



Figure 1. Influence on browning of three series of amino acids.

the monoamino acids containing the amino group in the alpha position caused only a small increase in color compared to those containing the amino group in the terminal position or to those containing two amino groups. The 6-carbon diamino acid, lysine, produced the greatest increase in color formation of all the 12 acids tested. This tremendous acceleration of browning by lysine does not seem to be wholly related to the general basicity which this compound would impart to a glucose solution. It was pointed out in the first paper of this series that arginine and histidine do not show the same positive effect as lysine. These two amino acids are basic in their reaction when in solution, but have different molecular structures than lysine. Another aliphatic diamino acid closely related to lysine, ornithine ( $\delta,\alpha$ -diamino valeric acid), also caused increased color development in the glucose solution. Apparently, then, the form of the molecular structure of the amino acid as well as its acid-base reaction in solution is a factor in determining its influence on browning.

The chain length of the amino acids produced an effect on the coloring of the glucose solution which varied with the type of amino acid. Figure 1 shows that the longer chained aliphatic alpha mono-amino acids, such as norvaline and norleucine, caused the production of only slightly more color than the shorter chained acids such as alanine. The absolute amounts of color which developed in the glucose solutions treated with monoamino aliphatic acids were only slightly more than the amount developed in the glucose control. Again it should be noted that the influence of the monoamino aliphatic acids as a whole was very slight compared to the terminal and diamino acids.

The monoamino acids containing the amino group in the terminal position

produced variable results on browning. Increasing their chain length from 3 to 4 carbons caused a significant increase in the amount of color in the sugar solution. However, the 5-carbon acid,  $\delta$ -amino valeric acid, produced a smaller amount of color than the 4-carbon acid. Further, the solution containing the 6-carbon acid,  $\epsilon$ -amino caproic acid, did not show any more color than the glucose control. One possible explanation of this reversal in effect with increase in chain length is that the terminal amino group in 3- and 4-carbon monoamino acids are kept at a distance from the carboxyl group, whereas the amino group in 5- and 6-carbon acids, because of the latter's ability to bend, is brought closer and closer to the carboxyl group, thus decreasing the effect of the amino group.

In the case of the diamino acids, the 2,4-diamino butyric acid caused only a small increase in color formation. However, the rate of increase in color formation with increase in chain length for this series was much greater than that for either of the two series of monoamino acids. Consequently the amount of color developed in all of the solutions tested was greater in the sugar solution containing lysine.

#### SUMMARY

In studying the effect of the positions of the amino groups in amino acids on the browning of glucose solutions it has been shown that the diamino acids caused the formation of the largest amount of additional color as compared to a glucose control. The greatest increase was caused by the 6-carbon diamino acid, lysine. The monoamino acids with the amino groups at the end of the chain caused varied changes in the color formation. Increased color formation occurred as the chain length of this series increased from 2 to 4 carbon atoms and then decreased as the chain length increased further to 5 and 6 carbon atoms. The monoamino acids with the amino group in the alpha position had almost no effect on the amount of color produced, regardless of chain length.

#### LITERATURE CITED

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1190

## BROWNING OF SUGAR SOLUTIONS

### II. Effect of the Position of Amino Group in the Acid Molecule in Dilute Glucose Solutions<sup>a</sup>

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In the previous paper of this series (2), it was shown that when glutamic acid, alanine, or lysine was present in a dilute glucose solution heated at 114° C., only the lysine, a basic amino acid, caused a significant increase in browning. This effect of lysine became more pronounced with increasing alkalinity of the sugar solution. Acidic and neutral amino acids did not show this accelerating effect, nor did arginine and histidine, other basic amino acids. Therefore, since lysine, in addition to being basic, is a simple diamino compound it was postulated that the number and the position of the amino groups in the amino acid molecule might determine its activity in the browning mechanism. The purpose of the present investigation was to determine the effect on the color developed in a glucose solution at pH 8 of several series of amino acids containing one or two amino groups. A first series contained the 4-, 5-, and 6-carbon homologs of lysine with amino groups attached to the alpha and terminal carbons. A second and a third series were composed respectively of the 3-, 4-, 5-, and 6-carbon examples of the alpha-amino and the terminal amino monocarboxylic acids.

#### EXPERIMENTAL

Weighed amounts of each amino acid, equivalent to 10 ml. of a 0.1 molar solution, were placed in separate beakers containing 2 ml. of the phosphate buffer solution, made according to Hawk, Oser, and Summerson (1). The pH was adjusted to 8 by titration (Beckman glass electrode pH meter<sup>d</sup>) with 0.1 N NaOH. To stabilize and keep the pH constant during the heating an additional 3 cc. of the buffer was added, and the solution quantitatively transferred to a 10-ml. volumetric flask containing 1 ml. of a stock glucose solution (0.1801 g. glucose per ml.) and the solution made to volume. The resulting solutions were 0.1 molar in respect to both the particular amino acid and to the glucose. Five-milliliter aliquots of each of these solutions were heated in sealed tubes at 114° C., at 10 p.s.i., as described previously (2). After cooling, the absorbance of the browned solutions was measured at 500 m $\mu$  in a Cary recording spectrophotometer. Data presented in Table 1 are averages of duplicate determinations.

#### RESULTS AND DISCUSSION

The amount of browning varied widely as influenced by both the type and chain length of the amino acid present. Graphic presentation of the influence of the 3 different series of amino acids tested is shown in Figure 1. In general,

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<sup>b</sup> From work done for a thesis to be submitted by H. G. Lento to the Graduate School of Georgetown University as partial fulfillment of the requirements for the degree of Doctor of Philosophy.

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<sup>d</sup> Mention of trade names does not imply endorsement by the U. S. Department of Agriculture over similar products not mentioned.

TABLE 1  
The effect of various amino acids on the color developed in a  
dilute glucose solution at pH 8

Amino acid	Formula	Number of carbon atoms	Terminal amino group	$\alpha$ -Amino group	Absorbance 500 m $\mu$
Glycine	$\text{NH}_2\text{CH}_2\text{COOH}$	2	+	+	1.56
DL- $\alpha$ -alanine	$\text{CH}_3\text{CHNH}_2\text{COOH}$	3	+	+	0.77
DL- $\beta$ -alanine	$\text{NH}_2\text{CH}_2\text{CH}_2\text{COOH}$	3	+	+	2.00
DL- $\alpha$ -aminobutyric acid	$\text{CH}_3\text{CH}_2\text{CHNH}_2\text{COOH}$	4	+	+	1.00
DL- $\gamma$ -aminobutyric acid	$\text{NH}_2\text{CH}_2(\text{CH}_2)_2\text{COOH}$	4	+	+	2.30
2,4-diaminobutyric acid.2 HCl	$\text{NH}_2\text{CH}_2\text{CH}_2\text{CHNH}_2\text{COOH}$	4	+	+	0.85
DL-norvaline	$\text{CH}_3(\text{CH}_2)_2\text{CHNH}_2\text{COOH}$	5	+	+	1.07
$\delta$ -aminovaleric acid.HCl	$\text{NH}_2\text{CH}_2(\text{CH}_2)_3\text{COOH}$	5	+	+	1.88
DL-ornithine.HCl	$\text{NH}_2\text{CH}_2(\text{CH}_2)_3\text{CHNH}_2\text{COOH}$	5	+	+	3.07
DL-norleucine	$\text{CH}_3(\text{CH}_2)_4\text{CHNH}_2\text{COOH}$	6	+	+	1.21
$\epsilon$ -aminocaproic acid	$\text{NH}_2\text{CH}_2(\text{CH}_2)_4\text{COOH}$	6	+	+	0.70
Lysine	$\text{NH}_2\text{CH}_2(\text{CH}_2)_3\text{CHNH}_2\text{COOH}$	6	+	+	4.10
Glucose	$\text{CH}_2\text{OH}(\text{CHOH})_4\text{CHO}$	6			0.64