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Synthesis and Characterization of New 5-Fluoroisatin-Chalcone Conjugates with Expected Antimicrobial Activity

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Abstract

Pathogenic microorganisms are becoming more and more resistant to antimicrobial agents. So the synthesis of new antimicrobial agents is very important. In this work, new 5-fluoroisatin-chalcone conjugates **5(a–g)** were synthesized based on previous research that showed the modifications of the isatin moiety led to the synthesis of many derivatives that have antimicrobial activity. 4-aminoacetophenone reacts with 5-fluoroisatin to form Schiff base **(3)**, which in turn reacts with two different groups of aromatic (carbocyclic and heterocyclic) aldehydes **4(a–g)** separately to form the final compounds **5(a–g)**. Proton-nuclear magnetic resonance (^{1}H-NMR) and Fourier-transform infrared (FT-IR) spectroscopy were used to confirm the chemical structures of the newly prepared compounds. Finally, the final compounds, 5-fluoroisatin-chalcone conjugates 5**(a–g)**, were screened for their antimicrobial activities and compared with three different references: vancomycin, ciprofloxacin, and fluconazole. They appeared to be good candidates as antibacterial agents against *E. coli* and *S. aureus* as well as antifungal agents against *C. albicans*. In general and for comparison, the antifungal activity of the final compounds **5(a–g)** was more potent than their antibacterial activity. Finally, for the antimicrobial activity, the most active compound of these series was compound **5e,** while compound **5g** was the least active one.

Keywords: 4-Aminoacetophenone, 5-fluoroisatin, Antimicrobial activity, Aromatic aldehydes, Chalcone, Schiff bases.

تخليق وتوصيف مقترنات جديدة من ٥-فلورو ايساتين-جالكون مع نشاط متوقع كمضادة للميكروبات

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الخلاصة

أصبحت الكائنات الحية الدقيقة المسببة للأمراض أكثر مقاومة للمركبات المضادة للميكروبات. لذا فإن تحضير مركبات جديدة مضادة للميكروبات اصبح مهم للغاية. في هذا العمل ، تم تصنيع مقترنات جديدة من ٥-فلورو ايساتين-شالكون **5(a–g)** بناءً على بحوث سابقة أظهرت أن التعديلات على جزيئة الايزاتين ادت إلى تخليق العديد من المشتقات التي لها نشاط مضاد للميكروبات. يتفاعل ٤-امينو اسيتوفينون(١) مع ٥-فلورو ايزاتين(٢) لتكوين قاعدة شيف (٣) ، والتي تتفاعل بدورها مع مجموعتين مختلفتين من الألدهيدات العطرية (الحلقية الكربوهيدراتية والحلقية الغير المتجانسة) **4(a–g)** بشكل منفصل لتشكيل المركبات النهائية **5(a–g)**. تم استخدام الرنين المغناطيسي النووي البروتوني (^{1}H-NMR) و مطيافية الأشعة تحت الحمراء باستخدام تحويل فوريه (FT-IR) لتأكيد البنية الكيميائية للمركبات المصنعة حديثًا. و اخيراً تم فحص المركبات النهائية ، مقترنات ٥-فلورو ايساتين-شالكون **5(a–g)**، بحثًا عن أنشطتها المضادة للميكروبات ومقارنتها مع مراجع قياسية (فانكومايسين، سيبروفلوكساسين، فلوكونازول). ولقد ظهرت كمركبات مرشحة للعمل ضد جراثيم الإشريكية القولونية *(E. coli)* والمكورات العنقودية الذهبية *(S. aureus)* وكذلك كمركبات مرشحة كمضادة للفطريات ضد *المبيضة البيضاء* (C. 1albicans). بصورة عامة و من باب المقارنة، كان النشاط المضاد للفطريات للمركبات النهائية أكثر قوة من النشاط المضاد للبكتيريا. و اخيراً، بالنسبة للنشاط المضاد للمكروبات، اكثر مركب فعال في هذه السلسلة كان المركب **5e**، بينما المركب **5g** كان أقل فعالية.

الكلمات المفتاحية: ٤-أمينواسيتوفينون ، ٥-فلورو ايساتين ، النشاط المضاد للميكروبات ، الألدهيدات العطرية ، الجالكون ، قواعد شيف.

Introduction

Invasive infections of any kind, whether bacterial, fungal, or viral, are considered a major and growing public health issue. Antibiotic overuse and abuse, combined with the natural evolutionary processes of microbes, have resulted in an increase in drug resistance , in addition to other contributing factors, including a lack of research and development of novel antimicrobial agents [(1)]. In contrast to the rapid development of antibiotics between 1940 and 1962, only a few significant novel classes have been marketed since 1962 [(2)]. In recent decades, brand pharmaceutical companies have given few resources to antibiotics development, as medications treating lifestyle diseases generate more

sustainable income [3]. These are significant problems that should be handled.Therefore, considerable efforts have been made to synthesize and investigate new antimicrobial agents that are safe, effective, and broad-spectrum [4].Isatin, a derivative of indole, is a heterocyclic compound that contains a nitrogen atom and is used as a building block in drug synthesis, its IUPAC name is 1*H*-indole-2,3-dione. Erdmann and Laurent first isolated isatin as an oxidation product of indigo by using nitric and chromic acids [5]. The chemical structure of isatin is composed of two planar cyclic rings, one with six members and the other with five, there is a nitrogen atom at position 1, and two carbonyl groups at positions 2 and 3 [6]. 5-fluoroisatin is an isatin analog and differs from isatin by having a fluorine atom at position 5 [7]. In previous studies, derivatives of isatin and its analogs showed many biological activities like antimicrobial [8], anticancer [9], antiviral [10], antituberculosis [11], antioxidant [12], anticonvulsant [13] and anti-inflammatory [14]. Because of the biological activities of many halogenated compounds, particularly aromatic compounds containing fluorine, have attracted considerable attention [15]. Fluorinated isatin derivatives have become a focus in the development of new biologically active compounds [16]. Sunitinib (Sutent), shown in Figure 1, is a 5-fluoroisatin derivative that was approved by the US FDA in 2006 for the treatment of advanced renal-cell carcinoma and gastrointestinal stromal tumors [17]. In veterinary medicine, toceranib (Figure 1), which is a 5-fluoroisatin derivative, is the only approved drug for the treatment of mast cell tumors in dogs [18].

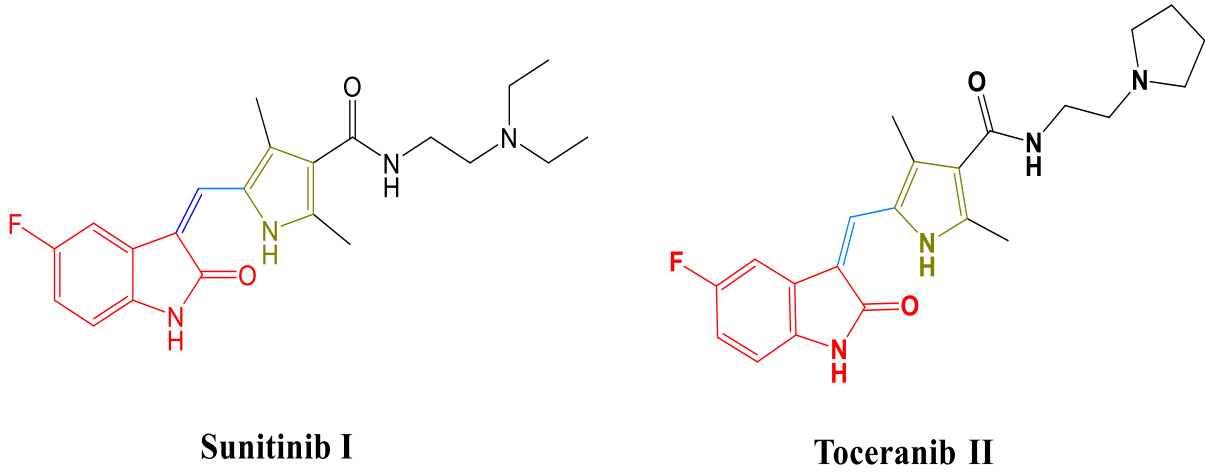


Figure 1. Structure of the 5-fluoroisatin-based approved anticancer drugs; Sunitinib I and Toceranib II [19,20]

Isatin and its derivatives are important and recommended in organic synthesis research due to the very reactive nature of the carbonyl group at the C-3 position [21]. Many new biologically active derivatives are produced by modification of the C3 carbonyl group of isatin, most commonly by nucleophilic additions such as the formation of Schiff bases or hydrazone when isatin reacts with a primary amine [22].

Chalcones are α,β–unsaturated ketones containing two fragrant rings with various arrangements of substituents that are connected by an aliphatic chain of three carbon atoms; they are an important component of natural substances as well as essential precursors for synthetic works [23]. There are two isomeric forms of chalcone, the cis and trans isomers; the trans isomer is more thermodynamically stable because it has the lowest heat of formation than the cis form, and the latter has substantial steric interactions [24]. Chalcone is usually prepared through an organic reaction called Claisen-Schmidt condensation [25]. Numerous previous studies proved that the biological activities of the drug molecules were enhanced after the introduction of the chalcone moiety, these findings are extremely significant and useful for the formulation of medications with varying biological actions [26,27]. This is primarily due to the presence of a double bond conjugated with a carbonyl group, as the removal of this functionality makes them inactive [28]. On the other hand, many derivatives of chalcone linked with isatin were synthesized and showed promising biological activities [29–31]. Different studies demonstrated the activity of isatin-chalcone derivatives as antimicrobial; Ezekwem *et al.* synthesized seven different isatin-chalcone derivatives, named 3-(3-substituted-phenyl)-*N*-(4-[2-oxo-1,2-dihydro-3*H*-indol-3-ylidene] amino} phenyl) prop-2-enamide. The antibacterial activity of the tested compound showed that the two derivatives having electron-withdrawing groups (-Cl and -Br) are the most active ones against four types of bacteria; *B. subtilis, E. coli, P. Aeruginosa*, and *S. aureus* [29]. In another study, different isatin−ferrocenyl chalcone conjugates were synthesized and evaluated for their inhibitory activity against *T. vaginalis*. It has been found that

the derivative of 5-chloroisatin- ferrocenyl chalcone is the most active one against this pathogen [31].

Based on the facts mentioned above about the problems that are associated with the increasing resistance of life-threatening microbes to existing antimicrobial agents and the lack of research for discovering and developing new effective antimicrobial agents, combined with the promising antimicrobial properties of 5-fluoroisatin and the chalcone moiety and in an attempt to synthesize new effective antimicrobial agents, the authors hypothesized generating a class of 5-fluoroisatin-chalcone conjugates by keeping the 5-fluoroisatin group constant, and diversity was created by synthesizing various substituted chalcone moieties, and the target compounds were analyzed for their antimicrobial activity.

Materials and Methods

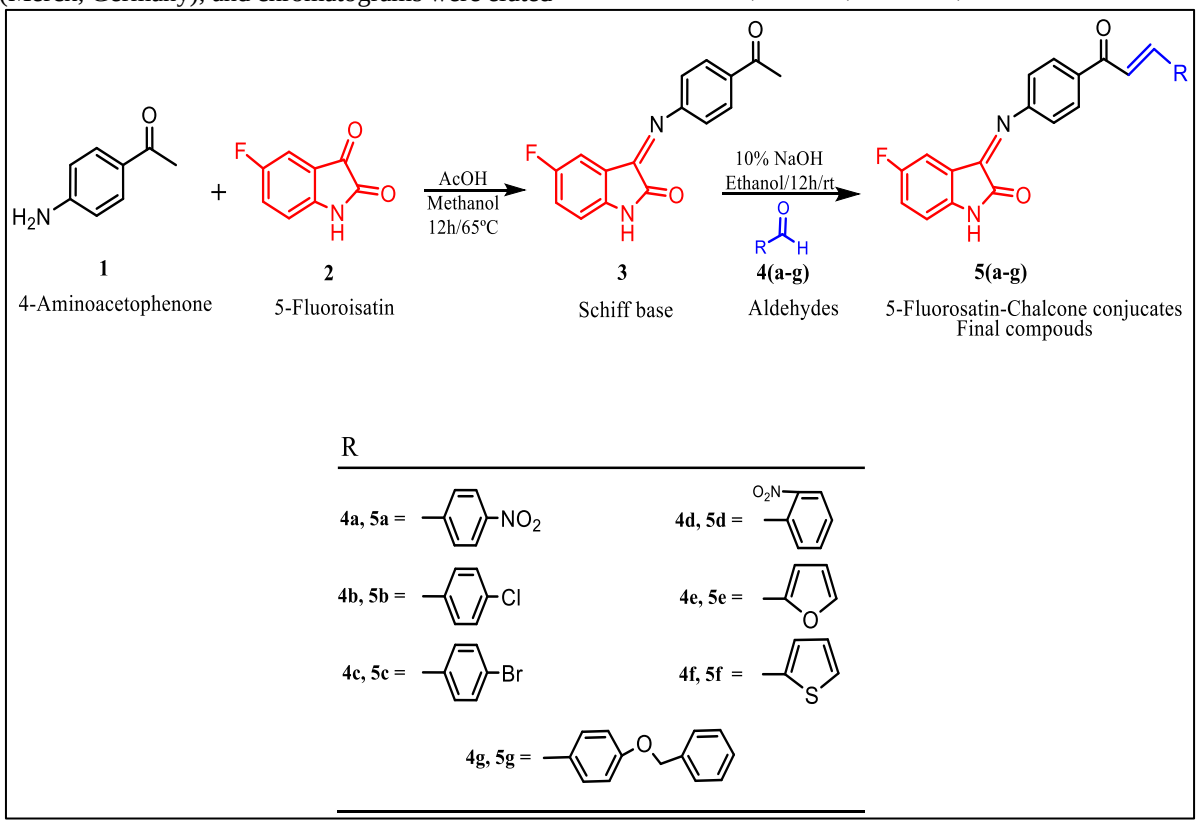
Chemicals and solvents

The solvents and chemicals used were of analytical quality and didn't need to be further purified. 5-fluoroisatin was purchased from BDH Chemicals Ltd., 4-aminoacetophenone was obtained from Alfa Aesar, Thermo Fisher Scientific/ UK, and aldehydes from Hangzhou Hyper Chemicals Limited (China). Thin-layer chromatography (TLC) was used for monitoring the reaction and checking the purity of the products by using aluminum plates (Merck, Germany), and chromatograms were eluted by using one or more of the following two mobile phases: A: toluene: ethyl acetate: n-hexane 1:2:1 and B: ethyl acetate: n-hexane 2:1. The Stuart SMP130 machine was used to measure the melting points of the used compounds by the open capillary method, and the results were used without any correction. Infrared spectra were done using the FT-IR spectrometer, Shimadzu, Japan. ^{1}H-NMR spectra were recorded on a Bruker Ascend 400 MHz NMR spectrometer, Germany.

Chemical synthesis

General Procedure for synthesis of 3-((4-acetylphenyl)imino)-5-fluoroindolin-2-one (3) [32–34]

5-fluoroisatin (2) (1.65 g, 0.01 mol.) was dissolved in 15 ml of hot methanol with a few drops of glacial acetic acid and stirred well for 10 minutes. For this mixture, 4-aminoacetophenone (1) (1.35 g, 0.01 mol.) was added, and after refluxing for 12 hours at 65 ºC, the precipitate was formed. It was filtered, and the obtained solid was recrystallized by 70% ethanol. Chemical formula; ($C_{16}H_{11}FN_2O_2$), color and appearance: red-orange powder, m.p: 233-236˚C, yield 60%; FT-IR (υ, cm^{-1}): 3248 (N-H, 5-fluorisatin), 3105 (C-H aromatic), 3040 (C-H aliphatic), 1739,1681 (for 2C=O), 1620 (N=C, imine), 1593, 1473 (C=C aromatic); ^{1}H-NMR (400 MHz, DMSO-d_6) δ 2.6 (s, 3H, CH_3), 7.07–8.10 (m, 7H, Ar-H), 11.07 (s, 1H, NH).



Scheme 1. Synthesis of 5-fluoroisatin-chalcone conjugates

General procedure for the synthesis of compounds 5(a–g) [35]

Compound (3) (2.82 g, 0.01 mol.) was dissolved in 30 ml of ethanol with 3 ml of 10% NaOH and stirred for 10 minutes. To this mixture in an ice bath, (0.01 mol.) of different aromatic aldehydes 4(a–g) were added gradually and separately. The mixture was left to stir for 30 minutes, followed by 12 hours of stirring at room temperature. The reaction was monitored using TLC. To get the precipitate, cool water was added, and the mixture was filtered, then recrystallized by 70% ethanol.

3-((4-(3-(4-nitrophenyl) acryloyl) phenyl) imino)-5-fluoroindolin-2-one (5a)

Chemical formula; ($C_{23}H_{14}FN_3O_4$), color and appearance: red powder, m.p: 285-287˚C, yield 83%; FT-IR (υ, cm^{-1}): 3231 (N-H, 5-fluoroisatin), 3113 (=C-H aromatic), 3088 (=C-H aliphatic), 1724, 1654 (for 2C=O), 1622 (N=C, imine), 1589, 1473 (C=C aromatic), 1512, 1342 (for -NO_2); ^{1}H-NMR (400 MHz, DMSO-d_6) δ 7.15–8.38 (m, 13H, Ar-H, and HC=CH), 11.10 (s, 1H, NH).

3-((4-(3-(4-chlorophenyl) acryloyl)phenyl)imino)-5-fluoroindolin-2-one (5b)

Chemical formula; ($C_{23}H_{14}ClFN_2O_2$), color and appearance: orange powder, m.p: 292-294˚C, yield 85%; FT-IR (υ, cm^{-1}): 3182 (N-H, 5-fluoroisatin), 3140 (=C-H aromatic), 3082 (=C-H aliphatic), 1720, 1658 (for 2C=O), 1624 (N=C, imine), 1589, 1569 (C=C aromatic); ^{1}H-NMR (400 MHz, DMSO-d_6) δ 7.13–8.34 (m, 13H, Ar-H, and HC=CH), 11.06 (s, 1H, NH).

3-((4-(3-(4-bromophenyl) acryloyl) phenyl)imino)-5-fluoroindolin-2-one (5c)

Chemical formula; ($C_{23}H_{14}BrFN_2O_2$), color and appearance: orange powder, m.p: 255-258˚C, yield 88%; FT-IR (υ, cm^{-1}): 3217 (N-H, 5-fluoroisatin), 3075 (=C-H aromatic), 3058 (=C-H aliphatic), 1728, 1658 (for 2C=O), 1620 (N=C, imine), 1589, 1473 (C=C aromatic); ^{1}H-NMR (400 MHz, DMSO-d_6) δ 7.15–8.36 (m, 13H, Ar-H, and HC=CH), 11.10 (s, 1H, NH).

3-((4-(3-(2-nitrophenyl) acryloyl) phenyl) imino)-5-fluoroindolin-2-one (5d)

Chemical formula; ($C_{23}H_{14}FN_3O_4$), color and appearance: orange powder, m.p: 154-156˚C, yield 72%; FT-IR (υ, cm^{-1}): 3232 (N-H, 5-fluoroisatin), 3209 (=C-H aromatic), 3095 (=C-H aliphatic), 1724, 1662 (for 2C=O), 1616 (N=C, imine), 1593, 1477 (C=C aromatic), 1523,1334 (for -NO_2); ^{1}H-NMR (400 MHz, DMSO-d_6) δ 7.15–8.35 (m, 13H, Ar-H, and HC=CH), 11.06 (s, 1H, NH).

3-((4-(3-(furan-2-yl) acryloyl) phenyl) imino)-5-fluoroindolin-2-one (5e)

Chemical formula; ($C_{21}H_{13}FN_2O_3$), color and appearance: orange powder, m.p: 199-201˚C, yield 77%; FT-IR (υ, cm^{-1}): 3232 (N-H, 5-fluoroisatin), 3124 (=C-H aromatic), 3094 (=C-H aliphatic), 1747, 1658 (for 2C=O), 1620 (N=C, imine), 1597, 1473 (C=C aromatic); ^{1}H-NMR (400 MHz, DMSO-d_6) δ 6.70–8.25 (m, 12H, Ar-H, and HC=CH), 11.09 (s, 1H, NH).

3-((4-(3-(thiophen-2-yl) acryloyl) phenyl)imino)-5-fluoroindolin-2-one (5f)

Chemical formula; ($C_{21}H_{13}FN_2O_2S$), color and appearance: red powder, m.p: 222-224˚C, yield 71%; FT-IR (υ, cm^{-1}): 3228 (N-H, 5-fluoroisatin), 3101 (=C-H aromatic), 3055 (=C-H aliphatic), 1724, 1651 (for 2C=O), 1620 (N=C, imine), 1593, 1473 (C=C aromatic); ^{1}H-NMR (400 MHz, DMSO-d_6) δ 7.12–8.28 (m, 12H, Ar-H, and HC=CH), 11.02 (s, 1H, NH).

3-((4-(3-(4-(benzyloxy) phenyl) acryloyl)phenyl) imino)-5-fluoroindolin-2-one (5g)

Chemical formula; ($C_{30}H_{21}FN_2O_3$), color and appearance: orange powder, m.p: 142-144˚C, yield 79%; FT-IR (υ, cm^{-1}): 3259 (N-H, 5-fluoroisatin), 3059 (=C-H aromatic), 3035 (=C-H aliphatic), 1747, 1658 (for 2C=O), 1627 (N=C, imine), 1597, 1508 (C=C, aromatic);^{1}H-NMR (400 MHz, DMSO-d_6) δ 5.20 (s, 2H, OCH_2-) 7.18–8.32 (m, 18H, Ar-H, and HC=CH), 11.09 (s, 1H, NH).

In vitro antimicrobial evaluation

By using the well-diffusion technique, the newly synthesized compounds 5-fluoroisatin-chalcone conjugates 5(a–g) were assessed for antimicrobial activity against gram-positive (*Streptococcus pyogenes* and *Staphylococcus aureus*), gram-negative *(Klebsiella pneumonia* and *Escherichia coli*) bacteria, and fungi (*Candida albicans*), as well as the inhibition zone (IZ) measured in millimeters (mm) and compared with three different reference medications: vancomycin, ciprofloxacin, and fluconazole, respectively. Dimethyl sulfoxide (DMSO) was used to dissolve all of the investigated final compounds and reference medications to create a concentration of (100 mg/ml) [36]. The Al Jazeera Lab Company/Iraq evaluated the *in vitro* antimicrobial activity.

Results and Discussion

Chemical synthesis

As illustrated in Scheme 1, the designed final compounds 5(a–g) were obtained through two reaction steps. The first reaction step involves the formation of Schiff's base (imine) by the nucleophilic addition of the primary amino group of 4-aminoacetophenone (1) to the carbonyl group (C=O) at position 3 of 5-fluoroisatin (2), in slightly acidic media (*p*H 4–6), and reflux to generate the compound (3) [37]. Mechanistically, as shown in Figure 2, the formation of an imine occurs in two steps, addition, and elimination (dehydration). The first steps involved the acid-catalyzed addition of amine nitrogen (which acts as a nucleophile) to the carbon of the carbonyl group of an aldehyde or ketone, then the deprotonation of nitrogen to give carbinolamine, which is an unstable compound. In the second step, by either acid- or base-catalyzed pathways, the carbinolamine loses a water molecule.

As carbinolamine is an alcohol, it is dehydrated by acid catalysis. Dehydration of the carbinolamine leaves a compound with a C=N double bond (an imine) [38].

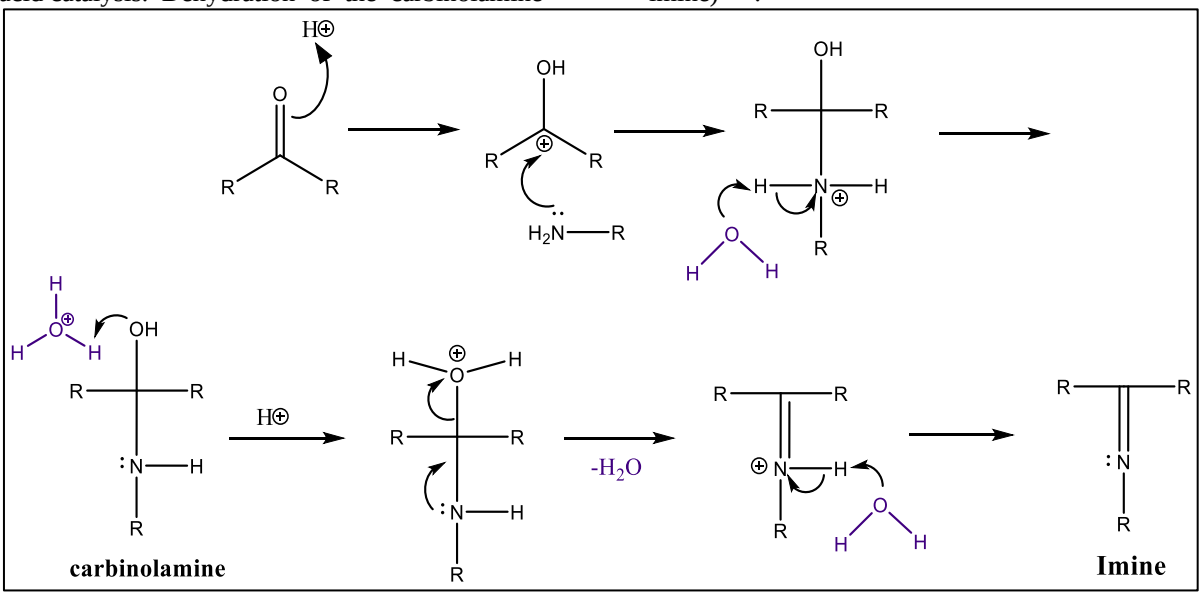


Figure 2. The steps for the mechanism of imine formation [38].

In the second reaction step, 5-fluoroisatin-chalcone conjugates 5(a–g) will be formed by treating compound (3) with different aromatic aldehydes 4(a–g) separately, in ethanol as a solvent, with the use of a diluted solution of NaOH as a catalytic base, by Claisen–Schmidt condensation [35]. The mechanism of this reaction, as shown in Figure 3, includes four main steps. The first step of the reaction begins with the abstraction of an α-hydrogen atom from ketone (acetophenone) by the catalyst OH^-, leading to the formation of a carbanion ion (intermediate I). The second step of the reaction is the nucleophilic addition of the carbanion ion to the carbon atom of the aldehyde's carbonyl group to form an alkoxide ion (intermediate II). In the third step, alkoxide ions take protons from solvent molecules, H_2O, resulting in the formation of β-hydroxy ketone (intermediate III). In the fourth step, the dehydration of β-hydroxy ketone takes place by the loss of water molecules that end with the formation of a chalcone compound [39].

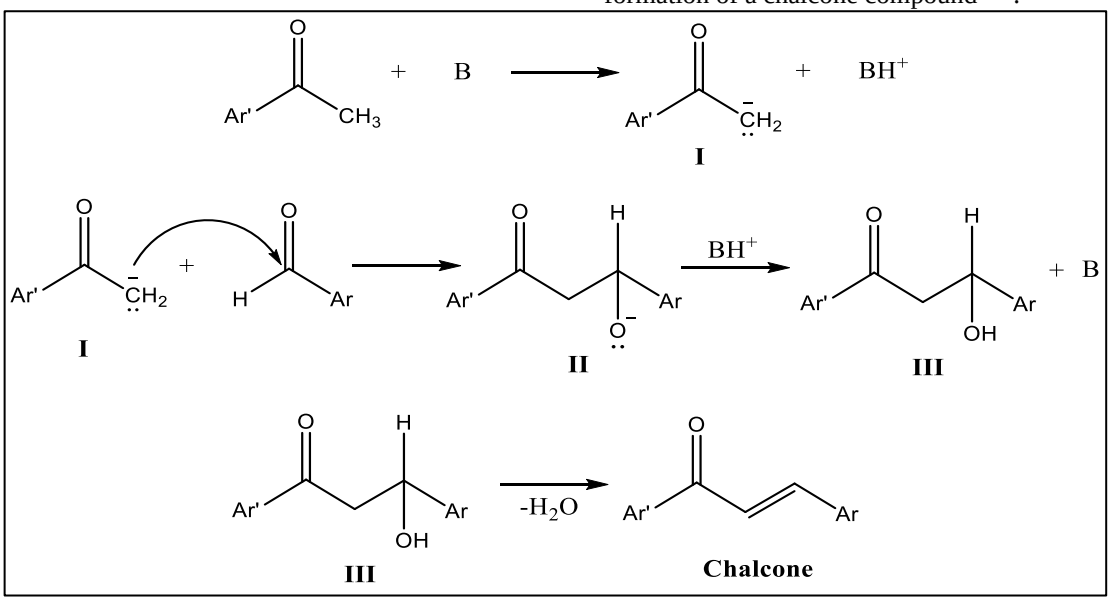


Figure 3. Mechanism for the formation of α,β–unsaturated ketone [39].

The chemical structures of the newly synthesized 5-fluoroisatin-chalcone conjugates 5(a–g) were confirmed by FT-IR and ^{1}H-NMR. In the FT-IR spectrum, compound (3) (Schiff's base) showed two IR absorption peaks attributed to stretching vibrations for two carbonyl groups at 1739 cm^{-1} (amide of 5-fluoroisatin) and 1681 cm^{-1} (4-aminoacetophenone), as well as an absorption

peak at 1620 cm^{-1} for (C=N) that indicates the formation of Schiff's base (imine). While the IR bands stretching vibration of (NH_2) primary amine of 4-aminoacetophenone disappeared. As in compound (3), in the IR spectra for the final compounds; 5(a–g), 5-fluoroisatin-chacone conjugates, there are two absorption peaks attributed to stretching vibrations of two carbonyl groups and a peak for (C=N). In the IR spectra of compound (3) and final compounds 5(a–g) 5-fluoroisatin-chacone conjugates, the 5-fluoroisatin N-H functions are found in the 3236–3163cm^{-1} region.

In the ^{1}H-NMR spectra, compound (3) showed a single peak assigned to one proton of (N-H) of the 5-fluoroisatin nucleus that appeared at 11.07 ppm, and another single peak appeared at 2.6 ppm as a singlet related to three protons of (-CH_3) and disappearance of the (NH_2) peak of 4-aminoacetophenone (1), this confirms the coupling of 4-aminoacetophenone with 5-fluoroisatin, while the peaks from (7.07–8.10) ppm related to the presence of aromatic rings. The final compounds, 5-fluoroisatin-chalcone conjugates 5(a–g), showed a single peak assigned to one proton of (N-H) of the 5-fluoroisatin nucleus that appeared at 11.02–11.10 ppm, while the peaks from (6.70–8.38) ppm related to the presence of aromatic rings and two protons of α,β–unsaturated ketone. The disappearance of the (-CH_3) peak of compound (3) indicates the formation of these final compounds 5(a–g). The compound 5g has a characteristic peak at 5.20 ppm as a singlet related to two protons of (O-CH_2-).

In vitro antimicrobial evaluation

From the results in Table 1, for gram-positive bacteria in comparison with the standard drug (vancomycin), most of the synthesized compounds showed moderate antibacterial activity against *S. aureus*, while compound 5c was slightly active and compound 5g was inactive. On the other hand, for antibacterial activity against *S. pyogenes*, the compounds (5a, 5b, and 5e) were moderately active, with slight activity for compounds (5d and 5f), while the compounds (5c and 5g) were inactive. While, for gram-negative bacteria in comparison with the standard drug (ciprofloxacin), all of the synthesized compounds were moderately active against *E. coli*, with the exception of compounds (5c and 5g) that were slightly active, the compounds (5b, 5d, and 5e) were slightly active against *K. pneumoniae*, with no activity for compounds (5a, 5c, 5f, and 5g). In general, all of them were less effective than the standard drugs, vancomycin and ciprofloxacin.

For antifungal activity against *C. albicans* in comparison with fluconazole as a standard drug, most of the synthesized compounds showed moderate-to-high activity, which was comparable with a standard drug, mainly for compound 5e that contains a furan ring, while compound 5c was slightly active and compound 5g was inactive. These results matched with other previous research, which showed that the introduction of heteroaromatic rings like furan or thiophene for molecules led to an increase in their antimicrobial activity, in addition to the presence of halogens or electron-withdrawing groups substituting on the phenyl ring, while substitution with electron-donating groups gave the least activity [40,41].

Table 1. Antimicrobial activities of 5-fluoroisatin-chacone conjugates 5(a–g**) in concentrations (100 mg/mL).**

Compound No.	Inhibition Zone (IZ) in millimeters				
	Bacterial strains				Fungi
	Gram-positive		Gram-negative		
	S. aureus	*S. pyogenes*	*E. coil*	*K. pneumonia*	*C. albicans*
5a	13	10	14	-	13
5b	12	10	13	6	13
5c	8	-	8	-	8
5d	11	8	14	8	16
5e	14	11	16	6	21
5f	11	8	12	-	15
5g	-	-	8	-	-
Vancomycin*	20	22	20	16	-
Ciprofloxacin*	16	18	26	18	-
Fluconazole**	-	-	-	-	18
DMSO	-	-	-	-	-

* Standard drugs for antibacterial activity, ** Standard drug for antifungal activity, IZ = inhibition zone.Highly active (IZ = more than 15 mm), moderately active (IZ = 10–15 mm), slightly active (IZ = 5–10 mm), and no activity (IZ = (-)).

Conclusion

A new seven of 5-fluoroisatin-chalcone conjugates in this study were synthesized. The structures and purity of the final compounds 5(a–g) have been confirmed by ^{1}H-NMR and FT-IR spectroscopy. The antimicrobial activity of 5-fluoroisatin is increased by a modification on carbonyl group at position 3 and condensation with chalcone moiety to give 5-fluoroisatin-chalcone conjugates 5(a–g). The target compounds 5(a–g) appeared to be good candidates as antibacterial agents against *E. coli* and *S. aureus* as well as antifungal agents against *C. albicans*. In general, and for comparison, the antifungal activity of the final compounds 5(a–g) was more potent than their antibacterial activity. Finally, for the antimicrobial activity, the most active compound of this series was compound 5e, while compound 5g was the least active one.

Acknowledgments

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Conflicts of Interest

The authors declare no conflicts of interest. Ethics Statements The authors state that as this was an in- vitro investigation, no ethics committee permission was required for the synthesis and evaluation of new compounds.

Author Contribution

The first author; (Hamza) assisted with the synthesis of desired compounds; analysis of IR and 1 H-NMR data, discussion of the antimicrobial activity, manuscript's drafting; and critical revision. The second author; (May) contributed to the design of the study, analyzed the final results, authorized the manuscript in its final form, and participated in the study's design.

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