

Efficient Synthesis of 1-Substituted-4-phenyl-1, 4-dihydro-5*H*-tetrazole-5-thione and (1-Phenyl-1*H*-tetrazol-5-yl)thioacetyl Derivatives

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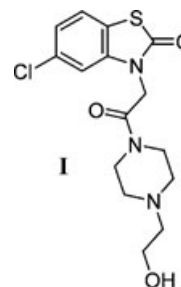
ABSTRACT: A new and convenient synthesis of a variety of *N*- and *S*-substituted tetrazoles has been developed via azide and Mannich reaction methods. Compounds were characterized by elemental analysis, MALDI MS, and ¹H NMR data. © 2007 Wiley Periodicals, Inc. *Heteroatom Chem* 18:637–643, 2007; Published online in Wiley InterScience (www.interscience.wiley.com). DOI 10.1002/hc.20353

INTRODUCTION

Substituted tetrazoles [1,2] are well known as an excellent nonclassical carboxylic acid bioisosteres in biological molecules [3,4]. Recently, X-ray crystal structure revealed that the tetrazolate moiety of several nonpeptide antagonists was proved to form a hydrogen bond with protonated lysine and histidine at the active site of the angiotensin II receptor [3,5,6].

Waisser et al. [7] reported that substituted benzyl sulfanyl group enhances the antimycobacterial activity of 1-aryl-5-alkylsulfanyltetrazoles. *Tiaramide I* and other benzothiazolone acetamides are shown to

inhibit prostoglandin synthesis and have significant analgesic and anti-inflammatory activities [8–10].



RESULTS AND DISCUSSION

In this study, we report an efficient synthesis of a series of tetrazoles containing a variety of functional methyl amino and acetamide moieties at positions 1 and 5, respectively, as represented in Schemes 1–3, for a significant structure–activity relationship study.

Our synthetic strategy is dependent on the regioselective reactions of thioamides, which gives *S*- and *N*-substituted derivatives [11–13]. Thus, the reaction of 1 molar equivalent of the selected 1-phenyl-4,5-dihydro-1*H*-1,2,3,4-tetrazole-5-thione (**1**) with 1 molar equivalent of secondary amines and excess formaldehyde regioselectively gives a reasonable amount of *N*-methyl amino tetrazole **2a–c**. The

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N-substitution is due to strong Coulombic attraction between the hard part of the ambident nucleophile and the electrophile [12]. However, treatment of 2 molar equivalents of tetrazole **1** with 1 molar equivalent of piperazine and excess formaldehyde, under the same reaction conditions, afforded bis methyl amino piperazine **3**.

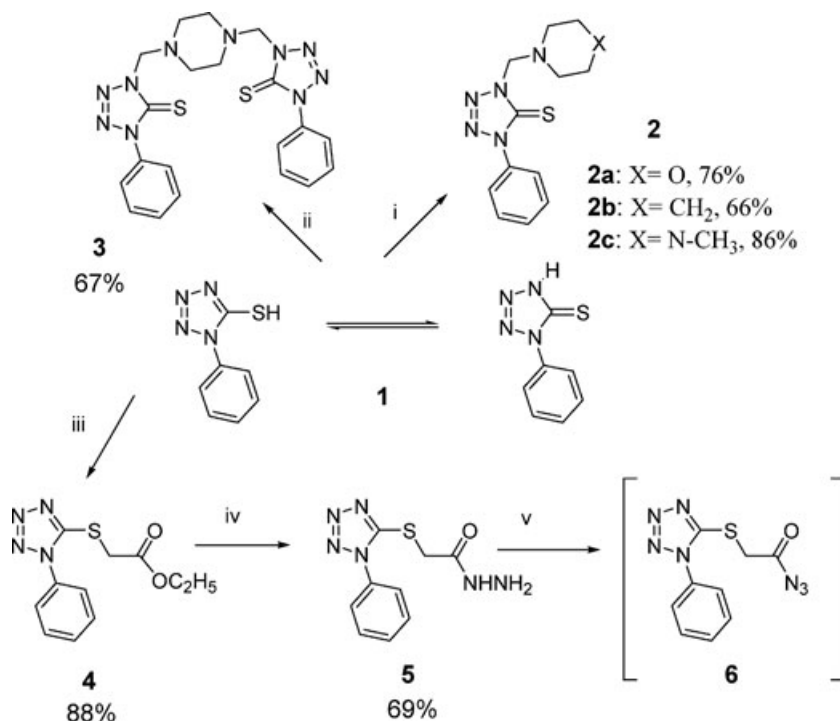
The elemental analysis together with MALDI MS of both structures **2a–c** and **3** agreed with the molecular formula of these compounds. The MALDI of **2a** gave 300.3 corresponding to $(M + Na)^+$. The ^1H NMR spectra were used as a major tool in identifying the *N*-substitution. Thus, ^1H NMR spectra showed a singlet at 5.33 ppm, characteristic for NCH_2N , and other peaks from secondary amine and tetrazole moieties are also clearly shown in the Experimental section.

The azide derivative **6** was proved to be highly versatile intermediate, forming a variety of acetamides. This extensive structure modification of the selected tetrazole molecules **1** could affect the biological activity. The synthesis of tetrazole acetamides via azide-coupling method is shown in Schemes 1–3. The reaction of 1-phenyl-4,5-dihydro-1*H*-1,2,3,4-tetrazole-5-thione (**1**) with chloroethyl acetate in the presence of K_2CO_3 furnished the regioselective *S*-alkylation reaction product **4** [13,14]. The expected

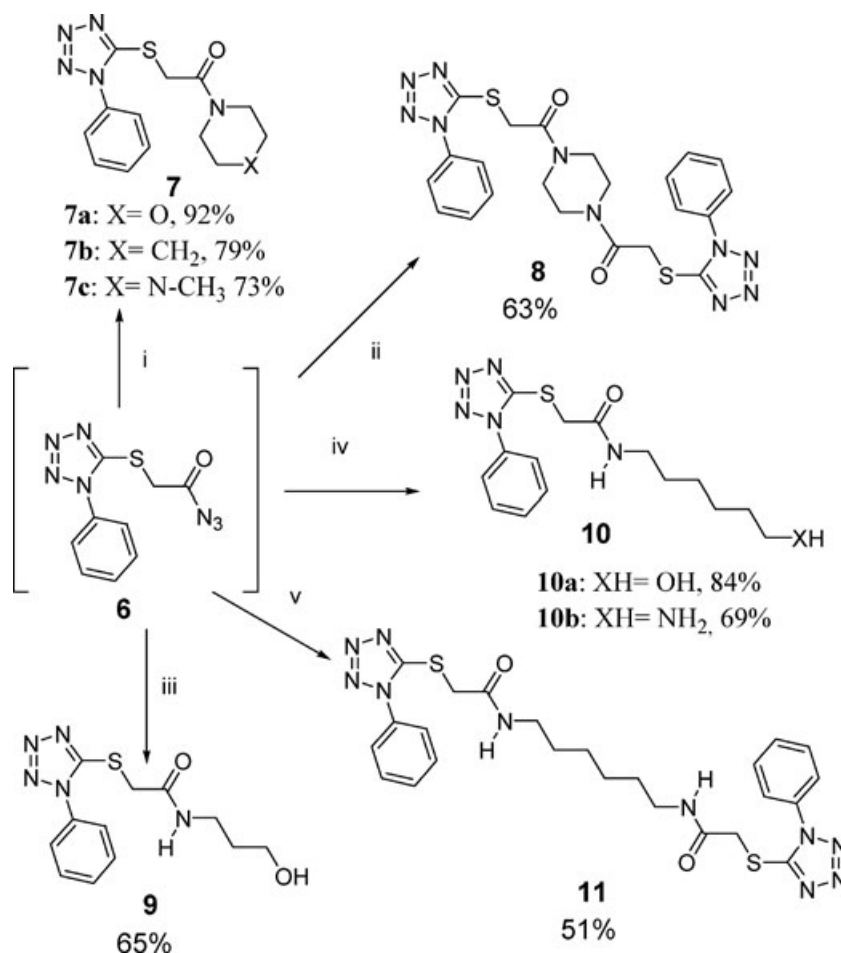
S-alkylation reaction was favored because of an interaction between the HOMO of the nucleophile and the LUMO of the electrophile to produce the *S*-attack [13]. Hydrazinolysis of **4** afforded hydrazide **5**, which was subsequently converted into azide **6** by treatment with NaNO_2 and HCl mixture (Scheme 1).

The in situ generated azide **6** solution extracted with ethyl acetate was used without isolation and purification. One molar equivalent of the amine was treated with 1 molar equivalent of the in situ generated ethyl acetate solution of azide **6** at -5°C . A variety of *S*-substituted acetamides of tetrazoles **7a–c**, **9**, and **10a,b** were obtained in moderate to good yields. However, employing 1 molar equivalent of the amine (piperazine and 1,6-diamino hexane) with 2 molar equivalents of the intermediate azide produced bistetrazoles derivatives **8** and **11**, respectively (Scheme 2). The long-chain derivatives **9–11** were designed to increase the lipophilicity of the tetrazole moiety, which is an important factor when designing a drug molecule able to pass through the cell membrane.

The ^1H NMR spectra of the *S*-acylated derivatives showed a singlet, typically associated with a SCH_2CO group appeared at 4.50 ppm for short-chain derivatives **9**, **7a–c**, **8** and 4.06 ppm for long-chain substituents **10a,b**, and **11**. The structure of hydroxy



SCHEME 1 Reagents and conditions: (i) CH_2O , secondary amine, ethanol, 25°C , 2 h; (ii) CH_2O , piperazine, ethanol, 25°C , 2 h; (iii) $\text{ClCH}_2\text{COOC}_2\text{H}_5$, Et_3N , EtOH , 85°C , 6 h; (iv) $\text{NH}_2\text{NH}_2 \cdot \text{H}_2\text{O}$, EtOH , 85°C , 3 h; and (v) NaNO_2 , HCl , -5°C , 30 min.



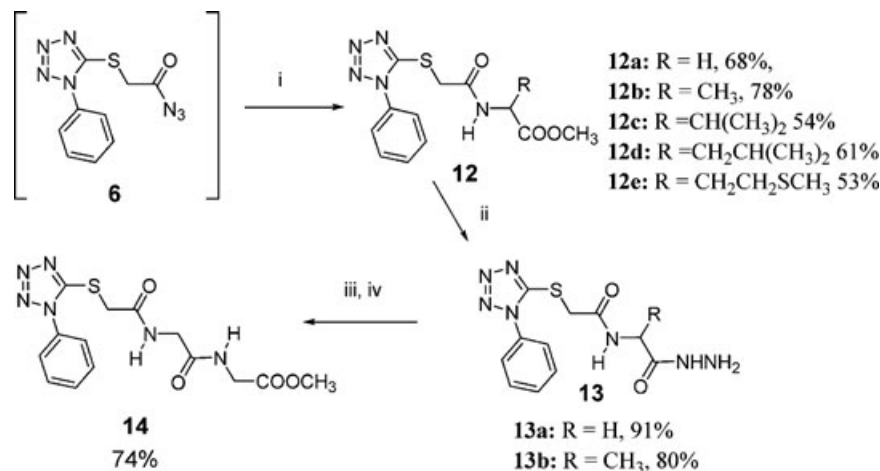
SCHEME 2 Reagents and conditions: (i) secondary amine, ethyl acetate, 25°C, 24 h; (ii) piperazine, ethyl acetate, 25°C, 24 h; (iii) 1-propanol amine, ethyl acetate, 25°C, 24 h; (iv) 1,6-diamino hexane or 1-hexanol amine, ethyl acetate, 25°C, 24 h; (v) 1,6-diamino hexane, ethyl acetate, 25°C, 24 h.

derivatives **9** and **10a** was confirmed by measuring the ¹H NMR spectrum in CDCl₃ and CDCl₃/D₂O. The ¹H NMR spectrum of **9** in CDCl₃ showed a quartet at 3.42 ppm, while giving a triplet at 3.42 ppm in CDCl₃/D₂O characteristic for NHCH₂CH₂. Furthermore, an exchangeable hydroxyl group appeared at 2.83 ppm. The structure of the bis derivatives **8** and **11** was simply deduced by comparing the intensity of bi-nucleophilic amine residue to a tetrazole thioacetyl moiety.

In addition, our next target was the elongation of the side chain at the thioacetyl group by coupling with amino acid. It was designed to increase the binding affinity of tetrazole moiety by interacting with the receptor recognition sites. Several coupling reagents were employed in the literature to introduce peptide bonds by the reaction of acid derivatives with amino acid methyl ester. Among those were 1-hydroxy-benzotriazole (HOBt) [15,16] and *N,N*-dicyclohexylcarbodiimide (DCC)

[17]. HOBt is widely used as an additive to decrease racemization in the carbodiimide peptide coupling. Our choice was azide coupling following Sahin et al.'s method [18–20], which was reported to minimize the degree of racemization in amino acid coupling. Thus, azide **6** reacts with amino acid methyl ester. HCl (Gly, Ala, Val, Leu, Meth) in the presence of Et₃N afforded peptides **12a–e** in good yield. Hydrazinolysis of **12a,b** afforded hydrazides **13a,b** (Scheme 3).

The structure assignments of the amino acid derivatives **12a–e** were based on elemental analysis, MALDI, and ¹H NMR spectra. The MALDI spectra of **12b** gave 330.5 corresponding to (M+Na)⁺. Significantly, *S*-alkylation site (SCH₂CO group) was indicated as a singlet in the ¹H NMR spectrum at 4.03 ppm for all amino acid derivatives **12a–e**. The ¹H NMR spectrum of methyl *N*-{[(1-phenyl-1*H*-tetrazol-5-yl)thio]acetyl}alaninate (**12b**) displayed multiplet resonance at δ 4.62–4.47 ppm



SCHEME 3 (i) amino acid methyl ester hydrochloride, Et₃N, ethyl acetate, 25°C, 24 h; (ii) NH₂NH₂·H₂O, EtOH, 85°C, 3 h; (iii) NaNO₂, HCl, -5°C, 30 min; (iv) glycine methyl ester hydrochloride, Et₃N, ethyl acetate, 25°C, 24 h.

characteristic for NHCH group, whereas a broad signal was observed at 7.48 ppm for a NH group. Additional evidence for the structure of glycine acetamide derivative **12a** was the dipeptide **14** formation via the azide-coupling method as described above.

CONCLUSION

In summary, we describe a simple, convenient, and practical method for the extensive modification of the structure of the selected 1-phenyl-4,5-dihydro-1H-1,2,3,4-tetrazole-5-thione (**1**) regioselectively at positions 1 and 5 via the azide-coupling method and the Mannich reaction, respectively, with lipophilic amines, secondary amines, and amino acids.

EXPERIMENTAL

Solvents were purified and dried in the usual way. The boiling range of the petroleum ether used was 35–65°C. Thin layer chromatography (TLC): silica gel 60 F₂₅₄ plastic plates (E. Merck, layer thickness 0.2 mm) detected by UV absorption. Melting points were determined on a Büchi 510 melting-point apparatus, and the values are uncorrected. NMR spectra were measured with Bruker AC 250 (250 MHz). TMS (0.00 ppm) or the signal of the deuterated solvent was used as internal standard. FAB-MS modified Finnigan MAT 312/ AMD 5000 spectrometer was used at 790 eV and $T = 70$ MALDI-MS, the mass spectra were measured with a KRATOS Analytical Kompact spectrometer. Microanalyses were performed at the Microanalytical Center, Chemistry Department, Konstanz University, Germany.

The starting compounds **1**, **5** were prepared according to the method described by Waisser et al. [14].

Mannich Reaction

General Procedure. To the suspension of tetrazole **1** (1.78 g, 0.01 mol) in ethyl alcohol (30 mL, absolute), formaldehyde (30%, 0.9 mL, 0.03 mol), and the desired secondary amine (0.01 mol) were added dropwise with stirring. The reaction mixture was further stirred for 1–2 h at room temperature and was concentrated under reduced pressure and cooled. The solid obtained was filtered and crystallized from ethyl alcohol.

1-(Morpholin-4-yl-methyl)-4-phenyl-1,4-dihydro-5H-tetrazole-5-thione (2a). (Amine = morpholine), white powder (2.1 g, 76%), mp 154°C. ¹H NMR (CDCl₃): δ = 8.03–7.91 (m, 2H, Ar-H), 7.66–7.47 (m, 3H, Ar-H), 5.33 (s, 2H, CH₂), 3.71 (t, 4H, $J = 4.94$ Hz, 2OCH₂), 2.89 (t, 4H, $J = 4.94$ Hz, 2NCH₂). (MALDI, positive mode, matrix: DHB): m/z = 300.3 (M + Na)⁺. C₁₂H₁₅N₅OS (277.3): C, 51.97; H, 5.45; N, 25.25; S, 11.56. Found: C, 51.76; H, 5.24; N, 25.12; S, 11.43.

1-Phenyl-4-(piperidin-1-yl-methyl)-1,4-dihydro-5H-tetrazole-5-thione (2b). (Amine = piperidine), white powder (1.8 g, 66%), mp 112°C. ¹H NMR (CDCl₃): δ = 8.01–7.84 (m, 2H, Ar-H), 7.65–7.44 (m, 3H, Ar-H), 5.32 (s, 2H, CH₂), 2.78 (t, 4H, $J = 5.1$ Hz, 2NCH₂), 1.69–1.54 (m, 4H, 2CH₂), 1.47–1.32 (m, 2H, CH₂). (MALDI, positive mode, matrix: DHB): m/z = 298.4 (M + Na)⁺. C₁₃H₁₇N₅S (275.4): C, 56.70; H,

6.22; N, 25.43; S, 11.64. Found: C, 56.64; H, 6.21; N, 25.33; S, 11.52.

1-[(4-Methylpiperazin-1-yl)methyl]-4-phenyl-1,4-dihydro-5H-tetrazole-5-thione (2c). (Amine = *N*-methyl piperazine), white powder (2.5 g, 86%), mp 139°C. ¹H NMR (CDCl₃): δ = 8.01–7.86 (m, 2H, Ar-H), 7.62–7.35 (m, 3H, Ar-H), 5.33 (s, 2H, CH₂), 3.29–3.12 (m, 8H, 4CH₂), 2.87 (s, 3H, CH₃). (MALDI, positive mode, matrix: DHB): *m/z* = 313.4 (M + Na)⁺. C₁₃H₁₈N₆S (290.4): C, 53.77; H, 6.25; N, 28.94; S, 11.04. Found: C, 53.75; H, 6.24; N, 28.91; S, 11.04.

1,1'-[Piperazine-1,4-diyl-bis-(methylene)]-bis-(4-phenyl-1,4-dihydro-5H-tetrazole-5-thione) (3). To the suspension of tetrazole **1** (1.78 g, 0.01 mol) in absolute ethyl alcohol (30 mL), formaldehyde (30%, 0.9 mL, 0.03 mol) and the desired secondary amine (0.005 mol) were added dropwise with stirring. The reaction mixture was further stirred for 2 h at room temperature, concentrated under reduced pressure, and cooled. The solid obtained was filtered and crystallized from ethyl alcohol. White powder (3.1 g, 67%), mp 187°C. ¹H NMR (CDCl₃): δ = 7.96–7.84 (m, 4H, Ar-H), 7.61–7.42 (m, 6H, Ar-H), 5.28 (s, 4H, 2CH₂), 2.88 (s, 8H, 4CH₂). (MALDI, positive mode, matrix: DHB): *m/z* = 489.6 (M + Na)⁺. C₂₀H₂₂N₁₀S₂ (466.6): C, 51.48; H, 4.75; N, 30.02; S, 13.74. Found: C, 51.46; H, 4.69; N, 29.99; S, 13.73.

Ethyl[(1-phenyl-1H-tetrazol-5-yl)thio] Acetate (4). To a mixture of tetrazole **1** (1.78 g, 0.01 mol) and K₂CO₃ (1.38 g, 0.01 mol) in dry acetone (30 mL), ethyl chloroacetate (1.1 mL, 0.01 mol) was added. The reaction mixture was heated under reflux for 6 h, filtered, then concentrated under reduced pressure. The solid obtained was filtered and recrystallized from ethyl alcohol. White powder (2.3 g, 88%), mp 113°C. ¹H NMR (CDCl₃): δ = 7.66–7.47 (m, 5H, Ar-H), 4.31 (q, 2H, *J* = 6.9 Hz, OCH₂), 4.26 (s, 2H, SCH₂), 1.29 (t, 3H, *J* = 7.1 Hz, CH₃). (MALDI, positive mode, matrix: DHB): *m/z* = 287.3 (M + Na)⁺. C₁₁H₁₂N₄O₂S (264.3): C, 49.99; H, 4.58; N, 21.20; S, 12.13. Found: C, 49.63; H, 4.37; N, 21.11; S, 11.94.

2-[(1-Phenyl-1H-tetrazol-5-yl)-thio]Acetohydrazide (5) [14]. To a solution of the ester derivatives **4** (1.78 g, 0.01 mol) in absolute ethyl alcohol (30 mL), hydrazine hydrate (0.74 mL, 0.015 mol) was added and the reaction mixture was heated under reflux for 3 h. The reaction mixture was cooled and evaporated under reduced pressure till dried. The precipitated residue was crystallized from ethyl alcohol. White crystal (2.1 g, 69%), mp 164°C. ¹H NMR (CDCl₃):

δ = 8.23 (bs, 2H, NH₂) 7.68–7.45 (m, 5H, Ar-H), 3.98 (s, 2H, SCH₂); (MALDI, positive mode, matrix: DHB): *m/z* = 273.3 (M + Na)⁺. C₉H₁₀N₆OS (250.3): C, 43.19; H, 4.03; N, 33.58; S, 12.81. Found: C, 43.12; H, 3.84; N, 33.18; S, 12.69.

[(1-Phenyl-1H-tetrazol-5-yl)thio] Acetyl Azide (6). To a cold solution (~ -5°C) of hydrazide **5** (2.50 g, 0.01 mmol) in 1 N HCl solution (20 mL) and acetic acid 96% (3 mL), 5 mL water solution of NaNO₂ (1.38 g, 0.02 mmol) was added. The reaction mixture was stirred at 0°C for 30 min. The reaction mixture was extracted three times with 30 mL ethyl acetate. The combined yellow syrup extract was washed several times with 3% solution of NaHCO₃ and water till it became neutral and was finally dried over Na₂SO₄. The in situ generated azide **6** was used without further isolation and purification.

4-[[[(1-Phenyl-1H-tetrazol-5-ptyl)thio]] Acetamide Derivatives

General Procedure. To the previously prepared cold ethyl acetate solution of azide **6** (from 0.01 mol hydrazide), a solution of appropriate amines (0.01 mol) in ethyl acetate (20 mL) was added dropwise with stirring. The reaction mixture was kept at -5°C for 12 h, then at room temperature for another 12 h. The reaction mixture was washed with 0.5 N HCl, water and 3% solution of NaHCO₃ and dried over Na₂SO₄. The solution was evaporated to dryness, and the residue was recrystallized from ethyl acetate/petroleum ether to give the desired product.

4-[[[(1-Phenyl-1H-tetrazol-5-yl)thio]acetyl] Morpholine (7a). (Amine = morpholine), white powder (2.8 g, 92%), mp 138°C. ¹H NMR (CDCl₃): δ = 7.67–7.52 (m, 5H, Ar-H), 4.49 (s, 2H, SCH₂), 3.79–3.55 (m, 8H, 4CH₂). (MALDI, positive mode, matrix: DHB): *m/z* = 328.3 (M + Na)⁺. C₁₃H₁₅N₅O₂S (305.4): C, 51.13; H, 4.95; N, 22.94; S, 10.50. Found: C, 51.12; H, 4.95; N, 22.87; S, 10.49.

1-[[[(1-Phenyl-1H-tetrazol-5-yl)thio]acetyl] Piperidine (7b). (Amine = piperidine), white powder (2.4 g, 79%), mp 73°C. ¹H NMR (CDCl₃): δ = 7.67–7.48 (m, 5H, Ar-H), 4.53 (s, 2H, SCH₂), 3.68–3.46 (m, 4H, 2NCH₂), 1.86–1.57 (m, 6H, 3CH₂). (MALDI, positive mode, matrix: DHB): *m/z* = 326.4 (M + Na)⁺. C₁₄H₁₇N₅OS (303.4): C, 55.42; H, 5.65; N, 23.08; S, 10.57. Found: C, 55.41; H, 5.64; N, 23.08; S, 10.55.

1-Methyl-4-[[[(1-phenyl-1H-tetrazol-5-yl)thio]acetyl] Piperazine (7c). (Amine = *N*-methyl piperazine), white powder (2.2 g, 73%), mp 112°C. ¹H

NMR (CDCl₃): δ = 7.65–7.52 (m, 5H, Ar-H), 4.51 (s, 2H, CH₂), 3.74–3.61 (m, 4H, 2NCH₂), 2.51 (t, 2H, J = 4.94 Hz, CH₂NCH₃), 2.48 (t, 2H, J = 4.94 Hz, CH₂NCH₃), 2.34 (s, 3H, CH₃) (MALDI, positive mode, matrix: DHB): m/z = 341.4 (M + Na)⁺. C₁₄H₁₈N₆OS (318.4): C, 52.81; H, 5.70; N, 26.39; S, 10.07. Found: C, 52.69; H, 5.63; N, 26.29; S, 10.01.

N-(3-Hydroxypropyl)-2-[(1-phenyl-1H-tetrazol-5-yl)thio] Acetamide (**9**). (1,3-Amino propanol), white powder (1.9 g, 65%), mp 74°C. ¹H NMR (CDCl₃): δ = 7.62–7.49 (m, 5H, Ar-H), 4.06 (s, 2H, SCH₂), 3.76 (t, 1H, J = 7.1 Hz, NH), 3.64 (t, 2H, J = 7.1 Hz, OCH₂), 3.42 (q, 2H, J = 7.1 Hz, NCH₂), 2.82 (bs, 1H, OH), 1.78–1.66 (m, 2H, CH₂), CDCl₃/D₂O, δ = 7.62–7.51 (m, 5H, Ar-H), 4.06 (s, 2H, SCH₂), 3.64 (t, 2H, J = 7.1 Hz, OCH₂), 3.42 (t, 2H, J = 7.1 Hz, NCH₂), 1.78–1.66 (m, 2H, CH₂) (MALDI, positive mode, matrix: DHB): m/z = 316.3 (M + Na)⁺. C₁₂H₁₅N₅O₂S (293.3): C, 49.13; H, 5.15; N, 23.87; S, 10.93. Found: C, 49.05; H, 5.12; N, 23.52; S, 10.79.

N-(6-Hydroxyhexyl)-2-[(1-phenyl-1H-tetrazol-5-yl)thio] Acetamide (**10a**). (1,6-Aminohexanol), white powder (2.8 g, 84%), mp 216°C. CDCl₃: δ = 7.59–7.48 (m, 5H, Ar-H), 7.18 (bs, 1H, NH), 4.04 (s, 2H, SCH₂), 3.59 (t, 2H, J = 7.0 Hz, OCH₂), 3.27 (q, 2H, J = 7.1 Hz, NCH₂), 2.33 (bs, 1H, OH), 1.62–1.26 (m, 8H, 4CH₂). CDCl₃/D₂O δ = 7.59–7.48 (m, 5H, Ar-H), 4.04 (s, 2H, SCH₂), 3.59 (t, 2H, J = 7.1 Hz, OCH₂), 3.27 (t, 2H, J = 7.1 Hz, NCH₂), 1.78–1.66 (m, 2H, CH₂), (MALDI, positive mode, matrix: DHB): m/z = 358.4 (M + Na)⁺. C₁₅H₂₁N₅O₂S (335.4): C, 53.71; H, 6.31; N, 20.88; S, 9.56. Found: C, 53.66; H, 6.31; N, 20.79; S, 9.54.

N-(6-Aminohexyl)-2-[(1-phenyl-1H-tetrazol-5-yl)thio] Acetamide (**10b**). (1,6-Aminohexane), white powder (2.3 g, 69%), mp 105°C. ¹H NMR (CDCl₃): δ = 7.63–7.52 (m, 5H, Ar-H), 7.16 (bs, 1H, NH), 3.92 (s, 2H, SCH₂), 3.27 (q, 2H, J = 7.1 Hz, NCH₂), 1.58–1.29 (m, 10H, 5CH₂), (MALDI, positive mode, matrix: DHB): m/z = 357.4. (M + Na)⁺. C₁₅H₂₂N₆OS (334.4): C, 53.87; H, 6.63; N, 25.13; S, 9.59. Found: C, 53.81; H, 6.63; N, 25.06; S, 9.58.

N,N'-(Alkyldiamino)bis{2-[(1-phenyl-1H-tetrazol-5-yl)thio]acetamide}

General Procedure. To a recently generated azide solution **6** in ethyl acetate (from 0.01 mol hydrazide), a solution of the appropriate diamine (0.005 mol) in ethyl acetate (20 mL) was added dropwise with stirring. The reaction mixture was kept at –5°C for 12 h, then at room temperature for an-

other 12 h. The reaction mixture was washed with 0.5 N HCl, water, and 3% solution of NaHCO₃ and dried over Na₂SO₄. The solution was evaporated to dryness, and the residue was recrystallized by ethyl acetate/petroleum ether to give the desired product.

1,4-Bis{[(1-phenyl-1H-tetrazol-5-yl)thio]acetyl}piperazine (**8**). (Amine = piperazine), white powder (3.3 g, 63%), mp 95°C. ¹H NMR (CDCl₃): δ = 7.65–7.52 (m, 10H, Ar-H), 4.48 (s, 4H, 2CH₂), 3.92–3.81 (m, 4H, 2NCH₂), 3.75–3.63 (m, 4H, 2NCH₂). (MALDI, positive mode, matrix: DHB): m/z = 545.6 (M + Na)⁺. C₂₂H₂₂N₁₀O₂S₂ (522.6): C, 50.56; H, 4.24; N, 26.80; S, 12.27. Found: C, 50.49; H, 4.21; N, 26.77; S, 12.14.

N,N'-(Iminodihexane-6,1-diyl)bis{2-[(1-phenyl-1H-tetrazol-5-yl)thio]acetamide} (**11**). (Amine = 1,6-hexandiamine), white powder (2.8 g, 51%), mp 145°C. ¹H NMR (CDCl₃): δ = 7.63–7.49 (m, 8H, Ar-H), 7.28–7.12 (m, 3H, Ar-H, NH), 3.95 (s, 4H, 2SCH₂), 3.26 (q, 4H, J = 7.0 Hz, 2NCH₂), 1.59–1.43 (m, 8H, 2CH₂), 1.38–1.20 (m, 8H, 2CH₂), (MALDI, positive mode, matrix: DHB): m/z = 575.7 (M + Na)⁺. C₂₄H₂₈N₁₀O₂S₂ (552.7): C, 52.16; H, 5.11; N, 25.34; S, 11.60. Found: 52.13; H, 5.09; N, 25.28; S, 11.59.

Methyl N-{[(1-phenyl-1H-tetrazol-5-yl)thio]acetyl} Amino Ester

General Procedure. To a recently generated azide solution **6** in ethyl acetate (from 0.01 mol hydrazide), appropriate amino ester hydrochloride (0.01 mol) and triethyl amine (1 mL, 0.01 mol) was added. The reaction mixture was kept at –5°C for 12 h, and then at room temperature for another 12 h. The reaction mixture was washed with 0.5 N HCl, water, and 3% solution of NaHCO₃ and dried over Na₂SO₄. The solution was evaporated to dryness, and the residue was recrystallized by ethyl acetate–petroleum ether to give the desired product.

Methyl N-{[(1-phenyl-1H-tetrazol-5-yl)thio]acetyl} Glycinate (**12a**). (Amino acid = glycine), white powder (2.3 g, 68%), mp 87°C. ¹H NMR (CDCl₃): δ = 7.63–7.43 (m, 6H, Ar-H, NH), 4.08 (s, 2H, SCH₂), 4.03 (s, 2H, NHCH₂), 3.72 (s, 3H, OCH₃), (MALDI, positive mode, matrix: DHB): m/z = 330.3 (M + Na)⁺. C₁₂H₁₃N₅O₃S (307.3): C, 46.90; H, 4.26; N, 22.79; S, 10.43. Found: C, 46.89; H, 4.24; N, 22.78; S, 10.41.

Methyl N-{[(1-phenyl-1H-tetrazol-5-yl)thio]acetyl} Alaninate (**12b**). (Amino acid = L-alanine), white powder (2.5 g, 78%), mp 67°C. ¹H NMR (CDCl₃):

δ = 7.58–7.53 (m, 5H, Ar-H), 7.48 (bs, 1H, NH), 4.62–4.47 (m, 1H, CHCH₃), 4.03 (s, 2H, SCH₂), 3.71 (s, 3H, OCH₃), 1.42 (d, 3H, J = 12.0 Hz, CHCH₃), (MALDI, positive mode, matrix: DHB): m/z = 344.3 (M + Na)⁺. C₁₃H₁₅N₅O₃S (321.4): C, 48.59; H, 4.70 N, 21.79; S, 9.98. Found: C, 48.56; H, 4.68 N, 21.73; S, 9.90.

Methyl N-[(1-phenyl-1H-tetrazol-5-yl)thio]acetyl Valinate (12c). (Amino acid = L-valine), white powder (1.9 g, 55%), mp 59°C. ¹H NMR (CDCl₃): δ = 7.63–7.58 (m, 5H, Ar-H), 7.50 (bs, 1H, NH), 4.59–4.48 (m, 1H, NHCH), 4.04 (s, 2H, SCH₂), 3.72 (s, 3H, OCH₃), 2.29–2.12 (m, 1H, CHCH₃), 0.95–0.82 (m, 6H, 2CH₃), (MALDI, positive mode, matrix: DHB): m/z = 372.4 (M + Na)⁺. C₁₅H₁₉N₅O₃S (349.4): C, 51.56; H, 5.48 N, 20.04; S, 9.18. Found: C, 51.56; H, 5.47 N, 20.02; S, 9.17.

Methyl N-[(1-phenyl-1H-tetrazol-5-yl)thio]acetyl Leucinate (12d). (Amino acid = L-leucine), white powder (2.2 g, 61%), mp 51°C. ¹H NMR (CDCl₃): δ = 7.63–7.53 (m, 5H, Ar-H), 7.36 (bs, 1H, NH), 4.63–4.51 (m, 1H, NHCH), 4.08 (s, 2H, SCH₂), 3.74 (s, 3H, OCH₃), 1.68–1.52 (m, 2H, CHCH₂), 1.80–1.26 (m, 1H, CHCH₃), 1.03–0.84 (m, 6H, 2CH₃), (MALDI, positive mode, matrix: DHB): m/z = 386.4 (M + Na)⁺. C₁₆H₂₁N₅O₃S (363.4): C, 52.88; H, 5.82 N, 19.27; S, 8.82. Found: C, 52.86; H, 5.82 N, 19.24; S, 8.80.

Methyl N-[(1-phenyl-1H-tetrazol-5-yl)thio]acetyl Methioninate (12e). (Amino acid = L-methionine), yellow oil (2.0 g, 53%), ¹H NMR (CDCl₃): δ = 7.68 (bs, 1H, NH), 7.61–7.45 (m, 5H, Ar-H), 4.76–4.64 (m, 1H, NHCH), 4.08 (s, 2H, SCH₂), 3.71 (s, 3H, OCH₃), 2.51 (t, 2H, J = 7.1 Hz, SCH₂), 1.92–2.22 (m, 5H, SCH₃, CH₂CH), (MALDI, positive mode, matrix: DHB): m/z = 404.5 (M + Na)⁺. C₁₅H₁₉N₅O₃S₂ (381.5): C, 47.23; H, 5.02 N, 18.36; S, 16.81. Found: C, 47.18; H, 4.85 N, 18.26; S, 16.79.

N²-[(1-Phenyl-1H-tetrazol-5-yl)thio]acetyl Glycine Hydrazide (13a). (Amino acid = glycine), white powder (2.8 g, 91%), mp 189°C. ¹H NMR (CDCl₃): δ = 9.05 (bs, 1H, NH), 8.60 (bs, 1H, NH), 7.63–7.55 (m, 5H, Ar-H), 4.24 (s, 2H, SCH₂), 3.66 (bs, 2H, NH₂), 3.33–3.23 (m, 2H, NHCH₂), (MALDI, positive mode, matrix: DHB): m/z = 330.3 (M + Na)⁺. C₁₁H₁₃N₇O₂S (307.3): C, 42.99; H, 4.26 N, 31.90; S, 10.43. Found: 42.85; H, 4.19 N, 31.87; S, 10.38.

N²-[(1-Phenyl-1H-tetrazol-5-yl)thio]acetyl Alanine Hydrazide (13b). (Amino acid = L-alanine), white powder (2.6 g, 80%), mp 163°C. ¹H NMR (CDCl₃): δ = 9.18 (bs, 1H, NH), 8.66 (bs, 1H, NH), 7.71–7.57 (m, 5H, Ar-H), 4.31–4.15 (s, 3H, SCH₂,

NHCH), 3.49 (bs, 2H, NH₂), 1.29–1.16. (m, 3H, CHCH₃), (MALDI, positive mode, matrix: DHB): m/z = 344.4 (M + Na)⁺. C₁₂H₁₅N₇O₂S (321.4): C, 44.85; H, 4.70 N, 30.51; S, 9.98. Found: C, 44.61; H, 4.63 N, 30.49; S, 9.87.

Methyl N-[(1-phenyl-1H-tetrazol-5-yl)thio]acetyl Glycylglycinate (14). White powder (2.7 g, 74%), mp 150°C. ¹H NMR (CDCl₃): δ = 7.70 (bs, 1H, NH), 7.58–7.53 (m, 5H, Ar-H), 7.33 (bs, 1H, NH), 4.14–4.08 (m, 4H, 2NHCH₂), 4.01 (s, 2H, SCH₂), 3.71 (s, 3H, OCH₃), (MALDI, positive mode, matrix: DHB): m/z = 387.4 (M + Na)⁺. C₁₄H₁₆N₆O₄S (364.4): C, 46.15; H, 4.43 N, 23.06; S, 8.80. Found: C, 46.13; H, 4.37 N, 23.04; S, 8.78.

REFERENCES

- [1] Thornber, C. W. *Chem Soc Rev* 1979, 8, 563–580.
- [2] Butler, R. N. In *Comprehensive Heterocyclic Chemistry II*; Katritzky, A. R.; Rees, C. W.; Scriven, E. F. V. (Eds.); Pergamon: Oxford, 1996; Vol. 4, p. 621.
- [3] Herr, R. J. *Bioorg Med Chem* 2002, 10, 3379–3393.
- [4] Lipinski, C. A. *Annu Rep Med Chem* 1986, 27, 283–291.
- [5] Noda, K.; Saad, Y.; Kinoshita, A.; Boyle, T. P.; Graham, R. M.; Husain, A.; Karnik, S. S. *J Biol Chem* 1995, 270, 2284–2289.
- [6] Goldgur, Y.; Craigie, R.; Cohen, G. H.; Fujiwara, T.; Yoshinaga, T.; Fujishita, T.; Sugimoto, H.; Endo, T.; Murai, H.; Davies, D. R. *Proc Natl Acad Sci USA* 1999, 96, 13040–13043.
- [7] Adamec, J.; Waissner, K.; Kunes, J.; Kaustova, J. *Arch Pharm Chem Life Sci* 2005, 338, 385–389.
- [8] Nakamura, H.; Shimoda, A.; Ishii, K.; Kadokawa, T. *Arch Int Pharma-codyn* 1986, 282, 16–25.
- [9] Çakir, B.; Dogruer, D.; Yesilada, E.; Sahin, M. F. *J Fac Pharm Gazi* 1997, 14, 92–98.
- [10] Dogruer, D.; Ünlü, S.; Yesilada, E.; Sahin, M. F. *Farmaco* 1997, 52, 745–750.
- [11] El-Tamany, S. H.; Abd El-Fattah, M.; Ibrahim, A. I.; Salem, M. E. *J Indian Chem Soc* 1997, 74, 772–776.
- [12] Fathalla, W.; Čajan, M.; Pazdera, P. *Molecules* 2000, 5, 1210–1223.
- [13] Fathalla, W.; Čajan, M.; Pazdera, P. *Molecules* 2001, 6, 557–573.
- [14] Waissner, K.; Vanzura, J.; Hrabalek, A.; Vinsova, J.; Gresak, S. *Collect Czech Chem Commun* 1991, 56, 2389–2394.
- [15] Davis, J. S.; Mohammed, A. K. *J Chem Soc, Perkin Trans I* 1981, 2982–2990, references therein.
- [16] König, W.; Geiger, R. *Chem Ber* 1970, 103, 788–798.
- [17] Pennigton, R. M.; Fischer, R. R. *J Biol Chem* 1981, 256, 8963–8969.
- [18] Sahin, G.; Palaska, E.; Ekizoglu, M.; Özalp, M. *Farmaco* 2002, 57, 539–542.
- [19] Ali, I. A. I.; Al-Masoudi, I. A.; Saeed, B., Al-Masoudi, N. A.; La Colla, P. *Heteroatom Chem* 2005, 16, 148–155.
- [20] Al-Masoudi, N. A.; Al-Masoudi, I. A.; Ali, I. A. I.; Saeed, B.; La Colla, P. *Heteroatom Chem* 2005, 16, 576–586.