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Biological evaluation and synthesis of new pyrimidine-2(1H)-ol/-thiol derivatives derived from chalcones using the solid phase microwave method

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Abstract: Twenty-five new hydroxy- and methoxy-substituted 4,6-diarylpyrimidin-2(1H)-ol (**20**–**34**) and 4,6-diarylpyrimidine-2(1H)-thiol derivatives (**35**–**44**) were synthesized from the reaction of the corresponding 1,3-diaryl-2-propene-1-one compounds (**1**–**19**) with urea or thiourea using the solid-phase microwave method. All the new synthetic compounds (**20**–**44**) were evaluated with regard to their α-glucosidase activity. However, only compounds **22**–**25**, **27**, **31**, **34**, **35**, **37**, and **40** exhibited a greater inhibitory effect than standard acarbose. The IC_{50} values of the active compounds ranged between 2.36 and 13.34 μM. The 25 new compounds were also screened for their in vitro pancreatic lipase activity and compounds **20**–**27** and **35**–**39** were found to be active. Of these compounds **26**, **27**, and **39** exhibited the best antilipase activities at concentrations of 0.40 $\pm$ 0.06, 0.26 $\pm$ 0.07, and 0.29 $\pm$ 0.026 μM. All the new compounds (**20**–**44**) were evaluated for their in vitro antimicrobial activity for nine test microorganisms. Compounds **20**–**24** and **35**–**39** were determined to possess a significant broad spectrum against the gram-positive bacteria *Escherichia faecalis*, *Staphylococcus aureus*, and *Bacillus cereus* among the tested bacterial agents. Compounds **20**–**24** and **35**–**39** exhibit the best activity against *Mycobacterium smegmatis*, with minimum inhibitory concentrations of 62.5–500 μg/mL, indicating their potential use as antituberculous agents.

Key words: Pyrimidine-2(1H)-ol/-thiol, chalcone, α-glucosidase, lipase, antimicrobial, enzyme inhibition

1. Introduction

Pyrimidines are the most important pharmacological chemical scaffolds widely present in many natural products and a basic building block of DNA and RNA. Pyrimidine derivatives have been shown to be associated with a variety of chemotherapeutic effects, including antiviral,[1–3] antitumor,[4–6] antimicrobial,[7–9] antitubercular,[10–16] and antifungal[17,18] activities. A number of useful drugs contain pyrimidine derivatives associated with anticancer,[19–22] antimalarial,[23,24] diuretic,[25] antibiotic,[10,26] and antihypertensive[26,27] pharmaceutical activities. Various pyrimidine derivatives have been synthesized from many different reagents.[10,18,20,28–34]

One of the most widely used methods for the synthesis of diaryl-substituted pyrimidine-2(1H)-ol/-thiol derivatives involves starting from chalcone by cyclization with urea and thiourea.[35] Chalcones are natural compounds and important precursors for the synthesis of many biologically active compounds. This class of com-

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pounds is reported to possess diverse chemotherapeutic properties, such as antituberculous,[30,36] antitumor,[37–39] and antifungal[33,40,41] activities. Tangeretin and nobiletin type polymethoxy chalcone derivatives are reported to exhibit highly cytotoxic activity against a variety of human cancer cell lines.[42–44] Many oxygen- and sulfur-containing heterocyclic compounds are known for their therapeutic activities and occupy a unique place in medicinal chemistry. In the light of the wide range of biological activities observed in substituted pyrimidine, the purpose of the present study was to synthesize 25 new hydroxy- and methoxy-substituted 4,6-diarylpyrimidine-2(1H)-ol (**20**–**34**) and 4,6-diarylpyrimidine-2(1H)-thiol (**35**–**44**) compounds from different substituted chalcone with urea or thiourea and to test their antimicrobial and enzyme activities.

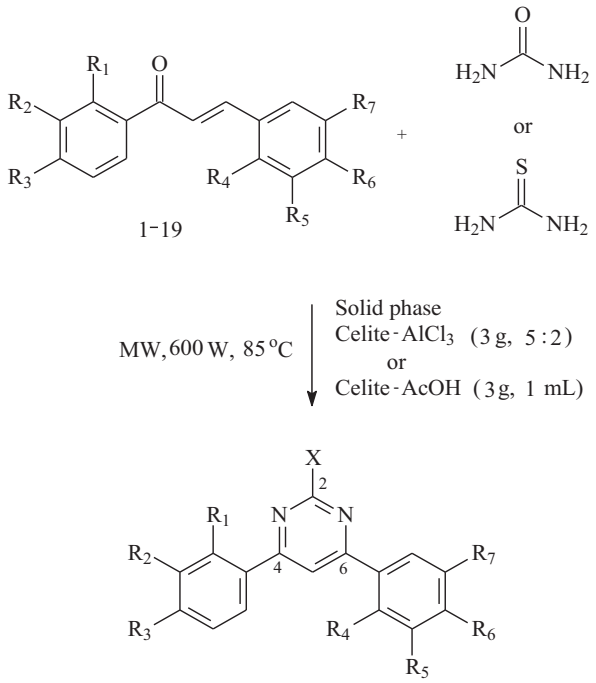
2. Results and discussion

2.1. Synthesis

Many classic synthetic methods of pyrimidine synthesis have been used in the literature,[28–34] although these are problematic in terms of time, yields, and harsh reaction conditions. We synthesized substituted pyrimidine-2(1H)-ol/-thiol derivatives (**20**–**44**) starting from hydroxy- or methoxy-substituted chalcone and urea or thiourea using the solid-phase microwave method, a practical technique for the synthesis of substituted pyrimidine-2(1H)-ol/-thiol derivatives. We also observed 4,6-diarylypyrimidin-2(1H)-one/thione and 4,6-diaryl-3,4-dihydropyrimidin-2(1H)-thione compounds under the same reaction conditions in very low yields. The reaction sequences used for the synthesis of target compounds **20**–**44** are outlined in the Figure. Compound **30** is mentioned in the literature as the tautomer form of methoxy-substituted 4,6-diarylypyrimidin-2(1H)-one,[45] but no spectral data are given. The structures of compounds **20**–**44** were identified by means of ^{1}H NMR, ^{13}C/APT-NMR, ^{1}H-^{1}H COSY, UV-Vis, FT-IR, LC-MS/MS, and LC-MS/TOF spectroscopic data, which were consistent with the proposed structure. The infrared (IR) spectra of these pyrimidine compounds showed the presence of a weak absorption band in the region of ∼3380 and ∼3100 cm^{-1} due to a –OH and hydrogen-bonded –OH group, indicating the transformation of carbonyl functionality into cyclic pyrimidines. The ^{13}C NMR result gave the heterocyclic aromatization ring. The ^{1}H and ^{13}C NMR spectra of compounds **20**–**44**, particularly H5–, showed peaks at δ_H 7.7–8.0 (1H, bs) and C_5 at δ_C 107–117 ppm, an indication of pyrimidine ring systems.[45] The mass spectral data of pyrimidine compounds (**20**–**44)** showed the presence of molecular ion peaks at the appropriate m/z values with reasonable $[M + H]^+$, $[M + Na]^+$, $[M + K]^+$, and $[M + CH_3COOH]^+$ intensities. However, LC-MS/TOF of the methoxy substituted pyrimidine derivatives (**25**–**34**, **40**–**44**) gave the molecular ion peaks as $[M + 4H_2O]^+$, $[M + 5H_2O]^+$, $[M + 3H_2O + Na - 1]^+$, and $[M + 4H_2O + 2]^+$ in a very high percentage ratio. The physical properties, spectral information, and mass analysis of all the synthesized compounds (**20**–**44**) are illustrated in the experimental section.

2.2. α-Glucosidase inhibitory activity of compounds 20–44

All the new synthetic compounds (**20**–**44**) evaluated with regard to their α-glucosidase activity exhibited better and reasonable activities. Compounds **22**–**25**, **26**, **30**, **33**, **34**, **36**, and **39** exhibited a greater inhibitory effect than acarbose, a known α-glucosidase inhibitor used as an antidiabetic drug (Table 1).[46–48] No significant inhibitory effect was detected for compounds **37** or **40**–**44**. The IC_{50} values of acarbose compounds, **22**–**25**, **26**, **30**, **33**, **34**, **36**, and **39**, were calculated as 13.34 ± 1.26, 9.56 ± 1.33, 3.09 ± 0.42, 10.02 ± 2.93, 12.30 ± 0.81, 3.15 ± 0.60, 13.09 ± 2.40, 6.10 ± 1.31, 2.36 ± 1.18, 3.38 ± 1.36, and 4.38 ± 0.36 μM, respectively (Table 1).



20-44

Products	R_1–R_7	–X
20	R_2 = –OH, R_1, R_3, R_4, R_5, R_6, R_7 = –H	–OH
21	R_3 = –OH, R_1, R_2, R_4, R_5, R_6, R_7 = –H	–OH
22	R_2, R_5 = –OH, R_1, R_3, R_4, R_6, R_7 = –H	–OH
23	R_3, R_5 = –OH, R_1, R_2, R_4, R_6, R_7 = –H	–OH
24	R_3 = –OH, R_5 = – OCH_3, R_1, R_2, R_4, R_6, R_7 = –H	–OH
25	R_1 = –OCH_3, R_2, R_3, R_4, R_5, R_6, R_7 = –H	–OH
26	R_1, R_5 = –OCH_3, R_2, R_3, R_4, R_6, R_7 = –H	–OH
27	R_1, R_4 = –OCH_3, R_2, R_3, R_4, R_5, R_7 = –H	–OH
28	R_3, R_4 = –OCH_3, R_1, R_2, R_5, R_6, R_7 = –H	–OH
29	R_3, R_5 = –OCH_3, R_1, R_2, R_4, R_6, R_7 = –H	–OH
30	R_3, R_6 = –OCH_3, R_1, R_2, R_4, R_5, R_7 = –H	–OH
31	R_3, R_4, R_5 = –OCH_3, R_1, R_2, R_6, R_7 = –H	–OH
32	R_3, R_4, R_7 = –OCH_3, R_1, R_2, R_5, R_6 = –H	–OH
33	R_1, R_4, R_5, R_6 = –OCH_3, R_2, R_3, R_7 = –H	–OH
34	R_3, R_4, R_5, R_6 = –OCH_3, R_1, R_2, R_7 = –H	–OH
35	R_2 = –OH, R_1, R_3, R_4, R_5, R_6, R_7 = –H	–SH
36	R_3 = –OH, R_1, R_2, R_4, R_5, R_6, R_7 = –H	–SH
37	R_2, R_5 = –OH, R_1, R_2, R_4, R_6, R_7 = –H	–SH
38	R_3, R_5 = –OH, R_1, R_2, R_4, R_6, R_7 = –H	–SH
39	R_3 = –OH, R_5 = –OCH_3, R_1, R_2, R_4, R_6, R_7 = –H	–SH
40	R_3, R_5 = –OCH_3, R_1, R_2, R_4, R_6, R_7 = –H	–SH
41	R_3, R_6 = –OCH_3, R_1, R_2, R_4, R_5, R_7 = –H	–SH
42	R_3, R_4, R_5 = –OCH_3, R_1, R_2, R_6, R_7 = –H	–SH
43	R_3, R_4, R_7 = –OCH_3, R_1, R_2, R_5, R_6 = –H	–SH
44	R_3, R_4, R_5, R_6 = –OCH_3, R_1, R_2, R_7 = –H	–SH

Figure. Synthesis of hydroxy- and methoxy-substituted 4,6-diarylpyrimidin-2(1H)-ol (**20**–**34**), and 4,6-diarylpyrimidine-2(1H)-thiol derivatives (**35**–**44**).

Table 1. The remaining α-glucosidase activities and IC_{50} values at the concentration of 10 μM among the all synthesized compounds (**20**–**44**).

Comp.	Residue activity %	IC_{50} (μM) ± SD
20	88.59 ± 2.88	36.51 ± 0.94
21	48.84 ± 0.31	15.04 ± 2.18
22	37.41 ± 3.40	9.56 ± 1.33
23	3.30 ± 2.15	3.09 ± 0.42
24	49.27 ± 10.38	10.02 ± 2.93
25	46.65 ± 4.45	12.30 ± 0.81
26	26.31 ± 10.10	3.15 ± 0.60
27	63.74 ± 9.10	14.51 ± 2.81
28	7.86 ± 2.18	47.49 ± 16.44
29	96.35 ± 0.85	36.11 ± 17.48
30	76.01 ± 9.43	13.09 ± 2.40
31	68.21 ± 17.00	17.25 ± 3.52
32	54.72 ± 4.20	25.47 ± 6.00
33	38.66 ± 7.79	6.10 ± 1.31
34	9.46 ± 2.68	2.36 ± 1.18
35	77.88 ± 7.30	34.86 ± 2.68
36	45.33 ± 2.87	3.38 ± 1.36
38	55.34 ± 1.86	15.34 ± 0.92
39	12.89 ± 2.08	4.38 ± 0.36
Acar. **	63.44 ± 9.61	13.34 ± 1.26

Acarbose**: Reference compound

2.3. Pancreatic lipase inhibitory activity of compounds 20–44

All compounds were evaluated with regard to pancreatic lipase activity and **26**, **27**, and **39** exhibited antilipase activities at various concentrations (Table 2). No significant inhibitory effect was detected for the other compounds. Of the tested compounds, **38** exhibited the best antilipase activity. This compound inhibited pancreatic lipase activity by 93.7% at a concentration of 10 μM (Table 2). Orlistat, a known pancreatic lipase inhibitor used as an antiobesity drug, exhibited an inhibitory effect of 99.3% at a concentration of 312 nM. The IC_{50} values of compound **27** and orlistat were determined as 0.26 ± 0.07 and 0.003 ± 0.0005 μM. Orlistat is the only approved antiobesity medication,[49] but side-effects have been reported, such as fecal incontinence, flatulence, and steatorrhea.[50]

2.4. Antimicrobial activity of compounds 20–44

All the synthesized compounds (**20**–**44**) with concentrations ranging from 10.000 to 18.200 μg/mL, were tested against gram-negative, gram-positive, and antifungal bacteria for antimicrobial activity using a known method.[51,52] Ampicillin, streptomycin, and fluconazole were used as standards. The experimental results showed that antimicrobial activity was more effective on the gram-positive bacteria *Enterococcus faecalis*, *Staphylococcus aureus*, and *Bacillus cereus* for compounds **20**, **21**, **22**, **23**, **24**, **35**, **36**, **37**, **38**, and **39**,

Table 2. The remaining lipase activities and IC_{50} values at the concentration of 10 μM among the all synthesized compounds, except for orlistat.

Comp.	Residue activity %	IC_{50} (μM) ± SD
20	9.59 ± 3.00	0.85 ± 0.11
21	26.31 ± 2.60	2.97 ± 0.52
22	18.21 ± 1.97	4.47 ± 0.74
23	30.60 ± 6.00	0.59 ± 0.06
24	7.30 ± 2.30	0.56 ± 0.17
25	3.50 ± 0.40	0.62 ± 0.06
26	5.70 ± 0.60	0.40 ± 0.06
27	7.50 ± 1.50	0.26 ± 0.07
35	8.78 ± 1.62	2.59 ± 0.42
36	58.08 ± 3.44	-*
37	25.91 ± 2.57	6.79 ± 1.08
38	19.69 ± 2.49	1.39 ± 0.05
39	6.30 ± 1.20	0.29 ± 0.02
Orlistat**	0.70 ± 0.02	0.003 ± 0.0005

-*: Not observed, Orlistat**: Reference compound

which were hydroxyl substituted 4,6-diarylypyrimidin-2(1H)-ol/thiol (**20**–**24** and **36**–**40**) compounds. The MIC values of compounds **20**, **21**, **24**, **35**, **36**, **37**, and **39** against *B. cereus* were 3.5, 2.2, 4.3, 2.3, 1.95, 2.1, and 8.6 μg/mL, respectively, better than those of the standard used (ampicillin, 15 μg/mL). Furthermore, compounds **20**, **21**, **22**, **23**, **24**, **35**, **36**, **37**, **38**, and **39** were observed to exhibit antimicrobial activity against tuberculosis bacterium type *M. smegmatis* with the MIC values within a range of 0.10–4.3 μg/mL. However, the MIC value for compound **22** was 18 μg/mL. The antimicrobial active compounds are listed in Table 3.

3. Experimental

3.1. Materials and equipment

All starting chemical reagents (methoxy- and hydroxy-substituted acetophenone, methoxy- and hydroxy-substituted benzaldehyde, urea, thiourea, Celite, $AlCl_3$, sodium hydroxide, and acetic acid used in the synthesis were high grade commercial products purchased from Aldrich, Fluka, or Sigma and were used without further purification. The solvents (n-hexane, chloroform, ethyl acetate, methanol, diethyl ether, and dimethyl sulfoxide) used were either analytical grade or bulk solvents distilled before use.

Thin-layer chromatography (TLC) and column chromatography were performed on Merck precoated 60 Kieselgel F_{254} analytical aluminum acidic plates and silica gel 60 (0.040–0.063 mm), respectively. All reactions were monitored using TLC. A Milestone microwave (MW) oven was used for solid-phase MW reactions. ^{1}H and ^{13}C NMR spectra were recorded on a Varian Mercury 200 MHz and Bruker 400 MHz NMR with tetramethylsilane (TMS) as an internal standard. The mass spectral analyses were carried out on a Micromass Quattro LC-MS/MS, and LC-MS/TOF spectrophotometer. Infrared spectra were obtained using a PerkinElmer 1600 FT-IR, KBr (4000–400 cm^{-1}) spectrometer. Melting points were determined using a Thermo-var apparatus

Table 3. Antimicrobial activity of compounds **20**–**24** and **35**–**39**.

Comp.	Stock solution (μg/mL)	Microorganism and minimal inhibitory concentrations (MIC, μg/mL) of the active compounds.								
		Ec	Yp	Pa	Ef	Sa	Bc	Ms	Ca	Sc
20	18.200	-	-	227	14.2	28.4	3.5	< 3.5	14.2	14.2
21	11.300	-	-	141	< 2.2	< 2.2	< 2.2	< 2.2	8.8	35.3
22	11.800	-	-	73	18	18	18	18	73	147
23	11.100	-	555	-	277.5	138.8	17.3	< 4.3	555	555
24	11.000	-	-	137	137	8.6	4.3	< 4.3	-	-
35	11.700	-	-	146	< 2.3	9.1	< 2.3	< 2.3	73.1	18.3
36	10.000	-	-	31.3	< 1.95	< 1.95	< 1.95	< 1.95	31.3	7.8
37	11.000	-	-	68.8	< 2.1	< 2.1	2.1	0.13	137	137
38	12.200	-	-	9.5	19.1	19.1	19.1	0.15	76.3	76.3
39	11.000	-	-	550	68.7	17.2	8.6	< 4.3	-	-
Amp.		10	18	> 128	10	35	15	-	-	-
Strep.		-	-	-	-	-	-	4	-	-
Fluc.		-	-	-	-	-	-	-	< 8	< 8

Ec*: Escherichia coli* ATCC 25922, Yp: *Yersinia pseudotuberculosis* ATCC 911, Pa: *Pseudomonas aeruginosa* ATCC 43288, Ef: *Enterococcus faecalis* ATCC 29212, Sa: *Staphylococcus aureus* ATCC 25923, Bc: *Bacillus cereus* 702 Roma, Ms: *Mycobacterium smegmatis* ATCC607, Ca: *Candida albicans* ATCC 60193, *Saccharomyces cerevisiae* RSKK 251, Amp.: Ampicillin, Strep.: Streptomycin, Fluc.: Fluconazole, (-): no activity.

fitted with a microscope and are uncorrected. UV-Vis absorbance measurements and spectral analyses were carried out on a Unicam UV2-100 at 25 °C.

3.2. Synthesis

3.2.1. General method for the synthesis of substituted 1,3-diaryl-2-propene-1-one (1–19)

1,3-Diaryl-2-propene-1-one derivatives (**1**–**19**) were prepared according to the procedure described elsewhere in the literature.[53–61] Equimolar quantities of substituted acetophenone (10 mmol) and substituted benzaldehyde (10 mmol) in ethanol (10 mL) were stirred at 0–5 °C for 30 min. Sodium hydroxide (40%, 4 mL) was then added dropwise. Stirring was continued for 12 h at room temperature and the reactions were stopped after TLC monitoring. The reaction mixture was poured into crushed ice and acidified if necessary with 6 N HCl. The precipitated solid mass was filtered, washed with water, and crystallized from ethanol to give white, orange, or yellow products purified by column chromatography if necessary.

3.2.2. General method for the synthesis of substituted 4,6-diaryl-pyrimidine-2-ol or 4,6-diaryl-pyrimidine-2-thiol (20–44)

Methoxy- and hydroxy-substituted 1,3-diaryl-2-propene-1-one (0.004 mol each) and urea or thiourea (0.004 mol) were dissolved in chloroform (15 mL) and uniformly adsorbed on the surface of Celite–$AlCl_3$ (3 g, 5:2 ratio, each) in a Pyrex round- bottomed flask. The solvent was evaporated under vacuum. The adsorbed material

was transferred to a Pyrex tube (2 cm diameter, 30 mL) and inserted inside the Milestone MW oven. The mixtures were heated using a fixed power of 600 W for 10 min at 85 °C. The brownish crude reaction mixture was dissolved in methanol (3 × 15 mL) and then neutralized with 2 N HCl and filtered off. The eluate was evaporated and the residue dissolved in water (50 mL) and then extracted with chloroform (3 × 30 mL) to give a crude mixture. This was purified by column chromatography over silica using hexane (50 mL) and a hexane–diethyl ether solvent mixture with a gradient of 9:1, 4:1, and 3:1 ratio (100 mL each) to elicit the pure corresponding products (**20**–**44)** in the range of 30% to 75% yields, respectively. The assigned structures were confirmed by their spectral properties (^{1}H, ^{13}C/APT, 2D-COSY NMR, FT-IR, LC-TOF, and LC-MS/MS) and by comparison with data in the literature.[28–34]

3.2.3. 4-(3-Hydroxyphenyl)-6-phenylpyrimidin-2-ol (20)

Yield: 54%, mp = 191–194 °C, R_f: 0.74 (hexane–diethyl ether, 3:2); IR (KBr, cm^{-1}): 3338 (–OH), 3056 (=CH), 1546 (C=C, aromatic ring), 1581 (C=N); UV-vis λ nm (log): 256 (1.68), 308 (0.59); ^{1}H NMR (400 MHz, $CDCl_3$–CD_3OD (20:1, δ, ppm): [ar-H: 6.91 (d, 1H, J = 8.0 Hz), 6.99 (m, 1H), 7.33 (d, 2H, J = 8.0 Hz), 7.43–7.50 (m, 1H), 7.71–7.73 (m, 1H)], 7.53 (d, 2H, J = 7.8 Hz), 7.64 (bs, 1H)], 7.84 (bs, 1H, pyrimidine-CH). APT-NMR (100 MHz, $CDCl_3$–CD_3OD (20:1, δ, ppm): 114.16 (pyrimidine-CH), 150.52, 157.21 (pyrimidine-C *2/4/6*), ar-C [116.33 (CH), 117.64 (CH), 118.56 (CH), 129.09 (CH), 129.74 (2CH), 129.08 (2CH), 130.00 (CH), 140.61 (C), 138.62 (C), 157.45 (C)]. $C_{16}H_{12}N_2O_2$ = 264.27, calculated $[M + 2K - 2H]^+$: 340.47; found, LC-MS/MS (m/z) (%) $[M + 2K - 2H]^+$: 340.44 (100).

3.2.4. 4-(4-Hydroxyphenyl)-6-phenylpyrimidin-2-ol: (21)

Yield: 36%, mp = 150–dec °C, R_f: 0.65 (hexane–diethyl ether, 3:2); IR (KBr, cm^{-1}): 3231 (–OH), 3026 (=CH), 1595 (C=C, aromatic ring), 1611 (C=N); UV-vis λ nm (log): 256 (1.68), 308 (0.59); ^{1}H NMR (400 MHz, $CDCl_3$–CD_3OD (20:1, δ, ppm): [ar-H: 6.94 (d, 2H, J = 8.0 Hz), 7.52–7.53 (m, 2H), 7.98 (d, 2H, J = 8.0 Hz), 7.72 (bs, 1H), 7.72 (m, 2H)], 7.80 (bs, 1H, pyrimidine, –CH). APT-NMR (100 MHz, $CDCl_3$-CD_3OD (20:1, δ, ppm): 115.72 (pyrimidine-CH), 152.56, 160.06 (pyrimidine-C *2/4/6*), ar-C [115.72 (2CH), 128.97 (2CH), 128.81 (2CH), 130.42 (2CH), 131.28 (CH), 140.03 (2C), 160.06 (C)]. $C_{16}H_{12}N_2O_2$ = 264.27, calculated $[M + 1]^+$: 265.28; found, LC-MS/MS: (m/z) (%) $[M + 1]^+$: 265.35 (78).

3.2.5. 4,6-Bis(3-hydroxyphenyl)-pyrimidin-2-ol: (22)

Yield: 57%, mp = 151–153 °C, R_f: 0.68 (hexane–diethyl ether, 3:2); IR (KBr, cm^{-1}): 3284 (–OH), 3050 (=CH), 1547 (C=C, aromatic ring), 1584 (C=N); UV-vis λ nm (log): 257 (1.11), 304 (0.48); ^{1}H NMR (400 MHz, $CDCl_3$–CD_3OD (20:1, δ, ppm): [ar-H: 6.91 (d, 2H, J = 8.0 Hz), 6.99 (m, 1H), 7.18–7.21 (m, 1H), 7.33 (d, 2H, J = 8.0 Hz), 7.52–7.53 (m, 1H), 7.44 (bs, 1H)], 7.80 (bs, 1H, pyrimidine, –CH). APT-NMR (100 MHz, $CDCl_3$–CD_3OD (20:1, δ, ppm): 117.59 (pyrimidine-CH), 150.35, 157.09 (pyrimidine-C *2/4/6*), ar-C [113.96 (CH), 114.12 (CH), 116.08 (CH), 116.23 (CH), 118.50 (CH), 118.61 (CH), 129.74 (CH), 130.16 (CH), 140.09 (C), 140.85 (C), 150.35 (C), 157.47 (C)]. $C_{16}H_{12}N_2O_3$ = 280.27, calculated $[M + 1]^+$ 281.27; found, LC/MS-TOF: (m/z) (%) $[M + 1]^+$: 281.13 (100).

3.2.6. 4-(3-Hydroxyphenyl)-6-(4-hydroxyphenyl)pyrimidin-2-ol (23)

Yield: 48%, mp = 133–135 °C, R_f:0.78 (hexane–diethyl ether, 3:2); IR (KBr, cm^{-1}): 3284 (–OH), 3050 (=CH), 1547 (C=C, aromatic ring), 1584 (C=N); UV-vis λ nm (log): 324 (1.09), 330 (0.96); ^{1}H NMR (400 MHz, $CDCl_3$–CD_3OD (20:1, δ, ppm): [ar-H: 6.90 (m, 1H), 6.90–6.95 (m, 2H), 6.94 (d, 2H, J = 8.0 Hz), 7.30 (bs, 1H), 7.98 (d, 2H, J = 8.0 Hz)], 7.72 (bs, 1H, pyrimidine, –CH). APT-NMR (100 MHz, $CDCl_3$–CD_3OD (20:1, δ, ppm): 115.72 (pyrimidine-CH), 152.56, 160.06 (pyrimidine-C *2/4/6*), ar-C [113.89 (CH), 115.39 (CH), 115.66 (CH), 115.79 (CH), 115.86 (CH), 128.30 (2CH), 130.03 (CH), 131.26 (CH), 128.81 (C), 140.03 (2C), 160.06 (C)]. $C_{16}H_{12}N_2O_3$ = 280.27, calculated $[M + 1]^+$ 281.27; found, LC/ MS-TOF: (m/z) (%) $[M + 1]^+$: 281.38 (100).

3.2.7. 4-(4-Hydroxyphenyl)-6-(3-methoxyphenyl)pyrimidin-2-ol (24)

Yield: 51%, mp = 177–179 °C, R_f: 0.71 (hexane–diethyl ether, 3:2); IR (KBr, cm^{-1}): 3296 (–OH), 1545 (C=C, aromatic ring), 1596 (C=N) 1235 (–OCH_3); UV-vis λ nm (log): 266 (1.99), 329 (0.38); ^{1}H NMR (400 MHz, $CDCl_3$–CD_3OD (20:1, δ, ppm): 4.01 (s, 3H, –OCH_3), [ar-H: 6.90–7.00 (m, 1H), 6.93 (d, 2H, J = 9.0 Hz), 7.00–7.42 (m, 2H), 7.22 (s, 1H), 7.98 (d, 2H, J = 9.0 Hz)], 7.71 (bs, 1H, pyrimidine, –CH). APT-NMR (100 MHz, $CDCl_3$–CD_3OD (20:1, δ, ppm): 55.32 (–OCH_3), 112.87 (pyrimidine-CH), 150.00, 160.06 (pyrimidine-C *2/4/6*), ar-C [114.11 (CH), 115.47 (2CH), 119.56 (CH), 128.29 (CH), 128.52 (CH), 130.01 (CH), 131.23 (CH), 140.67 (C), 157.48 (C), 158.01 (C), 160.06 (C)]. $C_{17}H_{14}N_2O_3$ = 294.30, calculated $[M + 2Na]^+$: 340.28; found, LC-MS/MS: (m/z) (%) $[M + 2Na]^+$: 340.30(100).

3.2.8. 4-(2-Methoxyphenyl)-6-phenylpyrimidin-2-ol (25)

Yield: 36%, mp = 156–159 °C, R_f: 0.69 (hexane-diethyl ether, 3:1) ; IR (KBr, cm^{-1}): 3100 (w, hydrogen-bonded –OH), 3033 (=CH), 1670 (C=N), 1580 (C=C, aromatic ring), 1239 (–OCH_3); UV-vis λ nm (log): 250 (1.58, 2.02), 305 (0.49, 1.52); ^{1}H NMR (200 MHz, $CDCl_3$ (δ, ppm): 3.92 (s, 3H, –OCH_3), [ar-H: 7.03–7.13 (m, 1H), 7.14–7.17 (m, 1H), 7.03–7.23 (m, 2H)], 7.37–7.41 (m, 1H), 7.30–7.55 (m, 2H), 7.70–7.78 (m, 1H), 7.94 (m, 1H)], 8.20 (bs, 1H, pyrimidine, –CH). APT-NMR (50 MHz, $CDCl_3$ (δ, ppm): 56.67 (–OCH_3), 111.30 (pyrimidine, –CH), 147.50, 156.97 (pyrimidine-C *2/4/6*), ar-C [121.31 (CH), 128.53 (CH), 121.42 (CH), 127.32 (2CH), 128.90 (2CH), 129.75 (CH), 129.42 (C), 131.52 (CH), 139.37 (C), 155.89 (C)]. $C_{17}H_{14}N_2O_2$ = 278.30, calculated $[M + 5H_2O]^+$: 368.38; found, LC/MS-TOF: (m/z) (%) $[M + 5H_2O]^+$: 368.25 (100).

3.2.9. 4-(2-Methoxyphenyl)-6-(3-methoxyphenyl)pyrimidin-2-ol (26)

Yield: 42%, mp = 127–129 °C, R_f : 0.67 (hexane–diethyl ether, 3:1); IR (KBr, cm^{-1}): 3100 (w, hydrogen-bonded –OH), 3063 (=CH), 1598 (C=N), 1580 (C=C, aromatic ring); UV-vis λ nm (log): 251 (1.39, 1.96), 294 (0.68, 1.66); ^{1}H NMR (200 MHz, $CDCl_3$ (δ, ppm): 3.84 (s, 6H, –OCH_3), [ar-H: 6.97–7.05 (m, 1H), 7.05–7.15 (m, 1H), 7.18–7.20 (m, 1H), 7.28–7.37 (m, 1H), 7.30 (s, 1H), 7.39–7.87 (m, 1H), 7.44–7.51 (m, 1H), 7.84 (d, 1H, J = 6.0 Hz)], 8.06 (bs, 1H, pyrimidine, –CH). APT-NMR (50 MHz, $CDCl_3$ (δ, ppm): 55.78, 55.14 (2 × –OCH_3), 115.27 (pyrimidine, –CH), 152.33, 160.10 (pyrimidine-C *2/4/6*), ar-C [116.77 (CH), 118.01 (CH), 123.41 (CH), 125.51 (CH), 124.65 (CH), 133.96 (CH), 132.64 (C), 134.05 (CH), 135.00 (CH), 143.98 (C), 160.05 (C), 164.10 (C)]. $C_{18}H_{16}N_2O_3$ = 308.33, calculated $[M + 5H_2O]^+$, 398.40; found, LC/ MS-TOF: (m/z) (%) $[M + 5H_2O]^+$: 398.31 (100).

3.2.10. 4-(2-Methoxyphenyl)-6-(2-methoxyphenyl)pyrimidin-2-ol (27)

Yield: 36%, mp =181–184 °C, R_f: 0.68 (hexane–diethyl ether, 3:1); IR (KBr, cm^{-1}): 3100 (w, hydrogen-bonded –OH), 3074 (=CH), 1580 (C=C, aromatic ring), 1600 (C=N), 1237 (–OCH_3); UV-vis λ nm (log): 259 (2.15); ^{1}H NMR (200 MHz, $CDCl_3$ (δ, ppm): 3.86, 3.93 (s, 6H, –OCH_3), [ar-H: 7.00–7.30 (m, 1H), 7.05–7.45 (m, 2H), 7.18–7.20 (m, 1H), 7.44–7.51 (m, 1H), 7.32–7.42 (m, 1H), 7.74 (d, 1H, J = 6.0 Hz), 7.90 (d, 2H, J = 8.6 Hz))], 8.03 (bs, 1H, pyrimidine, –CH). APT-NMR (50 MHz, $CDCl_3$ (δ, ppm): 55.41, 55.79 (2x -OCH_3), 111.46 (pyrimidine-C_5), 155.76, 160.27 (pyrimidine-C *2/4/6*), ar-C [114.37 (3CH), 121.00 (CH), 128.10 (CH), 129.35 (2CH), 129.35 (C), 131.60 (CH), 147.54 (C), 157.11 (2C)]. $C_{18}H_{16}N_2O_3$ = 308.33, calculated [M + $5H_2O$]$^+$, 398.40; found, LC/MS-TOF: (m/z) (%) [M + $5H_2O$]$^+$: 398.22 (100).

3.2.11. 4-(2-Methoxyphenyl)-6-(4-methoxyphenyl)pyrimidin-2-ol (28)

Yield: 48%, mp =137–139 °C, R_f: 0.72 (hexane–diethyl ether, 3:1); IR (KBr, cm^{-1}): 3100 (w, hydrogen-bonded –OH), 3062 (=CH), 1583 (C=C, aromatic ring), 1605 (C=N), 1241 (–OCH_3); UV-vis λ nm (log): 265 (1.64, 2.04); ^{1}H NMR (200 MHz, $CDCl_3$ (δ, ppm): 3.87 (s, 6H, –OCH_3), [ar-H: 7.00–7.60 (m, 2H), 7.14 (d, 2H, J = 8.6 Hz), 7.44–7.51 (m, 1H), 8.20 (m, 1H), 8.22 (d, 2H, J = 8.6 Hz)], 7.80 (bs, 1H, pyrimidine, –CH). APT-NMR ^{1}H NMR (50 MHz, $CDCl_3$ (δ, ppm): 55.94, 55.63 (2 × –OCH_3), 111.69 (pyrimidine-CH), 156.49, 160.60 (pyrimidine-C *2/4/6*), ar-C [114.24 (3CH), 118.62 (CH), 128.67 (CH), 128.96 (2CH), 130.78 (CH), 132.82 (C), 148.02 (C), 156.49 (C), 156.87 (C)]. $C_{18}H_{16}N_2O_3$ = 308.33, calculated [M + $5H_2O$]$^+$, 398.40; found, LC/MS-TOF: (m/z) (%) [M + $5H_2O$]$^+$: 398.22 (100).

3.2.12. 4-(3-Methoxyphenyl)-6-(4-methoxyphenyl)pyrimidin-2-ol (29)

Yield: 30%, mp = 153–155 °C, R_f: 0.78 (hexane–diethyl ether, 3:1); IR (KBr, cm^{-1}): 3100 (w, hydrogen-bonded –OH), 3063 (=CH), 1539 (C=C, aromatic ring), 1597 (C=N), 1239 (–OCH_3); UV-vis λ nm (log): 266 (1.27, 1.92); ^{1}H NMR (100 MHz, $CDCl_3$ (δ, ppm): 3.87, 3.88 (s, 6H, –OCH_3), [ar-H: 7.02 (d, 2H, J = 7.0 Hz), 7.24–7.40 (m, 3H), 8.14 (m, 1H), 8.15 (d, 2H, J = 7.0 Hz)], 7.78 (bs, 1H, pyrimidine, –CH). APT-NMR (50 MHz, $CDCl_3$ (δ, ppm): 55.29, 55.29 (2 × –OCH_3), 112.87 (pyrimidine-CH), 156.82, 159.99 (pyrimidine-C *2/4/6*), ar-C [113.93 (2CH), 115.65 (CH), 119.49 (CH), 119.50 (CH), 128.28 (2CH), 130.03 (CH), 132.16 (C), 140.71 (C), 159.97 (C), 160.38 (C)]. $C_{18}H_{16}N_2O_3$ = 308.33, calculated [M + $5H_2O$]$^+$, 398.40; found, LC/MS-TOF: (m/z) (%) [M + $5H_2O$]$^+$: 398.17 (100).

3.2.13. 4,6-Bis(4-methoxyphenyl)pyrimidin-2-ol (30)

Yield: 30%, mp = oily, R_f : 0.66 (hexane–diethyl ether, 3:1); IR (KBr, cm^{-1}): 3100 (w, hydrogen-bonded –OH), 3097 (=CH), 1573 (C=C, aromatic ring), 1595 (C=N), 1245 (–OCH_3); UV-vis λ nm (log): 264 (1.32, 1.87); ^{1}H NMR ^{1}H NMR (200 MHz, $CDCl_3$ (δ, ppm): 3.85 (s, 6H, –OCH_3), [ar-H: 6.99 (d, 2H, J = 8.4 Hz), 6.99 (d, 2H, J = 8.4 Hz), 8.12 (d, 2H, J = 8.4 Hz), 8.14 (d, 2H, J = 8.4 Hz)], 7.71 (bs, 1H, pyrimidine, –CH). APT-NMR ^{1}H NMR (50 MHz, $CDCl_3$ (δ, ppm): 55.26 (2 × –OCH_3), 113.68 (pyrimidine-CH), 156.77, 160.30 (pyrimidine-C *2/4/6*), ar-C [113.88 (4CH), 128.27 (4CH), 149.30 (2C), 160.30 (2C)]. $C_{18}H_{16}N_2O_3$ = 308.33, calculated [M + $5H_2O$]$^+$, 398.40; found, LC/ MS-TOF: (m/z) (%) [M + $5H_2O$]$^+$: 398.22 (100).

3.2.14. 4-(2,3-Dimethoxyphenyl)-6-(4-methoxylphenyl)pyrimidin-2-ol (31)

Yield: 36%, mp = oily, R_f: 0.81 (hexane–diethyl ether, 3:1); IR (KBr, cm^{-1}): 3100 (w, hydrogen-bonded –OH), 3065 (=CH), 1539 (C=C, aromatic ring), 1597 (C=N), 1239 (–OCH_3); UV-vis λ nm (log): 312 (1.36, 1.96); ^{1}H NMR (200 MHz, $CDCl_3$ (δ, ppm): 3.70, 3.90 (s, 9H, –OCH_3), [ar-H: 7.03–7.18 (m, 2H), 7.04 (d, 1H, J = 8.6 Hz), 7.20 (d, 2H, J = 7.6 Hz), 8.19 (d, 2H, J = 8.6 Hz)], 7.81 (bs, 1H, pyrimidine, –CH). APT-NMR (100 MHz, $CDCl_3$) δ, ppm: 55.38, 55.38, 56.00 (3 × –OCH_3), 113.68 (pyrimidine-CH), 156.27, 160.43 (pyrimidine-C *2/4/6*), ar-C [114.2 (2CH), 117.91 (CH), 124.47 (CH), 128.34 (2CH), 134.06 (C), 146.77 (C), 147.36 (C), 153.28 (C), 156.27 (C)]. $C_{19}H_{18}N_2O_4$ = 338.35, calculated $[M - H_2O + 1]^+$, 321.35; $[M + 5H_2O]^+$, 428.42; found, LC/ MS-TOF: (m/z) (%) $[M - H_2O + 1]^+$: 321.18 (100), $[M + 5H_2O]^+$: 428.18 (40).

3.2.15. 4-(2,5-Dimethoxyphenyl)-6-(4-methoxyphenyl)pyrimidin-2-ol (32)

Yield: 39%, mp = 186–188 °C, R_f : 0.65 (hexane–diethyl ether, 3:1); IR (KBr, cm^{-1}): 3100 (w, hydrogen-bonded –OH), 3072 (=CH), 1541 (C=C, aromatic ring), 1599 (C=N), 1243 (–OCH_3); UV-vis λ nm (log): 318 (0.61, 1.61); ^{1}H NMR (200 MHz, $CDCl_3$ (δ, ppm): 3.83, 3.85, 3.89 (s, 9H, –OCH_3), [ar-H: 6.90–7.10 (m, 3H), 7.05 (d, 2H, J = 8.4 Hz), 6.94–6.96 (m, 1H) , 8.16 (d, 2H, J = 8.4 Hz)], 7.78 (bs, 1H, pyrimidine, –CH). APT-NMR (50 MHz, $CDCl_3$ (δ, ppm): 55.32, 55.81, 56.32 (3 × –OCH_3), 113.68 (pyrimidine-CH), 156.18, 160.28 (pyrimidine-C *2/4/6*), ar-C [113.90 (2CH), 114.25 (CH), 116.24 (CH), 118.21 (CH), 128.35 (2CH), 129.45 (C), 147.52 (C), 150.75 (C), 153.75 (C), 156.18 (C)]. $C_{19}H_{18}N_2O_4$ = 338.35, calculated $[M + 5H_2O]^+$, 428.42; found, LC/MS-TOF: (m/z) (%) $[M + 5H_2O]^+$: 428.24 (100).

3.2.16. 4-(2-Methoxyphenyl)-6-(2,3,4-trimethoxyphenyl)pyrimidin-2-ol (33)

Yield: 39%, mp = 152–154 °C, R_f: 0.70 (hexane–diethyl ether, 3:1) ; IR (KBr, cm^{-1}): 3100 (w, hydrogen-bonded –OH), 3070 (=CH), 1580 (C=C, aromatic ring), 1600 (C=N), 1237 (–OCH_3); UV-vis λ nm (log): 256 (0.87, 1.76); ^{1}H NMR (200 MHz, $CDCl_3$ (δ, ppm): 3.85, 3.96 (s, 12H, –OCH_3), [ar-H: 6.80 (d, 1H, J = 6.0 Hz), 7.00–7.30 (m, 2H), 7.44–7.51 (m, 1H), 7.80 (m, 1H), 8.00–8.80 (m, 1H)], 7.92 (bs, 1H, pyrimidine, –CH). APT-NMR (50 MHz, $CDCl_3$ (δ, ppm): 55.97, 55.63, 60.98, 61.25 (4 × –OCH_3), 111.29 (pyrimidine-CH), 153.90, 157.00 (pyrimidine-C *2/4/6*), ar-C [107.48 (CH), 120.94 (CH), 124.92 (CH), 123.36 (CH), 129.57 (CH), 126.53 (C), 131.54 (CH), 142.42 (C), 145.04 (C), 151.82 (C), 155.10 (C), 157.00 (C)]. $C_{20}H_{20}N_2O_5$ = 368.38, calculated $[M + 5H_2O]^+$, 458.45; found, LC/MS-TOF: (m/z) (%) $[M + 5H_2O]^+$: 458.21 (100).

3.2.17. 4-(4-Methoxyphenyl)-6-(2,3,4-trimethoxyphenyl)pyrimidin-2-ol (34)

Yield: 36%, mp = 122–125 °C, R_f: 0.82 (hexane–diethyl ether, 3:1); IR (KBr, cm^{-1}): 3100 (w, hydrogen-bonded –OH), 3067 (=CH), 1539 (C=C, aromatic ring), 1597 (C=N), 1239 (–OCH_3); UV-vis λ nm (log): 277 (1.67, 2.05); ^{1}H NMR (200 MHz, $CDCl_3$ (δ, ppm): 3.75, 3.82, 3.87, 3.95 (s, 12H, –OCH_3), [ar-H: 6.74 (d, 1H, J = 8.8 Hz), 7.00 (d, 2H, J = 9 Hz), 7.15 (d, 1H, J = 8.8 Hz), 8.14 (d, 2H, J = 9.0 Hz)], 7.75 (bs, 1H, pyrimidine, –CH). APT-NMR (50 MHz, $CDCl_3$ (δ, ppm): 55.13, 55.85, 60.93, 61.13 (4 × –OCH_3), 117.51 (pyrimidine-CH), 156.09, 160.18 (pyrimidine-C 2/4/6), ar-C [107.48 (CH), 113.80 (2CH), 124.43 (CH), 126.31 (C), 128.13 (2CH), 147.09 (C), 156.10 (C), 160.17 (C), 142.42 (C), 153.96 (C)]. $C_{20}H_{20}N_2O_5$ = 368.38, calculated $[M + 5H_2O]^+$, 458.45; found, LC/ MS-TOF: (m/z) (%) (%) $[M + 5H_2O]^+$: 458.21 (100).

3.2.18. 3-(2-Mercapto-6-phenylpyrimidin-4-yl)phenol (35)

Yield: 69%, mp = 241–243 °C, R_f: 0.79 (hexane–diethyl ether, 3:2); IR (KBr, cm^{-1}): 3321 (–OH), 3070 (=CH), 2600 (w, –SH), 1549 (C=C, aromatic ring), 1583 (C=N), 757 (–SH); UV-vis λ nm (log): 266 (0.63), 313 (0.20); ^{1}H NMR (400 MHz, $CDCl_3$–CD_3OD (20:1, δ, ppm): [ar-H: 6.9 (d,d, 1H, J = 2.4, 8.0 Hz), 6.97 (d, 2H, J = 8.0 Hz), 7.45 (m, 1H), 7.45–7.50 (m, 5H), 7.68 (bs, 1H), 7.73 (d, 2H, J = 8.0 Hz)], 7.85 (bs, 1H, pyrimidine, –CH). APT-NMR (100 MHz, $CDCl_3$–CD_3OD (20:1, δ, ppm): 114.15 (pyrimidine-CH), 157.23, 157.47 (pyrimidine-C *2/4/6*), ar-C [116.26 (CH), 117.60 (CH), 118.57 (2CH), 129.07 (CH), 129.09 (CH), 129.71 (3CH), 138.61 (C), 140.03 (C), 150.54 (C)]. $C_{16}H_{12}N_2OS$ = 280.34, calculated $[M + AcOH + 1]^+$: 341.40; found, LC-MS/MS: (m/z) (%) $[M + AcOH + 1]^+$: 341.51 (50).

3.2.19. 4-(2-Mercapto-6-phenylpyrimidin-4-yl)phenol (36)

Yield: 75%, mp = 130–dec °C, R_f: 0.65 (hexane–diethyl ether, 3:2); IR (KBr, cm^{-1}): 3321 (–OH), 3039 (=CH), 2590 (w, –SH), 1543 (C=C, aromatic ring), 1595 (C=N), 829 (–SH); UV-vis λ nm (log): 265 (1.43), 325 (0.42); ^{1}H NMR (400 MHz, $CDCl_3$–CD_3OD (20:1, δ, ppm): [ar-H: 6.93 (d, 2H, J = 8.0 Hz), 7.30–7.70 (m, 5H), 7.97 (d, 2H, J = 8.0 Hz)], 7.72 (bs, 1H, pyrimidine, –CH). APT (100 MHz, $CDCl_3$–CD_3OD (20:1, δ, ppm): 115.83 (pyrimidine-CH), 157.41, 158.07 (pyrimidine-C *2/4/6*), ar-C [115.52 (2CH), 128.56 (4CH), 128.89 (CH), 131.04 (2CH), 132.22 (C), 139.00 (C), 150.30 (C)]. $C_{16}H_{12}N_2OS$ = 280.34, calculated $[M + AcOH]^+$: 340.40, $[M + AcOH + 1]^+$: 341.38; found, LC- MS/MS: (m/z) (%) $[M + AcOH]^+$: 340.38(100), $[M + AcOH + 1]^+$: 341.38 (30).

3.2.20. 3,3'-(2-Mercaptopyrimidin-4,6-diyl)diphenol (37)

Yield: 72%, mp = 219–222 °C, R_f: 0.69 (hexane–diethyl ether, 3:2); IR (KBr, cm^{-1}): 3311 (–OH), 2590 (w, –SH), 1546 (C=C, aromatic ring), 1584 (C=N), 772 (–SH); UV-vis λ nm (log): 257 (0.85), 305 (0.35); ^{1}H NMR (400 MHz, $CDCl_3$–CD_3OD (20:1, δ, ppm); [ar-H: 6.9 (m, 1H), 7.15–7.40 (m, 3H), 7.20 (bs, 1H), 7.39 (m, 1H), 7.45 (d, 2H, J = 8.0 Hz), 7.65 (bs, 1H)], 7.80 (bs, 1H, pyrimidine, –CH). APT-NMR (100 MHz, $CDCl_3$–CD_3OD (20:1, δ, ppm); 116.13 (pyrimidine-CH), 157.15, 157.46 (pyrimidine-C *2/4/6*), ar-C [114.12 (CH), 113.97 (CH), 116.29 (CH), 118.49 (CH), 129.74 (3CH), 130.16 (CH), 140.03 (C), 140.66 (C), 150.50 (C), 157.36 (C)]. $C_{16}H_{12}N_2O_2S$ = 296.34, calculated $[M + AcOH]^+$: 356.40; found, LC-MS/MS: (m/z) (%) $[M + AcOH]^+$: 356.40 (80).

3.2.21. 3-[6-(4-Hydroxyphenyl)-2-mercaptopyrimidin-4-yl]phenol (38)

Yield: 39%, mp = oily, R_f: 0.81 (hexane–diethyl ether, 3:2); IR (KBr, cm^{-1}): 3193 (–OH), 2600 (w, –SH), 1547 (C=C, aromatic ring), 1590 (C=N), 830 (–SH); UV-vis λ nm (log): 294 (0.78), 353 (0.66); ^{1}H NMR (400 MHz, $CDCl_3$–CD_3OD (20:1, δ, ppm): [ar-H: 6.9 (m, 1H), 7.15–7.40 (m, 3H), 7.20 (bs, 1H), 7.39 (m, 1H), 7.45 (d, 2H, J = 8.0 Hz), 7.65 (bs, 1H)], 7.80 (bs, 1H, pyrimidine, –CH). APT-NMR (100 MHz, $CDCl_3$–CD_3OD (20:1, δ, ppm): 116.83 (pyrimidine-CH), 160.12, 161.14 (pyrimidine-C *2/4/6*), ar-C [120.13 (2CH), 121.12 (CH), 121.64 (CH), 121.98 (CH), 131.12 (2CH), 131.93 (CH), 132.34 (C), 144.00 (C), 152.34 (C), 161.14 (C)]. $C_{16}H_{12}N_2O_2S$ = 296.34, calculated $[M + SH - H]^+$: 328.31; $[M + 3H_2O]^+$: 350.32; found, LC-MS/MS: (m/z) (%) $[M + SH - H]^+$: 328.60(45), $[M + 3H_2O]^+$: 350.45 (80).

3.2.22. 4-[2-Mercapto-6-(3-methoxyphenyl)pyrimidin-4-yl]phenol (39)

Yield: 69%, mp = oily, R_f: 0.73 (hexane–diethyl ether, 3:2); IR (KBr, cm^{-1}): 3193 (–OH), 2600 (w, –SH), 1516 (C=C, aromatic ring), 1597 (C=N), 830 (–SH); UV-vis λ nm (log): 268 (0.80), 330 (0.19); ^{1}H NMR (400 MHz, $CDCl_3$–CD_3OD (20:1, δ, ppm): (δ, ppm), 3.86 (s, 6H, –OCH_3), [ar-H: 6.92 (d, 2H, J = 8.0 Hz), 6.97 (d, 1H, J = 8.0 Hz), 7.20 (bs, 1H), 7.28 (d, 1H, J = 8.0 Hz), 7.96 (d, 2H, J = 8.0 Hz), 7.44 (t, 1H, J = 8.0 Hz)], 7.68 (bs, 1H, pyrimidine, –CH). APT-NMR (100 MHz, $CDCl_3$–CD_3OD (20:1, δ, ppm): 55.33 (–OCH_3), 112.86 (pyrimidine-CH), 157.45, 160.04 (pyrimidine-C *2/4/6*), ar-C [115.48 (2CH), 119.56 (CH), 119.56 (CH), 124.06 (CH), 130.06 (2CH), 131.19 (C), 132.08 (C), 140.66 (C), 150.00 (C), 158.00 (C)]. $C_{17}H_{14}N_2O_2S$ = 310.37, calculated $[M]^+$: 310.37; found, LC/ MS-TOF: (m/z) (%) $[M]^+$: 310.36 (70).

3.2.23. 4-(3-Methoxyphenyl)-6-(4-methoxyphenyl)pyrimidine-2-thiol (40)

Yield: 33%, mp = 141–143 °C, R_f: 0.77 (hexane–diethyl ether, 3:1); IR (KBr, cm^{-1}): 3067 (=CH), 2651(w, –SH), 1578 (C=C, aromatic ring), 1597 (C=N), 1244 (–OCH_3), 830 (–SH); UV-vis λ nm (log): 283 (1.35); ^{1}H NMR (200 MHz, $CDCl_3$ (δ, ppm): 3.85 (s, 6H, –OCH_3), [ar-H: 6.98–7.40 (m, 4H), 6.96 (d, 2H, J = 7.8 Hz), 6.94–6.96 (m, 1H), 8.13 (d, 2H, J = 7.8 Hz)], 7.75 (bs, 1H, pyrimidine, –CH). APT-NMR (50 MHz, $CDCl_3$ (δ, ppm): 55.26 (2 × –OCH_3), 112.87 (pyrimidine-CH), 156.77, 160.38 (pyrimidine-C *2/4/6*), ar-C [115.65 (CH), 113.91 (2CH), 119.49 (CH), 128.30 (2CH), 130.03 (CH), 132.08 (C), 140.66 (C), 149.79 (C), 159.98 (C)]. $C_{18}H_{16}N_2O_2S$ = 324.39, calculated $[M + 1]^+$, 325.39; found, LC-MS/MS: (m/z) (%) $[M + 1]^+$: 325.47 (60).

3.2.24. 4,6-Bis(4-methoxyphenyl)pyrimidine-2-thiol (41)

Yield: 36%, mp = oily, R_f: 0.74 (hexane–diethyl ether, 3:1); IR (KBr, cm^{-1}): 3057 (=CH), 2600 (w, –SH), 1540 (C=C, aromatic ring), 1597 (C=N), 1240 (–OCH_3), 832 (–SH); UV-vis λ nm (log): 276 (1.23), 293 (0.76); ^{1}H NMR (200 MHz, $CDCl_3$ (δ, ppm): 3.82 (s, 6H, –OCH_3), [ar-H: 6.94 (d, 2H, J = 7.6 Hz), 6.94 (d, 2H, J = 7.6 Hz), 8.03 (d, 2H, J = 7.6 Hz), 8.03 (d, 2H, J = 7.6 Hz)], 7.77 (bs, 1H, pyrimidine, –CH). APT-NMR (50 MHz, $CDCl_3$ (δ, ppm): 55.69 (2 × –OCH_3), 113.70 (pyrimidine-CH), 156.97, 160.76 (pyrimidine-C *2/4/6*), ar-C [113.3 (4CH), 128.71 (4CH), 132.27 (2C), 160.76 (2C)]. $C_{18}H_{16}N_2O_2S$ = 324.39, calculated $[M + 4H_2O + 2]^+$: 398.45; found, LC/ MS-TOF: (m/z) (%) $[M + 4H_2O + 2]^+$: 398.17 (100) or $[M$ + thiourea + $2]^+$: 398.17 (100).

3.2.25. 4-(2,3-Dimethoxyphenyl)-6-(4-methoxyphenyl)pyrimidine-2-thiol (42)

Yield: 39%, mp = 113–115 °C, R_f: 0.88 (hexane–diethyl ether, 3:1); IR (KBr, cm^{-1}): 3067 (=CH), 2544 (w, –SH), 1542 (C=C, aromatic ring), 1599 (C=N), 1242 (–OCH_3), 830 (–SH); UV-vis λ nm (log): 267 (0.76), 287 (1.32); ^{1}H NMR (200 MHz, $CDCl_3$ (δ, ppm): 3.70, 3.90 (s, 9H, –OCH_3), [ar-H: 7.03–7.30 (m, 3H), 7.06 (d, 2H, J = 7.6 Hz), 8.23 (d, 2H, J = 7.6 Hz)], 7.78 (bs, 1H, pyrimidine, –CH). APT-NMR (50 MHz, $CDCl_3$ (δ, ppm): 55.39, 56.00, 61.02 (3 × –OCH_3), 114.02 (pyrimidine-CH), 156.23, 160.45 (pyrimidine-C *2/4/6*), ar-C [114.21 (2CH), 117.89 (CH), 121.99 (CH), 124.50 (C), 128.39 (CH), 128.84 (CH), 134.00 (C), 134.40 (CH), 146.45 (C), 156.23 (C), 160.45 (C)]. $C_{19}H_{18}N_2O_3S$ = 354.42, calculated $[M + 4H_2O]^+$: 426.17; found, LC/MS-TOF: (m/z) (%) $[M + 4H_2O]^+$: 426.27 (100) or $[M$ + thiourea$]^+$: 426.26 (100).

3.2.26. 4-(2,5-Dimethoxyphenyl)-6-(4-methoxyphenyl)pyrimidine-2-thiol (43)

Yield: 42%, mp = oily, R_f: 0.73 (hexane–diethyl ether, 3:1); IR (KBr, cm^{-1}): 3072 (=CH), 2540 (w, –SH), 1541 (C=C, aromatic ring), 1599 (C=N), 1243 (–OCH_3), 830 (–SH); UV-vis λ nm (log): 289 (1.45); ^{1}H NMR (200 MHz, $CDCl_3$ (δ, ppm): 3.82, 3.85, 3.90 (s, 9H, –OCH_3), [ar-H: 6.80–7.10 (m, 3H), 7.02 (d, 2H, J = 8.0 Hz), 8.18 (d, 2H, J = 8.0 Hz)], 7.76 (bs, 1H, pyrimidine, –CH). APT-NMR (50 MHz, $CDCl_3$ (δ, ppm): 55.38, 55.90, 56.43 (3 × –OCH_3), 112.90 (pyrimidine-CH), 156.31, 160.35 (pyrimidine-C *2/4/6*), ar-C [113.98 (2CH), 114.35 (CH), 116.32 (CH), 118.21 (CH), 128.38 (2CH), 129.65 (C), 132.58 (C), 147.52 (C), 150.87 (C), 153.88 (C)]. $C_{19}H_{18}N_2O_3S$ = 354.42, calculated $[M + 4H_2O]^+$: 426.17; found, LC/MS-TOF: (m/z) (%) $[M + 4H_2O]^+$: 426.17 (100) or $[M$ + thiourea$]^+$: 426.16 (100).

3.2.27. 4-(4-Methoxyphenyl)-6-(2,3,4-trimethoxyphenyl)pyrimidine-2-thiol (44)

Yield: 42%, mp = oily, R_f: 0.80 (hexane–diethyl ether, 3:1); IR (KBr, cm^{-1}): 3074 (=CH), 2552 (w, –SH),1543 (C=C, aromatic ring), 1600 (C=N), 1243 (–OCH_3), 832 (–SH); UV-vis λ nm (log): 256 (0.87), 305 (0.88); ^{1}H NMR (200 MHz, $CDCl_3$ (δ, ppm): 3.80, 3.90, 3.95, 4.00 (s, 12H, –OCH_3), [ar-H: 6.81–7.20 (m, 2H), 7.08 (d, 2H, J = 7.8 Hz), 8.28 (d, 2H, J = 7.8 Hz)], 7.80 (bs, H, pyrimidine, –CH). APT-NMR (50 MHz, $CDCl_3$ (δ, ppm): 55.38, 55.47, 61.15, 61.33 (4 × –OCH_3), 107.21 (pyrimidine-CH), 156.32, 160.40 (pyrimidine-C *2/4/6*), ar-C [114.01 (2CH), 117.78 (CH), 126.60 (C), 128.36 (3CH), 132.47 (C), 142.70 (C), 147.32 (C), 151.72 (C), 154.19 (C). $C_{20}H_{20}N_2O_4S$ = 384.44, calculated $[M + 4H_2O + 2]^+$, 458.51; found, LC/MS-TOF: (m/z) (%) $[M + 4H_2O + 2]^+$: 458.20(100) or $[M$ + thiourea + 2$]^+$: 458.19 (100).

3.3. α-Glucosidase inhibition assay of compounds 20–44

The α-glucosidase inhibition assay was performed spectrophotometrically. α-Glucosidase from *Saccharomyces cerevisiae* (Sigma-Aldrich) was dissolved in phosphate buffer (pH 6.8, 50 mM). The test compounds were dissolved in DMSO. Next 20 μL of the test sample, 20 μL of the enzyme (20 mU/mL), and 135 μL of buffer were added to 96-well microtiter plates and incubated for 15 min at 37 °C. After incubation, 25 μL of p-nitrophenyl-α-D-glucopyranoside (2 mM, Sigma-Aldrich) was added, and the change in absorbance was monitored for 30 min at 400 nm. The test compound was replaced by DMSO (7.5% final) as a control and acarbose (Sigma-Aldrich) was used as a standard inhibitor.[46–48]

3.4. Antilipase activity assay of compounds 20–44

The inhibitory effects of the compounds were evaluated against porcine pancreatic lipase (PPL) (Applichem, Germany) (15 mg/mL). Lipase activity assays were performed as described in the literature.[62] Activity was measured using 4-methylumbelliferyl oleate (4-MU oleate) as a substrate. Briefly, compounds were mixed with PPL 1:3 (v/v) and incubated for 30 min. Microtiter plates containing 50 μL of 0.1 mM 4-MU oleate, 25 μL of diluted compound–lipase solution, 25 μL of dH_2O and assay buffer (13 mM Tris-HCl, 150 mM NaCl, and 1.3 mM $CaCl_2$, pH 8.0) were incubated at 37 °C for 20 min. After incubation, 0.1 mL of 0.1 M citrate buffer was added to stop the reaction. The amount of 4-methylumbelliferone released by the lipase was measured using a spectrofluorometer (SpectraMax M5, Molecular Devices) at an excitation wavelength of 355 nm and an emission wavelength of 460 nm. The inhibitory activity levels of the compounds and orlistat (Xenical, Hoffman, La Roche, Segrate, Italy), an inhibitor control of pancreatic lipase, were measured at various concentrations.

Residual activities were calculated in comparison to a control without inhibitor. The assays were done in triplicate. IC_{50} values were determined as the concentration of a compound giving 50% inhibition of maximal activity.[49,50,61]

3.5. Antimicrobial activities of pyrimidine compounds (20–44)

The newly synthesized compounds (**20**–**45**) were screened for their in vitro antibacterial activity against *Escherichia coli* ATCC35218, *Yersinia pseudotuberculosis* ATCC911, *Pseudomonas aeruginosa* ATCC43288, *E. faecalis* ATCC29212, *S. aureus* ATCC25923, *B. cereus* 709 Roma, *Mycobacterium smegmatis* ATCC607, *Candida albicans* ATCC60193, and *Saccharomyces cerevisiae* RSKK 251, all obtained from the Refik Saydam Hygiene Center (Ankara, Turkey). All tested compounds were dissolved in hexane and diluted with dimethyl sulfoxide (DMSO) to prepare 10.0 mg/mL solutions. The antimicrobial effects of the compounds were tested quantitatively in respective broth media using double dilution, and minimal inhibition concentration (MIC) values (μg/mL) were determined.[51] The antibacterial and antifungal assays were performed in Mueller-Hinton broth (MH) (Difco, Detroit, MI, USA) at pH 7.3 and buffered yeast nitrogen base (Difco) at pH 7.0, respectively. Brain–heart infusion broth (BHI) (Difco) was used for *M. smegmatis*.[52] The MIC was defined as the lowest concentration showing no growth. Ampicillin (10.0 mg/mL), streptomycin (10.0 mg/mL), and fluconazole (5.0 mg/mL) were used as standard antibacterial and antifungal drugs, respectively. DMSO at a dilution of 1:10 was used as solvent. The smallest concentration at which the growth of the test microorganism was totally inhibited is reported as the MIC (μg/mL) value (Table 3).

4. Conclusion

We have successfully developed an efficient and simple method for preparing a variety of new substituted 4,6-diarylpyrimidin-2-ol (**20**–**34**) and 4,6-diarylpyrimidine-2-thiol (**35**–**44**) compounds with the solid-phase MW reaction. The pyrimidine compounds (**20**–**44**) were screened against nine bacterial species as well as against *C. albicans* and *S. cerevisiae*. Compounds **20**–**24** and **35**–**39** exhibited promising antimicrobial activities against the gram-positive bacteria (*E. faecalis*, *S. aureus*, and *B. cereus*). MIC values for *E. faecalis*, *S. aureus*, and *B. cereus* were within the range of 1.95–277.5 μg/mL, 2.1–138.8 μg/mL, and 1.95–19.1 μg/mL, respectively. Of all the synthesized new pyrimidine derivatives, compounds **20**, **21**, **23**, **24**, **35**, **36**, **37**, **38**, and **39** were more active than the standard (streptomycin, 4 μg/mL), and can be used as tuberculosis agents.

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