A NOTE ON CARBOHYDRATES IN THE 11S GLOBULIN OF SOYBEAN SEEDS

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The presence of carbohydrate in soybean proteins has been confirmed in the 7S globulins (β-conglycinin (1–3) and γ-conglycinin (4)) and hemagglutinin (5). The carbohydrate components of a 7S globulin (β-conglycinin) and hemagglutinin were D-mannose and N-acetyl-D-glucosamine in the proportion of about 3 to 1. Further, the glycoprotein nature of two proteins was established by isolation of glycopeptides containing the two sugars (6–8).

However, Wolf *et al.* (9) have recognized that several soybean protein fractions gave a positive test for carbohydrates with phenol-sulfuric acid and that the sugar content of the 11S component was 0.2% as glucose. The 11S component prepared by us contained 0.78% carbohydrate as glucose in our experiment (1). Although there was a slight difference in the analytical values, the carbohydrate content was remarkably less than that of a 7S globulin (β-conglycinin). But, it has not yet been confirmed whether the small amount of carbohydrate in the 11S globulin is covalently bound to the protein or not. Here we report the nature of the carbohydrates in the 11S globulin.

In this experiment, the 11S globulin was prepared by the previous method (10) and further purified by preparative-scale polyacrylamide gel electrophoresis. The purified 11S contained 0.22% carbohydrate as β-D-mannose and 0.18% hexosamine as 2-amino-D-glucose (D-glucosamine). Even electrophoresis could not eliminate the carbohydrates in the protein.

When the 11S globulin was precipitated by heat denaturation, the precipitate gave a negative carbohydrate reaction with phenol-sulfuric acid (11) and Elson and Morgan's reaction (12). The results are shown in Table I. Similar results were also obtained from acid denaturation of the protein with trichloroacetic acid, as shown in Table II. Molisch's reaction was also negative in the protein precipitate. Accordingly, the carbohydrate and hexosamine in the 11S globulin were

TABLE I
Carbohydrate Content after Heat Denaturation of the 11S Globulin[a]

Treatment	Absorbance	
	Supernatant	Precipitate
100° C, 5 min		
Total carbohydrate	0.050	0.000
Hexosamine	0.057	0.000
100° C, 10 min		
Total carbohydrate	0.052	0.000
Hexosamine	0.060	0.000

[a]Heat denaturation was performed by using a mixture of 1.0% protein solution dissolved in water (0.5 ml), water (0.5 ml), and 1 *M* sodium phosphate buffer, pH 6 (0.5 ml). Absorbance was measured at 490 nm for total carbohydrate and at 512 nm for hexosamine. All the protein was recovered from the precipitated fractions and not detected in the supernatant fractions.

TABLE II
Carbohydrate Content after Acid Denaturation of the 11S Globulin[a]

	Absorbance	
	Supernatant	Precipitate
Molisch's test	++	–
Total carbohydrate	0.030	0.000
Hexosamine	0.027	0.000

[a]Acid denaturation was carried out by using a mixture of 1.0% protein solution dissolved in water (0.5 ml), water (0.5 ml), and 20% trichloroacetic acid (1.0 ml). Absorbance was measured at 490 nm for total carbohydrate and at 512 nm for hexosamine. All the protein was recovered from the precipitated fractions and not detected in the supernatant fractions.

concluded to be strongly absorbed to protein, but not bound to protein covalently.

The carbohydrates in the 11S globulin consisted of glucose, xylose, arabinose, mannose, galactose, and several unknown polysaccharides as determined by sugar analyzer using a column of anion-exchange resin equilibrated with borate buffer.

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