Polysaccharide Composition of a Gel Fraction Derived from Fenugreek and Its Effect on Starch Digestion and Bile Acid Absorption in Rats

Zecharia Madar*,‡ and Ilan Shomer§

Department of Biochemistry and Human Nutrition, Faculty of Agriculture, Hebrew University of Jerusalem, Rehovot 76100, Israel, and Department of Food Science, Agricultural Research Organization, The Volcani Center, P.O. Box 6, Bet Dagan 50250, Israel

The soluble gel fraction of fenugreek seeds constituted the major portion of the seed coat (including the endosperm) polysaccharides, most of which consisted of galactomannan with mannose:galactose ratio of 1.5:1. The relatively small amount of the insoluble cell wall included mainly cellulose (as glucose) and pectin (as galacturoinic acid). In vivo nutrition experiments in rats and in vitro studies using inverted gut showed that the gel fraction decreased both digestion and absorption of starch and uptake of bile acid (taurocholate and deoxycholate). Whereas 600 mg of the gel fraction was required to inhibit 50% of starch digestion, as little as 80 mg inhibited 50% of bile salt uptake. The present study indicated that the gel fraction, i.e., galactomannan, in the fenugreek seed is the factor which may be of potential benefit of fenugreek seeds in controlling plasma glucose and cholesterol levels.

INTRODUCTION

The seeds of fenugreek (*Trigonella foenum-graecum*), an annual plant of the legume family, are reported to have hypoglycemic and hypocholesterolemic effects (Shani et al., 1974). Ground seeds of fenugreek offered to diabetic rats reduced the postprandial glucose levels (Madar, 1984). Similar results were reported in alloxan-diabetic dogs (Ribes et al., 1986). Recently, it was shown that supplementation of ground seed reduced the plasma glucose levels in normal and diabetic subjects (Madar et al., 1988; Sharma, 1986a). The mechanism by which fenugreek may modulate plasma glucose is by delaying gastric emptying and direct interference with intestinal glucose absorption (Madar, 1984). In addition, a significant improvement in insulin response and a reduction in cholesterol levels were obtained in diabetics but not in healthy subjects (Sharma, 1986b).

Several investigators have reported that the ground seed of fenugreek has hypoglycemic and hypocholesterolemic effect, but the factors responsible for this activity have not been identified. Sharma (1986b) showed that the whole seed had the greatest effect in reducing plasma glucose, followed by gum isolate and extracted seeds. Fenugreek seeds were studied in relation to formation, metabolism, and enzymatic hydrolysis of galactomannan (Dey, 1978; Meier and Reid, 1977; Reid, 1971; Reid and Davis, 1977; Reid and Meier, 1970). It seems that the galactomannan is the factor which reduces the plasma glucose. In the present study the polysaccharide composition of fenugreek was determined, considering the specific fraction involved with the effect on both in vitro (using inverted gut) and in vivo (using rats) starch digestion and bile salts absorption using the inverted gut technique. In addition, the hypoglycemic effect of this fraction in vivo was evaluated in normal rats.

* Address correspondence to this author.
‡ Hebrew University of Jerusalem.
§ Agricultural Research Organization.

MATERIALS AND METHODS

Isolation of the Seed Fractions for Sugar Analyses. Dry seeds of fenugreek were purchased from the local market, soaked in distilled water for 24 h at room temperature, and used for the various experiments (see Scheme I). The polysaccharides of the soaking water fraction (fraction B), as well as of the other soluble fractions, were obtained as a precipitate of alcohol-insoluble solids (AIS) in a solution of 70% ethanol stirred for 15 min and centrifuged at 3000*g* for 15 min. The soaked seeds (fraction A) were separated by hand and treated as follows: the seed coat (including the endosperm) was crushed with a mortar in the presence of water (1:4, respectively) and filtered through a nylon sieve of 40 mesh, diluted to achieve complete dissolution, and centrifuged at 50000*g*. The AIS of the supernatant combined with the washing water of the nonfiltrated matter was freeze-dried (fraction C). The precipitate of the filtrate (fraction D) was also freeze-dried. The nonfiltrated matter of the seed coat, including the endosperm, was washed several times with distilled water and then freeze-dried (fraction E). The AIS fractions and the insoluble matter were freeze-dried, and ≈10 mg of the dry matter was used for sugar analysis. The alditol acetates of the acid-hydrolyzed polysaccharides were analyzed by gas–liquid chromatography (GLC) according to the method of Slonker (1971). Pectin (as galacturonic acid) was analyzed according to the method of Blumenkrantz and Asboe-Hansen (1973).

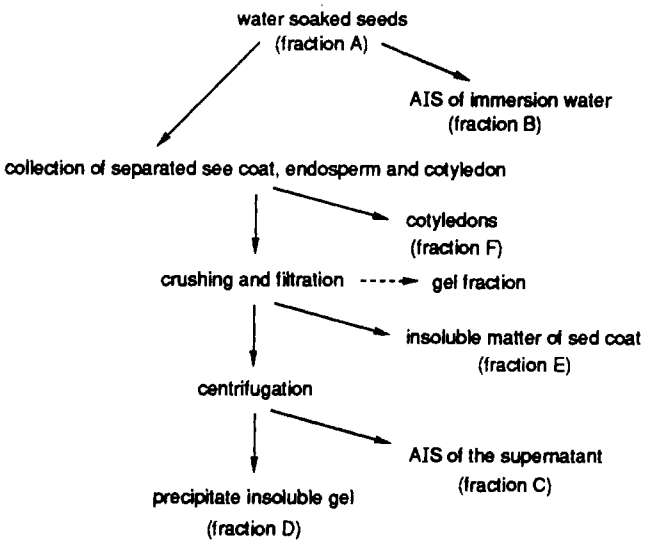
Isolation of Gel Fraction and Alcoholic Gel Fraction for in Vitro and in Vivo Studies. Fraction A was separated into two fractions: the seed coat and cotyledons. The seed coat was crushed in the presence of water (1:4) and filtrated through a sieve of 40 mesh. The resultant filtrate was called the "gel fraction". Ethanol was added to the gel fraction to yield a final concentration of 70% ethyl alcohol, and the mixture was stirred for 15 min and centrifuged for 15 min at 3000*g*. The alcoholic precipitate was lyophilized for later use.

Animals. Male Hebrew strain rats (Sabra), weighing 150–180 g, were used for the experiment. The animals were housed in a controlled environment (22 ■ 2 °C and 12-h light–dark cycle). A regular laboratory diet that met the American Institute of Nutrition Recommendations (1980) was provided ad libitum.

Determination of Bile Acid Absorption Using the Inverted Sac Technique. The preparation of the inverted sac was described previously (Madar, 1983). Briefly, male rats were decapitated, and the intestine was removed by cutting of both the upper end of the duodenum and the lower end of the ile-

0021-8561/90/1438-1535$02.50/0

Scheme I. Procedure for Isolation of Fractions



um. The entire intestinal content was washed with cold NaCl solution (0.9% w/v). The intestine was divided into 7-cm-long fragments and turned inside out. The inverted sac was lightly tied at one end, and 1 mL of Krebs-Henselteit buffer (KHB) was introduced into the inverted sac, which was then ligated. The sac was placed in an Erlenmeyer flask (25 mL) containing 6 mL of KHB with or without varying amounts (0–600 mg of dry matter/6 mL) of gel fraction isolated from fenugreek. Labeled (0.05 μCi) and unlabeled (0.3 mM) [^{14}C]taurocholic acid (sodium salt) or [^{14}C]deoxycholic acid (sodium salt) was also included in the reaction mixture. The flasks were gassed with O_2:CO_2 (95:5) for 30 s, tightly capped, and incubated in a shaking bath at 37 °C for 2 h. At the termination of the incubation period, the inverted sac was removed and the inner fluid was collected. The volumes of the inner (serosal side) and the outer (mucosal side) content were measured. One milliliter of each was centrifuged for 2 min at 10000*g* (minifuge), and then 200 μL of each supernatant was transferred into minivials. Scintillation liquid (5 mL) containing Triton X-100 was used for radioactivity measurement. The total radioactivity in the serosal side and in the mucosal side was calculated.

Determination of Starch Digestibility. The digestibility of starch in the presence of the gel fraction was determined by using the inverted sac technique as described above. Six milliliters of potato starch (2% w/v in KHB) in the presence or absence of various amounts of gel fraction was placed in the mucosal side. One milliliter of pancreatin (10 mg/mL) was also included in the reaction mixture. The sacs were incubated at 37 °C for 2 h, and the glucose liberated was measured in the serosal and mucosal sides.

In Vivo Effect of the Gel Fraction on Rats. *Meal Tolerance Test.* Following an 18-h fast, the rats were given orally, via a stomach tube, starch (0.5 g/100 g bw) with or without gel or alcoholic fractions (2 g). The starch was heated with water for 5 min. Blood samples were collected from the tail vein of the rats into tubes that had been prewashed with heparin (400 units/mL) and 0.1 mM NaF and dried. The blood samples were withdrawn from each rat preloading and 30, 60, 90, 120, and 180 min following the intubation.

Pancreatin, heparin, and NaF were obtained from Sigma Chemical Co. (St. Louis, MO); [*carbonyl*-^{14}C]taurocholic acid, sodium salt (specific activity 55.7 mCi/mmol), and [^{14}C]-deoxycholic acid, sodium salt (specific activity, 50 mCi/mmol), were purchased from Amersham Inc. (England).

Measurement and Analyses. Plasma glucose was determined by the glucose oxidase method using a Beckman glucose analyzer (Bun 2, Palo Alto, CA). All the data are presented as mean ± SEM, and the statistical analysis was carried out by the Student *t*-test.

RESULTS

The predominant sugars in the AIS matter of immersion water (fraction B) separated from the soaked seeds were

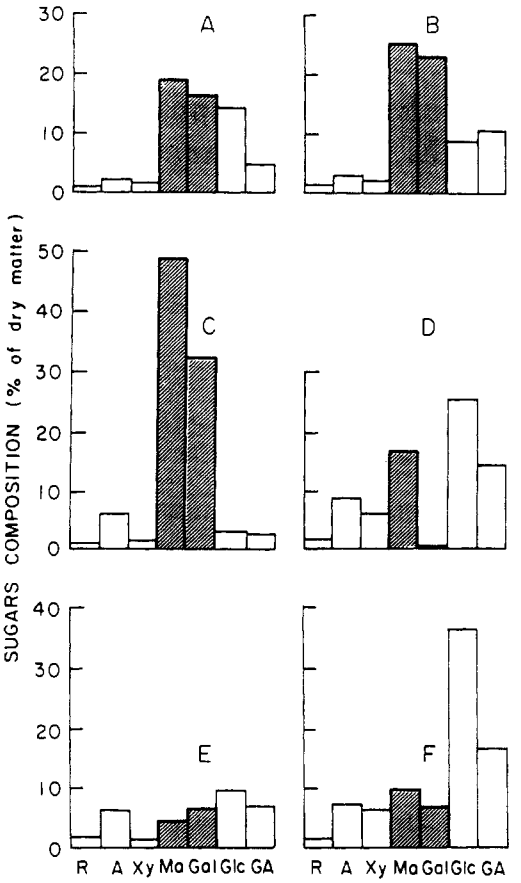


Figure 1. Content (percent of dry matter) of the neutral sugars rhamnose (R), arabinose (A), xylose (XY), mannose (Ma), galactose (Gal), and glucose (Glc) and pectin as galacturonic acid (GA) in the various fractions isolated from natural dry seeds of fenugreek (A), alcohol-insoluble solids of the soaking water (B), alcohol-insoluble solids of the gel (C), precipitate of the gel fraction (D), gel-extracted seed coat (E), and the cotyledons (F).

mannose and galactose in a ratio of 1.1:1, with the other sugars appearing in relatively low content (Figure 1B). This indicates leakage of galactomannan from the seed coat into the immersion water. Microscope observations showed that most of the cell wall of the seed coat dissolved during the soaking, and this accounts for the galactomannan found in the soaking water. Most of the dry matter (38%) was found in the cotyledons, and the main sugars of their polysaccharides are glucose as cellulose and galacturonic acid as pectin (Figure 1F). The polysaccharides of the fibrous fraction were found mainly in the seed coat (including the endosperm). The seed coat consisted of about 14% of the seed dry matter, most of which was composed of polysaccharides. Fraction C derived from the seed coat consisted of mostly mannose and galactose, in a ratio of 1.5:1. This sugar composition indicates galactomannan polysaccharides and is in agreement with other studies of fenugreek (Dey, 1978). The other sugars, such as rhamnose, arabinose, glucose, and galacturonic acid, were found to constitute several percentage units each. The insoluble matter of fraction E included relatively small amounts of sugars. The other fractions, such as the cotyledon (fraction F), the insoluble seed coat (fraction E) and fraction D, consisted of polysaccharides in which the cellulose (as glucose) and pectin were the main components (Figure 1D,F). Some of the soluble polysaccharides leaked from the seed coat during the latter's immersion in water. It seems that there are two fractions of galactomannan, the main one found in the endosperm

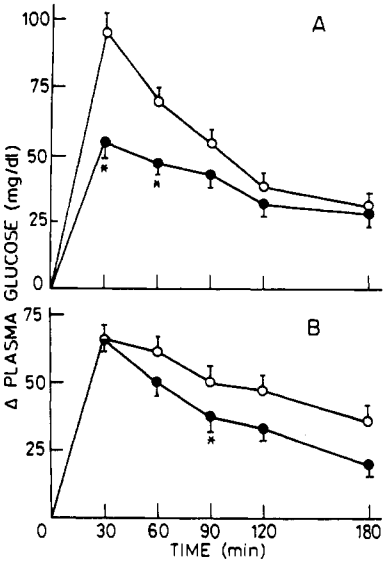


Figure 2. Change in plasma glucose level in response to starch (○) or starch plus gel fraction (●) derived from fenugreek. (A) The starch (0.5 g/100 g b.w.) was intubated along with the gel fraction (2 g/100 g b.w.). (B) The gel fraction was given 30 min prior to starch. Results are the mean ± SEM of 10 rats $p < 0.05$.

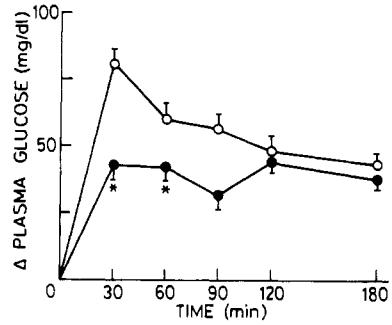


Figure 3. Change in plasma glucose level in response to starch (○) or starch plus ethanolic fraction (●) derived from gel fraction. Results are mean ± SEM of 10 rats. $p < 0.05$.

(with a mannose:galactose ratio of 1.5:1) and the second one found in the seed coat cell wall (with a ratio of 1.1:1).

Effect of Gel Fraction on Postprandial Glucose Levels in Rats. Figure 2A demonstrates the effect of the gel fraction on the digestion of starch intubated at the same time on postprandial glucose level in rats. The gel fraction significantly reduced the change in glucose levels at 30 and 60 min following the ingestion of the starch and the gel fraction. The glucose curve was blunted, and the glucose level in rats given the gel fraction continued to be lower at 90, 120, and 180 min but without significant difference. When the gel fraction was given 30 min prior to the starch intubation (Figure 2B), only at 90 min was the glucose level significantly lower than in rats given starch without gel fraction derived from fenugreek. Nevertheless, the glucose levels in rats that ingested starch concurrently with the gel fraction tended to be lower than the corresponding group which ingested starch without the gel fraction. The alcoholic fraction was also given orally with starch. The plasma glucose following the ingestion was measured and is illustrated in Figure 3. The ethanolic fraction decreased the glucose level following starch ingestion but reached a significant difference only at 30 and 60 min.

Effect of the Gel Fraction Derived from Fenugreek Seed on Potato Starch Digestion. Table I presents the effect of the gel fraction on the digestion of starch using inverted gut from rat. The gel fraction

Table I. Effect of the Gel Fraction Derived from Fenugreek Seed on Potato Starch Digestion[a]

gel fraction concn, mg	glucose (mg) liberated in serosal side	glucose (mg) liberated in mucosal side	glucose (mg) liberated in total	inhibition,[b] %
0	3.5 ± 1.50	43.8 ± 1.5	47.3 ± 1.5	
80	1.5 ± 0.30	35.2 ± 1.1	36.7 ± 0.9	23
160	0.3 ± 0.10	28.3 ± 0.8	28.6 ± 0.9	39
320	0.4 ± 0.03	27.7 ± 0.9	28.1 ± 1.2	41
600	0.2 ± 0.12	21.8 ± 0.7	21.8 ± 0.6	54

[a] Results are expressed as means ± SEM of five experiments (5 rats), with total of 15 gut sections. [b] The inhibition percentage was calculated as the ratio of total glucose in the presence and absence of the gel fraction.

Table II. Effect of Different Concentrations of the Gel Fraction Derived from Fenugreek Seed on Uptake of [^{14}C]Deoxycholic Acid (0.3 mM) and [^{14}C]Taurocholate (0.3 mM) in Rat Inverted Sac (Lower Part of the Ileum)

gel concn, mg of dry matter	radioactivity found (dpm × 10^3) of [^{14}C]deoxycholic acid serosal side	[^{14}C]deoxycholic acid mucosal side	[^{14}C]taurocholate serosal side	[^{14}C]taurocholate mucosal side
0	21 ± 4.3	213 ± 1.9	8 ± 0.2	85.4 ± 0.9
5	20 ± 0.2	184 ± 6.9	10.7 ± 1.6	76.1 ± 2.2
10	19 ± 1.7	198 ± 3.2	9.1 ± 0.9	74.9 ± 0.3
20	16 ± 3.2	167 ± 0.5	4.2 ± 0.5	68.8 ± 3.2
40	9.5 ± 2.1	153 ± 16.8	4.0 ± 0.7	61.8 ± 4.6
80	7.4 ± 1.9	106 ± 2.8	2.7 ± 0.1	40.7 ± 1.3
160	1.9 ± 0.1	103 ± 6.4	1.2 ± 0.3	34.1 ± 1.1

[a] Results are expressed as mean ± SEM of five experiments.

inhibited the starch digestion in the mucosal side and also inhibited glucose absorption into the serosal side. A high content of gel fraction (600 mg) inhibited the glucose absorption almost completely (3.5 ± 1.5 vs 0.2 ± 0.12 mg/dL), but only 50% starch inhibition was reached with this concentration when the results are expressed as total inhibition of glucose found in the serosal and mucosal sides. The inhibition effect in both sides of the inverted sac was not linear with the gel concentration. When the total inhibition effect of the gel fraction on absorption (mucosal side) and digestion (serosal side) was calculated, 23%, 39%, and 41% inhibition was found when 80, 160, and 320 mg of gel was used, respectively.

Effect of the Gel Fraction Derived from Fenugreek Seed on Bile Acid Salts Absorption. The uptake of bile salts in the presence of different concentrations of gel fraction in the mucosal side of the inverted sac is shown in Table II. The uptake of both bile acids was inhibited by the gel fraction. The inhibition is concentration dependent, but not in a linear fashion. The amount of radioactivity was decreased in both sides, mucosal and serosal, in the presence of the gel fraction. The reduction in the radioactivity was dependent on the concentration of the gel fraction used. When the results were expressed as residual of radioactivity inside the inverted sac, the effect of the gel fraction on both bile salts was similar (Figure 4). A residual of 50% in the bile salts was found in the mucosal side when 40 mg of gel fraction was used, whereas 80 mg of gel fraction was required to inhibit 50% of bile salts absorption from the serosal side. A higher concentration of gel fraction (above 80 mg) did not have a significant effect on bile salts absorption from the serosal side. However, 160 mg of gel fraction inhibited 90% bile salts uptake in the mucosal side.

DISCUSSION

Fenugreek seed was found to be a hypoglycemic agent in animals and humans (Madar, 1984). The aim of this

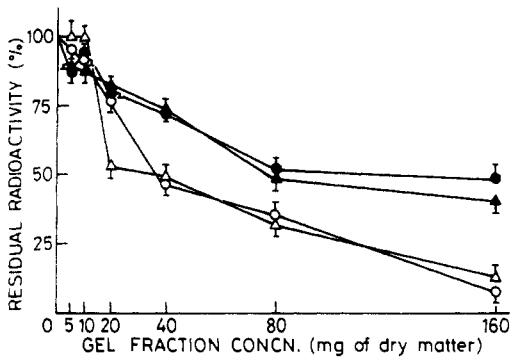


Figure 4. Effect of gel fraction on [^{14}C]deoxycholic acid (0.3 mM) and [^{14}C]taurocholate (0.3 mM) absorption in rat inverted sac. Sacs were incubated with different concentrations of the gel fraction. Following 2 h of incubation, the radioactivity in the serosal side and the mucosal side was measured. The results are expressed as a residual radioactivity of deoxycholic acid in the mucosal (○) and in the serosal (●) sides and of taurocholate in the mucosal (△) and in the serosal (▲) sides.

study was to identify which fraction derived from fenugreek is responsible for reducing the plasma glucose level and to evaluate the effect of this fraction on bile acid uptake. It is well-known that the seeds of *T. foenum-graecum* (fenugreek) contain relatively large amounts of galactomannan in the endosperm (Reid and Meier, 1970). Our unpublished microscope observations showed that water-soluble galactomannan comprises most of the cell walls of the seed coat cells. Some of the galactomannan, mainly from the outer cells of the seed coat, leaked into the immersion water.

Two main fractions were found in the cell wall of the seed coat: fraction C (AIS of the supernatant), which is the galactomannan, and the insoluble cellulose and pectin. Both fractions C and D (precipitate insoluble cell wall) included the neutral sugars such as rhamnose, arabinose, xylose, and galacturonic acid. Fraction C included also glucose and galacturonic acids in addition to the galactomannan, and the insoluble cell wall included also mannose and galactose in addition to the cellulose (glucose) and pectin. This composition indicates that each polysaccharide fraction included other sugar units in the main backbone of the specific polysaccharides, such as galactomannan cellulose and pectin. Nevertheless, the relative composition of the sugars mannose and galactose changes from one fraction to another (Figure 1). According to previous reports (Sharma, 1986a) the soluble dietary fiber is the main component attributed to the hypoglycemic effect. In the present study, experiments were conducted to see whether the gel fraction derived from seed coat has an effect on starch digestion in normal rats. The present study showed that the gel fraction reduced significantly the plasma glucose. The high capacity of the gel fraction to absorb water (16-fold) and its effect in shortening the transit time and inhibiting the gastric emptying, as shown previously, are the main contributors to the hypoglycemic effect (Madar, 1984). The timing of the intubation of the gel fraction plays an important role in the hypoglycemic effect (Figure 2, parts A vs part B). Giving the gel fraction simultaneously with the starch was more effective in reducing the plasma glucose than giving the gel fraction prior to the starch.

We have found that the gel fraction reached the upper intestine after 45 min of intubation (data not shown). This might be the reason for the moderate effect of the gel fraction when given separately. Hence, it appears that gel fraction and starch should reach the intestine at the same time. The mechanisms underlying the inhibitory effect of the gel fraction on starch absorption may be partly mechanical—by changing transit time and gastric emptying—and partly by interference with the digestive enzymes, which slows the starch digestion and leads to reduction in plasma glucose. The shorter intestinal residence time may promote a greater fraction of starch which escapes absorption, as it has been shown that changes in transit time affect starch output in the ileal effluent (Hamberg et al., 1989). Furthermore, the gel fraction also inhibited glucose liberation. The inhibition of glucose uptake and its lower level in the serosal side indicate that the gel fraction has an inhibitory effect on starch digestion and absorption. These results may explain the in vivo effect, that the gel fraction led to a reduction of the plasma glucose in rats, following starch ingestion (Figure 2). These results, however, did not indicate that the gel fraction has a direct effect on digestive enzymes. Nevertheless, the gel fraction has a high viscosity, thereby reducing enzyme–substrate contact (Edwards et al., 1988; Isaksson et al., 1982; Wong et al., 1985). In vitro experiments and perfusion studies have suggested that the fiber gelling agents may directly inhibit certain digestive and transport functions in the rat intestine (Elsenhans et al., 1984). In general, inhibition of nutrient transport and hydrolysis of disaccharides directly were related to the viscosity of the medium and therefore to the concentration of the gelling agent. As shown in this study (Figure 1), the gel fraction is a soluble galactomannan that fits the definition of a dietary fiber which increases the viscosity of the gut contents. Thus, the simultaneous ingestion of the gel fraction with food led to a reduction in the rate of nutrient absorption. However, it is not yet known how the gel fraction reduces the uptake of bile salts; this may be a result of several factors such as adsorption, bulking, gel forming, and water solubility. The water-holding capacity and the viscosity formed by the galactomannans appear to be important factors accounting for trapping of the bile acids and inhibition of their absorption (Story, 1985; Topping et al., 1988). The present results may indicate that the effect of the gel fraction on bile acids in the small intestine (in vivo) is to reduce the efficiency of the enterohepatic circulation, resulting in an increase in bile acid excretion and leading to a decrease in plasma cholesterol level.

In conclusion, the galactomannan derived from fenugreek seed leads to inhibition of starch digestion and bile acid absorption. Hence, it might be concluded that the fenugreek galactomannan may have a potential benefit to modulate carbohydrate (controlling plasma glucose) and lipid metabolism via alteration of starch digestion and bile acid absorption.

ACKNOWLEDGMENT

We thank A. Harel and R. Variliver for their skilled and conscientious technical assistance. Financial support for this study was partly provided by the Hebrew University and from the Agricultural Research Organization, The Volcani Center, Bet Dagan, Israel, No. 2780-E, 1989 series.

LITERATURE CITED

American Institute of Nutrition. Second report of the Ad Hoc committee on standards for nutritional studies. *J. Nutr.* **1980**, *11*, 1726–1730.

Blumenkrantz, N.; Asboe-Hansen, G. New method for quantitative determination of uronic acid. *Anal. Biochem.* **1973**, *54*, 484–489.

Dey, P. M. Biochemistry of plant galactomannans. *Adv. Carbohydr. Biochem.* **1978**, *35*, 341–376.

Edwards, C. A.; Johnson, I.; Read, N. W. Do viscous polysaccharides slow absorption by inhibiting diffusion or conversion. *Eur. J. Clin. Nutr.* **1988**, *42*, 307–312.

Elsenhans, B.; Zenker, D.; Caspary, W. F.; Blume, R. Guaran effect on rat intestinal absorption. *Gastroenterology* **1984**, *86*, 635–645.

Hamberg, O.; Rumessen, J. J.; Gudmund-hoyer, E. Inhibition of starch absorption by dietary fibre. A comparative study of wheat bran, sugar-Beet fibre and pea fibre. *Scand. J. Gastroenterol.* **1989**, *24*, 103–109.

Isaksson, G.; Lundquist, I.; Ihse, I. Effect of dietary fiber on pancreatic enzyme activity in vitro. *Gastroenterology* **1982**, *82*, 918–924.

Madar, Z. Demonstration of amino acid and glucose transport in chick small intestine everted sac as a student laboratory exercise. *Biochem. Educ.* **1983**, *11*, 9–11.

Madar, Z. Fenugreek (Trigonella foenum-graecum) as a means of reducing postprandial glucose level in diabetic rats. *Nutr. Rep. Int.* **1984**, *29*, 1267–1273.

Madar, Z.; Abel, R.; Samish, S.; Arad, J. Glucose-lowering effect of fenugreek in non-insulin dependent diabetics. *Eur. J. Clin. Nutr.* **1988**, *42*, 51–54.

Meier, H.; Reid, J. S. G. Morphological aspects of the galactomannan formation in the endosperm of Trigonella foenum-graecum L. (leguminosae) *Planta* **1977**, *133*, 243–248.

Reid, J. S. G. Reverse carbohydrate metabolism in germinating seeds of Trigonella foenum-graecum L. (Leguminosae) *Planta* **1971**, *100*, 131–142.

Reid, J. S. G.; Davis, C. Endo-β-mannanase, the leguminous aleurone layer and the storage galactomannan in germinating seeds of Trigonella foenum-graecum L. *Planta* **1977**, *133*, 219–222.

Reid, J. S. G.; Meier, H. Formation of reserve galactomannan in the seeds of Trigonella foenum-graecum. *Photochemistry* **1970**, *9*, 513–520.

Ribes, G. Y.; Sauvaire, Y.; Baccou, J. C.; Valett, G.; Chenon, D.; Trimble, E. R.; Loubatiers-Mariani, M. M. Antidiabetic effects of subfraction from fenugreek seeds in diabetic dogs. *Proc. Soc. Exp. Biol. Med.* **1986**, *182*, 159–166.

Shani, J.; Goldschmied, A.; Joseph, B.; Aharonson, Z.; Sulman, F. G. Hypoglycemic effect of Trigonella foenum-graecum and lupinus termis (Leguminosae) seed and their major alkaloids in alloxan diabetic and normal rats. *Arch. Int. Pharmacodyn. Ther.* **1974**, *210*, 27–36.

Sharma, R. D. An evaluation of hypocholesterolemic factor of fenugreek seeds (T. *foenum-graecum*) in rats. *Nutr. Rep. Int.* **1986a**, *33*, 669–677.

Sharma, R. D. Effect of fenugreek seed and leaves on blood glucose and serum insulin responses in human subjects. *Nutr. Res.* **1986b**, *6*, 1353–1364.

Slonker, J. H. Determination of cellulose and apparent hemicellulose in plant tissue by gas liquid chromatography. *Anal. Biochem.* **1971**, *43*, 539–546.

Story, J. A. Dietary fiber and lipid metabolism. *Proc. Soc. Exp. Biol. Med.* **1985**, *180*, 447–452.

Topping, D. L.; Oakenfull, D.; Trimble, R. P.; Illeman, R. A. A viscous fibre (methylcellulose) lowers blood glucose and plasma triacylglycerols and increases liver glycogen independently of volatile fatty acid production in the rat. *Br. J. Nutr.* **1988**, *59*, 21–30.

Wong, S.; Traianedes, K.; O'Dea, K. Factors affecting the rate of hydrolysis of starch in legumes. *Am. J. Clin. Nutr.* **1985**, *42*, 38–43.

Received for review October 5, 1989. Accepted March 12, 1990.

Registry No. Galactomannan, 11078-30-1; starch, 9005-25-8; sodium taurocholate, 145-42-6; sodium deoxycholate, 302-95-4; glucose, 50-99-7; pectin, 9000-69-5.