets a limit on the discharge energy. The TS, which was first described as a stopped spark, is an effective tool for scientists and researchers across several disciplines [23].

This research aims to illustrate how bacteria are affected by cold plasmas. The direct and indirect effects of two distinct sources of cold atmospheric plasma will be compared. The first is the transient spark discharge that is created in the air and pulses on its own [24]. It is known to be an efficient producer of Reactive Oxygen and Nitrogen Species (RONS) and can be applied to water solutions and biomolecules. We will investigate how this discharge interacts with water solutions containing both bacteria and normal cells [25]. Additionally, we will conduct a comparative analysis of the direct exposure of water solutions to plasma and their exposure to the plasma-activated gas flow indirectly [26].

Materials and methods

The transient spark (TS) discharge is a self-pulsing repetitive streamer to spark transition discharge driven by DC [27]. This discharge was operated in ambient atmospheric pressure air and brought in contact with liquid that circulated by a peristaltic pump repetitively through a discharge zone. All the experiments were performed at a constant applied voltage of around 14 kV and a current pulse frequency of approximately 2 kHz [28]. The TS discharge was applied to planktonic suspensions of S. aureus and E. coli bacteria. The effect of cold atmospheric plasma (CAP) on cell membrane integrity was evaluated in different bacterial growth phases, such as stationary and exponential phases, and correlated with the bactericidal effect [29].

Experiment method

A. Discharge mechanism

1. Air transient spark discharge

In our experiments, we used a positive DC-driven TS discharge as the first type of discharge. This discharge is generated in ambient air and transitions from streamers to sparks in a repetitive manner [30-32]. We generated the discharge using two different configurations in ambient air: the systems that use water spray and electrodes.

The water electrode system (WE), seen in Figure (1), is made up of an inclined grounded plane electrode positioned in point-to-plane geometry above a high-voltage needle electrode. The water solution in this system was supplied by a thin channel in the plane electrode and was subsequently pumped at a steady flow rate of 5–10 L/min by a peristaltic pump [33-35].

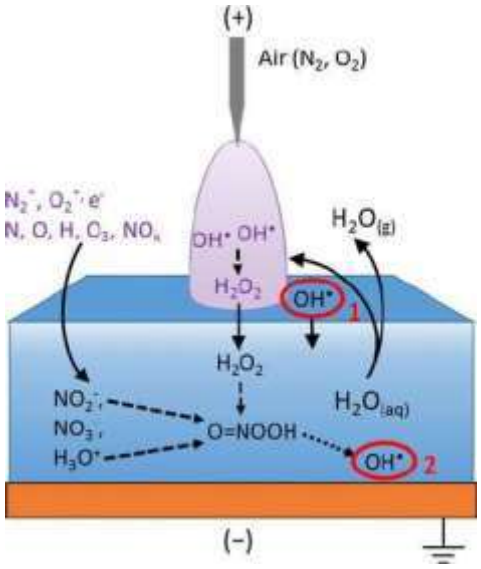

Figure 01- An illustration of a transient spark discharge in air [36]

2. Ar plasma jet

In this experiment, a DBD discharge chamber was used, as shown in Figure 2. A similar discharge chamber had been used in previous studies [37, 38]. The chamber was a quartz tube with a 2 mm inner diameter and 4 mm outer diameter, acting as a dielectric barrier. The quartz tube was filled with a high-voltage tubular electrode, while the second electrode was grounded and positioned close to the tube's end.

The gas used to create plasma was argon with a purity of 99.99%. This gas was blown through a quartz tube at flow rates of 1.5 and 10 L/min. To create the discharge, a laboratory pulsed high-voltage power source with a step-up transformer capable of providing an output voltage of up to 4.5 kV was used. A plasma jet formed as a result of the plasma produced in the DBD being transported into the surrounding medium by the flow of plasma-forming gas [39-41].

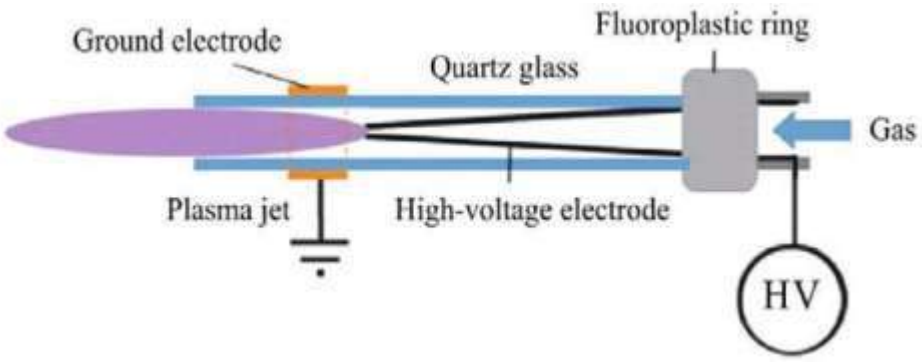

Figure 02- Diagram showing the DBD plasma jet's discharge chamber [42]

B. Measurements of Diagnosing Plasma Jets Optically

Emission spectra were recorded using a THORLABS spectrometer with plate number 1 and a resolution of 0.2 nm/pixel. The measurements were taken in the range of 200-1000 nm and were orthogonal to the axis of the plasma jet. The quartz tube's cross-section was used to take the measurements. Figure (3) Ar plasma jet spectrum included significant vibrational-rotational bands of the first negative N system N2+(B-X) and second positive N system N_2(C-B), as well as lines for Ar at 587 and 706 nm and an O I line at 777 nm. The existence of spectral components that are not associated with Ar was caused by the infiltration of ambient air into the jet. The air plasma jet's exceedingly weak emission at the discharge tube cross-section made it difficult to record its emission spectrum [43].

Figure 03- Photographic of Optical Emission Spectroscopy diagnostic tools

C. Evaluations of the bactericidal properties

The Gram-negative bacterium E. coli was used to investigate the plasma discharges' bactericidal properties. The initial populations of the planktonic bacteria, which were suspended in deionized water, ranged beginning 10^9 to 10^9 colony-forming units per milliliter (CFU ml^{-1}). The following procedures were used to cultivate the bacteria in a sterile environment: sterile liquid nutrients were used to create an overnight bacterial culture in a shaker. To achieve the appropriate concentrations, the culture was then centrifuged many times and diluted in saline or water solutions [44].

Bacterial suspensions were used in plasma tests, which were repeated ten to fifteen times in both WS and WE systems. PJ was used in bactericidal tests that were run up to three times. Colony-forming units (CFUs), which are cultivated on agar plates for 12–24 hours at 37°C, were used to count the amount of bacterium cells in the solution. To obtain the ideal number of

CFUs (colony-forming units) cultivated on agar plates, a series of tenfold dilutions were used. This was particularly important for low inactivation rates and controls, and the ideal range was between 30 and 300 CFUs. However, for high plasma-treated water inactivation rates, dilutions were not necessary. Typically, each sample produced three to four agar plates, which were then used for statistical analysis. The ratio of bacteria that survived in samples treated with plasma in reference samples relative to the entire population was used to assess the viability of the bacteria.

Results and discussion

A. Discharge systems

This study examined the chemical and biological impacts of two distinct cold atmospheric plasma sources. These included the DBD PJ and the self-pulsing TS produced in the air.

1. Air transient spark discharge

Figure (4) shows the relationship between Optical Emission Spectra and gas flow rate for TS where the magnetron voltage (200) V for pressure gas flow (1, 5, and 10) L/min. From Figure 4, we notice a group of emission peaks belonging to the argon, oxygen, and nitrogen atoms, as an indication of each peak. We note that the emission caused by the argon atoms is mostly caused by non-ionized atoms ArI. The spectrum produced by oxygen atoms is produced by atoms ionized to higher degrees, up to the loss of three electrons.

This indicates that TS has a high ability to produce molecules and atoms with a high ionization degree compared to other types of plasma. From the emission spectrum of TS, we conclude that this plasma contains high active oxygen, which can produce RONS compounds when this plasma interacts with the atmospheric air. we notice that the emission spectrum has been preserved with the same number of emission peaks, with the same locations and the same general shape, except that the intensity of the spectral lines has decreased when the diameter of the discharge tube decreases, due to the shortening in the number of atoms and molecules contributing to the generation of this spectrum [45].

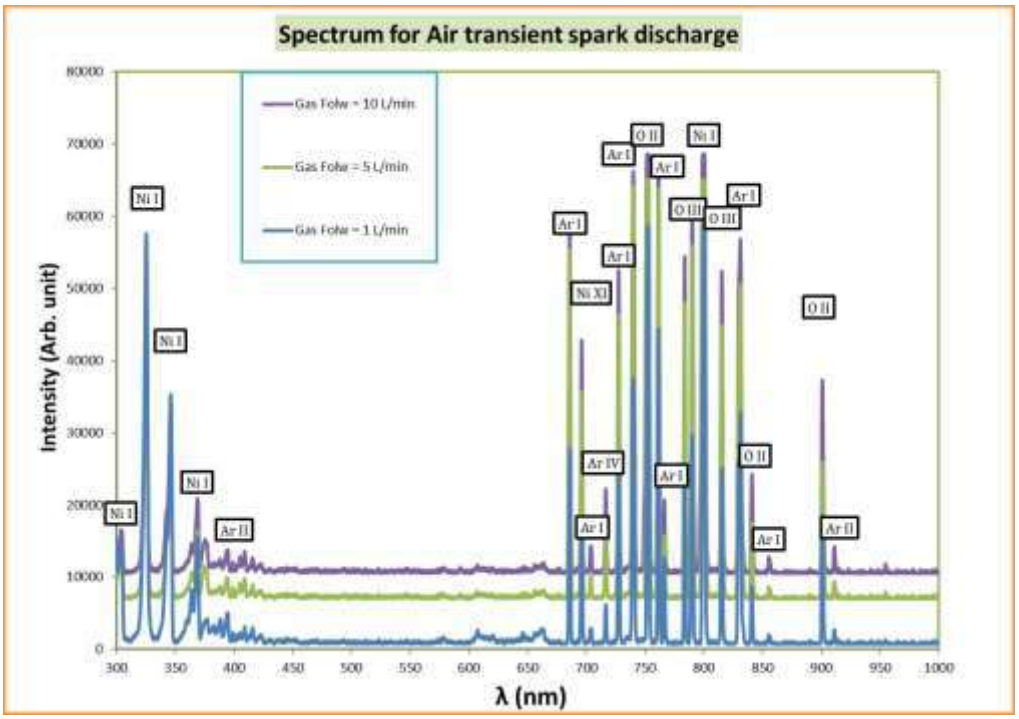

Figure 04- The relationship between Optical Emission Spectra and gas flow rate for TS where the voltage (200) V

We note from the figure that the electron temperature and electron density increase with the increase in flux when the discharge tube diameter is constant (10 mm) when the voltage is increased with a constant tube diameter, T_e was at 10 L/min (1.27) eV and (n_e = (5.58E +18)) and 5 L/min it was T_e (1.199 eV) and one (3.54 E + 18) and at 1 L/min it was T_e (1.144 eV) and one (2.45 E +18)) indicates This indicates that when the gas flow rates increases, Te increases due to more energy being supplied, and thus the kinetic energy of the electron increases as shown in figure (5) [46].

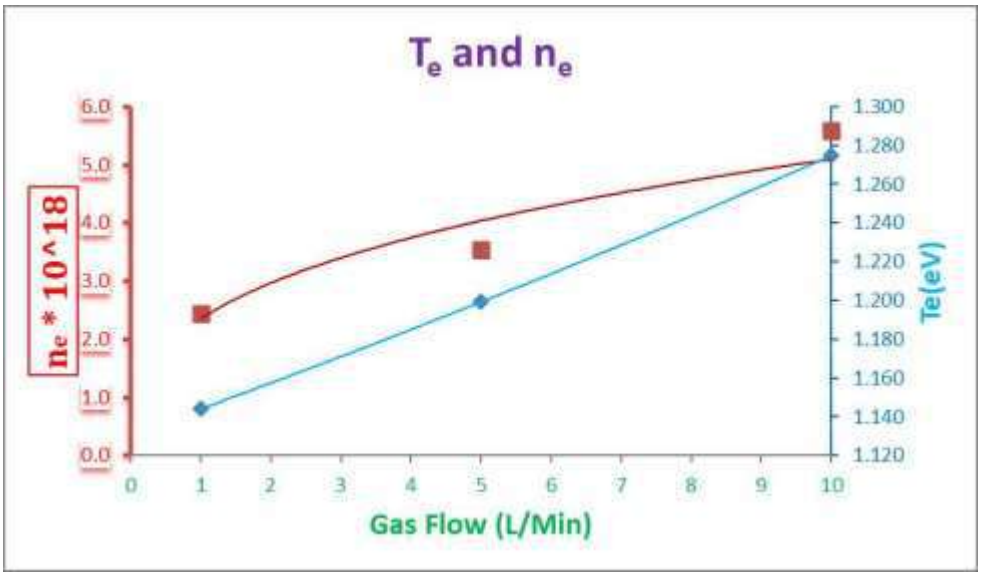

Figure 05- TS electron temperature T_e and density n_e as a function of gas flow rate for (1, 5 and 10) L/min inner diameter discharge tube at a voltage of 200

Table (1) shows the calculated electron temperature (T_e), electron density (n_e), Debye length (λ_D), plasma frequency (f_p), and Debye number (N_D) for Argon gas at different gas flow rates. All plasma parameters computed (λ_D, f_p, and N_D) showed that the Argon plasma generated satisfies the plasma conditions. It shows that f_p decreases with the gas flow because it's proportional to n_e, while λ_D and Nd increase with it [47].

Table 01- Plasma parameters for TS at different argon gas flow rates at voltages (200 V)

Gas Flow (L/Min)	T_e (eV)	n_e (cm^{-3})	f_p (Hz)	λ_D (cm)	N_d
10	1.275	5.58E+18	2.1E+13	3.3E-05	8.4E+05
5	1.199	3.54E+18	1.7E+13	4.0E-05	9.6E+05
1	1.144	2.45E+18	1.4E+13	4.7E-05	1.1E+06

2. Plasma jet DBD discharge

Plasma emissions were recorded by the plasma jet system at different flow rates, produced by air pressure. The resulting amplitude of the spectrum distribution was displayed against wavelength. The Optical Emission Spectroscopy (OES) technique was employed to obtain the argon gas optical emission spectrum for generating cold plasma. Each atom and Ar ion has a distinct set of spectral lines that make up its spectrum. Figure (6) displays the argon outflow spectrum gas to form a cold plasma in the frequency range of 600–950 nm. The stream rates used were 1, 5, and 10 L/min. The standard lines for ArI and ArII are used to represent the spectra. The Ar optical emission spectra for the cold plasma were measured using the OES method.

According to Figure 6, the peak flow rate of argon has a significant and consistent impact on the intensity of emission lines. The gas flow rate increases the intensity of spectral lines when additional molecules pass through the tube and the common of them ionize the gas passing through the plasma needle, resulting in an increased number of excited atoms and consequently, an increase in spectral line intensity peaks.

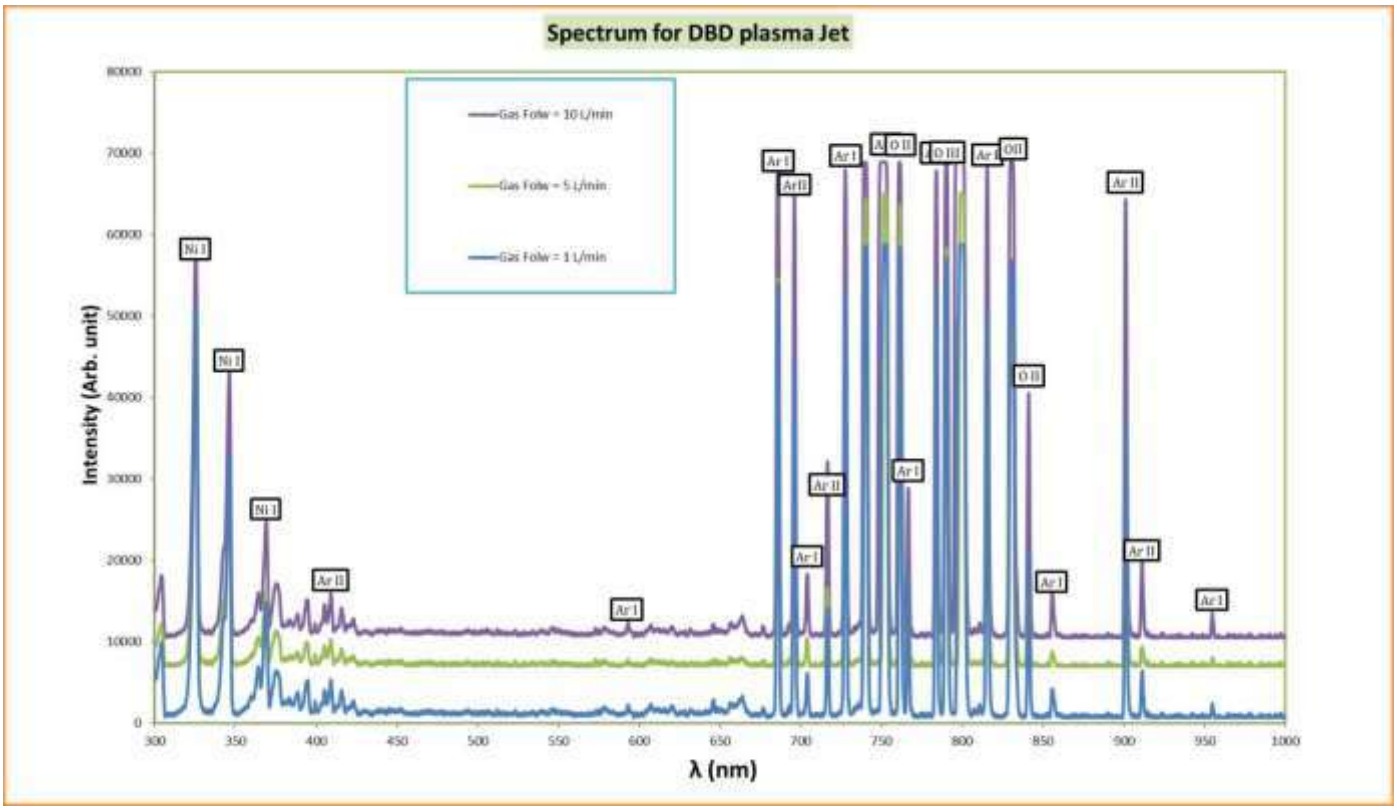

Figure 06- The relationship between Optical Emission Spectra and gas flow rate for DBD where the voltage (200) V

The temperature of the electron (T_e) was calculated using the line strength ratio, as shown in Figure 7. The NIST data was used to extract the positive aspects of some barriers, by utilizing the spectra from the OES. The line graph in the figures depicts the correlation between the change in gas flow rate and the temperature (T_e) and density (n_e) of electrons. As per the graph, both T_e and ne in the cold plasma system increase with an increase in flow rate. Electron temperature and density are 1.001 eV and 1.73 x10^{17} cm^{-3} at 1 L/min flow rate, respectively, and 1.173 eV and 6.49 x10^{17} cm^{-3} at 10 L/min, respectively [48].

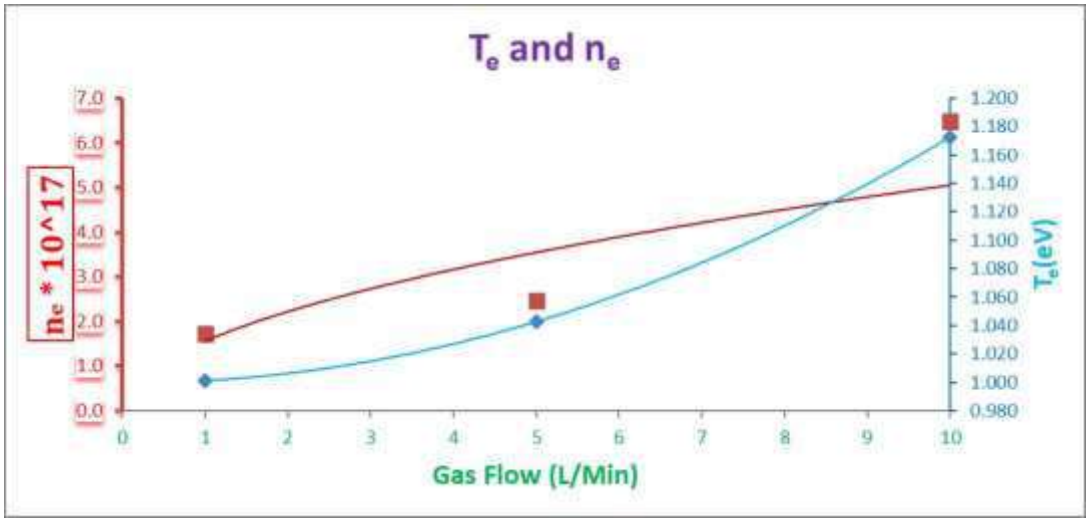

Figure 07- DBD electron temperature T_e and density n_e as a function of gas flow rate for (1, 5, and 10) L/min

Tables (2) show the predicted For Ar gas at various flow rates, electron density (n_e), electron temperature (T_e), plasma frequency (f_p), Debye length (λ_D), and Debye number were measured (N_D). Every estimated plasma parameter (λ_D, f_p, and N_D) satisfied the plasma requirements. Since it is proportional to n_e, it shows that f_p decreases as laser energy increases, but f_p and N_D have the opposite (decreasing) tendency [49].

Table 02- Plasma parameters for DBD at different argon gas flow rates at voltages 200 V

Gas Flow (L/Min)	T_e (eV)	n_e (cm^{-3})	f_p (Hz)	λ_D (cm)	N_d
10	1.173	6.49E+17	7.2E+12	9.3E-05	2.2E+06
5	1.043	2.47E+17	4.5E+12	1.4E-04	2.9E+06
1	1.001	1.73E+17	3.7E+12	1.7E-04	3.3E+06

B. Bactericidal actions

The two discharge types used to treat E. Coli suspended in water solutions with varying electrolytic conductivity levels were TS and DBD. Figures 8 and 9 display the results of measuring and expressing the bactericidal efficiency as a logarithmic decline. Both the TS and DBD systems showed a larger bactericidal effect; the TS system's efficiency was marginally better than that of the DBD system. The reason for this might be the electrospray effect, which involves spraying the solution of treated water into tiny, measured droplets. This increases the surface-to-volume ratio and makes it easier for reactive oxygen and nitrogen species (RONS) to mass transfer from the gas phase into the solution. The altered RONS chemistry was connected to the greater efficiency found in non-buffered solutions. Table 3 displays the expected efficacy using the discharge systems both before and following treatment [50].

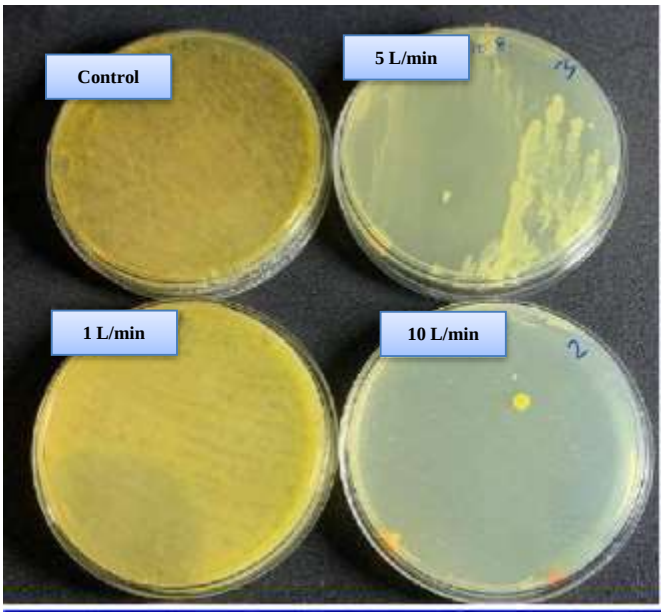

Figure 08- Inactivation of E. coli by Argon with TS discharge after various treatment flow rates

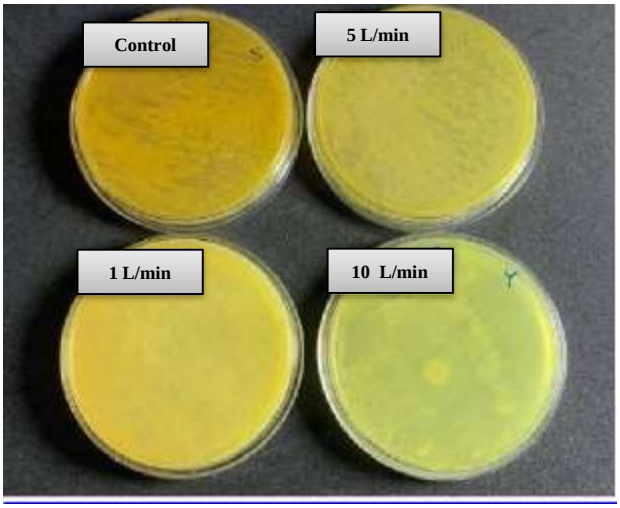

Figure 09- Inactivation of E. coli by Argon with DBD discharge after various treatment flow rates

Table 03- The number of *E. coli* with damaged cell membranes treated by TS and DBD discharges in the stationary growth phase.

No.	Plasma System	After effect	Before effect	Voltage	Plasma condition
1	TS	$60 * 10^8$	$5 * 10^9$	200V	10 L/min
2		$140 * 10^8$	$5 * 10^9$	200V	5 L/min
3		$280 * 10^8$	$5 * 10^9$	200V	1 L/min
1	DBD	$3* 10^8$	$5 * 10^9$	200V	10 L/min
2		$24 * 10^8$	$5 * 10^9$	200V	5 L/min
3		$76 * 10^8$	$5 * 10^9$	200V	1 L/min

Conclusion

Ar was used as the plasma-forming gas in the construction of a small device that could produce plasma jets in a TS and DBD. Emission and absorption spectroscopy was used to investigate the jet's component makeup. The number of nitrogen species and reactive oxygen species (RONS) formed were an order of magnitude more in water solutions than those generated by the plasma jet (PJ), were equivalent across the two TS systems of direct exposure, as were the chemical impacts and acidification of the water solution. Comparing the PJ's impact to the exposure to TS plasma indirectly, it was also lessened.

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Characterization of Metal Doped MEH-PPV Nanocomposite Thin Films for Environmental Gas Sensors

Mustafa M. Ali Hussein [1], Noor M. Jabbar [2]

Abstract

In this study, drop casting and spin coating techniques were both used to create MEH-PPV thin films. Some physical properties of MEH-PPV thin films produced by doping (0.03%, 0.06%, 0.09%, 0.12%) of FeCl3 were investigated using Hall Effect, UV-Vis and gas sensors. When the electrical properties were examined, it was found that the carrier mobility and conductivity increased when the FeCl3 concentration was increased. The band gap of the thin films was identified when the optical properties were investigated; the energy gap shrunk as the quantity of doping rose by 0.03%, 0.06%, 0.09%, and 0.12%. For sensors based on MEH-PPV, a noticeable response was observed when the sensor was exposed to H2S gas at the focus of 25 ppm. In contrast, MEH-PPV/FeCl3 (0.06%) had a maximum sensitivity of 57.36% at 175°C, with a reaction time of 9 seconds and a recovery time of 55.8 seconds.

Key Words

MEH-PPV; Thin film; Gas sensor; FeCl3 doping, UV-Vis, Hall Effect.

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Introduction

Poly (p-phenylene vinylene) and its related compounds are always of interest [1-3]. Multi-coupled organic polymers with electrical conductivity and nonlinear optical properties, as well as PPV, have attracted interest in both fundamental and practical materials science [4-7]. This interest is due to several reasons. Thus, PPV is informally produced with good purity and high partial weight. Also, it is completely insoluble and is relatively stable. However, as a water-soluble precursor polymer, it can be processed to generate films and fibers. It exhibits a relatively large optical bandgap (2.6 eV) and a bright yellow fluorescence [1]. Therefore, it is a formidable rival in light-emitting diodes and photovoltaic devices. Its ability to generate electrically conductive materials is easily deactivated.

By incorporating functional side groups, their physical and electrical characteristics can be extensively characterized [1]. Polymer screening has a number of benefits over metallic conductors. They are lightweight, practical, and, in most cases, chemically resistant at ambient temperature. Additionally, their conductivity, which is sometimes comparable to that of metals, is divisible by combining the active factors' concentrations [8]. Consideration of polymer conductivity is largely due to the breadth of conceivable applications enabled by their ease of synthesis, high interfacial stability, and long-term conductivity stability. They do, however, have the drawback of being insoluble and non-fusible [9]. Electric conductivity, photoluminescence, and photoconductivity are only a few of the many photoelectric features that the green electroactive polymer PPV exhibits. Over the years, a number of PPV compilation techniques have been documented. The initial test involved producing PPV by a direct chemical polymerization reaction. This method yields an insoluble powder as the final result, which has hampered the usage of polymers in many applications. [10].

Poly(p-phenylene) s, demonstrate nonlinear electrical, electrical, and optical properties to be excellent applications candidates in supercapacitors, transistors with a field effect , rechargeable batteries, electrical display devices, sensors and LEDs [11]. J. S. Shankar et al. conduct a scientific investigation on the effects of ZnO Nanoparticles and Poly's optical characteristics [2 Methoxy (5, 2′ Ethylhexyloxy)-P-Phenylenevinylene] (MEH-PPV). FTIR spectroscopy and X-ray diffraction were used to examine the hybrid composites' structural features. To investigate the optical properties of nanocomposite, the photoluminescence emission and UV absorption spectra were examined. ZnO nanorods allowed for a shift in emission wavelength, color-tuning capability, and multicolor emission. [12]. I. M. Ibrahim et

al. enhanced the conductive polymer's (MEH-PPV) H_2S gas sensing performance by adding TiO_2. First, chloroform solvent was used to dissolve the polymer of organic conjugated (2-methoxy-5- (2'-ethythexyloxy) - 1, 4 phenylenevinylene) and TiO_2. To create simple and affordable sensors, the two solutions (MEH-PPV) and TiO_2 were combined in a volume ratio of (0.002 and 0.008), respectively.

When exposed to H_2S gas at a concentration of 25 ppm, the (MEH-PPV/TiO2) based sensors also demonstrated notable reactions. The MEH-PPV/ (0.008) TiO_2 maximum sensitivity was 528.1 at operating temperature, with response and recovery times of 21.5 and 3.8 seconds, respectively [13]. In poly (p-phenylene vinylene) [PPV] doped by $FeCl_3$, the evolution of the density of states (DOS) and conductivity as functions of well regulated doping levels has been studied. It continues to be non-metallic at doping levels as high as 0.2 holes per monomer. According to the electrochemical gated transistor data, a comparable, almost linear increase in DOS has been seen in this system as a function of charges per unit volume. [14].

Experimental Part

Materials

The main polymeric material MEH-PPV supplied (American Dye source, Inc.Canad) was used with a purity of (99.99%). $FeCl_3$ was also used, and the supplier was (THOMAS BAKER-INDIA) with a purity of (98%).

Preparations of samples

The basis material for the created thin films in this study was a glass slide. The glass slides were divided into pieces measuring (2.5 * 2.5) cm^2 and (1 * 1) cm^2 before being cleaned in accordance with the prescribed steps (using distilled water to rinse, soaking in an organic ultrasonic solvent, and drying in a centrifuge or hot air) to prepare them for film coating. While the doping agent (210 mg MEH-PPV) was created by dissolving it in a solution of toluene (25 mg), the doping agent (80 mg FeCl3) was created by dissolving it in a mixture of tolueneand methanol. $FeCl_3$ was added to the MEH-PPV solution in varying weight percentages (0.03%, 0.06%, 0.09%, and 0.12%). The produced solutions were heated to room temperature, agitated for 10 hours on a stirrer, and then left to rest for 5 days. To check the UV, drop-casting was initiated. and gas sensor after the solution preparation process was finished. Additionally, the

spin coating procedure is for assessing the Hall Effect. Use clean glass bases to develop evenly thin coatings on the slides. The thin films created using the drop casting technique were dried for an hour in the oven at a temperature of 60°C. For the thin films created using the spin coating technique, the glass substrates were placed correctly on the magnetic platform that can rotate at high speeds. A micropipette was used to dry the required volume of solution, which was then placed in the center of the glass base and started rotating at a speed of 1000 (rotation per minute). The glass substrates are taken out of the device after the rotating period is finished.

Figure 01- In the present experiment, a spin coating apparatus was used.

Results and Disscution

Electrical Properties

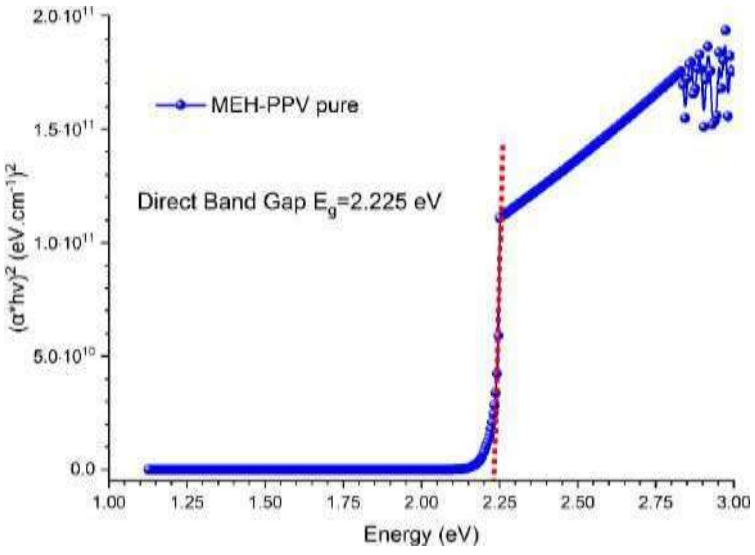
The Hall's effect on the samples was studied, and it was noted that the carrier mobility begins to increase when adding the doped material ($FeCl_3$) at different rates (0.03, 0.06, 0.09, and 0.12%). The conductivity also increases when adding $FeCl_3$. It was found that the Hall coefficient was positive for the first two samples and negative for the rest of the samples. This indicates that the samples with type p holes are the majority charge carriers, and the n-type samples with electrons are the majority charge carriers. The samples serve as p-type and n-type semiconductors. Table 1 shows the Hall Effect measurements for all samples.

Table 01- Hall Effect Measurements for All Samples

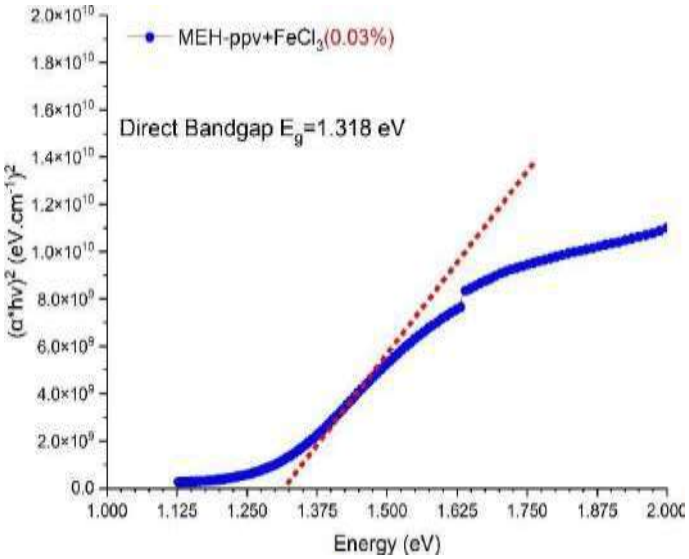
Doping percentage	nH(cm) $^{-3}$ *(E+15)	RH ($cm^{-3}C^{-1}$) *(E+02)	σ (Ω.cm) $^{-1}$	μ(cm^2/Vs) *(E+03)	Type
Pure	6.89	9.05	1.25	2.4	**P**
0.03%	5.70	1.10	1.71	2.85	**P**
0.06%	5.62	-1.11	2.57	2.86	**N**
0.09%	4.07	-1.53	2.60	3.07	**N**
0.12%	3.92	-1.59	2.69	3.33	**N**

Optical Properties

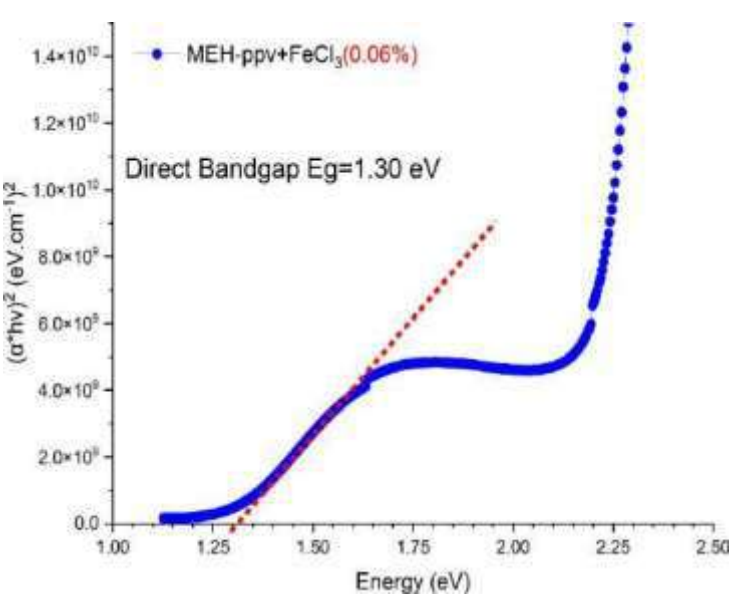
To check the optical properties that were obtained, UV-visible spectroscopy measurements were carried out on adevice (UV-Visible spectrophotometer) in the wavelength range (200-1200) nm. The basic absorption spectra data obtained from the analysis measurements of the visible and ultraviolet radiation are used to build variation graphs for each film $(\alpha h\nu)^2$ in order to compute the film band gap. The band gap values for the films are given as the energy values for the linear intersection of these graphs with the hν axis at $(\alpha h\nu = 0)$. This strategy is known as Tauc [15]. The comparative change of $(\alpha h\nu)^2$ vs. (hν) is shown in Figure 2. Additionally, Table (2) includes the frequency of the thin films band gap values.



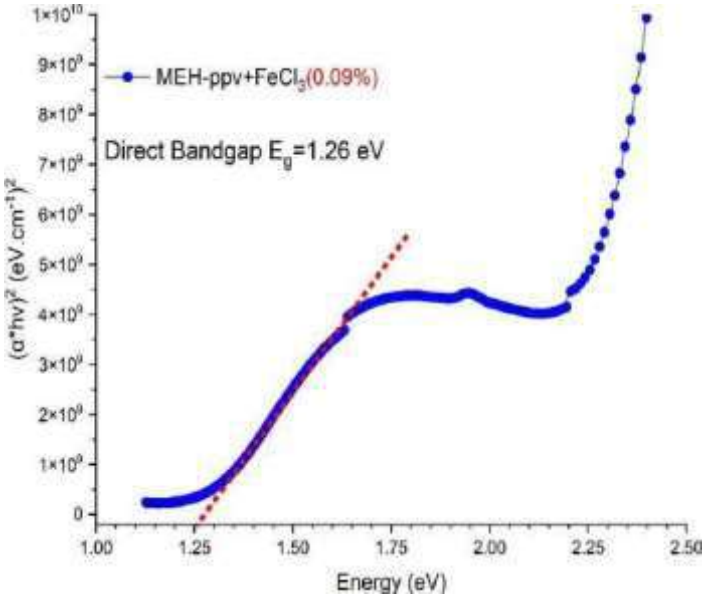
(a) MEH- PPV (pure)



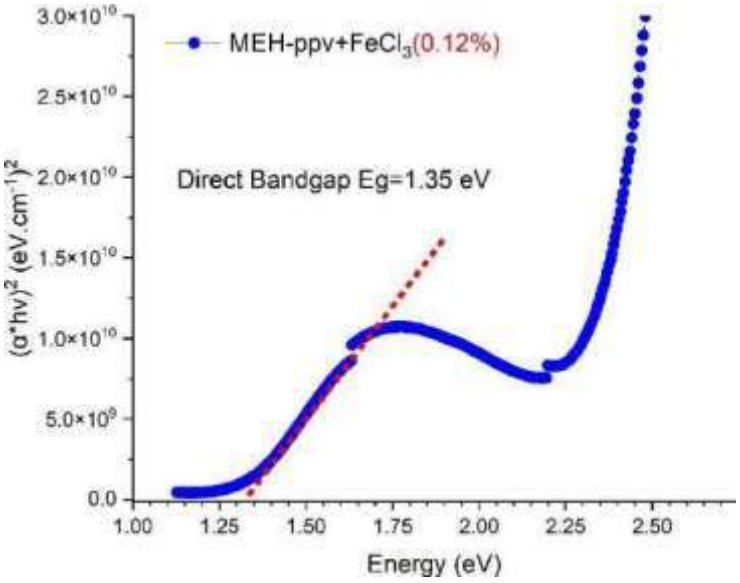
(b) MEH-PPV/FeCl3 (0.03%)



(c) MEH-PPV/FeCl3 (0.06%)



(d) MEH-PPV/FeCl3 (0.09%)



(e) MEH-PPV/FeCl3 (0.12%)

Figure 02- The change of (αhυ)2 versus hυ: (a)-(e)

Table 02- Band Gap Values of Thin Films

Doping percentage	Band Gap (eV)
Pure	2.225
0.03%	1.318
0.06%	1.30
0.09%	1.26
0.12%	1.35

Through the diagram and the table, we can see the energy gap in the MEH-PPV series, and it was (2.225 eV), and this value began to decrease when adding doping in different proportions, and this means that with the increase in the amount of doping, the energy gap decreases, and this decline is brought on by an increase in the crystal size values, which is consistent with previous literary studies [12].

Measurements from gas sensors

The sensor system for MEH-PPV/$FeCl_3$ is deposited on a microscope slide in order to sense H_2S gas at a concentration of 25 parts per million at different operating temperatures of 125, 150, and 175 °C. The figures (3,4, 5 and 6) show the resistance variation as a function of time with the gas valve on/off MEH-PPV/$Fecl_3$ in different weight percentages (0.03, 0.06, 0.09, and 0.12%). It turns out that there is an increase in the conductivity or a decrease in the resistance value of the devices when exposed to H_2S gas, the gas ON. Also, the resistance increases when the gas is turned off (gas off). Thus, the membrane sensing mechanism of MEH-PPV/$FeCl_3$ nanocomposites for molecular gas can return to the adsorption of molecular gases on the surface of the membranes.

Molecular gases interact with the polymer's electron network (π) to filter the nanoparticles and thus result in electron capture, depending on the nature of the gaseous molecule's reduction. This leads to a decrease in the resistance [16], and increased conductivity or decreased resistance due to the interaction of protons. The (H^+) polymer is protonated after the dissociation of H_2S. From the reaction of the proton, sime- quinone radicals are generated, and these radicals are conjugated, thus the conductivity increases [17]. Table (3) illustrates the response and recovery times for various MEH-PPV/$FeCl_3$ ratios and operating temperatures, whereas Table (4) illustrates the sensitivity of MEH-PPV/$FeCl_3$ for various ratios and operating temperatures.

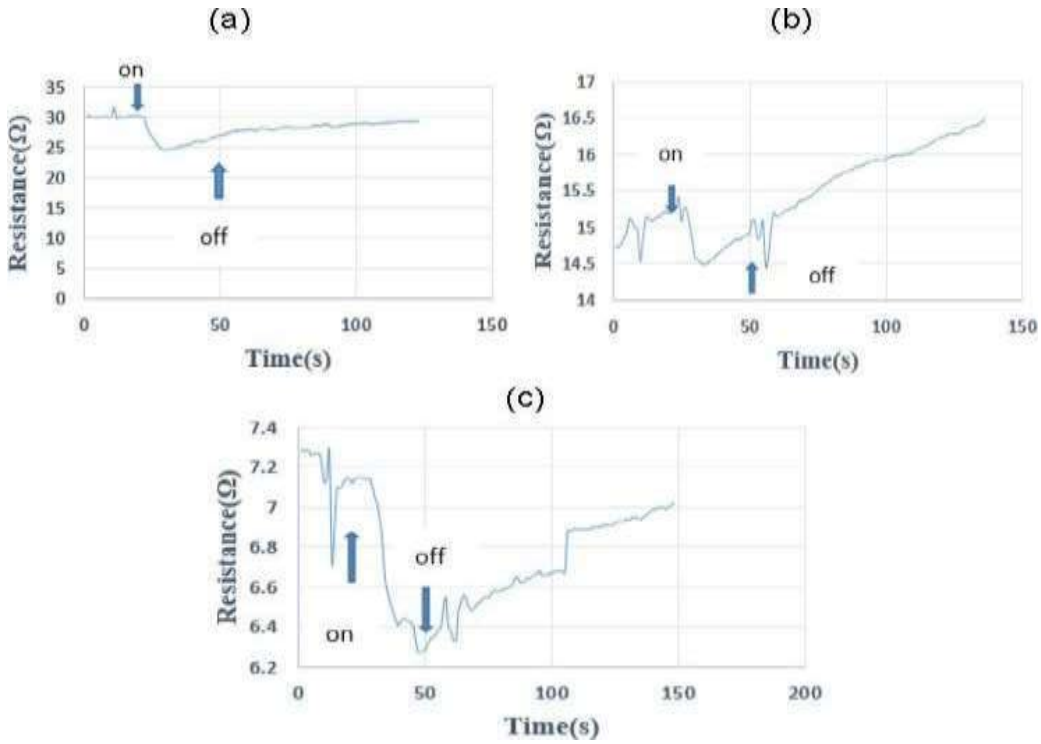


Figure 03- Resistance for thin films made of MEH-PPV/FeCl3 (0.03%) as a function of time at 125°C, 150°C, and 175°C

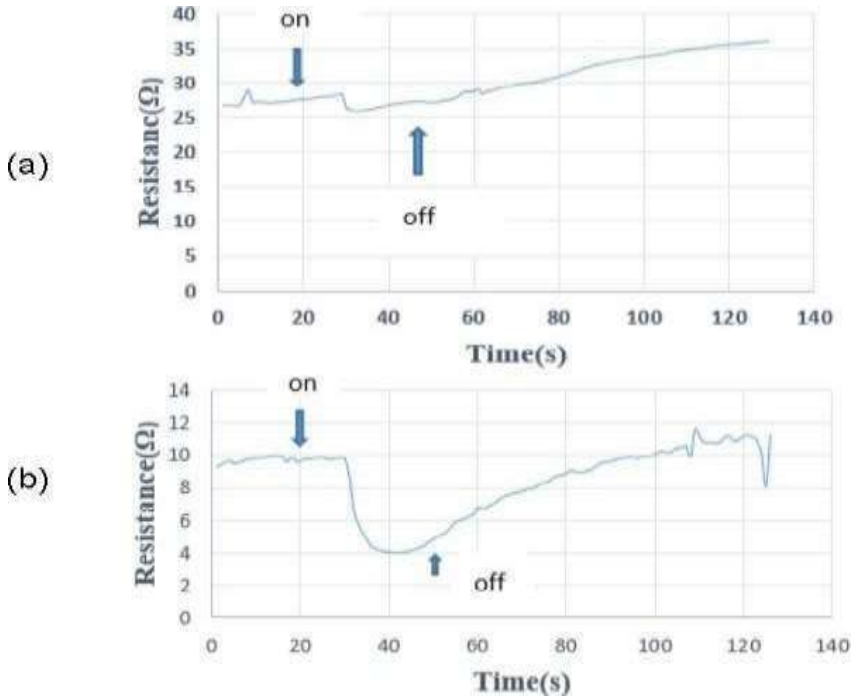


Figure 04- Resistance for thin films made of MEH-PPV/FeCl3 (0.06%) as a function of time at 150°C, and 175°C

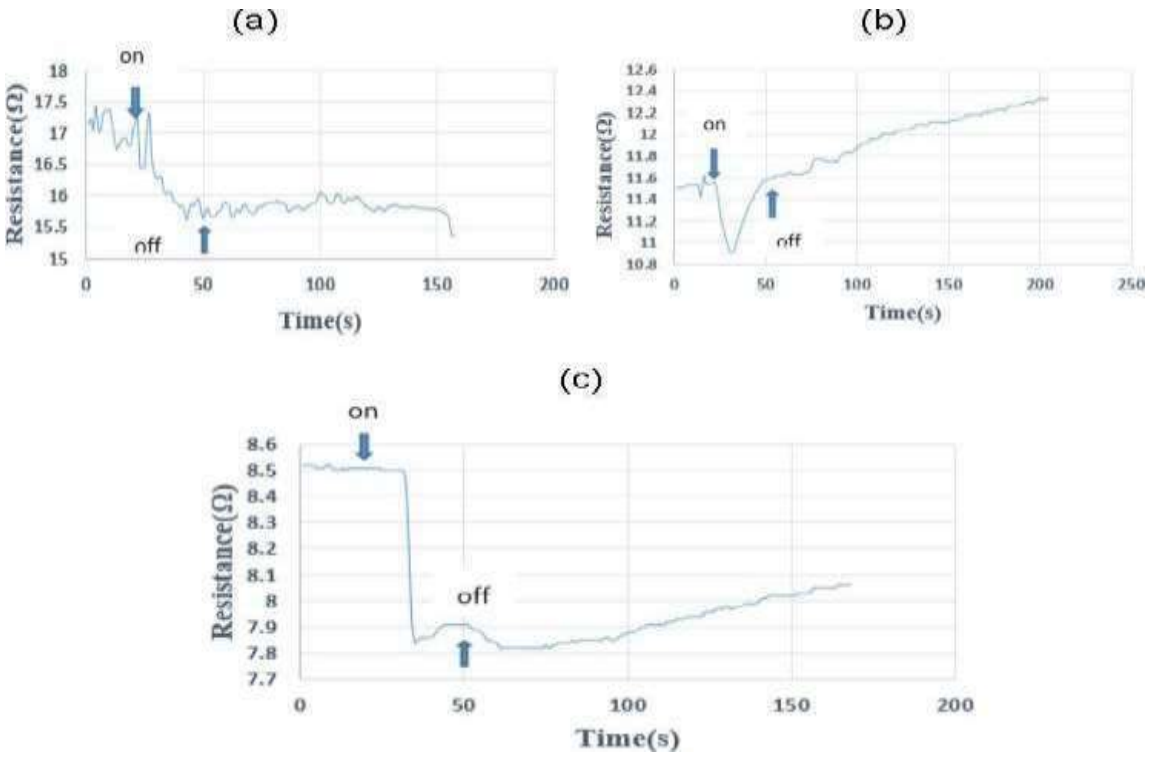


Figure 05. Resistance for Thin Films made of MEH-PPV/FeCl3 (0.09%) as a function of time at 125°C, 150°C, and175°C

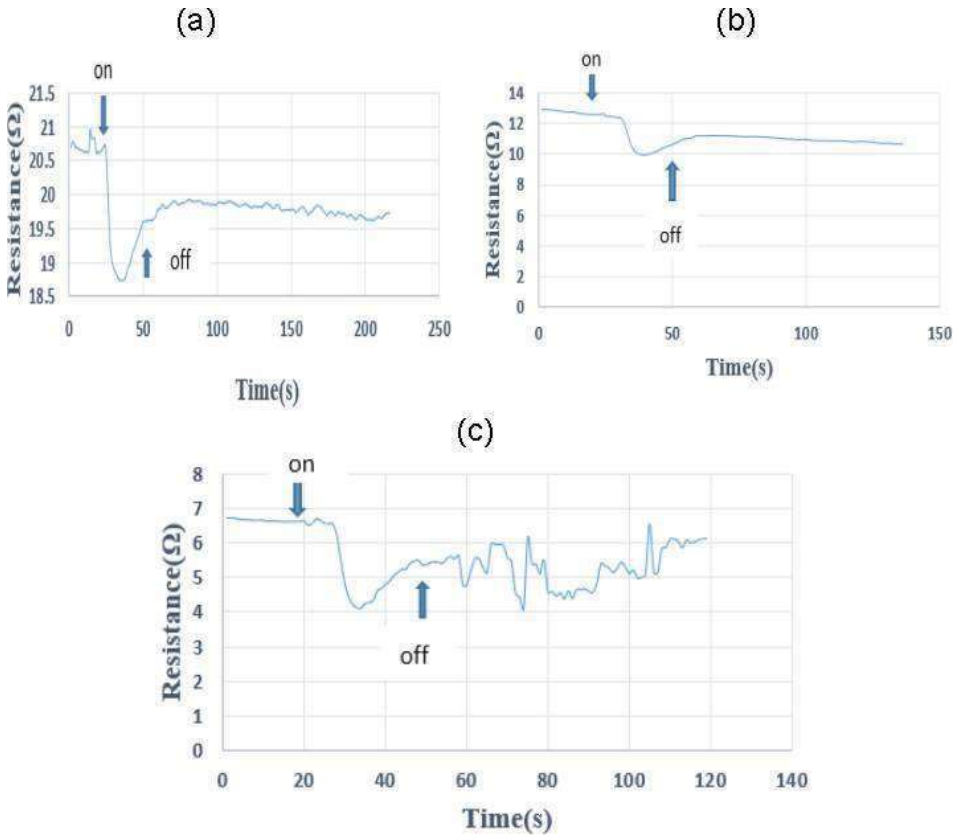


Figure 06- Resistance for Thin Films made of MEH-PPV/FeCl3 (0.12%) as a function of time at 125°C, 150°C, and 175°C

Table 03- Results of MEH-PPV/FeCl3 Thin Films for H2S Gas at Different Working Temperatures with DifferentFeCl3 Ratios (0.03, 0.06, 0.09 and 0.12%)

Operation Temp.	125°C		150°C		175°C	
Doping percentage	ζRes.(s)	ζ Rec. (s)	ζRes.(s)	ζ Rec. (s)	ζRes.(s)	ζ Rec. (s)
0.03%	7.2	64.8	5.4	61.2	18	92.7
0.06%	-	-	4.5	62.1	9	55.8
0.09%	16.2	94.5	9	63	9.9	73.8
0.12%	**9**	**61.2**	**7.2**	**57.6**	**7. 2**	61.2

Table 04- Sensitivity Results of MEH-PPV/FeCl3 Thin Films with Varying Aspect Ratios (0.03, 0.06, 0.09% and 0.12%) FeC l3 for H2S Gas at Different Operating Temperatures. Sensitivity %

Doping percentage	125°C	150°C	175°C
0.03%	17.815	4.849	12.184
0.06%	-	6.609	57.361
0.09%	8.261	5.372	7.529
0.12%	9.002	18.372	36.390

Figure 7, shows the absorbance changes vs. wavelength for pure and doped samples to compare the results with those of energy gap stated in figure 2.

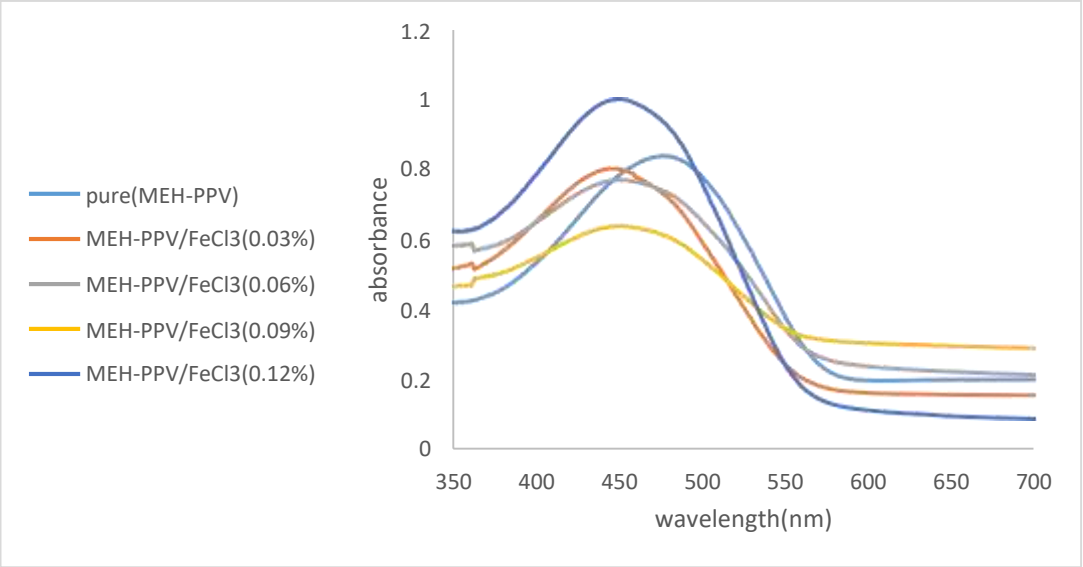


Figure 07- The absorbance of UV-Vis Spectra of all studied states

Conclusıons

Spin coating and drop casting techniques were used to successfully create thin films of the MEH-PPV/$FeCl_3$ nanocomposite. Hall Effect samples for all films created by combining MEH-PPV and $FeCl_3$ at various weight ratios are displayed. The findings show that there is an increase in carrier mobility and an increase in conductivity when $FeCl_3$ concentration increases. It was also observed from the UV-Vis measurement that there was an increase in the crystal size values and that the increase started with the increase in the amount of dopants.Thus, the energy gap obtained from UV-visible spectroscopy decreased with the increase in the amount of doping, and this is consistent with previous literature studies. For gas sensor sensitivity enhancement by adding $Fecl_3$ in different proportions to the MEH-PPV organic polymer, the highest sensitivity level is 57.361%, with a response time of 9 seconds, and the recovery time is 55.8 seconds for MEH-PPV/$FeCl_3$ (0.06%) at 175 °C.

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Characterizations of Hydroxyapatite Nanoparticles Synthesized from Fishbone Using the Calcination and Grinding Method: Structural and Mechanical Analysis

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Ahmed Namah Mohamed [3] , Jafer Fahdel Odah [4]

Abstract

Hydroxyapatite (HAp) is derived from fish bones, which serve as a natural reservoir of calcium, due to its resemblance to the inorganic mineral constituent present in bone and teeth. As a result, it is becoming a prominent bio ceramic material with a wide range of biomedical uses, especially in the fields of orthopedics and dentistry. The calcination and grinding process was used to successfully manufacture nanoparticles from carp spine bone. Not only is it highly valuable on international markets, but it is also one of the most essential species for fish farms. Nano-hydroxyapatite ceramics were examined for their size, shape, and chemical composition. The optical microscopy revealed that the main particles are round. XRD scatter plot and Fourier transform infrared spectra verified the hydroxyapatite structure. The average Vickers hardness value was 55 MPa, which is significantly higher than the hardness of human femoral cortical bone. Processing bio-ceramics derived from natural sources in an environmentally friendly, straightforward manner free of hazardous chemicals is the subject of this groundbreaking research.

Key Words

Hydroxyapatite; Calcination Method; Vickers Hardness; Fishbone; Nanoparticles.

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Introduction

A lot of work has gone into studying how to synthesize different bio ceramics for use in medicine during the last several decades. Orthopedics and dentistry are two of the many biomedical fields that make extensive use of hydroxyapatite (HAp), the most rapidly growing class of bio ceramics. Hydroxyapatite (HAp) has been widely utilized in several applications involving the replacement and rebuilding of hard tissues. These applications include coatings for implants, substitutes for bones, and dental uses [1, 2]. This is because it closely resembles the inorganic mineral component found in these tissues.Antibiotics, hormones, enzymes, antibody fragments, steroids, and many more pharmacological compounds bind strongly to its porous nature [3]. As a result, HAp has the potential to be used to administer pharmacological compounds in various therapeutic applications, with the added benefit of prolonged release. These applications include the treatment of osseous malignancies, osteomyelitis, osteoporosis, and other conditions were filling skeletal deficiencies necessitate local administration and is successful. The significance of this substance for numerous biomedical applications makes new, cost-effective, and flexible methods of HAp production appealing.

Though it is most commonly expressed as $Ca_{10}(PO_4)_6(OH)_2$, hydroxyapatite's chemical formula is $Ca_5(PO_4)_3OH$. The inorganic component hydroxyapatite is naturally present in human hard tissues like bone and teeth. Implants inside the human body often make use of these materials. Natural hydroxyapatite can be made from a variety of sources, including coral, poultry, fish, and eggshells. [4].

Equations (1) and (2) show the dehydroxylation that occurs during the heating transformation of the HAp ($Ca_{10}(PO_4)_6(OH)_2$) phase, while equations [5] show the breakup of HAp and oxyapatite ($Ca_{10}(PO_4)_6O$).

$Ca_{10}(PO_4)_6(OH)_2 \quad Ca_{10}(PO_4)_6(OH)2{-}2xO_xA_x + xH_2O$ ……. (1)

$Ca_{10}(PO_4)_6(OH)_{2-2x}O_xA_x \quad Ca_{10}(PO_4)_6O + (1\text{-}x)\ H_2O$ ……. (2)

$Ca_{10}(PO_4)_6O \quad 2Ca_3(PO_4)_2 + Ca_4(PO_4)_2O$ ……. (3)

$Ca_{10}(PO_4)_6(OH)_2 \quad 2Ca_3(PO_4)_2 + Ca_4(PO_4)_2O + H_2O$ ……. (4)

Oxy hydroxyapatite (OHA) is represented as $Ca_{10}(PO_4)_6(OH)_{2-2x}O_xA_x$, and A stands for hydrogen vacancies [6]. Eq. (5) [7] shows an alternative breakdown of oxy apatite.

$Ca_{10}(PO_4)_6O \quad 3Ca_3(PO_4)_2 + CaO$ .....(5)

Sobczak-Kupiec et al. discovered that the structure and physicochemical characteristics of HAp differ according on the method employed. The synthetic hydroxyapatite had properties of reduced crystallinity, substantial porosity, and increased surface area. Conversely, HAp exhibits maximum crystallinity when it is derived from animal bone through calcination at a temperature of 800˚C [8]. By creating a HAp interfacial layer, it enables the chemical bonding of hydroxyapatite with the adjacent hard tissues [9]. Due to its similarity in physical and chemical qualities to bone, natural hydroxyapatite is considered biocompatible [11].

For polarized bone replacements, Bowen and colleagues investigated the connection between composition, dielectric, and piezoelectric composites. Adding $BaTiO_3$ to a material raises its permittivity and ac conductivity, as previously noted [12]. In conclusion, polarized bone substitutes made of HAp-$BaTiO_3$ composites are a viable option [13]. Gao et al. produced three porous scaffolds using the three-dimensional gel-lamination process and sintering bovine bone. The results showed that three different kinds of HAp scaffolds promoted osteoblast adhesion, proliferation, and differentiation effectively [14]. Ceramic hydroxyapatite, made by sintering bovine bone, mimics the structure and porosity of natural bone. Ceramics can be impregnated with antibiotics or chemicals that enhance bone regeneration because of their porous nature and exceptional ability to absorb water and organic solvents [15].

Zhang and Darvell have stated that the initial Ca/P ratio, pH, and calcium concentration have an influence on the form and structural properties of hydroxyapatite whiskers [16]. The structure, which had the same crystallographic properties as HAp, remained unchanged despite modifications in these values. While [Ca] had little effect on the Ca/P ratio, it did improve with increasing pH and Ca/p. If the Ca/P was high and the pH was low, or vice versa, then the whiskers would be uniform. If the Ca/P was high and the pH was low, or vice versa, then the whiskers would be uniform. The formation of lath-like hydroxyapatite (HAP) was promoted under conditions of high initial pH and high calcium to phosphorus ratio (Ca/P), whereas the generation of branch-like whiskers and irregular plate-like particles occurred under conditions of low initial Ca/P and low initial pH. Lower [Ca] and higher Ca/P and pH increased preferred c-axis growth [14]. Werner and his colleagues used regulated pore-shaped HAp tapes in many layers to create osteo implants with different porosities [15].

The data show that sintering temperature affects HAp phase density, microstructure, and stability. The optimal temperature for sintering three-layered structures is 1250˚C for maximum flexural strength. A homogeneous three-layer structure with a single pore size has 40% lower flexural strength than a pore-graded structure. Macroporous hydroxyapatites (HAPs) allow

osteoblast-like cells to enter, attach, migrate, and proliferate in macropores and network linkages [17].

This work sought to calcine Carp scales and bones to make Hydroxyapatite (HAp), a natural substance. The HAp samples were then examined using scanning electron microscopy, FTIR, and XRD to further understand their microstructure and physical properties. Additionally, the completed goods' mechanical properties will be assessed.

Experimental part

In the experimental part of current work will illustrate in the Figure (1), which is a schematic diagram.

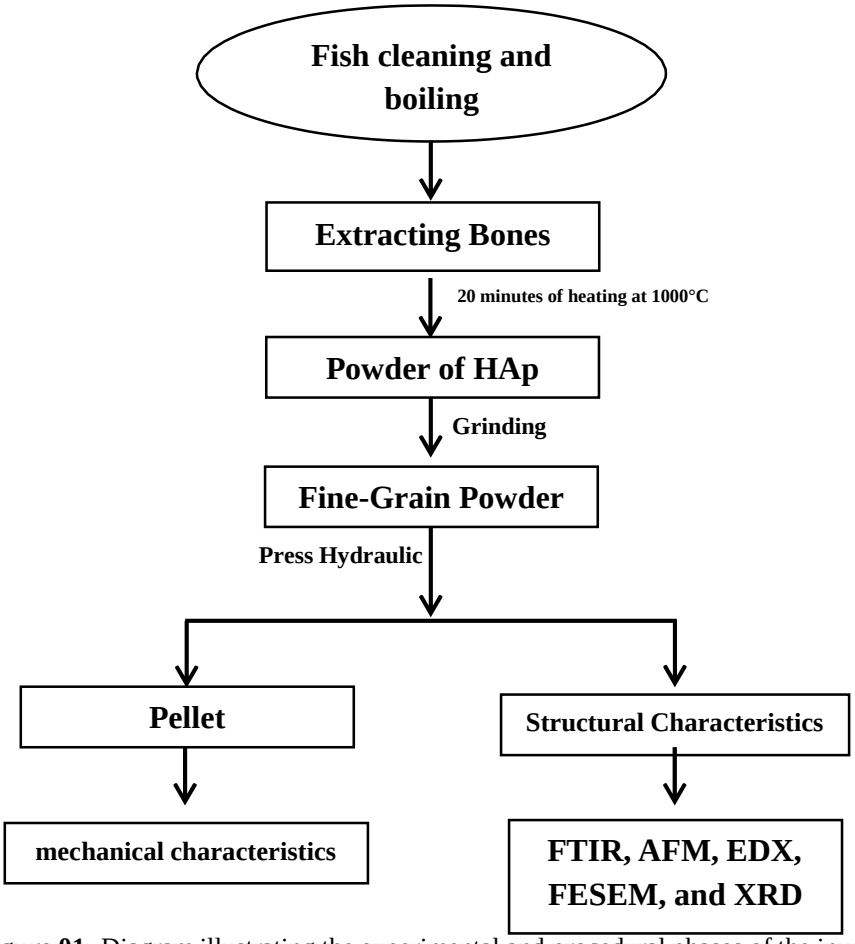


Figure 01- Diagram illustrating the experimental and procedural phases of the investigation

At first, the fish was cooked for 60 minutes in a domestic oven, which was set to maintain a constant temperature of approximately 220 ^{0}C. The bones of the cooked fish were extracted by simmering it in distilled water for two hours on a gentle heat following its natural cooling process. Lastly, ethanol was used to clean the boiled bones, removing any contaminants or suspended meat. The last step was to place the cleaned fish bones in a homemade ceramic

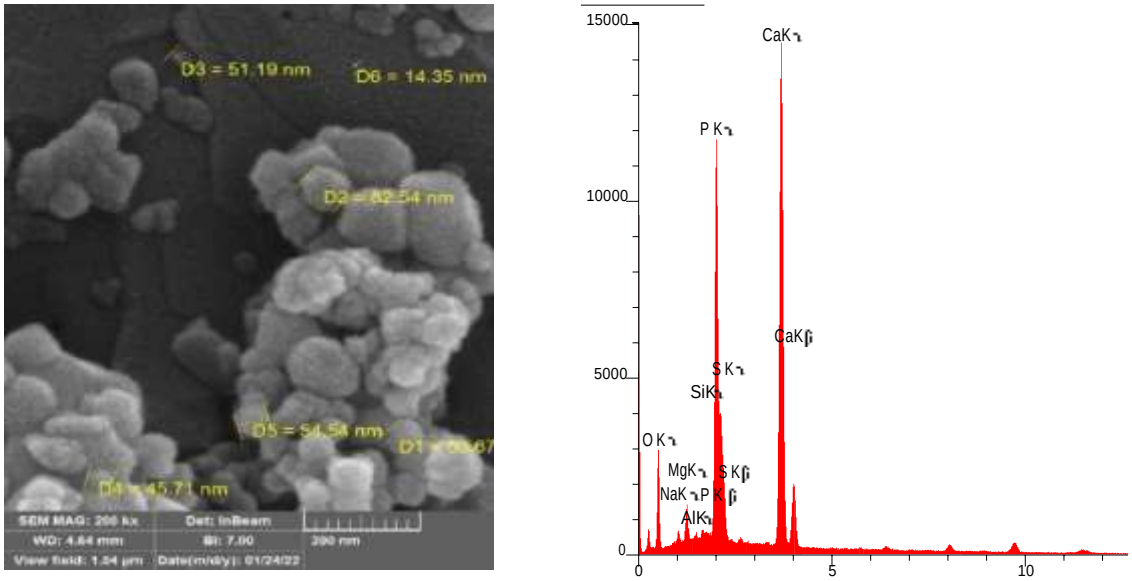
crucible and heat them to approximately 1000^{o}C using an oxy-acetylene flame held directly beneath the crucible. It will take 20 minutes for the fish bones to turn into a powdery, grayish consistency. Prior to conducting studies utilizing characterization techniques like XRD, FESEM, EDX, FTIR, and AFM, the powder was ground using a mortar to obtain a final fine sample. This sample was then transferred to a clean test tube to ensure it remained free of contaminants.

In order to obtain a consistent and uncontaminated sample, we employed a 2-ton hydraulic compressor to compact a portion of the finalized powder onto a pallet with a radius of 1 centimeter. Subsequently, we subjected it to calcination at a temperature of 1000 degrees Celsius, with the temperature rising at a rate of 10 degrees per minute over a duration of four hours. Subsequently, we allow the furnace to cool down organically without any attempt to open the door. It is now possible to conduct mechanical and characterization tests on the pellet.

The HAp structural and mechanical properties will be examined by the application of X-Ray Diffraction (XRD), Fourier Transform Infrared Spectroscopy (FTIR), Atomic Force Microscope (AFM), Scanning Electron Microscope (SEM), and the Vickers Hardness Test.

Results and Discussion

Using FESEM, we examined the produced hydroxyapatite's surface shape and crystal size. Images of FESEM and EDX spectra of raw hydroxyapatite from fishbones heated to 1000^{o}C are shown in Figure (2).



(a) FESEM **(b) EDX spectrum**

Figure 02- EDX and FESEM spectra of the finished product

Figure (2-a) shows that the hydroxyapatite microcrystals found in natural fishbone are incredibly tiny, measuring 45-55 nm in size, 18-25 nm in width, and several micrometers in length. The chemical compounds in raw fish bone gave the impression that the nanostructures were dense. Particles' propensity to crystallize and agglomerate at elevated temperatures is responsible for the development of these microstructures in the thermal process of generated hydroxyapatite. A powder that has been heated to a certain temperature will have spherical particles that are well defined, distributed, and oriented in a random fashion. The presence of aggregated nanoparticles was seen in certain areas, indicated by the presence of a small number of clusters of doped HAp nanoparticles. Figure (2-a) shows that the hydroxyapatite microcrystals found in natural fishbone are incredibly tiny, measuring 45-55 nm in size, 18-25 nm in width, and several micrometers in length. The chemical compounds in raw fish bone gave the impression that the nanostructures were dense. Particles' propensity to crystallize and agglomerate at elevated temperatures is responsible for the development of these microstructures in the thermal process of generated hydroxyapatite.

A powder that has been heated to a certain temperature will have spherical particles that are well defined, distributed, and oriented in a random fashion. Figure (2-b) shows that the EDX signals allowed us to calculate the Ca/P weight ratio for derived HAp, which was shown to be 1.67 at 1000 °C. In some areas, we could see clusters of doped HAp nanoparticles, which could mean that they agglomerated. When it comes to nanostructures, EDX analysis provides both qualitative and quantitative information about the elements that could be involved. The energy-dispersive X-ray (EDX) analysis revealed that the HAp nanostructures that were created solely included calcium, phosphorus, and oxygen. Through the use of atomic force microscopy, the morphology of the HAp surface topography has been examined. Figure (3) shows the outcomes of AFM examinations concerning the HAp's surface morphology.

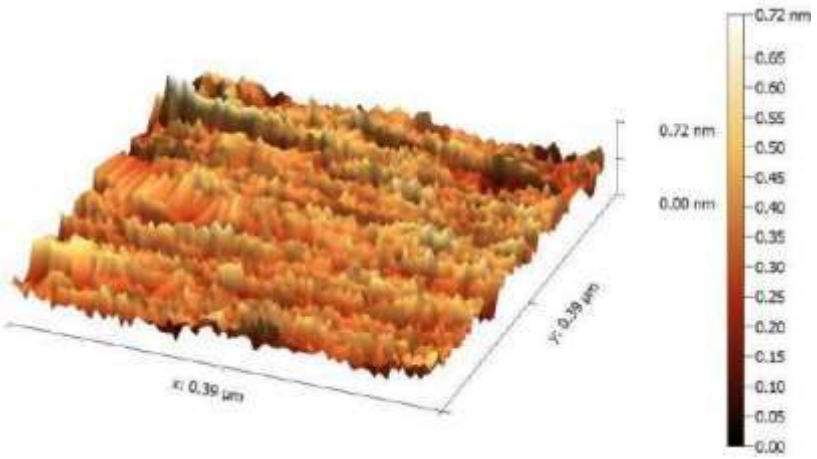


Figure 03- Advanced Fluorescence Microscopy of the Complete Sample

The AFM experiments highlighted that the HAp powder under investigation had the appearance of a layer that has been uniformly deposited. Also, the deposited sample is composed of evenly distributed nano-aggregates, and the 2D surface representation showed that there are no visible signs of cracks or fissures. The findings from AFM experiments corroborate those from SEM studies, which also showed that the coatings are homogeneous and consistent.

This is the XRD pattern of unprocessed fishbone from Figure 4. Files for calcite (5-0586) with the (JCPD card number) relate to the X-ray reflections.

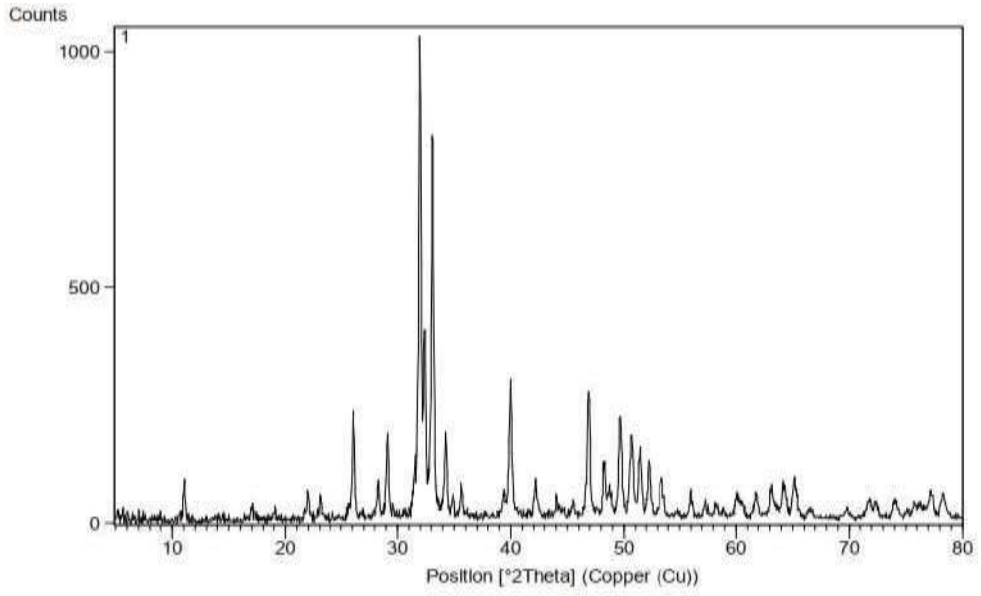


Figure 04- XRD analysis of the finished product

Figure. 4 shows that the solid substance that results from heating to 1000 °C is very porous, whitish, and fragile. There are reflections in the X-ray diffraction pattern that match the CaO files (JCPD-card number (37-1497)); picture (4) does not indicate any other species. These findings could be attributed to calcite not undergoing a complete transition because either the annealing temperature was too low or the duration was too short. The primary diffraction peaks closely match those of a normal hexagonal phase Ca $(OH)_2$ crystal, as indicated by the JCPDS card No. (87-0674). The crystal has a lattice parameter of a = b = 3.589 °A and c = 4.911 °A. The sole disparity lies in the heightened intensity of the (101) diffraction peak compared to the (001) peak, indicating a probable inclination of Ca $(OH)_2$ particles to grow along the c-axis. Both the particles' outer layers and the prevailing (101) surfaces are surrounded by (101) surfaces. Additionally, small quantities of $CaCO_3$ were identified, most likely formed through a chemical reaction between Ca $(OH)_2$ and CO_2 present in the atmosphere.

Fourier transform infrared spectroscopy (FTIR) was used to confirm the lack of organic components in the manufacturing process of HAp and to obtain reliable data on the vibrational source of phosphate and carbonate. Figure.5 displays the FTIR spectra of the raw fishbone and the resulting HAp at a temperature of 1000^{o}C.

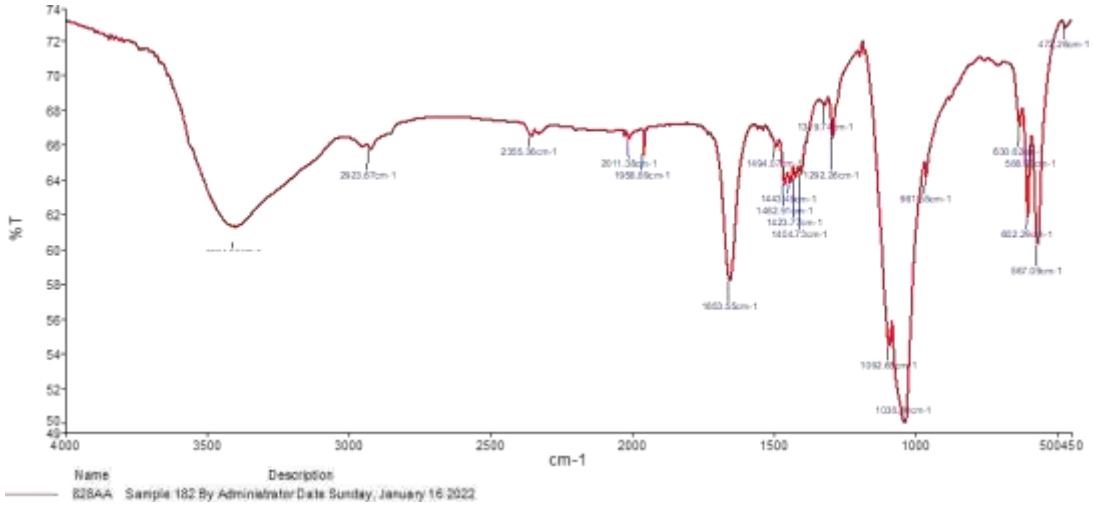


Figure 05- FTIR analysis of the finished product

The FT-IR spectrum of calcined bones, scales, and shells at 1000 °C showed the sole distinctive peak of HAp, as shown in figure.5. A wide range of spectra bands (600–3600 cm^{-1} and others at (601, 631, 873, 962, 962, 1027, 1088, 1413, 1454, 2034, 2157) cm^{-1} were in agreement with the corresponding bands in the HAp reference spectrum. At 1088 cm_{-1}, more beaks were noticed, which relate to the phosphate (PO_4) group. The creation of HAp crystals and the subsequent elimination of all organic matter from the source material (fishbone) could be to blame. Derived HAp has a significant quality: thermal stability. In addition, because the hydroxyl group was present, the FTIR spectra sample showed a peak at 631 cm^{-1} and a broad peak at 3200–3700 cm^{-1}. The CO_3 is responsible for the strong peaks at 1413 and 1457 cm^{-1} in the calcined fishbone spectra, as shown by the equation [18]:

HV=1.854 Pdav

This is where: HV stands for Vickers hardness, P for shed load (in N), and dav for average value of the two diagonals' lengths (d2, d1). We took hardness readings from the sample's edges and center, as well as from other locations, to establish a ballpark estimate of the hardness rate. Table (1) shows the average hardness as well as the results of three separate tests.

Table 01- The average Vickers hardness value for hydroxyapatite made from fishbone by thermal processing is 55 MPa.

Sample	First test (HV) (MPa)	Second test (HV) (MPa)	Third test (HV) (MPa)	Average Value of (HV) (MPa)
HAp	92.03	35.61	38.16	55.20

The result has a Vickers hardness that is significantly higher than that of human femoral cortical bone. The cortical bone is somewhat tougher by 10-15% compared to the neighboring fishbone. The calcium composition of the two bone varieties is the underlying cause of this distinction [19]. After the re-crushing process, the Vickers hardness readings showed an improvement.

Conclusions

The underutilization of leftover fish bones is a problem that stems from activities related to fishing. Hydroxyapatite (HAp) is produced by the synthesis of calcium, a naturally occurring element present in fish bones.

In this study, hydroxyapatite nanoparticles were produced using a fishbone as a natural source material. The process of calcination and grinding, utilizing a Carps fish, was effectively employed to produce nanoparticles. An analysis was conducted on the nano-hydroxyapatite ceramics to assess their dimensions, morphology, and chemical makeup. The results suggest that the fishbone synthesis undergoes calcination at a temperature of 1000 °C, resulting in a significant production of hydroxyapatite. SEM analysis confirmed that the primary particles have a spherical morphology with a diameter ranging from 18 to 50 nanometers. The mean Vickers hardness value was 55 MPa, which is much greater than the hardness of human femoral cortical bone. In comparison to fishbone, cortical bone is 10-15% more resilient. The disparity between the two types of bones can be attributed to the differing calcium composition. Following the procedure of re-crushing, the Vickers hardness values demonstrated enhancement.

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DC Electrical Discharge in a Metal Pin–Water Electrode System at Atmospheric Pressure

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Abstract

The discharge-liquid interactions represent one important topic in plasma technology and applications. In this research, the influence of added NaCl to distilled water on the discharge characteristics that formed in the gap between pin-water surfaces was investigated in more detail. Its electrical and optical characteristics define them. The optical results detected that the emission intensity of atomic and ionic nitrogen peaks decreased with increases in NaCl concentrations. the changing of the liquid conductivity (by for adding NaCl) causes to change in the discharge dynamics formed in the gap of the pin-water surface.

Key Words

Plasma liquid interaction; Optical Emission Spectroscopy; NaCl Concentrations; Current-Voltage Curve, Plasma parameters.

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Introduction

In plasma science and technology, interactions between plasma and liquid are becoming more and more significant. The generation of nitric acid in the air by an electric spark was the subject of Cavendish's well-known 1785 paper "Experiments on Air," which may be the earliest report of plasma-liquid interaction [1-4]. Complex physical and chemical processes arise from the interactions between plasma and liquid. The aforementioned processes commence with gas-phase chemistry and subsequently advance to encompass multi-phase species transport, as well as mass and heat transfer [5-12]. Furthermore, they involve interfacial reactions and very intricate liquid phase chemistry. The chemical processes occurring in low-temperature plasma sources operating in atmospheric air conditions, commonly referred to as cold atmospheric plasmas, are predominantly governed by reactive oxygen and nitrogen species [13-16].

These species are either produced within the liquid phase through secondary reactions or transferred from the plasma phase/gas to the liquid phase [17, 18]. Commonly, these discharges can be initiated within the liquid medium through the utilization of a dielectric barrier discharge [19-21] or a gliding arc discharge. In this configuration, one electrode is positioned above the liquid while the liquid's surface as the opposing electrode. They can also be started near the water's surface [22]. Investigated configurations included electro-spraying [23], high-voltage-fed hollow needles, and plasma jets. DC, AC, or pulsed high voltage can be used to energize the discharges [29-31]. The interplay between non-equilibrium plasmas and liquid states is crucial for a wide range of applications, including environmental remediation, material research, and medical care [32-38]. Extensive research has been conducted by numerous scholars on the subject of non-thermal plasmas (NTPs) both within liquids and in contact with them. The fundamental factor contributing to this phenomenon is the atypical chemical reactivity exhibited by liquids upon contact with plasma. Strong UV radiation, shock waves, and reactive radicals (OH, atomic oxygen, hydrogen peroxide, etc.) are produced by discharges in and in contact with liquids, and these substances are thought to be useful in the inactivation or conversion of a variety of biological and chemical materials [39-42].

NTPs have been identified as a new and promising technique with a wide range of applications, including the degradation of pollutants in water and biomedical applications [43, 44]. Research on plasma medicine, which combines biology, medicine, and plasma science and technology, is a fast-growing field [45]. Atmospheric pressure plasma jets (APPJs) are widely recognized as the most prevalent and valuable devices within this domain. Extensive research has been conducted to comprehensively investigate various aspects of APPJs, including species

generation, transportation, fundamental physics, and interactions with liquid media. This observation holds, although the existence a diverse range of atmospheric pressure plasma generators that have been developed for various uses [46-50].

This study aims to examine the impact of different NaCl concentrations on the discharge characteristics of the Pin-Liquid arrangement at atmospheric pressure in a more comprehensive manner. Furthermore, the experimental configuration is examined, and Optical Emission Spectroscopy (OES) is used to analyze plasma characteristics by studying the spectral lines emitted by nitrogen atoms in the vicinity of the plasma.

Theoretical consideration

Utilizing Optical Emission Spectroscopy (OES) One of the most widely used techniques for diagnosing plasma is the measurement of continuum, half-width lines, line shifts, and emission or absorption intensities. The emission of light from plasma occurs primarily through the release of photons when atoms or molecules, which have been excited to a higher energy state by electrons, subsequently transition to a lower energy state. This phenomenon is well-documented in the literature [19-22]. The composition of the producing species can be inferred by analyzing photon energy, the wavelength of light, and spectral emission data. Optical Discharge Spectroscopy falls into both the ultraviolet (130-800 nm) and electromagnetic light spectrums [5,9]. OES is used to gather information about the density, type, and electron temperature of the plasma [23].

1. Electron Temperature (T_e)

The electron temperature (T_e) in (eV) was one of the most crucial parameters used to study the plasma parameters. T_e was computed using the Boltzmann plot method as follows

$$\ln\left(\frac{I_z\lambda_{ki,z}}{g_{k,z}A_{ki,z}}\right) = -\frac{1}{k_B T_e}E_{k,z} + \ln\left(\frac{hcL_{n,z}}{4\pi P_Z}\right) \qquad (1)$$

Where the index Z, c, k_B, L, h, E_{kZ} and g_{kZ} shows the ionization state linked to the species, the speed of light, the Boltzmann constant, the plasma's characteristic length, the Planck constant, the energy and degeneracy of the upper energy level k, and the integrated intensity I_Z of a species in the ionization stage Z. In ionization stage Z, P_Z stands for the partition function for species. In terms of energy units, the electron temperature (T_e) is linked to the slope ($-1/T_e$) of the line that fits the relationship between $\ln\left(\frac{hcL_{n,z}}{4\pi P_Z}\right)$ and $E_{k,z}$ [27].

2. Electron Number Density

Lorentzian fitting and finding the full width at half maximum (FWHM) were used to determine number density of electron (n_e) (in cm^{-3}) with a variation of NaCl concentrations using stark broadening that are collected through the following relation [30, 43, 44]:

$$n_e\,(cm)^{-3} = \left[\frac{\Delta\lambda}{2\omega_S(\lambda_D T_e)}\right] N_r \qquad (2)$$

This method is more efficient than others and offer facts with high accuracy. In equation (2), $\Delta\lambda$ refers to the Stark full-width at half-maximum (FHWM), N_r represents the reference number density of electrons that equals ($10^{16}\ cm^{-3}$) for neutral atoms and ($10^{17}\ cm^{-3}$) for singly charged ions, and ω_s is the electron impact parameter, tabulated in Griem [22].

3. Plasma Frequency

According to the n_e, the plasma frequency (ω_p) was calculated as [11,25]:

$$\omega_p = \sqrt{\frac{e^2 n_e}{m_e \varepsilon_0}} \qquad (3)$$

Where m_e, ε_0, n_e, and e are electron mass, vacuum permittivity, electron number density and electronic charged, respectively [27].

4. Debye Length

The electron Debye length (λ_D) (in m) was defined as a microscopic maximum spatial scale for the charge-separation which represents the shielding distance or the plasma sheath thickness which is calculated as [24-27]:

$$\lambda_D = \sqrt{\frac{\varepsilon_0 k_B T_e}{e^2 n_e}} = 7430\ \left(\frac{T_e\,(eV)}{n_e}\right)^{1/2} \qquad (4)$$

5. Plasma Parameter

Plasma parameter (N_D) means the number of particles in a Debye sphere, that is, in a sphere of radius equal to Debye length (λ_D), using the equation [25-27]:

$$N_D = \frac{4\pi}{3} n_e\,(\lambda_D)^3 \qquad (5)$$

Experimental Part

Figure (1) illustrated the single pin-to-liquid discharge device used in this work (which is, highly efficient, simple, easy to operate, and low cost). The cathode pin electrode (iron, 100mm length, 4mm radius) is supplied by a variable a high voltage D.C. power source (model HLD-20C Hi-Rikesuta, 0-25kV, 100μA) and below this cathode is placed a glass vessel which contains a 100ml of tap water with different NaCl concentrations (300,400,500) mg. The liquid interface and pin electrode are 5 mm apart. In the tap water, a circular anode disc with a 20mm radius and 2mm thickness is positioned at the bottom of the vessel.

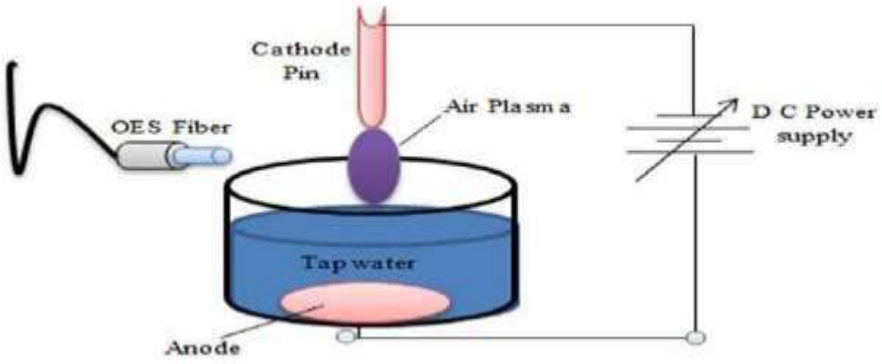

Figure 01- Experimental setup of Pin–Liquid configuration.

The discharge water container ignited between the metal pin electrode and the tap water surface. Photons with energy equal to the difference between the two energy states are released when electrons excite atoms or molecules and cause them to relax to a lower energy state. This process is observed by using OES (THOR model manufactured in Germany) to measure the emission light spectra from the air plasma [28]. The spectrometer was placed at the angle of 45^0 from the plasma column. The species' composition can be deduced through the analysis of photon energy, or the wavelength of light, and spectral emission data that are generated at the gap between the pin and the water's surface and are detected with varying dissolved concentrations of NaCl.

Results and Discussion

1. Emission Spectra

In general, there are three primary categories into which electrical discharges involving liquids can be separated: electrical discharges that originate above the liquid's surface, direct electrical discharges within the liquid, and electrical discharges that occur inside bubbles or vapor. Because of their comparable breakdown strengths in atmospheric gases, plasma

generation and gas phase breakdown above the liquid surface in the zone of electrical discharges that arise are largely comparable to gas electrical discharges. However, the presence of the liquid surface influences both the physical characteristics of the discharge and the chemical reactions taking place in the gas-liquid interface when it comes to electrical discharges above liquid-surfaces that use the liquid as an electrode. The reason is that ions, which have far less mobility than electrons in metals, carry the discharge current through the water electrode. In addition, compared to most metals, water is easier to deform and evaporate, and it has a lower secondary electron emission coefficient.

The emission spectra of air plasma in pin to liquid-discharge configuration at different NaCl concentrations are demonstrated in Figure (2). It may be pointed out from theses spectra that there are many emission peaks of neutral nitrogen lines (N2 I) corresponding to wavelengths of 337.13, 357.69, and 380.49nm. An emission-neutral atomic nitrogen peaks (N I) appear also at wavelengths 674.17 and 734.76 nm. The ionic nitrogen lines (N II) were also detected in the spectra at wavelengths 435.22, 631.88, 659.66, 697.56, 701.47, and 715.68 nm. All emissions peak intensity is decreased with increasing NaCl concentrations. As well as, the emission intensity of neutral emission peaks is much higher than that of the ionic emission peaks. These results indicated the plasma produced contained more atomic N than that of ionic N.

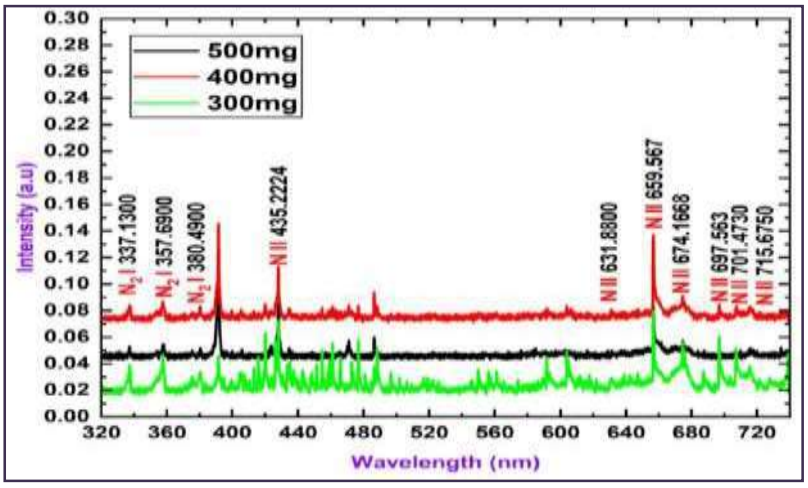

Figure 02- Emission spectra of glow discharge in pin-liquid configuration at different NaCl concentrations for wavelength range 320-740 nm.

2. Effect of NaCl Concentration on the Discharge Channel

A D.C. high-voltage was applied to the pin using a power source, to create a discharge between the cathode pin and the metal anode which is submerged in water with different NaCl concentration. The emission light of the resulting air plasma discharge was detected via a digital camera with higher resolutions. and illustrated in figure (3).

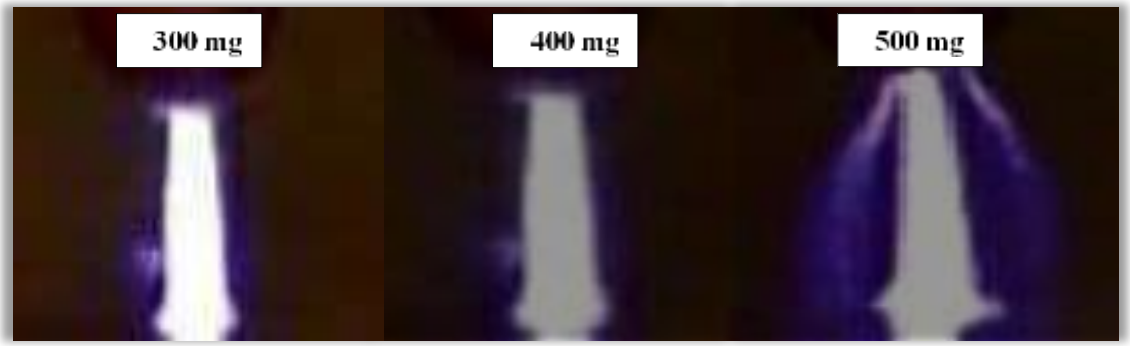

Figure 03- Typical images of the influence of NaCl concentration on the discharge channel in the gap between pin–liquid surface.

It is clear from the figure that the increasing NaCl concentration in water causes the dynamic change of the air plasma characteristics that formed in the gap between the pin electrode and the water surface. This result means that the conductivity of the liquid (water) will effect on the plasma characteristics that formed (where the increasing of NaCl concentration causes to increase in water conductivity).

Figure (4) illustrates the intensity of light emitted in the air gap between the pin electrode and surface water at different NaCl concentrations. One can be observed from this figure that the light intensity reduced with the increased of the NaCl concentration.

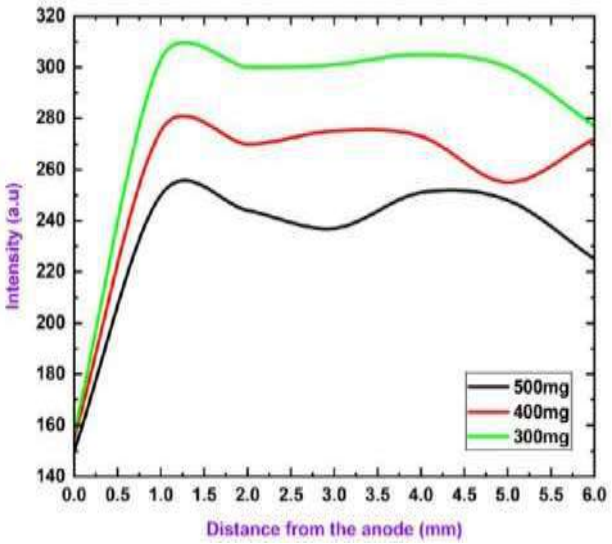

Figure 04- Influence of NaCl concentrations on the intensity of light emitted in the air gap between the pin electrode and water surface.

3. Current- Voltage Curve

Figure (5) demonstrates the effect of NaCl concentrations on the I-V curve. It can be observed that the abnormal glow discharge was established which causes increase in the discharge voltage with the current. We also observed that when the NaCl concentration is increased, the discharge current increases. This can be explained as follows; As the concentration of NaCl

increases, more Na+ and Cl- ions are present in the gas. This increased ion density can enhance the ionization process of the discharge, resulting in a higher discharge current.

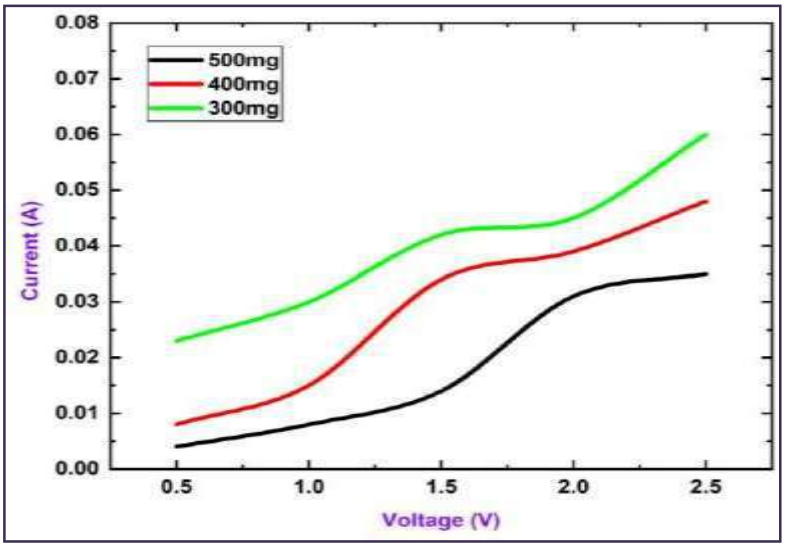

Figure -5 Influence of NaCl concentration on the I-V curve.

4. Influence of NaCl Concentrations on the Electron Temperature

In this section, the electric discharge that formed along the water surface with added of NaCl has been investigated. The T_e represents one of the most plasma parameters that calculate to detect of the plasma state. In this work, according to Boltzmann plot method (equation (1)) with the data tabulated in the table (1), the T_e was calculated (for all concentrations). Figure (6) shows the Boltzmann plots using the selected ionic nitrogen lines (N II) for the air plasma between pin electrode and water surface at three different NaCl concentrations. On the other hand, the n_e was determined. The variation of the T_e and electron number n_e with NaCl concentrations was plotted in figure (7). The data detected the fact that the T_e and n_e are reduced with increasing NaCl concentration.

Table 1- N II standard lines are used to calculate T_e, and their characteristics [29].

λ(nm)	$A_{mn} \times g_m$	E_i(eV)	E_j(eV)
631.8800	15.1×10^4	23.239296	25.200902
659.567	2.11×10^4	24.374310	26.253573
697.563	1.03×10^4	23.415330	25.192229
701.4730	175×10^4	23.425232	25.192229
715.6750	11.4×10^4	24.531351	26.263284

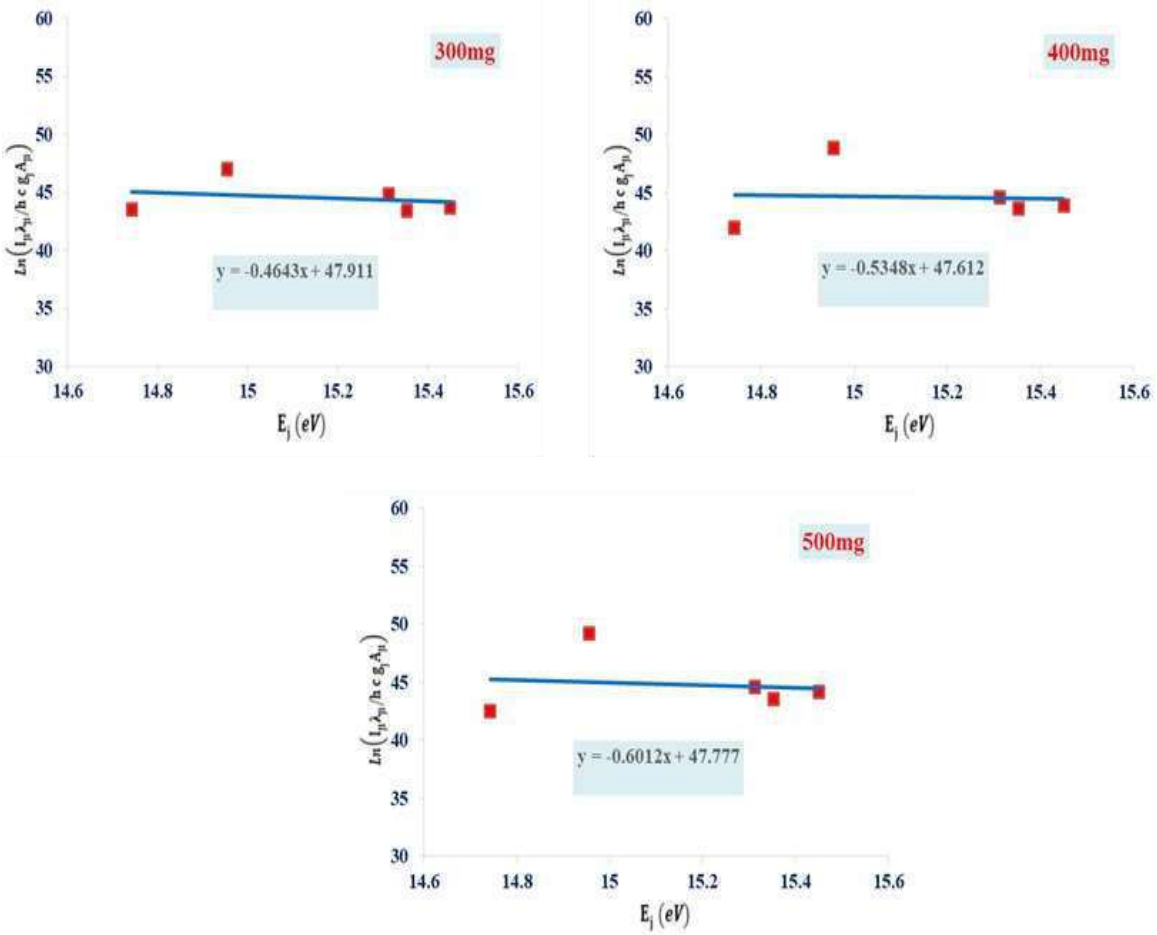

Figure 06- Boltzmann plot for N II peaks at three different NaCl concentrations.

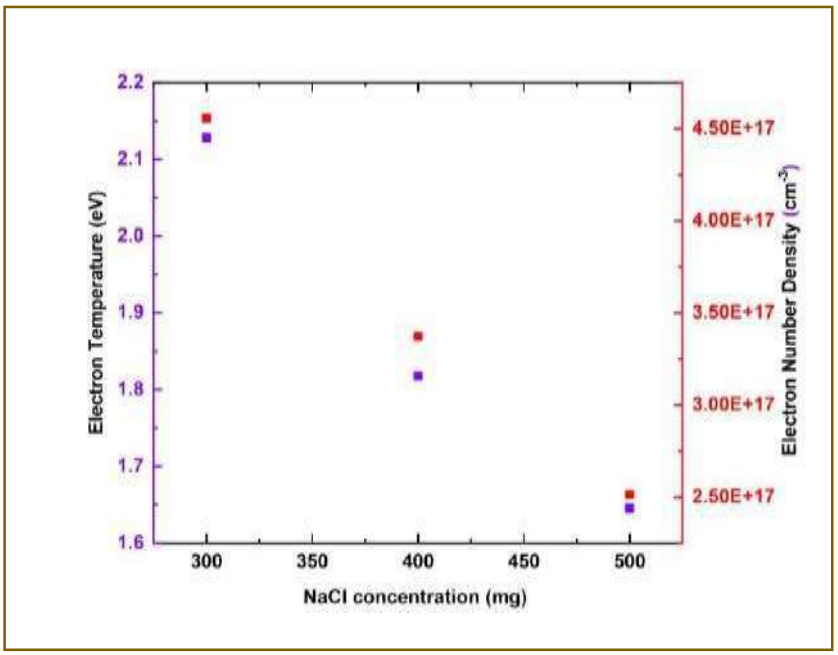

Figure 07- influence of NaCl concentrations on the electron temperature and electron number density.

Furthermore, table (2) depicts the effect of the NaCl concentrations on the air discharge parameters. The data detected that the plasma frequency and the plasma parameter decreased with increases in NaCl concentrations. While the Debye length showed a different behavior with an increase of NaCl concentrations (i.e., increases of water conductivity). The increase in ion concentration due to NaCl can increase the collisions in the flow. Collisions between electrons and neutral ions or particles can cause energy to be transferred and redistributed. In some cases, these collisions can effectively transfer energy to the electron, helping to lower the temperature of the electron. Like resistive electrodes, liquid electrodes stabilize the discharge to stop it from contracting at the electrode. Although distributed resistivity may be the cause of the stabilization, other mechanisms are most likely at play as well.

Table 2- the variation of plasma parameters of air plasma with NaCl concentrations.

Food slat weight (mg)	$\mathbf{T_e}$ $(\mathbf{eV})$	$\mathbf{n_e \times 10^{17}}$ $(\mathbf{cm^{-3}})$	$\boldsymbol{\omega_{pe} \times 10^{11}}$ $(\mathbf{rad/sec})$	$\boldsymbol{\lambda_D \times 10^{-6}}$ $(\mathbf{cm})$	$\mathbf{N_D}$
300	2.1537	4.4519	377.1176	1.6342	8
400	1.8698	3.1577	332.3521	1.7278	7
500	1.6633	2.4401	279.1951	1.93989	7

Conclusion

A DC discharges in atmospheric pressure air in a pin- with one electrode submerged in water at different NaCl concentrations were investigated. The influence of added NaCl to the distilled water on the air discharge was investigated in more detail. The findings demonstrate that the conductivity of the electrode liquid significantly affects the properties of the air discharge occurring in the space between the pin electrode and the surface of distilled water. Hence, it is evident that altering the conductivity of the liquid leads to a modification in the dynamics of the discharge occurring within the gap between the pin and the surface of distilled water.

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Serological Investigation of Equine Toxoplasmosis and Association to Prevalence Factors in Basra Province, Iraq

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Abstract

Background: *Toxoplasmosis is one of the most prevalent and neglected diseases which caused by a single-celled parasite called Toxoplasma gondii resulting in economically variable looses in different animals and marked public health problem in human, particularly women.*

Aims: *This study aims to identify serological prevalence of acute and chronic infection in native equine, horse and donkey, in Basra province (Iraq) using the qualitative enzyme-linked immunosorbent assay (ELISA).*

Materials and methods: *A total of 184 equine animals including 39 horses and 145 donkeys were selected randomly from different areas in Basra province (Iraq) during May-December (2023), and subjected to sampling the jugular venous blood to obtain sera that tested by ELISA.*

Results: *An overall 5.98% and 26.09% study animals were positively infected with acute and chronic infections, respectively. According to animal species, the prevalence of toxoplasmosis in horses and donkeys, respectively, was 5.13% and 6.21% for acute as well as 28.21% and 25.22% for chronic. Significantly, no significant variation (P>0.05) was seen between titers of acute and chronic infection in both horses and donkeys. Regarding age, acute infections were increased significantly in horses and donkeys of <5 years age old while chronic infection was elevated in both study animals of >5 years old. Concerning sex, acute infection was markedly higher in female horses than males; while for chronic infection, male horses were showed higher values than females. In contrast, male donkeys were reported a higher acute values but lower chronic values when compared to females.*

Conclusion: *This study revealed that the acutely and chronically stages of disease are found in both equine animals that might act as a source of toxoplasmosis for animals and humans. However, furthermore studies in other areas with using of molecular assays appeared necessary to providing additional data.*

Key Words

Toxoplasma Gondii; Horse; Donkey; ELISA; Zoonotic Disease.

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Introduction

Marked advances in controlling schemes toward various infectious pathogens, especially those that spend part of their life cycle inside host cells have taken place currently (Kumar et al., 2021; de Barros et al., 2022). Toxoplasmosis is a parasitic disease resulted by *Toxoplasma gondii* that classified scientifically under the Sarcocystidae Family in the Eucoccidiorida Order of Apicomplexa Phylum (Xue et al., 2020; Razooqi et al., 2022). The lifecycle of *T. gondii* is unique and involving three different infective stages involving rapidly dividing invasive tachyzoites, slowly dividing bradyzoites in tissue cysts, and environmental stage or sporozoite (Martorelli et al., 2020; Delgado et al., 2022).

Transmission of toxoplasmosis occurs asexually between intermediate hosts (carnivorism), sexually (definitive hosts) and by the oral-fecal route (Farhat and Hakimi, 2022). However, the asexual and sexual cycles and disease transmission patterns in a survival environment vary based the physical characters and structures of both definitive and intermediate host populations (Kennard, 2018; Gissot, 2022). Also, the presence of different *T. gondii* genotypes exhibit different behaviors and virulence; e.g., the genotypes detected in Asia, Africa and South America are considered more virulent than those predominating in North America and Europe (Shwab et al., 2014; Calero-Bernal et al., 2022). Therefore, the clinical signs depend on the affected tissue, the number of tachyzoites released and the ability of the host immune system to limit replication and transmission (Munoz et al., 2011). Because adult immunocompetent animals control tachyzoite transmission efficiently, toxoplasmosis is usually subclinical illness (Almeria and Dubey, 2021). Whilst, the tachyzoites are transmitted in susceptible species, young and immunocompromised animals systemically to cause different diseases and corresponding clinical symptoms such as high fever, dyspnea, cough, diarrhea, icterus, seizures and even death (Ibrahim, 2017; Stanić and Fureš, 2020).

In equine animals in particular horse, clinical signs of toxoplasmosis are rarely seen in naturally and experimentally infected animals, usually nonspecific and are not sufficiently characteristic for a definitive diagnosis (Hill et al., 2005; Jokelainen, 2013; Opsteegh et al., 2016). For these reasons, indirect diagnosis by serological testing in addition to postmortem diagnosis via histology, bioassay and molecular assays are the main methods used to diagnosis the disease (Casartelli-Alves et al., 2014; Ramírez et al., 2017; Uddin et al., 2021).

In Iraq, toxoplasmosis was surveyed serologically in Nineveh (Alshahery and Mansour, 2012), Baghdad (Altaee et al., 2014), Wasit (Asal and Al Zubaidy, 2016), Dhi-Qar (Gatie and

Hasso, 2017) and Duhok (Mikaeel and Al-Saeed, 2020) but not in Basra. Hence, this represents the first study aims to detect the seroprevalence of acute and chronic toxoplasmosis in equine animals, horse and donkey, with estimating the association both type of infection to host risk factors, age and sex.

Materials and methods

Ethical approval

This study was licensed by the Department of Parasitology and Department of Veterinary Public Health in the College of Veterinary Medicine (University of Basrah).

Samples

A total of 184 equine animals including 39 horses and 145 donkeys aged > 1 year and weighted approximately >150 kg, were selected randomly from different areas in Basra province (Iraq) during May-December (2023). Each study animal was subjected to sampling 5 ml jugular venous blood under aseptic conditions into free-anticoagulant glass-gel tubes using the disposable syringes. After centrifugation (5000 rpm, 5 minutes), the obtained sera was kept frozen in labeled Eppendorf tubes at 4°C (Razooqi et al., 2022).

Serology

According to manufacturer instructions of the qualitative ELISA; Toxo-IgM (Cat No: SL0016Ho) and Toxo-IgG (Cat No: SL0017Ho), the kit contents and sera were prepared, processed, and the optical density (OD) of control solutions and sera were measured at 450 nm using the Automated Microplate Photometer (BioTek, USA). Then, the test effectiveness and critical value (CUT OFF), in addition to the negative and positive judgments were determined. The CUT OFF values for acute and chronic infections were calculated at 311 and 339, respectively.

Statistical analysis

Data concerned the study animals and ELISA's kits were documented using the Microsoft Office Excel (version 2013) Software, and examined statistically using the GrphPad Prism (*version 6.0.1*) Software to evaluate significant differences between values of stage of infection (acute and chronic) and association of infection to age and sex of study animals. Values were represented either as number and percentage [No. (%)], or as Mean ± Standard Error (M ± SE),

and considered significant (S) if P<0.05 or non-significant (NS) if P>0.05 (Gharban et al., 2023).

Results

Among 184 equine animals, 59 (32.07%) were seropositively infected toxoplasmosis, including 33.33% (13/39) horses and 31.72% (46/145) donkeys (Figure 1).

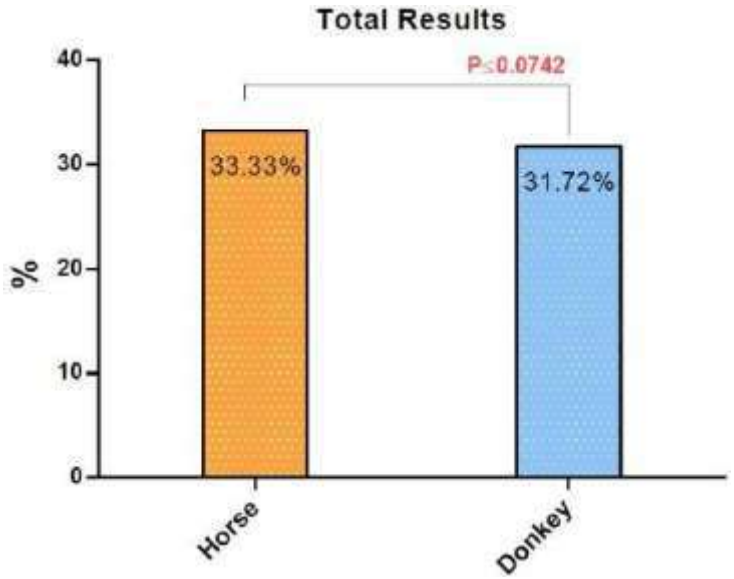

Figure 01- Total results of equine animals to detect the prevalence of toxoplasmosis

Regarding the stage of disease, an overall 5.98% was infected acutely with toxoplasmosis while 26.09% were infected chronically (P<0.0235). Also, significant increases (P<0.05) were identified in chronic infection of both horses (28.21%) and donkeys (25.22%) when compared to acute infections of both animals; horses (5.13%) and donkeys (6.21%), (Table 1). Concerning the titer of positive infections, insignificant variation (P>0.05) was seen between acute and chronic infection of both equine animals as well as between positive horses and donkeys in both infections (Table 2).

Table 01- Total positive acute and chronic infections in equine animals, horses and donkeys

Animal	Total No.	Acute	Chronic	p-value
Horse	39	2 (5.13%)	11 (28.21%) *	0.0091 S
Donkey	145	9 (6.21%)	37 (25.22%) *	0.0104 S
p-value	-	0.0733 NS	0.0611 NS	-
Total	184	11 (5.98%)	48 (26.09%)	0.0235 S

Table 02- Titer of positive acute and chronic infections in equine animals, horses and donkeys

Animal	Titer (M±SE)		p-value
	Acute	Chronic	
Horse	0.384 ± 0.047	0.392 ± 0.008	0.0968 NS
Donkey	0.393 ± 0.014	0.388 ± 0.005	0.0984 NS
p-value	0.0963 NS	0.0989 NS	-
Total	0.391 ± 0.013	0.389 ± 0.004	0.099 NS

Associations of seropositive acutely and chronically infected horses and donkeys to risk factors, age and sex, were reported a significant variation (P<0.05) in their values. For age, horses and donkeys aged <5 years were showed a higher rate of acute positivity but a lower rate of chronic positivity when compared to those aged >5 years (Tables 3, 4).

Table 03- Association of positive acute and chronic infections to age of study horses

Group	Total No.	Acute	Chronic	p-value
< 5	6	2 (33.33%)	4 (66.67%)	0.0089 S
>5	7	0 (0%)	7 (100%)	0.005 S
p-value		0.0051 S	0.0098 S	-

Table 04- Association of positive acute and chronic infections to age of study donkeys

Group	Total No.	Acute	Chronic	p-value
< 5	21	6 (28.57%)	15 (71.43%)	0.0084 S
>5	25	3 (12%)	22 (88%)	0.0019 S
p-value		0.0352 S	0.0442 S	-

Relation to titers of positive infections, horses and donkeys of <5 years old were showed a significant elevation in their values (P<0.05) compared to those of >5 years; however, values of chronic infections in both horses and donkeys were recorded an insignificant difference (P>0.05) between values of <5 and >5 years old (Tables 5, 6).

Table 05- Association of titers of positive acutely and chronically infected horses to age

Group	Titer (M±SE)		p-value
	Acute	Chronic	
< 5	0.354 ± 0.047	0.37 ± 0.011	0.0919 NS
>5	0 ± 0	0.405 ± 0.009	0.0095 S
p-value	0.0097 S	0.0628 NS	-

Table 06- Association of titers of positive acutely and chronically infected donkeys to age

Group	Titer (M±SE)		p-value
	Acute	Chronic	
< 5	0.416 ± 0.009	0.362 ± 0.005	0.0429 S
>5	0.347 ± 0.02	0.405 ± 0.006	0.0414 S
p-value	0.0355 S	0.0522 NS	-

Distribution of seropositive results among groups of sex factors observed that female horses were having a significant higher rate of acute infection (P<0.0058), but a lower rate of chronic infection (P<0.0196) when compared to males (Table 7). In contrast, male donkeys were revealed a significant elevation in acute infection and a reduction in chronic infection when compared to females (Table 8).

Table 07- Association of positive acute and chronic infections to sex of study horses

Sex	Total No.	Acute	Chronic	p-value
Male	5	0 (0%)	5 (100%)	0.0019 S
Female	8	2 (25%)	6 (75%)	0.007 S
p-value		0.0058 S	0.0196 S	-

Table 08- Association of positive acute and chronic infections to sex of study donkeys

Group	Total No.	Acute	Chronic	p-value
Male	19	5 (26.32%)	14 (73.68%)	0.0324 S
Female	27	4 (14.81%)	23 (85.19%)	0.0214 S
p-value		0.0428 S	0.0466 S	-

Although the titers of acutely infected female horses were significantly higher than males (P<0.05), insignificant variation (P>0.05) was seen in chronically infected male and female horses (Table 9), as well as in acutely and chronically infected male and female donkeys (Table 10).

Table 09- Association of titers of positive acutely and chronically infected horses to sex

Group	Titer (M±SE)		p-value
	Acute	Chronic	
Male	0 ± 0	0.383 ± 0.015	0.0096 S
Female	0.384 ± 0.047	0.399 ± 0.009	0.0932 NS
p-value	0.0096 S	0.0926 NS	-

Table 10- Association of titers of positive acutely and chronically infected donkeys to sex

Group	Titer (M±SE)		p-value
	Acute	Chronic	
Male	0.387 ± 0.026	0.394 ± 0.009	0.0973 NS
Female	0.401 ± 0.008	0.385 ± 0.006	0.0926 NS
p-value	0.0935 NS	0.0961 NS	-

Discussion

Toxoplasmosis is an obligate intracellular potentially zoonotic parasitic disease which causes different symptoms in various animals as well as humans. There was no doubt that equine animals can harbor viable *T. gondii*, which could be isolated from tissues of both naturally or experimentally infected animals (Stelzer et al., 2019). In this study, qualitative analysis of equine animals revealed the seroprevalence of toxoplasmosis was 32.07%, composing 33.33% horses and 31.72% donkeys. In comparison to other works in Iraq, prevalence rate of toxoplasmosis in horses was 72.2% in Nineveh (Alshahery and Mansour, 2012), 32.6-67.39% in Baghdad (Altaee et al., 2014), 18.4% in Wasit (Asal and Al Zubaidy, 2016), 21% in Dhi-Qar (Gatie and Hasso, 2017) and 17.7% in Duhok (Mikaeel and Al-Saeed, 2020); whereas in donkeys, the only one study done in Mosul was reported 46.15% as a total rate of positivity, in which, 8.33% for acute infection and 91.67% for chronic infection (Hussain, 2011). This variation may be attributed to the environmental contamination of pasture and water arising from oil spills in Basra regions, which are capable of inducing immunosuppression among animals consuming such pasture and water. Other responsible factors may include levels of environmental contamination by oocysts of the parasite, and possible contacts of other animals with the feces of infected cats. Worldwide, a relative several reports in horses, mules and donkeys have detected the great serological prevalence of anti- *T. gondii* antibodies (Rodrigues et al., 2019; Li et al., 2020; Cano-Terriza et al., 2023). Reported seroprevalence for equine animals range in 0-71% in Asia, 14-45% in Africa, 2% in Australia, 0-73% in North America, 3-90% in South America, and 0-55% in Europe (Stelzer et al., 2019; Karshima et al., 2020).

However, the obtained findings are complicated for comparison due to various, not always valid serological assays with different cut-offs which served (Aroussi et al., 2021; Olsen et al., 2022). Additionally, equine animals selected in different surveys are varied widely in numbers, ages, origins and purposes. Recently, no reference standards are existed to support serological methods in equine animals precisely, and the previous attempts for correlation the findings of

serology with those of molecular assays were failed largely (Aroussi et al., 2015). In almost cases, there a proper absence for the relationship between molecular diagnosis of toxoplasmosis by the high sensitive polymerase chain reaction and serological information (Botein et al., 2019; Amairia et al., 2023; Jeske et al., 2024). Nonetheless, seroppositive response may refer for exposing the equine animals to toxoplasmosis and thus can apply for identification the risk factors of toxoplasmosis (Arruda et al., 2020). The titeration of antibodies in positive study equine animals were showed an obvious variation in their values between acutely and chronically infected animals as well as between different age and sex groups. In one study, prevalence of toxoplasmosis among cases and controls which determined at standard serum cut-offs and the findings revealed that horses with clinical signs had 3.55 times the odds of a seropositive test compared to those without clinical signs (James et al., 2017).

According to age, Gatie and Hasso (2017) reported that toxoplasmosis was reported in horses aged <6 months (3%) and in those of 7 months-25 years (2%). Altaee et al. (2014) recorded that the disease is found in 55.55-72.22% of <20 years, 65.38-76.92% of 20-30 years, 54.16% in 30-40 years, and 55.55-66.66% of 40-50 years. (Touma et al., 2020) showed that the positivity was seen in 28.5% of horses aged ≤ 10 years and in 20% of horses aged > 10 years old. Based on these data, sex appears to be an important factor to influence the prevalence of the infection.

Concerning the sex, the prevalence rate of toxoplasmosis respectively in female and male horses was 71.4% and 77.8% in Nineveh (Alshahery and Mansour, 2012), 28-58% and 38-78.57% in Baghdad (Altaee et al., 2014), 20.2% and 0% in Wasit (Asal and Al Zubaidy, 2016), 24.39% and 5.55% in Dhi-Qar (Gatie and Hasso, 2017), and 20% and 26.6% in Baghdad (Touma et al., 2020); while in donkeys, it was 51.16% and 22.22% (Hussain, 2011). The fact that immune suppression associated with pregnancy can increase the chances of infection. Other studies suggested that higher concentration of testosterone plays a behavioral shift in Toxoplasma-infected subjects (Hodková et al., 2007; Flegr et al., 2008). In a recent study, the authors found that seropositivity seems to be a risk for infection in females but not in males, suggesting the role of sexual transmission of *T. gondii* from male to female (Hlaváčová et al., 2021).

Conclusion

Although many studies are available in literatures about the prevalence of toxoplasmosis in field animals, there are still large gaps in our current knowledge. Our finding reported that the acutely and chronically stages of toxoplasmosis are found in equine animals, horses and donkeys that might act as a source of infection in field for both animals and humans. Therefore, furthermore epidemiological studies using different serological and molecular assays appeared to be necessary for providing additional data.

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Authors' contribution: IME: Collection of blood samples and obtaining the sera. NAK: Serology for IgM ELISA kit. JYM: Serology for IgG ELISA kit. GJKA: Statistical analysis of obtained results. All authors were approved the final copy of the manuscript.

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Data Processing: An Approach Based On Indexing

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Abstract

The way things operate on the web has shifted because of advancements, in the Semantic Web and linked data. The use of the Standard Resource Description Framework (RDF) is widespread for storing data. Since RDF follows a graph based data model organizing and accessing information poses challenges. This necessitates the development of large scale systems that offer ways to search and retrieve information for use by both humans and machines. Our proposed approach seeks to address these issues by introducing an indexing engine tailor made for RDF structures. To enhance data retrieval speed and query efficiency this research introduces a technique for indexing. By creating indexes that capture relationships, in RDF graphs quick and targeted query results can be achieved. The system streamlines the search process by structuring RDF data elements through a sequence based indexing approach. The approach also examined how semantic knowledge can be integrated into query processing to improve it and enable more sophisticated and context-aware information retrieval. The proposed subgraph-based indexing and query processing strategy is effective as evidenced by experimental results showing higher accuracy and speed compared to traditional techniques. This project addresses challenges, in managing RDF data. Sets the stage for advancing graph based information retrieval systems and semantic web technologies in the future.

Key Words

Data processing; RDF; Information Retrieval; Storage; Indexing.

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Introduction

The additional web is undergoing a transformation with the emergence of the Semantic Web and networked data. The aim of these improvements is to make information on the Internet not only readable by humans, but also understandable by machines. There are different types of web, as shown in Figure 1. Early static websites (a: Web 1.0) are static webs (1990s to early 21st century) characterized by predominantly static content and limited interactivity with the user. Social networks (b: Web 2.0) are dynamic, interactive and user-centered. Knowledge Graphs (c: Web 3.0) is a semantic web focused on automatic data understanding and integrating artificial intelligence (AI) and machine learning (ML).

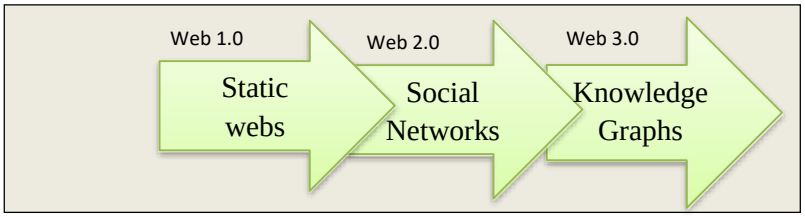

Figure 01- a: static websites web 1.0, b: Social networks web 2.0, c: Knowledge Graphs web 3.0

RDF is an essential part of the Semantic Web and provides a standardized way to represent and connect data on the web(Duan, Street and Xu, 2011). RDF contributes to the vision of the web, where data is not only intended for human use but is also understandable and interoperable by machines. It provides a simple graphical format for describing resources on the Internet and the relationships between them. With graphically structured data such as the Resource Description Framework (RDF), indexing becomes crucial for efficient data query and retrieval.

The RDF data structure index is essentially a graph created from the data itself (Gelling, Fletcher and Schmidt 2023). Different methods of indexing can be used for RDF data, like stores, inverted indexes and graph databases. Optimized query can be obtained from exploring the graph structure of RDF.

Structures RDF

RDF format is used in web and other data management methods to integrate data sources by arranging data in a graph-like manner with entities, predicates, and objects. Figure 2.explain triples of RDF graph.

Figure 02- Triples for RDF graph.

This makes it easier for humans and machines to search, analyze, and combine information across different systems and applications. In the big data environment, this seamless interaction is crucial for businesses dealing with massive volumes of data daily(Moffat and Mackenzie, 2022).

RDF data plays a role, in enhancing data integration query processes and inference ultimately improving the efficiency of information management. Moreover RDF data adds a layer that imparts meaning to the connections between entities enriching the data context and enabling data analysis and knowledge discovery. Embracing RDF data in information management systems can markedly enhance data quality, accessibility and usability empowering decision making. The future trajectory of RDF data is anticipated to emphasize advancements in data integration, scalability and interoperability. With the escalating volume of generated data there is a growing demand, for effective methods to integrate and handle RDF data from diverse origins. Scalability will be pivotal as organizations seek to manage datasets and accelerate data processing speeds.

To promote sharing of information and teamwork it is crucial to enhance the compatibility, among systems and software applications. As the technology evolves, these trends are likely to shape the future RDF data landscape and drive new innovations in data management(Darmont *et al.*, 2022). In this study we concentrate on data partitioning and query handling. To tackle these duties we introduce a method that utilizes the patterns displayed by the data stored in the structural index of one of the triples to implement clustering, on the elements stored in the structural index.

Related Work

To address the challenge of handling RDF graphs during search some suggested approaches involve generating a summarized version of the graph while conducting indexing. Strategies, for storing RDF data and querying methods for HBase and MySQL Cluster are put forward. A previous research paper by (Atre *et al.*, 2010). explored the use of bitmap indices for processing RDF joins on a machine. This study focuses on RDF graphs in contrast, to earlier work that dealt with much larger graphs requiring partitioning. This distinction enables us to implement optimizations in our storage, indexing and querying methodologies.

A TensorFlow based function is then employed to evaluate the subgraphs that are retrieved. The distributed RDF 3X system has detailed documentation, on RDF querying techniques in (Huang, Abadi and Ren, 2011). Proposed in the study(Franke *et al.*, 2011). are

storage strategies based on RDF and search methods, for HBase and MySQL Cluster. (Luo, Fletcher and Hidders, 2012) In addition it is important to consider how RDF data is stored and indexed in RDF databases taking into account relational, entity and graph viewpoints. Storing quantities of data, for indexing poses challenges especially when dealing with graphs.(Le *et al.*, 2014) ,((Coffman and Weaver, 2014)). According to (Ouksili *et al.*, 2017) papers serve as a literal or resource, and relationships between the keywords and the dataset's components are investigated using outside knowledge. (Faye, Curé and Blin, 2021) The focus is on both storage and indexing schemes for local RDF engines, split into native and non-native storage schemes. (Sagi et al., 2022) in this study a fresh approach, to evaluating query workload access patterns and a new design concept called Subdivision Compression Redundancy (SCR) for data representations in RDF databases were presented. By integrating these two elements researchers can now delve into the decision making process behind the structure of RDF stores specifically identifying the data representations, for specific workloads.

Martial and dataset

The information that has been extracted from the file talks about a particular resource that is designated with the URI[*], The Europeana schema indicates that this resource is of type 'ProvidedCHO'. Additional information about this resource, including its description, subject, title, and type, is also included in the data in Portuguese. For example, one of the triples indicates that the resource is affiliated with 'Angola' and has the title 'Antherotoma gracilis'. It is also related to 'ACTD › Biology › Herbarium › Melastomataceae'. The content in this file appears to be related to biodiversity and botanical specimens, as it reflects details. There are twenty-four unique pieces of information in the collection, each represented as an RDF triple. Subject, predicate, and object make up a triple in RDF; these three components work together to construct a statement about a resource.

Methodology

The standard format, for representing data on the web is RDF. Advanced systems for organizing and indexing sequence RDF data are utilized. One common method of indexing RDF datasets involves using graph based indexing algorithms for RDF databases. Establishing an index for RDF data is crucial for information retrieval and querying. The recommended

[*] http://id.bnportugal.gov.pt/bib/rnod/100000.

graph based approach to indexing Semantic Web (SW) data enhances the effectiveness of data retrieval and exploration.

This technique enables graph exploration and ranking in SW queries through the indexing of graph structures addressing the limitations of current SW search engines that lead to increased overhead when indexing large RDF texts due to incomplete indexation of all row triples or graph structures. Specifically this method streamlines the querying process by providing a means to index graph structures in SW data thereby enhancing exploration capabilities within graphs. Our findings indicate that while an entire RDF graph may not typically exhibit a structure it is feasible to represent each segment when dividing the graph into subgraphs.

Consequently these hierarchical segments can be indexed as a collection within an RDF graph where triples in the RDF data signify relationships, among subjects, predicates and objects. Figure 3 generates a representation, by transforming sets of three elements, into a display.

To give a clear and understandable picture of the relationships between the various entities in the dataset, the RDF data is visualized using labeled nodes and edges. We can quickly comprehend the relationships and connections within the data by labeling the nodes with the titles of the subjects and objects and the edges with the predicates. Through the identification of patterns, dependencies, and structures found in the RDF data, this visualization facilitates the interpretation and analysis of the data.

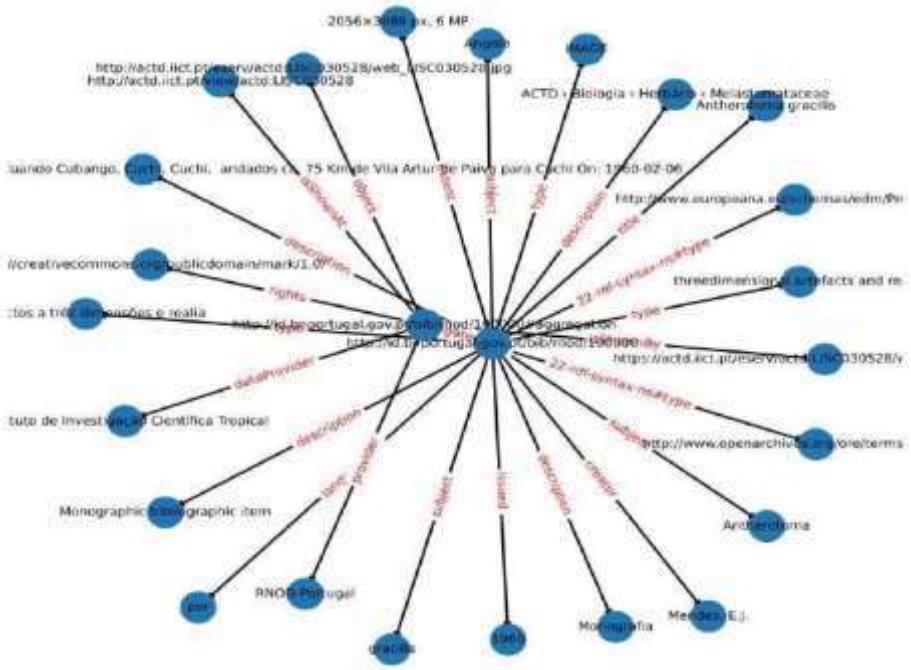

Figure 03- Example graph small data

The relationships between different topics and objects in RDF data are depicted in this graph, where the predicates signify the type of relationship. For clarity, each directed edge represents a predicate connecting two entities, identified with the type of relationship, and each node represents an entity. In creating an RDF index from a graph it is important to follow a process outlined in steps based on subgraphs;

- Step1: Providing URIs; Each resource and property, within the graph should be assigned an URI((Uniform Resource Identifier)). These URIs act as identifiers for RDF resources enabling them to be referenced and connected. Figure 4.a Show URI of dataset used.
- Step2: Indicate Triples: In RDF data relationships are depicted through triples (predicate object) with each triple denoting a connection, between two resources via a property. Figure 4.b Show URI of dataset used.

Figure 04-a Examples URI of dataset

Figure 04-b Explain examples triple of dataset

- Step3: Generate RDF Statements; Within a graph form RDF statements as triples in every connection, each statement affirms a detail about a resource using an attribute. . Each statement affirms a detail about a resource using an attribute.
- Step4: Identify Subgraph; Select the approach to define the subgraph for RDF data. This choice may vary based on factors, like the types of resources and predicates.
- Step5: Establish Index; Develop an index that links each subgraph identifier with the triples (statements comprising subject, predicate and object) belonging to that subgraph.
- Step6: Retrieve Subgraphs; Enable querying of the index to access subgraphs or individual triples associated with a subgraph.

To handle the subgraph indexing, we develop a class called RDF Subgraph Index. Based on a specified criterion predicate, the RDF data is indexed using the index subgraphs method. A particular subgraph can be retrieved using the get subgraph method by providing its identifier. Figure 5 illustrates subject-based indexing on a graph: To extract the subgraph, go to http://id.bnportugal.gov.pt/bib/rnod/100000. Using this method gives a versatile way to arrange and retrieve subgraphs from RDF data.

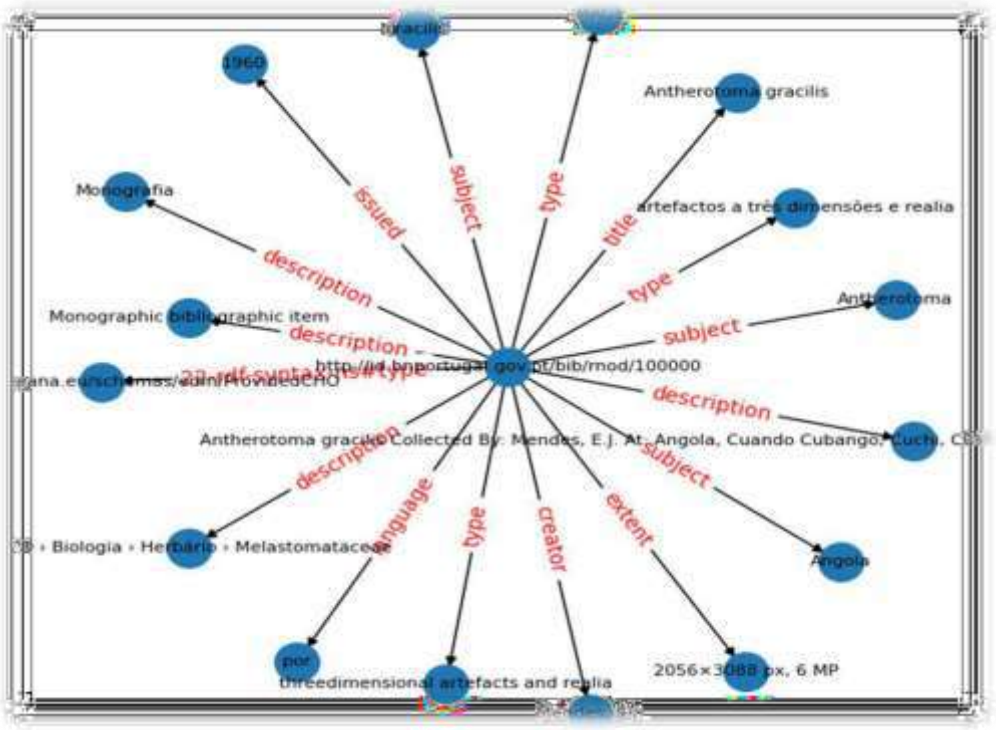

Figure 05- indexing on a graph

Efficiency, in queries can see improvements by breaking down the graph into subgraphs based on specific criteria. This approach allows focusing on subgraphs for a query of scanning the entire network, which proves especially beneficial for large graphs. Organizing graph data into subgraphs aids in management and understanding. Each subgraph represents a subset of data that can be examined or modified independently. Indexing subgraphs can enhance program scalability offering a memory alternative to loading the entire graph into memory particularly for very large graphs.

Customized criteria can be set to generate subgraphs based on requirements allowing flexibility, in adapting the graphs structure to suit anticipated query types and data formats. Indexing subgraphs may improve the performance of graph operations and algorithms potentially reducing complexity and boosting application efficiency by confining operations to subgraphs as needed.

Conclusion

In conclusion, RDF data provides a strong and adaptable means of linking and representing structured data on the internet. We may build an easily query able and shareable network of interconnected data across many platforms and applications by defining relationships between things using RDF triples. Although RDF data presents a unique set of problems, including those related to interoperability and scalability, its advantages in knowledge representation and data integration make it an invaluable resource for developing the next wave of intelligent online application. As the web develops further, RDF data will become more crucial for facilitating data-driven services. In summary, partitioning a network into subgraphs and indexing them offers an effective and structured method of organizing and querying graph data, with advantages including increased performance optimization, modularity, scalability, and adaptability.

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Studying Some Physical, Chemical and Biological Properties of the Locally Bottled Drinking Water in Mosul City – Iraq

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Abstract

The quality of bottled drinking water was evaluated for ten local brands that are most widely traded in local markets on the left side of the city of Mosul. This is done by assessing some of the physical and chemical characteristics that include (pH, electrical conductivity, turbidity, total dissolved salts, calcium, magnesium, sodium, potassium, nitrates, sulfates, chlorides, and phosphates), as well as the biological aspect, which includes examining bacterial contamination and comparing the results of the current study with drinking water specifications issued by the Iraqi Environmental Determinants and those of the World Health Organization.

The results showed that all samples under study were free of bacteria, and calcium and potassium ion concentrations ranged between (32 - 40), (0.24 - 0.4) mg/L respectively. and the sodium ion reached higher concentrations than the potassium Ion in the current study, the total dissolved salts did not exceed (143) mg/L, and the turbidity reached the highest values in the water quality of Ruffian and the Alwaha, recording (2.03-2.18) NTU, respectively. The pH values for all samples under study were within the limits permitted by the World Health Organization and did not exceed the range (6.5-8.5), while the sulphate ion recorded in the Life water sample had the highest concentration of (407) mg/L, thus exceeding the ion Iraqi and international limits amounting to (400) mg/L. The CWQI results indicated that most of the water samples under study were of excellent water quality, with the index value reaching (100).

Key Words

Water quality; bottled drinking; NTU; CWQI.

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Introduction

Water is an essential element for all living organisms, It covers 71% of the Earth's area, although water is the backbone of life, however, we deal with it poorly, misusing it and contributing to Its pollution with our own hands, and we know full well that this pollution will reach us directly or indirectly(Hassan and Ibrahim, 2022).Drinking water is one of the necessary human needs that cannot be dispensed with for any reason, and it must meet water quality standards in terms of taste and color, In addition to physical, chemical, and biological properties (Al-Obaidi and Al-Mahdawi, 2016).

In our current era, most water sources have been polluted, starting from the oceans, seas, and rivers, and ending with groundwater and rainwater. The pollutants present in the water that reach the human body every day pose a real threat to their health and Providing drinking water has become not an easy matter, due to the large number of pollutants that reach it in different ways and cause many serious health problems that affect living organisms, especially humans(Aoun and Kamoka, 2019).The bottled water industry has flourished in recent decades, and demand for it has increased in most parts of the world, and In Iraq, the bottled water industry has expanded greatly In the last ten years, and the Iraqi consumer has become mainly dependent on water bottled in various plastic containers (Al-Obaidi and Al-Mahdawi, 2016), the industry has become widespread in Iraq, this increase in the number of factories was accompanied by a sharp decline in quality and a failure to apply Iraqi standard specifications for bottled drinking water, Which led to the flooding of local markets with many brands without taking into account their conformity with standard standards for drinking water, and failure to adhere to the chemical, physical and microbial characteristics that determine the quality and suitability of this water (Al-Amiri et al. 2013), so drinking water must be examined on the basis of microbiological, physical, and chemical quality (Almnehlawi, 2013).

Aims of the study:

1. The current study aimed to evaluate the quality of bottled water in the city of Mosul by analyzing a number of chemical, physical and biological properties and matching them with Iraqi and international standard specifications.
2. Application of the Canadian Water Quality Index to determine the quality of bottled water Is under study in the city of Mosul.

Materials and methods

The study included 10 samples of locally bottled drinking water, as shown in Table (1) , samples were collected from September until December 2023. They were obtained from commercial stores at random, from the left side of the city of Mosul, all of which originated in Iraq. Three bottles of each type were purchased to conduct a number of physical, chemical, and biological test methods adopted by (APHA,2017) and (Abawi and Hassan, 1990) for the purpose of determining the quality of bottled water. The following tests were conducted:

The pH was evaluated using a pH meter. The electrical conductivity and total dissolved solids Were measured using the TDS-C˚-meter- conductivity model YL-TDS2-A. Turbidity was estimated in a Nephelometric Turbidity Unit (NTU) using a device type (LP 2000 Turbidity Meter). Dissolved oxygen concentrations were Measured based on the Winkler method of Azidemodifieation. The concentration of nitrate ions was measured using the Ultraviolet Screening method with wavelengths of 220 and 275nm. On the other hand, The concentration of phosphate ions was measured using the Staunous Chlorid method, using a spectrophotometer at a wavelength of 690nm. The concentration of sulfate Ions was estimated according to the Turbid matric method using a spectrophotometer at a wavelength of 420nm. Via using the Mohar method, the concentration of chloride ions was measured.

Additionally, the total hardness and Calcium ion concentrations were Measured based on the Na2EDTA titration method. The arithmetic method was used to estimate the magnesium ion concentration by subtracting the value of calcium hardness from the total number, in units of mg/L .As for the concentrations of sodium and potassium ions, they were estimated using a digital flame analysis Gallen Kamoy spectrometer, in mg/L.The biological tests included the total number of bacteria using the standard plate count method on nutrient agar media, which was specified by the World Health Organization, in units of cell/ml.

Calculating the Water Quality Index: The Canadian Water Quality Index (CWQI) was used which many researchers referred to (Lumb et al,2006 and Salman et al, 2015) and applied the model to forteen cases; as shown in (Table 7), the water quality index was calculated. According to the following stage:

1- Calculate the Scope value (F1) the number of factors that exceeded the standard criteria divided by the number of quantitative factors under study

$$F_1 = \{\frac{Number\ of\ Failed\ variables}{Total\ Number\ of\ variables}\} \times 100$$

2- Calculating the Frequency value (F2), which is the number of readings that exceeded the standard standards divided by the number of factors quantity studied.

$$F_2 = \{\frac{Number\ of\ Failed\ Tests}{Total\ Number\ of\ variables}\} \times 100$$

3- Calculate the value of Amplitude (F3) according to the following steps:

a. Excursion calculation according to the following equation

$$\text{Excursion} = \{\frac{Failed\ Tests\ Value}{Objective}\} - 1$$

b. The sum of the standard deviations (nse), and the sum of the readings that do not conform to the standard standards is calculated by the sum of the deviations divided on the quantitative total of the tests according to the following equation:

$$\text{nse} = \frac{\sum_i^n = \text{Excursion}}{Number\ of\ Tests}$$

that, F_3 is calculated according to the following equation:

$$F_3 = \frac{\text{nse}}{0.1\text{nse} + 0.01}$$

The Canadian Water Quality Index is calculated according to the following equation:

$$\text{CWQI} = 100 - \frac{\sqrt{F1^2 + F2^2 + F2^2}}{1.732}$$

The state of the water body is expressed as indicated in (Table 01).

Table 01- Water quality index values (Lumb et al., 2006)

(WQ1) value for water quality	Estimation according to index	Description
Excellent	100-95	Water quality is protected assuming there are no sources that threaten or pollute the water
Good	94-80	Water quality is protected but may be threatened or slightly weak
Moderate	79-60	Water quality is often protected, and the water state sometimes deviates from the required or desired level
Marginal	59-45	Water quality is often threatened or poor
Bad	44-0	Water quality is always threatened or weak, and the water state constantly deviates from the required or desired level

Results and discussion

The pH is an important criterion for estimating the suitability of water for drinking and various civil uses and the possibility of pollution (Al-Safawi et al., 2018),the results of the study (Table 2) showed a variation between the pH values mentioned on the plastic containers by the producing companies (Table 3) and what was measured during this study. The results showed that the Information mentioned on the containers for the pH values does not reflect the actual content in the water samples Packaged .It is worth noting that there is a difference In the pH value of the Larin sample written by the company, as the value on the plastic package Is 7, while the value written on the box is 7.8, and it is not known which one is correct. After testing, the results showed in Table (2) that the pH value is (7.5).

The reason for this difference between companies may be due to the nature of the water sources used in production and the concentration of salts in them, and it may also be due to technical factors related to the quality of the treatment processes used in modern laboratories

Table (2) indicates the basicity of all types of bottled water, and when comparing the pH values with the Iraqi and international standards (specifications table) , table (1), they are suitable for drinking.

Electrical conductivity values express the total concentration of dissolved salts in the water, as the strength of water's electrical conductivity increases when a very small amount of salts is present, Table (2) shows that the electrical conductivity values ranged between (289-110) µM/cm in the bottled water of Larin and Defaf, respectively, and from Table (3) it Is clear that all bottled water did not exceed the standards specified for drinking water.The difference in the EC value for bottled water samples may be due to the different composition of the water in the source area, in terms of the diversity of soil composition and the difference in the composition of the mineral rocks. An increase in the electrical conductivity of the water is accompanied by an Increase in the total

Total dissolved salts, this means that there is a close relationship between EC and total dissolved salts, this relationship has been noted by many researchers, including Abawei (2010), Al-Saadi (2014) and Shihab and Kannah (2023),.as shown in Figure (1).

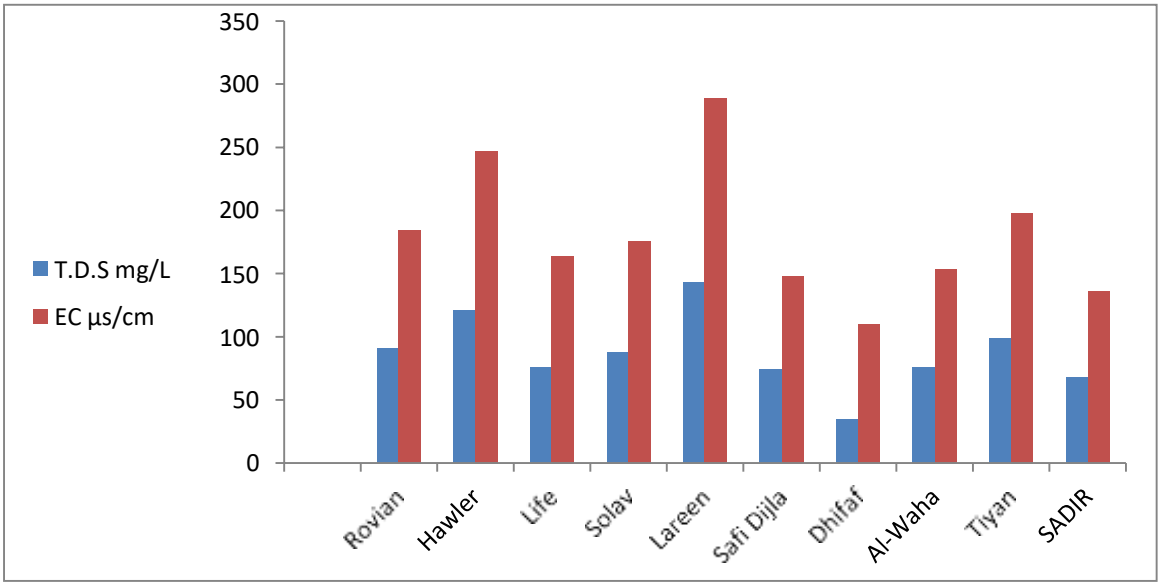

Figure 01- The relationship between electrical conductivity and total dissolved

Salinity is a distinct criterion in determining water quality, and high concentrations give the water an unpalatable taste, while low concentrations have a negative impact on human health. Water is divided according to its total dissolved salt content (TDS) into fresh water, moderately saline water, salty water, and very salty water (Hassan and Ibrahim, 2022; Abu Jalida et al., 2022)

The results of the study, Table (2), indicate that the concentrations of total dissolved salts ranged between (35 – 143) mg/L In Lareen and Al-Waha, respectively, and they differ from the concentrations found on the sample containers packaged by the manufacturers, Table (1)

When comparing the results of the current study with the Iraqi and international standards, Table (3), it is suitable for drinking.

Turbidity is a measure of the purity of a water sample and is one of the basic physical properties of water(Ahmed and Qasim, 2021).

The results of the current study show that the highest value of turbidity was (2.18) NTU and the lowest value was (0.60) NTU in each of the Ha, Lier, and Al-Waha samples, respectively, and when comparing the results with drinking water determinants based on the World Health Organization and Iraqi, Table (3) is suitable for drinking.

The concentration of dissolved oxygen in water is one of the most important criteria for evaluating water quality and the degree of Its pollution (Abawei and Hassan,1990) Many researchers, including Al-Wattar (2009) and Al-Sarraj (2013), confirmed that the abundance of

dissolved oxygen in water depends on the concentration of salts and organic materials present in it, as well as the temperature of the wate.

The results of the current study showed in Table (2) that the dissolved oxygen values ranged between (7.8-3.6) mg/L In the Life and Al-Waha samples, respectively. The results of the current study for dissolved oxygen did not conform to the standard specifications for human drinking, which were set by the Iraqi and international standards, which range between (4-6.5) mg/L (WHO,2008), with the exception of ruffian, Hah, Lear, and Solaf. Low concentrations of dissolved oxygen in water lead to an increase in the activity of anaerobic bacteria, which negatively affects water quality (Al-Safawi and Al-Assaf, 2018), while a high concentration of dissolved oxygen increases the activity of aerobic bacteria (Madan et al., 2018).

Nitrates are important for plant growth and are sometimes added to the soil to improve productivity ,Most of the nitrates found in the environment are from organic and inorganic sources, such as waste discharge and synthetic fertilizers ,Many people have become accustomed to using large amounts of nitrates to increase soil fertility, some of whichh drifts into water bodies and through the food chain reaches living organisms (Hassan and Ibrahim, 2022),the results we obtained from the current study are shown in Table (2), and when compared with the original concentrations written on the ten bottles of bottled water in Table (1), It was found that there is a discrepancy in the concentrations, as well as a difference In the concentrations found on the containers with the concentrations found on the box, as is the case in the company. Produced by Lareen.Both Holler and Solaf products exceeded Table (2) Determinants of the Iraqi Drinking Environment Table (3).

Table (2) shows that the lowest concentration of phosphate ion in the studied samples is (0.2) mg/L in the product from Life and Tian companies, and the highest concentration is (0.68) mg/L in Safi Degla. When the results are compared with Table (3), they are suitable for drinking, except for The product is from Safi Degla Company.

While the sulfate concentrations in most of the samples under study were within the permissible limits set by the World Health Organization (WHO) (2017) amounting to (250) mg/L, with the exception of the Life water sample, which recorded the highest concentration of sulfate formation, amounting to (407) mg/L, which is the highest. Among the Iraqi and international standards for drinking water, as shown in Table (3), It is possible that It comes from biological reactions through the oxidation of hydrogen sulfide by a number of bacteria

called sulfur bacteria, and then the resulting sulfur is oxidized by sulfur bacteria to sulfate (Arslan et al., 2016).

Rivers and groundwater are polluted with chloride, which gives them a salty taste and can make the water unsuitable for drinking, as high levels of chloride kill plants and wildlife (Abu Jalida etal,2022).The chloride concentration in the samples of the current study ranged between (19.9-11.9) mg/L, and when comparing the chloride concentration in all samples with the Iraqi standard specifications, they are suitable for drinking.

The results of the current study show Table (2) a clear difference in the concentration of calcium and magnesium ions. The highest and lowest values were (32-40) and (20-68) mg/L, respectively, from the concentrations found on the packaging of the samples placed by the companies. Table (1)) In addition to the difference in concentrations on the cover of the sample and the cover of the box, as is evident in the Larin sample.The results of the current study, Table (2), showed that the concentration of calcium and magnesium ions did not exceed the standard limits for drinking, Table (3).

The sodium Ion concentration ranged between (4 – 29.3) mg/L. The sodium concentration in bottled drinking water is low, and does not pose a danger to the human body. However, when the percentage of sodium In drinking water increases above 200 mg/L, the taste of the water becomes unpalatable. It is not acceptable to the user, and sodium, especially in bottled drinking water, has no harm to the human body (Al-Amiri et al., 2013), as It is very small amounts.

The percentage of potassium is usually significantly lower than sodium, and the concentration of potassium ranged between (0.024 - 0.4) mg/L. The reason may be attributed to the fact that potassium is present In sedimentary rocks such as potassium feldspar, and since these minerals are poorly soluble in water (AL-Layla et al.,1990). This is confirmed by the current study (Table 2), which shows that the sodium concentration in all samples is much greater than the potassium concentration. When comparing the potassium ion concentration with international standards and the Iraqi environment, it is suitable for drinking.

Bacterial pollution Is one of the determinants of water quality, and evidence of pollution causing diseases includes the total number of bacteria, total plate count, fecal coliform, and Escherichia Coli.The results of Table (2) indicated that all samples were free of bacteria.

The results of applying the Canadian Water Quality index showed that the water samples under study were of excellent quality in the water of Ruffian, Larin, Defaf, Al-Wahah, Tian and Sudair, as the value of the Index for each of them reached (100%) and that the water was of a protected quality assuming the absence of sources that threaten or pollute the water, While the water quality was good, but it was slightly threatened or weak in each of the water of Hawler, Life, Solaf, and Safi Tigris, as the index values ranged between (87.8-94.6%), as shown in Table (2).

Table (2) Values of parameters on the plastic bottle measured by the manufacturer

Sample	Ph	EC µs/cm	TDS ppm	Turbidity NTU	Total hardness mg/L	Mg^{+2} mg/L	Ca^{+2} mg/L	Nitrate mg/L
Rovian	7.3	178	121	***	***	2.3	28	0
Hawler	7.3	178	121	***	***	2.3	28	0
Life	7.2	***	***	0.2	40	4.4	24.2	0.5
Solav	7.3	178	121	***	***	2.3	28	0
Lareen	7 on bottle 7.8 on box	***	100	0	10	12 on bottle 7.5 on box	6 on bottle 26 on box	6.5 on bottle 0 on box
Safi Dijla	7.2	***	***	0.2	40	4.4	24.2	0.5
Dhifaf	7.2	***	***	0.2	40	4.4	24.2	0.5
Al-Waha	6.7	***	***	Not found	83	20	1.8	5
Tiyan	7.3	***	***	0	10	7.5	26	0.2
SADIR	7.2	***	***	0.2	40	4.4	24.4	0.3

Table 03- Physical, chemical and biological characteristics of locally bottled drinking water

Sample	Negative ions					Positive ions				Turbidity NTU	T.D.S mg/L	EC µs/cm	Ph	No. of bacteria cells/ml	CWQI
	D.O mg/L	NO_3^{-2} mg/L	PO_4^{-3} mg/L	Cl^{-1} mg/L	SO_4^{-2} mg/L	K^{+1} mg/L	Na^{+1} mg/L	Mg^{+2} mg/L	Ca^{+2} mg/L						
Rovian	6	8.8	0.4	11.9	18.6	0.4	16.6	68	36	2.03	91	184	7.8	0.0	100
Hawler	4.8	32.6	0.49	15.9	9.1	0.024	4	20	36	0.60	121	247	8	0.0	94.6
Life	3.6	8.9	0.2	11.9	407	0.4	16.6	20	4	1.8	76	164	7.6	0.0	87.8
Solav	4.4	33.5	0.39	13.9	22.5	0.024	4	44	36	1.29	88	176	7.6	0.0	94
Lareen	7.4	14.7	0.28	19.9	144	0.4	29.3	55	40	082	143	289	7.5	0.0	100
Safi Dijla	7.6	7.4	0.68	13.9	57.9	0.024	4	60	40	0.72	74	148	7.5	0.0	93
Dhifaf	7.2	7.4	0.28	15.9	47.1	0.024	4	65	40	0.92	35	110	7.3	0.0	100
Al-Waha	7.8	4.2	0.49	15.9	178	0.4	29.3	59	36	2.18	76	153	7.8	0.0	100
Tiyan	7.3	7.4	0.2	13.9	157	0.4	16.6	55	40	0.79	99	198	7.8	0.0	100
SADIR	6.9	9.3	0.27	15.9	98.8	0.024	4	68	32	1.03	68	136	7.3	0.0	100

Table 04- Drinking water determinants based on the World and Iraqi Health Organization for the year 2014 (Al-Shiblawi and Jassim, 2017)

No.	Chemical analyses	measuring unit	Determinants of the Iraqi environment	WHO determinants
			Highest permissible limit	
1	Turbidity	NTU	5	5
2	E.C	µS/cm	2000	2000
3	T.D.S	mg/L	1000	1200
4	pH	No unit	8.5-6.5	8.5-6.5
5	Ca	mg/L	150	200
6	Mg	mg/L	100	150
7	Na	mg/L	200	40
8	K	mg/L	-	10-12
9	PO_4	mg/L	0.5	0.4
10	SO_4	mg/L	400	400
11	Cl	mg/L	250	-
12	NO_3	mg/L	15	-
13	Total No. bacteria	cells/ml	50	-

Conclusion

1- The study showed a discrepancy in the values of some of the parameters under study, including pH, water hardness, and concentrations of the elements nitrate, sodium, chloride, sulfate, and potassium, compared to the concentrations that were written by the companies on the packages. The values of each of the electrical conductivity (EC) were not written. And the total dissolved salts (T.D.S.) on the containers by the companies for the samples of Safi Degla, Defaf, Al-Waha, Tian, Sudair, and Life.

2- The fact that all samples are free of bacteria cells is a good indicator of water quality

3- All water samples studied were of good to excellent quality for drinking purposes.

Recommendations

1. Conducting tests to measure the concentrations of heavy metals in bottled water samples.
2. Conducting an examination of the total number of colon bacteria (total coliform) and examining for the presence of fecal coliform bacteria (fecal coliform), in addition to conducting an examination for the presence of fecal streptococci bacteria (Streptococci coliform) and conducting an examination for the presence of Pseudomonas aeruginosa bacteria

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Protective Role of Cranberry Extract Against Zovirax-Induced Spleen Dysfunction in Adult Female Wistar Rats

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Abstract

This research explored a possible protective function of antioxidants, such as cranberries, against zovirax-induced spleen dysfunction in adult female Wistar rats. A collection of 24(adult female Wistar rats) was haphazardly appointed to four equal sets, each including six animals. They received the following treatment for (0–22–44) days per day. The first group, known as group (C), was obtained in (tap water), and served as a control. The second set (A1) obtained orally 150 mg/kg B.W. of cranberry alone. The third group (A2) received zovirax (450mg/kg B.W.) during the trial to elicit spleen toxicity. In the fourth group (A3), zovirax (450mg/kg B.W.) was administered, plus cranberry (150mg/kg B.W.) was used to alleviate symptoms. Blood samples taken from the orbital sinus were obtained on days (0, 22, and 44) of the experiment, after a fast, to test the serum levels of albumin, total bilirubin (T.B.), globulin, and total serum protein (T.S.P.). After the experiment, splenic slices were taken out for histological analysis. The findings showed that the quantities of total blood protein, albumin, and total bilirubin significantly decreased in rats given zovirax (A2) at a significance level of ($P \leq 0.01$), accompanying histopathological investigation alterations regarding the histological composition of spleen tissue sections to treat all other sets. However, the protecting function of cranberries was elucidated in the set (A1), encompassing both the histopathological changes and correction of the spleen function mentioned above. Based on these facts, we have determined the results of this study confirmed that cranberries, as an antioxidant, can protect the spleen against zovirax-induced damage in adult females.

Key Words

Spleen; Cranberry Extract; Zovirax; Adult Female Wistar Rats.

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Introduction

The spleen is a ubiquitous organ present in all vertebrates. It performs several crucial functions that maintain bodily health [1]. Its primary function is blood filtration, eliminating bacteria and shattered or damaged red blood cells. Its anatomy is akin to that of a sizable lymph node. [2], [3]. That organ plays a significant role in the immune system by generating white blood cells that combat infection and produce antibodies. Numerous illnesses, traumas, diseases, and abnormalities impact the spleen's function. In rat models, zovirax consumption results in necrosis, which then progresses to fibrosis and cirrhosis before becoming spleen cellular carcinoma. According to [4], zovirax is an antiviral and nucleoside analog used to treat varicella-zoster virus infections and herpes. Adults and children with severe infections brought on by the varicella-zoster virus (chickenpox virus), HSV-1, and HSV-2 are treated intravenously with zovirax [5].

Additionally, it is the recommended medication for managing herpes simplex encephalitis. Zovirax is often administered orally for treating adult and pediatric cases of chickenpox, managing the initial and recurring instances of mucocutaneous herpes in a specific patient, and treating cases of herpes zoster (shingles). Although systemic therapy is far more effective, mucocutaneous HSV infections are treated topically with zovirax. The body uses antioxidants as its first defense against free radicals and other oxidants. An antioxidant is a substance that can inhibit or delay the oxidation of other molecules. Avoiding oxidative stress in health and disease has garnered more attention recently. Oxidative stress denotes an initial occurrence in the pathology and physiology of chronic and non-communicable diseases [6]. Antioxidants effectively reduce the harmful effects of free radicals on oxidative damage to cells, which impacts the development and management of various disorders [7].

The World Health Organization reports that because plants have a wide range of bioactive chemicals that function as antioxidants in biological systems, 85% of nations, including those in Asia and Africa, use natural medicines derived from plants. An investigation into the effect of polyphenols on human health demonstrated that cranberry effectively suppressed oxidation lipids and prevented the creation of peroxidation products [8]. Eliminating free radicals, such as hydroxyl radicals(H.R.), superoxide radicals(S.R.), and singlet oxygen, by cranberry antioxidants is responsible for these benefits. This ultimately prevents the oxidation of biomolecules [9]. Among these plants are cranberries, which have long since valued for their therapeutic qualities. Native Americans Utilized (NAU) them to cure renal and bladder issues. Cranberries contain several bioactive components used widely in medicine because of their health benefits and ability to fight cancer. The components mentioned include fructose,

triterpenoids, vitamin C, flavonoids, anthocyanidins, catechins, and phenolic compounds [10]. According to research, cranberry flavonoids can remove harmful molecules called superoxide radicals, hydroxyl radicals, free radicals, and lipid peroxidation. Cranberry helps prevent damage to the mitochondria and loss of membrane integrity [11]. Powdered cranberry extract is added to food to strengthen the body's defenses against free radicals. Numerous biological characteristics of cranberries include their impact on microbial growth, biofilm formation, bacterial adhesion, immunomodulatory, and anti-inflammatory activity [12].

Moreover, cranberries were recognized to possess antioxidant qualities, as evidenced by their ability to protect red blood cells from hemolysis, maintain an antioxidant status in plasma, and exhibit cellular antioxidant activity [13]. Using this straightforward technique, it is possible to evaluate cranberry extract's antioxidant and protective properties against the oxidative stress that Zovirax causes in rats' spleen as part of an intriguing research program on the pharmaceutical importance of natural products, especially cranberry extract.

Aim of the Research

The objective of the current investigation was to assess the influence of cranberry on oxidative stress induced by zovirax in adult female Wistar rats. Explain the impact of Zovirax on specific biochemical indicators, including total protein, globulin, albumin, and total bilirubin. Additionally, discuss the effects of Zovirax on spleen tissue.

Materials and Methods

1. **Sampling:** In the animal house of the (College of Veterinary Medicine), adult female Wistar rats weighing between 190 and 250 grams were kept in cages with adequate lighting and ventilation. They were also provided with complimentary availability of water and regular rodent feed from the University of Mosul and Alnoor University College. They were given two weeks to get used to the experimental setup. For 44 days, twenty-four adult female rats were handled in the following ways each day.
2. **Experiment Design:** They were split up into four equal groups at random (6 rats in each group):
    1. The first set: designated as the control(C), was doused with tap water.
    2. The second set: (group A1) was administered orally with cranberry only (150mg/kg B.W.)]14].
    3. The third set (group A2): received zovirax (450mg/kg B.W.) to induce spleen toxicity during the experiment [15].

4. The fourth set (group A3): was treated with zovirax (450mg/kg B.W.) and oral cranberry (150mg/kg B.W.) to alleviate symptoms.

Fasting blood samples were obtained from anesthetized subjects using the retro-orbital sinus technique [16] at 22 and 44 days into the study period. The subjects were given an (i/m injection of ketamine 90mg/kg B.W. and Xylazine 40mg/kg B.W.) of serum was separated and refrigerated at -18 °C until analysis, after which samples were centrifuged for (15 minutes at 3000rpm). After that, the following parameters were assessed using kits (a BioSystems, Agappy, Switzerland product); these included albumin as reported by [17], and total serum protein (TSP) as calculated by [18]. State that the calculation of globulin involved subtracting (serum albumin) from (total blood protein), and total bilirubin(TB). After the experiment was over, the animals were killed, and portions of the spleen were taken out for histological analysis [19].

3. **Impact of Treatment on Biochemical Parameters:** Examinations Several ready tests were utilized to estimate the concentrations of biochemical parameters, including proteins, from multinational corporations such as the German Biocon Company, the French Biolabo Company, and the English Randox Company. Using the measuring techniques mentioned in Table 1, the amounts of various blood and biochemical parameters were estimated from the whole blood and serum (1). Manual techniques were employed to estimate the test of biochemical parameters.
4. **Histopathology studies:** Spleen tissue was fixed for a whole day in formal saline (10%). The tissue was then embedded in paraffin wax at (56ºC) after being dehydrated using (methyl, ethyl, and absolute ethyl alcohols). The wax tissue blocks were sectioned using a sliding microtome after hematoxylin and eosin staining, and they were then prepared for histological slide analysis using a light electric microscope (Olympus Cx21 with attached digital camera). Two separate investigators carried out morphometric analysis on ten sections of histology. Vacuolated hepatocyte counts (per square millimeter) and the percentage of lymphocyte-infiltrated surface (defined as the ratio of the infiltration area to the entire studied surface of the sample) were measured at ×100 and ×200 magnifications.
5. **Analytical statistics:** Results were analyzed using the complete randomized design(C.R.D.). Analyzed data was subjected to a thorough randomized design(C.R.D.). The groups were differentiated using the Duncan multiple range test, with findings evaluated at a concentration of ($P \leq 0.01$) and the statistical analysis conducted using the Microsoft Excel 365 software [20].

Results

1. Treatment's Impact on Biochemical Parameters:

Female rats were given zovirax (450 mg/kg B.W.) for 44 days. The statistics in tables 1, 2, and 3 demonstrate that their blood serum's albumin, total protein, and globulin levels were statistically significantly lower than those in the control(C) set. The concentration of total protein in treatment (A2) during the exposure period (22 and 44) days (4.99±0.17, 4.63±0.78 g/dl), respectively, decreases significantly at the level (P≤0.01) as indicated by (Table 1). When cranberries were added with the antibiotic and during the exposure period (22, 44) days, there was a modest rise in the level of total protein concentration in treatment (A3) to (5.60±0.32, 5.89±0.81 g/dl), respectively. Then, following treatment with cranberry antioxidants, a notable increase in the level of total protein concentration was seen during the same periods (22, 44) days in treatment (A1) to (6.17±0.97, 6.30±0.15g/dl), respectively. These outcomes closely resemble the control group's level.

Table 01- The protective effect of cranberry extract on total serum protein (TSP) concentration(g/dl) of adult female Wistar rats exposed to zovirax

Mean total serum protein (TSP) concentration (g/dl)				
Time	**(C)**	**Groups (A1)**	**(A2)**	**(A3)**
zero	6.18±0.38 A	6.26±0.45 A	6.33±0.48 A	6.26±0.36 A
22 days	6.34±0.42 A	6.30±0.15 A	4.99±0.17 BC	5.60±0.32 AB
44 days	6.41± 0.50 A	6.17±0.97 A	4.63±0.78 C	5.89±0.81 AB

Average ± Standard Error for three replicates. Different letters are positioned vertically before the numbers, signifying a significant difference at the probability level (P≤ 0.01) for each set of six animals. according to Duncan test.

When compared to the control(C) set, treatment (A2) during the exposure period (22, 44) days had a significant decrease in total albumin concentration(2.51±0.27, 2.25±0.056 g/dl) at the level (P≤0.01), according to the results of the measurement of total albumin concentration in the rats' blood serum, as shown in Table (2). while treatment (A3) zovirax with cranberry treated group showed a slight increase in total albumin concentration during the exposure period (22, 44) days (2.92±0.74, 3.14±0.83 g/dl) respectively. As its concentration was near that of the control group, treatment (A1) demonstrated a substantial rise in albumin concentration during the exposure period (22, and 44) days to the level of (3.67±0.33, 3.60±0.577 g/dl), respectively.

Table 02- The protective effect of cranberry extract on total serum albumin (g/dl) . of adult female Wistar rats exposed to zovirax

Mean total serum albumin concentration (g/dl)				
	Groups			
Time	**(C)**	**(A1)**	**(A2)**	**(A3)**
zero	3.53± 0.44 A	3.56±0.41 A	3.42±0.19 A	3.45±0.35 A
22 days	3.61±0.37 A	3.67±0.33 A	2.51±0.27 BC	2.92±0.74 ABC
44 days	3.57±0.46 A	3.60±0.577 A	2.25±0.056 C	3.14±0.83 AB

Average ± Standard Error for three replicates. Different letters are positioned vertically before the numbers, signifying a significant difference at the probability level (P≤ 0.01) for each set of six animals according to Duncan's test.

The efficacy of globulin content in rat serum is displayed in Table (3). The concentration of globulin in treatment (A2) decreased somewhat throughout the 22 and 44-day periods, coming in at (2.46±0.44 g/dl and 2.32±0.29 g/dl), respectively. Over the same period, the results were 2.64±0.21 g/dl for the control group and 2.61±0.15 g/dl for therapy (A1).

Table 03- The protective effect of Cranberry extract on total serum globulin (g/dl) of adult female Wistar rats exposed to zovirax

Mean total serum globulin (g/dl)				
	Groups			
Time	**(C)**	**(A1)**	**(A2)**	**(A3)**
zero	2.67±0.31 A	2.66±0.15 A	2.93±0.28 A	2.87±0.84 A
22 days	2.72±0.38 A	2.64±0.21 A	2.46±0.44 A	2.61±0.14 A
44 days	2.83±0.42 A	2.61 ± 0.15 A	2.32±0.29 A	2.80±0.14 A

Average ± Standard Error for three replicates. Different letters are positioned vertically before the numbers at the probability level (P≤ 0.01) for each set of six animals, according to Duncan's test.

2. Effect of Cranberry extract on total serum bilirubin

When compared to the control(C) set, the results in table (4) demonstrated a significant rise in bilirubin concentration at the probability level(P≤0.01) in the serum of rats treated with the antibiotic zovirax (A2) throughout periods (22, and 44). In treatment (A1), it dropped to 0.33±0.043 g/dl and 0.31±0.045 g/dl at the same exposure periods (22, 44), and the concentration resembled that of the control group. It climbed to 0.49±0.07 and 0.55±0.106 g/dl, respectively. The breakdown of red blood cells over the usual limit causes a rise in the percentage of total bilirubin in the blood, which is the cause of the high concentration of bilirubin in treatment (A2).

Table 04- The protective effect of cranberry extract on total serum bilirubin(TB) concentration(mg/dl) of adult female Wistar rats exposed to zovirax.

Mean total serum bilirubin (TB) concentration (mg/dl)				
	Groups			
Time	(C)	(A1)	(A2)	(A3)
zero	0.34±0.032 C	0.35±0.049 C	0.32±0.023 C	0.33±0.045 C
22 days	0.33±0.029 C	0.33±0.043 C	0.49±0.075 AB	0.40± 0.091 BC
44 days	0.34±0.032 B	0.31±0.045 C	0.55±0.106 A	0.42±0.056 BC

Average ± Standard Error for three replicates. Different letters are positioned vertically before the numbers, signifying a significant difference at the probability level (P≤ 0.01) for each set of six animals. according to Duncan test.

3. The histopathological results of the spleen tissue in various rat groups show:

The spleen's normal anatomy is depicted in Figure 1 of the histological evaluation. Sections of adult female Wistar rats in the control group showed visible white pulp cells and red pulp cells with a central artery.

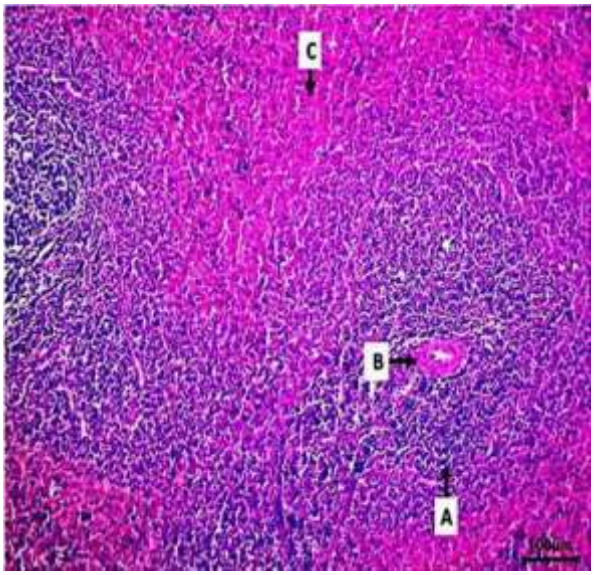

Figure 01- Photomicrograph of rat's spleen of the control group showing normal architecture of white pulp (A) with central artery (B) and red pulp (C) .H & E stain, 100X [21].

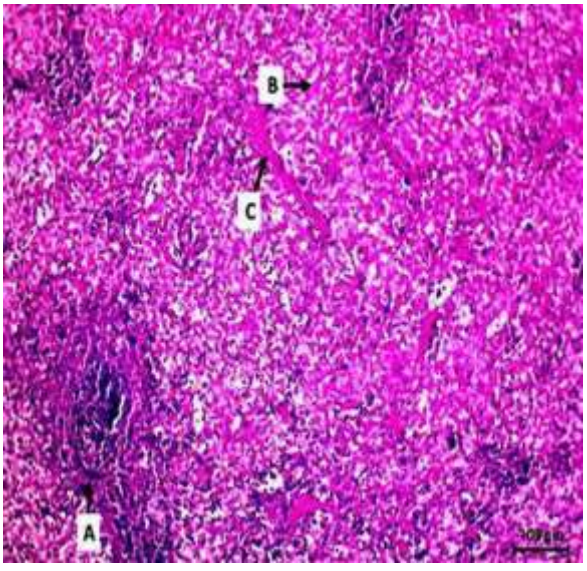

Figure 02- A photomicrograph of a rat's spleen from the zovirax-treated group demonstrates cell loss and necrosis in the red and white pulps (A and B), together with fibrinoid necrosis in the ellipsoidal sheath shown in (C).Stain H&E, 100X [22].

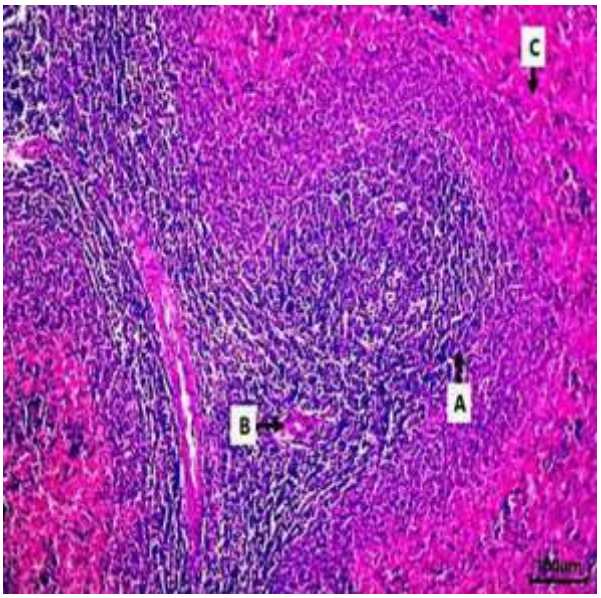

Figure 03- Photomicrograph of rat's spleen of the Zovirax with Cranberry treated group showing normal architecture of white pulp (A) with central artery (B) and red pulp (C) .H & E stain, 100X [23].

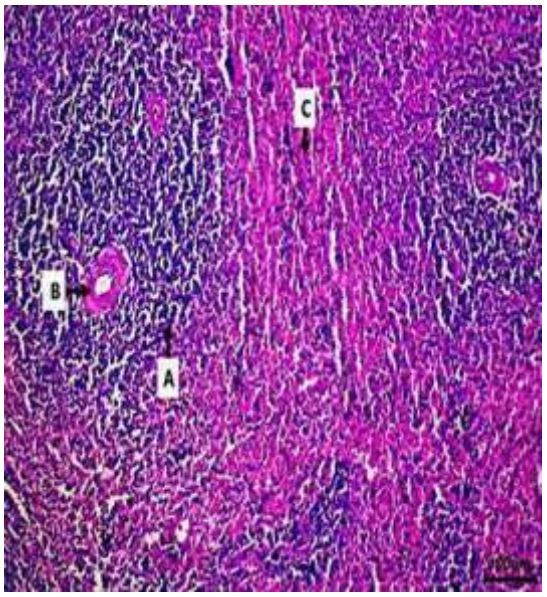

Figure 04- A photomicrograph of the rat spleen from the Cranberry group demonstrates the typical architecture of the red pulp (C) and white pulp (A) with the central artery (B).100X H&E stain [24].

Discussion

The current manuscript investigates the anti-fibrotic properties of cranberries by evaluating different pharmacological, biochemical, and pathomorphological factors in rats with spleen fibrosis produced by Zovirax. This model is extensively accepted and used to study hepatic fibrosis and screen hepatoprotective agents [25]. Solutions containing berry polyphenols have been demonstrated to have cytoprotective effects on the myocardium [26]. Interestingly, the current study's findings support cranberry's protective effects against spleen damage. Cranberry extracts reduce fibrosis and inflammation to lessen Zavriax-induced spleen damage. Compared to the control(C) set, the current study's results indicate a significant decrease in (blood serum protein) levels during the 0, 22, and 44-day treatment periods with the antibiotic zovirax. This decrease may result from the antibiotic interfering with the body's processes of protein synthesis and metabolism, or it could result from an osmotic imbalance brought on by the release of high concentrations of lipid peroxide [27].

The protein degradation that occurs due to the antibiotic's impact on the structural metabolism of proteins in the liver and spleen could be the cause. Because cranberries contain flavonoids, which are very good at reducing spleen damage, using them resulted in a rise in protein levels. These flavonoids prevent or suppress free radicals, which also maintain proteins

[28]. In an investigation of the cranberries' active ingredients and a test of the spleen's blood proteins.

This is a crucial sign for bolstering the defenses against illness and determining the organism's state of health. The findings of this investigation support those of numerous other studies, which show that cranberry extract's potency stems from its high phenolic component content [29]. Furthermore, anthocyanins are critical in lowering the pathogenic activity of root growth. Free oxygen is released into the bloodstream when the body is subjected to substances that damage its tissues or normal metabolic processes occur, according to [30]. Phenolic phytochemicals include phenolic acids (benzoic, hydroxycinnamic, and ellagic acids). Moreover, cranberries have abundant flavonoids (anthocyanins, flavonols, and flavan-3-ols). Cranberries and their products have been found to possess some of the most significant antioxidant capacities among fruits, according to studies that have employed several antioxidant activity measurements [31] [32].

The albumin, globulin, and total protein concentrations significantly increased in the rats treated with cranberries. Therefore, cranberries can be a characteristic to several factors, including the ability of antioxidants such as flavonoids, phenolic compounds, and vitamin C to promote the process of protein synthesis in many organs, especially the liver and spleen, or to the role of cranberries in this regard. That is something that the cranberry contains and is significant. Because they all aim to reduce the use of albumin as an antioxidant, stop the oxidation of proteins and amino acids, and eliminate free radicals, oxidative stress is reduced [33].

The two main components of the spleen are the red and white pulps [34]. The white pulp mainly comprises white blood cells that are important at the start of the adaptive immune response. It is composed of lymphatic tissue that surrounds a central arteriole. B-cells are found in the germinal core, the deepest part of the white [35]. T-cells are present in the periarteriolar lymphoid sheath, which envelops the marginal zone. Red pulp encircles the white pulp seen throughout the spleen. The structure's distinct red appearance under a microscope is caused by a significant number of venous sinuses and splenic cords, or Cords of Billroth.

The reticulin and fibrils produced by the splenic cords provide the organ structure. Additionally, a supply of monocytes that promote wound healing is present in the cords. The splenic cords lead to the splenic sinuses, home to macrophages. They react to antigens and remove aged or aberrant erythrocytes from the bloodstream. The dorsal mesogastrium, referred to as the larger omentum, is home to a mass of mesenchymal cells from which the spleen is

derived. [36]. To compare with the control group, the treatment (A2) showed changes in fatty tissue, vacuolar degeneration, and cell death in the ellipsoidal sheath (Figure) of the red pulp (B) and white pulp (A). In contrast, these tissue characteristics vanished following (A1) cranberry treatment. The rationale is that, as shown in Figure (4), cranberries' ability to shield tissue from harmful substances to which it is exposed because of consuming antibiotics helps to shield the body and its tissues from the harmful effects of an antibiotic. Cranberry extract has been shown to maintain the histological integrity of damaged spleen tissue with parenchyma necrosis, tubular extension, and hyperemic conditions [37].

Cranberry extract was used to improve the structure and reduce the rigidity of the histopathological change in treated rats. It also partially restored the studied parameters to standard values (Figure 4). Flavonoids were discovered during a phytochemical preliminary investigation of cranberry extract. Bioflavonoids, also known as flavonoids, are chemical molecules that can impact many cellular processes and regulate the SOD through the enzymatic activity of superoxide dismutase and catalase. In many research investigations. Cranberries have been linked to antibacterial, antiviral properties, antiangiogenic, anticarcinogenic, antimutagenic, antioxidant, and anti-inflammatory effects [38].

Conclusions

We concluded that the results of this study supported the notion that cranberries, as an antioxidant, can shield adult female Wistar rats' spleens against harm caused by zovirax. Zovirax impacts biochemical measurements such as total protein, globulin, albumin, and total bilirubin. Cranberry extract's spleen-protective properties against oxidative stress brought on by zovirax in female rats. As a component of a fascinating study on the value of natural goods in medicine, particularly cranberry extract. The antibiotic's effects on the liver's and spleen's structural metabolism of proteins may cause protein degradation. Using cranberries increases protein levels because they contain flavonoids, which are excellent at reducing spleen damage. These flavonoids preserve proteins and completely prevent or reduce free radicals. The female rats given cranberry treatment had far higher concentrations of albumin, globulin, and total protein. Cranberries help protect the body and its tissues from adverse effects by shielding tissue from toxins to which it is exposed due to eating antibiotics.

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Conflict of Interest: We confirm that this document is free from any conflict of interest.

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Numerical Investigation On Combustion Characteristics in Compression Ignition Engine Fueled with Hydrogen - Biogas Blends

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Ameer K.alhilo [3]

Abstract

In the current paper, uncontrolled exploitation of the fossil fuel leads to the depletion of energy resources and the environmental degradation of hazard, the researchers tracked to investigate for the alternatives fuel. Term of biogas greenhouse gas, sustainability and obtainability influence of enriched H_2 to synthetic biogas compared with bassline diesel fuel on compression ignition (CI) engines to revealed its characteristics impacted. CFD numerical model based on the chemkine reaction has been procced. In the current research, a mixture of biogas, which contains fixed amounts of (70% CH_4 and 30% CO_2), is mixed with variable amounts of H_2 (up to 20, 40 and 60% by volume) were injected as a main injection along with different of operation load conditions (10% to 90%) on compression ignition (CI) engine working under dual fuel (DF) mode. The extracted results from a numerical model for a single-cylinder, gaseous fuel and pilot fuel injected directly into engine cylinder validated against the data taken from experimental study in identical engine characteristics. Numeral results observed a good agreement for three parameters; in-cylinder pressure (ICP), heat release rate (HRR) and CO_2. The numerical results showed that, peak of ICP slightly increased from (74.2 bar) with BH40 to (77.6 bar) with BH60, also the ICP increased along increased gaseous fuel load. Besides, the first peak of HRR at 10^o CA ATDC is increased by 7.8, 8.8 ,9.8 % for BH20, BH40, BH60 compared for the D100.

Key Words

Compression Ignition Engines; Hydrogen-Biogas Blends; Diesel.

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Introduction

In recent years, internal combustion engine (IC) engines have become widely popular due to their high thermal efficiency and output power. Most of the compression ignition (CI) engines use fossil fuels, which are causing distracting environmental pollution on our planet. In addition to this, gasoline fuel has been restricted because of the persistent presence of sulfur, which results in corrosion and pollutes the environment in a negative way[1]. As a result of limited resources of the fossil fuel , Many developed countries are greatly affected by the import of gasoline because their economic structures have been affected by the import of gasoline [2]. Globally, there has been a significant increase in energy demand as a result of rapid industrialization, population growth, expanding development, and economic growth around the world. To reduce the power demand problem, CI engines from a variety of gasoline sources have been widely used to solve this problem. In the context of addressing these problems, it has been proven that the use of biogas has a positive impact on the problem. As an alternative fuel source for fossil fuel-powered CI engines, biogas can be used as a raw material for the production of CI engines.

The chemical composition of biogas fuel depends mainly on the production method used, as it usually consists of a large proportion of methane gas in terms of the volumetric analysis of the fuel component, which is classified as a greenhouse gas (GHG) instead of carbon dioxide (CO_2). The impact of greenhouse gases on the environment is reduced due to the conversion of methane into carbon dioxide during the biogas combustion process [3]. However, the production of decomposers from organic waste leads to the decomposition of methane resulting in an environmental reduction of greenhouse gases. The presence of CH_4 gas in the biogas composition increases the octane number, which makes it possible to resist knocking, and also makes it suitable for use in internal combustion (IC) engines that require a high compression ratio (CR)[4]. The self-ignition temperature of the biogas is (1087K), While, the temperature of the compressed air in the combustion chamber at the end of the compression stroke typically reaches 850 K in which making pilot liquid fuels initiate the first steps to ignite the biogas fuel. In addition, due to the lower C/H ratio of biogas compared to conventional diesel fuel, in this way, the use of biogas as a base fuel with a small amount of pilot diesel leads to a significant reduction in pollutant emissions (CO, CO_2 and particulates)[5]. Besides, this gives a promoting the in-cylinder temperature reduction during the burning process under very lean conditions or diluted air-biogas mixture which consequently attains lower NO_x emissions and expanding flammable limits. Hence, the mixture composed of diesel-biogas is considered suitable for use

in CI engines according to two factors, firstly, biogas economical production from an organic waste source. And secondly, reducing the emissions pollutants produced from combustion while the efficiencies stay maintaining in comparable fuel[6]. In the past few years, many studies have been conducted regarding the use of biogas in CI engines, as well as studying the combustion characteristics of fuels with biogas produced from different sources [7]. Bari *et al.* [8] investigated the effect of CO_2 concentration in mixtures of biogas-fossil fuel in the CI engines, and the authors reported that increased BSFC corresponded to CO_2 concentrations of more than 20-30% by volume in biogas mixtures. Also, the authors state that the presence of CO_2 within biogas mixtures leads to a weaker mixture in the combustion chamber which in turn reduces the local flame temperature due to lower energy absorption. Moreover, the researchers observed a decrease in BSFC when the CO_2 concentration was less than 20%, which was due to the oxygen concentration generated by the dissociation of CO_2 resulting in a shorter ID period and improved combustion oxidation process. Yoon & Lee [8] conducted an experimental comparative study to evaluate the combustion and emission characteristics of CI engines operating on single mode (primary diesel fuel) and dual mode (biogas-diesel blend) mode.. For the engine operating in dual-fuel mode, the ID was observed higher than that for the single mode fueled by diesel-only engines. Due to the slightly lower local flame temperature of biogas and diesel mixture and higher specific heat capacity corresponding to lower exhaust gas temperature, and NO_x emissions [9]. Hence, biogas constituents that containing have a higher concentration of CO_2 reducing calorific value, flame propagation speed, and longer ID which contributing to reduction in thermal efficiency. In addition, removing CO_2 fraction from the biogas constituents is costly in process consumption and required substantial energy.

Henham *et al.*[10]investigated the CI engines performance and emissions characteristics with variations in CH_4-CO_2 ration, authors report increasing the CO_2 concentration at the expenses of CH_4 in the biogas constitution renders obvious thermal efficiency reduction alongside of reduction of CO emissions. Furthermore, it was demonstrated that increasing the concentrations of CO_2 in biogas blends resulted in the slight increase of carbon dioxide emissions, while the exhaust gas temperature decreased due to the decrease of CO emissions. There is a study by Bari *et al.*[11] that examines the performance of the CI engines dual fuel mode and evaluation of emission sensitivity to CO_2 concentrations. The authors declared that the performance of the CI engines dual fuel mode was improved by the addition of 20-30% CO_2 by volume to blends of biogas, in other words, it reveals the reduction in brake specific fuel consumption associated with sweeping CO_2 percentages greater than these limits the performance deterioration. They also reported that at concentrations of less than 20% CO_2, the

process of dissociation into CO and O_2 leads to improved combustion characteristics and performance. While, when the % CO_2 is higher than acceptable limits, major of CO_2 amounts stay undissociated and act as the diluted gases restrict the flame propagation of the burnt mixture.[12]performed study of biogas composition having different ratio of CH_4-CO_2 blends 95:5, 85:15 and 60:35 . The authors declare that, at low load there is noticeable increasing BSFC for dual fuel mode. Also, they suggested to procced engine parameter modification like as advancing start of injection (SOI) and exhaust gas recirculation (EGR) impetrate to improve combustion and performance characteristics. According to al.,[13] there were lower emissions of particulate matter (PM) and NO_x, and higher emissions of CO and HC. However, when CH_4 concentrations varied from 70-60%, no significant differences were observed.

It has been noted in recent years that a wide variety of alternative fuels have been investigated, including hydrogen. In addition to being environmentally friendly and cost-effective for the production of hydrogen, the water splitter can also be used as a method of generating hydrogen for a variety of other environmentally friendly uses, such as thermoplastics, thermal processes, petrochemicals, and photovoltaics[14]. There is no doubt that water electrolysis can contribute to addressing today's environmental and energy concerns by providing a clean, efficient, and cost-effective alternative to hydrogen production. Hydrogen is characterized by higher laminar flame speeds and a broader range of flammability limits than other gases.

It is suitable for use in IC engines as a result of its molecular structure and properties[15]. Compared to conventional fossil fuels, hydrogen is a clean and carbon-free energy source. As opposed to hydrocarbon fuels, hydrogen combustion produces no hydrocarbons (HC), carbon dioxide (CO_2), or even carbon monoxide (CO). Increasing the hydrogen share in a hydrogen-diesel dual fuel mode will result in a reduction in the production of CO_2 and particulate matter emissions as well. There is a correlation between higher NO_x emissions in this case and a greater proportion of hydrogen in the energy input. It is found that NO_x emissions are higher when hydrogen shares exceed 10%. It is reported that in [16], a hydrogen augmentation system that can be controlled by the accelerator pedal is developed. In terms of cost, this results in a reduction of CO_2 and NO_x emissions, but when it comes to the environmental impact, the particulate matter emissions are increased.

Several studies have been conducted on the co-combustion of hydrogen with dual fuels, including Juknelevicius *et al*.[17]. A study is being conducted to examine the effect of hydrogen energy sharing with a variety of loads on auto ignition delay, combustion intensity, and

emissions at various loads. AVL boost engines are being developed to study and validate the effect of varying hydrogen share rates at fixed injection timings on combustion in CI engines. The low hydrogen energy share has no significant effect on combustion duration in low load conditions, while a reduction in in-cylinder pressure has a substantial effect on BTE and BSFC. According to Karagoz et al., a method of injecting hydrogen into the intake manifold by using an LPG-CNG injector that is controlled by the ECU has been proposed for this application. Experiments are conducted to determine the effects of adding 0%, 22%, and 53% hydrogen to the total energy supply on emission and performance characteristics[18]. When hydrogen is provided at low load, heavy-duty engines using diesel-hydrogen blends produce reduced carbon and NO_x emissions by over 90%, while soot emissions are reduced by 85%. The NO_x emissions are increasing at medium loads, which is the primary challenge that needs to be addressed *et al.*[19]. A single-cylinder direct injection diesel-hydrogen engine has been studied by Yadav *et al.* [20]in regard to its performance and emissions characteristics. A great deal of performance can be improved and emissions can be reduced by using hydrogen-enriched air in the fresh charge intake along with 10% and 20% EGR.

A prominent alternative fuel that is produced domestically is biogas, which is often used as a dual fuel with diesel to equate to premium fuel. However, changing diesel fuel with a higher-rate fuel does come with certain challenges, including significant reductions in the engine performance and rough running of the CI engines during the operation process. The hydrogen enrichment of biogas has been shown to be one of the most promising technical approaches to improving biogas combustion. In fact, hydrogen is one of the cleanest fuels on the market for IC engines. In addition to the fact that it contains no carbon as it burns without releasing any carbon dioxide pollution into the atmosphere. There are many advantages of hydrogen, in addition to its high calorific value (120 MJ/kg for hydrogen, compared with 23 MJ/kg for biogas), including its fast combustion rate, a wide range of flammability, and a high diffusivity in air, a short ignition delay, and a wide range of flammability, as well as a small quenching distance. The purpose of this study was to investigate the impact of adding hydrogen gas to single-cylinder CI engines powered by biogas-diesel blends on their performance. Diesel, biogas-diesel, and hydrogen-biogas-diesel were compared based on their performance and emissions characteristics. This paper examines a direct injection, single cylinder diesel engine operating in dual fuel mode with very small amounts of diesel fuel as pilot fuel and biogas with composition of $CH_4 + CO_2 + H_2$, as the engine's main fuel. A study was conducted to evaluate the improvement in combustion efficiency of biogas when the H_2 ratio was enriched with

various levels (10,20,30% by volume), while maintaining constant the baseline fuel rate of the pilot diesel fuel.

For all of the cases examined in this study, we used the conventional compression combustion mode with diesel as the pilot fuel. while, the main fuel direct injected into engine cylinder was biogas composed which containing CH_4 (70%) and CO_2 (30% by volume). Meanwhile, one of the main objectives of the present paper was to investigate the effects of H_2 addition on the quality of biogas combustion characteristics, adding hydrogen could be practically enriched by a ratio (up to 0,10,30%).

Numerical methods

An adequate pool of scientific results was compiled by utilizing three significant online databases in order to collect satisfactory list: Web of Since, Science Direct and Scopus. There were two steps that were taken as part of this process. To begin with, a search was conducted using the keywords "biogas", "compression ignition engine" and "sustainability" in the largest database (Science Direct). Articles for this publication should address both reducing emissions and introducing sustainability innovations simultaneously, focusing on more than just cost and production sustainability. We received more than 200 results during our initial screening process. It was therefore decided to limit the search to English-language scientific articles that have been published within the last five years, between 2016 and the present date. This was to keep up with the latest achievements in the review. The outcome of this step was the reduction of 89 items to 25 items, while the selection was based on the methodology strategy and the operation of the fuel test.

An analysis of engine performance and emission characteristics under variable biogas-hydrogen ratios has been conducted using a computational fluid dynamic model. A three-dimensional domain was used in order to simulate the injection and combustion processes in CI engines. By determining the boundary conditions within a combustion chamber of a diesel engine that are time-dependent, one can determine the thermophysical properties of mixtures within the combustion chamber of the engine. A numerical study was conducted on direct biogas-hydrogen injection and injector systems. It has been examined in detail how different biogas-hydrogen injection ratios and timings affect engine performance and emissions. In this research, a four-stroke, single-cylinder, direct-injection engine is used to carry out the test. From literature provide in-depth analyses of the specifications of the diesel engine that is used for current numerical analysis, as well as a description of its construction. A numerical analysis of

hydrogen injection in CI engines has been carried out by modifying design parameters of the diesel engine. The following table (1) shows the major engine parameters and operating conditions of diesel engines in accordance with the information contained in the table. Biogas-hydrogen fuel has also been injected directly into engines alongside diesel fuel. When compared with fossil fuels and biogas, hydrogen and biogas have many differences in terms of their physical and chemical properties. Biogas, hydrogen, and diesel fuel are listed in Table 1 as their most significant physical properties. Due to its carbon-free composition, hydrogen is used in internal combustion engines as fuel, which is a safe and clean fuel. A primary reason for hydrogen's popularity as an alternative fuel is its high energy density and low heating value (LHV)[21].

Table 01- Diesel, biogas, H_2 properties

Property	Diesel fuel	Biogas	Hydrogen
Density (kg/m^3)	852	1.46	0.083
Calorific value (MJ/kg)	42.35	22.97	120
Cetane number	49	-	-
Octane number	0.5	0.2	2.7

Numerical model

Numerical studies require the selection of the computational domain and the modeling of it in a proper manner. The numerical analysis of combustion is optimized by choosing a geometry model, spray physics, and chemical processes that are appropriate for internal combustion engines in order to achieve an optimized outcome.

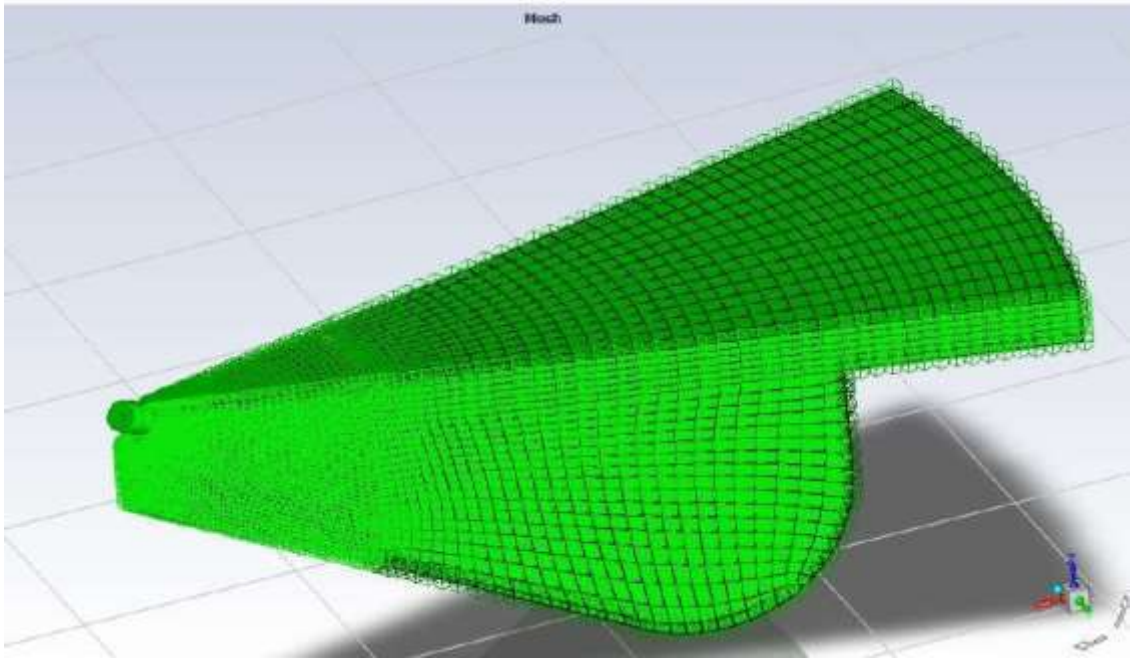


Figure 01- Illustrates the diesel engine injector design expressed in the plane of axial cross-section.

Fig.1 illustrates a three-dimensional sector mesh which is determined by the number of injector nozzles in diesel engine injectors. Based on a sixty-degree sector of meshes which are either structured or unstructured elements, a computational model with approximate 505,890 elements is constructed [22]. In the combustion chamber, the geometry is shaped like a bowl to improve the quality of the air-fuel mixture by creating a high level of turbulence. There are various components that need to be assembled in a symmetrical 60-degree geometry, including the cylinder head, the liner, the piston, and the nozzle that will be used for biogas-hydrogen and diesel fuel injection. The position of injectors and nozzles plays an important role in combustion efficiency as a result of the interaction between diesel and biogas-hydrogen jets[21].

Boundary and initial conditions

Under different injected ratio of biogas-hydrogen blends, combustion analysis was performed during compression stroke, there was a close condition on both the exhaust and intake valves at the same time. With the exception of fuel injection, the combustion chamber is considered a closed system during this period. The combustion model was obtained at 1200 rpm speed engine under constant low load operation. It takes into consideration solid walls, periodic and symmetrical wall motions as well as internal flow during a numerical analysis. It is therefore crucial to determine the optimal boundary conditions for the accurate analysis of a three-dimensional numerical model to ensure the most precise results. It is possible to predict the heat flow between combustion chambers and the rest walls using the theory of conduction and convection based on computational models. Heat transfer between engine internal walls due to convection heat flow significantly affects diesel engines' performance and efficiency. In this study could determine the temperature of the combustion chamber, piston, and internal liner wall as well as chemical species and solvents present in the combustion chamber. A facilitated nheptane (C_7H_{16}) mechanism was used in this study to identify combustion characteristics of a pilot diesel engine. It is estimated that there are 35 species and 173 reactions involved in the n-heptane reduction mechanism. This gets converted into diesel fuel as part of the combustion process. Tetradecane ($C_{14}H_{30}$) is one of several chemical formulas used to describe diesel physical properties. From the Ansys chemkin-subroutine software, the thermodynamic properties of fuel-reduced mechanisms are extracted.

From the literature review ,the k-ε RNG turbulence model prove reliability and producing astilbe accurate result in the internal combustion (IC) engine problem compared to the other model such as k-ε , Reynolds-averaged Navier-Stokes (RANS)[23]. The air-fuel mixture statues play a very important factor which it improved by creating eddies withing combustion

chamber geometry. Using k-ε model to simulating combustion problem producing high accuracy result due to adding term for the adaptive turbulent flow equation (ε). In addition, user-defined k-ε standard equation and k-ε RNG models have two variables of imperative conditional terms which give the potential to rigid boundary walls[24].

The approach to calculating the distribution of air and fuel within a combustion zone was used Euler-Lagrange flow modelers. Additionally, Kelvin-Helmholtz/Rayleigh-Taylor (KH-RT) has also been proving to be one of the most important models for demonstrating the distribution and atomization of liquid pilot fuel. The numerical model and methods utilized in this simulation could be summarized in the following Table 1.

Table 02- Summary of the models used in current numerical stud models

Description	Models used
Solver	Pressure based type (Transient)
Dynamic mesh	Layering (In-cylinder options)
Spatial discretization schemes	Higher order upwind
Turbulence	k-ε RNG
Flamelet models	turbulent Flamele approach
Turbulence chemistry interaction (Combustion)	Non-Premixed Model
Diesel pilot fuel injection Spray model	Discrete phase model (DPM) Solid cone model
NO_x emission	Thermal NO_x and Prompt NO_x

For the CFD combustion solver, Table 2 contains the initial boundary condition to proceed with the simulation of the CI engine. Figure 1 shows the combustion chamber geometry is defined for the CFD solver, which represents the air-fuel mixture domain. It is necessary to validate the results from the CFD solver with experimental data. Then, the numerical model is reliable and ready for CI engine simulation.

Table 03- The specification of standard diesel engine

Parameter	Value
Type of engine	four strokes, water cooled, single-cylinder
Displacement	566 cc
Bore & stroke	88 mm&110mm
Compression ratio (CR)	17.5 mm
Length of connection rod	303 mm
Fuel	Diesel(D100), biogas- hydrogen (BH20,BH40,BH60) blends
Load operations	10,30,50,70%
Fuel injection timing	23° CA before TDC
Inlet valve closing (Ivc)	35.5° CA after BDC
Exhaust valve opining (Evo)	35.5° CA before BDC

Governing equations

Simulation of a compression ignition (CI) engine by numerical model requires appropriate governing equations that incorporate spray pilot fuel dynamics, turbulent heat transfer, and thermodynamic fuel properties. The Navier-Stokes equations have been used to describe gas-phase flow properties in this study. In this equations, momentum and energy transport equations, as well as continuity equations, are expressed together. An expression for the continuity equation of a combustion chamber that is set for a specific control volume is given below[25].

$$\frac{\partial \rho}{\partial t} + \frac{\partial (\rho U_j)}{\partial X_i} = S_k \quad (1)$$

The ρ in the equation is fluid density, $U_j$the internal flow velocity, while the spray evaporation and chemical reaction represented by the S_k . The Navier-Stokes momentum equation is defined as follows.

$$\frac{\partial \rho U_i}{\partial t} + \frac{\partial (\rho U_i U_j)}{\partial X_j} = -\frac{\partial p}{\partial X_i} + \frac{\partial c_{ij}}{\partial X_j} + F_i^s + F_i^b \quad (2)$$

Where F_i^b, F_i^s,$\boldsymbol{\tau}$, and p body force, the induced spray source term, and tensor of viscous stress, respectively. It is common practice to express energy conservation equations numerically as a function of the thermodynamics of the combustion chamber. There are a number of thermodynamic properties that can be defined as enthalpy, energy, and specific

internal energy, which come from chemical energy, the following equation represented the transport energy equation.

$$\frac{\partial \rho e}{\partial t} + \frac{\partial (\rho e U_j)}{\partial X_j} = -P \frac{\partial U_j}{\partial X_i} + \frac{\partial J_j}{\partial X_j} + Q^s + Q^c \qquad (3)$$

The combination of enthalpy diffusion and heat conduction results in total heat transfer, which is referred to as sensible energy e. Q^s, Q^c are symbolics of the chemical reaction that occurs within the reaction and the source of diesel fuel, respectively.In this study, the turbulence model has been determined by using k-ε RNG which combines turbulent kinetic and turbulent dispassion energy together as shown in Equation () [26]. In addition , the k-ε RNG mathematical driven technique is based on the "Renormalization group".

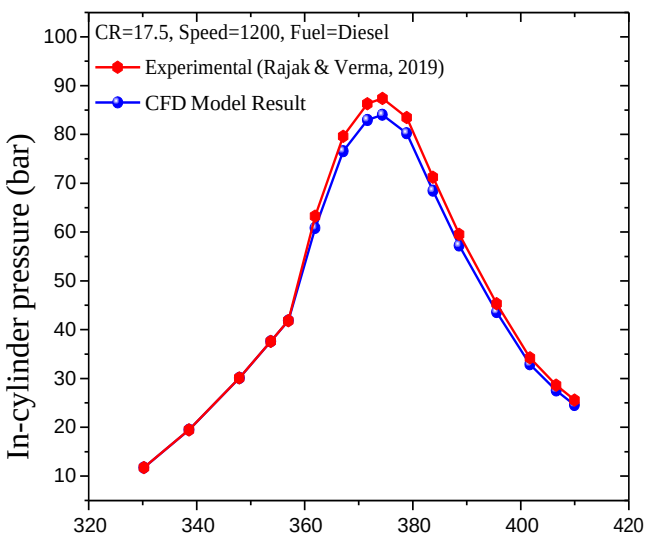
$$\frac{\partial}{\partial t}(\rho k) + \frac{\partial}{\partial X_i}(\rho k u_i) = \frac{\partial}{\partial X_i}(a_k \mu_{eff} \frac{\partial k}{\partial X_j}) + G_k + G_b - \rho Y_m + S_k \quad (4)$$

$$\frac{\partial}{\partial t}(\rho \varepsilon) + \frac{\partial}{\partial X_i}(\rho \varepsilon u_i) = \frac{\partial}{\partial X_j}(a_s \mu_{eff} \frac{\partial s}{\partial X_j}) + C'_{1s} \frac{s}{k} G_k + C_{3s} G_b) - C_{2s} \rho \frac{s^2}{k} - R_e + S_s \quad (5)$$

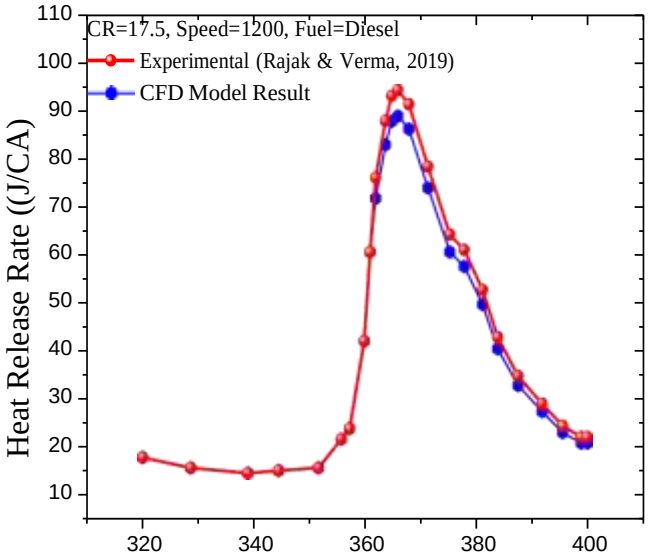
The turbulent kinetic energy energy determined in the equation by the two terms G_k and G_b driven from the bouncy effect and velocity average gradient. Moreover, in through of turbulence incompressible flow, the effect of fluctuating dilation on the rate of total dissipation represented by the Y_m. $a_k and$ a_s represented the Prandtl number effective for the kinetic and dissipation energy equations respectively. while, the source term for the kinetic and dissipation energy equations, respectively. There are some of variables that used in the equations could be define as the following.

Model validation

The CFD model needs to be reliable. For this purpose, reference studies should be used to verify numerical results. Validation of numerical studies relies on a variety of methods as described in the literature. Experimental results were used as benchmark reference results by the [27] carried on the CI engine, single cylinder, constant engine speed at (1200 rpm) , four stroke and the operation run under 25% load condition . For the identical operation condition, Fig.2 shows the comparison experimental and CFD model results of the very important parameters in-cylinder pressure, and heat release rate versus crank angle degree. There has been a comparative analysis conducted generally between the crank angle degree period between intake valve closing (IVC) and exhaust valve opening (EVO) . In light of the results of the analysis, it is evident that there is a good agreement between the experimental and numerical results.



(a) Crank Angle(deg)
In-cylinder pressure as function of the crank angle



(b) Crank Angle (deg)
Heat release rate as function of the crank angle degree

Figure 02- The X value represent higher value obtained, while the minimum value represented by Y

For numerical and experimental results with constant compression ratios at low load operation condition, error deviation analysis was conducted by the following Equation (2), the result of deviation error analyses obtained which are 3.6%, 4.2% for in-cylinder temperature, and heat release rate .

$$ED = \left(\frac{X-Y}{X}\right) \qquad (6)$$

Results

The CI engines suffer from knocking and excessive engine temperatures caused by pure hydrogen, which negatively impacts the components of the engine. Thus, blending the hydrogen with fuel has a higher-octane number is an effective method that is being investigated as a method of overcoming these negative impacts. CFD simulations were conducted on a diesel engine to investigate hydrogen injection with different biogas-hydrogen fuel ratios and timings, and diesel fuel was directly injected into the combustion chamber and kept at a constant ratio throughout the test. The performance and emissions of diesel engine were investigated at specific boundary conditions imposed by various biogas-hydrogen ratios and injection timings.

Performance characteristics

In comparison to other gases, hydrogen has a higher diffusivity speed rate which increasing air-fuel mixture quality. Because of its ability make it a suitable fuel for diesel engines, as it enhances their performance and efficiency.

In-cylinder peak pressure

Figure (3) demonstrates the variation of the in-cylinder pressure (ICP) over the crank angle degree (CAD) for the pure diesel (D100) and different ratio of biogas- hydrogen (BH20, BH40, BH60) blends. It is observed that, for all the test condition the peak of ICP was enclosed between 358^o and 375^o CAD. For the current dual-fuel combustion mode, the combined of SI and CI have been carried out. Biogas-hydrogen blends are directly injected into engine cylinders during the last stage of the compression stroke. Due to its higher cetane number, injected pilot diesel fuel provides significant ignition source during the premixed phase. Though, air-fuel charge surrounding the sprayed pilot diesel spontaneously combusts. The ICP reach the maximum value in this range due to the fuel injected at 15^o CAD BTDC which take about 12^o CAD to initiating the ignition process of fuel. From the Fig.3 it can be seen the peak of in-cylinder pressure for the D100, BH20, BH40, BH60 fuel blends were calculated 66.4, 70.2, 74.3, 78.5 bar, respectively. Fig.3 shows clearly, in diesel engines with a higher ratio of hydrogen injection, pressure increases by at least 9% compared to pure diesel engines (D100). It has been suggested that adding H_2 fuel to diesel-fueled CI engines will result in better combustion of the fuel. This increase in ICP is attributed to the better mixing of fuel blends with air, driven by a higher diffusivity of H_2 (0.63 cm2/s) compared to D100.

The current study is designed for medium load, where pilot diesel injection quantity is higher than low load in conventional CI engines. In this engine of dual-fuel mode, pilot diesel quantity remains constant unlike to the hydrogen ratio in biogas-hydrogen blends which is continuously changing. The findings of santoso *et al.*[28] a higher hydrogen amount was found to have a positive effect on combustion starting early and increasing ICP [29]. According to a study conducted by Swaja *et al,* [30]. to investigate the effect of hydrogen addition on diesel engine characteristics, they reported that pilot diesel fuel at medium load and in dual-fuel mode (DFM) formed a greater number of ignition source centers due to its significant spray characteristics and pilot diesel fuel mixture characteristics.

Thereby increasing combustion rate and ICP excessively. Therefore, increasing hydrogen energy share increases flame velocity propagation, which leads to increased burning turbulence

and complete oxidation. The peak of ICP increased from (67.4 bar) based on single diesel fuel (D100) to (68.9 bar) based on dual-fuel mode where the testing blend is BH20. Similarly, the peak of ICP slightly increased from (74.2 bar) with BH40 to (77.6 bar) with BH60 as shown in Fig.3. Liew et al.[31] conducted an experimental study to investigate hydrogen addition on the variation of in-cylinder pressure at medium load (60%) and low load (15%). Based on the authors' conclusion, the peak of in-cylinder pressure decreased corresponded to increasing the hydrogen volume share (HVS), this trend was evident under low-load operation conditions. This previous literature analysis suggested that adding hydrogen to dual-fuel engines at high and moderate loads cause to increase in-cylinder pressure, in contrast to low loads which it causes to decrease in-cylinder pressure.

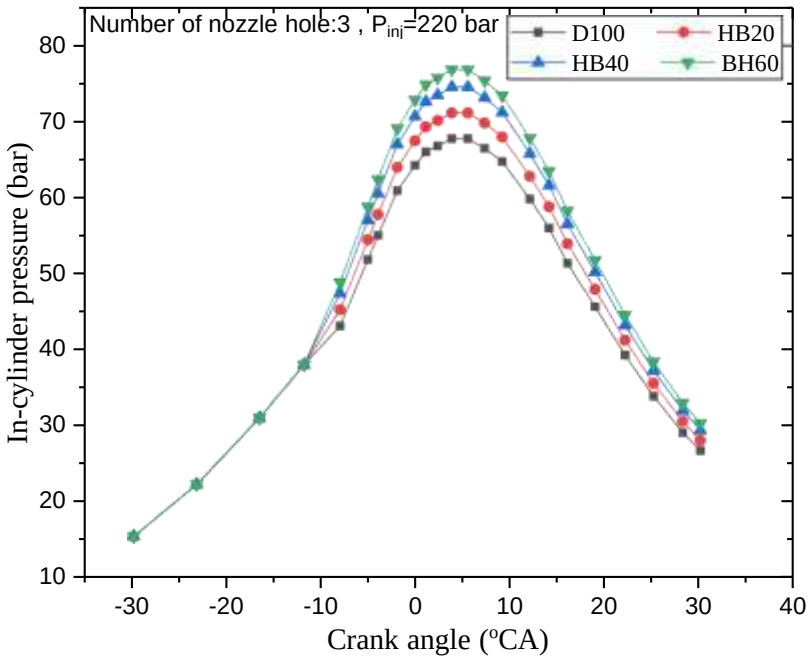


Figure 03- The In-cylinder pressure as function of crank angle degree

Heat release rate

It has been found that the heat release rate (HRR) of CI engines in Dual Fuel Mode (DFM) is divided into three major phases based on the fuel combustion stage. The heat released from premixed combustion of major quantity of pilot diesel fuel. It has been clearly observed that the HRR profile curve is divided into three distinct phases, as discussed previously. The first stage is characterized by a high rate of HRR, a sudden increase in pressure, and an increase in the rate of combustion. Whenever the fuel-air mixture in the combustion engine reaches the ideal level, the combustion engine will operate at its peak efficiency. Increasing combustion performance at early injection timings can be attributed to the fact that most of the fuel that is injected at advanced crank angles is being consumed by the engine. Upon reaching a sufficient

amount of oxygen in the fuel cylinder, the flame ignites, resulting in the temperature and pressure in the cylinder both increasing, resulting in the second peak in the rate of HRR. As long as there is a sufficient amount of oxygen present in a combustion chamber and the unburned fuel or parts that are partially burned remain in the combustion chamber, combustion will continue and heat will be released. According to the results of this study, the HRR increases with an increase in H_2 injection, which corresponds to an increase in combustion efficiency.

Accordingly, this situation can be explained by the fact that the early ignition delay period is shorter as a result of increasing hydrogen injection, a slow pre-ignition reaction, and the rapid combustion properties of H_2. As shown in the Fig.4 , varying the hydrogen energy share has a positive effect on the heat release rate. It is observed that as the hydrogen energy share increases the HRR increases simultaneously at each stage of combustion, as well during combustion, when fuel is in contact with a sufficient amount of oxygen, an increase in the rate of heat release occurs in the combustion chamber. The first peak of HRR at 10^o CA ATDC is increased by 5.4, 7.3, 8.4 %, for BH20, BH40, BH60 compared for the D100. while, the second peak of HRR at 30^o CA ATDC increased by 7.8, 8.8 ,9.8 % compared for D100. In both the first and second combustion periods, hydrogen addition to the gaseous fuel contribute to an increase in the peak of HRR and intensity. It has been found that this phenomenon is resulted from the effect of H_2 properties (calorific value, flame speed) in which improving combustion reaction process in account of heat released. It could be attributed to an enhanced diffuse hydrogen-enriched flame that burns more fuel during the early stages of combustion, resulting in a reduced heat transfer rate at the end of combustion.

Contrary to the biogas operation at the same conditions, the H_2 introduced mixture has an earlier peak HRR value and a higher PRR value than the biogas mixture, this is caused by the fact that H_2 burns at a higher flame speed than other gases[28]. Talibi *et al.*[3] they found that H_2 addition to biogas led to a slight increase in the peak HRR value, in addition to the peak HRR sweep. As a consequence of the introduction of H_2, a higher flame propagation speed on a mass basis can be achieved, as well as a higher heating value.

Due to absorbing the surrounding fuel -air mixture's heat, the engine working under dual-fuel modes have negative HRR profile curve during the ignition delay period. It is also important to note that the pre-mixed fuel also has a high specific enthalpy, which contributes to the fact that it has a negative HRR value. Immediately after starting the pre-injection, the pilot fuel is ignited along with a small amount of surrounding air-fuel mixture and a very little

quantity of gaseous fuel that entraining into the pilot fuel with just a few seconds to go after starting the pre-injection[32].

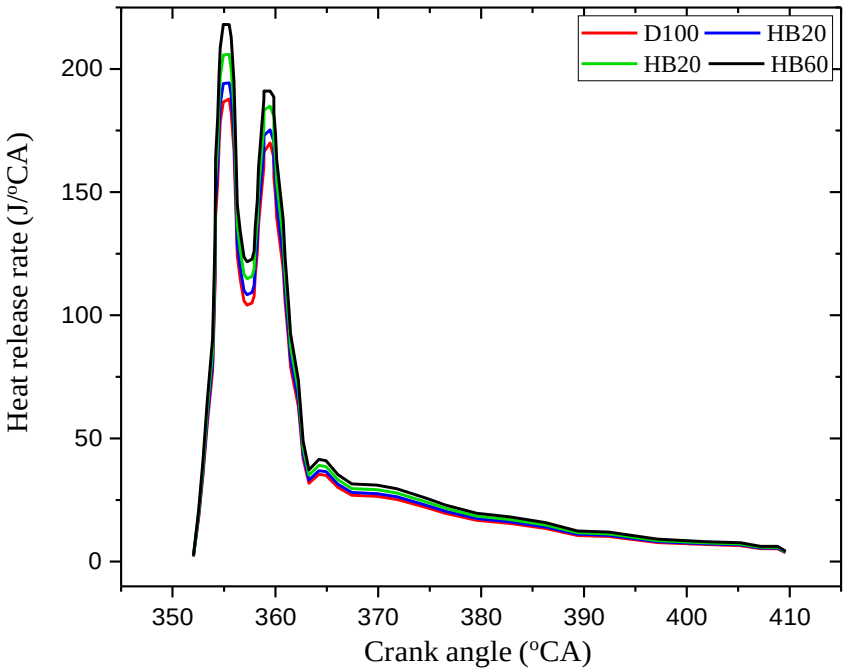


Figure 04- The heat release rate as function of crank angle degree

As a function of crank angle, the diesel line pressure curves in the bottom boxes clearly show the pre-injection stage to be the initiation point of combustion in the dual-fuel modes, While, the main-injection strategy is dominating the combustion of conventional diesel fuel. The chemical energy of the air-fuel mixture is released as soon as the dual fuel modes ignite due to the homogeneous mixture of fuel and air. The multiplicity of burning cores in the homogeneous charge explains most of the abrupt changes in the combustion characteristics caused by the multiple burning cores.

Brake specific fuel consumption

Based on the simulation data shown in Fig .5, it can be concluded that the effect of blending biogas with H_2 on the performance of an engine can be described by the brake-specific energy consumption of the engine (BSEC). Diesel engine working under DF mode for H2/biogas is compared with the conventional engine diesel mode for this performance characteristic. According to the BSEC results, it appears to be true that the results obtained under low to medium engine loads, regardless of the type of primary fuel used, are consistent with the hypothetical dual fuel results that would normally be obtained. Based on the figure, it is evident that the BSFC for conventional diesel engines (CDE) with baseline diesel fuel at 20% of the full engine has a higher BSEC by 30% than that for diesel engines with H2-Biogas blends working in DF mode. In these circumstances, the failure of the engine is primarily a result of a

combination of factors, primarily low quantities of pilot fuel, low biogas characteristics, and low concentrations of biogas in the air, as well as local operating conditions, such as slow propagation of flames throughout the whole charge within the available timeframe, are the main reasons why the engine fails. This means that the flames generated by the auto-ignition of the pilot fuel cannot sufficiently spread into any surrounding lean mixtures of gaseous fuel in order to complete the oxidation process. This results in weak oxidation of the gaseous fuel mixture. If the engine load is increased further, the BSEC values will differ slightly between the values for the CDE mode in comparison with those for the DF mode until both combustion modes are reverted at full engine load, resulting in a slight variation in BSEC values.

While, the BSFC for the CDE mode lower than DF mode with BH20 by 18, 24, 30% for operation engine load 10%, 30, 50, 70, 90%, respectively. By increasing the percentage of gaseous fuel in an engine while keeping the mass of diesel fuel constant, the engine load can be increased by increasing the volume of gaseous fuel. This leads to the production of higher temperatures and the propagation of flames as a consequence of such richer mixtures. It should be noted that when the dual-fuel mode is employed, the main gaseous fuel burns inefficiently, as well as that gas fuels have a much lower volumetric heating value than diesel fuels.

To achieve the same amount of power from the gaseous fuel, it is necessary to load the cylinders with a greater amount of main fuel injection in order to get identical output power. Both synthetic biogas and baseline diesel fuel(D100) operations have approximately similar BSFC values at low and intermediate loads, which indicates that the same amount of fuel is required to produce the same amount of power. In contrast to the D100, the biogas BSFC slightly decreases at high loads. Due to the dissociation of CO_2 at high temperatures, the BSFC will drop as the CO_2 dissolves into CO and O_2, which is the cause of the drop in BSFC.

When the energy share rate (ESR) of hydrogen increasing in the blends of biogas the BSFC decreased at low load as observed from Fig 5, in this manner the BSFC was (625 : BH20),(616 : BH40) ,(600 g/kWh: BH60) at 10% of fuel operation load. This is relative to the H_2 ability to improving combustion process reaction due to hydrogen wide the limit of lean flammability. Since the H_2 has the capability of widening the lean flammability limit, it is able to improve the combustion reaction as a result. Numerous studies investigated the effect of H_2 on BSFC[33][5][34], and they found a correlation between an increase in ESR of H_2 and a reduction of BSFC. It is important to understand that turbulence generated within the combustion chamber is one of the most important factors that influence the homogeneous distribution of fuel within the chamber.

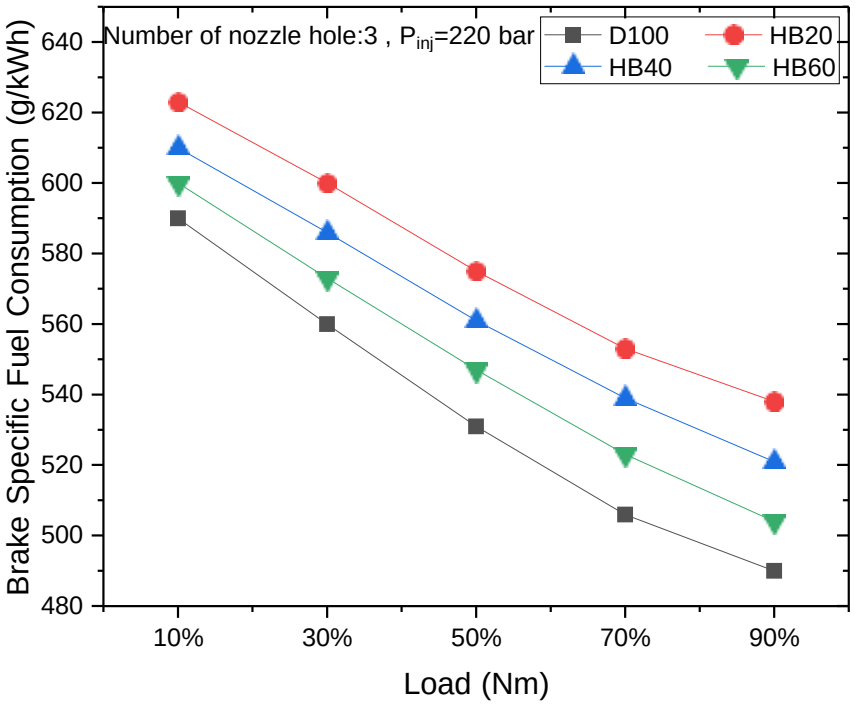


Figure 05- The brake specific fuel consumption as function of load

Emissions characteristics

HC emissions

As a result of the incomplete combustion reaction process for engines that operate on hydrocarbon fuels, there are HC emissions that are produced. Fig.6 shows the variation of HC concentration as a function of operation load for H_2/biogas blende and bassline diesel fuel.

It is clear that the emitted HC is generated clearly and significantly when the fuel is in the gaseous phase, and its rates change between (78.1) and (74.8g/Kwh) for BH20 and BH60. The larger amount of hydrocarbon compounds emitted from the combustion reaction process of fuel which leads to drop the efficiency of combustion in all its stages, as well as to the escape of hydrocarbon compounds that do not go through the burning combustion process to the piston bowel and squashing region [28]. As these areas do not reach the combustion flame, which are considered as areas of accumulated fuel charge loss. Thus, the unburned fuel charge accumulated in the piston bowl constitutes the majority of the researchers' interest.

At the end of the combustion process, unburned fuel is discharged along with the gases that result from combustion. When operating under low load conditions, as this is more common and this condition is more noticeable. The combustion conditions proceeding under low temperature, which is one of the results of the lean fuel mixture and low injection pressure,

which reduces the rate of fuel oxidation. Therefore, the most significant unburned HC emissions are methane gas, often in very excessive concentrations[35].

Simultaneously with the increase in engine load, the equivalence ratio of the fuel increases proportionally, the cores that initiate ignition will also improve, the intensity of turbulence of the fuel-air charge increases, and therefore the richer mixture will combust at the highest temperature. The fuel spray characteristics also improve significantly, which significantly enhances the combustion rate and increases the oxidation process of the unburned HC emissions. as shown in Fig.6, The HC as a function of operation load, its clear obvious for the DF mode the HC emissions was lowest compared to the conventional diesel mode . A higher diffusivity of gaseous fuel helps fill the combustion chamber's gap. In this manner, increasing the engine operation load leads to increasing gaseous fuel quantity in the manifold intake charge which promoting combustion reaction process. It has been noted that increase in relative concentration of gaseous fuel continues to enhance the combustion area and increase the spreading of flame in the largest part of the gaseous charge, which consumes a large amount of fuel. The result of this is a significant improvement in the combustion process, which is evident when the engine is running at high loads, as HC emissions have been reduced by approximately 50% when compared with diesel emissions. Researchers in literature reached the same conclusion[36][37][38][6].

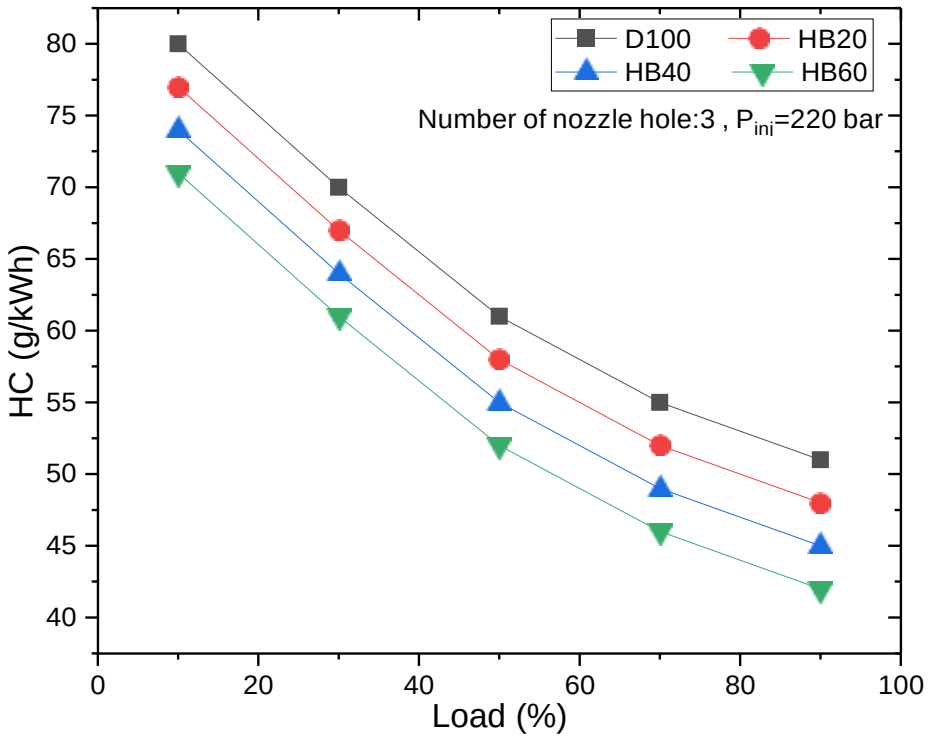


Figure 06- The HC as function of load

As far as the effect of adding hydrogen to biogas on the release of HC has been demonstrated, it has been shown that over the entire engine load range, the levels of HC released by the engine are often significantly lower than those produced by pure diesel fuel. It is obvious from Fig.6 that, increasing the energy share rate (ESR) of H_2 in the biogas blended reducing the HC emission. For BH20, the HC decreased by 8.3, 9.4, 10.5, 13.5, 12.4 % for all the load, compared to the D100. The reasons for what was previously mentioned are that increasing the percentage of hydrogen leads to a clear reduction in the percentage of replaced carbon atoms, and thus the blends of H_2/biogas with low concentration of CO_2. In addition, the H_2 improves the combustion reaction process on account of higher flame speed which increases corresponding to the H_2 presence leading to the increasing combustion temperature and HC emissions reduction.

CO_2 emissions

Efficiencies of combustion reactions are indicated by the levels of CO_2 emissions. In the case of a combustion engine that runs under a stoichiometric fuel-air mixture condition, the system products can reach their maximum level. Fig.7 shows the level of CO_2 emissions as function of different fuel blends along with various operation loads.it is clear, increasing operation engine load leads to increase emissions of CO_2, the reason of this relevant to fact the mixture promoting with fuel enrichment of the mixture. While the fuel-air mixture trend to the stoichiometric condition, the concentration of the CO_2 emissions fetch the maximum levels. Increasing operation load results in an improved of combustion reaction process, a drastic temperature increase and a higher amount of O_2 available to initiate the oxidation reaction. This results in a significant amount of CO to convert to CO_2 produced.

For the CI engine DF mode working with baseline diesel fuel (D100) at all load, the concentration of the CO_2 emissions higher than H_2/biogas mixture. It can be concluded from what was mentioned that, replacing diesel with hydrogen reducing the carbon content of the mixture, which makes the carbon/hydrogen ratio decrease to the lowest levels. leads to a significant decrease in the concentration of pollutants associated with the carbon atoms. Fig.7 shows that, the concentration of CO_2 emissions or the BH20, BH40, BH60 decreasing by 9.2 ,12.4, 14.3% lower than D100 for the different operation load. As mentioned previously, the presence of H_2 in the mixture contributes to boosting the combustion reaction in term of increasing the flame speed propagation. In other hand, H_2 has no carbon atoms, so that, increasing H_2 concentration in cylinder decline carbona content leads to reduction of the CO_2 emissions formation. the conclusion associated with presence of H_2 match with literature.

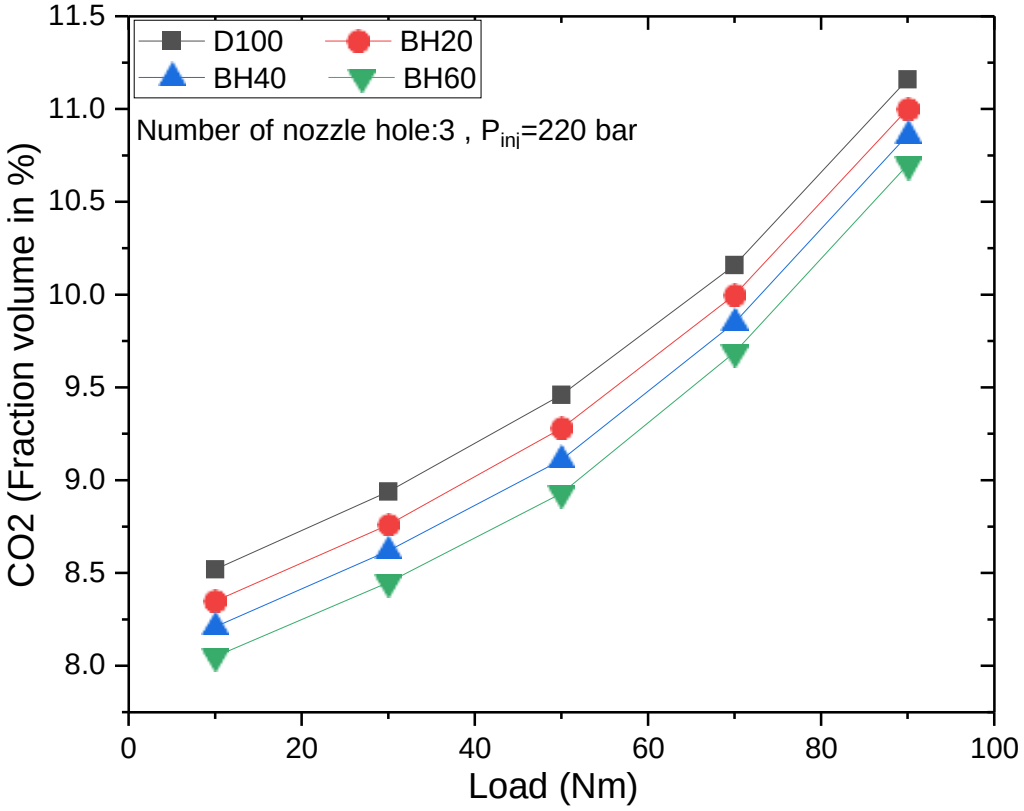


Figure 07- The CO_2 as function with load

NO_x emissions

The NO_x emissions from combustion are among the most significant environmental pollutants. There is a chemical union that occurs between NO and a very small amount of NO_2, resulting in the formation of NO_x. The amount of O_2, in the combustion chamber plays an important role in controlling the formation of nitrogen oxides emitted from the combustion ignition (CI) engines. NO_x emissions are also determined, the residence time necessary for the oxidation process to be completed, as well as the peak temperature at which the combustion process completes. The variations of the NO_x emissions for baseline diesel fuel and various H_2/biogas blended for CI engines DF mode at different load are shown in Fig.8. It is clear, that there are identical upward trends for NO_x emissions of both the diesel and H_2/biogas blended when the operation load increasing. When the operation load increased from 30 to 90%, the NO_x emissions increased from 650 ppm to 1050 ppm. This is could be explained by, when the CI engines working under a high operation loads to formation a richer mixture in which contributes to increase the charge intensity of the turbulency result in enhancing the combustion reaction process and temperature and pressure reach to peak value. Besides, as operation load increase the injection pilot diesel and gaseous fuel corresponding increasing driving to increasing the injection pressure which leading to enhancing the fuel-air atomization. Thus, the zone under the injection pole, has a promotion of charge mixture characteristics consequent to

elevate the generation rate of NO_x emissions obtained[39]. From the Fig.8, regarding H_2 addition to the blends of biogas, NO_x was (600, 648, 687, 745 ppm) for the D100, BH20, BH40, BH60, respectively, at 10% test operation load. From the figure, presence of H_2 leads to higher NO_x emissions compared to the baseline diesel fuel for all the test operation loads (TOL).

The formation of NO_x emissions primarily has been governed by various factors, CO_2 is one most important of them and is a major of artificial biogas consists [40]. The authors of the study, De Serio et al.,[41] stated that even a small amount of CO_2 present in the biogas increases the concentrations of HC and slows down the combustion reaction rate. They also confirmed that the presence of CO_2 contributes effectively to the decrease in combustion temperature due to its thermal properties and chemical effects. Thus, the combustion reaction process breaks down resulting in a higher amount of unburnt fuel releasing. It has become urgent to add fuel that overcomes biogas' negative factors and improves its characteristics. hydrogen has no carbon atoms within its structure chemical formula, and replaced with CO_2 drop carbon content in the mixture by the increasing the H/C ratio. In addition, hydrogen has a higher flame speed compared to CO_2 leads to improve combustion quality and increasing the temperature of the mixture inside the engine cylinder result in elevating the formation of NO_x emissions.

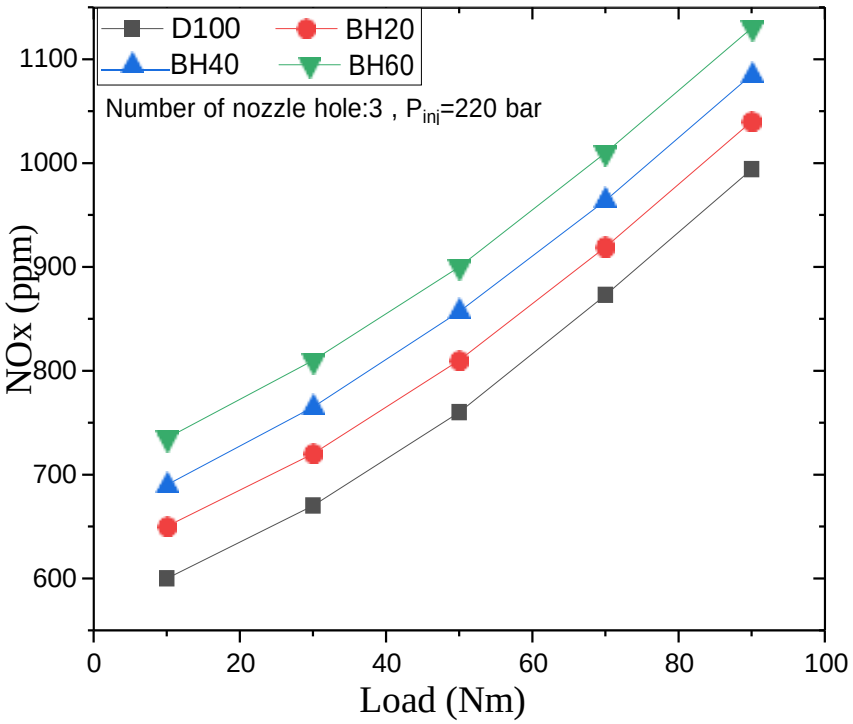


Figure 08- The NO_x as function of load

Conclusion

1. From the results extracted from the current study, it was obvious that the peak of in-cylinder pressure for the D100, BH20, BH40, and BH60 fuel blends was calculated at 66.4, 70.2, 74.3, and 78.5 bar, respectively.

2. The first peak of HRR at 10^{o} CA ATDC is increased by 5.4, 7.3, and 8.4 %, for BH20, BH40, and BH60 compared to pure diesel fuel (D100). while, the second peak of HRR at 30^{o} CA ATDC increased by 7.8, 8.8,9.8 % compared to D100.

3. the BSFC for the CDE mode lower than DF mode with BH20 by 18 , 24, 30% for operation engine load 10%, 30 , 50 , 70 , 90% , respectively.

4. When the energy share rate (ESR) of hydrogen increased in the blends of biogas the BSFC decreased at low load, in this manner, the BSFC was (625: BH20), (616: BH40), (600 g/kWh: BH60) at 10% of fuel operation load. This is relative to the H_2 ability to improve combustion process reaction due to hydrogen wide the limit of lean flammability. Since the H_2 has the capability of widening the lean flammability limit, it can improve the combustion reaction as a result

5. The emitted HC is generated clearly and significantly when the fuel is in the gaseous phase, and its rates change between (78.1) and (74.8g/Kwh) for BH20 and BH60.

6. The increasing of the energy share rate (ESR) of H_2 in the biogas blended reducing the HC emission. For BH20, the HC decreased by 8.3, 9.4, 10.5, 13.5, 12.4 % for all the load, compared to the D100. The reasons for what was previously mentioned are that increasing the percentage of hydrogen leads to a clear reduction in the percentage of replaced carbon atoms, and thus the blends of H2/biogas with low concentration of CO2.

7. The concentration of CO_2 emissions or the BH20, BH40 , BH60 decreasing by 9.2 ,12.4, 14.3% lower than D100 for the different operation load.

8. Regarding H_2 addition to the blends of biogas, NO_x was (600, 648, 687, 745 ppm) for the D100, BH20, BH40, BH60, respectively, at 10% test operation load.

9. It is clear, that there are identical upward trends for NO_x emissions of both the diesel and H_2/biogas blended when the operation load increases. When the operation load increased from 30 to 90%, the NO_x emissions increased from 650 ppm to 1050 ppm.

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Detection of Particle Pollution in Selected Areas of Baghdad

Nahla Shadeed Ajeel [1]

Abstract

The aim of this research was conducted to determine the levels of particulate matters PM in ambient air of selected dense areas of Baghdad city. Air samples were collected from two studied sites, located in the University of Baghdad campus, air samples were collected from sampling sites by portable device. Sampling points were selected by taking into account the industrial sources of pollutants, the strength of traffic, prevalent winds direction and density of population. In addition to measuring particulates matters PM with different diameters (micrometer), including PM2.5 $\mu g/m^3$ and PM10 $\mu g/m^3$, larger suspended particles matters TSP $\mu g/m^3$ also measured. Measurements of PM2.5, PM10, and SPM are presented for the months November and December, 2023. Concentrations of PM2.5 in November ranged from 55 to 58 $\mu g/m^3$ in site one, while in site two ranged between 55 to 57 $\mu g/m^3$, PM10 concentrations ranged from 81 to 83 in site one and 73 to 75 $\mu g/m^3$ in site two. As well as the concentration of TSP ranged from 8466 to 8559 $\mu g/m^3$ in site one 7389 to 7513 $\mu g/m^3$ in site two. While In December concentration of PM2.5, PM10, and TSP more than that recordrd in November and this due to meteorological factors. The results of this research showed that the lowest level recorded in November in site two; while the largest levels recorded in December in site one. The averages of PM2.5, PM10, and SPM in selected sampling areas were sometimes within and sometimes exceeded levels from the WHO suggested standards for ambient air. This may be as a result of frequent occurrence of heavy traffic, the existence of relevant industries near the city. Therefore, effective strategies such as traffic management, industrial movement, can be effective reducing PM2.5, PM10 and SPM concentrations.

Key Words

Air Pollution; Particulate Matter; Meteorological Parameters; Spm.

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Introduction

Air pollution has become a major environmental and health concern worldwide, particulate matter PM is a central concern for public health (1). According to WHO, air pollution refers to contamination/or changes in the natural environment by physical, chemical or biological agent (pollutants), which may be contributed by natural or anthropogenic sources (2). Epidemiological and toxicological studies have linked inhalation of particulate matter PM to a broad spectrum of acute and chronic health problems, causing different diseases like respiratory and cardiovascular diseases, dermal cancer and premature death (3), also PM can impact the climate, interfering in the Earth's radiation balance by scattering and absorbing solar radiation, acidifying oceans and lakes, and contributing to acid rain (4). Regarding the latter effect, PM can cause deterioration and blackening of materials, with associated economic costs (5). PM can be emitted from vary sources, natural and artificial sources, natural sources include volcanic eruptions and mineral dust, while anthropogenic sources are non-exhaust and exhaust vehicle emissions, agricultural activities, biomass burning and fossil fuel combustion, mining activities (6). The aim of this research was conducted to determine the levels of particulate matters PM in ambient air of selected dense areas of Baghdad city.

Materials and Methods

Sampling Site

Air samples were collected from studied areas by use portable device Figures (1) and (2) Which placed on a stand at about 1.5 – 2m high above the ground to avoid contamination and get accurate readings (7).

Figure 01- Portable particle mass counter device

Figure 02- Map show studied points (Google earth, 2024)

Study Relationship of PM with Meteorological Parameters during Sampling Periods

Throughout the sampling periods, the mean values of relative humidity, air temperature, and wind speed measured. Wind speed and direction have been termed the fundamental parameters in the movement of air pollutants and all these are interred linked to temperature (8). That mean, the greater the wind speed, the greater the turbulence and the more rapid and complete the dispersion of contaminants in the air (9). Therefore, mean value of relative humidity RH%, temperature, wind speed variables measured in ambient air during study periods of examined sites.

Results and discussions

Air quality variables

Average meteorological parameters values such as relative humidity RH%, temperature, wind speed and particulate matter concentrations PM like PM2.5, PM10, and TSP measured during study period in sampling sites are given in table (1)

Table 01- Average of meteorological parameters and particulate matter concentrations in sampling sites

Variable	Site 1		Site 2	
	Nov.	Dec.	Nov.	Dec.
RH %	57	43	57	43
Temp. C°	25	24	25	24
Wind spee. Km/h	33	8	33	8
PM 2.5 µg/m³	57	108	56	103
PM 10 µg/m³	82	160	74	145
TSP µg/m³	8513	15619	7451	15124

Weather factors

Quality of air can affect by prevailing weather conditions, improve or weaken air quality. Strong winds can rapidly transport pollutants hundreds of kilometers, while during weak wind pollutants can collect near and around the source of the release. Also the rain can either clean or pollute the environment depending on harmful substances in the air.

Relative Humidity RH%

According to the values that obtained from this study, relative humidity RH% in November recorded 57%, while in December 43%. That means the maximum value found through November and minimum value during December as shown in figure (3). Relative Humidity RH% one of weather factors that has an importance effects by decreasing scale of pollutants in the air, as water vapor in the air works to rid it of a large percent of which suspended in it (10). High humidity enhance the negative effects of harmful air pollutants such as, gases and smog due to it helps to accumulate pollutants and condense them in the air and this effects on public health (11).

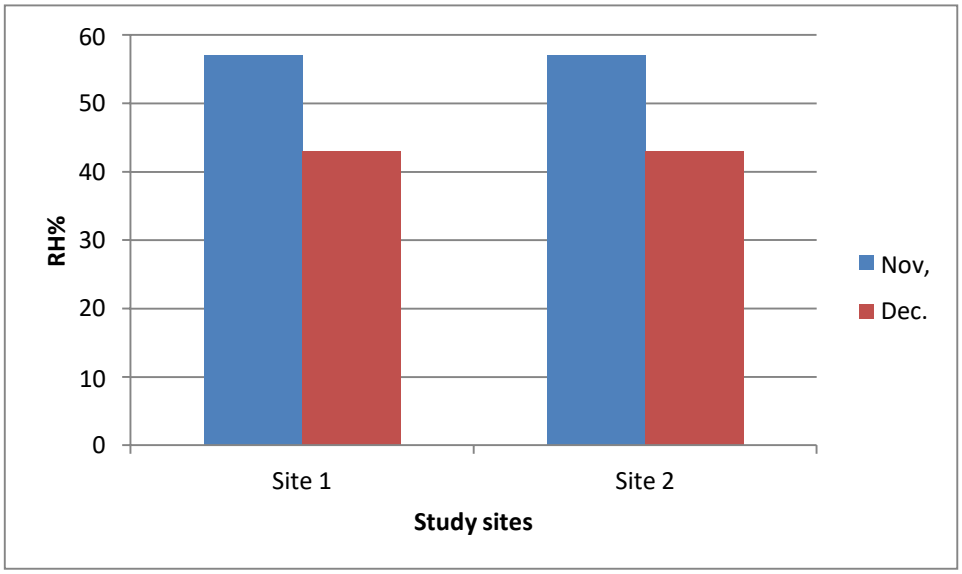

Figure 03- Mean values of RH% during study periods

Temperature

In case of temperature, the present data had shown that mean values of temperature during November is 25 Co, while during December recorded 24 Co as shown in figure (4). In general, minimum degree recorded in December 22Co, while the maximum value found in November was 24 Co, these varies in temperature contribute in spread and dense of pollutant in the air. Temperature plays an important role in the formation and disappearance of air pollution, it is one of atmospheric conditions that strongly affected on air pollution, such as PM tend to increase during winter months, temperature inversions, occur when air near the surface of earth is colder than air above it. Cold air is heavier than warm air, therefore temperature inversions make vertical mixing limited and locked up pollutants near surface of earth (12).

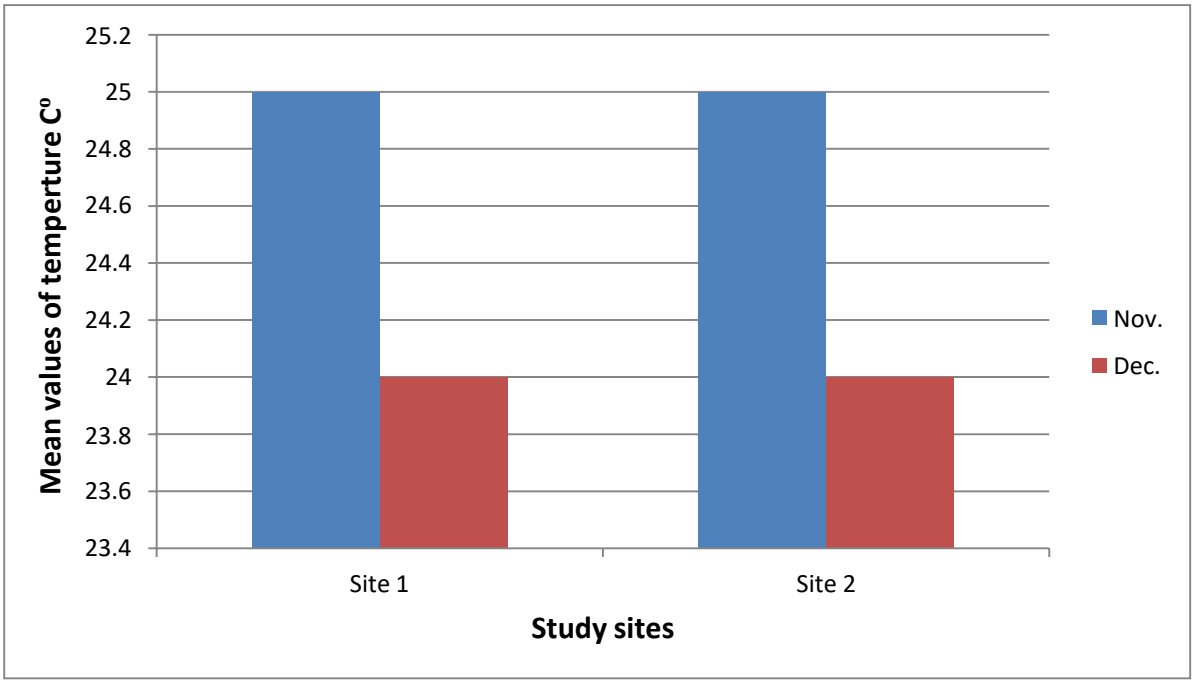

Figure 04- Mean values of temperature C° during study periods

4.2.3 Wind speed

During November, highest value of wind speed was recorded 33 km/h in both study sites, while the lowest value was 8 km/h. as shown in figure (5). According to the results, there are significant differences between means of wind speed during study months. These different in wind speed effects in collection of pollutants in air near surface of earth, when wind speed rise that led to reduce in concentrations of pollutants including PM by dispersion and transport of pollutants from source and spread on large space that led to reduced it and decrease the dangerous of pollution (13,14). While when wind speed is low that makes pollutants such as PM and other air pollutants to accumulate in the air near the surface of earth (14).

In general, increase in wind speed accompanied with a decrease in the levels of pollutants and vice versa. The wind speed has an important role in the spread of pollutants where the wind transported contaminants from emitted sources toward the prevailing wind. (15,16). From above results about the mean values of relative humidity, air temperature, and wind speed it is clear that there is no vary significantly during study period.

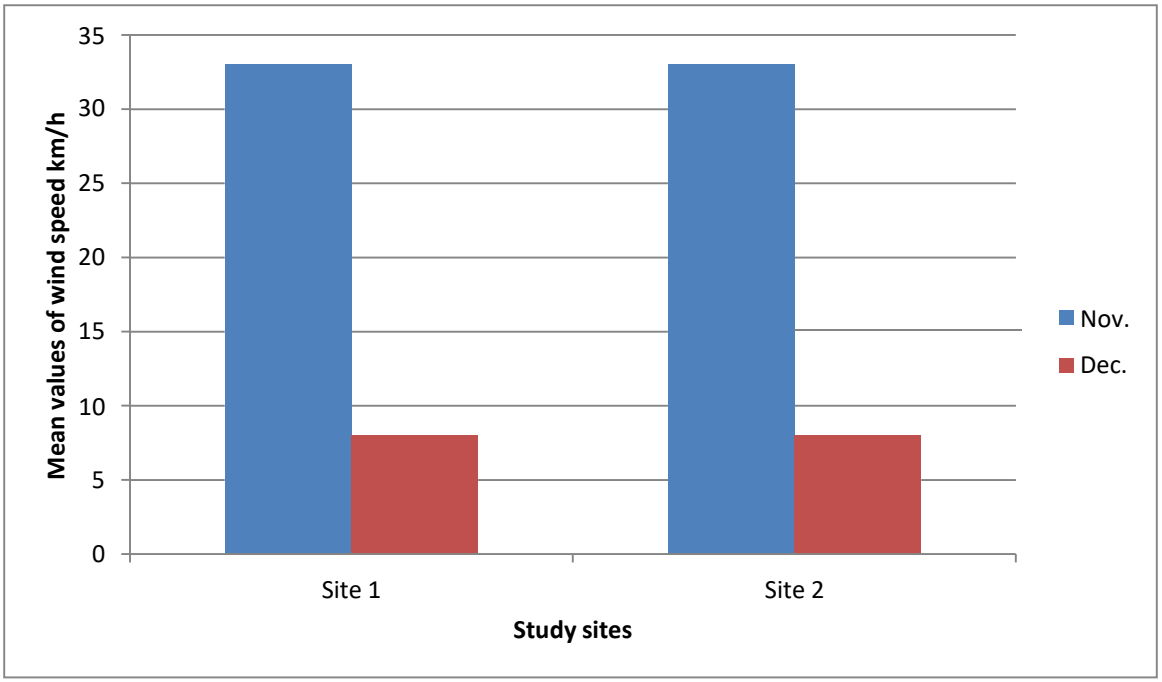

Figure 05- Mean values of wind speed during study periods

Concentrations of PM2.5

Concentrations of PM2.5 in November ranged from 55 to 58 µg /m^3 in site one, while in the other site ranged between 55 to 57 µg /m^3 as shown in figure (6).

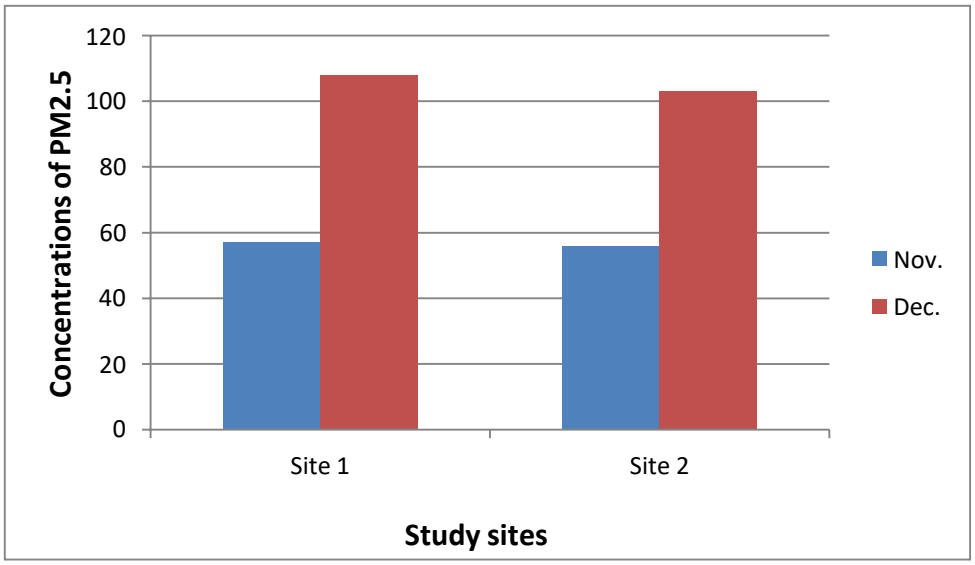

Figure 06- Mean concentrations of PM2.5 µg/m^3 during study periods

Concentrations of PM10

PM10 concentrations ranged from 81 μg/m^3 to 83 μg/m^3 in site one and 73 μg/m^3 to 75 μg/m^3 in site two as shown in figure (7). From results was observed that most of the values exceeded WHO limits. WHO PM2.5 -25μg/m^3 and PM10 50μg/m^3

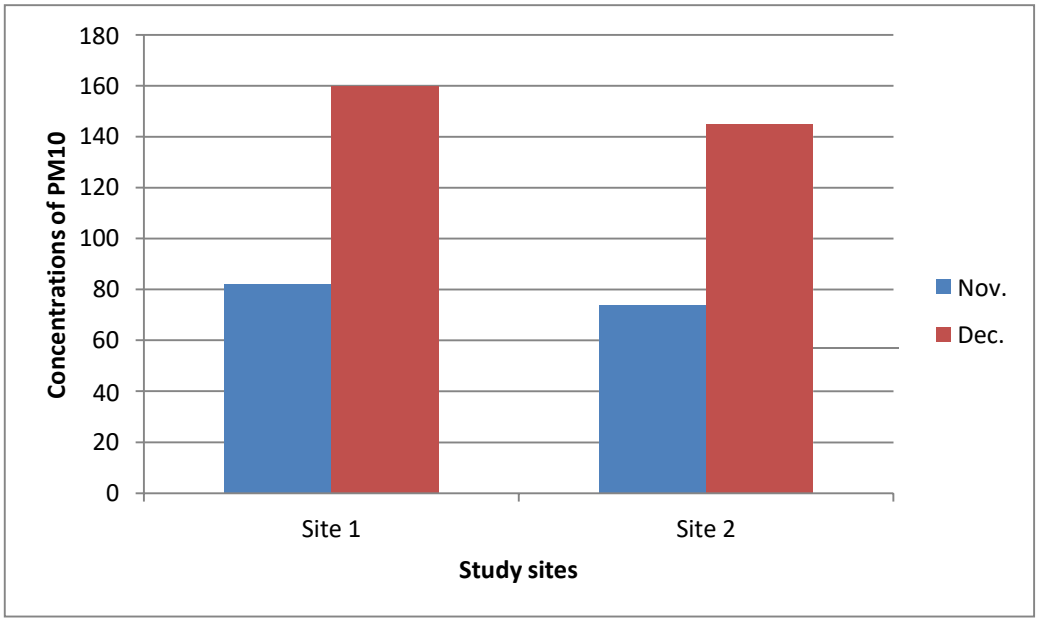

Figure 07- Mean concentrations of PM10 μg/m^3 during study periods

Concentration of TSP

Concentration of TSP ranged from 8466 to 8559 μg/m^3 in site one 7389 to 7513 μg /m^3 in site two. In December concentration of PM2.5, PM10, and TSP more than that recorded in November and this due to meteorological factors that effects on accumulation and concentration of PM as shown in figure (8).

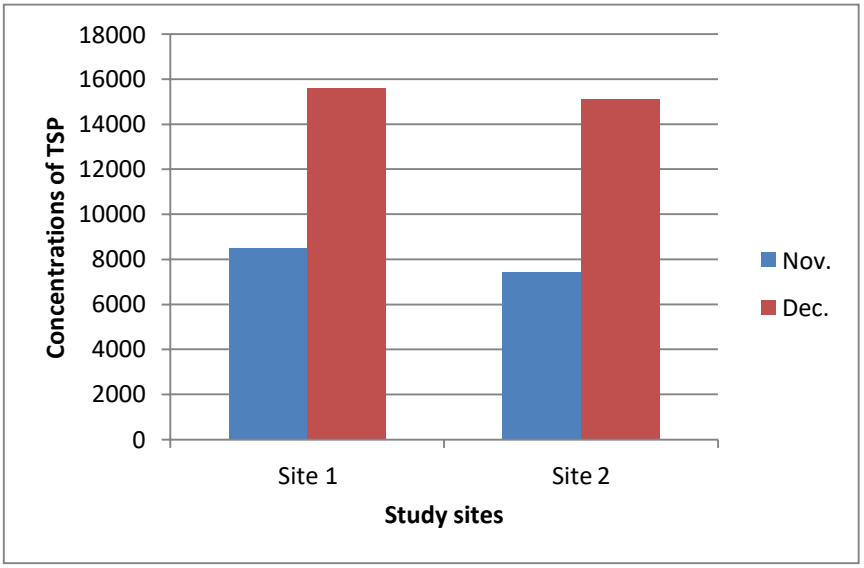

Figure 08- Mean concentrations of TSP μg/m^3 during study periods

The mean PM1 and PM2.5 concentrations measured in present study exceeded the limits of the World Health Organization WHO (25 μg/m^3).The measured PM2.5 and levels are considered in the unhealthy to very unhealthy (17). The mean TSP concentrations measured in present study exceeded the World Health Organization WHO (150 μg/m^3 /hour).

Environmental protection agency EPA in 1997 has evaluated thousands of new studies on PM and, in September 2006; EPA revised the standards of PM by lowering the level of the 24-hour $PM_{2.5}$ standard to 35 μg/m^3 and retaining the level of the annual $PM_{2.5}$ standard at 15 μg/m^3 (18,19). The Agency retained the 24-hour PM_{10} standard of 150 μg/m^3

Conclusion

Based on the results of this study, PM concentration are high and usually exceeded the recommended standards limits of WHO and EPA, therefore must be reducing and eliminating sources of pollution, as well as move factories and car garages away from residential areas to decease dangerous on public health. The issue requires more studies to cover more areas and then estimate the extent of the impact of the concentrations of these pollutants on the environment and humans alike.

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Synthesis of Anthracene and Benzene Derivatives from Glycerol and Study of the Effectiveness of Biological

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Abstract

This research includes the synthesis of benzene and poly hydroxyl anthracene derivatives from the reaction of glycerol with ethanoic acid using the catalyst poly allyl sulfonic acid benzene and produces glycerol tri acetate and this latter compound is reacted with Benzene, anthracene and their derivatives in the presence of copper silicate (catalyst)and then analyze these compounds using the catalyst agent hydrochloric acid and these compounds were diagnosed using infrared, element analysis and thin-layer chromatography technology of these compounds and measure the degree of Boiling point and physical properties.

Key Words

Aromatic compounds; Alcohols; Glycerin; Poly Hydroxyl Compounds.

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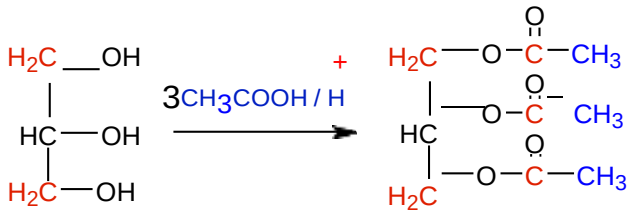
Introduction

The compound glycerol was discovered by scientist Scheele in 1979(1) and obtained as a result of lipolysis by base and hydroxylation which is widely used in oil plants (2) as well as in detergent production (3,4) Which is the result of the process of alcohol fermentation of sugar (5) and is of great importance due to its polar group (OH) which combine with the polar group containing oxygen and nitrogen and others and also produces polymer chains (6) . Casserole is a non-toxic substance and for this reason it is used in cosmetics and medicine(7,8,9), lubricating oil (10,11), antifreeze or antifreeze (12) and is also used in the manufacture of vegetable perfumes in addition to industries Textile (13,14,15).

In the field of use in the medical field is used in the production of ointments for the treatment of burns (16) and in the industrial field is used in the manufacture of printer colors and other dyes (17,18) and has wide applications of which use it as a solvent for materials Non-lumbar (19) and to forget its importance in the synthesis of polymers such as polymer (Polly such as terphthalene polly methyline triphthalates) used in the plastic industries (20). Glycerol interacts with aromatic compounds in many pharmacological compounds as well as cosmetics and others (21). To prepare the esters are from the interaction of alcohols with acid and these esters are called the ester of Fesher, which has many applications, including natural and industrial applications (22) and as shown in the following equation:

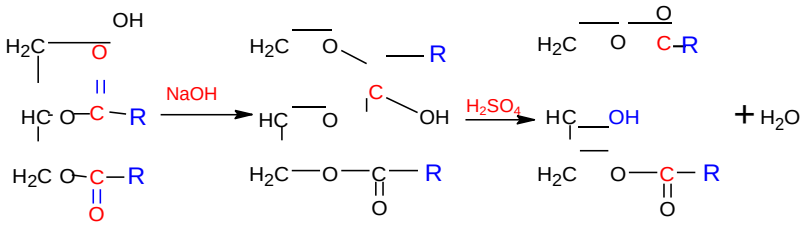


Glycerol **Glycerol Triacetate**

The glycerol esters are multiple esters called aromatic oils that are created from high vegetable oils and oligosaccharides (23,24) . Many detergents are esters consisting of multiple alcohols (25) They are considered good solvents such as almond oil, which are involved in the production of pastes. It is not secret that we use them as polar compounds in interactions that include the protection of hydroxyl groups in the synthesis of aromatic compounds as well as used as ion exchangers in the separation process (26) ,that have the potential for slow diffusion.

They are small granules (27) on which the negative charge due to the SO3 group appears and the positive charge is also due to H+ (28) resins that do not lose their activity and this

enables us to use them several times (29). It regains its activity using HCL (30) through the interaction of glycerol with lipids and with the presence of acid as in the equation below.



Experimental

Preparations of 2 Aryl glycerol:

Put 1g of poly allyl benzene sulphonic acid is a catalyst and we put it in a beaker attached to a condenser, adding 30g of glycerol, 100 g of ethanoic acid, 100ml of benzene and leaving it for 4 hours Continuous stirring with magnetic stir with 80 degree centigrade. After the reaction is over, the excess solvent is drawn by the rotary device and the output is diluted with anhydrous copper sulfate, left to dry and left, then we take 1g of copper silicate in a three-hole percussion and contact the capacitor and heated For 15 minutes, add the compound 30 g glycerol tri- ethanoate with 2 moles of benzene derivatives, stir the mixture continuously and leave it for an hour at a temperature of 80 degrees Celsius. After we cool the mixture with the water droplet to it, we leave it at room temperature, where we observe the separation of two layers from each other . And we remove the solvent using the rotary advice, and then we dry it with anhydrous copper sulfate, and we filter and use the same method to prepare the rest of the compounds.

Added 4 g of Last ester with 65 ml , 0.1 M of hydrochloric acid in a beaker connected to a condenser and we reflux for an hour and a half with a water bath boiling 100 degrees Celsius, we get an oil layer and we do the separation process using ether, we use magnesium sulfate anhydrous .We filter the solution and get rid of the excess solvent through the rotary advice. An oily substance for prepared compounds and the rate of velocity of the runoff in the thin-layer chromatographyA1-A10 have their physical properties in a table of (1).

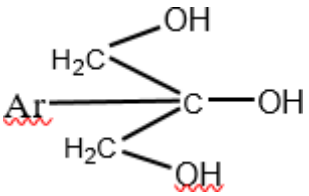
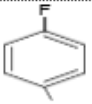
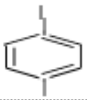
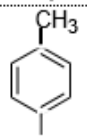
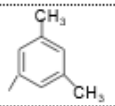
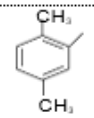
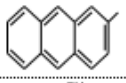
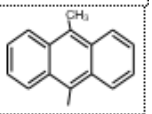
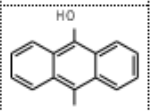


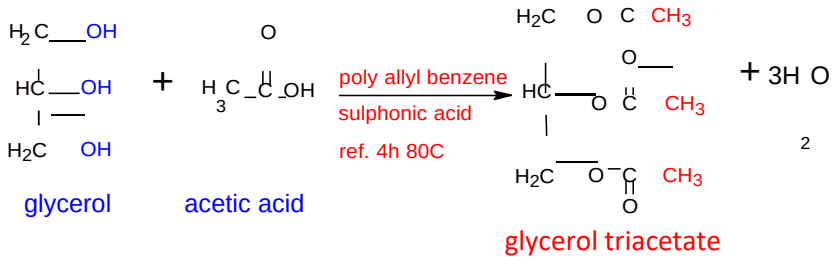
Table 01-

No. of compounds	Molecula rFormula	Color of compounded	Molecula rWeight	Boiling point	percentage	Rf	structure
A1	$C_9H_{12}O_3$	Yellow-Orange	168	259-257	84	0.85	
A2	$C_{12}H_{18}O_6$	Yellow-Orange	258	319-317	82	0.81	
A3	$C_9H_{11}O_3F$	Yellow-Orange	186	326-324	79	0.82	
A4	$C_9H_{11}O_3$ I	Yellow-Orange	294	295-290	82	0.79	
A5	$C_{10}H_{14}O_3$	Yellow-Orange	182	254-250	87	0.78	
A6	$C_{11}H_{16}O_3$	Yellow-Orange	196	286-284	68	0.81	
A7	$C_{11}H_{16}O_3$	Yellow-Orange	196	296-294	62	0.78	
A8	$C_{17}H_{17}O_3$	Yellow-Orange	269	352-351	61	0.86	
A9	$C_{17}H_{20}O_3$	Orange	272	354-352	65	0.87	
A10	$C_{17}H_{18}O_4$	Brown	286	349-345	68	0.93	

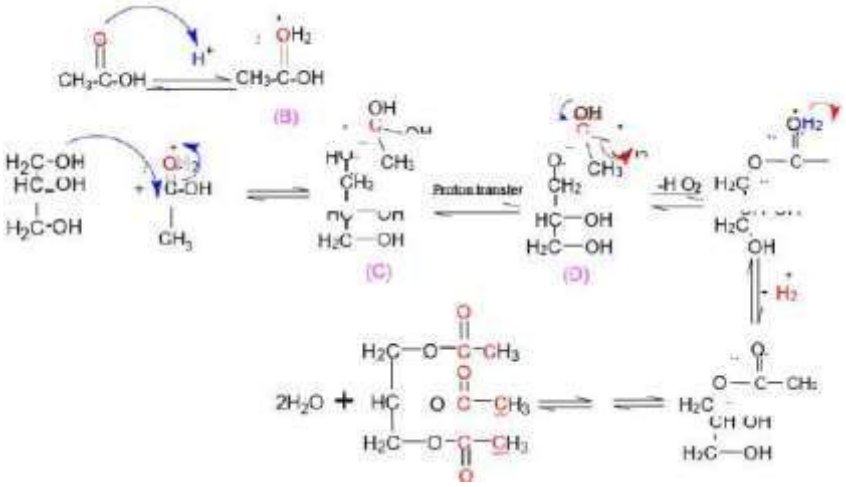
Results and discussion

The reaction was done using the glycerol compound and for two reasons, the first is poly hydroxyl alcohol and the second is the glycerol triacetate compound to protect hydroxyl chemicals according to the equation below:

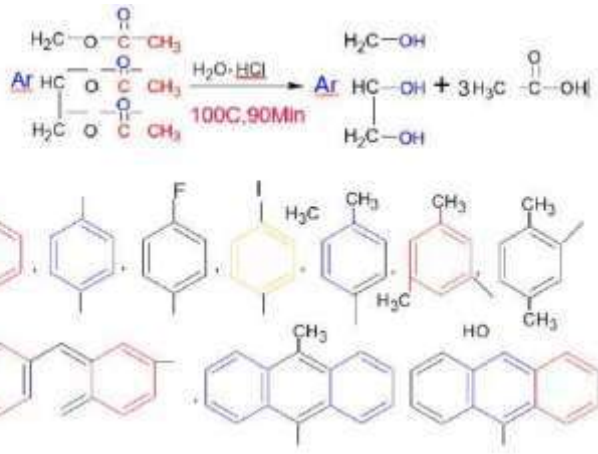
The mechanism of the esterification reaction and the formation of triacetateglycerol.



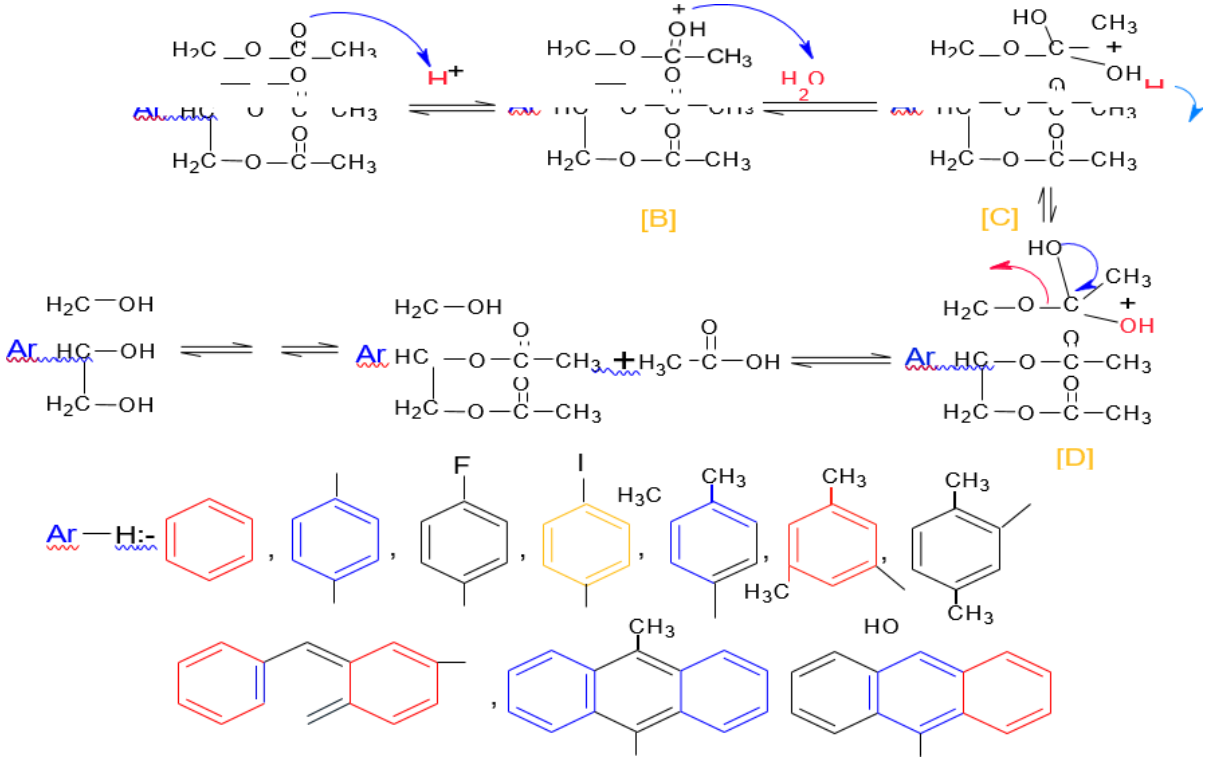
The reaction occurred using the catalyst poly-allyl sulfonic benzene acid for its economic cost, easy of separation, access to a high product.



These compounds were prepared from the Ester reaction found with diluted hydrochloric acid as in the equationbelow:

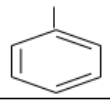
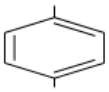
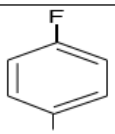
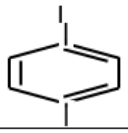
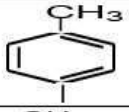
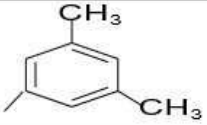
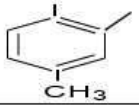
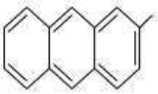
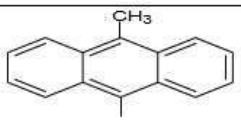
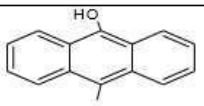


The reaction occurs when a proton is added to the carbonyl group, the oxonium ion, a fast electron looking at the electrons, attacks the hydroxyl group, turns into a cation, and then the proton moves from oxygen to oxygen and the cation is formed and occurs in this reaction is loss a water molecule in the following equation)[31](:



We use the thin layer chromatography technique and the use of methanol-chloroform solvent at a different rate the table(2)showed the rate of flow velocity per Compound and we took the infrared spectrum of the prepared compounds its appearance peak (OH)hydroxyl group at 3400-3550cm^{-1} and a clear package appeared at 3530 cm^{-1} in the compound A3 and an absorption pack at 3030 cm^{-1} returned to(C-H) (Aromatic) and the appearance of an absorption pack at (1478 and 1505cm^{-1}) [(22)], which is due to the double-bond in the benzene-anthracene compound as well as the appearance of an absorption pack at 1038cm^{-1} and 1270 cm^{-1} due to the presence of (C-O) alcoholic The table (3) shows the red-ray spectrum of the prepared compounds.

Table 03- IR (infrared spectrum of compounds)

Co. No.	Compound	V(O-H)	V(C-H) ar.	V(C-H) al.	V CH2bn	V(C-O) alc	V(C-H) OOP.	V(C=C) cm	V(others) cm-1
A1		3400	3030	2932asy. 2883sy.	1495 1462	1227 1035	699 726	1510 1495 1462	*V* C-H ip 1227,1035
A2		3550	3030	2928 2873	1475	1145 1062	812	1470	*V* C-H ip 1146,1062
A3		3530	3038	2987 2893	1460	1150 1120 1260	898	1598 1455	*V*C-F ar.1120 *V* C-F ar. 520 *V* C-H ip 1022,1165
A4		3467	3007	2873	1440 1420	1230 1145 1032	880	1610 1520 1400	*V* C-I ar.St. 1030 ,*V* C-I ar.500
A5		3452	3055	2940 2870	1380	1129 1270	785	1485	*V* C-H ip 1129, 959
A6		3498	3038 3090	2885	1472 1410	1248 1135 1021	678 885	1640 1420 1477	*V* C-H ip1022 *V*OH ,,V.CH3 1357
A7		3460	3028	2910 2838	1464	1238 1137 1032	776 878	1573 1468	*V* C-H ip1032 *V*OH ,, VCH3 1365
A8		3428	3009	2805 2853	1468	1169 1088	835 789 728	1505 1470	*V*C-H at α position1275 *V* C-H bend ip 959 1170,1085 *V*(C-C-C) bend.620
A9		3505	3080	2887	0	1209 1166	850 785	1509	*V* C-H ip *958,1168* *V* OH ,V.CH3 1380,*V* C-C-C 615,*V* C-H at β-position 1209
A10		3453	3090	2989	1478	1206 1164 1038	698 729	1598 1478	VO-H 1385, 1335,*V* C-H at β-position1210

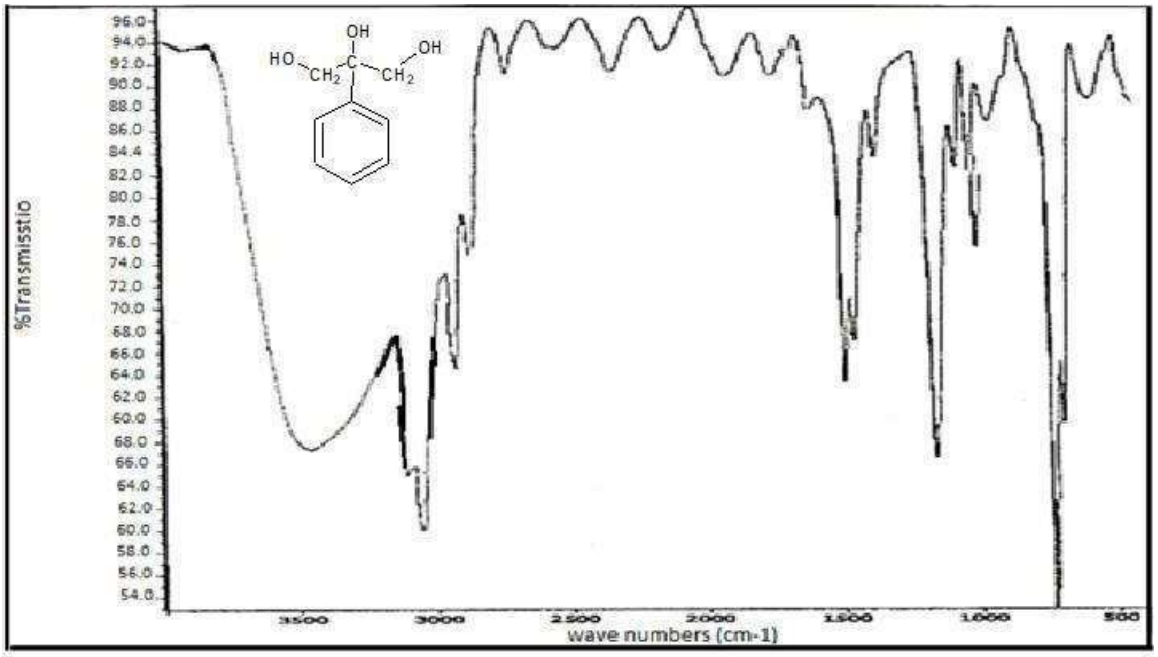


Figure 01- IR for compound A1

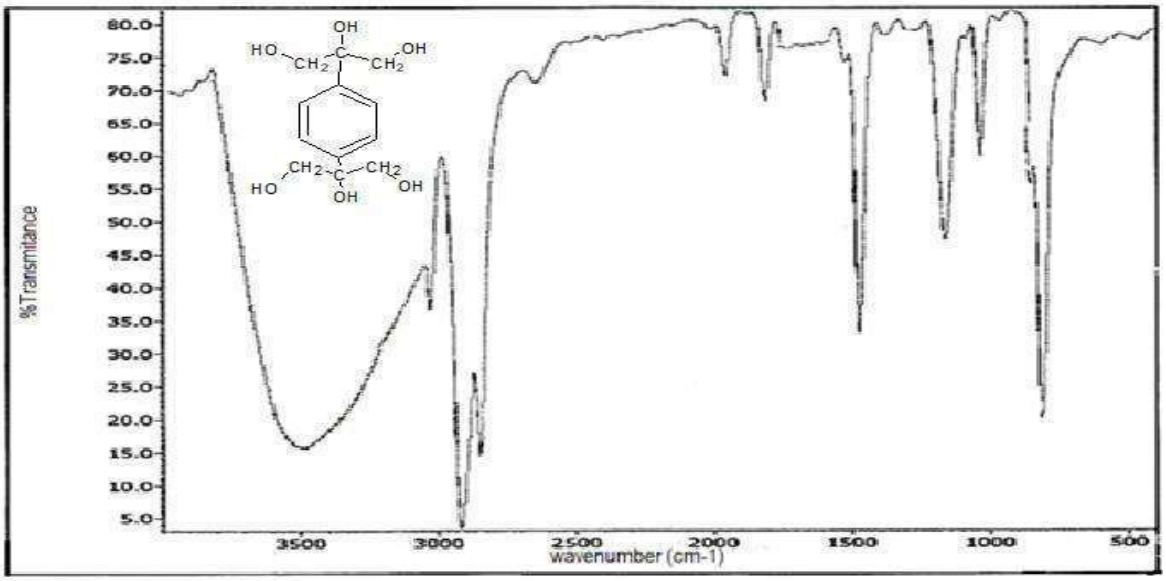


Figure 02- IR for compound A

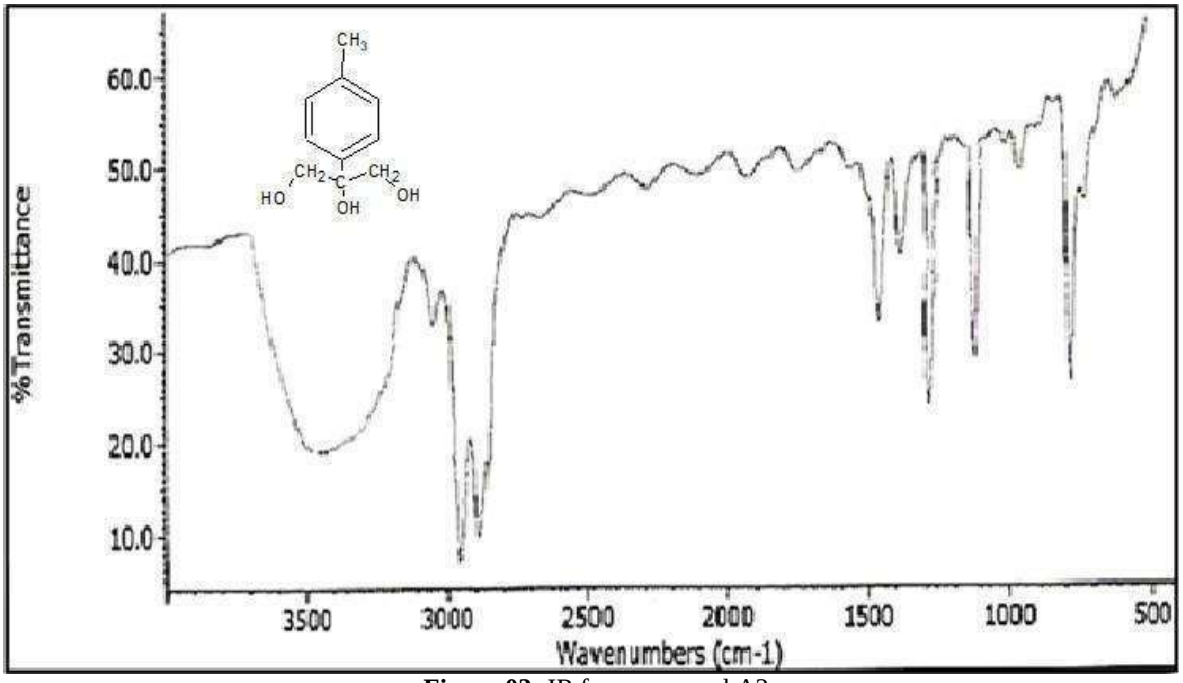


Figure 03- IR for compound A3

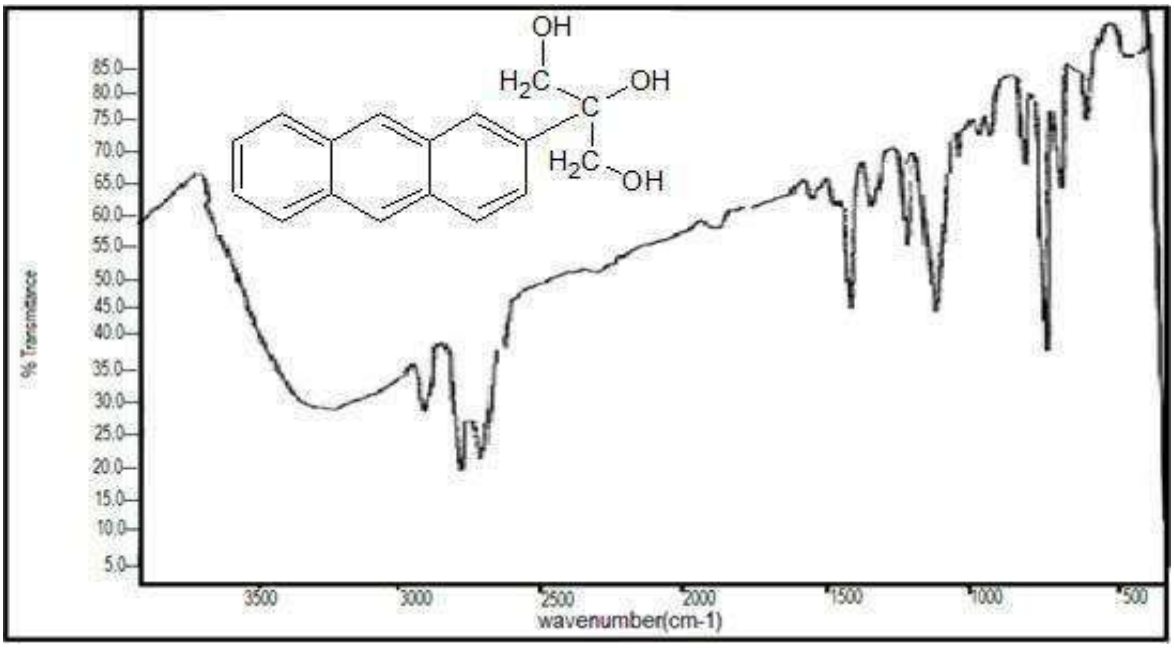


Figure 04- IR for compound A4

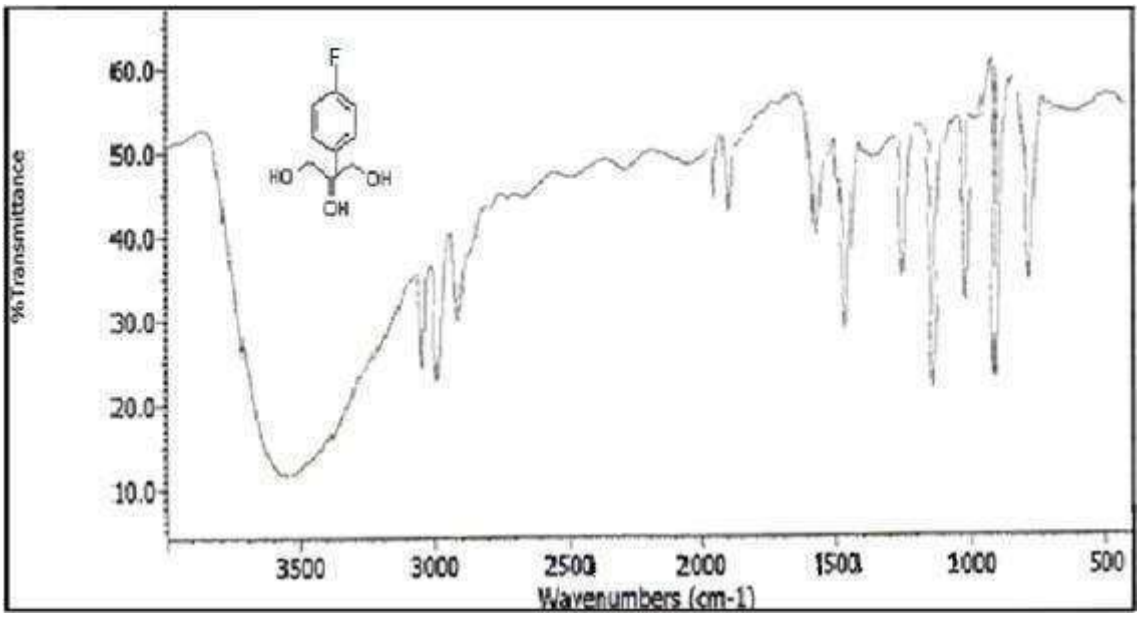


Figure 05- IR for compound A5

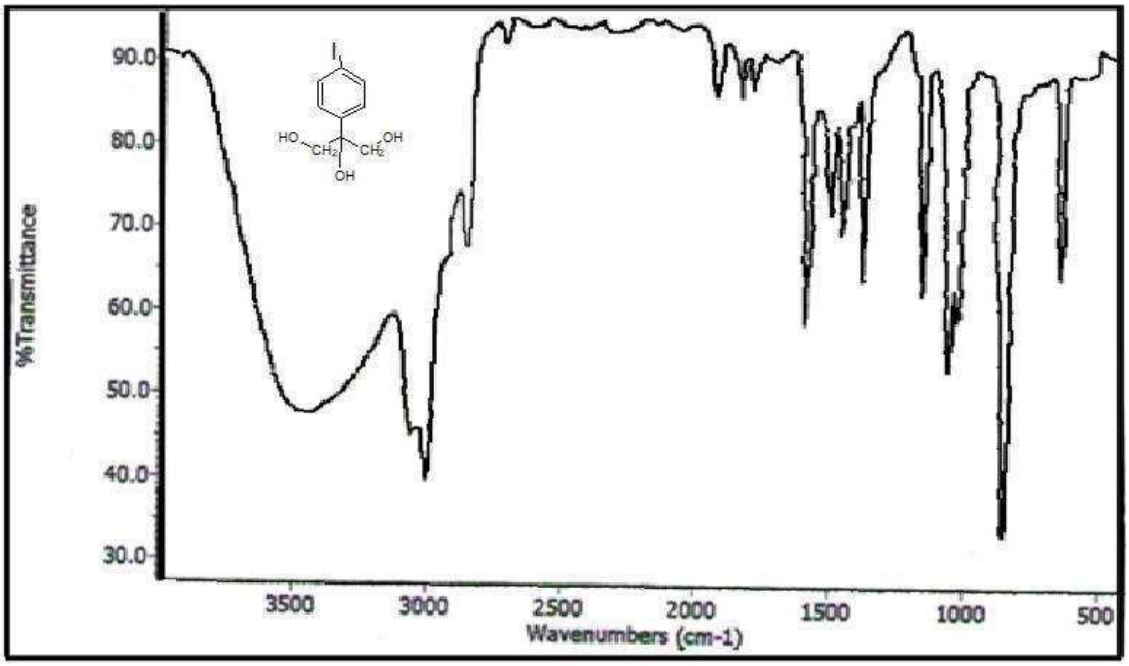


Figure 06- IR for compound A6

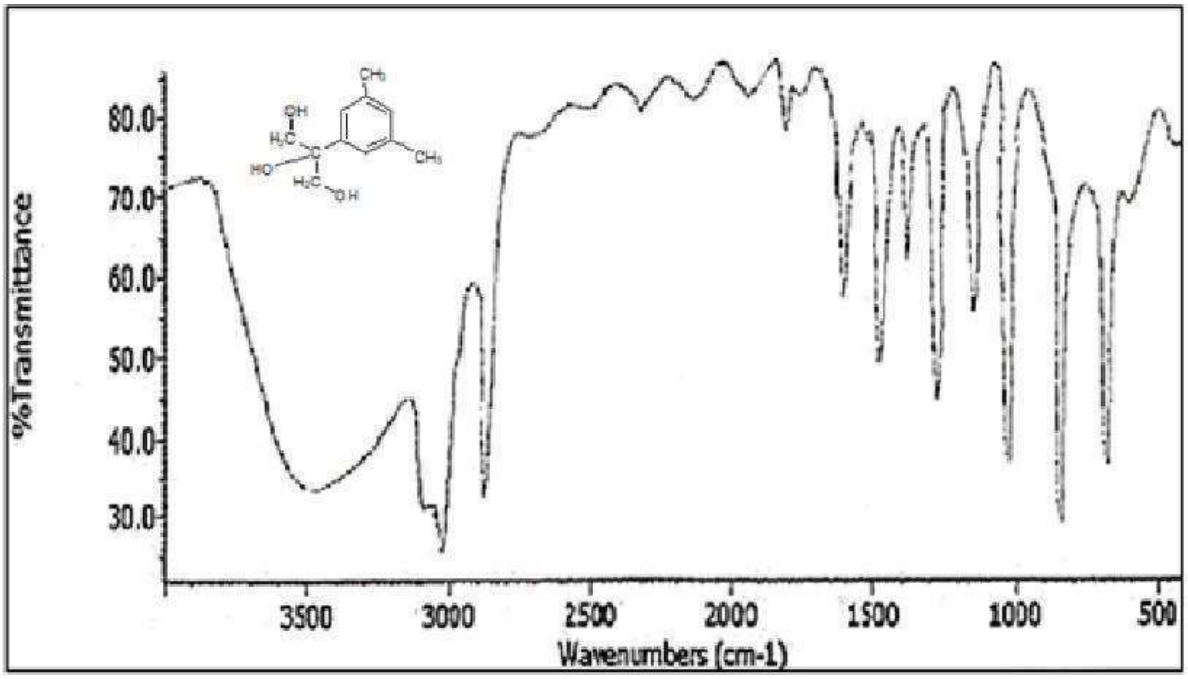


Figure 07- IR for compound A7

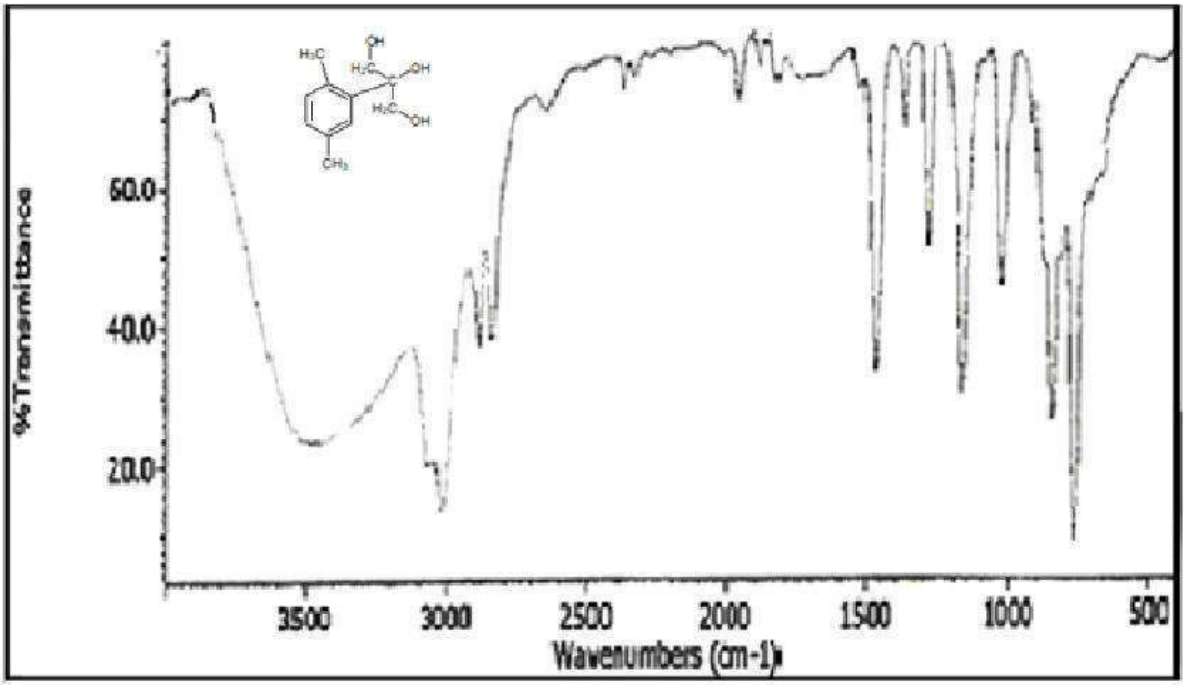


Figure 08- IR for compound A8

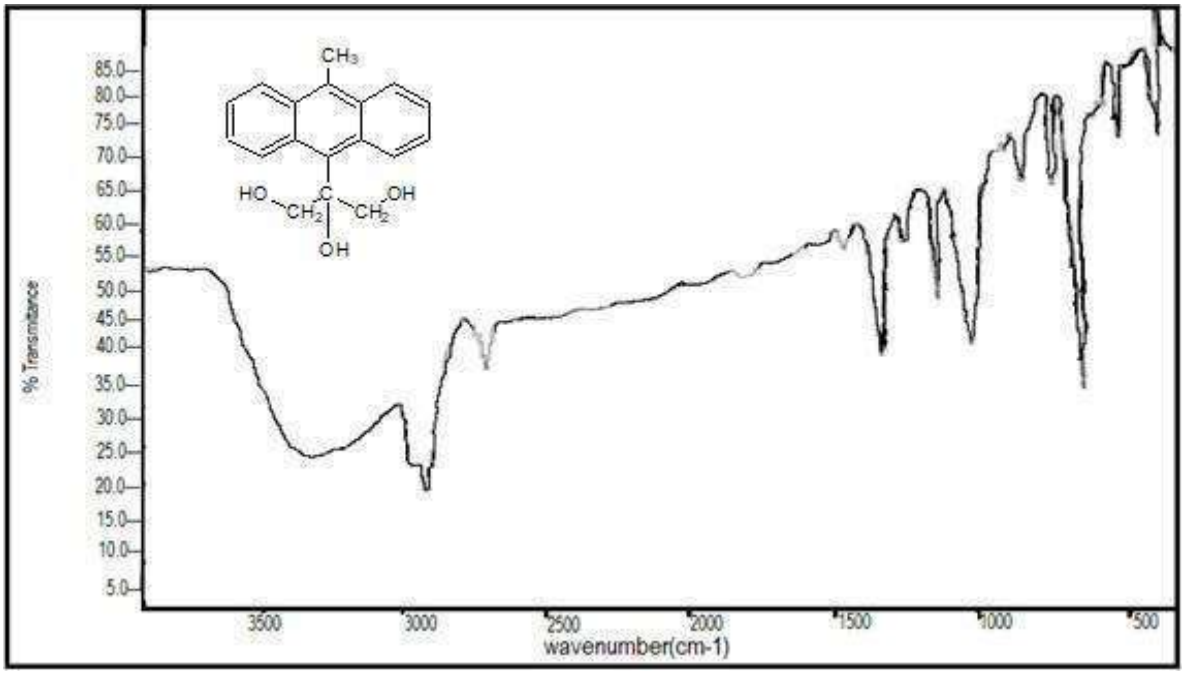


Figure 09- IR for compound A9

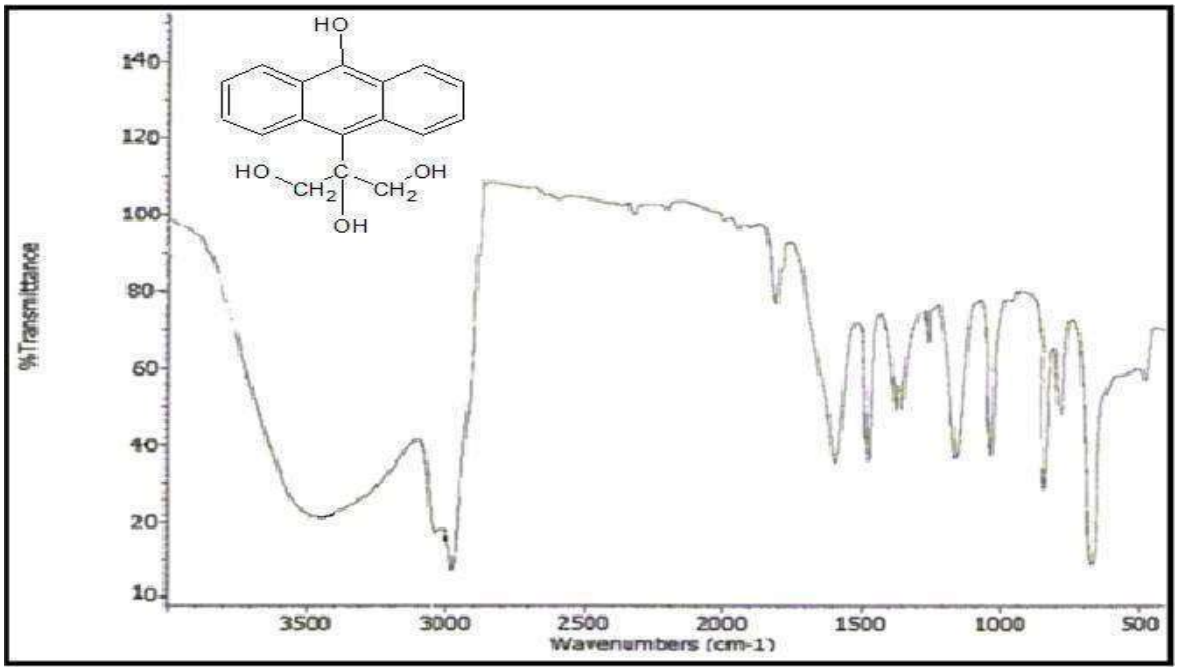


Figure 10- IR for compound A10

Table 04- C. H.N. of compounds.

Co. No.	C%		H%		N%	
	Calc.	Fou.	Calc.	Fou.	Calc.	Fou.
A1	64.28	64.25	7.14	7.08	**0**	**0**
A2	55.81	55.77	6.97	6.90	**0**	**0**
A3	58.06	55.25	5.91	5.40	**0**	**0**
A4	36.73	30.70	3.74	3.36	**0**	**0**
A5	65.95	65.90	7.69	7.65	**0**	**0**
A6	69.38	67.40	8.16	8.12	**0**	**0**
A7	69.38	67.40	8.16	8.12	**0**	**0**
A8	75.83	72.70	6.31	6.30	**0**	**0**
A9	75.00	72.32	7.35	7.12	**0**	**0**
A10	71.32	70.20	6.29	6.25	**0**	**0**

Conclusion

A high percentage of the product was obtained without any by-products. It was easy, cheap, and economically inexpensive. The ester we obtained was preserved in.Under dry conditions, away from moisture, so that the ester does not disintegrate into undesirable compounds. During the preparation of the compounds, the various hydroxyl groups that are involved in the synthesis of the glycerol compound were protected. The prepared compounds have biological activity.

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Comparison of Some Diagnostic Techniques for *Toxoplasma Gondii* Detection in Aborted Women in Babylon Province, Iraq

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Abstract

Toxoplasma gondii is an important apicomplexian parasites with worldwide distribution, its can infect all Mammalia included humans. This study aimed to detect Toxoplasma gondii by different diagnostic technique and determine the most accurate technique. One hundred blood samples were collected from aborted women of different age groups (<20- 40) years old in Babylon province. Different methods used to detect Toxoplasma gondii infection; Latex agglutination test, Combo Rapid test, ELISA and real time-PCR.The overall prevalence of Toxoplasma gondii infection in aborted women was 27% by Latex agglutination test, (35% IgG and 9% IgM) by Combo Rapid test, (42% IgG and 16% IgM) by ELISA and 48% by real time-PCR. Varying diagnostic techniques (sensitivity and specificity); real time-PCR showed that it's highly sensitive (92%) and specific (100%) technique in comparative with other techniques.

Conclusion: The most accurate and sensitive method of detecting toxoplasmosis in clinical specimens (blood) in both acute and chronic cases is real-time PCR, which uses the B1 gene and is suitable for routine screening. It provides a fast, quantitative method with no false-positive results, enabling targeted treatment.

Key Words

Abortion; Combo Rapid Test; Elisa; Latex Agglutination Test; Real Time-Pcr; Toxoplasma Gondii; Woman.

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Introduction

The obligate intracellular parasite protozoa *Toxoplasma gondii* is the cause of the zoonotic disease toxoplasmosis. It is likely contagious to humans and all warm-blooded animals, causing a range of clinical symptoms such as encephalitis, chorioretinitis, mental retardation, congenital infection-related blindness, and miscarriage in mammals[1,2]. Contaminated food and water with *Toxoplasma gondii* oocysts, or the direct contact with cats as well as ingesting tissue cysts in meat can cause infection to humans [3]

Toxoplasmosis also transmitted congenitally, in addition to foodborne transmission. As the parasite departs from its intermediate hosts that have warm blood, its life cycle is complete. The parasite only known to replicate sexually in Felidae, producing oocysts that are excreted in domestic cats' feces [4]

Toxoxplasma gondii can be detected using a variety of techniques, including indirect methods using serological techniques like the latex agglutination test (LAT), modified agglutination test (MAT), ELISA, and direct procedures such producing impression smears from infected organs and histological inspection [5,6,7]. Different molecular methods and quantitative Real Time PCR technique could be use different samples such as blood and tissue biopsy for detection of *T. gondii* infection [8,9]. The objective of this study to detect *Toxoplasma gondii* by different diagnostic technique and determine the most accurate technique.

Materials and methods

Ethical approval

This study was approved by College of Medicine/ Babylon University ethical committee (No. BMS/0231/016) to collect blood samples from aborted women in different hospitals of Babylon Province, Iraq. One hundred blood samples were collected from aborted women of different ages (<20 – 40 years) from four hospitals of Babylon province (Al-Hilla Teaching Hospital for Women and Children, Al-Hashimiya General Hospital, Al-Mahaweel General Hospital and Al-Qasim General Hospital(during the period from 1 September 2022 to 31 August 2023.

Five ml of blood were drawn carefully from vein and divided to: 1- Three ml of blood samples was collected with sterilized plain tube, left 30 min. at 25 °C to clot, centrifuged was done in 3000 rpm /5 minutes after that the sera were gotten, which were separated and pipetted

into another sterilized tube and stored at -20 °C till used. 2- Two ml placed in EDTA tube for plasma collected and stored in -20 °C for DNA extraction. Serological methods: Latex agglutination test (LAT); the kit was providing from Atlas Medical Company – USA and the test principle is depending on antibody - antigen reaction directly.

Toxo IgG/IgM Combo Rapid Test (CTK/ biotech USA); this test provides an initial test results to aid diagnosis of *Toxoplasma gondii.*

EnzymelinkedImmunosorbent Assay Toxo IgG ELISA kit ID. Screen ®Toxoplasmosis indirect- Multi species ID-VET (innovative Diagnostic Toxo- Msver 1013 GB, French) Toxo IgG ELISA kit (Human Gesellschaft Fur Biochemica und Diagnostica) mbh Max –Plank- Ring 21.65205 Wiesbaden Germany).

IgG/IgM (ELISA IgG/IgM) (ACON kit USA); Used to determine *Toxoplasma gondii* antibodies in serum samples. The serological tests were performed according to the manufacturer's instructions. Real time- PCR amplification; DNA extracted from aborted women blood samples by use gSYAN DNA mini kit extraction kit Geneaid. USA, that implemented depending on company instructions. Real time PCR was done for detection of *Toxoplasma gondii* by using the primers and specific TaqMan probe for *B1* gene. This technique was performed in relation to technique designated by [10].

Table 01- The primers and probe were used for amplification of DNA

Primers	Sequences		Product size
B1 primer	F	TCCCCTCTGCTGGCGAAAAGT	96 bp
	R	AGCGTTCGTGGTCAACTATCGATTG	
B1gene probe	FAM-TCTGTGCAACTTTGGTGTATTCGCAG-TAMRA		

PCR master mix components transported to exispin vortex centrifuge at 3000 / rpm for 3 min. Subsequently in Real-time PCR Thermocycler. Real-Time PCR thermocycler situations have been fixed in the relation to according to Taq Man kit instructions as in the follows (Table-2)..

Table 02- Taq Man kit instructions

Steps	Time/Temperature	Cycles
Pre-Denaturation	5 min in 95°C	1
Denaturation	20 sec in 95°C	45
Annealing Extension	30 sec in 60°C	45
Revealing	30 sec in 60°C	45

Real-Time PCR calculation data: The analyses of data have been done by CT value (threshold series number) that obtains positive amplification of *Toxoplasma gondii* B1 gene in Real-Time PCR cycle number.

Statistical analysis

Computerized analyses were performed by use version 17 SPSS, Chi-square test used to evaluate the variables (Joda, 2008).

Results

The infection rate of *Toxoplasma gondii* according to diagnostic methods

The results showed that the infection rates of *T. gondii* differ according to the method of diagnosis; the lowest rate (27%) with latex agglutination test and the highest (48%) by Real-time PCR test with significant differences (P≤0.05) (Table- 3).

Table 03- Four diagnostic methods show the infection rates of *T gondii* in aborted women

Type of Test		No. of sample examined	No. of positive samples	Percentage %
Latex agglutination test		100	27	27%
Rapid test	**IgG**	100	35	35%
	IgM	100	9	9%
ELISA	**IgG**	100	42	42%
	IgM	100	16	16%
RT-PCR	**B1 gene**	100	48	48%

*P≤0.05

Relationship between age groups of aborted women with diagnostic methods

The results revealed that females at age group 31- 40 years old had a higher rate of infection (P≤0.05) in comparable with other age group <20-30 that recorded lowest infection rate (Table-4)

Table 04- The infection rate of *Toxoplasma gondii* in aborted women in relation to age groups

Age group / Type of test			<20-30		31-40		Total	
			50 examined		50 examined		No.	%
			No.	%	No.	%		
Latex agglutination test		+ve	11	22	16	32	27	27
Rapid test	IgG	+ve	14	28	21	42	35	35
	IgM	+ve	3	6	6	12	9	9
ELISA	IgG	+ve	13	26	29	58	42	42
	IgM	+ve	5	10	11	22	16	16
RT-PCR		+ve	19	36	29	58	48	48

*P ≤0.05

Infection rate of *T. gondii* in the relation to residency

The frequency distribution of aborted women with *T. gondii* infection was lower in urban than in rural areas with significant differences (P ≤0.05) (Table-5).

Table 05- The infection rate of *Toxoplasma gondii* in aborted women in relation to residency

Residency / Type of test			Urban		Rural		Total	
			47 examined		53 examined			
			No.	%	No.	%	No.	%
Latex agglutination test		+ve	11	23.40	16	30.18	27	27
Rapid test	IgG	+ve	9	19.14	26	49.05	35	35
	IgM	+ve	4	8.51	5	9.43	9	9
ELISA	IgG	+ve	14	29.78	28	52.83	42	42
	IgM	+ve	3	6.38	13	24.52	16	16
RT-PCR		+ve	18	38.29	30	56.60	48	48

*P≤0.05

Specificity and Sensitivity of Real Time PCR compared to other metods for detection *Toxoplasma gondii* infection in aborted women.

The results showed that Real time PCR in highly sensitive (92%), specific (100%) and accurate, while the lowest sensitivity, specificity and accuracy is latex agglutination test (Table-6).

Table 06- Specificity and Sensitivity of Real Time PCR compared to other metods for detection *T. gondii* in aborted women.

Type of Test		Sensitivity	Specificity
Latex agglutination test		82	48%
Rapid test	IgG	64.55	78.17%
	IgM	31.48	91.25%
ELISA	IgG	60	88%
	IgM	82	94%
RT-PCR	*B1* gene	92	100%

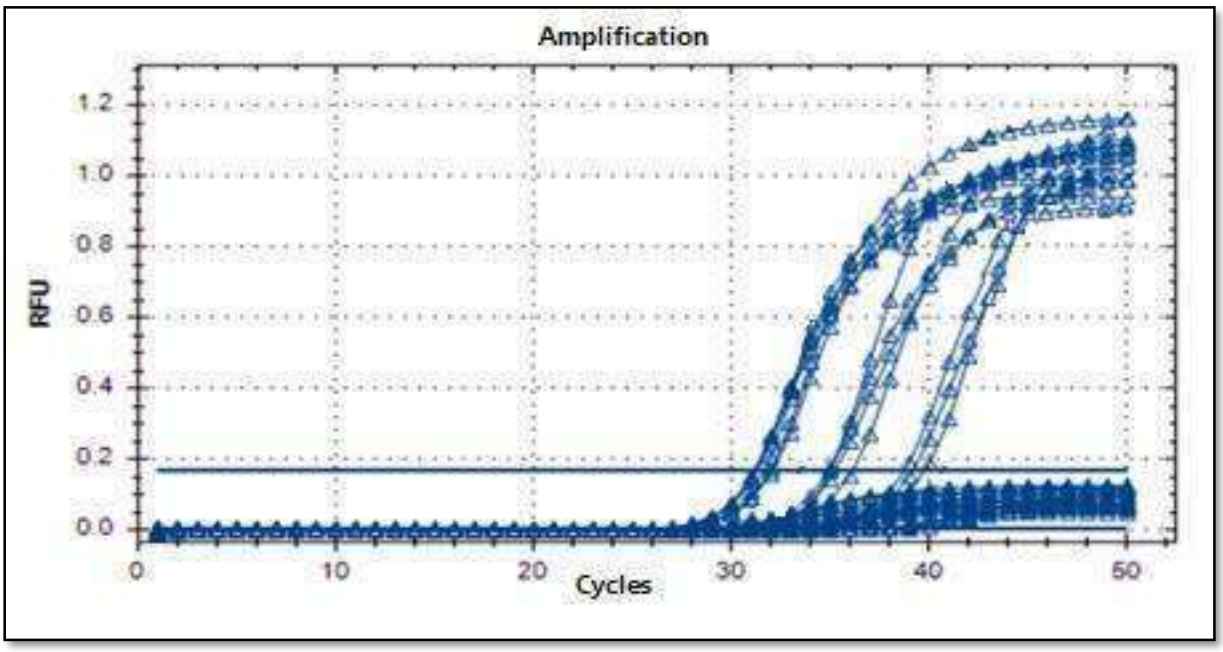

Figure 01- Positive results of toxoplasmosis in aborted women by qPCR based B1 gene. The positive amplification showed in threshold cycles 30-40 Ct values.

Discussion

The results of present study showed that the infection rates in aborted women varied according to diagnostic method used; The most sensitive and specific technique is Real-time PCR, while the lowest diagnostic method was Latex agglutination test. These results were in agreement with the results of [11] . whom showed that even only single parasite could be detected by PCR. [12] showed the highest diagnostic value is PCR. But disagree with [13] whom recorded latex test positive cases (62%) and molecular test PCR (51.61%). Serological

results are specific but may be give false positive results and this confirmed by the results, when small number obtained by PCR results [14,15,13]. In serological tests immunoglobulin IgM start to appear after 1- 2 weeks of the infection and reaches to the peak in 4th week and then decreases IgG begin appear in continue for longer time [16] .

The results of study showed high infection rate in 30-40 years old aborted women in comparative with young; this can be attributed to longer exposure to *Toxoplasma gondii*. These results were in agreement with [17,18,19,20,21] .that recorded the same reasons, *Toxoplasma gondii* infection increased with age. But disagree with [22] whom recorded the highest infection rate at age group 27-32 years old (40.90%).

Our results recorded *Toxoplasma gondii* prevalence in rural areas higher than in urban. These results were in accordance to [23 , 22, 16] that recorded that toxoplasmosis prevalence in aborted women was higher in rural areas than in urban. In contrast (AL-Taei *et al.,* 2015) recorded that there was close relation between increased *T. gondii* infection and residence in urban. Several causes for the spread of Toxoplasmosis in the rural more than urban is due to poor hygiene, lack of clean drinking water, low economic and social level, failure to conduct periodic examinations, and crowded living conditions in addition to livestock breeding, cats final host of parasite which increase chance of diseases transmission.

The results showed that Real time PCR test is significantly highly sensitive, specific and more accurate than the serological test, which in consistent with the results of (14,15, 22, 24, 12,16) that reported Real time PCR test is highly sensitive, specific and more accurate than the serological test. Real time PCR test is the best method for detection of *T. gondii* infection [20]

Conclusion

Real-time PCR is very sensitive, specific and accurate method for detection of pathogenic microorganisms in comparative with serological test, RT-PCR method detect acute, chronic and latent stages of infection in different samples (blood, tissue biopsy) and no possible for false positive results; that could be help detect the treatment.

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Molecular detection of *nif* H gene in plasmids of *Microbacterium* sp. bacteria isolated from the root nodules of *Medicago Sativa*

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Abstract

Medicago sativa L. root nodules were used as the source of an endophytic bacteria isolated on Yeast Extract Mannitol (YEM) media that has solidified. After 48 hours, the unique strain of the bacterium isolate took on a white color, a round form with a small convexity, and was Gram positive. The bacteria were resistant to the antibiotics 10µg/ml Ampicillin, 25µg/ml Amoxicillin, 30µg/ml Cefotaxime and 30µg/ml Tetracycline. The isolate from this study was determined to be at least 99% similar to the genus Microbacterium by DNA sequencing technique for analysis of the sequence of the nitrogenous bases of 16S rRNA gene with the world database, National Center for Biotechnology Information (NCBI). As a result, it was recorded for the first time as Microbacterium sp. ZAJ strain in NCBI and The isolated bacteria that produced these better findings had the nif H gene, which expressed one of the proteins for the nitrogen-fixing Nitrogenase enzyme.

Key Words

Medicago Sativa L.; Microbacterium Sp. Zaj; 16s Rrna; Nif H Gene.

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Introduction

The genus *Microbacterium* belongs to the phylum: Actinobacteria, the class: Actinobacteria, the order: Actinomycetales ,and family: *Microbacteriaceae* (Wang *et al*., 2014; Hadjadj *et al*., 2016), which is positive for the Gram stain. *A* non-symbiotic endophytic bacteria called *Microbacterium* colonizes the root nodules of legume plants. (Peng *et al*., 2015). About 90 species in this genus have been isolated from a variety of environments, including those linked with plants., *Microbacterium phyllosphaerae* and *Microbacterium foliorum*, for instance, were found in the phyllosphere of grasses (Behrendt *et al*., 2001). Azadirachta indica seedlings' rhizoplanes contain the plant-growth-promoting bacterium *Microbacterium azadirachtae*. (Madhaiyan *et al*., 2010). *Microbacterium yannicii* isolated from roots of *Arabidopsis thaliana* (Karojet *et al*., 2012). Additionally, *Microbacterium* strains have been discovered in a wide range of plants, including *Tagetes erecta*, *T. patula*, cotton, *Gossypium hirsutum*, sweet maize (*Zea mays*), (McInroy and Kloepper, 1995), (Sturz and Kimpinski, 2004), and wild legumes (Zakhia *et al*., 2006). Endophytic bacteria, in addition to rhizobia, have been found in root nodules of many legume plants, including *Medicago sativa* L. (Stajkovic *et al*., 2009) and *Ornithopus compressus* (Muresu *et al*., 2008). Which, through direct or indirect methods, play a significant impact in improving plant development and raising agricultural yield. (Peix *et al*., 2015). Numerous studies have demonstrated their ability to coexist while assisting nodulation even though they do not symbiotically induce nodules. (Martínez-Hidalgo and Hirsch, 2017; Zhang *et al*., 2018).

The enzyme *Nitrogenase* is responsible for biological nitrogen fixing, and it is a crucial stage in the entire nitrogen cycle. (Hu and Ribbe, 2015). Dinitrogenase (MoFe-protein) and dinitrogenase reductase (Fe-protein) are the two subunits that make up the *Nitrogenase* enzyme. (Hu and Ribbe, 2015). Four different *Nitrogenases* have been identified to yet. (Eady, 1996; Hofmann-Findeklee *et al*., 2000). So there were four different *Nitrogenase* enzymes identified; whether there are any other classes of *Nitrogenase* is unknown. (Hofmann-Findeklee *et al*., 2000), Three are oxygen-sensitive, and one is not; only Streptomyces *thermoviolaceus autotrophicus* has been recorded for them. (Zhao *et al*., 2006). The three varieties that are sensitive to oxygen are alternate vanadium *Nitrogenase* (*Vnf*), alternative iron only *Nitrogenase* (*Anf*), and conventional molybdenum (*nif*). (Hu and Ribbe , 2015) .

The Species *Medicago sativa* L. belongs to the Kingdom: *Plantae* , the Subkingdom: *Tracheobionta*, the Superdivision: *Spermatophyta* , the Division: *Magnoliophyta* , the Class:

Magnoliopsida, the Subclass: *Rosidae,* the Order: *Fabales,* the Family *Fabaceae* Lindl and the Genus: *Medicago* L. (Latifi and Akan 2020).

The ability of the *Medicago sativa* plant to form a symbiotic relationship with the bacterium *S. meliloti* results in the development of nodules that fix nitrogen increase soil fertility and enhance its quality, making it one of the most significant and widespread leguminous crops. (Saidi *et al.*, 2021). The *Medicago* genus includes several species, including *M.orbicularis, M.lupulinal, M. falcate, M.sativa* ,and *M. minima* (Crespi and Galvez, 2000).

This study's objectives included isolating bacteria from the root nodules of the *Medicago sativa* plant and diagnosing it at the molecular and microbiological levels. the *nif* H gene was also found.

Materials and Methods

Isolation of the bacteria from the nodules of *Medicago sativa* L. roots and their purification.

The seeds of the *Medicago sativa* L. (classification mentioned in the introduction) plant were obtained from Mosul, Iraq, local markets, and they were grown in soil from a home garden. The *Medicago sativa* plants were completely uprooted after 25 days of germination, with a little soil surrounding their roots to prevent any parts from being lost in the soil. They were checked periodically after germination for days to make sure that they started to form the root nodules, which were on the 18^{th} day. A portion of the root was taken with the fresh nodules, and the soil was then washed out of them by running water several times. Then, 5-7 nodules were removed, picked up, and surface sterilized by being soaked in 96% ethanol alcohol for 2 minutes, followed by 3% (NaOCl) Sodium hypochlorite for 15 minutes, before being washed four times with sterile DW to remove any remaining traces of sterile materials. The surface-sterilized nodules were cultured the surface of a solidNutrient Agar (NA) medium and then transferred to growth incubation at 28°C. (Aneja, 2003), After incubation crushed with 3.0 ml of normal saline using a sterile glass rod and then 0.1 ml of nodule suspension was spread on sterilized plates of solid YEM medium. At 28°C, the samples were incubated. A single well-isolated colony was selected to solid YEM medium in a petri plate and incubated at 28C° after the purity and uniformity of colony type were thoroughly evaluated through several re-streaked experiments (Kapembwa, 2016)

Preparation of isolated bacteria cell suspension

A lob full of the isolated bacteria was taken after 48 hours and put in a vial with 20 ml of liquid YEM medium. This suspension was then incubated at 28 °C for 48 hours in a shaker incubator with a rotational speed of 100 r/min (Godschalx *et al*., 2017).

Characterization of isolated bacteria from fenugreek root nodules

For bacteria isolated from fenugreek root nodules, the colony morphology of the bacterium was visually characterized.

Cultural characters

Observed for cell colony color, shape, Gram stain (Aneja, 2003).

Sensitive test for antibiotics

1.0 ml of a 48-hour-old suspension of isolated bacteria was cultured by spreading it across the 25 ml of solid YEM medium surface in a 9.0 cm petri dish containing the antibiotics ampicillin (10 g/ml), amoxicillin (25 g/ml), cefotaxime (30 g/ml), gentamycin (40 g/ml), and tetracycline (30 g/ml) (Platt, 2020).

16S rRNA amplification and sequencing by polymerase chain reaction (PCR)

Using a genomic DNA purification kit (Geneaid, Korea), genomic DNA was isolated in accordance with the manufacturer's instructions. A bacterial primer was used to amplify and sequence the 16S rRNA gene. Set F=5′- GACCTCGGTTTAGTTCACAGA -3′, R=5′- CACACGCTGACGCTGACCA-3′.

Mix the reaction components to create the reaction mixture in the Premix tubes. In a thermal cycler, DNA was amplified, employing the following steps: An initial 6 minutes of denaturation at 95 °C is followed by 35 cycles, each of which comprises of a denaturation cycle at 95 °C for 45 seconds, an annealing cycle at 56 °C for 1 minute, and an extension cycle at 72 °C for 1 minute. The amplified products were discernible on an agarose gel stained with 3.0 L Safe red stain at 1.0% w/v. Sequencing was done on purified PCR products at the 3130 genetic analyzer (Japan). Using the BLAST program at the National Center for Biotechnology Information (NCBI), the sequence acquired from isolated bacteria was compared to the nucleotide database available at the Gene Bank.

Detection of *nif* H gene in the DNA plasmid of seedlings nodules bacteria

The Wizard genomic DNA Purification kit from Promega (USA) is used. DNA plasmid of seedling nodules bacteria that generated from re-inoculation at the aforesaid procedure was separated, and its concentration and purity were determined by nanodrop. The *nif* H gene was amplified using PCR using the following primers:

F (5′CGTAGGTTGCGACCCTAAGGCTGA -3′)

R (5′-GCATACATCGCCATCATTTCACC-3′).

The PCR mixture, amplification conditions, and sequencing of the PCR products were carried out in accordance with (Mazard *et al*., 2004). The *nif* H gene was amplified using PCR as follows.: initial denaturation at 95C^{o} for 6 min for 1cycles, (denaturation at 95 C° for 45 Sec., annealing at 52C^{o} for 1 min, extension at 72C° for1 min) with 35 cycles and final extension at 72C^{o} ,5 min, for one cycle. The amplified products were evident on 1.0% agarose gel stained with Safe red stain.

Results and Discussion

Cultural characteristics of endophytic bacteria isolated from root nodules of *Medicago sativa* L.

After 48 hours of growth on solid YEM medium, the colonies of a unique strain of endophytic bacteria that was isolated from nodules on the roots of *Medicago sativa* L. were white in color and took on a slightly convex, round form (Fig. 1). The results showed that Gram's staining had a positive reaction. All of the aforementioned findings are consistent with what was stated regarding the endophytic Actinobacteria bacteria isolated from various crops. (Yu *et al*., 2013 ; Peng, 2015; Fidalgo *et al*., 2016).

Figure 01- Endophytic bacteria isolated from the root nodules of *Medicago sativa* L. grown on the YEM medium after 48 hours of incubation

Resistance and sensitivity to antibiotics

Results of this study's isolate growth on solid YEM medium that contained several antibiotic kinds demonstrated its capacity for resistance. 10 g/ml of Ampicillin, 25 g/ml of Amoxicillin, 30 g/ml of Ceftaxime, 30 g/ml of Tetracycline, and sensitive to 40 g/ml of Gentamycin (Table 1).

Table 01- Resistance and sensitivity to antibiotic test

Antibiotics (µg/ml)	Results
Ampicillin (10)	R
Amoxicillin (25)	R
Cefotaxime (30)	R
Gentamicin (40)	S
Tetracycline (30)	R

16s rRNA sequencing

Data revealed that the endophytic bacteria's DNA had a purity of 1.7 and a concentration of 244 g/ml. The electrophoresis in 1% agarose gel of the chromosomal DNA amplified product by PCR using the particular primer of the 16s rRNA gene resulted in the separation of a single band with a molecular weight of 585 bp (Fig. 2), it is identical to the molecular weight of the particular primer utilized.

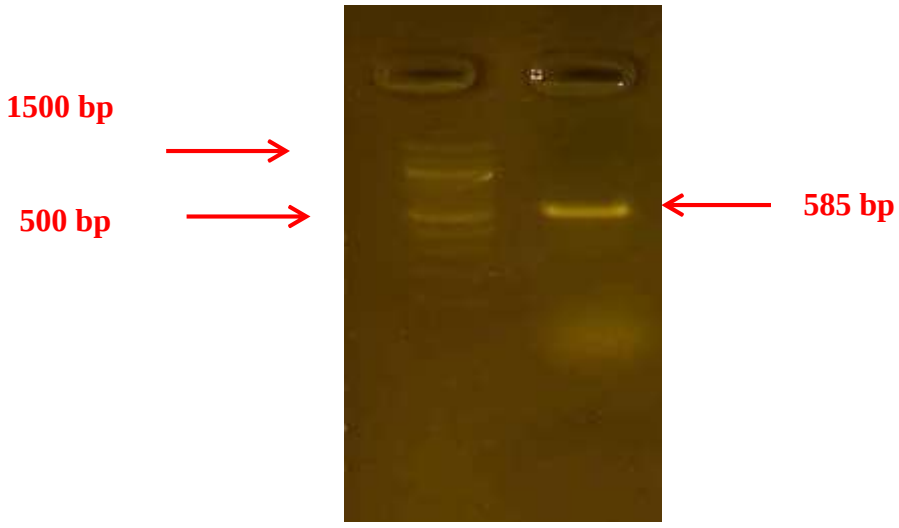

Figure 02- Electrophoresis of the DNA amplified to the 16s rRNA region by PCR and isolated from the studied bacteria in 1.0% agarose.

The study was able to register it for the first time as a novel strain and was given its number ON248047.1 and was known as *Microbacterium* sp. AJZ (Fig. 3). This was accomplished by transmitting the results of the nitrogenous bases sequences analysis to the gene bank at the NCBI.

Microbacterium sp. strain ZAJ 16S ribosomal RNA gene, partial sequence

GenBank: ON248047.1

FASTA Graphics

Go to:

```
LOCUS       ON248047                  550 bp    DNA     linear   BCT 21-APR-
2022
DEFINITION  Microbacterium sp. strain ZAJ 16S ribosomal RNA gene, partial
            sequence.
ACCESSION   ON248047
VERSION     ON248047.1
KEYWORDS    .
SOURCE      Microbacterium sp.
  ORGANISM  Microbacterium sp.
            Bacteria; Actinomycetota; Actinomycetes; Micrococcales;
            Microbacteriaceae; Microbacterium.
REFERENCE   1  (bases 1 to 550)
  AUTHORS   Qaddawi,Z.T., Mohammed,A.A. and Ahmed,J.A.
  TITLE     Molecular and family specifically diagnosis of bacteria
isolated
            from Medicago sativa root nodules
  JOURNAL   Unpublished
REFERENCE   2  (bases 1 to 550)
  AUTHORS   Qaddawi,Z.T., Mohammed,A.A. and Ahmed,J.A.
  TITLE     Direct Submission
  JOURNAL   Submitted (16-APR-2022) Biology, University of Mosul, Al-
majmoaa
            street, Mosul 41002, Iraq
COMMENT     Sequences were screened for chimeras by the submitter using
            Chromas.

            ##Assembly-Data-START##
            Sequencing Technology :: Sanger dideoxy sequencing
            ##Assembly-Data-END##
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                     /strain="ZAJ"
                     /db_xref="taxon:51671"
     rRNA            <1..>550
                     /product="16S ribosomal RNA"
ORIGIN
        1 ggtcaaggat gagctcgcgg cctatcagct tgttggtgag gtaatggctc accaaggcgt
       61 cgacgggtag ccggcctgag agggtgaccg gccacactgg gactgagaca cggcccagac
      121 tcctacggga ggcagcagtg gggaatattg cacaatgggc gcaagcctga tgcagcaacg
      181 ccgcgtgagg gacgacggcc ttcgggttgt aaacctcttt tagcagggaa gaagcgaaag
      241 tgacggtacc tgcagaaaaa gcaccggcta actacgtgcc agcagccgcg gtaatacgta
      301 gggtgcaagc gttatccgga attattgggc gtaaagagct cgtaggcggt ttgtcacgtc
      361 tgctgtgaaa actggaggct caacctccag cctgcagtgc gtacgggcag actagagtgc
      421 ggtaggggag attggaattc ctggtgtagc ggtggaatgc gcagatatcc cgaggaacac
      481 cgatggcgaa cggcagatct ctgggccgta actgacgctg agcagcgaaa gggtggggag
      541 caaacaggct
//
```

Figure 03- Register the novel *Microbacterium* sp. ZAJ with code ON248047.1 in NCBI

The researcher (Grison *et al*., 2015) isolated 56 isolates of rhizobia bacteria from the leguminous host *Anthyllis vulneraria* in southern France. These findings are consistent with many studies within the possibility of identifying the genetic variation between the studied isolates through the technique of analyzing nitrogenous bases sequences, where 16S rRNA region DNA sequencing method was applied by the researchers. The findings indicated that the strain ChimEc512T belongs to the genus *Rhizobium* and is, with a percentage of 98.4%, closer to the reference strain *Rhizobium* endophyticum CCGE2052T (NR-116477.1). There were two groups of isolates in a different study by (El-Zanaty *et al*., 2014) that examined the genetic

diversity using molecular diagnosis of the 16s rRNA region between eleven isolates of the symbiotic rhizobacterium *Rhizobium leguminosarum* symbiovar. viciae were taken from the root nodes of the *Vicia faba* L. plant, which was grown in 11 fields spread across various governorates in the Arab Republic of Egypt.

Detection of presence *the nif* H gene in the DNA plasmids of the bacteria isolated from M. sativa seedlings root nodules

Results of DNA plasmid electrophoresis in 1% agarose using specialized primers for the *nif* H gene, that encodes one of the proteins for the *Nitrogenase* enzyme, revealed the presence of a single band with a molecular weight of 370 bp, This is comparable to the particular primer employed in this study's molecular weight. (Figure, 4). These results are in agreement with (Qaddawi and Mohammed, 2021; Alqusaimy, 2020) and show that the isolated bacteria had the *nif* H gene, that encodes one of the proteins for the *Nitrogenase* enzyme that fixes atmospheric nitrogen.

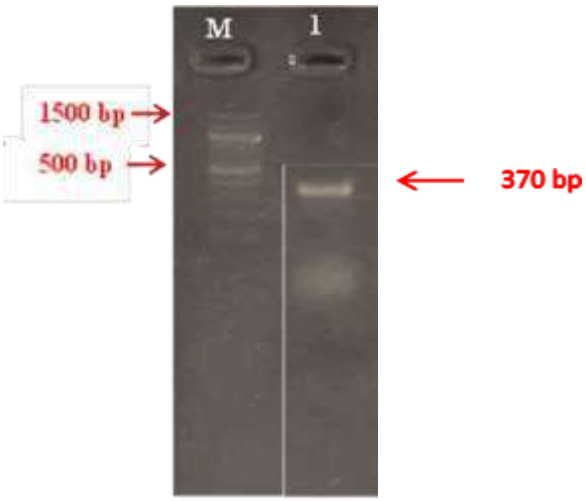

Figure 04- Presence the *nif* H gene in the DNA plasmid amplified and isolated from the *Microbacterium* sp. ZAJ by electrophoresese in 1.0% agarose. (M= Ladder , 1= Sample)

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Nuclear Apoptosis Inducing Factor 1 (NAIF1) Gene Expression In Non-Tumoral Human Tissues

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Abstract

Background: The nuclear apoptosis inducing factor1 (NAIF1) is frequently inactivated or not expressed in advanced human malignancies. Fascinatingly, no much data is available regarding the natural impression of NAIF1 expression in normal human tissue.

The purpose of this work is to describe how the NAIF1 protein is expressed in normal human tissues.

Materials and procedures: 50 samples of normal human tissue were examined using western blot and immunohistochemical method to determine the expression of NAIF1. Normal or inflammatory tissues without signs of cellular atypia were included as inclusion criteria in this work.

Results: NAIF1 was positively expressed by western blot and stained positively on all examined specimens by immunohistochemistry, which displayed either nuclear or cytoplasmic expression. It's interesting to note that NAIF1 staining patterns were consistent across diverse organ specimens. Although some cell types also exhibit cytoplasmic staining, NAIF1 is mostly found in the nucleus. Strongly expressing the nuclear protein was found in the epithelial layer of oesophagus, duct of breast. In connective tissue, NAIF1 has been found to express strongly in the cytoplasm of histocytes such as macrophages and dendritic cells.

Conclusion: By documenting the aspect of NAIF1 expression in healthy tissues, we may better understand how this protein works normally and assess the possibility of changed NAIF1 expression in human cancers.

Key Words

Expression; NAIF1; Immunohistochemistry; Western Blot; Normal Tissues.

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Introduction

On human chromosome 9q34.11, nuclear apoptosis-inducing factor 1 gene (NAIF1) located, encodes 327 amino acids [1,2].

This gene encode a protein which is expressed in multi organ of human tissues. NAIF1 has been identified to slow the growth, impede progress, and be associated with the prevention of development of numerous human cancers. NAIF1 overexpression inhibits the advancement of prostatic cancer cells.[3,4] NAIF1 has been discovered to be down regulated or missing and has an inhibitory role in the gastric carcinogenesis.[5] NAIF1 expression was down regulated in lung cancer, it's restoration in lung cancer cells decrease their capability to proliferate and survive without anchorage.[6]On the other hand, it is down regulated in osteosarcoma tissue and cell lines and can suppress their propagation and invasion.[7]

Although NAIF1 expression previously researched in numerous human tumors [3-11], NAIF1 expression in healthy human tissues not greatly explored. Identifying the pattern of NAIF1 expression probably help us in better understanding how this protein works in healthy tissues and give a scheme for interpreting unusual NAIF1 expression in human cancers. By western blot and immunohistochemical techniques, we clarified different localization of the NAIF1 protein both in nucleus and cytoplasm. In order to link NAIF1 expression with different stages of development within various organs, we thus want to carry out an extensive immunohistochemistry investigation of NAIF1 expression in healthy human tissues.

Materials and methods

Tissue samples

From surgical specimens, 50 samples fixed by formalin and paraffinized of non tumoral human tissues (Table I) were collected from the histopathology unit of the Trousseau hospital in Tours, France, have all been studied tissues. Normal/inflammatory tissue without signs of cellular atypia was one of the inclusion criteria. To validate the inclusion of each instance, a typical hematoxylin-eosin histological analysis was performed. At least two pathologists independently evaluated each slide.

Table 01- The partition of the examined normal specimens

No.	Tissue sort	Number of samples analyzed
1	Oral	5
2	Oesophagus	5
3	Breast	5
4	Skin	5
5	Liver	5
6	Spleen	5
7	Kidney	5
8	Bowel	5
9	Stomach	5
10	Hyaline cartilage	5
Total		50

Protein extraction for western blot technique

Protein extractions from tissue samples: sections of tissue samples kept at -80°C were prepared using a cryostat. Protein lysates were created from these tissue sections similar to the procedure that was already established by Arnaoty et al.[8] The Bradford technique was used to measure the protein extracts, and they were stored at -20 °C.

Utilizing acrylamide gel electrophoresis to analyze proteins

Using acrylamide gel electrophoresis to analyze proteins SDS-PAGE, a method previously described by Arnaoty et al. [8] Each well of a polyacrylamide gel received 50 micrograms of tissue-specific protein extracts.

Western Blot

Arnaoty et al previously described the protocol in details. [8] Using anti-NAIF1 primary antibody (In Cell Art, Nantes, France) at a dilution of 1: 250. A secondary antibody linked to goat anti-mouse IgG-HRP (Amersham, GE Healthcare) was then incubated for an hour at room temperature. Images were captured using the FUGI LAS4000 imager at which point the membranes were subjected to a chemiluminescence reaction for analysis (Amersham ECL Advance Western Blotting Detection Kit, GE Healthcare).

Immunohistochemical technique

Three formalin-fixed, paraffin-embedded slices from each organ under investigation were deparaffinized, rehydrated, and submerged in 0.5 % H2O2-methanol for five minutes. After

being microwaved in 0.01 M sodium citrate buffer (pH 6.0) for 10 minutes, sections were blocked in 10% normal horse serum in 5% milk for 30 minutes at room temperature. Using a 1:200 dilution in 1 % bovine serum albumin-phosphate buffered saline (BSA-PBS), the NAIF1 antibody (In-Cell-Art, Nantes, France) [8] was employed overnight at 4 °C. After that, sections were treated for an additional hour at room temperature with the avidin-biotin complex and biotinylated horse anti-mouse IgG (1:1000 dilution; Amersham, GE Healthcare). As a chromogen, diaminobenzidine was utilized, and then hematoxylin was used as a counterstain. The main antibody was left out as a negative control. The positive control was a previously identified positive normal human colon tissue.

Statistical analysis

Strong staining scores where immunoreactivity in >50% of cells stained, moderate staining scores where immunoreactivity in 10-50%, Negative and low where immunoreactivity less than 10%. All statistical analyses were performed using the program SPSS 22.0. The statistical analysis includes the use of proportions, frequency distributions, and descriptive statistics.

Results

Western blot results:

Applying anti-NAIF1 antibody (In-Cell-Art, Nantes,France)[12] showed a distinct NAIF1 expression product in each sample of different tissue tested at a molecular weight of 35 kDa, which is the same as the NAIF1 transposase[13] (Fig. 1).

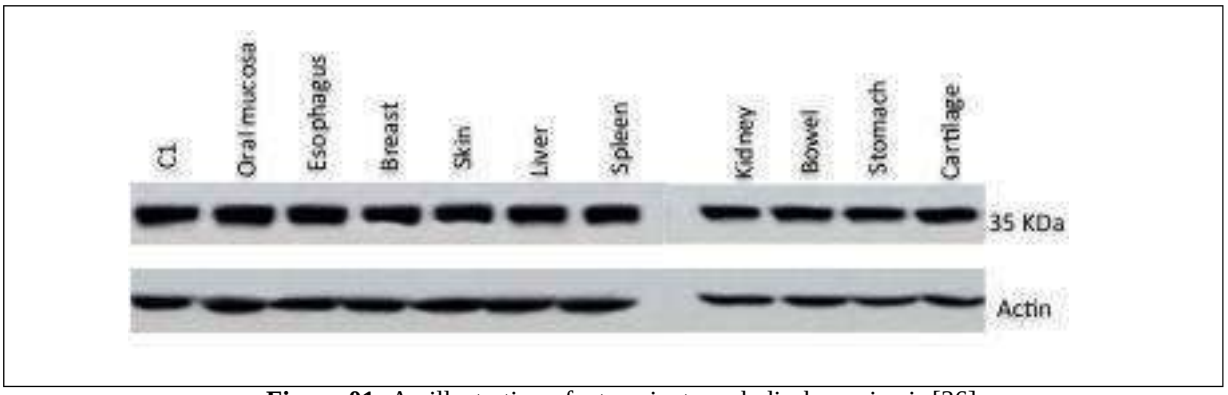

Figure 01- An illustration of a transient spark discharge in air [36]

Immunohistochemistry results:

The study included 50 samples that met the inclusion requirements. Table I lists the number and classification of the examined organs. All examined tissues had either nuclear or

cytoplasmic expression and stained positively with NAIF1. Surprisingly, regardless of fixation period, the NAIF1 staining pattern remained consistent across various specimens from each organ. The dissemination of NAIF1 expression in many sorts of cells and organs is shown in Table II.

Strong nuclear NAIF1 expression is found in ectoderm-derived skin, and oral mucosa, among other epithelial and ductal cells (Fig. 2). The majority of cell types that express NAIF1 have cytoplasmic staining as well, however the nucleus is where it is most commonly seen. Only basal and parabasal cell layers in the oral squamous epithelium expressed the NAIF1 protein, which is a striking pattern. On the other hand, the epithelial surface and glands of mesoderm or endoderm-derived organs, such as the stomach and intestine have demonstrated a preference for high cytoplasmic expression. In connective tissue, histocytes such as macrophages, dendritic cells, fibroblasts, and myofibroblasts have demonstrated significant cytoplasmic expression of NAIF1 (Fig. 1A). Nuclear staining was also present in these cells. Lymphocytes, plasma cells, and neutrophils expressing the NAIF1 protein could not be distinguished. In the lacunae of the hyaline cartilage, NAIF1 protein was shown to have cytoplasmic and nuclear localisation.

Table 02- NAIF1 expression in organs and cell types

Organ	Cell type	NAIF1 Cytoplasmic	NAIF1 nuclear
Oral	Mucosa	0	3
Oesophagus	Squamous epithelia	0	3
Breast	Ducts	0	3
Skin	Squamous epithelium	0	2
Liver	Hepatocytes	3	0
	Kupffer cells	0	2
Spleen	Macrophages	2	0
	Lymphocytes	0	0
Kidney	Tubular epithelium	0	1
Bowel	Mucosa (epithelium)	3	1
Stomach	Surface epithelium	2	1
Hyaline cartilage	Lucanae	3	1

Strong positive cells (3); moderate positive cells (2); few positive cells (1); Negative cells (0).

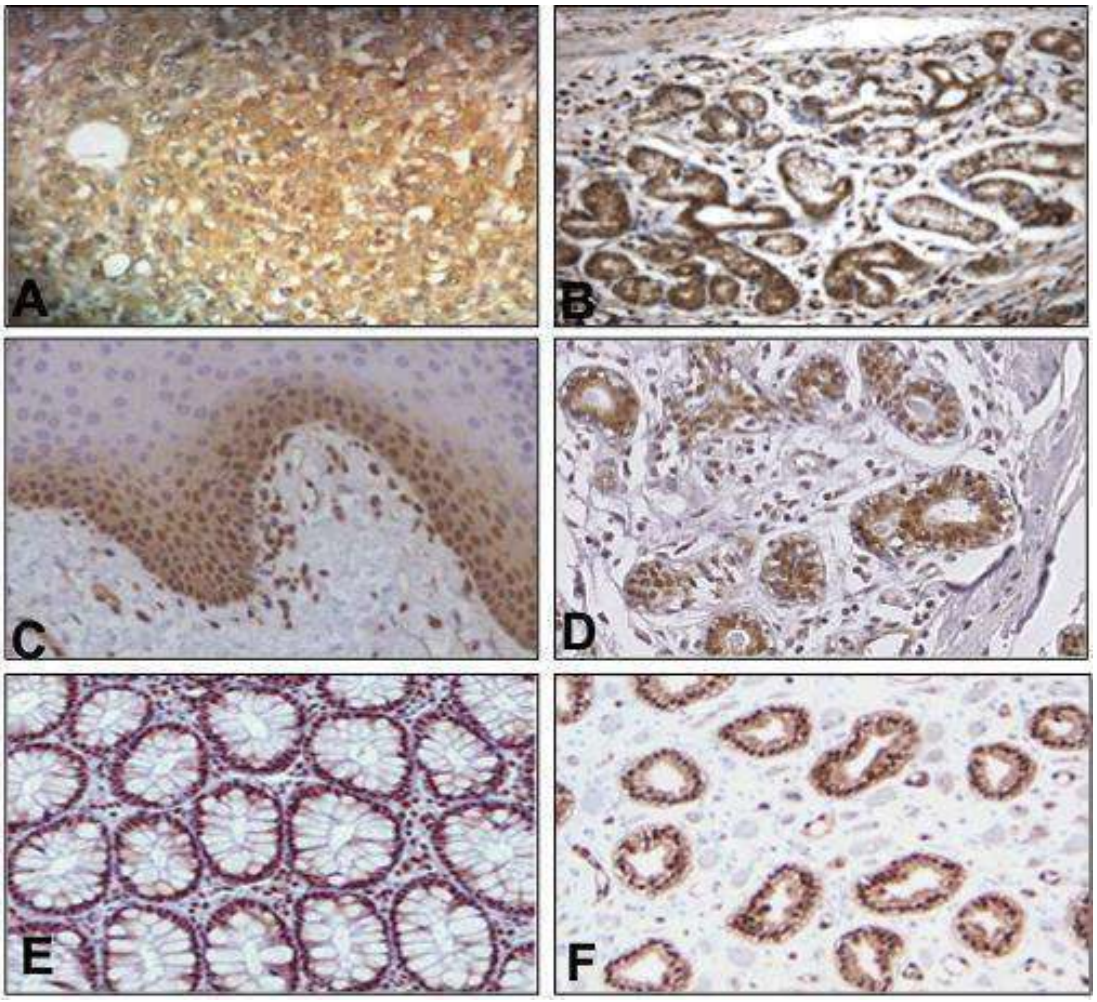

Figure 02- NAIF1 expression Immunohistochemistry in diverse human normal tissues. A) NAIF1 negatively expressed in lymphocytes and significantly expressed in cytoplasm of histocytes, phagocytes in spleen (×200). B) NAIF1 greatly expressed in the stomach epithelium (×100). C) NAIF1 was identified oral squamous epithelium. D) NAIF1 expressed in the nucleus of breast duct (×200). E) NAIF1 expressed actively in the surface epithelium of colon (×200). F) NAIF1 expressed firmly in the distal convoluted tubule of kidney (×200).

Discussion

NAIF1 has been shown to inhibit the growth of a number of human malignancies [3-11]. Numerous studies have documented the NAIF1 proteomic expression in human tissue either in cancer or healthy tissue [3-11]. Regarding its function in carcinogenesis, its considered as tumor suppressor gene by inducing apoptosis. The NAIF1 gene and the protein it encodes have been the topic of recent debate. Our findings in western blot (expression in multi organ tissues) are in accordance with earlier research concerning the expression of the NAIF1, which demonstrated that NAIF1 is expressed in cancer (low expression) and in normal or non-cancerous tissue (high expression) [3-11]. These results would suggest that NAIF1 is most likely to function as a tumor suppressor gene in different malignancies, potentially via activating apoptotic pathways. Furthermore, the high level of NAIF1 expression in healthy tissues may

indicate that NAIF1 contributes to the control of genes with anti-tumor action [8]. Numerous studies suggested utilizing immunohistochemistry to gauge the level of NAIF1 expression in a primary tumor because it enables the determination of the number of cells expressing the protein along with the intensity of expression [3,8]. Exploring intracellular localisation of NAIF1 and its role in the development or inhibition of tumors is a relatively unexplored subject. Our research revealed that NAIF1 protein appear in both nucleus and cytoplasm, with the latter possibly being in the plasma membranous system comparable findings to past studies showing that NAIF1 was mostly found in the cytoplasm [4] or in nucleus [1]. Our finding may be explained according to Lv B. et al. indicating that the NAIF1 protein's amino acids (1-70) may be a crucial area for nuclear localization as well as apoptotic induction [1,4]. Additionally, it's probable that amino acids (71-327) of NAIF1 are responsible for its location in the cytoplasm rather than the nucleus and diminish its role to inducing apoptosis [1,4].

Actually, there is considerable NAIF1 nuclear expression in the cells of the epithelia, including the cutaneous squamous epithelium, oral epithelium, esophageal epithelium and breast parenchyma. The glands and epithelial surface of organs that either evolved from mesoderm or endoderm, such as the stomach and colon, however, showed a profusion of NAIF1 cytoplasmic expression.

The most important feature is that the presence of NAIF1 protein in dendritic and macrophage nuclei within a visible microscopic backdrop occurs consistently and most visibly, which strongly implies that NAIF1 protein's nuclear signal function is related to the activity of histiocytes.

The monocytes, macrophages, Kupffer cells and the dendritic cells are all notable sources of stable nuclear NAIF1 protein. The neutrophil did not express NAIF1 protein in either the nucleus or the cytoplasm. Different NAIF1 roles in the cell may account for these differences in NAIF1 expression patterns. NAIF1 has been shown through in vitro research to decrease tumor cell development and trigger apoptosis at nuclear level. [3-5,9]. The role of NAIF1 nuclear localization in the histiocytes is currently unknown, but it is possible that it is not related to phagocytosis and migration because neutrophils and lymphocytes do not express NAIF1. The precise link connecting NAIF1's nuclear localization and its biological importance in histiocytes, however, has to be clarified in more extensive research. Further investigation is necessary to fully comprehend the physiological role of NAIF1.

Conclusion

Recording the pattern of NAIF1 expression in healthy tissues will help us understand how this protein normally behaves and how to interpret NAIF1 expression patterns that may be changed in human cancers.

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Ethical Clearance: Patients who agreed to take part in this sort of study, provided samples from the histopathology department of the Trousseau hospital in Tours, France.

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Study The Effects of *Foeniculum Vulgare* On Serological and Biochemical Traits in Broiler Chicks

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Abstract

The present study was carried out to investigate the protective role and beneficial effects of different concentrations of Foeniculum vulgare (fennel seed) powder in chicks. Thirty Cobb broiler chicks at one-day old with an average weight 43 ± 5 g were sexed and randomly divided into 3 equal experimental groups; the first group represented as control group, male chicks in this group were fed the basal diet. The second (TF1) group and the third group (TF2): male chicks in these groups were fed a powdered Foeniculum vulgare at 5% and 10%, respectively, daily for the 42 days. At 42 days old, the live body weight (LBW) and blood samples were collected for hematological parameters estimation. While sera separated after centrifugation to estimate some biochemical parameters. The current results showed a significant increase ($p<0.05$) in body weight, RBC, WBC counts, Hb, total protein, and globulin concentrations in TF2 and TF1 groups, respectively, compared with the control group. The current results showed a significant decline ($P<0.05$) in Alkaline Phosphatase, and malondialdehyde levels in the TF2 and TF1 groups compared to control chicks. On the other hand, the results recorded a significant elevation ($p<0.05$) in GSH-Px, FSH, LH, and testosterone concentrations in chicks that received 10% and 5% of fennel seed compared with the control group.

Conclusion: It can be concluded from present findings, that dietary Foeniculum vulgare seed supplementation can improve the general health of chicks via hepatoprotective and antioxidant activity that is reflected positively on all studied parameters in broiler chicks that encourage economic and agricultural production.

Key Words

Fennel seeds; Physiological; Live body weight; Immunity; Broiler chicks.

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Introduction

The poultry industry is highly productive and rapidly expanding, and various strategies have been taken to prevent disease outbreaks and enhance immunity. Herbs and herbal products are commonly utilized in this industry to encourage the efficient utilization of feed nutrients, leading to faster and higher weight gain, increased production rates, and better feed quality. Moreover, the active compounds found in herbs may boost digestion and the immune system of broilers (Khafaji, 2018).

Several studies have identified medicinal plants as substances that promote growth. Due to their natural and non-toxic properties, it is acceptable to include medicinal herbs in animal and poultry feed(AL-Kafajii, 2013) (Khan et al., 2022). Botanicals and their constituents have the potential to enhance the development and health of chickens without causing any negative effects on their blood profiles or inducing pathological disorders, indicating that botanicals can serve as effective antibiotic growth promoters (Hafeez and Khan, 2020) and (Abd El-Hack et al., 2022). The seeds of fennel (*Foeniculum vulgare*) are a commonly used agricultural product in the Asia Mediterranean region, known for their gastrointestinal benefits (Khan et al., 2022).

The *Foeniculum vulgare* mill, also known as fennel, belongs to the Apiaceae family and is known for its properties such as stomach tonic, carminative, milk secretion-enhancing, bracing, laxative, and appetizer (Dhayalan et al., 2015). The use of fennel seeds (*Foeniculum vulgare*) can help prevent gastrointestinal problems such as flatulence and colic because it has the ability to stimulate and enhance the digestive enzyme, while also improving the performance and production of poultry in Iraq (Mohammed and Abbas, 2009) and (Kazak et al., 2020)

Additionally, studies have shown that fennel seed oil a rich antioxidants and exhibits antifungal and antibacterial properties, can suppress the growth of Clostridium perfringens and Enterococcus in the rat colon. Furthermore, fennel seeds have been found to have hepatoprotective properties, as determined by extensive research (Ruberto et al., 2000) (Renjie et al., n.d.)

With the increasing demands of livestock and poultry breeding with the growing number of diseases, it is necessary to develop strategies to produce healthy poultry using natural additives. Fennel (*Foeniculum vulgare* Mill.) has been used as a natural dietary additive in poultry production by some researchers, but there are few studies on the beneficial effects of dietary fennel seeds powder supplementation on broilers chicks in Iraq, so the current research was aimed to explore the impact of this study aimed to investigate the beneficial effects of

different levels of dietary fennel seeds powder supplementation on body weight, hematological, biochemical, some oxidant and anti-oxidant parameters in broiler chicks.

Materials and methods

The seeds of fennel (Foeniculum vulgare L.) were collected from the local market of herbal medicine in AL-Qassim city. 500 g of seeds was ground by blender. The powder seeds were stored dry in plastic bags for this study.

Animals

Thirty-one-day old of Cobb broiler chicks with an average weight 43 ± 5 g were used in this study and obtained from the agricultural hatchery of Agriculture College / Kerbala University. After determining the sex of the chicks by wings band were randomly divided into 3 equal experimental groups. All laboratory analyses of the experiment were carried out in the College of Veterinary Medicine, Al-Qasim Green University from 22/12/2022 to 15/3/2023.

All chicks were reared in cages with dimensions 9×4 m under controlled conditions about lighting, which was provided 24 hours daily during the first week, after that, it was reduced to 23 hours until the end of the experimental period, the temperature in chicks' cages was about 36°-37° C for the first 5 days of age, after that, it was reduced 2 degrees weekly up to 27° C. All chicks vaccinated as follows:

1. Gumboro vaccine (Fatro, Italy) at ages 7^{th} , 14^{th}, and 23^{rd} with drinking water.
2. Newcatle vaccine (Fatro, Italy) at ages 10^{th}, 20^{th}, 30^{th} with drinking water.

Free access water and feed were available at all times. All chicks were fed a starter diet until from 1st day to 21st day age with crude protein 22.21% and metabolized energy 2911Kcal/kg diets, then chicks were fed a finisher diet from 22nd day to 42nd day with crude protein 19.23% and metabolized energy 3130.68 Kcal/kg diets (table 1).

Table 01- Composition of experimental diets in different periods of the experiment.

Ingredient (%)	**Starter (1st - 21st day)**	**Finisher (22nd – 42nd day)**
Yellow corn	58	64
Soybean meal (45% protein)	38.0	32.0
*Premix	3.0	3.0
Oil	0.5	0.5
Salt	0.3	0.3
Methionine	0.1	0.1
Lysine	0.1	0.1
Total	100	100
Composition		
Metabolized energy (Kcal/kg)	2911	3130.68
Crude protein (%)	22.21	19.23
Calcium (%)	0.13	0.23
Avail. Phosphorus (%)	0.17	0.16
Methionine + cysteine	0.80	0.75
Lysine	1.22	1.15

Premix (1%) provided the following (per kg of complete diets): 1400 IU Vit. A, 3000 IU Vit. D3, 50 mg Vit. E, 4 mg Vit. K, 3 mg Vit. B6, 6 mg Vit. B12, 60 mg niacin, 20 mg pantothenic acid, 0.2 mg folic acid, 150 mg choline, 4.8 mg Ca, 3.18 mg P,100 mg Mn, 50 mg Fe, 80 mg Zn, 10 mg Cu, 0.25 mg Co and 1.5 mg iodine.

Experimental Design

Thirty Cobb broiler male chicks were randomly divided into 3 experimental groups, 10 birds in each group. Each group contained 2 replicates of 5 birds:

- First group: Control group, chicks in this group fed basal diet.
- Second group (TF1): Chicks in this group were supplemented powdered Foeniculum vulgare seeds as 5% daily for 42th day (Al-Sagan et al., 2020).
- Third group (TF2): Chicks in this group were supplemented powdered Foeniculum vulgare seeds as 10% daily for 42th day (Al-Sagan et al., 2020).

Blood collection:

At 1st and 42nd days old the live body weight of each group will be estimated. At the end of the experimental period, blood samples were taken from the wing vein, and divided into two parts, 1st part 1.5 ml put into Ethylene Diamine Teteraacetic Acid (EDTA) tubs for hematological estimation: Red blood cells count (RBCs), White blood cells count (WBCs), Packed cells volume (PCV) and Hemoglobin concentration (Hb) according to the methodology of (Campbell and Ellis, 2007). 2nd part 3.5 ml put in tubs without anticoagulant. Isolation of serum had been done by centrifugation of blood samples at 5000 rpm for 10 min to obtain serum for biochemical analysis that are total Protein, Albumin, Globulin, Alanine aminotransaminase (ALT), Asparatate aminotransaminase (AST) and Alkaline Phosphatase (ALP),

Malondialdehyde (MDA), Glutathione peroxidase (GSH-Px) and some hormones testosterone,FSH, and LH.

Determination of total protein, Albumin, and Globulin:

A total protein was determined by Biuret method via using spectrophotometer, while albumin is estimated by Bromocresol Green Method according to (N. Tietz, 1995), Globulin was calculated (total protein- albumin).

Determination of AST, ALT, and ALP Activity in Serum:

It depends on colorless methods to detect the activity of Alanine aminotransaminase, Asparatate aminotransaminase and Alkaline Phosphatase (ALP) activity in serum were performed by kit (Bio Merieux, France) (Belfield and Goldberg, 1971).

Determination of malondialdehyde and glutathione peroxidase concentration: Plasma enzyme activities were determined using commercial kits and following the manufacturer's instructions, particularly malondialdehyde level (Biosystems-Spine) and glutathione peroxidase activity (Biosystems-Spine) according to (Buege and Aust, 1978).

Determination serum LH and testosterone:

For determined serum LH, FSH, and testosterone concentrations by using ELIZA technique according to procedure of instruction of kits (Biotech Fine Co., Ltd., China)

Statistical analysis:

The study data were analyzed with SPSS software version 16. All results parameters are presented as mean ± SD. Differences between quantitative data were analyzed with one - way ANOVA, followed by the LSD test. P - Value less than 0.05 were considered significant for all data shown in our results (Bryman and Cramer, 2011).

Results and Discussion

1. **Effect of fennel seeds on Live Body weight of broiler chicks:**

The date represented in figure (1) showed a significant elevation ($p<0.05$) on the live body weight of chicks fed 10% fennel seeds at 42nd day when compared with chicks fed 5% and control chicks. As well as, chicks of TF1 reported a significant increment ($p<0.05$) in their body weight in comparison with control chicks.

The current results showed an increment in body weight in chicks taken fennel seeds compared with control chicks that may be some components of the essential oils in fennel are stimulants and they stimulate secretion of digestive and gastric juices, while reducing inflammation of the stomach and intestines and facilitating proper absorption of nutrients from the food (Al-Sagan et al., 2020). Anethole and estrgole have digestive stimulating and appetizing effects (Çabuk et al., 2014).

Furthermore, (Liu et al., 2021) indicated that fennel stimulates the flow of digestive juice in the stomach and intestine and increase the efficiency of broken fats to fatty acids. They also noted that the anethole 'active component in fennel seeds' affected pathogen microorganisms in digestive system and increased live body weight and improved feed conversion ratio and summarized that supplementation of broiler diets with different levels of fennel was improved weight gain, feed efficiency and some carcass and blood characteristic (Liu et al., 2021).

2. Effect of fennel seeds on hematological parameters of broiler chicks:

The results in this study showed a significant increase in red blood cells (5.223±0.109) and white blood cells counts (27.248±0.959) ($p<0.05$) in TF2 group that fed grinding fennel seed at percentage 10 % when compared with TF1 and control group, figure (2 and 3) for RBC and WBC, respectively. However, there was a significant ($p<0.05$) increase in RBCs (4.917±0.212) and WBCs (21.839±1.619) in TF1 group that fed grinding fennel seed at percentage 5%, when compared with control group (3.984±0.143) and (15.783±1.19) for RBC and WBC respectively, as shown in figure (2 and 3).

The results in this study reported a significant increase in PCV (38.505±0.806) and HB concentration (12.800 ±0.600) ($p<0.05$) in TF2 chicks that fed 10% of grinding fennel seed when compared with TF1 and control group. However, there was significant increase in PCV (33.917 ± 0.852) and HB concentration (10.776±0.500) ($p<0.05$) in TF1 group that fed grinding fennel seed at percentage 5% , when compared with control group (28.692±0.783) and (8.160±0.78) for PCV and HB respectively, as shown in figure (4 and 5).

The results of hematological characteristics have been demonstrated in figure (2,3,4 and 5). The chicks that fed 5 and 10 % fennel seeds showed a significant increment in RBCs counts, Hb and PCV compared with the control group, these changes may be due to the biological active components of fennel seed such as lignin and flavonoids, as well as, the fennel seed improved nutrients absorption and metabolism, as well as, it stimulate erythropoiesis in bone marrow by enhancing erythropoietin secretion from kidney cells as a result elevated a counts of red blood

cells in consequence increment hemoglobin concentration and packed cell volume(Mansouri et al., 2015).

Besides, the supplementation of fennel seeds could enhance the absorption iron from digestive system that may be promoted erythrocytosis and hemoglobin formation by available the principle elements for these processes (Mansouri et al., 2015). The current results agreement with the findings of (Mohammed and Abbas, 2009) who used fennel seeds as dietary supplementation on broiler chick diets with levels 1, 2, and 3 g/kg of fennel seeds, and found an elevated in erythrocyte counts, hemoglobin concentration, and percentage of packed cells volume that was attributed to biologically active components of fennel seeds that promoted iron absorption, as well as, fennel seeds are concentrated source of minerals like copper, calcium, iron, potassium, magnesium, selenium, and zinc, manganese. Copper and iron required for hematopoiesis positively reflected on the above blood parameters (Singh et al., 2006).

Furthermore, the present study concluded the beneficial effect of fennel seed on blood leukocyte count due to the biological components of seed that enhanced WBC formation, which is agreement with the findings of (Mohammed and Abbas, 2009) Leukocytes constitute the most important defensive cells of the blood, Leukocytes are transported by a regular circulation of blood to lymph, from the inside of the vessel to the outside and from within the tissue toward the blood, these cells apply full and comprehensive supervision on various parts and the proprietary tissues of the body and protect the body against pathological factors (Mansouri et al., 2015). In the present study, an increasing number of WBC was observed in the experimental group.(Cherng et al., 2008) investigated the immunomodulatory effects of fennel seed extract on peripheral blood mononuclear cells (PBMC) and indicated that the extract of fennel has a strong stimulating effect on PBMC cells of experimental models(Cherng et al., 2008).

Some compositions of the fennel seed extract are Trans-Anethole, myrcene, fenchone, limonene, and alpha-Phellandrene (Cherng et al., 2008). In addition, the components of seeds like sterols, triglycerides, flavonoids, and coumarins have pharmacological properties such as analgesic, anti-fever, and anti-microbial effects, the entimicrobial properties of ethanolic extract of fennel have been proven (Saeedi et al., 2010). Anethole has antioxidant and anti-inflammatory effects and wound-healing activity in experimental models (Mansouri et al., 2015). Therefore, a significant increase in WBC by the dietary addition of fennel seeds in this study can confirm the anti-inflammatory and anti-microbial effects of this herb.

3. Effect of fennel seeds on Biochemical parameters of broiler chicks:

A. Effect of fennel seeds on total protein, albumin and globulin concentrations of broiler chicks:

The results in this study showed significant increases ($p<0.05$) in serum total protein (5.828 ± 0.371) and globulin concentrations (3.061± 0.379) in the TF2 group when compared with chicks fed 5% fennel seed and control group. As well as, there were a significant ($p<0.05$) increases in total protein (4.928± 0.608) and globulin concentrations (2.588± 0.155) in the TF1 group when compared with the control group (3.969±0.262) and (2.163±0.098) for total protein and globulin concentrations respectively, as shown in figure (6 and 7). While there were non-significant differences ($p>0.05$) in albumin concentration among treatment groups, as shown in Figure (8).

The recent study confirmed that dietary intake of chicks with 5 &10% fennel seeds for 42 days could be enhancing peptide and amino acids uptake by a cell that supports the synthesis of serum proteins that enter in the synthesis of many varies serum proteins or globulins that reflected on current results (Ruberto et al., 2000). This may be attributed to the biological components of fennel seed that have immunomodulatory effects on the stimulation of peritoneal macrophages of mice and the production of nitric oxide and ROS. On the other hand, fennel oil supplementation caused a significant increase in total protein, globulin, IgM, and IgG but has no effect on albumin (Roby et al., 2013), these results agreement with current results.

B. Effect of fennel seeds on ALT, AST, and ALP concentrations of broiler chicks:

The results of this study showed that there were non-significant differences ($p>0.05$) among all experimental groups in the activity of alanine aminotransaminase and asparatate aminotransaminase concentrations figure (9 and 10), respectively. Whereas, there was a significant ($p<0.05$) decline in Alkaline Phosphatase, ALP (241.666±3.918) in the TF2 group when compared with TF1 and control chicks, in addition, there was a significant ($p<0.05$) decrease in ALP (249.226±3.971) in TF1 group when compared with the control group, as shown in the figure (11).

The current results showed a non-significant decrement in AST and ALT levels, while significant decrement in ALP concentration, this indicated the important hepatoprotective role of Foeniculum vulgaris seeds constitutes to protect hepatocytes from any toxic as recorded by (Özbek et al., 2003), who clarified that the essential oil roles of fennel seeds to reduce and inhibit the CCl4 effects that induced acute hepatotoxicity by decreasing levels of serum aspirate

aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase (ALP) and bilirubin. D-limonene and β-myrcene of the fennel oil might be the potential candidates.

Besides, the current results constant with (Özbek et al., 2003) showed that significant decrement in AST, ALT, and ALP, which are attributed to anethole, D-limonene and β-myrcene compounds found in fennel seeds have a potent hepatoprotective action; D-limonene increases the concentration of liver glutathione (GSH) which is required by several enzymes that participate in the formation of the correct disulfide bonds of many proteins. β-myrcene elevates the levels of apoproteins, which are subtypes of the P450 enzyme system that catalyze the oxidative metabolism of a wide variety of exogenous chemicals including drugs, toxins, and endogenous compounds such as fatty acids (Özbek et al., 2003). Additionally, the Foeniculum vulgare (fennel) seeds have hepatoprotective activity via a decline in hepatic enzymes as illustrated by(Huang et al., 2010) who reported a decline in serum AST, ALT and ALP after induced toxicity by CCl4 due to actions of Foeniculum vulgare seeds.

C. Effect of fennel seeds on malondialdehyde (MDA), Glutathione peroxidase (GSH-Px), and reproductive hormones concentrations of broiler chicks:

The results in this study showed that there was a significant ($p<0.05$) decrease in malondialdehyde level (1.949 ±0.104) and (2.451±0.171) in TF2 and TF1 groups, respectively, when compared with the control group (3.788 ± 0.548), as figure (12). On the other hand, the results in this study reveal that there was a significant elevation ($p<0.05$) in Glutathione peroxidase (GSH-Px) concentration in chicks that received 10% of fennel seed with a mean value (39.851±0.409) ($p<0.05$) when compared with TF1 and control group. In addition, there was a significant ($p<0.05$) increase above the parameter in chicks fed 5% fennel seed (35.803± 0.785) when compared with the control group (30.317 ± 0.441), as shown in figure (13).

In addition, the results recorded significant ($p<0.05$) elevation in LH, FSH, and testosterone concentrations in TF2 and TF1 when compared with control group figure 14,15,16.

The results of this study revealed that there was a significant elevation ($p<0.05$) Glutathione peroxidase (GSH-Px) concentration in chicks fed fennel seeds and a significant decline in malondialdehyde, that changes may be due to the ability of Fennel to exhibit a good radical scavenging activity. A significant enhancement in the activities of antioxidant enzymes were observed in diets containing fennel (Nickavar and Abolhasani, 2009).

Moreover, (Rezq, 2013) reported that feeding supplemented diets with different levels of fennel seeds significantly increased the serum activity of Superoxide dismutase (SOD) and

GSH-Px enzyme compared to the control group. Feeding chicks with high fat-diet supplemented with fennel seeds significantly reduced serum MDA levels compared with positive control (Rezq, 2013).

Fennel seeds reduced oxidative stress and improve antioxidant defense. This action of fennel may be related to its antioxidant activity (Ruberto et al., 2000). This effect may be due to the fennel content of phenolic and flavonoid compounds which enhance the activity of the antioxidant system (Özbek et al., 2003). Flavonoid and phenolic compounds possess antioxidants that have a radical scavenging mechanism, resulting in a potential alteration of physiological antioxidant status as elevation in GSH-Px and decline in malondialdehyde, as current results (Zayachkivska et al., 2005). The biological activities of the flavonoids are related to their antioxidant activity by various mechanisms, e.g. by scavenging or quenching free radicals, by chelating metal ions, or by inhibiting enzymatic systems responsible for the generation of free radicals (Mojžišová and Kuchta, 2001). Fennel seeds can improve the reproductive hormones that enhanced fecundity via the bioactive constituents of flavonoids and phytoestrogenic compounds that improve the enzyme responsible for the biosynthesis GnRH in the hypothalamus in turn increasing synthesis and release of FSH and LH from pituitary glands. Fennel seed increased the synthesis FSH and LH in turn improved spermatogenesis in Sertoli cells and induced the synthesis of testosterone in Leydig cells, respectively (Shahsavari et al., 2022).

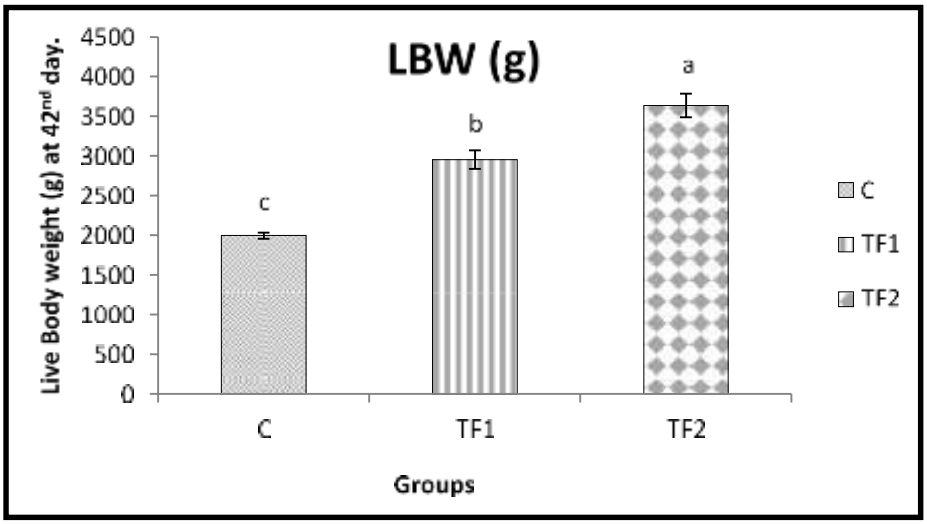

Figure 01- Effect of fennel seeds powder on live body weight of broiler chicks at the end of the experiment (42nd day). Different letters denote significant differences ($p<0.05$) between groups. The data represented mean ± standard deviation. C: chicks fed basic diets. TF1: chicks fed basic diets mixed with 5% ground fennel seed and TF2: chicks fed basic diets mixed with 10% ground fennel seed

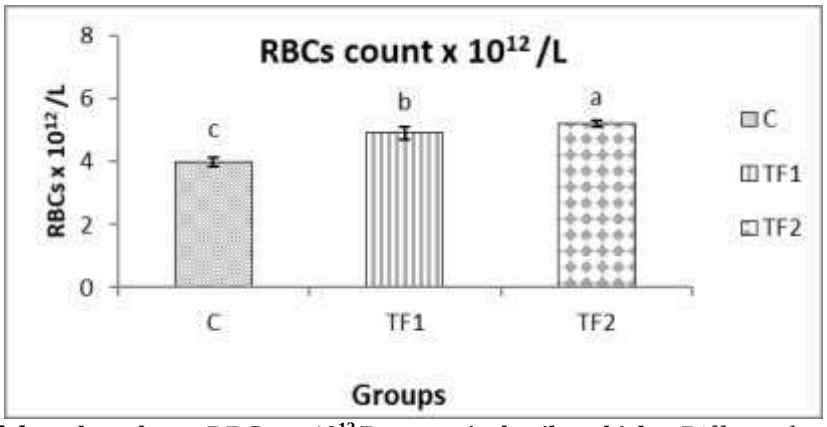

Figure 02- Effect of fennel seeds on RBCs x 10^{12}/L count in broiler chicks. Different letters denote significant difference ($p<0.05$) between groups. The data represented mean ± standard deviation. C: chicks fed basic diets. TF1: chicks fed basic diets mixed with 5% ground fennel seed and TF2: chicks fed basic diets mixed with 10% ground fennel seed.

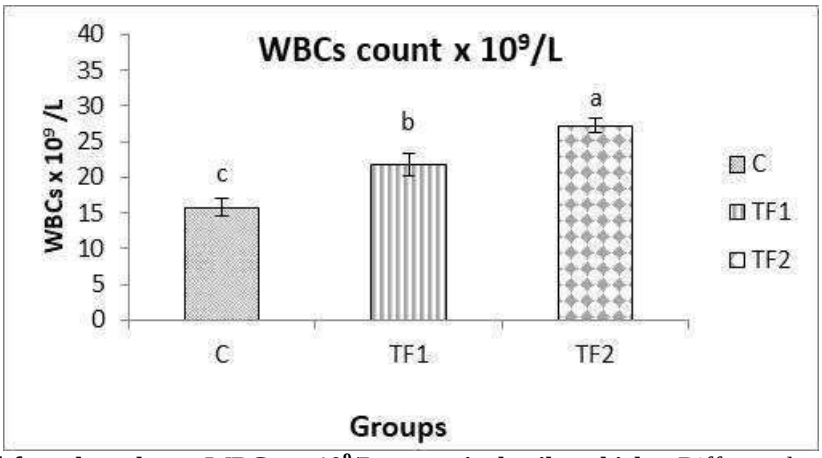

Figure 03- Effect of fennel seeds on WBCs x 10^{9}/L count in broiler chicks. Different letters denote significant difference ($p<0.05$) between groups. The data represented mean ± standard deviation. C: chicks fed basic diets. TF1: chicks fed basic diets mixed with 5% ground fennel seed and TF2: chicks fed basic diets mixed with 10% ground fennel seed.

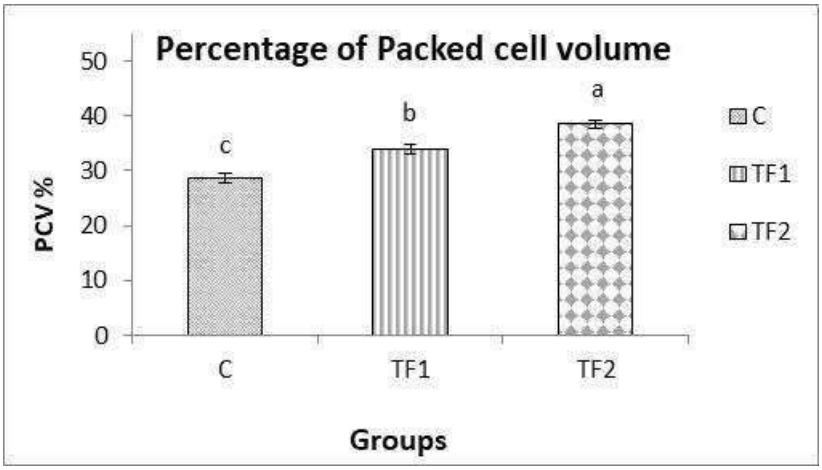

Figure 04- Effect of fennel seeds on packed cell volume % in broiler chicks. Different letters denote significant difference ($p<0.05$) between groups. The data represented mean ± standard deviation. C: chicks fed basic diets. TF1: chicks fed basic diets mixed with 5% ground fennel seed and TF2: chicks fed basic diets mixed with 10% ground fennel seed.

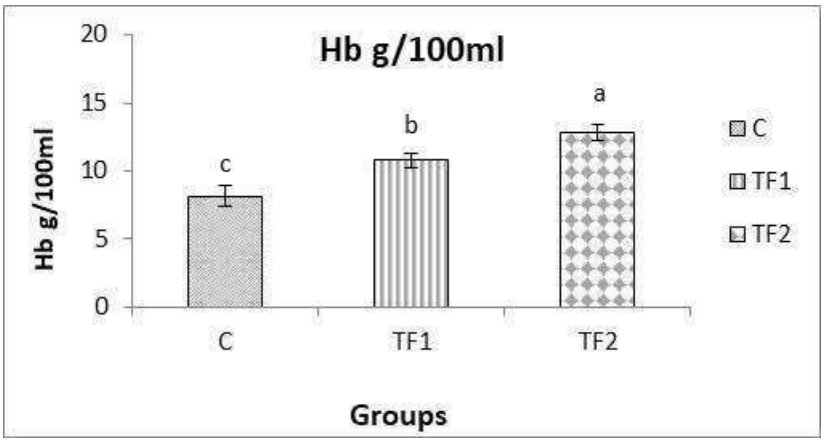

Figure 05- Effect of fennel seeds on hemoglobin concentration (g/100ml) in broiler chicks. Different letters denote significant difference ($p<0.05$) between groups. The data represented mean ± standard deviation. C: chicks fed basic diets. TF1: chicks fed basic diets mixed with 5% ground fennel seed and TF2: chicks fed basic diets mixed with 10% ground fennel seed.

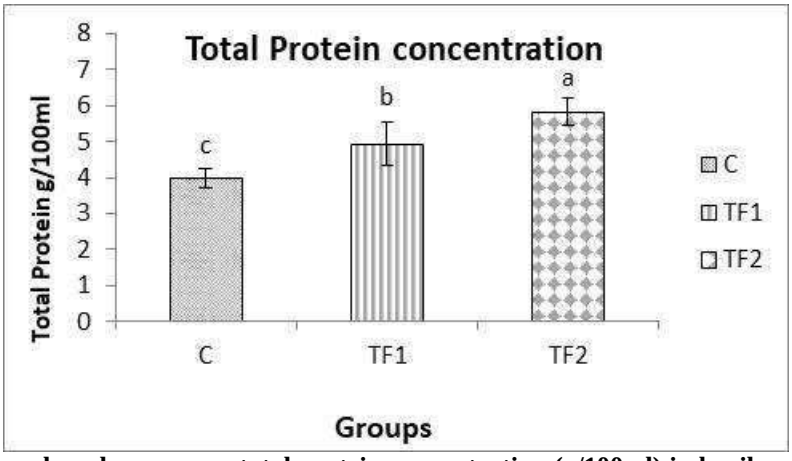

Figure 06- Effect of fennel seeds on serum total protein concentration (g/100ml) in broiler chicks. Different letters denote significant differences ($p<0.05$) between groups. The data represented mean ± standard deviation. C: chicks fed basic diets. TF1: chicks fed basic diets mixed with 5% ground fennel seed and TF2: chicks fed basic diets mixed with 10% ground fennel seed.

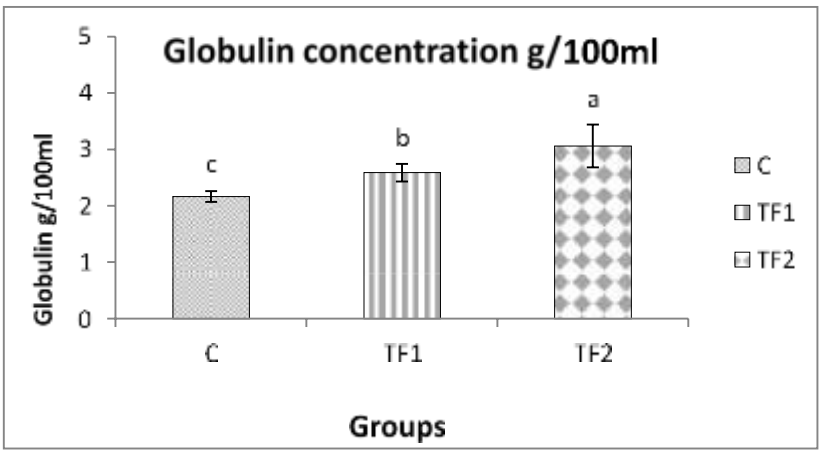

Figure 07- Effect of fennel seeds on serum globulin (g/100ml) concentration. Different letters denote significant differences ($p<0.05$) between groups. The data represented mean ± standard deviation. C: chicks fed basic diets. TF1: chicks fed basic diets mixed with 5% ground fennel seed and TF2: chicks fed basic diets mixed with 10% ground fennel seed.

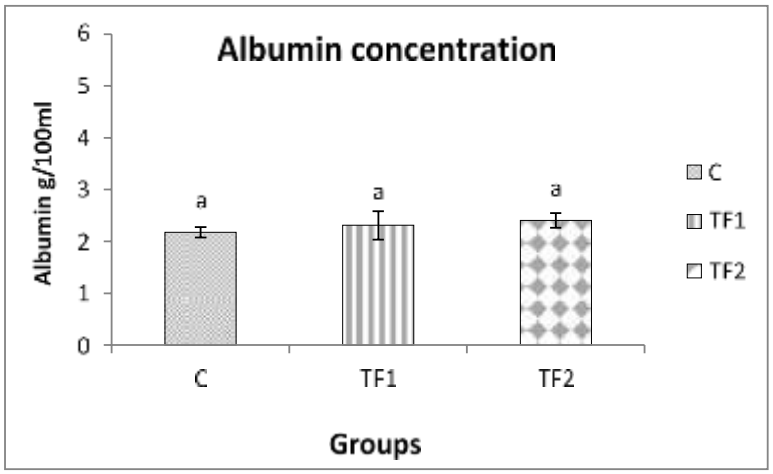

Figure 08- Effect of fennel seeds on serum albumin (g/100ml) concentration in broiler chicks. Different letters denote significant differences ($p<0.05$) between groups. The data represented mean ± standard deviation. C: chicks fed basic diets. TF1: chicks fed basic diets mixed with 5% ground fennel seed and TF2: chicks fed basic diets mixed with 10% ground fennel seed.

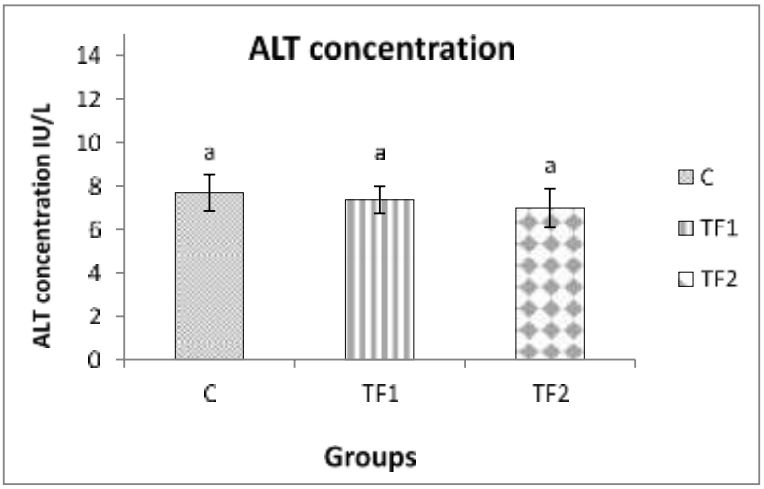

Figure 09- Effect of fennel seeds on serum alanine aminotransaminase, ALT (IU/L) concentration in broiler chicks. Different letters denote significant differences ($p<0.05$) between groups. The data represented mean ± standard deviation. C: chicks fed basic diets. TF1: chicks fed basic diets mixed with 5% ground fennel seed and TF2: chicks fed basic diets mixed with 10% ground fennel seed.

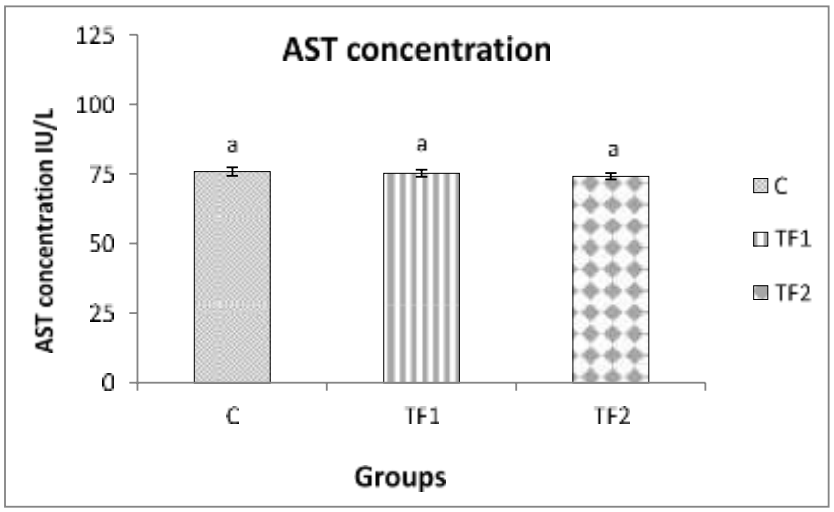

Figure 10- Effect of fennel seeds on serum asparatate aminotransaminase, AST (IU/L) concentration in broiler chicks. Different letters denote significant difference ($p<0.05$) between groups. The data represented mean ± standard deviation. C: chicks fed basic diets. TF1: chicks fed basic diets mixed with 5% ground fennel seed and TF2: chicks fed basic diets mixed with 10% ground fennel seed.

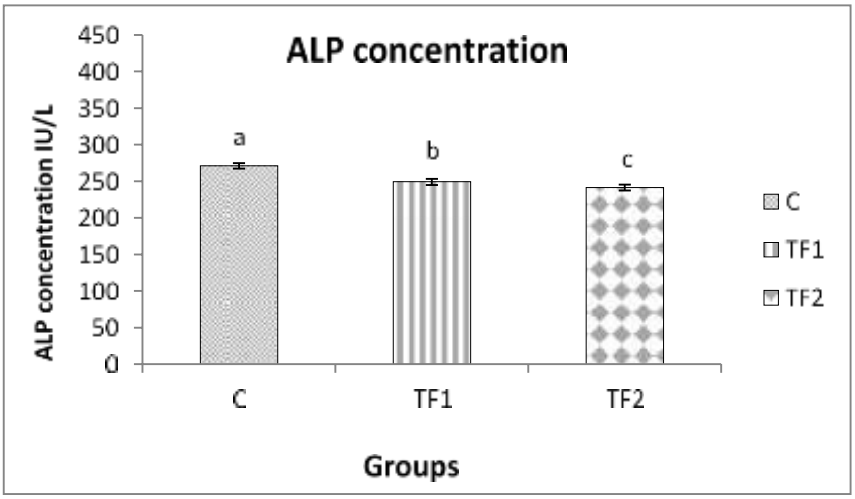

Figure 11- Effect of fennel seeds on serum Alkaline Phosphatase , ALP (IU/L) concentration in broiler chicks. Different letters denote significant differences ($p<0.05$) between groups. The data represented mean ± standard deviation. C: chicks fed basic diets. TF1: chicks fed basic diets mixed with 5% ground fennel seed and TF2: chicks fed basic diets mixed with 10% ground fennel seed.

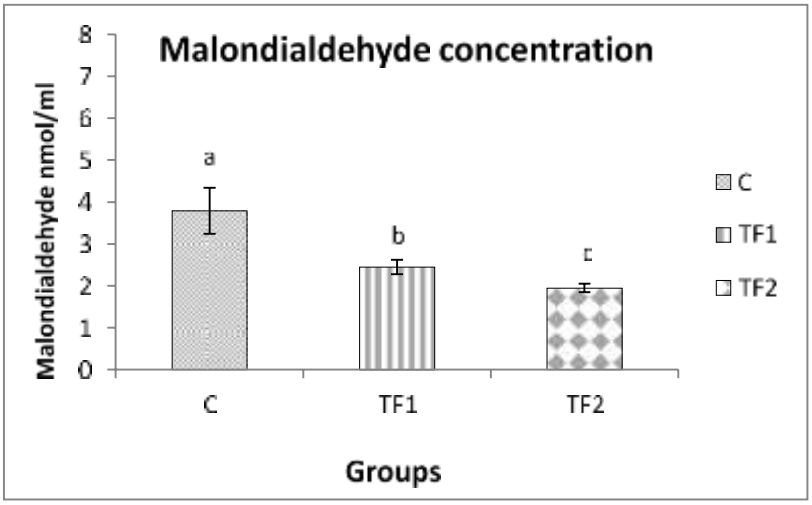

Figure 12 Effect of fennel seeds on serum malondialdehyde (nmol/ml) concentration in broiler chicks. Different letters denote significant differences ($p<0.05$) between groups. The data represented mean ± standard deviation. C: chicks fed basic diets. TF1: chicks fed basic diets mixed with 5% ground fennel seed and TF2: chicks fed basic diets mixed with 10% ground fennel seed.

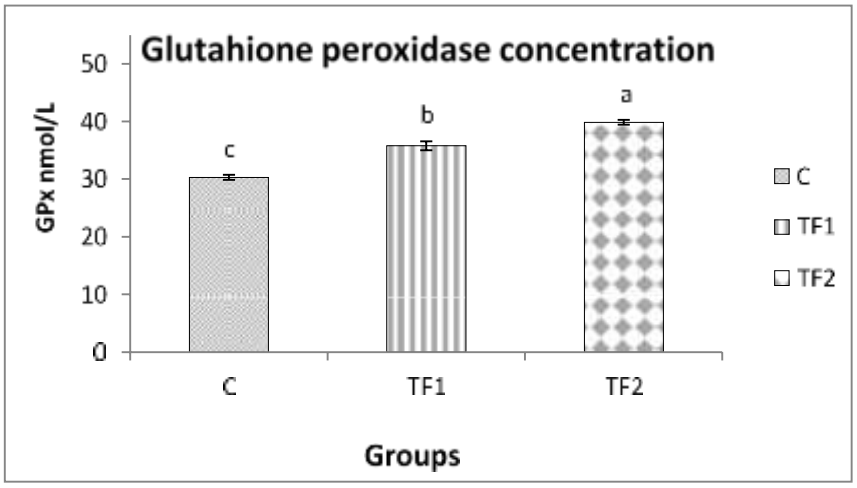

Figure 13- Effect of fennel seeds on serum Glutathione peroxidase, GSH-Px, (nmol/L) concentration in broiler chicks. Different letters denote significant difference ($p<0.05$) between groups. The data represented mean ± standard deviation. C: chicks fed basic diets. TF1: chicks fed basic diets mixed with 5% ground fennel seed and TF2: chicks fed basic diets mixed with 10% ground fennel seed.

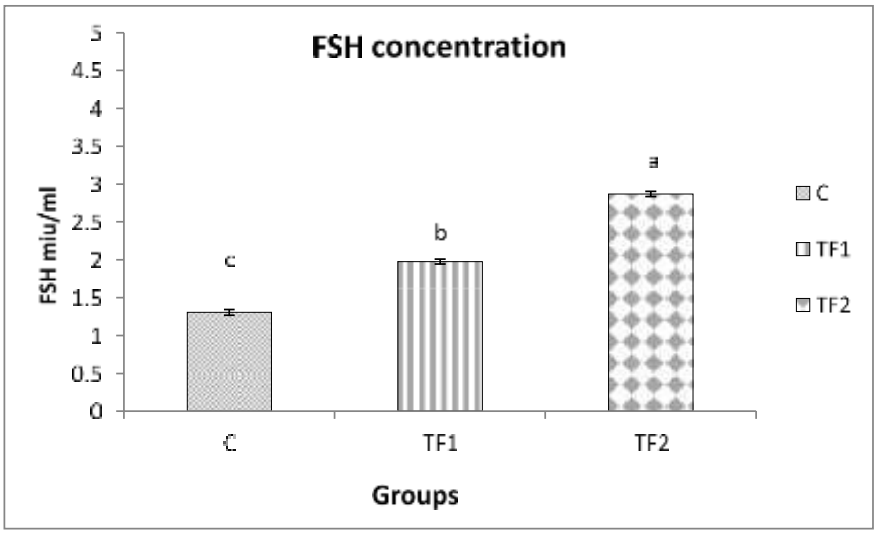

Figure 14- Effect of fennel seeds on serum FSH (miu/ml) concentration in broiler chicks. Different letters denote significant difference ($p<0.05$) between groups. The data represented mean ± standard deviation. C: chicks fed basic diets. TF1: chicks fed basic diets mixed with 5% ground fennel seed and TF2: chicks fed basic diets mixed with 10% ground fennel seed.

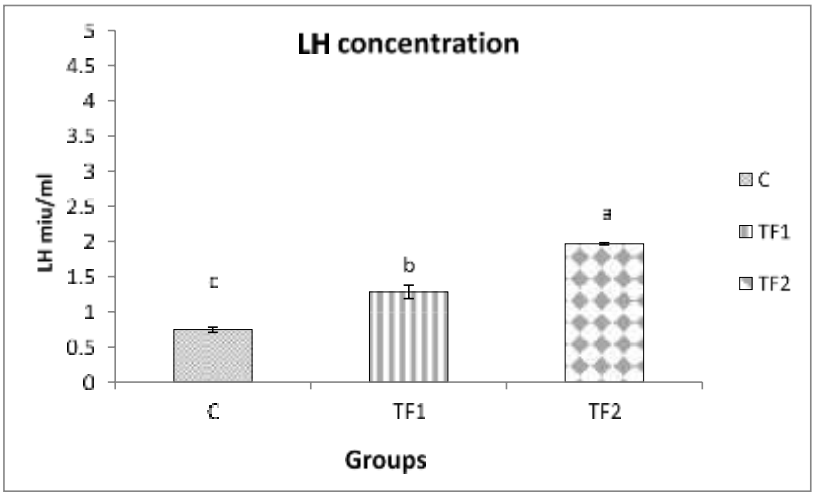

Figure 15- Effect of fennel seeds on serum LH (miu/ml) concentration in broiler chicks. Different letters denote significant difference ($p<0.05$) between groups. The data represented mean ± standard deviation. C: chicks fed basic diets. TF1: chicks fed basic diets mixed with 5% ground fennel seed and TF2: chicks fed basic diets mixed with 10% ground fennel seed.

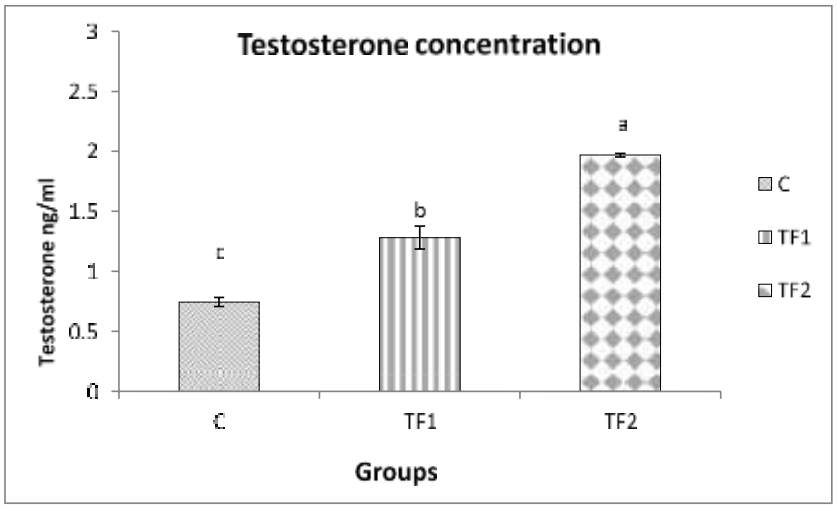

Figure 16- Effect of fennel seeds on serum testosterone (ng/ml) concentration in broiler chicks. Different letters denote significant difference ($p<0.05$) between groups. The data represented mean ± standard deviation. C: chicks fed basic diets. TF1: chicks fed basic diets mixed with 5% ground fennel seed and TF2: chicks fed basic diets mixed with 10% ground fennel seed.

Conclusion

The dietary supplementation of fennel seed powder at a 10 % level was beneficial to obtain higher body weight in broiler chicks. As well as supplementing with fennel seeds can improve the general health of chicks by increasing red blood cell count and PVC which in turn raises hemoglobin concentration in broiler chicks. Intake of fennel seeds at 5% and 10% could be enhanced immunity by encouraging white blood cells count and globulin concentration in chicks. Fennel seeds at 10% have hepatoprotective properties better than at 5%, that is by lowering the hepatic enzyme due to their antioxidant activity via elevation in GSH-Px and decline in malondialdehyde in broiler chicks, As well as, fennel seeds have the ability to improve the reproductive hormones of chocks that positively reflected on spermatogenesis. The fennel seed powder can be used as a natural photogenic growth promoter to enhance production performance by improving all studied parameters in chicks-fed fennel seeds at 10% positively reflected on economic agricultural production.

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Authors Contribution: Dr. Sura Safi Khafaji designed was designed the current work, conceptualized, reared the animals, supplemented the fennel seeds to animals and all analytical laboratory. Mohanad Obayes Abdullah Al-Jubouri was performed the data analysis and interpretation of data. Areej Gh. Hasan wrote the manuscript.

Conflict of interest: The authors declare that there is no concaflict of interests regarding the publication of this article.

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Checklist of Critical Thinking Process in Architectural Learning

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Rousl Hameed Razaq [4], Khalid abdul Wahab al Mudares [5], Omer Khalid Fayadh [6]

Abstract

Thinking is a mental activity that individuals engage in when faced with a situation or problem, attempting to find suitable solutions. It is an integral part of the human behavioral pattern that determines compatibility with the external environment. Critical thinking aims to shape the mind to judge ideas, perceptions, and other judgments to assess their logical consistency before adoption. This research sought to investigate the components of critical thinking in the educational process within the field of architectural engineering at two levels: the competence of curriculum implementation and the competence of the teaching staff across all academic stages. The research utilized theoretical conceptual frameworks that addressed the concept of critical thinking, its components, and criteria, considering its significance in the academic educational process and aligning it with quality performance standards at both levels. The findings revealed clear deficiencies in activating critical thinking components at the researched levels, as indicated by the comprehensive questionnaire included in the practical study and the identified indicators in the research results.

Key Words

Thinking; Critical Thinking; Architectural Engineering; Curriculum Implementation; Teaching Staff.

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Research Problem - "Lack of a clear understanding of the extent to which critical thinking components are realized in the educational effectiveness of the Department of Architectural Engineering at the University of Al-Nahrain."

Research Hypothesis - "The research assumes that by activating critical thinking components in educational effectiveness (in the curriculum and the performance of the teaching staff) ‹the deficiencies in this area in the Department of Architectural Engineering at the University of Al-Nahrain can be addressed."

Research Objective - "The research aims to investigate the extent to which critical thinking components are realized in the educational effectiveness of the Department of Architectural Engineering at the University of Al-Nahrain at two levels: first, formulating the curriculum vocabulary, and second, the performance of the teaching staff, for all subjects and academic stages."

Mechanism for Achieving the Research Objective –

- Study the concept of thinking as a mental process in its general form.
- Study the concept of critical thinking, its characteristics, components, and criteria specifically.
- Study the self-assessment according to the IBT standards for the curriculum vocabulary and all academic stages, as well as for faculty members and all academic stages in a selected department of architectural engineering.
- Align the performance evaluation criteria (specific to the curriculum on one hand and the teaching staff on the other) for the relevant department with the criteria and components of critical thinking.
- From this alignment, draw conclusions about the stance of the adopted curriculum and the teaching staff regarding the extent to which they achieve critical thinking standards and components in the results.

Introduction

Human beings distinguish themselves from other creatures by being entities driven to acquire knowledge to satisfy their material and spiritual needs. To attain knowledge, humans utilize their minds, engaging in various and multiple stages of deductive reasoning through sequential premises derived from their mental intuition. Ancient civilizations, recognizing the importance of thinking, invented rules and mental regulations called logic, which became

universal constants in human knowledge. This is because humans often fail to reach knowledge that reveals reality, or they may construct knowledge that aligns with their desires and interests, leading to errors in acquiring or using knowledge correctly.

As rational beings, human life patterns are shaped by the type of cognitive movement they adopt and their reasoning methods. Changing many of the errors we experience and transforming our reality into a healthier one heavily relies on uncovering the thinking style we employ. If our reality is bleak and stagnant, it indicates that we are thinking incorrectly, and we must change our cognitive and reasoning methods to alter this backward reality.

Many things in life may appear sound and free of flaws on the surface, prompting us to shape our lives and subsequent actions based on these perceptions. However, these ideas may be nothing more than errors we have become accustomed to over time until stagnation, inertia, and backwardness take over. Continuously adhering to error leads to the creation of successive accumulations of complex errors, posing a methodological problem that individuals, groups, or societies must consider. The crucial lesson here is that intellectual methodology determines the course of an individual, a nation, and the vitality of a nation is linked to the quality of the thinking it follows. Many nations have disappeared and perished when immersed in stagnant swamps of rigid ideas, while others have risen with the revival of their life-giving and spirited thoughts by adopting a sound approach in their intellectual system.

Concept of Thinking*:*

1. **Thinking Process**

Thinking is a continuous mental process performed by the brain when exposed to stimuli received through one or more of the five senses: touch, sight, hearing, smell, and taste. In its broadest sense, thinking is a series of mental activities that the brain engages in, such as problem-solving, comprehension, application, deduction, and directing and controlling cognitive activities. According to Barell (1991), thinking requires deep contemplation and a thorough examination of the components of a situation or experience.

Based on these definitions, it is clear that the elements of the thinking process include complex cognitive operations (such as problem-solving), less complex operations (like comprehension, application, and deduction), and directive and controlling operations over cognitive knowledge. Additionally, thinking involves specific knowledge about the content of the subject.

2. Concept of Critical Thinking

The term "critique" in Arabic means to distinguish coins by separating genuine ones from counterfeit, and it also means to reveal the flaws or merits in poetry or prose. In the context of art criticism, it refers to evaluating the artistic work, distinguishing between its good and bad aspects and verifying its accuracy or falsehood.

Critical thinking is considered one of the most complex forms of thinking, given its connection to various behaviors such as logic, problem-solving, and its close association with abstract and contemplative thinking in terms of shared characteristics. Scholars have shown clear interest in this type of thinking due to its implications for the learning process and problem-solving ability. Critical thinking has gained prominence in various fields of education and scientific research, from pre-school to higher education and beyond.

3. Critical Thinking

Critical thinking is a form of high-order thinking that requires the use of advanced thinking skills or components. High-order thinking is a combination of skills akin to creative thinking, as critical and creative thinking are considered together (Libman, 1994:103-113).

Critical thinking involves "examining and evaluating proposed solutions" and is described as "reflective, reasonable thinking focused on decision-making in what is being thought or done." It is the process of using the rules of logical inference, avoiding common errors in judgment. Critical thinking can also be defined as the thinking that "relies on analysis, sorting, choosing, and testing of an individual's information with the aim of distinguishing between sound and erroneous ideas".

Based on the above, critical thinking aims to arrive at the truth after eliminating doubt by studying logical evidence and available facts, involving careful examination and scrutiny.

The relationship between critical thinking and creative thinking is very strong. The presence of critical thinking is a requirement for the ability to think creatively. Critical thinking includes creative thinking as it involves formulating hypotheses, asking questions, testing, and planning experiments. Practicing creativity employs critical thinking to compare the solutions obtained to choose the most suitable and appropriate solution for the presented problem. Critical thinking challenges the ideas and actions of others, supporting them with evidence, logical reasoning, without bias, and refusing to follow others blindly without thought.

4. Definition of Critical Thinking

Numerous definitions of critical thinking have emerged due to the multitude of perspectives and various theories that have addressed critical thinking. Although most scholars agree that critical thinking is a form of thinking that allows individuals to engage in logical and realistic thinking, there is variation when it comes to defining the characteristics of critical thinking and its relationship to other forms of thinking, such as creative, contemplative, and abstract thinking.

The linguistic connotation of the word "critic" in Greek is considered to be the same traditional view of thinking that laid its foundations and was adopted by the three philosophers Socrates, Plato, and Aristotle. This view holds that analytical skills, judgment, and argumentation are sufficient to reach the truth. Returning to the English word "Critical," we find that it is derived from the Latin "Criti-cus" or the Greek "Kritikos," which simply means the ability to distinguish or pass judgments (Davis, 2004:61).

Critical thinking is contemplative thinking governed by the rules of logic and analysis. It is a result of various cognitive aspects such as knowledge of assumptions, interpretation, discussion evaluation, induction, deduction, and critical thinking is an evaluative process that uses the rules of logical inference in dealing with variables. It is a complex mental process consisting of skills and inclinations. The critical thinker relies on a detailed examination of the available information based on the rules of logic gradually to achieve accurate and precise results.

Critical thinking is one of the most complex forms of thinking, and it has captured the interest of researchers and thinkers who are known for their writings in the field of thinking. The expression "critical thinking" is often misused by many to describe thinking processes and skills. In reality, the term is used to indicate new meanings, including revealing flaws and errors, skepticism, analytical thinking, contemplative thinking, problem-solving, clear thinking, alert thinking, independent thinking, and identifying biases, contradictions, and inconsistencies.

4.1 Common Grounds in Definitions of Critical Thinking

Despite apparent differences in the treatments of the concept of critical thinking by many authors, there are common elements among them that can be summarized as follows:

1. **Not Synonymous with Decision-Making or Problem Solving:** Critical thinking is not synonymous with decision-making or problem-solving. It goes beyond mere recall or

summoning of information and is not bound by following an organized strategy for handling a situation.

2. **Begins with a Claim or Inference:** Critical thinking begins with a claim, inference, information, or a specific theory. The central question is, "What is the value or validity of this thing?" In contrast, problem-solving starts with a problem, and the central question is, "How can it be solved"?
3. **Not a Sequential Strategy:** Critical thinking is not a strategy like problem-solving or decision-making. It does not consist of a series of processes and methods that can be used sequentially to address a situation.
4. **Two Formulations:** Two formulations can organize the diverse definitions of critical thinking:
    a. Personal and Self-focused: Focuses on the individual's personal goals behind critical thinking. It is contemplative, reasonable thinking that aims to develop and control one's thinking for clarity, accuracy, and advocacy.
    b. Social Aspect: Emphasizes the social aspect of critical thinking, involving mental processes when asked to judge an issue, discuss a topic, or conduct an evaluation. It helps achieve accuracy in making objective judgments about others' opinions and beliefs.

4.2 Critical Thinking as a Complex Concept

Critical thinking is a complex concept with connections to an unlimited number of behaviors in various situations and contexts. It intertwines with other concepts such as logic, problem-solving, learning, and the theory of knowledge. According to Mayer, John Dewey expresses the essence of critical thinking in his book "How We Think" by saying: "It is deliberation in giving judgments and suspending them until verification". Some see critical thinking as corresponding to abstract thinking according to Piaget (Ward Zorth, 1990, p. 17), consisting of three mechanisms:

- **Crafting Generalizations with Caution:** Being cautious in formulating generalizations.
- **Considering Possibilities and Alternatives:** Reflecting on possibilities and alternatives.
- **Withholding Judgment**: Postponing judgment about something until sufficient information and evidence are available.

5. Criteria for Critical Thinking

Criteria for critical thinking refer to the general specifications agreed upon by researchers in the field of thinking. These criteria serve as guidelines for both researchers and evaluators, students, and instructors. Some of the prominent criteria presented by Elder and Paul (2001:40) are:

1. **Clarity:** The expression should be clear, allowing for accurate understanding through detailed explanations and examples.
2. **Accuracy**: Statements should exhibit a high degree of accuracy and reliability, supported by evidence and proof.
3. **Precision:** Expressing the subject matter with a high degree of precision, specificity, and detail.
4. **Relevance:** Elements of the problem or situation should exhibit a high degree of clarity in the interconnection between elements or between data and the problem.
5. **Depth** Processing the problem or phenomenon should involve a high degree of depth in thinking, interpreting, and predicting to move beyond surface-level treatment.
6. **Breadth:** Taking all aspects of the problem or situation comprehensively and broadly, considering different perspectives and ways of dealing with the problem.
7. **Logic:** Critical explanations should be logical, organizing thoughts and their interconnections in a way that leads to clear and specific meanings.
8. **Significance**: Recognizing the importance of the problem or situation compared to other problems and situations encountered by the individual.
9. These criteria collectively contribute to distinguishing between elements related to the problem and those unrelated to it, ensuring clarity, precision, relevance, depth, breadth, logic, and significance in the critical thinking process.

6. Critical Thinking Skills

There are several classifications of critical thinking skills based on the diversity of definitions and theoretical frameworks that explain them. Among the most famous classifications are:

Firstly, the classification by Dunlosky and Grasserhacker, which divides critical thinking skills into the following (Hacker, Dunlosky, Grasser, 1998:38):

1. Recognizing Assumptions: Refers to the ability to distinguish between the accuracy of specific information and its falsehood. It involves differentiating between facts and opinions and understanding the purpose of given information.
2. Interpretation: Involves the ability to identify problems, recognize logical interpretations, and assess whether generalizations and conclusions based on specific information are acceptable.
3. Inference: Refers to the individual's ability to identify some of the outcomes resulting from premises or previous information.
4. Deduction: Refers to the individual's ability to extract a conclusion from specific observed or assumed facts, and to assess the validity of the result in light of the given facts.
5. Argument Evaluation: Means the ability to evaluate an idea, accept or reject it, distinguish between primary and secondary sources, strong and weak arguments, and make judgments about the sufficiency of information.

Secondly, the Facione classification (Facione, 1998:29) explains that critical thinking consists of the following basic cognitive skills:

1. Interpretation: Comprehending and expressing a broad range of situations, data, experiences, rules, standards, and procedures. It includes sub-skills such as classification, extracting meaning, and clarification.
2. Analysis: Refers to identifying inductive and deductive relationships between statements, questions, concepts, and attributes. It has sub-skills like examining opinions and discovering and analyzing arguments.
3. Evaluation: Involves assessing the credibility of statements or a person's perception (experience, qualification, judgment, belief, and opinion). It includes skills in evaluating claims and arguments.
4. Inference: Identifying the elements necessary to draw reasonable conclusions. Sub-skills include examining evidence, guessing alternatives, and reaching inferences.
5. Explanation: Declaring the results of thinking and justifying them in light of evidence, concepts, measurement, context, and persuasive arguments. Sub-skills for explanation include declaring results, justifying actions, and presenting arguments.
6. Self-Regulation: Defined as an individual's ability to question, ensure credibility, organize thoughts, and results. It involves the sub-skills of self-testing and self-organization.

Thirdly, Beyer (Beyer, 1999:84) specifies ten main critical thinking skills:

1. Distinguishing between proven facts and claims made by others.
2. Distinguishing between objective and random evidence related to claims.
3. Ability to determine the credibility of news or narration.
4. Identifying or verifying the credibility of the news source or information source.
5. Distinguishing between provable facts or verifiable truths and claims, self-proclaimed statements, or values.
6. Differentiating vague claims and evidence from objectivity or recognizing ambiguous claims, evidence, and arguments.
7. Ability to assess the bias of others.
8. Recognizing seemingly logical fallacies.
9. Identifying implicit assumptions in the text.
10. Recognizing inconsistencies or inconsistencies in the course of the inference process from premises or facts.

Fourthly, Stevens (Stevens, 1990:32) believes that critical thinking skills require the ability to accomplish the following:

1. Formulating and defining hypotheses.
2. Deducting and extracting information.
3. Distinguishing between fact, opinion, and assertion.
4. Differentiating between information related to the subject and unrelated information.
5. Recognizing logical inconsistencies (fallacies).
6. Determining the accuracy of information and comprehending it, exercising caution in judging it.
7. Identifying vague or intertwined claims and arguments.
8. Predicting decision or solution outcomes.
9. Reporting the difficulty of evidence.
10. Determining the strength of a specific argument or evidence.

Fifthly, Wilberg (Wilberg, 1995, p. 45) identified twelve skills that enable researchers to practice critical thinking. These skills include:

1. Grasping all aspects of the presented issue and understanding its substance by giving equal importance to all aspects of the position.

2. Testing the results reached.
3. Recognizing contradictions in statements and clearly identifying the issue.
4. Providing justifications for the result reached.
5. The ability to judge whether something is an assumption.
6. Formulating statements acceptably.
7. Attempting to separate emotional from logical thinking.
8. Being deliberate in issuing judgments.
9. Taking a stance and changing it when evidence is available.
10. Knowing the difference between a result that "might be true" and a result that "must be true".
11. Objectivity and distancing oneself from personal factors.
12. Knowing that people have different ideas about the meanings of vocabulary terms.
13. Knowing when one needs new information about something.

Sixthly, Edward de Bono (1989:19) formulated twelve skills for developing critical thinking. These abilities include:

1. Identifying central problems and issues, contributing to the main parts of arguments and evidence.
2. Identifying information related to the subject, capable of making comparisons between provable or verifiable matters, and distinguishing basic information from less relevant information.
3. Distinguishing similarities and differences, contributing to the ability to identify distinctive parts and categorize information for different purposes.
4. Formulating questions that contribute to a deeper understanding of the problem.
5. Providing a standard for judging the quality of observations and conclusions.
6. Determining whether phrases or symbols are connected with each other and with the overall context.
7. Achieving implicit issues not explicitly evident in the argument and evidence.
8. Distinguishing recurring formulas.
9. Determining the reliability of sources.
10. Distinguishing different trends and perceptions of a specific situation.
11. Assessing the data's capacity, sufficiency, and quality in addressing the subject.
12. Anticipating possible or likely outcomes of an event or set of events.

Seventhly, Al-Harthi (1999:7) identifies six integrated strategies for critical thinking:

1. Understanding the idea, thing, event, or action and determining its meaning.
2. Identifying causes and factors.
3. Knowing the purposes or objectives it aims to achieve.
4. Evaluating by knowing the criteria and adhering to their application.
5. Understanding future consequences and sequences built on that idea, thought, or event.

Through the presentation of critical thinking skills, we find that most of them are repeated in these classifications. Therefore, they can be summarized as follows:

1. Assumptions: Recognizing the degree of truth in information, facts, fallacies, formulating hypotheses, and making predictions.
2. Interpretation and Understanding: The ability to identify, understand, and explain the problem, recognize logical interpretations, practice classification, extract meaning from data, and determine the accuracy of information and its sources.
3. Logic: Utilizing deductive and inductive reasoning and various forms of inference.
4. Evaluation: Assessing arguments, evidence, claims, and determining their strength and refuting them. Distinguishing between facts and claims.

7. **Procedural Elements of Critical Thinking*:***

Some thinkers, such as Al-Jabri (1976:42), have identified the practical procedural characteristics of critical thinking as follows:

1. Knowledge of assumptions.
2. Interpretation.
3. Evaluation of discussions.
4. Deduction.
5. Assessment.

Additionally, Al-Jabri (1976:42) outlined the goals of curricula that develop critical thinking, categorizing them into capabilities and capacities. He assumed that critical thinking includes three aspects:

1. Identifying logical research methods contributing to determining values and weighing different types of evidence that contribute to acceptable results.
2. The need for evidence and support for opinions and results before judging their reliability.

3. Skills in using all previous orientations and skills.

Table 01- illustrates the components and criteria of critical thinking accordingto the previous literary sources - Part One of the sources.

Components, standards, or characteristics of critical thinking according to sources (previous literature). Part One/			
(1)	***)2(***	**(3)**	***)4(***
Elder & Paul ,2001:40	**hacker,Dunlosky ,Grasser,1998:38**	**Facione,1998:29**	**Beyer**
1/ Clarity	The ability to recognize assumptions.	1/Interpretation	Distinguishing between provable facts and claims made by others.
2/ Accurac	The ability to interpret.	2/Analysis	Differentiating between evidence and objective or random pieces of information related to claims.
3/ Precision	The ability to deduce.	3/Evaluation	The ability to determine the credibility of news or a narrative.
4/ Depth	The ability to infer.	4/ Inference	Identifying or verifying the credibility of the source of news or information. Distinguishing between facts that can be proven or verified and claims, self-assertions,
5/Relevance	The ability to evaluate arguments.	5/ Explanation	The ability to assess the degree of bias in others or to detect bias or prejudice.
6/ Breadth	The ability to recognize assumptions.	6/ Self — Regulation	The ability to identify logical fallacies.
7/Logic	The ability to interpret.		Recognizing implicit assumptions in the text.
8/ Significance			Identifying inconsistencies or lack of coherence in the process of inference from premises or facts.
			Assessing the strength of evidence, proof, or claims. Distinguishing between information and claims and between reasons related to the subject and those that are irrelevant."

Table 01- illustrates the components and standards of critical thinking according to previous literary sources – the second section of the sources. (Source / Researchers).

(5)	(6)	(7)	(8)
(Stevens, 1990:32)	**(Wilberg, 1995, p. 45)**	**(De Bono, 1989, p. 19)**	**(Al-Harithi, 1999, p. 7)**
Formulating hypotheses and defining them.	Surrounds the aspects of the presented issue and understands its essence, considering all aspects of the situation equally important.	The ability to identify central problems and issues, contributing to the main parts of the argument and evidence.	Understanding the idea, thing, event, or action and defining its meaning.
Deduction and extraction of information.	Can test the results that are reached.	The ability to identify information related to the subject, which has the capacity to make comparisons between matters that can be proven or verified, distinguishing basic information from less relevant information.	Recognizing the causes and factors.
Differentiating between fact, opinion, and claims.	Recognizes contradictions in statements and clearly defines the issue.	The ability to distinguish similarities and differences, contributing to the ability to identify distinct parts and categorize information for various purposes.	Understanding the purposes or goals aimed at.
Distinguishing between information related to the subject and unrelated information.	Provides justifications for the reached conclusion.	The ability to formulate questions that contribute to a deeper understanding of the problem.	The ability to assess through knowledge of standards and committing to their application.
Recognizing logical contradictions (fallacies).	Has the ability to judge whether something is an assumption.	The ability to provide a standard for judging the quality of observations and conclusions.	Understanding future consequences and the sequences built upon that idea, thing, or event.
Assessing the accuracy of information, comprehending it, and exercising caution in judgment.	Formulates expressions in an acceptable manner.	The ability to determine whether phrases or symbols are connected to each other and to the general context.	(8) (Al-Harithi, 1999, p. 7)
Identifying vague and overlapping claims or arguments.	Attempts to separate emotional thinking from logical thinking.	The ability to address implicit issues that did not explicitly appear in the argument and evidence.	
Anticipating the outcomes of decisions or solutions.	Exercises caution in making judgments.	The ability to distinguish recurring patterns.	Understanding the idea, thing, event, or action and defining its meaning.
Assessing the difficulty of evidence.	Takes a position and changes it when evidence is available.	The ability to assess the reliability of sources.	Recognizing the causes and factors.
Determining the strength of a specific argument or claim.	Knows the difference between a result that "might be true" and a result that "must be true."	The ability to distinguish different trends and perceptions regarding a specific situation.	Understanding the purposes or goals aimed at.

	Objectivity and distancing from personal factors.	The ability to determine the capacity, sufficiency, and quality of data in addressing the subject.	The ability to assess through knowledge of standards and committing to their application.
	Recognizes that people have different ideas about the meanings of vocabulary.	The ability to anticipate possible or probable outcomes of an event or a set of events.	Understanding future consequences and the sequences built upon that idea, thing, or event.
	Knows when new information is needed about something.		

For the purpose of studying the extent to which critical thinking standards or components were applied and used in a real case, the standards used to evaluate the self-performance of the Department of Architectural Engineering at the University of the Tigris for the academic year 2011-2012 were utilized. This was based on the standards set by ABET for the curriculum of each course in the Architectural Engineering Department at the University of the Tigris. The same standards were also used to evaluate the performance of the faculty members in the same institution and for the same year. These evaluations were conducted for students at all academic levels, and the results were documented in Tables (5) and (6) and the accompanying graphs.

After aligning the critical thinking standards and skills with the criteria used to determine the quality of teaching each course in the Architectural Engineering Department, as well as students' perceptions of those criteria (whether achieved or not, or somewhere in between) for all academic levels, and similarly for those used to determine the quality of the curriculum for all subjects and all academic levels, the results showed gaps in activating critical thinking skills during class sessions.

The results were analyzed in detail, providing an accurate interpretation of the correlation between the failure to achieve critical thinking skills and the stance of the academic curriculum, as well as the position of faculty members.

The scientific study was conducted on results obtained from two matrices documented in Tables (5) and (6), along with graphical representations. These results were derived from a general questionnaire that included all students in the Department of Architectural Engineering at the University of the Tigris, spanning all academic levels. The study aimed to investigate the realization of critical thinking components in the educational process within the specialization of architectural engineering.

Table 03- illustrates the correlation between critical thinking components and the criteria for evaluating academic curriculum performance. Source/Researcher: (Utilizing curriculum components from the source: Aljumaily, Al-Ukaily, 2011).

	Criteria for Assessing the Curriculum Vocabulary	Critical Thinking Criteria
1	Overall, this Curriculum subject is good and useful.	To achieve the standards listed to the left in this table, we find that they can be derived from all the criteria of critical thinking, which have been gathered in various previous studies. The analysis of the questionnaire results illustrates this.
2	Lecture time is sufficient to cover the contents of the article.	
3	The content of article commensurate with the objective of Curriculum.	
4	Subject content is anointer dependent information.	
5	Text books and references are available and meaningful.	
6	available of References helpful for stimulate and thinking.	
7	The book is free of grammatical errors Printing.	
8	Contents of the book are of outdated information.	
9	The book contains a variety of examples and exercises	
10	The evaluation of the subject system is appropriate (test method).	
11	Exams reflect the content of the subject.	
12	Number of exams be exhaustive of the content subject.	
13	Examinations and assignments helped to absorb the subject.	
14	Examinations and exercises are in line with the objectives of the subject.	
15	Examinations and exercises help to think of more conservation.	
16	Number of exams and the their recurrence are appropriate.	
17	The case of equipped lecture hall satisfactory.	

Table 04- illustrates the alignment between the criteria of critical thinking and the criteria for evaluating the performance of the teaching staff. Source/Researcher (By relying on the curriculum criteria from the source: Aljumaily, Al-Ukaily, 2011).

	Criteria for Evaluating Teaching Staff	Critical Thinking Criteria
1	Has the ability to communicate scientific material in a smooth and easy manner.	To achieve the standards listed on the left side of this table, it can be deduced from each of the critical thinking criteria, which have been gathered in all previous studies. The analysis of the questionnaire results illustrates that
2	Keep to use the tools and techniques of modern education.	
3	Illustrates the theoretical aspects in the subject with examples from the reality.	
4	Gives the scientific material in a manner covering the time of the lecture.	
5	Committed to the dates of lectures.	
6	Improve in the management ranks and give equal opportunities to students in dialogue and discussion.	
7	Motivates students and encourages them to think and research.	
8	Respects the different views of the students.	
9	Through self-learning encourages students to search for what is new and modern.	
10	Accept criticism and suggestions with an open mind.	
11	Be objective and fair in his / her evaluation of students.	
12	Uses a variety of methods to assess the performance of students(such as reports, research, and quizzes(.	
13	Follow up activities and duties to put the evaluation weights.	
14	Has the ability to discuss all issues of the subject	
15	Working to increase the knowledge of the outcome requested.	

Table 05- illustrates the results of the survey conducted with students from all academic stages regarding the performance of the educational curricula. (Source / (Aljumaily, Al-Ukaily, 2011)

Score		1	2	3	4	5
No.	**Question**	**Strongly Agree**	**Agree**	**I Don't Know**	**Disagree**	**I Don't Agree At All**
1	Overall, this Curriculum subject is good and useful.	**300**	**216**	**9**	**23**	**12**
2	Lecture time is sufficient to cover the contents of the article.	**134**	**231**	**35**	**75**	**32**
3	The content of article commensurate with the objective of Curriculum.	**137**	**254**	**42**	**57**	**12**
4	Subject content is anointer dependent information.	**136**	**250**	**47**	**44**	**18**
5	Text books and references are available and meaningful.	**83**	**216**	**75**	**91**	**48**
6	Available of References helpful for stimulate and thinking.	**106**	**239**	**90**	**67**	**16**
7	The book is free of grammatical errors Printing.	**74**	**188**	**192**	**43**	**19**
8	Contents of the book are of outdated information.	**63**	**161**	**201**	**77**	**20**
9	The book contains a variety of examples and exercises.	**64**	**196**	**168**	**55**	**21**
10	The evaluation of the subject system is appropriate (test method).	**93**	**262**	**77**	**49**	**19**
11	Exams reflect the content of the subject.	**121**	**255**	**51**	**53**	**9**
12	Number of exams is exhaustive of the content subject.	**126**	**263**	**41**	**57**	**14**
13	Examinations and assignments helped to absorb the subject.	**110**	**287**	**35**	**51**	**19**
14	Examinations and exercises are in line with the objectives of the subject.	**116**	**293**	**34**	**32**	**24**
15	Examinations and exercises help to think of more conservation.	**120**	**297**	**22**	**42**	**26**
16	Number of exams and the their recurrence are appropriate	**114**	**273**	**35**	**42**	**30**
17	The case of equipped lecture hall satisfactory.	**113**	**269**	**25**	**76**	**30**
18	Capabilities and laboratories are appropriate and effective.	**100**	**246**	**22**	**74**	**45**

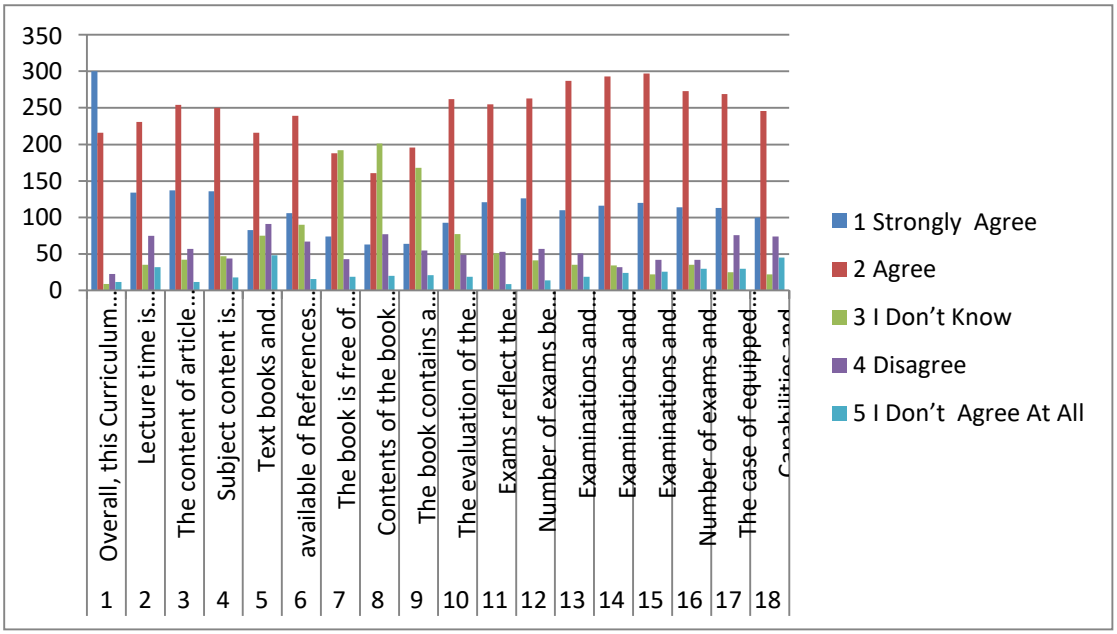
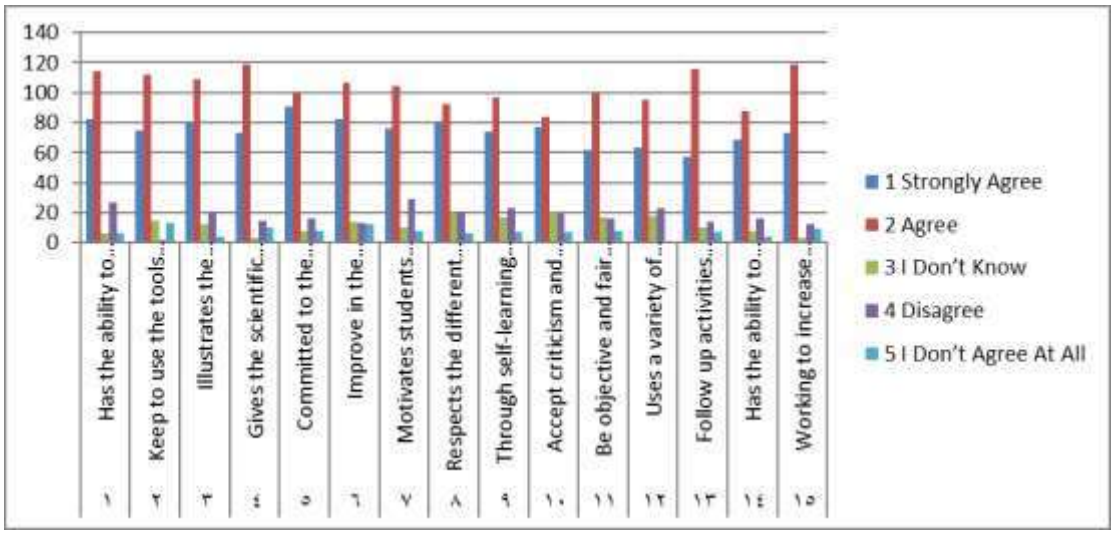


Chart 01- illustrates the evaluation results of the students of the Department of Architecture - University of al-Nahrain for the curricula of their academic terms at all stages. The chart is a graphical representation of the information presented in Table No. (5) above. (Source / Aljumaily, Al-Ukaily, 2011)

Table 06- illustrates the survey results of students from all academic stages regarding the performance of their teaching staff. (Source / Aljumaily, Al-Ukaily, 2011).

Score		1	2	3	4	5
	Question	Strongly Agree	Agree	I Don't Know	Disagree	I Don't Agree At All
1	Has the ability to communicate scientific material in a smooth and easy manner.	82	114	6	27	6
2	Keep to use the tools and techniques of modern education.	75	112	15	2	13
3	Illustrates the theoretical aspects in the subject with examples from the reality.	81	109	12	21	4
4	Gives the scientific material in a manner covering the time of the lecture.	73	119	3	15	10
5	Committed to the dates of lectures.	91	100	8	16	8
6	Improve in the management ranks and give equal opportunities to students in dialogue and discussion.	82	107	14	13	12
7	Motivates students and encourages them to think and research.	76	104	10	29	8
8	Respects the different views of the students.	80	92	20	21	6
9	Through self-learning encourages students to search for what is new and modern.	74	97	17	23	7
10	Accept criticism and suggestions with an open mind.	77	84	20	20	7
11	Be objective and fair in his / her evaluation of students.	62	100	17	16	8
12	Uses a variety of methods to assess the performance of students(such as reports, research, and quizzes(,	63	95	18	23	
13	Follow up activities and duties to put the evaluation weights.	57	116	10	14	7
14	Has the ability to discuss all issues of the subject.	69	88	8	16	4
15	Working to increase the knowledge of the outcome requested.	73	119	3	12	9



Graph 02- illustrates the results of the evaluation of the teaching staff by students of the Department of Architecture at Al-Nahrain University for all academic stages. The graph represents a graphical translation of the information presented in Table No. (6) above. (Source / Aljumaily, Al-Ukaily, 2011)

Results

Considering the abundance of result sections that can be derived from the questionnaire conducted on students of all academic stages regarding the evaluation of their curriculum content and the performance of the teaching staff, these results have been summarized to address the main topics of the research and its objectives as follows:

Regarding the evaluation of the curriculum designed for each academic unit at that academic stage, the assessment is conducted through the following questions:

Regarding the evaluation of the curriculum designed for each academic subject in that academic stage, the assessment is based on the following questions:

a. Is the overall subject matter of the curriculum good and useful?

- Out of 560 responses (covering all subjects and academic stages), 44 responses indicated uncertainty about the quality and usefulness of the curriculum, constituting 8% of the total survey responses. This is an indicator of a failure to activate critical thinking skills that would allow students to identify assumptions, interpretations, deductions, and conclusions accurately.

b. Is the lecture time sufficient to cover the lecture topic?

- Out of 507 responses, 142 indicated either a lack of awareness of the importance of lecture time or deemed it insufficient or entirely inadequate, representing over 28% of survey responses. These results indicate a failure to activate critical thinking and

undermine the ability to identify central problems and issues, hindering the efficient delivery of course material.

c. Does the lecture content align with the educational or cognitive objective of the curriculum for that academic subject?

- Out of 502 responses, 111 indicated a lack of awareness of the educational objective, and others decisively claimed that the lecture content does not align with the educational objective, constituting 22% of the total survey responses. This points to a failure to activate critical thinking skills in students, hindering their ability to identify if statements or symbols are connected within the general context and the overall purpose of the topic.

d. Does the lecture content provide independent and specialized information?

- Out of 495 responses, 109 stated a lack of awareness of the importance of having specialized information related to the academic subject, or claimed that such information is absent or entirely lacking, accounting for 22% of the total survey responses. This reflects a lack of awareness among students regarding the necessity of specialized information supporting the aspects of the academic subject.

e. Are there instructional books or references available to support and enhance the curriculum?

- Out of 513 responses, 214 indicated a lack of awareness of the need for instructional books or references to support the curriculum, claiming their absence decisively, totaling 42% of the survey responses. This is another indicator of the failure to activate critical thinking skills that should expand students' knowledge base and understanding of the purposes or objectives they aim for.

f. Are the assigned textbooks free from printing errors and grammatical mistakes?

- Out of 519 responses, 254 indicated a lack of awareness or knowledge of whether there are printing errors in the assigned textbooks, while others decisively claimed the presence of printing errors, constituting 49% of the total survey responses. This paragraph does not address critical thinking elements as it concerns mechanical printing errors.

g. Are the contents of the textbook for that academic subject in the curriculum outdated?

- Out of 522 responses, 298 were uncertain whether the given information was old or recent, while others claimed that the information in the textbook is outdated, totaling 57% of the total survey responses. This section highlights the outdatedness of the information and its lack of integration with recent developments, contradicting the activation of critical thinking elements, such as enhancing the student's ability to distinguish between repeated formulas, determine the reliability of sources, and identify the strength of a specific argument or claim.

Secondly, regarding the evaluation of the performance of the lecturer responsible for that academic subject in the curriculum, the assessment is based on the following questions:

1. **Does the lecturer have the ability to convey the scientific material in an easy and simple manner?**

- Out of 235 responses, 39 indicated uncertainty about whether the lecturer conveyed the material in an easy and simple manner, while others claimed failure on the part of the lecturer. A total of 16% of the responses acknowledged this, serving as an indicator of a lack of critical thinking skills in delivering the lesson due to poor explanation and inadequate comprehension, especially given the modest academic level of the majority of students, resulting in deficiencies in achieving general critical thinking standards mentioned in Tables (1) and (2) in the text.

2. **Did the lecturer use tools and techniques specific to modern education?**

- Out of 213 responses, 30 stated they were unsure whether the lecturer used modern techniques in teaching, while others claimed that the lecturer did not or decisively stated a failure in using modern techniques in presenting the lecture. This accounts for 14% of the total responses. These results contribute to weakening the overall critical thinking skills of students and reducing their ability to improve their educational performance.

3. **Does the lecturer represent the theoretical concepts he addresses in his subject with real-life examples?**

- Out of 227 responses, 37 indicated they did not know whether the lecturer represented the theoretical concepts he invoked in his lecture with real-life examples, while others claimed he did not or decisively stated that he did not. This constitutes 16% of the total responses. This can undermine many critical thinking skills; for example, students may struggle to deduce and extract information, recognize contradictions in statements, clearly identify the issue, distinguish between fact and opinion or assertion, recognize

inconsistencies in the process of inference, and verify the credibility of the source of news or information.

In summary, the above results point to significant challenges and weaknesses in the activation of critical thinking skills among students, impacting their learning experience and overall academic performance.

4. **Does the lecturer cover the scientific material of the academic subject within the allotted time for the lecture or otherwise?**

- Out of 220 survey responses, 28 participants stated that they didn't know if the lecturer covers the scientific material during the entire lecture time or finishes before time or needs more time. Another group indicated that the lecturer does not adhere to the allotted time for the lecture (either exceeding or falling short), with a total of 12% of the responses. This certainly affects critical thinking elements in both cases of extended or shortened lecture time.

5. **Is the lecturer committed to attending lectures on the scheduled dates?**

- Out of 223 responses, 32 participants expressed uncertainty about whether the lecturer adheres to the lecture schedules, while others claimed non-compliance. Some asserted this definitively. This represents 14% of the total responses, leaving a negative impact on the critical thinking components mentioned in Tables (1) and (2) in the research text.

6. **Does the lecturer manage the lecture well, providing opportunities for dialogue and discussion for all students?**

- Out of 227 responses, 39 participants indicated they didn't know if the lecturer manages the lecture well and provides opportunities for dialogue and discussion for all students. Another group claimed that the lecturer failed to do so. This accounts for 17% of the total responses and contradicts the enhancement of overall critical thinking skills for students.

7. **Does the lecturer motivate students and encourage them to engage in thinking and research activities?**

- Out of 227 responses, 37 participants expressed uncertainty about whether the lecturer motivates students and encourages them to engage in thinking and research activities. Another group claimed that the lecturer does not do this, with a total of 16% of the responses. This directly impacts the activation of critical thinking skills, especially the

students' ability to deduce and extract information, recognize contradictions, and engage in research activities.

8 Does the lecturer respect the different viewpoints of students?

- Out of 219 responses, 47 participants stated that they didn't know whether the lecturer respects the different viewpoints of students. Another group claimed that the lecturer does not respect diverse viewpoints, with a total of 14% of the responses. This is undoubtedly a significant factor inhibiting the activation of critical thinking skills among students.

9. Does the lecturer encourage students to learn on their own and search for what is new and modern?

- Out of 218 responses, 37 participants indicated they didn't know if the lecturer encourages students to learn on their own and search for what is new and modern. Another group highlighted the lecturer's shortcomings in this regard, with others affirming the lecturer's complete failure in encouraging independent learning. This constitutes 17% of the total responses, conflicting with the promotion of various critical thinking elements.

10. Does the lecturer accept criticism and suggestions with an open mind?

- Out of 208 responses, 47 participants indicated that they didn't know whether the lecturer accepts criticism and suggestions with an open mind. Another group claimed that the lecturer does not accept criticism and suggestions, with a total of 22% of the responses. This contradicts the goal of enhancing critical thinking skills among students, as it inhibits their ability to distinguish between provable facts and assertions and to evaluate arguments objectively.

11. Is the lecturer objective and fair in evaluating students?

- Out of 208 responses, 41 participants indicated that they didn't know if the lecturer is objective and fair in evaluating students or not. Another group asserted that the lecturer is not objective and fair, with some stating this definitively. This represents 20% of the total responses, affecting the activation of critical thinking skills among students, especially in evaluating their ability to present evidence and make inferences or deductions in an objective and scientific manner.

12. Does the lecturer use diverse methods to assess students' performance, such as assigning reports, research, and surprise exams?

- Out of 199 responses, 41 participants indicated that they didn't know if the lecturer uses diverse methods to assess students' performance or not. Another group highlighted the lecturer's shortcomings in this regard, with some claiming that the lecturer does not use such methods at all. This constitutes 20% of the total responses, negatively impacting the activation of critical thinking skills, particularly those related to students' self-organization.

13. Does the lecturer track activities and assignments to determine grades?

- Out of 204 responses, 31 participants indicated that they didn't know if the lecturer tracks activities and assignments to determine grades or if they have other methods for grading. Another group asserted that the lecturer does not track what is submitted for grade reporting, and a third group confirmed this decisively. This is 15% of the total responses, conflicting with the goal of activating critical thinking in the academic and scientific environment.

14. Does the lecturer have the ability to discuss all topics related to the academic subject?

- Out of 212 responses, 24 participants stated that they didn't know if the lecturer has the ability to discuss all topics related to the academic subject or not. Another group claimed that the lecturer lacks this ability, with others asserting definitively that the lecturer is incapable of doing so. This accounts for 11% of the total responses, negatively impacting the activation of critical thinking skills in various aspects.

15. Does the lecturer work to increase the required knowledge or not?

- Out of 219 responses, 24 participants indicated that they didn't know if the lecturer works to increase the required knowledge or not. Another group asserted that the lecturer does not work on that, with some stating this categorically. This constitutes 10% of the total responses, further weakening the possibility of enhancing critical thinking skills in various components, such as the student's ability to identify when new information is needed, assess the sufficiency and quality of data in addressing the topic, and anticipate future consequences and implications based on the acquired knowledge.

Conclusions

1. Motivating students and researchers to take into account critical thinking mechanisms that in turn affect the quality of the educational process, including:
   - Use detailed explanations and examples that are clear and understandable.
   - Use accuracy and reliability when formulating sentences and choosing phrases.
   - The logical and contextual connection of phrases and elements contained in sentences.
   - Deepening thinking, interpretation, and prediction to move beyond superficial treatment and realize the importance of the topic or goal based on that idea, thought, or event.
   - Taking all aspects of the problem or situation comprehensively and broadly, considering different perspectives and ways of dealing with the problem.
   - Identifying logical research methods that contribute to determining values and weighing different types of evidence that contribute to acceptable results.
   - The need for evidence and support for opinions and results before judging their reliability.
   - Knowledge of assumptions and evaluation of discussions.
   - Identifying information related to the subject, capable of making comparisons between proven or verifiable matters, and distinguishing basic information from relevant less information.
   - Formulating questions that contribute to a deeper understanding of the problem, with providing a standard for judging the quality of observations and conclusions.
   - Determining the reliability of sources, In order to achieve the general educational goals of establishing academic institutions.
2. Achieving quality in education, including the unknown experiences and problems it contains, while employing constant flexibility in providing a lot of new information.
3. There is a relationship between the components of critical thinking skills and the standards set by the Individualized Education Program to diagnose the quality of academic outputs and institutions in order to disseminate these skills and components to administrative agencies, curriculum planning, and quality control committees to adopt them in reviewing the value of efforts and directing them to ensure better and higher results.

Recommendations

1. The research recommends reviewing the alignment of critical thinking skills with each academic subject in architectural engineering departments for all study levels. This aims to activate these skills during the limited class time to achieve

greater efficiency in accomplishing the educational goal of each subject in a simpler and shorter time.

2. Study the meaning of cognitive strategies and their relationship with different thinking styles to align thinking styles with appropriate cognitive strategies. This will activate that thinking style specifically, considering the nature of problem-solving related to that academic subject at that study level.
3. Review the educational objectives of academic subjects in architectural engineering departments for all study levels in light of the required thinking styles to achieve those objectives. Assess cognitive strategies that activate the required thinking style to deliver the subject or academic term to the student in the simplest and fastest way.

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