Formation of Pyrroles, 2-Pyrrolidones, and Pyridones by Heating of 4-Aminobutyric Acid and Reducing Sugars

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The Maillard reaction of 4-aminobutyric acid with reducing sugars (D-arabinose, D-glucose, L-rhamnose, D-fructose, maltose) was investigated in slightly acidic aqueous media. Eight pyrroles, 14 2-pyrrolidones, and 2 4-pyridones were characterized and quantified by capillary GC/MS. For structure elucidation the compounds were separated by preparative GC or synthesized and investigated by MS, IR, and ^{1}H NMR spectroscopy. Due to a blocked Strecker degradation, the 4-aminobutyric acid specific Maillard products are comparable to those of peptide-bound L-lysine. Thus, the title compounds generated in D-glucose and maltose/4-aminobutyric acid model experiments are structurally related to ϵ-pyrrolonorleucine and maltosine, respectively.

INTRODUCTION

4-Aminobutyric acid is a major free amino acid in coffee, cereals, and fruits. Up to now, only a few specific Maillard products with pyrrole and pyrrolidone structures were derived from this amino acid. In contrast to primary and secondary α-amino acids, the Strecker degradation of 4-aminobutyric acid with α-dicarbonyls is almost entirely suppressed. 4-Aminobutyric acid catalyzes the degradation of reducing sugars comparable to peptide-bound L-lysine (Kersten, 1991). Among the Maillard reaction products from D-glucose and amino acids, 4-aminobutyric acid specific products possess the strongest reducing ability (Ninomiya et al., 1992). So far, the structures of these compounds are unknown.

The major products from 4-aminobutyric acid and D-glucose, compounds **8** [4-[2-formyl-5-(hydroxymethyl)-1-pyrrolyl]butanoic acid], **6** [4-[2-(hydroxyacetyl)-1-pyrrolyl]butanoic acid], and **23** [4-(1,4-dihydro-3-hydroxy-2-methyl-4-oxo-1-pyridyl)butanoic acid], correspond to title compounds as characterized from peptide-bound L-lysine/D-glucose and maltose, respectively (Nakayama et al., 1980; Heyns et al., 1968; Ledl et al., 1989). The investigation of L-lysine-specific products from peptides and N^{α}-acetyl-L-lysine is more complicated. Therefore, the results of 4-aminobutyric acid/reducing sugar reactions may give further insight into the Maillard reaction of L-lysine. In addition, the identification of specific Maillard products is a prerequisite for mechanistic studies with ^{13}C-labeled sugars. This could be demonstrated with [1-^{13}C]-D-glucose, L-hydroxyproline, and L-proline, respectively (Tressl et al., 1993a).

In a series of model experiments 4-aminobutyric acid was heated with D-xylose, D-glucose, L-rhamnose, maltose, and D-fructose for 1.5 h at 100 and 160 °C, respectively. Eight pyrroles, 14 2-pyrrolidones, and 2 4-pyridones were identified by MS, IR, and ^{1}H NMR spectroscopy as described earlier (Tressl et al., 1985a; Helak et al., 1989). To a certain extent the pattern of the 2-pyrrolidones was similar to that of the corresponding pyrrolyl derivatives in L-hydroxyproline/D-glucose and pyrrolidines in the L-proline/D-glucose experiments (Tressl et al., 1985b). The flavor qualities of these compounds changed from nutty/cereal to roasty aromas.

EXPERIMENTAL PROCEDURES

Sample Preparation. Reaction of 4-Aminobutyric Acid with Monosaccharides, Disaccharides, and α-Dicarbonyls. Equimolar amounts (0.02 mol) of 4-aminobutyric acid and D-xylose (D-glucose, D-fructose, L-rhamnose, maltose; diacetyl, 2,5-hexanedione) dissolved in water (25 mL) were refluxed for 2 h and then autoclaved for 1.5 h at 160 °C in a stainless steel laboratory autoclave (Roth, I series) equipped with a 100-mL duran glass tube and heated by an electric heater with magnetic stirrer. After cooling to room temperature, the compounds were extracted three times at pH 5 with freshly distilled diethyl ether. The combined ether extracts were dried over anhydrous sodium sulfate and concentrated to a volume of about 1 mL on a 20-cm Vigreux column. Aliquots (250 μL) of the ether extracts were used to separate the components by liquid–solid chromatography (see below). The obtained fractions were concentrated to a volume of 1 mL and investigated by capillary GC/MS. According to the method of Tressl et al. (1970) the pyrroles were analyzed as methyl esters. Individual components were isolated by preparative GC and identified by MS, IR, and ^{1}H NMR spectroscopy.

Column Chromatography. The extracts of the 4-aminobutyric acid/reducing sugar (α-dicarbonyl compound) model experiments were separated by liquid–solid chromatography on silica gel (activity IV, column 20 × 9cm) into six fractions with pentane (F1), pentane–dichloromethane 4:1 (F2), 1:1 (F3), and 1:4 (F4), pentane–diethyl ether 1:1 (F5), and diethyl ether (F6) in 40-mL portions. Compound **3** could be isolated directly from fraction F6 without further purification. On basic alumina (activity IV) the extracts were separated into five fractions with pentane–dichloromethane 9:1 (F1) and 3:1 (F2), pentane–diethyl ether 9:1 (F3) and 1:1 (F4), and diethyl ether (F5) in 40-mL portions. Compound **15** could be isolated directly from fraction F5 without further purification.

Gas Chromatography (GC)/Mass Spectrometry (MS). The extracts prepared according to the described techniques were analyzed by GC/MS using a 25 m × 0.32 mm i.d. CP-Wax fused silica capillary column Chrompack (column A, temperature was programmed from 70 to 220 °C at 2 °C/min) and a 50 m × 0.32 mm i.d. CP-Sil fused silica capillary column Chrompack (column B, temperature was programmed from 80 to 280 °C at 4 °C/min). Compound **24** was analyzed on a 60 m × 0.32 mm i.d. DB-1 fused silica capillary column (column C, temperature was programmed to 180 °C at 4 °C/min). Columns A–C were coupled with a double-focusing mass spectrometer CH 5-DF (Varian MAT), ionization voltage 70 eV, resolution 2000 (10% valley).

0021-8561/93/1441-2125$04.00/0

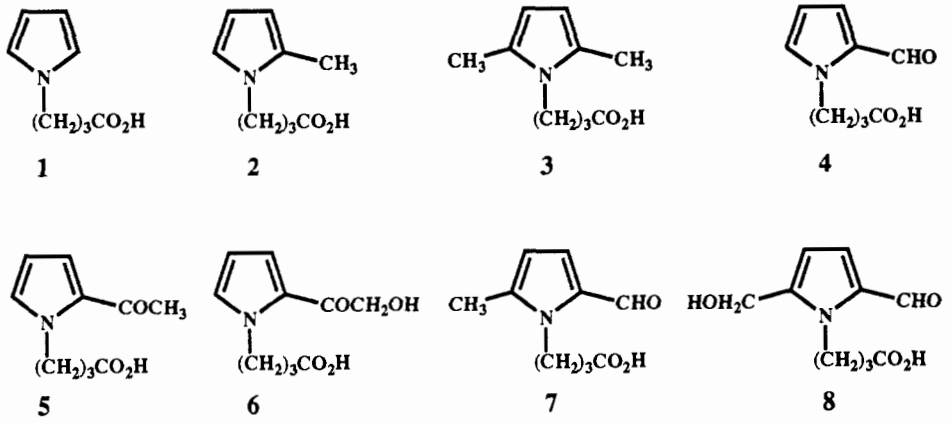


Figure 1. Pyrroles characterized in reducing sugar/4-aminobutyric acid model experiments.

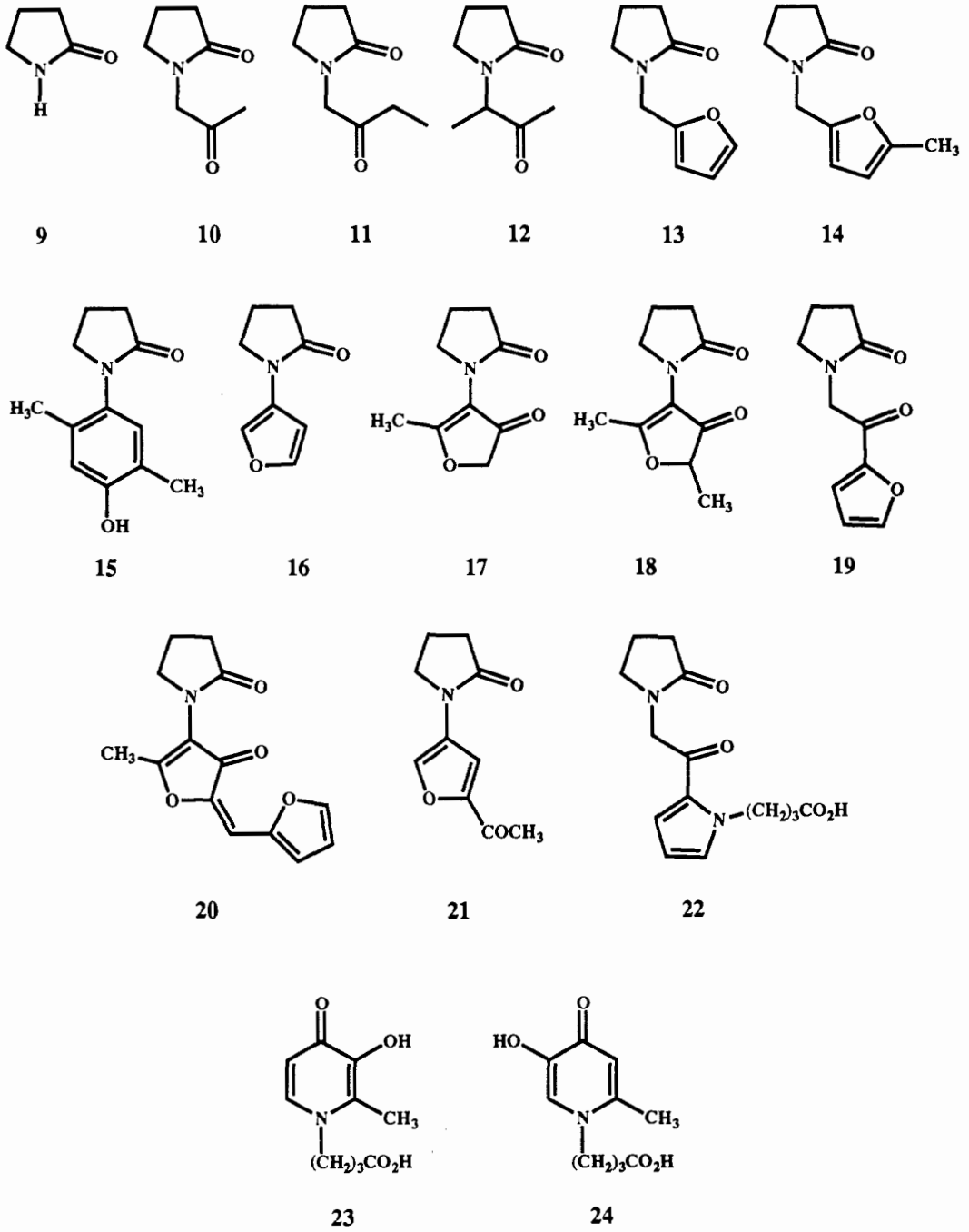


Figure 2. 2-Pyrrolidones and 4-pyridones characterized in reducing sugar/4-aminobutyric acid model experiments.

Preparative Gas Chromatography. For the isolation of individual compounds from the fractions of column chromatography, preparative GC was used. On a Varian Aerograph 2800 equipped with a 3 m × 0.25 in. glass column, compounds **10** and **18** was isolated on 15% Carbowax 20M on 80–90-mesh Chromosorb WAW/DMCS (column D) and compounds **4**, **5**, **7**, and

8 on 5% SP 2401 DB on 100–120-mesh Supelcoport. The column temperature was elevated from 60 to 230 °C at a rate of 4 °C/min.

^{1}H NMR and IR Spectroscopy. ^{1}H NMR spectra were recorded at 270 MHz on a Bruker WH 270 NMR spectrometer in $CDCl_3$ solution; chemical shifts are referenced to tetramethylsilane (TMS) as internal standard. Coupling constants (J) are in hertz. Infrared spectra (4000–400 cm^{-1}) were obtained from $CDCl_3$ or CCl_4 solutions.

Synthesis of 1-Furfuryl-2-pyrrolidone and -1-(5-Methylfurfuryl)-2-pyrrolidone. According to the method of La Londe et al. (1977) compounds **13** and **14** were synthesized by reaction of 4-aminobutyric acid with furfuraldehyde and 5-methyl-2-furfuraldehyde, respectively, by addition of CaO to a benzene solution of both components at room temperature over 24 h. The Schiff bases were reduced with $NaBH_4$. The isolated compounds were characterized by MS, IR, and ^{1}H NMR spectroscopy.

RESULTS AND DISCUSSION

The compounds generated by the reaction of 4-aminobutyric acid and reducing sugars at 100 and 160 °C in aqueous medium (Figures 1 and 2) were extracted with ether and investigated by capillary GC/MS. Further separation and purification of individual compounds was carried out by liquid–solid chromatography and preparative GC (**4**, **5**, **7**, **8**, **18**, **23**) and synthesis (**3**, **10**, **12–15**). The pyrroles **1–8** and the pyridones **23** and **24** were analyzed as methyl esters. Compounds **1**, **2**, **6**, **11**, **16**, **17**, **19**, **20**, and **22** were tentatively identified by their analogous mass spectrometric fragmentations to corresponding or structurally related reference samples. The MS and ^{1}H NMR spectroscopic data are summarized in Table I. The observed concentrations of the pyrroles and 2-pyrrolidones (Table II) demonstrate the strong influence of the reducing sugar on their formation.

Pyrroles. Compound **3** was synthesized from 2,5-hexanedione and 4-aminobutyric acid. Spectroscopic data allowed the identification of the homologous pyrroles **1** and **2**, which are formed from D-xylose, D-glucose (**1**), and L-rhamnose (**2**), respectively. Title compounds in the D-xylose/4-aminobutyric acid experiments are **4** and **17**. From the corresponding L-rhamnose system the structurally related compounds **5**, **7**, and **18** were isolated by preparative GC and identified by MS, IR, and ^{1}H NMR spectroscopy. The spectroscopic data of **4** isolated from D-xylose model experiments are in agreement with 4-(2-formyl-1-pyrrolyl)butanoic acid data. The mass spectrometric fragmentations of **4** [m/z (%) 195 (53, M^+), 167 (63, M – CO), 136 (82, M – CO_2CH_3, α-cleavage), 122 (100, M – $CH_2CO_2CH_3$, β-cleavage), 108 (56, M – CO – CO_2CH_3)] correspond to an N-alkyl-2-formylpyrrole. The fragment ions m/z 95, 94, 81, 80, and 53 result from rearrangements and hydrogen transfers. Compounds **5** and **7** were isolated from the L-rhamnose/4-aminobutyric acid model experiment. The spectroscopic data confirm **5** as 4-(2-acetyl-1-pyrrolyl)butanoic acid. The mass spectrometric fragmentation of the methyl ester [m/z (%) 209 (63, M^+), 193 (13, M – CH_3), 166 (11, M – $COCH_3$), 162 (11, M – CH_3 – CH_2OH), and 43 (87, CH_3CO)] is in agreement with N-alkyl-2-acetylpyrrole. α-, β-, and γ-cleavages of the alkyl chain form major ions at m/z 150 (13, M – CO_2CH_3), 136 (100, M – $CH_2CO_2CH_3$), and 122 (31, M – $CH_2CH_2CO_2CH_3$). Compound **6** was identified by its mass spectrometric fragmentation as a 2-(hydroxyacetyl)pyrrole. It showed an analogous fragmentation to the synthesized homologous 2-(2-(hydroxyacetyl)pyrrole-1-acetic acid (Podgorski et al., 1991) [m/z (%) 225 (31, M^+), 194 (72, M – CH_2OH), 162 (77, M – CH_2OH – CH_3OH), 150 (13, M – 75), and 136 (100, M – 89)]. The ions m/z 101, 94, 80, 53, and 59 are comparable to those of the structurally related **5**. The ^{1}H NMR and MS spectroscopic data of **7** correspond to an N-alkyl-2-formyl-5-methylpyrrole. The title compound of the D-glucose/4-aminobutyric acid model experiment was recently identified and possessed weak reducing ability (Ninomiya et al., 1992). All spectroscopic data of compound **8** are in agreement with those of 4-(2-formyl-5-(hydroxymethyl)-1-pyrrolyl)butanoic acid. N-Substituted 2-formyl-5-(hydroxymethyl)- and 2-(hydroxyacetyl)pyrroles were characterized as title compounds in model experiments of D-glucose with primary amines under mild conditions (Jurch and Tatum, 1970; Olsson et al., 1977; Njoroge et al., 1988; Beck et al., 1989). Because of their reactivities with nucleophiles, they are involved in the cross-linking reaction of proteins. Additionally, primary amino acids form pyrrole lactones and the corresponding pyrroles as identified in the 4-aminobutyric acid experiments (Shigematsu et al., 1971; Kato, 1967).

2-Pyrrolidones. Fourteen 2-pyrrolidones were characterized as 4-aminobutyric acid specific Maillard products. Some of these compounds possess roasty cereal aroma and flavor qualities and are stable at moderate pH ranges. In alkaline aqueous media 2-pyrrolidones are transformed into nonvolatile compounds, which are still under investigation. Compounds **10** and **12** were synthesized from 4-aminobutyric acid and lactaldehyde and acetoin, respectively. The spectroscopic data are in good agreement with 1-(2-oxoalkyl)-2-pyrrolidone structure. According to its analogous mass spectrometric fragmentation, **11** was identified. Compounds **13** and **14** were synthesized from 4-aminobutyric acid and furfuraldehyde (or 5-methylfurfurylaldehyde) by $NaBH_4$ reduction of the initially formed Schiff bases. Compound **15** is generated as title compound from 4-aminobutyric acid and diacetyl. Compound **18** was isolated from the L-rhamnose model experiment. The MS and NMR spectroscopic data are in agreement with those of 1-(2,5-dimethyl-3-oxo-2H-furan-4-yl)-2-pyrrolidone. According to its analogous mass spectrometric fragmentation, **17** was characterized as the corresponding 5-methyl-3-oxo-2H-4-furanyl derivative. The methylene active **17** formed colored products during isolation from the D-xylose system. One of these products was identified as the 2-furylidene derivative **20**. Compound **16** is formed as a minor constituent and was tentatively identified by its mass spectrometric fragmentation, chromatographic separation, and chemical reactions. Compound **19** is generated as a reactive intermediate during heating of D-glucose and 4-aminobutyric acid. The mass spectrum of **19** is consistent with that of 1-(2-furoylmethyl)-2-pyrrolidone [m/z (%) 193 (12, M^+), 110 (64, M – 83), 98 (100, M – 95), 95 (46, M – 98)]. In alkaline aqueous medium **19** is transformed into the corresponding furosine derivative. According to its analogous mass spectrometric fragmentation, **22** was tentatively characterized as a structurally related pyrrole derivative 4-[2-[[(2-oxo-1-pyrrolidinyl)methyl]carbonyl]-1-pyrrolyl]butanoic acid. The mass spectrometric data of **22** are in agreement with the fragmentation of **19** and **6**. Compound **21** is formed during heating of D-fructose and 4-aminobutyric acid as major product. The mass spectrometric data of **21** [m/z (%) 193 (85, M^+), 150 (60, M – CH_3CO), 138 (100, M – 55), 110 (6, M – 83), 43 (70, CH_3CO)] are in agreement with those of 1-(2-acetyl-4-furyl)-2-pyrrolidone.

4-Pyridones. Compound **23** was isolated from the maltose/4-aminobutyric acid experiment and synthesized from maltol and 4-aminobutyric acid. Corresponding pyridones had been characterized in model experiments of maltose, glycine, and N-acetyl-L-lysine, respectively (Loidl, 1976; Ledl et al., 1989). The MS and ^{1}H NMR spectroscopic data are consistent with those of 1-(3-carboxypropyl)-3-hydroxy-2-methyl-4-pyridone. After methylation with diazomethane, **23** was analyzed as a mixture

Table I. MS Data and ^{1}H NMR Data of Selected Products, Characterized in γ-Aminobutyric Acid Model Experiments with Reducing Sugars and α-Dicarbonyls[a]

	Pyrroles
1 4-(1-pyrrolyl)butanoic acid (identified as methyl butanoate)	167 (43), 136 (36), 108 (14), 106 (97), 101 (4), 94 (36), 93 (22), 81 (100), 80 (67), 68 (19), 67 (18), 59 (20), 53 (74), 41 (43), 39 (17) (I_K CP-Wax 1946)
2 4-(2-methyl-1-pyrrolyl)butanoic acid (identified as methyl butanoate)	181 (42), 150 (18), 122 (18), 108 (10), 94 (100), 80 (20), 67 (18), 59 (20) (I_K CP-Wax 2055)
3 4-(2,5-dimethyl-1-pyrrolyl)butanoic acid (identified as methyl butanoate)	195 (84), 180 (7), 164 (43), 136 (14), 120 (11), 109 (72), 108 (100), 101 (47), 94 (63), 59 (52), 53 (11), 41 (26), 39 (11); δ 1.95 (qui, J = 7 Hz, 2H, NCH_2CH_2), 2.04 (s, 6H, 2,5-CH_3), 2.38 (t, J = 7 Hz, $CH_2CO_2CH_3$), 3.70 (s,3H, CO_2CH_3), 3.79 (m, 2H, NCH_2), 5.78 (s, 2H, pyrrole H-3/H-4) (I_K CP-Sil 1525)
4 1-(2-formyl-1-pyrrolyl)butanoic acid (identified as methyl butanoate)	195 (53), 167 (63), 136 (82), 122 (100), 108 (56), 106 (23), 95 (23), 94 (66), 93 (19), 81 (56), 80 (42), 69 (19), 68 (20), 67 (18), 59 (43), 41 (48), 39 (49); δ 2.10 (qui, J = 7 Hz, 2H, NCH_2CH_2), 2.30 (t, J = 7 Hz, 2H, CH_2CO_2R), 3.66 (s, 3H, CO_2CH_3), 4.38 (t, J = 7 Hz, 2H, NCH_2), 6.22 (dd, J = 3.8, 2.6 Hz, 1H, pyrrole H-4), 6.92 (mc, 2H, pyrrole H-3,H-5), 9.52 (d, J = 0.8 Hz, 1H, CHO) (I_K CP-Wax 2400)
5 4-(2-acetyl-1-pyrrolyl)butanoic acid (identified as methyl butanoate)	209 (63), 194 (13), 166 (11), 162 (11), 150 (13), 136 (100), 122 (31), 109 (12), 108 (12), 106 (17), 101 (11), 94 (41), 80 (22), 59 (30), 43 (87), 41 (23), 39 (23); δ 2.07 (qui, J = 7 Hz, 2H, NCH_2CH_2), 2.29 (t, J = 7 Hz, 2H, CH_2CO_2R), 2.45 (s, 3H, $COCH_3$), 3.67 (s, 3H, CO_2CH_3), 4.36 (t, J = 7 Hz, 2H, NCH_2), 6.14 (dd, J = 3.8, 2.6 Hz, 1H, pyrrole H-4), 6.84 (m, 1H, pyrrole H-5), 6.97 (dd, J = 3.8, 1.4 Hz, 1H, pyrrole H-3) (I_K CP-Wax 2433)
6 4-[2-(hydroxyacetyl)-1-pyrrolyl]butanoic acid (identified as methyl butanoate)	225 (31), 210 (21), 194 (72), 162 (77), 150 (83), 136 (87), 135 (37), 107 (26), 106 (31), 101 (36), 94 (29), 82 (23), 80 (25), 69 (38), 59 (100) (I_K CP-Sil 1690)
7 4-(2-formyl-5-methyl-1-pyrrolyl)butanoic acid (identified as methyl butanoate)	209 (100), 194 (9), 192 (16), 181 (32), 180 (32), 150 (89), 148 (35), 136 (67), 122 (41), 120 (40), 108 (53), 101 (26), 95 (32), 94 (44), 80 (50), 69 (55), 59 (50), 53 (37), 41 (37), 39 (23); δ 2.02 (qui, J = 7 Hz, 2H, NCH_2CH_2), 2.31 (s, 3H, 5-CH_3), 2.38 (t, J = 7 Hz, 2H, $CH_2CO_2CH_3$), 3.68 (s, 3H, CO_2CH_3), 4.34 (m, 2H, NCH_2), 6.02, 6.85 (each d, J = 3.6 Hz, 1H, pyrrole H-3/H-4), 9.39 (s, 1H, CHO) (I_K CP-Wax 2525)
8 4-[2-formyl-5-(hydroxymethyl)-1-pyrrolyl]butanoic acid (identified as methyl butanoate)	225 (56), 208 (5), 207 (5), 196 (77), 168 (44), 166 (37), 164 (64), 148 (25), 136 (100), 124 (29), 120 (18), 108 (73), 106 (19), 101 (24), 96 (17), 94 (13), 93 (11), 80 (32), 69 (18), 68 (25), 59 (81), 41 (49), 39 (33); δ 2.10 (qui, J = 7.3 Hz, 2H, NCH_2CH_2), 2.45 (t, J = 7.3 Hz, CH_2COCH_3), 2.73 (br s, 1H, OH), 3.70 (s, 3H, CO_2CH_3), 4.42 (mc, 2H, NCH_2), 4.70 (s, 2H, CH_2OH), 6.25 (d, J = 3.6 Hz, 1H, pyrrole H-3), 6.88 (d, J = 3.6 Hz, 1H, pyrrole H-4), 9.50 (s, 1H, CHO) (I_K CP-Sil 1740)
	2-Pyrrolidones
9 2-pyrrolidone	85 (100), 84 (44), 56 (18), 42 (80), 30 (60) (I_K CP-Wax 2068)
10 1-(2-oxopropyl)-2-pyrrolidone	141 (13), 126 (3), 99 (25), 98 (100), 84 (23), 70 (58), 69 (33), 43 (49), 41 (49); δ 2.08 (qui, J = 7.5 Hz, 2H, H-4), 2.16 (s, 3H, $COCH_3$), 2.44 (t, J = 7.5 Hz, 2H, H-3), 3.46 (t, J = 7.6 Hz, 2H, H-5), 4.12 (s, 2H, NCH_2CO) (I_K CP-Wax 2252)
11 1-(2-oxobutyl)-2-pyrrolidone	155 (12), 140 (4), 126 (10), 112 (9), 98 (100), 84 (69), 70 (60), 69 (29), 56 (11), 43 (25), 41 (37) (I_K CP-Sil 1167)
12 1-(3-oxo-2-butyl)-2-pyrrolidone	155 (26), 112 (100), 99 (15), 98 (55), 84 (18), 70 (26), 69 (40), 56 (8), 41 (37) (I_K DB1 1286)
13 1-furfuryl-2-pyrrolidone	165 (58), 148 (6), 137 (10), 136 (14), 120 (10), 109 (18), 94 (16), 84 (23), 82 (32), 81 (100), 69 (12), 53 (44), 41 (23), 39 (15); δ 1.94 (qui, J = 7.5 Hz, 2H, H-4), 2.34 (t, J = 7.5 Hz, 2H, H-3), 3.28 (t, J = 7.5 Hz, 2H, H-5), 4.20 (s, 2H, NCH_2), 6.17 (dd, J = 0.8, 3.3 Hz, 1H, furan H-3), 6.25 (dd, J = 1.9, 3.3 Hz, furan H-4), 7.28 (dd, J = 0.8, 1.9 Hz, 1H, furan H-5) (I_K CP-Wax 2366)
14 1-(5-methylfurfuryl)-2-pyrrolidone	179 (50), 164 (8), 151 (4), 150 (8), 136 (47), 110 (8), 109 (19), 108 (24), 95 (100), 82 (17), 80 (11), 69 (52), 43 (56), 41 (45); δ 2.00 (qui, J = 7.5 Hz, 2H, H-4), 2.28 (s, 3H, CH_3), 2.42 (br t, J = 7.5 Hz, 2H, H-3), 3.35 (t, J = 7.5 Hz, 2H, H-5), 4.38 (s, 2H, NCH_2), 5.88, 6.11 (each d, J = 3.3 Hz, 1H, furan H-3/4) (I_K CP-Wax 2434)
15 1-(4-hydroxy-2,5-dimethylphenyl)-2-pyrrolidone (identified as 4-methoxy compound)	219 (100), 204 (8), 188 (28), 177 (8), 164 (90), 149 (40), 148 (32), 136 (17), 135 (12), 134 (32), 120 (10), 107 (10), 106 (12), 91 (30), 86 (42), 79 (18), 77 (19), 69 (13), 65 (15), 53 (8), 51 (8), 46 (12), 39 (25); δ 2.15 (m, 2H, H-4), 2.16, 2.32 (each s, 3H, CH_3), 2.55 (br t, J = 7 Hz, 2H, H-3), 3.71 (t, J = 7.5 Hz, 2H, H-5), 3.79 (s, 3H, OCH_3), 6.73, 6.98 (each s, 1H, furan H-3/6) (I_K CP-Wax >2700)
16 1-(3-furyl)-2-pyrrolidone	151 (34), 122 (5), 96 (100), 95 (11), 94 (13), 80 (3), 68 (5), 67 (7), 55 (4), 53 (4), 52 (8), 42 (18), 41 (51), 40 (10) (I_K CP-Wax 2424)
17 1-(5-methyl-3-oxo-2H-furan-4-yl)-2-pyrrolidone	181 (100), 153 (9), 126 (60), 110 (11), 84 (49), 55 (11), 43 (38), 41 (28) (I_K CP-Sil 1406)
18 1-(2,5-dimethyl-3-oxo-2H-furan-4-yl)-2-pyrrolidone	195 (100), 180 (1), 152 (6), 140 (42), 124 (13), 122 (8), 112 (15), 110 (8), 98 (33), 84 (11), 69 (16), 68 (16), 55 (10), 43 (49), 42 (15), 41 (25); δ 1.50 (d, J = 6.8 Hz, 3H, $CHCH_3$), 2.17 (qui, J = 7.5 Hz, 2H, H-4), 2.24 (s, 3H, $=CCH_3$), 2.47 (t, J = 7.5 Hz, 2H, H-3), 3.73 (t, J = 7.5 Hz, 2H, H-5), 4.55 (br q, J = 6.8 Hz, 1H, $CHCH_3$) (I_K CP-Wax 2500)
19 1-(2-furoylmethyl)-2-pyrrolidone	193 (12), 110 (64), 98 (100), 95 (46), 84 (39), 70 (70), 69 (37), 43 (46), 41 (53), 40 (42) (I_K CP-Sil 1600)
20 1-(2-furfurylidene-5-methyl-3-oxo-2H-furan-4-yl)-2-pyrrolidone	259 (100), 204 (83), 176 (66), 174 (27), 162 (12), 146 (22), 106 (24), 84 (7), 78 (15), 69 (27), 55 (10), 41 (57) (I_K DB-1 2310)
21 1-(2-acetyl-4-furyl)-2-pyrrolidone	193 (85), 178 (5), 164 (1), 150 (60), 138 (100), 136 (5), 122 (45), 110 (6), 95 (3), 94 (9), 80 (3), 69 (7), 68 (11), 67 (8), 55 (12), 43 (70) (I_K DB-1 1820)
22 4-[2-[[(2-oxo-1-pyrrolidinyl)methyl]carbonyl]-1-pyrrolyl]butanoic acid (identified as methyl butanoate)	292 (1), 209 (97), 194 (48), 162 (52), 150 (100), 136 (4), 135 (15), 126 (23), 107 (8), 106 (10), 101 (15), 98 (97), 94 (8), 80 (11), 79 (9), 70 (30), 69 (27), 59 (38), 41 (50) (I_K DB-1 2348)

Table I (Continued)

4-Pyridones	
23 4-(1,4-dihydro-3-hydroxy-2-methyl-4-oxo-1-pyridyl)butanoic acid	δ 1.96 (qui, J = 7.5 Hz, 2H, NCH_2CH_2), 2.33 (t, J = 7 Hz, 2H, CH_2CO_2H), 2.39 (s, 3H, 2-CH_3), 3.99 (t, J = 7.6 Hz, 2H, NCH_2), 6.23, 7.43 (each d, J = 7.3 Hz, each 1H, H-5/H-6) (I_K CP-Sil 1987); further identification was made after methylation to a mixture of **23a**/**23b**
23a methyl 4-(1,4-dihydro-3-methoxy-2-methyl-4-oxo-1-pyridyl)butanoate	239 (67), 224 (11), 208 (8), 207 (31), 180 (39), 166 (42), 162 (11), 153 (21), 150 (16), 148 (31), 139 (56), 136 (24), 124 (18), 121 (40), 109 (29), 101 (66), 67 (20), 59 (100), 53 (20), 41 (45); δ 2.04 (m, 2H, NCH_2CH_2), 2.42 (t, J = 7 Hz, 2H, $CH_2CO_2CH_3$), 2.60 (s, 3H, 2-CH_3), 3.71 (s, 3H, CO_2CH_3), 3.86 (s, 3H, OCH_3), 3.90 (t, J = 7 Hz, NCH_2), 6.38, 7.24 (each d, J = 7.4 Hz, each 1H, H-5/H-6) (I_K DB-1 2365)
23b 1-[3-(methoxycarbonyl)propyl]-2-methyl-4-methoxypyridinium-3-olate	δ 2.13 (m, 2H, NCH_2CH_2), 2.39 (s, 3H, 2-CH_3), 2.42 (t, J = 7 Hz, 2H, $CH_2CO_2CH_3$), 3.72 (s, 3H, CO_2CH_3), 3.92 (s, 3H, OCH_3), 4.12 (t, J = 7 Hz, 2H, NCH_2), 6.67, 7.08 (each d, J = 6.3 Hz, each 1H, H-5/H-6)
24 4-(1,4-dihydro-5-hydroxy-2-methyl-4-oxo-1-pyridyl)butanoic acid (identified as methyl butanoate)	225 (96), 208 (10), 196 (20), 166 (60), 164 (32), 152 (39), 138 (32), 124 (53), 110 (26), 101 (72), 69 (41), 59 (100), 55 (49), 43 (69), 41 (97) (I_K DB-1 1930)

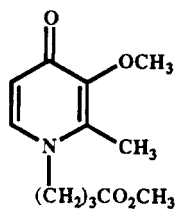
[a] MS data: m/z (relative intensity). ^{1}H NMR data: s, singlet; d, doublet; t, triplet; qui, quintet; m, multiplet; mc, center of a multiplet, br s, broad singlet; br t, broad triplet; br q, broad quartet, dd, doublet of doublets.

Table II. Selected 2-Pyrrolidones and 4-(1-Pyrrolyl)butyric Acids Characterized in 4-Aminobutyric Acid/Sugar Model Experiments[a]

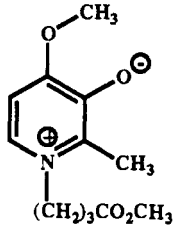
component	formation with[b] Xyl	Glu	Rha	Mal
1	225	225	–	20
2	–	–	50	–
4	1870	215	–	15
5	35	1250	2350	100
6	–	+	–	+
7	–	90	8235	10
8	–	3000	–	1000
9	+	2370	+	70
10	40	865	445	+
11	+	+	–	–
13	+	85	+	30
14	–	–	180	–
16	–	85	+	–
17	600	–	–	–
18	–	105	755	–
19	–	+	–	+

[a] 1.5 h, 160 °C; data represent concentrations in ppm. Internal standards: 2-(methoxycarbonyl)pyrrole, 1-methyl-2-pyrrolidone.[b] +, <1 ppm; –, not detectable.

of **23a** (75%) and **23b** (25%) by NMR spectroscopy, demonstrating the tautomeric equilibrium between the 3-hydroxy-4-pyridone and the 4-hydroxypyridinium betaine form of this compound as well as the strong chelate-forming tendency of **23**. According to its analogous MS spectrum, **24** was characterized in D-glucose/4-aminobutyric acid model experiments.



23a



23b

The results of Table II demonstrate that the formation of pyrroles is dependent on the monosaccharides. The formation pathways by the postulated 3-deoxyaldoketose/1-deoxydiketose routes cannot be proved without labeling experiments. Thus, compound **5** is formed as a major product from D-glucose/4-aminobutyric acid as well as from L-rhamnose/4-aminobutyric acid. The 6-deoxyaldose product should be generated via 3-deoxyaldoketose and the D-glucose product via 1-deoxydiketose as reactive intermediates. On the other hand, the low formation of pyrrole **8** from D-fructose/4-aminobutyric acid experiments cannot be explained. In a subsequent paper of this series (Tressl et al., 1993b) the chemical pathways will be rationalized on the basis of analogous experiments starting with labeled monosaccharides.

ACKNOWLEDGMENT

This work was supported by the Deutsche Forschungsgemeinschaft, Bonn-Bad Godesberg, and by the Arbeitsgemeinschaft Industrieller Forschungsvereinigungen e.V., Köln. We are grateful to H. Köppler for assistance in mass spectrometric analysis.

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Received for review June 3, 1993. Revised manuscript received August 19, 1993. Accepted August 30, 1993.[⊗]

[⊗] Abstract published in *Advance ACS Abstracts,* October 15, 1993.