Chemistry of Natural Compounds, Vol. 44, No. 4, 2008

SYNTHESIS AND CHARACTERIZATION OF *N*-TOLYLGLYCOSYLAMINES

R. I. Kublashvili, M. O. Labartkava, K. P. Giorgadze, and D. Sh. Ugrekhelidze*

UDC 547.917

N-*Tolylglycosylamines were synthesized from* o-, m-*, and* p-*toluidines and aldoses (D-glucose, D-galactose, D-mannose, L-rhamnose, D-xylose, and L-arabinose). The anomeric and isomeric compositions of the synthesized products were established using* ^{13}*C NMR methods.*

Keywords: *N*-tolylglycosylamines, aldoses, D-glucose, D-galactose, D-mannose, L-rhamnose, D-xylose, L-arabinose.

Toluidines characteristically have high physiological activity [1, 2]. One of the possible cellular metabolic pathways of toluidines is their *N*-glycosylation, as a result of which the activity is considerably changed [3]. The *N*-glycosylation products of toluidines have been investigated. Their anomers were identified using PMR and gamma-resonance spectroscopy [4, 5]. ^{13}C NMR spectroscopy made it possible to study the rotational isomerism around the *N*-glycoside bonds. The principal parameter of ^{13}C NMR spectroscopy is the chemical shift, which provides important information about the conformation [6]. Our goal was to investigate the *N*-glycosylation of *o*-, *m*-, and *p*-toluidines by aldoses (D-glucose, D-galactose, D-mannose, L-rhamnose, D-xylose, and L-arabinose) and to study the anomeric and isomeric compositions of the synthesized products using ^{13}C NMR methods.

Of all methods for preparing *N*-glycosides with aromatic aglycons (direct reaction of sugars and amines, synthesis from derivatives of sugars and amines, *trans*-glycosylation), direct synthesis by reaction of unsubstituted monosaccharides and aromatic amines of moderate basicity (pK_a 1-6) is a convenient method for preparing these compounds [7-9]. The pK_a values for *o*-, *m*-, and *p*-toluidines are 4.39, 4.60, and 5.12, respectively. It is known that the yield of the target *N*-glycoside is strongly influenced by the basicity of the starting amine. The higher the basicity of the amine is, the easier the resulting *N*-glycoside undergoes various transformations (hydrolysis, Amadori—Heyns rearrangement, melanoidine formation, etc.) [10]. As a result, the preparative yield of the target *N*-glycoside decreases as the basicity of the starting amine increases. We selected the optimum conditions for the *N*-glycosylation considering the properties of the amine involved. This made it possible to isolate quantitatively the target products from the reaction mixtures. After the appropriate workup (recrystallization, paper chromatography and thin-layer chromatography over silica gel) and identification (melting point, IR spectrum, elemental analysis), the synthesized *N*-glycosides were investigated using ^{13}C NMR spectra.

IR spectra of the *N*-tolylglycosylamines (KBr, ν, cm^{-1}): 3380-3325 (OH stretch), 2910-2860 (CH stretch), 1660-1600 (C_6H_6), 1530-1520 (*N*-glycoside), 1550-1540 (CH deformation, C–OH stretch), 1375-1370 (CN stretch), 1290-1250 (OH deformation, CO stretch), 1180-1170 (CN stretch, anomeric C1), 1060-1000 (carbohydrate ring), 850-800 and 780-750 (*N*-glycosylamine pyranose ring), 650-640 (CN stretch of anomeric C1).

Tables 1 and 2 give the ^{13}C NMR spectra of the *N*-tolylglycosylamines. Carbon atoms of the carbohydrate part that are bonded to primary and secondary alcohols of the aldoses resonate at the strongest field. Carbon atoms of the carbohydrate part that are bonded to two electronegative atoms (O–C–N) resonate at the weakest field. It is known that the chemical shifts of β-conformers of most monosaccharides (except mannose, rhamnose, and arabinose) are greater than those of the α-conformers [6, 11, 12]. Therefore, resonances located at comparatively weak field belong to the β-anomers; at comparatively strong field, to the α-anomers. Resonances of C atoms of the aromatic part are situated in the range 110-149 ppm. The ratios of β- to α-anomers were calculated taking into account the relaxation time of the anomeric C and the signal strength. These data were used to calculate the isomeric and anomeric compositions of equilibrium mixtures of *N*-glycosides of isomorphous aldose pairs (Table 2).

I. Javakhishvili Tbilisi State University, 0128, Georgia, Tbilisi, pr. I. Chavchavadze, 3, e-mail: devi_ugrekhelidze@hotmail.com. Translated from Khimiya Prirodnykh Soedinenii, No. 4, pp. 333-335, July-August, 2008. Original article submitted September 19, 2007.

0009-3130/08/4404-0413

TABLE 1. Isomeric and Anomeric Compositions of *N*-Tolylglycosylamines and Chemical Shifts of ^{13}C Atoms of the Carbohydrate Fragments

Tolyl-*N*-glycoside	Yield, %	mp, °C	β/α, %	^{13}C chemical shift, ppm					
				C1	C2	C3	C4	C5	C6
N-*o*-Tolyl-β-D-glucopyranosylamine	78.0	83-84	100	85.4	73.0	73.1	70.5	77.3	61.2
N-*m*-Tolyl-β-D-glucopyranosylamine	89.0	123-124	100	85.3	73.1	77.3	70.3	77.7	61.1
N-*p*-Tolyl-β-D-glucopyranosylamine	81.0	101-102	100	85.5	73.2	77.1	70.4	77.7	61.1
N-*o*-Tolyl-β-D-galactopyranosylamine	70.6	122-123	100	85.9	68.4	73.9	70.3	75.5	60.6
N-*m*-Tolyl-β-D-galactopyranosylamine	81.7	107-108	100	82.5	72.2	73.5	70.2	76.1	62.9
N-*p*-Tolyl-β-D-galactopyranosylamine*			84.3	86.0	70.4	74.3	69.5	75.5	60.6
N-*p*-Tolyl-β-D-galactofuranosylamine*	66.8	134-135	15.7	88.2	82.5	76.1	83.2	70.8	62.8
N-*o*-Tolyl-β-D-mannopyranosylamine	92.8	143	100	80.9	71.2	74.4	67.1	77.8	61.2
N-*m*-Tolyl-β-D-mannopyranosylamine	93.0	155-156	100	81.4	71.2	74.5	67.1	77.8	61.3
N-*p*-Tolyl-β-D-mannopyranosylamine**			75.8	81.7	71.3	74.6	67.2	77.8	61.3
N-*p*-Tolyl-α-D-mannopyranosylamine**	85.4	158-159	24.2	84.1	70.8	71.1	67.9	72.6	61.3
N-*o*-Tolyl-β-L-rhamnopyranosylamine	79.0	128-129	100	80.6	72.4	71.3	74.1	67.7	17.2
N-*m*-Tolyl-β-L-rhamnopyranosylamine	87.0	126-127	100	81.1	72.5	72.1	74.2	71.3	18.1
N-*p*-Tolyl-β-L-rhamnopyranosylamine**			73.3	81.3	72.4	72.1	74.2	71.3	18.0
N-*o*-Tolyl-α-L-rhamnopyranosylamine**	79.0	112-113	26.7	83.8	72.4	72.9	70.9	70.8	18.0
N-*o*-Tolyl-β-D-xylopyranosylamine	72.0	103-104	100	86.0	74.9	76.9	70.2	65.8	-
N-*m*-Tolyl-β-D-xylopyranosylamine	75.0	115-117	100	86.0	73.4	77.8	70.1	66.5	-
N-*p*-Tolyl-β-D-xylopyranosylamine	70.0	122-123	100	86.4	73.2	77.9	70.4	66.5	-
N-*m*-Tolyl-β-L-arabinopyranosylamine**			65.1	85.5	70.8	70.6	70.4	63.5	-
N-*m*-Tolyl-α-L-arabinopyranosylamine**	74.0	94-95	34.9	88.5	70.3	73.5	69.7	67.8	-
N-*p*-Tolyl-β-L-arabinopyranosylamine**			71.4	85.7	70.8	70.6	70.4	63.5	-
N-*p*-Tolyl-α-L-arabinopyranosylamine**	68.0	97-98	28.6	88.7	69.7	73.5	69.5	67.8	-

Yield and mp for the forms of sugars (*): for isomeres (**).

TABLE 2. ^{13}C Chemical Shifts of Atoms in Aromatic Fragments of *N*-Tolylglycosylamines and Their Anomeric Compositions, %

Tolyl-*N*-glycoside	^{13}C chemical shift, ppm							%
	C1	C2	C3	C4	C5	C6	CH_3	
N-*o*-Tolyl-β-D-glucopyranosylamine	146.2	123.5	131.1	118.8	128.1	113.0	17.4	100
N-*m*-Tolyl-β-D-glucopyranosylamine	147.3	114.0	137.7	118.0	128.7	110.5	21.4	100
N-*p*-Tolyl-β-D-glucopyranosylamine	145.0	113.3	129.2	125.4	129.2	113.3	20.2	100
N-*o*-Tolyl-β-D-galactopyranosylamine	144.8	122.1	129.7	117.2	126.6	111.5	17.4	100
N-*m*-Tolyl-β-D-galactopyranosylamine	149.0	113.3	137.6	116.6	128.6	110.1	21.4	100
N-*p*-Tolyl-β-D-galactopyranosylamine	144.5	113.3	129.2	125.3	129.2	113.3	20.2	84.3
N-*p*-Tolyl-β-D-galactofuranosylamine	144.8	113.7	129.3	125.6	129.3	113.7	20.2	15.7
N-*o*-Tolyl-β-D-mannopyranosylamine	143.6	122.0	129.9	117.4	126.8	111.8	17.2	100
N-*m*-Tolyl-β-D-mannopyranosylamine	146.2	114.3	137.7	118.2	128.6	110.9	21.4	100
N-*p*-Tolyl-β-D-mannopyranosylamine	143.9	113.8	129.3	125.7	129.3	113.8	20.2	75.8
N-*p*-Tolyl-α-D-mannopyranosylamine	144.8	113.5	129.2	125.4	129.2	113.5	20.2	24.2
N-*o*-Tolyl-β-L-rhamnopyranosylamine	143.6	121.7	129.9	117.4	126.7	111.7	18.1	100
N-*m*-Tolyl-β-L-rhamnopyranosylamine	146.2	114.2	137.8	118.2	128.7	110.9	21.4	100
N-*p*-Tolyl-β-L-rhamnopyranosylamine	143.8	113.7	129.2	125.7	129.2	113.7	20.1	73.3
N-*o*-Tolyl-α-L-rhamnopyranosylamine	144.6	113.2	129.2	125.3	129.2	113.2	20.1	26.7
N-*o*-Tolyl-β-D-xylopyranosylamine	144.8	122.5	130.0	117.7	126.8	111.7	17.7	100

TABLE 2. (continued)

Tolyl-*N*-glycoside	^{13}C chemical shift, ppm							%
	C1	C2	C3	C4	C5	C6	CH_3	
N-*m*-Tolyl-β-D-xylopyranosylamine	147.0	114.0	137.9	118.2	131.9	110.7	21.4	100
N-*p*-Tolyl-β-D-xylopyranosylamine	144.9	113.5	129.4	125.7	129.4	113.5	20.4	100
N-*m*-Tolyl-β-L-arabinopyranosylamine	147.2	114.3	137.8	118.3	128.8	110.8	21.6	65.1
N-*m*-Tolyl-α-L-arabinopyranosylamine	146.7	113.8	137.9	118.5	128.8	111.2	21.6	34.9
N-*p*-Tolyl-β-L-arabinopyranosylamine	144.8	113.9	129.4	125.6	129.4	113.9	20.3	71.4
N-*p*-Tolyl-α-L-arabinopyranosylamine	144.4	113.5	129.4	125.9	129.4	113.5	20.3	28.6

The furanose ring is unstable due to *cis*-interactions between neighboring substituents. The cyclic furanose forms of mannose, rhamnose, glucose, and xylose are especially unstable because of these interactions. For example, the number of 1,2-*cis*-interactions for mannose and rhamnose is two, where the interaction involves O_2/O_3 and O_3/C_5. The number of 1,2-*cis*-interactions for glucose and xylose is one involving O_3/C_5. Only *trans*-interactions are possible for the furanose forms of galactose and arabinose [13]. According to Table 1, the yields of the synthesized *N*-tolylglycosylamines decreased in the order mannose, rhamnose, glucose, xylose, galactose, and arabinose. Thus, the stable furanose forms of aldoses produce *N*-glycosides that are largely converted to melanoidines under the *N*-glycosylation reaction conditions.

EXPERIMENTAL

IR spectra in KBr were recorded on a Specord 75 IR spectrophotometer; ^{13}C NMR spectra, on a Bruker NM-250 MGH instrument using $(CD_3)_2SO$ with a shift of the central resonance of 39.505 ppm with full C–H decoupling, weighed samples (30 mg), and 60°C.

Synthesis of *N*-tolylglycosylamines [11, 12]. A mixture of aldose (0.01 mol) and toluidine (0.012 mol) in water (0.5 mL) was heated to boiling on a water bath with stirring until complete dissolution of the starting materials. The reaction mixture was cooled to room temperature, treated with diethylether (40 mL), stirred, and left overnight at room temperature. The resulting crystalline precipitate was filtered off, ground with ethanol (96%), treated with diethylether, and thoroughly mixed. The precipitate was filtered off. The resulting *N*-glycoside was purified by reprecipitation from alcoholic diethylether. The purity of the products was monitored by TLC on Silufol UV-254 plates (for toluidines, benzene:ethanol, 9:1; for aldoses, $CHCl_3$:CH_3OH, 19:5; for *N*-tolylglycosylamines, dioxane:benzene, 1:4). Sugars were developed by a basic solution of potassium permanganate and sodium metaperiodate; toluidines, by alcoholic *p*-dimethylaminobenzaldehyde.

REFERENCES

1. N. Danford, *Mutat. Res.*, **258**, 207 (1991).
2. M. Loidl, C. Hinteregger, J. Stockinger, and F. Streichsbier, *J. Basic Microbiol.*, **41**, 159 (2001).
3. G. Legler, *Pure Appl. Chem.*, **59**, 1457 (1987).
4. N. I. Kaletina, *N-Glycosylamines and Microelements* [in Russian], Izd. Arm. SSR, Erevan (1988), 161.
5. D. Gomes de Anderez, H. Gil, M. Helliwell, and J. Mata Segreda, *Acta Crystallogr. Sect. C: Cryst. Struct. Commun.*, **52**, 252 (1996).
6. A. S. Shashkov and O. S. Chizhov, *Bioorg. Khim.*, **2**, 437 (1976).
7. B. N. Stepanenko, N. I. Kaletina, and V. V. Zelenkova, *Dokl. Akad. Nauk SSSR*, **200**, 609 (1971).
8. R. I. Kublashvili, *Khim. Prir. Soedin.*, 484 (2003).
9. R. Kublashvili, M. Labartkava, and D. Ugrekhelidze, *J. Biol. Phys. Chem.*, **5**, 25 (2005).
10. H. D. Belitz and W. Grosch, *Food Chemistry*, Springer-Verlag, Berlin, Heidelberg, New York (1999), 263-318.
11. K. Bock and C. Pedersen, *Adv. Carbohydr. Chem. Biochem.*, **41**, 27 (1983).
12. E. Breitmeier and C. Voelter, *^{13}C NMR Spectroscopy*, Verlag Chemie, Weinheim (1978), 183-263.
13. J. F. Stoddart, *Stereochemistry of Carbohydrates*, Interscience, New York (1971).