



## Biomass production potential in the Batini barley landrace from Oman

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Received 12 January 2005, accepted 25 March 2005.

### Abstract

Diversity available in seven subpopulations of the Batini barley landrace was quantified for pre- and post-anthesis thermal time, filling period, biological and grain yield, number of tillers and plant height. We identified grain, forage and dual-purpose types and selected elite germplasm for breeding purposes. Patterns of phenotypic variation and co-variation in the phenological and agronomic traits, measured on 2,040 landrace accessions, subdivided the collection into 14 phenological classes in three (grain, dual-purpose and forage) end-use types, each with distinct phenological trait combinations. The dual-purpose type was more polymorphic and significantly had higher total diversity and differentiation than the grain and forage types. Directional selection, especially in the phenological traits, resulted in large, idiosyncratic changes in plant phenotypes. Targeting traits that have a disproportionately large influence on differences in the mean covariance will help identify sources of variation for breeding purposes.

**Key words:** Barley landrace, diversity, phenology, biomass, Oman.

### Introduction

With the increased interest in indigenous plant resources for forage and biomass production, information is required on the phenology and biomass production potential of barley landraces <sup>1</sup> as a reliable forage source in countries lacking feed for increasing livestock. The availability of locally-grown, high quality grain and forage is a major limitation to successful livestock husbandry in the Middle East <sup>2</sup>. Barley landraces, the feed source of choice for sheep throughout the Middle East, have been used by subsistence farmers for millennia to produce grain, forage or both <sup>2</sup>. Inter- and intra-specific variations for vigorous early growth in small grains have been exploited for high winter biomass <sup>3</sup> and straw production as an important summer feed source <sup>2</sup>. Farmers traditionally select visually for high biomass, tall plants, profuse tillering and high grain yield <sup>1</sup>. Early-maturing genotypes, visually selected by farmers <sup>1</sup> or breeders <sup>4</sup>, produce higher grain per unit time, up to anthesis, than late-maturing genotypes. Total biomass, however, is little affected by earliness <sup>5</sup> and is maximized in genotypes having tall plants and profuse tillering capacity <sup>6</sup>. Diversity for biomass, grain yield and phenological traits in this landrace is unknown, but would aid in determining selection and breeding priorities. The objectives of this study were to quantify the diversity available in the germplasm collection for phenological and agronomic traits, and identify grain, forage, or dual-purpose genotypes.

### Materials and Methods

A field experiment was conducted during the 2001/2002 growing seasons at the Experiment Station of The International Center for Biosaline Agriculture (ICBA), Dubai, United Arab Emirates (25°13'N and 55°17'E). We planted ten seeds from each of 2,308 accessions representing seven subpopulations in the Batini barley landrace <sup>7</sup> at a distance of 25 cm between rows and 15 cm

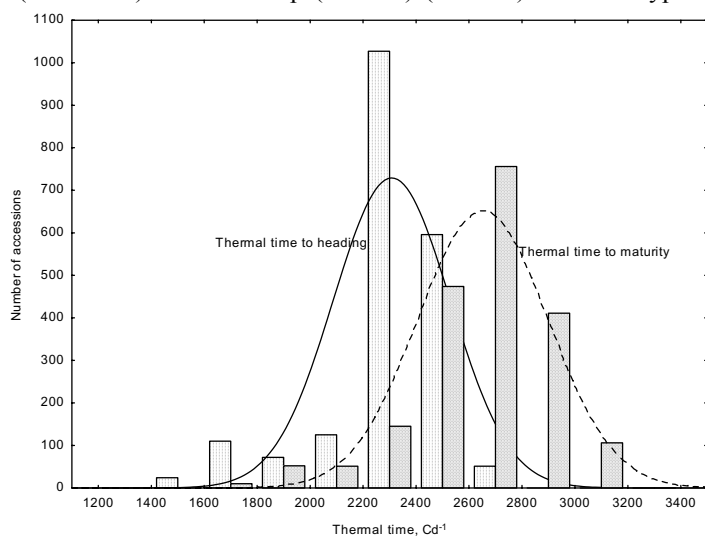
between seeds within rows. A local check variety was planted in every 20th row throughout the nursery, and its variation was used as a measure of soil heterogeneity, to estimate error variance, and for the reliability of categorical data analysis. Planting date was November 11-13, 2001 and physiological maturity extended from early March to late April 2002. Recommended management practices for irrigation, fertilization and weed control were followed throughout the growing season. Field observations on phenological development were recorded on the primary tillers of each accession according to Tottman <sup>8</sup>. Single plants were harvested at full maturity, and data were collected on a minimum of five plants per accession of only 2,040 accessions for plant height, number of tillers, biological yield and grain yield. Each accession was visually classified by three independent raters, and the combined visual classification was used to classify the germplasm collection into grain-, dual purpose- and forage-types.

**Statistical analyses:** Length in days of pre- and post-anthesis developmental stages was converted to thermal time ( $^{\circ}\text{C d}^{-1}$ ) using a base temperature of 0.0 $^{\circ}\text{C}$  for the period from sowing to anthesis, and 9.0 $^{\circ}\text{C}$  for the period from anthesis to maturity <sup>9</sup>. Quantitative data for the three phenological stages were used to identify phenotypic classes <sup>9</sup> of accessions with similar phenological stages. Fourteen phenological classes were delineated within the three end-use types (Table 1) using the likelihood ratio test <sup>10</sup>. This "optimal" number of phenological classes was selected when the log-likelihood function reached its highest increase, and was verified through a two-stage clustering analysis procedure to minimize the Euclidean sum of squares using the K-means cluster module in ClustanGraphics <sup>11</sup>. The mean and standard deviation (s.d.) calculated for continuous variables were used to categorize data into three discrete classes according to Zar <sup>12</sup>. For the six

traits in Table 1, variation of the local check on no occasion exceeded that of the accessions. Categorical data (i.e., early, medium and late) for thermal time to anthesis ( $^{\circ}\text{C d}^{-1}\text{H}$ ), and to maturity ( $^{\circ}\text{C d}^{-1}\text{M}$ ), and the length of filling period ( $^{\circ}\text{C d}^{-1}\text{Fp}$ ) were used to quantify available diversity in this germplasm collection. Relative phenotypic frequencies of categorical traits were used to calculate a polymorphic diversity index ( $I$ ) for each subpopulation, end-use type and phenological class as described by Zhang and Allard<sup>13</sup>. These estimates were zero, or not significantly different from zero for the check variety, but were significantly greater than zero ( $P<0.05$ ) for the accessions (data not presented). Total diversity ( $H_T$ ), average diversity ( $H_S$ ), and diversity among subpopulations, diversity among end-use types and diversity among phenological classes relative to total diversity ( $G_{ST}$ ) were calculated using frequencies of all categorical data<sup>14</sup>. A dimension reduction and perceptual mapping statistical procedure<sup>12</sup> was employed to reduce the dimensionality of a matrix of three end-use types, 14 phenological classes and three levels of each of six categorical traits. A two-dimensional plot was developed where associations among objects (end-use types and phenological classes) and levels of descriptive traits can be identified. All statistical analyses were conducted using several modules in the statistical packages STATISTICA<sup>15</sup>, unless otherwise specified.

## Results

**Phenology and end-use types:** Frequency distributions of accessions based on their thermal time to anthesis and to maturity are presented in Fig. 1. Thermal time to anthesis and to maturity peaked between 2200 and 2300 and between 2700 and 2800  $^{\circ}\text{C d}^{-1}$ , respectively. The skewness ( $g_1$ ) values for  $^{\circ}\text{C d}^{-1}\text{H}$ ,  $^{\circ}\text{C d}^{-1}\text{M}$  and  $^{\circ}\text{C d}^{-1}\text{Fp}$  were -1.589, -1.098 and 0.135, respectively, and the Shapiro-Wilks statistic to test for normality was significant ( $W = 0.715$ ,  $P<0.05$ ). The combined visual classification by three independent raters partitioned the 2,040 accessions into 10, 75 and 15% grain dual-purpose and forage types, respectively. Results of the likelihood ratio test<sup>10</sup>, as confirmed by the K-means clustering procedure<sup>11</sup> separated the 2,040 accessions into 14 phenological classes with a combination of short, medium and long  $^{\circ}\text{C d}^{-1}\text{H}$  (mean $\pm$ standard deviation 2306 $\pm$ 219),  $^{\circ}\text{C d}^{-1}\text{M}$  (2649 $\pm$ 246) and  $^{\circ}\text{C d}^{-1}\text{Fp}$  (343 $\pm$ 92) (Table 1). End-use types



**Figure 1.** Frequency distribution of 2,040 accessions of the Batini barley landrace from Oman based on thermal time to heading and to maturity.

exhibited highly variable relationships among their phenological stages. The grain, dual purpose and forage end-use types on average allocated 89, 87 and 82% of their total  $^{\circ}\text{C d}^{-1}$  to pre-anthesis growth stage, respectively, with a concomitant increase in biological yield and decrease in grain yield (Table 1). Across all phenological classes,  $^{\circ}\text{C d}^{-1}\text{H}$  and  $^{\circ}\text{C d}^{-1}\text{M}$  were positively and significantly correlated ( $r = 0.91$ ;  $P<0.01$ ), whereas  $^{\circ}\text{C d}^{-1}\text{Fp}$  was only negatively and significantly correlated ( $r = -0.74$ ;  $P<0.05$ ) with grain yield. Significant correlations among agronomic traits were limited to those between plant height and each of biological yield ( $r = 0.84$ ;  $P<0.01$ ) and number of tillers ( $r = 0.80$ ;  $P<0.05$ ), and between biological yield and number of tillers ( $r = 0.97$ ;  $P<0.01$ ). All phenological and agronomic traits were influenced by end-use type and phenological class (Table 1). Level of variation among end-use types and among phenological classes is reflected on the percent pairwise significant differences among the least square means of each trait. There were significant pairwise differences among all three end-use types for all six traits (100%), whereas there were a minimum of 67% for  $^{\circ}\text{C d}^{-1}\text{H}$ , to a maximum 84% pairwise significant differences among means of phenological classes for grain yield.

**Polymorphism analyses:** Mean polymorphic index ( $I$ ) for all subpopulations was  $0.78\pm 0.11$ . High polymorphisms in Batini 1 ( $I = 0.74\pm 0.15$ ) and in Batini 2 ( $I = 0.73\pm 0.26$ ) were associated with high standard deviation. Polymorphic indices for Batini 5 ( $I = 0.63\pm 0.10$ ), Batini 7 ( $I = 0.61\pm 0.17$ ), and Batini 4 ( $I = 0.56\pm 0.18$ ) did not differ significantly from the remaining subpopulations. However, Batini 6 was the only subpopulation to differ significantly from the remaining subpopulations. Mean polymorphic indices for phenological ( $0.85\pm 0.1$ ) and agronomic ( $0.88\pm 0.09$ ) traits did not differ significantly (Table 2). Germplasm in the grain type displayed the highest polymorphism for  $^{\circ}\text{C d}^{-1}\text{Fp}$  ( $I = 0.98$ ), followed by the dual-purpose type (0.92) and the forage type ( $I = 0.66$ ). However, except for the polymorphic index of  $^{\circ}\text{C d}^{-1}\text{M}$  (0.83) in the dual-purpose type, the remaining  $I$ -estimates were comparably similar. Relatively similar and high polymorphic indices were displayed by grain (0.92), dual-purpose (0.91) and forage types (0.98) for plant height.  $I$ -estimates for the remaining agronomic traits ranged from 0.52 for tiller number in the forage-type to  $>0.80$  for grain yield in the dual-purpose and forage types.

**Diversity analyses:** Total diversity ( $H_T$ ) was estimated for all subpopulations as  $0.43\pm 0.06$ , and the average diversity within subpopulation ( $H_S$ ) was  $0.37\pm 0.03$ . Hence, the low average diversity among subpopulations as a percentage of total diversity ( $G_{ST} = 0.14$ ). However,  $G_{ST}$  was relatively higher than average for plant height (0.19), intermediate for each of  $^{\circ}\text{C d}^{-1}\text{H}$ ,  $^{\circ}\text{C d}^{-1}\text{M}$  and  $^{\circ}\text{C d}^{-1}\text{Fp}$  (0.15), and lower than average for biological yield (0.13), number of tillers (0.12) and grain yield (0.07). Mean total diversity for all end-use types ( $H_T = 0.44\pm 0.02$ ), when partitioned into its components, reflected an extremely high (81.1%) level of differentiation.  $H_T$  estimates differed significantly among the three end use-types, and the level of differentiation was lowest (72.4%), intermediate (82.2%) and highest (88.6%) for forage, dual-purpose and grain types, respectively. Mean  $H_T$  based on phenological ( $0.55\pm 0.09$ ) and agronomic ( $0.57\pm 0.05$ ) traits did not differ significantly; however, levels of differentiation for individual traits

**Table 1.** Number of accessions in each of 14 phenological classes and their phenological and agronomic trait means identified in three end-use types of the Batini barley landrace from Oman.

End use	Phenological class	Number of accessions	Days to Anthesis	Days to Maturity	Filling period	Phenological traits , °Cd <sup>1</sup>			Agronomic traits			
						Anthesis (% of total °Cd <sup>1</sup> )	Maturity	Filling period	PH† (cm)	TL (number)	BY (Kg ha <sup>-1</sup> )	GY
Grain		204										
	EES	32	Early	Early	Short	2169 (89)	2433	264	85	11.6	3042	1622
	EEM	54	Early	Early	Medium	2065 (85)	2421	356	97	13.8	3388	1125
	MES	118	Medium	Early	Short	2389 (92)	2590	201	95	15.6	3659	1458
Mean					2207 (89)	2481	274	92	13.7	3363	1401	
Dual purpose		1530										
	EMM	126	Early	Medium	Medium	2311 (86)	2673	362	104	14.5	3524	1215
	MMS	180	Medium	Medium	Short	2424 (89)	2727	302	115	19.4	4043	1462
	MMM	287	Medium	Medium	Medium	2426 (86)	2822	396	85	10.7	2843	732
	MLM	248	Medium	Late	Medium	2507 (85)	2935	428	89	13.6	3259	967
	LMS	178	Late	Medium	Short	2574 (90)	2847	273	103	16.5	3667	1231
	LLS	274	Late	Late	Short	2603 (88)	2956	352	114	18.0	3991	1532
	LLM	237	Late	Late	Medium	2600 (86)	3029	429	108	17.6	3818	1310
	Mean					2492 (87)	2855	363	103	15.8	3592	1207
Forage		306										
	EEL	85	Early	Early	Long	1844 (79)	2312	467	109	23.7	4544	893
	EML	73	Early	Medium	Long	2259 (83)	2726	467	89	15.9	3348	882
	MML	58	Medium	Medium	Long	2390 (83)	2855	465	99	15.1	3429	965
	MLL	90	Medium	Late	Long	2455 (82)	2988	533	108	17.6	3745	1012
Mean					2237 (82)	2720	483	101	18.0	3766	938	
Percent significant differences among												
End-use types						100	100	100	100	100	100	100
Phenological classes						67	71	82	71	71	77	84

†: PH=Plant height; TL= Tillers per plant; BY= Biological yield; GY= Grain yield.

**Table 2.** Phenotypic diversity indices and components of genetic diversity in phenological and agronomic traits of three end-use types identified in the Batini barley landrace from Oman.

End-use	GD Statistic	Phenological traits					Agronomic traits					
		Number of Sub-population	°Cd <sup>-1</sup> H	°Cd <sup>-1</sup> M	°Cd <sup>-1</sup> Fp	Mean (s.d.)	Number of Sub- population	PH†	TL	BY	GY	Mean (s.d.)
Batini landrace	<i>Na</i> ‡	7.0					7.0					
	<i>Ne</i>	5.8					4.5					
	<i>I</i>		0.73	0.91	0.91	0.85 (0.10)		0.99	0.82	0.84	0.87	0.88 (0.09)
	<i>H<sub>T</sub></i>		0.53	0.61	0.50	0.55 (0.09)		0.62	0.55	0.58	0.52	0.57 (0.05)
	<i>G<sub>ST</sub></i>		0.10	0.15	0.22	0.16		0.12	0.10	0.12	0.11	0.09
Grain	<i>Na</i>	5.0					5.0					
	<i>Ne</i>	1.9					1.4					
	<i>I</i>		0.65	0.69	0.98	0.78 (0.18)		0.92	0.72	0.63	0.79	0.77 (0.12)
	<i>H<sub>T</sub></i>		0.51	0.49	0.63	0.55 (0.01)		0.50	0.49	0.43	0.52	0.48 (0.01)
	<i>G<sub>ST</sub></i>		0.74	0.45	0.44	0.53b†		0.40	0.42	0.46	0.27	0.38c
Dual Purpose	<i>Na</i>	7.0					7.0					
	<i>Ne</i>	5.6					4.2					
	<i>I</i>		0.64	0.83	0.92	0.79 (0.14)		0.91	0.78	0.76	0.86	0.87 (0.14)
	<i>H<sub>T</sub></i>		0.61	0.66	0.52	0.59 (0.01)		0.64	0.61	0.61	0.57	0.61 (0.01)
	<i>G<sub>ST</sub></i>		0.82	0.75	0.73	0.77a		0.58	0.71	0.77	0.65	0.68a
Forage	<i>Na</i>	5.0					4.0					
	<i>Ne</i>	2.6					1.7					
	<i>I</i>		0.75	0.63	0.66	0.68 (0.06)		0.98	0.52	0.69	0.89	0.77 (0.21)
	<i>H<sub>T</sub></i>		0.53	0.59	0.57	0.56 (0.01)		0.66	0.51	0.58	0.62	0.59 (0.01)
	<i>G<sub>ST</sub></i>		0.16	0.62	0.48	0.43c		0.48	0.71	0.51	0.52	0.55b

† See Table 1 for abbreviations

‡ Genetic Diversity (GD) statistics: *Na* = Number of subpopulations; *Ne* = Number of effective subpopulations; *I* = Phenotypic diversity index; *H<sub>T</sub>* = Total diversity, *G<sub>ST</sub>* = Diversity among subpopulations relative to total diversity.

were very high. Mean total diversity, based on phenological traits, differed significantly among the dual purpose-type (0.59±0.01) and each of the forage- (0.56±0.01) and the grain-type (0.55±0.01). This was reflected on the relative population differentiation with the dual-purpose type being highly differentiated (0.77), followed by the grain type (0.53) and forage type (0.43). All three *G<sub>ST</sub>* estimates differed significantly from each other (*P*<0.01). Mean total diversity, based on agronomic traits, was similar for dual-purpose (0.61±0.01) and forage types (0.59±0.01), and significantly lower (0.48±0.01) for grain-type. However, levels of differentiation among the three end-use types were highly significant (Table 2). Mean genetic distance among end-use types

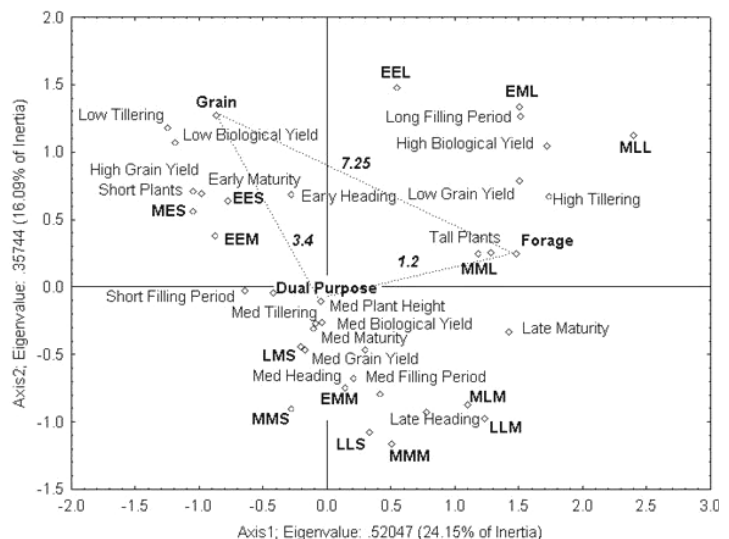
was 0.49. Mean genetic distance among phenological classes within each end-use types was highest (0.67), intermediate (0.31) and lowest (0.17) in the dual purpose-, forage- and grain-types, respectively. The phenological classes EML and MML in the forage-type were the closest (0.13), whereas MMS and MLM in the dual purpose-type were the farthest (0.97).

**Perceptual mapping of phenological classes and their attributes:** Associations among different levels of phenological and agronomic traits, along with the three end-use types and the 14 phenological classes are displayed in a two-dimensional plot (Fig. 2). The first axis separated the grain type from the dual

purpose- and forage-types, and accounted for 24.15% of the variance. Axis 1 totally separated the grain from the forage types, however, three phenological classes (LMS, LLS and MMS), visually classified as grain type, were plotted closer to the dual-purpose type. Axis 2 separated the grain- and forage-types from the dual-purpose type, however, it accounted for a smaller (16.09%) portion of total variation.

### Discussion

The persistence of landraces in subsistence farming systems of Oman<sup>16</sup> and other parts of the Middle East<sup>17</sup> implicitly suggests that farmers value them as crops and as genetic resources. This suggestion is confirmed by research on farmers' knowledge<sup>1</sup> and their use of different landraces and old cultivars<sup>17</sup>. Deviation from normality (Fig. 1) and the high phenotypic diversification in phenological and agronomic traits (Table 1) are primarily caused by directional selection<sup>18</sup>, with phenology traits being more strongly selected than agronomic traits<sup>19</sup> as evidenced by  $G_{ST}$  values (Table 2). The large genotypic variation in barley phenology found in this and other studies<sup>6,24</sup>, especially at the pre-anthesis growth stage<sup>20</sup>, effected major changes in subsequent growth stages and in biological yield, seed yield and their components. Differences as high as 41 and 31% in °C d<sup>-1</sup> H and °C d<sup>-1</sup> M, respectively, were found in the Batini landrace. Such differences have been identified, albeit at a lower magnitude (17-27%), in barley landrace germplasm of diverse origins<sup>21</sup> and helped explain the inconsistent relationships among phenological stages, among grain yield and phenological stages and among agronomic traits. Grain yield advantages of the grain type over the dual-purpose and forage types were 16 and 49.4%, respectively. However, forage type had 4.8 and 12.0% biological yield advantage over grain and dual-purpose types, respectively. The grain, dual-purpose and forage phenological classes allocated 89, 87 and 82% of their thermal time to pre-anthesis growth stage, respectively. These ratios were comparable to values (86-89%) reported for barley under similar environmental conditions in the Persian Gulf<sup>22</sup>. The few positive and significant correlations found among agronomic traits across all phenological classes, in this and other studies, point to the importance of plant height and tillering capacity as sources of biomass in dual purpose or forage barley<sup>9</sup> and winter wheat<sup>23</sup> genotypes. Forage and dual-purpose types were ~12.0% taller than the grain type. However, forage types had 31.4 and 13.9% more tillers than the grain and dual purpose types, respectively, and the dual purpose types have 15.3% more tillers than the grain types. Nevertheless, a large biomass due mainly to tall plants cannot be useful for grazing as it is mainly realized after stem elongation<sup>2,5</sup>. Total biomass was little affected by earliness in the grain type. The negative correlation between °C d<sup>-1</sup> H and °C d<sup>-1</sup> Fp ( $r = -0.25$ ,  $P < 0.05$ ) suggests that gains from longer °C d<sup>-1</sup> H are lost by a shorter °C d<sup>-1</sup> Fp<sup>24</sup>. However, the respective r-value in the forage-type was positive and highly significant ( $r = 0.63$ ,  $P < 0.001$ ). Extreme negative and significant correlation ( $r = -0.95$ ,  $P < 0.001$ ) between °C d<sup>-1</sup> H and °C d<sup>-1</sup> Fp in barley resulted in low biological yield and was due not only to shorter °C d<sup>-1</sup> Fp, but also to longer °C d<sup>-1</sup> H, associated with taller but fewer tillers per plant<sup>24</sup>. Agronomic diversity in the Batini barley landrace, similar to other landraces can be impacted by a combination of natural and anthropogenic factors. The high level of diversification in this and other landraces<sup>6,21</sup> within



**Figure 2.** Mahalanobis distances among three end-use types, association among 14 phenological classes and variants of phenological and agronomic traits in the Batini barley landrace from Oman.

subsistence farming systems can be explained on the basis of environmental, socioeconomic and cultural factors, or as a result of farmers' selection being in favor of heterozygosity<sup>25</sup>. Patterns of covariation of phenological and agronomic traits proved to be more relevant for phenotypic evaluation than patterns of univariate variation. The former offers an effective method of stratifying and sampling variation in germplasm collections and identifying germplasm for breeding programs<sup>26</sup>. The two-dimensional plot of end-use types, phenological classes and their attributes, along with the  $D^2$  values among the end-use types (Fig. 2) furnished the basic characteristics of end-use types and provided simple selection criteria for specific end-use germplasm.

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