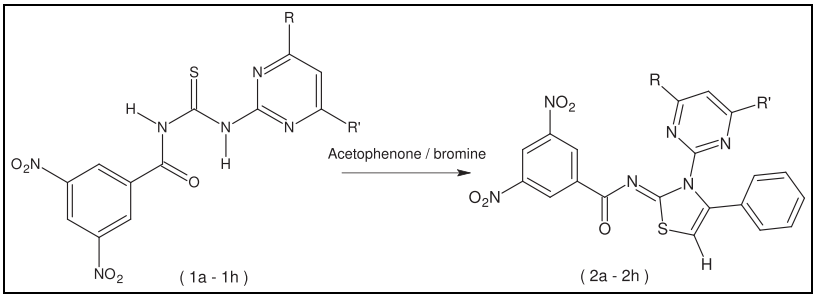
Synthesis and Antimicrobial Activity of *N*-[(2*Z*)-3-(4,6-Substitutedpyrimidin-2-yl)-4-phenyl-1,3-thiazol-2(3*H*)-ylidene]-3, 5-dinitrobenzamide Analogues

Sohail Saeed,[a,b]* Rizwan Hussain,[b] and Muhammad Ali[c]

[a]Department of Chemistry, Research Complex, Allama Iqbal Open University, Islamabad 44000, Pakistan
[b]National Engineering and Scientific Commission, P.O. Box 2801, Islamabad, Pakistan
[c]Department of Pharmacy, University of the Punjab, Lahore-54590, Pakistan
*E-mail: sohail262001@yahoo.com

Additional Supporting Information may be found in the online version of this article.
Received March 3, 2011
DOI 10.1002/jhet.964
Published online 29 March 2013 in Wiley Online Library (wileyonlinelibrary.com).



In this study, we have synthesized 1-(4,6-disubstitutedpyrimidin-2-yl)-3-(3,5-dinitrobenzoyl)-thiourea derivatives (**1a-1h**) and *N*-[(2Z)-3-(4,6-disubstitutedpyrimidin-2-yl)-4-phenyl-1,3-thiazol-2(3*H*)-ylidene]-3, 5-dinitrobenzamide (**2a-2h**) analogues and characterized by IR spectroscopy, NMR spectroscopy, elemental analysis, and single crystal X-ray diffraction data. The compounds (**2a-2h**) were screened for antimicrobial activity against Gram positive, Gram negative, and fungal species. The results of antimicrobial study indicated that compounds showed most potential and appreciable antibacterial and antifungal activities.

J. Heterocyclic Chem., **50**, 237 (2013).

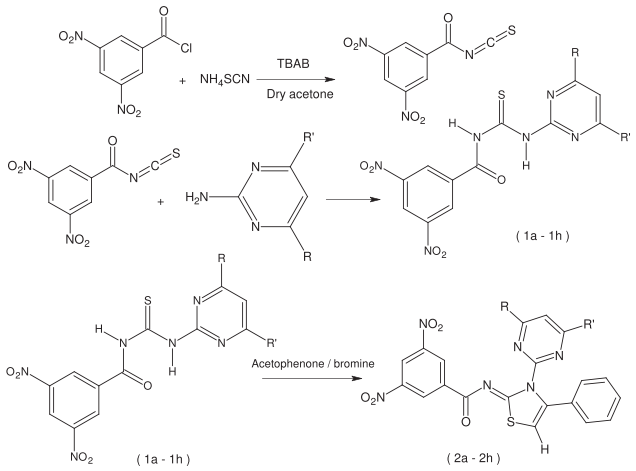
INTRODUCTION

In the family of heterocyclic compounds, nitrogen-containing heterocycles with a sulfur atom are the important class of compounds in medicinal chemistry. Pyrimidine moiety is an important class of N-containing heterocycles widely used as key building blocks for pharmaceutical agents when attached with thiazole functional group. It exhibits a wide spectrum of pharmacophore as it acts as bactericidal, fungicidal [1], analgesic [2], antihypertensive [3], and antitumor agents [4]. Among these, thiouracils are similarly used as anti-inflammatory and virucidal agents [5]. In addition, preclinical data from literature survey indicate continuing research in polysubstituted pyrimidine as potential antitumor agents [6]. The biological and synthetic significance places this scaffold at a prestigious position in medicinal chemistry research.

Thiazole heterocycle is a commonly occurring substructure in organic chemistry, found in a variety of structurally and biologically interesting natural products, such as mirabazoles, thiangazole, and tantazoles, which display selective cytotoxicity against murine solid tumors and are potent and selective inhibitors of HIV-1 [7, 8]. Synthetic thiazoline derivatives are useful medicaments showing antifungal [9, 10], antitumor [11], antiallergic [12], anti-inflammatory [13], antihypertension [14], analgesic, antibacterials, antirheumatic, antipyretic, and antiHIV [15] activities. Some fused thiazolines find applications in the treatment of allergies, hypertension, inflammation, schizophrenia, and bacterial and HIV infections [16]. 2-Thiazolylimino-5-arylidene-4-thiazolidinones show marked antimicrobial activity against bacteria, yeasts, and molds [17]. Some cyclic chiral oligothiazolines, having a wheel-like architecture containing a linear array of thiazoline rings, are well known supramolecules [18]. 2-(Tetrahydronaphthalen-1-yl)iminothiazolidine exhibits pronounced antidepressant activity [19], and β-(hydrooxyethyl)thiazolidines are effective antihypertensives [20]. 3-Substituted 2-(cyanoimino)thiazolidines can be used in agriculture because of their neonicotinoid insecticidal activity [21]. 3-Substituted thiazolidines show radioprotective properties against γ-radiations [22]. 2-Imino-1,3-thiazolines derivatives have shown antifungal activity against the rice blast fungus *Pyricularia oryzae* [23], and thus, can be used as fungicides. 2-Imino-1,3-thiazoline derivatives significantly inhibits melanin production in a dose-dependant manner, thus acting as a skin whitening agent [24], and pifithrin-α is a reversible inhibitor of p53-mediated apoptosis and p53-dependant gene transcription [25].

The importance of such work lies in the possibility that the next generation thiazole derivatives bearing pyrimidine nucleus might be more efficacious as antimicrobial agents.

Scheme 1. Synthesis of *N*-[(2*Z*)-3-(4,6-substitutedpyrimidin-2-yl)-4-phenyl-1,3-thiazol-2(3*H*)-ylidene]-3,5-dinitrobenzamide analogues.



	R	R'
1a, 2a	CH_3	CH_3
1b, 2b	OCH_3	OCH_3
1c, 2c	C_2H_5	CH_3
1d, 2d	C_2H_5	C_2H_5
1e, 2e	OC_2H_5	OCH_3
1f, 2f	OCH_3	CH_3
1g, 2g	OC_2H_5	C_2H_5
1h, 2h	OC_2H_5	OC_2H_5

However, a thorough investigation relating the structure and the activity of the thiazole derivatives as well as their stability under biological conditions is required. These detailed investigations could be helpful in designing more potent antimicrobial agents. Since varying substituents is a common method for drug design in medicinal chemistry and a useful medical value of substituted thiazole derivatives containing pyrimidine moiety, we aimed to synthesize new thiazole derivatives and to investigate their antimicrobial activities. On the basis of these reports, we herein report the synthesis, characterization, and antimicrobial activity of a series of novel thiazole derivatives bearing pyrimidine moiety.

RESULTS AND DISCUSSION

Synthesis and spectral properties. *N*-[(4,6-disubstituted-pyrimidin-2-yl)carbamothioyl]-3,5-dinitrobenzamide (**1a-1h**) were prepared according to the published procedure involving treatment of 3,5-dinitrobenzoyl chloride with ammonium thiocyanate in anhydrous acetone followed by reaction with suitably substituted anilines [26–29]. The use of phase transfer catalysts as a method of agitating a heterogeneous reaction system is gaining recognition [30, 31]. In search of improved methods to prepare the *N*-[(4,6-disubstitutedpyrimidin-2-yl) carbamothioyl]-3, 5-dinitrobenzamide by reacting isothiocyanates with nucleophiles, we have found the use of tetrabutylammonium bromide (TBAB) as a phase transfer catalyst (PTC) can afford aroyl isothiocyanates in good yield, as reported here (Scheme 1).

The compounds (**1a-1h**) showed absorptions at 3351 and 3200 cm^{-1} for free and associated NH, at 1660–1685 for carbonyl, and at 1230 cm^{-1} for thiocarbonyl groups in IR spectra and singlets at δ 9.0–9.5 and 11.2–12.1 (ppm) for NH (1) and NH (3) and peaks at 168–170 and 178–180 (ppm) for carbonyl and thiocarbonyl were observed in ^1H and ^{13}C NMR spectra, respectively.

The cyclocondensation of *N*-[(4,6-disubstitutedpyrimidin-2-yl)carbamothioyl]-3,5-dinitrobenzamide (**1a-1h**) with acetophenone was achieved in the presence of bromine and base. Thus, triethyl amine was added to a solution of *N*-[(4,6-disubstitutedpyrimidin-2-yl) carbamothioyl]-3,5-dinitrobenzamide in anhydrous dichloromethane, followed by the treatment with a mixture of acetophenone and bromine under an inert atmosphere to afford heterocyclic *N*-[(2*Z*)-3-(4,6-substituted-pyrimidin-2-yl)-4-phenyl-1,3-thiazol-2(3*H*)-ylidene]-3,5-dinitrobenzamide derivatives (**2a-2h**).

The absence of N-H peaks at 3200–3400 cm^{-1}, slight shifting of C═O absorptions to 1630–1675 cm^{-1}, and appearance of characteristic C═N at 1450–1495 cm^{-1} were observed in the IR spectra. ^1H NMR spectra showed the disappearance of N-H peaks and emergence of a ^1H characteristic singlet at δ 6.7–6.9 because of C═C-H of the thiazole ring.

In ^{13}C NMR the characteristic peak for olefinic carbon at δ107.5–107.8 (ppm) confirmed the formation of *N*-[(2*Z*)-3-(4,6-substitutedpyrimidin-2-yl)-4-phenyl-1,3-thiazol-2(3*H*)-ylidene]-3,5-dinitrobenzamide derivatives. Although the same mesomeric anionic thiourea intermediate may furnish either imino-1,3-thiazoles (*S*-cyclization products) or isomeric imidazole-2-thiones (*N*-cyclization products), it has already equally demonstrated [32] that under these conditions the thermodynamically stable imino-1,3-thiazoles are the exclusive products as reported in previous papers [33, 34].

Crystal structure study. The compound, 1-(4,6-dimethylpyrimidin-2-yl)-3-(3,5-dinitrobenzoyl)-thiourea (**1a**), crystallizes in a monoclinic primitive space group, P 2_1/n (Table 1). The packing is three-dimensional, but a representative two-dimensional view along the short axis and a molecular diagram is provided in the Supporting Information.

Unlike its other analog, the molecule is almost planar. The nitro groups are 5.05 (11)° and 9.40 (15)° from the phenyl ring plane of C1-C6. The thiourea group is almost coplanar with the amido group. The 4, 6-dimethyl-pyrimidinyl ring plane, C9-C14/N5/N6, makes a dihedral angle of 1.31 (5) with the the plane, C7/O5/N3/C8/S1/N4. There are intramolecular N-H···O and N-H···N H-bond interactions. The intermolecular O-H···O H-bond interactions link the molecules to form 2D networks in the crystal lattice (Table 2). There are also weak π···π between neighboring benzene rings in the crystal lattice. There is no residual solvent accessible void volume in the unit cell.

Table 1

Crystallographic data, data collection and structure refinement for 1-(4,6-dimethylpyrimidin-2-yl)-3-(3,5-dinitrobenzoyl)-thiourea (**1a**).

Empirical formula	$C_{14}H_{14}N_6O_6S$
Formula weight	394.37
Temperature	297 (2) K
Wavelength	1.54184 Å
Crystal system	Monoclinic
Space group	$P2_1/n$
Unit cell dimensions	a = 6.7892 (6) Å b = 10.1823 (9) Å c = 24.267 (2) Å β= 92.901 (1)°
Volume	1675.4 (3) $Å^3$
Z	4
Density (calculated)	1.563 g/cm3
Absorption coefficient	0.24 mm^{-1}
F(000)	816
Crystal size	0.40 × 0.16 × 0.14 mm^3
Reflections collected	9067
Independent reflections	2942
Absorption correction	multI-scan, SADABS (Sheldrick, 2004)
Max. and min. transmission	0.909 and 0.967
Refinement method	Full-matrix least-squares on F2
$(\Delta/\sigma)_{max}$	0.003
$\Delta\rho_{max}$	0.20 e $Å^{-3}$
$\Delta\rho_{min}$	–0.23 e $Å^{-3}$
CCDC	805048

Antimicrobial activities. In the light of interesting antimicrobial activities of thiazole derivatives were screened for antibacterial and antifungal activity against *Staphylococcus aureus, Staphylococcus epidermidis*, *Escherichia coli*, *Proteus valgaris*, *Candida albicans,* and *Candida glabrata* by the broth microdilution procedure.

The Gram-positive antibacterial agent, amikacin, the Gram-negative antibacterial agent, gentamycin, and the antifungal agent, nystatin, were used as controls. The *in vitro* antimicrobial properties against a number of Gram-negative, Gram-positive bacteria, and yeasts are presented in Table 3.

All the compounds inhibited the growth of bacteria with MIC values ranging between 40 and 100 μg/mL and showed antiyeast activity with MICs between 30 and 50 μg/mL. Compounds which showed MIC of >100 μg/mL or above have not been included for the discussion. According to the antimicrobial studies, all the compounds showed such activity, albeit lower than their antiyeast efficacy. This difference may be due to the differences between the cell structure of bacteria and yeast. The cell wall of fungi contain chitin, whereas the cell wall of bacteria contains murein [35]. In addition, fungi contain ergosterol in their cell membranes instead of the cholesterol found in the cell membranes of animals [36]. When all the antiyeast MIC values are compared, two of eight compounds showed good activity against *C.albicans* and seven showed low activity against *C.glabrata*. According to the antibacterial studies, the efficacy against Gram-positive is higher than Gram-negative bacteria. Two of eight compounds showed good activity against *S.epidermidis*. In addition, **2e** and **2h** showed high activity against Gram-positive, Gram-negative bacteria, and fungi.

Table 2

Hydrogen bonds [Å and °] for compound **1a**.

D-H…A	d (D-H)	d (H… A)	D (D… A)	<(DHA)
N4—H4N···O6	0.88(2)	1.97(2)	2.849(2)	171.9(19)
N3—H3N···N6	0.83(2)	2.00(2)	2.705(2)	141.8(19)
O6—H6A···O3[a]	0.82 (4)	2.32 (4)	3.119 (3)	3.119 (3)
O6—H6B···O2[b]	0.73 (4)	2.44 (4)	3.143 (3)	164 (4)
O6—H6B···O1[b]	0.73(4)	2.62(4)	3.248(3)	146(4)

Symmetry codes:
[a] *x*-1/2, -*y*+1/2, *z*+1/2.
[b] *x*-1/2, -*y*+3/2, *z*+1/2.

CONCLUSIONS

The straightforward approach, simplicity and first one step method make it an interesting approach for the synthesis of thiazole-pyrimidine core derivatives. An *in vitro* screen led to the identification of compounds **2e** and **2h** as potential antifungal candidates worthy of further structural modification and pharmacological evaluation. Moreover, the antimicrobial activity of this series suggests the thiazole-pyrimidine core offers a novel template for the development of a new class of antimicrobial agents.

EXPERIMENTAL

Melting points were recorded on Electrothermal IA9000 series digital melting point apparatus. The proton NMR and ^{13}C spectra

Table 3

MIC values (μg/mL) of the synthesized thiazole derivatives against the tested Gram-positive, Gram-negative bacteria and yeasts.

Compounds	1	2	3	4	5	6
2a	100	>100	100	>100	50	50
2b	100	100	50	100	50	50
2c	100	>100	50	100	50	50
2d	100	100	50	80	50	50
2e	50	80	80	100	30	30
2f	80	100	80	80	50	50
2g	80	80	100	50	50	50
2h	40	50	50	80	50	30
Ref. drugs	0.5[a]	2[a]	0.5[b]	2[b]	2[c]	1[c]

1: *S.epidermidis* (ATCC 12228), 2: *S.aureus* (ATCC 29213), 3: *E.coli* (ATCC 25922), 4: *P.vulgaris*(ATCC 13315), 5: *C.glabrata* (ATCC 32554), 6: *C.albicans* (ATCC 90028)
[a] Amikacin.
[b] Gentamycin.
[c] Nystatin.

were recorded in DMSO-d_6 solvent on Bruker 300 MHz spectrophotometer using tetramethylsilane as an internal reference, respectively. The apparent resonance multiplicity is described as s (singlet), br s (broad singlet), d (doublet), dd (doublet of doublets), t (triplet), q (quartet), and m (multiplet). Infrared measurements were recorded in the range 400–4000 cm^{-1} on spectrum 2000 by Perkin Elmer. Elemental analysis was carried out using Perkin Elmer CHNS/O 2400. Obtained results were within 0.4 % of the theoretical values. Thin layer chromatography (TLC) analysis were carried out on 5 × 20 cm plate coated with silica gel GF_{254} type 60 (25–250 mesh) using an ethyl acetate-dichloromethane mixture (1:2) as solvent. Synthetic starting material, reagents, and solvents were of analytical reagent grade or of the highest quality commercially available and were purchased from Aldrich Chemical Co., Merck Chemical Co. and were dried when necessary. 4, 6-disubstituted 2-amino-pyrimidines were prepared by the literature method [37].

General procedure for preparation of compounds. ***1-(4,6-Disubstitutedpyrimidin-2-yl)-3-(3,5-dinitrobenzoyl)-thiourea (1a-1h).*** A solution of 3, 5-dinitrobenzoyl chloride (26 mmol) in anhydrous acetone (80 mL) and 3% tetrabutyl ammonium bromide (TBAB) in acetone was added dropwise to a suspension of ammonium thiocyanate in acetone (50 mL) and the reaction mixture was refluxed for 45 min. After cooling to room temperature, a solution of 4,6-disubstituted-2-aminopyrimidines of equimolar quantity in acetone (25 mL) was added and the resulting mixture refluxed for 2.5 h. The reaction mixture was poured into five times its volume of cold water, whereupon the thiourea precipitated. The solid product was washed with water and purified by recrystallization from ethyl acetate.

N-[(2Z)-3-(4,6-Disubstitutedpyrimidin-2-yl)-4-phenyl-1,3-thiazol-2(3H)-ylidene]-3,5-dinitrobenzamide (2a-2h). Triethylamine (0.01 mol) was added to a stirred solution of 1-(4,6-disubstitutedpyrimidin-2-yl)-3-(3,5-dinitrobenzoyl)-thioureas (0.01 mol) in dry dichloromethane (30 mL), followed by drop-wise addition of a solution of bromine (0.01 mol) in acetophenone (0.01 mol) under nitrogen. The reaction mixture was stirred for 1–2 h and progress of the reaction was monitored by thin layer chromatography (hexane: ethyl acetate, 4:1). After the reaction was complete, the mixture was filtered; the filtrate was concentrated to afford thiazole derivatives, which were purified by recrystallization from ethanol.

Spectroscopic data of representative compounds. ***1-(4,6-Dimethyl-pyrimidin-2-yl)-3-(3,5-dinitrobenzoyl)-thiourea (1a).*** Elemental analysis for $C_{14}H_{12}N_6O_5S$ (MW = 376.3) in wt % calc. C = 44.68, H = 3.21, N = 22.33, S = 8.52, found C = 44.71, H = 3.25, N = 22.30, S = 8.51. m.p. 155–156°C, yield 86 %. IR (KBr pellet) in cm^{-1}: 3352 (free NH), 3208 (assoc. NH), 1686 (C=O), 1615 (C=N stretching), 1592 (aromatic C=C); ^{1}H NMR (300 MHz, DMSO-d_6) in δ (ppm): 12.80 (1H, s, broad, NH), 11.63 (1H, s, broad, NH), 8.54–8.14 (3H, m, Ar CH), 6.56(1H, s, pyrimidine-5-H),1.21(6H,s, pyrimidine-C<u>H</u>$_3$); ^{13}C NMR (300 MHz, DMSO-d_6) in δ (ppm): 180.1, 168.4,164.5, 162.4,153.5,140.2, 128.0, 123.4, 25.4.

1-(4,6-Dimethoxypyrimidin-2-yl)-3-(3,5-dinitrobenzoyl)-thiourea (1b). Elemental analysis for $C_{14}H_{12}N_6O_7S$ (MW = 408.3) in wt % calc. C = 41.18, H = 2.96, N = 20.58, S = 7.85, found C = 41.16, H = 2.99, N = 20.57, S = 7.85. m.p 147–149°C, yield 82 %. IR (KBr pellet) in cm^{-1}: 3355 (free NH), 3207 (assoc. NH), 1682 (C=O), 1615 (C=N stretching), 1590 (aromatic C=C); ^{1}H NMR (300 MHz, DMSO-d_6) in δ (ppm): 12.78 (1H, s, broad, NH), 11.60 (1H, s, broad, NH), 8.51–8.11 (3H, m, Ar CH), 6.57(1H, s, pyrimidine-5-H), 3.19 (6H,s, pyrimidine-OC<u>H</u>$_3$); ^{13}C NMR (300 MHz, DMSO-d_6) in δ (ppm): 180.3, 168.2,164.5, 162.4, 153.5,140.2, 128.0, 123.4, 53.1.

1-(4-Ethyl-6-methylpyrimidin-2-yl)-3-(3,5-dinitrobenzoyl)-thiourea (1c). Elemental analysis for $C_{15}H_{14}N_6O_5S$ (MW = 390.3) in wt % calc. C = 46.15, H = 3.61, N = 21.53, S = 8.21, found C = 46.18, H = 3.65, N = 21.50, S = 8.20. m.p. 161–162°C, yield 79 %. IR (KBr pellet) in cm^{-1}: 3351 (free NH), 3205 (assoc. NH), 1684 (C=O), 1613 (C=N stretching), 1590 (aromatic C=C); ^{1}H NMR (300 MHz, DMSO-d_6) in δ (ppm): 12.77 (1H, s, broad, NH), 11.64 (1H, s, broad, NH), 8.55–8.11 (3H, m, Ar CH), 6.55 (1H, s, pyrimidine-5-H), 1.79 (2H,m, pyrimidine-C<u>H</u>$_2$CH$_3$); 1.28 (3H,t, pyrimidine-CH$_2$C<u>H</u>$_3$); 1.21 (3H,s, pyrimidine-C<u>H</u>$_3$); ^{13}C NMR (300 MHz,DMSO-d_6) in δ(ppm):180.2, 168.0, 164.5, 162.4, 153.5, 140.2, 128.0, 123.4, 35.1, 24.3, 21.3.

1-(4,6-Diethylpyrimidin-2-yl)-3-(3,5-dinitrobenzoyl)-thiourea (1d). Elemental analysis for $C_{16}H_{16}N_6O_5S$ (MW = 404.4) in wt % calc. C = 47.52, H = 3.99, N = 20.78, S = 7.93, found C = 47.56, H = 3.99, N = 20.75, S = 7.92. m.p. 158–159°C, yield 86 %. IR (KBr pellet) in cm^{-1}: 3352 (free NH), 3207 (assoc. NH), 1686 (C=O), 1618 (C=N stretching), 1596 (aromatic C=C); ^{1}H NMR (300 MHz, DMSO-d_6) in δ (ppm): 12.79 (1H, s, broad, NH), 11.64 (1H, s, broad, NH), 8.50–8.17 (3H, m, Ar CH), 6.54 (1H, s, pyrimidine-5-H),1.87 (4H,m, pyrimidine-C<u>H</u>$_2$CH$_3$); 1.28 (6H,t, pyrimidine-CH$_2$C<u>H</u>$_3$); ^{13}C NMR (300 MHz, DMSO-d_6) in δ (ppm): 180.0, 168.0, 164.5, 162.4, 153.5, 140.2, 128.0, 123.4, 34.6, 23.8.

1-(4-Ethoxy-6-methoxypyrimidin-2-yl)-3-(3,5-dinitrobenzoyl)-thiourea (1e). Elemental analysis for $C_{15}H_{14}N_6O_7S$ (MW = 422.3) in wt % calc. C = 42.65, H = 3.34, N = 19.90, S = 7.59, found C = 42.67, H = 3.39, N = 19.89, S = 7.59. m.p. 139–140°C, yield 80 %. IR (KBr pellet) in cm^{-1}: 3352 (free NH), 3201 (assoc. NH), 1688 (C=O), 1615 (C=N stretching), 1589 (aromatic C=C); ^{1}H NMR (300 MHz, DMSO-d_6) in δ (ppm): 12.75 (1H, s, broad, NH), 11.66 (1H, s, broad, NH), 8.54–8.11 (3H, m, Ar CH), 6.56(1H, s, pyrimidine-5-H),3.85 (2H,m, pyrimidine-OC<u>H</u>$_2$CH$_3$);2.15 (3H,t, pyrimidine-OCH$_2$C<u>H</u>$_3$); 2.28 (3H,s, pyrimidine-OC<u>H</u>$_3$); ^{13}C NMR (300 MHz, DMSO-d_6) in δ (ppm): 180.2, 168.2,164.5, 162.4, 153.5,140.2, 128.0, 123.4, 56.4, 54.2, 28.5.

1-(4-Methoxy-6-methylpyrimidin-2-yl)-3-(3,5-dinitrobenzoyl)-thiourea (1f). Elemental analysis for $C_{14}H_{12}N_6O_6S$ (MW = 392.3) in wt % calc. C = 42.86, H = 3.08, N = 21.42, S = 8.17, found C = 42.84, H = 3.12, N = 21.41, S = 8.18. m.p. 153–154°C, yield 79 %. IR (KBr pellet) in cm^{-1}: 3354 (free NH), 3206 (assoc. NH), 1686 (C=O), 1614 (C=N stretching), 1596 (aromatic C=C); ^{1}H NMR (300 MHz, DMSO-d_6) in δ (ppm): 12.78 (1H, s, broad, NH), 11.63 (1H, s, broad, NH), 8.53–8.09 (3H, m, Ar CH), 6.59(1H, s, pyrimidine-5-H), 3.19 (3H,t, pyrimidine-OC<u>H</u>$_3$); 1.23 (3H,s, pyrimidine-C<u>H</u>$_3$); ^{13}C NMR (300 MHz, DMSO-d_6) in δ (ppm): 180.3,168.2,164.5,162.4,153.5,140.2, 128.0, 123.4, 56.3, 23.8.

1-(4-Ethoxy-6-ethylpyrimidin-2-yl)-3-(3,5-dinitrobenzoyl)-thiourea (1g). Elemental analysis for $C_{16}H_{16}N_6O_6S$ (MW = 420.3) in wt % calc. C = 45.71, H = 3.84, N = 19.99, S = 7.63, found C = 45.74, H = 3.83, N = 19.96, S = 7.62. m.p. 136–138°C, yield 73 %. IR (KBr pellet) in cm^{-1}: 3351 (free NH), 3209 (assoc. NH), 1685 (C=O), 1613 (C=N stretching), 1590 (aromatic C=C); ^{1}H NMR (300 MHz, DMSO-d_6) in δ (ppm): 12.81 (1H, s, broad, NH), 11.60 (1H, s, broad, NH), 8.55–8.10 (3H, m, Ar CH), 6.52 (1H, s, pyrimidine-5-H),3.85(2H,m,pyrimidine-OC<u>H</u>$_2$CH$_3$);2.45(3H,t, pyrimidine-OCH$_2$C<u>H</u>$_3$); 1.46 (2H,s, pyrimidine-C<u>H</u>$_2$CH$_3$),1.24 (3H,s,

pyrimidine-$CH_2\underline{CH_3}$); ^{13}C NMR (300 MHz, DMSO-d_6) in δ (ppm): 180.1,168.3,164.5,162.4,153.5,140.2, 128.0, 123.4, 56.7, 45.4,35.4, 22.1.

1-(4,6-Diethoxypyrimidin-2-yl)-3-(3,5-dinitrobenzoyl)-thiourea (1h). Elemental analysis for $C_{16}H_{16}N_6O_7S$ (MW = 436.3) in wt % calc. C = 44.04, H = 3.70, N = 19.26, S = 7.35, found C = 44.00, H = 3.72, N =19.25, S = 7.38. m.p. 131–132°C, yield 81%. IR (KBr pellet) in cm^{-1}: 3350 (free NH), 3209 (assoc. NH), 1681 (C═O), 1613 (C═N stretching), 1596 (aromatic C═C); ^{1}H NMR (300 MHz, DMSO-d_6) in δ (ppm): 12.77 (1H, s, broad, NH), 11.61 (1H, s, broad, NH), 8.50–8.13 (3H, m, Ar CH), 6.56(1H, s, pyrimidine-5-H),3.78(4H,m,pyrimidine-$OC\underline{H_2}CH_3$);2.41(6H, t, pyrimidine-$OCH_2C\underline{H_3}$); ^{13}C NMR (300 MHz, DMSO-d_6) in δ (ppm): 180.0,168.2,164.5,162.4,153.5,140.2,128.0, 123.4, 56.1, 45.8.

N-[(2Z)-3-(4,6-Dimethylpyrimidin-2-yl)-4-phenyl-1,3-thiazol-2(3H)-ylidene]-3,5-dinitrobenzamide (2a). Elemental analysis for $C_{22}H_{16}N_6O_5S$ (MW = 476.46) in wt % calc. C = 55.46, H = 3.38, N = 17.64, S = 6.73 and found to be C = 55.11, H = 3.45, N = 17.65, S = 6.70. m.p.251°C, yield 65 %.IR (KBr pellet) in cm^{-1}: 1670 (C═O), 1525 (Ar-C═C), 1456 (C═N stretching), 1275 (C-S), 1155 (C-N). ^{1}H NMR (300 MHz, DMSO-d_6) in δ (ppm): 8.50–8.13 (3H, m, Ar CH), 7.65–7.28 (5H, m, phenyl-H), 6.73 (1H, s, thiazole CH), 6.45 (1H, s, pyrimidine-5-H), 1.23 (6H,s, pyrimidine-$C\underline{H_3}$); ^{13}C NMR (300 MHz, DMSO-d_6) in δ (ppm) : 171.02, 168.31, 139.62, 138.04, 137.75, 136.25, 135.70, 134.30, 132.47, 130.10, 129.56, 129.10, 128.64, 128.26, 126.81, 126.08,107.68, 21.81,21.15.

N-[(2Z)-3-(4,6-Dimethoxypyrimidin-2-yl)-4-phenyl-1,3-thiazol-2(3H)-ylidene]-3,5-dinitrobenzamide (2b). Elemental analysis for $C_{22}H_{16}N_6O_7S$ (MW = 358.39) in wt % calc. C = 51.97, H = 3.17, N = 16.53, S = 6.31 and found to be C = 51.92, H = 3.22, N = 16.52, S = 6.35. m.p.245°C, yield 62 %.IR (KBr pellet) in cm^{-1}: 1659 (C═O), 1528 (Ar-C═C), 1459 (C═N stretching), 1271 (C-S), 1152 (C-N). ^{1}H NMR (300 MHz, DMSO-d_6) in δ (ppm): 8.25–8.05 (3H, m, Ar CH), 7.65–7.28 (5H, m, phenyl-H), 6.78(1H, s, thiazole CH), 6.54 (1H, s, pyrimidine-5-H), 3.21 (6H,s, pyrimidine-$OC\underline{H_3}$); ^{13}C NMR (300 MHz, DMSO-d_6) in δ (ppm) : 176.02, 169.50, 139.67, 138.62, 137.75, 136.05, 134.31, 132.49, 130.17, 130.1, 129.84, 129.10, 128.61, 128.08, 126.64, 126.08, 107.61, 55.10.31,53.13.

N-[(2Z)-3-(4-Ethyl-6-methylypyrimidin-2-yl)-4-phenyl-1,3-thiazol-2(3H)-ylidene]-3,5-dinitrobenzamide (2c). Elemental analysis for $C_{23}H_{18}N_6O_5S$ (MW = 490.49) in wt % calc. C = 56.32, H = 3.70, N = 17.13, S = 6.54 and found to be C = 56.36, H = 3.74, N = 17.11, S = 6.53. m.p. 265–266°C, yield 59 %.IR (KBr pellet) in cm^{-1}: 1668 (C═O), 1525 (Ar-C═C), 1456 (C═N stretching), 1275 (C-S), 1155 (C-N). ^{1}H NMR (300 MHz, DMSO-d_6) in δ (ppm): 8.19–8.05 (3H, m, Ar CH), 7.65–7.28 (5H, m, phenyl-H), 6.77(1H, s, thiazole CH), 6.46 (1H, s, pyrimidine-5-H), 1.85 (2H,m, pyrimidine-$C\underline{H_2}CH_3$); 1.25 (3H,t, pyrimidine-$C\underline{H_3}$); 1.21 (3H,s, pyrimidine-$C\underline{H_3}$); ^{13}C NMR (300 MHz, DMSO-d_6) in δ (ppm): 171.02, 168.31, 139.62, 138.04, 137.75, 136.25, 135.70, 134.30, 132.47, 130.10, 130.0, 129.56, 129.10, 128.64, 128.26, 126.81, 126.08, 107.8, 45.5,29.31,21.15.

N-[(2Z)-3-(4,6-Diethylpyrimidin-2-yl)-4-phenyl-1,3-thiazol-2(3H)-ylidene]-3,5-dinitrobenzamide (2d). Elemental analysis for $C_{24}H_{20}N_6O_5S$ (MW = 504.51) in wt % calc. C = 57.14, H = 4.00, N = 16.66, S = 6.36 and found to be C = 57.13, H=4.25, N=16.65, S= 6.35. m.p. 248–249°C, yield 60 %.IR (KBr pellet) in cm^{-1}: 1672 (C═O), 1523 (Ar-C═C), 1481 (C═N stretching), 1268 (C-S), 1153 (C-N). ^{1}H NMR (300 MHz, DMSO-d_6) in δ (ppm): 8.19–8.05 (3H, m, Ar CH), 7.65–7.28 (5H, m, phenyl-H), 6.76(1H, s, thiazole CH), 6.42 (1H, s, pyrimidine-5-H), 1.85 (4H,m, pyrimidine-$C\underline{H_2}CH_3$),1.20 (6H,t, pyrimidine-$CH_2C\underline{H_3}$); ^{13}C NMR (300 MHz, DMSO-d_6) in δ (ppm): 171.02, 168.31, 139.62, 138.04, 137.75, 136.25, 135.70, 134.30, 132.47, 130.10, 130.0, 129.56, 129.10, 128.64, 128.26, 126.81, 126.08,107.54, 46.12, 21.15.

N-[(2Z)-3-(4-Ethoxy-6-methoxypyrimidin-2-yl)-4-phenyl-1,3-thiazol-2(3H)-ylidene]-3,5-dinitrobenzamide (2e). Elemental analysis for $C_{23}H_{18}N_6O_7S$ (MW = 522.49) in wt % calc. C = 52.87, H = 3.47, N = 16.08, S = 6.14 and found to be C = 52.90, H = 3.50, N = 16.05, S= 6.12. m.p. 236–238°C, yield 65 %.IR (KBr pellet) in cm^{-1}: 1672 (C═O), 1525 (Ar-C═C), 1495 (C═N stretching), 1272 (C-S), 1153 (C-N). ^{1}H NMR (300 MHz, DMSO-d_6) in δ (ppm): 8.19–8.05 (3H, m, Ar CH), 7.65–7.28 (5H, m, phenyl-H), 6.81(1H, s, thiazole CH), 6.56 (1H, s, pyrimidine-5-H), 2.81 (2H,m, pyrimidine-$OC\underline{H_2}CH_3$); 2.75 (3H,s, pyrimidine-$OC\underline{H_3}$), 1.95 (3H,t, pyrimidine-$OCH_2C\underline{H_3}$); ^{13}C NMR (300 MHz, DMSO-d_6) in δ (ppm) : 171.02, 168.31, 139.62, 138.04, 137.75, 136.25, 135.70, 134.30, 132.47, 130.10, 130.0, 129.56, 129.10, 128.64, 128.26, 128.0, 126.81, 107.8, 56.7, 45.8, 29.31.

N-[(2Z)-3-(4-Methyl-6-methoxypyrimidin-2-yl)-4-phenyl-1,3-thiazol-2(3H)-ylidene]-3,5-dinitrobenzamide (2f). Elemental analysis for $C_{22}H_{16}N_6O_6S$ (MW = 492.46) in wt % calc. C = 53.66, H = 3.27, N = 17.07, S = 6.51 and found to be C = 53.64, H = 3.30, N = 17.10, S = 6.50. m.p. 241–242°C, yield 69 %.IR (KBr pellet) in cm^{-1}: 1674 (C═O), 1523 (Ar-C═C), 1490 (C═N stretching), 1269 (C-S), 1151 (C-N). ^{1}H NMR (300 MHz, DMSO-d_6) in δ (ppm): 8.19–8.05 (3H, m, Ar CH), 7.65–7.28 (5H, m, phenyl-H), 6.79(1H, s, thiazole CH), 6.51 (1H, s, pyrimidine-5-H), 2.57 (3H,s, pyrimidine-$OC\underline{H_3}$); 1.28 (3H,s, pyrimidine-$C\underline{H_3}$); ^{13}C NMR (300 MHz, DMSO-d_6) in δ (ppm) : 171.02, 168.31, 139.62, 138.04, 137.75, 136.25, 135.70, 134.30, 132.47, 130.10, 130.0, 129.56, 129.10, 128.64, 128.26, 128.0, 126.81, 107.70, 54.25, 24.16.

N-[(2Z)-3-(4-Ethoxy-6-ethylpyrimidin-2-yl)-4-phenyl-1,3-thiazol-2(3H)-ylidene]-3,5-dinitrobenzamide (2g). Elemental analysis for $C_{24}H_{20}N_6O_6S$ (MW = 520.51) in wt % calc. C = 55.38, H = 3.87, N = 16.15, S = 6.16 and found to be C = 55.39, H = 3.90, N = 16.13, S = 6.18. m.p. 251–253°C, yield 61 %.IR (KBr pellet) in cm^{-1}: 1671 (C═O), 1528 (Ar-C═C), 1488 (C═N stretching), 1270 (C-S), 1151 (C-N). ^{1}H NMR (300 MHz, DMSO-d_6) in δ (ppm): 8.19–8.05 (3H, m, Ar CH), 7.65–7.28 (5H, m, phenyl-H), 6.80(1H, s, thiazole CH), 6.48 (1H, s, pyrimidine-5-H), 2.74 (2H,m, pyrimidine-$OC\underline{H_2}CH_3$);1.98 (3H,t, pyrimidine-$OCH_2C\underline{H_3}$),1.85 (2H,m, pyrimidine-$C\underline{H_2}CH_3$), 1.20 (3H,t, pyrimidine-$CH_2C\underline{H_3}$); ^{13}C NMR (300 MHz, DMSO-d_6) in δ (ppm) : 171.02, 168.31, 139.62, 138.04, 137.75, 136.25, 135.70, 134.30, 132.47, 130.10, 130.0, 129.56, 129.10, 128.64, 128.26, 128.0, 126.81, 107.52, 54.3, 46.34, 35.40, 21.31.

N-[(2Z)-3-(4,6-Diethoxypyrimidin-2-yl)-4-phenyl-1,3-thiazol-2(3H)-ylidene]-3,5-dinitrobenzamide (2h). Elemental analysis for $C_{24}H_{20}N_6O_7S$ (MW = 536.51) in wt % calc. C = 53.73, H = 3.76, N = 15.66, S = 5.98 and found to be C = 53.71, H = 3.81, N = 15.63, S = 5.98. m.p. 232–233°C, yield 65 %.IR (KBr pellet) in cm^{-1}: 1674 (C═O), 1522 (Ar-C═C), 1468 (C═N stretching), 1271 (C-S), 1158 (C-N). ^{1}H NMR (300 MHz, DMSO-d_6) in δ (ppm): 8.19–8.05 (3H, m, Ar CH), 7.65–7.28 (5H, m, phenyl-H), 6.81(1H, s, thiazole CH), 6.50 (1H, s, pyrimidine-5-H), 2.81 (4H, m, pyrimidine-$OC\underline{H_2}CH_3$);1.93 (6H,t, pyrimidine-$OCH_2C\underline{H_3}$); ^{13}C NMR (300 MHz, DMSO-d_6) in δ (ppm): 171.02, 168.31, 139.62, 138.04, 137.75, 136.25, 135.70, 134.30, 132.47, 130.10,

130.0, 129.56, 129.10, 128.64, 128.26, 128.0, 126.81, 107.41, 56.41, 45.32.

X-ray data collection and structure refinement. 1a: A yellow prism crystal of 1-(4,6-Dimethylpyrimidin-2-yl)-3-(3,5-dinitrobenzoyl)-thiourea monohydrate, $C_{14}H_{12}N_6O_5S_1.H_2O$, having approximate dimensions of 0.14 mm × 0.16 mm × 0.40 mm was mounted on glass fiber. All measurements were made on a Bruker *SMART* 1000 CCD detector with graphite monochromated Mo-Kα radiation. Indexing was performed from 60 images that were exposed for 10 s for a preliminary unit cell determination. Of which, 86 out of a total of 110 reflections were successfully indexed. The crystal-to-detector distance was 50.00 mm. Crystal data: $C_{14}H_{12}N_6O_5S_1.H_2O$, monoclinic, space group $P2_1/n$, a = 6.7892 (6) Å, b = 10.1823 (9) Å, c = 24.267 (2) Å, β = 92.901 (1)°, V = 1675.4 (3) Å^3, T = 297(2) K, Z = 4, F (000) = 816, D_x = 1.563 g cm^{-3}, μ = 0.24 mm^{-1}. The data were collected at a temperature of 24(1)°C to a maximum 2θ value of 50.05°. A total of 1421 oscillation images were collected in 4 runs. A sweep of data was done using ω scans from 330.0 to 148.2° in –0.3° step, at χ = 54.7° and φ = 0.0°. The exposure rate was 30.0 [sec./°]. The detector swing angle was –30.00°. A second sweep was performed using ω scans from 330.0 to 201.5° in –0.3° step, at χ = 54.7° and φ = 90.0°. The detector swing angle was –30.00°. Another sweep was performed using ω scans from 330.0 to 261° in –0.3° step, at χ = 54.7° and φ = 180.0°. A final sweep was performed using ω scans from 330.0 to 285° in –0.3° step, at χ = 54.7° and φ = 270.0°. The crystal-to-detector distance was 50.00 mm. Of the 11914 reflections that was collected, 2942 reflections were unique. (R_{int} = 0.0200); equivalent reflections were merged. The structure was solved by direct methods (*SHELXS97*) and expanded using Fourier techniques. All non-H atoms were refined anisotropically. All of the C-bound H atoms are observable from difference Fourier map but are all placed at geometrical positions with C-H = 0.93 and 0.96Å for phenyl and methyl H-atoms. All C-bound H-atoms are refined using riding model with U_{iso}(H) = 1.2U_{eq}(Carrier). Both the N- and O-bound H-atoms were located from difference Fourier map and refined isotropically. CCDC 805048 contains the supplementary crystallographic data for this article. [Copies of this information may be obtained free of charge from the Director, CCDC, 12 Union Road, Cambridge, CBZ IEZ, UK. Facsimile (44) 01223 336 033, E-mail: deposit@ccdc.cam.ac.uk or http//www.ccdc.com.ac.uk/deposit].

Microbiology. ***Antibacterial and antifungal screening.*** For the bacterial organisms, both Gram-positive and Gram-negative bacteria were used. Gram-positive and Gram-negative bacteria can be differentiated in the physical appearance of their cell envelopes. The compounds were screened for their *in vitro* antibacterial and antiyeast activities. Antimicrobial activities were determined by the broth microdilution procedures and principles of the Clinical and Laboratory Standards Institute (CLSI) [38, 39]. Minimal inhibitory concentrations for each compound were investigated against standard bacterial strains; *S.aureus* (ATCC 29213), *E.coli* (ATCC 25922), *S.epidermidis* (ATCC 12228), *P.vulgaris* (ATCC 13315) and yeast-like fungi, *C.albicans* (ATCC90028), *C.glabrata* (ATCC 32554). Bacterial and fungal colonies of the test organisms were suspended directly into a small volume of 0.9% saline and further diluted until turbidity matched the Mc Farland Standard no: 0.5 Petri dishes containing Mueller–Hinton agar for bacteria and Sabouraud and Dextrose agar for fungi were impregnated with these microbial suspensions. The stock solutions of the synthesized compounds were prepared in dimethyl sulfoxide (DMSO), which had no effect on the organisms in the concentrations studied. The initial concentration was 200 mg/mL. All of the dilutions were done with distillated water. The concentrations of tested compounds were 100, 50, 25, 12.5, 6.25, 3.125 μg/mL. DMSO was used as negative control. Gentamycin, amikacin, and nystatin were used as reference drugs for Gram-negative antibacterial activity, Gram-positive antibacterial activity, and anti-fungal activity, respectively. All the inoculated plates were incubated at 37°C and results were evaluated after 24 h for bacteria and 48 h for fungi. The lowest concentration of the compounds that prevented visible growth was considered minimal inhibitor concentrations (MICs).

Acknowledgments. Mr. Sohail Saeed is thankful to National Engineering and Scientific Commission, Pakistan for providing the facility of spectroscopic and microanalytical techniques free of cost.

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