A STUDY OF THE RELATION BETWEEN THE PEPTIZABILITY OF THE PROTEIN BY INORGANIC SALT SOLUTIONS, ASH CONTENT AND BAKING QUALITY OF WHEAT AND WHEAT FLOUR.

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JESSIE S. ROBERTS

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INTRODUCTION.

Of the many qualities of wheat flour, that designated as flour strength is, perhaps, the most interesting. From both the practical standpoint, and from the purely theoretical it is of first importance. To the miller and baker it means the ability the flour possesses to make large well piled loaves. To the chemist it means a combination of chemical and physico-chemical characteristics of probably a group of constituents, the sum total of which determines the type of reaction the flour will give in its various treatments.

Flour strength as measured by baking quality is definitely associated with protein content, other constituents being equal. High protein wheats as a rule give large, well piled loaves. But total protein content alone cannot be the determining factor for in many cases erratic results are obtained when it is used as a standard for judgment. Especially in the case of frost damaged wheats do we find high protein wheat producing poor loaves. Therefore there must be differences in quality of the various proteins.

The term quality is ambiguous; whether it refers to the physical or the chemical properties of the protein is still a question. From the chemical viewpoint, quality would be dependent on the chemical constitution and on variation in amount of the groups of protein present. From the physical, it would depend only on the state of the protein particle, as found in the flour.

As yet the chemical constitution of the flour protein is unknown. Variations in amount extracted by the con-

mon solvents show no relationship to flour strength. From the physical standpoint more surprising results are obtained.

Sharpe and Gortner (1927) found that the amount of protein dissolved in salt solutions varied both with the kind of salt used, and with the flour. Their results showed that the so-called globulin fraction could not be a separate entity. Such variation in results could not be explained in terms of solubility but could be accounted for if the protein particle were considered as a micelle, and solubility as peptization of the colloidal particle. The degree of peptization seemed related to strength. The proteins of those flours designated as weak peptized more readily and more completely than did those of the strong.

From several centres work relating ease of peptization of protein to the baking quality of various types and conditions of wheat has been reported. In some instances high correlations are shown between the amount peptized and baking quality. In others, baking quality is correlated more closely with non peptized fraction. More recently it has been suggested that the electrolyte content as determined by total ash is responsible for the degree of peptization of the flour. Rich (1933) has suggested that though the total ash may not be wholly responsible in itself, yet it must be related to the same factor as is peptizability. It was felt that it would be desirable at this time to investigate further this relationship between peptization ash content and baking quality, and to determine the effect of soluble ash

on the degree of peptization. At this time a series of frost damaged wheats was available. This was used for the investigation.

HISTORICAL

For the past 40 years, intensive research on the subject of flour proteins has been carried on. In 1893 Osborne and Voorhees established the composition of wheat flour as containing five distinct proteins--an albumin (leucosin); a globulin; an almost insignificant amount of proteose; a prolamin (gliadin) and a glutelin (glutenin). The gliadin and glutenin constitute the gluten. The albumin is soluble in water and both it and the globulin are soluble in dilute saline solutions. The gliadin is slightly soluble in water and freely soluble in 50 - 70 % alcohol. The glutenin is insoluble in water, in saline solutions, and in alcohol, but is dispersed by dilute acid and alkali.

These results were confirmed by other investigators when identical technique was followed, but it was soon observed that different amounts of these fractions were obtained from the same flour if the order of procedure was altered. Chamberlain found that considerable quantities of proteins which were supposedly soluble in only the one reagent could be extracted by another. He was able to remove part of the alcohol soluble nitrogen by extracting first with salt solution.

Greaves (1911) showed the effect of environmental conditions on degree of extraction. He found that the per-

centage nitrogen extracted with 70 - 74 % alcohol varied with the proportion of flour to alcohol used, that hot alcohol extracted a considerably larger proportion than did the cold, and that flour previously dried at 96° C yielded less protein to the alcohol than did the untreated flour.

Olson (1914) pointed out that substances allied to gliadin which closely merge into one another are present in the alcohol soluble fraction. Like Chamberlain he found that the 1 % salt (NaCl) extracts a part of the gliadin in addition to the globulin and albumin. In a later paper (1914) comparing the fractions obtained in 1 % and 10 % NaCl extractions he states:--

> "That the amount of nitrogenous material separated varies in any one group depending upon the concentration of the solvent used for extracting these bodies."

The 10 % NaCl concentration gave higher results than were found possible with any technique with a 1 % salt solution.

As in the instance of the alcohol soluble and salt soluble fractions similarly glutenin shows variation in degree of solubility. Blish, et al, (1927) showed that if the ratio of flour to sodium hydroxide is altered variations in percentage of glutenin extracted occurs. Thus a study of the literature points strongly to the fact that the accepted separations cannot be exact owing to these overlapping solubilities of the proteins in the solvents used.

The major portion of the work of this period was concentrated on the prolamine and glutelin fractions. More recently, attention has been turned in another direction. In 1927 Hoffman and Gortner further confirmed the work on the gliadin, glutenin and albumin, but were unable to substantiate former results on the globulin fraction. Their data show that extraction with 5 % K_2SO_4 and 10 % NaCl do not yield similar fractions. In 1928 Gortner furthering their data raised the question, "What salts and what concentrations are implied in the definition of globulin (as given by the Physiological and Biochemical Committee on nomenclature) which states that 'globulins are simple proteins insolubble in pure water, but soluble in neutral solutions of strong bases with strong acids' ?" On extracting flour proteins with a variety of such salts, a surprising difference in amount of protein removed resulted. Thus with 1.0 N concentrations of a series of KI, KCl, KBr, KF, protein ranging from 13.07 % - 63.89 % total protein was removed. Expressing these fractions in terms of the non gluten proteins they mount from 69.2 % to 340.2 %. Therefore, as Gortner states, the salt soluble protein fraction does not represent a mixture of albumin and globulin nor can it represent the non gluten proteins. Equally striking variability of the individual flours toward a single salt solution was observed. With 1.0 N KF the range of non gluten protein extracted from various flours ranged from 51 - 95 %, with KI from 266 - 391 %.

To explain these results the protein particle must

be considered as a micelle, and solubility as the peptization of this micelle. Then, varying degree of peptization would result from varying enfironmental conditions. Its extent would be governed by the nature of the anions and cations present and by the concentration of the solvent used. This viewpoint is not new. In 1907 Wood, and 1908 Wood and Hardy claimed that the colloidal properties of the flour proteins were of primary importance. But with the publication of the above results attention was re-turned to the hypothesis of the earlier investigators. The majority of the work in the field was now directed away from a study of the solubility of the protein as a whole. Since the flours showed a difference in degree of peptization by the same solution it was thought that peptizability might lend itself as a measure of strength for results indicated that certain weak flours were more readily dispersed than were the strong.

Gortner (1920) studied the relationship existing between peptization and loaf volume and obtained high negative correlations between the two, indicating that the larger the loaf volume the smaller would be the percentage of total protein peptized. MacLeod (1929) determined the percentage of total protein peptized by 0.5 N $MgSO_4$ and 0.5 N KBr. His data confirmed that of Gortner showing a significant negative correlation between loaf volume and total protein peptized.

Geddes (1930) found with increasing severity of heat treatment a progressive decrease in peptization of flour proteins with N $MgSO_4$ and N KI; this suggested a relationship between

peptization and degree of denaturation of the protein micelle. Moreover he found that the percentage of protein extractable was positively correlated with viscosity, indicating that the degree of protein peptization was dependent on colloidal properties. High positive correlations were obtained between loaf volume and percentage of protein peptized by 0.5 N KI.

Harris (1930) after a study of 44 flours of the 1929 crop peptized by 0.5 N K Br and $MgSO_4$ directed his attention towards the relationship between the non peptized fraction and loaf volume. He found that the correlation between non peptized protein and improver loaf volume was not significantly different from that found between total protein and improver loaf volume. The corresponding relationship between basic loaf volume and peptized protein was of even less practical importance.

In the same year, 1930, Geddes and Goulden made a comprehensive study of 102 straight grade flours experimentally milled from commercial samples of hard red spring wheat grown in Western Canada in 1928. This series represented wheat of low average quality, grading from No. 1 Northern to Feeds. The series was characterized by green and immature kernels resulting from improper germination and frost damage. Such a collection was used to determine whether flour proteins would exhibit differences in ease of peptization as a result of damaged kernels, and if so, whether these differences would show any relation to baking quality; also to ascertain by detailed statistical analysis whether the peptized or the non peptized

fraction is the more valuable portion in such a relationship. Their data show no difference between peptizability of flour proteins from sound wheat and that from immature and frost damaged. They found that with 0.5 N $MgSO_4$ the non peptized fraction is of more value than the peptized for estimating loaf volume, whereas with 0.5 N KI the peptized fraction is of greater worth. Thus they concluded that the relative value of the two fractions depends on the peptizing solution used. They explain their results on the basis of the Kent Jones Theory of Optimum Coagulation. The author of the theory himself states:--

> "It would appear that some weak flours such as English are too dispersed, (too much in the sol state), while others, e. g., Indians, are too coagulated. Strength then depends on the proteins being within certain limits of dispersion, and beyond these limits in either direction poor baking quality will result."

This theory suggests that the first protein fractions removed would consist of the more highly dispersed protein. Those salt solutions which show low peptizing powers, e. g., Na_2SO_4, would remove this part most readily. As this part of the protein has the least value in baking the residual or non peptized fraction would show a greater relationship to baking quality. On the other hand, such solutions as KI which possess a strong peptizing power, remove more and more of the more highly coagulated protein as the percentage of total protein increases. In cases of such strong solutions the peptized fraction shows the higher correlation. Moreover there will be a point reached in certain flours where the degree of coagulation is such that the peptized and the non peptized

fraction will be of equal value in relation to baking quality. Flour proteins in a too highly coagulated condition will show poor baking quality despite the high protein content.

Harris (1931, 1931_a, 1931_b, 1931_c, 1932) has done additional work on commercially milled wheat and flour. His data substantiate the interpretation of Weddes and Goulden. He claims that as much information relative to baking strength may be gained from a knowledge of the total and non peptized protein of the wheat as from similar knowledge regarding flour.

Sorensen (1930) discusses this whole problem of soluble proteins from a different angle. He considers that each protein system consists of a series of complexes which are feebly interlinked and readily interchangeable. The author himself writes:--

> "Within each complex all the atoms or atom groups are interlinked by main valencies, whereas the various complexes or components are reversibly interlinked by means of residual valencies. The linkage between the components is comparatively feeble, and of such a character that alterations in the composition of the solution (salt content, Hydrogen ion activity, alcohol content, temperature), can cause reversible dissociations of and exchange of components between the component systems present. When these alterations in the composition of the solution are so adapted as to render possible the formation in sufficient amount of a component system insoluble or sparingly soluble under the new conditions, this system will be formed and precipitated."

Whether these "reversible dissociations and exchanges of compounds between systems" have to do with degree of peptization is still a question. Sinclair and Gortner claim that they are identical phenomena. These workers (1933) extracted gliadin by different solvents and chemically analyzed the fract-

ions thus obtained to determine whether or not they were the same chemically. They found no increase in carboxyl groups of the proteins on treatment with different solvents, therefore no hydrolysis can take place during peptization. Moreover, the nitrogen distribution as determined by the Hausmann numbers of the various gliadin fractions was found to correspond to that of gliadin previously recorded by other investigators. They conclude that though the physical properties of the protein systems are explainable only on the basis of physical heterogeneity that Sorensen's "reversible - dissociable component systems of proteins," Svedberg's "decomposition of protein molecules," and their "protein peptization" are identical phenomena described in different words by three groups of workers.

The whole system of protein classification must then be considered as simply arbitrary. Sandstedt and Blish (1933) claim three main dissociable component systems in flour protein, each system in turn consisting of a group of lesser dissociable components or complexes. The purified fractions of gluten probably consist of a group of these complexes having similar properties. Preparations of constant properties are obtained only as long as the same method of extraction is used, for variation in method causes variation in properties of complexes due to their reversible dissociations as solubility, viscosity, susceptibility to denaturation, etc, are modified.

In an investigation of the hydration capacity

of flour proteins. Rich (1933) showed that ash content played a part in determining the capacity for hydration of these flours. Since hydration capacity, viscosity and peptizability all show a relationship to protein content, and since total ash was shown to have an effect on viscosity, Rich investigated a series of flours to determine whether any relationship existed between ash content and total protein. He concluded that "The percentage of wheat flour proteins peptized by a solution of a neutral salt of a strong acid and a strong base is very highly and positively correlated with the ash content of the sample."

Mangels (1934) found no consistent difference in peptizable protein in two groups of common wheats, one of which was of good, the other of poor baking quality. Wheats grown under different environmental conditions show as great variation as that found between the varieties.

From this brief review of the literature it would seem that the problem which has been approached from many angles is still far from being solved. Causes of variation in degree of peptization must be deep seated. That these are definitely associated with baking quality would seem questionable. Whether these variations are dependent on reversible dissociating complexes or on degree of coagulation, and also what is meant by degree of coagulation must still be answered. Might it not be possible to describe coagulation simply as a definite condition of these complexes and thus view the whole subject of peptization from a more extensive field?

EXPERIMENTAL.

The present work was undertaken originally to further test the Kent Jones Theory of Optimum Coagulation. A series of 59 frozen wheats which might be expected to show differences in the state of coagulation of the proteins was chosen. Straight grade flours experimentally milled from this series were peptized by four different inorganic salt solutions shown to have marked difference in ability to peptize. These were N Na_2SO_4, 0.5 N Ca Br_2, 0.5 N K Br and 0.5 N KI. As no work had been reported on peptizability of wheat at this time two of the peptizing solutions, N Na_2SO_4 and 0.5 N KI were used on the finely ground whole wheat meal as well. This enabled a comparison of the extent of peptization of the wheat and of the flour milled therefrom to be made. Before the completion of the work the data of Rich, showing high positive correlations between total ash and percentage peptized protein, were published. Therefore it was deemed advisable to carry on the work to investigate this relationship of ash to peptizable protein in the flours of this series. Since the fraction of the ash that would have any effect on the degree of peptization would be soluble ash this was determined as well. Moreover, soluble ash was added to the peptizing solutions to find out whether or not an increase in the amount of protein peptized would result.

(a) Relationship between protein of wheat peptized by 0.5 N KI and N Na_2SO_4, and loaf volume of the corresponding flours.

Before engaging in the work of extracting the wheat

by the above solvents it was necessary to determine the effect of the size of the wheat particle on the degree of peptization. To determine this the wheat was ground in three ways: (1) In a Wiley mill using a sieve of round holes 1 m.m. in diameter, (2) in the same mill using a sieve of round holes 0.5 m.m. in diameter, and (3) in the small electric Lab. Con Co mill set at the finest adjustment that could be maintained without excessive heating of the mill.

Samples were weighed in duplicate, suspended in 50 c.c. of N Na_2SO_4 solution and shaken mechanically for 30 minutes. The flour residue was packed in the bottom of the tube by whirling in a centrifuge and the supernatant liquid removed. The process was repeated six times and the nitrogen content of the combination of the last three extractions determined by the Kjeldahl-Gunning Procedure. If the wheat is ground finely enough the first three extractions with the salt solution should remove the bulk of the peptizable protein and the last three should contain very little.

The peptized protein found in three last extractions of wheat ground in the Wiley mill, using 1 m.m. sieve was 0.33 per cent protein; of wheat ground in the Wiley mill using 0.5 m.m. sieve 0.34 per cent protein, and of wheat ground in the Lab. Con Co 0.28 per cent protein. Although the amount of protein peptized is the same within experimental error in each case the saving in time with the Lab. Con Co mill was a deciding factor in its favour. Wheat ground in this mill was stored in stoppered bottles until used. The wheat meal was then ex-

tracted using the peptizing method of MacLeod (1929). This method consists of a single extraction of 6 gm. of flour with 200 c.c. of the salt solution by agitation in a mechanical shaker for 2 hours. This is centrifuged and the protein in a 50 c.c. aliquot of the supernatant liquid determined by the Kjeldahl-Gunning Procedure. The results are summarized in Tables I and II.

From Table I it will be observed that 0.5 N KI extracts a much higher percentage of total wheat protein than N Na_2SO_4. On the average 0.5 N KI removed 6.5 gms. of protein whereas N Na_2SO_4 peptized only 2.6 gms. per 100 gm. of sample. In Table II the data for the various grades represented in this series are tabulated separately. This shows no significant variation in degree of peptization for the grades 3rd - No.6, the grades represented in this series.

The results were submitted to a statistical analysis which is summarized in Table III.

Protein peptized by 0.5 N KI is highly correlated with total protein and with non peptized protein. Also a high positive correlation between non peptized fraction of the wheat protein and loaf volume of flour milled from the wheat is shown. This value however, is lower than that found when total protein and loaf volume are correlated. Protein peptized by N Na_2SO_4 is not related to total protein nor to loaf volume. The non peptized fraction is highly correlated to both. In this case too, total protein shows a higher degree of correlation with loaf volume than does either the peptized or the non peptized fraction.

TABLE I

ANALYTICAL DATA ON GROUND WHEATS OF FROZEN WHEAT SERIES - 1930

Lab. No.	Grade	Weight per bus. in lbs.	Total Protein 13.5% Moisture Basis	Extractions calculated as percentage of sample: 0.5 N KI Peptised Protein	0.5 N KI Non Peptised Protein	N Na_2SO_4 Peptised Protein	N Na_2SO_4 Non Peptised Protein	Loaf Volume Bromate Method c.c.
302.30	3°	62.5	13.1	7.0	6.1	2.7	10.4	575
303		66.0	12.9	6.6	6.3	2.5	10.4	565
304		64.5	12.9	6.6	6.3	2.6	10.3	578
305		64.5	10.6	5.4	5.2	2.0	8.6	570
306		64.0	14.6	7.8	6.8	2.7	11.9	680
307		66.5	13.8	7.3	6.5	2.5	11.3	620
308		65.0	12.4	6.6	5.8	2.7	9.7	598
309		63.0	11.9	6.2	5.7	2.4	9.5	548
310		62.0	14.1	7.2	6.9	2.6	11.5	630
311	4°	64.0	13.0	6.8	6.2	2.7	10.3	632
312		64.0	11.8	6.4	5.4	2.6	9.2	585
313		65.0	12.4	6.6	5.8	2.3	10.1	615
314		66.0	12.4	6.6	5.8	2.4	10.0	600
315		64.0	12.8	6.9	5.9	2.5	10.3	595
316		64.5	12.7	6.7	6.0	2.7	10.0	602
317		65.0	13.4	6.9	6.5	2.5	10.9	578
318		65.0	10.8	5.3	5.5	2.0	8.8	572
319		64.0	12.3	6.1	6.2	2.3	10.0	620
320		65.0	11.6	5.0	6.6	2.1	9.5	618
321		64.0	10.7	5.1	5.6	2.2	8.5	592
322	No. 5	65.0	11.5	6.4	5.1	2.6	8.9	565
323		64.0	13.1	6.1	7.0	2.1	11.0	592
324		62.0	13.0	7.1	5.9	2.9	10.1	665
325		62.5	10.2	5.1	5.1	2.2	8.0	445
326		63.0	12.5	6.5	6.0	2.9	9.6	655
327		62.5	14.3	7.3	7.0	2.8	11.5	680
328		58.5	13.2	6.9	6.3	3.0	10.2	565
329		62.0	12.5	6.4	6.1	2.7	9.8	608

TABLE I (CONTD.)

Lab. No.	Grade	Weight per bus. in lbs.	Total Protein 13.5% Moisture Basis	Extractions calculated as percentage of sample				Loaf Volume Bromate Method c.c.
				0.5 N KI		.N Na_2SO_4		
				Peptized Protein	Non Peptized Protein	Peptized Protein	Non Peptized Protein	
330.30		63.5	12.2	6.2	6.0	2.5	9.7	580
331		63.5	12.9	6.3	6.6	2.5	10.4	632
332	No. 6	59.0	12.5	6.2	6.3	2.8	9.7	465
333		58.5	12.5	6.5	6.0	3.8	8.7	498
334		59.5	11.5	6.0	5.5	2.8	8.7	430
335		59.0	13.4	7.1	6.3	3.0	10.4	582
336		59.0	11.4	6.1	5.3	2.9	8.5	485
337		61.0	12.1	6.2	5.9	2.6	9.5	555
338		61.5	12.4	6.0	6.4	2.6	9.8	572
339		59.5	12.9	6.8	6.1	2.8	10.1	620
340		64.0	12.5	6.7	5.8	2.9	9.6	572
341		59.0	12.7	6.6	6.1	2.8	9.9	568
342	3°	66.0	11.4	6.2	5.2	2.5	8.9	582
343		66.0	12.5	6.0	6.5	2.3	10.2	612
344		66.0	12.7	6.9	5.8	2.7	10.0	645
345		65.0	12.5	7.1	5.4	2.7	9.8	425
346		65.5	12.0	6.2	5.8	2.6	9.4	588
347	4°	65.0	12.1	6.2	5.9	2.5	9.6	632
348		66.0	11.8	6.5	5.3	2.6	9.2	525
349		64.0	13.3	7.0	6.3	2.7	10.6	690
350		64.0	11.4	6.0	5.4	2.6	8.8	620
351		64.0	13.0	6.5	6.5	2.6	10.4	635
352		64.0	12.4	6.9	5.5	2.9	9.5	622
353		64.0	12.1	6.6	5.5	2.7	9.4	570
354		65.0	10.6	5.9	4.7	2.4	8.2	412
355		64.0	12.9	6.8	6.1	2.6	10.3	675
356		63.0	12.9	6.7	6.2	2.6	10.3	625
357		64.0	12.3	6.9	5.4	2.6	9.7	665
358		64.0	12.3	6.9	5.4	2.6	9.7	665
359		63.5	12.4	7.2	5.2	2.7	9.7	618
360		61.0	10.7	5.6	5.1	2.4	8.3	400

TABLE II

ANALYTICAL DATA ON GROUND WHEATS OF FROZEN WHEAT SERIES

SEPARATED FOR DIFFERENT GRADES

Lab. No.	Weight per bus. lbs.	Total Protein 13.5% Moisture Basis	Extraction Calculated as Percentage of Sample: Peptized Protein 0.5 N KI	Non Peptized Protein 0.5 N KI	Peptized Protein M Na_2SO_4	Non Peptized Protein M Na_2SO_4	Loaf Volume Bromate Method c.c.
No. 3 Northern							
302.30	62.5	13.1	7.0	6.1	2.7	10.4	575
303	66.0	12.9	6.6	6.3	2.5	10.4	565
304	64.5	12.9	6.6	6.3	2.7	10.2	578
305	64.5	10.6	5.4	5.2	2.0	8.6	570
306	64.0	14.6	7.8	6.8	2.7	11.9	680
307	66.5	13.8	7.3	6.5	2.5	11.3	620
308	65.0	12.4	6.6	5.8	2.7	9.7	598
309	63.0	11.9	6.2	5.7	2.4	9.5	548
310	62.0	14.1	7.2	6.9	2.7	11.4	630
342	66.0	11.4	6.2	5.2	2.5	8.9	582
343	66.0	12.5	6.0	6.5	2.3	10.2	612
344	66.0	12.7	6.9	5.8	2.7	10.0	645
345	65.0	12.5	7.1	5.4	2.7	9.8	425
346	65.5	12.0	6.2	5.8	2.6	9.4	588
Average		12.67	6.65	6.02	2.55	10.12	586.8

TABLE II (CONTD.)

ANALYTICAL DATA ON GROUND WHEATS OF FROZEN WHEAT SERIES

SEPARATED FOR DIFFERENT GRADES

Lab. No.	Weight per bus. lbs.	Total Protein 13.5% Moisture Basis	Extraction Calculated as Percentage of Sample: Peptized Protein 0.5 N KI	Non Peptized Protein 0.5 N KI	Peptized Protein N Na_2SO_4	Non Peptized Protein N Na_2SO_4	Loaf Volume Brometo Method c.c.
No. 4 Northern							
311.30	64.0	13.0	6.8	6.2	2.7	10.3	632
312	64.0	11.8	6.4	5.4	2.6	9.2	585
313	65.0	12.4	6.6	5.8	2.3	10.1	615
314	66.0	12.4	6.6	5.8	2.4	10.0	600
315	64.0	12.8	6.9	5.9	2.5	10.3	595
316	64.5	12.7	6.7	6.0	2.7	10.0	602
317	65.0	13.4	6.9	6.5	2.5	10.9	578
318	65.0	10.8	5.3	5.5	2.0	8.8	572
319	64.0	12.3	6.1	6.2	2.3	10.0	620
320	65.0	11.6	5.0	6.6	2.1	9.5	618
321	64.0	10.7	5.1	5.6	2.2	8.5	592
347	65.0	12.1	6.2	5.9	2.5	9.6	632
348	66.0	11.8	6.5	5.3	2.6	9.2	525
349	64.0	13.3	7.0	6.3	2.7	10.6	690
350	64.0	11.4	6.0	5.4	2.6	8.8	620
351	64.0	13.0	6.5	6.5	2.6	10.4	635
Average		12.21	6.29	5.93	2.45	9.96	606.9

TABLE II (CONTD.)

ANALYTICAL DATA ON GROUND WHEATS OF FROZEN WHEAT SERIES

SEPARATED FOR DIFFERENT GRADES

Lab. No.	Weight per bus. lbs.	Total Protein 13.5% Moisture Basis	Extraction Calculated as Percentage of Sample				Loaf Volume Brom te Method
			Peptized Protein 0.5 N KI	Non Peptized Protein 0.5 N KI	Peptized Protein N Na_2 SO_4	Non Peptized Protein N Na_2 SO_4	c.c.
No. 5 Northern							
322	65.0	11.5	6.4	5.1	2.6	8.9	565
323	64.0	13.1	6.1	7.0	2.1	11.0	592
324	62.0	13.0	7.1	5.9	2.9	10.1	665
325	62.5	10.2	5.1	5.1	2.2	8.0	445
326	63.0	12.5	6.5	6.0	2.9	9.6	655
327	62.5	14.3	7.3	7.0	2.8	11.5	680
328	58.5	13.2	6.9	6.3	3.0	10.2	565
329	63.0	12.5	6.4	6.1	2.7	9.8	608
330	63.5	12.2	6.2	6.0	2.5	9.7	580
331	63.5	12.9	6.3	6.6	2.5	10.4	632
352	64.0	12.4	6.9	5.5	2.9	9.5	622
353	64.0	12.1	6.6	5.5	2.7	9.4	570
354	65.0	10.6	5.9	4.1	2.4	8.2	412
355	64.0	12.9	6.8	6.1	2.7	10.2	675
356	63.0	12.9	6.7	6.2	2.6	10.3	625
Average		12.42	6.48	5.94	2.6	10.0	592.7

TABLE II (CONTD.)

ANALYTICAL DATA ON GROUND WHEATS OF FROZEN WHEAT SERIES

SEPARATED FOR DIFFERENT GRADES

Lab. No.	Weight per bus. lbs.	Total Protein 13.5% Moisture Basis	Extraction Calculated as Percentage of Sample				Loaf Volume Brometo Method c.c.
			Peptized Protein	Non Peptized Protein	Peptized Protein	Non Peptized Protein	
			0.5 N KI		N Na_2 SO_4		
No. 6 Northern							
332	59.0	12.5	6.2	6.3	2.8	9.7	465
333	58.5	12.5	6.5	6.0	3.8	8.7	498
334	59.5	11.5	6.0	5.5	2.8	8.7	430
335	59.0	13.4	7.1	6.3	3.0	10.4	582
336	59.0	11.4	6.1	5.3	2.9	8.5	495
337	61.0	12.1	6.2	5.9	2.6	9.5	555
338	61.5	12.4	6.0	6.4	2.6	9.8	572
339	59.5	12.9	6.8	6.1	2.9	10.0	620
340	64.0	12.5	6.7	5.8	2.9	9.6	572
341	59.0	12.7	6.6	6.1	2.8	9.9	568
357	63.0	12.2	6.6	5.6	2.6	9.6	538
358	64.0	12.3	6.9	5.4	2.6	9.7	665
359	63.5	12.4	7.2	5.2	2.7	9.7	618
360	61.0	10.7	5.6	5.1	2.4	8.3	400
Average		12.25	6.46	5.78	2.81	9.43	540.5

TABLE III

STATISTICAL ANALYSIS OF DATA ON GROUND WHEAT.

	Means	Standard Deviations	Total Correlation Coefficients	Partial Correlations
a	12.38	.895	rab_1 + .3974	r b_1 g.c_1 = .0517
b_1	2.60	.276	rac_1 + .8936	r c_1 g.b_1 = .5189
c_1	9.80	.797	rag + .5709	
g	582.81	66.198	rb_1g - .012	
			rc_1g + .5172	
			rb_1c_1 + .0622	
b_2	6.46	.568		
c_2	5.92	.518		
g	582.81	66.198	rab_2 + .8413	r b_2 g.c_2 = .3013
			rac_2 + .8047	r c_2 g.b_2 = .4356
			rb_2g + .4254	
			rc_2g + .5197	
			rb_2c_2 + .3555	

Note:--

a = Per cent total protein in flour.

b_1, b_2 = Per cent peptized protein by N Na_2SO_4 and 0.5 N KI respectively.

c_1, c_2 = Per cent non peptized protein by N Na_2SO_4 and 0.5 N KI respectively.

g = Loaf Volume by Bromate Method.

TABLE III

TEST OF SIGNIFICANCE OF PARTIAL CORRELATIONS BY Z VALUE.

	Sum of squares	Degrees of freedom	Mean square	1/2 log e	Z	5 % point
$r_{b_1\ g.c} = .0517$						
$r^2\ b_1\ g.c_1 =$	.0027	2	.0013	.1312	-1.3084	1.484
$1-r^2\ b_1 g.c_1 =$	.9973	56	.0178	1.4396		
$r_{c_1\ g.b_1} = .5189$						
$r^2\ c_1\ g.b_1 =$	.2692	2	.1346	1.2998	1.1686	.5960
$1-r^2\ c_1 g b_1 =$	.7308	56	.0130	.1312		
$r_{b_2\ g.c_2} = .3013$						
$r^2 b_2 g.c_2 =$	.0908	2	.0454	.7565	.5153	.5960
$1-r^2 b_2\ g.c_2 =$	.9092	56	.0162	.2412		
$r_{c_2\ g.b_2} = .4356$						
$r^2 c_2\ g.b_2 =$	.1897	2	.0948	1.1016	.9185	.5960
$1-r^2 C_2\ g.b_2 =$	.8103	56	.0145	.1856		

The simple correlation coefficient rbg, is affected in part by (c), the non peptized protein, similarly rcg is partially dependent on (b) the peptized fraction. For that reason partial correlations rcg, b and rbg, c were determined and compared. If c is the more valuable fraction in relation to loaf volume, g, we would expect rcg.b to be greater than rbg.c. Such is obviously the case for peptization with both 0.5 N KI and N Na_2SO_4. We conclude therefore that with these solutions the non peptized fraction of the wheat protein is of greater value as an estimate of baking quality than is the peptized protein.

(b) Relationship between protein of flour peptized by Inorganic Salt Solutions and Loaf Volume.

Table IV gives a summary of data accumulated on the flours milled from the wheats discussed previously. Peptizability of the flour proteins was studied using the four inorganic salt solutions, 0.5 N KI, N Ca Br_2, 0.5 N KBr and N Na_2SO_4.

The results show that as in the instance of the wheat, there is a great variability in the amount of protein that can be extracted by the different inorganic salt solutions. With flour as with wheat 0.5 N KI extracts the largest amount of the total protein, removing on the average 6.2 gms. per 100 gms. sample; N Ca Br_2 averaging 5.1 gms. and 0.5 N K Br averaging 3.9 gms. are intermediate in peptizing ability. The difference in peptizing effect of the anions I and Br is shown. This confirms the work of Gortner (1929). It is not feasible to make a comparison between the effect of the cations Ca and K for different strength solutions were used in the two cases.

A statistical analysis of the results is given in Tables V and VI. The correlation coefficients between total protein and the non peptized fraction with all four solutions is higher than that found between total protein and the peptized fraction. With 0.5 N KI, N Ca Br_2 and 0.5 N K Br peptized protein is more highly correlated with loaf volume than is non peptized. With N Na_2SO_4 the opposite is the case. The value for the partial correlations

show that the protein peptized by 0.5 N KI, N Ca Br_2 and 0.5 N K Br is significantly correlated with loaf volume. With N Na_2SO_4 it is the non peptized fraction that shows such a relationship. The peptized and non peptized fractions with N Ca Br_2 seem of equal value in relation to loaf volume.

These data support the theory of optimum coagulation. Strong peptizing agents like KI and K Br remove much of the protein, the amount removed depending on the total amount present. Both the total protein and the peptized protein in these instances are correlated with loaf volume. Solutions of low peptizing power remove only a very small percentage of the total protein. This would appear to consist mainly of the more highly dispersed fraction which is presumably of lower value in baking. The non peptized fraction would then contain the more highly coagulated protein and it is this that shows a high correlation with baking strength. This theory presupposes some point where the two protein fractions would be of equal value. With Ca Br_2 such a point of equality is reached. It is obvious from the above data that the value of the two fractions in predicting baking strength is entirely dependent on the peptizing solution used. It is evident too, that the amount of peptized protein is of less value than total protein in predicting baking quality.

TABLE IV

ANALYTICAL DATA ON FLOURS MILLED FROM FROZEN WHEAT SERIES

Extractions calculated as per cent of sample

Lab. No.	Total Protein 13.5% Moisture Basis	0.5 N KI		N $CaBr_2$		0.5 N KBr		N Na_2SO_4		Loaf Volume Bromate Method c.c.
		Peptized Protein	Non Peptized Protein	Peptized Protein	Non Peptized Protein	Peptized Protein	Non Peptized Protein	Peptized Protein	Non Peptized Protein	
302.30	12.2	6.2	6.0	5.7	6.5	4.0	8.2	1.6	10.6	575
303	12.0	6.4	5.6	5.5	6.5	3.7	8.3	1.4	10.6	565
304	12.1	6.2	5.9	5.4	6.7	4.5	7.6	1.6	10.5	578
305	9.8	5.7	4.1	4.6	5.2	3.5	6.3	1.4	8.4	570
306	13.8	7.3	6.5	6.1	7.7	4.3	9.5	1.6	12.2	680
307	13.2	7.4	5.8	5.9	7.3	4.2	9.0	1.5	11.6	620
308	11.7	6.1	5.6	5.4	6.3	4.0	7.7	1.5	10.2	598
309	11.0	5.7	5.3	4.9	6.1	3.7	7.3	1.4	9.6	548
310	13.4	7.0	6.4	5.8	7.6	4.3	9.1	1.5	11.9	630
311	12.3	6.4	5.9	5.4	6.9	4.0	8.3	1.5	10.8	632
312	11.1	6.1	5.0	5.0	6.1	3.7	7.4	1.5	9.6	585
313	11.7	6.3	5.4	5.3	6.4	3.9	7.8	1.5	10.2	615
314	11.7	6.2	5.5	5.2	6.5	3.9	7.8	1.5	10.2	600
315	11.9	6.4	5.5	5.3	6.6	4.0	7.9	1.5	10.4	595
316	12.2	6.7	5.5	5.5	6.7	4.0	8.2	1.7	10.5	600
317	12.7	6.7	6.0	5.5	7.2	4.1	8.6	1.6	11.1	578
318	9.9	5.7	4.2	4.2	5.7	3.4	6.5	1.4	8.5	572
319	11.5	6.1	5.4	5.0	6.5	3.9	7.6	1.5	10.0	620
320	10.8	5.4	5.4	4.8	6.0	3.7	7.1	1.6	9.2	618
321	9.9	5.7	4.2	4.6	5.3	3.7	6.2	1.6	8.3	592
322	10.8	5.9	4.8	6.0	3.7	7.1	1.1	1.6	9.2	565
323	12.0	6.3	5.7	5.3	6.7	3.9	8.1	1.5	10.5	592
324	12.5	6.7	5.8	5.4	7.1	4.1	8.4	1.8	10.7	665
325	9.4	5.1	4.3	4.1	5.3	3.3	6.1	1.5	7.9	445
326	11.4	6.0	5.4	4.8	6.6	4.3	7.1	1.8	9.6	655
327	13.8	7.3	6.5	5.9	7.9	4.4	9.4	1.7	12.1	680
328	12.6	6.5	6.1	5.3	7.3	4.1	8.5	1.4	11.2	565
329	11.8	6.2	5.6	5.3	6.5	4.0	7.8	1.7	10.1	608

TABLE IV (CONTD.)

Lab. No.	Total Protein 13.5% Moisture Basis	Pep-tized Protein	Non Pep-tized Protein	Pep-tized Protein	Non Pep-tized Protein	Pep-tized Protein	Non Pep-tized Protein	Pep-tized Protein	Non Pep-tized Protein	Loaf Volume Brome te Method c.c.
330.30	11.7	6.2	5.5	5.2	6.5	3.9	7.8	1.8	9.9	580
331	11.8	6.4	5.4	5.2	6.6	4.0	7.8	1.7	10.0	632
332	11.5	6.0	5.5	5.0	6.5	3.9	7.6	1.7	9.8	465
333	11.4	5.9	5.5	4.9	6.5	3.8	7.6	1.5	9.9	498
334	10.7	5.9	4.8	4.7	6.0	3.7	7.0	1.3	9.4	430
335	12.5	6.6	5.9	5.5	7.0	4.0	8.5	1.8	10.7	582
336	10.8	5.9	4.9	4.6	6.2	3.6	7.2	1.8	9.0	485
337	11.1	5.9	5.2	5.1	6.0	3.7	7.4	1.5	9.6	555
338	11.4	6.2	5.2	5.2	6.2	3.9	7.5	1.8	9.6	572
339	12.1	6.7	5.4	5.5	6.6	4.1	8.0	1.8	10.3	620
340	11.8	6.5	5.3	4.7	7.1	3.9	7.9	1.7	10.1	572
341	11.7	6.3	5.4	5.0	6.7	3.8	7.9	1.7	10.0	568
342	10.5	5.8	4.7	4.9	5.6	3.7	6.8	1.4	9.1	582
343	11.9	6.4	5.5	5.5	6.4	4.0	7.9	1.6	10.3	612
344	11.7	6.6	5.1	5.7	6.0	3.9	7.8	1.7	10.0	645
345	11.7	6.3	5.4	5.6	6.1	4.0	7.7	1.5	10.2	425
346	11.1	6.0	5.1	5.3	5.8	3.8	7.3	1.5	9.6	588
347	11.5	6.2	5.3	3.0	8.5	4.2	7.3	1.8	9.7	632
348	10.8	5.9	4.9	5.1	5.7	3.8	7.0	1.6	9.2	525
349	12.5	6.7	5.8	5.8	6.7	4.3	8.2	1.8	10.7	690
350	10.6	6.0	4.6	4.9	5.7	4.0	6.6	1.7	8.9	620
351	12.1	6.5	5.6	5.6	6.5	4.1	8.0	1.7	10.4	635
352	11.6	6.5	5.1	5.6	6.0	4.2	7.4	1.9	9.7	622
353	11.2	4.9	6.3	5.1	6.1	3.8	7.4	1.4	9.8	570
354	9.7	5.5	4.2	4.1	5.6	3.4	6.3	1.6	8.1	412
355	11.8	6.6	5.2	5.5	6.3	4.0	7.8	1.5	10.3	675
356	11.8	6.3	5.5	5.2	6.6	4.0	7.8	1.8	10.0	625
357	11.1	6.0	5.1	6.0	3.8	7.3	1.8	9.3	9.3	538
358	12.0	6.7	5.3	5.7	6.3	7.7	7.7	1.8	10.2	665
359	11.7	6.3	5.4	5.1	6.6	4.1	7.6	1.8	9.9	618
360	9.7	5.4	4.3	4.2	5.5	3.5	6.2	1.6	8.1	400

TABLE V.

STATISTICAL ANALYSIS OF ANALYTICAL DATA ON FLOUR.

	Means	Standard Deviations	Total Correlations Coefficients		Partial Correlation Coefficients
a	11.57	.932	r_{ag}	+ .5934	$r_{b_1g.c_1}$ = .3061
g	582.81	66.198	r_{ab_1}	+ .1956	
b_1	1.60	.151	r_{ac_1}	+ .9877	$r_{c_1g.b_1}$ = .5671
c_1	9.96	.914	r_{b_1g}	+ .2786	
			r_{c_1g}	+ .5564	
			$r_{b_1c_1}$	+ .0441	
b_2	3.92	.250	r_{ab_2}	+ .8304	$r_{b_2g.c_2}$ = .4481
c_2	7.64	.746	r_{ac_2}	+ .9934	
			r_{b_2g}	+ .6587	$r_{c_2g.b_2}$ = .1044
			r_{c_2g}	+ .5472	
			$r_{b_2c_2}$	+ .7521	
b_3	5.15	.526	r_{ab_3}	+ .7585	$r_{b_3g.c_3}$ = .4071
c_3	6.42	.635	r_{ac_3}	+ .8415	
			r_{b_3g}	+ .4790	$r_{c_3g.b_3}$ = .4025
			r_{c_3g}	+ .4754	
			$r_{b_3c_3}$	+ .2856	
b_4	6.22	.482	r_{ab_4}	+ .8854	$r_{b_4g.c_4}$ = .4385
c_4	5.35	.556	r_{ac_4}	+ .9130	
			r_{b_4g}	+ .6975	$r_{c_4g.b_4}$ = .1758
			r_{c_4g}	+ .4781	
			$r_{b_4c_4}$	+ .6139	

Note:-- (Over)

TABLE V

STATISTICAL ANALYSIS OF ANALYTICAL DATA ON FLOUR.

Explanatory Note:--

a = Per cent of total protein in flour.

b_1, b_2, b_3, b4 = Per cent of peptized protein by N Na_2SO_4, 0.5 N K Br, N Ca Br_2 0.5 N Ki respectively.

c_1, C_2, c_3, c_4 = Per cent non peptized protein by N Na_2SO_4, 0.5 N Br, N Ca Br_2, 0.5 N KI respectively.

g = Loaf Volume by Bromate method.

TABLE VI

SIGNIFICANCE OF PARTIAL CORRELATIONS LISTED IN TABLE V

	Sums of squares:	Degrees of freedom:	Mean squares	1/2 log e	Z	5 % point
$rb_1g.c_1$ =.3061						
$r^2b_1g.c_1$ =	.0937	2	.0468	.7716	.5304	.5960
$1-r^2b_1g.c_1$=	.9063	56	.0162	.2412		
$rc_1g.b_1$ =.5671						
$r^2c_1g.b_1$=	.3216	2	.1608	1.3887	1.2934	.5960
$1-r^2c_1g.b_1$ =	.6784	56	.0121	.0953		
$rb_2g.c_2$ =.4481						
$r^2b_2g.c_2$ =	.2008	2	.1004	1.1532	.9744	.5960
$1-r^2b_2g.c_2$ =	.7992	56	.0143	.1788		
$rc_2g.b_2$ =.1044						
$r^2c_2g.b_2$ =	.0109	2	.0054	.8432	.5936	.5960
$1-r^2c_2g.b_2$ =	.9891	56	.0177	1.4368		
$rb_3g.c_3$ =.4071						
$r^2b_3g.c_3$ =	.1657	2	.0828	1.0569	.8575	.5960
$1-r^2b_3g.c_3$ =	.8343	56	.0149	.1994		
$rc_3g.b_3$ =.4025						
$r^2c_3g.b_3$ =	.1620	2	.0810	1.0460	.8433	.5960
$1-r^2c_3g.b_3$ =	.8380	56	.0150	.2027		

TABLE VI

SIGNIFICANCE OF PARTIAL CORRELATIONS LISTED IN TABLE V

	Sum of squares	Degrees of freedom	Mean squares	1/2 log e	z	5 % point
$r_{b_4 g \cdot c_4} = .4385$						
$r^2_{b_4 g \cdot c_4}$ =	.1923	2	.0962	1.1320	.9496	.5960
$1-r^2_{b_4 g \cdot c_4}$ =	.8077	56	.0144	.1824		
$r_{c_4 g \cdot b_4} = .1758$						
$r^2_{c_4 g \cdot b_4}$ =	.0309	2	.0154	.2159	-.0581	1.484
$1-r^2_{c_4 g \cdot b_4}$ =	.9691	56	.0173	.2740		

(c) Comparison of extent of Peptization of Proteins of Wheat and of Flour milled therefrom by 0.5 N KI and N Na_2SO_4

The data for this comparison are summarized in Table VII. It will be observed that, on the average, the percentage protein extracted from the wheat by 0.5 N KI is lower than that extracted from the flour milled from it. The variation within the series is probably due to variation in initial protein for it is a well known fact that solubility in a definite volume of salt solvent is closely related to initial amount of protein present.

Peptization with N Na_2SO_4 shows a much greater difference in amount removed in favour of the wheat protein. These results suggest that the protein of the wheat is in the more highly dispersed state. In this condition it is extracted more readily by N Na_2SO_4 and evidently not so readily by 0.5 N KI. This necessitates a slightly different interpretation of the Kent Jones Theory of Optimum Coagulation. Geddes and Goulden (1930) on the basis of this theory, suggest that KI the "strong" peptizing agent removes both the highly dispersed and the more coagulated fractions of the protein. If this were true, the percentage of protein peptized should remain the same whether the degree of dispersion of the protein is increased or not. But the variation in the amount of protein extracted by the two solutions in the above instances suggests that as the protein is changed to the more highly dispersed state from the more coagulated, less is removed by KI. Therefore the so-called "strong" peptizing solutions are strong only in that

they peptize a larger amount of the protein which is evidently in a state that is not touched by the "weak" peptizing solutions. The change in state may be due to the loss of some constituent in the milling process which effects the degree of dispersion of the protein ,or it may be due simply to the removal of the protein and nitrogenous extractions of the bran and germ which are generally considered to be easily extracted. To prove which of these suggestions is true an investigation of the solubility of nitrogen compounds of bran and germ in the solutions would be required. This has been left for future work.

TABLE VII

COMPARISON OF EXTENT OF PEPTIZATION OF WHEAT AND CORRESPONDING FLOUR PROTEINS BY 0.5 N KI AND N Na_2SO_4.

All results calculated to 13.5 % moisture basis.

Lab. No.	Total protein: Wheat	Total protein: Flour	Per cent protein. Peptized by 0.5 N KI: Wheat	Per cent protein. Peptized by 0.5 N KI: Flour	Percent protein peptized by N Na_2SO_4: Wheat	Percent protein peptized by N Na_2SO_4: Flour
30...30	13.1	12.2	53.4	50.8	20.6	13.2
	12.9	12.0	51.2	53.3	19.4	11.7
	12.9	12.1	51.2	51.2	20.2	13.0
	10.6	9.8	50.9	58.2	18.9	14.1
	14.6	13.8	53.4	52.9	18.5	11.4
	13.8	13.2	52.9	56.1	18.1	11.4
	12.4	11.1	53.2	52.1	21.8	12.8
	11.9	11.0	52.1	51.8	20.2	12.7
	14.1	13.4	51.1	52.2	18.4	11.9
	13.0	12.3	52.3	52.0	20.8	11.8
	11.8	11.1	54.2	55.0	22.0	13.1
	12.4	11.7	53.2	53.8	18.5	12.4
	12.4	11.7	53.2	53.0	19.4	12.4
	12.8	11.9	53.9	53.8	19.5	12.7
	12.7	12.2	52.8	54.9	21.2	13.8
	13.4	12.7	51.5	52.8	18.6	12.2
	10.8	9.9	49.1	57.6	18.5	14.3
	12.3	11.5	49.6	53.0	18.7	12.7
	11.6	10.8	43.1	50.0	18.1	15.2
	10.7	9.9	47.7	57.6	20.6	16.6
	11.5	10.8	55.6	54.6	22.6	14.4
	13.1	12.0	46.6	52.5	15.6	12.8
	13.0	12.5	54.6	53.6	22.3	14.5
	10.2	9.4	50.0	54.2	21.6	16.2
	12.5	11.4	52.0	52.6	23.2	15.8
	14.3	13.8	51.0	57.9	19.6	12.3
	13.2	12.6	52.3	51.6	22.7	11.4
	12.5	11.8	51.2	52.5	21.6	14.2
	12.2	11.7	50.8	53.0	20.5	15.5
	12.9	11.8	48.8	54.2	19.4	14.2
	12.5	11.5	49.6	52.2	22.4	15.0
	12.5	11.4	52.0	51.8	30.4	12.9
	11.5	10.7	52.2	55.1	24.3	12.4
	13.4	12.5	53.0	52.8	22.4	14.4
	11.4	10.8	53.5	54.6	25.4	16.7
	12.1	11.1	51.2	53.2	21.5	13.7

TABLE VII

(Continued)

Lab. No.	Total Protein: Wheat	Total Protein: Flour	Percent Protein Peptized by 0.5 N KI: Wheat	Percent Protein Peptized by 0.5 N KI: Flour	Percent Protein Peptized by N Na_2SO_4: Wheat	Percent Protein Peptized by N Na_2SO_4: Flour
	12.4	11.4	48.4	54.4	21.0	15.4
	12.9	12.1	52.7	55.4	21.7	15.1
	12.5	11.8	53.6	55.1	23.2	14.3
	12.7	11.7	52.0	53.8	22.0	14.3
	11.4	10.5	54.4	55.2	21.9	13.6
	12.5	11.9	48.0	53.8	18.4	13.2
	12.7	11.7	54.3	56.4	21.2	14.2
	12.5	11.7	56.8	53.8	21.6	12.8
346.30	12.0	11.1	51.7	54.0	21.7	14.0
	12.1	11.5	51.2	53.9	20.7	15.9
	11.8	10.8	55.1	54.6	22.0	14.8
	13.3	12.5	52.6	53.6	20.3	14.4
	11.4	10.6	52.6	56.6	22.8	15.8
	13.0	12.1	50.0	53.7	20.0	14.1
	12.4	11.6	53.6	56.0	23.4	16.6
	12.1	11.2	54.5	43.8	22.3	12.7
	10.6	9.7	55.7	56.7	22.6	16.1
	12.9	11.8	52.7	55.9	20.2	12.9
	12.9	11.8	51.9	53.4	20.2	14.9
	12.2	11.1	54.1	54.0	21.3	15.9
	12.3	12.0	56.1	55.8	21.1	15.4
	12.4	11.7	58.1	53.8	21.8	15.1
	10.7	9.7	52.3	55.7	22.4	16.3
Average	12.4	11.6	52.0	53.8	21.0	14.0

(d) Relationship between ash content and peptized protein.

Total ash of this series of flours was determined and correlations between ash and peptized protein calculated. The results are given in Tables VIII and IX. Protein peptized by N Na_2SO_4 shows a high positive correlation with total ash, that peptized by N Ca Br_2 a significant negative value. Correlation coefficients between protein peptized by 0.5 N KI and by 0.5 N K Br and total ash are not significant. But the total correlation coefficient does not give us enough information regarding the relation between these two. It will be observed by reference to Table I that the amount of peptized protein varies with the total amount of protein present. In other words, as the total protein increases the relative proportions of peptized and non peptized protein vary. Therefore "b" is affected by "c", the non peptized fraction. If the multiple correlation coefficient R b. cx is compared with R bc (which is equal to rbc) we will discover whether or not total ash (x) is of importance in estimating peptized protein (b). If "x" is adding to our information Rb.cx will be significantly greater than rbc. These statistics are calculated for protein peptized by the four solutions and are presented in Table X.

Total ash is not of value in estimating flour protein peptized by either 0.5 N KI or 0.5 N K Br. The value rbc is not significantly different from that of Rb.cx in either case. Total ash does contribute much however, to our estimation of protein peptized by N Na_2SO_4. The variable "x" has greatly increased the value of the correlation coefficient in this

case. This is true also, of protein peptized by N Ca Br_2, although as pointed out above, the correlation between this peptized protein and total ash is negative in character.

Any direct effect on peptized protein must be due not to total ash but to the soluble fraction of the ash. Therefore a study of the water soluble ash of the series in its effect on peptization was made. The data are summarized in Tables IX and XI . The correlation coefficient between soluble ash and protein peptized by N Na_2SO_4 shows the only significant positive value. Between protein peptized by 0.5 N KI and soluble ash and that peptized by N Ca Br_2 and soluble ash the values are significant but negative. Protein peptized by 0.5 N K Br is not significantly correlated with soluble ash. As above, multiple correlation coefficients were calculated and compared with the simple. Rb. es is significantly greater than rbe in the case of protein peptized by 0.5 N KI, N Ca Br_2 and N Na_2SO_4.

The negative values of the simple correlation coefficients show that as the soluble ash content of the flour increases the amount of protein extracted by the "strong" peptizing solutions KI and Ca Br_2 decreases. With Na_2SO_4 a "weak" peptizing solution, protein extracted increases with an increase in soluble ash content. The significant values for the difference between the multiple and simple correlation coefficients in the above cases show that soluble ash is contributing additional information regarding the peptized protein fraction in all three cases.

TABLE VIII

SUMMARY OF RESULTS ON ASH AND SOLUBLE ASH OF FROZEN WHEAT SERIES.

All Results Calculated to 13.5 per cent Moisture Basis.

Lab No.	Per cent Total Ash	Per cent Soluble Ash	Lab No.	Per cent Total Ash	Per cent Soluble Ash	Lab No.	Per cent Total Ash	Per cent Soluble Ash
302	.48	.30	322	.55	.48	341	.64	.53
303	.42	.30	323	.46	.45	342	.45	.38
305	.42	.36	324	.55	.44	343	.48	.40
306	.41	.31	325	.58	.54	344	.51	.40
307	.42	.12	326	.52	.42	345	.54	.36
308	.44	.20	327	.50	.40	346	.52	.37
309	.46	.37	328	.55	.52	347	.56	.38
310	.46	.37	329	.56	.44	348	.51	.42
311	.45	.37	330	.52	.44	349	.53	.44
312	.46	.41	331	.52	.46	350	.52	.48
313	.45	.35	332	.64	.45	351	.52	.38
314	.41	.33	333	.61	.52	352	.56	.53
315	.47	.37	334	.70	.55	353	.47	.37
316	.54	.43	335	.59	.44	354	.52	.36
317	.54	.39	336	.72	.63	355	.52	.44
318	.49	.34	337	.58	.52	356	.54	.42
319	.44	.35	338	.58	.42	357	.55	.38
320	.50	.38	339	.61	.52	358	.60	.55
						359	.60	.54
321	.53	.48	340	.61	.56	360	.62	.61

TABLE IX

STATISTICAL ANALYSIS OF DATA ON TOTAL ASH, SOLUBLE ASH AND PEPTIZED PROTEIN.

	Means	Standard Deviations	Total Correlation Coefficients	t.value	P	
x	.116	.069				
s	.122	.091				
b_1	.605	.153	rb_1x = +.4782	4.075	.01	Highly significant
			rb_1s = +.3454	2.7546	.01	Highly significant
b_2	.912	.240	rb_2x = -.1191	.8972	.4	Not significant
			rb_2s = +.1781	1.3545	.2	Not significant
b_3	.641	.529	rb_3x = -.2910	2.276	.05	Significant
			rb_3s = -.2899	2.267	.05	significant
b_4	.717	.486	rb_4x = -.1428	.1080	.4	Not significant
			rb_4s = -.2227	1.7091	.1	significant

b_1, b_2, b_3, b_4 = Per cent of peptized protein by N Na_2SO_4, 0.5 N K Br, N Ca Br_2, and 0.5 N KI

X = Total Ash

S = Soluble Ash.

TABLE X.

MULTIPLE AND TOTAL CORRELATION COEFFICIENTS OBTAINED FROM TOTAL ASH, PEPTIZED AND NON PEPTIZED PROTEIN OF FLOUR.

	PEPTIZING SOLVENTS.			
	0.5 N KI	N $CaBr_2$	0.5 N K Br	N Na_2SO_4
rbx	- .1428	- .2916	- .1205	+ .4782
rbx.c	- .0443	- .4278	+ .1016	+ .5123
Rb.cx	+ .6149	+ .4991	+ .7508	+ .5185
rbc	+ .6139	+ .2845	+ .7479	+ .0441

b = Peptized protein, c = Non peptized protein, x = total ash.

TESTS OF SIGNIFICANCE OF ADDITIONAL INFORMATION OBTAINED BY CALCULATING MULTIPLE CORRELATION COEFFICIENTS.

PEPTIZING SOLUTION 0.5 N KI

	Sum of squares	Degrees of freedom	Mean squares	1/2 log e	Z	5 % point
$1 - r^2bc$	.6231					
$1 - R^2b.cx$	.6219	55	.01131	.06155		
Difference	.0112	1	.0112	.05756	-.0399	2.7588

PEPTIZING SOLUTION N $CaBr_2$

$1 - r^2bc$	.9191					
$1 - R^2b.cx$	.7509	55	.01365	.1556		
Difference	.1682	1	.1682	1.4113	1.2557	.6933

TABLE X

TESTS OF SIGNIFICANCE OF ADDITIONAL INFORMATION OBTAINED BY CALCULATING MULTIPLE CORRELATION COEFFICIENTS.

PEPTIZING SOLUTION 0.5 N K Br.

	Sum of Squares	Degrees of Freedom	Mean squares	1/2 log e	Z	5% point
$1 - r^2bc$	.4407					
$1 - R^2b.cx$	.4362	55	.0079	1.0034	.2814	2.7588
Difference	.0045	1	.0045	.7520		
PEPTIZING SOLUTION	N Na_2SO_4					
$1 - r^2bc$	.9981					
$1 - R^2b.cx$	.7312	55	.0133	.1422		
Difference	.2669	1	.2669	1.6422	1.5000	.6933

TABLE XI

MULTIPLE AND TOTAL CORRELATION COEFFICIENTS OBTAINED FROM SOLUBLE ASH, PEPTIZED AND NON PEPTIZED PROTEIN OF FLOUR

	PEPTIZING SOLVENTS			
	0.5 N KI	N $CaBr_2$	0.5 N K Br	N Na_2SO_4
rbs	- .2227	- .2917	- .1789	+ .3466
rbs.c	+ .5936	+ .3672	+ .0870	+ .3850
Rb.cs	+ .7723	+ .4525	+ .7500	+ .3871
rbc	+ .6139	+ .2845	+ .7479	+ .0441

b = Peptized protein, c = Non peptized protein, s = soluble ash

TABLE XI

TESTS OF SIGNIFICANCE OF ADDITIONAL INFORMATION OBTAINED BY CALCULATING MULTIPLE CORRELATION COEFFICIENTS.

	Sum of Squares	Degrees of Freedom	Mean squares	1/2 log e	z	5% point
PEPTIZING SOLUTION 0.5 N KI						
$1 - r^2_{bc}$	.6231					
$1 - R^2_{b.cs}$	.4036	55	.0073	.9668		
Difference	.2196	1	.2196	2.6958	1.7291	.6933
PEPTIZING SOLUTION N Ca Br_2						
$1 - r^2_{bc}$	.9191					
$1 - R^2_{b.cs}$	.7952	55	.0144	.1823		
Difference	.1239	1	.1239	1.2469	1.0646	.6933
PEPTIZING SOLUTION 0.5 N K Br						
$1 - r^2_{bc}$	.4407					
$1 - R^2_{b.cs}$	.4374	55	.0019	.3209		
Difference	.0033	1	.0033	.5969	.2760	.6933
PEPTIZING SOLUTION N Na_2SO_4						
$1 - r^2_{bc}$	.9981					
$1 - R^2_{b.cs}$	.8502	55	.0155	.2178		
Difference	.1479	1	.1479	1.3469	1.1291	.6933

It was necessary then to find out whether soluble ash affects degree of peptization by simply increasing the electrolyte content of the peptizing solution, or whether the effect is indirect and due to some fundamental factor to which both the dispersion of the protein and the ash content are related. To determine this, soluble ash from the flour was used in the peptizing solutions 0.5 N KI and N Na_2SO_4 and peptized protein determined as above. As there was not sufficient flour of the original series remaining for this work, ten new flours ranging in protein content from 9.8 % - 15.6 % were chosen. Water soluble ash from 10 gm. of the flour was added to the salt and sufficient water added to make the solution the required strength. Six gm. of the same flour were then peptized by 200 c.c. of the solution.

The results from this study are reported in Table XII. It is evident that in the majority of cases a slightly larger proportion of the protein is removed when soluble ash extract is included in the 0.5 N KI peptizing solution, due evidently to the higher concentration of electrolyte. But this increase in concentration cannot be a determining factor for the degree of peptization of the protein, for, if such were the case, a positive correlation between protein peptized by KI and soluble ash content would result. As we have seen before this correlation is negative. On the other hand, there is no change in the peptizability of the protein when soluble ash is added to Na_2SO_4 solution. Therefore it appears that ash in itself is of too low a concentration to have any effect on

the amount of protein extracted in this instance, yet a high positive correlation between soluble ash and protein peptized by N Na_2SO_4 was obtained previously. Therefore change in electrolyte content cannot be the direct factor that s causing this variability in peptization of the protein by these salt solutions.

It has been observed previously that 0.5 N KI shows less ability to extract protein from wheat than from flour milled from it, whereas N Na_2SO_4 peptizes a much higher percentage of wheat protein than it does of the flour. Between protein peptized by KI and soluble ash content a significant negative correlation is found, between that peptized by Na_2SO_4 and soluble ash the correlation is positive and significant. Moreover it has been suggested that KI extracts more of the coagulated fraction of the protein particle and that Na_2SO_4 removes only the more dispersed. Therefore, it would appear that protein of flour of high ash content and of wheat, which also is high in ash is in a more highly dispersed state than that of flour of low ash. This highly dispersed protein is more readily extracted by so-called "weak" peptizing solutions like Na_2SO_4 and somewhat less readily removed by "Strong" solutions such as KI and $Ca\ Br_2$.

From the theoretical standpoint the effect might be explained using Sorensen's terminology, "to alterations in the composition of the solution which h ave caused reversible dissociations of, and exchange of components beteen the com-

ponent systems present resulting in a modification of both physical and chemical properties". Whether this condition is directly dependent on the ash content as it effects salt concentration or hydrogen ion activity, or whether both ash and degree of dispersion of protein are related to some other constituent which is the determining factor is still a question. In any case, the terms "Strong" and "Weak" used to describe peptizing ability of a salt solution would appear to be erroneous, for the power a solution possesses of extracting protein would seem to depend only on the state of the protein particle. As this state changes, degree of extraction by any individual peptizing solution is altered.

TABLE XII

EFFECT OF INCLUSION OF SOLUBLE ASH EXTRACTED FROM 10 GM. SAMPLE OF FLOUR IN PEPTIZING SOLUTION ON THE AMOUNT OF PROTEIN PEPTIZED.

All results expressed on 13.5 per cent moisture basis.

Flour Number	1	2	3	4	5	6	7	8	9	10
Total Ash Per cent	.64	.39	.41	.41	.53	.41	.41	.59	.63	.38
Soluble Ash Per cent	.40	.28	.32	.30	.34	.28	.30	.46	.41	.26
Protein Per cent	15.60	9.90	10.80	10.20	11.90	11.20	9.80	10.6	13.90	12.00
Protein peptized by 0.5 N KI including soluble ash	8.08	5.11	5.60	5.30	6.07	6.17	5.54	5.35	7.29	6.63
0.5 N KI alone	8.00	4.76	5.40	5.00	6.07	6.00	4.85	4.70	6.70	6.17
N Na_2SO_4 including soluble ash	1.76	1.07	1.09	1.33	1.68	1.41	1.50	1.32	1.92	1.75
N Na_2SO_4 alone	1.79	1.10	1.09	1.47	1.81	1.33	1.40	1.40	1.81	1.38

SUMMARY AND CONCLUSIONS

Fifty-nine samples of wheat varying in grade from 3° to No. 6 which had been damaged by frost, were subjected to peptization by inorganic salt solutions before and after milling. The non peptized fraction of the wheat protein using N Na_2SO_4 and 0.5 N KI is highly correlated with baking strength as determined by loaf volume of the flour milled from this wheat.

The flour proteins showed a great variation in the amount of protein peptized, 0.5 N KI showing the greatest ability to extract protein, followed by N Ca Br_2, 0.5 N K Br, and N Na_2SO_4 in the order named. The findings support the Kent Jones Theory of Optimum Coagulation. Protein peptized by "strong" peptizing agents like KI and K Br is highly correlated with loaf volume. Protein extracted by "weak" peptizing agents like N Na_2SO_4, which only removes the highly dispersed protein of low value as far as Baking Strength is concerned, shows no correlation with loaf volume. Rather the non peptized fraction shows such a relationship. The peptized and the non peptized fraction of protein extracted with N Ca Br_2 are of equal value in predicting Baking Strength. The value of the two fractions in predicting strength then is entirely dependent on the peptizing solution used. A survey of the data shows a big difference in value of the two fractions.

A higher percentage of wheat protein is extracted by N Na_2SO_4 than of flour protein. 0.5 N KI extracts slightly less of the wheat protein than of the flour. Therefore it

would appear probable that the proteins of the wheat are in a more highly dispersed state than are the proteins in the flours milled from the wheat. Some constituent present in the wheat to a greater extent than in the flour is affecting the reversible dissociations of the component systems present in such a way that the protein is more readily peptized by $Na_2 SO_4$.

This alteration may be due to ash. Therefore, total ash and soluble ash content were correlated with peptized protein. When protein is peptized by KI total ash shows no relationship with peptized protein, but soluble ash is negatively correlated with the peptized fraction. When peptized by N Na_2SO_4 total ash and soluble ash are both positively correlated with peptized protein. On another series of flour soluble ash was extracted and added to the salt in making up the solution for peptizing. No increase in peptized protein resulted when ash was included in Na_2SO_4 solution; a slight increase in the amount extracted occurred when it was added to KI.

It is concluded that:-

Amount of protein peptized by any one solution is of less value in estimating baking strength than is total protein.

The protein of the wheat is in a more highly dispersed state than is the protein of the flour milled therefrom.

The relationship between soluble ash content and peptized protein is not simply due to an increase in electrolyte content of the peptizing solution.

Ash content of wheat or flour or some factor to which it is related affects an exchange between the various complexes present, such that the resulting system is more readily extracted by the weak peptizing solutions and less readily removed by the strong.

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