hydrolysis would be expected to yield a large amount of orthophosphate and small amounts of imidodiphosphate and imidotriphosphate. Polymers of the type shown in Fig. 1 would be highly metachromatic due to the presence of few secondary phosphate ionizations, which inhibit metachromasy (*8*). They might also exhibit a completely different affinity for RNA than a true polyphosphate, thus resembling the naturally occurring "polyphosphate."

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Hemagglutination by Fava Bean Extract Inhibited by Simple Sugars

Abstract. *Hemagglutination by extract of fava bean was inhibited by 5-percent* d-*glucose,* d-*fructose, or maltose, but not by 5-percent* d-*galactose or lactose. Failure to inhibit seems to reflect the presence of a hydroxyl group at the carbon No. 4 position. Hemagglutination was enhanced by dextran of high molecular weight, but not by dextran of low molecular weight. The finding supports the hypothesis that large molecular size explains the enhancement by gum acacia of hemagglutination by fava bean.*

Agglutination of normal human erythrocytes by substances other than blood group–specific isoantibodies is well known (*1*). Inhibition of this hemagglutination by various complex sugars has been reported (*2*). The effects of simple sugars on hemagglutination by saline extract of fava bean are the subject of this report.

Hemagglutination by fava bean was inhibited by 5-percent *d*-glucose, *d*-fructose, or maltose, but not by 5-percent *d*-galactose or lactose (Table 1). The determining factor appears to reside in the configuration of the No. 4 carbon atom: presence of a hydroxyl group prevents inhibition of hemagglutination. Inhibition by normal human serum of hemagglutination by fava bean has been reported (*3*); the concentration of glucose in normal human serum, 0.6 to 1.05 mg ml^{-1}, cannot explain such inhibition. Inhibition was complete with 0.6-, partial with 0.55-, and none with 0.4-percent *d*-glucose. Agglutination disappeared if normal serum or 5-percent *d*-glucose was added after agglutination occurred. The inhibition in vitro by normal serum of hemagglutination by fava bean did not occur in the serum of a child with favism (*4*). These observations suggest that induced hyperglycemia or transfusion of plasma may have a therapeutic effect in human favism.

Hemagglutination by fava bean is enhanced by gum acacia (*3*). Similar studies with dextrans of varying molecular weights (*5*) show augmented titers (1:640) with dextran of high molecular weight (75,000), but not with dextran of lower molecular weight (41,500). The hypothesis that enhancement of agglutination by gum acacia is due to the gum's molecular size is strengthened. Agglutination by dextrans of high molecular weight also is inhibited by 5-percent *d*-glucose or normal human serum.

Table 1. Effects of simple sugars on hemagglutination by saline extract of fava bean. Degree of agglutination indicated by number of plus signs.

Sugar concentration (%)		Extract titer				
		1:10	1:20	1:40	1:80	1:160
None		+ + + +	+ + + +	+ + +	+ +	0
Lactose	5	+ + + +	+ + + +	+ + +	+ +	0
d-Galactose	5	+ + + +	+ + + +	+ + +	+ +	0
Maltose	5	0	0	0	0	0
d-Fructose	5	0	0	0	0	0
d-Glucose	5	0	0	0	0	0
d-Glucose	0.6	0	0	0	0	0
d-Glucose	.55	+ +	+ +	0	0	0
d-Glucose	.4	+ + +	+ + +	+ +	+ +	0

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Encephalitogenic Activity of Bovine Basic Proteins

Abstract. *Two basic proteins isolated from bovine white matter in connection with a study of the protein-bound phosphoinositides of central nervous system tissue have been tested for encephalitogenic activity. The biological activity of these proteins, which is equivalent to that of basic encephalitogenic proteins isolated in other laboratories, suggested that they are identical.*

The basic protein fraction of central nervous system (CNS) myelin has been investigated intensively because it can induce experimental allergic encephalomyelitis (EAE) in various animals (*1*). Although the encephalitogen is part of the myelin proteolipid, it is not found in the chloroform-methanol (2:1 mixture) extract of the whole tissue. Its absence from the chloroform-methanol extract of whole tissue is presumably caused by some interaction between the encephalitogenic proteolipid and other constituents of the tissue

Table 1. Encephalitogenic activity of bovine basic proteins.

Amount tested (μg)	Day of onset*	Disease index†
	S-7 total fraction‡	
20	13	9.1
5	18	6.0
1		2.6
	S-7 major protein‡	
20	12	8.4
5	15	7.8
1	20	4.4
	Component IV‡	
20	14	8.9
5	18	7.4
1		2.2

* Day on which the third of the group of five guinea pigs showed definite clinical signs of EAE. † Combined clinico-pathologic index of severity of EAE (average of five animals), the maximum being ten (*6*). ‡ Prepared by Martenson and LeBaron (*5*).

homogenate. This interaction may result in cleavage of the protein-lipid bonds (*2*).

The major portion of the encephalitogenic activity of whole CNS tissue is obtained by acid extraction of tissue which has been previously extracted with chloroform-methanol. These acid extracts may be further purified, yielding highly encephalitogenic as well as nonencephalitogenic basic proteins (*3, 4*). In addition, small amounts of acidic and neutral protein contaminants have been identified immunochemically. These are nonencephalitogenic.

Martenson and LeBaron (*5*) have reported purification of a group of basic proteins isolated in the course of an investigation of protein-bound phosphoinositides in the chloroform-methanol insoluble residue of CNS white matter. Chemical similarities between two of these basic proteins, which they call "major S-7 component" and "component IV," and the encephalitogenic myelin basic protein suggested that these preparations were identical with the encephalitogen. Consequently arrangements were made to test the unknown preparations under the same conditions used for bioassay of the previously reported encephalitogenic proteins (*6*). Equally susceptible inbred strain 13 guinea pigs were used instead of the random-bred NIH animals. The fractions were prepared from bovine white matter that had been extracted with chloroform and methanol. They were S-7 (that part of the acid extract soluble at *p*H 7), the major protein component of S-7, and component IV which is a subfraction of the material insoluble at *p*H 7. When 0.02 mg of each fraction was tested, maximum EAE resulted in all of the guinea pigs injected. Each fraction was again tested at 0.001 and 0.005 mg (Table 1). All three are encephalitogenic with 50 percent effective dose (ED_{50}) of about 2 μg. The major component of fraction S-7 probably accounts for all of its encephalitogenic activity. Component IV, possessing approximately the same activity as the major S-7 component, appears to be closely related to the latter even though the two were originally obtained from two fractions of the acid extract with different solubilities.

The major protein of the fraction soluble at *p*H 7, as well as component IV of the fraction insoluble at *p*H 7, can thus be added to the increasing list of encephalitogenic proteins reported (*2, 4, 7*). It is unlikely that all of these preparations are identical. Quite probably they are closely related, at least as indicated by their chemical and biological properties. Gel filtration on Biogel P-10 of component IV and of the major S-7 component indicates that they are similar in size (35,000 to 40,000 molecular weight) and that they fall into the category of "large" encephalitogens characteristically obtained from CNS tissue which had been previously extracted with chloroform-methanol in contrast to "small" encephalitogens (about 10,000 molecular weight) frequently obtained from tissue that had previously been extracted with acetone.

These findings are all the more interesting in that the same purified protein was obtained in two independent studies. In one, bioassay was the criterion of progressive purification. In the other, chemical analysis alone was the criterion. Furthermore, in both cases, we have been impressed by the relative abundance of this protein in CNS tissue and its remarkable stability in the presence of chloroform-methanol and of relatively strong acid.

The bioassays strengthen the opinion of Martenson and LeBaron (*5*) that the major component of the S-7 fraction is the same protein as component IV. Encephalitogenic activity in more than one fraction has also been observed during isolation of the encephalitogen (*8*). A possible explanation is that there is an equilibrium between free and combined protein, the bound forms being less soluble than free protein at neutral *p*H. If so, solution of the insoluble complex in dilute acetic acid and chromatography on carboxymethyl cellulose appears to cause dissociation of the complex. Free protein is then isolated as component IV.

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Intracellular Localization of Growth Hormones in Plants

Abstract. *Autoradiographic studies of* Allium cernuum *and* Vicia faba *root-tip cells treated with indoleacetic acid-methyl-C^{14} or 2,4-dichlorophenoxyacetic acid-carboxyl-C^{14} revealed nuclear and cytoplasmic labeling of the cells. The cytoplasmic labeling decreased with time after the removal of the labeled auxin, but nuclear and chromosomal labeling was retained for at least 120 hours.*

There is evidence that auxin-induced growth in various plant tissues is accompanied by changes in the cell wall and cytoplasm, and in nucleic acid metabolism (*1*). However, it is unknown which of these is the direct effect of auxin action. It has been shown by autoradiography that the plant growth inhibitor maleic hydrazide-C^{14} was localized in the nuclei of root-tip cells for a short time interval after treatment. Subsequent time-course experiments indicated movement of the inhib-