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SYNTHESIS OF SOME NON-PROTEIN AMINO ACIDS DERIVED FROM SPIROHYDANTOINS

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Abstract: *This article presents a synthesis of non-protein amino acids derived from 6- and 8- substituted cyclohexanespiro-5-hydantoins, spiro(adamantane-2',4'-imidazolidine)-2,5-dione and 3',4'-dihydro-2H,2'H,5H-spiro[imidazolidine-4,1'-naphthalene]-2,5-dione. The target compounds were prepared by an alkaline hydrolysis of the corresponding spirohydantoins with barium hydroxide. The products obtained were characterized by physicochemical parameters, IR , ^{1}H and ^{13}C NMR spectral data.*

Key words: *Synthesis, Non-protein Amino Acids, Spirohydantoins*

I. Introduction

Different types of biological activity of non-protein cyclic amino acids have been established. One of the most important properties in this respect is the antitumor activity manifested by some representatives of such compounds [1-7].

Recently, the fungicidal [8] and insecticidal [9, 10] potential of 1-aminocyclopentane-1-carboxylic acid have been showed by us.

Among the known methods for the synthesis of non-protein amino acids, one of the most convenient and widely used procedure is the hydrolysis of hydantoins and spirohydantoins, resulting in the hydantoin ring degradation and the formation of $C^{\alpha,\alpha}$- symmetrically and asymmetrically disubstituted glycines [11].

In a previous study of ours, 1-aminocycloalkanecarboxylic acids with five-, six-, seven-, eight- and twelve-membered ring have been obtained by an alkaline hydrolysis of the corresponding cycloalkanespiro-5-hydantoins with barium hydroxide [12].

The purpose of this study is the synthesis of 2- and 4- substituted 1-aminocyclohexane-1-carboxylic acids, 2-aminoadamantane-2-carboxylic acid and 1-amino-1,2,3,4-tetrahydronaphthalene-1-carboxylic acid. The target non-protein cyclic amino acids are obtained from the relevant spirohydantoins by an application of the above cited technique [12].

II. Experimental

II.1. General

All chemicals used were purchased from Merck and Sigma-Aldrich. The melting points were determined with a Koffler apparatus. The elemental analysis data were obtained with an automatic analyzer Carlo Erba 1106. The purity of the compounds was checked by thin layer chromatography on Kieselgel 60 F_{254}, 0.2 mm Merck plates, eluent system (vol. ratio): n-butanol : glacial acetic acid : water = 4 : 1 : 5. The IR spectra were taken on Perkin-Elmer FTIR-1600 spectrometer in KBr discs. The NMR spectra were taken on a Bruker DRX-250 spectrometer, operating at 250.13 and 62.90 MHz for ^{1}H and ^{13}C, respectively, using the standard Bruker software. The chemical shifts were referenced to tetramethylsilane (TMS). The measurements in DMSO-d_6 + D_2O solutions were carried out at ambient temperature (300 K). Typical conditions for ^{1}H NMR spectra were: pulse width 30º, 1 s relaxation delay, 16K time domain points, zero-filled to 64K, hard pulse with 90º pulse width of 11.8 μs; ^{13}C NMR spectra: pulse width 30º, 1 s relaxation delay, 16K time domain points, zero-filled to 32K, hard pulse with 90º pulse width of 6.4 μs at a power level of 3 dB below the maximum output.

II.2. Synthesis of non-protein amino acids (2a-2f, Scheme 1) [12]

A suspension of 0.01 mol of the corresponding spirohydantoin (**1a-1f**) and 0.019 mol of $Ba(OH)_2.8H_2O$ in 40 ml water was heated at 160 °C in an autoclave for two hours. The reaction mixture was cooled to room temperature, filtered and the filtrate was treated with 0.021 mol of $(NH_4)_2CO_3$. The resulting solution was filtered, concentrated and the corresponding amino acid (**2a-2f**) crystallized after cooling. The compounds obtained were recrystallized from methanol.

III. Results and discussion

The non-protein amino acids (**2a-2f**) were obtained by an alkaline hydrolysis of the corresponding spirohydantoins (**1a-1f**) with barium hydroxide (Scheme 1), following the procedure previously reported by us [12]. The 6- and 8- substituted cyclohexanespiro-5-hydantoins (**1a-1d**, Scheme 1) were prepared *via* the Bucherer-Lieb method [13]. The synthesis was carried out by an interaction between the corresponding 2- and 4- substituted cyclohexanone,

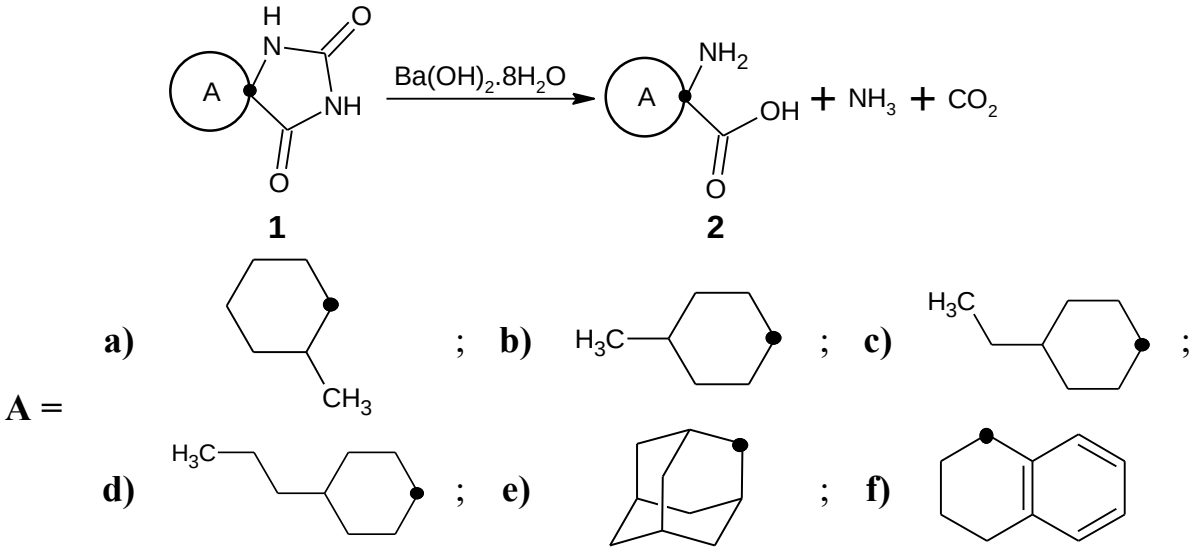
sodium cyanide, ammonium carbonate and ethanol. As a result of that reaction, the following compounds were obtained [14]:

- 6-methyl-1,3-diazaspiro[4.5]decane-2,4-dione (**1a**);
- 8-methyl-1,3-diazaspiro[4.5]decane-2,4-dione (**1b**);
- 8-ethyl-1,3-diazaspiro[4.5]decane-2,4-dione (**1c**);
- 8-propyl-1,3-diazaspiro[4.5]decane-2,4-dione (**1d**).

The spiro(adamantane-2,4'-imidazolidine)-2',5'-dione (**1e**) was obtained in accordance with Nagasawa et al. [15].

The 3',4'-dihydro-2*H*,2'*H*,5*H*-spiro[imidazolidine-4,1'-naphthalene]-2,5-dione (**1f**) was synthesized in accordance with Marinov et al. [16].

The spirohydantoins obtained (**1a-1f**) were subjected to an alkaline hydrolysis with barium hydroxide (see the Experimental part), resulting in the formation of the corresponding non-protein amino acids (**2a-2f**). The latter were characterized by physicochemical parameters, elemental analysis, IR and NMR spectral data. The results obtained from these analyses are listed in Tables 1-4 respectively.



Scheme 1. *Synthesis of non-protein amino acids*

Table 1. *Physicochemical parameters of compounds* ***2a-2f***

Compound[1]	Systematic name	Yield, %	M. p., °C	R_f[2]
2a	1-amino-2-methylcyclohexane-1-carboxylic acid	65	> 300 °C[3]	0.41
2b	1-amino-4-methylcyclohexane-1-carboxylic acid	77	> 300 °C[4]	0.48

Table 1. *Physicochemical parameters of compounds* ***2a-2f*** *(continuation)*

Compound[1]	Systematic name	Yield, %	M. p., ºC	R_f[2]
2c	1-amino-4-ethylcyclohexane-1-carboxylic acid	68	> 300 ºC	0.43
2d	1-amino-4-propylcyclohexane-1-carboxylic acid	64	> 300 ºC	0.42
2e	2-aminoadamantane-2-carboxylic acid	80	> 300 ºC[5]	0.36
2f	1-amino-1,2,3,4-tetrahydronaphthalene-1-carboxylic acid	86	253-254 ºC[6]	0.45

[1] The compounds numbering is in accordance with Scheme 1.
[2] Eluent system (vol. ratio): n-butanol : glacial acetic acid : water = 4 : 1 : 5.
[3] Ref. [17]: M. p. > 300 ºC.
[4] Ref. [18]: M. p. > 300 ºC.
[5] Ref. [15]: M. p. 308-310 ºC.
[6] Ref. [19]: M. p. 244-245 ºC for *rac*-**2f**.

Table 2. *Elemental analysis data of compounds* ***2a-2f***

Compound	Molecular formula	Elemental analysis, %					
		Calculated			Found		
		C	H	N	C	H	N
2a	$C_8H_{15}NO_2$	61.12	9.62	8.91	60.89	9.47	8.84
2b	$C_8H_{15}NO_2$	61.12	9.62	8.91	60.96	9.55	8.81
2c	$C_9H_{17}NO_2$	63.13	10.01	8.18	62.95	9.98	7.97
2d	$C_{10}H_{19}NO_2$	64.83	10.34	7.56	64.75	10.19	7.53
2e	$C_{11}H_{17}NO_2$	67.66	8.78	7.17	67.48	8.64	7.05
2f	$C_{11}H_{13}NO_2$	69.09	6.85	7.32	68.93	6.75	7.22

Table 3. *IR spectral data of compounds* ***2a-2f***

Compound	IR (KBr, cm^{-1})
2a	3026, 2955, 2868, 2928, 2568, 2077, 1615
2b	3035, 2961, 2871, 2931, 2570, 2075, 1616
2c	3040, 2959, 2870, 2929, 2921, 2571, 2081, 1598
2d	3044, 2957, 2872, 2933, 2927, 2566, 2084, 1595
2e	3031, 2926, 2564, 2081, 1562
2f	3028, 2931, 2571, 2085, 1573

Table 4. *NMR spectral data of compounds* ***2a-2f***

№	^{1}H NMR (DMSO-d_6 + D_2O), δ / ppm	^{13}C NMR (DMSO-d_6 + D_2O), δ / ppm	^{13}C DEPT 135 (DMSO-d_6 + D_2O), δ / ppm
2a	1.07 (s, CH_3), 1.39-1.90 (m, 8H, CH_2), 2.15 (s, H, CH), 8.28 (s, 2H, NH_2), 10.9 (s, H, OH)	11.8 (CH_3), 19.5-31.8 (CH_2), 36.1 (CH), 67.2 (C^1), 180.8 (C=O)	11.8 (CH_3), 19.5-31.8 (CH_2), 36.1 (CH)
2b	1.04 (s, CH_3), 1.40-1.88 (m, 8H, CH_2), 1.60 (s, H, CH), 8.33 (s, 2H, NH_2), 10.8 (s, H, OH)	19.6 (CH_3), 26.0-32.2 (CH_2), 28.6 (CH), 61.3 (C^1), 181.2 (C=O)	19.6 (CH_3), 26.0-32.2 (CH_2), 28.6 (CH)
2c	0.95 (s, CH_3), 1.28 (s, CH_2), 1.38-1.91 (m, 8H, CH_2, cyclohex. ring), 1.41 (s, CH), 8.42 (s, 2H, NH_2), 11.2 (s, H, OH)	12.1 (CH_3), 23.5-32.4 (CH_2, cyclohex. ring), 27.7 (CH_2), 34.6 (CH), 59.9 (C^1), 181.4 (C=O)	12.1 (CH_3), 23.5-32.4 (CH_2, cyclohex. ring), 27.7 (CH_2), 34.6 (CH)

Table 4. *NMR spectral data of compounds* ***2a-2f*** *(continuation)*

№	**^{1}H NMR (DMSO-d_6 + D_2O), δ / ppm**	**^{13}C NMR (DMSO-d_6 + D_2O), δ / ppm**	**^{13}C DEPT 135 (DMSO-d_6 + D_2O), δ / ppm**
2d	0.97 (s, CH_3), 1.23 (s, CH_2), 1.31 (s, CH_2), 1.39-1.88 (m, 8H, CH_2, cyclohex. ring), 1.42 (s, CH), 8.45 (s, 2H, NH_2), 11.6 (s, H, OH)	14.5 (CH_3), 21.1 (CH_2), 37.3 (CH_2), 24.1-32.6 (CH_2, cyclohex. ring), 31.3 (CH), 60.5 (C^1), 182.2 (C=O)	14.5 (CH_3), 21.1 (CH_2), 37.3 (CH_2), 24.1-32.6 (CH_2, cyclohex. ring), 31.3 (CH)
2e	1.37 (s, 10H, CH_2), 1.42 (s, 2H, CH), 1.99 (s, 2H, CH), 8.11 (s, 2H, NH_2), 11.5 (s, H, OH)	28.6 (CH), 29.9 (CH_2), 35.6 (CH), 63.4 (C^2), 181.7 (C=O)	28.6 (CH), 29.9 (CH_2), 35.6 (CH)
2f	1.57-2.87 (m, 6H, CH_2), 6.92-7.03 (m, 4H, CH), 8.27 (s, 2H, NH_2), 11. 8 (s, H, OH)	26.8-42.2 (CH_2), 65.1 (C^1), 125.4-128.8 (CH), 139.3 (C=C), 177.3 (C=O)	26.8-42.2 (CH_2), 125.4-128.8 (CH)

Conclusions

This study presents the synthesis of a series of non-protein cyclic amino acids (**2a-2f**), based on the interaction of the corresponding spirohydantoins (**1a-1f**) with barium hydroxyde. The structures of the compounds obtained have been proven through physicochemical parameters, IR, ^{1}H and ^{13}C NMR spectral data.

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References

[1]. Connors, T. A., L. A. Elson, A. Haddow, W. C. J. Ross, 1958. The tumor growth inhibitory activity of 1-aminocyclopentanecarboxylic acid and related peptides. Biochem. Pharmacol., 1, 239-240.

[2]. Martel, F., L. Berlinguet, 1959. Impairment of tumor growth by unnatural amino acids. Can. J. Biochem. Physiol., 37, 433-439.

[3]. Benefiel, W. W., J. T. Helsper, G. S. Sharp, 1960. Apparent control of multiple myeloma by 1-aminocyclopentane-1-carboxylic acid (NSC-1026). Cancer Chemotherapy Rept., 9, 21-22.
[4]. Goldin, A., J. M. Vendditi, I. Kline, N. Mantel, 1961. Evaluation of antileukemic agents employing advanced Leukemia L-1210 in mice IV. Cancer Res., 21, 27-39.
[5]. Ross, R. B., C. I. Noll, W. C. J. Ross, M. V. Nadkarni, B. H. Morrison, H. W. Bond, 1961. Cycloaliphatic amino acids in cancer chemotherapy. J. Med. Pharm. Chem., 3, 1-23.
[6]. Krant, M. J., D. M. Iszard, A. Abadi, R. W. Carey, 1962. Treatment of multiple myeloma by 1-aminocyclopentanecarboxylic acid (NSC-1026). Cancer Chemotherapy Rept., 22, 59-64.
[7]. Hayes, R. L., L. C. Washburn, B. W. Wieland, T. T. Sun, R. R. Turtle, T. A. Butler, 1976. Carboxy-labeled ^{11}C-1-aminocyclopentanecarboxylic acid, a potential agent for cancer detection. J. Nucl. Med., 17, 748-751.
[8]. Marinov, M., D. Ganchev, A Nikolov, P. Marinova, S. Krustev, V. Madzharova, N. Stoyanov, 2013. *In vitro* fungicidal activity of cyclopentanespiro-5-hydantoin and its derivatives towards *Blumeria graminis* f. sp. *tritici*. Agric. Sci., 12, 97-101.
[9]. Ganchev, D., M. Marinov, M. Zlateva, R. Prodanova, A. Nikolov, S. Krustev, N. Stoyanov, 2013. *In vivo* insecticidal activity of cyclopentanespiro-5-hydantoin and its two derivatives towards Mealy plum aphid (*Hyalopterus pruni*) and effect on *Prunus cerasifera*. University of Ruse "Angel Kanchev" Proc., 52 (10.2), 16-20.
[10]. Marinov, M. N., D. H. Ganchev, P. E. Marinova, A. S. Nikolov, R. Y. Prodanova, S. V. Krustev, M. R. Zlateva, N. M. Stoyanov, 2013. *In vivo* insecticidal activity of cyclopentanespiro-5-hydantoin and its two derivatives towards Oleander aphid (*Aphis nerii*) and effect on *Buddleja davidii*. J. Sci. Appl. Res., 4, 171-177.
[11]. Spatola, A., in: B. Weinstein (Ed.): Chemistry and Biochemistry of Amino Acids, Peptides and Proteins, Vol. 7, Dekker, New York, 1983, pp. 267-357.
[12]. Stoyanov, N., M. Marinov, 2012. Two methods for spirothiohydantoin synthesis. Acta Chim. Slov., 59, 680-685.
[13]. Bucherer, H. T., V. A. Lieb, 1934. Über die bildung substituierter hydantoine aus aldehyden und ketonen. Synthese von hydantoinen. J. Prakt. Chem., 141, 5-43.
[14]. Marinov, M., E. Naydenova, R. Prodanova, N. Markova, P. Marinova, I. Kostova, I. Valcheva, D. Draganova, M. Naydenov, P. Penchev, N. Stoyanov, 2016. Synthesis, characterization, theoretical calculations and

antimicrobial studies of substituted 3-aminocyclohexanespiro-5-hydantoins. Agric. Sci., 19, 117-122.
[15]. Nagasawa, H. T., J. A. Elberling, F. N. Shirota, 1973. 2-Aminoadamantane-2-carboxylic acid, a rigid, achiral, tricyclic α- amino acid with transport inhibitory properties. J. Med. Chem., 16 (7), 823-826.
[16]. Marinov, M., P. Marinova, N. Stoyanov, N. Markova, V. Enchev, 2014. Synthesis of 3',4'-dihydro-2*H*,2'*H*,5*H*-spiro[imidazolidine-4,1'-naphthalene]-2,5-dione and its derivatives. Acta Chim. Slov., 61, 420-424.
[17]. Rivero, I. A., E. A. Reynoso-Soto, A. Ochoa-Terán, 2011. Microwave-assisted synthesis of cycloalkanespirohydantoins and piperidinespirohydantoins as precursors of restricted α-amino acids. Arkivoc, ii, 260-271.
[18]. Skita, A., R. Levi, 1908. Über hydrocyclische α-aminosäuren. Ber., 41 (2), 2925-2937.
[19]. Obrecht, D., C. Spiegler, P. Schönholzer, K. Müller, H. Heimgartner, F. Stierli, 1992. A new general approach to enantiomerically pure cyclic and open-chain (*R*)- and (*S*)-α,α-disubstituted α-amino acids. Helv. Chim. Acta, 75, 1666-1696.