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THE USE OF 2-ACETYL PYRAZINE AS A BASIC NUCLEOPHILE FOR THE PREPARATION OF NEW DERIVATIVES OF THE 5,6-DIHYDROPYRIDINE-2(1H)-YLIDENE)CYANAMIDE RING, THEIR DIAGNOSIS, AND EVALUATION OF THEIR BACTERIAL EFFICACY

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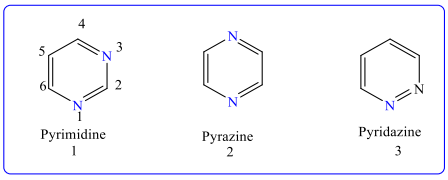
Abstract: Due to the great importance of heterocyclic rings, especially hexacyclic rings, in recent years, due to their great importance in the medical and pharmaceutical field, new derivatives of the ring of permidine, one of the heterocyclic hexacyclic rings prepared from the reaction of Chaluconate derivatives with cyanoguandine, have been prepared in this research, where the compound 2-Acetyl pyrazine was adopted as the primary nucleus in the preparation of gluconates used as intermediate compounds. The reactions took place in a base medium of sodium hydroxide. These compounds were diagnosed using physical properties such as color and melting point and spectral methods, including the infrared spectrum (FT-IR) and the proton-carbon (1H-NMR)(13C-NMR) nuclear magnetic resonance spectrum. The effectiveness of these compounds was measured against different types of gram-negative bacteria, such as (Escherichia coli) and gram-positive bacteria, such as (Staphylococcus aureus) and the use of amoxicillin as a control sample to compare with the results of inhibition of these compounds.

Key words: Chalcones, pyrimidine, Biological activity.

1. Introduction

pyrimidine They are heterocyclic compounds similar to pyridine[1], consisting of two nitrogen atoms and four carbon atoms in the ring. They are in the form of 1,3- hexacyclic heterocyclic diazines, as the location of the nitrogens [2]in the ring determines the type and name of the compound as follows:



Recently, researchers have been interested in pyrimidine compounds and their derivatives due to their importance in pharmacy[3], medicine, and industrial applications. And many previous studies have shown that it has great significance in the medical field, cytotoxic activity[4], activity Analgesics, antimicrobial activity [5][6], anti-inflammatory [7],[8], antioxidant activity [9],[10], antibacterial activity[11], and anticancer agents[12].

Chalcones are important compounds for the preparation of many heterocyclic compounds. The general formula of a chalcone consists of two aromatic rings linked through the α-β system of unsaturated carbonyl

compounds. Compounds from electrophilic and nucleophile addition reactions[13]. The importance of these compounds is due to the presence of two (active) functional groups, the double bond, and the carbonyl group, in succession. Due to the presence of these two groups, they are subject to two types of addition 1,4- on the double bond, which is Michael's addition, and Claysen's addition 1,2- on the carbonyl group, and this qualifies it to enter the reactions of preparing the nuclei of heterocyclic rings[14]. Chalcones have many uses in the fields of medicine and agriculture. They are used as antioxidants[15], antibacterial activity[16], antimicrobial activity [17], antioxidant activity[18], antileishmanial[19], and anti-cancer[20]

2. Experimental

2.1. Material: All chemicals were used through this work and purchased from Fluka, BDH Companies.

2.2. Devices used: Melting points are uncorrected and were recorded in an open capillary tube on Stuart's melting point apparatus. Infrared spectra have been recorded on a Shimadzu FTIR-8100 spectrophotometer using KBr discs–and ^{1}H-NMR Spectra have been measured on an MHZ spectrometer using DMSO-d^{6}) as solvent. TLC did reaction monitoring and verification of the purity of the compounds on silica gel-percolated alum sheets (type 60 F254 Merck, Darmstadt, Germany).

2.3. Preparation of chalcones derivatives (NA1-NA5) [21]

Dissolve (0.013 Mol 1.6,g) of (2_Acetyl pyrazine) in (6 ml) of ethanol and add to it (10 ml) of (10% NaOH) alcoholic solution while stirring, and add to it (0.013 Mol, 1.96 g) of one of the aromatic benzaldehyde compensators dissolved in (5 ml) of ethanol and leave the mixture while stirring for a period of 3-4) at a temperature of (40-20) m°, then added to the crushed ice and left in the refrigerator for 24hr, then equalized the medium using diluted HCL[21], collected the precipitate and filtered and recrystallized from absolute ethanol, where the course of the reaction was traced by TLC and table (1) shows the physical properties of compounds (NA1-NA5).

Table (1) Physical properties of prepared compounds (NA1-NA5)

Comp. No.	R	Molecular formula	m.p. °C	Yield %	Color
NA_1	4-NO_2	$C_{13}H_9N_3O_3$	130-140	87	Light yellow
NA_2	4-Cl	$C_{13}H_9N_2OCL$	150-160	85	Dark brown
NA_3	4-Br	$C_{13}H_9N_2OBr$	135-140	90	Light brown
NA_4	4-F	$C_{13}H_9N_2OF$	145-150	75	Yellow
NA_5	4-CH_3	$C_{13}H_9N_2OCH_3$	160-165	87	Off white

2.4. Preparation of 5,6-dihydropyrimidin-2(1H)-ylidene)cyanamide derivatives(NA6-NA10)[22]

(0.01 ML, 0.2 g) of galcon was dissolved in (10 ml) of (10% ethanol) in a round flask with a capacity of (100 ml), and then added to (0. 001 Mol 0.15, g) of cyanoquandine dissolved in (10 ml) of (ethanol 10%) with stirring for (10) minutes, then add (10 ml) of (NaoH 10%) to it. The mixture is escalated for (3-6hr) at a temperature of (70-110) m°; cool the solution and add to the crushed ice, equivalent to the solution, using (10% HCL). The precipitate is filtered[22], dried, and recrystallized from absolute ethanol G, where the course of the reaction is traced by (TLC) and table (2) shows the physical properties of the compounds(NA6-NA10).

Table (2) Physical properties of prepared compounds (NA6-NA10)

Comp. No.	R	Molecular formula	m.p. °C	Yield %	Color
NA_6	4-NO_2	$C_{14}H_{10}N_6O_2$	120-125	53	White

NA_7	4-Cl	$C_{14}H_{10}N_5CL$	115-120	61	Light Brown
NA_8	4-Br	$C_{14}H_{10}N_5Br$	116-118	55	Off White
NA_9	4-F	$C_{14}H_{10}N_5F$	153-155	63	Yellow
NA_{10}	4-CH_3	$C_{14}H_{10}N_5CH_3$	143-145	51	Orange

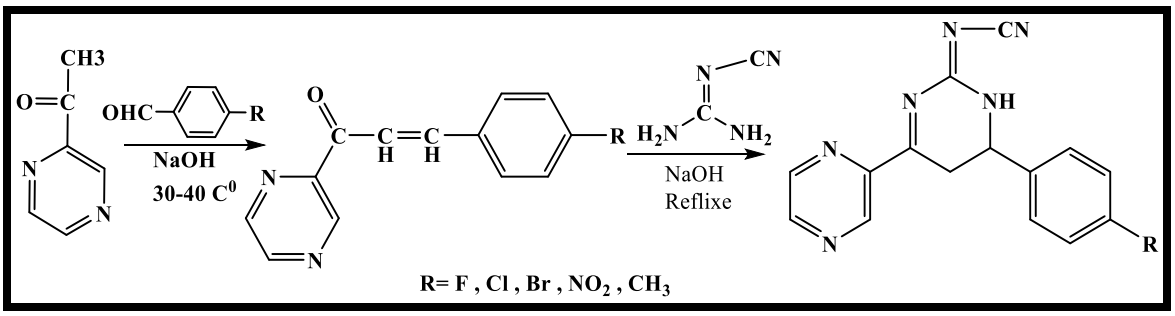
2.5. Evaluation of biological activity

The biological activity was estimated using the propagation method. In contrast, the biological activity has been assessed by the Kirby-Bauer movement[23],

[24], 0.1 ml of bacterial suspension was spread to the ager Muller Hinton dishes, and the mixture was left for 5 minutes to absorb the suspension[25],[26]. After that, holes were prepared for each dish using a Cork Porer and a diameter of (5) mm per hole (0.1 ml) of the prepared solutions of the fourth hole using (Amoxicillin) as a control sample. They incubated the dishes for (24) hours at 37. The inhibition zone diameters around each hole have been measured in millimeters, depending on the method of Prescott[27],[28].

3. Results and Discussion

The vehicles have been prepared (NA1-NA10) as shown in the following Scheme:



Scheme (1): Route of prepared compounds (NA1-NA10)

3.1. Characterization of chalcone (NA1-NA5)

When studying the infrared (IR) spectrum of compounds (NA1-NA5), it was observed that an absorption beam appeared at the range (3074 – 3039) cm-1 belonging to the aromatic sphincter (CH), with an apparent decrease in the wavenumber of the carbonyl group (C=O) ketone to appear at the range (1689-1662) cm-1 due to the sequence between the carbonyl group and the double sphincter that appeared at the range (1613 – The spectrum also showed a signal in the range (3176-3135) cm-1 belonging to the elastic (=C-H) olefin, and the appearance of an absorption beam was observed in the range (1647-1616) cm-1 belonging to the elastic (C=N) as well as the appearance of two absorption beams in the range (1558-1512) cm-1 and (1490-1454)cm-1 belong to elastic (C=C) aromatics. As shown in Table (3) and Figure (1,2), these results were comparable to what is found in the literature[29].

Table (3) results of infrared absorption of chalcones (NA1-NA5)

Comp. No.	R	IR (KBr) cm^{-1}						
		ν(C-H) Arom.	ν(C-H) Olph.	ν C=O	ν(C=C) Olph.	ν(C=C) Arom.	ν C=N	Others
NA_1	4-NO_2	3039	3136	1662	1595	1523-1485	1631	ν(N-O) 1346
NA_2	4-Cl	3045	3150	1689	1613	1548-1490	1647	ν(C-C) 744
NA_3	4-Br	3055	3165	1687	1611	1556-1483	1632	ν(C-Br) 675
NA_4	4-F	3074	3176	1674	1587	1512-1454	1616	ν(C-F) 943
NA_5	4-CH_3	3066	3135	1676	1610	1558-1470	1639	ν(C-H) 2929-2972

When studying the 1H-NMR Spectrum of the compound [NA3] using a solvent (DMSO-d6), the appearance of a second signal at the site (6.66-6.69) ppm attributed to the Proton of the group (O=C-CH) adjacent to the carbonyl group, the appearance of a second signal at the site (6.90 and 6.93) ppm attributed to the Proton of the second group (=CH) adjacent to the benzene ring, and the appearance of a single signal at the site (3.34) PPM is attributed to the Proton Group (HDO). The appearance of a signal at the site (2.49) PPM is attributed to the protons of the solvent (DMSO-D6), as in Figure (3).

When studying the 13C-NMR spectrum of the compound [N3] using a solvent (DMSO-d6), it was observed the appearance of a single signal at the site (185.64) ppm attributed to the carbon of the carbonyl group (C=O), the appearance of a single signal at the site (115.32) ppm attributed to the carbon group (=CH) adjacent to the carbonyl group, and the appearance of a single signal at the site (148.30) ppm attributed to the carbon group (=CH) adjacent to the benzene ring, the appearance of a single signal at the site (159.57) ppm attributed to the carbon group (CH3) of the permidine ring, the appearance of a single signal at the site (155.38) ppm attributed to the carbon group (CH3) of the second permidine ring, the appearance of multiple signals at (122.30-159.57) ppm is attributed to aromatic ring carbons, the appearance of a signal at the site (39.11-40.78) ppm is attributed to solvent carbon (DMSO-d6), the spectrum is shown in figures (4).

3.2. Characterization of 5,6-dihydropyridine-2(1H)-ylidene)cyanamide derivatives (NA6-NA10)

When studying the infrared (IR) spectrum of pyrimidine derivatives (NA_6-NA_{10}). Absorption beams in the range (3099-3035) cm-1 belonging to the aromatic (CH) emitter were shown in the radiation spectrum, and absorption beams were observed in the range (3378-3245) cm-1 belonging to the (NH) emitter. It was also noted the appearance of two absorption beams at the range (2935-2975) cm-1 and (2893-2844) cm-1 belonging to the automatic elastic (CH), in addition to the appearance of absorption beams at the range (2219-2123) cm-1 belonging to the elastic (CN), the appearance of an absorption beam at the range (1659-1629) cm-1 belonging to the elastic (C=N), and the appearance of two absorption beams at the range (1568-1529) cm-1 and (1515-1452) cm-1 belong to the elastic (C=C) aromatics, and the absorption beam showed at the range (1275-1234) cm-1 belongs to the elastic (C-N) group. Note Table (4) and Figure (5). These packages were close to what exists in the literature[30].

Table (4) Infrared absorption results of 2 - Cyanamide pyrimidine compounds (NA6-NA10)

Comp. No.	R	IR (KBr) cm^{-1}							
		ν(C-H) Arom.	ν(N-H)	ν(C-H) Aliph	ν(C-N)	ν(C=N)	ν(C=C) Arom.	ν(C-N)	Others
NA_6	4-NO_2	3048	3359	2945 2855	2206	1645	1551 1498	1274	ν(N-O) 1335
NA_7	4-Cl	3035	3350	2975 2850	2219	1637	1568 1485	1240	ν(C-C) 742
NA_8	4-Br	3050	3378	2935 2893	2185	1659	1560 1493	1234	ν(C-Br) 650
NA_9	4-F	3099	3345	2950 2844	2123	1629	1529 1452	1245	ν(C-F) 941
NA_{10}	4-CH_3	3043	3325	2940 2852	2173	1627	1557 1515	1237	ν(C-N)

When studying the 1H-NMR spectrum of the compound [N9], the appearance of a multiple signal at the range (7.01-7.94) ppm attributed to the protons of the aromatic ring, the appearance of a single signal at the position (1.17) ppm attributed to the Proton group (NH), the appearance of a triple signal at the range (3.73-3.78) ppm attributed to the Proton Group (CH) aliphatic, as well as the appearance of a binary signal at the positions (3.11, 3.13) PPM is attributed to the Proton Group (CH2), and the appearance of a signal at the site (2.49) ppm is attributed to the protons of the solvent (DMSO-D6). The spectrum is shown in the figure. (6).

When studying the 13C-NMR spectrum of the compound [N9], the appearance of an in-situ signal (152.78) ppm attributable to the carbon Group (C=N) adjacent to the (CN) Group, the appearance of an in-situ signal (163.85) ppm attributable to the carbon Group (C=N) pyrimidine ring, the appearance of an in-situ signal (119.94) ppm attributable to the carbon group (CN), as well as the appearance of 115.94-160.08) ppm attributed to aromatic ring Carbons, as well as the appearance of a signal in situ (62.10) ppm attributed to the Carbon Group (CH) of the permidine ring adjacent to the aromatic benzene ring, the appearance of a signal in situ (57.51) ppm attributed to the carbon group (ch2) of the permidine ring, the appearance of signals At the range (39.34-40.36) ppm is attributed to the solvent carbonate (DMSO-d6), as in Figure (7).

3.3. Evaluation of Biological Activity:

Some of the synthesized compounds (NA3, NA4, NA6, NA7, NA9) were tested against various strains of bacteria: Gram-positive bacteria Staphylococcus aureus and gram-negative bacteria Escherichia coli by cup plate agar diffusion method[31]. The microbial cultures were incubated at (37 ℃ for 8 hours) and diluted with 0.8% sterile saline[32]. The solution concentration for used drugs in DMSO was kept at 100µg/mL. Amoxicillin as a negative control was used. The biological activity was measured by measuring the inhibition diameter of the growth of bacteria around the disk in use [33]. as shown in Table (5).

Table (3): Inhibitory effectiveness of some prepared compounds (NA3, NA4, NA6, JA7, NA9) and control treatments (antibiotics) on the growth of several positive and negative bacteria

Comp. No.	Escherichia coli			Staphylococcus aureus		
	25%	50%	100%	25%	50%	100%
NA3	17	19	23	16	17	18
NA4	16	20	24	11	14	16
NA6	6	10	13	10	16	17
NA7	10	15	13	12	15	16
NA9	13	19	20	10	12	15
Amoxicillin	10	16	24	10	20	20

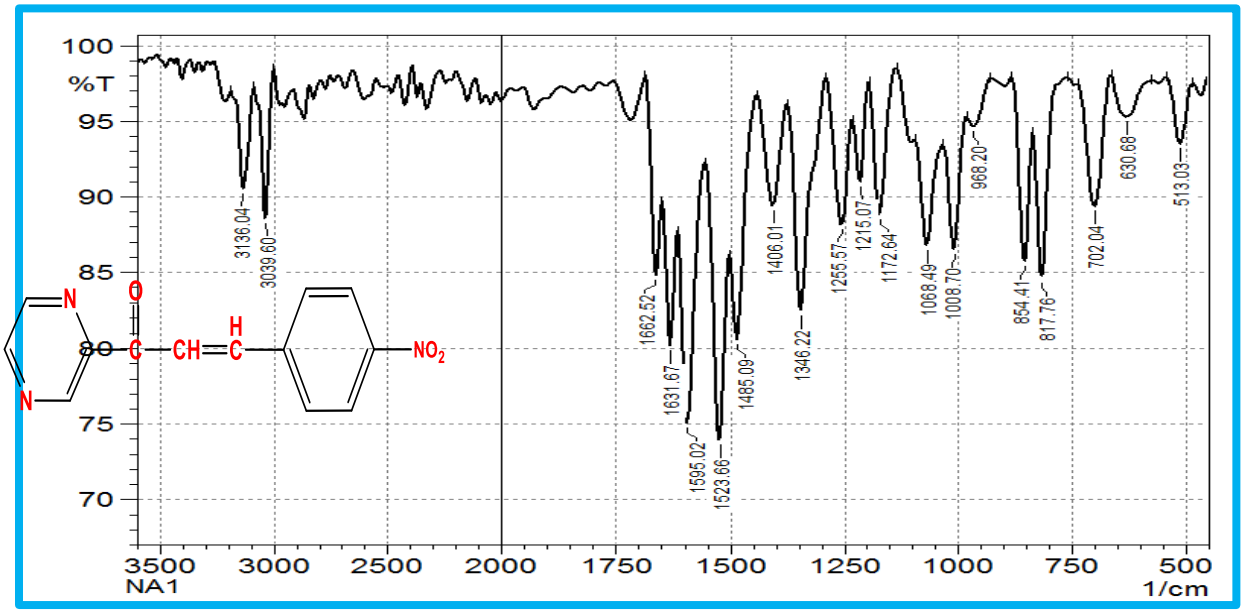


Figure (1): The infrared spectrum of the compound (NA1)

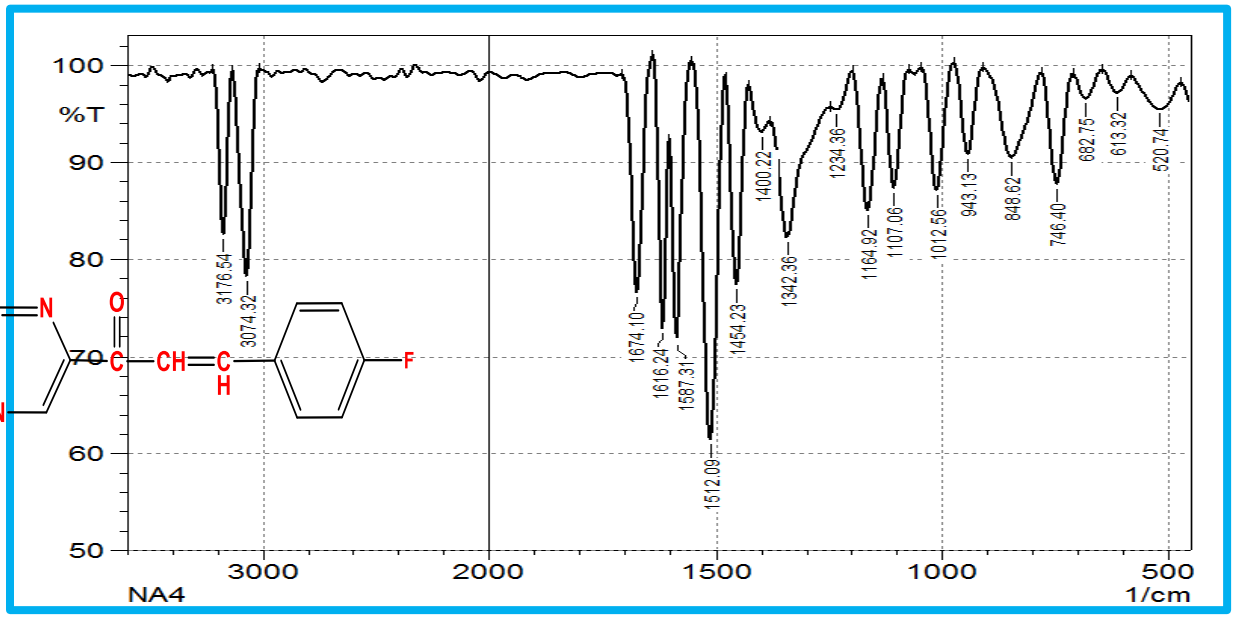


Figure (2): The infrared spectrum of the compound (NA4)

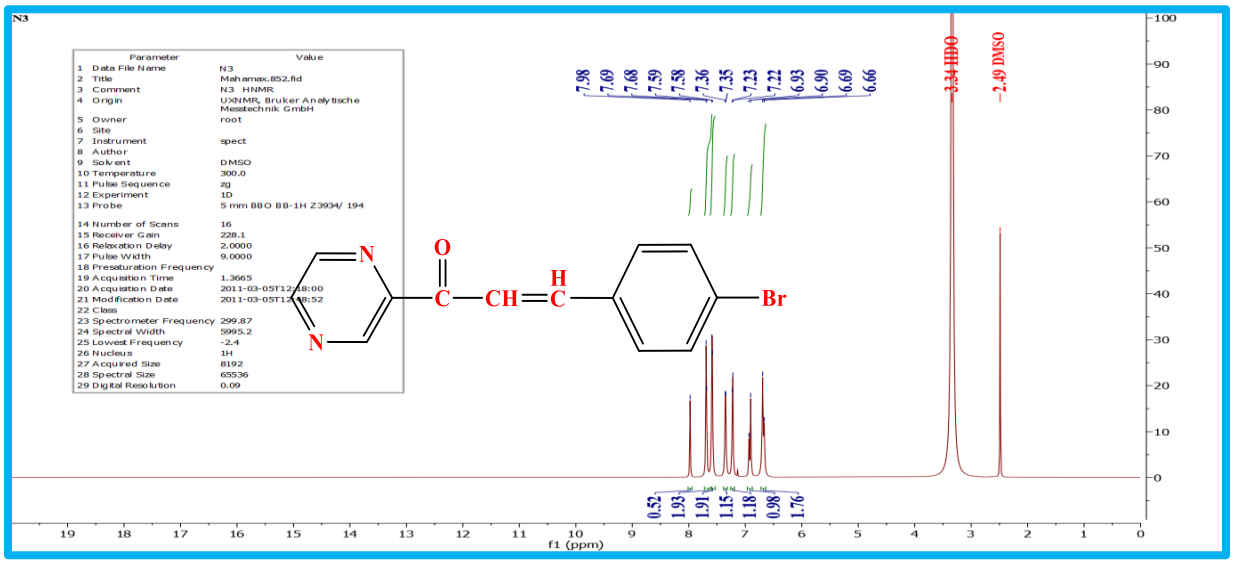


Figure (3): The ^{1}H-NMR spectrum of the compound (NA3)

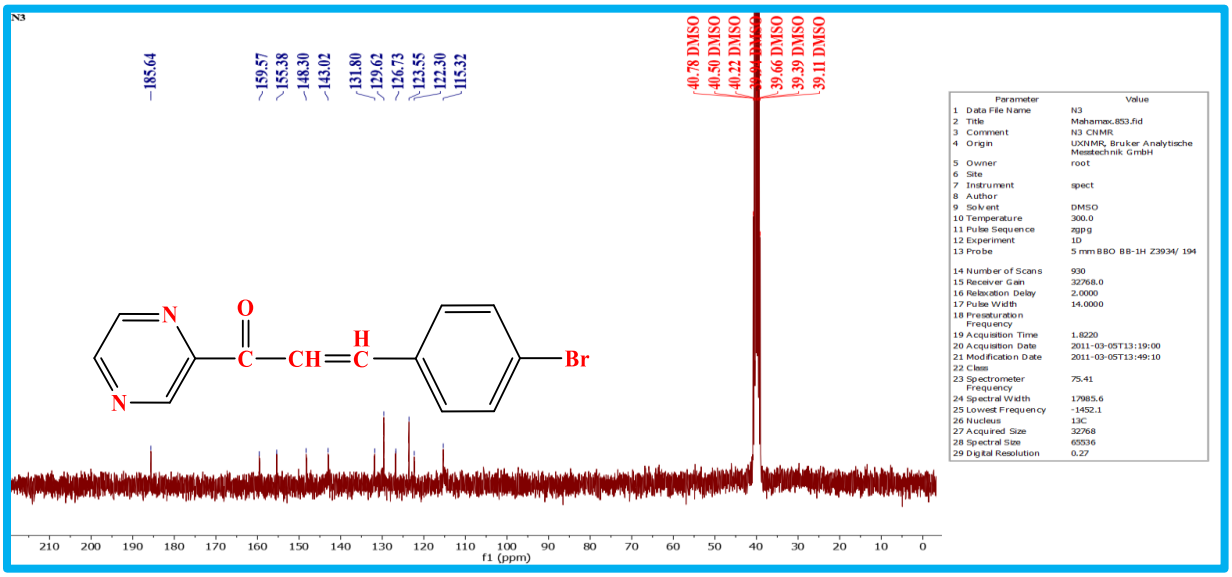


Figure (4): The ^{13}C-NMR spectrum of the compound (NA3)

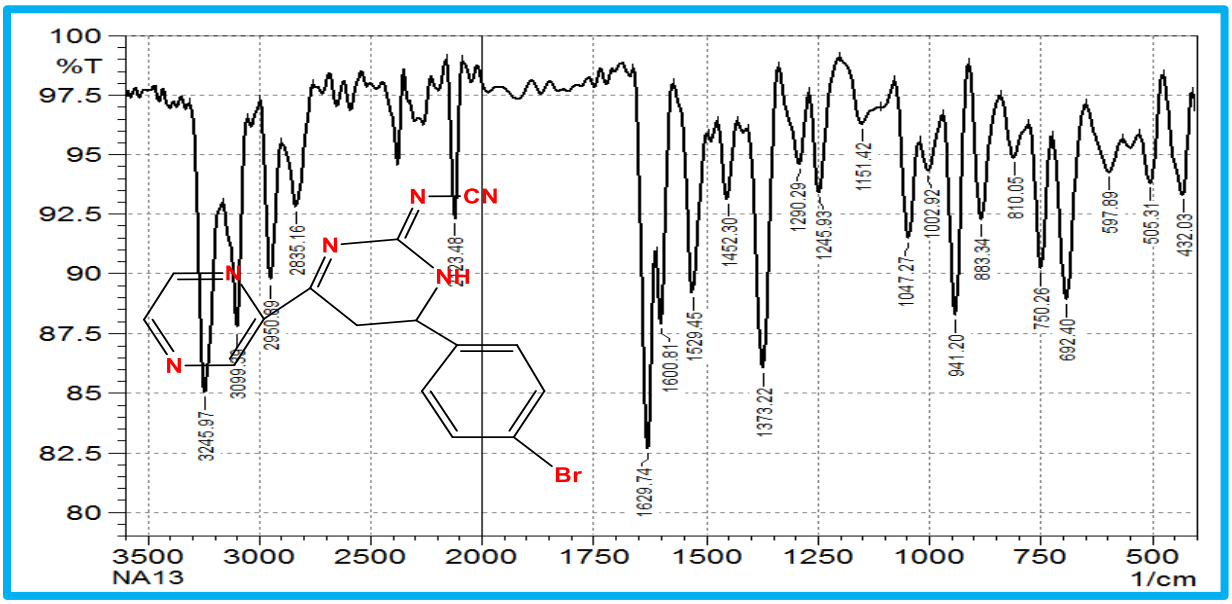


Figure (5): The infrared spectrum of the compound (NA8)

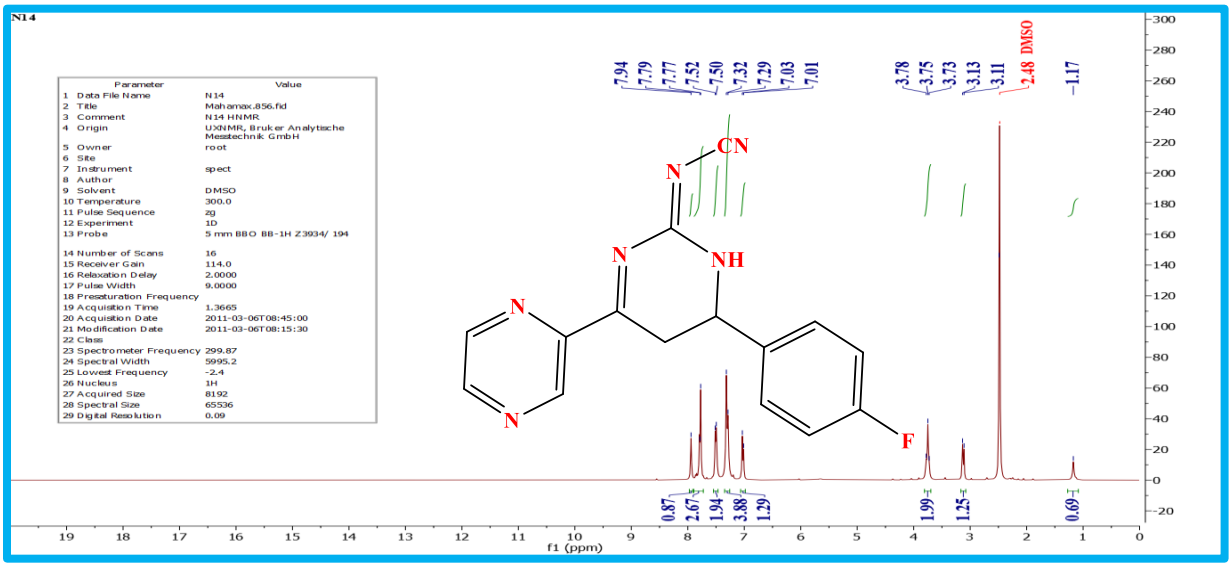


Figure (6): The ^{1}H-NMR spectrum of the compound (NA9)

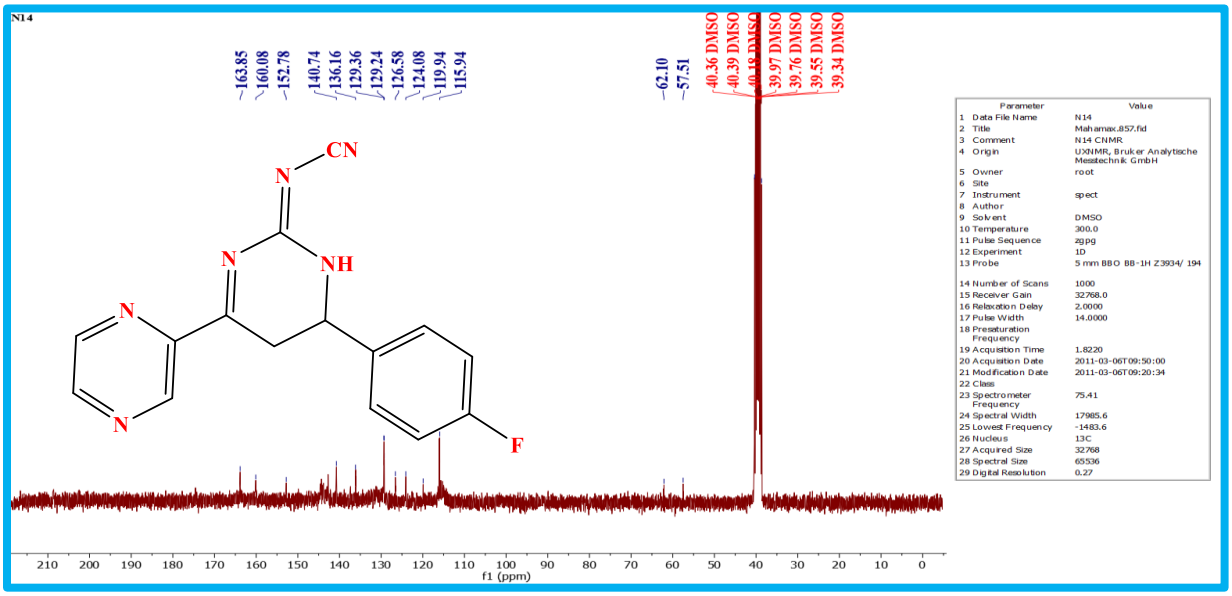
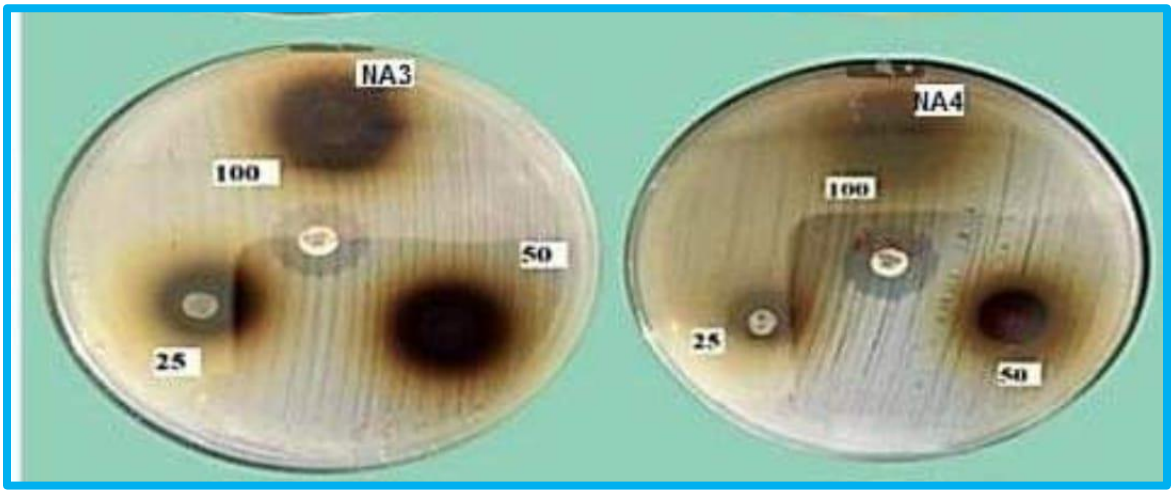


Figure (7): The ^{13}C-NMR spectrum of the compound (NA9)



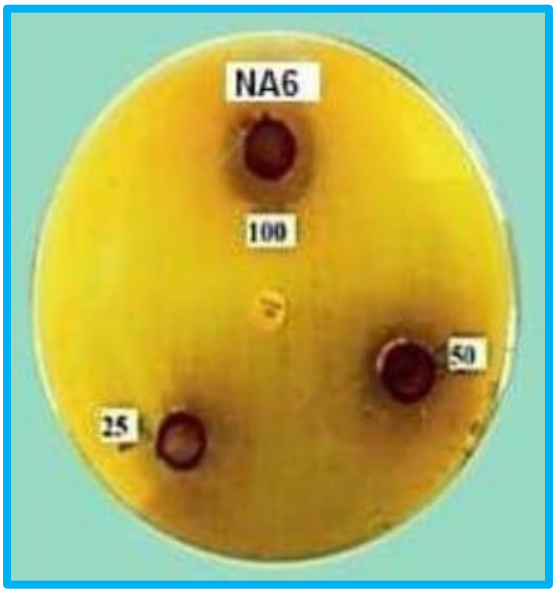


Figure (8) inhibitory effectiveness of some compounds against bacteria (Escherichia coli) and Staphylococcus aureus

4. Conclusions: The correctness and validity of the prepared compounds were ascertained by spectroscopic and physical measurements, where the infrared spectrum accurately proved the presence of active aggregates, and this confirmation increased the NMR spectrum of the proton and the carbon spectrum, which accurately agreed on the correctness of the structures of the prepared compounds. These compounds are stable at Laboratory temperature and do not decompose or change color. The prepared compounds showed high and good inhibitory activity against Gram-positive and Gram-negative bacteria, and the results were compared with those of amoxicillin, which was used as a control sample.

References:

1. T. L. Gilchrist, "Heterocyclic chemistry," *(No Title)*, 1985.
2. J. A. Joule, *Heterocyclic chemistry*. CRC Press, 2020.
3. A. Ahamed, I. A. Arif, R. Surendra Kumar, I. Akbar, and B. Shaik, "Antimicrobial and cytotoxic activities of novel pyrimidine-2, 4-dione connected with 2H-thiopyran derivatives," *J. King Saud Univ.*, vol. 35, no. 4, p. 102588, 2023.
4. T. Wang, F. Wu, L. Luo, Y. Zhang, J. Ma, and Y. Hu, "Efficient synthesis and cytotoxic activity of polysubstituted thieno [2, 3-d] pyrimidine derivatives," *J. Mol. Struct.*, vol. 1256, p. 132497, 2022.
5. N. V. Rani and R. Kunta, "PEG-400 promoted a simple, efficient and eco-friendly synthesis of functionalized novel isoxazolyl pyrido [2, 3-d] pyrimidines and their antimicrobial and anti-inflammatory activity," *Synth. Commun.*, vol. 51, no. 8, pp. 1171–1183, 2021.
6. M. A. A. Radwan, M. A. Alshubramy, M. Abdel-Motaal, B. A. Hemdan, and D. S. El-Kady, "Synthesis, molecular docking and antimicrobial activity of new fused pyrimidine and pyridine derivatives," *Bioorg. Chem.*, vol. 96, p. 103516, 2020.
7. E. H. El-Sayed and K. S. Mohamed, "Synthesis and anti-inflammatory evaluation of some new pyrazole, pyrimidine, pyrazolo [1, 5-a] pyrimidine, imidazo [1, 2-b] pyrazole and pyrazolo [5, 1-b] quinazoline derivatives containing indane moiety," *Polycycl. Aromat. Compd.*, vol. 41, no. 5, pp. 1077–1093, 2021.
8. H. Wang *et al.*, "Design and synthesis of novel indole and indazole-piperazine pyrimidine derivatives

with anti-inflammatory and neuroprotective activities for ischemic stroke treatment," *Eur. J. Med. Chem.*, vol. 241, p. 114597, 2022.

9. R. R. Gajjala *et al.*, "Ethyl-4-(aryl)-6-methyl-2-(oxo/thio)-3, 4-dihydro-1H-pyrimidine-5-carboxylates: Silica supported bismuth (III) triflate catalyzed synthesis and antioxidant activity," *Bioorg. Chem.*, vol. 129, p. 106205, 2022.
10. P. P. Mohire *et al.*, "An expedient four component synthesis of substituted pyrido-pyrimidine heterocycles in glycerol: proline based low transition temperature mixture and their antioxidant activity with molecular docking studies," *Polycycl. Aromat. Compd.*, vol. 42, no. 1, pp. 137–155, 2021.
11. N. Ahmadi, S. Z. Sayyed-Alangi, and A. Varasteh-Moradi, "Cu@ KF/clinoptilolite nanoparticles promoted green synthesis of pyrimidine derivatives: study of antioxidant activity," *Polycycl. Aromat. Compd.*, vol. 42, no. 7, pp. 4019–4033, 2022.
12. S. Dastmard, M. Mamaghani, L. Farahnak, and M. Rassa, "Facile Synthesis of Polyfunctional Indole-Pyrido [2, 3-d] Pyrimidine Hybrids Using Nickel-Incorporated Fluorapatite Encapsulated Iron Oxide Nanocatalyst and Study of Their Antibacterial Activities," *Polycycl. Aromat. Compd.*, vol. 42, no. 4, pp. 1747–1760, 2022.
13. D. W. Elias, M. A. Beazely, and N. M. Kandepu, "Bioactivities of chalcones," *Curr. Med. Chem.*, vol. 6, no. 12, p. 1125, 1999.
14. D. I. Batovska and I. T. Todorova, "Trends in utilization of the pharmacological potential of chalcones," *Curr. Clin. Pharmacol.*, vol. 5, no. 1, pp. 1–29, 2010.
15. A. Suyambulingam, S. Nair, and K. Chellapandian, "Synthesis, spectral characterization of novel chalcones based oxazines derivatives and screening of their antimicrobial and antioxidant activity," *J. Mol. Struct.*, vol. 1268, p. 133708, 2022.
16. C. H. P. Kumar, M. S. Katagi, and B. P. Nandeshwarappa, "Novel synthesis of quinoline chalcone derivatives-Design, synthesis, characterization and antimicrobial activity," *Chem. Data Collect.*, vol. 42, p. 100955, 2022.
17. A. Siddiqa *et al.*, "Synthesis, molecular docking, bio-evaluation and quantitative structure activity relationship of new chalcone derivatives as antioxidants," *J. Mol. Struct.*, vol. 1277, p. 134814, 2023.
18. J. Aucamp and D. D. N'Da, "In vitro antileishmanial efficacy of antiplasmodial active aminoquinoline-chalcone hybrids," *Exp. Parasitol.*, vol. 236, p. 108249, 2022.
19. S. Patel, N. Challagundla, R. A. Rajput, and S. Mishra, "Design, synthesis, characterization and anticancer activity evaluation of deoxycholic acid-chalcone conjugates," *Bioorg. Chem.*, vol. 127, p. 106036, 2022.
20. J. N. Saleh and A. Khalid, "Synthesis, Characterization and Biological Activity Evaluation of Some New Pyrimidine Derivatives by Solid Base Catalyst AL2O3-OBa," *Cent. Asian J. Med. Nat. Sci.*, vol. 4, no. 4, pp. 231–239, 2023.
21. S. A. Mohamed, K. A. Al-Badrany, and M. S. Huseen, "PREPARATION AND STUDY OF BIOLOGICAL ACTIVITY OF PYRIMIDINE COMPOUNDS DERIVED FROM 2-Acetyl pyrazine," *Vegueta. Anu. la Fac. Geogr. e Hist.*, vol. 22, no. 8, 2022.
22. W. M. Al-Joboury, K. A. Al-Badrany, and N. J. Asli, "N-alkylation of substituted 2-amino benzothiazoles by 1, 4-bis (bromo methyl) benzene on mixed oxides at room temperature and study their biological activity," in *AIP Conference Proceedings*, AIP Publishing, 2022.

23. O. A. F. Al-Hadidi, K. A. Al-Badrany, and S. A. Al-Bajari, “Synthesis some of thiazepine compounds from 2-carbboxyaldehyde-5-methyl thiophene and study their biological activity on infected male rats epileptic,” *J. Educ. Sci. Stud.*, vol. 2, no. 20, 2022.

24. M. J. Saleh and K. A. Al-Badrany, “Preparation, Characterization of New 2-Oxo Pyran Derivatives by AL2O3-OK Solid Base Catalyst and Biological Activity Evaluation,” *Cent. Asian J. Med. Nat. Sci.*, vol. 4, no. 4, pp. 222–230, 2023.

25. A. W. A. S. Talluh, “Preparation, Characterization, Evaluation of Biological Activity, and Study of Molecular Docking of Azetidine Derivatives,” *Cent. Asian J. Med. Nat. Sci.*, vol. 5, no. 1, pp. 608–616, 2024.

26. A. Talluh, J. N. Saleh, and M. J. Saleh, “Preparation, Characterization and Evaluation of Biological Activity and Study of Molecular Docking of Some New Thiazoli-dine Derivatives,” 2024.

27. A. W. A. S. Talluh, M. J. Saleh, J. N. Saleh, K. Al-Badrany, and H. mohammed saleh Al-Jubori, “Preparation, characterization, and evaluation of the biological activity of new 2, 3-dihydroquinazoline-4-one derivatives,” *Eur. J. Mod. Med. Pract.*, vol. 4, no. 4, pp. 326–332, 2024.

28. A. Khalid, A. A. Ahmed, and M. R. Wissam, “Synthesis and characterization of some azetidine Derivatives that derived from 6-Bromo-4-Fluoro-2-Aminobenzothiazole and Study Their biological activity,” *J. Educ. Sci. Stud.*, vol. 2, no. 20, 2022.

29. D. M. Najim, K. A. Al-badrany, and M. K. Saleh, “Synthesis of some new oxazepine compounds derived from cyanoethyl acetate and study their inhibitory activity against some pathogenic bacterial species”.

30. W. M. R. Al-Joboury, K. A. Al-Badrany, and N. J. Asli, “Synthesis of new azo dye compounds derived from 2-aminobenzothiazole and study their biological activity,” *Mater. Today Proc.*, vol. 47, pp. 5977–5982, 2021.

31. A. A. M. Al Rashidy, K. A. Al Badrany, and G. M. Al Garagoly, “Spectrophotometric determination of sulphamethoxazole drug by new pyrazoline derived from 2, 4-dinitro phenyl hydrazine,” in *Materials Science Forum*, Trans Tech Publ, 2020, pp. 350–359.

32. S. A. Mohamed, M. S. Hussein, and K. A. Al-badrany, “Synthesis and characterization of pyrazolines and oxazapine derivatives using chalcones as precursor and evaluation of their biological activity,” *Samarra J. Pure Appl. Sci.*, vol. 4, no. 4, 2022.