



Evaluation of the Antibacterial Potential of Cephalexin Schiff Bases against *Bacillus Pumilus* and *Candida albicans*

Khalid Zainulabdeen¹, Raghdha Alsayed¹, Sarah Mahdi¹, Dina Ahmed², Israa Salman³, Anaheed Yaseen⁴, Nany Hairunisa⁵, Salam Mohammed⁶, Emad Yousif^{1*}

¹ Department of Chemistry, College of Science, Al-Nahrain University, Baghdad, Iraq.

² Department of Chemical Industries, Institute of Technology-Baghdad, Middle Technical University, Baghdad, Iraq

³ College of Science, Al-Nahrain University, Baghdad, Iraq.

⁴ Department of Chemistry, College of Science, Tikrit University, Tikrit, Iraq.

⁵ Department of Occupational Medicine, Faculty of Medicine, Universitas Trisakti, Jakarta, Indonesia

⁶ Department of Chemical and Petrochemical Engineering, College of Engineering and Architecture, University of Nizwa, Oman

emad_yousif@hotmail.com

Received: 6 June 2023

Accepted: 11 September 2023

DOI: <https://doi.org/10.24237/ASJ.01.04.765I>

Abstract

In the current study, cephalexin and two beta-lactam antibiotics were combined with four distinct aldehydes: 3-hydroxybenzaldehyde, 4-dimethylaminobenzaldehyde, 4-methoxybenzaldehyde, and 4-bromobenzaldehyde, to synthesize Schiff bases. The reaction between the free amino group of the acylamino side chain and the aldehydes resulted in the formation of four Schiff bases within the medication. Comprehensive characterization and surface examination were performed on each Schiff base. The biological activity of the synthesized compounds against *Bacillus Pumilus* (Gram-positive bacteria) and *Candida*



albicans (fungus) was evaluated using the agar diffusion disc method. The antibacterial screening tests revealed that the Schiff bases exhibited enhanced antibacterial activity against these microorganisms, indicating superior efficacy compared to the parent drugs. However, no significant inhibitory effect on the tested fungus was observed.

Keywords: Schiff base; Cephalexin; Aldehydes; Bacillus Pumilus; Candida albicans.

النشاط المضاد للبكتريا باستخدام معقدات القصدير المشتقة من قاعدة شيف ضد

Candida albicans و Bacillus Pumilus

خالد زين العابدين¹ و رغبة السيد¹ سارة مهدي¹ و دينا احمد² و اسراء سلمان³ و اناهيد ياسين⁴ و ناني خير النساء⁵ و سلام محمد⁶ و عماد يوسف¹

¹ قسم الكيمياء-كلية العلوم-جامعة النهرين-العراق

² معهد التكنولوجيا-بغداد-العراق

³ كلية العلوم-جامعة النهرين-العراق

⁴ قسم الكيمياء-كلية العلوم-جامعة تكريت-العراق

⁵ قسم الطب المهني ، كلية الطب ، جامعة تريساكنتي ، جاكرتا ، إندونيسيا

⁶ قسم الهندسة الكيميائية والبتر وكيمائية ، كلية الهندسة والعمارة ، جامعة نزوى ، عمان

الخلاصة

في الدراسة الحالية ، تم الجمع بين سيفالكسين واثنين من المضادات الحيوية بينا لاكتام مع أربعة ألدهيدات مميزة: 3- هيدروكسي بنزالديهيد ، 4-ثنائي ميثيل أمينوبنزالديهيد ، 4-ميثوكسي بنزالديهيد ، و 4-بروموبنزالديهيد ، لتكوين قواعد شيف. نتج عن التفاعل بين المجموعة الأمينية الحرة من السلسلة الجانبية والألدهيدات تكوين أربع قواعد Schiff داخل الدواء. تم إجراء توصيف شامل وفحص السطح على كل قاعدة شيف. تم تقييم النشاط البيولوجي للمركبات المحضرة ضد (البكتيريا موجبة الجرام) والمبيضات البيضاء (الفطريات) باستخدام طريقة قرص انتشار الأجار. أظهرت اختبارات الفحص المضاد للبكتيريا أن قواعد شيف أظهرت نشاطاً مضاداً للبكتيريا معزراً ضد هذه الكائنات الدقيقة ، مما يشير إلى فعالية فائقة مقارنة بالأدوية الأم. ومع ذلك ، لم يلاحظ أي تأثير مثبت معنوي على الفطريات المختبرة.

الكلمات المفتاحية: قاعدة شيف. سيفالكسين. الألدهيدات. Bacillus Pumilus. Candida albicans.

Introduction

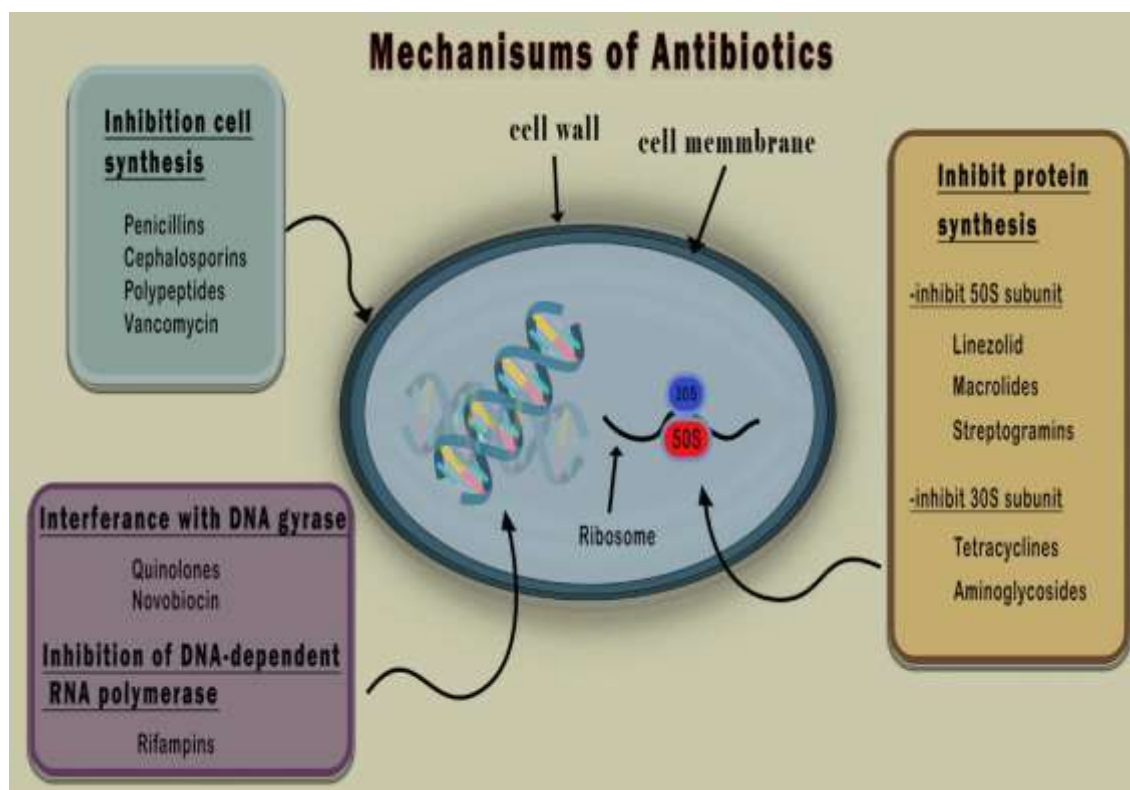
Antibiotics are substances that are released by microorganisms as natural metabolites that have a bactericidal or bacteriostatic effect on the growth of other microorganisms. The effect of the antibiotic on one of the main parts of the bacterial cell as shown in Scheme 1 [1]:

Cephalexin and all forms of penicillin inhibit the growth of the gram-positive germ wall on the cell wall.

Tetracyclines target the 30S ribosomal subunit, preventing t-RNA from binding to the A site.

The formation of the RNA, such as rifampin, streptomycin, tetracycline, and erythromycin.

On DNA, Quinolones, The fluoroquinolones (FQ) inhibit the enzyme bacterial DNA gyrase, which nicks the double-stranded DNA.



Scheme 1: Mechanisms of antibiotics [1]



Chemical therapies use inorganic substances to inhibit growth. The effects of several chemical agents on bacterial cells are unclear, but some, such as sulfonamides, disrupt the production of tetrahydrofolic acid, an important catalyst in bacteria metabolism. The uses of chemical therapeutic materials are limited due to their relatively high toxicity and many side effects [2, 3]. Derivatives of β -lactam antibiotics have been made to boost their potency and widen their antibacterial abilities. Schiff base ligands and their metal complexes generated from various aldehydes with cefadroxil [4], cephradine [5, 6], ceftriaxone [7], and cephalothin [8], as well as amoxicillin [9-13]. Metal complexes and Schiff bases were created by combining the drug molecule with various aldehydes. Metal complexes for the prepared Schiff bases have also been produced. The synthesized compounds' potential antibacterial qualities were examined [14–31]. Research has shifted towards metal-drug complexes to increase activity and reduce side effects. The work's goal is to investigate cephalixin Schiff bases' biological activities.

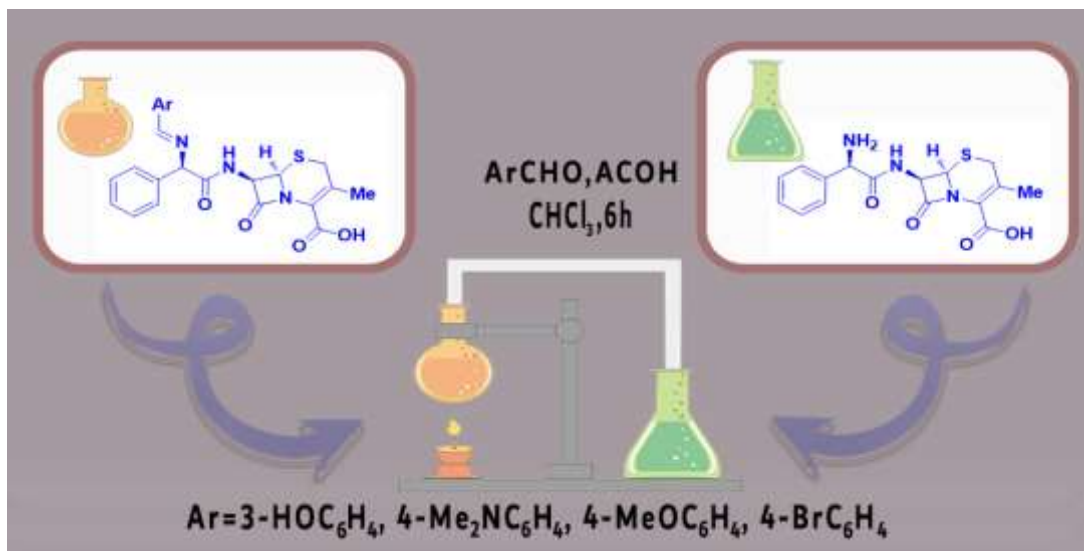
Experimental

1. Material.

All the chemicals and solvents such as CH_3Cl , AcOH , DMSO , cephalixin, nutrient agar were analytical grade (Fluka and Sigma-Aldrich) and weren't further purified before usage.

2. Synthesis of Cephalixin Schiff Bases.

The Schiff bases were made as previously described [32] by boiling CHCl_3 (25 mL) with AcOH (0.5 mL) and stirring in cephalixin (1.83 g, 5.0 mmol) and an aryl aldehyde (5.0 mmol). This mixture was refluxed for 6 hours (Scheme 2). The mixture was cooled to room temperature and the solid was collected, washed with CHCl_3 , and dried to obtain 1-4 ml in high yields.



Scheme 2: Synthesis of cephalixin Schiff bases 1–4.

3. Culture media preparation.

A - The clusters (28gm) were dissolved in a liter of distilled water to make the nutrient agar. The temperature of (45Co) was placed on a culture plate and left to solidify at room temperature for (15min). The culture media were then perforated and injected with both bacterial and fungal inoculums. Before pouring it onto dishes, it was heated to (37Co) for one hour in the incubator [33].

B- The weights of the aforementioned ingredients were dissolved in one liter of distilled water to create Media 1 [34]:

4. Preparing chemical solutions

The dimethyl sulfoxide solution was utilized to prepare the chemical solutions for the biological study (DMSO). The two concentrations of 10⁻² M and 10⁻⁴ M were created from each sample. Glass tubes utilized for the preparation were sterilized in an autoclave for 15 minutes at 120 Co and 15 atm of pressure. The antibiotic cephalixin served as a point of reference.



Under the same conditions, a control model using only (DMSO) was carried out, and its impact on bacteria and fungus was investigated [35].

5. Sensitivity examination technique for compound preparation.

The concentrations of the prepared compounds, the antibiotic, were prepared with two concentrations of each, 10^{-2} M and 10^{-4} M, and then 50 μ l into the holes that were made in the culture medium and allowed to absorb the material. The media was then preserved in the incubator at a degree (37 Co), which is the proper degree for bacterial growth, for a period of (24) hours.

6. Damping area measurements via (diameter determination)

The transparent region that included the hole and did not support bacterial growth (the zone of inhibition) was measured by millimeters. The synthesized compounds have a higher biological activity when the diameter of inhibition is larger.

Results and Discussion

1. The FESEM of cephalixin Schiff bases 1–4

The FESEM was used to examine the surface morphology of the sample (Fig. 1). Images revealed that layers 1-4 have porous and diverse structures. The particles' diameters ranged from 40.58 to 56.65 nm for Schiff base 1, 39.96 to 49.75 nm for Schiff base 2, 29.46 to 39.01 nm for Schiff base 3, and 42.29 to 47.16 nm for Schiff base 4. Additionally, the particles' morphologies were varied.

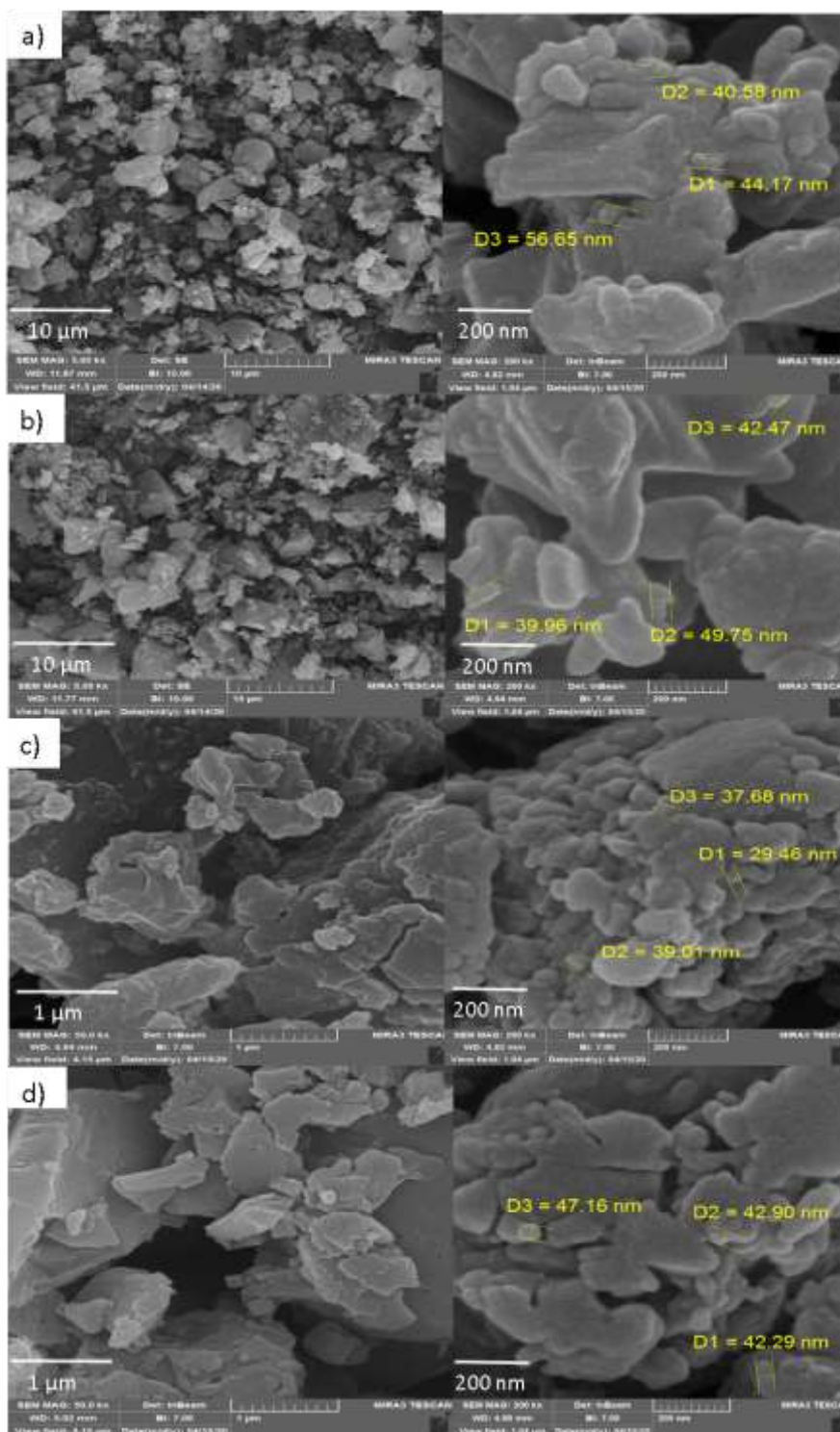


Figure 1: (a-d) SEM images of cephalixin Schiff bases 1-4, respectively.

This study used one type of bacteria, *Bacillus Pumilus*, and one type of fungi, *Candida albicans*, to upgrade the action of cephalexin and cephalexin Schiff bases. The compounds gave high inhibition for the bacteria, but no inhibition for the mushrooms, as shown in Table (1).

Table 1: Biological activity for cephalexin and cephalexin Schiff bases.

COMPOUND	CONC. (MG/ML)	INHIB. ZONE (MM) BACILLUS-PUMILUS	INHIB. ZONE (MM) CANDIDA-ALBICANS
Cephalexin(Ceph)	1×10 ⁻²	13.2	25.6
	1×10 ⁻⁴	12.2	25.5
Ceph+3-HOC6H4	1×10 ⁻²	28.5	0.0
	1×10 ⁻⁴	22.6	0.0
Ceph+4-Me2NC6H4	1×10 ⁻²	31.9	0.0
	1×10 ⁻⁴	28.1	0.0
Ceph+ 4-MeOC6H4	1×10 ⁻²	29.1	15.9
	1×10 ⁻⁴	28.7	11.7
Ceph+ 4-BrC6H4	1×10 ⁻²	27.6	12.9
	1×10 ⁻⁴	26.5	11.6

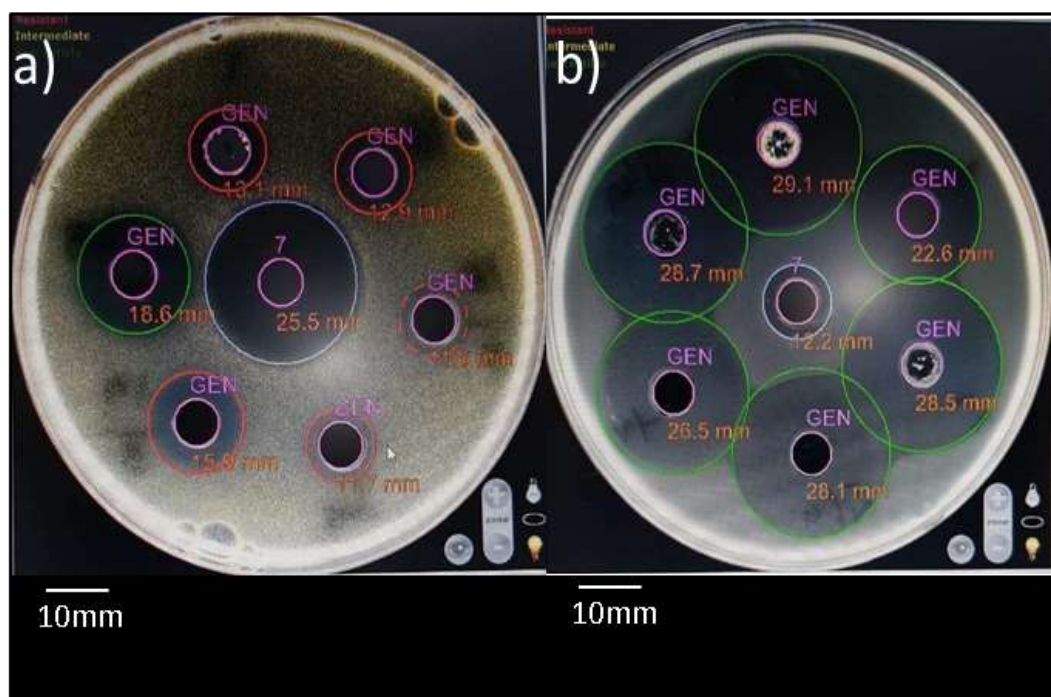


Figure 2: Biological activity for cephalexin Schiff bases against a) *Bacillus Pumilus* and b) *Candida albicans*.



Conclusions

In conclusion, benzaldehyde derivatives have been shown to be effective against microbes. These include three-hydroxybenzaldehyde, 4-dimethylaminobenzaldehyde, 4-methoxybenzaldehyde, and 4-bromobenzaldehyde. These substances have antibacterial qualities that make them effective at preventing bacterial and fungal diseases. Prospects for using benzaldehyde derivatives as powerful antibacterial agents in medical and pharmaceutical applications are bright with more study and development in this field.

References

1. Nicholls, D. (1974). Complexes and first-row transition elements. London: Macmillan.
2. Misra, H. K., & Gupta, A. K. (1982). Synthesis and pesticidal activities of some new substituted 3H-quinazolin-4-one derivatives; part XVIII. *Pesticide Science*, 13(2), 177-182.
3. Raman, N., Selvan, A., Sudharsan, S., et al. (2011). Metallation of ethylenediamine based Schiff base with biologically active Cu(II), Ni(II) and Zn(II) ions: Synthesis, spectroscopic characterization, electrochemical behaviour, DNA binding, photonuclease activity and in vitro antimicrobial efficacy. *Spectrochimica Acta Part A*, 79, 873-883.
4. Etaiw, H. E. S., El-Aziz, A. M. D., El-Zaher, A. H. E., et al. (2011). Synthesis, spectral, antimicrobial and antitumor assessment of Schiff base derived from 2-aminobenzothiazole and its transition metal complexes. *Spectrochimica Acta Part A*, 79, 1331-1337.
5. Abdulsada, A. H., Nasser, N. H., & Hussein, A. K. (2018). *Acta Chimica and Pharmaceutica Indica*, 03, 17-23.
6. Bukhari, I. H., Sher, M., & Arif, M. (2005). Synthesis, characterization and biological evaluation of Schiff base transition metal complexes with ampicillin. *Science International (Lahore)*, 17(3), 223-226.



7. Fareed, G., Kalhoro, M. A., Versiani, M. A., Afza, N., & Fareed, N. (2012). 7-[[2-(4-Hydroxyphenyl)methylidene]amino}-1,3-thiazol-4-yl)-2-(methoxyimino)acetyl]amino}-3-[[2-methyl-5,6-dioxo-1,2,5,6-tetrahydro-1,2,4-triazin-3-yl)sulfanyl]methyl]-8-oxo-5-thia-1-azabicyclo[4.2.0]oct-2-ene-2-carboxylic Acid. *Molbank*, 2012(2), M756.
8. Anacona, J., Noriega, N., & Camus, J. (2015, February). Synthesis, characterization and antibacterial activity of a tridentate Schiff base derived from cephalothin and sulfadiazine, and its transition metal complexes. *Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy*, 137, 16–22. <https://doi.org/10.1016/j.saa.2014.07.091>
9. Chaudhary, N. K., & Mishra, P. (2017). Metal Complexes of a Novel Schiff Base Based on Penicillin: Characterization, Molecular Modeling, and Antibacterial Activity Study. *Bioinorganic Chemistry and Applications*, 2017, 1–13.
10. Al Masoudi, W. A., Mohammad, H. T., & Hama, A. A. (2015, June 15). SYNTHESIS, CHARACTERIZATION AND PHARMACOLOGICAL STUDY OF NEW SCHIFF BASE DERIVED FROM AMOXICILLIN DRUG. *International Research Journal of Pharmacy*, 6(6), 386–389.
11. Al-Noor, T. H., Aziz, M. R., & AL-Jeboori, A. T. (2014). Synthesis, characterization and antimicrobial activities of [Fe (II), Co (II), Ni (II), Cu (II) and Zn (II)] mixed ligand complexes schiff base derived from amoxicillin drug and 4-(dimethylamino) benzaldehyde with nicotinamide. *Journal of Chemical and Pharmaceutical Research*, 6(4), 1225-1231.
12. Joshi, S., Pawar, V., & Uma, V. (2011). Synthesis characterization and biological studies of Schiff base metal complexes Co (II), Zn (II) Ni (II) and Mn (II) derived from amoxicillin trihydrate and various aldehydes. *International Journal of Pharma and Bio Sciences*, 2(1), 240-250.
13. Anacona, R. J., & Marquez, V. (2013). Synthesis of Some Salicylidene-β-Lactam Antibiotics and Their Antibacterial Activity. *Latin American Journal of Pharmacy*, 32, 887-891.



14. Kaushal, R., Sheetal, & Kaushal, R. (2014). Synthesis characterization and biological screening of titanium (IV) complexes derived from antibiotic drugs. *International Journal of Pharmaceutical Sciences and Research*, 6, 374-378.
15. Naz, N., Khatoon, S., Ajaz, H., et al. (2013). Synthesis, Spectral Characterization and Biological Evaluation of Schiff Base Transition Metal Complexes Derived from Ampicillin with D-Glucose. *Asian Journal of Chemistry*, 25(4), 2239-2242.
16. AL-Mudhafar, J. M. M. (2016). Synthesis characterization and preliminary antimicrobial evaluation of new Schiff bases of ampicillin and amoxicillin derived from isatin derivatives. *International Journal of Pharmaceutical Sciences*, 8, 113-116.
17. Al-Noor, H. T., Aziz, R. M., & AL-Jeboori, T. A. (2014). Synthesis characterization and antimicrobial activities of {Fe(II), Co(II), Ni(II), Cu(II), and Zn(II)} mixed ligand complexes Schiff base derived from ampicillin drug and 4(dimethylamino)benzaldehyde with nicotinamide. *International Journal of Technical Research and Applications*, 2(4), 187-192.
18. Kaushal, R., & Thakur, S. (2013). Syntheses and Biological screening of Schiff base complexes of Titanium (IV). *Chemical Engineering Transactions*, 32, 1801-1806.
19. Al-Noor, H. T., AL-Jeboori, A. T., & Aziz, R. M. (2013). Preparation, Characterization and Antimicrobial activities of {Fe(II),Co(II),Ni(II),Cu(II),and Zn(II)}Mixed Ligand Complexes Schiff base derived from Cephalexin drug and 4(dimethylamino)benzaldehyde with Nicotinamide. *Advanced Physics Theories and Applications*, 18, 1-8.
20. Elgellal, A. A. (2014). Preparation Characterization and Biological Evaluation of Schiff-Base of Some Drug Substances. *Journal of Medical and Bioengineering*, 3, 187-189.
21. Iqbal, S. M., Bukhari, H. I., & Arif, M. (2005). Preparation, characterization and biological evaluation of copper(II) and zinc(II) complexes with Schiff bases derived from amoxicillin and cephalexin. *Applied Organometallic Chemistry*, 19, 864-869.



22. Narjis, N., Zubi, S., & Zafar, M. I. (2013). Synthesis, spectral and biological studies of novel Schiff base metal complexes derived from cephalixin and sugars. *Asian Journal of Chemistry*, 25, 297-300.
23. Al-Noor, H. T., Jarad, J. A., & Hussien, O. A. (2014). Synthesis, physico-chemical and antimicrobial properties of some metal (II) mixed ligand complexes of tridentate Schiff base derived from B-lactam antibiotic {(cephalexin monohydrate)-4-chlorobenzophenone} and saccharin. *International Journal of Chemical and Process Engineering Research*, 1, 109-120.
24. Anacona, J., Pineda, Y., Alina Bravo, A., et al. (2016). Synthesis, characterization and antibacterial activity of a tridentate Schiff base derived from cephalixin and 1,6-hexanediamine and its transition metal complexes. *Medicinal Chemistry*, 6, 467-473.
25. Joshi, S., Pawar, V., & Uma, V. (2011). Antibacterial and antioxidant properties of Mn(II), Co(II), Ni(II) and Zn(II) complex of Schiff base derived from cephalixin. *Research Journal of Pharmaceutical, Biological and Chemical Sciences*, 2(1), 61-70.
26. Srivastava, K. P., Singh, A., Singh, K. S., et al. (2014). Green and efficient synthesis, characterization and anti-bacterial activity of copper(II) complexes with unsymmetrical bidentate Schiff base ligands. *Journal of Applied Chemistry*, 7, 16-23.
27. Chohan, H. Z., Pervez, H., Khan, M. K., et al. (2004). Antifungal cobalt(II), copper(II), nickel(II) and zinc(II) complexes of furanyl-, thiophenyl-, pyrrolyl-, salicylyl- and pyridyl-derived cephalixins. *Journal of Enzyme Inhibition and Medicinal Chemistry*, 19, 85-90.
28. Iqbal, S. M., Abid, R., Sabir, M. A., et al. (1999). Preparation, characterization and biological evaluation of copper(II) and zinc(II) complexes with cephalixin. *Journal of Pharmacy and Pharmacology*, 51, 371-375.
29. Deepak, A., Chauhan, S., Verma, K. K., & Garg, S. (2017). Preparation, characterization and biological studies on pyridoxylidene-cephalexin Schiff base complexes of aryltellurium (IV). *International Journal of Chemical Sciences*, 15(4), 182.



30. Anacona, J. R., Rodriguez, J. L., Camus, J. (2014). Synthesis, characterization and antibacterial activity of a Schiff base derived from cephalixin and sulphathiazole and its transition metal complexes. *Spectrochimica Acta*, 129, 96-102.
31. Anacona, J. R., Rangel, V., Loroño, M., Camus, J. (2015, October 5). Tetradentate metal complexes derived from cephalixin and 2,6-diacetylpyridine bis(hydrazone): Synthesis, characterization and antibacterial activity. *Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy*, 149, 23-29.
32. Yaseen, A. A., Al-Tikrity, E. T., Yousif, E., Ahmed, D. S., Kariuki, B. M., & El-Hiti, G. A. (2021, September 2). Effect of ultraviolet irradiation on polystyrene containing cephalixin Schiff bases. *Polymers*, 13(17), 2982.
33. Gillies, R. R., & Dodds, T. C. (1968). *Bacteriology illustrated* (2nd ed.).
34. Lourenço, F. R., & Pinto, T. D. (2011). Antibiotic microbial assay using kinetic-reading microplate system. *Brazilian Journal of Pharmaceutical Sciences*, 47, 573-584.
35. Kelava, T., Čavar, I., & Čulo, F. (2011, October 31). Biological actions of drug solvents. *Periodicum Biologorum*, 113(3), 311-320.