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Original article

The Antimicrobial Effect of Some Metal Concentrations on the Growth of *Staphylococcus* and *Klebsiella* Bacteria Species

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Abstract
This study aimed to isolate and identify 110 samples from some rooms of different departments in Derna and Al-Bayda hospitals in order to determine the bacterial species that are likely to be present inside the hospitals. Some known microbiological methods were used to identify the isolated bacterial species, such as the spectrophotometer, the minimum inhibitory concentration (MIC), and the agar diffusion method. Concentrations of some elements (nickel, iron, copper) were used as inhibitors for the isolated bacterial species. Statistical analysis was performed using Tukey analysis, MLNLTAB program, version 2021. The results showed that the most isolated bacterial species were *Staphylococcus* at a rate of 20% and *Klebsiella* at a rate of 8.9%. The effectiveness of the concentrations of some of the elements under study on the growth of the isolated bacterial species was studied. The results showed that the nickel element was the most effective inhibitor at a concentration of 100% among the elements, followed by copper at a concentration of 100%, and the least effective element was iron on Staphylococcus bacteria, while on Gram-negative bacteria *Klebsiella* Copper was the most effective element at 100% concentration, followed by nickel, and iron had no effective effect on *Klebsiella*.
Keywords. Bacterial Isolation, Staphylococcus, Klebsiella; Antimicrobial, Hospital.

Introduction

Nosocomial infection can be caused by any organism, yet certain organisms are more likely to be the source of hospital-acquired infections [1]. The results of a study done in Europe show that the prevalence of HAIs. Numerous bacteria can cause nosocomial infections, and each one can result in infection in medical environments. Bacteria cause ninety percent of illnesses, with less of an influence from fungus. Hospital-acquired infections are typically caused by a variety of agents, such as *Streptococcus* species, Acinetobacter species, *Enterococci*, *Pseudomonas aeruginosa, coagulase-negative Staphylococci*, *Staphylococcus aureus*, *Bacillus cereus, Legionella*, and members of *Klebsiella pneumonia*, and the numerous species of *Staphylococcus* bacteria, *Staphylococcus aureus* is considered a very important pathogen responsible for many infections acquired in healthcare. It is more than just that we are more likely to contract *S. aureus* infections in communities. Not only can *S. aureus* infect surface tissues, but it can also infect deep tissues and local abscess lesions. Food poisoning from ingesting enterotoxins is one of the toxin-mediated disorders caused by S. aureus, whereas exfoliative toxins produce *staphylococcal* scalded skin syndrome [2]. *K. pneumoniae* is the seventh major pathogen in healthcare settings, accounting for three to seven percent of hospital-acquired bacterial infections. It belongs to the Enterobacteriaceae family and is an opportunistic, Gram-negative bacillus. It typically colonizes the skin, throat, and digestive tract. It is associated with illnesses like septicemia, wound infections, pneumonia, and newborn septicemia. One of the main factors contributing to nosocomial infection consequences is resistance to β-lactam medicines. *K. pneumoniae* and E. coli are two of the microorganisms that are resistant to medications that include β-lactamase. Third- and fourth-generation cephalosporins exhibit *K. pneumoniae.* [3] .It most likely plays a part in maintaining the stability of RNA structure. Divalent nickel works in tandem with the enzyme Glyoxalase I to facilitate the breakdown of methylglyoxal, lactate, and water. An octahedral high-spin Ni (II) complex called acereductonedioxygenase catalyzes the breakdown of the peroxo intermediate. Coordination connections can be formed between metal ions [4] .

There is a clear correlation between the rise in infectious disease-related fatalities in Africa and bacterial multi-antibiotic resistance. The primary cause of this problem is the dearth of potent antibacterial drugs. Recent research has focused mostly on the synthesis of nickel complexes with ligands due to their biological properties [5] .There is no doubt that developing new antibacterial agents through a range of methods is a pressing medical need [6]. The development of the latest nickel-based pharmaceuticals has been greatly aided by nickel's ability to increase the inhibitory power of chemotherapeutic medications. Coordination has been found to improve the efficacy of a range of medical therapies [7]. In the past few years, research has been conducted to create the metal copper, which has been shown to have self-sanitizing properties that prevent human infections from surviving exposure to copper or copper alloy surfaces for any appreciable amount of time. There is a significant reason for concern in the current pandemic context because this feature is not observed with other common surface materials such as plastic, stainless steel, or aluminum

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[7]. Three hundred different copper surfaces were registered with the US Environmental Protection Agency (USEPA) in 2008 as a result of testing conducted in an independent microbiology laboratory about the efficacy of copper surfaces. The following is stated in the registration: "When routinely cleaned, the antimicrobial copper metals have antimicrobial qualities that have been understood for many years [8].
In addition, the evaluation of their combination with antibiotics has been done in multiple trials. For instance, research has demonstrated that β-lactams combined with silver or zinc oxide nanoparticles have excellent results [9]. In the form of a metal complex, iron is the most prevalent transition element in the human body and a potentially effective antibacterial agent. Iron can affect bacterial cells by inducing oxidative stress, blocking respiration and ATP synthesis, increasing cell hydrophobicity, and facilitating penetration through the cell wall. Consequently, combination treatments provide a number of benefits, including a decreased risk of developing [10].

Methods

Study locations

This study was conducted at Al-Bayda Medical Center in Al-Bayda and Al-Wehda Hospital in Derna, (Libya), between October 2022 and December 2023. Al-Bayda and Derna are cities located in the northeastern region of Libya, as shown in Figure 1.

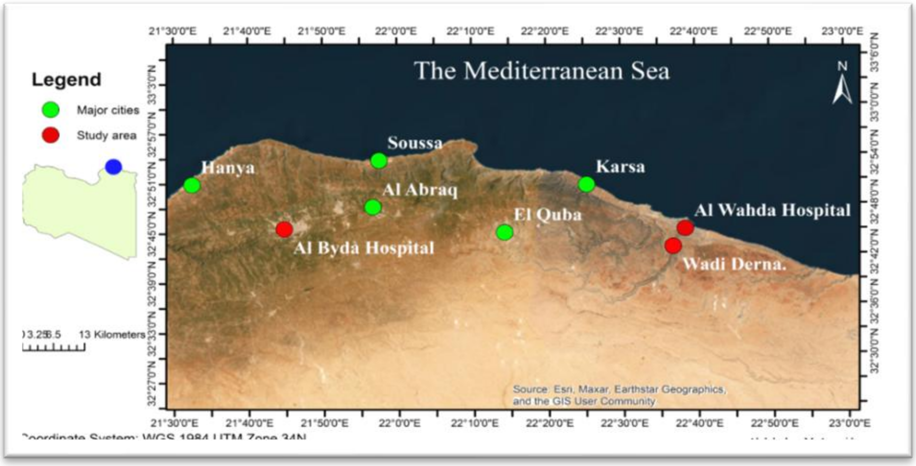

Figure 1. The location of Derna and Al-Bayda hospitals

Sampling

The samples were collected from two different hospitals (Al-Bayda Medical Center and Al-Wahda Derna) from some departments, Benches, and rooms. The samples were selected randomly, where 112 samples were taken from the pediatric department, incubators, some rooms, sterilization devices, monitoring devices, cauterization devices, emergency rooms, surgical department rooms, doors, handles, and ventilators. For Al-Bayda Hospital, the samples were collected from the women's Department only, which consisted of cauterization devices, monitoring devices, emergency carts, sterilization devices, and seats.

Isolation

The samples were isolated from the selected collection sites. A conventional Nutrient Broth bacterial medium was prepared according to the manufacturer's instructions on the package. Then the medium was sterilized in sterile tubes under sterile conditions. Swabs were taken from the locations, placed in test tubes under sterile conditions, and transferred to the laboratory to conduct the known microbiological tests [11].

Identification

Bacteria were identified using conventional diagnostic microbiological laboratory techniques as well as morphological appearance. They were cultured on selective media to determine the growth type of each bacterial isolate. Blood agar, MacConkey agar, mannitol agar, NB, and NA nutrient agar were used. They were incubated for 24 h at 37°C. Gram stain was then performed to determine whether they were Gram-positive or Gram-negative. Biochemical tests, including catalase and oxidase tests, were performed [11].

Antimicrobial investigation

Spectrophotometric method

The spectrophotometric method, which was carried out in this study was involved measuring the mixture of isolated bacteria with the suggested metal ion solutions. A suspension of 1.5×108 CFU ml was prepared for bacterial species of *Klebsiella, Pseudomonas, and S.aureus.* where 1 ml of the bacterial suspension and 1 ml of the element or plant extract used in this study were taken, which are (Different concentrations of iron, copper, and nickel Solutions) and extracts of the dodder plant, and 1 ml of sterile nutrient broth, and

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incubated at a temperature of 37 C° for 24 hours. Then, a spectrophotometer was used to estimate the turbidity values. The optical density (OD) of the designated broth media was measured using a spectrophotometer set at a wavelength of 600 nm to improve accuracy, then slightly diluted before taking the readings. These measurements were then repeated throughout the experiment at selected time points. Different concentrations of (5, 10,25,75,50, and 100 ppm) for metal ions in addition to the extract plant Dodder and minerals at all tested species of organisms [12].

Agar well propagation techniques

Antibacterial activity was confirmed using the agar diffusion method and using three bacterial colonies, namely *Klebsiella, Pseudomonas,* and *S. aureus*, using nutrient agar (NA) and sterile cotton swabs. A bacterial suspension was prepared using the McFarland method. The suspension was distributed on the plates, then using a sterile drill to create a bacterial well with a diameter of 4 mm in each plate; concentrations of mineral elements and different extracts Dodder plant were added at a rate of about 100 microliters. After 24 hours in the Incubation, the results were read using a ruler by measuring the diameter of the Inhibition zone [13].

Minimum inhibitory concentration (MIC) test

Different concentrations of metal elements (Iron, Copper and Nickel) and extracts of the plant Dodder according to the method of the National Committee for Clinical Laboratory Standards (NCCLS,2000), In this study, each one of the concentrations 25, 50, 75, and 100 ppm to the Nutrient Agar (NA) medium at 50°C. The suspension was distributed on plates and then left for a few minutes to dry. The plants were 24 hours at 37°C. A bacterial suspension was prepared without any antimicrobial agents. It is used to determine the actual growth of bacteria. The results were recorded as the presence or absence of growth.

Results

The obtained results were described according to the applied methods in this study (spectrophotometer and diffusion method). Colonies appeared on transitional media and were confirmed by biochemical tests and Gram stain, where under the microscope, a large, spherical, bright red colony appeared, non-motile, Gram-negative *Klebsiella*, while a blue colony appeared under the microscope, spherical, resembling a bunch of grapes, irregular balls. The result of the coagulase test showed blood clotting, and the colonies appeared yellow on Mannitol salt agar medium, due to its ability to completely decompose red blood cells, anaerobically. The results of isolation bacteria for the 110 samples which were used in this study showed that the species of bacteria *S. aureus* 20% and *Klebsiella* 8.9%, as shown in Table 1.

Table 1. Distribution values of bacterial isolates at sampling sites.

Isolation bacteria	Average
Staphylococcus aureus	20.719b±.1239
klebsiella	8.944c±7.87
Tukey's	0.2652 ***

The high spectrometer was set to measure the transmittance 600 nm which refer to the optical density at a wavelength of 600nm transmittance measurement was taken without adding coefficients at 100% and when adding substances at zero high turbidity little or no light transmittance occurs indicating high bacterial growth because the growth intercepts the light and it is absorbed instead of going into The other way and transmittance tells us how light passes through the sample that contains a large number of cells and the transmittance reading will be lower than the sample which contains a lower number of cells gives a higher reading and this is what we indicated in concentration 100 where the reading was higher and the number of microbial cells was lower .Showed the effect of different concentrations of metal ions (50, 75 and 100%) showed that the low concentrations of metal ion solutions (5, 10, 25%) were the readings respectively iron (5, 10, and 25%) (0.2, 4.5, and 2.4) copper (0.9, 4.6, and 2.2) nickel (1.2, 2.3, and 1.7) dodder (0.1, 0.5 and 0.9) did not show any bacterial activity on the other side the metal concentrations of 50, 75, and 100% recorded different antibacterial activities (6.7, 4.9, 2.1, and 35.96 nm) respectively the metal concentration of 100% gave high antibacterial permeability of (35.96 nm) the results were illustrated Table (2).

For the Nickel concentrations (50, 75, and 100 %) effect, the results recorded that the high concentration of 100 % gave antibacterial activities of (87.133, 16.2, and 96.76 nm) on Staphylococcus *aureus*, *Klebsiella* species, respectively. While the Nickel concentrations of (75 %) recorded inhibition, Bacteria values of (4, 20.66, and 14.07 nm) on *Staphylococcus aureus*, *Klebsiella* species, respectively. On the other side, the Nickel concentrations of (50 %) recorded lower antibacterial values of (8.06, 1.1, and 10.20 nm), on *Staphylococcus aureus*, *Klebsiella* species, respectively. The same manner was recorded for the effect of Iron solution concentrations on the selected bacterial species. Also, the concentrations of copper solutions of (50, 75 and 100 %) recorded antibacterial activities on the studied specie's (*Staphylococcus aureus*, *klebsieiia*),

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where the inhibition values (nm) were as following: (49, 27 and 44nm), (15, 7 and 11 nm) and (7, 2, and 9 nm) on the above concentrations and bacteria species, respectively.

Table 2. Effect of metal concentrations on antibacterial activity.

Concentrations	Average (mm)
100%	35.961a±29.64
75%	9.217b±5.702
50%	6.747c±4.080
Tukey's	0.4593 ***

The results of the first experiment containing three treatments, the first factor isolates used in the study, the second factor is elements, minerals, the third factor is concentrations at a significant level of 0.001, the coefficient of estimation of the difference was 99.65%, evidence of the accuracy of the experiment according to the statistical analysis of the normal distribution using Completely Randomized Design (CRD) and using the Turkey ś test using the MlNlTAB program by version 2021, Among the metal concentrations the one with the highest permeability was nickel at a concentration of 100% and copper at a concentration of 100%, then iron permeable according to the Tukey's test, which showed a significant difference between the averages, which were (a), (b), and then (c), as shown in Table (3).

Table 3. The antibacterial activity of different concentrations of metals

Isolation	*S. aureus*			*Klebsiella*		
Concentrations	100%	75%	50%	100%	75%	50%
Ni	87.133b	40	8.067	16.2	20.667f	1.1r
Fe	32.733d	3.2o	1.233q	2.733	6.067	6.6
Cu	49.067c	15.767f	7k	27.267 e	7.733	2.333
Tukey's	0.9185***					

For the agar diffusion method, the effect of metal concentrations on isolated bacteria inhibition by the Agar diffusion method. The solutions of metals (Fe, Ni, and Cu) for concentrations of (50, 75, and 100 %) showed small antibacterial activities compared with the values of values obtained by spectrophotometric methods. In addition, the concentrations of (Fe) did not show any antibacterial activities. The concentrations (100 %) of Nickel solutions showed (11, 9, and 11 mm), while the isolated bacteria S. *aureus* and *Klebsiella* whereas the values at (75 %) gave inhibition values. In addition, for the concentrations of (10, 8 and 10 mm) and the Copper concentrations of the same concentrations of (50, 75 and 100 %) for copper recorded values (11, 9 and 8) for 100 % against *S. aureus*, in addition to values of (10,7 and 6 mm) for (75 %) against.

Table 4. Effect of antibacterial activity of different metal concentrations and isolation by (Agar diffusion method) mm.

Isolation	*S. aureus*			*Klebsiella*		
Conc.	100	75	50	100	75	50
Ni	11a	10 ab	8 bc	9 abc	8 bc	7 c
Cu	11a	10 ab	9 abc	8 b	7 c	6 d
Tukey's	0.458					

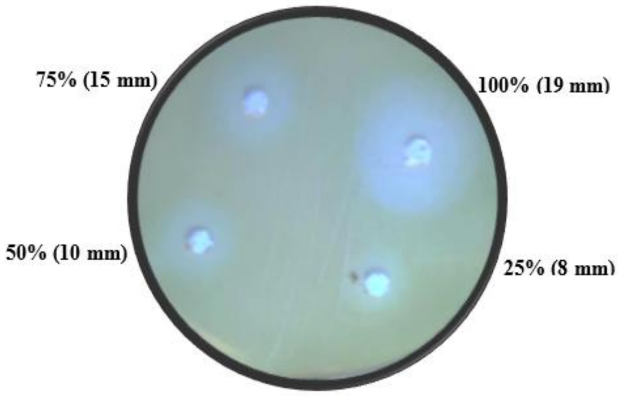

Figure 2. Antibacterial activity of nickel against S. aeruginosa

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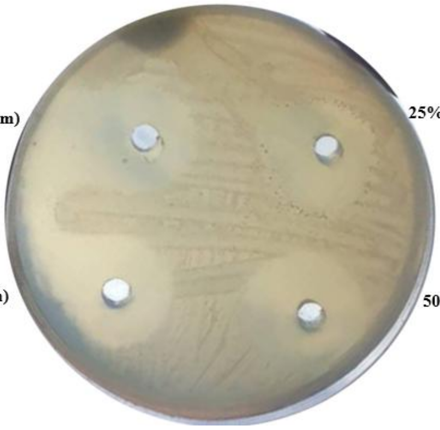

Figure 3. Antibacterial activity of copper against S. aeruginosa

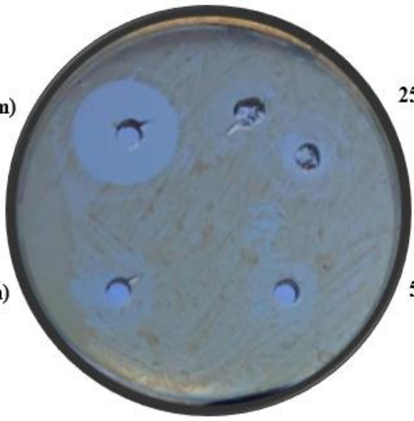

Figure 4. Antibacterial activity of nickel against klebsiella

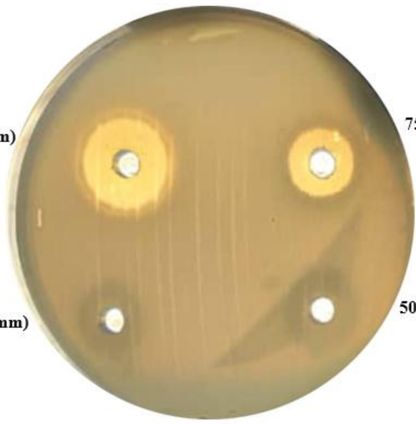

Figure 5. Antibacterial activity of copper against Klebsiella

Effect of different concentrations of metals on the growth of bacterial species

The effect of different concentrations (100%, 75%, and 50%) of metal solutions (Ni, Cu, and Fe) on isolated bacteria was illustrated in (Table 5 - 7). The results showed that the antibacterial activity, no growth of bacteria at a high concentration of (100%), whereas there is our presence of bacterial growth at concentrations (50 & 75 %). In addition, showed bacterial growth for the species (*Klebsiella* and *S.aureus*).

Table 5. Effect of Nickel concentrations on special bacteria.

Ni	**100%**	**75%**	**50%**
Klebsiella	_	+	+
S. aureus	_	+	+

**Positive (+), and negative (-)*

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Table 6. Effect of copper concentrations on selected bacteria

Cu	**100%**	**75%**	**50%**
Klebsiella	-	+	+
S. aureus	-	+	+

Table 7. Effect of iron concentrations on selected bacteria.

Fe	100%	75%	50%
Klebsiella	-	+	+
S. aureus	-	+	+

Discussion

In this investigation, about 112 samples were collected from different departments of Al Bayda and Derna city hospitals. The isolated bacterial species were identified, and then different concentrations of metal ions and Dodder extracts were used to study their effect on the isolated bacterial species.

The results of this study showed different bacteria species, but most of the studied samples exhibited the presence of *Pseudomonas* is the most common [14]. As per the results of this investigation, the element nickel exhibited antibacterial activity at a concentration of 100, as demonstrated by its effective activity measured by spectrophotometry, MIC, and the agar diffusion methods. The discovery was made that the enhanced antibacterial activity that permeates the bacterial cell and reaches the membrane surface is due to the properties of both the metal ion and the ligand surrounding it. The microbial cell and its internal milieu. The impact of this flow on bacterial metabolism is the destruction of the organelles. The findings of this study were in agreement with the results recorded by some studies [15]. Which they were examined the MlC and UV spectroscopy methods for assaying the antibacterial activity of some bacteria using electron microscopy on *E. coli* and *S. aureus*. In addition, some studies recorded that the Nickel (Ni) solutions showed antibacterial activity on some species of bacteria. Both Gram-positive and Gram-negative bacteria did nickel complexes effectively combat. In this study, the results of copper concentrations (Cu) on the isolated bacteria agreed with those results that were shown by some studies [16]. which they reported that different Copper (Cu) showed a good effect on the inhibition of bacterial growth. It was revealed that the effect of Meal solutions on the anti-bacterial activity is due to their great capacity to pierce bacterial membranes; copper is thought to possess the greatest bactericidal potential. The capacity to attain a very high killing effectiveness for bacteria that are in close contact for a brief length of time is what makes metallic copper or copper surfaces unique. This is because of the way that copper atoms are structured, particularly how quickly they can give or receive electrons, which is what makes copper an electrical conductor. unbound electrons throughout Microbes' activity can be suppressed by interactions with microbial proteins during contact [16].

It was reported that cell disruption occurs when there is a leakage of cellular components due to damage to the cell membrane. Copper destroys MRSA through DNA damage, corroborating [17]. Their study declared that copper exposure renders many microorganisms nonviable. It is suggested that frequently touched surfaces in healthcare institutions be replaced with copper or copper pipes, either immediately or over a predetermined period. This will significantly reduce the spread of illness. It was reported that the velocity and iron uptake in vivo are intimately related. Pseudomonas growth is induced by bacterial infection, which also generates bacteria with enhanced iron use. It is now known that iron metabolism and bacterial pathogenicity are related. Different chemical compounds and metal complexes were used as anti-bacterial inhibitors; most of these studies concluded that the effect depended on the linkage between metal and ligand used in complexes and their attack of bacterial cell membranes [18-26]. Also, numerous studies used plant extracts as anti-bacterial agents; they considered the presence of different metals and different chemical compounds for the anti-bacterial activities [27-55].

Conclusion

This study used crude dodder extract and certain element concentrations to demonstrate antibacterial efficacy against both Gram-positive and Gram-negative bacteria. This study demonstrates that nickel metal exhibits 100% concentration-dependent bactericidal and antibacterial action. Similarly, copper's capacity to penetrate bacterial membranes, which results in damage to DNA, cell membrane damage, and cell death, had an effective effect. Furthermore, the alcoholic dodder extract is thought to have no effect Pseudomonas. At high concentrations 100%, the extract has a greater impact on Gram-positive bacteria than on Gram-negative bacteria. All of the data used in this study it has different contents of phytochemical substances that, at varying, medium to low concentrations, contain phenols, alkaloids, amino acids, and minerals and have antioxidant, antibacterial, and anticancer effects.

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Conflict of interest. Nil

References

1. Hu Y, Li D, Xu L, Hu Y, Sang Y, Zhang G, Dai H. Epidemiology and outcomes of bloodstream infections in severe burn patients: a six-year retrospective study. Antimicrob Resist Infect Control. 2021;10:1-8.
2. Grass G, Rensing C, Solioz M. Metallic copper as an antimicrobial surface. Appl Environ Microbiol. 2011;77(5):1541-7.
3. Giacometti A, Cirioni O, Schimizzi AM, Del Prete MS, Barchiesi F, D'Errico MM, et al. Epidemiology and microbiology of surgical wound infections. J Clin Microbiol. 2000;38(2):918-22.
4. Chivers T, Laitinen RS. Tellurium: a maverick among the chalcogens. Chem Soc Rev. 2015;44(7):1725-39.
5. Wilfred CD, Kiat CF, Man Z, Bustam MA, Mutalib MIM, Phak CZ. Extraction of dibenzothiophene from dodecane using ionic liquids. Fuel Process Technol. 2012;93(1):85-9.
6. Adams C, Ide T, Barnett J, Detges A. Sampling bias in climate–conflict research. Nat Clim Chang. 2018;8(3):200-3.
7. Carson JB, Tesluk PE, Marrone JA. Shared leadership in teams: An investigation of antecedent conditions and performance. Acad Manage J. 2007;50(5):1217-34.
8. Ogunniran KO, Ajanaku KO, James OO, Ajani OO, Adekoya JA, Omonhimin CA, et al. Synthesis, physical properties, antimicrobial potentials of some mixed antibiotics complexed with transition metals and their effects on alkaline phosphatase activities of selected rat tissues. Sci Res Essays. 2008;3(8):348-54.
9. Alekshun MN, Levy SB. Molecular mechanisms of antibacterial multidrug resistance. Cell. 2007;128(6):1037-50.
10. Claudel M, Schwarte JV, Fromm KM. New antimicrobial strategies based on metal complexes. Chemistry. 2020;2(4):849-99.
11. Benson HJ. Microbiological applications: A laboratory manual in general microbiology. 5th ed. Dubuque (IA): WCB Publishers; 1990.
12. Wadhwani AR, Affaneh A, Van Gulden S, Kessler JA. Neuronal apolipoprotein E4 increases cell death and phosphorylated tau release in Alzheimer disease. Ann Neurol. 2019;85(5):726-39.
13. Mahmoudi M, Sant S, Wang B, Laurent S, Sen T. Superparamagnetic iron oxide nanoparticles (SPIONs): development, surface modification and applications in chemotherapy. Adv Drug Deliv Rev. 2011;63(1-2):24-46.
14. Haghshenas L, Faraji A. Evaluation of the effect of Gold and Nickel nanoparticles on Escherichia coli and Staphylococcus aureus bacteria in milk. J Micro Nano Biomed. 2016;1:1-6.
15. Santos F, DeCastro M, Gómez-Gesteira M, Álvarez I. Differences in coastal and oceanic SST warming rates along the Canary upwelling ecosystem from 1982 to 2010. Cont Shelf Res. 2012;47:1-6.
16. Abraham J, Dowling K, Florentine S. Can Copper Products and Surfaces Reduce the Spread of Infectious Microorganisms and Hospital-Acquired Infections? Materials (Basel). 2021;14(13):3444.
17. Hamad MAH, Hanan AAK, Fatima A. Infrared (IR) characterization and physicochemical properties of Schiff base compound obtained by the reaction between 4-hydroxy-3-methoxy benzaldehyde and 2-amino-3. J Res Pharm Sci. 2021;7(3):8-12.
18. Hamad MIH, Aaza IY, Safaa SH, Mabrouk MS. Biological study of transition metal complexes with adenine ligand. Proceedings. 2019;41(1):77.
19. Ahmed O, Ahmed NH, Hamad MAH, Fatin ME. Chemical and biological study of some transition metal complexes with guanine as ligand. Int J New Chem. 2023;10(3):172-83.
20. Hamad MAH, Enas UE, Hanan AK, Hana FS, Somia MAE. Synthesis, characterization and antibacterial applications of compounds produced by reaction between Barbital with Threonine, glycine, lycine, and alanine. Afr J Biol Sci. 2024;6(4).
21. Ashraf AA, Hamad MAH, Hanan AAK, Hana FS, Somaia MAE, Taffaha AA, et al. Molecular docking studies of some Schiff base compounds. Afr J Biol Sci. 2024;6(3):3324-34.
22. Mohmed GB, Zainab SH, Hamad MAH, Hanan AKA, Mounera AAE, Mohammed MY, et al. IR analysis and some biological applications for some Schiff base compounds prepared between (4-dimethyl amino benzaldehyde) and some amino acids (Tryptophan, Phenylalanine). Eur Chem Bull. 2024;12(5):887-906.
23. Mabrouk MS, Moussa SF, Hamad MAH, Hasan H. Synthesis, characterization, and antibacterial studies of metal complexes with tyrosine ligand. Int J New Chem. 2023;10(5):323-39.
24. Hamad Hasan. Biological study of some first series transition metal complexes with adenine ligand. In: Proceedings of the 23rd International Electronic Conference on Synthetic Organic Chemistry; 2019.
25. Siddiqui AA, Mojahid I, Hasan HH. Synthesis and antituberculostic activity of some novel 1,3,4-oxadiazole. Hamdard Medicus. 2011;54(1):82-9.
26. Eltawaty SA, Abdalkader GA, Hasan HM, Houssein MA. Antibacterial activity and GC-MS analysis of chloroform extract of bark of the Libyan Salvia fruticosa Mill. Int J Multidiscip Sci Adv Technol. 2021;1(1):715-21.
27. Aljamal MA, Hasan HM, Al Sonosy HA. Antibacterial activity investigation and antibiotic sensitivities for different solvents (Ethanol, propanol, DMSO and diethyl ether) extracts of seeds, leaves and stems of (Laurus azorica and Avena sterilis) plants. Int J Curr Microbiol Appl Sci. 2024;13(11):175-90.
28. Hamade MH, Abdelraziq SA, Gebreel AA. Extraction and determination of the beta-carotene content in carrots and tomato samples collected from some markets at El Beida City, Libya. EPH Int J Appl Sci. 2019;1(1):105-10.

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29. Hasan HM, Ibrahim H, Gonaid MA, Mojahidul I. Comparative phytochemical and antimicrobial investigation of some plants growing in Al Jabal Al-Akhdar. J Nat Prod Plant Resour. 2011;1(1):15-23.
30. Hasan H, Jadallah S, Zuhir A, Ali F, Saber M. The anti-cancer, anti-inflammatory, antibacterial, antifungal, anti-oxidant and phytochemical investigation of flowers and stems of Anacyclus clavatus plant extracts. AlQalam J Med Appl Sci. 2025:415-27.
31. Hasan H, Zuhir A, Shuib F, Abdraba D. Phytochemical investigation and exploring the Citrullus colocynthis extracts as antibacterial agents against some Gram-negative bacteria species. AlQalam J Med Appl Sci. 2025:392-400.
32. MdZeyaullah RA, Naseem A, Badrul I, Hamad MIH, Azza SA, Faheem AB, et al. Catechol biodegradation by Pseudomonas strain: a critical analysis. Int J Chem Sci. 2009;7(3):2211-21.
33. El-Mehdawy MF, Eman KS, Hamad MIH. Amino acids contents of leaves and stems for two types of herbal plants (Marjoram and Hybrid tea rose) at AL-Gabal AL-Akhder region. Der Pharma Chem. 2014;6(6):442-7.
34. Gonaid MH, Hamad HH, Ibrahim HH, Mojahidul I. Comparative phytochemical and antimicrobial investigation of some plants growing in Al Jabal Al-Akhdar. J Nat Prod Plant Resour. 2011;1(1):15-23.
35. El-Mehdawy MF, Eman KS, Hamad MIH. Amino acid contents of leaves and stems for three types of herbal plants at Al-Gabal Al-Akhder region. World J Chem. 2014;9(1):15-9.
36. Hamad MH, Noura AAM, Salem AM. Phytochemical screening, total phenolic, anti-oxidant, metal and mineral contents in some parts of Plantago albicans grown in Libya. World J Pharm Res. 2024;13(3):1-17.
37. Anees AS, Hamad MIH, Mojahidul I. Antifungal potential of 1,2,4-triazole derivatives and therapeutic efficacy of Tinea corporis in albino rats. Der Pharm Lett. 2011;3(1):228-36.
38. Hamad H, Marwa M, Amal H. Determining the contents of antioxidants, total phenols, carbohydrate, total protein, and some elements in Eucalyptus gomphocephala and Ricinus communis plant samples. Libyan Med J. 2015:222-31.
39. Hamad H, Zuhir A, Farag S, Dala A. Efficiency of Cynara cornigera fruits on antibacterial, antifungal and its phytochemical, anti-oxidant screening. Libyan Med J. 2025:120-8.
40. Hanan MA, Hamida E, Hamad MAH. Nitrogen, phosphorus and minerals (Sodium, Potassium and Calcium) contents of some algae's species (Anabaena and Spirulina platensis). Int J Curr Microbiol Appl Sci. 2016;5(11):836-41.
41. Hasan H, Mariea FFE, Eman KS. The contents of some chemical compounds of leaves and stems of some herbal plants (Thyme, Rosemary, Salvia, Marjoram and Hybrid Tea Rose) at Al-Gabal Al-Akhder region. EPH Int J Appl Sci. 2014;6(3).
42. El-Mehdawe MF, Eman KS, Hamad MIH. Heavy metals and mineral elements contents of leaves and stems for some herbal plants at AL-Gabal AL-Akhder region. Chem Sci Rev Lett. 2014;3(12):980-6.
43. Hamad H, Ashour S, Ahmed A. Estimation of amino acid composition, total carbohydrate, and total protein content in Ballota pseudodictamnus plant extracts from Al Jabal Al Akhdar region, Libya. Libyan Med J. 2015:266-71.
44. Hamad H, Ahmed H, Wafa A. Evaluation of anti-oxidant capacity, total phenol, metal, and mineral contents of Ziziphus lotus plant grown at some regions of Al Gabal Al Khder, Libya. Libyan Med J. 2015:137-43.
45. Hesien RA, Amira AKA, Ahlaam MA, Hamad MAH. Determination of the anti-oxidant capacity, total phenols, minerals and evaluation of the anti-bacteria activity of leaves and stems of Gaper plant extracts. Sch J Appl Med Sci. 2024;12(4):451-7.
46. Hamad MAH, Noura AAM, Salem AM. Total carbohydrate, total protein, minerals and amino acid contents in fruits, pulps and seeds of some cultivars of muskmelon and watermelon fruit samples collected from Al. Der Pharma Chem. 2024;16(3):330-4.
47. Ben Arous NAA, Naser ME, Hamad MAH. Phytochemical screening, anti-bacterial and anti-fungi activities of leaves, stems and roots of C. parviflorus Lam and C. salviifolius L plants. Int J Curr Microbiol Appl Sci. 2014;13(11):262-80.
48. Anas FAE, Hamad MAH, Salim AM, Azza MH. Phytochemical screening, total phenolics, antioxidant activity and minerals composition of Helichrysum stoechas grown in Libya. Afr J Biol Sci. 2024;3(6).
49. Naseer RE, Najat MAB, Salma AA, Hamad MAH. Evaluation of metal and mineral contents of leaves, stems and roots of C. parviflorus Lam and C. salviifolius L plants growing at Al Ghabal Al-Khder (Libya). Int J Adv Multidiscip Res Stud. 2024;4(5):191-4.
50. Hamad MAH, Salem AM. Total carbohydrate, total protein, minerals and amino acid contents in fruits, pulps and seeds of some cultivars of muskmelon and watermelon fruit samples collected from Algabal Alkhder region. Sch J Appl Med Sci. 2024;12(1):1-7.
51. Haroon A, Hamad MAH, Wafa AAS, Baset ESM. A comparative study of morphological, physiological and chemical properties of leaves and stem samples of (E. gomphocephala) (Tuart) plant growing at coastal (Derna city) and… J Res Environ Earth Sci. 2024;9(12):10-8.
52. Enam FM, Wesam FAM, Hamad MAH. Detection of the contents of minerals of (Sodium, Potassium and Calcium) and some metals of (Iron, Nickel and Copper) in some vegetable and soil samples collected from Al-Marj. Int J Adv Multidiscip Res Stud. 2023;5(3):304-9.
53. Rinya FMA, Hamad MAH, Ahlam KA, Hammida MEH. Phytochemical screening of some herbal plants (Mentha, Origanum and Salvia) growing at Al-Gabal Al-Akhder Region-Libya. Afr J Basic Appl Sci. 2017;9(3):161-4.
54. Ali RFA, Hamad MAH, Ahlam KA, Hammida MEH. Phytochemical screening of some herbal plants (Mentha, Origanum and Salvia) growing at Al-Gabal Al-Akhder Region-Libya. Int J Pharm Life Sci. 2017;8(4):5500-3

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