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The composition of 2-keto aldoses in organic solvents as determined by NMR spectroscopy

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Abstract—The composition of the 2-keto aldoses D-glucosone (**1**), 6-deoxy-D-glucosone (**2**), D-allosone (**3**), and D-galactosone (**4**) in organic solvents has been determined using NMR spectroscopy. Whereas these keto aldoses form mixtures with up to 15 different isomers in water, the number of forms is significantly decreased in organic solvents. Equilibrium mixtures of **1**, **2**, and **4** in Me_2SO, DMF, and pyridine consist to 70–90% of the prevailing α-1,5-pyranose form. Two bicyclic forms with a proportion of 80% are the main isomers of **3** in pyridine. Generally, forms with non-hydrated keto functions prevail in non-aqueous solutions.

Keywords: 2-Keto aldoses; Isomeric forms; Composition in solution; NMR spectroscopy

1. Introduction

The composition of carbohydrates in solution is of major importance for their chemical, physical, and biological properties. The equilibrium mixture of common reducing sugars in aqueous solution is well documented.[1,2] It has been studied extensively by NMR spectroscopy, and some data have also been determined in organic solvents like Me_2SO, DMF, or pyridine.[3–6] Solutions of aldoses and ketoses mainly consist of two pyranose and two furanose anomers, which are rarely accompanied by open chain or other forms. An analysis of the NMR spectra of these common sugars is therefore relatively straightforward. This is not the case for carbohydrates with a second carbonyl function. In keto aldoses, ring closure can take place at two different positions, and, accordingly, these compounds show complex mixtures of isomeric forms. This might be the reason why the composition of keto aldoses has been analyzed only in relatively few cases.[7]

2-Keto aldoses are important intermediates in biological and chemical processes. In mammals, D-glucosone (**1**) is believed to be formed by autooxidation of glucose,[8] whereas in several plants, the conversion of **1** to L-ascorbic acid has been observed.[9] 3-Deoxy-D-glucosone plays an important role in the complex *Maillard* reaction.[10,11] D-Galactosone (**4**) is an efficient glucuronide inhibitor in hepatocytes.[12] Likewise, 2-keto aldoses are interesting compounds for chemical syntheses since they combine a high number of functional groups and the inherent chiral information of sugars with a higher degree of diversity than common aldoses. They are consequently very helpful synthons for a variety of chemical procedures,[13–16] and some of them were effectively used for the synthesis of antibiotics[17–20] and aminosugars.[21,22] The composition of 2-keto aldoses in solution is therefore of concern for the chemistry of these compounds. Since the feasibility of many chemical reactions suffer from the presence of manifold forms in water, it is especially interesting to deduce the composition in organic solvents, what might eventually help to establish an attractive chemistry for 2-keto aldoses in non-aqueous solvents.[23]

We recently obtained some 2-keto aldoses by enzymatic oxidation[24] and determined their solution structures in water applying NMR spectroscopy.[7] Aqueous

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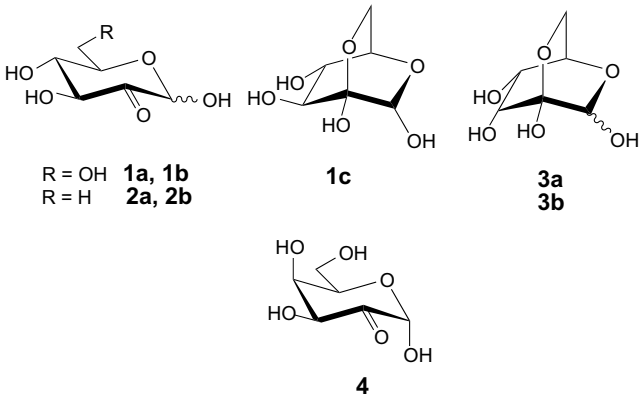
solutions of these compounds show a profound complexity, that is, some of the investigated keto aldoses show up to 10 or 15 different isomers. In the organic solvents Me_2SO, DMF, and pyridine, however, we found a significantly decreased number of forms. This obviously facilitates the analysis of the NMR spectra but, moreover, may eventually be advantageous in the development of useful chemical reactions in these solvents.

2. Results and discussion

2.1. Structural assignment of isomers by NMR spectroscopy

The structural assignment of the isomers of 2-keto aldoses in organic solvents is easier than in water since there are less equilibrium forms. The assignments of compounds **1**–**4** were performed applying standard NMR methods. ^1H–^1H connectivities were deduced from phase sensitive DQF-COSY and TOCSY spectra and ^1H–^{13}C connectivities from HMQC spectra. In a previous paper, we reported a number of ^1H and ^{13}C NMR data for various C-1 and C-2 cyclized forms of 2-keto aldoses including pyranose, furanose, and bicyclic forms.[7] These data and data from the literature helped to establish and to verify the NMR assignments of the isomers in organic solvents (Tables 1 and 2), which are in all cases in agreement with the published values and will not be discussed explicitly.



2.2. Composition in organic solvents

In contrast to aqueous solutions of underivatized 2-keto-aldoses,[7] examinations in organic solvents have to our knowledge up to now not been reported. We found the number of isomers of 2-keto aldoses (Table 3) in the solvents Me_2SO, DMF, and pyridine to be effectively decreased compared to the composition in water. It is conspicuous that the 2-keto group is not hydrated in these solvents although the compounds **1**–**4** were originally obtained as hydrates.[24] D-Glucosone (**1**) shows the

Table 1. ^1H NMR shifts (δ, ppm) and $^3J_{H,H}$ values (Hz) of D-*arabino*-hexos-2-ulose (**1**), 6-deoxy-D-*arabino*-hexos-2-ulose (**2**), D-*ribo*-hexos-2-ulose (**3**), and D-*lyxo*-hexos-2-ulose (**4**)

Compound	H-1	$J_{3,4}$	H-3	$J_{4,5}$	H-4	$J_{5,6a}$	H-5	$J_{5,6b}$	H-6a	$J_{6a,6b}$	H-6b
1a[a]	4.873	9.3	4.297	9.8	3.340	1.7	3.912	5.9	3.665	11.9	3.529
1b[a]	5.016	8.9	4.136	9.0	3.293	1.0	3.510	6.2	3.713	11.7	3.453
1c[a]	4.675	n.d.	3.923[d]	0.0	3.643[d]	1.0	3.700	0.0	3.767	10.9	3.226
1a[b]	5.006	9.0	4.524	9.7	3.556	2.2	4.116	5.4	3.846	12.3	3.715
1b[b]	5.198	8.7	4.362	n.d.	3.514	n.d.	3.688	n.d.	3.894	12.5	3.660
1c[b]	4.911	4.7	4.155[d]	0.0	3.882[d]	1.6	3.830	1.0	3.914	10.7	3.313
1a[c]	5.788	9.2	5.510	9.9	4.514	2.1	5.056	5.2	4.570	11.7	4.449
2a[a]	4.821	9.4	4.246	9.4	3.028	6.2	4.021		1.172		
2b[a]	4.941	9.2	4.085	9.2	3.028	6.1	3.587		1.192		
3a[c]	5.345	6.5	4.536	0.0	4.762	1.4	4.438	0.7	4.393	11.3	3.503
3b[c]	5.514	6.4	5.028	0.0	4.762	1.5	4.385	0.8	3.914	11.2	3.848
4[a]	4.820	3.2	4.522	0.4	4.136	6.5	4.204	6.5	3.539	10.7	3.459
4[b]	4.976	3.7	4.709	0.5	4.367	6.5	4.402	5.9	3.750	11.0	3.646
4[c]	5.800	3.6	5.511	0.5	5.101	6.2	5.235	6.2	4.513	11.0	4.479

	OH-1	$J_{1,OH-1}$	OH-2	$J_{3,OH-3}$	OH-3	$J_{4,OH-4}$	OH-4	$J_{6,OH-6}$	OH-6
1a[a]	7.470	3.7		5.6	5.314	6.2	5.519	4.9/6.0	4.563
1b[a]	6.928	8.7		5.8	5.346	4.7	5.538	5.8	4.644
1c[a]	6.202	6.5	5.980	4.7	5.131	5.5	5.230		
1a[b]	7.722	8.4		9.0	5.307	9.9	5.707	6.5	4.602
1b[b]	7.028	8.6		9.0	5.372	n.d.	n.d.	6.6	4.836

[a] In Me_2SO-d_6.
[b] In DMF-d_7.
[c] In pyridine-d_5.
[d] Signal assignment may be inverted.

Table 2. ^{13}C NMR chemical shifts (δ, ppm) of D-*arabino*-hexos-2-ulose (**1**), 6-deoxy-D-*arabino*-hexos-2-ulose (**2**), D-*ribo*-hexos-2-ulose (**3**), and D-*lyxo*-hexos-2-ulose (**4**)

Compound	C-1	C-2	C-3	C-4	C-5	C-6
1a[a]	93.63	202.31	76.22	74.13	72.59	60.49
1b[a]	93.89	201.56	78.85	74.91	77.02	60.76
1c[a]	89.08	101.71	78.92[d]	84.00[d]	81.60	61.09
1a[b]	95.07	202.90	77.58	75.85	73.59	61.98
1b[b]	95.07	202.16	80.21	76.53	78.15	61.98
1c[b]	90.50	102.66	80.44[d]	85.30[d]	83.01	62.15
1a[c]	95.48[e]	203.96	78.47	76.48	74.27	62.46
2a[a]	93.65	202.48	76.05	79.33	67.31	17.54
2b[a]	93.86	201.72	78.61	80.00	71.59	17.71
3a[c]	94.20[f]	102.85	71.30	72.91	83.11	61.80
3b[c]	97.03[g]	104.35	68.54	74.00	81.86	68.14
4[a]	93.92	204.28	73.29	74.31	71.56	60.15
4[b]	95.28	204.49	74.23	75.80	72.30	61.63
4[c]	95.68	205.94	75.30	76.50	73.10	62.27

[a] In Me_2SO-d_6.
[b] In DMF-d_7.
[c] In pyridine-d_5.
[d] Signal assignment may be inverted.
[e] $J_{C\text{-}1,H\text{-}1} = 170.0$ Hz.
[f] $J_{C\text{-}1,H\text{-}1} = 165.5$ Hz.
[g] $J_{C\text{-}1,H\text{-}1} = 161.9$ Hz.

Table 3. Isomers of D-*arabino*-hexos-2-ulose (**1**), 6-deoxy-D-*arabino*-hexos-2-ulose (**2**), D-*ribo*-hexos-2-ulose (**3**), and D-*lyxo*-hexos-2-ulose (**4**) and their proportions (%) in organic solvents at 300 K

Solvent	Structure	D-Glucosone (**1**)	6-Deoxy-D-glucosone (**2**)	D-Allosone (**3**)	D-Galactosone (**4**)
Me_2SO-d_6	α-1,5-Pyr	75 (**1a**)	79 (**2a**)	~10 forms	90
	β-1,5-Pyr	16 (**1b**)	21 (**2b**)		
	α-2,6-Bicycl.	9 (**1c**)			
DMF-d_7	α-1,5-Pyr	70 (**1a**)			80
	β-1,5-Pyr	20 (**1b**)			
	α-2,6-Bicycl.	10 (**1c**)			
Pyridine-d_5	α-1,5-Pyr	80 (**1a**)			80
	α-2,6-Bicycl.			30 (**3a**)	
	β-2,6-Bicycl.			50 (**3b**)	

three isomeric forms **1a**–**c** in Me_2SO. The predominating form is the α-1,5-pyranose **1a** accompanied by minor amounts of the β-anomer **1b** and the bicyclic 2,6-bridged form **1c**, which resembles the bicyclic forms found for D-allosone (**3**) and 3-deoxy-D-glucosone in water.[7] The composition of **1** in DMF shows comparable amounts of the three forms; in pyridine, we only assigned the major compound **1a**, which represents 80% of the equilibrium. In a similar way, the equilibrium mixture of 6-deoxy-D-glucosone (**2**) in Me_2SO consists of the 1,5-pyranose anomers **2a** and **2b** but of no 2,6-cyclic structures due to the lack of a hydroxyl group at C-6.

D-Galactosone (**4**) consists of approximately 15 to 18 isomers in aqueous solution. Their number is too large to allow an exact determination, and only seven of these forms were unequivocally assigned.[7] In organic solvents, however, the number of isomers is much smaller. In Me_2SO, **4** mainly consists of one anomer with an proportion of 90%. A similar behavior was found for solutions in DMF and pyridine both consisting to 80% of the same non-hydrated α-1,5-pyranose form.

D-Allosone (**3**) is the only 2-keto aldose investigated that does not have a reduced number of isomers in organic solvents. This compound shows in Me_2SO approximately six major and four minor forms. The NMR spectra were to poorly resolved to allow an exact determination, but with the help of the data for the bicyclic structures in pyridine, these forms could be detected in Me_2SO as well. In pyridine, the bicyclic forms **3a** and **3b** prevail that were previously described for D-glucosone (**1**) and other compounds.[24] It has to be mentioned that the basic character of pyridine in all cases caused some degradation leading to approximately 10% of unknown forms.

The investigation of the composition of the 2-keto aldoses **1**–**4** in methanol did not lead to any definite results, because the NMR spectra were even more

complicated and showed more forms compared to the aqueous solutions. Whether there are hemiacetals formed or not, remained unclear.

In 1994, Angyal has published the so far most extensive comparison of the composition of reducing sugars in water and in Me_2SO.[5] As an important result he found, that the proportion of furanose forms in Me_2SO is generally higher than in water. It is not clear why, but the influence of solvation by water or other solvents has been suggested and is still under discussion.[5,25,26] These possible mechanisms, however, do not seem to have a significant effect in the case of the 2-keto aldoses **1**–**4** since furanose forms have not been detected. This is surprising considering the proportions of furanoidic isomers in aqueous solutions of **1**–**4** ranging from 8% to 33%.[7] The lack of furanose forms might possibly be due to the non-hydrated 2-keto group and is in fact one reason for the advantageous simpleness of the equilibria in Me_2SO compared to water.

The preference of 1,5-pyranose forms of the 2-keto aldoses **1**, **2**, and **4** reflects—similar to most common reducing sugars—the higher tendency of aldoses than of ketoses to cyclize. Prevailing of the α-anomer is due to the anomeric effect being more efficient in organic than in aqueous solution. Astonishingly, D-allosone (**3**) behaves differently. Comparable to some of its derivatives,[24] **3** shows a high tendency to form 2,6-bridged structures, which in pyridine seem to be stabilized in such a way that they predominate over all other forms. Further work is necessary to clarify this interesting finding.

3. Experimental

The 2-keto aldoses **1**–**4** were obtained by enzymatic oxidation of the corresponding aldoses.[24,27] Solutions of **1**–**4** were equilibrated for 1 day at ambient temperature. NMR spectra were recorded on a Bruker AMX 500 spectrometer at 500.14 MHz for ^1H and 125.76 MHz for ^{13}C at 300 K applying standard 1D and 2D spectroscopy (gated decoupling, phase sensitive and DQF-COSY, TOCSY, HMQC). Chemical shifts are referenced to the residual proton signal ($\delta = 2.490$, 2.740, 8.710) and the carbon signal ($\delta = 39.50$, 162.70, 149.90 ppm) of Me_2SO-d_6, DMF-d_7, and pyridine-d_5, respectively. Abbreviation 'n.d.' means not determined.

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References

1. Angyal, S. J. *Adv. Carbohydr. Chem. Biochem.* **1984**, *3*, 15–68.
2. Angyal, S. J. *Adv. Carbohydr. Chem. Biochem.* **1992**, *49*, 19–35.
3. Dais, P.; Perlin, A. S. *Carbohydr. Res.* **1987**, *169*, 159–169.
4. Jaseja, M.; Perlin, A. S.; Dais, P. *Magn. Reson. Chem.* **1990**, *28*, 283–289.
5. Angyal, S. J. *Carbohydr. Res.* **1994**, *263*, 1–11.
6. Angyal, S. J.; Christofides, J. C. *J. Chem. Soc., Perkin Trans. 2* **1996**, 1485–1491.
7. Köpper, S.; Freimund, S. *Helv. Chim. Acta* **2003**, *3*, 827–843.
8. Wolff, S. P.; Dean, R. T. *Biochem. J.* **1987**, *245*, 243–250.
9. Saito, K. *Phytochemistry* **1996**, *41*, 145–149.
10. Hayase, F. *Food Sci. Technol. Res.* **2000**, *6*, 79–86.
11. Liedke, R.; Eichner, K. *ACS Symp. Ser.* **2002**, *807*, 69–82.
12. Ullrich, D.; Bock, K. W. *Biochem. Pharmacol.* **1984**, *33*, 1827–1830.
13. Lichtenthaler, F. W.; Schwidetzky, S.; Nakamura, K. *Tetrahedron Lett.* **1990**, *31*, 71–74.
14. Freimund, S.; Huwig, A.; Giffhorn, F.; Köpper, S. *J. Carbohydr. Chem.* **1996**, *15*, 115–120.
15. Choque, J.-C.; Fleche, G. Eur. Pat. Appl. EP 1048671, 2000; *Chem. Abstr.* **2000**, *131*, 321214.
16. Andersen, S. M.; Lundt, I.; Marcussen, J.; Søtofte, I.; Yu, S. *J. Carbohydr. Chem.* **1998**, *17*, 1027–1035.
17. Volc, J.; Kubátová, E.; Sedmera, P.; Daniel, G.; Gabriel, J. *Arch. Microbiol.* **1991**, *156*, 297–301.
18. Koths, K.; Halenbeck, R.; Moreland, M. *Carbohydr. Res.* **1992**, *232*, 59–75.
19. Gabriel, J.; Volc, J.; Kubá tová, E.; Palková, Z.; Pospíšek, M. *Carbohydr. Res.* **1994**, *252*, 297–301.
20. Gabriel, J.; Volc, J.; Sedmera, P.; Daniel, G.; Kubátová, E. *Arch. Microbiol.* **1993**, *160*, 27–34.
21. Baxter, E. W.; Reitz, A. B. *Tetrahedron Lett.* **1990**, *31*, 6777–6780.
22. Baxter, E. W.; Reitz, A. B. *Bioorg. Med. Chem. Lett.* **1992**, *2*, 1419–1422.
23. Lichtenthaler, F. W.; Klotz, J.; Flath, F.-J. *Liebigs Ann.* **1995**, 2069–2080.
24. Freimund, S.; Huwig, A.; Giffhorn, F.; Köpper, S. *Chem. Eur. J.* **1998**, *4*, 2442–2455.
25. Berger, S.; Diaz, M. D.; Hawat, C. *Polish J. Chem.* **1999**, *73*, 193–197.
26. López de la Paz, M.; Ellis, G.; Pérez, M.; Perkins, J.; Jiménez-Barbero, J.; Vicent, C. *Eur. J. Org. Chem.* **2002**, *1*, 840–855.
27. Huwig, A.; Danneel, H. J.; Giffhorn, F. *J. Biotechnol.* **1994**, *32*, 309–315.