

ENZYMATIC EVALUATION OF CHANGES IN PROCESSED GRAIN

by

An-chein Sung

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Major Professor

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TABLE OF CONTENTS

	Page
INTRODUCTION	1
REVIEW OF LITERATURE	2
Starch	2
Fractions of Starch	2
Gelatinization of Starch	3
Retrogradation of Starch	5
Chemical Gelatinization	6
Heat-Moisture Treated Starch	7
Processed Grain	8
MATERIALS AND METHODS	10
RESULTS	14
DISCUSSION	27
SUMMARY	29
ACKNOWLEDGMENTS	30
BIBLIOGRAPHY	31

INTRODUCTION

Lately, a number of studies have indicated that processed grain in high concentrate rations improves feed utilization (5, 8, 22, 29). Therefore, processing grain has received more attention and is becoming more and more important. Grain processing has taken many forms, from simple grinding to complex high-pressure, moist-heat treatments. The effects of various grain treatments on improving feed efficiency are different. A simple and economical test method to evaluate processing effects is needed.

It is well known that starch is the major constituent of cereal grain. Any change in the starch due to processing should therefore play an important role in the utilization of the grain. Hence, a method quantitatively measuring the changes in the starch could be used to detect effects of treatment on the grain. Instead of a feeding trial, a simple and rapid laboratory procedure might thus be adapted for determining the improvement in efficiency of utilization of processed grain.

The objective of this study was to evaluate a procedure for determining the degree of change in the starch of the grain due to the effects of pressure, moisture, duration of treatment and urea following moist-heat treatment. Corn and sorghum grain were chosen as test materials, because they are the most dominant feed grains. Urea was included in the study because it is commonly used as a source of non-protein nitrogen for ruminants (27). Other studies have also indicated that grain-urea mixtures processed under certain conditions moisture, pressure and time are better than unprocessed material as regards the utilization of non-protein nitrogen.

REVIEW OF LITERATURE

Starch

Starch is the principal source of carbohydrate available for animals. It occurs in leaves, seeds, roots, fruits, tubers and other parts of most green plants. The physical appearance and properties of starch granules from different sources show a great variety. The size of starch granules varies widely from one type to another (14). Because of the great variation in physical properties of starch from different species and the differences in chemical constitution, different starches can be distinguished by the size and shape of the granules, the temperatures at which they gelatinize in water, the degree of isotropism evident when viewed on the polarizing microscope and the extent to which they combine with iodine (33).

The structure of starch granules as reported by Leach (12) shows them to be composed of linear and branched molecules associated by hydrogen bonding either directly or through water bridges to form radially oriented micelles or crystalline areas of various degrees of order. An interconnected three-dimensional micellar lattice is considered to be formed by the participation of segments of individual molecules in several micellar areas. The over-all strength of the micellar network depends on the degree of association and the molecular arrangement.

Fractions of Starch

Ordinary starches are a mixture of high polymeric molecules. Amyloses with structures of linear chains, are composed of α -D-glucopyranose units, linked by α -D-(1-4) bonds. Amylopectins are branched molecules

in which the branches have the same structure as the linear molecules but contain α -D-(1-6) bonds at the points where branching occurs (32). Some starches possess nearly identical ratios of amylose to amylopectin. The most dominant composition is 22-26% amylose and 74-78% amylopectin (35). Waxy varieties of corn, rice, sorghum and barley may contain only amylopectin or up to 6% amylose, depending on genetic factors. There are also other types of starches such as those from the lily, certain peas and sugar corn which contains two or more times the usual amount of amylose (34).

The linear, amylose fraction of starch shows a strong tendency to crystallize. The rather long uniform branches of the amylopectin fraction can also crystallize. The crystalline nature of the fractions is observed in all natural starch granules, even in those composed only of amylopectin molecules and known as waxy starches (32).

Amylose has a lower average molecular weight than amylopectin, and absorbs iodine to form a deep blue starch-iodine complex. It has a high digestibility with beta-amylase and retrogrades rapidly. In contrast, amylopectin does not develop a blue color with iodine and has much less tendency to retrograde and crystallize. The special properties of these two fractions are very useful in chemical analysis and industrial processes.

Gelatinization of Starch

When water suspensions of starch are heated above certain critical temperatures the starch granules start to swell and simultaneously lose their birefringence properties (i.e. polarization cross). This phenomenon has been referred to as "gelatinization" (12, 14). It is believed that heat weakens the intermicellar network within the starch granule by

disrupting hydrogen bonds, permitting further hydration and eventually irreversible granule swelling which results in "gelatinization of starch" (9).

Kerr suggested (11) that gelatinization of starch granules during heating in water has three distinct phases. He stated water was first slowly and reversibly taken up and limited swelling occurred. The viscosity of the suspension did not increase noticeably and the granule retained a characteristic appearance and birefringence. In the second stage the granule swelled rapidly, viscosity increased and there was a loss of birefringence. The third phase of swelling, which took place with increasing temperature resulted in granules becoming formless sacs, and the more soluble part of the starch leached out.

Since gelatinization of starch does not occur instantly for all granules in a sample, the gelatinization temperature of a starch is defined by the temperature range over which birefringence is progressively lost by the granules or by the temperature at which all granules have lost all birefringence (14). Starch gelatinization temperatures vary from one species to another. Gelatinization generally occurs within a range of 7° to 10° C (13). With a Köfler hot stage on the microscope, Schoch (25) found that corn and sorghum starches are gelatinized at temperature ranges of 60° - 72° C and 68.5° - 75° C, respectively. Normally the temperatures are not affected by slight variations in the amount of the linear starch fraction. High-amylose (amylomaize) corn starch and wrinkled-seeded pea starch show exceptional behavior because they are composed predominantly of linear molecules so highly associated that some of the granules resist gelatinization even in boiling water (12). Different methods have been used for detecting the gelatinization temperatures

of starches. These include measurements of the increase in optical transmittancy (16), determining changes of viscosity (24), and measurements of maltose produced by amylases (28). Measurement of the loss of birefringence has also been used in studying gelatinization (2). Recently the K f ler hot stage has been used (25, 30) to provide a simple and sensitive way to determine the temperature at which starch gelatinizes.

Retrogradation of Starch

A solution or dispersion of starch allowed to age aseptically will exhibit a gradual decrease in digestibility, swelling power, water sorbing capacity, and viscosity and increased cloudiness. Finally, the starch precipitates; this phenomenon is termed "retrogradation" (31). Retrogradation is believed to result from crystallization of large starch molecules. Studies (4) on recrystallizing potato starch from dilute aqueous solutions by the addition of suitable amounts of gelatin showed that granules formed by recrystallization have the same B-type X-ray diffraction pattern as retrograded potato starch.

The rate of retrogradation is dependent on temperature, concentration, molecular size, hydrogen ion concentration and the presence of other chemical reagents in the paste. Some studies (32) showed that amyloses from different sources retrograde at different rates depending upon their average molecular weights. For example corn and wheat amyloses retrograde rapidly from neutral solution in water, while potato amylose, with a much higher molecular weight, retrogrades slowly. Retrogradation rates are highly dependent on temperature. The rates are very rapid at 25  C and increase at lower temperatures. At temperatures above 60  - 70  C amylose solutions remain stable. Davis (3) pointed

out that starch gels held and dried at temperatures higher than 70° C have a strong molecular association and are difficultly dispersible in hot water. Amylose molecules, due to their linear nature, associate rapidly in solution to build up molecular aggregates, hence they retrograde more readily than branched amylopectin. Therefore, according to Davis (3), retrogradation of whole starch is related to the concentration of amylose and the presence of amylopectin. Studies by Jackel et al. (10) also showed that retrogradation may cause staling of bread resulting in a decrease in the starch available to beta-amylase action and also changes in the physical properties of the bread. Retrogradation also has important applications in the adhesive industry and in the fractionation of starch.

Chemical Gelatinization

Starch gelatinization may be accomplished by certain chemical compounds at room temperature. Mangels and Bailey (15) studied the cold gelatinization of wheat starch in aqueous solutions using various reagents over a range of concentrations. Sodium hydroxide, sodium salicylate, sodium, ammonium, or potassium thiocyanate, sodium or potassium iodide, and urea were used as starch-swelling reagents in cold water. Remarkable differences were found in the concentration of the chemical reagent required to induce gelatinization. In order to obtain sufficient swelling to increase the relative viscosity to a value of $4 t/t_w$ the molarity of NaOH needed was 0.09, but when urea was used the molarity required was 6.2. Radley (19) pointed out the use of certain salts and alkalis, such as ammonium nitrate, silver nitrate, calcium chloride, and sodium hydroxide to effect gelatinization. Microscopic

examination indicated that viscosity at lower concentrations of reagents was due to crowding of swollen granules. As concentration was increased the granules broke, and the amylopectin dispersed, forming a true colloidal dispersion (15).

Heat-moisture Treated Starch

At room temperature starch in equilibrium with atmospheric moisture conditions contains 10-17% moisture (12). Water absorption by corn, tapioca, potato and waxy corn starches from saturated vapor has been reported to be 39, 42.9, 50.9 and 51.4 grams per 100 grams, dry basis, respectively (9).

Excess water is needed for the starch granule to swell freely without mechanical disintegration through contact with other granules. Leach reported (13) that one hundred grams of water are required for 4.4 and 4.8 grams of corn and sorghum starch, respectively, to produce a paste at 90° C in which the swollen granules occupy the entire volume. There was almost no "free water" between the swollen granules under these conditions. Novelle and Schutte (18) studied the effect of temperature, time and salts on the gelatinization of pure sorghum starch and starch in sorghum grain. It was found that gelatinization increased rapidly during the first 5 -15 minutes of heating but soon reached a maximum value. This value increased with increasing temperature. Pure starches gelatinized more readily than starch in whole grain.

Heat-treatment of potato and corn starch under high moisture conditions greatly affected their gelatinization behavior. The temperature range for gelatinization was broader and higher than that of the untreated starch (21). Sair's study (21) showed that physical properties of potato

and corn starches change upon treatment at or below 95° or 100° C. There was an increased degree of association of the starch molecules, and a decreased extent to which the granules swelled. Above 100° C degradation of starch was appreciable. The moisture content of starch was an important factor in affecting the physical change (20). It was observed that heating starch under pressure with a limited amount of water brought about a "toughening" action. Thus the solubility ranges were narrower and confined to higher temperatures than those of untreated starches.

Processed Grain

The exact physical and chemical changes in the structure of starch of grain after various treatments are not well known. It has been generally recognized that moist-heat treated grain when fed to certain animals will affect the utilization. Hale et al. reported (5) that steam processing and flaking milo improved gains and feed intake and reduced the feed requirement. Gains were increased by approximately 10% and feed requirements reduced by 5% when compared to dry rolled milo. Ground shelled corn was compared with pelleted, flaked, crumbled and heat-treated corn for fattening lambs and steers by Newland et al. (17). They found that neither pelleting nor the various other methods of heat-processing of corn significantly influenced gains in steers and lambs. However, there were consistent improved feed efficiencies in both steers and lambs with all forms of processed corn when compared with ground shelled corn. Haenlein and Mitchell (8) observed that rates of daily gain and efficiency of feed conversion were significantly

improved by steam-processing of the grain fed. But Garrett (7) pointed out that similar results were obtained in comparisons of steam-rolled and ground grain when feeding beef steers. Gain, carcass yield and feed consumption indicated no significant differences.

Since variable results have occurred with processed grain, there are disagreements concerning the true effects of processing. This work was initiated to evaluate a method for determining starch gelatinization that might be sufficiently accurate to be a useful guide for studying and predicting the effect of moist-heat treatments on grain.

MATERIALS AND METHODS

Sorghum and corn grain were used in experiments to test effects of moisture, pressure, urea and time on starch gelatinization (damage) following moist-heat treatments of grain.

Sorghum was used in Experiment 1 and corn was used in Experiment 2. Heat-moisture treatments were performed as follows. A thin layer of finely ground sorghum or corn was placed in an aluminum dish. Distilled water was added to the sample with a wash bottle where moisture additions were made. Moisture additions of 0, 10 and 20% were made to the grain by varying the amount of added water. The samples were mixed thoroughly prior to heat treatments. Each of the samples with different moisture contents was placed in a pressure chamber and treated with saturated steam under 0, 25 and 50 psi gauge pressure. The pressure chamber was preheated prior to treatments and then flushed with live steam after samples were placed in the chamber and before the valves were closed. The temperatures of the steam were calculated to be 100°, 130.5° and 147.7° C at 0, 25 and 50 psi gauge pressure, respectively. Samples were subjected to moist-heat treatment for periods of 1, 5 and 10 minutes. All the treated samples were air-dried and ground through a Wiley mill following cooking treatments and prior to assays. The samples were packed in plastic sacks and kept in the refrigerator at 10° C until analyses could be completed.

In Experiment 3, urea was added to ground corn prior to heat treatment to determine its effect on starch changes. Urea was added to the ground corn at 0, 5 and 10% levels and mixed thoroughly before cooking. Distilled water was added at 0, 10 and 20% levels in the same manner as

in Experiments 1 and 2. Each of the samples were treated with saturated steam in the pressure cooker at 0, 25 and 50 psi gauge pressure. All samples were processed for a 5-minute period, air-dried when removed, ground and kept in the refrigerator until analyzed.

Sorghum grain was mixed with urea in Experiment 4. The samples were prepared at 0, 5 and 10% levels of urea and water additions were made in the same manner as in Experiments 1 and 2. Moisture addition levels of 0, 10 and 20% and pressures of 0, 25 and 50 psi were studied. All samples were processed for a 5-minute period, the processed samples were prepared for analysis as described for the previous experiments.

In order to observe effects of higher pressures on changes in grain two additional experiments were designed. Sorghum and corn were used in Experiments 5 and 6, respectively. The samples were prepared exactly as the samples of Experiments 1 and 2, except processing pressures were 50 psi (147.7° C) and 75 psi (177.7° C) gauge pressure and treatment times were 1 and 5 minutes.

Determining Starch Damage

The procedure used for determining the degree of starch gelatinization (damage) was based on the availability of starch to enzyme hydrolysis (23, 26, 28). The method used was a modification of the ferricyanide method for the determination of flour maltose values (1). The amount of enzyme used was the major modification. The effect of different quantities of beta-amylase (product of Wallerstein Co., New York, N. Y.) when assaying one gram of processed grain is shown in Figure 1. Fifty milligrams of beta-amylase appeared adequate as there was no obvious

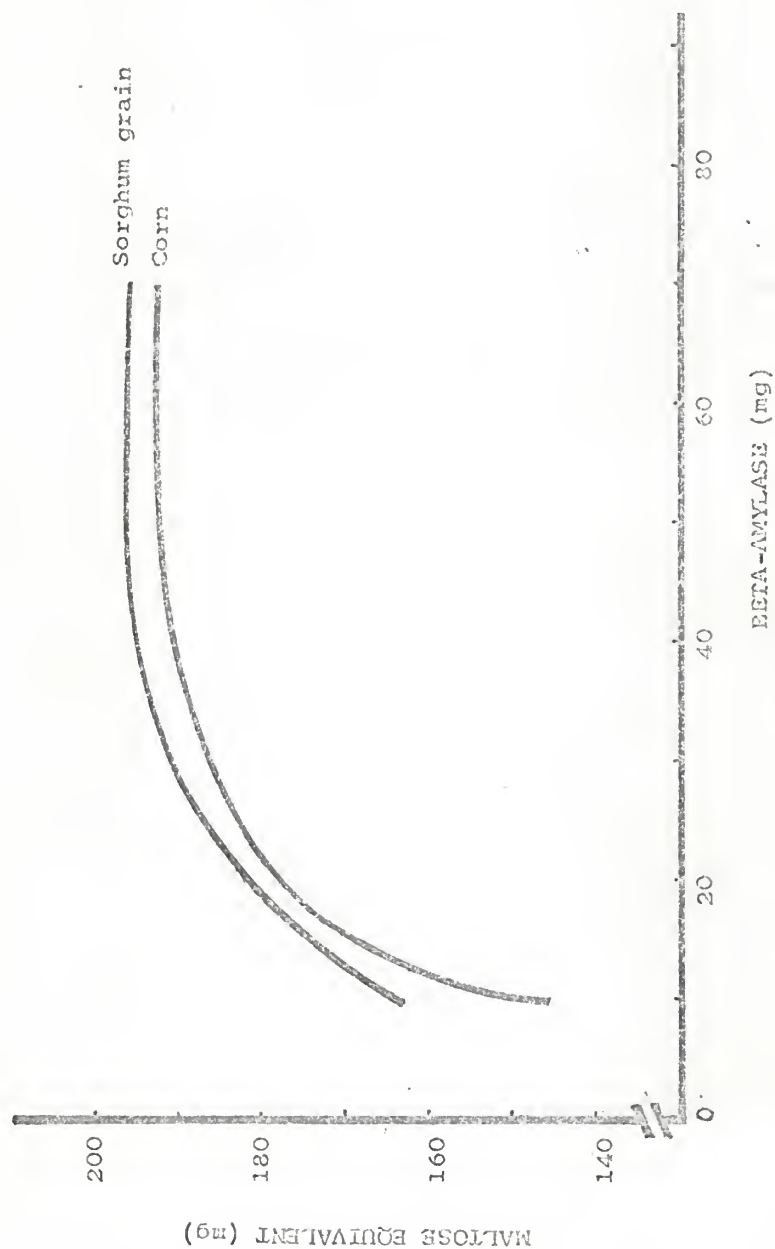


Fig. 1. Effect of Varying Enzyme Concentration on the Amount of Maltose Equivalent

increase in maltose value with increased enzyme concentration above this level. Sixty milligrams of beta-amylase was used in the tests however to insure an excess of the enzyme.

One gram of processed sample was used for each test. Forty milliliters sodium acetate-acetic acid buffer solution, at pH 4.6-4.8 and 30° C was added to the sample. Six milliliters of enzyme solution containing 60 mg of beta-amylase was then added, and mixed thoroughly. The digestion mixture was incubated in a thermostatically controlled water bath at 30° C for exactly two hours. At the end of the 2-hour period 2 ml of 3.58 N sulfuric acid was added and well mixed. Two milliliters of sodium tungstate was then added and mixed thoroughly. The solution was allowed to stand for 2 minutes and was then filtered. A 5 ml aliquot of the filtrate was transferred into a clean test tube and 10 ml of 0.1 N alkaline ferricyanide reagent was added, mixed and the test tube was placed in a boiling water bath for exactly 20 minutes. The test tube and contents were cooled rapidly and 25 ml acetic-acid salt solution were added and 1 ml of soluble starch potassium iodine indicator. The contents were mixed thoroughly and titrated with 0.1 N thiosulfate solution. The ml of 0.1 N ferricyanide reduced by the liberated reducing sugars was converted to mg maltose equivalent.

Analysis of variance and LSD means separation were made by standard methods (6).

RESULTS

The data from the experiments are shown in the following tables and figures. In Experiments 1 and 2, Tables I, II, III, IV, V and VI, it was found that pressure and moisture had significant effects on the gelatinization of starch in both corn and sorghum grain. The effect of treatment time under the conditions of these studies was significant ($\alpha = .05$) interactions between moisture and pressure, but not between time and moisture. Significant pressure and time interactions were found in the studies with corn samples. The interaction effects between pressure and moisture and pressure and time are shown in Figures 3, 4 and 5. Fisher's LSD test was used to determine the significance ($\alpha = .05$) of differences among three levels of pressure, moisture and time. The increase in damaged starch of either corn or sorghum grain due to the change in pressure from 0 to 25 psi was not significant. Increased moisture (10% added moisture) and time, from 1 to 5 minutes, did not result in significant changes in maltose equivalents produced by the action of beta-amylase.

In Experiment 3, Tables VII, VIII and IX, the effects of urea, pressure and moisture were studied. Data in Table VIII show no significant interaction between any two of the three factors. The three levels of added urea did not result in significant changes in the starch of corn. Pressure and moisture had the same effect on the starch as in Experiments 1 and 2.

In Experiment 4, Tables X, XI and XII, significant urea, pressure and moisture effects were found when 0, 5 and 10% urea was added to sorghum grain at added moisture levels of 0, 10 and 20%. There were no

TABLE I

Effect of Moisture, Pressure and Duration of Treatment
on Starch Gelatinization of Moist-Heat Treated Sorghum Grain

Duration of treatment, minutes	Pressure, psi								
	0			25			50		
	Moisture, %								
	0	10	20	0	10	20	0	10	20
	maltose equivalent mg								
1	33	28	31	24	25	34	25	67	137
5	28	21	24	20	44	87	39	127	119
10	18	21	31	20	30	59	33	103	157

TABLE II

Analysis of Variance for the Effect of Moisture, Pressure and
Duration of Treatment on Starch Gelatinization
of Moist-Heat Treated Sorghum Grain

Source of Variation	DF	Mean Square	F
Pressure	2	10261.9	38.6*
Moisture	2	5354.9	20.1*
Time	2	315.1	1.2
Pressure Moisture	4	2181.9	8.2*
Time x Moisture	4	155.8	0.5
Time x Pressure	4	265.0	0.9
Samples	8	265.5	

* Significant at $\alpha = .05$

TABLE III

Fisher's LSD Test for Significance Between Main Effects
on the Treated Sorghum Grain in Table I

Pressure, psi	0	25	50
	26.1	38.1	89.6
Moisture, %	0	10	20
	26.1	51.7	76.5

Values which are underlined are significantly different ($\alpha = .05$)

TABLE IV

Effect of Pressure, Moisture and Duration of Treatment on Starch
Gelatinization of Moist-Heat Treated Corn

Duration of treatment, minutes	Pressure, psi								
	0			25			50		
				Moisture, %					
	0	10	20	0	10	10	0	10	20
	maltose equivalent mg								
1	35	32	34	28	29	34	28	42	66
5	23	19	22	16	31	34	39	97	117
10	20	24	31	48	32	46	42	88	139

TABLE V

Analysis of Variance for the Effect of Pressure, Moisture and Duration of Treatment on Starch Gelatinization of the Moist-Heat Treated Corn

Source of Variation	DF	Mean Square	F
Pressure	2	5697.9	52.8*
Moisture	2	1655.5	15.3*
Time	2	560.1	5.1*
Pressure x Moisture	4	1103.3	10.2*
Time x Pressure	4	751.8	6.9*
Time x Moisture	4	144.2	1.3
Sample	8	107.8	

* Significant at $\alpha = .05$

TABLE VI

Fisher's LSD Test for Significance Between Main Effect on Treated Corn in Table VI

Pressure, psi	0	25	50
	26.6	<u>33.1</u>	<u>73.1</u>
Moisture, %	0	10	20
	31.0	<u>43.7</u>	<u>58.1</u>
Time, min.	1	5	10
	<u>36.4</u>	<u>44.2</u>	<u>55.2</u>

Values which are underlined are significantly different ($\alpha = .05$)

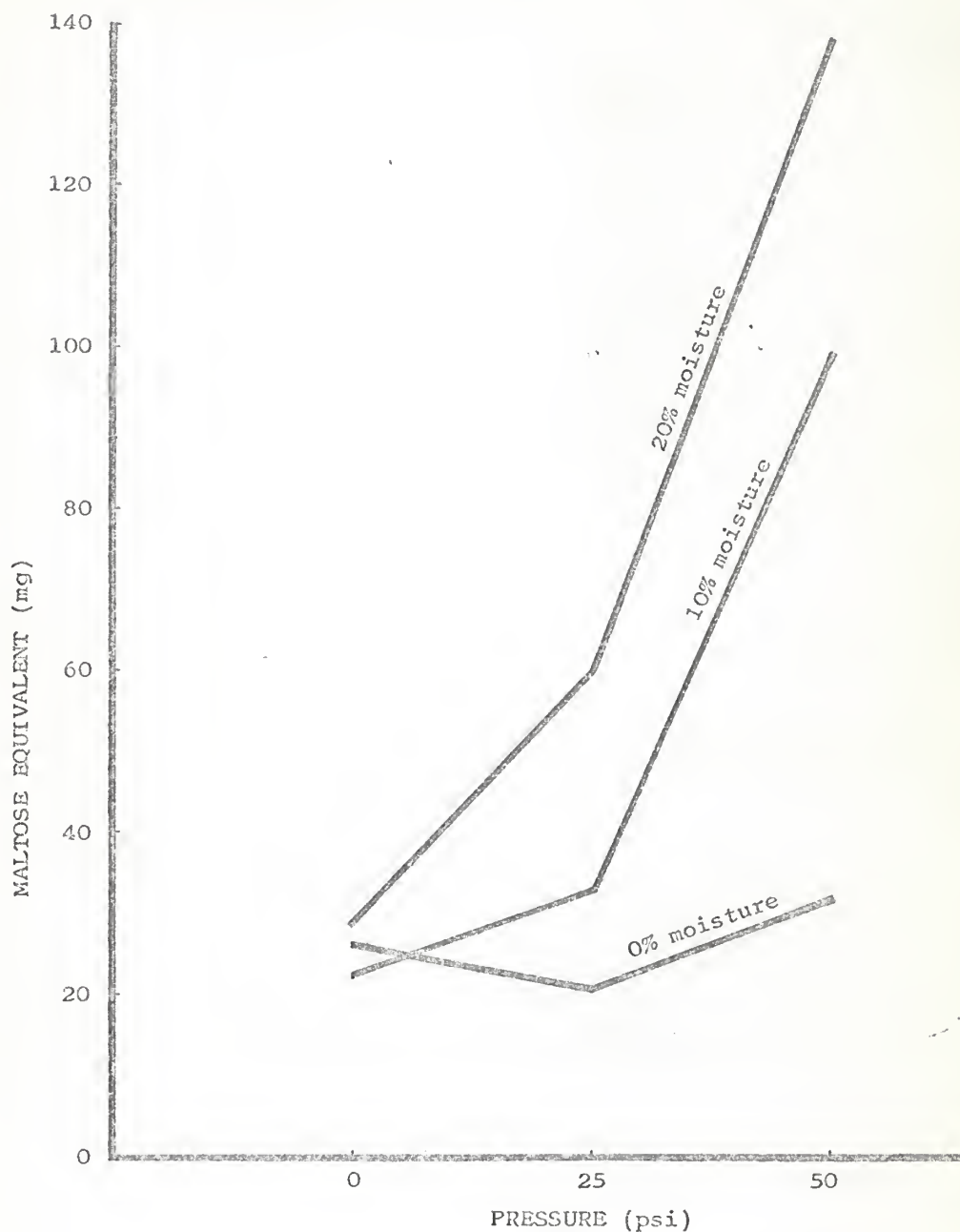


Fig. 2. Effects of Pressure and Moisture During Treatment on Maltose Equivalent of Sorghum Grain

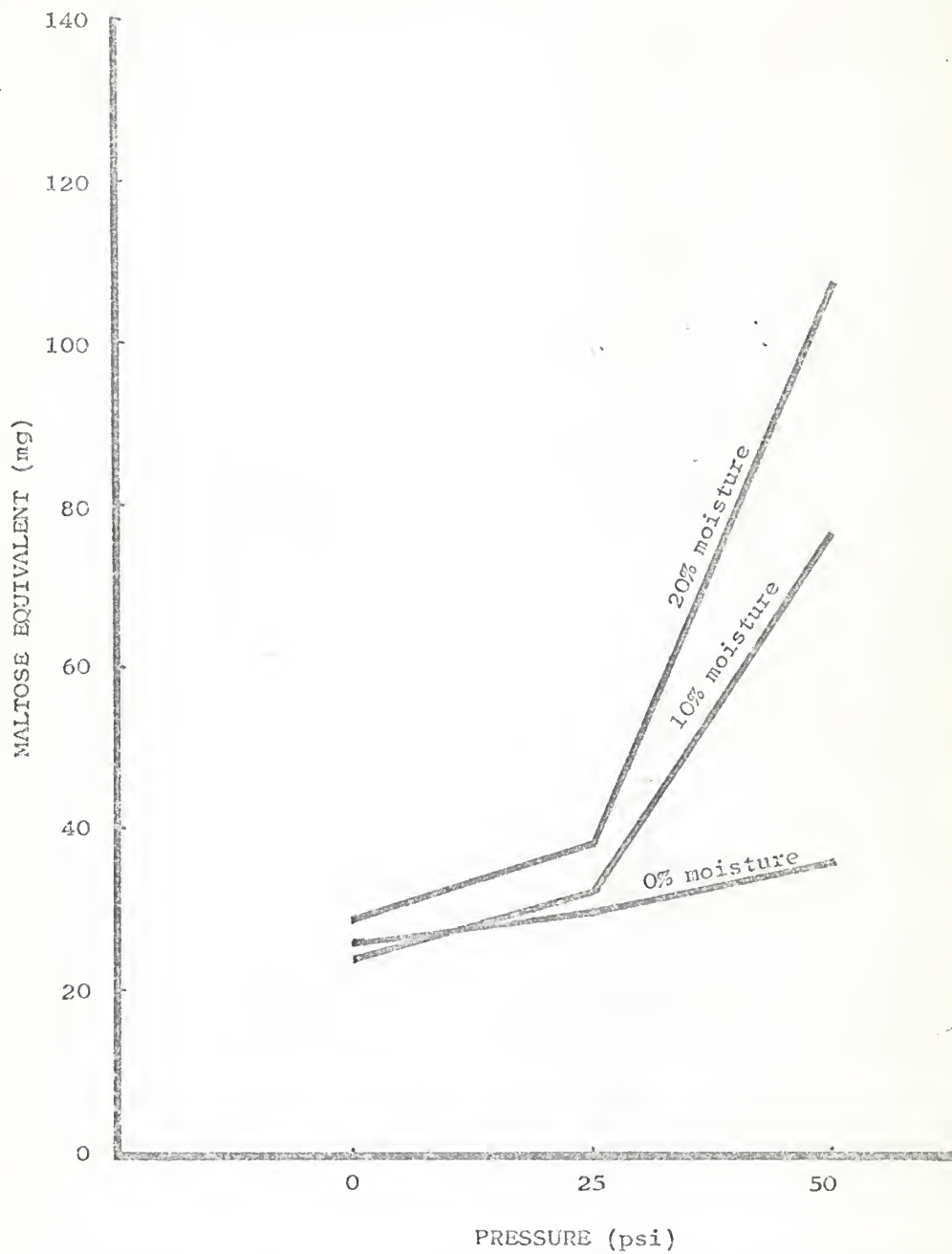


Fig. 3. Effects of Pressure and Moisture During Treatment on Maltose Equivalent of Corn

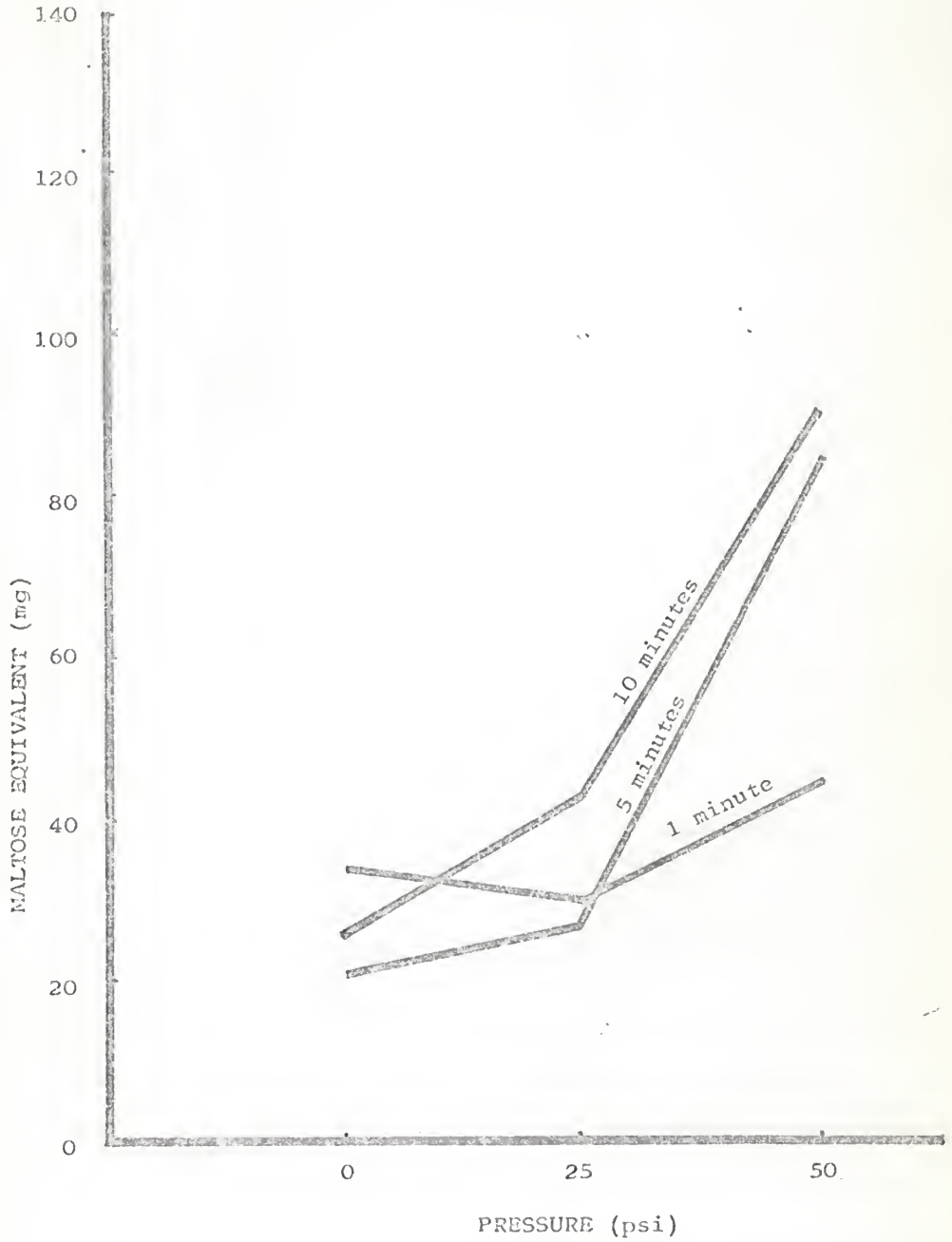


Fig. 4. Effects of Pressure and Duration of Treatment on Maltose Equivalent of Starch in Corn

TABLE VII

Effect of Pressure, Moisture and Urea on Starch Gelatinization
of Corn Moist-Heat Treated for 5 Minutes

Urea, %	Pressure, psi								
	0			25			50		
	Moisture, %								
	0	10	20	0	10	20	0	10	20
	maltose equivalent mg								
0	23	19	22	16	31	34	39	97	117
5	36	42	43	28	37	47	38	41	87
10	39	55	57	38	49	50	46	74	102

TABLE VIII

Analysis of Variance for the Effect of Pressure, Moisture and
Urea on Starch Gelatinization of Moist-Heat Treated Corn

Source of Variation	DF	Mean Square	F
Pressure	2	3514.38	21.25*
Urea	2	451.48	2.73
Moisture	2	1818.70	10.99*
Pressure x Moisture	4	618.42	3.74
Pressure x Urea	4	554.98	3.55
Moisture x Urea	4	68.54	0.41
Samples	8	165.35	

* Significant at $\alpha = .05$

TABLE IX

Fisher's LSD Test for the Significance Between Main Effect
on Treated Corn in Table VII

Pressure, psi	0	25	50
	37.3	<u>36.6</u>	<u>71.2</u>
Moisture, %	0	10	20
	<u>33.6</u>	<u>49.4</u>	62.1

Values which are underlined are significantly different ($\alpha = .05$).

TABLE X

Effect of Pressure, Moisture and Urea on Starch Gelatinization
of Moist-Heat Treated Sorghum Grain

Urea, %	Pressure, psi								
	0			25			50		
	Moisture, %			Moisture, %			Moisture, %		
	0	10	20	0	10	20	0	10	20
	maltose equivalent mg								
0	15	14	19	18	70	56	119	101	108
5	26	33	40	47	76	91	91	174	236
10	42	38	76	50	85	119	218	198	258

TABLE XI

Analysis of Variance for the Effect of Pressure, Moisture and Urea on Starch Gelatinization of Moist-Heat Treated Sorghum Grain

Source of Variation	Df	Mean Square	F
Pressure	2	43136.3	71.7*
Moisture	2	3972.1	6.6*
Urea	2	8841.3	14.7*
Pressure x Moisture	4	559.2	.9
Pressure x Urea	4	1570.6	2.6
Moisture x Urea	4	860.4	1.4
Samples	8	600.9	

*Significant at $\alpha = .05$

TABLE XII

Fisher's LSD Test for the Significance Between Main Effect on the Treated Sorghum Grain

Pressure, psi	0	25	50
	<u>33.7</u>	<u>68</u>	<u>167</u>
Moisture, %	0	10	20
	<u>69.5</u>	<u>87.6</u>	<u>111.4</u>
Urea, %	0	5	10
	<u>57.7</u>	<u>90.4</u>	<u>120.4</u>

Values which are underlined are significantly different ($\alpha = .05$)

significant interaction effects between pressure and urea, pressure and moisture or moisture and urea.

In Experiments 5 and 6, Tables XIII, XIV and XVI, there were significant differences in starch damage occurring at 50 and 75 psi. Effects of the levels of moisture on starch gelatinization were significant in both corn and sorghum grain. These effects were also found in Experiments 1 and 2. It can be seen (Tables XIII and XIV) that increasing the time of treatment from 1 to 10 minutes resulted in a significant increase in maltose equivalents produced by the action of beta-amylase on the starch of the grain. There were no interaction effects however, between moisture and pressure, time and pressure or time and moisture.

TABLE XIII

Effect of Moisture, Pressure and Duration of Treatment on Starch Gelatinization of the Moist-Heated Grain Sorghum

Duration of Treatment, Minutes	Moisture, %					
	0		10		20	
			Pressure, psi			
	50	75	50	75	50	75
	maltose equivalent mg					
1	45	118	112	190	133	237
10	150	199	225	270	195	307

TABLE XIV

Analysis of Variance for the Effect of Moisture, Pressure and Duration of Treatment on Starch Gelatinization of the Moist-Heat Treated Sorghum Grain

Source of Variation	DF	Mean Square	F
Pressure	1	17710.08	152.55*
Moisture	2	9018.75	77.68*
Time	1	21760.09	187.44*
Pressure x Moisture	2	728.59	6.27
Time x Moisture	2	278.58	2.39
Time x Pressure	1	200.08	1.72
Samples	2	116.09	

* Significant at $\alpha = .05$

TABLE XV

Effect of Moisture, Pressure and Duration of Treatment on Starch Gelatinization of the Moist-Heat Treated Corn

Duration of Treatment, Minutes	Moisture, %					
	0		10		20	
	Pressure, psi					
	50	75	50	75	50	75
maltose equivalent mg						
1	47	131	101	187	123	249
10	96	184	190	251	200	257

TABLE XVI

Analysis of Variance for the Effects of Moisture, Pressure and Duration of Treatment on Starch Gelatinization of the Moist-Heat Treated Corn

Source of Variation	DF	Mean Square	F
Pressure	1	21020.49	58.90*
Moisture	2	9211.75	25.81*
Time	1	9653.49	27.05*
Pressure x Moisture	2	76.03	0.21
Time x Moisture	2	303.03	0.84
Time x Pressure	1	634.68	1.78
Samples	2	356.87	

* Significant at $\alpha = .05$.

DISCUSSION

Pressure is an important factor in grain processing. Within the limitations of these studies, increasing the pressure caused an increase in change in the starch. However, the first 25 psi pressure failed to produce significant changes in the starch. This may have been because 25 psi pressure did not provide enough energy to weaken the overall strength of the micellar network of the starch granule (12). There was little difference in the effect of pressure on corn and on sorghum grain.

Moisture is considered extremely important to the gelatinization of starch (13). As the temperature increases above the gelatinization range, the amount of water available to the starch influences the swelling of the starch granule and directly affects the extent of starch solubilization. It is known that starch heated under high pressure with a limited amount of water has decreased solubility. The results of this study indicate that the higher the moisture content the greater the amount of gelatinized or damaged starch available to beta-amylase action. The first 10% moisture addition resulted in slightly different effects on corn and sorghum grain. This may have been due to variation of the grain constituents or to moisture movement in the grain structure. At 0% added moisture significant effects on starch damage for both corn and sorghum were not found when the treatment pressure was increased. After the addition of moisture however, at 10 to 20% levels the effect of increasing pressure was highly significant.

The length of time during which the grain was under treatment also affected the amount of gelatinized starch; the longer the treatment time, the higher the amount of starch gelatinized. The first 10 minutes did not appear to be a significant factor.

Mangels and Bailey (15) reported that starch was found to be gelatinized at room temperature when a concentration of 6.2 M urea solution was added to a 2% starch dispersion. The effect of urea on the starch in grain at a high temperature was tested. The extent of starch gelatinization was not significantly affected by the urea in corn samples. But in sorghum grain, a significant urea effect did exist. The different behavior of corn and sorghum grain with urea may have been brought about by different physical and chemical properties of these two grains. It is known that the gelatinization temperature (13), retrogradation rate (32), swelling power, solubility (25), and water absorption (9) of starch vary from one species to another. The structure and constituents of grain are highly complex and the exact reaction between urea and the grain is not clear. Therefore, different effects of urea on the intact starch of corn and sorghum grain might be expected.

SUMMARY

The effect of moisture, pressure, time and urea on the gelatinization of starch in moist-heat treated sorghum grain and corn was studied. Susceptibility to beta-amylase of the starch in the heat-moisture treated grain was used for measuring the amount of gelatinized starch.

The effect of pressure on starch gelatinization was to significantly increase it as pressure increased from 25 to 75 psi. At 25 psi, pressure was not an important factor. Significant effects of time could be found after the first 10 minutes. Generally, moisture effects were found to be highly significant after added water was raised above 10%. Within the limitations of this study, amount of urea in the sample of sorghum grain had a great deal of influence on the starch gelatinization, but this effect was not found in the case of corn.

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ENZYMATIC EVALUATION OF CHANGES IN PROCESSED GRAIN

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An-chein Sung

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AN ABSTRACT OF A MASTER'S THESIS

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MASTER OF SCIENCE

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Finely ground sorghum grain or corn, with different moisture contents were treated with saturated steam under different pressures in a pressure chamber. Various amounts of urea were added in some of the samples. Samples were treated for varying time periods. The effect of moisture, pressure, time and urea on the starch damage in moist-heat treated sorghum and corn was studied. Susceptibility to beta-amylase digestion of the starch in heat-moisture treated grain was used to determine the amount of damaged starch.

Within the limitations of these studies, it was found that pressure was an important factor in grain processing. Increasing the pressure from 25 to 75 psi resulted in significant increase in damage to the starch. The first 25 psi pressure however failed to produce significant changes in the starch. There was little difference in the effect of the pressure on corn and on sorghum grain.

Moisture was found to be extremely important in the gelatinization of starch. This study indicated that higher moisture contents resulted in greater amounts of gelatinized or damaged starch available to beta-amylase action. Moisture effects were found to be highly significant when added water was raised above 10%. The first 10% moisture addition resulted in slightly different effects on corn and sorghum grain. At 0% added moisture no significant effects on starch damage for both corn and sorghum grain were found when the treatment pressure was increased. But after the addition of moisture at 10 to 20% levels the effect of increasing pressure was highly significant.

The length of time during which the grain was under treatment also affected the amount of gelatinized starch: the longer the treatment time, the higher the amount of starch gelatinized. The first 10 minutes did

not appear to be a significant factor when the effect of urea on the starch in grain at a high temperature was tested. The extent of starch gelatinization was not significantly affected by the urea in the corn samples, but in sorghum grain a significant urea effect was found.