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Synthesis of new imidazolone derivatives and evaluated their biological activities

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Abstract

Some novel imidazolone derivatives were synthesized in excellent yields and evaluated for their antifungal activities. The structures of the prepared compounds were proved by IR, ^{1}H-NMR, Mass spectra. The antifungal activities of imidazolone derivatives were evaluated by the agar well diffusion method. From the result of biological activity 2-((4-(4-chlorobenzylidene)-5-oxo-4,5-dihydrooxazol-2-yl)methyl)isoindoline-1,3-dione **(1)** showed high activity against *Aspergillus flavus* and *Aspergillus ochrachous*.

1. Introduction

Imidazolone and its derivatives are very important heterocyclic class because of their biological activities such as anticancer , hemeoxygenase inhibitors , antiaging agents , anticoagulants , anti-inflammatory , antibacterial , antifungal , antiviral , antitubercular , antidiabetic and antimalarial [1-14].

The amphoteric nature of imidazolone derivatives promoted us to study its behavior towards some nucleophiles and electrophiles. Many drugs have an imidazole ring, like certain antifungal drugs, the nitroimidazole chain of antibiotics, and the sedative midazolam.[15-19].

Imidazole is an important group of compounds that have various biological activities and the current study was undertaken to prepare some new derivatives of imidazole and related fused heterocyclic compounds and screen for their antimicrobial activity [20- 26].

Based on the above facts, herein we synthesized new imidazolone derivatives and evaluated their biological activities.

2. Experimental

2.1. Synthetic methods, spectral and analytical data

Melting points are uncorrected and were determined by the open capillary methode using Gallen Kamp melting point apparatus. FT-IR spectra (KBr disk) were recorded on a JASCO FT-IR 660 Plus spectrometer. ^{1}HNMR spectra were recorded on a Bruker

Avance 400 (400 MHz) by using $CDCl_3$ and DMSO as the solvents. Mass spectra were obtained using a Shimadzu GCMS-QP 1000 EX instrument (70 ev EI mode). All microanalysis (1HNMR ,IR ,MASS) were carried out at cairo university, micro analytical center, Ain Shams and Benha university, Egypt.

Biological activity of some the synthesized compounds were done in the Microbiological laboratory, Botany Department, Benha University.

2.1.1. General procedure for preparation of oxazolone

A mixture of phthalylglycylglycine (0.01 mol) and *p*-chlorobenzaldehyde (0.01 mol), fused sodium acetate (0.015 mol) were dissolved in acetic anhydride (20ml) was refluxed for 3hrs. After cooling the reaction mixture was poured onto ice water/dilute HCl and the solid product was filtered off and crystallized from benzene formed (**1**). Yield (97%) ; m.p. 170-172 ^{0}C; IR spectrum showed absorption bands at υ 1683 cm^{-1} attributable to CO of cyclic imides , at υ 1591 cm^{-1} attributable to C=N .

Anal. Calcd. For $C_{19}H_{11}ClN_2O_4$;C, 62.50; H, 3.00; N, 7.30 .Found: C, 62.22; H, 3.02; Cl, 9.67; N, 7.64 .

2.1.2. General procedure for preparation of imidazolone derivative

A mixture of (1), (0.01 mol) and *p*-aminoacetophenone (0.01 mol), fused sodium acetate (0.015 mol) were dissolved in glacial acetic acid (30ml) was refluxed for 5hrs. Then, the reaction mixture was cooled and poured onto ice cold water. The obtained solid was filtered off, washed with water and crystallized from benzene gave (**2**). Yield (95%) ; m.p. 178-180 ^{0}C; IR spectrum showed absorption bands at υ 1682 cm^{-1} attributable to CO of cyclic imides , at υ 1592 cm^{-1} attributable to CO. ^{1}H NMR spectrum (DMSO-d6) showed signals at δ ppm 2.5 (s, 3H, CH_3), 3.4 (s, 2H, CH_2) and 3.3 (s, 1H, CH), 7.4-8.8 (m ,14H , ArH and olefinic protons).

Anal. Calcd. For $C_{27}H_{18}ClN_3O_4$;C, 67.25; H, 3.60; N, 8.52 .Found: C, 67.02; H, 3.75; N, 8.68 .

2.1.3. Synthesis of 2-((4-(-4-chlorobenzylidene)-1-(4-(3-(4-nitrophenyl)acryloyl)phenyl)-5-oxo-4,5-dihydro-1H-imidazol-2-yl)methyl)isoindoline-1,3-dione

A mixture of (**2**), (0.01 mol) and *p*-nitrobenzaldehyde (0.01 mol) was dissolved in 50 ml ethanol (absolute). To the stirred 6.2 gm of KOH dissolved in 60 ml water was added drop by drop within 30 minutes. The stirring was continued onto ice bath for 3 hr and left overnight at room temperature. The solid obtained was filtered off and crystallized from benzene gave (**3**). Yield (81%) ; m.p. 180-183

^{0}C; IR spectrum showed absorption bands at υ 1767 cm^{-1} attributable to CO of cyclic imides, at υ 1616 cm^{-1} attributable to CO and at υ 1422 , 1379 cm^{-1} attributable to NO_2.

Anal. Calcd. For $C_{34}H_{21}ClN_4O_6$;C, 66.00; H, 3.32; N, 9.19. Found: C, 66.19; H, 3.43; N, 9.08 .

2.1.4. General procedure for preparation of pyrazoline derivative

A mixture of (**3**), (0.01 mol) and hydrazine hydrate (0.01 mol) were dissolved in ethanol or/ semicarbazide (0.01 mol) in pyridine was refluxed for 5 hrs. Then, the reaction mixture was cooled and poured onto ice water /dilute.HCl. The solid product was filtered off and crystallized from ethanol gave **4(a,b)**.

2.1.4.a. 2-((4-(4-chlorobenzylidene)-1-(4-(5-(4-nitrophenyl)-4,5-dihydro-1H-pyrazol-3-yl)phenyl)-5-oxo-4,5-dihydro-1H-imidazol-2-yl)methyl)isoindoline-1,3-dione

Yield (58%) ; m.p. 140-145 ^{0}C ; IR spectrum showed absorption bands at υ 3300 cm^{-1} attributable to NH and at υ 1594 cm^{-1} attributable to CO of cyclic imides, at υ 1520 cm^{-1} attributable to CO.

Anal. Calcd. For $C_{35}H_{24}ClN_7O_6$; C, 64.63; H, 3.75; N, 13.41 . Found: C, 64.71; H, 3.67; N, 13.32 .

2.1.4.b. 3-(4-(4-(4-chlorobenzylidene)-2-((1,3-dioxoisoindolin-2-yl)methyl)-5-oxo-4,5-dihydro-1H-imidazol-1-yl)phenyl)-5-(4-nitrophenyl)-4,5-dihydro-1H-pyrazole-1-carboxamide

Yield (87%) ; m.p. 148-150 ^{0}C ; IR spectrum showed absorption bands at υ 3435 cm^{-1} attributable to NH_2 and at υ 1796 cm^{-1} attributable to CO of cyclic imides, at υ 1594 cm^{-1} attributable to CO.

Anal. Calcd. For $C_{34}H_{23}ClN_6O_5$; C, 62.24; H, 3.32; N, 14.40. Found: C, 62.37; H, 3.59; N, 14.55.

2.1.5. General procedure for preparation of isoxazoline

A mixture of (**3**), and hydroxylamine hydrochloride (0.01 mol) were dissolved in pyridine was refluxed for 5 hrs. After cooling the mixture, the produced solution was poured onto ice water /dilute.HCl. The solid obtained was filtered off and crystallized from ethanol formed (**5**). Yield (83%) ; m.p. 122-125 ^{0}C; IR spectrum showed absorption bands at υ 3341 cm^{-1} corresponding to NH, at υ 1343 cm^{-1} corresponding to NO_2 , at υ 1645 cm^{-1} corresponding to CO of cyclic imides and at υ 1604 cm^{-1} corresponding to CO.

Anal. Calcd. For $C_{34}H_{22}ClN_5O_6$; C, 64.91; H, 3.67; N, 11.19 . Found: C, 64.61; H, 3.51; N, 11.08.

2.1.6. General procedure for preparation of pyrimidines

A mixture of (**3**), (0.01 mol) and thiourea /or urea (0.01 mol) were dissolved in sodium ethoxide (15ml) was refluxed for 6hrs. After that, the reaction mixture was concentrated and cooled then poured onto ice cold water the solid product was filtered off and crystallized from ethanol gave **6(a,b).**

2.1.6.a. 2-((4-(4-chlorobenzylidene)-1-(4-(2-mercapto-6-(4-nitrophenyl)pyrimidin-4-yl)phenyl)-5-oxo-4,5-dihydro-1H-imidazol-2-yl)methyl)isoindoline-1,3-dione

Yield (70%) ; m.p. 160-164 ^{0}C; IR spectrum showed absorption bands at υ 3351 cm^{-1} corresponding to NH, at υ 1593 cm^{-1} corresponding to CO of cyclic imides, at υ 1514cm^{-1} corresponding to CO, at υ 1410 corresponding to NO_2 and at υ 1332 cm^{-1} corresponding to CS. Mass spectrum showed molecular ion peak at m/z = 672 ($M^{\cdot+}$, 100.00%).

Anal. Calcd. For $C_{35}H_{21}ClN_6O_5S$; C, 62.44; H, 3.01; N, 12.59 . Found: C, 62.46; H, 3.14; N, 12.49 .

2.1.6.b. 2-((4-(4-chlorobenzylidene)-1-(4-(2-hydroxy-6-(4-nitrophenyl)pyrimidin-4-yl)phenyl)-5-oxo-4,5-dihydro-1H-imidazol-2-yl)methyl)isoindoline-1,3-dione

Yield (59%) ; m.p. 150-154 ^{0}C; IR spectrum showed absorption bands at υ 3353 cm^{-1} corresponding to NH at υ 1593 cm^{-1} corresponding to CO of cyclic imides, at υ 1513 cm^{-1} corresponding to CO and at υ 1376 corresponding to NO_2 . Mass spectrum showed molecular ion peak at m/z = 656 ($M^{\cdot+}$, 100.00%).

Anal. Calcd. For $C_{35}H_{21}ClN_6O_6$; C, 63.77; H, 3.43; N, 12.65. Found: C, 63.98; H, 3.22; N, 12.79.

2.1.7. General procedure for preparation of anilides

A mixture of (**3**), (0.01 mol) and aromatic amines (0.01 mol) namely, aniline, *p*-toluidine, *p*-anisidine were dissolved in ethanol (30ml) was refluxed for 3 hrs. After cooling, the solid obtained was collected and crystallized from ethanol formed **7(a-c)**.

2.1.7.a. 2-((4-(4-chlorobenzylidene)-1-(4-(3-(4-nitrophenyl)-3-(phenylamino)propanoyl)phenyl)-5-oxo-4,5-dihydro-1H-imidazol-2-yl)methyl)isoindoline-1,3-dione

Yield (75%) ; m.p. 180-182 ^{0}C; IR spectrum for **7(a-c)** revealed the presence of absorption bands at υ 3341 cm^{-1} corresponding to NH at υ 1657-1646 cm^{-1} corresponding to CO of cyclic imides, at υ 1629 cm^{-1} corresponding to CO and at υ 1344 cm^{-1} corresponding to NO_2 . ^{1}H NMR spectrum for **(7a)** showed signals at δ ppm 2.1 (s, 2H, CH_2) , 2.5 (s, 2H, CH_2) and 3.4 (s, 1H, CH) , 7.5-8.1 (m ,17H , ArH , olefinic protons) and 10.33 (s,1H,NH, exchangeable) .

Anal. Calcd. For $C_{40}H_{28}ClN_5O_6$; C, 67.87; H, 3.89; N, 9.98 . Found: C, 67.65; H, 3.97; N, 9.86.

2.1.7.b. 2-((4-(4-chlorobenzylidene)-1-(4-(3-(4-nitrophenyl)-3-(p-tolylamino)propanoyl)phenyl)-5-oxo-4,5-dihydro-1H-imidazol-2-yl)methyl)isoindoline-1,3-dione

Yield (82%) ; m.p. 115-120 ^{0}C.

Anal. Calcd. For $C_{41}H_{30}ClN_5O_6$; C, 68.01; H, 4.29; N, 9.54 . Found: C, 68.00; H, 4.18; N, 9.67.

2.1.7.c. 2-((4-(4-chlorobenzylidene)-1-(4-(3-((4-methoxyphenyl)amino)-3-(4-nitrophenyl)propanoyl)phenyl)-5-oxo-4,5-dihydro-1H-imidazol-2-yl)methyl)isoindoline-1,3-dione

Yield (84%) ; m.p. 135-140 ^{0}C.

Anal. Calcd. For $C_{41}H_{30}ClN_5O_7$; C, 66.29; H, 4.25; N, 9.57 . Found: C, 66.53; H, 4.09; N,9.46 .

2.1.8. General procedure for preparation of pyridine

A mixture of (**3**), (0.01 mol), malononitrile or /and ethylcyanoacetate (0.01 mol) and ammonium acetate (0.02 mol) can be heated in oil bath for 8hrs. Then, the reaction mixture was cooled and poured onto ice water. The solid product was filtered off and crystallized from ethanol gave **8(a,b).**

2.1.8.a. 6-(4-(4-(4-chlorobenzylidene)-2-((1,3-dioxoisoindolin-2-yl)methyl)-5-oxo-4,5-dihydro-1H-imidazol-1-yl)phenyl)-2-hydroxy-4-(4-nitrophenyl)nicotinonitrile

Yield (69%) ; m.p. 90-95 ^{0}C; IR spectrum for **8(a,b)** showed absorption bands at υ 3388, 3447 cm^{-1} corresponding to (OH, NH_2), at υ 2209, 2207 corresponding to CN, at υ 1317,1373 cm^{-1} corresponding to NO_2 , at υ 1710, 1716 cm^{-1} corresponding to CO of cyclic imides and at υ 1592, 1697 cm^{-1} corresponding to CO .

Anal. Calcd. For $C_{37}H_{21}ClN_6O_6$; C, 65.13; H, 3.65; N, 12.26. Found: C, 65.25; H, 3.11; N, 12.34 .

2.1.8.b. 2-amino-6-(4-(4-(4-chlorobenzylidene)-2-((1,3-dioxoisoindolin-2-yl)methyl)-5-oxo-4,5-dihydro-1H-imidazol-1-yl)phenyl)-4-(4-nitrophenyl)nicotinonitrile

Yield (84%) ; m.p. 170-173 ^{0}C.

Anal. Calcd. For $C_{37}H_{22}ClN_7O_5$; C, 65.59; H, 3.55; N, 14.49. Found: C, 65.35; H, 3.26; N, 14.42 .

2.1.9. General procedure for preparation of 2-((4-(4-chlorobenzylidene)-1-(4-(3-(4-nitrophenyl)oxirane-2-carbonyl)phenyl)-5-oxo-4,5-dihydro-1H-imidazol-2-yl)methyl)isoindoline-1,3-dione

A solution of (**3**), (0.01 mol) was dissolved in acetone and methanol was reacted with aqueous sodium hydroxide (12 ml) then hydrogen peroxide (5 ml , 30%) was added drop by drop. Then, the solution was shaken and left overnight at room

temperature. A yellow crystalline product was collected that recrystallized from light petroleum-ether (60-80%) formed (**9**).

Yield (85%) ; m.p. 177-182 ^{0}C; IR spectrum showed absorption bands at υ 1884 cm^{-1} corresponding to CO of cyclic imides, at υ 1654 cm^{-1} corresponding to CO and at υ 1304 cm^{-1} corresponding to NO_2 .

Anal. Calcd. For $C_{34}H_{21}ClN_4O_7$; C, 64.60; H, 3.23; N, 8.72 . Found: C, 64.51; H, 3.34; N, 8.85 .

2.1.10. General procedure for preparation of 2-((4-(4-chlorobenzylidene)-1-(4-(4-hydroxy-5-(4-nitrophenyl)-4,5-dihydro-1H-pyrazol-3-yl)phenyl)-5-oxo-4,5-dihydro-1H-imidazol-2-yl)methyl)isoindoline-1,3-dione

A solution of (**9**), (0.01 mol) was dissolved in ethanol then reacted with hydrazine hydrate (0.05 mol). The reaction mixture was refluxed for 6 hrs. After cooling, the solid obtained was collected by filteration and crystallized from ethanol gave (**10**).

Yield (93%) ; m.p. 190-192 ^{0}C; IR spectrum showed absorption bands at υ 3470 cm^{-1} corresponding to (OH, NH) , at υ 1394-1336 cm^{-1} corresponding to NO_2 , at υ 1685 cm^{-1} corresponding to CO of cyclic imides, at υ 1600 cm^{-1} corresponding to CO and at υ 1580 corresponding to (C=N). ^{1}H NMR spectrum showed signals at δ ppm 2.4 (s, 2H, CH_2) , 4.2 (s, 1H, CH) , 7.2 (m ,13H , ArH and olefinic protons). Mass spectrum showed molecular ion peak at m/z = 646 ($M^{\cdot+}$, 100.00%).

Anal. Calcd. For $C_{34}H_{23}ClN_6O_6$; C, 63.29; H, 3.85; N, 12.91 . Found: C, 63.11; H, 3.58; N, 12.99 .

2.1.11. General procedure for preparation of 2-((4-(4-chlorobenzylidene)-1-(4-(4-hydroxy-5-(4-nitrophenyl)-4,5-dihydroisoxazol-3-yl)phenyl)-5-oxo-4,5-dihydro-1H-imidazol-2-yl)methyl)isoindoline-1,3-dione

A mixture of (**9**), (0.01 mol) and hydroxylamine hydrochloride (0.02 mol) were dissolved in ethanol (20ml) was refluxed for 6 hrs. Then, the reaction mixture was cooled and poured onto ice water. The solid product was filtered off and recrystallized from ethanol gave (**11**).

Yield (63%) ; m.p. 180-183^{0}C; IR spectrum showed absorption bands at υ 3442 cm^{-1} corresponding to OH, at υ 1700 cm^{-1} corresponding to CO of cyclic imides, at υ 1597 cm^{-1} corresponding to CO and at υ 1384,1345 cm^{-1} corresponding to NO_2 . Mass spectrum showed molecular ion peak at m/z = 647 ($M^{\cdot+}$, 100.00%).

Anal. Calcd. For $C_{34}H_{22}ClN_5O_7$; C, 63.12; H, 3.64; N, 10.67 . Found: C, 63.02; H, 3.42; N, 10.81 .

2.1.12. General procedure for preparation of 2-((4-(4-chlorobenzylidene)-1-(4-(4-(4-nitrophenyl)-2-thioxooxazolidine-5-carbonyl)phenyl)-5-oxo-4,5-dihydro-1H-imidazol-2-yl)methyl)isoindoline-1,3-dione

A mixture of (**9**), (0.01 mol) and thiourea (0.01 mol) were dissolved in DMF (20 ml) was refluxed for 3hrs. After cooling, the reaction mixture was poured onto ice water and extracted with ether. After evaporation of ether, the solid product was crystallized from ethanol gave (**12**).

Yield (77%) ; m.p. 154-159 ^{0}C; IR spectrum showed absorption bands at υ 3404 cm^{-1} corresponding to NH, at υ 1633 cm^{-1} corresponding to CO of cyclic imides, at υ 1600 cm^{-1} corresponding to CO and at υ 1403 cm^{-1} corresponding to CS.

Anal. Calcd. For $C_{35}H_{22}ClN_5O_7S$; C, 60.96; H, 3.01; N, 10.28 . Found: C, 60.74; H, 3.20; N, 10.12 .

2.1.13. General procedure for preparation of 2-((4-(4-chlorobenzylidene)-1-(4-(3-(4-nitrophenyl)-6-oxomorpholine-2-carbonyl)phenyl)-5-oxo-4,5-dihydro-1H-imidazol-2-yl)methyl)isoindoline-1,3-dione

Equimolar amounts of epoxide (**9**), (0.01 mol) and glycine (0.01 mol) in DMF (15ml) was refluxed for 3hrs. After cooling, the reaction mixture was poured onto ice water and extracted with ether. After evaporation of ether, the solid product was crystallized from benzene gave (**13**).

Yield (68%) ; m.p. 190-192^{0}C; IR spectrum showed absorption bands at υ 3420 cm^{-1} corresponding to NH, at υ 1670 cm^{-1} corresponding to CO of cyclic imides, at υ 1592 cm^{-1} corresponding to CO and at υ 1388,1351 corresponding to NO_2 . Mass spectrum showed molecular ion peak at m/z = 689 ($M^{\cdot+}$, 100.00%).

Anal. Calcd. For $C_{36}H_{24}ClN_5O_8$; C, 62.64; H, 3.37; N, 10.02 . Found: C, 62.66; H, 3.51; N, 10.15.

2.1.14. General procedure for preparation of 2-(4-(4-chlorobenzylidene)-2-((1,3-dioxoisoindolin-2-yl)methyl)-5-oxo-4,5-dihydro-1H-imidazol-1-yl)acetic acid

A mixture of (**1**), (0.01 mol), [glycine (0.01 mol) was dissolved in 3 ml H_2O] in pyridine (10 ml) was refluxed for 5 hrs. Then, the reaction mixture was concentrated. After cooling the produced mixture was poured onto ice cold water /dilute.HCl. The solid product was filtered off then washed with water, dried and crystallized from ethanol formed (**14**).

Yield (93%) ; m.p. 220-223 ^{0}C; IR spectrum showed absorption bands at υ 3565 cm^{-1} corresponding to OH, at υ 1770, 1732 cm^{-1} corresponding to CO of cyclic imides, at υ 1609 cm^{-1} corresponding to CO and at υ 1391-1319 cm^{-1} corresponding to NO_2 . ^{1}H NMR spectrum showed signals at δ ppm 2.5 (s, 3H, CH_3), 3.3 (s, 2H, CH_2) and 4.3 (s, 1H, CH), 7.1-9.4 (m ,14H , ArH and olefinic protons) and 13.0

(s,1H,OH). Mass spectrum showed molecular ion peak at m/z = 423($M^{\cdot+}$, 100.00%).

Anal. Calcd. For $C_{21}H_{14}ClN_3O_5$; C, 59.68; H, 3.54; N, 9.71 . Found: C, 59.52; H, 3.33; N, 9.92 .

2.1.15. General procedure for preparation of 2-(4-(4-chlorobenzylidene)-2-((1,3-dioxoisoindolin-2-yl)methyl)-5-oxo-4,5-dihydro-1H-imidazol-1-yl)acetyl isothiocyanate

Compound (**14**), was dissolved in dry acetone (20 ml) then solid ammonium thiocyanate (0.01mol) was added within 30 min. with stirring. The reaction mixture was continued stirring for 2 hrs. at room temperature , then the product was filtered off leaved a clear solution of (**15**).

2.1.16. General procedure for preparation of 2-((4-(4-chlorobenzylidene)-5-oxo-1-((5-thioxo-2,5-dihydro-1H-1,2,4-triazol-3-yl)methyl)-4,5-dihydro-1H-imidazol-2-yl)methyl)isoindoline-1,3-dione

A solution of isothiocyanate (**15**), (0.01 mol) and hydrazine hydrate (0.01 mol) in dry acetone was refluxed for 1 hr. Then, the reaction mixture was concentrated. After cooling, the solid product was filtered off and crystallized from dry toluene formed (**16**).

Yield (80%) ; m.p. 155-160^{0}C; IR spectrum showed strong absorption bands at υ 3444-3015 cm^{-1} corresponding to NH, at υ 1658 cm^{-1} corresponding to CO of cyclic imides, at υ 1604 cm^{-1} corresponding to CO and at υ 1375 cm^{-1} corresponding to NO_2 and at υ 1330 cm^{-1} corresponding to CS.

Anal. Calcd. For $C_{22}H_{15}ClN_6O_3S$; C, 55.33; H, 3.00; N, 17.61 . Found: C, 55.18; H, 3.16; N, 17.55 .

2.1.17. General procedure for preparation of 2-(4-(4-chlorobenzylidene)-2-((1,3-dioxoisoindolin-2-yl)methyl)-5-oxo-4,5-dihydro-1H-imidazol-1-yl)-N-((2-mercaptophenyl)carbamothioyl)acetamide

A solution of (15), (0.01 mol) in dry acetone (25ml) was added to o-aminothiophenol (0.01 mol). The reaction mixture was heated under reflux for 1 hr. After cooling, the solid obtained was collected and recrystallized from benzene gave (**17**).

Yield (59%) ; m.p. 157-160 ^{0}C; IR spectrum showed strong absorption bands at υ 3462-3375 cm^{-1} ,3100 cm^{-1} attributable to (OH, NH), at υ 1750,1721 cm^{-1} attributable to CO of cyclic imides, at υ 1609 cm^{-1} attributable to CO and at υ 1397-1308 cm^{-1} attributable to NO_2 .

Anal. Calcd. For $C_{28}H_{20}ClN_5O_4S_2$; C, 56.90; H, 3.12; N, 11.93 . Found: C, 56.99; H, 3.42; N, 11.87.

2.1.18. General procedure for preparation of 2-(3-(2-(4-(4-chlorobenzylidene)-2-((1,3-dioxoisoindolin-2-yl)methyl)-5-oxo-4,5-dihydro-1H-imidazol-1-yl)acetyl)thioureido)benzoic acid

A mixture of (**15**), (0.01 mol) and anthranilic acid (0.01 mol) in dry acetone (20 ml) was refluxed for 1 hr . Then, the reaction mixture was cooled at room temperature and poured onto ice water. The solid obtained was filtered off and crystallized from toluene gave (**18**).

Yield (72%) ; m.p. 200-202 ^{0}C; IR spectrum showed strong absorption bands at υ 3472,3371 cm^{-1}, 3028 cm^{-1} attributable to (OH, NH), at υ 1721 cm^{-1} attributable to CO of cyclic imides, at υ 1611 cm^{-1} attributable to CO, at υ 1421 cm^{-1} attributable to NO_2 and at υ 1310 cm^{-1} attributable to CS. Mass spectrum showed molecular ion peak at m/z = 601 ($M^{\cdot+}$, 60.65%).

Anal. Calcd. For $C_{29}H_{20}ClN_5O_6S$; C, 57.56; H, 3.10; N, 11.28 . Found: C, 57.86; H, 3.35; N, 11.63 .

2.1.19. General procedure for preparation of 2-((4-(4-chlorobenzylidene)-5-oxo-1-((3-phenyl-2,4-dithioxo-3,4-dihydro-2H-1,3,5-oxadiazin-6-yl)methyl)-4,5-dihydro-1H-imidazol-2-yl)methyl)isoindoline-1,3-dione

To a solution of (**15**) , (0.01 mol) in dry acetone (20ml), phenylisothiocyanate (0.01 mol) was added and the mixture was refluxed for 1 hr . The solid obtained was crystallized from ethanol gave (**19**) .

Yield (85%) ; m.p. 162-167 ^{0}C; IR spectrum showed strong absorption bands at υ 1720 cm^{-1} attributable to CO of cyclic imides , at υ 1600 cm^{-1} attributable to CO , at υ 1535 cm^{-1} attributable to (C=N)and at υ 1375,1312 cm^{-1} attributable to NO_2 .

Anal. Calcd. For $C_{29}H_{18}ClN_5O_4S_2$; C, 58.24; H, 3.18; N, 11.98 . Found: C, 58.05; H, 3.02; N, 11.67 .

2.1.20. General procedure for preparation of 2-((1-((5-acetyl-6-methyl-4-thioxo-5,6-dihydro-4H-1,3-oxazin-2-yl)methyl)-4-(4-chlorobenzylidene)-5-oxo-4,5-dihydro-1H-imidazol-2-yl)methyl)isoindoline-1,3-dione

To a solution of isothiocyanate (15), (0.01 mol) in dry acetone (30 ml) , acetylacetone (0.01 mol) was added and the mixture was heated under reflux for 3 hrs . The reaction mixture was cooled and poured onto ice cold water . The solid precipitated was recrystallized from toluene formed (**20**).

Yield (86%) ; m.p. 189-193 ^{0}C; IR spectrum showed strong absorption bands at υ 1721 cm^{-1} attributable to CO of cyclic imides , at υ 1612 cm^{-1} attributable to CO , at υ 1422 cm^{-1} attributable to NO_2 and at υ 1312 cm^{-1} attributable to CS.

Anal. Calcd. For $C_{27}H_{21}ClN_4O_5S$; C, 59.24; H, 3.97; N, 10.35 . Found: C, 59.07; H, 3.86; N, 10.21 .

2.1.21. General procedure for preparation of 2-(((2-(4-(4-chlorobenzylidene)-2-((1,3-dioxoisoindolin-2-yl)methyl)-5-oxo-4,5-dihydro-1H-imidazol-1-yl)acetyl)carbamothioyl)thio)acetic acid

To a solution of (**15**) , (0.01 mol) in dry acetone (20ml) , thioglycolic acid (0.01 mol) was added . The mixture was refluxed for 1 hr . Then, the reaction mixture was cooled and poured onto ice cold water . The obtained precipitate was recrystallized from acetic acid formed (**21**).

Yield (64%) ; m.p. 199-204 ^{0}C; IR spectrum showed strong absorption bands at υ 3746, 3463 cm^{-1} ,3028 cm^{-1} attributable to (OH, NH) , at υ 1721 cm^{-1} attributable to CO of cyclic imides, at υ 1600 cm^{-1} attributable to CO, at υ 1422 cm^{-1} attributable to NO_2 and at υ 1312 cm^{-1} attributable to CS.

Anal. Calcd. For $C_{24}H_{17}ClN_4O_6S_2$; C, 51.34; H, 3.37; N, 10.52 . Found: C, 51.75; H, 3.08; N, 10.06 .

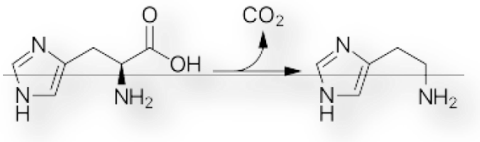
2.1.22. General procedure for preparation of 2-((4-(4-chlorobenzylidene)-5-oxo-1-(2-oxo-2-(4-oxo-2 thioxothiazolidin-3-yl)ethyl)-4,5-dihydro-1H-imidazol-2-yl)methyl)isoindoline-1,3-dione

A solution of (**25**) , (0.01 mol) was dissolved in acetic anhydride (15ml) and refluxed for 1 hr . After cooling, the solid precipitated was crystallized from ethanol gave (**22**) .Yield (80%) ; m.p. 166-168 ^{0}C; IR spectrum showed strong absorption bands at υ 1773, 1750, 1725 cm^{-1} attributable to CO of cyclic imides, at υ 1611 cm^{-1} attributable to CO, at υ 1422 cm^{-1} attributable to NO_2 and at υ 1313 cm^{-1} attributable to CS.

Anal. Calcd. For $C_{24}H_{15}ClN_4O_5S_2$; C, 53.68; H, 2.39; N, 10.14 . Found: C, 53.48; H, 2.81; N, 10.40 .

2.2. Biological activity of synthesized compounds

Imidazole is combined into many important biological molecules. The most popular is the amino acid histidine, that has an imidazole side-chain. Histidine is existing in many proteins and enzymes and plays a very important part in the structure and binding functions of hemoglobin. Imidazole-based histidine compounds play a vital role in intracellular buffering[27]. Histidine decarboxylated to histamine, that is also a popular biological compound. Histamine causes urticaria (hives) when it is formed during allergic reaction. The connection between histidine and histamine can be shown below:



Some of the applications of imidazole is in the purification of His-tagged proteins in immobilised metal affinity chromatography (IMAC). Imidazole can be used to elute tagged proteins bound to nickel ions linked

to the surface of beads in the chromatography column. An excess of imidazole is passed across the column, that separates the His-tag from nickel coordination, freeing the His- tagged proteins.

Imidazole has an important part of many pharmaceuticals. Synthetic imidazoles are existing in many antifungal and fungicides, antiprotozoal and antihypertensive medications. Imidazole is part of the theophylline molecule, present in tea leaves and coffee beans, that stimulates the central nervous system. It is existing in the anticancer medication mercaptopurine, that fights leukemia by interfering with DNA activities.

A set of substituted imidazoles, involving clotrimazole, are selective inhibitors of nitric oxide synthase, that makes them interesting drug targets in sore, neurodegenerative diseases and neoplasms of the nervous system[28][29]. Other biological activities of the imidazole pharmacophore connect to the downregulation of intracellular Ca2+ and K+ fluxes, and interference with translation initiation[30].

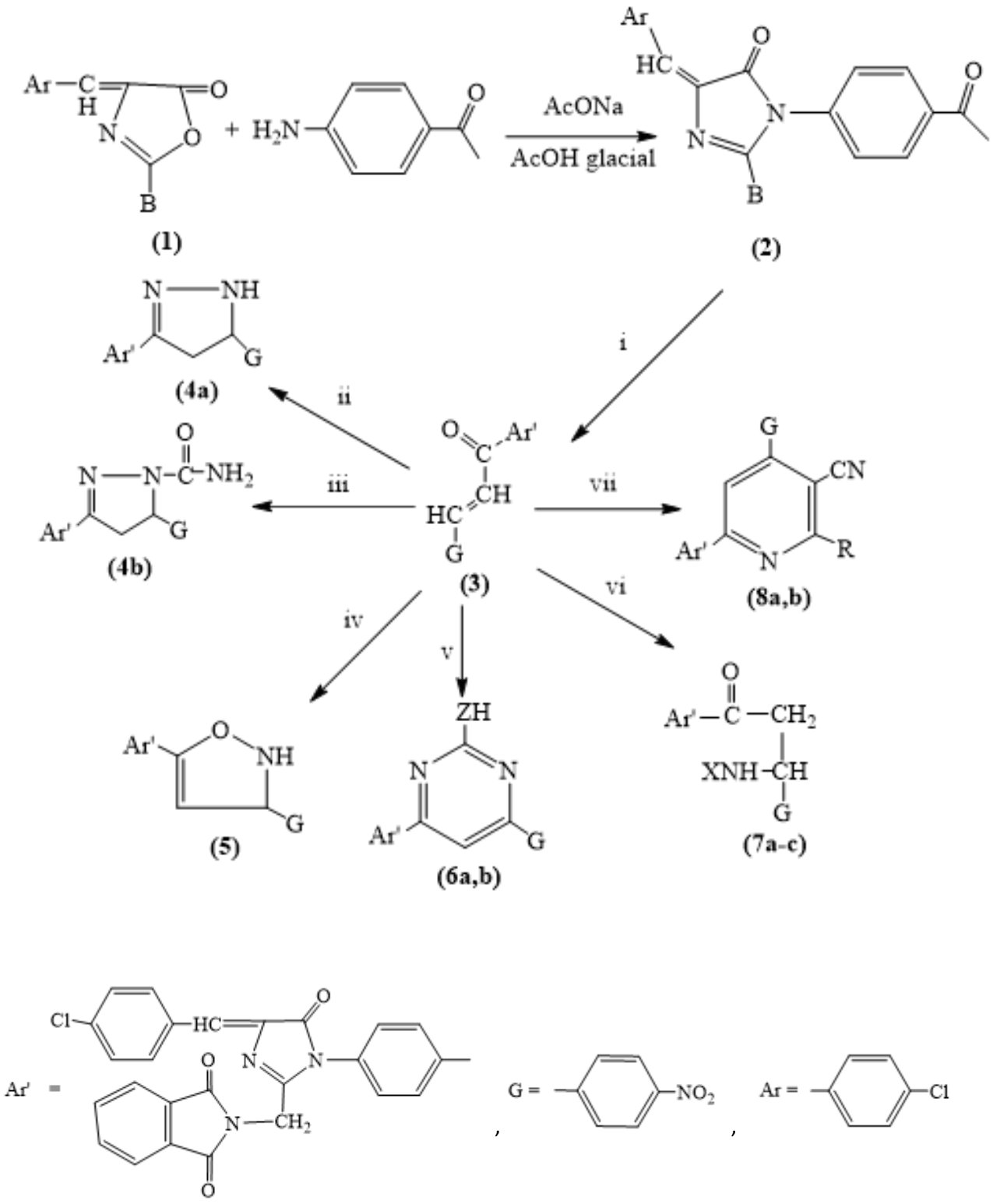
3. Result and discussion

2-((1-(4-acetylphenyl)-4-(4-chlorobenzylidene)-5-oxo-4,5-dihydro-1H-imidazol-2-yl)methyl)isoindoline-1,3-dione **(2)** has been prepared via the condensation of 2-((4-(4-chlorobenzylidene)-5-oxo-4,5-dihydrooxazol-2-yl)methyl)isoindoline-1,3-dione **(1)** with *p*-aminoacetophenone in glacial acetic acid . Its chemical structure was confirmed by showed absorption band at 1682 and 1592 cm^{-1} attributed to CO of cyclic imides and CO respectively. ^{1}H NMR spectrum (DMSO-d6) showed signals at δ ppm 2.5 (s, 3H, CH_3) , 3.4 (s, 2H, CH_2) and 3.3 (s, 1H, CH) , 7.4-8.8 (m ,14H , ArH and olefinic protons).

2-((4-(4-chlorobenzylidene)-1-(4-(3-(4-nitrophenyl)acryloyl)phenyl)-5-oxo-4,5-dihydro-1H-imidazol-2-yl)methyl)isoindoline-1,3-dione **(3)** was synthesized by dissolving a mixture of 2-((1-(4-acetylphenyl)-4-(4-chlorobenzylidene)-5-oxo-4,5-dihydro-1H-imidazol-2-yl)methyl)isoindoline-1,3-dione **(2)** and *p*.nitrobenzaldehyde in absolute ethanol . Its chemical structure was confirmed by showed absorption band at 1767 and 1616 attributed to CO of cyclic imides and CO respectively.

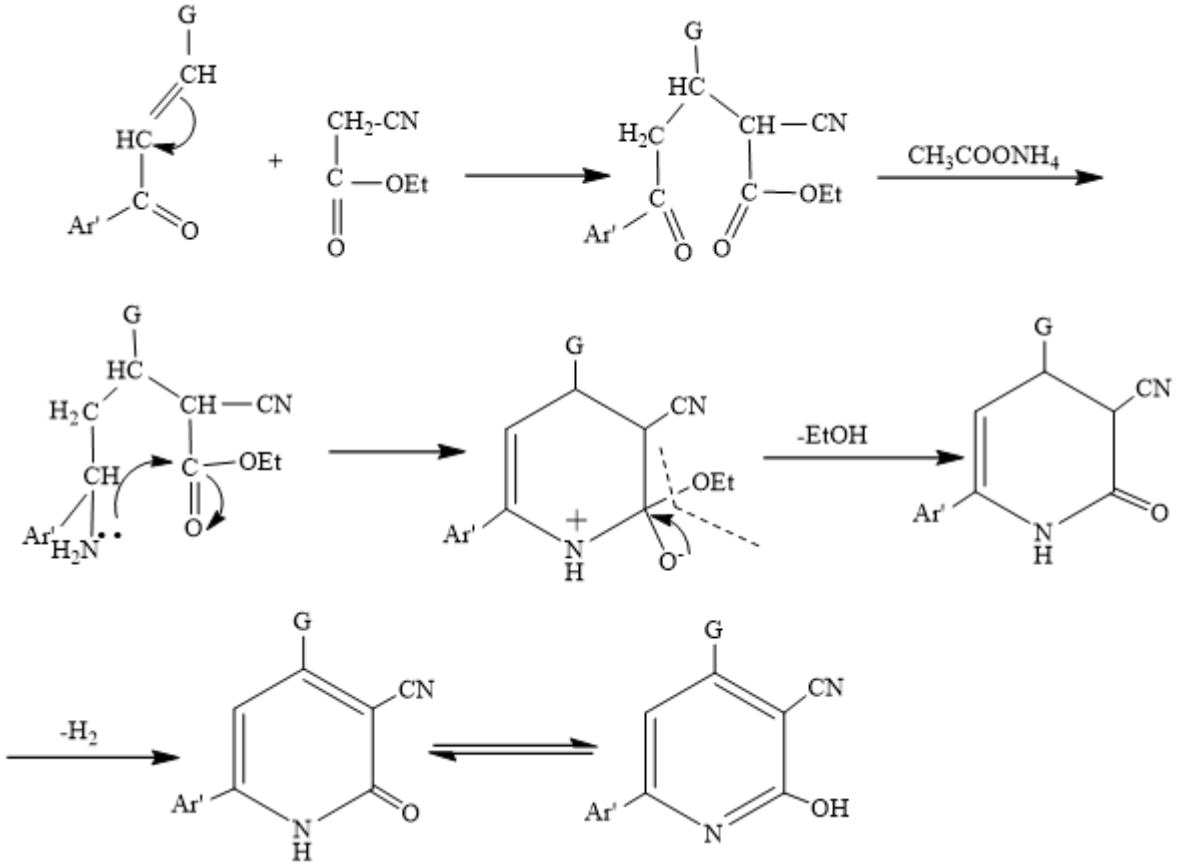
Imidazolone derivative was reacted with various nucleophiles to afford new imidazolone derivatives of biological interest. Consequently, the reaction of compound **(3)** with hydrazine hydrate in boiling ethanol gave the corresponding pyrazoline derivative **(4a)**. The chalcone **(3)** treated also with semicarbazide in pyridine to form the corresponding pyrazoline

carboxamide derivative **(4b)**. Reaction of chalcone **(3)** with hydroxylamine hydrochloride in boiling pyridine afforded the isoxazoline derivative **(5)**. Also, refluxing imidazolone derivative **(3)** with thiourea or/ and urea in boiling sodium ethoxide gave the pyrimidine derivatives **6(a,b)** respectively. Moreover , when compound **(3)** reacted with aromatic amines namely, aniline, *p*-toluidine, *p*-anisidine in boiling ethanol afforded the imidazole derivatives **7(a-c)** respectively. Furthermore, Fusion of compound **(3)** with active methylene compounds namely, ethyl cyanoacetate, malononitrile in presence of ammonium acetate yielded the pyridine derivatives **8(a,b)** respectively. Formation of compounds **(4(a,b) – 8(a,b))** excepting **7(a,b)** takes place via cyclization scheme **(1)**.

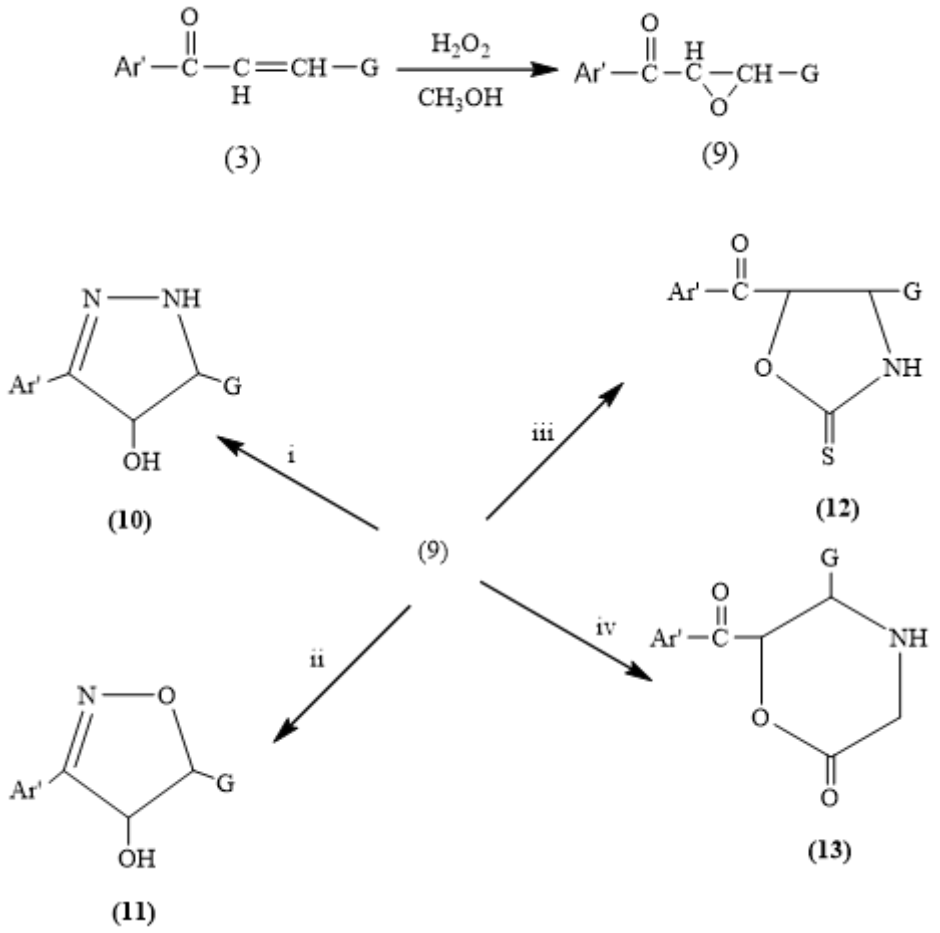


Scheme (1) : (i) CHO-C_6H_4.NO_2-P , KOH, EtOH, sttiring 3h ; (ii) N_2H_4.H_2O ,EtOH,reflux 5h ; (iii) $NH_2CONHNH_2$, pyridine, reflux 5h ; (iv) NH_2OH.HCl, pyridine reflux 5h ; (v) NH_2CZNH_2 [Z=a;S , b;O],C_2H_5ONa, 6h ; (vi) NH_2-X [X= a ; C_6H_5 , b ; C_6H_4.CH_3-P , c ; C_6H_4.OCH_3-P],reflux 3h ; (vii) CH_3COONH_4 , [a; $CNCH_2COOC_2H_5$,{R= OH} , b; NC-CH_2-CN, {R=NH_2}] reflux 8h.

The reaction of (vii) probably occurs through the following mechanism:



Imidazolone derivative **(3)** acts as arow material for preparing oxirane. In details, reaction of **(3)** with mixture of aqueous sodium hydroxide and hydrogen peroxide afforded the derivative of oxirane **(9)** .Hydrazinolysis of oxirane in presence of ethanol gave **(10)**. Similarly, compound **(9)** treated with hydroxyl amine hydrochloride in boiling ethanol formed 2-((4-(4-chlorobenzylidene)-1-(4-(4-hydroxy-5-(4-nitrophenyl)-4,5-dihydroisoxazol-3-yl)phenyl)-5-oxo-4,5-dihydro-1H-imidazol-2-yl)methyl)isoindoline-1,3-dione **(11)**. Compound **(9)** treated also with thiourea in boiling DMF formed **(12)**. Furthermore, compound **(9)** reacted with glycine in boiling DMF gave 2-((4-(4-chlorobenzylidene)-1-(4-(3-(4-nitrophenyl)-6-oxomorpholine-2-carbonyl)phenyl)-5-oxo-4,5-dihydro-1H-imidazol-2-yl)methyl)isoindoline-1,3-dione **(13)**. Formation of compounds **(10-13)** takes place via cyclization scheme **(2)**.

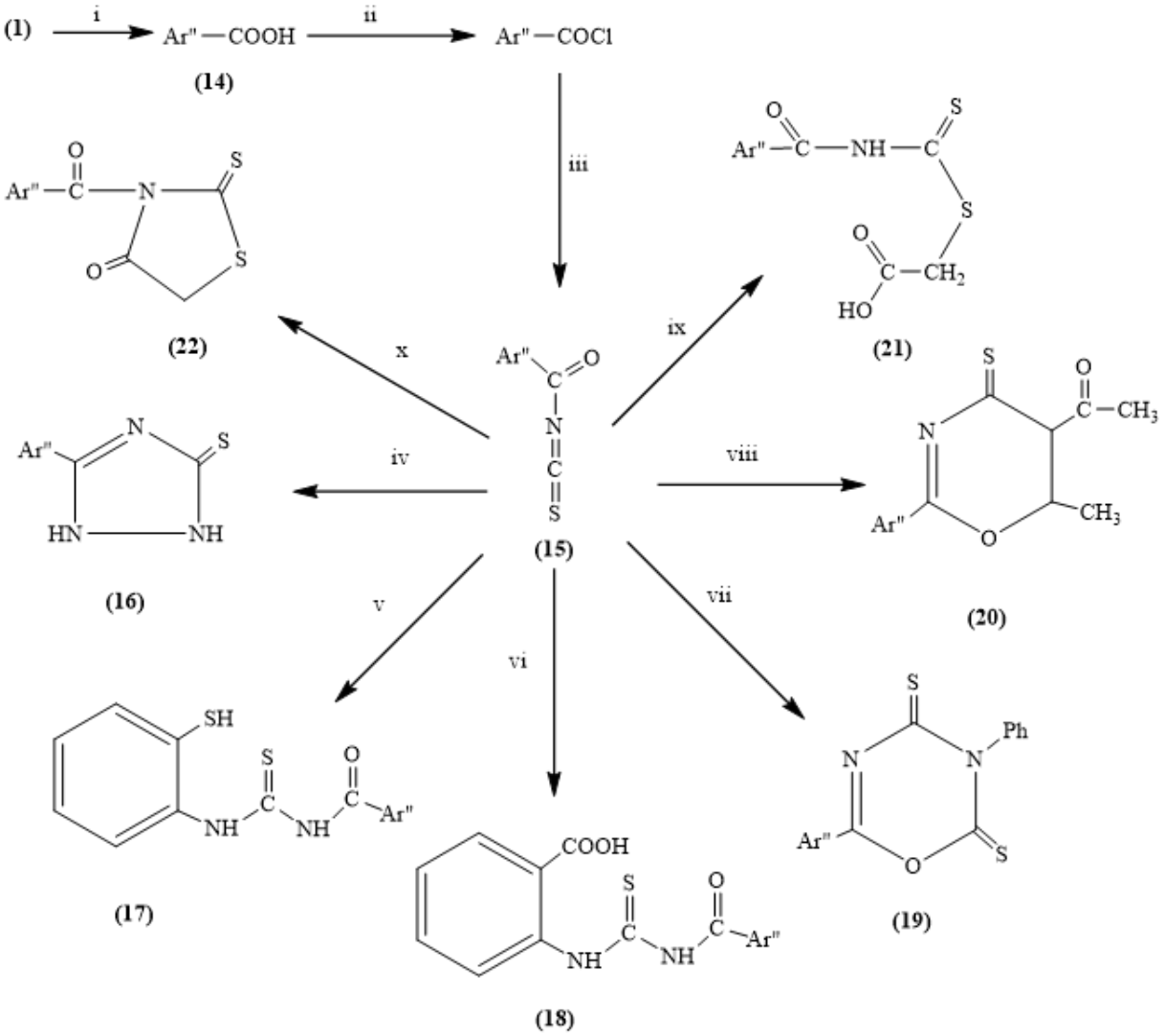


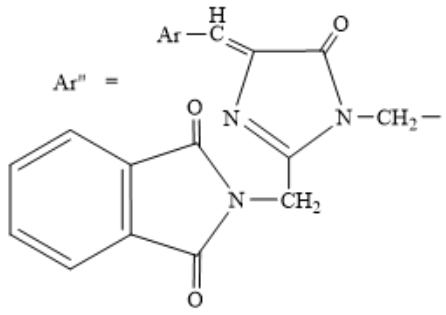
Scheme (2) : (i) H_2N-NH_2 , EtOH, reflux 6h ; (ii) $NH_2OH.HCl$, EtOH, reflux 6h ; (iii) NH_2- CS-NH_2 , DMF, reflux 3h ; (iv) NH_2-CH_2-COOH, DMF, reflux 3h.

We continued our work in this part to prepare a new set of derivatives and studied their biological properties. So, 2-(4-(4-chlorobenzylidene)-2-((1,3-dioxoisoindolin-2-yl)methyl)-5-oxo-4,5-dihydro-1H-imidazol-1-yl)acetic acid **(14)** was prepared from the treatment of 2-((4-(4-chlorobenzylidene)-5-oxo-4,5-dihydrooxazol-2-yl)methyl)isoindoline-1,3-dione **(1)** with glycine in pyridine. Then compound **(14)** reacted with thionyl chloride to give 2-(4-(4-chlorobenzylidene)-2-((1,3-dioxoisoindolin-2-yl)methyl)-5-oxo-4,5-dihydro-1H-imidazol-1-yl)acetyl chloride.

The key precursor 2-(4-(4-chlorobenzylidene)-2-((1,3-dioxoisoindolin-2-yl)methyl)-5-oxo-4,5-dihydro-1H-imidazol-1-yl)acetyl isothiocyanate **(15)** was prepared from the treatment of 2-(4-(4-chlorobenzylidene)-2-((1,3-dioxoisoindolin-2-yl)methyl)-5-oxo-4,5-dihydro-1H-imidazol-1-yl)acetyl chloride with ammonium thiocyanate in presence of dry acetone , and the obtained solution was used to block its decomposition scheme **(3)**.The isothiocyanate derivative has reactivity to some electrophilic and nucleophilic reagents which was investigated to form new heterocyclic systems of expected biological

activity. consequently, compound **(15)** reacted with hydrazine hydrate in boiling dry acetone gave **(16)**. Compound **(15)** reacted also with o-aminothiophenol as amphoteric nucleophile in boiling dry acetone gave thiourea derivative **(17)**. Also, compound **(15)** treated with anthranilic acid in boiling dry acetone to give thiourea derivative **(18)**. Like to the stated behaviour of isothiocyanate derivatives , the 2-((4-(4-chlorobenzylidene)-5-oxo-1-((3-phenyl-2,4-dithioxo-3,4-dihydro-2H-1,3,5-oxadiazin-6-yl)methyl)-4,5-dihydro-1H-imidazol-2-yl)methyl)isoindoline-1,3-dione **(19)** was synthesized from the cycloaddition reaction of compound **(15)** with phenylisothiocyanate in boiling dry acetone. Compound **(15)** reacted also with some active methylene compounds under Michael reaction conditions, such as the treatment of compound **(15)** with acetylacetone in boiling dry acetone and in the presence of triethylamine gave the oxazine derivative **(20)**. When the isothiocyanate **(15)** treated with thioglycolic acid as a sulphur nucleophile in boiling dry acetone gave the adduct **(21)**, that cyclized to thiazolidine derivative **(22)**, via the evaporation of H_2O when boiled with acetic anhydride. Formation of compounds **(16, 19, 20, 22)** takes place via cyclization scheme **(3)**.





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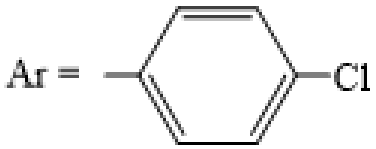
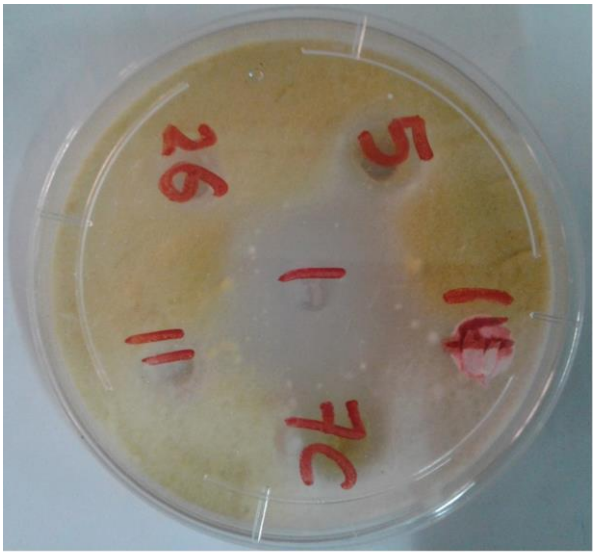
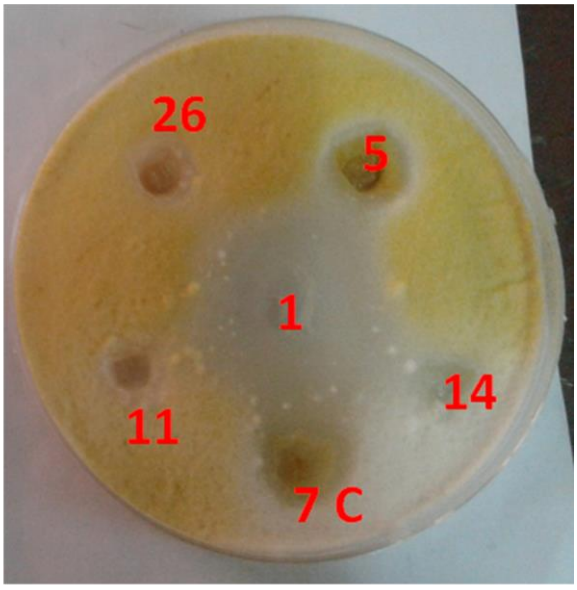
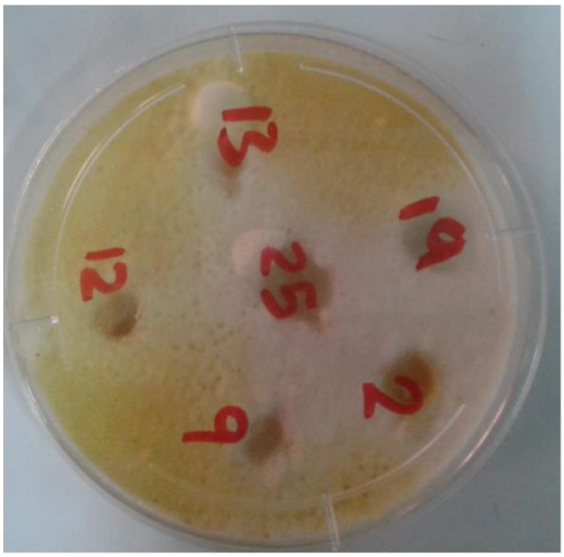


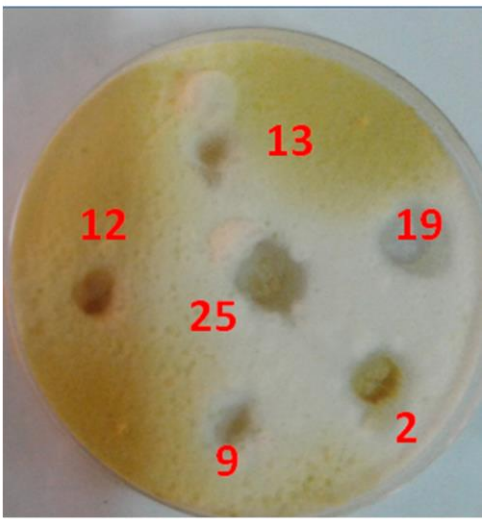
Scheme (3): (i) NH_2-CH_2-COOH , pyridine, reflux 5h ; (ii) $SOCl_2$,reflux 1.5h ; (iii) NH_4SCN ,dry acetone , stirring 2 h . (iv) NH_2NH_2 ,dry acetone, reflux 1h; (v) HS-C_6H_4-NH_2 , dry acetone, reflux 1h; (vi) HOOC-C_6H_4-NH_2 ,dry acetone, reflux 1h; (vii) Ph-NCS , dry acetone, reflux 1h; (viii) $CH_2(CH_3CO)_2$, dry acetone, reflux 3h; (ix) HS-CH_2-COOH , dry acetone, reflux 1h; (x) Ac_2O ,reflux 1h .



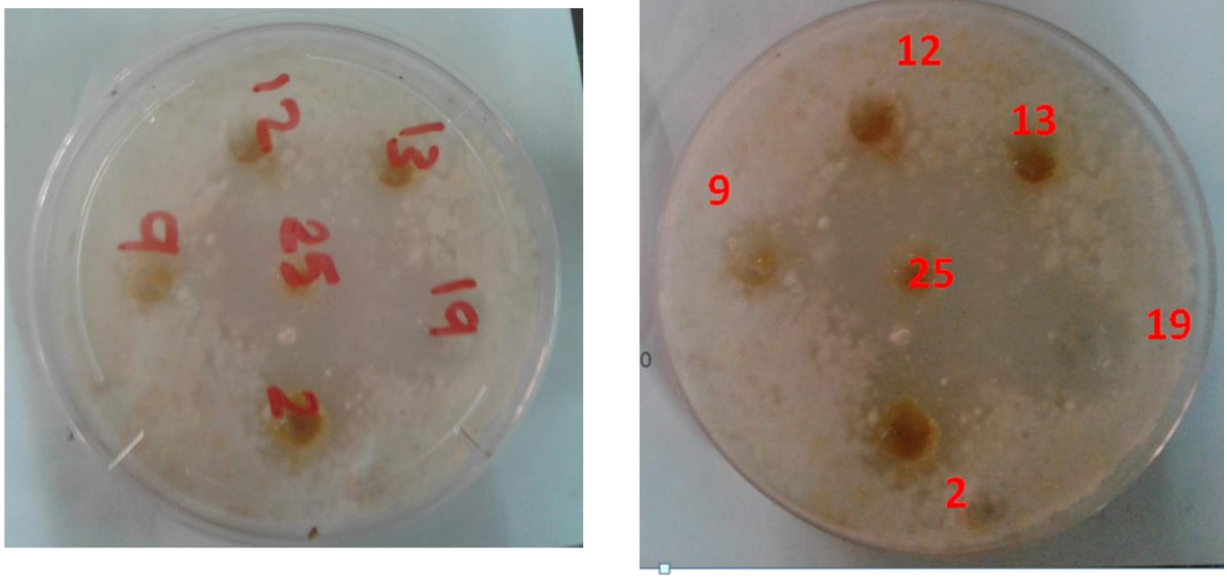


Growth of *Aspergillus flavus* showing the effect of samples 1 is strong positive 26 , 7 C , 5 , 14 are weak positive while 11 is negative . Table 1

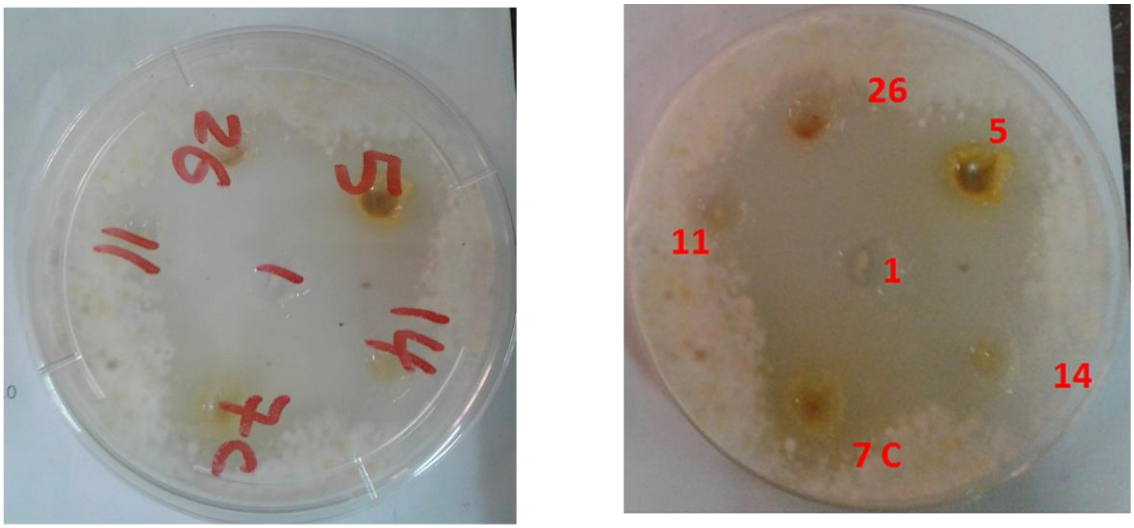




Growth of *Aspergillus flavus* with samples 19 , 25, 2 are positive while 13 , 9 , 12 are negative. Table 1



Growth of *Aspergillus ochrachous* with samples 25 and 19 are strong positive while 2 is weak positive and 12 , 13 and 9 Are negative we think that 25 and 19 are synergized with each other. Table 2



Growth of *Aspergillus ochrachous* with samples 1 , 14 and 5 are strong positive while 11 , 7 C and 26 Are weak positive we think that all of these samples are synergized with each other. Table 2

In case of *Aspergillus flavus*

Inhibition zone diameter (mm)

samples	1	2	3	average
19	1.2	1.3	1.4	1.30
13	Nil	Nil	Nil	Nil
25	1.2	1.3	1.3	1.27
12	Nil	Nil	Nil	Nil
9	Nil	Nil	Nil	Nil
2	1.2	1.3	1.4	1.30
11	Nil	Nil	Nil	Nil
26	1.5	1.2	1.3	1.33
5	1.5	1.4	1.4	1.43
1	3	3	3	3.00
14	1	1	1	1.00
7c	1.4	1.5	1.5	1.47

In case of *Aspergillus ochrachous*

Inhibition zone diameter (mm)

samples	1	2	3	average
5	2.5	2.4	2.5	2.47
14	2.7	2.8	2.9	2.80
7c	2.3	2.7	2.6	2.533333
11	1.6	1.4	1.7	1.566667
26	2	1.9	2	1.966667
1	3.8	3.9	4	3.9
12	Nil	Nil	Nil	Nil
2	2	1.9	2	1.966667
19	2.2	2.1	2	2.1
13	Nil	Nil	Nil	Nil
9	Nil	Nil	Nil	Nil
25	2.5	2.7	2.4	2.533333

Note : (19) =(18) , (25) =(21) , (26) =(22)

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