The Synthesis and Characterization Of Several Heterocyclic Compounds Derived From Chalcone Derivatives, As Well As The Investigation Of Their Biological Activity

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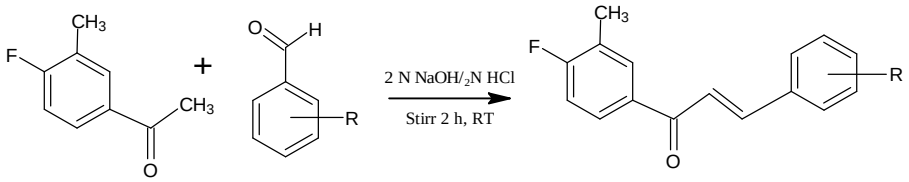
Abstract
In this work, 4-fluoro-3-methylacetophenone derivatives (**1a-e**) were prepared via aldol condensation with 4-fluoro-3-methylacetophenone and various aromatic aldehydes (**a-e**) in the presence of a base in ethanol to yield chalcones derivatives (**1a-e**), which are a valuable component for the synthesis of a wide variety of heterocyclic derivatives via reactions with hydrazine to yield pyrazole derivatives (**2a-e**), react with hydroxylamine hydrochloride to get isoxazole (**3a-e**), also react with urea to get oxazine derivatives (**4a-e**). All these compounds are characterized using FT-IR, ^{1}H-NMR, and mass, and follow reaction by TLC and measurements of melting points. After that, we analyzed the compounds' biological activity against two different types of bacteria.

Keywords: Chalcone, Pyrazole, Isoxazole, Pyrimidine, Biological activity

Introduction:
Chalcones are a type of flavonoid, and flavonoids have a variety of essential applications in the field of medicine. These compounds have been found to exhibit a wide variety of biological actions, including antimalarial, antioxidant, anticancer, antitumor, and antibacterial properties [1-4].
Pyrazoles are important members of heterocyclic compounds because they feature two nitrogen atoms that are adjacent to one another in a five-member ring structure. Pyrazole derivatives have good pharmacological effects or have biological activities such as anti-inflammatory, antibacterial, anticancer, and antifungal [5-7]. Isoxazole derivatives are well known for their value to medicine; they also exhibit a wide range of biological activities, including those that are analgesic, anticancer, antibiotic, anti-inflammatory, and anti-inflammatory, as well as antioxidant, antibacterial, and antifungal (8-11).

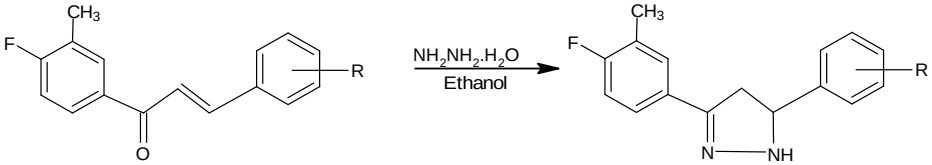
Materials:
High-purity synthetic chemical reagents and solvents were purchased from reputable vendors and dried or purified according to industry standards for use in this study. The uncorrected melting points of the synthetic compounds were determined using open capillary tubes. TLC, with silica gel as the stationary phase and pet ether and ethyl acetate as the mobile phases, was employed to confirm the reaction progress and product conversion. A Brucker FT-IR spectrometer and a Brucker 400 MHz spectrometer were used to acquire the infrared and nuclear magnetic resonance spectra, respectively.

Chalcone Synthesis General Method:
In 50 ml of ethanol, dissolved 0.03 mol each of 4-fluoro-3-methylacetophenone (**1**) and benzaldehyde derivatives (*o*-iodobenzaldehyde, *m*-iodobenzaldehyde, *p*-iodobenzaldehyde, 2,3-diiodobenzaldehyde, and 2, 4-diiodobenzaldehyde). After vigorously swirling the aforesaid mixture for 30 minutes, 10ml of 2N NaOH solution was added drop by drop. The mixture was let to stand for 8 hours before being neutralized with 2N HCl to bring about the precipitation. The resulting precipitate was air-dried and re-crystallized using ethanol as the solvent [12].



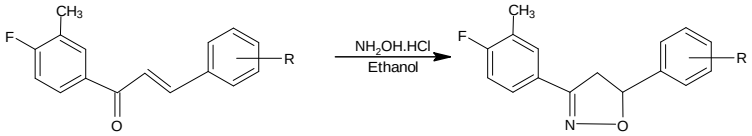
Scheme 1: Chalcone Synthesis General Method

Pyrazole Derivatives (2a-e) Synthesis:
The chalcone (**1a-e**) (0.005 mol) and hydrazine hydrate (0.5 ml) were mixed in ethanol (25 mL), and the mixture was allowed to reflux for 3 hours. After the mixture from the reaction was cooled, it was filtered, dried, and recrystallized from the ethanol solvent [12].



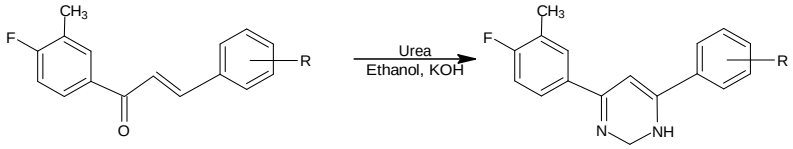
Scheme 2: Pyrazole Derivatives (**2a-e)** Synthesis

Synthesis of Isoxazole Derivatives (3a-e):
A mixture of Chaconne (**1a-e**) (0.005 mol), hydroxylamine hydrochloride (0.005 mol), and aqueous sodium hydroxide (2N, 0.5 ml) was dissolved in 30 mL of ethanol. The resulting solution was then refluxed for a duration of 5-6 hours. After refluxing, the solution was poured into ice-cold water and subsequently filtered. The obtained precipitate was washed and recrystallized using ethanol. This recrystallization process yielded isoxazole derivatives (**3a-e**) [13].



Scheme 3: Isoxazole Derivatives (**3a-e**) Synthesis

Synthesis of Oxazine Derivatives (4a-e):
A solution was prepared by dissolving a mixture of chaconne (0.003 mol) and urea (0.003 mol) in 25 mL of ethanol sodium hydroxide solution. The resulting solution was agitated for a duration of 12 hours. Subsequently, the solution was put into 50 mL of cold water while continuously stirring for a period of 30 minutes. The resulting mixture was then left undisturbed overnight. The resulting solid underwent filtration, followed by a washing process and then recrystallization from ethanol [14].



Scheme 4: Oxazine Derivatives (**4a-e**) Synthesis

Preparation of Microbiology Culture Media:
Dissolve 28 g of nutritional agar in 1 L of distillation water, then sterilize in an autoclave for 15 minutes at 121 °C. At 37 °C, the media is poured into Petri dishes that have been prepared for bacterial streaking. Escherichia coli and staphylococcus aureus were among the hospital-isolated pathogens being used. DMF was used as a solvent to produce solutions for the various compounds investigated (0.02 g of compounds in 5mL DMF), and then the inhibitory zones were examined for all of the compounds under test [15]. The bacteria were cultivated, and the plates were incubated at 37 °C for 24 hours.

Results and Discussion:
FT(IR) Sepctra:
The infrared spectrum data of compounds **1a-b** exhibited characteristic bands within specific regions. These regions include 1629-1648 cm^{-1}, which corresponds to the (C=O) stretching vibrations of the chaconne group. Additionally, bands were observed at 3244-3190 cm^{-1}, indicating the presence of (Ar-H) aromatic hydrogen stretching vibrations. The region 1325-1345 cm^{-1} corresponds to the (C-I) stretching vibrations of the iodo group. The methine group (-CH=) stretching vibrations were observed within the range of 2928-2993 cm^{-1}. Furthermore, the (C-H) stretching vibrations of the methyl group (CH_3) were observed between 3048-3112 cm^{-1}. Aromatic (C=C) stretching vibrations were detected at 1591-1596 cm^{-1}, while aliphatic (C=C) stretching vibrations were observed within the range of 1546-1550 cm^{-1}.
The infrared spectrum data of compounds (**2a-e**) exhibited characteristic bands in specific regions. These included a band at 1644-1661 cm^{-1}, indicating the presence of a (C=N) pyrazol group. Another band appeared at 3094-3099 cm^{-1}, indicating the presence of a (Ar-H) group. Additionally, a band at 3226-3230 cm^{-1} indicated the presence of a (N-H) group in imidazole, which overlapped with the band for (N-H) pyrazol that appeared at 3390-3395 cm^{-1}. Furthermore, bands at 1325-1340 cm^{-1} were observed, indicating the presence of a (C-I) group. Bands at 2986-2990 cm^{-1} and 2924-2975 cm^{-1} were indicative of (C-H) groups in (CH_3) and (-CH=) respectively. Finally, a band at 1565-1598 cm^{-1} was observed, suggesting the presence of aromatic (C=C) bonds.
The infrared spectrum data of compounds (**3a-e**) revealed the presence of specific bands within certain regions. These bands were observed at 1665-1692 cm^{-1}, indicating the presence of the (C=N) isoxazol functional group. Additionally,

bands at 3088-3095 cm^{-1} were observed, indicating the presence of (Ar-H) aromatic hydrogen groups. Bands at 2975-2981 cm^{-1} were observed, indicating the presence of (-CH=) methine groups. The presence of (C-I) iodo groups was indicated by bands at 1344-1346 cm^{-1}. Bands at 2922-2925 cm^{-1} were observed, indicating the presence of (C-H) groups in the (CH_3) methyl group. Bands at 1411-1425 cm^{-1} were observed, indicating the presence of (N=N) groups. Aromatic (C=C) bonds were indicated by bands at 1560-1565 cm^{-1}. Lastly, bands at 1141-1452 cm^{-1} were observed, indicating the presence of (C-O) bonds within the isoxazol ring.

The infrared spectrum data of compounds (**4a-e**) revealed specific bands at various regions. A band was observed at the region of 1652-1661 cm^{-1}, corresponding to the presence of a C=N bond in the oxazine compound. Another band was observed at 3077-3087 cm^{-1}, indicating the presence of an Ar-H bond. The range of 2911-2978 cm^{-1} was observed for the (-CH=) methine group. A distinct band at 3318-3325 cm^{-1} was observed for the (NH_2) group in the oxazine compound. Additionally, a band at 1621-1635 cm^{-1} was observed for the (C=N) bond within the imidazole ring. The range of 2923-2948 cm^{-1} was observed for the (C-H) bond in the (CH_3) group. Bands at 1342-1355 cm^{-1} and 1620-1640 cm^{-1} were observed due to the presence of (N=N) and aromatic (C=C) bonds, respectively.

1H NMR Spectra:

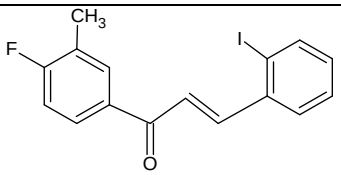
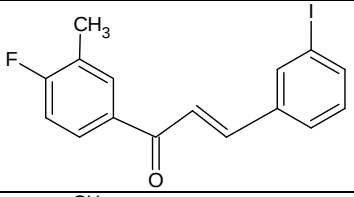
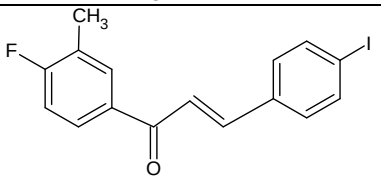
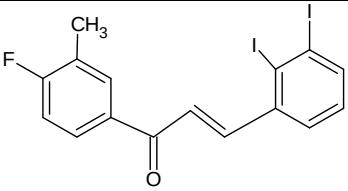
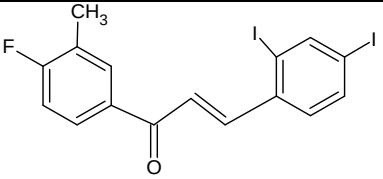
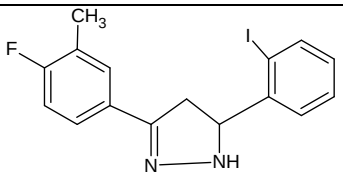
The ^1H NMR spectral data of compounds (**1a-e**) indicate chemical shifts (δ) at 6.99-7.89 (*m*, 7H, aromatic protons), 0.95-1.22 (*s*, 3H, methyl group), 8.5 (*s*, 1H, vinyl group), and 3.5 (*d*, 1H, aldehyde group). The solvent used for the NMR measurements was dimethyl sulfoxide (DMSO).

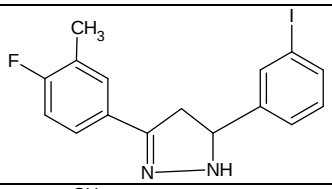
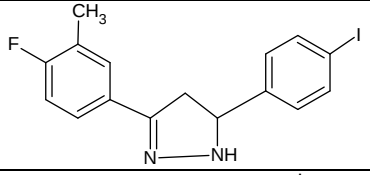
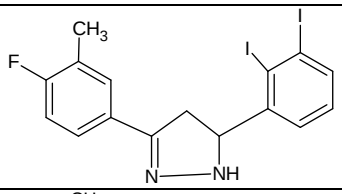
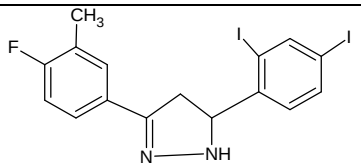
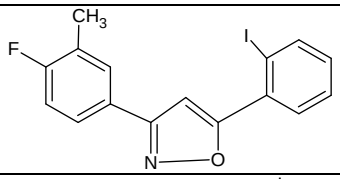
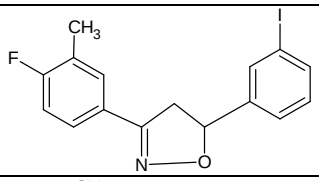
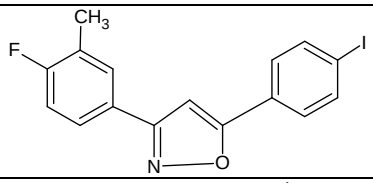
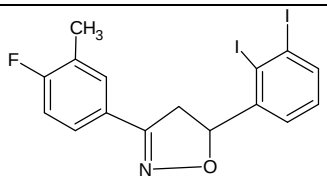
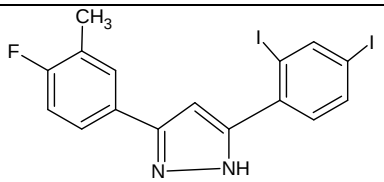
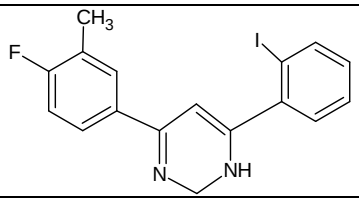
Compounds (**2a-e**) were analyzed by ^1H NMR in DMSO, and the results showed values of 6.95-8.00 (*m*, 7H, Ar-H), 0.99-1.25 (*s*, 3H, CH_3), 2.77 (*d*, 2H, CH_2 pyrazole ring), 3.83 (*t*, 1H, CH pyrazole ring), and 5.75 (*s*, 1H, NH imidazole ring).

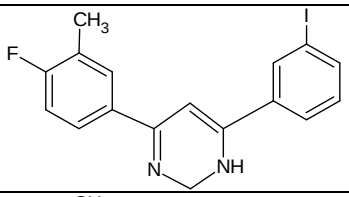
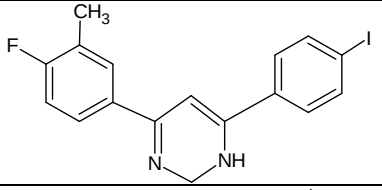
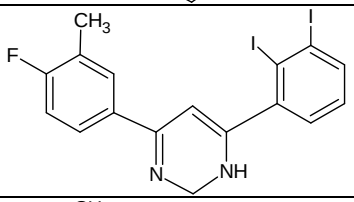
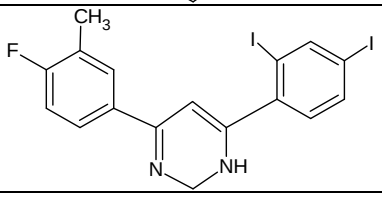
Compounds (**3a-e**) have ^1H NMR (DMSO) spectra displaying:7.05-8.04 (*m*, 7H, Ar-H), 0.95-1.11(*s*, 3H, CH_3), 11.25 (*s*, 2H, CH imidazol ring), 4.66 (*d*, 2H, CH_2 isoxazol ring), and 4.21 (*t*,1H, O-CH isoxazol ring).

^1H nuclear magnetic resonance (DMSO) spectra of compounds (**4a-e**) exhibit :6.85-7.92 (*m*, 8H, Ar-H), 1.05-1.33 (*s*, 3H, CH_3), 8.11-8.25 (*s*, 2H, CH imidazol ring), 5.75 (*s*,1H, NH_2, oxazine ring), and 3.61 (*d*, 1H, O-CH, oxazine ring).

Table 1: Physical properties of prepared compounds

Comp Code	MW	Formula	MP	Structure
1a	366	$C_{16}H_{12}FIO$	169	
1b	366	$C_{16}H_{12}FIO$	171	
1c	366	$C_{16}H_{12}FIO$	176	
1d	492	$C_{16}H_{11}FI_2O$	183	
1e	492	$C_{16}H_{11}FI_2O$	182	
2a	380	$C_{16}H_{14}FIN_2$	180	

2b	380	$C_{16}H_{14}FIN_2$	183	
2c	380	$C_{16}H_{14}FIN_2$	178	
2d	506	$C_{16}H_{13}FI_2N_2$	192	
2e	506	$C_{16}H_{13}FI_2N_2$	189	
3a	381	$C_{16}H_{13}FION$	177	
3b	381	$C_{16}H_{13}FION$	180	
3c	381	$C_{16}H_{13}FION$	189	
3d	507	$C_{16}H_{12}FI_2ON$	195	
3e	507	$C_{16}H_{12}FI_2ON$	196	
4a	392	$C_{17}H_{14}FIN_2$	177	

4b	392	$C_{17}H_{14}FIN_2$	180	
4c	392	$C_{17}H_{14}FIN_2$	189	
4d	518	$C_{17}H_{13}FI_2N_2$	199	
4e	518	$C_{17}H_{13}FI_2N_2$	203	

Conclusion:

From the results of the aforementioned studies, it can be deduced that the synthesized compounds exhibit noteworthy antibacterial efficacy against the bacteria *Staphylococcus aureus* and *Escherichia coli*. The compounds that demonstrated substantial activity against *Staphylococcus aureus* are **1a, 1c, 1e, 2a, 2b, 2d, 2e, 3b, 3c, 3d, 4a**, and **4b**. Conversely, compounds **1b, 1d, 1e, 2b, 2c, 2d, 2e, 3a, 3c, 3e, 4b**, and **4d** exhibited commendable activity against *Escherichia coli*.

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