The Chemical Characterization of Favin, a Lectin Isolated from *Vicia faba*  

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We have determined the subunit structure of the glucose- and mannose-binding lectin favin, from *Vicia faba*. The molecule is composed of two nonidentical polypeptide chains held together by noncovalent interactions. We have determined the complete amino acid sequence of the smaller \( \alpha \) chain (\( M_r = 5,571 \)) and shown that it is homologous to the \( \alpha \) chain of the lectins from lentil and pea and to residues 72 to 120 of concanavalin A (Con A). The larger \( \beta \) chain (\( M_r = 20,000 \)) contains carbohydrate and is homologous to the \( \beta \) chain of lentil, pea, soybean, peanut, and red kidney bean lectins and is homologous to a portion of the Con A molecule beginning at residue 122. Favin also contains a minor component, \( \beta' \) (\( M_r = 18,700 \)), that closely resembles the \( \beta \) chain but lacks carbohydrate and may, on the basis of amino acid composition, be derived from the \( \beta' \) chain of lentil lectin. Favin binds glucose and mannose and is homologous to Con A. The larger \( \beta \) chain of favin is homologous to the \( \beta \) chain of Con A, which is isolated from *Canavalia ensiformis*, con also bind hydrophobic moieties (4, 5), including the plant auxin 3-indoleacetic acid (6). Extensive studies have been directed toward elucidating the molecular basis for these diverse properties. For example, the amino acid sequence (7, 8) and three-dimensional structure (4, 8-10) of Con A have been determined and the carbohydrate (11, 12) and metal binding (4, 8, 10) sites defined. Other studies have shown extensive sequence homologies between Con A and lentil lectin (13, 14) and between various lectins from other leguminous plants (13). Neither the complete amino acid sequence nor the three-dimensional structure of any of these homologous lectins has been described.

We have begun a study of the primary and three-dimensional structure of the lectin favin from fava beans (*Vicia faba*) (15). This lectin resembles Con A and the pea and lentil lectins in that it binds glucose and mannose. Initial studies of this protein by different investigators have given disparate results. The report from this laboratory described the purification of favin and demonstrated that it crystallizes as a molecular species of 53,000 daltons in a form suitable for x-ray crystallography (15). In subsequent studies from other laboratories the same native molecular weight for favin was reported, but different isolation procedures were used and a different organization of the peptide chains was suggested (16, 17). The complete amino acid sequence of the smaller \( \alpha \) chain of favin and have partially sequenced the larger \( \beta \) chain. Our studies indicate that favin closely resembles the lentil lectin and is homologous to Con A.

**EXPERIMENTAL PROCEDURES**

**Materials**—Dried fava beans, with or without testae, were purchased from local grocers. *Sephadex* and CH-Sepharose were purchased from Pharmacia. Bio-Gel polyacrylamide gel filtration media and gel electrophoresis supplies were from Bio-Rad Laboratories. Diethylaminoethyl cellulose (DE-52) was from Whatman. 3-O-Methylglucosamine was synthesized by the procedure of Roth et al. (17). The protease inhibitor Tranexyl was purchased from Mobay Chemical Corp. (New York City, N. Y.). All other chemicals were of reagent grade and were used as supplied.

**Methods**—Favin was isolated from a single lot of dried fava beans according to three procedures: 1) affinity chromatography of a dialyzed bean extract on *Sephadex* G-75 (15); 2) 80% ammonium sulfate precipitation of bean extract followed by affinity chromatography on *Sephadex* G-150 (16); and 3) extraction of delipidated meal followed by 65% ammonium sulfate precipitation and subsequent affinity chromatography on 3-O-methylglucosamine-derivatized *Sepharose* (17). Favin was also isolated from four different unrelated lots of dried fava beans according to three procedures: 1) affinity chromatography of a dialyzed bean extract on *Sephadex* G-75 (15); 2) 80% ammonium sulfate precipitation of bean extract followed by affinity chromatography on *Sephadex* G-150 (16); and 3) extraction of delipidated meal followed by 65% ammonium sulfate precipitation and subsequent affinity chromatography on 3-O-methylglucosamine-derivatized *Sepharose* (17).
began by the first method (15). In all cases, the beans were initially ground to a coarse meal in a Waring blender. Favin prepared by affinity chromatography on Sephadex was eluted with glucose, di-
alyzed against water, and stored as lyopholized powder. The material prepared by elution from 3-O-methylglucosamine-Sepharose with a-methyl glucoside was stored as a suspension in 1 m NaCl.

SDS-polyacrylamide gel electrophoresis was performed as described by Laemmli (19) using a final concentration of 18% or 20% acrylamide. Proteins used as molecular weight standards for SDS-polyacrylamide gel electrophoresis were: Con A isolated from jack bean meal (Schwarz/Mann) as previously described (20), egg white lysozyme (Worthington), sperm whale myoglobin (Miles), and bovine pancreatic trypsin inhibitor (Worthington). Periodate-Schiff reagent staining of SDS-polyacrylamide gels was performed as described by Glossman and Neville (21).

The phenol-sulfuric acid assay was used for the determination of total hexose content (22), referred to mannose as a standard.

Protein and peptide hydrolysates were performed in vacuo with constant boiling HCl for 24, 48, and 72 h at 110°C. Amino acid analysis was carried out on a Beckman 121M analyzer equipped with a model AA automatic integrator. The tryptophan/tyrosine ratio was estimated spectrophotometrically (23).

Protein sequence determinations were carried out as described in the miniprint supplement.

Sedimentation equilibrium ultracentrifugation was performed by the short column method of Yphantis (24) using a Spinco model E ultracentrifuge operated at 20,000 or 30,000 rpm.

The β and β' components of favin (see below) were separated by preparative SDS-polyacrylamide gel electrophoresis. Included in the gel were fluorescamine-labeled Con A and lysozyme and 3H-labeled favin prepared with chloramine-T and Na125I (25). After electrophoresis of fluorescamine-labeled lysozyme to the bottom of an 18% acrylamide slab gel (0.8 × 11 × 18 cm), the protein bands were visualized by staining in 0.1% Coomassie brilliant blue in 25% methanol, 7% acetic acid for 10 min. The β and β' bands were excised and soaked in 50 mM Tris-HCl (pH 8.6), 0.1% SDS for 1/2 h at room temperature. The gels were then minced and electroeluted into the same buffer. The recovery was 95% for β and 88% for β' as determined by monitoring the radioactivity. The material was dialyzed against 0.05 M NH4HCO3 and against water, and the solution lyopholized. The resulting mixture was extracted with anhydrous methanol. The residue was dried and used for sequence determination by NH2-terminal Edman degradation and for amino acid analysis.

RESULTS

Fava lectin isolated by affinity chromatography on Sephadex G-75 gave three components on SDS-polyacrylamide gel electrophoresis (Fig. 1). This pattern closely resembles that obtained by Wang et al. (15) with a major band of apparent molecular weight 20,000 and a minor 18,700-dalton com-
ponent; these molecular weights are interpolated from the mobilities of Con A, myoglobin, and lysozyme. Attempts to apply this calibration curve to the other major band (showing a bow shape near the bottom of this particular gel) yielded an apparent molecular weight of 8,900; this material most likely corresponds to the 9,000-dalton component reported by Wang et al. (15). However, this apparent 8,900-dalton component shows a faster mobility than bovine pancreatic trypsin inhibitor, a protein of molecular weight 6,512, which also gave an incorrect higher apparent molecular weight on the same calibration curve. This finding is consistent with the anomalous mobilities of smaller proteins on SDS-polyacrylamide gel electrophoresis (26).

Sedimentation equilibrium in 6 M guanidine HCl, 0.1 M Tris-HCl (pH 8.6) yielded a molecular weight of 5,473 ± 107 for the smaller molecular weight component of favin. A value of 5,571 was calculated from the amino acid sequence given below. In keeping with the nomenclature used for the subunits of other legume lectins (13), and based on the studies described below, we have designated the 20,000-dalton component the β chain of favin, the 5,571-dalton component the α chain, and the minor 18,700-dalton component the β' chain.

To test for the presence of other possible forms of favin or its subunits, the lectin was isolated by other procedures (16,17), from four apparently unrelated lots of fava beans (with or

These gels were selected primarily to show any variation in the high molecular weight material. On 20% gels, the lower molecular weight material did not migrate as close to the bottom of the gel and gave a normal horizontal band, with approximately the same apparent molecular weight as was obtained on 18% gels.
without testae), and in the presence of the protease inhibitors Trasylol and phenylmethanesulfonyl fluoride. The results of these studies are shown in Fig. 1. In all cases only bands corresponding to the $\beta$, $\beta'$ and $\alpha$ chains were seen by SDS-polyacrylamide gel electrophoresis; in addition, the amino acid composition of favin isolated by all three procedures was very similar (data not shown).

Separation of Favin Chains—Fractionation of favin on Sephadex G-100 in the presence of 6 M guanidine HCl yielded three fractions (Fig. 2), and the material in each fraction was analyzed by SDS-polyacrylamide gel electrophoresis. Material in Fraction A consisted of small and variable amounts of high molecular weight components. Even SDS-polyacrylamide gel electrophoretically purified material showed small amounts of high molecular weight components; presumably these are undissociated aggregates of the $\beta$ and $\beta'$ chains. Fraction B contained predominantly the $\beta$ and $\beta'$ chains and could be further purified from the $\alpha$ chain by rechromatography on the same column. Fraction C contained only the $\alpha$ chain. After salt was removed by dialysis through cellulose tubing with a molecular weight cutoff of 4,700, the fractions were lyophilized. The amino acid compositions of these fractions are compared with that of whole favin in Table I. The sum of the compositions of the $\alpha$ and $\beta$ plus $\beta'$ chains agrees very closely with the composition of the isolated lectin. The average weight ratio of isolated material from Fraction B to that from Fraction C was 4.0 to 1. Assuming the molecular weights to be 50,000 (15-17), suggesting that the native lectin consists of two $\alpha$ chains and two $\beta$ or $\beta'$ chains.

The $\beta'$ component appeared to represent about 10% of the total B fraction (Fig. 2) as estimated by densitometer traces of Coomassie brilliant blue-stained SDS gels. The $\beta$ and $\beta'$ components were not readily separated by gel filtration or ion exchange chromatography in 6 M urea. These components were separated by preparative SDS-polyacrylamide gel electrophoresis as described under "Experimental Procedures." Analytical gel electrophoresis on the separated components showed that they were not cross-contaminated (Fig. 3).

Carbohydrate Content of the $\alpha$ and $\beta$ Chains—The carbo-

![Fig. 2. Gel filtration of favin on a column (2.6 × 93.5 cm) of Sephadex G-100 equilibrated with 6 M guanidine HCl, 0.1 M Tris-HCl (pH 8.6). Each tube contained 3 ml of effluent. A, B, and C designate fractions that were pooled for further characterization.](image-url)
hydrate contents of material in Fractions B and C (Fig. 2) were analyzed on SDS-gels by periodate-Schiff reagent staining (21) and in solution by the phenol-sulfuric acid assay for neutral sugars. PAS staining of rigorously washed gels indicated that only the $\beta$ component was positive for carbohydrate. The phenol-sulfuric acid assays on the same materials were consistent with this result except that $\beta$ and $\beta'$ were not distinguished in this assay. Material in Fraction B contained 3% carbohydrate and material in Fraction C contained 1% carbohydrate. For the $\alpha$ chain, 1% carbohydrate is less than 1 mol of sugar/mol of protein, and this value most likely represents protein background. The estimate of 3% carbohydrate for the $\beta$ chain is consistent with the data obtained by A. Allen et al. (17), who isolated a glycopeptide from the total favin molecule that contained an average of 3 residues of sugar, and also agrees with the value of 3% carbohydrate we found for whole favin from which noncovalently bound sugar was removed by gel filtration on a column of Bio-Gel P-2.

Amino Acid Sequence of the $\alpha$ Chain of Favin—The strategy for the determination of the amino acid sequence of the $\alpha$ chain (Fig. 4) included automated Edman degradation of the polypeptide from the NH$_2$ terminus, isolation and sequence analysis of a tryptic peptide obtained after treatment of the $\alpha$ chain with succinic anhydride, and isolation and characterization of three peptides obtained by tryptic digestion of the unmodified $\alpha$ chain. The details of these determinations are given in the miniprint supplement. Carboxypeptidases A, B, and Y did not release any amino acids from the COOH terminus of the $\alpha$ chain. The sequence (Fig. 4) is in excellent agreement with the amino acid composition of the $\alpha$ chain (Table I).

Characterization and NH$_2$-terminal Sequence of the $\beta$ and $\beta'$ Chains—The amino acid compositions of material from Fraction B (Fig. 2) and of the $\beta$ and $\beta'$ chains purified by
**DISCUSSION**

Our studies demonstrate that the lectin favin is composed of two polypeptide chains, an α chain (M_r = 5,600) that contains 51 amino acid residues and a β chain (M_r = 20,000) that contains about 180 amino acid residues, as well as some carbohydrate. The observed stoichiometry of these chains suggests that the native favin molecule (M_r = 50,000) consists of two α chains and two β chains held together by noncovalent interactions. As normally isolated, the lectin contains a small amount of a third chain, β'. The β' chain (M_r = 18,700) closely resembles the β chain in amino acid composition and has the same NH2-terminal amino acid sequence, but probably lacks most or all of the carbohydrate and, on the basis of the apparent molecular weight, may lack some part of the COOH-terminal region of the polypeptide chain. Previous studies on favin have suggested different molecular properties of the lectin. While Wang et al. (15) reported that favin isolated by affinity chromatography on Sephadex G-75 consists of subunits with molecular weights similar to those found in this study, Allen et al. (16) determined that favin isolated by affinity chromatography on Sephadex G-150 is composed of two subunits of apparent molecular weights 17,300 and 14,300. Allen et al. (17), using favin isolated by affinity chromatography on 3-O-methylglucosamine-Sepharose, characterized only one subunit of molecular weight 22,500; they did not exclude the possibility that another smaller subunit might be present. We found that samples of favin isolated from the same lot of fava beans by the procedures described in each of these studies were identical with one another by SDS-polyacrylamide gel electrophoresis (Fig. 1) and amino acid composition. Thus it would seem that the method of isolation is not the cause of these reported variations. We have also screened several apparently unrelated lots of dried beans and have found no variations in the isolated lectins (Fig. 1). On the basis of apparent molecular weight, the homogeneous 22,500 ± 1,500-dalton material reported by Allen et al. (17) may correspond to the β chain. They report that this chain has an NH2-terminal leucine, whereas we found threonine as the NH2-terminal residue of the β and β' chains. However, leucine is the NH2-terminal residue of the much smaller α chain, suggesting the possibility that Allen et al. (17) may have isolated a higher molecular weight material composed of α and β or β' joined as an uncleaved precursor molecule. To test the possibility of extensive proteolytic degradation during our isolation procedures, we isolated favin in the presence of the protease inhibitors Trasylol and phenylmethylsulfonyl fluoride. The same 20,000-, 18,700-, and 5,600-dalton materials were obtained, and no significant amounts of higher molecular weight materials were detected (Fig. 1). Inasmuch as these inhibitors do not block all proteases, we cannot exclude the possibility that different forms of the molecule exist prior to extraction. In addition, larger forms of the lectin may well exist during different stages of seed development and growth. Our data, however suggest that the differences in favin structure reported previously are most likely due to differences in the methods used to characterize the molecule and that favin as described here is a well defined and reasonably homogeneous lectin.

The results presented here also confirm the previous observations (15-17) that favin is a glycoprotein and suggest that the carbohydrate is located on the β chain of the molecule. However, we have not found either any covalently bound carbohydrate or the (alanine, asparagine) glycopeptide reported by Allen et al. (17) within the first 35 residues of the β chain.

The subunit structure of favin closely resembles that of other glucose- and mannose-binding lectins such as those from pea (30,31) and lentil (13,32). This similarity is more apparent on comparison of the sequences of the α chains and the partial sequences of the β chains of these molecules. In Fig. 5, the sequences of all these lectins are aligned with that of Con A, which contains a single polypeptide chain of 237 amino acids. As with the lectins from the lentil and pea, the α chain of favin is highly homologous to residues 72 to 120 of Con A, and the β chain resembles a region beginning at residue 122 of Con A. As shown previously (13), the β chains of the N-acetylgalactosamine-binding soybean and red kidney bean lectins and the galactose-binding peanut lectin are also similar to Con A beginning at residue 123. By inspection, the sequences of all the various lectins shown (Fig. 5) are highly conserved. Pavin appears to be more closely related to the lentil and pea lectins than to Con A or the other lectins.

The sequence homologies suggest that all of these lectins
may have similar three-dimensional structures. Residues 72 to 120 of Con A, which are homologous to the α chain of favin, include portions of each of the two large β structures, as well as a loop of peptide associated with the specific carbohydrate binding site (4, 10–12). Residues 122 to 141 of Con A, which are homologous to the NH₄-terminal 20 residues of the β chain of favin, comprise a large part of the region of contact between the two monomers making up a Con A dimer. This similarity suggests that the 50,000-dalton favin molecule, made up of two α and two β subunits may be stabilized by β structure-like hydrogen bonds between the first 20 residues of one β chain and similar residues in the second β chain, an assembly similar to that formed by residues 120 to 139 of Con A (9).

It is particularly noteworthy that the region of the chain (Con A residues 120 to 123) where the α chains of the various lectins end and the β chains begin is the site at which a natural cleavage of Con A is known to occur (33). These results suggest that all of these lectins may be initially synthesized as larger precursor molecules and that the observed forms derive from post-translational modification. Immunological evidence for a possible precursor molecule has been obtained for the Dolichos biflorus lectin (34), but no direct demonstration of such a precursor has been obtained.

The fact that regions homologous to residues 1 to 71 of Con A have not been found in favin or other homologous lectins raises a question about the carbohydrate binding sites of these proteins. X-ray crystallographic data (11, 12) suggest residues 1 to 71 of Con A contribute a significant number, but not all, of those atoms involved in sugar binding by Con A. The other lectins may have a much different binding site. Alternatively, since the estimate of the molecular weight of the β chain of favin implies that it, as with the β chain of other lectins, extends beyond the COOH terminus of Con A by 40 to 50 residues, this region may contribute residues in favin that form a three-dimensional binding site similar to that of Con A. (See Note Added in Proof, below.) Binding sites with similar three-dimensional structure made up from different portions of the polypeptide chain in proteins of homologous function are also found in the serine proteases (35).

Although lectins are used as cell surface probes on mammalian cells, the functions of these proteins in plants are unknown. Recently some lectins have been localized in the matrices of seed protein bodies (36, 37). In preliminary studies we have found both favin and Con A associated with the protein body fraction of fava beans and jack beans, respectively, but have not obtained definitive data to show that the lectins are, in fact, incorporated in the protein bodies. These organelles are known to contain the storage proteins vicilin and legumin, as well as many hydrolytic enzymes (38), and to be disrupted under the conditions used for lectin isolation. It seems unlikely that favin and Con A are merely storage proteins, and it is possible that the lectins are associated with some hydrolytic activity either as hydrolases or as processing agents for other hydrolytic products. For example, the lectin from the mung bean has been reported to have α-galactosidase activity (39). We have tested favin and Con A for glycosidase activity toward simple mannose-, glucose-, and galactose-containing saccharides. Although the extracts of fava beans and jack beans have such activity (40),4 no such activity has been shown to be associated with these lectins.

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Note Added in Proof—Completion of the amino acid sequence of the β chain (Cunningham, B. A., Hemperly, J. J., Hopp, T. H., and Edelman, G. M. (1979) Proc. Natl. Acad. Sci. U. S. A. 75, in press) indicates that residues homologous to residues 1 to 69 of Con A are located at the COOH terminus of the favin β chain and that the carbohydrate is attached to residue 171 of this chain.

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Chemical Characterization of Favin

SYNOPSIS

The material was prepared in the following manner: Favin 6809 was decolored by heating at 100°C for 30 min to destroy color, then extracted with chloroform. The extract was dried with anhydrous sodium sulfate and concentrated to a small volume. The solid residue was then subjected to column chromatography on silica gel. The fractions were collected and analyzed for their chemical content. The major components were identified as fatty acids, amino acids, and carbohydrates.

MATERIALS AND METHODS

Favin 6809 was extracted with chloroform. The crude extract was fractionated by column chromatography on silica gel. The fractions were analyzed by gas chromatography and thin-layer chromatography.

RESULTS

The major components of Favin 6809 were fatty acids, amino acids, and carbohydrates. The fatty acids were mainly saturated and unsaturated long-chain fatty acids. The amino acids were mainly glycine, alanine, and serine. The carbohydrates were mainly sucrose and glucose.

DISCUSSION

The chemical characterization of Favin 6809 provides important information about its functional components. The presence of fatty acids suggests that Favin 6809 may have applications in food and agricultural products. The presence of amino acids and carbohydrates indicates that Favin 6809 may also have applications in pharmaceuticals and cosmetics.

TABLE I

| Cycle Number | Residue | Field of View (mm²) |
|--------------|---------|-------------------|
| 1            | Thy      | 138.8              |
| 2            | Thy      | 146.0              |
| 3            | Thy      | 186.3              |
| 4            | Thy      | 288.9 (F 9.3)     |
| 5            | Ala      | 168.7              |
| 6            | Ala      | 266.3 (F 29.4)    |
| 7            | Ala      | 237.1              |
| 8            | Ala      | 196.4              |
| 9            | Thr      | 158.0              |
| 10           | Thr      | 152.3              |
| 11           | Thr      | 17.9 (F 14.4)     |
| 12           | Ala      | 64.4               |
| 13           | Ala      | 104.3              |
| 14           | Ala      | 50.0               |
| 15           | Ala      | 31.5 (G 1.3)      |

*The values in parentheses indicate the field of view in square millimeters. The values were determined by gas chromatography.

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*The reference cited is not in the manuscript.*
TABLE III

| Amino Acid | T1 | T2 |
|------------|----|----|
| Asp        | 2.6| 1.2|
| Thr        | 2.1| 1.9|
| Ser        | 1.5| 1.3|
| Gly        | 1.2| 1.2|
| Ala        | 2.5| 2.2|
| Val        | 0.7| 0.4|
| Leu        | 2.6| 2.7|
| Pro        | 0.9| 0.9|
| Ile        | 0.3| 0.3|
| Thr        | 2.5| 2.2|
| Ser        | 0.7| 0.4|
| Leu        | 2.7| 3.3|
| Pro        | 0.9| 0.9|
| Ile        | 0.3| 0.3|

Total residues: 32 22 24 9

*Values are based on micromoles of isolated peptide compared with micromoles of calcium digested with tryptic-EDTA.

TABLE IV

| Automatic Sequence Analysis of n-Chain Fragment T1 |
|--------------------------------------------------|
| **Cycle Number** | **Residue** | **FTD Amino Acid** |
|------------------|-------------|--------------------|
| 1                | Lys         | 11.1               |
| 2                | Thr         | 2.2 (1.4)          |
| 3                | Gly         | 2.4                |
| 4                | Thr         | 1.4 (1.6)          |
| 5                | Val         | 2.2                |
| 6                | Pro         | 0.9 (0.3)          |
| 7                | Leu         | -                  |

*Values are based on micromoles of isolated peptide compared with micromoles of calcium digested with tryptic-EDTA.

TABLE V

| Automatic Sequence Analysis of n-Chain Fragment T2 |
|--------------------------------------------------|
| **Cycle Number** | **Residue** | **FTD Amino Acid** |
|------------------|-------------|--------------------|
| 1                | Ile         | 31.2               |
| 2                | Gly         | 29.2               |
| 3                | Thr         | 30.1               |
| 4                | Ser         | 9.1                |
| 5                | Ala         | 33.7               |
| 6                | Thr         | 18.5               |
| 7                | Ile         | 10.9               |
| 8                | Ala         | 14.8               |
| 9                | Ser         | 4.4                |
| 10               | Glu         | 18.3               |
| 11               | Thr         | 11.4               |
| 12               | Ala         | 11.5               |
| 13               | Lys         | 4.8 (4.2)          |
| 14               | His         | 5.0                |
| 15               | Val         | 1.9                |
| 16               | Leu         | 3.8                |
| 17               | Ser         | 0.6 (0.4)          |
| 18               | Thr         | 2.0                |
| 19               | Pro         | 0.9 (0.3)          |
| 20               | Lys         | 3.0                |
| 21               | Leu         | 2.1                |
| 22               | Val         | 1.0 (0.1)          |
| 23               | Glu         | 2.0                |
| 24               | Lys         | 0.9 (0.3)          |
| 25               | Thr         | 0.6 (0.1)          |
| 26               | Glu         | 0.9                |
| 27               | Pro         | 0.4 (0.1)          |

*Values are based on micromoles of isolated peptide compared with micromoles of calcium digested with tryptic-EDTA.

TABLE VI

| Automatic Sequence Analysis of n-Chain Fragment T3 |
|--------------------------------------------------|
| **Cycle Number** | **Residue** | **FTD Amino Acid** |
|------------------|-------------|--------------------|
| 1                | Thr         | 13.4               |
| 2                | Arg         | 13.8               |
| 3                | Gln         | 13.1               |
| 4                | Thr         | 14.1               |
| 5                | Ser         | 8.4                |
| 6                | Phe         | 10.8               |
| 7                | Lys         | 11.8               |
| 8                | Pro         | 11.5               |
| 9                | Lys         | 12.6               |
| 10               | Lys         | 13.9               |
| 11               | Pro         | 11.2               |
| 12               | Lys         | 11.2               |
| 13               | Pro         | 11.2               |

*Values are based on micromoles of isolated peptide compared with micromoles of calcium digested with tryptic-EDTA.

Chemical Characterization of Favin

![Graph of chemical data](image)

**Figure 4:**

Nel filtration of a tryptic digest of monobiotylyl n-chain of fava on a column (0.8 x 100 cm) of Sephadex G-50 equilibrated with 0.15M HCl (pH containing 0.01 M-mercaptoethanol) each tube contained approximately 2 ml of effluent. Neutral solution was further purified by repeated gel filtration on the same column.

![Graph of chemical data](image)

**Figure 5:**

Nel filtration of a tryptic digest of n-chain of fava on a column (1.7 x 100 cm) of Sephadex G-50 equilibrated with 0.15M HCl containing 0.01 M-mercaptoethanol. Each tube contained approximately 2 ml of effluent. Neutral solution was further purified as described in the text.

![Graph of chemical data](image)

**Figure 6:**

Ion exchange chromatography of neutral from peak A (Figure 4) on a column (1 x 100 cm) of NAD-agarose in 0.2 M Tris HCl (pH 8.5). The neutral solution passed through the column at a flow rate of 0.5 M HCl was used. Each tube contained approximately 2 ml of effluent. The letters A, B, and C denote material pooled for analysis as described in the text. FT5, T11, and T12 detected from each fraction were noted.
The chemical characterization of favin, a lectin isolated from Vicia faba.
J J Hemperly, T P Hopp, J W Becker and B A Cunningham

J. Biol. Chem. 1979, 254:6803-6810.

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