Effect of Soak Time, Soak Temperature, and Lactic Acid on Germ Recovery Parameters

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ABSTRACT

To recover high valued corn germ from dry-milling ethanol facilities, corn was hydrated in under 12 hr and germ was recovered using traditional wet-milling recovery techniques. The optimal soak time and temperature for germ recovery was 12 hr at 59°C. Germ was recovered by flotation and skimming. Recovery levels of germ were near that of traditional wet milling. Results showed that soak time decreased with increasing soak temperature up through 59°C. A reduction in germ quality and quantity occurred at soak temperature of 75°C.

Two main processes are used for ethanol production from corn: 1) wet milling (Fig. 1), and 2) dry milling (Fig. 2). Wet milling accounts for ≈60% of total ethanol production (Rendleman and Hohmann 1993). Dry-milling ethanol plants cost less to build and produce higher yields of ethanol, ≈387 L/MT (2.6 gallons per bushel) compared to 372 L/MT (2.5 gallons per bushel) for wet mills, but the coproduct are valued lower (Rendleman and Hohmann 1993). The reason dry-milling ethanol plants give higher yields of ethanol (≈0.1 gal/bushel more than wet milling) is because, in wet mills, some amount of starch is lost in fiber and gluten fractions. Ethanol producers receive substantial revenues from the sale of coproducts which offset the purchase cost of corn. From 1981 to 1991, the average net corn cost was $0.44/gal in a wet-milling plant and $0.53/gal in a dry-milling ethanol plant (Rendleman and Hohmann 1993). Lewis and Gimes (1988) reported that the net corn cost difference between wet-milling and dry-milling ethanol plants was $0.15/gal.

Dry-milling ethanol production is expected to benefit from germ recovery before fermentation. Corn oil is separated in wet mills by recovering the germ, but in the dry milling process, the nonfermentable germ, meal, and oil is lost to the distillers dried grains (DDG) livestock feed product. Watson et al (1969) reported that corn germ can be recovered by kernel hydration or soaking for short times (3–6 hr), followed by conventional wet milling. Since the germ is the first part of the kernel to fully hydrate, it is not necessary to soak the corn for 24–36 hr as used in conventional steeping. It is also not necessary to soak the kernels in sulfur dioxide because the role of sulfur dioxide in steeping is in enhancing starch protein separation, not germ separation.

The new process, which we call dry-grind process (Fig. 3), will recover the germ in a dry-milling process using the degermination facility of the wet-milling process. This dry-grind process is a combination of dry milling and wet milling processes. To commercialize the dry-grind process, we need information on the optimal soak time and temperature to maximize the germ recovery and oil content in germ.

The objectives of this study were: 1) to determine the effect of soak time and temperature on germ yield, germ oil content, soak water solids, and specific gravity required to float the germ; and 2) to determine the effect of lactic acid on germ yield, germ oil content, soak water solids, and specific gravity required to float the germ.

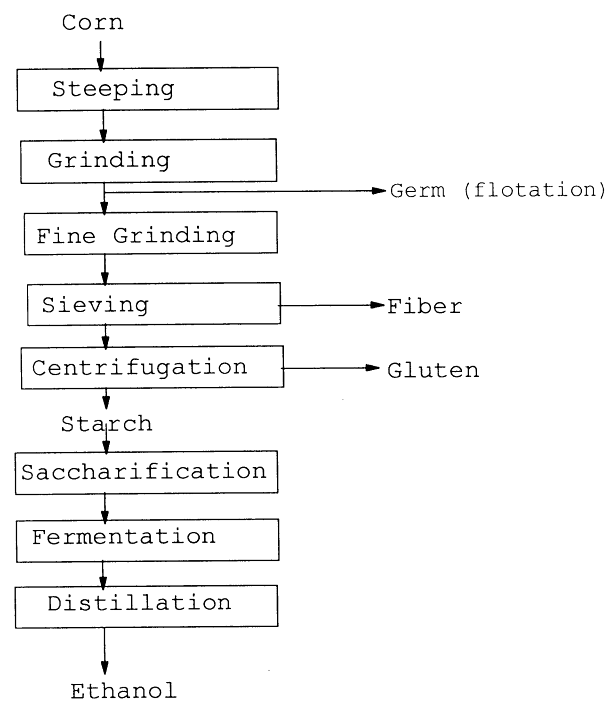

Fig. 1. Wet-milling process for ethanol production.

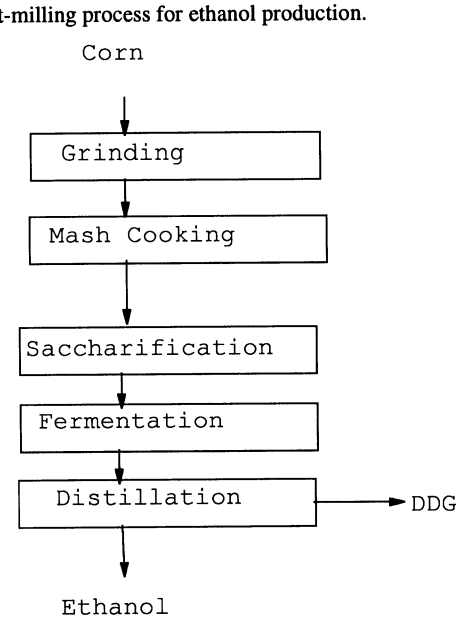

Fig. 2. Dry-milling process for ethanol production.

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[2]Professor, Department of Agricultural Engineering. University of Illinois, Urbana, IL 61801. Mention of product or trade names does not imply endorsement by the University of Illinois.

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MATERIALS AND METHODS

Yellow dent corn (FR600×Mo17), grown during the 1993 season at the agricultural engineering farm at the University of Illinois at Urbana-Champaign, was combine-harvested at 16.5% mc (wb) and ambient dried to 14% mc (wb). The corn was stored in 190 L (55 gallon) sealed steel drums at ambient conditions for approximately six months (September 1993 through February 1994) before the start of testing. Corn was mixed in drums and ≈1,200 g of sample was taken and manually sieved over a 4.8-mm (12/64") round-hole screen. Molded corn and foreign material were hand-picked and discarded; such corn generally represented <2 g per sample. From this screened, hand-picked sample, 1 kg was weighed and placed in a plastic bag and equilibrated overnight to ambient temperature before steeping. The remaining hand-picked corn was used for triplicate moisture content determination using standard methods (AACC 1983).

Effect of Soak Time and Soak Temperature

Two full-factorial experiments were performed to determine the effect of soak time and soak temperature on germ recovery parameters. Both the experiments were done in duplicate. In the first experiment, 1-kg samples were batch-soaked in 2,000 ml of distilled water for 3, 6, 9, or 12 hr at 45, 52, 59, or 75°C. Germ was recovered using the laboratory wet-milling procedure of Eckhoff et al (1993). The germ yield was then compared statistically across the levels of soak time and soak temperature. At 45°C, low germ yields were obtained as compared to soak temperatures of 52 and 59°C. At 75°C, effects of starch gelatinization could be observed and there was difficulty in floating the germ. Based on the these results, a second experiment was done in which 1-kg corn samples were batch-soaked in 2,000 ml of distilled water for 4.5, 6, 7.5, or 9 hr at 52 or 59°C. Germs were recovered using the laboratory wet-milling procedure of Eckhoff et al (1993). The data sets from both experiments were merged and analyzed as one set.

After soaking, the soak water was pumped out of the tank into a 2,000-ml graduated cylinder using a variable speed Cole-Parmer peristaltic pump and the soak water volume was recorded. Soak water samples were placed in preweighed aluminum cups and dried for dry solids content determination using a two-staged forced air-oven method (AACC 1983). Solids content were expressed as percentage of the sample weight.

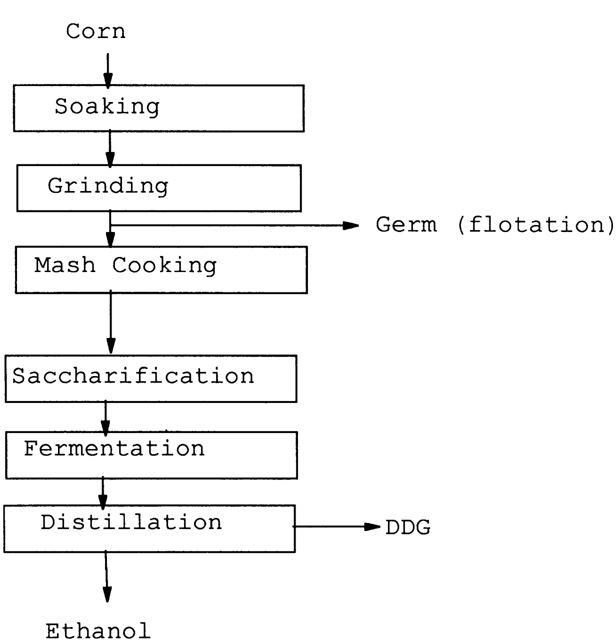

Fig. 3. Dry-grind process for ethanol production.

The soaked corn was ground based on the procedures outlined by Eckhoff et al (1993). One modification to this method was setting the speed of the blender at 40% of full power for 3 min (medium blender speed) and 46% full power for another 3 min (medium blender speed). A powerstat transformer (model 116, Superior Electric Co., Bristol, CT) was used to control motor speed. These settings are based on preliminary tests that gave the best quality of germ. In these preliminary tests, soaked corn was ground with equal volumes of water at various speed settings. The qualitative measure used to determine the speed settings was the germ quality, which is defined as minimal broken germ and no whole kernels at the end of the grind.

The initial specific gravity of the slurry was measured by hygrometer, and dry starch was added to the slurry until good germ flotation was observed (when all the corn germ floating on the surface have no endosperm particles floating along). The final specific gravity of slurry was then recorded.

The germ was skimmed using the procedure outlined by Eckhoff et al (1993). The germ samples were collected in a weigh boat and then dried for moisture content determination by the two-stage forced-air oven procedure (AACC 1983). Oil content of germ samples was determined by solvent extraction method (AOCS 1988).

Effect of Lactic Acid

Samples (1 kg) of corn were batch soaked for 7.5 hr at 59°C in 2,000 ml of distilled water using one of three levels of lactic acid concentration (0, 0.1, or 0.5%,w/w). The soaked corn was milled and the germ was recovered as previously described, except the speed of the blender had to be reduced to get the best quality results (minimal broken germ and no whole kernels at the end of the grind). A preliminary study for speed setting was also done for the corn soaked in distilled water with lactic acid. The blender speed that gave good quality germ for corn soaked in distilled water with lactic acid was 36% of its full power for 3 min and 42% of full power for another 3 min (medium blender speed) compared to 40 and 46% for 3 min each in the soak time and soak temperature study. Germ quality is same as defined for soak time and soak temperature. Germ yield, germ oil content, soak water soluble content, and the specific gravity required to float the germ were measured for each condition.

Statistical Analysis

Analysis of variance (ANOVA) and Duncan's multiple range test were used (SAS 1985) with $\alpha = 5\%$ ($P < 0.05$). Regression analysis was also done to determine the relationship between the average germ yield and soak time and soak temperature. Variable selection for model was done using forward selection with regression procedure (SAS 1985). Three-dimensional response surface for average germ yield, average oil content, soak water solids, and specific gravity required to float the germ with the independent variables of time and temperature were generated using SAS/GRAPH software (SAS 1985).

RESULTS AND DISCUSSION

Effect of Soak Time and Soak Temperature

Tables I–IV summarize the effect of soak time and soak temperature on germ yield, germ oil content, soak water solids, and specific gravity required to float the germ.

Germ Yields

The effect of time and temperature on germ yield were statistically significant ($P < 0.05$) (Table I). Results showed that germ yield was generally highest at soak temperature of 59°C. Only germ yield at 9 hr of soak time at 75°C was actually at the same level as the germ yield at 59°C. All the other soak times at 75°C showed germ yields at levels similar to those at soak temperature of 52°C. At 75°C, effects of starch gelatinization could be observed and there was difficulty in floating the germ. Soaking at 45°C resulted in a germ yield 1.8–2.5 percentage points lower. Surprisingly, soaking at 52°C (normal steeping temperature used in wet milling [Watson 1984]) showed a germ yield 0.82–1.2 percentage points lower when compared to soaking at 59°C. In fact, average germ yield for 52°C at 12 hr of soaking was not statistically different from germ yield for 3 hr of soaking at 59°C.

Germ yields increased with increasing soak time at all soak temperatures except 75°C. Soaking at 75°C for 12 hr resulted in a lower germ yield than did soaking for 6 or 9 hr. This was due to excessive starch gelatinization and difficulty in floating the germ.

The interaction effect between soak time and soak temperature was significant. Thus, it was not possible to determine the effect of soak time independently of soak temperature. The factorial structure was ignored in the statistical analysis and a one-way ANOVA was performed for the time-temperature combination and germ yield.

The maximum average germ yield of 6.76% was obtained at a time and temperature of 12 hr and 59°C followed by average germ yields of 6.41 and 6.27% at time-temperature combinations of 9 hr, 59°C and 7.5 hr, 59°C, respectively. No significant difference was found for germ yields obtained at 9 hr and 7.5 hr at 59°C.

The lowest average germ yield of 4.03% was obtained at a time temperature combination of 3 hr and 45°C. Response surface of germ yield with independent variables of soak time and soak temperature (Fig. 4) shows the maximum average germ yield of 6.76% at soak time of 12 hr and soak temperature of 59°C. At 59°C, germ yield reduced as the soak time decreased. A change in temperature is much more drastic to germ yield than a change in soak time (Fig. 3).

A statistical model to predict germ yield based on soak time and temperature was obtained using the regression analysis ($R^2 = 0.93$):

$$\text{Germ yield} = 20.8128 + 0.1556 \times time + 0.8242 \times temp - 0.0064 \times temp^2 - 0.0014 \times timetp$$

where *time* is soak time (hr); *temp* is soak temperature (°C); *timetp* is time-temperature interaction term (hr, °C).

Germ Oil Content

A time-temperature combination of 12 hr and 59°C gave the highest average recoverable oil content (30 g/kg) compared to all other time-temperature combinations studied (Table II). Response surface analysis of average recoverable oil content shows the same results (Fig. 5).

Soak Water Solids

Table III presents the average solids content at varying soak times and temperatures. Soak water solids increased with increase in soak time. As the soak time increases from 3 to 12 hr, soak water solids increased by 0.3, 0.79, 0.79, and 1.24% for 45, 52, 59, and 75°C soak temperatures, respectively (Table III). The maximum soak water solids content of 1.71% was at 12 hr, 59°C. The solids content at 12 hr, 59°C is not statistically different for solids at 6 hr, 75°C. The increase in solids content with increase in time is due to greater exposure of the corn kernel to the soak water. The increase in the solids content with increase in temperature is due to the increased diffusion of the solubles into the water brought about by higher temperature. Figure 6 illustrates the effect of soak time and temperature on soak water solids.

Specific Gravity Required to Float the Germ

Specific gravity required to float the germ decreased with increased soak time, except that it increased from 1.068 at time-temperature combination of 7.5 hr at 59°C to 1.070 at time-temperature combination of 9 hr, 59°C (Table IV). The highest value of 1.078 was obtained at time-temperature combination of 3 hr, 45°C. The lowest value of 1.060 was obtained at time-temperature combination of 12 hr, 75°C. Decreased value of specific gravity indicates that less recycled slurry will be required at each soak condition to float the germ. The range of specific gravity values obtained for different combinations of soak time and temperature (1.060-1.078) are comparable to specific gravity values of 1.058–1.066, which are recommended for first germ filtrate to achieve good germ recovery (Blanchard 1992). Figure 7 illustrates the effect of soak time and temperature on specific gravity required to float the germ.

TABLE I
Average Germ Yields at Varying Soak Times and Soak Temperatures

	Germ Yields (%)[a–c]			
Time (hr)	45°C	52°C	59°C	75°C
3	4.03 J	4.74 H	5.91 CD	5.24 G
4.5	···	4.76 H	5.96 CD	···
6	4.15 IJ	5.26 G[b]	6.08 C[b]	5.56 E
7.5	···	5.30 GF	6.27 B	···
9	4.18 IJ	5.48 EF[b]	6.41 B[b]	5.84 D
12	4.26 I	5.90 CD	6.76 A	5.36 GF

[a] Yields expressed on % dry solids basis and are mean of two observations.
[b] Mean of four observations.
[c] Values followed by the same letter are not significantly different at the 95% confidence level.

TABLE II
Average Recoverable Oil Content at Varying Soak Times and Soak Temperatures

	Recoverable Oil Content (g/kg)[a–c]			
Time (hr)	45°C	52°C	59°C	75°C
3	14.29 K	17.67 J	23.76 EFG	21.97 G
4.5	···	19.74 I	24.95 CDEF	···
6	15.49 K	22.45 GH[b]	26.07 BCD[b]	24.40 DEF
7.5	···	23.24 GF	26.73 BC	···
9	17.35 J	24.47 DEF[b]	26.93 B[b]	25.62 BCDE
12	17.35 J	26.89 B	30.05 A	21.19 HI

[a] Yields expressed on % dry solids basis and are mean of two observations.
[b] Mean of four observations.
[c] Values followed by the same letter are not significantly different at the 95% confidence level.

TABLE III
Average Solids Content at Varying Soak Times and Soak Temperatures

	Solids Content (%)[a–c]			
Time (hr)	45°C	52°C	59°C	75°C
3	0.15 L	0.21 KL	0.25 K	0.47 GH
4.5	···	0.36 J	0.36 IJ	···
6	0.23 KL	0.54 FG[b]	0.55 EFG[b]	0.95 C
7.5	···	0.57 EF	0.64 E	···
9	0.41 HIJ	0.77 D[b]	0.77 D[b]	1.21 B
12	0.45 HI	1.00 C	1.04 C	1.71 A

[a] Yields expressed on % dry solids basis and are mean of two observations.
[b] Mean of four observations.
[c] Values followed by the same letter are not significantly different at the 95% confidence level.

TABLE IV
Specific Gravity of the Slurry at Varying Soak Times and Soak Temperatures

	Specific Gravity of Slurry[a–c]			
Time (hr)	45°C	52°C	59°C	75°C
3	1.078 A	1.077 A	1.074 B	1.070 DEF
4.5	···	1.073 BC	1.075 B	···
6	1.079 A	1.074 B[b]	1.068 FGH[b]	1.068 GH
7.5	···	1.074 B	1.068 FGH	···
9	1.072 CD	1.069 EFG[b]	1.070 DE[b]	1.068 FGH
12	1.067 H	1.063 I	1.061 J	1.060 J

[a] Yields expressed on % dry solids basis and are mean of two observations.
[b] Mean of four observations.
[c] Values followed by the same letter are not significantly different at the 95% confidence level.

Effect of Lactic Acid

Results of soak time and temperature study showed that at 59°C, there is no considerable change in germ yields (0.85%) as we increase the soak time from 3 hr to 12 hr. Therefore, central value of the range for soak time variable (3–12 hr), which is 7.5 hr, was chosen to study the effect of lactic acid on dependent variables.

When lactic acid was used (both 0.1 and 0.5% levels), the blender speed could be decreased to 36% full power for the first 3 min and 42% full power for the last 3 min as compared to 40% and 46% for 3 min each for corn soaked without added lactic acid. This is consistent with the comments of Watson (1984) and Earp et al (1988), who reported that the lactic acid softens the kernel and made grinding easier.

Germ Yields

The presence of lactic acid (0.1%) in the soak water increased the average germ yield by 0.3 percentage points (Table V). However, the average germ yield for 0.5% lactic acid concentration did not show any significant difference when compared to steeps without added lactic acid. A more detailed study on the effect of lactic acid is warranted.

Germ Oil Content

The germ oil content for 0, 0.1, and 0.5%, lactic acid concentration was 27.5, 29.1, and 28.9 g/kg, respectively (Table V). Germ oil

TABLE V
Germ Yields, Germ Oil Content, Soak Water Solids, Specific Gravity of Slurry at Three Lactic Acid Levels for a Soak Time of 7.5 hr and Soak Temperature of 59°C

	Lactic Acid Levels (%, w/w)		
	0.0	0.1	0.5
Germ yields (%)	6.51 a[a]	6.83 b	6.50 a
Germ oil content (g/kg)	27.54 b	29.05 a	28.86 a
Soak water solids (%)	0.70 c	0.87 b	1.28 a
Specific gravity of slurry	1.064 a	1.063 a	1.063 a

[a] Averages with the same letter within same row are not significantly different at the 95% confidence level.

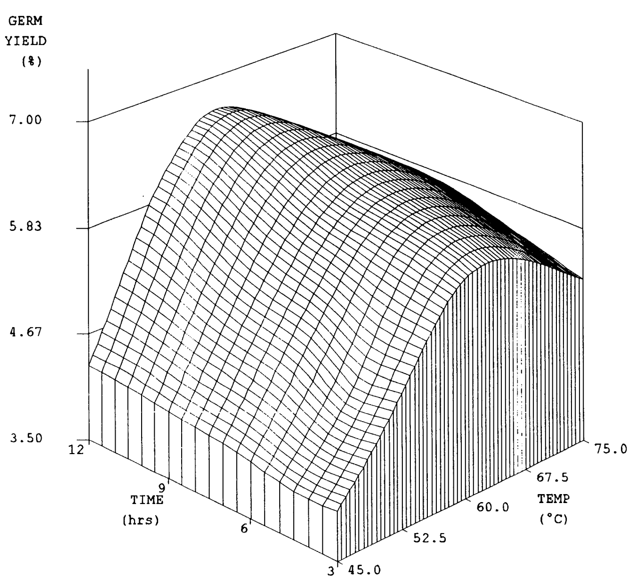

Fig. 4. Three-dimensional response surface of the germ yield (%) at all combinations of time and temperature.

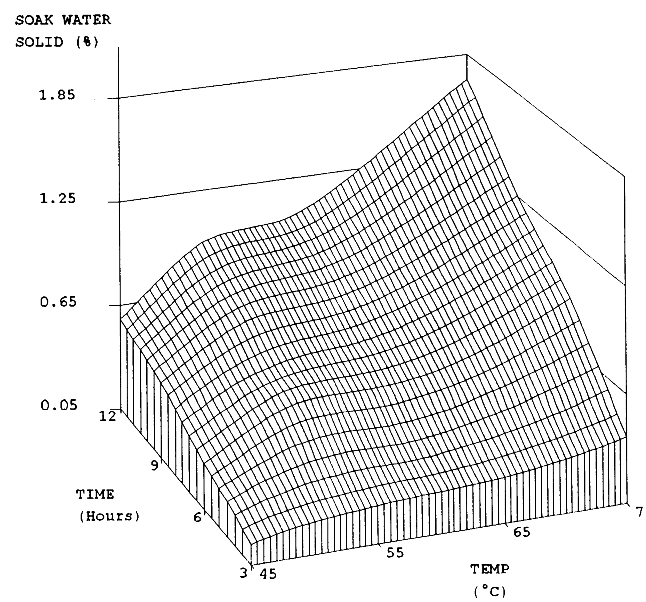

Fig. 6. Three-dimensional response surface of the soak water solids (%) at all combinations of time and temperature.

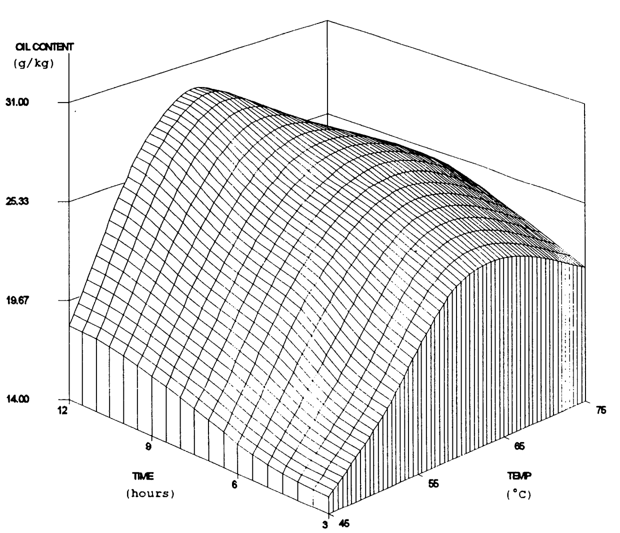

Fig. 5. Three-dimensional response surface of the germ oil content (g/kg) at all combinations of time and temperature.

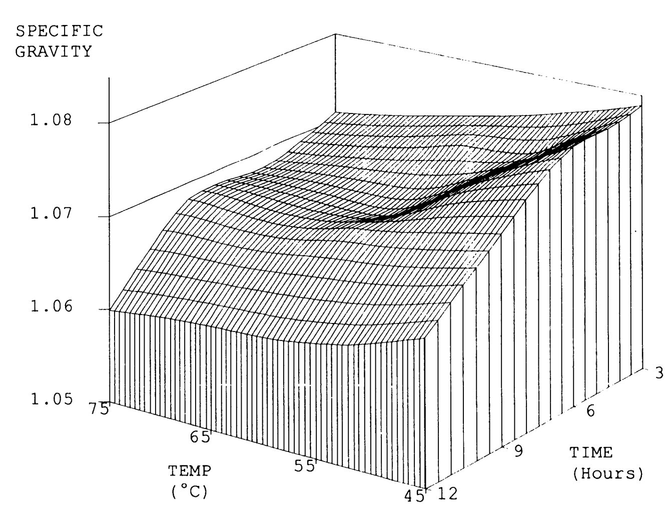

Fig. 7. Three-dimensional response surface of specific gravity required to float germ at all combinations of time and temperature.

content for steeps with 0% lactic acid was significantly lower than steeps with 0.1 and 0.5% lactic acid.

Oil content, however, was not statistically different for the lactic acid concentrations of 0.1 and 0.5%. Results showed that lactic acid increased oil content recovery in the germ, but the concentration of lactic at levels used in this study did not significantly affect the amount of total oil recovered.

Effect of lactic acid on soak water solids was also significantly different for all levels of lactic acid concentration. The average solids content increased by 0.170% at lactic acid concentration of 0.1, and 0.584% at lactic acid concentration of 0.5% compared to 0% lactic acid (Table V). This suggests that lactic acid may have role in the creation of soluble solids or increasing the diffusivity of solubles. Further research is needed in this area.

The specific gravity of the slurries with 0, 0.1, and 0.5% lactic acid concentration were 1.0641, 1.0635, and 1.0635, respectively (Table V). No significant difference was observed with variations in amount of lactic acid added to the soaking media.

CONCLUSIONS

Soak time and temperature affect germ yield, germ oil content, soak water solids, and specific gravity of slurry. Germ can be recovered at levels comparable to those of wet milling from corn going into a dry-grind ethanol process and using a soak time of 12 hr and soak temperature of 59°C. Results for effect of lactic acid were inconsistent and more detailed study is recommended.

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