Wet-Milling Properties of Four Sorghum Parents and Their Hybrids[1]

J. R. NORRIS[2] and L. W. ROONEY[2], Texas A&M University, College Station, Texas

ABSTRACT

A laboratory wet-milling technique was used to separate starch and peripheral endosperm fractions from Martin, Combine Kafir-60, R7078, RTx414, and their hybrids RS608, RS625, RS610, and RS626. Lower starch recovery from Martin than from the other parents had been indicated in preliminary studies. This study was made to determine the influence of Martin and Combine Kafir-60 on the wet-milling properties of hybrids resulting from crosses with R7078 and RTx414. The starch content of the whole grain of the parents and hybrids ranged from 66 to 68%, but differences were not significant. However, differences in starch recovery from parents and hybrids by the wet-milling technique were significant. Hybrids of Martin with R7078 and RTx414 gave significantly lower starch recovery and higher protein content of the starch than hybrids of Combine Kafir-60 with R7078 and RTx414. Martin and Combine Kafir-60 had significantly greater amounts of peripheral endosperm than R7078 and RTx414. Peripheral endosperm content was significantly positively correlated with protein content of the starch and negatively related to starch yield and recovery. Peripheral endosperm content of the hybrids was intermediate between the female and male parents. The protein content of the peripheral endosperm of the hybrids was not significantly influenced by the parents. This study indicates that the female parents, Martin and Combine Kafir-60, significantly influenced starch recovery and protein content of the starch.

Laboratory wet-milling of sorghum grain has been discussed in detail by Watson et al. (1,2). Their techniques have been utilized to detect differences in starch recovery, peripheral endosperm cell content (PEC), and ease of separating protein from starch in sorghums. Although variations among samples of a single variety were great, there appeared to be real varietal differences in PEC content. Martin and Combine Kafir-60 had relatively thick, dense peripheral layers, which presumably led to greater problems in starch purification (3). Watson and Hirata (4) also indicated that hard-seeded varieties such as Martin gave starches of higher protein content.

Martin and Combine Kafir-60 have been used as parents of several sorghum hybrids. Preliminary studies as part of a research program on sorghum quality at the Cereals Laboratory, Texas A&M University, indicated Martin and Combine Kafir-60 were lower in starch recovery and higher in PEC content than R7078 and RTx414. Since these varieties are often crossed to produce hybrids of grain sorghum, this study was designed to determine the influence of the parents on wet-milling characteristics of the hybrids. Information on the heritability of wet-milling traits may be important in the development of high-quality sorghum for processing.

MATERIALS AND METHODS

Samples

The parents and hybrids of grain sorghums studied are presented in Table I.

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[2]Graduate Research Assistant and Assistant Professor, respectively, Cereals Laboratory, Soil and Crop Sciences Department, Texas A&M University, College Station, Texas.

TABLE I. SORGHUM GRAIN PARENTS AND HYBRIDS UTILIZED FOR WET-MILLING STUDIES

Parents	Common Name and Description	Hybrids
BTx398	Martin	RS608 (ATx398 X R7078)
BTx3197	Combine Kafir-60	RS625 (ATx398 X RTx414)
R7078		RS610 (ATx3197 X R7078)
RTx414		RS626 (ATx3197 X RTx414)

Sorghum hybrids are produced by crossing a male-sterile cultivar with a fertility-restoring cultivar. Male-sterility is designated by the prefix "A" preceding the cultivar number, as in ATx398 and ATx3197. Nonrestoring male-fertile lines used to maintain the seed of male-sterile lines contain the prefix "B," and lines which restore fertility to male-sterile cultivars contain the prefix "R," as in R7078 and RTx414. Tx in the designation refers to Texas, the state in which these cultivars were developed. RS is a designation given to sorghum hybrids developed co-operatively by two or more state experiment stations.

Grain for these experiments was produced in 1966 under comparable conditions in sandy loam test plots on the Texas A&M University Plantation, College Station, Texas. The plots were replicated in the field and bulked after harvest. Only clean, sound grain was used. All samples had a test weight greater than 56 lb. per bu. and met all requirements for No. 1 market grade.

Laboratory Wet-Milling

The experimental milling technique employed was essentially the method described by Watson et al. (3). The procedure used in our laboratory is outlined in Fig. 1. Grain (300 g. oven-dry basis) was subjected to two-phase steeping. Phase I

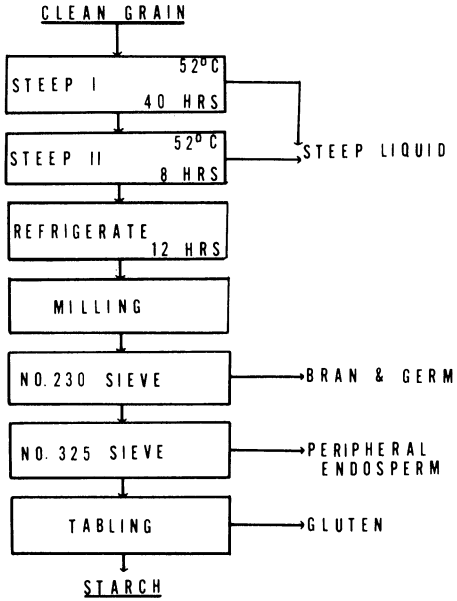

Fig. 1. Laboratory wet-milling procedure.

consisted of steeping for 40 hr. at 52°C. in 1,500 ml. of a solution containing 0.05% SO_2 and 1.5% lactic acid. Phase I solution was replaced by phase II solution which contained 0.10% SO_2 and 0.5% lactic acid, and the grain was steeped for 8 hr. more. The steeped grain was refrigerated overnight and warmed to 40°to 45°C. prior to milling.

The sample (200 g. wet basis) was ground in 250 ml. of water in a Waring Blendor for 1.5 min. Blades of the blender were reversed and the speed was controlled by a voltage regulator set at 85 volts.

The slurry was sieved over U.S. Nos. 230- and 325-mesh sieves. Material remaining on the No. 230 sieve was primarily bran and germ. The overs of the No. 325 were chiefly PEC's.

Starch and gluten (throughs of No. 325) were separated on a starch table constructed from an aluminum I-beam 4 in. wide, 8 ft., 9 in. long. Pitch of the starch table was 1 in. for its entire length. The starch was washed with distilled water, dried, and weighed. Appropriate corrections for steeping solubles and moisture content of the steeped grain and isolated starch were included in the calculations. All times and procedures were rigorously maintained during milling. All determinations of wet-milling characteristics were made in duplicate on samples from the same dry grain lot. The laboratory wet-milling technique proved extremely reproducible, varying little between duplicate samples or operators.

Chemical Determinations

Protein, ash, and ether extract were determined in duplicate by AACC procedures (5).

Starch was determined by a modified fungal amylase-acid-hydrolysis technique (6).

Moisture content of the steeped grain was determined by drying in a forced-draft oven at 130°C. for 16 hr. Loss in weight was calculated as moisture.

TABLE II. COMPOSITION OF THE GRAIN FROM PARENTS AND HYBRIDS

Parents	Starch[a] %	Protein[a] %	Ether Extract[a] %	Ash[a] %
BTx398	65.8	13.8	3.3	1.6
BTx3197	68.4	13.2	2.7	1.6
R7078	66.8	12.4	2.7	1.5
RTx414	67.7	13.0	2.6	1.7
Mean	67.2	13.1	2.8	1.6
Range	65.8-68.4	12.4-13.8	2.6-3.3	1.5-1.7
Hybrids				
RS608	66.3	13.4	2.9	1.5
RS625	67.2	14.0	2.3	1.5
RS610	67.8	11.3	2.8	1.5
RS626	66.6	11.8	3.1	1.6
Mean	67.0	12.6	2.8	1.5
Range	66.3-67.8	11.3-14.0	2.3-3.1	1.5-1.6

[a]Dry-weight basis.

TABLE III. WET-MILLING ATTRIBUTES OF PARENTS AND HYBRIDS[a]

Parents	Starch Yield %	Starch Recovery %	PEC Content %	Protein Content of Starch %	Protein Content of PEC %
BTx398	44.6a	67.7a	1.9d	1.9f	26.0c
BTx3197	48.4b	70.8b	1.6c	1.0c	21.8a
R7078	48.2b	72.2bc	1.2b	0.9b	25.8c
RTx414	53.4c	78.9d	0.8a	0.9b	30.4d
Hybrids					
RS608	49.5b	74.7c	1.4bc	1.2e	25.6c
RS625	49.1b	73.1bc	1.3b	1.1d	24.5b
RS610	52.9c	78.0d	1.2b	0.8a	24.7b
RS626	53.8c	80.8d	1.4bc	1.0c	25.5c

[a]If the small letters beside the mean are different, the means are significantly different at the 0.05 level.

RESULTS AND DISCUSSION

Chemical Composition

Means of compositional data of the parents and hybrids are shown in Table II. No statistically significant differences in starch content of the grain were noted among parents or hybrids. The amounts of protein, ether extract, and ash also are very similar for the parents and hybrids.

Starch Recovery

Recovery of starch is defined as the amount of starch obtained by wet-milling (starch yield, %) divided by the amount of starch present in the whole kernel (starch, %). This is a relative measure of wet-milling performance. Large differences in percentage of starch recovered are apparent in Table III. The statistical significance of many of these differences is indicated in Table IV. Hybrids as a

TABLE IV. ANALYSIS OF VARIANCE OF STARCH YIELD, STARCH RECOVERY, AND PEC CONTENTS

	Mean Square Values		
Source, d.f.	Starch Yield	Starch Recovery	PEC Yield
Reps 1	0.6	1.4	<0.1
Varieties 7	20.9**	39.4**	1.3**
Parents 3	28.3**	44.5**	3.0**
Female vs. male 1	40.8**	78.3**	7.5**
Between females 1	16.3**	9.5*	0.3
Between males 1	27.9**	45.5**	1.0**
Hybrids 3	11.7**	23.5**	<0.1
Martin vs. Combine Kafir-60 1	33.6**	59.7**	<0.1
Between Martin 1	0.4	2.7	<0.1
Between Combine Kafir-60 1	1.1	7.9	<0.1
Parents vs. Hybrids 1	26.2**	71.7**	<0.1
Error 7	0.7	1.5	<0.1

group gave significantly greater starch recovery than the parents. Martin was significantly lower than any other parent and RTx414 was significantly higher than the other parents. No statistically significant differences were apparent between Martin hybrids (RS608 and RS625) and between Combine Kafir-60 hybrids (RS610 and RS626), but Martin hybrids were significantly lower in recovery of starch than Combine Kafir-60 hybrids. Martin gave a lower recovery of starch, and its presence in a hybrid resulted in decreased starch recovery from the hybrid.

Peripheral Endosperm Cells

PEC's have been indicated as contaminants of starch responsible for extra purification steps and may be important indicators of wet-milling potential. Although the screened fraction called PEC may contain other portions of the kernel, it is chiefly the unbroken cells from the outer endosperm. The female parents, Martin and Combine Kafir-60, contained significantly more PEC than R7078 and RTx414. This is in agreement with findings of Watson et al. (3), who indicated varietal differences in PEC content. The PEC content of the grain from the hybrids appears to be intermediate to the PEC of the grain from their parents.

Protein Content of the Starch

Protein content of starch is indicative of the relative ease of separation of starch granules from the proteinaceous matrix during wet-milling. Martin starch contained significantly more protein than the starch from any other parent sorghum variety. Martin hybrids, RS608 and RS625, were significantly higher in starch protein content than Combine Kafir-60 hybrids. This indicates that the parents of a hybrid may influence the relative ease of separating protein and starch.

Protein Content of PEC

The protein content of PEC fraction appeared to be related inversely to the amount of PEC present. Correlation analysis for PEC protein content vs. percent PEC yielded $r = -0.63$. No significant differences in PEC protein content existed among the hybrids. However, among the parents, the PEC fraction of Combine Kafir-60 was significantly lower in protein than the PEC fractions of the other parents. RTx414 was significantly higher.

Relationships of Wet-Milling Attributes

Simple correlation coefficients of wet-milling attributes (Table V) indicate starch yield and starch recovery were highly related, $r = 0.98^{**}$. PEC content was significantly related to yield, recovery, and protein content of starch. The r values

TABLE V. SIMPLE CORRELATIONS AMONG WET-MILLING ATTRIBUTES

	Starch Yield %	Starch Recovery %	PEC Yield %	Protein Content of PEC %	Protein Content of Starch %
Starch, %	0.41	0.22	−0.37	−0.23	−0.70*
Starch yield, %		0.98**	−0.73*	0.34	−0.74*
Starch recovery, %			−0.70*	0.39	−0.64
PEC yield, %				−0.63	0.74*
Protein content of PEC, %					−0.84**

were -0.73*, -0.70*, and 0.74*, respectively. Starch yield was significantly negatively related with protein content of the starch, r = -0.74*. As the protein content of the PEC fraction increased, the protein content of the starch decreased, r = -0.84**. Varieties with low quantities of PEC, R7078 and RTx414, also had high protein in the PEC and the recovered starch was low in protein.

CONCLUSIONS

During laboratory wet-milling, sorghum hybrids yielded more starch than their parents. Starch yield, starch recovery, and protein content of the starch were influenced significantly by the choice of female parents. Martin decreased the starch yield and increased the protein content of the starch when it was present in a hybrid.

Martin and Combine Kafir-60 contained considerably more PEC than R7078 and RTx414. Hybrids appeared to be intermediate in PEC content. Considerable variation in protein content of PEC occurred among the parents. PEC fractions from the hybrids were intermediate in protein content.

It appears that wet-milling quality of sorghum hybrids can be influenced by choice of parents for hybrids. Optimum wet-milling quality in sorghums may perhaps be achieved by hybridization of parents well suited for recovery of starch with low protein.

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