

# Behaviour of 4-[4-methoxy-3-methylphenyl]-4-oxobutenoic acid towards nitrogen-containing nucleophiles

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**Abstract.** A series of novel amino acid derivatives has been synthesized by the reaction of 4-[4-methoxy-3-methylphenyl]-4-oxobutenoic acid with primary and secondary amines. The treatment of amino acids with hydrazine afforded pyridazine. Phenylhydrazone was obtained from the reaction of the acid with phenyl hydrazine in ethanol. On the other hand, the acid underwent heterocyclization upon the treatment with 2-aminopyridine, *o*-phenylenediamine, aryldithiocarbamates and thiourea derivatives to give the corresponding pyridopyrimidine, quinoxalone, 2-thioxo-1,3-thiazole and 4-hydroxy-1,3-thiazole, respectively. The thiazolopyridazine derivatives were obtained from the reaction of 4-hydroxy-1,3-thiazole with hydrazine and phenylhydrazine, respectively. The behaviour of the 4-hydroxy-1,3-thiazole toward acetic anhydride and bromine was also studied. The proposed structures of the products were based on microanalytical and spectroscopic data. Some of the synthesized compounds also exhibited anti-microbial activities.

**Keywords.**  $\beta$ -aroylacrylic acid; amino acid; pyridazine; quinoxalone; thiazole; thiazolopyridazine; Aza-Michael addition

## 1. Introduction

It was found that  $\beta$ -Aroylacrylic acids and their derivatives showed effective biological activities including antibacterial activity,<sup>1–4</sup> anti-ulcer and cytoprotective properties,<sup>5</sup> kynurenine-3-hydroxylase,<sup>6</sup> and antiproliferative activity toward Human cervix carcinoma (HeLa).<sup>7,8</sup> Besides their broad spectrum of biological activity,  $\beta$ -aroylacrylic acids are versatile and convenient precursors which have been extensively utilized in the synthesis of a wide variety of heterocyclic compounds.<sup>9–12</sup> The  $\alpha$ ,  $\beta$ -unsaturated carbonyl moiety of these compounds is a favorable unit for addition of different nucleophiles which occurs exclusively at the olefin electrophilic centre of the molecule and usually results in the formation of various nitrogen-containing heterocyclic compounds of different ring sizes with one or more heteroatom.

In this regard, the present work deals with the reactivity of 4-[4-methoxy-3-methylphenyl]-4-oxobutenoic acid towards some nucleophilic reagents in order to synthesize a novel series of heterocyclic compounds such as quinaxoline, thiazole, thiazolopyridazine, pyridazine bearing 4-methoxy-3-methylphenyl moiety for biological evaluation.

## 2. Experimental

All melting points reported are uncorrected and determined by the open capillary tube method on a Gallen Kamp melting point apparatus. <sup>1</sup>H-NMR spectra were measured on Bruker (200 MHz) with DMSO-d<sub>6</sub> as the solvent and TMS was used as internal standard (chemical shifts are expressed as  $\delta$ , ppm), IR spectra were recorded on a Perking Elmer 1430 ratio recording infrared spectrophotometer with CDS data station using KBr Wafer technique and Mass spectra were measured on a GC-MSQP 1000EX Shimadzu at microanalytical laboratory, Cairo University, Cairo, Egypt.

### 2.1 General procedure for the synthesis of 2-substitutedamino-4-[4-methoxy-3-methylphenyl]-4-oxobutanoic acids (2a-e)

To a stirred solution of 4-[4-methoxy-3-methyl]-4-oxobutenoic acid (**1**) (0.01 mol) in benzene (40 mL) was added the appropriate amines, namely benzyl amine; cyclohexyl amine; morpholine; or/ and piperidine (0.01 mol). The reaction mixture was stirred at room temperature for 2 hrs., then kept overnight, the resultant precipitate was filtered off, washed with

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benzene and crystallized from suitable solvent to give (**2a-e**) as white solid.

**2.1a 2-benzylamino-4-[4-methoxy-3-methylphenyl]-4-oxobutanoic acid (2a):** From benzene/ethanol; M.p. 170°C; Anal. Calcd. for C<sub>19</sub>H<sub>21</sub>NO<sub>4</sub> (327.38): C, 69.71; H, 6.47; N, 4.28. Found: C, 70.00; H, 6.67; N, 4.40; IR (KBr,  $\nu$ , cm<sup>-1</sup>): 3436 (OH), 1668 (C=O). <sup>1</sup>H-NMR (DMSO-*d*<sub>6</sub>,  $\delta$ /ppm): 2.18 (3H, s, CH<sub>3</sub>Ar), 3.31–3.36 (2H, m, CH<sub>2</sub>-CH), 3.61–3.64 (H, m, CH-CH<sub>2</sub>), 3.86 (3H, s, OCH<sub>3</sub>), 3.88 (2H, s, N-CH<sub>2</sub>Ph), 6.65–7.85 (10H, m, Ar-H, NH and OH); MS (*m/z*, 327 (0.03%)) (M<sup>+</sup>).

**2.1b 2-cyclohexylamino-4-[4-methoxy-3-methylphenyl]-4-oxobutanoic acid (2b):** From toluene, M.p. 180°C; Anal. Calcd. for C<sub>18</sub>H<sub>25</sub>NO<sub>4</sub> (319.40): C, 67.69; H, 7.89; N, 4.39. Found: C, 67.72; H, 7.75; N, 4.51; IR (KBr,  $\nu$ , cm<sup>-1</sup>): 3438 (OH), 3337 (NH), 2929 (CH-aliph.), 1675 (C=O); <sup>1</sup>H-NMR (DMSO-*d*<sub>6</sub>,  $\delta$  ppm): 1.07–1.69 (11H, m, cyclohexyl group), 2.20 (3H, s, CH<sub>3</sub>Ar), 2.86–2.90 (2H, m, CH<sub>2</sub>-CH), 3.36–3.40 (H, m, CH-CH<sub>2</sub>), 3.88 (3H, s, OCH<sub>3</sub>), 6.64–7.86 (5H, m, Ar-H, NH and OH).

**2.1c 2-(2-hydroxyethylamino)-4-[4-methoxy-3-methylphenyl]-4-oxobutanoic acid (2c):** From butanol, M.p. 172°C. Anal. Calcd. for C<sub>14</sub>H<sub>19</sub>NO<sub>5</sub> (281.30): C, 59.78; H, 6.81; N, 4.98. Found: C, 59.80; H, 7.03; N, 5.10; IR (KBr,  $\nu$ , cm<sup>-1</sup>): 3275 (NH), 2958 (CH-aliph), 1670 (C=O); <sup>1</sup>H-NMR (DMSO-*d*<sub>6</sub>,  $\delta$  ppm): 2.16 (3H, s, CH<sub>3</sub>Ar), 2.83–2.98 (4H, 2t, CH<sub>2</sub>-CH<sub>2</sub>), 3.41–3.47 (2H, m, CH<sub>2</sub>-CH), 3.56–3.69 (H, m, CH-CH<sub>2</sub>), 3.88 (3H, s, OCH<sub>3</sub>), 6.63–7.86 (6H, m, Ar-H, NH and OH).

**2.1d 4-[4-methoxy-3-methylphenyl]-2-piperidino-4-oxobutanoic acid (2d):** From ethanol, M.p. 156°C. Anal. Calcd. for C<sub>17</sub>H<sub>23</sub>NO<sub>4</sub> (305.37): C, 66.86; H, 7.59; N, 4.59. Found: C, 66.95; H, 7.78; N, 4.38; IR (KBr,  $\nu$ , cm<sup>-1</sup>): 3437 (OH), 2941 (CH-aliph.), 1675 (C=O); <sup>1</sup>H-NMR (DMSO-*d*<sub>6</sub>,  $\delta$  ppm): 1.37–1.63 (6H, m, piperidine), 2.19 (3H, s, CH<sub>3</sub>Ar), 2.48–2.77 (4H, m, piperidine), 2.93–2.96 (2H, m, CH<sub>2</sub>-CH), 3.71–3.77 (H, m, CH-CH<sub>2</sub>), 3.88 (3H, s, OCH<sub>3</sub>), 6.65–7.85 (5H, m, Ar-H and OH); MS (*m/z*, 305 (5.7%)) (M<sup>+</sup>).

**2.1e 4-[4-methoxy-3-methylphenyl]-2-morpholino-4-oxobutanoic acid (2e):** From toluene, M.p. 172°C. Anal. Calcd. for C<sub>16</sub>H<sub>21</sub>NO<sub>5</sub> (307.34): C, 62.53; H, 6.89; N, 4.56. Found: C, 62.70; H, 7.02; N, 4.71; IR

(KBr,  $\nu$ , cm<sup>-1</sup>): 3433 (OH), 2973 (CH-aliph), 1675 (C=O); <sup>1</sup>H-NMR (DMSO-*d*<sub>6</sub>,  $\delta$  ppm): 2.19 (3H, s, CH<sub>3</sub>Ar), 2.34–2.83 (2H, m, CH<sub>2</sub>-CH), 3.04–3.10 (H, m, CH-CH<sub>2</sub>), 2.43–2.51 and 3.49–3.87 (8H, 2m, morpholin), 3.89 (3H, s, OCH<sub>3</sub>), 7.02–7.89 (4H, m, Ar-H and OH); MS (*m/z*, 306 (16.4%)) (M<sup>+</sup>-1).

## 2.2 Formation of pyridazinone (3)

Hydrazine hydrate 80% (0.015 mol) was added to a solution of 2-substituted amino-butanoic acids (2a) or (2b) or (2c) or (2d) (0.01 mol) in ethanol (30 mL) and the reaction mixture was heated under reflux for 6 hrs., after concentration and cooling, the product obtained was collected and crystallized from ethanol to give pyridazinone (**3**) as white crystals with M.p. 142°C. Anal. Calcd. for C<sub>12</sub>H<sub>12</sub>N<sub>2</sub>O<sub>2</sub> (216.23): C, 66.65; H, 5.59; N, 12.95. Found: C, 66.83; H, 5.79; N, 12.87; IR (KBr,  $\nu$ , cm<sup>-1</sup>): 3443 (NH) or (OH), 1668 (C=O); <sup>1</sup>H-NMR (CDCl<sub>3</sub> - *d*<sub>6</sub>,  $\delta$  ppm): 2.28 (3H, s, CH<sub>3</sub>), 3.89 (3H, s, CH<sub>3</sub>O), 6.88–6.91 (1H, d, H-5 pyridazinone, *J* = 8.23 Hz), 7.01–7.05 (1H, d, H-6 benzene ring, *J* = 9.85 Hz), 7.26 (1H, s, H-2 benzene ring), 7.55–7.56 (1H, d, H-5 benzene ring, *J* = 9.80 Hz), 7.71–7.75 (1H, d, H-4 pyridazinone, *J* = 9.75 Hz), 10.9 (1H, broad, N-H pyridazine); MS (*m/z*, 216 (100%)) (M<sup>+</sup>).

## 2.3 Synthesis of 4-[4-methoxy-3-methylphenyl]-4-oxobutenoic acid phenyl hydrazone (4)

To a solution of the acid (**1**) (0.01 mol) in ethanol (20 mL), phenyl hydrazine (0.01 mol) was added and the reaction mixture was heated under water bath for 1/2 hr., the solid product separated after cooling was filtered off and crystallized from ethanol to give orange crystals (**4**) M.p. 209°C. Anal. Calcd. for C<sub>18</sub>H<sub>18</sub>N<sub>2</sub>O<sub>3</sub>(310.35): C, 69.66; H, 5.85; N, 9.03. Found: C, 69.83; H, 5.90; N, 9.14; IR (KBr,  $\nu$ , cm<sup>-1</sup>): 3587 (OH), 3294 (NH), 1679 (C=O); <sup>1</sup>H-NMR (DMSO-*d*<sub>6</sub>,  $\delta$  ppm): 2.21 (3H, s, CH<sub>3</sub>Ar), 3.87 (3H, s, OCH<sub>3</sub>), 5.29–5.34 (H, d, CH=CH, *J* = 15.9 Hz), 6.82–7.46 (9H, m, Ar-H and CH=CH), 9.35 (H, s, NH), 12.14 (H, s, OH); MS (*m/z*, 310 (19.6%)) (M<sup>+</sup>).

## 2.4 Synthesis of 4-[6-chlorobenzothiazol-2-ylimino]-4-[4-methoxy-3-methylphenyl]-4-oxobutenoic (5)

A mixture of the acid (0.01 mol) and 6-chloro-2-aminobenzothiazol (0.01 mol) in ethanol (20 mL) was refluxed for 8 hrs. The solid product separated after cooling was filtered off and crystallized from

ethanol to give the compound (**5**). Anal. Calcd. for  $C_{19}H_{15}ClN_2O_3S$  (386.85): C, 58.99; H, 3.91; N, 7.24. Found: C, 59.02; H, 3.77; N, 7.31. IR (KBr,  $\nu$ ,  $cm^{-1}$ ): 3300 (OH), 3172 (NH), 1652 (C=O);  $^1H$ -NMR (DMSO- $d_6$ ,  $\delta$  ppm): 2.18 (3H, s,  $CH_3$ Ar), 3.89 (3H, s, OCH<sub>3</sub>), 6.62–7.97 (9H, m, Ar-H, CH=CH and OH).

### 2.5 Synthesis of 2-[4-methoxy-3-methylphenyl]-4H-pyrido[1,2-a]pyrimidine-4-carboxylic acid (**6**)

A mixture of the acid (**1**) (0.01 mol) and 2-aminopyridine (0.01 mol) in ethanol (40 mL) was refluxed for 2 hrs., the product separated after cooling was filtered off, washed with cold ethanol and crystallized from ethanol to give yellow crystals of (**6**) with M.p. 145°C. Anal. Calcd. for  $C_{17}H_{16}N_2O_3$  (296.32): C, 68.91; H, 5.44; N, 9.45. Found: C, 68.85; H, 5.53; N, 9.52; IR (KBr,  $\nu$ ,  $cm^{-1}$ ): 3451 (OH), 1649 (C=O);  $^1H$ -NMR (DMSO- $d_6$ ,  $\delta$  ppm): 2.14 (3H, s,  $CH_3$ Ar), 3.77 (3H, s, OCH<sub>3</sub>), 4.82–4.83 (H, d, CH-COOH), 6.17–7.55 (8H, m, Ar-H); MS ( $m/z$ , 252 (51.9%)) ( $M^+ - CO_2$ ).

### 2.6 Synthesis of 3,4-dihydro-3-(2-(4-methoxy-3-methylphenyl)-2-oxoethyl)-1H-quinoxalin-2-one (**7**)

A mixture of *o*-phenylenediamine (0.005 mol) and the acid (**1**) (0.005 mol) in ethanol (15 mL) was heated under reflux for 6 h. The solid product obtained after cooling was filtered off and crystallized from ethanol to give the product (**7**) with M.p. 180°C. Anal. Calcd. for  $C_{18}H_{18}N_2O_3$  (310.35): C, 69.66; H, 5.85; N, 9.03. Found: C, 69.66; H, 5.90; N, 8.91; IR (KBr,  $\nu$ ,  $cm^{-1}$ ): 3369 (NH), 3196 (NH), 1688 (C=O), 1663 (C=O);  $^1H$ -NMR (DMSO- $d_6$ ,  $\delta$  ppm): 2.20 (3H, s,  $CH_3$ Ar), 3.47–3.45 (2H, m, CH<sub>2</sub>-CH), 3.89 (3H, s, OCH<sub>3</sub>), 4.30–4.38 (H, m, CH-CH<sub>2</sub>), 2.63 (H, s, NH), 6.60–7.89 (7H, m, Ar-H), 10.28 (H, s, NH); MS ( $m/z$ , 310 (6.29%)) ( $M^+$ ).

### 2.7 General procedure for the synthesis of 3-Aryl-5-[2-(4-methoxy-3-methylphenyl)-2-oxoethyl]-2-thioxo-1,3-thiazolidin-4-ones (**8a-c**)

To a stirred solution of the acid (**1**) (0.005 mol) in EtOH (10 mL), ammonium N-aryldithiocarbamate (0.005 mol), namely ammonium N-benzyl dithiocarbamate; ammonium N-phenyldithiocarbamate, ammonium N-4-methylphenyldithiocarbamate, was added portion wise and stirred at room temperature for 30 min. Then, the mixture was acidified with conc. HCl (3 mL), boiled for 5 min and left to cool. The precipitated solid was filtered off, washed with H<sub>2</sub>O, air dried, and the crude product was crystallized from the proper solvent to give the product.

2.7a 5-[2-(4-methoxy-3-methylphenyl)-2-oxoethyl]-3-phenyl-2-thioxo-1,3-thiazolidin-4-one (**8a**): From ethanol, M.p. 145°C. Anal. Calcd. for  $C_{19}H_{17}NO_3S_2$  (371.46): C, 61.43; H, 4.61; N, 3.77. Found: C, 61.83; H, 4.58; N, 4.15; IR (KBr,  $\nu$ ,  $cm^{-1}$ ): 1740 (C=O), 1659 (C=O), 1254 (C=S);  $^1H$ -NMR (DMSO- $d_6$ ,  $\delta$  ppm): 2.07 (3H, s,  $CH_3$  Ar), 3.91(3H, s, OCH<sub>3</sub>), 3.94–4.17 (2H, m, CH<sub>2</sub>-CH), 4.95–4.99 (H, m, CH-CH<sub>2</sub>), 7.07–7.91 (8H, m, Ar); MS ( $m/z$ , 371 (17.6%)) ( $M^+$ ).

2.7b 3-benzyl-5-[2-(4-methoxy-3-methylphenyl)-2-oxoethyl]-2-thioxo-1,3-thiazolidin-4-one (**8b**): From ethanol, M.p. 142°C. Anal. Calcd. for  $C_{20}H_{19}NO_3S_2$  (385.49): C, 62.31; H, 4.97; N, 3.63. Found: C, 62.55; H, 5.00; N, 3.58; IR (KBr,  $\nu$ ,  $cm^{-1}$ ): 1716 (C=O), 1664 (C=O), 1240 (C=S);  $^1H$ -NMR (DMSO- $d_6$ ,  $\delta$  ppm): 2.16 (3H, s,  $CH_3$  Ar), 3.29 (2H, s, CH<sub>2</sub>N), 3.88 (3H, s, OCH<sub>3</sub>), 4.06–4.13 (2H, m, CH<sub>2</sub>-CH), 4.94–4.97 (H, m, CH-CH<sub>2</sub>), 7.05–7.88 (8H, m, Ar); MS ( $m/z$ , 385 (17.0%)) ( $M^+$ ).

2.7c 5-[2-(4-methoxy-3-methylphenyl)-2-oxoethyl]-3-(4-tolyl)-2-thioxo-1,3-thiazolidin-4-one (**8c**): From ethanol, M.p. 163–165°C. Anal. Calcd. for  $C_{20}H_{19}NO_3S_2$  (385.49): C, 62.31; H, 4.97; N, 3.63. Found: C, 62.19; H, 4.88; N, 3.55. IR (KBr,  $\nu$ ,  $cm^{-1}$ ): 1733 (C=O), 1662 (C=O), 1251 (C=S);  $^1H$ -NMR (DMSO- $d_6$ ,  $\delta$  ppm): 2.20–2.37 (6H, 2s,  $CH_3$  Ar), 3.89 (3H, s, OCH<sub>3</sub>), 3.98–4.14 (2H, m, CH<sub>2</sub>-CH), 4.94–4.97 (H, m, CH-CH<sub>2</sub>), 7.07–7.90 (7H, m, Ar); MS ( $m/z$ , 385 (32.2%)) ( $M^+$ ).

### 2.8 General procedure for the synthesis of 2-substitutedamino-4-hydroxy-5-[2-(4-methoxy-3-methylphenyl)-2-oxoethyl]-1,3-thiazoles (**9a-d**)

A mixture of the acid (0.01 mol); appropriate thioureas (0.01 mol); namely thiourea; phenyl thiourea; p-tolyl thiourea and/or 4-chlorophenyl thiourea and ethanol (20 mL) in the presence of few drops of glacial acetic acid, was refluxed for 4 hrs., the solid product that obtained after cooling was filtered, washed by ethanol and crystallized from the suitable solvent to give (**9a-d**).

2.8a 2-amino-4-hydroxy-5-[2-(4-methoxy-3-methylphenyl)-2-oxoethyl]-1,3-thiazole (**9a**): From butanol, M.p. 240°C. Anal. Calcd. for  $C_{13}H_{14}N_2O_3S$  (278.32): C, 56.10; H, 5.07; N, 10.06. Found: C, 56.00; H, 5.00; N, 10.12; IR (KBr,  $\nu$ ,  $cm^{-1}$ ): 3219 (NH), 2964 (CH aliph.), 1665 (C=O);  $^1H$ -NMR (DMSO- $d_6$ ,  $\delta$  ppm):

2.14 (3H, s, CH<sub>3</sub>), 3.29–3.45 (2H, m, CH<sub>2</sub>-CH), 3.80 (3H, s, OCH<sub>3</sub>), 4.34–4.39 (1H, m, CH-CH<sub>2</sub>), 7.03–7.80 (3H, m, Ar-H), 8.76, 9.00 (2H, 2s, NH); MS (*m/z*, 278 (20%)) (M<sup>+</sup>).

2.8b *2-anilino-4-hydroxy-5-[2-(4-methoxy-3-methylphenyl)-2-oxoethyl]-1,3-thiazole (9b)*: From butanol, M.p. 210°C. Anal. Calcd. for C<sub>19</sub>H<sub>18</sub>N<sub>2</sub>O<sub>3</sub>S (354.42): C, 64.39; H, 5.12; N, 7.90. Found: C, 64.40; H, 5.00; N, 8.03; IR (KBr,  $\nu$ , cm<sup>-1</sup>): 3260 (NH), 1664 (C=O); <sup>1</sup>H-NMR (DMSO-*d*<sub>6</sub>,  $\delta$  ppm): 2.11 (3H, s, CH<sub>3</sub>), 3.29–3.64 (2H, m, CH<sub>2</sub>-CH), 3.86 (3H, s, OCH<sub>3</sub>), 4.40–4.50 (H, m, CH-CH<sub>2</sub>), 6.96–7.89 (8H, m, Ar-H), 11.15 (H, s, NH); MS (*m/z*, 354 (42%)) (M<sup>+</sup>).

2.8c *4-hydroxy-5-[2-(4-methoxy-3-methylphenyl)-2-oxoethyl]-2-(4-toluidino)-1,3-thiazole (9c)*: From butanol, M.p. 225°C. Anal. Calcd. for C<sub>20</sub>H<sub>20</sub>N<sub>2</sub>O<sub>3</sub>S (368.45): C, 65.20; H, 5.47; N, 7.60. Found: C, 65.51; H, 5.63; N, 7.91; IR (KBr,  $\nu$ , cm<sup>-1</sup>): 3181 (NH), 1684 (C=O); <sup>1</sup>H-NMR (DMSO-*d*<sub>6</sub>,  $\delta$  ppm): 2.11, 2.26 (6H, 2s, 2CH<sub>3</sub>-Ar), 3.28–3.55 (2H, m, CH<sub>2</sub>-CH), 3.86 (3H, s, OCH<sub>3</sub>), 4.44–4.48 (H, m, CH-CH<sub>2</sub>), 6.88–7.89 (7H, m, Ar-H), 11.08 (1H, s, NH); MS (*m/z*, 368 (98%)) (M<sup>+</sup>).

2.8d *2-(4-chloroanilino)-4-hydroxy-5-[2-(4-methoxy-3-methylphenyl)-2-oxoethyl]-1,3-thiazole (9d)*: From acetic acid, M.p. 228°C. Anal. Calcd. for C<sub>19</sub>H<sub>17</sub>ClN<sub>2</sub>O<sub>3</sub>S (388.45): C, 58.69; H, 4.41; N, 7.20. Found: C, 59.06; H, 4.92; N, 6.97; IR (KBr,  $\nu$ , cm<sup>-1</sup>): 3173 (NH), 1667 (C=O); <sup>1</sup>H-NMR (DMSO-*d*<sub>6</sub>,  $\delta$  ppm): 2.17 (3H, s, CH<sub>3</sub>), 3.28–3.67 (2H, m, CH<sub>2</sub>-CH), 3.86 (3H, s, OCH<sub>3</sub>), 4.40–4.60 (H, m, CH-CH<sub>2</sub>), 6.91–7.98 (7H, m, Ar-H), 11.32 (H, s, NH); MS (*m/z*, 388 (62%)) (M<sup>+</sup>), 390 (27%) (M<sup>+</sup>+2).

## 2.9 General procedure for the synthesis of 6-arylamino-1,4-dihydro-3-[4-methoxy-3-methylphenyl]-1,3-thiazolo[4,5-*c*]pyridazines (10a-c)

To a solution of the thiazoles (**9b-d**) (0.01 mol) in ethanol (20 mL); hydrazine hydrate 80% (0.015 mol) was added and the reaction mixture was refluxed for 6 h. The solid that separated after cooling was crystallized from suitable solvent to give the corresponding thiazolopyridazine (**10a-c**).

2.9a *6-anilino-1,4-dihydro-3-[4-methoxy-3-methylphenyl]-1,3-thiazolo[4,5-*c*]pyridazine (10a)*: From ethanol, M.p. 290°C. Anal. Calcd. for C<sub>19</sub>H<sub>18</sub>N<sub>4</sub>OS (350.43):

C, 65.12; H, 5.18; N, 15.99. Found: C, 65.33; H, 5.16; N, 16.11; IR (KBr,  $\nu$ , cm<sup>-1</sup>): 3349 (NH), 1601 (C=N) or (C=C); <sup>1</sup>H-NMR (DMSO-*d*<sub>6</sub>,  $\delta$  ppm): 2.19 (3H, s, CH<sub>3</sub>), 3.24 (2H, s, CH<sub>2</sub>), 3.89 (3H, s, OCH<sub>3</sub>), 6.75–7.85 (8H, m, Ar-H); MS (*m/z*, 350 (1.3%)) (M<sup>+</sup>).

2.9b *1,4-dihydro-3-[4-methoxy-3-methylphenyl]-6-(4-toluidino)-1,3-thiazolo[4,5-*c*]pyridazine (10b)*: From ethanol, M.p. 240–242°C. Anal. Calcd. for C<sub>20</sub>H<sub>20</sub>N<sub>4</sub>OS (364.46): C, 65.91; H, 5.53; N, 15.37. Found: C, 66.05; H, 5.76; N, 15.29; IR (KBr,  $\nu$ , cm<sup>-1</sup>): 3342 (NH), 1603 (C=N) or (C=C).); <sup>1</sup>H-NMR (DMSO-*d*<sub>6</sub>,  $\delta$  ppm): 2.16, 2.19 (6H, 2s, 2CH<sub>3</sub>-Ar), 3.33 (2H, s, CH<sub>2</sub>), 3.80 (3H, s, OCH<sub>3</sub>), 6.93–7.89 (7H, m, Ar-H), 8.96 (H, s, NH), 13.1 (H, s, NH); MS (*m/z*, 363 (14.7%)) (M<sup>+</sup>-H).

2.9c *6-(4-chloroanilino)-1,4-dihydro-3-[4-methoxy-3-methylphenyl]-1,3-thiazolo[4,5-*c*]pyridazine (10c)*: From butanol, M.p. 256–258°C. Anal. Calcd. for C<sub>19</sub>H<sub>17</sub>ClN<sub>4</sub>OS (384.88): C, 59.29; H, 4.45; N, 14.56. Found: C, 59.00; H, 4.56; N, 14.48; IR (KBr,  $\nu$ , cm<sup>-1</sup>): 3331 (NH), 1601 (C=N) or (C=C); <sup>1</sup>H-NMR (DMSO-*d*<sub>6</sub>,  $\delta$  ppm): 2.16 (3H, s, CH<sub>3</sub>), 3.30 (2H, s, CH<sub>2</sub>), 3.80 (3H, s, OCH<sub>3</sub>), 6.93–7.55 (7H, m, Ar-H), 9.2 (H, s, NH), 13.2 (H, s, NH); MS (*m/z*, 384 (4.5%)) (M<sup>+</sup>).

## 2.10 Synthesis of 1,4-dihydro-3-[4-methoxy-3-methylphenyl]-1-phenyl-6-(4-toluidino)-1,3-thiazolo[4,5-*c*]pyridazine (11)

A mixture of 4-hydroxy-5-[2-(4-methoxy-3-methylphenyl)-2-oxoethyl]-2-(4-toluidino)-1,3-thiazole (**9b**) (0.005 mol) and phenylhydrazine hydrochloride (0.006 mol) in pyridine (5 mL) was heated under reflux for 6 hrs. the solid separated after evaporation of the most of the solvent and cooling, was washed by benzene and crystallized from the proper solvent to give (**11**) with M.p. 316°C. Anal. Calcd. for C<sub>26</sub>H<sub>24</sub>N<sub>4</sub>OS (440.56): C, 70.88; H, 5.49; N, 12.72; Found: C, 71.32; H, 5.42; N, 12.66; IR (KBr,  $\nu$ , cm<sup>-1</sup>): 3018 (CH-Ar), 2839–02918 (CH aliph.), 1620 (C=C); <sup>1</sup>H-NMR (DMSO-*d*<sub>6</sub>,  $\delta$  ppm): 2.23 and 2.25 (6H, 2s, 2CH<sub>3</sub>-Ar), 2.36 (2H, s, CH<sub>2</sub>), 3.89 (3H, s, OCH<sub>3</sub>), 7.20–8.03 (12H, m, Ar-H) 9.23 (H, s, NH); MS (*m/z*, 440 (10%)) (M<sup>+</sup>).

## 2.11 Synthesis of acetyl derivatives of thiazoles (12a-d)

A solution of thiazoles (**9a-d**) (1 gm) in acetic anhydride (5 mL) was refluxed for 4 h. The solid

product obtained, after evaporation of most of the solvent and cooling, was filtered off, washed with ethanol and crystallized from the proper solvent to give (**12a-d**).

2.11a *4-acetoxy-2-amino-5-[2-(4-methoxy-3-methylphenyl)-2-oxoethyl]-1,3-thiazole (12a)*: from butanol, M.p. 205°C. Anal. Calcd. for C<sub>15</sub>H<sub>16</sub>N<sub>2</sub>O<sub>4</sub>S (320.36): C, 56.24; H, 5.03; N, 8.74. Found: C, 56.47; H, 4.88; N, 8.91; IR (KBr,  $\nu$ , cm<sup>-1</sup>): 3439 (OH), 3237 (NH), 1779 (CO-CH<sub>3</sub>), 1672 (CO-Ar); <sup>1</sup>H-NMR (DMSO-*d*<sub>6</sub>,  $\delta$  ppm): 2.17 (3H, s, CH<sub>3</sub>Ar), 2.38 (2H, s, CH<sub>2</sub>), 2.43 (3H, s, CH<sub>3</sub>CO), 3.80 (3H, s, OCH<sub>3</sub>), 6.81–7.42 (3H, m, Ar-H), 12.34 (H, s, NH); MS (*m/z*, 320 (100%)) (M<sup>+</sup>).

2.11b *4-acetoxy-2-anilino-5-[2-(4-methoxy-3-methylphenyl)-2-oxoethyl]-1,3-thiazole (12b)*: From chloroform, M.p. 275°C. Anal. Calcd. for C<sub>21</sub>H<sub>20</sub>N<sub>2</sub>O<sub>4</sub>S (396.46): C, 63.62; H, 5.08; N, 7.07. Found: C, 63.90; H, 5.21; N, 6.66; IR (KBr,  $\nu$ , cm<sup>-1</sup>): 1781 (CO-CH<sub>3</sub>), 1679 (CO-Ar); <sup>1</sup>H-NMR (DMSO-*d*<sub>6</sub>,  $\delta$  ppm): 2.17 (2H, s, CH<sub>2</sub>), 2.27 (3H, s, CH<sub>3</sub>Ar), 2.51 (3H, s, CH<sub>3</sub>-CO), 3.87 (3H, s, OCH<sub>3</sub>), 7.05–7.88 (8H, m, Ar-H); MS (*m/z*, 396 (4.4%)) (M<sup>+</sup>).

2.11c *4-acetoxy-2-(4-toluidino)-5-[2-(4-methoxy-3-methylphenyl)-2-oxoethyl]-1,3-thiazole (12c)*: from ethanol, M.p. 215°C. Anal. Calcd. for C<sub>22</sub>H<sub>22</sub>N<sub>2</sub>O<sub>4</sub>S (410.48): C, 64.37; H, 5.40; N, 6.82. Found: C, 64.20; H, 5.02; N, 6.77; IR (KBr,  $\nu$ , cm<sup>-1</sup>): 1768 (CO-CH<sub>3</sub>), 1672 (CO-Ar); <sup>1</sup>H-NMR (DMSO-*d*<sub>6</sub>,  $\delta$  ppm): 2.18, 2.28 (6H, 2s, 2CH<sub>3</sub>-Ar), 2.38 (3H, s, CH<sub>3</sub>-C=O), 2.48 (2H, s, CH<sub>2</sub>), 3.81 (3H, s, OCH<sub>3</sub>), 6.72–7.41 (7H, m, Ar-H); MS (*m/z*: 410 (16.7%)) (M<sup>+</sup>).

2.11d *4-acetoxy-2-(4-chloroanilino)-5-[2-(4-methoxy-3-methylphenyl)-2-oxoethyl]-1,3-thiazole (12d)*: From butanol, M.p. 208°C. Anal. Calcd. for C<sub>21</sub>H<sub>19</sub>ClN<sub>2</sub>O<sub>4</sub>S (430.90): C, 58.54; H, 4.44; N, 6.50. Found: C, 58.55; H, 4.44; N, 6.38; IR (KBr,  $\nu$ , cm<sup>-1</sup>): 1768 (CO-CH<sub>3</sub>), 1672 (CO-Ar); <sup>1</sup>H-NMR (DMSO-*d*<sub>6</sub>,  $\delta$  ppm): 1.98 (3H, s, CH<sub>3</sub>-Ar), 2.20 (2H, s, CH<sub>2</sub>), 2.28 (3H, s, CH<sub>3</sub>-CO), 3.81 (3H, s, OCH<sub>3</sub>), 6.73–7.64 (7H, m, Ar-H); MS (*m/z*, 430 (60.6%)) (M<sup>+</sup>).

2.12 *Synthesis of 2-substituted amino-4-hydroxy-5-[2-(4-methoxy-3-methylphenyl)-1-bromo-2-oxoethyl]-1,3-thiazoles (13a-b)*

To a solution of thiazoles (**9b**) or (**9d**) (0.005 mol) in acetic acid (40 mL), a bromine solution (0.005 mol) in acetic acid (10 mL) was added slowly with stirring.

After addition was complete, the mixture was gently warmed until HBr gas evolution ceased. The precipitated solid was filtered off, washed with ethanol, air dried and crystallized from the proper solvent to give (**13a-b**).

2.12a *2-anilino-4-hydroxy-5-[2-(4-methoxy-3-methylphenyl)-1-bromo-2-oxoethyl]-1,3-thiazole (13a)*: from ethanol, M.p. 250–252°C. Anal. Calcd. for C<sub>19</sub>H<sub>17</sub>BrN<sub>2</sub>O<sub>3</sub>S (433.31): C, 52.67; H, 3.95; N, 6.46. Found: C, 52.49; H, 3.88; N, 6.52; IR (KBr,  $\nu$ , cm<sup>-1</sup>): 3254 (NH), 1696 (CO-Ar); <sup>1</sup>H-NMR (DMSO-*d*<sub>6</sub>,  $\delta$  ppm): 2.19 (3H, s, CH<sub>3</sub>-Ar), 3.32–3.45 (2H, m, CH-CH), 3.89 (3H, s, OCH<sub>3</sub>), 7.04–8.10 (8H, m, Ar-H), 11.76 (H, s, NH); MS (*m/z*, 433 (1.0%)) (M<sup>+</sup>).

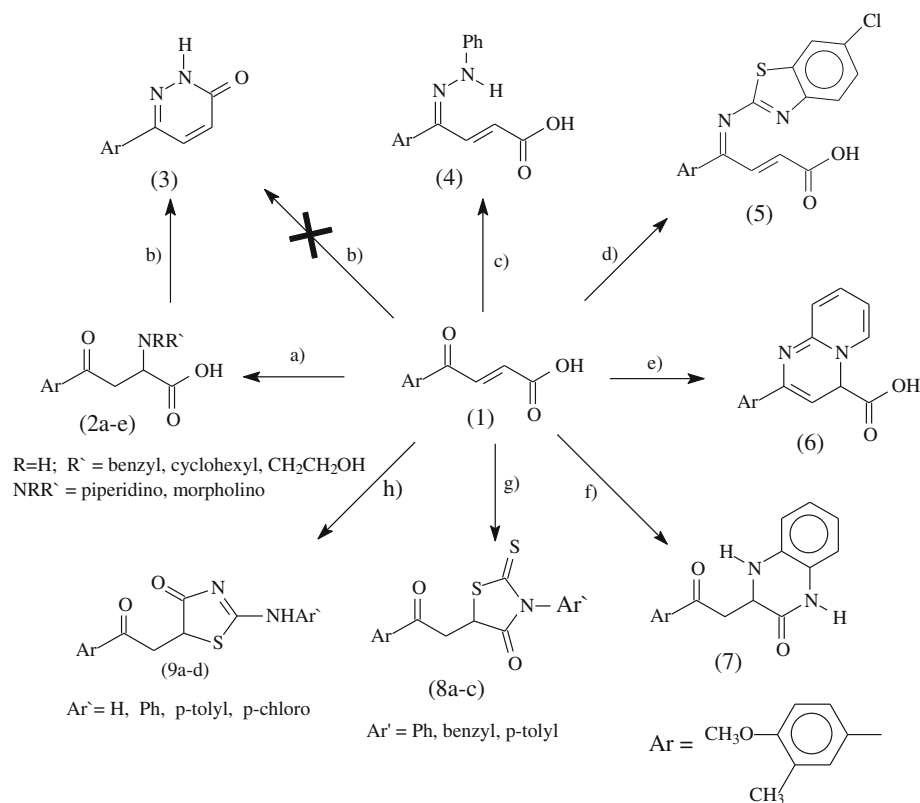
2.12b *2-(4-chloroanilino)-4-hydroxy-5-[2-(4-methoxy-3-methylphenyl)-1-bromo-2-oxoethyl]-1,3-thiazole (13b)*: From ethanol, M.p. 278–280°C. Anal. Calcd. for C<sub>19</sub>H<sub>16</sub>BrClN<sub>2</sub>O<sub>3</sub>S (467.76): C, 48.79; H, 3.45; N, 5.99. Found: C, 49.20; H, 3.55; N, 6.10; IR (KBr,  $\nu$ , cm<sup>-1</sup>): 3253 (NH), 1682 (CO-Ar); <sup>1</sup>H-NMR (DMSO-*d*<sub>6</sub>,  $\delta$  ppm): 2.18 (3H, s, CH<sub>3</sub>-Ar), 3.53–3.56 (2H, m, CH-CH), 3.89 (3H, s, OCH<sub>3</sub>), 7.39–8.10 (7H, m, Ar-H), 11.87 (H, s, NH); MS (*m/z*, 386 (18.3%)) (M<sup>+</sup>-HBr).

### 3. Results and Discussion

The synthetic routes of the prepared derivatives are shown in schemes 1, 2 and 3. The key compound, 4-[4-methoxy-3-methylphenyl]-4-oxobutenoic acid (**1**), was synthesized in good yield by Friedel-Crafts acylation of *o*-cresyl methyl ether with maleic anhydride according to reported method.<sup>13</sup>

The aza-Michael addition with primary and secondary amines was carried out by the treatment of the acid (**1**) with amines in dry benzene in good yields with the formation of the corresponding  $\alpha$ -substituted amino acids (**2a-e**). These compounds were formed through the attack of the amino group on the electron-deficient olefinic carbon atom,  $\alpha$ - to the carboxyl group. IR spectra showed prominent bands for OH groups at 3436–3438 cm<sup>-1</sup> and for C=O groups at 1668–1675 cm<sup>-1</sup>. <sup>1</sup>H-NMR spectra the aromatic protons were observed at expected ppm's. The signals for OH groups were observed as merged with the signals of aromatic protons in aromatic region.

Surprisingly, when the reaction of the  $\alpha$ -substituted amino acids (**2**) with hydrazine was carried out in refluxing ethanol, we unexpectedly obtained 6-(4-methoxy-3-methylphenyl)-3(2*H*)-pyridazinone (**3**)

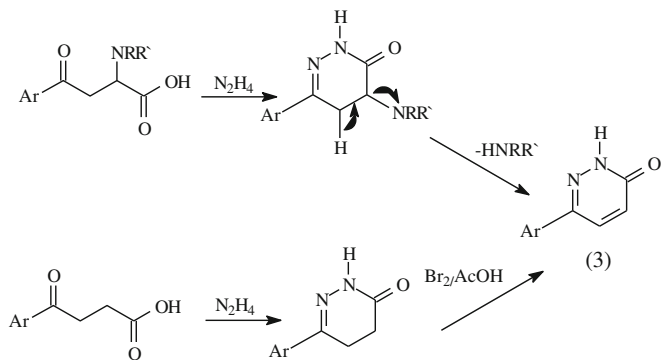


### Reaction conditions

- a) amines, dry benzen; b) hydrazine, ethanol, reflux; c) phenylhydrazine, ethanol, reflux;  
 d) 2-amino-6-chlorobenzothiazole, ethanol, reflux; e) 2-aminopyridine, ethanol, reflux;  
 f) o-phenylene diamine, ethanol, reflux; g) ammonium aryl dithiocarbamate, HCl  
 h) thioureas, ethanol and AcOH, reflux;

**Scheme 1.** Reactions of Oxobutenoic acid derivative with different nucleophiles.

(Scheme 2) instead of 4-alkylamino dihydropyridazinone. In the presence of hydrazine hydrate the acid (2) underwent cyclization with the formation of 4-alkylamino dihydropyridazinone followed by the fission of the benzylamino group in position 4 and aromatization of the pyridazine ring. The proposed structure of this compound was supported spectral data and by the identity of its m.p. with that of an authentic sample prepared by us<sup>14</sup> from the reaction of the corresponding



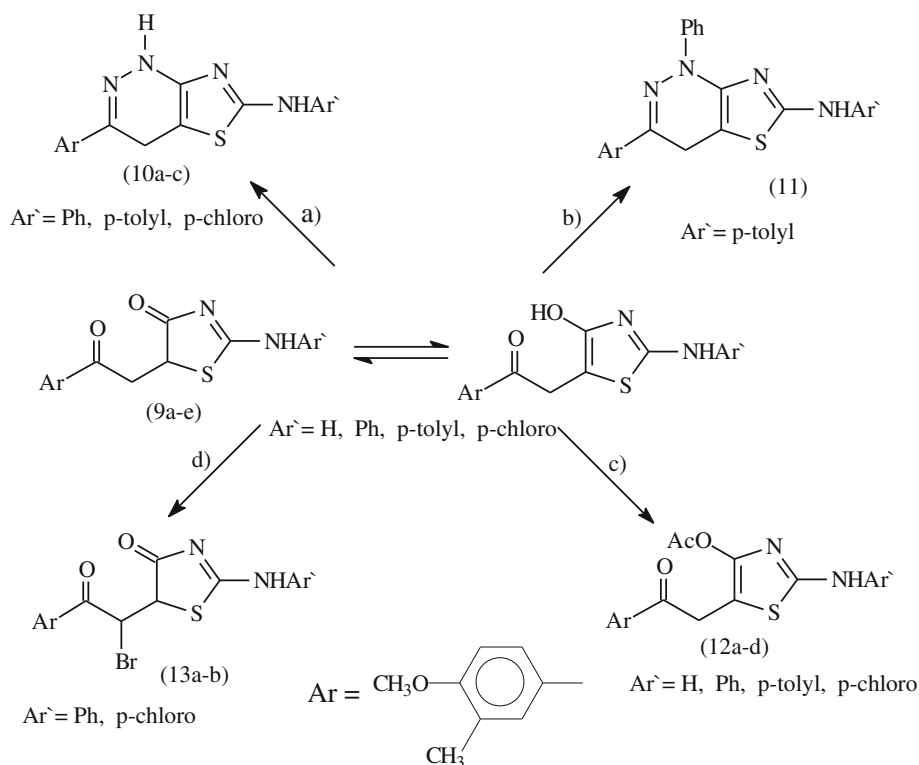
**Scheme 2.** Formation of 6-(4-methoxy-3-methylphenyl)-3(2H)-pyridazinone.

4-(4-methoxy-3-methylphenyl)-4-oxobutanoic acid with hydrazine hydrate and treating the resulting dihydropyridazinone with bromine.

Attempts to obtain a solid product through the reaction of the acid (1) with hydrazine hydrate under different conditions were failed. However, when the acid was treated with phenylhydrazine in ethanol afforded the corresponding 4-[4-methoxy-3-methylphenyl]-4-oxobutenoic acid phenyl hydrazone (4). The IR showed bands in the region of 3500 cm<sup>-1</sup>, 3294 cm<sup>-1</sup> and 1679 cm<sup>-1</sup> which are assigned to the vibrations of the OH, NH and C=O groups, respectively.

In similar way, when the acid (1) was submitted to react with 2-amino-6-chlorobenzothiazole in boiling ethanol it afforded 4-[6-chlorobenzothiazol-2-ylimino]-4-[4-methoxy-3-methylphenyl]-4-oxobutenoic (5). The reaction takes place via the condensation reaction between the amino and keto groups with the formation of the corresponding Schiff base.

The treatment of the acid (1) with 2-aminopyridine in boiling ethanol resulted in the formation of pyridopyrimidine derivative (6). The formation of compound (6) was assumed to proceed via the addition of the amine



### Reaction conditions

a) hydrazine, ethanol, reflux; b) phenylhydrazine hydrochloride, pyridine, reflux; c) acetic anhydride, reflux; d) bromine, acetic acid, stirring

**Scheme 3.** Reactions of 2-aminothiazole derivatives.

to the double bond followed by cyclocondensation with the keto group.

The interaction of the acid (1) with *o*-phenylene diamine in refluxing ethanol afforded the corresponding quinoxaline (7). The <sup>1</sup>H-NMR showed multiplets for the methylene and methine groups protons ( $\delta = 3.20$ – $3.88$ ), a signal for the amino group ( $\delta = 5.88$  ppm), a multiplet for the aromatic protons ( $\delta = 6.06$ – $7.89$ ) and also a downfield singlet ( $\delta = 10.2$  ppm) which is assigned to the amido NH proton.

When the acid (1) was allowed to react with ammonium aryldithiocarbamates in ethanol in the presence of hydrochloric acid gave the corresponding 2-thioxo-1,3-thiazolidin-4-one derivatives (8a-c) in good yield. The structures of these compounds were confirmed by the absence of the absorption bands corresponding to the hydroxyl group in their IR spectra and the appearance of two multiplets in the aliphatic regions corresponding to the methylene ( $\delta = 3.77$ – $4.14$ ) and methine ( $\delta = 4.93$ – $4.97$ ) groups in their <sup>1</sup>H-NMR spectra.

Similarly, the 2-substitutedamino-4-hydroxy-5-[2-(4-methoxy-3-methylphenyl)-2-oxoethyl]-1,3-thiazoles (9a-c) were obtained from the reaction of the acid (1) with the appropriate thiourea derivatives in boiling

ethanol containing catalytic amount of glacial acetic acid.

The 2-aminothiazole derivatives (9a-c) were considered as key starting material in which they reacted with various reagents (Scheme 2). Thus, the treatment of (9a-c) with hydrazine hydrate in refluxing ethanol or phenylhydrazine hydrochloride in pyridine afforded the corresponding thiazolopyridazine derivatives (10a-c) and (11), respectively. The structures of these compounds were confirmed by the absence of the characteristic absorption bands corresponding to the carbonyl group in their IR spectra.

The acetylation of the aminothiazolons (9a-c) was carried out by boiling them with acetic anhydride giving the 5-acetoxy-2-substitutedamino-thiazoles (12a-c). The IR spectra of these compounds showed the appearance of new absorption bands in the region  $1768$ – $1781$   $\text{cm}^{-1}$  corresponding to the acetate group which confirmed that the acetylation occurred at the hydroxyl groups and the appearance of new singlets in the aliphatic regions corresponding to the acetate groups ( $\delta = 2.28$ – $2.51$ ) in their <sup>1</sup>H-NMR spectra.

The 2-aminothiazolone derivatives (9b-c) underwent bromination upon treatment with bromine in acetic

**Table 1.** The anti-bacterial effects of the synthesized compounds.

Compound No.	Gram negative bacteria			Gram positive bacteria		
	<i>Pseudomonas aeruginosa</i>	<i>Serratia sp.</i>	<i>Escherichia coli</i>	<i>Micrococcus luteus</i>	<i>Bacillus subtilis</i>	<i>Staphylococcus aureus</i>
2a	+	+	-	+	+	++
2b	-	+	-	+	+	+
2c	++	+	+	+	+	+++
2d	+	+	+	-	+	+++
2e	+	+	-	-	+	+++
3	+	+	-	+	+	++
4	-	-	-	+	-	++
5	+	+	-	+	+	++
6	+	+	+	+	+	++
7	-	-	-	-	-	-
8a	+	-	-	-	-	-
8b	++	-	+	+	-	-
8c	-	-	-	-	-	-
9a	+	+	-	-	-	-
9b	+	+	-	-	-	-
9c	+	+	-	-	-	-
9d	+	+	-	-	-	+
10a	+	-	-	+++	+	-
10b	++	-	-	-	-	-
10c	-	-	-	+	-	-
11	+	-	-	-	-	-
12a	+	-	-	-	-	+
12b	+	+	+	-	-	+
12c	+	-	+	-	-	+
12d	+	-	-	+	+	-
13a	-	-	-	-	-	-
13b	+	-	-	-	-	-

Solvent: DMSO, C= 1mg/ml; (-) = no activity; (+) = mild activity (diameter 7-10 mm); (++) = moderate (diameter 10-17 mm); (+++) = high activity (diameter 18-22 mm).

acid affording the 2-substituted amino-4-hydroxy-5-[2-(4-methoxy-3-methylphenyl)-1-bromo-2-oxoethyl]-1,3-thiazoles (**13a-b**).

### 3.1 Biological activity

Using paper disk agar diffusion technique<sup>15</sup> all the newly synthesized compounds were tested in-vitro for anti-bacterial activity against strains of bacteria such as

*Bacillus subtilis*, *Staphylococcus aureus*, *Micrococcus luteus*, *Escherichia coli*, *Serratia sp.* and *Pseudomonas aeruginosa*. The compounds were tested at 1mg/mL concentration and the activity was determined by measuring Zone of inhibition. The screening indicated that some of the synthesized compounds exhibited antibacterial activities against one or the other type of bacteria (table 1). We expect that our results may be helpful in understanding the minimum structural requirements for these compounds acting as antibacterial agents. So,

**Table 2.** MIC's as ( $\mu$ g/ml) of a few newly synthesized compound.

Compound No	Gram positive bacteria		Gram negative bacteria	
	<i>Pseudomonas aeruginosa</i>	<i>Serratia sp.</i>	<i>Bacillus subtilis</i>	<i>Staphylococcus aureus</i>
2a	250	125	125	125
2d	125	125	250	125
2e	500	500	125	125
5	500	125	125	125
6	125	< 125	125	< 125



further investigation was tested to calculate MIC's as ( $\mu\text{g/ml}$ ) of those active compounds (table 2). The series of amino acid derivatives (**2a-e**) showed higher activity than the other compounds. The thiazole derivatives (**9a-d**) showed activity against Gram negative bacteria only (*Pseudomonas aeruginosa* and *Serratia sp.*). Actylation of these compound extends the activity to include also *Staphylococcus aureus*. While condensation with phenylhydrazine or addition to bromine lead to decrease its activity.

#### 4. Conclusion

We have shown that different heterocyclic compounds have been synthesized from the reaction of 4-[4-methoxy-3-methylphenyl]-4-oxobutenoic acid with different nucleophiles. Compounds which retain the carboxyl group (the series of amino acid, benzothiazole and pyridopyrimidine derivatives) showed highest and broad spectrum anti-bacterial activity compared to other compounds which lose the carboxyl group.

#### Supplementary Information

IR, NMR and mass spectra of compounds 2a, 2b, 2c, 2d, 2e, 3, 4, 5, 6, 7, 8a, 8b, 8c, 9a, 9b, 9c, 9d, 10a, 10b, 10c, 11, 12a, 12b, 12c, 12d, 13a and 13b are shown in Figures S1 to S78.

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