S-Thioribofuranoside of Mercaptotriazole

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In this work is presented the synthesis of the first S-riboside using 1H-3-(4-ethoxy-phenyl)-5-mercapto-1,2,4-triazole as aglycone, in the Ferrier reaction conditions. The ^{1}N-, ^{4}N-glycosilated derivatives were not observed during the reaction. The synthetised compounds were characterised by MS-EI, ^{1}H-NMR, I.R., UV and melting points.

Keywords: thioglycoside, S-ribofuranoside, glycosilation reaction, mercaptotriazoles

The thioglycosides of saccharides enjoyed a special attention during the last 20 years, being used as inhibitors, inductors or ligands in the affinity chromatography of enzymes and proteins [1]. On the other hand, the thioglycosides are valuable donors of glycosile in the synthesis of O-glycosides and glycoconjugates [2]. Among these donors are also the thioglycosides of the heterocycles which present an outstanding stability and can easily be converted into other chemical functions [3]. These thioglycosides may function both as acceptors and donors in the synthesis of olygosaccharides. The thioglycosides of heterocycles are found in both pyranosic and furanosic series, the first being by far the most numerous [4,5]. The thioglycosides of heterocycles with 5 atoms, having the ribose as a saccharide component (in the furanosic series) have been synthetised with only a few aglycones: 1-aminoimidazole 2(3H)-one [6], 5,6-dicloro-2-mercapto-benzimidazole [7], 1-(4-chlorophenyl) amino-2,3,-dihydro-4-methyl-5-phenyl-1H-imidazole-2-thiones [8]. The aim of this study is to present the results obtained in the synthesis of a thioglycoside of the ribose using 5-(4-ethoxyphenyl)-3-mercapto-1,2,4-triazole as glycosile acceptor.

Experimental part

Materials and methods

The mass spectra were recorded on a mass spectrometer Varian Finnigan MAT212 for EI= 40 eV which operate in positive ion mode. The proton spectra were recorded on a Varian Gemini 300MHz spectrometer. The chemical shifts are given in ppm, and the coupling constants in Hz. The infrared spectra were recorded in a KBr pellet on a Carl Zeiss-Jena Specord M-80 spectrometer. The ultraviolet spectra were recorded on a Perkin Elmer Lambda 12 spectrophotometer. The reactives and solvents used were purchased from: Fluka (D-ribose, the pyridine, acetic anhydride, 4-ethoxybenzoic acid, dimethyl-formamide), Chimopar (dichloromethane, acetic acid, ethylic alcohol, methylic alcohol), Riedel de Haen (sodium hydroxide) and Aldrich (4,4'-dimethylamino pyridine (DMAP)). The thin layer chromatography (TLC) was realised on Kieselgel (Merck) F_{254} plates. The separation of intermediates and products was done by flash chromatography, using silicagel 220-400 Mesh (Aldrich). The chromatograms were visualized with a solution of H_2SO_4 20% in EtOH and heating the plate at 120°C for 5 min. The solvents were dried over P_2O_5 or sodium and distilled under argon.

General procedures

The synthesis of 5-(4-ethoxiphenyl)-3-mercapto-1,2,4-triazole was realised following the reactions presented in figure 2. The syntheses were done according to the general procedure. steps of the mercaptotriazoles [20]. The synthesis of 5-O-trityl-1,2,3-tri-O-acetil-ribofuranose (**2**) was done by fitting to the procedure of the synthesis of the diethyldithioacetal of 2,3,4-tri-O–acetyl-5-O-trityl-D-arabinose, [19]. The trityl protecting group of the compound (**2**) was removed with acetic acid 60% at 80°C, according to the general method [19] The acetylation of the tri-O-acetyl compound (**3**) was done follow the typical procedure from the chemistry of carbohydrates (Ac_2O,Py) [19]. The synthesis of the S-(2',3',5'-tri-O-acetil-β-D-ribofuranosyl)-3-mercapto-5-p-ethoxi-phenyl-1,2,4-triazole, was done according to the previous method of the mercapto-triazoles from the pyranosic series [5].

(1) 5-O-trityl-ribofuranose white solid**; MS-EI**(40eV)m/z = 392 ($M^{+\cdot}$); **M.p. (°C)** (lit.)[9] = 125; (exp) **=** 127-128; **I.R.** (KBr, cm^{-1}): 704,766,1000,1010,1046,1078,1448,1492,3392; **UV**(MeOH)$\varepsilon_m(\lambda_{max}$,215nm) =21847.

(2) 5-O-trityl-1,2,3-tri-O-acetyl-ribofuranose: white solid**; MS-EI**(40eV)m/z=518($M^{+\cdot}$); **M.p.(°C)**(lit.)[9]=36-40; (exp) **=** 38-40; **I.R.** (KBr, cm^{-1}): 706,750,964,986,1028,1092,1220,1370, 1754 **UV** (MeOH)$\varepsilon_m(\lambda_{max}$,220nm) = 17910.

(3) 1,2,3-tri-O-acetyl-ribose: foam; **MS-EI**(40eV)m/z = 275 $[M-H]^+$;

(4)1,2.3,5-tetra-O-acetyl-ribose:white solid **MS-EI**(40eV)m/z =275$[M-Ac]^+$**; M.p. (°C)** (lit.) [10] = 81-83; (exp) **=** 82-84;**I.R.**(KBr, cm^{-1}): 976, 1004, 1020, 1074, 1138,1218,1756

(7); 1-(4-ethoxy-benzoyl)-tiosemicarbazide: yelow solid **MS-EI**(40eV)m/z=239 [$M^{+\cdot}$] ; **M.p. (°C)** (lit) [11] =205-206; (exp)=221-224;**I.R.**(KBr, cm^{-1}): 3364, 3266, 3179, 3072, 2988, 2930, 2879, 1665, 1607, 1502, 1266, 768, 695

(8) 1H-3-(4-ethoxy-benzoyl)-5-mercapto-1,2,4-triazole: white crystals **MS-EI**(40eV)m/z= 221[$M^{+\cdot}$];

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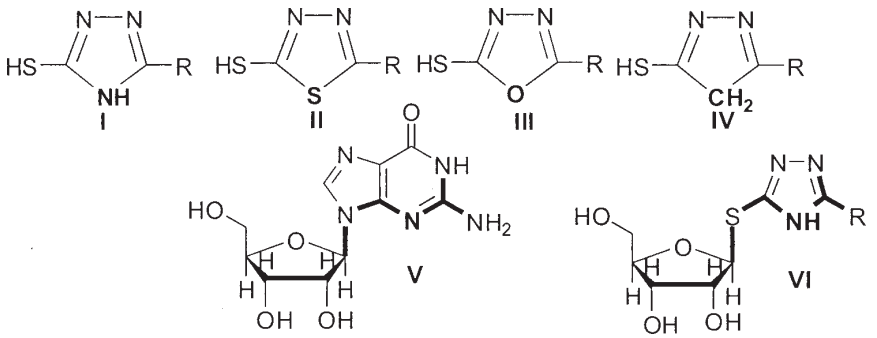

Fig. 1. Isostere heterocycles of mercaptotriazole. The S-glycoriboside of mercaptotriazole, possible or potential mimetic of guanosine.

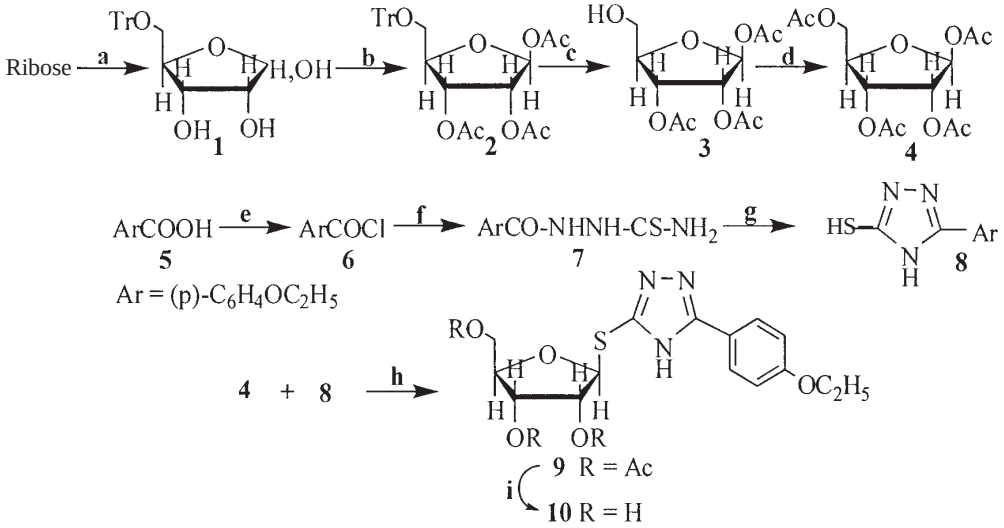

Fig. 2.a)TrCl,Py,4DMAP,r.t,12h.b)Ac_2O,Py,4DMAP,r.t.4h;c)AcOH(60%),80°C,0.5h; d)Ac_2O,Py,r.t,2h;e)$SOCl_2$,50°C,2h;f)$NH_2C(=S)NH_2NH_2$,DMF,Py,r.t. → 50°C,4h;g)EtOH, NaOH, 78°C; h)$BF_3.Et_2O$/DCM/Ar;0°C-r.t.,24h i)MeOH/Na/r.t.,3h

M.p.(°C) =261-264;**I.R.**(KBr, cm^{-1}): 3342, 3089, 2916, 2872, 2586, 1614, 1520, 1258, 1228, 1181, 823

(9) S-(2',3',5'-tri-O-acetyl-β-D-ribofuranosyl)-3-mercapto-5-p-ethoxy-phenyl-1,2,4-triazole: white crystals **^{1}H-NMR($CDCl_3$, δ ppm, *J* Hz):** 11.8(bs, 1H, HN); 7.91(d, 2H, H-10-14, 8.1); 6.94(d, 2H, H-11-13, 8.1); 5.75(m, 1H, H-1); 5.53(m, 1H, H-2); 5.43(m, 1H, H-3); 4.39(m, 2H, H-4, H^A-CH_2-5); 4.22(m, 1H, H^B-CH_2-5); 4.07(q, 2H, OCH_2CH_3); 2.12; 2.11; 2.09(3s, 3H,$OCOCH_3$);1.43(t, 3H, OCH_2CH_3); **M.p (°C) =** 92-94; **I.R.**(KBr,cm^{-1}): 976,1044, 1088, 1256, 1378, 1452,1502,1614,1746; **MS-EI** (40eV) m/z = 479 ($M^{+\cdot}$); **UV**(AcOEt)(λ_{max}263) ε_m =21847

(10)S-β-D-ribofuranosyl)-3-mercapto-5-p-ethoxy-phenyl-1,2,4-triazole: white crystals **^{1}H-NMR (DMSO-d_6, δ ppm, *J* Hz):** 7.90(d, 2H, H-10-14, 8.4); 7.05(d, 2H, H-11-13, 8.4); 5.59(m, 1H, H-1); 4.09(m, 3H, H-2, 2H-15); 3.99(m, 1H, H-3); 3.83(m, 1H, H-4); 3.49(m, syst.AB,2H-5)); 1.35(t, 3H,H-16,7.3);**M.p.°C) =** 74-76;**I.R.**(KBr,cm^{-1}): 1046,1076,1092,1260, 1290,1502,1616,2932,3408;**MS-EI**(40eV) m/z = 353 **($M^{+\cdot}$)**;**UV** (MeOH) ($\lambda_{max,}$263) ε_m=19871

Results and discussions

The derivatives of mercaptotriazoles (**I**) and their isosteres, mercaptothiadiazoles (**II**), mercaptooxadiazoles (**III**) and mercaptopyrazoles (**IV**) (fig. 1) are substances with physiological action. They are used as: anticonvulsives [12], hypoglycemics and diuretics, antiinflamatories [13,14] as well as insecticides [15] and fungicides [16].The coupling of 1,2,4-triazoles with a saccharide moiety allowed to obtain the first N-glycosidic compound with a large spectrum antiviral action: the ribavirine (VIRAZOL@,) [17]. Bioisosteres of guanosine(**V**), pseudonucleotides with mimetic action (**VI**) can compete in the formation of the ARNm (viral nature e.g.). They can be obtained either as N- or S-glycosides of mercaptotriazoles and their derivatives [18]. In order to obtain a compound with a potential mimetic action, the 5-(4-ethoxyphenyl)-3-mercapto-1,2,4-triazole was glycosylated with tetraacetyl ribofuranose (fig. 2).

The 1,2,3,5-Tetra-O-acetyl ribofuranose can not be obtained through the direct acetylation of ribose (Ac_2O/Py sau Ac_2O/AcONa) at room temperature conditions, because the 1,2,3,4-tetra-O-acetyl ribopyranose is obtained (the more stable thermodynamic product) [19]. The attempts at increasing the temperature for the acetylation reaction with the aim of obtaining exclusively the furanosic form lead invariably to a pyranosic and furanosic mixture which are difficult to separate. The protection of the hydroxylic group at C5 with the form of a tritylate derivative (**1**) followed by its full acetylation leads to the furanosydic triacetylate derivative (**2**). The deprotection of the hydroxyl group at C5, by removing the trityl group, followed by its full acetylation leads to the desired tetra-O-acetyl compound (**4**) (fig. 2). The intermediaries were characterised by mass spectrometry (MS), melting point (m.p.) and I.R spectroscopy. The global yield of reactions was 32%.

The synthesis of the aglycone was done according to the procedure for the mercaptotriazoles [20]. The 4-ethoxybenzoic acid (**5**) was transformed to the (**6**) acid chloride corresponding by treatment with thionyl chloride,

in chloroform as a solvent, at 50°C, for 2 h. The excess of thionyl chloride and solvent were removed at low pressure, and the crude product (**6**) was used in the next step without further purification. The cyclization of the acylthiosemicarbazide (**7**) was realised with sodium-ethanolic hydroxide, at reflux. The desired aglycone (**8**) was purified by recrystallization from water (fig.2). The intermediaries were characterised by melting point and mass spectrometry. The global yield of sequence was 85%.

The S-glycofuranoside (**9**) was obtained by the glycosilation reaction of 5-(4-ethoxyphenyl)-3-mercapto-1,2,4-triazole (**8**) with the tetra-O-acetyl ribofuranose (**4**) under the conditions of the Ferrier reaction with an yield of 78%. The TLC shows the formation of a crude single product which was separated and purified by flash column chromatography (Tol:AcOEt = 1:2). The examination of the sample by mass spectrometry confirms the formation of a glycoside having 5-(4-ethoxyphenyl)-3-mercapto-1,2,4-triazole as an aglycone. In order to confirm the formation of the S-glycoside and not the N-glycoside, the product was dissolved in a solution of acetone –water (4:1) and treated for 48h with $AgNO_3$. The Ag cation is well known as thiophyl agent which ensure the cleveage of the carbon –sulf bond [21]. The TLC examination of a sample indicates the formation of the 2,3,5-tri-O-acetyl ribofuranose as anomers mixture (compared to a standard), following the cleveage of the C_{anom}-S bond, which indicates clearly the formation of the S-glycoside (the N-glycosides do not present this reaction). The regioselectivity of reaction, by forming of β-S-glycoside is ensured due to the assistance of the protecting acetate group from C-2. This was confirmed by NMR experiments (H, HxH, APT, NOESY). In order to test the biologic activity, the S-glycoside was underwent to the deprotection of the alcoholic functions. The removing of the protecting acetate groups was performed according to the Zemplen method (MeONa/MeOH). The reaction was monitorized by TLC. After the deprotection, the thioriboside (**10**) was purified by recrystallization and the product analysed by **I.R.** The spectrum confirms the complete deprotection by the absence of the valence vibration $\nu_{C=O}$ from 1746 cm^{-1}, presented in the protected compound (**9**). The structure of compound **(10)** was then confirmed through mass spectrometry and proton nuclear magnetic resonance. ^{1}H-NMR spectrum with the numbering of the atoms is showed in the figure 3.

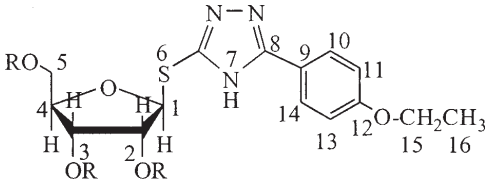

Fig. 3. The numbering of the atoms of (9) and (10) compounds used in the description of ^{1}H-NMR spectra

Aknowledgement This work was supported by the Romanian National Authority for Scientific Research through the grants CEx.111/2006 and PNII-41001.

References

1. LEVY,D.E., FUGEDI, P.(Eds), The Organic Chemistry of Sugar, CRC Press, Boca Raton London, New York, 2006
2.HANESSIAN,S.,(Ed.),Preparative Carbohydrate Chemistry, Marcel Dekker,Inc.,1997
3.CHU,C.K.,(Ed.),Antiviral nucleosides:chiral synthesis and chemoterapy,Elsevier BV,2003
4.El ASHRY, E.S.H., AWAD,L.F., ATTA, A.I., Tetrahedron **62**,2006,p.2943
5.ŞIŞU, E.,LASCU, A., BERCEAN, V., CĂPROIU, M. , ZAMFIR, A.,ŞISU, I., NEANU, C., CSUNDERLIK,C, RUSU V., KATALINIC, J. P., Rev.Chim. (Bucuresti), **54**, 2003,p.181
6.LAGOJA, I. M., AERSCHOT, A. V., HENDRIX, C., HERDEWIJN, P., Collect. Czech. Chem. Commun., **65,** 2000,p.1145.
7.MATHE, C., PERIGAUD, C., GOSSELIN, G., IMBACH, J.,L, Nucleosides Nucleotides **13,** 1994, p.437.
8.AL-MASOUDI,I A.,KHODAIR,A.I, AL-SOUD,Y.A., AL-MASOUDI, N. A.,Nucleosides Nucleotides Nucleic Acids, **22**, 2003, p.299.
9.BREDEREK, H., KOTHNING, M., BERGER, E., Ber.,**73**, 1940,p.956
10.MICHEEL,F.,Chemie der Zucker und Polysaccharide,**2**., Akademische Verlags gesellschaft, Leipzig, 1956
11.LORA-TAMAYO,M., Bull. Soc. Chim. Fr., 1962; p.1024
12.AMIR, M., HUSAIN, M.I., J.Indian Chem.Soc., **63**,1986,p.317
13.PATHAKA, U.S.,DEVANI, M.B.,SHISHOO, C.J.,SHAH, S.A., Indian J.Chem., sect.B**, 28,** 1989,p.183
14.TANDON, M.,BARTHWAL, J.P.,BHALLA, T.N.,BHARGAVA, K.P., Indian J.Chem., sect.B,**20**,1981,p.1017
15.GOSWAMI, B.N., KATAKY, J.C.S., BARUAH, J.N., J.Indian Chem.Soc.,**64**,1987,p.422
16.GOSWAMI, B.N., KATAKY, J.C.S., BARUAH, J.N., NATH, S.C., BORDOLOI, D.N. J.Indian Chem.Soc., **61**,1984,p.530
17.WITKOWSKI,.J.T.,ROBINS, R.K.,KHARE,G.P., SIDWELL,R.W., J.Med.Chem.,**16**, 1973**,** p.935
18.AL-MASOUDI,N.A.,ISSA,F.B.,AL-TIMARI,U.A.,Bull.Soc.Chim.Belg., **106,**1997, p.215
19. WHISTLER,ROY,L,WOLFROM,M,L(eds.).Methods in Carbohydrate Chemistry,vol.II, Academic Press Inc.,1963
20.BERCEAN,V,BADEA,V.,ŞIŞU, E., BINDILĂ, L. CSUNDERLICK, C., Rev.Chim. (Bucharest), **54**, nr. 4, 2003,p.368
21.GARREG, PER J., HULTBERG, H., LINDBERG, C.,Carbohydr. Res.,**83**,1980,p.157

Manuscript received: 29.09.2008