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SYNTHESIS OF SUBSTITUTED 5,6-DIHYDRO-4H-[1,2,4]TRIAZOLO-[4,3-*a*][1,5]BENZODIAZEPINES

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A one-pot synthetic approach to the novel 5,6-dihydro-4H-[1,2,4]triazolo[4,3-a][1,5]benzodiazepines by thermal cyclization of 4-acylhydrazino-2,3-dihydro-1H-1,5-benzodiazepines is described.

Keywords: 4-acylhydrazino-2,3-dihydro-1H-1,5-benzodiazepines, 4H-[1,2,4]triazolo[4,3-*a*][1,5]benzodiazepines, cyclization.

Benzodiazepines and their polycyclic derivatives are important medicinal agents for the treatment of CNS disturbances [1-4]. Moreover, some annulated benzodiazepines, *e.g.*, naturally occurring pyrrolo[2,1-*c*][1,4]benzodiazepines such as anthramycin [5], thiazolo[4,3-*c*][1,4]benzodiazepines [6] and substituted chromeno[4,3-*b*][1,5]benzodiazepine stereoisomers [7] have been found to exhibit antitumor and antineoplastic activities. The majority of these compounds have common general structures, comprising a tri- or tetracyclic almost planar chromophore [8]. In view of these benefits and our interest in the study of novel compounds bearing potential biological activity, our research group became interested in triazolo[4,3-*a*][1,5]benzodiazepine derivatives. We report here the synthesis and the physicochemical properties of new compounds.

The preparation of some analogous target compounds was first reported by E. Szarvasi [9, 10] through the one-step cyclocondensation procedure starting from the corresponding tetrahydro-1,5-benzodiazepine-2(1H)-thiones and arylhydrazides. This procedure involved heating the reactants at 200°C or refluxing them in higher boiling solvents (*e.g.*, trimethylbenzene). However, this approach can be restricted by the lack of reactivity of the thiolactams towards weak nucleophiles [11]. Besides, it was proved that the formation of the triazole ring in the 1,4-benzodiazepine series occurred through the intermediates obtained by phosphorylation with the moisture sensitive di(4-morpholinyl)phosphinic chloride (commercially not available) of lactams and subsequent reaction of an intermediate imine with hydrazides [2].

In the present work we used a procedure based on the thermal cyclization of 4-acylhydrazino-2,3-dihydro-1H-1,5-benzodiazepines (**1a**-**m**) for the synthesis of the desired tricyclic compounds of the general formula **2**. The procedure herein reported was chosen after a number of experiments in order to obtain the highest yields of the tricyclic derivatives. As a rule, the cyclization of **1** was achieved in refluxing anhydrous ethanol or butanol. Dehydration of 4-methyl-substituted derivatives **1l**,**m** was performed only at a higher temperature by refluxing in xylene and removing the formed water. Generally, it was demonstrated that cyclization of the starting materials proceeded within a separate time limit for each compound and depended on its structure, as well as on the solubility in an appropriate solvent. An attempt to prepare triazole **2e** from the corresponding thiolactam [12] and an excess of the acetylhydrazine in refluxing tetrahydrofuran in the presence of mercuric chloride [13] led to the mercury-benzodiazepine compound. The formation of a metal complex in

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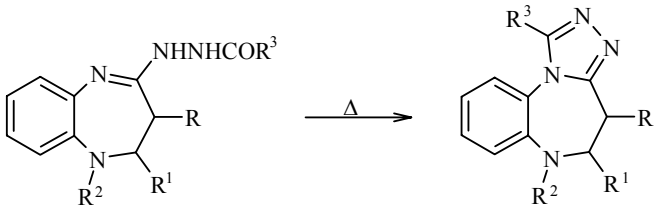
the nucleophilic substitution reaction of tetrahydro-1,5-benzodiazepinethiones with amines was also observed [14]. The key starting materials **1a-m** were prepared from the corresponding 4-methylthio-2,3-dihydro-1H-1,5-benzodiazepines and acetyl- or benzoylhydrazines, as previously described [15]. According to this procedure compounds **1b**,**d**,**f**,**g**,**k**,**l** have been obtained for the first time.



1a–m **2a–m**

1, 2	R	R^1	R^2	R^3
a	H	H	H	Me
b	H	H	H	Ph
c	H	H	CH_2Ph	Me
d	H	H	CH_2Ph	Ph
e	Me	H	H	Me
f	Me	H	H	Ph
g	Me	H	Me	Ph
h	Me	H	CH_2Ph	Me
i	Me	H	CH_2Ph	Ph
j	H	Me	H	Me
k	H	Me	H	Ph
l	H	Me	CH_2Ph	Me
m	H	Me	CH_2Ph	Ph

The structures of new compounds **1** and **2** were assigned by analytical and spectral data. For triazolobenzodiazepines **2a-m**, the IR spectra are characterized by C=N stretching absorption bands at 1600-1520 cm^{-1}. The ^1H NMR spectra of these compounds, registered in $CDCl_3$, exhibit signals corresponding to the diazepine skeleton protons. It can be noted that the $C_{(1)}$–CH_3 signals of **2a**,**c**,**e**,**h**,**j**,**l** ($R^3 = CH_3$) appear as sharp singlets at 2.45-2.51 ppm, thus indicating the formation of the triazole ring.

The *in vitro* anticancer activity of some newly synthesized tricyclic compounds has been tested at the National Cancer Institute (NCI, USA). According to the NCI's Developmental Therapeutics Program the compounds **2c**,**e**,**h**,**j,k** have been evaluated in the 3-cell line, one dose primary anticancer assay. Only the compound **2c** which passed criteria for activity in this assay has been scheduled for testing against the full panel of 60 human tumor cell lines. The compound **2c**, bearing a lipophilic benzyl substituent at the $N_{(6)}$ atom of the tricyclic heterosystem, exhibited noteworthy activity and remarkable selectivity for leukemia and breast cancer cell lines.

EXPERIMENTAL

Melting points were determined in open glass capillaries and are not corrected. IR spectra were recorded on a Specord IR-75 spectrometer. ^1H NMR spectra were measured at 80 MHz on a Tesla BS-587A spectrometer with TMS as an internal reference. TLC analyses were carried out on Silufol UV-254 silica gel plates in the butanol–acetic acid–water, 4:2:1, system. Compounds **1a,c,e,h-j,m** were synthesized following the reported method [15].

TABLE 1. Physical and Analytical Data of the Newly Synthesized Compounds

Compound	Reaction time, h	Empirical formula (MW)	Found, % / Calculated, %			mp, °C (solvent)	Yield*, %
			C	H	N		
1b	—	$C_{16}H_{16}N_4O$ (280.33)	68.34 / 68.55	5.59 / 5.75	20.23 / 19.98	199-200 (EtOH)	57
1d	—	$C_{23}H_{22}N_4O$ (370.45)	74.69 / 74.57	5.81 / 5.99	15.22 / 15.12	206-208 (EtOH)	87
1f	—	$C_{17}H_{18}N_4O$ (294.36)	69.51 / 69.37	6.26 / 6.16	19.23 / 19.03	166-168 (EtOH)	48
1g	—	$C_{18}H_{20}N_4O$ (308.38)	70.18 / 70.11	6.36 / 6.54	18.21 / 18.17	192-195 (EtOH)	70
1k	—	$C_{17}H_{18}N_4O$ (294.36)	69.66 / 69.37	6.02 / 6.16	19.15 / 19.03	174-176 (EtOH)	74
1l	—	$C_{24}H_{24}N_4O$ (384.48)	75.12 / 74.98	6.17 / 6.29	14.42 / 14.57	178-181 (EtOH)	75
2a	7	$C_{11}H_{12}N_4$ (200.24)	65.77 / 65.98	5.91 / 6.04	28.02 / 27.98	174-176 (EtOAc)	41
2b	10	$C_{16}H_{14}N_4$ (262.32)	73.39 / 73.26	5.43 / 5.38	21.31 / 21.36	183-185*[2] (EtOAc)	49
2c	20	$C_{18}H_{18}N_4$ (290.37)	74.29 / 74.46	6.51 / 6.25	19.27 / 19.29	138-139 (EtOAc)	82
2d	8	$C_{23}H_{20}N_4$ (352.44)	76.66 / 76.80	6.02 / 6.14	16.91 / 17.06	146-147 (EtOAc)	70
2e	5	$C_{12}H_{14}N_4$ (214.27)	67.38 / 67.27	6.91 / 6.59	26.30 / 26.15	186-188 (EtOAc)	65
2f	8	$C_{17}H_{16}N_4$ (276.34)	74.12 / 73.89	6.01 / 5.84	20.39 / 20.27	240-242*[3] (EtOH)	71
2g	8	$C_{18}H_{18}N_4$ (290.37)	74.67 / 74.46	6.53 / 6.25	19.20 / 19.29	162-165 (Et_2O)	74
2h	28	$C_{19}H_{20}N_4$ (304.40)	74.95 / 74.97	6.93 / 6.62	18.59 / 18.40	120-122 (EtOAc)	58
2i	15	$C_{24}H_{22}N_4$ (366.47)	78.74 / 78.66	6.17 / 6.05	15.42 / 15.29	174-176 (EtOAc)	69
2j	25	$C_{12}H_{14}N_4$ (214.27)	67.49 / 67.27	6.79 / 6.59	26.26 / 26.15	194-196 (EtOH)	62
2k	18	$C_{17}H_{16}N_4$ (276.34)	73.61 / 73.89	6.12 / 5.84	19.93 / 20.27	134-136 (EtOH)	65
2l	6	$C_{19}H_{20}N_4$ (304.40)	75.11 / 74.97	6.47 / 6.62	18.32 / 18.40	128-130 (EtOAc)	84
2m	16	$C_{24}H_{22}N_4$ (366.47)	78.53 / 78.66	6.00 / 6.05	15.20 / 15.29	190-193 (EtOAc)	86

* Yields are based on purified products.

*[2] Lit. [9] 183-185°C.

*[3] Lit. [9] 239-240°C.

5,6-Dihydro-4-R-5-R'-6-R"-4H-[1,2,4]triazolo[4,3-*a*][1,5]benzodiazepines (2a-m) (General Procedure). A suspension of the appropriate acylhydrazinobenzodiazepine **1a-m** (5.0 mmol) in 60 ml of anhydrous EtOH (for compounds **1a**,**b**,**h** in 50 ml of anhydrous BuOH and for compounds **1l**,**m** in 50 ml of *o*-xylene employing Dean–Stark trap) was heated at reflux. After a while the reaction mixture became clear and refluxing was continued. The reaction was monitored by TLC analysis. When the starting material was no

TABLE 2. IR and ^{1}H NMR Spectral Data for the Newly Synthesized Compounds

Com-pound	IR (nujol), ν, cm^{-1}	^{1}H NMR, δ, ppm (*J*, Hz)*
1b	3350, 3180, 3135 (NH), 1660 (CO)	2.55 (2H, m, CH_2); 3.50 (2H, m, CH_2N); 5.40 (1H, s, $NHCH_2$); 6.80-8.10 (9H, m, H–Ar); 8.35 (1H, br. s, NH); 9.98 (1H, br. s, NH)
1d	3280, 3185 (NH), 1665 (CO)	2.43 (2H, m, CH_2); 3.34 (2H, m, CH_2N); 4.32 (2H, s, CH_2Ph); 6.85-8.10 (14H, m, H–Ar); 8.71 (1H, br. s, NH); 10.21 (1H, br. s, NH)
1f	3325, 3220 (NH), 1655 (CO)	1.12 (3H, d, CH_3); 2.30-3.80 (3H, m, CH_2CH); 5.44 (1H, br. s, $NHCH_2$); 6.80-8.05 (9H, m, H–Ar); 8.27 (1H, br. s, NH); 10.01 (1H, br. s, NH)
1g	3300, 3220 (NH), 1660 (CO)	1.10 (3H, d, CH_3CH); 2.30-3.80 (3H, m, CH_2CH); 2.72 (2H, s, CH_3); 6.85-8.05 (9H, m, H–Ar); 8.60 (1H, br. s, NH); 10.10 (1H, s, NH)
1k	3270, 3175 (NH), 1660 (CO)	1.20 (3H, d, CH_3); 2.00-2.60 (2H, m, CH_2); 3.82 (1H, m, CH); 5.04 (1H, br. s, NHCH); 6.85-8.05 (9H, m, H–Ar); 10.21 (2H, br. s, NHNH)
1l	3280, 3180, 3150 (NH), 1645 (CO)	1.01 (3H, d, CH_3CH); 1.93 and 2.03 (3H, two s, CH_3); 2.00-2.80 (2H, m, CH_2); 3.67 (1H, m, CH); 4.26 (1H, AB-q, *J* = 15.2, CH_2Ph); 4.37 (1H, AB-q, *J* = 15.2, CH_2Ph); 6.80-7.40 (9H, m, H–Ar); 8.34, 8.43, 9.58, 9.72 (2H, four br. s, NHNH)
2a	3260 (NH), 1600, 1585, 1550	2.51 (3H, s, CH_3); 3.04 (2H, m, CH_2); 3.49 (1H, br. s, NH); 3.72 (2H, m, CH_2N); 6.85-7.50 (4H, m, H–Ar)
2b	3300 (NH), 1600, 1580, 1525	3.11 (2H, m, CH_2); 3.50 (1H, br. s, NH); 3.78 (2H, m, CH_2N); 6.65-7.55 (9H, m, H–Ar)
2c	1600, 1535	2.46 (3H, s, CH_3); 2.88 (2H, m, CH_2); 3.35 (2H, m, CH_2N); 4.21 (2H, s, CH_2Ph); 6.85-7.40 (9H, m, H–Ar)
2d	1600, 1575, 1535	3.03 (2H, m, CH_2); 3.52 (2H, m, CH_2N); 4.36 (2H, s, CH_2Ph); 6.65-7.55 (14H, m, H–Ar)
2e	3255 (NH), 1605, 1535	1.49 (3H, d, CH_3CH); 2.50 (3H, s, CH_3); 3.00 (1H, br. s, NH); 3.06 (1H, m, CH); 3.15-3.85 (2H, m, CH_2); 6.90-7.40 (4H, m, H–Ar)
2f	3250, 3175 (NH), 1660, 1520	1.52 (3H, d, CH_3CH); 2.95-3.90 (3H, m, CH_2CH); 3.81 (1H, br. s, NH); 6.65-7.50 (9H, m, H–Ar)
2g	1600, 1535, 1525	1.53 (3H, d, CH_3); 2.70-3.80 (3H, m, CH_2CH); 2.86 (3H, s, CH_3N); 6.60-7.45 (9H, m, H–Ar)
2h	1600, 1535, 1525	1.38 (3H, d, CH_3CH); 2.51 (3H, s, CH_3); 2.70-3.31 (3H, m, CH_2CH); 4.07 (1H, AB-q, *J* = 14.8, CH_2); 4.36 (1H, AB-q, *J* = 14.8, CH_2); 6.95-7.30 (9H, m, H–Ar)
2i	1595, 1520	1.49 (3H, d, CH_3); 2.70-3.80 (3H, m, CH_2CH); 4.18 (1H, AB-q, *J* = 14.8, CH_2); 4.47 (1H, AB-q, *J* = 14.8, CH_2); 6.60-7.50 (14H, m, H–Ar)
2j	3270 (NH), 1600, 1535	1.26 (3H, d, CH_3CH); 2.48 (3H, s, CH_3); 2.72 (1H, dd, *J* = 6.3, 14.5, CH_2); 3.07 (1H, dd, *J* = 5.4, 14.5, CH_2); 3.37 (1H, br. s, NH); 3.98 (1H, m, CH); 6.90-7.35 (4H, m, H–Ar)
2k	3240 (NH), 1600, 1640	1.27 (3H, d, CH_3CH); 2.70 (1H, dd, *J* = 6.6, 14.6, CH_2); 3.07 (1H, dd, *J* = 5.2, 14.6, CH_2); 3.74 (1H, br. s, NH); 4.03 (1H, m, CH); 6.60-7.50 (9H, m, H–Ar)
2l	1600, 1545, 1530	1.14 (3H, d, CH_3CH); 2.28 (1H, dd, *J* = 10.4, 14.3, CH_2C=); 2.45 (3H, s, CH_3); 3.24 (1H, dd, *J* = 6.8, 14.8, CH_2C=); 3.87 (1H, m, CH); 4.17 (1H, AB-q, *J* = 15.2, CH_2Ph); 4.35 (1H, AB-q, *J* = 15.2, CH_2Ph); 6.70-7.40 (9H, m, H–Ar)
2m	1595, 1535	1.24 (3H, d, CH_3CH); 2.40 (1H, dd, *J* = 11.2, 14.4, CH_2C=); 3.38 (1H, dd, *J* = 6.0, 14.4, CH_2C=); 3.98 (1H, m, CH); 4.24 (1H, AB-q, *J* = 14.4, CH_2Ph); 4.54 (1H, AB-q, *J* = 14.4, CH_2Ph); 6.70-7.40 (14H, m, H–Ar)

* Solvents: compounds **1b**,**d**,**f**,**g**,**k**,**l** recorded in DMSO-d_6, **2a-m** in $CDCl_3$.

longer detectable (reaction time for each material is presented in Table 1) the work up of the reaction mixture for the separate compound was performed differently. The mixture was concentrated by evaporation under reduced pressure to 20 ml and left to cool. The precipitated product (**2d**,**f**,**j**) was filtered. In the case of compounds **2a**,**b**,**g-i**,**k** the solvent was evaporated to dryness. The thick residue was triturated with diethyl ether and the resultant product was filtered. After the reaction solvent was evaporated to dryness, the compounds **2l,m** were obtained as solids. The dark reaction mixture (compound **2c,e**) after being cooled to room temperature was filtered through a short plug of silica gel (Chemapol L 40/100, 8 g). After the column was eluted with ethanol (20 ml), the combined organic fractions were evaporated to give a solid residue. Compounds **2a-m** were obtained as white or light yellow crystals by recrystallization from the proper solvent.

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