Research Article

Synthesis and Microbial Activity of Novel 3-Methyl-2-pyrazolin-5-one Derivatives

Mohamed S. Mostafa,¹ Nasser M. Abd El-Salam,² and Othman Y. Alothman³

¹ Chemistry Department, Faculty of Science, Jazan University, P.O. Box 2079, Saudi Arabia

² Riyadh Community College, King Saud University, P.O. Box 28095, Riyadh 11437, Saudi Arabia

³ Department of Chemical Engineering, College of Engineering, King Saud University, P.O. Box 800, Riyadh 11421, Saudi Arabia

Correspondence should be addressed to Mohamed S. Mostafa; msmostafaa@yahoo.com

Received 18 December 2012; Revised 14 March 2013; Accepted 27 March 2013

Academic Editor: Philippe Jeandet

Copyright © 2013 Mohamed S. Mostafa et al. This is an open access article distributed under the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

2-Oxo-2*H*-chromene-3-carbohydrazide derivatives **2a,b** react with 2-[[4-(substituted thiazol-2-yl)iminoethyl]-phenyl]hydrazono-3-oxo-butiric acid ethyl esters **4a-c** to give 3-methyl-1-[(2-oxo-2*H*-chromen-3-yl) carbonyl]-4-[[4-(substituted thiazol-2-yl)iminoethyl]-phenyl]hydrazono-2-pyrazolin-5-one derivatives **5a-f**. A considerable increase in the reaction rate had been observed with better yield using microwave irradiation for the synthesis of compounds **2a, b, 3a-c**, and **5a-f**. The synthesized products were tested against *B. subtilis*, *S. aureus*, and *E. coli* as well as *C. albicans* compared with tetracycline and nystatin as reference drugs.

1. Introduction

Pyrazolones are an increasingly popular functionality with wide-range applications. They are used as antibacterial [1], antifungal [2], anti-inflammatory [3], cytotoxic [4], analgesic [5], antiviral [6], and SARS-corona virus 3C-like protease inhibitors [7] agents. Furthermore, 4-(1,3-benzothiazol-2-yl)hydraziny lidene-2,4-dihydro-3*H*-pyrazol-3-one derivatives have shown good antioxidant activity [8]. On the other hand, the iminothiazoline derivatives have been reported to exhibit antibacterial and antifungal activities [9, 10]. Coumarin derivatives possess various biological activities namely antibacterial [11, 12], antifungal [13], antitumor [14], anticoagulant [15], and anti-inflammatory agents [16]. These compounds are used as additives in food and cosmetics [17], dispersed fluorescent brightening agents, and as dyes for tuning lasers [18]. Microwave irradiation is well known to promote the synthesis of a variety of organic compounds [19–21] where chemical reactions are accelerated because of selective absorption of microwave by polar molecules. In view of these observations we report herein the synthesis of some pyrazolone derivatives, using conventional and microwave irradiated methods, with the hope to get better antimicrobial agents.

2. Experimental

Melting points were estimated using a Stuart apparatus in open capillaries. Purity of compounds was checked by TLC. IR spectra were recorded as KBr pellets on a Jasco FTIR 460 plus spectrophotometer. ¹H-NMR spectra were recorded using a Bruker AV 500 MHz spectrometer using DMSO-*d*₆ as solvent and TMS as an internal standard. The mass spectral data were obtained with a Micro Spectrometer model 7070 at 70 eV and a 90°C inlet temperature. Elemental analyses were performed on a Perkin-Elmer 240 microanalyser in the Faculty of Science, Cairo University. Microwave irradiations (MWIs) were carried out in a SANYO EM-700T domestic oven (700 W).

3. Synthesis of 2-Oxo-2*H*-Chromene-3-carbohydrazides (**2a,b**)

3.1. Method I. A mixture of ethyl-2-oxo-2*H*-chromene-3-carboxylates (**1a,b**) (0.01 mole) and hydrazine hydrate 98% (0.5 mole) was refluxed for 2 h. The reaction mixture was poured into water and then the separated solid was filtered off and recrystallized from ethanol to give **2a, b** (Tables 1 and 2).

TABLE 1: Characterization data of synthesized compounds.

Comp	M.P. (°C)	Solvent of crystn.	Yield (%)	Formula (M.W.)	Analysis calcd./found			
					C	H	N	S
2a	144–146	Ethanol	75	C ₁₀ H ₈ N ₂ O ₃	58.82	3.95	13.71	—
				(204.185)	58.81	3.97	13.75	—
2b	210–212	Ethanol	60	C ₁₀ H ₇ BrN ₂ O ₃	42.43	2.49	9.89	—
				(283.081)	42.48	2.46	9.91	—
3a [23]	201–204	Ethanol	62	C ₁₁ H ₁₁ N ₃ S	60.80	5.10	19.33	14.75
				(217.29)	60.81	5.13	19.38	14.71
3b	230–232	Ethanol	58	C ₁₂ H ₁₃ N ₃ S	62.85	5.71	18.32	13.98
				(229.301)	62.86	5.73	18.37	14.01
3c	271–273	EtOH-H ₂ O	50	C ₁₅ H ₁₃ N ₃ S	67.38	4.90	15.71	11.99
				(267.35)	67.40	4.91	15.69	11.96
4a [23]	165–167	Ethanol	66	C ₁₇ H ₁₈ N ₄ O ₃ S	56.96	5.06	15.63	8.94
				(358.416)	56.98	5.04	15.67	8.98
4b	186–188	Ethanol	62	C ₁₈ H ₂₀ N ₄ O ₃ S	58.04	5.41	15.04	8.60
				(372.443)	58.09	5.40	15.07	8.66
4c	140–142	Ethanol	60	C ₂₁ H ₂₀ N ₄ O ₃ S	61.74	4.93	13.71	7.84
				(408.476)	61.78	4.96	13.69	7.88
5a	208–210	Ethanol	60	C ₂₅ H ₁₈ N ₆ O ₄ S	60.23	3.63	16.85	6.43
				(498.517)	60.20	3.66	16.80	6.47
5b	232–234	Ethanol	54	C ₂₆ H ₂₀ N ₆ O ₄ S	60.92	3.93	16.39	6.25
				(512.544)	60.94	3.98	16.42	6.30
5c	222–224	DMF	57	C ₂₉ H ₂₀ N ₆ O ₄ S	63.49	3.67	15.32	5.84
				(548.577)	63.51	3.62	15.38	5.88
5d	280–282	DMF	50	C ₂₅ H ₁₇ BrN ₆ O ₄ S	52.00	2.96	14.55	5.55
				(577.413)	52.08	3.99	14.51	5.59
5e	250–252	DMF	48	C ₂₆ H ₁₉ BrN ₆ O ₄ S	52.80	3.23	14.20	5.42
				(591.44)	52.78	3.20	14.28	5.45
5f	264–267	Pet-ether	37	C ₂₉ H ₁₉ BrN ₆ O ₄ S	55.51	3.05	13.39	5.10
				(627.473)	55.50	3.03	13.41	5.12

TABLE 2: Comparison of conventional and microwave synthesis.

Compd.	Conventional		Microwave	
	% yield	t/hrs	% yield	t/min
2a	75	2	94	3
2b	60	2	98	3
3a	64	4	82	3
3b	58	4	86	3.5
3c	50	4	89	2
5a	60	8	93	3
5b	54	8	88	3
5c	57	8	82	3
5d	50	9	91	3
5e	48	8	83	3
5f	37	8	81	4

3.2. *Method II.* Hydrazine hydrate (0.01 mole), **1a,b** (0.01 mole), and absolute ethanol (2 mL) were irradiated in an Erlenmeyer flask under MWI for 3 min. The reaction mixture

was cooled, and the separated solid was filtered off and washed with water to yield **2a,b** (Table 3).

4. Synthesis of 1-(4-Aminophenyl)-1-(substituted thiazol-2-yl)iminoethanes **3a–c**

4.1. *Method I.* A mixture of 4-aminoacetophenone (0.01 mole) and the appropriate amine, namely, (2-aminothiazole, 2-amino-5-methylthiazol and 2-aminobenzothiazole) (0.01 mole for each) in DMF (30 mL) was heated under reflux for 4 h. The precipitated solid formed after cooling was collected by filtration. Then, it was washed with water, dried, and crystallized to afford **3a–c** (Tables 1 and 2).

4.2. *Method II.* A solution of 4-aminoacetophenone (0.01 mole) in methanol (30 mL) and 2-aminothiazoles (0.01 mole) was put in round-bottomed flask placed in a microwave oven and irradiated for 2.0–3.5 min, and then the solvent was removed by vacuum distillation. The solid product was filtered, dried, and recrystallized from ethanol to give **3a–c** (Table 3).

TABLE 3: Spectral data of synthesized compounds.

Comp.	IR cm^{-1}	$^1\text{H-NMR}$ δ ppm	MS m/z
2a	3304–3245 (NH_2), 3078 (NH), 1730 (lactone C=O) and 1682 cm^{-1} (amidic CO).	7.01–7.48 (m, 3H, ArH), 9.36 (s, 1H, CH-4), 11.14 (s, 1H, NH) and 12.01 (s, 2H, NH_2).	M^+ : 204 (100%)
2b	3311–3239 (NH_2), 3046 (NH), 1721 (lactone C=O) and 1680 cm^{-1} (amidic CO)	7.12–7.54 (m, 3H, ArH), 9.41 (s, 1H, CH-4), 11.21 (s, 1H, NH) and 12.11 (s, 2H, NH_2).	$\text{M}+1$: 284 (32.67%), M^+ : 283 (61.06%)
3b	3400–3312 (NH_2), 1640 (C=N)	2.01 (s, 3H, CH_3), 2.72 (s, 3H, CH_3), 6.88 (s, 2H, NH_2 , D_2O -exchangeable, 7.22–7.82 (m, 5H, ArH).	M^+ : 229 (100%), 97 (55.98%).
3c	3400–3312 (NH_2), 1640 (C=N)	2.78 (s, 3H, CH_3), 6.10 (s, 2H, NH_2 , D_2O -exchangeable, 7.35–7.68 (m, 8H, ArH).	M^+ : 267 (46.11%), 133 (100%).
4b	1760 (C=O), 1630 (C=N), 1560 (N=N).	0.91 (s, 3H, CH_3), 1.21 (t, 3H, OCH_2CH_3), 2.60 (s, 3H, CH_3), 2.88 (s, 3H, CH_3), 3.0 (s, 1H, CH), 4.20–4.22 (q, 2H, OCH_2CH_3), 6.70–7.60 (m, 5H, ArH).	M^+ : 372 (88.34%), 328 (22.56%), 254 (43.87%), 156 (52%).
4c	1755 (C=O), 1622 (C=N), 1560 (N=N).	1.27 (t, 3H, OCH_2CH_3), 2.57 (s, 3H, CH_3), 2.80 (s, 3H, CH_3), 3.22 (s, 1H, CH), 4.27–4.29 (q, 2H, OCH_2CH_3), 6.66–7.98 (m, 8H, ArH).	M^+ : 408 (22.11%), 364 (56.54%), 290 (78.23%), 156 (23.11%).
5a	3202 (NH), 1710 (lactone CO), 1677 (CO), 1586 (C=N).	2.03 (s, 3H, CH_3), 2.92 (s, 3H, CH_3), 4.58 (s, 1H, CH), 7.31–7.52 (m, 10H, ArH), 9.72 (s, 1H, CH-4), 11.40 (s, 1H, NH, disappeared after D_2O exchange)	M^+ : 498 (23.11%), 267 (52.34%), 229 (26.87%), 121 (7.08%), 92 (17.58%), 84 (39.76%).
5b	3106 (NH), 1719 (lactone CO), 1680 (CO), 1582 (C=N).	1.44 (s, 3H, CH_3), 2.03 (s, 3H, CH_3), 2.88 (s, 3H, CH_3), 4.61 (s, 1H, CH), 7.31–7.52 (m, 9H, ArH), 9.66 (s, 1H, CH-4), 11.07 (s, 1H, NH, disappeared after D_2O exchange)	M^+ : 512 (42.26%), 267 (65%), 242 (36.98%), 121 (19.78%), 97 (14.44%), 92 (22.32%)
5c	3135 (NH), 1713 (lactone CO), 1654 (CO), 1567 (C=N).	2.45 (s, 3H, CH_3), 2.99 (s, 3H, CH_3), 4.28 (s, 1H, CH), 6.78–8.89 (m, 12H, ArH), 9.21 (s, 1H, CH-4), 11.11 (s, 1H, NH, disappeared after D_2O exchange)	M^+ : 548 (100%), 279 (15.47%), 267 (25.96%), 133 (44.11%), 121 (45.34%), 92 (13.67%).
5d	3065 (NH), 1722 (lactone CO), 1682 (CO), 1560 (C=N).	2.19 (s, 3H, CH_3), 2.86 (s, 3H, CH_3), 4.65 (s, 1H, CH), 7.11–7.95 (m, 9H, ArH), 9.70 (s, 1H, CH-4), 11.32 (s, 1H, NH, disappeared after D_2O exchange)	$\text{M}+1$: 578 (22.52%), M^+ : 577 (78.93%), 344 (32.33%), 229 (21.45%), 84 (76.23%).
5e	3116 (NH), 1709 (lactone CO), 1673 (CO), 1551 (C=N).	1.31 (s, 1H, CH_3), 2.73 (s, 3H, CH_3), 2.99 (s, 3H, CH_3), 4.95 (s, 1H, CH), 7.11–7.54 (m, 8H, ArH), 9.72 (s, 1H, CH-4), 11.98 (s, 1H, NH, disappeared after D_2O exchange)	$\text{M}+1$: 520 (98%), M^+ : 519 (38%), 344 (16.95%), 242 (68.23%), 121 (19%), 97 (23.31%).
5f	3109 (NH), 1725 (lactone CO), 1686 (CO), 1509 (C=N).	2.19 (s, 3H, CH_3), 2.86 (s, 3H, CH_3), 4.65 (s, 1H, CH), 6.91–8.12 (m, 11H, ArH), 9.04 (s, 1H, CH-4), 10.56 (s, 1H, NH, disappeared after D_2O exchange)	$\text{M}+1$: 628 (15.12%), M^+ : 627 (34%), 344 (71.71%), 279 (51.11%), 133 (55.04%).

5. Synthesis of 2-[[4-(Substituted thiazol-2-yl)iminoethyl]-phenyl]Hydrazono}-3-oxo-Butyric Acid Ethyl Ester **4a–c**

A solution of sodium nitrite (0.01 mole) in water (10 mL) was added to an ice-cooled mixture of **3a–c** (0.01 mole) in concentrated HCl (10 mL) and water (10 mL). The diazotized compound was dropped while cooling with stirring over a cold mixture of ethyl acetoacetate (0.01 mole) and sodium acetate (2 g in 10 mL water) in ethanol (20 mL). The reaction mixture was stirred at room temperature for 8 h. The precipitated solid was collected by filtration. Then it was washed with water, dried, and recrystallized to afford **4a–c** (Tables 1 and 2).

6. Synthesis of 3-Methyl-1-[(2-oxo-2H-chromen-3-yl) carbonyl]-4-[[4-(substituted thiazol-2-yl)iminoethyl]-phenyl]hydrazono}]-2-pyrazolin-5-one Derivatives (**5a–f**)

6.1. Method I. A mixture of **4a–c** (0.002 mole for each) and (**2a,b**) (0.002 mole for each) in acetic acid (15 mL) was refluxed for 8 h. The reaction mixture was then allowed to stand overnight. After conclusion of the reaction (TLC), the reaction mixture was poured onto crushed ice to give sticky product which was extracted from ether for three times.

FIGURE 1

FIGURE 2

FIGURE 3

The organic layer was washed with water. Then, it was dried with MgSO_4 and filtered. After that, the solvent was removed under vacuum to give **5a-f** (Tables 1 and 2).

6.2. Method II. A mixture of **4a-c** (0.01 mole for each), **2a,b** (0.01 mole for each), and glacial acetic acid (2 mL) in an Erlenmeyer flask was exposed to pulsed microwave irradiation using microwave oven for 3 min. The reaction mixture was poured onto crushed ice; the separated solid was filtered, then washed with water, and dried to give the pyrazolin-5-ones **5a-f** (Table 3).

7. Results and Discussion

Treatment of ethyl-2-oxo-2H-chromene-3-carboxylates (**1a,b**) [22] with hydrazine hydrate under heating reflux afforded 2-oxo-2H-chromene-3-carbohydrazide derivatives **2a,b**. Again, compounds **2a,b** were obtained in a 94–98% yield by irradiation of (**1a,b**) with hydrazine hydrate in ethanol under MWI for 3 min (Figure 1).

The IR (KBr) of compounds **2a,b** displayed absorption bands at 3321(NH_2), 3078(NH), 1730(lactone $\text{C}=\text{O}$), and

1682 cm^{-1} for amidic CO. The structure of **2b** was also confirmed by its mass spectrum that shows molecular ion peaks (M^+) at m/z 283(61.06%, ^{79}Br) and m/z 284(32.67%, ^{81}Br).

It has been found that the 4-aminoacetophenone reacted with 2-amino-5-methylthiazole or 2-aminobenzothiazole in DMF to give the imino derivatives (**3b,c**), similar to compound (**3a**) [23]. It was found that the higher yields of compounds **3a-c** were obtained at 500 watt for 2-3 min of microwave irradiation (Figure 2).

IR spectrum (KBr) of **3b**, as an example, showed bands at 3400–3388 (NH_2), and 1640 cm^{-1} for $\text{C}=\text{N}$. The ^1H -NMR spectrum ($\text{DMSO}-d_6$) of **3b** showed signals at δ 2.01 (s, 3H, CH_3), 2.72 (s, 3H, CH_3), 6.88 (s, 2H, NH_2 , D_2O -exchangeable), and 7.22–7.82 ppm (m, 5H, ArH). Diazotization of **3a-c** followed by coupling with ethyl acetoacetate in presence of sodium acetate gave 2-[[4-(substituted thiazol-2-yl)iminoethyl]-phenyl]hydrazono-3-oxo-butyric acid ethyl esters **4a-c** (Figure 3).

IR spectra of **4a-c** revealed no absorption band in amino group region; furthermore, it displayed absorption bands at 1760 and 1560 cm^{-1} for ester group and ($\text{N}=\text{N}$), respectively.

FIGURE 4

The $^1\text{H-NMR}$ spectrum (DMSO-d_6) of compound **4c**, as an example, exhibited signals at δ 1.27 (t, 3H, OCH_2CH_3), 2.57 (s, 3H, CH_3), 2.80 (s, 3H, CH_3), 3.22 (s, 1H, CH), 4.27–4.29 (q, 2H, OCH_2CH_3) and 6.66–7.98 ppm (m, 8H, ArH).

Refluxing compounds **2a,b** and **4a–c** in acetic acid for 8 hrs lead to 3-methyl-1-[(2-oxo-2H-chromen-3-yl) carbonyl]-4-[(4-(substituted thiazol-2-yl)iminoethyl)-phenyl]hydrazono]-2-pyrazolin-5-one derivatives **5a–f**. These pyrazolin-5-one derivatives were obtained in good yield through the irradiation of a mixture of **2a,b** and **4a–c** using microwave oven for 3–4 min (Figure 4).

The $^1\text{H-NMR}$ spectrum (DMSO-d_6) of compounds **5a**, as an example, showed signals at δ 2.03 (s, 3H, CH_3), 2.92 (s, 3H, CH_3), 4.58 (s, 1H, CH), 7.31–7.52 (m, 10H, ArH), 9.72 (s, 1H, CH-4), and 11.40 ppm (s, 1H, NH, disappeared after D_2O exchange). The infrared spectrum of compounds **5a–f** revealed absorption bands characteristic for NH, lactone CO, CO, and $\text{C}=\text{N}$. The mass spectra of compounds containing bromine atoms (**5d, 5e**, and **5f**) showed fragments corresponding to the typical bromine isotope (^{79}Br and ^{81}Br) patterns. Thus, the mass spectrum of **5d** shows its $\text{M}+1$ and M^+ peaks at m/z 578 (22.52%) and 577 (78.93%), respectively (Table 3).

8. Antimicrobial Activity

The antimicrobial activity of new compounds was investigated against a variety of microorganisms, including the gram-positive bacteria *Bacillus subtilis* and *Staphylococcus aureus* and the gram-negative bacteria *Escherichia coli* as well as the unicellular fungi *Candida albicans*. The agar plate disc-diffusion method [24] was used to assess the activity of the compounds. Sterilized filter paper discs (5 mm in diameter) were wetted with 10 μL each of a solution of the

tested compound (10 mg/mL of the compound in DMF). The discs were then allowed to dry and were placed on the surface of agar plates seeded with the test organism. Nutrient agar was used for bacterial plating and sabourauds dextrose agar for fungi. Each plate contained 15 mL of the agar medium, previously seeded with 0.2 mL of an 18 h-old broth culture of each organism. The inoculated plates were incubated at 37°C for 48 h with the test discs in place, and the inhibition zones were measured in mm. Controls including the use of the solvent DMF without test compounds that showed no antimicrobial activity for this solvent. The antibacterial reference tetracycline discs and the antifungal reference nystatin discs were tested concurrently as standards.

Concerning the data of antimicrobial activity in Table 4, some of the synthesized compounds showed antibacterial activity comparable to that of tetracycline (the reference drug used). Concerning the activity against gram-positive bacteria (*Bacillus subtilis*), 3-methyl-1-[(2-oxo-2H-chromen-3-yl)carbonyl]-4-[(4-thiazol-2-yl)iminoethyl)phenyl]hydrazon]-2-pyrazolin-5-one derivative (**5a**) showed excellent activity, and compounds **5b** and **5d** exhibited good activity, whereas compounds **4c, 5e**, and **5f** showed moderate activity. All synthesized compounds exhibited moderate activity against *Staphylococcus aureus*. Compounds **5a** and **5f** showed excellent activity against gram-negative bacteria *Escherichia coli*. On the other hand, the unicellular fungi *Candida albicans* showed high responses to **5d**.

9. Conclusion

This paper describes the synthesis, spectral characterization, and screening of antimicrobial activity of some 3-methylpyrazolin-5-one derivatives bearing side chains, imino-thiazole-phenylhydrazono derivatives, and coumarin

TABLE 4: Antimicrobial activity of prepared compounds.

Compd. no.	Inhibition zones (mm)			
	Gram +ve bacteria <i>B. subtilis</i>	<i>S. aureus</i>	Gram -ve bacteria <i>E. coli</i>	Fungi <i>C. albicans</i>
2a	18	14	12	19
2b	13	10	11	20
3b	16	14	10	13
3c	14	12	15	11
4b	17	18	18	22
4c	22	17	16	20
5a	29	10	21	19
5b	23	10	11	18
5c	19	15	20	10
5d	23	10	19	25
5e	22	14	13	15
5f	20	18	22	13
Tetracycline	28	26	20	—
Nystatin	—	—	—	24

moiety. Microwave-assisted synthesis was also used to improve the yield and reaction time. The synthesized compounds showed a wide range of potentially promising antimicrobial activities.

Acknowledgment

The authors extend their appreciation to the Deanship of Scientific Research at King Saud University for funding the work through the research group project no. RGP-VPP-133.

References

- [1] P. T. Pavlov, A. F. Goleneva, A. E. Lesnov, and T. S. Prokhorova, "Biological activity of some pyrazolone derivatives," *Pharmaceutical Chemistry Journal*, vol. 32, no. 7, pp. 370–372, 1998.
- [2] D. Singh and D. Singh, "Synthesis and antifungal activity of some 4-arylmethylene derivatives of substituted pyrazolones," *Journal of the Indian Chemical Society*, vol. 68, no. 3, pp. 165–167, 1991.
- [3] G. Mariappan, B. P. Saha, L. Sutharson, and A. Haldar, "Synthesis and bioactivity evaluation of pyrazolone derivatives," *Indian Journal of Chemistry B*, vol. 49, no. 12, pp. 1671–1674, 2010.
- [4] H. P. Devnath and R. Islam, "Synthesis of some pyrazolone derivatives from ciprofloxacin and study of their cytotoxicity," *Bangladesh Journal of Pharmacology*, vol. 5, no. 1, pp. 30–34, 2010.
- [5] M. Amir and S. Kumar, "Synthesis and anti-inflammatory, analgesic, ulcerogenic and lipid peroxidation activities of 3,5-dimethyl pyrazoles, 3-methyl pyrazol-5-ones and 3,5-disubstituted pyrazolines," *Indian Journal of Chemistry B*, vol. 44, no. 12, pp. 2532–2537, 2005.
- [6] A. N. Evstropov, V. E. Yavorovskaya, E. S. Vorobev et al., "Synthesis and antiviral activity of antipyrine derivatives," *Pharmaceutical Chemistry Journal*, vol. 26, no. 5, pp. 426–430, 1992.
- [7] R. Ramajayam, K. P. Tan, H. G. Liu, and P. H. Liang, "Synthesis and evaluation of pyrazolone compounds as SARS-coronavirus 3C-like protease inhibitors," *Bioorganic and Medicinal Chemistry*, vol. 18, no. 22, pp. 7849–7854, 2010.
- [8] K. Siva and A. Rajasekharan, "Synthesis and Characterisation, in vitro antioxidants activity of N-mannich base of pyrazolone derivatives," *International Journal of Research of Pharmacy and Chemistry*, vol. 2, no. 2, pp. 327–337, 2012.
- [9] A. Gürsoy and N. Karali, "Synthesis, characterization and primary antituberculosis activity evaluation of 4-(3-Coumarinyl)-3-benzyl-4-thiazolin-2-one benzylidenehydrazones," *Turkish Journal of Chemistry*, vol. 27, no. 5, pp. 545–551, 2003.
- [10] S. Hiremath, K. Swamy, and B. Mruthyunjayaswamy, "Synthesis and antimicrobial activity of 4-amino-3-(2'-phenylindol-3'-yl)-2-imino-4-thiozoline-5-carbonitriles and their derivatives," *Journal of the Indian Chemical Society*, vol. 69, p. 87, 1992.
- [11] O. A. Abd Allah, "Synthesis and biological studies of some benzopyrano[2,3-c]pyrazole derivatives," *Farmaco*, vol. 55, no. 9-10, pp. 641–649, 2000.
- [12] A. M. El-Agrody, M. S. Abd El-Latif, N. A. El-Hady, A. H. Fakery, and A. H. Bedair, "Heteroaromatization with 4-Hydroxycoumarin Part II: synthesis of some new pyrano[2,3-d]pyrimidines, [1,2,4]triazolo[1,5-c]pyrimidines and pyrimido [1,6-b]-[1,2,4]triazine derivatives," *Molecules*, vol. 6, no. 6, pp. 519–527, 2001.
- [13] M. A. Al-Haiza, M. S. Mostafa, and M. Y. El-Kady, "Synthesis and biological evaluation of some new coumarin derivatives," *Molecules*, vol. 8, no. 2, pp. 275–286, 2003.
- [14] F. Belluti, G. Fontana, L. D. Bo, N. Carenini, C. Giommarelli, and F. Zunino, "Design, synthesis and anticancer activities of stilbene-coumarin hybrid compounds: identification of novel proapoptotic agents," *Bioorganic and Medicinal Chemistry*, vol. 18, no. 10, pp. 3543–3550, 2010.
- [15] H. El-Diwani and H. El-Sahrawi, "Synthesis and anticoagulant, antipyretic and analgesic activities of some 4H-1benzopyran-4-one derivatives," *Indian Journal of Chemistry B*, vol. 34, pp. 27–31, 1995.
- [16] K. N. Venugopala and B. S. Jayashree, "Synthesis of carbamides of 2-amino-4-(6-bromo-3-coumarinyl) thiazole as analgesic and antiinflammatory agents," *Indian Journal of Heterocyclic Chemistry*, vol. 12, no. 4, pp. 307–310, 2003.
- [17] R. O'Kennedy and R. Thornes, *Coumarins: Biology, Applications and Mode of Action*, John Wiley & Sons, Chichester, UK, 1997.
- [18] M. Zahradnik, *The Production and Application of Fluorescent Brightening Agent*, John Wiley & Sons, Chichester, UK, 1992.
- [19] P. Gothwal, G. Malhotra, and Y. K. Srivastava, "Microwave Assisted Synthesis and antimicrobial activities of some 3-[4'-(4'-nitrophenoxy)-phenyl]-5-(substituted aryl)-2-pyrazoline-1-thiocarbamides," *International Journal of Green and Herbal Chemistry*, vol. 1, no. 1, pp. 39–45, 2012.
- [20] B. Parashar, S. Bhardwaj, S. Sharma, G. Gupta, V. Sharma, and P. Punjabi, "Comparative conventional and microwave assisted synthesis of some pyrazoline derivatives and their antimicrobial activity," *Journal of Chemical and Pharmaceutical Research*, vol. 2, no. 3, pp. 33–42, 2010.
- [21] M. Srinivas, A. Tejasri, N. Anjaneyulu, and K. Satyanarayana, "Microwave assisted synthesis and antimicrobial activity of some novel quinoxaline compounds," *International Journal of Pharma Sciences*, vol. 3, no. 1, pp. 142–146, 2013.
- [22] K. Suresh, M. Veeresh, A. Prashant et al., "Synthesis and pharmacological evaluation of a novel series of 5-(substituted) aryl-3-(3-coumarinyl)-1-phenyl-2-pyrazolines as novel anti-inflammatory and analgesic agents," *European Journal of Medicinal Chemistry*, vol. 30, pp. 1–7, 2008.

- [23] F. E. Goda, A. R. Maarouf, and E. R. EL-Bendary, "Synthesis and antimicrobial evaluation of new isoxazole and pyrazole derivatives," *Saudi Pharmaceutical Journal*, vol. 11, no. 3, pp. 111–117, 2003.
- [24] C. H. Collins and P. M. Lyne, *Microbiological Methods*, Butterworths, London, UK, 4th edition, 1976.