WET MILLING OF CORN USING GASEOUS SO_2 ADDITION PRIOR TO STEEPING AND THE EFFECT OF LACTIC ACID ON STEEPING

by

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INTRODUCTION

Corn is the only important cereal crop indigenous to the Americas. In the United States, corn production is more than double the production of any other crop. Corn has a high starch content (more than 70%) and protein of acceptable quantity (about 10%) and quality. Thus, it is useful for processing into valuable food and industrial products.

Approximately 15% of the annual U.S. production is processed by wet milling to produce food starch and sweetener products. Nonfood products, such as industrial starch, corn gluten feed, and corn gluten meal, are also produced by the process. These products have shown steady market growth in recent years.

Corn wet milling involves steeping under well controlled conditions to soften the kernels, milling, and separating components by screening, centrifuging and washing to produce starch, oil, and feed by-products. It is an energy and capital intensive process, in which considerable energy use is for steeping, the soaking of corn kernels in water under controlled processing conditions of temperature, time, concentration of SO_2, lactic acid content, etc.

Traditionally, whole kernels are steeped for 20 to 35 hours in a dilute SO_2 solution, which acts to control undesirable fermentations, softens the kernel to facilitate germ separation, removes corn solubles, cleaves disulfide bonds in the protein, and activates indigenous protolytic enzymes which solubilize the protein matrix encasing the starch granules. Preliminary studies have indicated that a gaseous SO_2 steep may induce similar changes on dry corn kernels. Vapor steeping would appear to reduce steep time by getting the gaseous SO_2 into the endosperm rapidly and considerable energy savings could be realized.

Lactic acid, the chief product of fermentation from soluble sugars in the SO_2 solution, was reported to be able to lower the pH value of the SO_2 solution, restrict the growth of other organisms, cause some softening of kernel components, and produce the most favorable conditions for separation of the kernel components, especially protein, after milling.

The objectives of this study are to determine the relationship between starch yield, steep time, concentration of SO_2, and the addition of lactic acid to steep water when a gaseous SO_2 treatment prior to steeping is applied, and to help determine the most economical method of steeping in corn wet milling.

LITERATURE REVIEW

Corn kernels can be separated structurally into four parts: the endosperm, germ, pericarp, and tip cap. Table 1 shows the weight and composition of component parts of dent corn kernels. Foreign materials, such as water, solution, etc., enter corn kernels through the tip cap, diffuse into the spaces in the pericarp, and penetrate into the endosperm.

The endosperm of corn kernels constitutes 82% to 84% of the kernel dry weight and is 86% to 89% starch by weight. It is comprised of elongate cells packed with starch granules embedded in a continuous protein matrix. To recover starch by wet milling, the granules must be released from the matrix. The traditional method is to treat corn kernels with a reducing agent, commonly a dilute SO_2 solution (0.1% to 0.2%, based on the weight of corn kernels), at 45^oC to 50^oC (Watson, 1984) for 40 to 48 hours (now about 20 to 35 hours).

The SO_2 causes the matrix structure to weaken by breaking the disulfide bonds and forming soluble S-sulfoproteins, which prevent disulfide bonds reformation (Boundy et al., 1967).

After steeping, the kernel components, including germ, fiber, starch, and gluten, can be separated by processing methods, including centrifuging, screening, and washing.

Eckhoff (1983) showed that gaseous SO_2 penetrates the corn kernel about two orders of magnitude faster than Fan

Table 1. Weight and Composition of Component Parts of Dent Corn Kernels.

Part	Percent dry weight of whole kernel	Composition of kernel parts (%, d.b.) Starch	Fat	Protein	Ash	Sugar	Unaccounted for
Endosperm							
Mean	82.9	87.6	0.8	8.0	0.3	0.62	2.7
Range	81.8	86.4	0.7	6.9	0.2	0.50	
	83.5	88.9	1.0	10.4	0.5	0.80	
Germ							
Mean	11.1	8.3	33.2	18.4	10.5	10.8	8.8
Range	10.2	5.1	31.1	17.3	9.9	10.0	
	11.9	10.0	35.1	19.0	11.3	12.5	
Pericarp							
Mean	5.3	7.3	1.0	3.7	0.8	0.34	86.7
Range	5.1	3.5	0.7	2.9	0.4	0.2	
	5.7	10.4	1.2	3.9	1.0	0.4	
Tip Cap							
Mean	0.8	5.3*	3.8	9.1	1.6	1.6	78.6
Range	0.8	----	3.7	9.1	1.4	----	
	1.1		3.9	10.7	2.0		
Whole Kernel							
Mean	100	73.4	4.4	9.1	1.4	1.9	9.8
Range	----	67.8	3.9	8.1	1.37	1.61	
		74.0	5.8	11.5	1.50	2.22	

* Composite

Source: Watson (Corn: Chemistry And Technology) et al. (1965) found that an SO_2 solution diffuses into the kernel. For example, Cox et al. (1944) found that it requires 8 hours to completely penetrate the corn kernels by the SO_2 solution steep. The time required for penetration is reduced from 8 hours to 4 to 5 minutes with a gaseous SO_2 steep. This shortened penetration was confirmed by Eckhoff (1983). Eckhoff also showed that corn kernels at a moisture content of 30%, exposed to gaseous SO_2 for 2 minutes, were almost completely penetrated by the SO_2. Watson and Sanders (1961) showed that starch release could be achieved in as little as 2 to 6 hours of steeping if the diffusion limitation were removed.

Eckhoff (1983) observed that the gaseous SO_2 entered through the tip of the cap, moved rapidly up through the germ, and then penetrated the endosperm from the inside out. The SO_2 rapidly moved through the space between the pericarp and the seed coat concurrently. However, penetration into the endosperm through the seed coat was slower than the movement of the SO_2 up through the germ into the endosperm. The SO_2 solution was found by Cox et al. (1944) to spread rapidly through the germ in less than 1 hour, but no mention was made of its penetration into the endosperm via this path. Contrary to this, Wolf et al. (1952), Fan et al. (1965), and others, had considered that the SO_2 solution penetrated the endosperm primarily through the seed coat.

The application of gaseous SO_2 will increase not only the rate of penetration into the endosperm but also the amount of SO_2 that can potentially be added. Increased concentration levels of SO_2 have been found to increase starch yield. Watson and Sanders (1961) showed that increasing SO_2 concentration from 0.05% to 0.2% resulted in an increased rate and extent of starch release. Cox et al. (1944) found that protein dispersion was faster and starch extraction was more complete as the SO_2 concentration increased up to 0.4%. Similarily, Roushdi et al. (1981a) showed that batch steeping for 36 hours in a high level of fresh SO_2 (250 ppm) at 50^oC inhibited lactic acid formation, but still gave higher starch yields with lower protein contents than steeping with SO_2 levels which promoted lactic acid formation. Roushdi et al. (1981b) found that, in a semi-batch system where the steep water was changed after 10 hours and 30 hours (50 hours total steep time), low levels of SO_2 (100 ppm) with added lactic acid had higher starch yields and lower protein content than using 250 ppm SO_2 steep water which inhibited lactic acid formation. Krochta et al. (1980) also showed that increasing the SO_2 concentration from 0.1% to 0.4% increased the starch milling yield from 66.6% to 72.6%.

The main deterrent to the use of SO_2 levels above 0.2% in the steep has been concern that the delicate balance of lactic acid fermentation would be destroyed (Watson and Sanders, 1961). However, the importance of the lactic acid fermentation phase of the process has never been fully established. Contrary to the assumptions made by Watson et al. (1955), lactic acid does not produce the proteolytic enzymes which solubilize the protein and enhance starch release (Wall and Paulis, 1977).

The kernel softening effect necessary in wet milling can be divided into two parts. The first is chemical softening, which is the SO_2 disruption of the protein structure in the endosperm. This phenomena does not appear to need water to occur (or at least not more than normal storage moisture contents). The second part of softening is the physical softening that results principally in a change in the solid mechanical characteristics of the germ and fiber. This aids in the subsequent separation processes. This physical softening appears to occur primarily as a result of water absorption. Removal of solubles from the germ via the water also causes physical changes in the germ. There is some indication that lactic acid is beneficial in this physical softening process. Roushdi et al. (1981a) found that high levels of SO_2 alone gave better starch yields than lower levels of SO_2 with lactic acid production. However, in a follow-up study they found that steeping with lactic acid aided in lowering the protein content in starch and increased starch yields. Table 2 shows the results of their experiments.

After these quantitative analyses, quality of products is to be considered. For starch, it is a question of the residual protein content and rheological qualities: maximum viscosity, and temperature at the start of gelatinization (Le Bras, 1982).

Table 2. Steeping Agents During Steeping Process, and Starch Recovery, Protein Content in the Starch and Starch Content of Hulls and Fibers.

	Treatment no.						
	1	2	3	4	5	6	7
Steep agents*							
First stage (10 hrs)	F	O	O	O	Low F and L	Low F and L	Low F and L
Second stage (20 Hrs)	F	F	O	O	F	Low F and L	Low F and L
Third stage (20 Hrs)	F	F	F	O	F	F	Low F and L
Isolated starch							
Recovery (%)	58.5	63.0	61.8	63.4	63.1	60.7	63.8
Protein content(%)	1.11	0.87	0.99	0.82	0.85	0.87	0.84
Starch content in							
Hulls (%)	10.5	7.0	8.1	6.9	7.9	8.5	7.4
Fibers (%)	31.3	28.1	29.4	28.0	28.0	29.5	27.5

* F = Fresh SO_2 solution contained 300 ppm SO_2
O = Old SO_2 solution was used before in the steeping process and contained 100 ppm SO_2
Low F = Fresh low SO_2 solution contained 100 ppm SO_2
L = 0.55% lactic acid (based on the weight of SO_2 solution)

Source: Roushdi et al. (1981b)

OBJECTIVES

The objectives of this research project were to:

1. Compare the product yields and starch quality of corn kernels treated with gaseous SO_2 or sulfurous acid and steeped for different periods of time.
2. Investigate the influence of the addition of lactic acid in steep water on starch yield.
3. Investigate the changes of concentration and amount of SO_2 in steep water and in corn kernels steeped with gaseous SO_2 and SO_2 solution.

METHODS AND PROCEDURES

Corn Wet Milling

Experimental Design

A 4*3 experimental design with two replicates of each condition was utilized. Steep time, concentration level of SO_2, and the addition of lactic acid to steep water were the variables in the experimental design.

Steep times of 6 hours, 12 hours, and 24 hours were evaluted for corn kernels treated with gaseous SO_2 at concentration levels of 0.4% (based on the weight of corn kernels) and 0.2% with and without the addition of 0.55% lactic acid (based on the weight of steep water) while steeping. A 0.4% gaseous SO_2 treatment corresponded to a 0.2% SO_2 solution steep. A 48-hour traditional steep in which SO_2 was added to distilled water to make an SO_2 solution served as the control. The concentration levels of the SO_2 solutions were also 0.2% and 0.4%. Table 3 shows this experimental design.

Procedures

Corn Wet Milling with a Gaseous SO_2 Steep: Yellow dent corn kernels (Dekalb 636) with a moisture content of 13.5% to

Table 3. Experimental Design of Corn Wet Milling with Different Types of Steep.

Starch Yields

Steep time	Concentration of SO_2 (based on weight of corn kernels): 0.2% with lactic acid*	0.2%	0.4%
6 Hrs	S_1 S_2**	S_1 S_2	S_1 S_2
12 Hrs	S_1 S_2	S_1 S_2	S_1 S_2
24 Hrs	S_1 S_2	S_1 S_2	S_1 S_2
48 Hrs***	S_1 S_2	S_1 S_2	S_1 S_2

* The concentration of lactic acid is 0.55% based on the weight of steep water.

** S_1 and S_2 refer to the starch yields for replication 1 and replication 2, respectively.

*** Corn kernels are steeped traditionally with SO_2 solution to serve as a control.

14.5% w.b. were run through a dockage tester and inspected visually.

Fifteen hundred grams of corn kernels and six 25-gram samples were placed in nylon mesh for further analysis into a heavy polyethylene bag and sealed. The gaseous SO_2 treatment was performed by using a procedure involving the metering of gaseous SO_2 from a compressed gas tank through a rotameter and into the bag with an injection needle. The length of time of treatment at a constant flow rate was measured to determine the total dosage of SO_2 in the bag. The length of time for flow of the gaseous SO_2 into the sample bag was determined by the following method.

flow rate of SO_2 = 1.59 g/min,

weight of corn kernels = 1500 g + 6*25 g

= 1650 g,

weight of SO_2 to be added to make a concentration level of 0.2% = 0.2%*1650 g

= 3.3 g,

length of time of SO_2 treatment to make a concentration level of 0.2% = 3.3 g/(1.59 g/min)

= 2 min 4.5 sec,

The bag was held for 15 minutes at room temperature for SO_2 adsorption and shaken for uniformity. Three samples were removed, and the direct titrimetric method (Eckhoff and Okos, 1983) was used to estimate water-extractable SO_2 in the corn kernels. The corn kernels in the bag were placed with or without 0.55% lactic acid (based on the weight of steep water) into a temperature-controlled container, which contained 2800 milliliters of distilled water preheated to 45^oC to 50^oC.

After storage in the container for a scheduled steep time of 6, 12, or 24 hours at 45^oC to 50^oC, the steep water was drained off and saved. The direct titrimetric method was used for the remaining three samples and 30 milliliters of steep water to estimate the water-extractable SO_2 in the corn kernels and in the steep water. The weight and pH value of the steep water were measured, and then the steep water was dried to determine the weight of soluble solids.

The corn kernels were ground and some distilled water was added during the grinding. The germ was separated from the slurry by the removal of germ floating on the surface of the slurry with a screen. Some starch was added to float the germ if necessary. The germ was then washed with distilled water on a 0.039-inch screen, and dried at 49^oC for 16 hours. The slurry was ground finely. Coarse fiber was separated from the slurry with a 0.039-inch screen, washed, and dried at 49°C for 16 hours. Fine fiber was separated from the slurry with a 200-mesh screen, washed, and dried at 49°C for 16 hours.

The specific gravity of the slurry was measured and adjusted to 1.04 by the addition of starch or distilled water to increase or decrease it respectively. Starch was then separated from gluten by passing the slurry to a starch table in a discontinuous flow. The starch on the starch table was collected and dried at 49°C for 16 hours. Water was removed from the overflow of the gluten slurry by filtration with a vacuum pump and the gluten slurry was dried at 49°C for 16 hours. The weights of all products, soluble solids, germ, coarse fiber, fine fiber, starch and gluten, were recorded.

Corn Wet Milling with a Traditional SO_2 Solution Steep: Yellow dent corn kernels with a moisture content of 13.5% to 14.5% w.b. were run through a dockage tester and inspected visually. The concentration of a concentrate SO_2 solution, made by dissolving the gaseous SO_2 into distilled water, was estimated by the direct titrimetric method to determine the weight of the concentrate SO_2 solution to be added to steep water.

concentration of the concentrate SO_2 solution = 74800ppm, weight of corn kernels = 1500 g + 3*25 g = 1575 g,

weight of the concentrate SO_2 solution to be added to distilled water to make a concentration level of 0.2%
= (0.2%*1575 g)/74800ppm
= 42.11 g,

The concentrate SO_2 solution and corn kernels were added with or without 0.55% lactic acid to a temperature-controlled container which contained 2800 milliliters of distilled water preheated to 45°C to 50°C.

After storage for 48 hours at 45°C to 50°C in the container, the steep water was drained off and saved. The three samples and 30 milliliters of steep water were removed and the direct titrimetric method was used to estimate water-extractable SO_2 in the corn kernels and in the steep water. The weight and pH value of the steep water were measured, and then the steep water was dried to determine the weight of soluble solids in it.

The corn kernels were ground and some distilled water was added during the grinding. The germ was separated from the slurry by the removal of germ floating on the surface of the slurry with a screen. Some starch was added to float the germ if necessary. The germ was then washed with distilled water on a 0.039-inch screen, and dried at 49°C for 16 hours.

The slurry was ground finely. Coarse fiber was separated from the slurry with a 0.039-inch screen, washed, and dried at 49°C for 16 hours. Fine fiber was separated from the slurry with a 200-mesh screen, washed, and dried at 49°C for 16 hours.

The specific gravity of the slurry was measured and adjusted to 1.04 by the addition of starch or distilled water to increase or decrease it, respectively. Starch was then separated from gluten by passing the slurry to a starch table constructed according to the specifications reported by Anderson (1963) in a discontinuous flow. The starch on the starch table was collected and dried at 49°C for 16 hours. Water was removed from the overflow of the gluten slurry by filtration with a vacuum pump and the gluten slurry was dried at 49°C for 16 hours. The weights of all products, soluble solids, germ, coarse fiber, fine fiber, starch and gluten, were recorded.

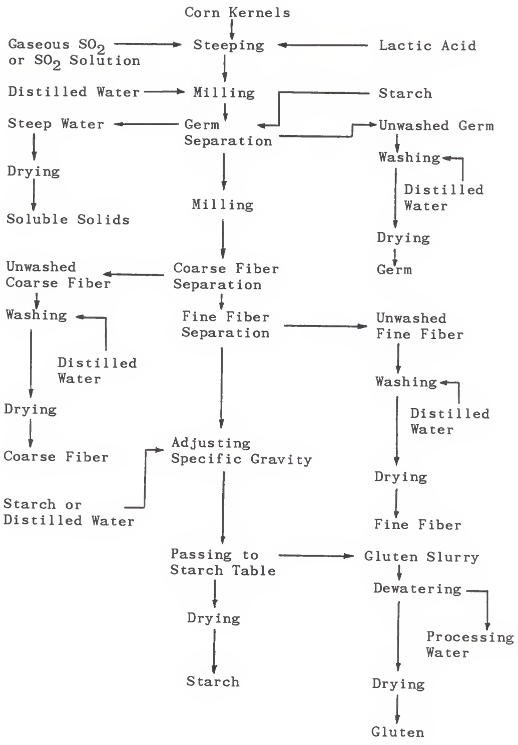
Figure 1 shows the laboratory flow of corn wet milling.

Analysis of Starch Quality

Protein Content in Starch

All of the starch samples were sent to the Analytic Center in the Animal Science Department, Weber Hall, Kansas

Figure 1. Laboratory Flow of Corn Wet Milling.



State University for analysis of the protein content in each starch sample.

Consistency of Starch Slurry

A Brabender viscoamylograph was used to measure the consistencies of slurries of all the starch samples. Fifty grams of starch was added to distilled water to make a 500-milliliter starch slurry. The starch slurry was poured into the viscoamylograph bowl and the instrument, set at 75 rpm, was switched on. The drawing instrument connected to the viscoamylograph was switched on when the temperature of the slurry was increased to 50^{o}C, and switched off when the temperature reached 95^{o}C. The instrument was allowed to run until the red heat indicating pilot light went off, and then the instrument was turned off.

The maximum value of the heating portion of the curve (pasting peak) was recorded, and the value was considered the consistency of the starch slurry. The unit of the consistency was b.u. (Brabender unit), and 1 b.u. indicated that 1 millivolt was needed to keep the instrument operating at 75 rpm during the experiments.

Changes of Concentration and Amount of SO_2 During the Steeping Process

Experimental Design

In order to investigate the concentration change of SO_2 in corn kernels and in steep water while steeping, the procedures of steeping were performed with 0.2% gaseous SO_2 for 24 hours and 0.2% SO_2 solution for 48 hours, respectively, and the direct titrimetric method was used to estimate water-extractable SO_2 in corn kernels and in steep water. Table 4 shows this experimental design.

Procedures

<u>Changes of Concentration and Amount of SO_2 While Steeping with Gaseous SO_2</u>: Fifteen hundred grams of corn kernels (including eleven 25-gram samples in nylon mesh) were treated with gaseous SO_2 of a concentration level of 0.2%. The steeping procedures were performed and one sample and some steep water were removed after a period of time to estimate the water-extractable SO_2 in corn kernels and in steep water with the direct titrimetric method.

The sample was weighed after removal, and the amount of steep water to be removed was determined by the method of estimation given below, so that the ratio of weight of corn kernels to weight of steep water remained the same.

Table 4. Experimental Design of SO_2 Concentration Change During the Steeping Process.

Water-Extractable SO_2

Steep	Gaseous SO_2 steep		SO_2 Solution steep	
time	Corn kernels*	Steep water	Corn kernels	Steep water
0 Min	W_1	----	----	W_2
15 Min	W_1	W_2	W_1	W_2
30 Min	W_1	W_2	W_1	W_2
60 Min	W_1	W_2	W_1	W_2
2 Hrs	W_1	W_2	W_1	W_2
4 Hrs	W_1	W_2	W_1	W_2
8 Hrs	W_1	W_2	W_1	W_2
12 Hrs	W_1	W_2	W_1	W_2
16 Hrs	W_1	W_2	W_1	W_2
20 Hrs	W_1	W_2	W_1	W_2
24 Hrs	W_1	W_2	W_1	W_2
32 Hrs	----	----	W_1	W_2
40 Hrs	----	----	W_1	W_2
48 Hrs	----	----	W_1	W_2

* The concentration of water-extractable SO_2 in corn kernels is on the basis of the weight of dry corn kernels.

after a period of time,

weight of sample = M_1 g,

estimated weight of corn kernels = $(M_1/25)*1500$ g

= $60M_1$ g,

estimated weight of steep water = 2800 g + 1500 g − $60M_1$ g

= 4300 g − $60M_1$ g,

weight of steep water to be removed = M_2 g,

$$M_1/M_2 = (60\ M_1)/(4300 - 60M_1),$$

$$M_2 = 71.67 - M_1$$

<u>Changes of Concentration and Amount of SO_2 While Steeping with SO_2 Solution</u>: Fifteen hundred grams of corn kernels (including thirteen 25-gram samples) and some concentrate SO_2 solution were prepared to make a concentration level of 0.2%. The steeping procedures were performed and one sample and some steep water were removed after a period of time to estimate the water-extractable SO_2 in corn kernels and in steep water with the direct titrimetric method.

The sample was weighed after removal, and the amount of steep water to be removed was determined by the method of estimation as before, so that the ratio of weight of corn kernels to weight of steep water remained the same.

RESULTS AND DISCUSSION

Corn Wet Milling

Starch Yield

The experimental results of starch yield of corn kernels treated with gaseous SO_2 of different concentration levels and steeped conventionally for different periods of time are presented in Table 5 and Figure 2, which show that starch yield increased with increased steep time and concentration of SO_2 when gaseous SO_2 was applied. The addition of lactic acid in steep water also increased starch yield.

Table 5 also presents the results of statistical analysis of all sample means using Duncan's new multiple range test. It shows how much steep time can be saved to derive the same starch yield and how much starch yield can be increased with an increase of SO_2 concentration level, the addition of lactic acid, and the application of gaseous SO_2. For example, the starch yield of steeping with 0.4% gaseous SO_2 for 6 hours is within the same range as that of steeping with 0.2% gaseous SO_2 for 12 hours. Eighteen hours can be saved to obtain starch yields within the same range if corn kernels are steeped with 0.2% gaseous SO_2 and lactic acid rather than 0.4% gaseous SO_2; furthermore, 42 hours can be saved when it is applied instead of a 0.4% SO_2 solution.

Table 5. Starch Yields of Steeping with Different Concentration Levels of SO_2 and Different Periods of Time (Weight of Starch/Weight of Corn Kernels).

Starch Yield* (%, d.b.)

Steep time	Concentration level of SO_2 0.2% with lactic acid**	0.2%	0.4%
6 Hrs	67.43 ± 0.42 b***	61.79 ± 1.15 g	63.23 ± 0.24 f
12 Hrs	69.06 ± 0.25 a	64.10 ± 0.13 e f	66.18 ± 0.24 c
24 Hrs	69.98 ± 0.18 a	65.54 ± 0.58 c d	67.89 ± 0.18 b
48 Hrs****	69.12 ± 0.30 a	64.87 ± 0.28 d e	67.26 ± 0.17 b

* Each value represents the mean of two replicates with the standard deviation.

** The concentration of lactic acid is 0.55% based on the weight of steep water.

*** Mean comparisons followed by the same letter are not significantly different ($p<0.05$).

**** Corn kernels are steeped traditionally with SO_2 solution to serve as a control.

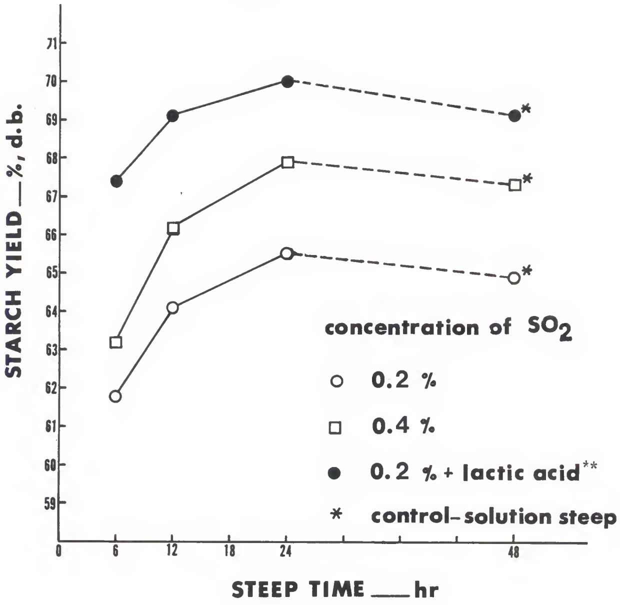


Figure 2. Starch Yields of Steeping with Different Concentration Levels of SO_2 For Different Periods of Time (Weight of Starch/Weight of Corn Kernels).

** The concentration of lactic acid is 0.55% based on the weight of steep water.

Effect of Steep Time on Starch Yields: Table 6 shows the effect of steep time on starch yields by statistical analysis. Duncan's new multiple range test was used for the four average starch yields of steeping for 6, 12, and 24 hours with gaseous SO_2 and 48 hours with SO_2 solution.

It was observed that the starch yield when steeping for 12 hours was in the same range as for the control when 0.2% gaseous SO_2 and lactic acid were applied. Also, there was no significant difference between the starch yields of steeping for 12 hours, for 24 hours or for the control when corn kernels were treated with 0.2% gaseous SO_2. However, the starch yields when steeping for 12 hours and for 24 hours with 0.4% gaseous SO_2 and for the control were significantly different.

In conclusion, when corn kernels were treated with gaseous SO_2 prior to steeping, steep time had a significant effect on starch yields, and 24 to 36 hours could be saved to obtain the same starch yield as the control. That is, 1/2 to 3/4 of the steep time could be saved by the application of gaseous SO_2 rather than SO_2 solution under same concentration level with or without the addition of lactic acid.

Effect of Concentration Level of SO_2 and Addition of Lactic Acid on Starch Yields: Table 7 presents the effect of the concentration level of SO_2 and the addition of lactic acid

Table 6. Effect of Steep Time on Starch Yields of Steeping with Same Concentration Level of SO_2 (Weight of Starch/Weight of Corn Kernels).

Concentration level of SO_2	Steep time	Starch yield* (%, d.b.)
0.2% with lactic acid**		
	6 Hrs	67.43 c***
	12 Hrs	69.06 b
	24 Hrs	69.98 a
	48 Hrs****	69.12 b
0.2%		
	6 Hrs	61.79 b
	12 Hrs	64.10 a
	24 Hrs	65.54 a
	48 Hrs****	64.87 a
0.4%		
	6 Hrs	63.23 d
	12 Hrs	66.18 c
	24 Hrs	67.89 a
	48 Hrs****	67.26 b

* Each value represents the mean of two replicates.

** The concentration of lactic acid is 0.55% based on the weight of steep water.

*** Mean comparisons within the same group followed by the same letter are not significantly different ($p<0.05$).

**** Corn kernels are steeped traditionally with SO_2 solution to serve as a control.

Table 7. Effect of Concentration Level of SO_2 and Addition of Lactic Acid on Starch Yields of Steeping For Same Period of Time (Weight of Starch/Weight of Corn Kernels).

Steep time Concentration level of SO_2	Starch yield* (%, d.b.)
6 Hrs	
0.2% with lactic acid**	67.43 a***
0.4%	63.23 b
0.2%	61.79 b
12 Hrs	
0.2% with lactic acid**	69.06 a
0.4%	66.18 b
0.2%	64.10 c
24 Hrs	
0.2% with lactic acid**	69.98 a
0.4%	67.89 b
0.2%	65.54 c
48 Hrs****	
0.2% with lactic acid**	69.12 a
0.4%	67.26 b
0.2%	64.87 c

* Each value represents the mean of two replicates.

** The concentration of lactic acid is 0.55% based on the weight of steep water.

*** Mean comparisons within the same group followed by the same letter are not significantly different ($p<0.05$).

**** Corn kernels are steeped traditionally with SO_2 solution to serve as a control.

on starch yield by Duncan's new range test, which was used for the three average starch yields of steeping with 0.2% SO_2, 0.4% SO_2 and 0.2% SO_2 with lactic acid.

Consistently, corn kernels steeped with 0.2% SO_2 with lactic acid had the highest starch yields, and those steeped with 0.2% SO_2 had the lowest starch yields, with the exception of starch yields of steeping with 0.2% and 0.4% gaseous SO_2 for 6 hours, which were not significantly different.

It can also be observed from Table 7 that the difference between starch yields when steeping with 0.2% and 0.4% SO_2 increased as steep time increased. However, the difference between starch yields when steeping with 0.2% SO_2 plus lactic acid compared to SO_2 only decreased when steep time was increased.

This indicates that the addition of lactic acid can release starch granules from the protein matrix faster, and the effect of the level of SO_2 treatment becomes more significant when steep time is increased.

Starch Quality

Protein content in starch and consistency of the starch slurry are the two important criteria to determine starch quality. Starch is considered to be of high quality when it has a low protein content and a high consistency of its slurry.

Protein Content in Starch: Table 8 gives the protein content in starch recovered from corn kernels when steeping with different levels of SO_2 treatments for different periods of time. Compared to the average protein content of bench-scale wet milling given in Anderson (1963), of which the percentage is 0.54 ± 0.02, the protein contents in Table 8 are satisfactorily lower.

However, no relationship was observed between protein content and steep time, concentration level of SO_2, the application of gaseous SO_2, or the addition of lactic acid, which can reduce the protein content in starch according to previous work (Roushdi et al., 1981b). On the other hand, it did not show that the protein content was increased with the application of gaseous SO_2 rather than SO_2 solution.

Consistency of Starch Slurry: The consistencies of slurries of starch recovered from corn kernels by steeping with different concentration levels of SO_2 for different periods of time are shown in Table 9 and Figure 3. It was observed that the consistency of starch slurry decreased with an increase of steep time and concentration level of SO_2 and the addition of lactic acid.

Table 10 shows the effect of steep time on the

Table 8. Protein Contents in Starch Recovered from Corn Kernels When Steeping with Different Concentration Levels of SO_2 For Different Periods of Time.

Protein Content in Starch* (%, d.b.)

Steep time	Concentration level of SO_2		
	0.2% with lactic acid**	0.2%	0.4%
6 Hrs	0.255 0.300	0.356 0.332	0.387 0.375
12 Hrs	0.332 0.342	0.315 0.281	0.427 0.383
24 Hrs	0.404 0.386	0.460 0.366	0.352 0.380
48 Hrs***	0.362 0.332	0.369 0.395	0.331 0.317

* Each value is the mean of two samples from starch of one replicate.

** The concentration of lactic acid is 0.55% based on the weight of steep water.

*** Corn kernels are steeped traditionally with SO_2 solution to serve as a control.

Table 9. Consistencies of Slurries of Starch Recovered from Corn Kernels When Steeping with Different Concentration Levels of SO_2 For Different Periods of Time.

Consistency* (Brabender unit)

Steep time	Concentration level of SO_2		
	0.2% with lactic acid**	0.2%	0.4%
6 Hrs	1175 b*** c	1255 a	1130 c
12 Hrs	1165 b c	1210 a	1060 d e
24 Hrs	1160 b c	1190 b	1030 d e
48 Hrs****	1010 e	1070 d	935 f

* Each value represents the mean of two replicates.

** The concentration of lactic acid is 0.55% based on the weight of steep water.

*** Mean comparisons followed by the same letter are not significantly different ($p<0.05$).

**** Corn kernels are steeped traditionally with SO_2 solution to serve as a control.

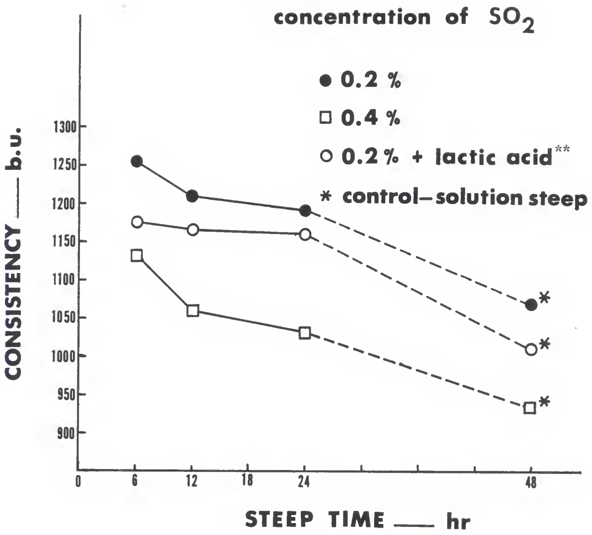


Figure 3. Consistencies of Slurries of Starch Recovered from Corn Kernels by Steeping with Different Concentration Levels of SO_2 For Different Periods of Time.

** The concentration of lactic acid is 0.55% based on the weight of steep water.

Table 10. Effect of Steep Time on Consistencies of Slurries of Starch Recovered from Corn Kernels When Steeping with Same Concentration Level of SO_2.

Concentration level of SO_2	Steep time	Consistency* (Brabender unit)
0.2% with lactic acid**		
	6 Hrs	1175 a***
	12 Hrs	1165 a
	24 Hrs	1160 a
	48 Hrs****	1010 b
0.2%		
	6 Hrs	1255 a
	12 Hrs	1210 a b
	24 Hrs	1190 b
	48 Hrs****	1070 c
0.4%		
	6 Hrs	1130 a
	12 Hrs	1060 a b
	24 Hrs	1030 b
	48 Hrs****	935 c

* Each value represents the mean of two replicates.

** The concentration of lactic acid is 0.55% based on the weight of steep water.

*** Mean comparisons within the same group followed by the same letter are not significantly different ($p<0.05$).

**** Corn kernels are steeped traditionally with SO_2 solution to serve as a control.

consistencies of starch slurries. Duncan's new multiple range test was used for the four average consistencies of slurries of starch recovered from corn kernels steeped for 6, 12, and 24 hours with gaseous SO_2 and 48 hours with SO_2 solution. In each of the three treatments, there was no significant difference between consistencies of slurries of starch recovered from corn kernels steeped for 6 and 12 hours, and steeped for 12 and 24 hours; however, they were all higher than their controls.

The effect of the concentration level of SO_2 and the addition of lactic acid on consistencies of starch slurries is shown in Table 11. Duncan's new range test was used for the three average consistencies of slurries of starch recovered from corn kernels steeped with 0.2% SO_2, 0.4% SO_2 and 0.2% SO_2 with lactic acid. For the six-hour steeping, those steeped with 0.2% SO_2 with lactic acid had lower consistencies of starch slurries than those steeeped with 0.2% SO_2 only. They also had higher consistencies of starch slurries than those steeped with 0.4% SO_2 for 12 and 24 hours. When steep time was increased, the addition of lactic acid showed less effect on the consistencies of starch slurries than the increase of the concentration level of SO_2.

Table 11. Effect of Concentration Level of SO_2 and Addition of Lactic Acid on Consistencies of Slurries of Starch Recovered from Corn Kernels When Steeping For Same Period of Time.

Steep time / Concentration level of SO_2	Consistency* (Brabender unit)
6 Hrs	
0.2%	1255 a**
0.2% with lactic acid***	1175 b
0.4%	1130 b
12 Hrs	
0.2%	1210 a
0.2% with lactic acid***	1165 a
0.4%	1060 b
24 Hrs	
0.2%	1190 a
0.2% with lactic acid***	1160 a
0.4%	1030 b
48 Hrs****	
0.2%	1070 a
0.2% with lactic acid***	1010 b
0.4%	935 c

* Each value represents the mean of two replicates.

** Mean comparisons within the same group followed by the same letter are not significantly different ($p<0.05$).

*** The concentration of lactic acid is 0.55% based on the weight of steep water.

**** Corn kernels aie steeped traditionally with SO_2 solution to serve as a control.

Changes of Concentration and Amount of SO_2

Changes of Concentration and Amount of SO_2 While Steeping with Gaseous SO_2

When corn kernels were steeped with 0.2% gaseous SO_2 for 24 hours, the concentrations of water-extractable SO_2 in corn kernels and in steep water varied, as shown in Table 12 and Figure 4. The estimated weights of corn kernels and steep water are given in Table 13 and were used to determine the total amount of SO_2, which is shown in Table 14 and Figure 5.

Changes of Concentration and Amount of SO_2 While Steeping with SO_2 Solution

When corn kernels were steeped traditionally with 0.2% SO_2 solution for 48 hours, the concentrations of water-extractable SO_2 in corn kernels and in steep water varied, as shown in Table 15 and Figure 6. The estimated weights of corn kernels and steep water are given in Table 16 and are used to determine the total amount of SO_2, which is shown in Table 17 and Figure 7.

From Figures 4 and 6, it was observed that the concentrations of water-extractable SO_2 in steep water and in corn kernels varied in a similar way when gaseous SO_2

Table 12. Concentrations of Water-Extractable SO_2 in Corn Kernels and Steep Water While Steeping with Gaseous SO_2.

	Concentration of Water-Extractable SO_2 (ppm)	
Steep time	In corn kernels (d.b.)	In steep water
0 Min	1785	---
15 Min	610	560
30 Min	555	545
60 Min	680	455
2 Hrs	720	345
4 Hrs	760	325
8 Hrs	720	295
12 Hrs	620	310
16 Hrs	510	375
20 Hrs	490	390
24 Hrs	485	395

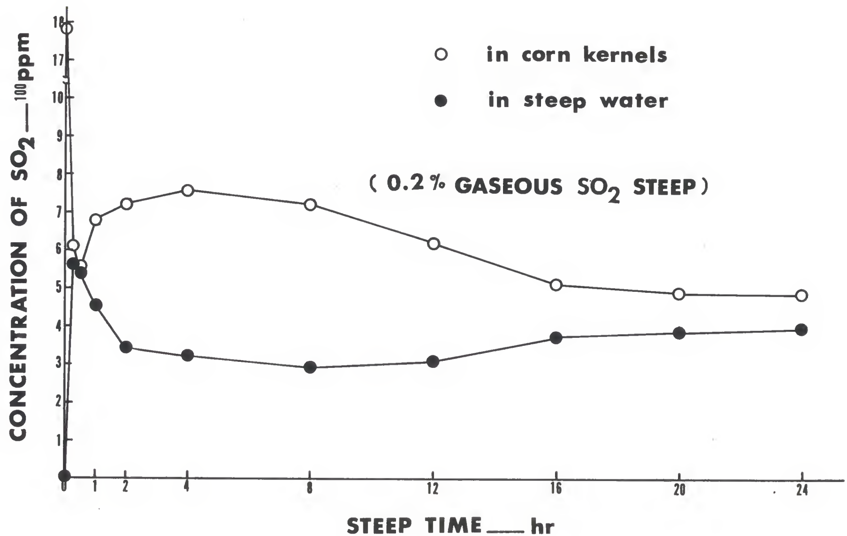


Figure 4. Concentrations of Water-Extractable SO_2 in Corn Kernels and Steep Water While Steeping with Gaseous SO_2.

Table 13. Estimated Weights of Corn Kernels and Steep Water While Steeping with Gaseous SO_2.

	Estimated Weight (gram)	
Steep time	Corn kernels	Steep water
0 Min	1500	2800
15 Min	1670	2630
30 Min	1715	2585
60 Min	1755	2545
2 Hrs	1875	2425
4 Hrs	1980	2320
8 Hrs	2130	2170
12 Hrs	2170	2130
16 Hrs	2195	2105
20 Hrs	2205	2095
24 Hrs	2215	2085

Table 14. Weights of Water-Extractable SO_2 in Corn Kernels and Steep Water While Steeping with Gaseous SO_2.

	Weight of Water-Extractable SO_2 (gram)		
Steep time	In corn kernels	In steep water	Total
0 Min	2.31	----	2.31
15 Min	0.79	1.47	2.26
30 Min	0.71	1.41	2.12
60 Min	0.88	1.16	2.04
2 Hrs	0.93	0.84	1.77
4 Hrs	0.98	0.75	1.73
8 Hrs	0.93	0.64	1.57
12 Hrs	0.80	0.66	1.46
16 Hrs	0.66	0.79	1.45
20 Hrs	0.63	0.82	1.44
24 Hrs	0.63	0.82	1.44

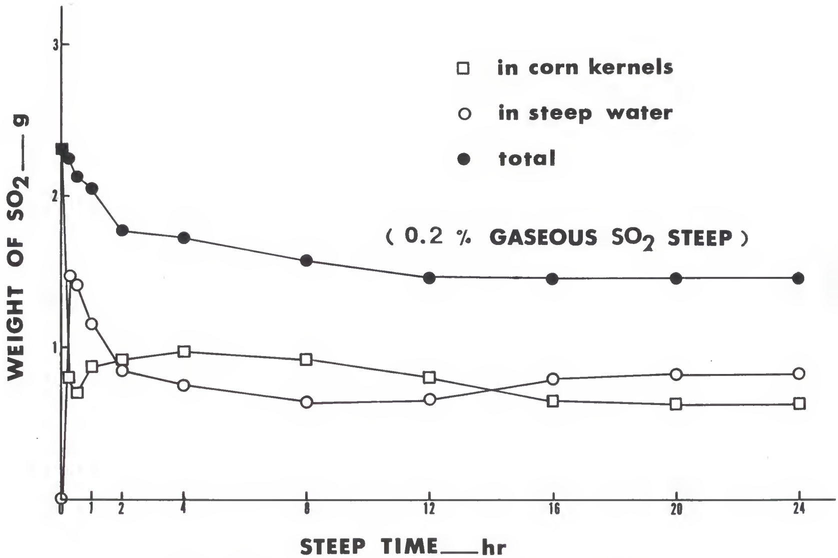


Figure 5. Weights of Water-Extractable SO_2 in Corn Kernels and Steep Water While Steeping with Gaseous SO_2.

Table 15. Concentrations of Water-Extractable SO_2 in Corn Kernels and Steep Water While Steeping with SO_2 Solution.

	Concentration of Water-Extractable SO_2 (ppm)	
Steep time	In corn kernels (d.b.)	In steep water
0 Min	---	1070
15 Min	345	760
30 Min	550	580
60 Min	640	445
2 Hrs	730	335
4 Hrs	810	235
8 Hrs	780	185
12 Hrs	640	265
16 Hrs	555	320
20 Hrs	485	325
24 Hrs	430	325
32 Hrs	400	325
40 Hrs	390	330
48 Hrs	390	330

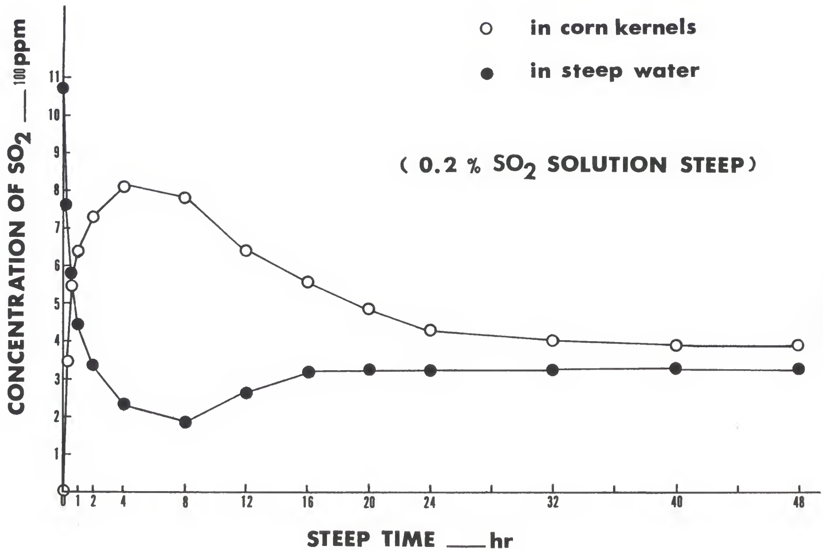


Figure 6. Concentrations of Water-Extractable SO_2 in Corn Kernels and Steep Water While Steeping with SO_2 Solution.

Table 16. Estimated Weights of Corn Kernels and Steep Water While Steeping with SO_2 Solution.

	Estimated Weight (gram)	
Steep time	Corn kernels	Steep water
0 Min	1500	2800
15 Min	1705	2595
30 Min	1710	2590
60 Min	1785	2515
2 Hrs	1860	2440
4 Hrs	1930	2370
8 Hrs	2080	2220
12 Hrs	2155	2145
16 Hrs	2210	2090
20 Hrs	2215	2085
24 Hrs	2220	2080
32 Hrs	2225	2075
40 Hrs	2230	2070
48 Hrs	2230	2070

Table 17. Weights of Water-Extractable SO_2 in Corn Kernels and Steep Water While Steeping with SO_2 Solution.

Weight of Water-Extractable SO_2 (gram)

Steep time	In corn kernels	In steep water	Total
0 Min	----	3.00	3.00
15 Min	0.45	1.97	2.42
30 Min	0.71	1.50	2.21
60 Min	0.83	1.12	1.95
2 Hrs	0.94	0.82	1.76
4 Hrs	1.05	0.56	1.61
8 Hrs	1.01	0.41	1.42
12 Hrs	0.83	0.57	1.40
16 Hrs	0.72	0.67	1.39
20 Hrs	0.63	0.68	1.31
24 Hrs	0.56	0.68	1.24
32 Hrs	0.52	0.67	1.19
40 Hrs	0.50	0.68	1.18
48 Hr	0.50	0.68	1.18

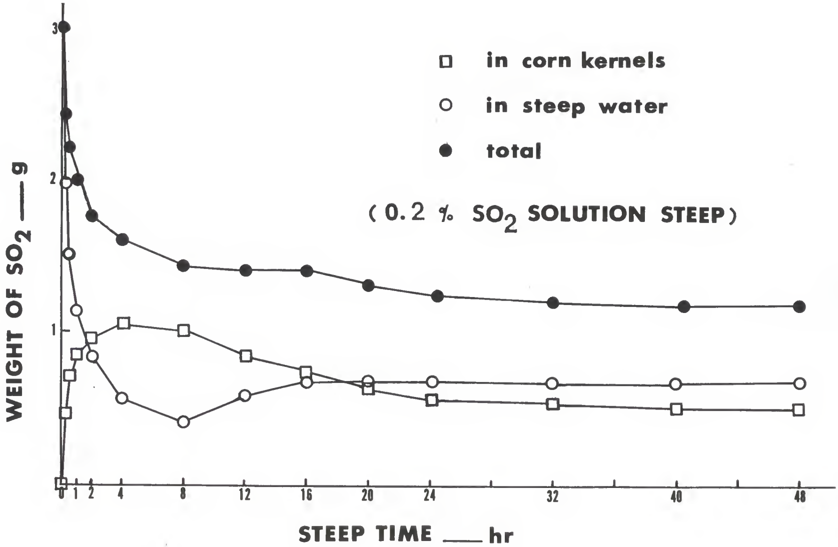


Figure 7. Weights of Water-Extractable SO_2 in Corn Kernels and Steep Water While Steeping with SO_2 Solution.

and SO_2 solution were applied. The concentration of SO_2 in corn kernels reached their highest values after 4 hours and gradually decreased. However, for the same steep time, the concentration of SO_2 in steep water was higher when gaseous SO_2 was applied rather than SO_2 solution. In corn kernels, the reverse was true. This indicated that the amount of gaseous SO_2 which diffused into the endosperm was larger than that of SO_2 solution for the same steep time. It also confirmed the increase of speed of penetration into the endosperm with the application of gaseous SO_2.

From Figures 5 and 7, it is shown that, for the same steep time, the total amounts of SO_2 in corn kernels and in steep water were about the same when corn kernels were treated with gaseous SO_2 and SO_2 solution, even though about 23% of gaseous SO_2 was lost to the air before steeping. In addition, the total amount of SO_2 remained constant after 16 to 20 hours when gaseous SO_2 was applied, and it remained constant after 32 to 40 hours when SO_2 solution was applied. That is, about one-half of the steep time could be saved to complete the SO_2 reaction in steeping if 0.2% gaseous SO_2 was used rather than 0.2% SO_2 solution.

CONCLUSIONS AND SUGGESTIONS FOR FUTURE RESEARCH

The application of gaseous SO_2 in corn wet milling by treating corn kernels prior to steeping could save steep time significantly. Generally, with the same amounts of steeping agents, starch yields of steeping traditionally for 48 hours with SO_2 solution were statistically the same as those of steeping for 12 hours, and were all less than those of steeping for 24 hours with gaseous SO_2.

The increase of the concentration level of gaseous SO_2 resulted in the increase of the starch yield. When the concentration level of SO_2 was increased from 0.2% to 0.4%, starch yield increased 1.44%, 2.08%, and 2.35% for 6-, 12-, and 24-hour steeps, respectively.

Lactic acid made a greater contribution to starch yields than the increase of the concentration level of gaseous SO_2. With the addition of 0.55% lactic acid (based on the weight of steep water) to steep water after 0.2% gaseous SO_2 treatment, starch yield was raised from 61.79% to 67.43% for a 6-hour steep, 64.10% to 69.06% for a 12-hour steep, and 65.54% to 69.98% for a 24-hour steep.

The use of gaseous SO_2 does not have a detrimental effect on the quality of the starch. In addition, due to the reduction of steep time, consistencies of slurries of starch recovered from corn kernels treated with gaseous SO_2 were increased significantly when compared to their controls. However, no effect was found on the protein content in starch made by the applications of gaseous SO_2 and lactic acid, but the protein contents in starch of all of the experiments in this research work were rather low and varied in a range from 0.255% to 0.460%.

In conclusion, steeping, the energy intensive and rate determining step of corn wet-milling processes, can be improved by reducing the steep time and increasing the starch yield achieved from the application of gaseous SO_2 and the addition of lactic acid.

In future research, more details should be taken into consideration so that a model could be established to predict the most economical way to recover starch with an expected quality to an optimum amount. Since consistency and starch yield vary with steep time, concentration level of SO_2, and the addition of lactic acid, there are some limits in the selections of them. If an accurate and precise model can be established, the most economical way can be figured out with the considerations of steep time, costs of steeping agents, etc.

Furthermore, it is justifiable to investigate whether steep time and the amount of SO_2 could be reduced by performing the treatment sometime during the steeping process. It is observed in this study that about 20% of gaseous SO_2 was lost to the air before steeping, and about 30% of SO_2 was consumed (reacting or lost to the air) during the steeping process when a 0.2% gaseous SO_2 treatment was applied. Obviously, steep time and the amount of SO_2 can saved if the treatment is performed at a particular time during the steeping process, at which the protein matrix is under the most favorable condition for reacting with SO_2.

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APPENDIX

Records of Experiments

	Experiment no.			
	1	2	3	4
M.C. (%. w.b.)	13.78	14.01	13.98	13.77
Steeping agent*	G	G	G	G
SO_2 concentration**	0.2%	0.2%	0.2%	0.2%
Concentration of lactic acid***	0.55%	0.55%	----	----
Steep time (hr)	6	6	6	6
pH of steep water	2.89	2.97	4.03	4.12
Product yields (%. d.b.)				
Germ	5.99	5.85	5.89	6.02
Fiber	10.67	10.52	9.41	10.58
Starch	67.13	67.73	60.98	62.60
Gluten	10.52	10.27	14.88	13.98
Soluble solids	4.71	4.66	1.12	1.51
Consistency (b.u.)	1180	1170	1270	1240
Protein content in starch (%, d.b.)	0.255	0.300	0.356	0.332

* S = SO_2 solution, G = Gaseous SO_2
** SO_2 concentration is based on the weight of corn kernels.
*** Concentration of lactic acid is based on the weight of step water.

(Continued)

	Experiment no.			
	5	6	7	8
M.C. (%. w.b.)	13.48	13.69	14.21	13.95
Steeping agent*	G	G	G	G
SO_2 concentration**	0.4%	0.4%	0.2%	0.2%
Concentration of lactic acid***	----	----	0.55%	0.55%
Steep time (hr)	6	6	12	12
pH of steep water	3.65	3.71	3.28	3.22
Product yields (%. d.b.)				
Germ	7.63	7.45	6.34	6.11
Fiber	13.00	12.25	9.58	9.43
Starch	63.06	63.40	69.24	68.88
Gluten	9.74	10.32	10.64	10.91
Soluble solids	5.61	5.42	3.83	3.82
Consistency (b.u.)	1140	1120	1180	1150
Protein content in starch (%, d.b.)	0.387	0.375	0.332	0.342

* S = SO_2 solution, G = Gaseous SO_2
** SO_2 concentration is based on the weight of corn kernels.
*** Concentration of lactic acid is based on the weight of step water.

(Continued)

	Experiment no.			
	9	10	11	12
M.C. (%. w.b.)	13.70	13.59	13.66	14.10
Steeping agent*	G	G	G	G
SO_2 concentration**	0.2%	0.2%	0.4%	0.4%
Concentration of lactic acid***	----	----	----	----
Steep time (hr)	12	12	12	12
pH of steep water	4.48	4.56	4.04	4.00
Product yields (%. d.b.)				
Germ	5.50	5.96	6.95	6.22
Fiber	11.87	11.32	10.36	11.65
Starch	64.01	64.19	66.35	66.01
Gluten	11.38	11.23	10.05	10.25
Soluble solids	2.39	2.29	4.68	4.55
Consistency (b.u.)	1200	1220	1080	1040
Protein content in starch (%, d.b.)	0.315	0.281	0.427	0.383

* S = SO_2 solution, G = Gaseous SO_2

** SO_2 concentration is based on the weight of corn kernels.

*** Concentration of lactic acid is based on the weight of step water.

(Continued)

	Experiment no.			
	13	14	15	16
M.C. (%. w.b.)	13.60	13.55	13.62	13.87
Steeping agent*	G	G	G	G
SO_2 concentration**	0.2%	0.2%	0.2%	0.2%
Concentration of lactic acid***	0.55%	0.55%	----	----
Steep time (hr)	24	24	24	24
pH of steep water	3.69	3.79	5.23	5.36
Product yields (%. d.b.)				
Germ	6.49	6.92	5.92	6.10
Fiber	9.48	8.82	11.25	10.82
Starch	69.85	70.11	65.13	65.95
Gluten	9.83	9.77	12.07	11.26
Soluble solids	4.05	3.97	4.20	4.33
Consistency (b.u.)	1140	1180	1170	1210
Protein content in starch (%, d.b.)	0.404	0.386	0.460	0.366

* S = SO_2 solution, G = Gaseous SO_2
** SO_2 concentration is based on the weight of corn kernels.
*** Concentration of lactic acid is based on the weight of step water.

(Continued)

	Experiment no.			
	17	18	19	20
M.C. (%. w.b.)	13.85	13.50	14.05	13.78
Steeping agent*	G	G	S	S
SO_2 concentration**	0.4%	0.4%	0.2%	0.2%
Concentration of lactic acid***	----	----	0.55%	0.55%
Steep time (hr)	24	24	48	48
pH of steep water	4.55	4.45	3.81	3.90
Product yields (%. d.b.)				
Germ	5.90	5.93	6.37	5.86
Fiber	8.93	9.33	8.06	8.21
Starch	68.02	67.76	69.33	68.91
Gluten	10.51	10.89	10.07	9.80
Soluble solids	5.93	5.04	5.23	6.09
Consistency (b.u.)	1000	1060	1010	1010
Protein content in starch (%, d.b.)	0.352	0.380	0.362	0.332

* S = SO_2 solution, G = Gaseous SO_2
** SO_2 concentration is based on the weight of corn kernels.
*** Concentration of lactic acid is based on the weight of step water.

(Continued)

	Experiment no.			
	21	22	23	24
M.C. (%. w.b.)	14.50	13.96	13.86	13.92
Steeping agent*	S	S	S	S
SO_2 concentration**	0.2%	0.2%	0.4%	0.4%
Concentration of lactic acid***	----	----	----	----
Steep time (hr)	48	48	48	48
pH of steep water	5.72	5.64	4.56	4.50
Product yields (%. d.b.)				
Germ	5.95	5.88	6.12	5.85
Fiber	12.07	11.19	8.84	8.79
Starch	64.67	65.07	67.38	67.14
Gluten	8.86	9.42	9.22	10.27
Soluble solids	4.47	4.58	6.11	6.23
Consistency (b.u.)	1060	1080	950	920
Protein content in starch (%, d.b.)	0.369	0.395	0.331	0.317

* S = SO_2 solution, G = Gaseous SO_2
** SO_2 concentration is based on the weight of corn kernels.
*** Concentration of lactic acid is based on the weight of step water.

WET MILLING OF CORN USING GASEOUS SO_2 ADDITION PRIOR TO STEEPING AND THE EFFECT OF LACTIC ACID ON STEEPING

by

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ABSTRACT

Corn wet milling is a process for the recovery of starch from corn kernels. It is energy intensive, with a high percentage of the energy being used for steeping. The objectives of this study are to determine the relationship between starch yield, steep time, concentration of SO_2, and the addition of lactic acid to steep water when a gaseous SO_2 treatment prior to steeping is applied, and to help determine the most economical method of steeping in corn wet milling.

In this study, steep time (6, 12 and 24 hours), the concentration level of gaseous SO_2 (0.2 and 0.4%), and the addition of lactic acid were the variables in the experimental design while a 48-hour traditional steep served as a control.

It was observed that starch yield of the controls were statistically the same as those of steeping for 12 hours, and were all less than those of steeping for 24 hours with gaseous SO_2. That is, 1/2 to 3/4 of steep time was saved by the application of gaseous SO_2. The quality of starch was also shown as good by the application of gaseous SO_2 because of the reduction of steep time. Increasing the concentration level of gaseous SO_2 from 0.2% to 0.4% resulted in an increased starch yield of 1.44% to 2.35% for the 6- to 24-hour steeps. With the addition of 0.55% lactic acid to the

steep water after a 0.2% gaseous SO_2 treatment, starch yield was increased by 5.64%, 4.96%, and 4.44% for the 6-, 12-, and 24-hour steeps, respectively.