portive problem with her was one of supplying nutrition, particularly calories.

The one child to whom fat has been given was a four-year-old boy in whom the maintenance of an adequate caloric intake was the major supportive problem.

As is quite obvious, the clinical studies with intravenous fat are just beginning and at this time it is not possible to evaluate the results. It can be said, however, that fat emulsions have been prepared which can be given safely to man and which do not give rise to changes in temperature, pulse, blood pressure, or respiration.

Summary

Fat emulsions satisfactory for intravenous administration have an important role to play in parenteral nutrition. They offer an opportunity to provide adequate calories in a limited fluid volume. Fat emulsions given intravenously are utilized for energy requirements for growth and maintenance and are helpful in maintaining a positive nitrogen balance. They have been given successfully to man. They should play an important role in supportive and preventive therapy in pre- and post-operative care and in any disease characterized by serious weight loss or emaciation.

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Viscosity of Cottonseed Protein Dispersions[1]

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Introduction

IN a previous publication (1) a method for dispersing cottonseed proteins in concentrations as high as 25% protein to produce dispersions which were viscous, tacky, and did not gel was described. The preparation of the dispersions was accomplished by the inclusion of trichloracetate ion in the alkaline dispersing mixture, thereby preventing gel formation. The viscosity of the cottonseed protein dispersions was found to decrease on aging the dispersion and to be dependent upon the concentration of alkali used in its preparation and upon the concentration of protein. The instability of viscosity of the dispersions made it difficult to utilize such dispersions for manufacture of fibers, films, adhesives, sizes, and related products because the "working life" was too short for industrial operations.

This publication describes the results of an investigation of the effect of various methods of preparing the meal and of preparing the cottonseed protein dispersions upon their viscosity characteristics.

EXPERIMENTAL

Materials and Methods

Isolation of Protein. Cottonseed protein was isolated from oil-free meals which were obtained by three methods of solvent extraction: 1. removal of the oil by means of n-hexane (6), 2. removal of the oil by means of the mixed solvent flotation process (deglanding process) followed by a second extraction with n-hexane (2, 7), and 3. removal of the oil by means of isopropanol.[3] The maximum solvent temperature reached during the preparation of the cottonseed meals was less than the boiling point of hexane. Cottonseed meal, prepared as described, was suspended in a 0.2 N sodium sulfite solution at pH 7.5 in the ratio of 10 liters of solution to 1 kilogram of meal. The suspension was stirred for 2 hours at room temperature, after which the extract was separated from the insoluble residue by centrifugation, in a solid basket centrifuge, and the protein was precipitated by the addition of gaseous sulfur dioxide to the extract, lowering the pH to 4.0. The protein curd was washed several times with water and twice with acetone, after which it was air-dried at room tem-

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[1] Presented at the 113th National Meeting of the American Chemical Society, April 19-23, 1948, at Chicago, Illinois.

[2] One of the laboratories of the Bureau of Agricultural and Industrial Chemistry, Agricultural Research Administration, U. S. Department of Agriculture.

[3] Meal supplied through the courtesy of W. D. Harris, Agricultural and Mechanical College of Texas, College Station, Tex.

perature (5). The analyses of the meals and protein used in this investigation are given in Table 1.

TABLE I

Analyses of Meals and Proteins [1]

Material	Nitrogen, %	Ash, %	Lipids, %
Isopropanol-extracted meal	11.03	8.32	0.69
Degланded-hexane-extracted meal	9.33	......	0.60
Hexane-extracted meal	9.11	......	0.74
Protein	15.6	2.8	......

[1] Calculated on a moisture-free basis.

Apparatus. The absolute viscosities of the concentrated protein dispersions were measured with a Höppler rolling ball viscometer (3) and a MacMicheal torsion wire viscometer (4) at a temperature of 25° ± 0.02°C. The viscometers were calibrated by means of standard viscosity oils provided by the National Bureau of Standards. Prior to use, the viscometers and accessories were washed with water and ethanol and dried with air.

Results and Discussion

The factors previously shown to affect the viscosity characteristics of cottonseed protein dispersions are the concentration of the protein, the amount of sodium hydroxide used to disperse the protein, the age of the dispersion, and the presence of specific anions (1). These factors have been more extensively investigated with results as reported here. In addition, it has been found that the method of removal of oil from the meal has a pronounced effect on the properties of the protein. The effect of method of oil removal and the effect of addition of reducing sugars to the dispersion was also determined.

Effect of Aging of the Protein Dispersion. The data in Figure 1 show the effect of aging of the

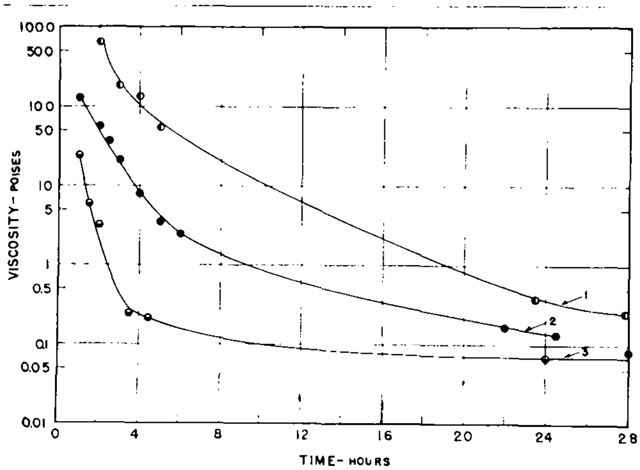

FIG. 1. Effect of aging and alkali concentration on the viscosity of 20% cottonseed protein dispersions at a temperature of 25°C. 1. 12% NaOH; 2. 14% NaOH; 3. 16% NaOH.

dispersions on their viscosity for a period of time greater than 24 hours. The properties of the protein used in this investigation differ from previously reported proteins in that the nitrogen percentage is slightly lower and the ash content, slightly higher. All three of the dispersions of 20% protein concentration were prepared with protein isolated from deglanded cottonseed meal by use of sodium hydroxide and to prevent gelation contained 2.5% (based on the weight of protein) of trichloracetate ion. The concentrations of sodium hydroxide used to effect dispersion were 12, 14, and 16% of the weight of protein for dispersions Nos. 1, 2, and 3 of Figure 1, respectively, and the pH of the dispersions was 12.5 to 12.6. The pH of the dispersions was measured using a glass electrode with a Beckmann pH meter, model G. No correction was made for the sodium ion effect. The viscosities of the dispersions were unstable, and for most practical uses the dispersions had a short "working life."

Effect of Protein Concentration. The relative effect of varying the protein concentration on the viscosity of dispersions containing 14% sodium hydroxide and 2.5% trichloracetate ion is shown in Figure 2. In this experiment the protein was also prepared from

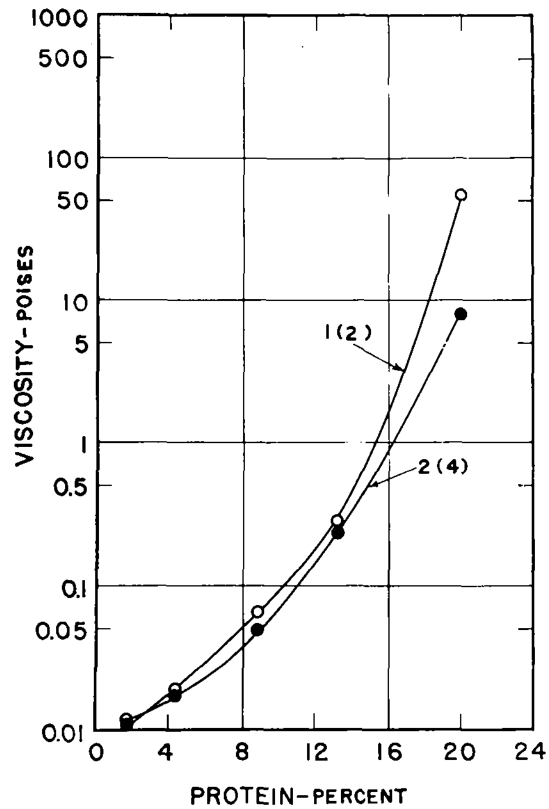

FIG. 2. Effect of protein concentration on the viscosity of the cottonseed protein dispersions aged 2 hours and 4 hours at a temperature of 25°C. (14% NaOH, 2.5% trichloroacetate, pH 12.5.)

deglanded meal. The viscosity obtained after 2 and 4 hours of aging is given for each concentration of protein. It is clear that the viscosity is very sensitive to small changes in protein concentration. The sensitivity is greatest when the dispersions are first made and decreases somewhat with their age.

Effect of Sodium Hydroxide Concentration. The effect of sodium hydroxide concentration on the viscosity characteristics was determined on dispersions prepared with protein made from hexane-extracted meal, isopropanol-extracted meal, and deglanded meal. In the case of the last two-named protein preparations, dispersions were tested at two concentrations of protein. From the results of this investigation, as given in Figure 3, it is clear that the viscosity of the dispersions decreased almost logarithmically with increasing sodium hydroxide concentration. The viscosities of 20% dispersions prepared from hexane-extracted meal and from deglanded cottonseed meal

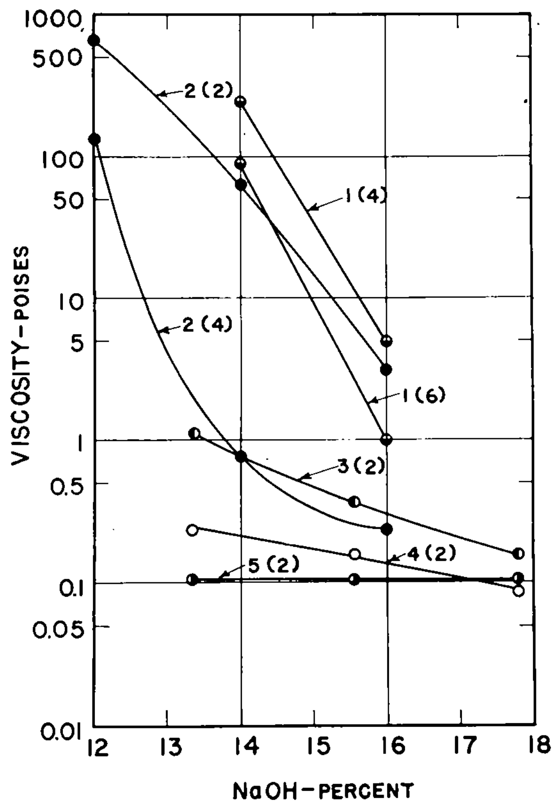

FIG. 3. Effect of NaOH concentration and method of solvent extraction of the meal on the viscosity of cottonseed protein dispersions. 1. hexane-extracted meal, 20% protein; 2. deglanded and hexane-extracted meal, 20% protein; 3. isopropanol-extracted meal, 18% protein; 4. deglanded and hexane-extracted meal, 13.5% protein; 5. isopropanol-extracted meal, 13.5% protein.

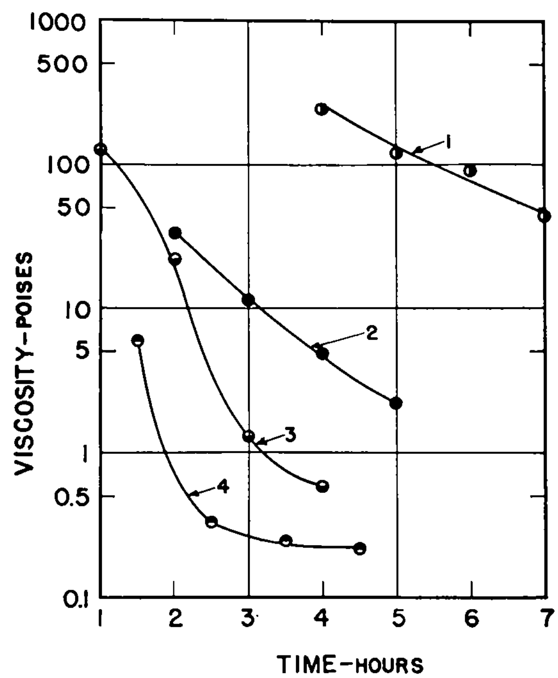

FIG. 4. Effect of method of solvent extraction of the meal on the viscosity of cottonseed dispersions.

1. Hexane-extracted meal, 20% protein and 14% NaOH.
2. Hexane-extracted meal, 20% protein and 16% NaOH.
3. Deglanded and hexane-extracted meal, 20% protein and 14% NaOH.
4. Deglanded and hexane-extracted meal, 20% protein and 16% NaOH.

were measured at the end of 4 and 6 hours and are plotted (see Curves 1 and 2, Figure 3) to show the effect of sodium hydroxide concentration upon dispersions of different ages. The decrease in viscosity which is observed at higher concentrations of sodium hydroxide is probably due to the hydrolysis of the protein.

Effect of Method of Removal of Oil. The extent of denaturation of cottonseed proteins by temperature, solvent, and mechanical agitation, which takes place when the oil is extracted from the cottonseed flakes, influences the viscosity of the dispersion of proteins prepared from such meals, as is shown in Figures 3 and 4. Proteins isolated from deglanded

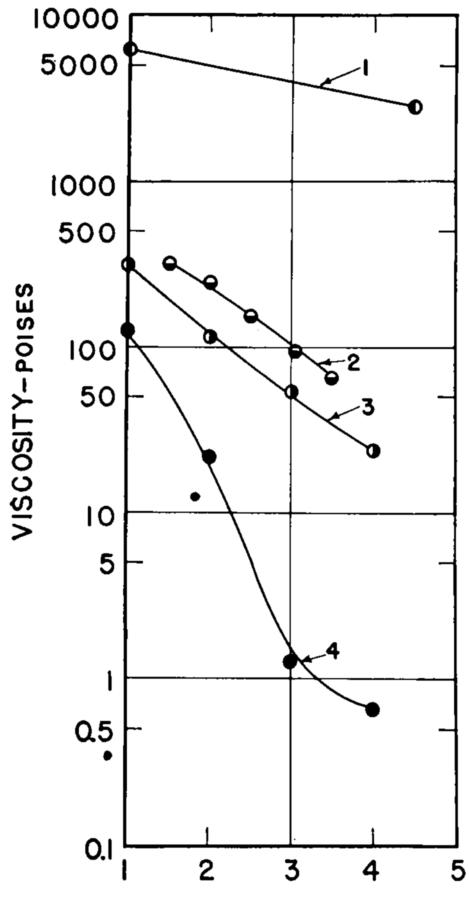

FIG. 5. Effect of dextrose on the viscosity of cottonseed protein dispersions. (Deglanded-hexane-extracted meal.) 1. 20% dextrose; 2. 12.5% dextrose; 3. 10% dextrose; 4. 0% dextrose.

meals seem to be modified to a greater degree than proteins isolated from hexane-extracted meals, as is indicated by the viscosity characteristics of the dispersions. In every case, as is shown in Figure 4, the proteins prepared from hexane-extracted meals gave more viscous and stable dispersions.

Effect of Sugars. The effect of sugars on the viscosity and stability of protein dispersions was determined by incorporating sugars with the protein and trichloracetate ion as the dispersions were made, with results as shown in Figures 5 and 6. In all of these experiments the protein concentration was maintained at 20%, and 14% sodium hydroxide and 2.5% trichloracetate ion were used to effect the dispersion. The addition of dextrose increased the initial viscos-

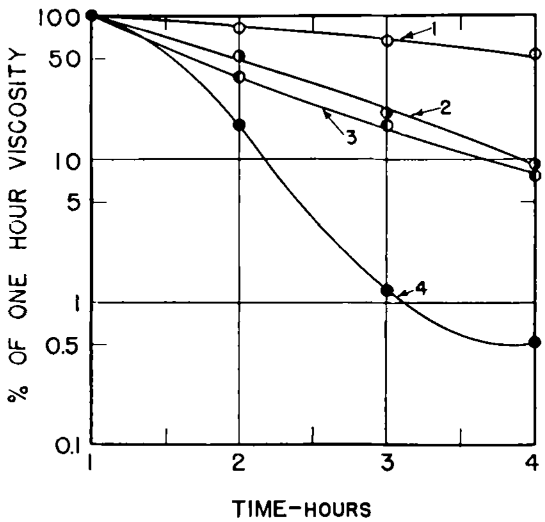

FIG. 6. Effect of dextrose on relative value of the viscosity of cottonseed protein dispersions. 1. 20% dextrose; 2. 12.5% dextrose; 3. 10% dextrose; 4. 0% dextrose.

ity of the dispersions and decreased the relative rate of decrease of viscosity with age, as is shown in Figure 6. The data in this figure have been plotted to show the effect of dextrose on the percentage change decrease in viscosity with time compared to the viscosity value obtained 1 hour after the dispersion was made.

Although the addition of sucrose to the dispersions increased the initial viscosity of the dispersions, the relative rate of decrease in viscosity was not affected, as is shown by the experiments given in Figure 7. A number of experiments on the effect of a variety of sugars on the viscosity and stability of cottonseed protein dispersions indicates that the ability to decrease the rate of change of viscosity of these dispersions with age is related to reducing sugars. The ability of the reducing sugars to stabilize the protein dispersions was always accompanied by a large increase in the initial viscosity upon addition of the sugar, thus indicating that a reaction probably occurs between the two types of molecules to form larger and more stable aggregates.

With the information now available on the effect of reducing sugars on the viscosity and stability of cottonseed protein dispersions, it is possible to prepare dispersions with a practical "working life" by the addition of 12.5 to 15% dextrose (on the basis of the weight of protein). The viscosity of such dispersions will not decrease below 50 poises during a 4-hour period.

Summary

Tacky and viscous cottonseed protein dispersions can be prepared from proteins isolated from hexane-extracted meals, deglanded-hexane-extracted meals, and isopropanol-extracted meals by means of sodium hydroxide and trichloracetate ion. The addition of sugars increases the viscosity of the dispersions, and dextrose significantly decreases the rate of change in

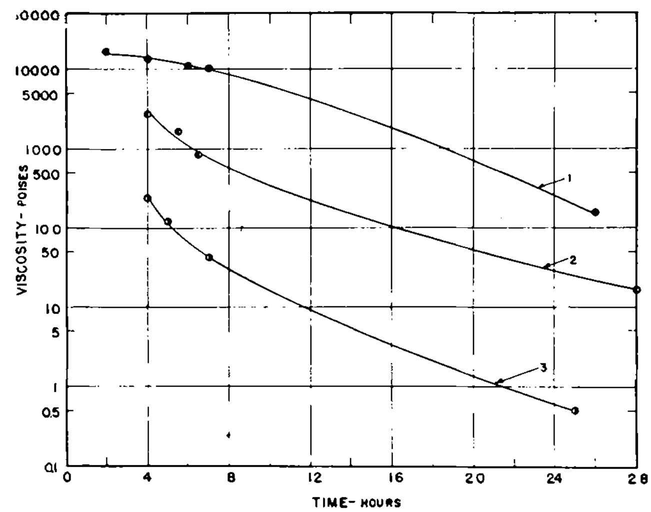

FIG. 7. Effect of sugars on viscosity of cottonseed protein dispersions. (Hexane-extracted meal.) 1. 10% dextrose; 2. 10% sucrose; 3. control, no sugar added.

the relative viscosity. Proteins isolated from hexane-extracted meals seem to be less modified than those isolated from deglanded-hexane-extracted meals, as indicated by the higher viscosity and stability of the dispersions.

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