Molybdic Acid-Catalyzed Mutual Interconversions of 2-*C*-(Hydroxymethyl)-D-glucose with D-*manno*-Hept-2-ulose and 2-*C*-(Hydroxymethyl)-D-mannose with D-*gluco*-Hept-2-ulose

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Nitromethane synthesis with D-fructose followed with the Nef reaction of intermediate 1-deoxy-2-*C*-hydroxymethylhexitol-1-nitronates afforded 2-*C*-(hydroxymethyl)-D-glucose and -D-mannose which were separated *via* their phenylhydrazones and by chromatography on a cation-exchange resin in the Ba^{2+} form. The treatment of individual branched-chain aldoses with catalytic amount of molybdic acid, *i.e.* under the conditions of the Bílik reaction afforded thermodynamic equilibrium mixtures of 2-*C*-(hydroxymethyl)-D-glucose and D-*manno*-hept-2-ulose in the mole ratio 2 55 and of 2-*C*-(hydroxymethyl)-D-mannose and D-*gluco*-hept-2-ulose in the ratio 2 23, respectively. The same equilibria were obtained also from the side of hept-2-uloses. Due to easy availability of 2-*C*-(hydroxymethyl)-D-mannose also from its di-*O*-isopropylidene derivative the interconversion was advantageously used for one-step synthesis of D-*gluco*-hept-2-ulose.

Effective synthetic procedures for naturally rare carbohydrates have become very important in preparation of compounds that are used in biochemistry and medicinal chemistry. The development of methods where metalic ions play a catalytic role in transformations of saccharides can significantly simplify often complex synthetic procedures. For example, *Bílik* has shown in a series of reports that, in mildly acidic solutions of molybdate, aldoses epimerize at carbon atom C-2 under the formation of thermodynamic equilibrium mixture of two epimers [1—3]. Later the transformation has been generalized to all aldoses and became known as the Bílik reaction [4].

There are two simple ways of approaching the synthesis leading to preparation of 2-*C*-hydroxymethyl derivatives of aldohexoses. One involves their *cis*-bidentate protection including C-2—OH group to obtain a suitable rigid structure of the C-2 nucleophile for its addition to formaldehyde [5]. The second, and more generally applicable one, is *via* nitromethane synthesis with 2-ketoses and subsequent Nef reaction which yields 2-*C*-branched derivatives [6]. In the latter preliminary study it was shown that a treatment of two branched-chain aldoses, namely 2-*C*-(hydroxymethyl)-D-mannose and -D-glucose, respectively, with diluted molybdic acid at elevated temperature produces corresponding hept-2-uloses, D-*gluco*- and D-*manno*-hept-2-ulose. A stereospecific rearrangement of these 2-*C*-branched aldoses is a probable mechanism of this reaction and is apparently analogical to that occurring with unbranched aldoses [7]. Here we present a more detailed study of the molybdic acid-catalyzed isomerization of 2-*C*-hydroxymethyl branched-chain aldoses to the corresponding 2-ketoses using two model sugars, 2-*C*-(hydroxymethyl)-D-mannose and 2-*C*-(hydroxymethyl)-D-glucose.

EXPERIMENTAL

300 MHz ^{1}H NMR and 75.46 MHz ^{13}C NMR spectra were recorded at 40°C in D_2O on a Bruker DPX 300 spectrometer. The values of chemical shifts were expressed relative to external TSP. Two-dimensional COSY and HSQC experiments were performed using z-gradients for coherence transfer. HSQC spectra were collected in phase sensitivity-enhanced pure-absorption mode. Both ^{1}H and ^{13}C NMR resonances were assigned from two-dimensional experiments and ^{1}H-decoupled ^{13}C NMR spectra. Optical rotations were obtained on a Perkin—Elmer 141 polarimeter at 20°C. Melting points were measured on a Kofler stage. Deionizations were carried out with ion-exchange resins Amberlite IRA-402 in the HCO_3^- form and Dowex 50W X-4, in the H^+ form. Solutions were concentrated under reduced pressure at temperatures below 40°C.

The composition of reaction mixtures and the purity of isolated saccharides were examined also by chromatography on Whatman No. 1 paper, using the

elution with solvent system S_1 butanol—ethanol—water (volume ratio = 5 1 4) for 18—20 h followed by visualization with alkaline silver nitrate.

Nitromethane Synthesis and the Nef Reaction with D-Fructose

D-Fructose (25 g) was dissolved in methanol (110 cm^3) and nitromethane (56 cm^3) and under occasional agitation, a cold solution of sodium methoxide (6.25 g of sodium in 175 cm^3 of methanol) was added in portions. Reaction mixture became turbid. After addition of butanol (500 cm^3) a voluminous precipitate occurred at once. The precipitate was filtered with suction and immediately added into vigorously stirred 4.5 M-H_2SO_4 (500 cm^3) at 25 °C and the reaction mixture was allowed to stand for 1 h. The acidic solution was neutralized with an excess of barium carbonate (695 g) by stirring the suspension for 2 h. The neutral mixture was filtered with suction. Clear filtrate was concetrated under reduced pressure to a sirup which was dissolved in tap water (2 dm^3) and baker's yeast was added. D-Fructose was removed by a week fermentation. The fermented solution was filtered, concentrated (to 200 cm^3), then methanol (200 cm^3) was added and the mixture was treated with charcoal (1 g) and filtered. Afterwards it was deionized with cation (H^+ form) and anion (HCO_3^- form) exchange resins and evaporated. The obtained yellow sirup (3.46 g, 18.2 %) contained branched-chain saccharides, namely 2-*C*-(hydroxymethyl)-D-mannose and -D-glucose.

2-*C*-(Hydroxymethyl)-D-mannose (*IV*)

a) The mixture of 2-*C*-(hydroxymethyl)-D-mannose and 2-*C*-(hydroxymethyl)-D-glucose (1 g; 4.76 mmol) dissolved in water (8 cm^3) was stirred with a solution of phenylhydrazine (0.5 cm^3, 4.76 mmol) in ethanol (2 cm^3) for 2 h at room temperature and then it was placed in a refrigerator for 24 h. After filtration of crystals these were washed with cold water (2 × 5 cm^3), and dried in desiccator in the presence of phosphorus pentoxide to give 2-*C*-(hydroxymethyl)-D-mannose phenylhydrazone (0.45 g, 77 %), m.p. = 184—185 °C.

A mixture of 2-*C*-(hydroxymethyl)-D-mannose phenylhydrazone (0.4 g), water (1.8 cm^3), methanol (0.15 cm^3), benzaldehyde (0.1 cm^3), and pyridine (0.5 cm^3) was heated at 100 °C for 3 h with stirring. The reaction mixture was filtered, washed with water (2 cm^3). The filtrate was extracted with ethyl acetate (3 × 5 cm^3), purified with charcoal and evaporated *in vacuo* to a sirupy *IV*. Yield = 0.25 g (89 %), [α](D, 20°C, ρ = 20 g dm^{-3}, water) = + 11.0°, R_{Fru} = 0.75 (S_1). ^{1}H NMR spectrum (D_2O), δ: 5.08 (s, H-1α), 4.87 (s, H-1β), 3.90 (dd, H-6aα), 3.84 (m, H-5α), 3.81 (dd, H-6bα), 3.76 (s, H-2′a,bβ), 3.75 (s, H-2′a,bα), 3.71 (dd, H-6aβ), 3.70 (d, H-3α), 3.68 (d, H-3β), 3.64 (m, H-4α,β), 3.59 (dd, H-6bβ), 3.39 (m, H-5β). ^{13}C NMR spectrum (D_2O), δ: 97.0 (C-1α), 96.9 (C-1β), 78.7 (C-5β), 78.4 (C-2α), 78.1 (C-2β), 74.9 (C-3β), 74.8 (C-5α), 74.2 (C-3α), 70.5 (C-4α), 70.5 (C-4β), 66.3 (C-2′α), 63.9 (C-6α), 63.9 (C-2′β), 63.3 (C-6β).

b) A mixture of 2-*C*-(hydroxymethyl)-2,3:5,6-di-*O*-isopropylidene-D-mannofuranose [5] (0.28 g), water (8 cm^3), and Dowex 50 W X-4 in H^+ form (1 cm^3) was stirred at 70 °C for 5 h. The resin was then filtered off, washed with water (3 × 5 cm^3), and the filtrates were purified with charcoal and evaporated to dryness to give sirupy 2-*C*-(hydroxymethyl)-D-mannose (0.19 g, 93 %).

2-*C*-(Hydroxymethyl)-D-glucose (*V*)

The mother liquor obtained after removal of 2-*C*-(hydroxymethyl)-D-mannose phenylhydrazone was evaporated *in vacuo* to sirup. A mixture of the sirupy residue (0.87 g), water (3.5 cm^3), methanol (0.3 cm^3), benzaldehyde (0.2 cm^3), and pyridine (0.1 cm^3) was heated at 100 °C for 3 h with stirring. The reaction mixture was filtered, washed with water (3 cm^3). The filtrate was extracted with ethyl acetate (3 × 10 cm^3), purified with charcoal and evaporated *in vacuo* to a sirupy residue (0.55 g, 60 %). Examination by ^{1}H NMR spectroscopy of the product showed a *ca.* 85 % purity of 2-*C*-(hydroxymethyl)-D-glucose with the admixture of 2-*C*-(hydroxymethyl)-D-mannose. Chromatography of the sirupy residue (0.2 g) on a column (95 cm × 1.6 cm) of Dowex 50W X-8 (37—75 μm) in the Ba^{2+} form afforded *V* (0.12 g, 70 %), sirup with [α](D, 20 °C, ρ = 27 g dm^{-3}, water) = + 27.4°, R_{Fru} = 0.65 (S_1). ^{1}H NMR spectrum (D_2O), δ: 5.18 (s, H-1α), 4.72 (s, H-1β), 3.96 (d, H-2′aβ), 3.89 (d, H-3α), 3.86 (d, H-2′bβ), 3.84 (m, H-5α), 3.75—3.84 (m, H-2′a,bα, H-6a,bα), 3.82 (dd, H-6aβ), 3.69 (dd, H-6bβ), 3.58 (d, H-3β), 3.52 (t, H-4α), 3.52 (t, H-4β), 3.46 (m, H-5β). ^{13}C NMR spectrum (D_2O), δ: 101.1 (C-1β), 94.8 (C-1α), 80.9 (C-3β), 79.6 (C-5β), 78.00 (C-2α), 77.3 (C-3α), 76.7 (C-2β), 74.6 (C-5α), 71.0 (C-4α), 71.0 (C-4β), 63.9 (C-6β), 63.6 (C-6α), 63.6 (C-2′α), 62.8 (C-2′β).

D-*gluco*-Hept-2-ulose (*VI*)

2-*C*-(Hydroxymethyl)-D-mannose (0.4 g) dissolved in 0.2 % aqueous solution of molybdic acid (20 cm^3) was heated at 80 °C for 1.5 h. The cold reaction mixture was then stirred with Amberlite IRA-400 in HCO_3^- form (10 cm^3), which was filtered off after 15 min and washed with water (3 × 10 cm^3). The filtrate was concentrated to a sirup, which was dissolved in a small quantity of water and then methanol was added. Seeding of the solution with authentic crystals of D-*gluco*-hept-2-ulose yielded crystalline *VI* (0.32 g, 80 %) which after recrystallization had m.p. = 171—173 °C, [α](D, 23 °C, ρ = 20 g dm^{-3}, water) = + 65.5° Ref.

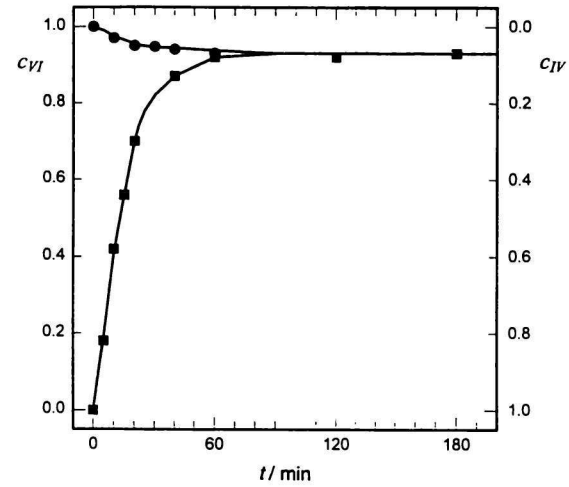


Fig. 1. Time dependence of the concentration of 2-*C*-(hydroxymethyl)-D-mannose (*IV*) (■) and D-*gluco*-hept-2-ulose (*VI*) (●) during the conversion to their mutual equilibrium in 0.2 % molybdic acid at 80 °C.

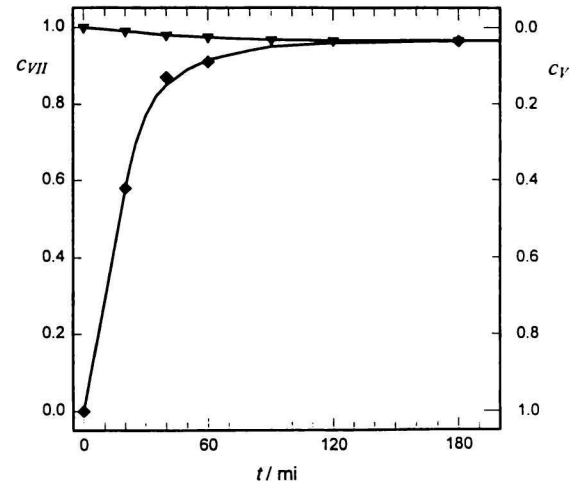


Fig. 2. Time dependence of the concentration of 2-*C*-(hydroxymethyl)-D-glucose (*V*) (♦) and D-*manno*-hept-2-ulose (*VII*) (▼) during the conversion to their mutual equilibrium in 0.2 % molybdic acid at 80 °C.

[8] gives m.p. = 171—174 °C and [α](D, 20 °C, ρ = 25 g dm^{-3}, water) = + 67.5°

Kinetic Analysis

A solution of 100 mg of 2-*C*-(hydroxymethyl)-D-mannose, 2-*C*-(hydroxymethyl)-D-glucose, D-*manno*-hept-2-ulose or D-*gluco*-hept-2-ulose, respectively, in 5 cm^3 of 0.2 % aqueous molybdic acid was kept at 80 °C. The 0.5 cm^3 samples of the reaction mixture were taken in time intervals, molybdic acid was removed with 3 cm^3 of Amberlite IRA-400 in the HCO_3^- form, and ^1H NMR spectra were measured to determine the mole ratio of *IV* and *VI* or *V* and *VII*, respectively, until the equilibria *IV* ⇌ *VI* and *V* ⇌ *VII* were reached (Figs. 1 and 2). The concentrations of the products at each time point c_t divided by their equilibrium concentrations c_∞ plotted *vs.* time are given in Figs. 3 and 4.

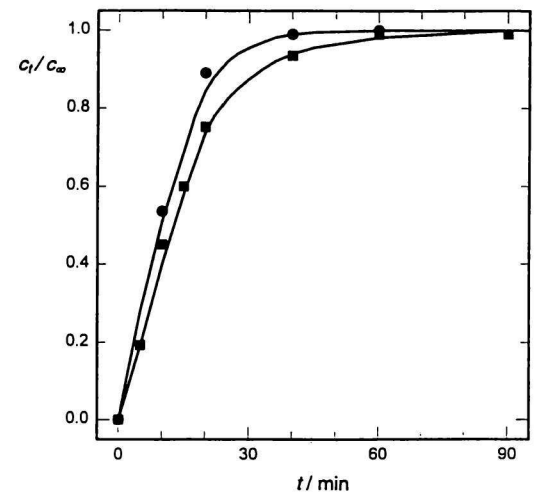


Fig. 3. Comparison of the rates of molybdic acid-catalyzed interconversions of 2-*C*-(hydroxymethyl)-D-mannose (*IV*) and D-*gluco*-hept-2-ulose (*VI*); conversion *IV* → *VI* (■), conversion *VI* → *IV* (●).

RESULTS AND DISCUSSION

Nitromethane synthesis with D-fructose followed by immediate Nef reaction of intermediate sodium 1-deoxy-2-*C*-(hydroxymethyl)hexitol-1-nitronates *II* and *III* (Scheme 1) afforded a mixture of 2-*C*-(hydroxymethyl)-D-mannose (*IV*) and -D-glucose (*V*) and starting D-fructose. After removal of D-fructose by fermentation with baker's yeast, 18 % of the branched-chain aldoses were obtained in the ratio x_r = 1.4 1 (determined by ^1H NMR). Branched-chain saccharides *IV* and *V* were separated as phenylhydrazones. 2-*C*-(Hydroxymethyl)-D-mannose phenylhydrazone was obtained by reaction of equimolar ratio of the saccharides mixture and phenylhydrazine at room temperature and *IV* was released from the hydrazone by reaction with benzaldehyde. Branched-chain sugar *IV* was prepared also by hydrolysis of its 2,3:5,6-di-*O*-isopropylidene derivative [5]. The residue after withdrawal of *IV via* its phenylhydrazone contained mainly branched-chain aldose *V* and about 15 % of *IV*. Sugar *V* free of *IV* was obtained by chromatographic purification of the residue on a column of Dowex 50W in the Ba^{2+} form.

The treatment of the 2-*C*-hydroxymethyl branched-chain hexoses *IV* and *V* with diluted molybdic acid as shown already in the preliminary communication [6] caused their rapid transformation to the corresponding hept-2-uloses. This transformation was now studied in detail under milder reaction conditions. Thus,

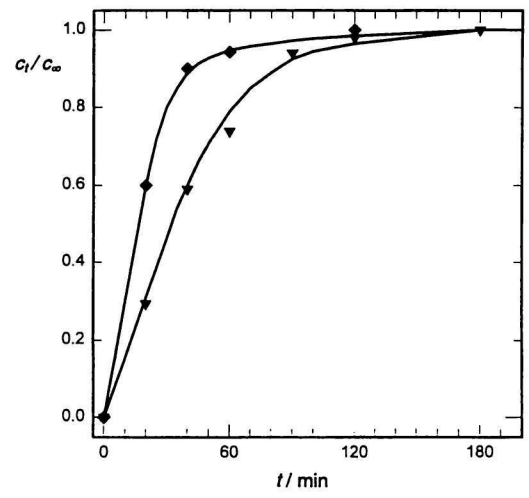
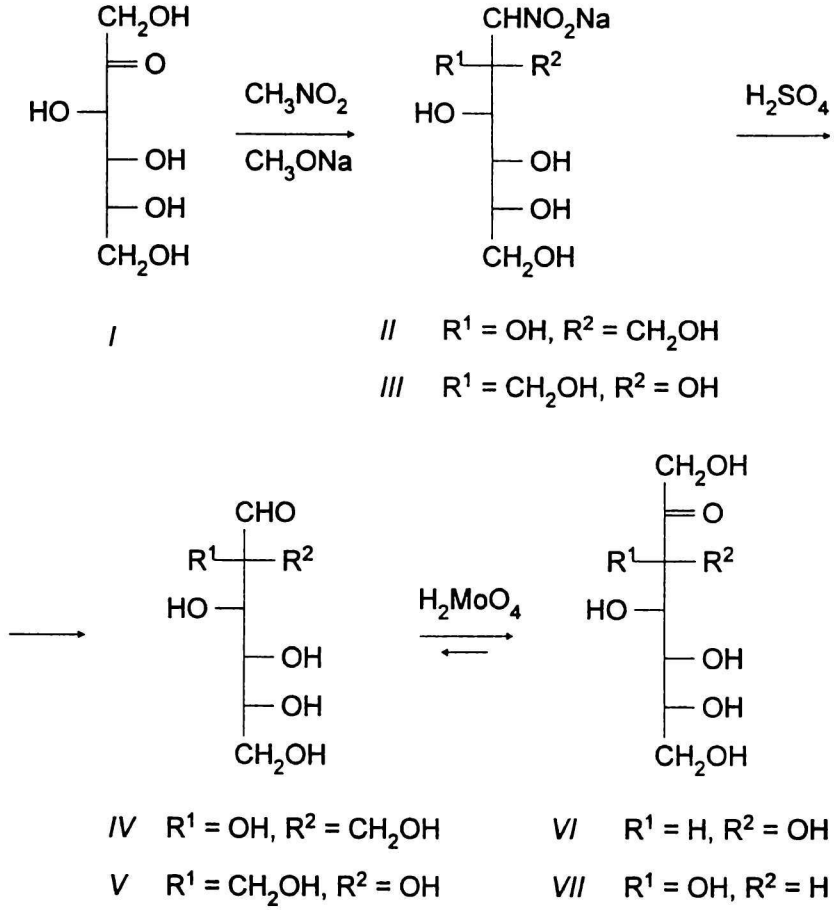


Fig. 4. Comparison of the rates of molybdic acid-catalyzed interconversions of 2-*C*-(hydroxymethyl)-D-glucose (*V*) and D-*manno*-hept-2-ulose (*VII*); conversion *V* → *VII* (♦), conversion *VII* → *V* (▼).

in 0.2 % aqueous solution of molybdic acid at 80 °C 2-*C*-(hydroxymethyl)-D-mannose reached equilibrium within 1 h (Fig. 1) that contained the starting sugar and D-*gluco*-hept-2-ulose (*VI*) in the ratio x_r = 2 23 (Fig. 1). The ratio was determined by integration of both H-1α and H-1β protons of *IV* and H-5 proton of *VI* in the reaction mixture. To confirm that the obtained ratio of sugars *IV* and *VI* is their thermodynamic equilibrium the transformation was carried out also from the side of heptulose *VI*. The same equilibrium of *IV* and *VI* (x_r = 2 23) was reached within 20 min under otherwise identical reaction conditions (Fig. 1). The molybdic acid-catalyzed mutual interconversion of another rearranging pair of sugars 2-*C*-(hydroxymethyl)-D-glucose (*V*) and D-*manno*-hept-2-ulose (*VII*) proceeded with slower reaction rate. The thermodynamic equilibrium of *V* and *VII* y_r = 2 55 determined by the integration of both H-1α and H-1β protons of *V* and H-5 and H-1a protons of *VII* was reached in 3 h under the same reaction conditions starting from the side of heptulose *VII* (Fig. 2). From the other side when branched-chain aldose *VI* was used as starting material the equilibrium of the reaction was reached within 1 h.

Unlike the preliminary study using ^{13}C NMR spectral analysis, the present more detailed analysis of

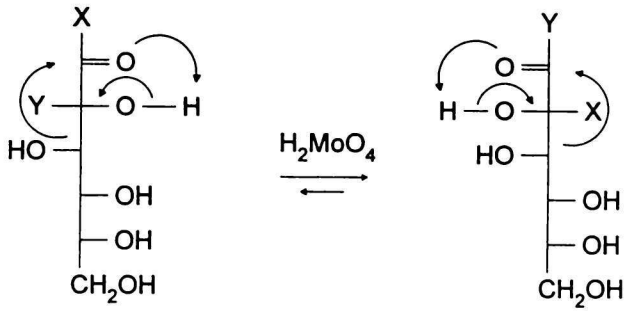


Scheme 1

the reaction mixtures including ^{1}H NMR spectra and 2D HSQC revealed the presence of small quantities of branched-chain aldoses *IV* and *V* in their reaction equilibria with the respective hept-2-uloses *VI* and *VII*. The obtained data suggest that the conversions of *IV* and *V* to respective *VI* and *VII* are not irreversible but their reaction equilibria are strongly shifted to the side of hept-2-uloses. These relatively low mole ratios of the interconverting branched-chain aldoses to ketoses (2 23 and 2 55, respectively) caused the former simplified interpretation of their ^{13}C NMR spectra and inaccurate conclusions [6].

The convenient spectral analysis of the reaction mixtures enabled to perform also a kinetic analysis of the transformations. Thus the rates of conversions of *IV*, *V*, *VI* or *VII* to equilibria were followed with time at identical reaction conditions. For a simple reversible reaction A $\rightleftharpoons$ B, the dependences c_t/c_∞ *vs.* time for both forward and reverse reactions should be the same. However, the comparison of the dependences for the respective pair conversions *IV* $\rightarrow$ *VI* and *VI* $\rightarrow$ *IV* (Fig. 3) as well as *V* $\rightarrow$ *VII* and *VII* $\rightarrow$ *V* (Fig. 4) shows significant differences. The differences between rearranging pairs can be explained by the formation of unreactive complexes of sugars with molybdic acid [7]. Thus, *e.g.* 2-*C*-(hydroxymethyl)-D-mannose can form similar unreactive tridentate or tetradentate molybdate complexes as D-mannose [9, 10], which is not possible for D-*gluco*-hept-2-ulose. Therefore in early stages of conversion *IV* $\rightarrow$ *VI* the effective concentration of catalyst is lower than the total concentration so that the rate of conversion is lower. On the contrary, conversion *IV* $\rightarrow$ *VI* from its early stages is not catalyst-deficient due to the formation of catalytically inactive species so that its reaction rate is high. Similar differences found between the rates of conversions *V* $\rightarrow$ *VII* and *VII* $\rightarrow$ *V* could be explained analogically. Due to these difficulties the reaction rates of conversions were not further analyzed.

The mechanism of the Bílik reaction has been reliably proved using ^{13}C and ^{2}H isotopically substituted aldoses [7, 11]. The results of an earlier study using only ^{3}H isotopically labelled aldoses [12] are in accordance with this mechanism. According to the mechanism the carbon skeleton of aldoses rearranges during the reaction in such a way that the carbon atom C-1 of the starting aldose becomes the carbon atom C-2 of the product aldose and *vice versa*, while the other carbon atoms C-3, C-4, *etc.* do not change their positions in the carbon skeleton. It means that during the process the C-2—C-3 bond is broken simultaneously with the formation of a new bond C-1—C-3. Thus the hydrogen, deuterium, or tritium atom originally linked to the carbonyl carbon atom of the respective starting D-glucose (X = ^{1}H, ^{2}H, or ^{3}H, respectively; Y = H; Scheme 2) becomes bound to the carbon atom C-2 of the formed D-mannose and *vice versa*. On the basis of the known mechanism of the Bílik reaction different isotopically substituted sugars have been prepared [13, 14].



Scheme 2

In this work studied sugars *IV*—*VII* can be formally considered as substituted D-glucoses (*V*: X = H, Y = CH_2OH; *VI*: X = CH_2OH, Y = H) and D-mannoses (*IV*: X = CH_2OH, Y = H; *VII*: X = H, Y = CH_2OH; Scheme 2). Then the observation of the mutual interconversion of sugars *IV* $\rightleftharpoons$ *VI* and *V* $\rightleftharpoons$ *VII* catalyzed by molybdic acid can be regarded as another structural proof of the mechanism of the Bílik reaction. On the other hand, thus the Bílik reaction can be a convenient preparative tool also for preparation of 2-ketoses from 2-*C*-hydroxymethyl branched-chain aldoses and *vice versa*. The former case is here exemplified by a simple, one-step synthesis of D-*gluco*-hept-2-ulose obtained in a 80 % yield from easily available 2-*C*-(hydroxymethyl)-D-mannose. A useful example of an opposite application, the preparation of D-hamamelose from D-fructose is shown elsewhere [15].

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REFERENCES

1. Bílik, V., *Chem. Zvesti 26*, 183 (1972).
2. Bílik, V., *Chem. Zvesti 26*, 187 (1972).
3. Bílik, V., *Chem. Zvesti 26*, 372 (1972).
4. Angyal, S. J., Bethell, S. J., and Beveridge, R. J. *Carbohydr. Res. 73*, 9 (1979).
5. Ho, P. T., *Can. J. Chem. 57*, 381 (1979).
6. Petruš, L., Hricovíniová, Z., Petrušová, M., and Matulová, M., *Chem. Papers 50*, 373 (1996).
7. Hayes, M. L., Pennings, N. J., Serianni, A. S., and Barker, R., *J. Am. Chem. Soc. 104*, 6764 (1982).
8. Austin, W. C., *J. Am. Chem. Soc. 52*, 2106 (1930).
9. Alföldi, J., Petruš, L., and Bílik, V. *Collect. Czech. Chem. Commun. 43*, 1476 (1978).
10. Chapelle, S. and Verchère, J. F., *Carbohydr. Res. 277*, 39 (1995).
11. Bílik, V. and Matulová, M., *Chem. Papers 44*, 257 (1990).
12. Bílik, V., Petruš, L., and Farkaš, V. *Chem. Zvesti 29*, 690 (1975).

13. Clark, E. L., Jr. and Barker, R., *Carbohydr. Res. 153*, 253 (1986).
14. Serianni, A. S., Vuorinen, T., and Bondo, P. B., *J. Carbohydr. Chem. 9*, 513 (1990).
15. Hricovíniová, Z., Hricovíni, M., and Petruš, L., *Chem. Papers*, in press.

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