SECTION 5 CASE STUDY 3

Integrating selection for drought tolerance into a breeding program: the Brazilian experience

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1. Our target environments (TPE)

Brazilian savannas have a well-defined rainy season, starting in October and ending in April. During this period, total rainfall ranges from 1,200 to 1,500 mm, with monthly averages sometimes higher than 200 mm. In spite of this abundance, rain distribution may be irregular and dry spells can occur, most frequently during January and February, when the upland rice crop, sown at the onset of the rainy season, undergoes reproductive development.

In the early years of the Cerrado (Brazilian savanna) opening, upland rice was the most attractive pioneer crop because of its rusticity and tolerance of soil acidity and low fertility. In the 1970s and '80s, a large area was deforested for agricultural activities and rice attained its peak of 4.5 million ha under cultivation in 1987-88. During this period, while area and production increased, upland rice yield was low and constant at around 1.2 t ha⁻¹. Yields remained low because of the combined effect of dry spells and low adoption of recommended technology. Thus, our *initial TPE* was for the drought-prone uplands with low fertility, with a focus on developing varieties with tolerance of midseason reproductive drought, relying on the japonica group as the major source of germplasm.

After the studies of Pinheiro et al (1985) and Steinmetz et al (1985), the breeding strategy was expanded to include selection for yield potential (modern plant type) to obtain genotypes to be grown under supplementary irrigation and in favorable microregions with desirable rainfall distribution. Initially, this new TPE, aimed at favorable upland conditions, required only a small share of human and financial resources and used predominantly indica germplasm. With time, the decline in savanna frontier land and concomitant migration from the southeast to northwest, that is, from a riskier toward a less risky environment (Steinmetz et al 1988), resulted in a decline in upland rice area (2.4 million ha in 2001) associated with increased average yield (1.9 t ha⁻¹). Thus, the *target domain has changed* to more favorable conditions (Photo 17) and the breeding priorities have shifted to include yield potential and improvement of grain quality, and to rely more and more on japonica by indica crosses. Plant architecture and grain appearance are now important requisites for variety release for both favorable and unfavorable climatic conditions, so the distinction between the two former upland breeding programs, aimed at the different TPE, has disappeared.

2. Our breeding approach for drought-prone areas

The dramatic shift of the crop to more favorable locations and the higher use of technology by farmers, which help minimize risk, led to a decrease in the priority for drought tolerance in

The TPE can change when farming practices change.



the upland rice breeding program. Now, because of the move in plant type from tropical japonica to japonica \times indica derivatives, yield potential has increased from 4.5 to 6 t ha⁻¹ and average yield has doubled. Accordingly, the support program on drought-tolerance evaluation has been reduced and drought-stress tolerance, considered previously as a major research priority in the national upland rice breeding program, now plays a secondary role to improvement of yield under favorable environments. However, here we describe the initial approach



Fig. 1. Schematic representation of germplasm flux in the drought-tolerance evaluation program and its relationship with the upland rice breeding program aimed at unfavorable conditions. used by the upland rice breeding program of EMBRAPA Rice and Beans (formerly the National Rice and Beans Research Center, CNPAF) to develop varieties for the TPE of unfavorable savanna areas with midseason drought. We discuss the successes as well as difficulties and limitations of the program.

From the start, we found that the unpredictable occurrence of drought and its timing with the critical stages of plant development made selection among segregating materials in a conventional breeding program inefficient. Therefore, we focused on (1) the careful evaluation of potential progenitors using drought-tolerant selections for crossing to elite germplasm and (2) testing advanced (fixed) breeding lines under controlled drought conditions. These two drought selection activities were conducted as support to the routine breeding program and involved a strong partnership between plant physiologists and breeders in three classes of experiments, designated as "preliminary evaluation," "second evaluation," and "final evaluation" (left side of Fig. 1). Such trials, described in more detail below, were conducted at CNPAF's headquarters in Goiânia, Goiás.

Figure 1 shows the overall approach of this breeding and evaluation work. The program began by focusing on selecting for drought-tolerant materials to be used in crossing. The genotypes used as parents were predominantly of tropical japonica extraction, from both national and African origin. They were screened in the preliminary and second evaluation to identify those for use in the crossing

program. The final evaluation for drought, in the original strategy, contained the best selections from the second evaluation and the most promising lines from the second year of testing in the advanced yield trials (right side of Fig. 1). The methodology of this drought evaluation trial, which is still part of the breeding program, is described in detail later.

3. Identifying the parents



The *preliminary evaluation* trial for drought tolerance (see Fig. 1) comprised 400 to 600 entries each year, including local varieties, some regionally collected ones, and elite germplasm from national and international programs. The experiments were planted late in the season, in January (recommended sowing time is 15 October to 15 November), to improve the probability of drought during reproductive development. The plots were kept well watered during the vegetative stage until a significant proportion of the entries were in the reproductive stage, when irrigation was discontinued to induce water stress. If stress occurred, entries were evaluated using the IRRI standard drought-tolerance visual scale (Chang et al 1974, Loresto et al 1976), with scores from 1 to 9. Entries that ranked equal to or better than the commercial varieties IAC 25 and IAC 47 (respectively, checks for early and late maturity) in this trial, as well as the outstanding entries from the preliminary yield trial, were selected for the second evaluation trial (see Fig. 1).

In this *second evaluation*, the entries were grouped into early (less than 80 days from sowing to flowering), medium (80–90 days), and late (more than 90 days) maturity classes to allow for a comparison among genotypes with similar maturity. The sowing date was staggered by maturity (i.e., the late group was sown 10 days before the medium group and 20 days before the early group). The field arrangement was similar to that of the preliminary evaluation, but the number of replications increased to at least three and a fully irrigated treatment was included. The protocol to induce water stress (in the drought treatment) was the same as for the preliminary evaluation. Again, no yield data were collected from the plots and drought evaluation was based on IRRI's visual scale for drought tolerance at the reproductive stage.

The topmost entries were recommended as parents for hybridization with elite varieties and advanced breeding lines.

4. Yield testing

The segregating lines or populations derived from these crosses of drought-tolerant with elite materials were evaluated for yield as part of the normal plant-breeding program. The selection also considered visual characters related to stress escape and avoidance such as a short growth cycle, moderate tillering ability, and moderate leaf area, as well as agronomic characters and reaction to biotic stresses. If drought stress occurred in the routine yield testing, individual lines were discarded on the basis of leaf rolling, panicle exsertion, and spikelet fertility.

The fixed lines were then evaluated for yield in three classes of experiments: observational nurseries, preliminary yield trials, and advanced yield trials (right side of Fig. 1). Selections from the advanced yield trials were then included in the final drought-tolerance trials (see left side of Fig. 1) before recommendations were made for varietal release. Genotypes with high yield potential or desirable grain traits, as identified in the yield trials, could be discarded in favor of more drought-tolerant genotypes, identified in the drought evaluation trials.

At first, all of these yield trials were conducted only at CNPAF's headquarters in Goiânia, Goiás. However, in 1983, the Regional Commission for Testing and Recommendation of Rice Varieties was established with 15 public research institutions and 18 sites to conduct preliminary and advanced yield trials of breeding lines from three institutions for the savanna region. In this network, trials are conducted during the normal growing season, without controlled irrigation.

In the original strategy, the final evaluation for drought contained the best selections from the second evaluation and the most promising lines from the second year of testing in the advanced yield trials (right side of Fig. 1). Since the main objective was to use the information to decide on variety release, this trial compared yield under irrigated and drought treatments of entries whose flowering period matched through staggered sowing. Desirable breeding lines and varieties for release were those that had relatively high yield under drought relative to that under irrigation and an absolute yield under drought equal to or greater than that of the check variety of a similar maturity class. The methodology of this drought evaluation trial is still part of the present breeding program.



Promising lines are evaluated under managed drought. 5. Managing the final drought evaluation trials

The *final drought evaluation trials* are planted late with multiple sowings to collect data on drought response despite the significant year-to-year variation at the onset of the dry season. This trial can accommodate 18 to 20 advanced lines and two to four drought-tolerant checks of different growth duration. Ideally, the experimental design is a randomized complete block with split plots, composed of water treatments (stress and sprinkler-irrigated control) and subtreatments (tested lines), with at least four replications. To prevent interference of one water treatment upon the other, there is a safe distance between irrigated and stressed plots, which means that water treatments within the same block need some spatial separation. For this reason, this fully randomized experimental design is not always used in our conditions. In most years, depending on area availability, the irrigated control is conducted as a complementary experiment in the same experimental area.

The entries tested are arranged in a minimum of four rows, 5 to 6 m long, spaced 0.40 m apart. The latest-maturing materials are sown from 2 to 6 weeks in advance in relation to the earliest ones, depending on the relative differences in their growth cycle duration, to allow for a reasonable synchronization of reproductive development. In the case of multiple sowing dates, the sequential sowings of the same entry are spaced 1 week apart.

Sowing dates are not considered as part of the experimental treatments in the analysis rather, one sowing date for each entry (the one that best matches the flowering time) is used to evaluate drought tolerance. Entries whose date of flowering falls within a deviation of more than 5 days from the average date of flowering are discarded. Depending on the homogeneity of the area, it is preferable to reduce the number of sowings and maintain the desired number of rows per plot. It is also highly recommended to have three extra rows surrounding the entire experiment to minimize border effects and provide some protection against insects and diseases. Since these experiments are grown out of season, they are especially prone to various pests, including birds. Sorghum rows or extra plots may act as attractants to birds and provide some degree of control.

We endeavor to provide a uniform drought stress by

- *Rotation of the site.* Upland rice yields decrease noticeably after the second consecutive sowing in the same area. To avoid this problem, a homogeneous experimental area of approximately 2 ha was divided into two modules of 1 ha each. The fields are rotated with soybean or maize, followed by rice, and the area is sown to pasture for 2 years after the rice. In this way, the field is sown to rice only once every 4 years.
- Uniform soil preparation. The occurrence of hard soil pans, superficial soil compaction, and any soil physical or chemical discontinuity must be avoided to minimize the already high spatial variation when drought stress is applied. Soil preparation begins at the onset of the rainy season, in early October, with deep plowing, using a moldboard plow, to incorporate previous crop residues, followed by repairing of the levees. The area is then left undisturbed until planting of the experiment. Depending on weed contamination, herbicide application may be necessary, followed 1 week later by a light harrow to level the soil and incorporate any vegetation residue.
- Uniform distribution of irrigation. Water is applied to both the irrigated treatment and during the vegetative stage of the drought treatment, whenever rainfall is below pan evaporation for a period of 4 consecutive days. The irrigation sprinklers are carefully placed to ensure uniformity. The distribution is checked by installing cans at various distances from the sprinklers, just above the plant canopy. It is desirable to saturate the soil by applying excess water at the last irrigation before beginning the drought treat-

Uniform conditions in the drought nursery are essential. ments. Soil samples are taken the day after the last irrigation and then at weekly intervals to determine soil moisture content.

- *Beginning of the stress.* The decision on when to induce stress has implications for the level of stress imposed and the timing of stress may be constrained by weather conditions. At our experimental site, rains decline substantially in April, but occasional showers can disturb the evaluation protocol. In contrast, almost no rain occurs in May, but low temperatures may occur. This temperature hazard, although occurring less frequently (usually one in four years) than the April showers, may cause the loss of data in some years.
- *Monitor the plant water status.* In addition to monitoring soil water content, we measure leaf water potential to monitor the average plant stress level (see Fig. 2). The combined measurements of leaf water potential and soil water content help determine when to terminate the stress for the targeted yield reduction. In our experiments, leaf water



Fig. 2. Measuring the water status of plants under the drought stress and in irrigated treatments.

Compare the performance of plants that flower at the same time and under the same drought conditions. potential is measured twice a week, from 1300 to 1500. We usually monitor the two checks plus a few entries, randomly chosen. We use a pressure chamber and measure a maximum of six genotypes per block with four leaf samples per genotype, measuring two samples at a time. We usually rewater before water potential values become lower than -2.0 MPa to avoid excess damage to the plants and to achieve our target yield.

At our site, under the prevailing climatic conditions of the dry season, and starting the stress imposition under full soil water saturation, it normally takes from 15 to 20 days to attain an adequate level of stress. An average vapor pressure

deficit of 18 to 22 millibars at 1500, kept for 18 to 22 days, is capable of inducing a 30% yield loss in the resistant check.

• *Comparing genotypes at the same plant development stage.* Adjustments of sowing dates allow for a certain degree of synchronization among the reproductive stages of most lines. However, even small differences in phenological development may represent a significant difference in drought response. Plant size also influences plant response to drought. Consequently, comparing genotypes of different growth cycles would normally favor the early ones that develop a smaller leaf area in relation to the late ones. For this reason, it is best to confine comparisons among those entries of the same maturity group and it is necessary to include two or more checks of different growth duration. We now use Guaraní as an early check and Rio Paranaíba as a late check, replacing IAC 25, IAC 165, and IAC 47, now highly susceptible to blast.

It is important to remember that drought takes place some time after the rains subside or irrigation is discontinued. At our site, under the prevailing evaporative demand of the dry season, it normally takes from 5 to 7 days for a stressed plot to be differentiated from the irrigated control. So, when selecting the desired target stage for drought induction, care must be taken to make sure that induction begins in time to allow for adequate drought pressure during the targeted period. We choose flowering and early grain filling as the critical period and normally provide the last irrigation when around 10% of the tillers are at booting.

Use direct and indirect measures of drought tolerance to evaluate lines.

6. Using direct and indirect evaluation criteria for drought tolerance

We found it very difficult to attribute a drought-tolerance score based strictly on visual criteria during the stress. However, in the early years of the program, when some thousands of entries were being evaluated, we relied on the visual score as the only possible alternative and it was very helpful.

To evaluate the response of promising advanced lines for variety release, we use yield under stress and the relative yield loss in relation to the irrigated control (drought index) as the main criteria. Such experiments, however, require a large and uniform experimental area in addition to good crop and water management to assure adequate plant growth and uniformity.

We use the indirect measures of drought tolerance of

- delay in flowering,
- leaf rolling and leaf drying,
- panicle exsertion and panicle size, and, especially,
- spikelet fertility.

Box 1. Measuring spikelet fertility.

Visual scoring

Differences among entries are detected and translated into numerical scores. We rate spikelet fertility from 1 to 9, using the scale below.

Score	Percent spikelet fertility					
1	Higher than 90%					
2	80-90%					
3	70–80%					
4	60–70%					
5	50–60%					
6	40–50%					
7	30–40%					
8	20–30%					
9	Lower than 20%					

Measure the percentage of unfilled grains

Sample a reasonable number of panicles per plot, separate the empty and filled grains (here both the complete and partially filled caryopsis are included), and count only the empty ones. Then, determine the number of filled grains in an adequate subsample of the total sample and weigh the whole sample. The number of filled grains is then determined by simple calculation.

In our experience, spikelet fertility is the most useful visual indicator of the response of upland rice during the reproductive stage and, whenever it is more easily assessed than yield (see Box 1 for how to measure spikelet fertility), we see no restriction to using it instead. It also has the advantage of not being influenced by factors other than drought in the grain-filling period.

The correlation of these indirect traits with grain yield under stress is shown in Table 1. Note that

- yield under drought is correlated (r = 0.6) with yield potential,
- spikelet fertility is highly correlated with yield under drought conditions,
- leaf rolling and leaf water potential have a low correlation (nonsignificant) with yield under drought, and
- there is a significant correlation between yield under stress and the number of days from stress imposition to flowering.

The drought response of the tested entries still has some confounding effect with growth stage even though we staggered the planting to match the reproductive development of the tested entries.



7. Some examples of the response of different rice lines under drought

Table 2 shows the performance of 18 varieties—ten from CNPAF (CNA lines), two from CIRAD (IRAT 216 and IRAT 335), two from the Agronomic Institute of Campinas (IAC

	Trait								
Trial	Stressed yield ^a	Straw yield	Stress timing ^b	Number of panicles	Number of spikelets panicle ^{_1}	Spikelet fertility	Weight of 100 grains	Leaf rolling	Leaf water potential
Irrigated yield	0.60**	0.328 ns	-	–0.067 ns	0.143 ns	0.339 ns	0.427 ns	–0.257 ns	–0.176 ns
Stressed yield	-	–0.353 ns	-0.541**	–0.226 ns	0.056 ns	0.842**	0.481*	–0.317 ns	0.174 ns

** and ** = significant at 5% and 1% level, respectively. Number of days from beginning of stress to date of 50% flowering in the stressed trial. ns = nonsignificant.

Table 2. Results of a final evaluation trial involving 14 upland advanced lines originating from the regional yield trial of the Brazilian breeding network. Numbers followed by the same letter are not significantly different at the 5% probability level.

Entry	Yield under drought stress (g m ⁻²)	Yield loss ^a (%)	Flowering date ^b	Stress timing ^c	Straw biomass (g m ⁻²)	Spikelet fertility (%)	Leaf rolling score	Midday leaf water potential (MPa)
IAC 84-198	235.3 a	19.8 cd	68.7	6.7 fg	304.1 de	71.2 ab	4.3 ab	–1.75 a
Guaraní	233.6 a	20.2 cd	69.3	7.3 fg	344.7 cde	81.1 a	4.3 ab	–1.88 a
CNA 6710 CNA 6891 IRAT 216 IAC 1176	221.7 ab 218.1 ab 211.4 abc 203.8 abc	25.8 bcd 18.9 cd 35.0 abcd 28.6 bcd	73.0 84.7 93.0 65.7	9.0 def 7.7 efg 9.0 def 5.7 g	350.9 cde 525.7 abcde 622.8 ab 380.5 abcd	63.0 abc 55.7 bcd 51.5 bcde 63.8 abc	4.3 ab 4.2 ab 4.3 ab 4.5 ab	–1.89 a –1.88 a –1.84 a –1.87 a
R. Paranaíba	190.2 abcd	38.7 abcd	98.3	14.3 a	516.4 abcde	56.5 bcd	4.5 ab	–1.68 a
CNA 4140 IAC 165 IAC 1175 IAC 47 CNA 6881 CNA 6187 CNA 7101 CNA 7127 CNA 7141 IRAT 335 CNA 7066 F value CV (%)	188.4 abcd 182.8 abcde 131.8 bcdef 123.8 cdef 97.3 def 92.0 ef 87.1 f 70.8 f 64.3 f 55.0 f 13.8** 20 3	36.5 abcd 9.8 a 47.4 abcd 52.9 abc 56.8 abc 64.6 ab 55.8 abc 66.1 ab 65.2 ab 54.9 abc 76.4 a 6.0**	94.7 69.7 71.3 98.0 94.0 96.7 88.0 87.0 95.0 72.0 95.5	10.7 cd 7.7 efg, 9.3 def 14.0 ab 7.7 efg 12.7 abc 11.0 cd 10.0 cde 11.0 cd 10.0 cde 11.3 bcd 20.1 ** 9 3	497.8 abcde 338.2 cde 386.5 bcde 480.6 bcde 632.0 ab 574.9 abc 521.7 abcde 631.1 ab 548.9 abcd 276.2 e 753.3 a 7.1 ** 18 1	50.7 bcde 67.9 ab 46.5 bcdef 20.5 g 35.0 defg 39.3 cdefg 23.7 fg 34.1 defg 24.3 fg 26.9 efg 13.9** 11 9	4.2 ab 4.5 ab 4.8 a 4.5 ab 3.8 b 4.5 ab 4.7 a 4.2 ab 4.8 a 4.5 ab 4.8 a 3.4 ** 7 1	-1.67 a -1.91 a -1.80 a -1.80 a -1.73 a -1.77 a -1.79 a -1.88 a -1.79 a -1.80 a 1.1 ns 6 6

"Percentage yield reduction of the stressed plots in relation to the irrigated plots. "Number of days from sowing to 50% flowering. "Number of days from beginning of stress to date of 50% flowering in the stressed trial.

lines), two resistant checks (Guaraní and Rio Paranaíba, of short and long growth duration), and traditional upland varieties IAC 47 and IAC 165—evaluated in the managed-drought experiment of the regional advanced yield trials of the national breeding network coordinated by EMBRAPA.

The 18 entries were separated into three groups according to growth duration and sowing took place on five dates, spaced 1 week apart, with three sowings per group, beginning with the latest group and ending with the earliest one. Drought stress began on 5 May and was relieved on 27 May. Most of the entries in the irrigated plots flowered (50% flowering) from 12 to 19 May. On the last day of stress imposition, water potential ranged from -1.6 to -2.0 MPa and leaf rolling attained values of 4 to 5 in the stressed plots. Note that

- The drought-tolerant checks Guaraní and Rio Paranaíba had a higher yield and spikelet fertility than most of the entries of corresponding growth duration.
- The yield loss of Rio Paranaíba, the medium-duration check, was more than that of the short-duration check, Guaraní. This effect of maturity is normally observed in this kind of experiment, that is, the response to drought has to be evaluated within the same

maturity group; otherwise, long-growth-duration genotypes are seldom classified as tolerant.

- Independent of the growth cycle duration, we consider entries that lose less than 30% yield in relation to the irrigated control as tolerant, from 30% to 50% as moderately tolerant, from 51% to 80% as moderately susceptible (MS), and from 80% onward as susceptible (S).
- The data from the managed-drought trials were used to recommend the release of CNA 4140, IRAT 216, and CNA 6187 (Rio Paraguai, Rio Verde, and Carajás, respectively) and to discard CNA 7066, although it has excellent grain appearance.
- The performance has to be tested for at least two years to establish drought response with reliability.

8. Lessons learned from the drought evaluation program

The main problem we face is assuring adequate levels of drought stress at the target growth stage. Our experimental farm is in a low to medium climatic risk zone for the normal cultivation season. Our strategy of delaying planting, although increasing the chances of inducing drought at the desired plant growth stages, is not completely rain-proof. Moreover, in some years, low night temperatures may induce spikelet sterility, thus masking results and contributing to failure as well. Delayed sowing may also cause some undesirable effects on plant size and growth duration, besides increasing the incidence of blast. Nevertheless, the strategy worked well in a reasonable number of years.

Because of their adequate performance under drought conditions, traditional upland genotypes (landraces) collected regionally during the 1970s were used extensively as progenitors in the early period of CNPAF's upland breeding program. However, the majority of the derived pure lines were subsequently discarded because of their high susceptibility to blast. In the same period, several African genotypes were also used as parents and the crosses with 63-83 and improved Brazilian varieties gave origin to the widely used cultivars Guaraní (IAC 25 × 63-83) and Rio Paranaíba (IAC 47 × 63-83), both released in 1986 (Table 3).

The strategy of confining crosses to progenitors of the japonica group showing adequate drought tolerance has proved useful. The varieties derived from such crosses possess a higher

Table 3. Year of release, progenitor group, growth cycle duration, plant and grain type, and response to drought of upland rice releases from EMBRAPA Rice and Beans to the savanna region of Brazil.

Cultivar	Year of release	Progenitor group	Growth duration	Plant type	Grain type	Drought toleranceª
Cuiabana	1985	Japonica, indica	Medium	Traditional	Long bold	MS
Guaraní	1986	Japonica	Early	Traditional	Long bold	Т
Rio Paranaíba	1986	Japonica	Medium	Traditional	Long bold	MT
Araguaia	1986	Japonica, indica	Medium	Traditional	Long bold	MS
C. América	1987	Japonica	Early	Traditional	Long bold	MT
Rio Paraguai	1992	Japonica	Medium	Traditional	Long bold	MT
Rio Verde	1992	Japonica	Medium	Traditional	Long bold	MT
Progresso	1993	Japonica, indica	Medium	Modern	Long slender	MT
Caiapó	1994	Japonica, indica	Medium	Traditional	Long	MT
Carajás	1994	Japonica	Early	Traditional	Long bold	Т
Maravilha	1996	Japonica, indica	Medium	Modern	Long slender	S
Primavera	1996	Japonica, indica	Early	Modern	Long slender	MS
Canastra	1996	Japonica, indica	Medium	Modern	Long slender	MT
Confiança	1996	Japonica, indica	Medium	Modern	Long slender	MS
Carisma	2000	Japonica, indica	Early	Modern	Long slender	MS
Bonança	2001	Japonica, indica	Medium	Modern	Medium slender	MS

^aT = tolerant, MT = moderately tolerant, MS = moderately susceptible, S = susceptible.

Stress timing is critical for making progress.

High-yielding drought-tolerant varieties can be developed by selecting for both yield and drought tolerance. degree of drought tolerance than those involving indica sources of blast resistance, such as Cuiabana and Araguaia, released in the same period. Moreover, releases after 1994, such as Maravilha and Primavera, developed after some changes were made in the breeding program strategy, show less drought tolerance.

Recovering the original level of drought tolerance in the new releases is feasible. The variety Canastra (Table 3) as well as some new advanced lines recently tested (data not shown), all japonica by indica derivatives, have shown the same level of drought tolerance as their original japonica progenitors. In this new generation of crosses, a more adequate balance among plant type, grain quality, and drought tolerance has been achieved.

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Notes

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