Synthesis of some new derivatives of thiazolopyrimidines and hydrolysis of its arylidene derivative

H NAGARAJAIAH^a, IMTIYAZ AHMED M KHAZI^b and NOOR SHAHINA BEGUM^{a,*}

^aDepartment of Chemistry, Bangalore University, Central College Campus, Bangalore 560001, India

^bDepartment of Chemistry, Karnatak University, Dharwad 580003, India

e-mail: noorsb05@gmail.com; noorsb@rediffmail.com

MS received 16 May 2014; revised 12 September 2014; accepted 7 October 2014

Abstract. A new ammonium acetate-assisted, convenient and efficient procedure for the synthesis of arylidene derivatives of thiazolopyrimidine is described. The main advantages of this protocol is that it is economical, short reaction time, commonly available chemicals, and ease of isolation of products. In addition, a new series of thiazole-fused pyrimidines were synthesized and hydrolysis of one of its arylidene derivative studied. All the compounds were characterized by analytical and spectroscopic methods. Further, the structure of hydrolyzed product and two other compounds were confirmed by X-ray crystal structure analysis. The crystal structures are stabilized by intermolecular C-H...O, C-H...N, C-H... π and π ... π weak interactions. The anti-microbial screening was done on the compounds in order to test their anti-bacterial and anti-fungal activities.

Keywords. Thiazolo[3,2-*a*]pyrimidines; hydrolysis of arylidene derivative; crystal structure; anti-microbial activity.

1. Introduction

Heterocycles containing sulphur and nitrogen are of great interest because they constitute an important class of natural and synthetic organic molecules, many of which exhibit useful biological activity. One such heterocyclic class is thiazolopyrimidines. These have generated interest in recent years due to their biological and pharmaceutical activities. These ring systems have shown anti-microbial,^{1–4} anti-malarial,⁵ anti-inflammatory,^{6,7} anti-oxidant,⁶ anti-nociceptive⁷ and anti-tubercular⁸ activity. In addition, they have been reported to possess anti-tumour activity^{9,10} and have also shown to possess anti-parkinsonian and anti-proliferative properties.¹¹

In view of all these observations, we have undertaken the synthesis of some new derivatives of thiazolo [3,2-*a*]pyrimidines. The traditional methods^{7,12–14} to synthesize arylidene derivative involve the use of 1:1 mixture of acetic acid and acetic anhydride in presence of sodium acetate. But this method is not satisfactory with regard to the isolation of the products and also due to the drastic reaction conditions that are employed. To overcome this limitation, we have developed a new protocol for the synthesis of arylidene derivative of thiazolopyrimidines using readily available, inexpensive and environment-friendly catalysts. We have made

use of ammonium acetate under warm condition with ethanol as a solvent medium. It is a convenient and fast protocol, although the synthetic route followed is a conventional one. However, the duration of the reaction is very short, as it goes to completion in 10 min. This is yet another advantage of the present protocol.

In continuation of our work with thiazolopyrimidines, we report herein the synthesis of some new derivatives and a tricyclic thiazolopyrimidine. It has been observed that the hydrolysis of arylidene derivative of thiazolopyrimidine occurs via rupture of imine bond under acidic condition. In this paper, the mechanism for hydrolysis has been proposed. Some of these derivatives exhibit promising anti-microbial activities. We have presented here the crystal structure analysis of the two derivatives. The structure analysis was undertaken to determine the configuration and conformation of the molecule and the orientation of the substituents around the thiazolopyrimidine framework.

2. Experimental

2.1 Materials and methods

All the chemicals were purchased from S D Fine and Spectrochem used as received without further purification. Melting point apparatus was used to determine the melting point in open capillaries and are uncorrected. IR spectra were recorded on Nicolet Impact 400D FT-IR

*For correspondence

spectrophotometer using KBr pellets. Bruker 400 MHz FT-NMR spectrometer was used to record ^1H and ^{13}C NMR in CDCl_3 and $\text{DMSO}-d_6$ with TMS as internal standard. The reactions and purity of the products were monitored by TLC silica gel plates. Mass spectra were recorded on LC-MSD-Trap-SL spectrometer and elemental analyses were carried out using CHNS Elimentar (Vario-micro cube).

2.2 Procedure for the synthesis of compounds 2(a–d)

A mixture of ethyl 3,5-dihydro-7-methyl-3-oxo-5-phenyl-2H-thiazolo[3,2-a]pyrimidine-6-carboxylate (**1a/1d/1e**) (10 mmol), aromatic aldehydes (10 mmol) and catalytic amount of ammonium acetate in ethanol (30 mL) was warmed for 5–10 min with constant stirring. The reaction mixture was allowed to stand at room temperature for 30 min. The solid thus separated was filtered and washed with water and recrystallized from ethanol to afford yellow compound with good yield 82–87%.

2.2a (2Z)-Ethyl 2-benzylidene-3,5-dihydro-7-methyl-3-oxo-5-phenyl-2H-thiazolo[3,2-a]pyrimidine-6-carboxylate (2a): Yellow solid, yield 87%, M.p.: 178–179 °C. IR (KBr) ν cm^{-1} : 2978 (CH), 1711 (C=O), 1608 (C=C), 1484 (C=N), 1169 (C-O). ^1H NMR (300 MHz, CDCl_3) δ ppm: 1.18 (t, $J=7.2$ Hz, 3H, CH_2CH_3), 2.52 (s, 3H, CH_3), 4.11 (q, $J=7.2$ Hz, 2H, CH_2CH_3), 6.22 (s, 1H, C5-H), 7.26–7.45 (m, 10H, ArH). 7.73 (s, 1H, arylidene-H). ^{13}C NMR (300 MHz, CDCl_3) δ ppm: 14.98, 23.42, 55.13, 60.35, 102.08, 108.88, 114.16, 127.41, 128.15, 129.62, 130.96, 132.43, 134.09, 144.51, 154.62, 159.62, 163.14, 165.62, 167.73. %CHNS found (calc): C 68.30 (68.41), H 4.98 (5.13), N 6.93 (6.82), S 7.93 (8.13).

2.2b (2Z)-Methyl 2-(3-cyanobenzylidene)-3,5-dihydro-5-(4-hydroxyphenyl)-7-methyl-3-oxo-2H-thiazolo[3,2-a]pyrimidine-6-carboxylate (2b): Yellow solid, yield 84%, M.p.: 234–235 °C. IR (KBr) ν cm^{-1} : 2987 (CH), 1698 (C=O), 1603 (C=C), 1486 (C=N), 1231 (C-O). ^1H NMR (400 MHz, $\text{DMSO}-d_6$) δ ppm: 2.38 (s, 3H, CH_3), 3.60 (s, 3H, OCH_3), 5.96 (s, 1H, C5-H), 6.95–7.95 (m, 9H, Ar-H), 9.58 (s, 1H, Ar-OH). ^{13}C NMR (400 MHz, $\text{DMSO}-d_6$) δ ppm: 23.31, 54.86, 61.15, 100.14, 122.96, 127.15, 128.4–137.63, 141.04, 145.78, 153.04, 164.99, 165.69, 167.47. Mass (m/z): 432.22 $[\text{M}+\text{H}]^+$. %CHNS found (calc): C 64.03 (64.32), H 3.97 (3.76), N 9.74 (9.63), S 7.43 (7.59).

2.2c 4-((1Z)-(6-(ethoxycarbonyl)-5-(4-hydroxyphenyl)-7-methyl-3-oxo-3H-thiazolo[3,2-a]pyrimidin-2(5H)-ylidene)methyl)benzoic acid (2c): Yellow solid, yield

82%, M.p.: 241–242 °C. IR (KBr) ν cm^{-1} : 2981 (CH), 1698 (C=O), 1599 (C=C), 1491 (C=N), 1215 (C-O). ^1H NMR (400 MHz, CDCl_3) δ ppm: 1.18 (t, $J=7.2$ Hz, 3H, CH_2CH_3), 2.37 (s, 3H, CH_3), 4.09 (q, $J=7.2$ Hz, 2H, CH_2CH_3), 6.19 (s, 1H, C5-H), 7.26–7.74 (m, 9H, Ar-H), 9.62 (s, 1H, OH), 13.10 (s, 1H, COOH). ^{13}C NMR (400 MHz, CDCl_3) δ ppm: 14.76, 23.30, 54.86, 60.09, 100.14, 109.91, 122.96, 127.15–141.04, 145.28, 153.04, 156.04, 164.99, 165.69, 166.23, 167.47. %CHNS found (calc): C 62.06 (62.12), H 4.34 (4.52), N 6.03 (6.31), S 6.90 (6.61).

2.2d (2Z)-methyl 2-(3-fluorobenzylidene)-3,5-dihydro-5-(4-hydroxyphenyl)-7-methyl-3-oxo-2H-thiazolo[3,2-a]pyrimidine-6-carboxylate (2d): Yellow solid, yield 84%, M.p.: 286–287 °C. IR (KBr) ν cm^{-1} : 2976 (CH), 1701 (C=O), 1598 (C=C), 1497 (C=N), 1192 (C-O). ^1H NMR (400 MHz, CDCl_3) δ ppm: 2.36 (s, 3H, CH_3), 3.67 (s, 3H, OCH_3), 6.02 (s, 1H, C5-H), 6.75–7.83 (m, 9H, Ar-H), 9.60 (s, 1H, OH). ^{13}C NMR (400 MHz, CDCl_3) δ ppm: 23.26, 55.44, 60.12, 100.14, 116.29, 122.96, 127.15, 128.4–137.63, 141.04, 145.78, 154.03, 162.31, 164.72, 165.59, 167.42. %CHNS found (calc): C 62.25 (62.42), H 4.04 (4.06), N 6.60 (6.83), S 7.55 (7.59).

2.3 Procedure for the hydrolyzed compound 3

2.3a Method 1: Compound **1a** was refluxed with benzaldehyde in presence of ammonium acetate in acetic acid for 5–6 h with constant stirring. The reaction mixture was allowed to stand at room temperature overnight. The solid thus separated was filtered, washed with water and recrystallized from ethanol to afford pale yellow crystals in good yield (82%).

2.3b Method 2: A mixture of compound **2a** and catalytic amount of conc. HCl in ethanol (100 mL) was refluxed for 5–6 h with constant stirring. The reaction mixture was allowed to stand at room temperature overnight. The solid thus separated was filtered, washed with water and recrystallized from ethanol to afford good quality crystals (yield 80%).

2.3c (Z)-5-Benzylidenethiazolidine-2,4-dione(3): Colourless solid, yield 82%, M.p.: 264–265 °C. IR (KBr) ν cm^{-1} : 3145 (NH), 3037 (CH), 1739 (SCONH), 1691 (CONH), 1606 (C=C), 1336 (C-N). ^1H NMR (300 MHz, $\text{DMSO}-d_6$) δ ppm: 7.42–7.58 (m, 5H, Ar-H), 7.75 (s, 1H, arylidene-H), 12.58 (s, 1H, NH), %CHNS found (calc): C 58.52 (58.63), H 3.44 (3.23), N 6.82 (6.67), S 15.62 (15.67).

2.4 General procedure for the synthesis compounds **4a** and **4b**

A mixture of ethyl 3,5-dihydro-7-methyl-3-oxo-5-substituted phenyl-2*H*-thiazolo[3,2-*a*]pyrimidine-6-carboxylate (5 mmol), morpholine (6 mmol), formalin (1 mL) and acetic acid (1 mL) in methanol (20 mL) was refluxed for 8 h (monitored by TLC). Reaction mixture was diluted with water and extracted with chloroform (3 × 30 mL). The combined chloroform extract was washed with water (3 × 30 mL) and dried over anhydrous sodium sulfate. The solvent was removed under vacuum and the residue was recrystallized from benzene and hexane mixture to yield colorless to yellow granules with good yield 82–87%.

2.4a (2*Z*)-ethyl 3,5-dihydro-7-methyl-2-(morpholinomethyl)-3-oxo-5-phenyl-2*H*-thiazolo[3,2-*a*]pyrimidine-6-carboxylate (4a**):** Pale yellow solid, yield 87%, M.p.: 84–85°C. IR (KBr) ν cm⁻¹: 2982 (CH), 1711 (C=O), 1613 (C=C), 1499 (C=N), 1180 (C-O). ¹H NMR (400 MHz, CDCl₃) δ ppm: 1.16 (t, *J*=7.2 Hz, 3H, CH₂CH₃), 2.38 (s, 3H, CH₃), 2.47 (t, *J*=4.5 Hz, 4H, C₃, C₅-H, morpholine), 3.00 (d, *J*=4.1 Hz, 2H, CH₂-N), 3.36 (t, *J*=4.3 Hz, 1H, S-CH), 3.63 (t, *J*=4.3 Hz, 4H, C₂, C₆-H, morpholine), 4.07 (q, *J*=7.2 Hz, 2H, CH₂CH₃), 5.99 (s, 1H, C5-H), 7.26–7.36 (m, 5H, Ar-H). ¹³C NMR (400 MHz, CDCl₃) δ ppm: 14.06, 22.27, 50.48, 54.11, 55.37, 57.23, 59.19, 60.51, 101.16, 117.23, 127.9–136.32, 140.07, 144.98, 155.13, 161.92, 164.29, 166.01, 167.13. %CHNS found (calc): C 60.70 (60.49), H 6.06 (5.76), N 10.11 (10.33), S 7.72 (7.59).

2.4b (2*Z*)-ethyl 3,5-dihydro-5-(3-methoxyphenyl)-7-methyl-2-(morpholinomethyl)-3-oxo-2*H*-thiazolo[3,2-*a*]pyrimidine-6-carboxylate (4b**):** Pale yellow solid, yield 82%, M.p.: 88–89°C. IR (KBr) ν cm⁻¹: 2996 (CH), 1707 (C=O), 1601 (C=C), 1512 (C=N), 1172 (C-O). ¹H NMR (400 MHz, CDCl₃) δ ppm: 1.17 (t, *J*=7.2 Hz, 3H, -CH₂CH₃), 2.36 (s, 3H, CH₃), 2.47 (t, *J*=4.5 Hz, 4H, C₃, C₅-H, morpholine), 3.08 (d, *J*=4.1 Hz, 2H, CH₂), 3.33 (t, *J*=4.3 Hz, 1H, S-CH), 3.65 (t, *J*=4.3 Hz, 4H, C₂, C₆-H, morpholine), 3.79 (s, 3H, OCH₃), 4.10 (q, *J*=7.2 Hz, 2H, CH₂CH₃), 6.05 (s, 1H, C5-H), 7.13–7.41 (m, 4H, Ar-H). ¹³C NMR (400 MHz, CDCl₃) δ ppm: 13.98, 22.19, 50.43, 54.17, 55.90, 57.71, 59.42, 60.18, 61.41, 100.43, 119.19, 126.36–138.50, 140.57, 144.02, 155.19, 161.92, 163.41, 165.97, 167.11. %CHNS found (calc): C 59.31 (59.23), H 6.11 (6.37), N 9.43 (9.22), S 7.20 (7.47).

2.5 General procedure for the synthesis of compounds **5 (a–d)**

A mixture of compound **1(a–d)** (10 mmol) and DMF–DMA (5 mL) was heated under reflux for 2 h. The reaction mixture was triturated with ethanol to give a solid product that was collected by filtration and crystallized from ethanol to give compounds **5(a–d)** as deep orange crystals with yield of 85–89%.

2.5a 2-Dimethylaminomethylene-7-methyl-3-oxo-5-phenyl-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyrimidine-6-carboxylic acid ethyl ester (5a**):** Orange yellow solid, yield 87%, M.p.: 204–205°C. IR (KBr) ν cm⁻¹: 2983 (CH), 1710 (C=O), 1618 (C=C), 1490 (C=N), 1179 (C-O). ¹H NMR (400 MHz, CDCl₃) δ ppm: 1.17 (t, *J*=7.2 Hz, 3H, CH₂CH₃), 2.47 (s, 3H, CH₃), 3.10 (s, 6H, 2XCH₃), 4.08 (q, *J*=7.2 Hz, 2H, CH₂CH₃), 6.16 (s, 1H, C5-H), 7.19–7.39 (m, 5H, Ar-H), 7.49 (s, 1H, =CH-NMe₂). ¹³C NMR (400 MHz, CDCl₃) δ ppm: 14.14, 23.02, 54.87, 59.99, 86.06, 106.03, 127.82–128.34, 141.71, 145.73, 154.32, 166.05, 166.15. %CHNS found (calc): C 61.44 (61.41), H 5.70 (5.76), N 11.31 (11.33), S 8.63 (8.59).

2.5b 2-Dimethylaminomethylene-5-(3-methoxy-phenyl)-7-methyl-3-oxo-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyrimidine-6-carboxylic acid ethyl ester (5b**):** Orange yellow solid, yield 89%, M.p.: 181–182°C. IR (KBr) ν cm⁻¹: 3033, 2987 (CH), 1714 (C=O), 1620 (C=C), 1499 (C=N), 1173 (C-O). ¹H NMR (400 MHz, CDCl₃) δ ppm: 1.19 (t, *J*=7.2 Hz, 3H, CH₂CH₃), 2.47 (s, 3H, CH₃), 3.11 (s, 6H, 2XCH₃), 3.75 (s, 3H, OCH₃), 4.09 (q, *J*=7.2 Hz, 2H, CH₂CH₃), 6.15 (s, 1H, C5-H), 6.75–7.26 (m, 4H, Ar-H), 7.53 (s, 1H, =CH-NMe₂). ¹³C NMR (400 MHz, CDCl₃) δ ppm: 14.19, 22.93, 54.59, 60.06, 61.32, 86.43, 106.27, 127.85–128.54, 140.36, 141.55, 145.82, 154.41, 165.89, 166.32. Mass (m/z): 402.3 [M+H]⁺. %CHNS found (calc): C 59.83 (60.12), H 5.77 (5.76), N 10.47 (10.33), S 7.99 (7.95).

2.5c 2-Dimethylaminomethylene-5-(4-methoxy-phenyl)-7-methyl-3-oxo-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyrimidine-6-carboxylic acid ethyl ester (5c**):** Orange yellow solid, yield 85%, M.p.: 165–167°C. IR (KBr) ν cm⁻¹: 3035, 2996 (CH), 1711 (C=O), 1600 (C=C), 1498 (C=N), 1215 (C-O). ¹H NMR (400 MHz, CDCl₃) δ ppm: 1.18 (t, *J*=7.2 Hz, 3H, -CH₂CH₃), 2.51 (s, 3H, CH₃), 3.13 (s, 6H, 2XCH₃), 3.75 (s, 3H, -OCH₃), 4.09 (q, *J*=7.2 Hz, 2H, CH₂CH₃), 6.14 (s, 1H, C5-H), 6.78–7.33 (m, 4H, Ar-H), 7.55 (s, 1H, =CH-NMe₂). ¹³C NMR (400 MHz, CDCl₃) δ ppm: 14.03,

23.21, 54.89, 59.93, 61.10, 87.33, 106.05, 127.52, 128.87, 139.59, 141.76, 145.32, 154.19, 166.27, 167.02. %CHNS found (calc): C 59.83 (60.12), H 5.77 (5.76), N 10.47 (10.33), S 7.99 (7.95).

2.5d 2-Dimethylaminomethylene-5-(4-hydroxy-phenyl)-7-methyl-3-oxo-2,3-dihydro-5H-thiazolo[3,2-a]pyrimidine-6-carboxylic acid ethyl ester (5d): Orange yellow solid, yield 87%, M.p.: 280–281°C. IR (KBr) ν cm⁻¹: 3321 (OH), 2979 (CH), 1712 (C=O), 1623 (C=C), 1492 (C=N), 1183 (C-O). ¹H NMR (400 MHz, CDCl₃) δ ppm: 1.21 (t, J =7.2 Hz, 3H, CH₂CH₃), 2.64 (s, 3H, CH₃), 3.22 (s, 6H, 2XCH₃), 4.12 (q, J =7.2 Hz, 2H, CH₂CH₃), 6.13 (s, 1H, C5-H), 6.74–7.09 (m, 4H, Ar-H), 7.59 (s, 1H, =CH-NMe₂), 9.60 (s, 1H, -OH). ¹³C NMR (400 MHz, CDCl₃) δ ppm: 13.90, 23.01, 54.47, 60.06, 86.88, 107.03, 124.72, 127.40, 143.76, 145.19, 154.62, 165.95, 166.31. %CHNS found (calc): C 58.90 (58.76), H 5.46 (5.59), N 10.85 (10.63), S 8.28 (8.31).

2.6 General procedure for the synthesis of compounds 6a and 6b

A mixture of 2-dimethylaminomethylene-7-methyl-3-oxo-5-substituted phenyl-2,3-dihydro-5H-thiazolo[3,2-a]pyrimidine-6-carboxylic acid ethyl ester (5 mmol), 2-amino benzoic acid (5 mmol) and acetic acid (1 mL) in methanol (30 mL) was refluxed for 8 h (monitored by TLC). The excess methanol was removed under reduced pressure. The compound was diluted with water. The solid thus separated was filtered and dried to give yellow compound with 60–63% yield.

2.6a 2-((1Z)-(6-(ethoxycarbonyl)-7-methyl-3-oxo-5-phenyl-3H-thiazolo[3,2-a]pyrimidin-2(5H)-ylidene)methylamino)benzoic acid (6a): Yellow solid, yield 60%, M.p.:244–245°C. IR (KBr) ν cm⁻¹: 3340 (OH), 2994(CH), 1712 (ester C=O), 1690 (COOH), 1510 (C=C), 1042(C-O). ¹H NMR (400 MHz, DMSO-*d*₆) δ ppm: 1.10 (t, J =7.2 Hz, 3H, CH₂CH₃), 2.21 (s,3H, CH₃), 3.99 (q, J =7.2 Hz, 2H, CH₂), 5.72 (s, 1H, C5-H), 6.63–8.03(m, 8H, Ar-H), 9.87 (d,1H, NH), 11.83(s, 1H, COOH). ¹³C NMR (400 MHz, DMSO-*d*₆) δ ppm:14.02, 17.70, 53.72, 59.14, 112.10, 112.35, 114.44–132.25, 145.41, 146.32, 159.61, 167.29. %CHNS found (calc): C 62.19 (62.34), H 4.57 (4.53), N 9.07 (8.88), S 6.92 (6.97).

2.6b 2-(-(6-(ethoxycarbonyl)-5-(3-methoxyphenyl)-7-methyl-3-oxo-3H-thiazolo[3,2-a]pyrimidin-2(5H)-yli-

dene)methylamino)benzoic acid (6b): Yellow solid, yield 63%, M.p.:220–221°C. IR (KBr) ν cm⁻¹:3342 (-OH), 2987 (CH), 1710 (ester C=O), 1691 (COOH), 1503 (C=C), 1052 (C-O). ¹H NMR (400 MHz, DMSO-*d*₆) δ ppm: 1.11 (t, J =7.2 Hz, 3H, CH₂CH₃), 2.23 (s,3H, CH₃), 3.79 (s,3H, OCH₃), 3.99 (q, J =7.2 Hz, 2H, CH₂), 5.73 (s, 1H, C5-H), 6.76–8.05(m, 9H, Ar-H), 9.88 (d, 1H, NH), 11.85 (s, 1H, -COOH). ¹³C NMR (400 MHz, DMSO-*d*₆) δ ppm:14.23, 17.68, 53.70, 55.25, 59.15, 112.10, 112.35, 115.26–134.39, 144.54, 146.30, 159.60, 167.20. %CHNS found (calc): C 60.84 (60.99), H 4.70 (4.63), N 8.51 (8.67), S 6.50 (6.38).

2.7 General procedure for the synthesis of compounds 7 (a–f)

To a solution of **5a/5b** (5 mmol) in ethanol (30 mL) secondary amines like morpholine, piperidine and pyrrolidine (5 mmol) was added and then refluxed for several hours. Completion of the reaction was monitored by TLC. The solid separated was filtered and dried to get the desired product.

2.7a 7-Methyl-2-morpholin-4-ylmethylene-3-oxo-5-phenyl-2,3-dihydro-5H-thiazolo[3,2-a] pyrimidine-6-carboxylic acid ethyl ester (7a): Pale orange yellow solid, yield 80%, M.p.:221–222°C. IR (KBr) ν cm⁻¹:2982 (CH), 1705 (C=O), 1612 (C=C), 1502 (C=N), 1192 (C-O). ¹H NMR (400 MHz, CDCl₃) δ ppm: 1.17 (t, J =7.2 Hz, 3H, CH₂CH₃), 2.50 (s, 3H, CH₃), 3.45 (t, J =4.8 Hz, 4H, CH₂CH₂), 3.73 (t, J =4.8 Hz, 4H, CH₂CH₂), 4.09 (q, J =7.2 Hz, 2H, CH₂CH₃), 6.17 (s, 1H, C5-H), 7.23–7.40 (m, 5H, Ar-H), 7.49 (s, 1H,=CH-N(CH₂)₂). ¹³C NMR (400 MHz, CDCl₃) δ ppm: 14.16, 23.02, 55.57, 57.03, 59.89, 60.15, 100.18, 117.54, 125.79–137.27, 140.04, 144.56, 155.29, 161.09, 164.61, 166.26, 167.01. %CHNS found (calc): C 61.00 (61.10), H 5.61 (5.66), N 10.16 (10.31), S 7.75 (7.93).

2.7b 5-(3-Methoxy-phenyl)-7-methyl-2-morpholin-4-ylmethylene-3-oxo-2,3-dihydro-5H-thia -zolo[3,2-a]pyrimidine-6-carboxylic acid ethyl ester (7b): Pale orange yellow solid, yield 78%, M.p.:214–215°C. IR (KBr) ν cm⁻¹: 3037, 2956 (CH), 1703 (C=O), 1610 (C=C), 1501 (C=N), 1198 (C-O). ¹H NMR (400 MHz, CDCl₃) δ ppm: 1.18 (t, J =7.2 Hz, 3H, CH₂CH₃), 2.51 (s, 3H, CH₃), 3.43 (t, J =4.8 Hz, 4H, CH₂CH₂), 3.73 (t, J =4.8 Hz, 4H, CH₂CH₂), 4.05 (q, J =7.2 Hz, 2H, CH₂CH₃), 6.12 (s, 1H, C5-H), 7.27–7.64 (m, 4H, Ar-H), 7.51 (s, 1H, =CH-N(CH₂)₂). ¹³C NMR (400 MHz, CDCl₃) δ ppm: 14.01, 23.12, 55.47, 57.17, 58.03,

59.67, 60.18, 100.04, 119.21, 125.63–137.37, 140.16, 144.37, 154.48, 160.59, 164.16, 166.66, 167.13. %CHNS found (calc): C 59.58 (59.62), H 9.47 (9.71), N 9.47 (9.38), S 7.23 (7.15).

2.7c *7-Methyl-3-oxo-5-phenyl-2-piperidin-1-ylmethylene-2,3-dihydro-5H-thiazolo[3,2-a]pyrimidine-6-carboxylic acid ethyl ester (7c)*: Pale orange red solid, yield 76%, M.p.: 242–243°C. IR (KBr) ν cm⁻¹: 2974 (CH), 1701 (C=O), 1605 (C=C), 1495 (C=N), 1188 (C-O). ¹H NMR (400 MHz, CDCl₃) δ ppm: 1.28 (t, J =7.2 Hz, 3H, CH₂CH₃), 1.64 (m, 6H, C₃, C₄, C₅-H, piperidine), 2.51 (s, 3H, CH₃), 3.42 (m, 4H, C₂, C₆-H, piperidine), 4.09 (q, J =7.2 Hz, 2H, CH₂CH₃), 6.11 (s, 1H, C₅-H), 6.93–7.25 (m, 5H, Ar-H), 7.52 (s, 1H, =CH-N(CH₂)₂). ¹³C NMR (400 MHz, CDCl₃) δ ppm: 14.16, 23.02, 24.42, 26.31, 54.49, 59.84, 60.30, 100.19, 118.03, 123.79–134.27, 140.44, 144.01, 155.57, 160.69, 164.72, 166.32, 167.11. %CHNS found (calc): C 64.21 (64.23), H 6.12 (6.31), N 10.21 (10.37), S 7.79 (7.61).

2.7d *5-(3-Methoxy-phenyl)-7-methyl-3-oxo-2-piperidin-1-ylmethylene-2,3-dihydro-5H-thiazolo[3,2-a]pyrimidine-6-carboxylic acid ethyl ester (7d)*: Pale orange red solid, yield 73%, M.p.: 237–238°C. IR (KBr) ν cm⁻¹: 2945 (CH), 1700 (C=O), 1611 (C=C), 1493 (C=N), 1196 (C-O). ¹H NMR (400 MHz, CDCl₃) δ ppm: 1.19 (t, J =7.2 Hz, 3H, CH₂CH₃), 1.66 (m, 6H, C₃, C₄, C₅-H, piperidine), 2.49 (s, 3H, CH₃), 3.41 (m, 4H, C₂, C₆-H, piperidine), 3.76 (s, 3H, OCH₃), 4.10 (q, J =7.2 Hz, 2H, CH₂CH₃), 6.16 (s, 1H, C₅-H), 6.76–7.26 (m, 4H, Ar-H), 7.56 (s, 1H, =CH-N(CH₂)₂). ¹³C NMR (400 MHz, CDCl₃) δ ppm: 14.10, 22.96, 24.12, 26.54, 55.55, 57.42, 59.91, 61.10, 101.12, 117.53, 127.12–136.19, 141.19, 144.23, 155.54, 161.03, 163.91, 166.60, 167.33. Mass (m/z): 441.8 [M]⁺. %CHNS found (calc): C 62.56 (62.39), H 6.16 (5.97), N 9.52 (9.38), S 7.26 (7.19).

2.7e *Ethyl 3,5-dihydro-7-methyl-3-oxo-5-phenyl-2-((pyrrolidin-1-yl)methylene)-2H-thiazolo[3,2-a]pyrimidine-6-carboxylate (7e)*: Pale orange yellow solid, yield 75%, M.p.: 262–263°C. IR (KBr) ν cm⁻¹: IR (KBr $\nu_{\max}$, cm⁻¹): 3052, 2976 (CH), 1707 (C=O), 1620 (C=C), 1502 (C=N), 1190 (C-O). ¹H NMR (400 MHz, CDCl₃) δ ppm: 1.22 (t, J =7.2 Hz, 3H, CH₂CH₃), 1.78 (m, 4H, C₃, C₄-H, pyrrolidine), 2.36 (s, 3H, CH₃), 2.65 (m, C₂, C₅-H, pyrrolidine), 4.09 (q, J =7.2 Hz, 2H, CH₂CH₃), 6.07 (s, 1H, C₅-H), 7.19–7.32 (m, 5H, Ar-H), 7.52 (s, 1H, =CH-N(CH₂)₂). ¹³C NMR (400 MHz, CDCl₃) δ ppm: 14.03, 23.10, 25.31, 54.43, 59.86, 61.01, 100.17, 118.33, 123.19–135.42, 139.94, 144.21,

155.37, 160.51, 164.62, 166.17, 167.02. %CHNS found (calc): C 63.45 (63.23), H 5.83 (5.67), N 10.57 (10.61), S 8.07 (7.97).

2.7f *Ethyl 3,5-dihydro-5-(3-methoxyphenyl)-7-methyl-3-oxo-2-((pyrrolidin-1-yl)methylene)-2H-thiazolo[3,2-a]pyrimidine-6-carboxylate (7f)*: Pale orange yellow solid, yield 73%, M.p.: 254–255°C. IR (KBr) ν cm⁻¹: 3048, 2977 (CH), 1703 (C=O), 1618 (C=C), 1506 (C=N), 1197 (C-O). ¹H NMR (400 MHz, CDCl₃) δ ppm: 1.24 (t, J =7.2 Hz, 3H, CH₂CH₃), 1.78 (m, 4H, C₃, C₄-H, pyrrolidine), 2.64 (m, C₂, C₅-H, pyrrolidine), 2.34 (s, 3H, CH₃), 3.79 (s, 3H, OCH₃), 4.11 (q, J =7.2 Hz, 2H, CH₂CH₃), 6.02 (s, 1H, C₅-H), 6.78–7.29 (m, 4H, Ar-H), 7.53 (s, 1H, =CH-N(CH₂)₂). ¹³C NMR (400 MHz, CDCl₃) δ ppm: 14.03, 22.99, 25.51, 55.53, 57.52, 59.71, 61.23, 100.96, 118.01, 129.36–135.49, 141.02, 144.19, 155.37, 161.29, 163.71, 166.32, 167.18. %CHNS found (calc): C 61.81 (61.67), H 5.89 (5.71), N 9.83 (9.92), S 7.50 (7.41).

2.8 Procedure for the synthesis of tricyclic thiazolopyrimidine

2.8a *Procedure for the synthesis of ethyl 3-amino-2-cyano-5-(3-methoxyphenyl)-7-methyl-5H-thiazolo[3,2-a]pyrimidine-6-carboxylate (9)*: To a warm solution of KOH (10 mmol) and pyrimidine derivative (**8**) (10 mmol) in ethanol (30 mL), bromomalononitrile (11 mmol) was added dropwise with constant stirring. The reaction mixture was stirred for 2 h. The solution was allowed to stand at room temperature for some time and the solid product precipitated. The precipitate was filtered and recrystallized from ethanol.

Yellow solid, yield 88%, M.p.: 238–239°C. IR (KBr) ν cm⁻¹: 3373, 3329 (NH₂) 2931 (CH), 2189 (C≡N) 1703 (C=O), 1655 (C=C), 1566 (C=N), 1195 (C-O). ¹H NMR (400 MHz, CDCl₃) δ ppm: 1.25 (t, J = 7.1 Hz, 3H, CH₂CH₃), 2.35 (s, 3H, CH₃), 3.78 (s, 3H, OCH₃), 4.12 (q, J = 7.0 Hz, 2H, CH₂CH₃), 4.75 (s, 2H, NH₂), 6.06 (s, 1H, C₅-H), 6.84–6.94 (m, 4H, Ar-H). ¹³C NMR (400 MHz, CDCl₃) δ ppm: 14.09, 23.06, 54.87, 60.06, 90.43, 102.32, 113.76, 118.42, 129.63, 132.63, 143.19, 155.35, 157.99, 163.16, 164.09, 165.23. %CHNS found (calc): C 58.36 (58.62), H 4.90 (5.06), N 15.12 (15.13), S 8.66 (8.59).

2.8b *Procedure for the synthesis 2-cyano-3-(1-ethoxyethylideneamino)-5-(3-methoxy-phenyl)-7-methyl-5H-thiazolo[3,2-a]pyrimidine-6-carboxylic acid ethyl ester (10)*: A mixture of **9** (1.85 g, 5 mmol) and triethylorthoacetate (2 mL) was heated under reflux in acetic anhydride for 6 h, excess triethylorthoacetate and acetic

anhydride was removed. The residue was treated with petroleum ether. The solid thus separated was filtered, washed and recrystallized with petroleum ether to afford light greenish yellow crystals.

Yellow green solid, yield 83%, M.p.:110–111°C. IR (KBr) ν cm^{-1} : 2977, 2935 (CH), 2224 ($\text{C}\equiv\text{N}$), 1703 ($\text{C}=\text{O}$), 1616 ($\text{C}=\text{C}$), 1541 ($\text{C}=\text{N}$), 1184 ($\text{C}-\text{O}$); ^1H NMR (400 MHz , CDCl_3) δ ppm: 1.21 (t, $J = 7.1$ Hz, 3H, CH_2CH_3), 1.36 (t, $J = 7.1$ Hz, 3H, CH_2CH_3),

1.43(s, 3H, CH_3), 2.00 (s, 3H, CH_3), 2.36 (s, 3H, CH_3), 3.73(s, 3H, OCH_3), 4.08 (q, $J = 7.0$ Hz, 2H, CH_2CH_3), 4.28 (q, $J = 7.0$ Hz, 2H, CH_2), 5.94 (s, 1H, C5-H), 6.76–7.23 (m, 4H, Ar-H). ^{13}C NMR (400 MHz , CDCl_3) δ ppm:14.11, 23.14, 25.08, 54.08, 54.89, 59.46, 90.48, 102.30, 112.89, 113.05, 118.68, 129.63, 143.17, 155.35, 158.97, 159.55, 163.23, 164.17, 165.46. %CHNS found (calc): C 58.96 (58.72), H 5.16 (5.33), N 11.96 (12.01), S 6.84 (7.03).

Code	R	R ₁	R ₂	Code	R	R ₁	R ₂
2a	H	H	Et	2c	4-OH	4-COOH	Et
2b	4-OH	3-CN	Me	2d	4-OH	3-F	Me

Mechanism for hydrolysis

Scheme 1. Synthesis and hydrolysis of arylidene derivatives of thiazolopyrimidine.

2.8c Procedure for the synthesis 7-amino-8-imino-4-(3-methoxy-phenyl)-2,6-dimethyl-7,8-dihydro-4H-9-thia-1,4a,5,7-tetraaza-fluorene-3-carboxylic acid ethyl ester (II): A mixture of **10** (2.2 g, 5 mmol) and hydrazine hydrate (7 mL) was stirred at room temperature for 2 h and then diluted with ethanol (20 mL). The resulting fine solid suspension was filtered, washed with ethanol and purified by recrystallization from ethanol-dioxane mixture to afford analytically (TLC) pure pale yellow granules.

Yellow solid, yield 62%, M.p.: 225–226°C. IR (KBr) ν cm^{-1} : 3361 (NH), 3232, 3190 (NH_2), 2923 (CH), 1697 (C=O), 1652 (C=C), 1492 (C=N), 1230 (C-O). ^1H NMR (400 MHz, $\text{DMSO}-d_6$) δ ppm: 1.15 (t, $J=7.2$ Hz, 3H, CH_2CH_3), 2.29 (s, 3H, CH_3), 2.33 (s, 3H, CH_3), 3.68 (s, 3H, OCH_3), 4.06 (q, $J=7.2$ Hz, 2H, CH_2CH_3), 4.81 (s, 2H, NH_2), 6.42 (s, 1H, C5-H of thiazolopyrimidine), 6.78–7.21 (m, 4H, Ar-H), 8.97 (s, 1H, =NH). ^{13}C NMR (400 MHz, $\text{DMSO}-d_6$) δ ppm: 14.11, 23.14, 25.08, 54.08, 54.89, 59.46, 90.48, 102.30, 112.89, 113.05, 118.68, 129.63, 143.17, 155.35, 158.97, 159.55, 163.23, 164.17, 165.46. %CHNS found (calc): C 56.32 (56.42), H 5.20 (4.99), N 19.70 (19.83), S 7.52 (7.59).

2.9 X-ray diffraction analysis

Good quality single crystals of compounds **3**, **5b** and **7a** were obtained by slow evaporation of ethanol. The X-ray diffraction data, for the compounds **3**, **5b** and **7a** were collected on a Bruker Smart CCD Area Detector System using $\text{MoK}\alpha$ (0.71073 Å) radiation. The data were reduced using SAINT-Plus.¹⁵ The structure was solved by Direct methods using SHELXS97¹⁶ and refined by difference Fourier syntheses using SHELXL97.¹⁶ The positions and anisotropic displacement parameters of all non-hydrogen atoms were included in the full-matrix least-square refinement using SHELXL97¹⁶ and the refinement procedures were carried out until convergence was reached.

The H atoms were placed at calculated positions in the riding model approximation, their temperature factors were set to 1.2 times those of the equivalent isotropic temperature factors of the parent atoms. All other non-H atoms were refined anisotropically. Molecular diagrams were generated using ORTEP.¹⁷ The mean plane calculation was done using the program PARST.¹⁸

2.10 Anti-microbial activity

Anti-microbial activities of the synthesized compounds **2d**, **4a**, **5a**, **5b**, **6a**, **7a**, **7d**, **9** and **10** were screened against the two standard bacterial strains *Staphylococcus aureus*, *E.coli* and two standard fungal strains *Aspergillus niger*, *Penicillium chrysogenum* by Agar diffusion method with 3 different concentrations (0.5, 1.0, 2.0 mg/mL).^{19,20}

For anti-bacterial activity test, media used was peptone 10 g, NaCl 10 g and yeast extract 5 g, agar 20 g in 1000 mL distilled water. Initially, the stock cultures of bacteria were revived by inoculating in broth media and grown at 37°C for 18 h. The agar plates of the above media were prepared and wells were made in the plate. Each plate was inoculated with 18 h old cultures (100 μL , 10^{-4}CFU) and spread evenly on the plate. After 20 min, the wells were filled with compound at different concentrations. The control wells with Gentamycin were also prepared. All the plates were incubated at 37°C for 24 h and the diameter of inhibition zone were noted.

For anti-fungal activities media used was Czapek-Dox Agar: Composition (g/L) Sucrose-30.0; Sodium nitrate-2.0; K_2HPO_4 -1.0, $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ -0.5; KCl-0.5; FeSO_4 -0.01; Agar-20: Initially, the stock cultures of fungi were revived by inoculating in broth media and grown at 27°C for 48 h.

The agar plates of the above media were prepared and wells were made in the plate. Each plate was

Scheme 2. Synthesis of some new derivatives of thiazolopyrimidines.

inoculated with 48 h old cultures ($100\ \mu\text{L}$, 10^4 CFU) and spread evenly on the plate. After 20 min, the wells were filled with different concentrations of samples. The control wells were filled with antibiotic. All the plates were incubated at 27°C for 72 h and the diameter of inhibition zone were noted. The results of the anti-microbial activities are shown in table 3.

3. Result and Discussion

3.1 Chemistry

Compounds were synthesized by following procedure depicted in the schemes 1, 2, 3 and 4. The starting materials 7-methyl-3-oxo-5-(substituted phenyl)-2,3-dihydro-5H-thiazolo[3,2-a]pyrimidine-6-carboxylic acid

Scheme 3. Synthesis of dimethyl amino-methylene derivatives of thiazolopyrimidines.

Scheme 4. Synthesis of tricyclic thiazolopyrimidine.

ethyl esters **1(a–e)** were prepared by treating 6-methyl-4-(substituted phenyl)-2-thioxo-1,2,3,4-tetrahydro-pyrimidine-5-carboxylic acid ethyl esters with chloro acetic acidin dioxane under reflux condition. These active methylene compounds **1a**, **1d** and **1e** were further allowed to react with substituted aromatic aldehydes in presence of ammonium acetate to give arylidene derivatives of thiazolopyrimidine. The ^1H NMR spectra of compounds **2(a–d)** indicate the absence of signals for the CH_2 proton around δ 3.7–3.9 ppm and additional signals in the aromatic region. Acid hydrolysis of arylidene derivative **2a** leads to the formation of thiazolid-2,4-one (**3**) via the rupture of endocyclic $\text{C}=\text{N}$ bond in pyrimidine ring by method **1**. The hydrolyzed component (**3**) was also obtained by an alternative method **2**, which involves *in situ* condensation followed by hydrolysis (scheme 1).

Compounds **1a** and **1b** undergo Mannich reaction with different secondary amines yielding compounds **4a** and **4b**. Further treatment of compounds **1(a–d)** with dimethylformamide–dimethylacetal (DMF–DMA) under reflux condition gave 2-dimethylaminomethylene-7-methyl-3-oxo-5-(substituted phenyl)-2,3-dihydro-5*H*-thiazolo[3,2-*a*]pyrimidine-6-carboxylic acid ethyl esters

5 (a–d). In order to confirm the spectral data, one of the derivative **5b** was subjected to single crystal X-ray diffraction technique. Crystal structure of compound **5b** showed that the product is *Z*-isomer. Treatment of compounds **5a** and **5b** with 2-amino benzoic acid gave the corresponding secondary amine derivative. Reaction of compounds **5a** and **5b** with secondary amines like morpholine, piperidine or pyrrolidine taken in ethanol, on refluxing gave corresponding tertiary amines **7 (a–f)** (scheme 2). In addition to spectral technique, one of the tertiary amine was subjected to single crystal X-ray diffraction technique. Crystal structure of compound **7a** showed the morpholine ring adopts chair conformation.

Compound **8** was synthesized from well-known Biginelli one pot cyclo-condensation reaction.²¹ The reaction of dihydropyrimidine (**8**) with bromomalononitrile in presence of strong base resulted in the formation of compound **9**. Product was confirmed by using spectral techniques. In the IR spectrum of compound **9** there is an appearance of sharp peak at 2189 cm^{-1} characteristic of nitrile group and two sharp peaks at $3300\text{--}3150\text{ cm}^{-1}$ correspond to $-\text{NH}_2$ group and in the ^1H NMR spectrum, appearance of signal at δ 4.75 ppm correspond to $-\text{NH}_2$ confirming ring closure. Compound

Table 1. Crystal data and refinement parameters for compounds **3**, **5b** and **7a**.

	3	5b	7a
Formula	$\text{C}_{10}\text{H}_7\text{NO}_2\text{S}$	$\text{C}_{20}\text{H}_{23}\text{N}_3\text{O}_4\text{S}$	$\text{C}_{21}\text{H}_{23}\text{N}_3\text{O}_4\text{S}$
<i>M</i>	205.23	401.47	413.48
<i>T</i> /K	296(2)	296(2)	296(2)
Crystal system	Monoclinic	Monoclinic	Monoclinic
Space group	$P2_1/c$	$P2_1/c$	$P2_1/c$
<i>a</i> /Å	9.4365(6)	15.4787(17)	12.3736(13)
<i>b</i> /Å	11.5294(8)	9.6946(11)	12.2454(13)
<i>c</i> /Å	8.1876(6)	13.0224(14)	13.0093(13)
β /°	95.5990(10)	90.836(2)	99.458(2)
<i>V</i> /Å ³	886.54(11)	1953.9(4)	1944.4(3)
<i>Z</i> , <i>d</i> _{calcd} /(g cm ^{−3})	4, 1.538	4, 1.365	4, 1.413
μ /mm ^{−1}	0.332	0.198	0.201
<i>F</i> (000)	424	848	872
θ range/°	2.80 to 25.00	1.32 to 27.00	1.67 to 27.00
Index ranges	−11 ≤ <i>h</i> ≤ 11 −13 ≤ <i>k</i> ≤ 13 −9 ≤ <i>l</i> ≤ 7	−15 ≤ <i>h</i> ≤ 19 −12 ≤ <i>k</i> ≤ 12 −15 ≤ <i>l</i> ≤ 16	−13 ≤ <i>h</i> ≤ 15 −14 ≤ <i>k</i> ≤ 15 −16 ≤ <i>l</i> ≤ 8
Reflections collected	4506	11280	11413
Independent reflections	1554 [<i>R</i> _{int} = 0.0214]	4209 [<i>R</i> _{int} = 0.0386]	4197 [<i>R</i> _{int} = 0.0356]
Completeness	99.4%	98.5 %	98.8%
Data/restraints/parameters	1554 /0/127	4209/0/258	4197/0/264
Goodness-of-fit on <i>F</i> ²	0.940	1.499	0.949
<i>R</i> ₁ , <i>wR</i> ₂ [<i>I</i> > 2σ(<i>I</i>)]	0.0347, 0.0998	0.0671, 0.1763	0.0619, 0.1644
<i>R</i> ₁ , <i>wR</i> ₂ (all data)	0.0369, 0.1059	0.0966, 0.2502	0.0865, 0.2418
Largest diff. peak, hole/(e Å ^{−3})	0.266, −0.363	0.947, −0.774	0.751, −0.761

9 and triethylorthoacetate was refluxed in acetic anhydride to give compound **10**. IR spectrum shows the absence of peaks around $3300\text{--}3150\text{ cm}^{-1}$ confirming the absence of -NH_2 group and from the ^1H NMR spectrum appearance of signals at δ 1.36, 1.45 and 4.28 ppm corresponds to additional -CH_3 , -CH_3 and -CH_2 groups respectively. Further treatment of compound **10** with hydrazine hydrate in ethanol gave tricyclic thiazolopyrimidine **11**. The ring system obtained was confirmed by IR spectrum. The absence of peak at 2189 cm^{-1} indicates that there is nonitrile group, the appearance of peak at 3361 and peaks at 3232 and 3190 cm^{-1} corresponds to -NH and -NH_2 group.

3.2 Crystallography

The details of crystal data and refinements are given in table 1 for the compounds **3**, **5b** and **7a**. Tables **2a** and **2b** give respective hydrogen bond interactions. The ORTEP view of the molecule with atomic labeling (thermal ellipsoids drawn at 50% probability) of **3**, **5b** and **7a** are shown in figures 1, 2a and 2b respectively. Figures 3a, 3b and 3c show the packing of molecules in the crystal structure.

Compounds **5b** and **7a** crystallize in the monoclinic space group containing one molecule each in the asymmetric unit. Both the compounds have almost similar substituents at C2, C3, C5, C6 and C7. In compounds **5b** and **7a**, the aryl ring at C5 is positioned axially to the pyrimidine ring²² with a dihedral angle of $89.88(8)$ and $89.66(8)^\circ$ respectively. Aryl ring in both the compounds is projecting away from the plane of the thiazolopyrimidine ring. The ethyl carboxylate group at C6 in compound **7a** is planar with respect to the thiazolopyrimidine ring whereas in compound **5b**

Figure 1. ORTEP diagram for the hydrolyzed compound **3**.

Figure 2. (a) ORTEP view of compound **5b**, showing 50% probability ellipsoids and the atom numbering scheme. (b) ORTEP view of compound **7a**, showing 50% probability ellipsoids and the atom numbering scheme.

it slightly deviates from the mean plane of thiazolopyrimidine with dihedral angle of $17.77(8)^\circ$ this may be attributed to the presence of bulky group at C5. The central pyrimidine ring in compounds **5b** and **7a** adopt twist boat conformation²² due to the deviation of C5 and N2 atom from the mean plane C6/C7/C9/N1 constituting ring by $-0.295/-0.297(3)$ and $-0.052/-0.066(3)\text{ \AA}$ respectively. The two values separated by '/' correspond to two different compounds *i.e.* **5b** and **7a**. In compound **5b**, dimethylaminomethylene group adopts *Z* configuration. The substituted morpholine ring at C17 of compound **7a** adopts chair conformation with the atoms N3 and O4 displaced from the mean plane of the remaining ring atoms (C18/C19/C20/C21) by $0.6863(2)$ and $-0.5735(2)\text{ \AA}$, respectively.

The crystal structure of the compound **5b** is stabilized by $\text{C-H}\cdots\text{O}$ and $\text{C-H}\cdots\text{N}$ interactions. There are two types of $\text{C-H}\cdots\text{O}$ interactions, the former C8-H8A $\cdots$ O4 interaction running along crystallographic 'b' axis, whereas the later C12-H12 $\cdots$ O1 interaction

running along crystallographic 'c' axis. Both resulted in two dimensional networks. The C-H...N interaction generates bifurcated bonds from two donors, C17 and C18, to the same acceptor N2 along 'c' axis. The crystal structure is further stabilized by C4-H4B...Cg1 (Cg1 is the centroid of aryl ring C11–C16) interaction with a distance of 2.842 Å (table 2a).

The crystal structure of the compound **7a** is also stabilized by C-H...O and C-H...N interactions. As in

the case of compound **5b** there are two types of C-H...O interactions. Here one of the C-H...O interaction generates bifurcated bonds from two donors, C4 and C14, to the same acceptor O4 along 'a' axis. The other two interactions C-H...O and C-H...N are resulting in a helical pattern running along the 'b' axis (figure 3c). The crystal structure is also stabilized by C19-H19B...Cg2 (Cg2 is the centroid of aryl ring C11–C16) interaction with a distance of 2.940 Å

Figure 3. (a) Packing of the compound **5b** showing intermolecular C-H...O and C-H...N interactions. (b) Packing of the compound **7a** showing intermolecular bifurcated C-H...O interactions. (c) Packing of the compound **7a** showing intermolecular C-H...O and C-H...N interactions.

Table 2a. C-H... π interaction and possible hydrogen bond distances and angles (Å, °) for compound **5b** (*D*-donor; *A*-acceptor; *H*-hydrogen).

D—H...A	D—H	H...A	D...A	D—H...A
C8-H8A...O4 ⁱ	0.970	2.428	3.302(4)	150
C12-H12...O1 ⁱⁱ	0.930	2.537	3.288(4)	138
C18-H18A...N2 ⁱⁱⁱ	0.970	2.725	3.639(5)	159
C17-H17...N2 ^{iv}	0.930	2.554	3.470(4)	168
C4-H4B...Cg1 ^v	0.960	2.842	3.733(4)	155

Symmetry code: (i) $x, +y - 1, +z$ (ii) $x, -y + 1/2, +z + 1/2$ (iii) $x, -y + 1/2, +z - 1/2$ (iv) $x, -y + 1/2, +z - 1/2$ (v) $x, 1/2 - y, z - 1/2$

Table 2b. C-H... π interaction and possible hydrogen bond distances and angles (Å, °) for compound **7a** (*D*-donor; *A*-acceptor; *H*-hydrogen).

D—H...A	D—H	H...A	D...A	D—H...A
C21-H21A...O1 ⁱ	0.970	2.670	3.499(4)	144
C14-H14A...O4 ⁱⁱ	0.930	2.566	3.477(4)	166
C18-H18B...N2 ⁱⁱⁱ	0.970	2.684	3.653(4)	177
C4-H4A...O4 ⁱⁱⁱ	0.960	2.577	3.534(4)	174
C19-H19B...Cg2 ^{iv}	0.969	2.940	3.715(4)	137

Symmetry code: (i) $-x + 1, +y - 1/2, -z + 1/2$ (ii) $x + 1, +y, +z$ (iii) $-x + 1, +y + 1/2, -z + 1/2$ (iv) $1 - x, 1 - y, 1 - z$

Table 3. Inhibition zone diameter (mm) of synthesized compounds against tested bacterial strains by Agar diffusion method.

Compounds Sample concentration	Bacteria						Fungi					
	<i>E. coli</i>			<i>S. aureus</i>			<i>A. niger</i>			<i>P. crysogenium</i>		
	0.5 mg	1 mg	2 mg	0.5 mg	1 mg	2 mg	0.5 mg	1 mg	2 mg	0.5 mg	1 mg	2 mg
2d	-	-	-	-	-	-	-	-	-	-	-	-
4a	7	10	17	4	5	7	-	-	-	-	-	-
5a	-	-	-	-	-	-	-	-	-	-	-	-
5b	-	-	-	-	-	-	-	-	-	-	-	-
6a	0	3	5	0	2	3	-	-	-	-	-	-
7a	-	-	-	-	-	-	0	1	3	-	-	-
7d	-	-	-	-	-	-	-	-	-	-	-	-
9	-	-	-	-	-	-	-	-	-	-	-	-
10	-	-	-	0	2	5	-	-	-	-	-	-
	200 μ g	400 μ g	800 μ g	200 μ g	400 μ g	800 μ g	200 μ g	400 μ g	800 μ g	200 μ g	400 μ g	800 μ g
Gentamicin	2.6	2.8	3.1	2.5	2.7	3.4	ND	ND	ND	ND	ND	ND
Amphotericin	ND	ND	ND	ND	ND	ND	3	5	7	0	3	5

Note: “-” Not Active, “ND” – Not Determined.

(table 2b). Finally, supramolecular assembly is further consolidated by π – π stacking interactions between the thiazolopyrimidine rings. This interaction is not observed in the case of compound **5b**.

3.3 Anti-microbial evaluation

The anti-microbial screening was done using agar diffusion method using different strains of bacteria and fungi. The results with the test compounds **2d**, **4a**, **5a**, **5b**, **6a**, **7a**, **7d**, **9** and **10** reveal that only few compounds exhibit antimicrobial activity. Gentamicin and Amphotericin were used as reference to evaluate the potency of tested compounds. Compounds **6a**, **7a** and **10** showed moderate anti-microbial activity, while compound **4a** showed very good activity even at low concentration. Compounds **2d**, **5a**, **5b**, **7d** and **9** did not show any activity against the tested microorganisms. The result of anti-microbial activity is tabulated in table 3.

4. Conclusion

This work reports the synthesis of a new series of thiazolopyrimidines. We have developed a new and convenient synthetic method for the preparation of arylidene derivatives of thiazolopyrimidine. During our studies with thiazolopyrimidines, we have also noticed that hydrolysis of arylidene derivative of thiazolopyrimidine via rupture of endocyclic C=N bonds in pyrimidine ring under acidic condition gives the same product as that obtained by *in situ* condensation followed by hydrolysis (scheme 1, method 1), for which a mechanism has been proposed. The structures of the products were confirmed by spectral techniques. In

addition, crystal structures of two of the derivatives that yielded good quality crystals were verified by single crystal X-ray diffraction. Crystal structures reveal that compounds **5b** and **7a** are Z-isomers. Crystal structure of compound **7a** showed the substituted morpholine ring adopts chair conformation. In both the derivatives, the ester at C6 adopts *cis* configuration with respect to C6-C7 double bond, which is a prerequisite for the molecule to exhibit calcium channel modulatory activity.²³ On screening all the compounds, only compounds **4a**, **6a**, **7a**, **9** and **10** exhibited good anti-microbial activities at very low concentration of 0.5–2.0 mg/mL against *Aspergillus niger*, *Penicillium chrysogenum*, *Staphylococcus aureus* and *Escherichia coli*. using Gentamicin and Amphotericin as standards.

Supplementary Information

The CIF files are deposited at the Cambridge Crystallographic Data Centre. The deposition number of compounds **3**, **5b** and **7a** are CCDC-966410, CCDC-975896 and CCDC-975899, respectively.

Acknowledgements

NSB and HN thank the University Grants Commission, New Delhi, India for the financial assistance.

References

1. Abu-Hashem A A, Gouda MA and Badria FA 2010 *Eur. J. Med. Chem.* **45** 1976
2. Youssef M S K, Ahmed R A, Abbady M S, Abdel-Mohsen S A and Omar A A 2008 *Monatsh Chem.* **139** 553

3. Sayed H H, Morsy E M H and Kotb E R 2010 *Synth. Commun.* **40** 2712
4. Yaragatti N B, Kulkarni M V, Ghate M D, Hebbar S S and Hegde G R 2010 *J. Sulfur Chem.* **31** 123
5. Fatima S, Sharma A, Saxena R, Tripathi R, Shukla S K, Pandey S K, Tripathi R and Tripathi R P 2012 *Eur. J. Med. Chem.* **55** 195
6. Abu-Hashem A A, Youssef M M and Hussein H A R 2011 *J. Chin. Chem. Soc.* **58** 41
7. Alam O, Khan S A and Ahsan N S W 2010 *Med. Chem. Res.* **19** 1245
8. Jean Kumar V U, Poyraz O, Saxena S, Schnell R, Yogeewari P, Schneider G and Sriram D 2013 *Bioorg. Med. Chem. Lett.* **23** 1182
9. Mohamed A M, Amr A E, Alsharari M A, Al-Qalawi H R M, Germoush M O and Al-Omar M A 2011 *Am. J. Biochem. & Biotech.* **7** 43
10. Fatmah A M, Al-Omary, Hassan G S, El-Messery S M and El-Subbagh H I 2012 *Eur. J. Med. Chem.* **47** 65
11. Amr A E E, Maigali S S and Abdulla M M 2008 *Monatsh Chem.* **139** 1409
12. Flefel E E, Salama M A and Shahat M E 2007 *Phosphorus, Sulfur Silicon Relat. Elem.* **182** 1739
13. Hu J, Wang Y, Wei X, Wu X, Chen G, Cao G, Shen X, Zhang X, Tang Q, Liang G and Li X 2013 *Eur. J. Med. Chem.* **64** 292
14. Kotaiah Y, Hari Krishna N, Naga Raju K, Rao C V, Jonnalagadda S B, Maddila S 2012 *J. Korean. Chem. Soc.* **56** 68
15. Bruker. SMART. SAINT-Plus. SADABS. Bruker AXS Inc. Madison, 1998
16. Sheldrick G M 2008 *Acta. Cryst. A* **64** 112
17. Farrugia L J, ORTEP-3. 1999 *J. Appl. Cryst.* **32** 837
18. Nardelli M 1983 *Acta Cryst. C* **39** 1141
19. Threlfall E J, Fisher I S T, Ward L, Tschape H and Gerner-Smidt P 1999 *Microb. Drug Resist.* **5** 195
20. Walker R D 2000 In *Antimicrobial Therapy in Veterinary Medicine* J F Prescott, J D Baggot and R D Walker (Eds.) (Ames, IA: Iowa State University Press) p. 12
21. (a) Biginelli P 1891 *Ber.* **24** 1317; (b) Biginelli P 1893 *Ber.* **26** 447; (c) Biginelli P 1893 *Gazz. Chim. Ital.* **23** 360
22. (a) Nagarajaiah H, Khazi I M and Begum N S 2012 *J. Chem. Sci.* **124** 847; (b) Nagarajaiah H and Begum N S 2012 *Acta Cryst. E* **68** o2878; (c) Nagarajaiah H, Fathima N and Begum N S 2012 *Acta Cryst. E* **68** o1257; (d) Nagarajaiah H and Begum N S 2011 *Acta Cryst. E* **67** o3444
23. (a) Rovnyak G C, Kimball S D, Beyer B, Cucinotta G, DiMarco J D, Gougoutas J, Hedberg A, Malley M, McCarthy J P, Zhang R and Moreland S 1995 *J. Med. Chem.* **38** 119; (b) Triggie D J and Padmanabhan S 1995 *Chemtracts: Org. Chem.* **8** 191