

## BRS 368RF: A glyphosate tolerant, midseason upland cotton cultivar for Northeast and North Brazilian cerrado

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**Abstract:** Cotton cultivar BRS 368RF was developed by EMBRAPA in a partnership with the Bahia Foundation, aiming to create high-yielding germplasm by adding transgenic traits addressed to Brazilian Savanna (cerrado) of the Northeast and North regions. BRS 368RF is a midseason cultivar and has yield stability and transgenic tolerance to glyphosate.

**Key words:** *Gossypium hirsutum*, glyphosate tolerance, and lint yield.

### INTRODUCTION

The Brazilian cerrado is located in central Brazil and has a great potential to produce meat, food and fiber crops. Efforts have been made to develop crop cultivars adapted to this environment (Morello et al. 2010, Amabile et al. 2013, Morello et al. 2015). Moreover, in recent years, cotton-growing areas have dramatically increased in the cerrado region located in the states of Bahia, Piauí, Maranhão, and Tocantins, in the Brazilian Northeast and North regions. In these areas, the total rainfall is lower than that in the central Brazil. Additionally, the cerrado of the North and Northeast regions is a distinct environment due to its high average temperature. Unfortunately, little effort has been made to develop upland cotton cultivars (*Gossypium hirsutum* L. r. *latifolium* Hutch) for this environment (Morello et al. 2012a, Morello et al. 2012b).

Embrapa's cotton-breeding program has been carried out in partnership with the private sector, which has provided financial and technical support by means of the Fund for the Development of Cotton Agribusiness (FUNDEAGRO - *Fundo para o Desenvolvimento do Agronegócio do Algodão*) and the Bahia Foundation. BRS 368RF has been released to meet the demand for high yield potential and transgenic traits cotton cultivars and it is recommended for all cotton growing areas of the North and Northeast Brazilian cerrado.

### GENETIC ORIGIN AND DEVELOPMENT

Biotechnological tools have been applied in cultivar development, in order to add to these cultivars specific traits that would be difficult to be obtained by using conventional breeding (Parentoni et al. 2013). An important transgenic trait in cotton production is tolerance to herbicide, especially to glyphosate, due to the effective control of a wide range of annual and perennial weeds that

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this herbicide provide to the plant. This trait has been successfully introduced into cotton plants, and has been regularly used. The action mode of the herbicide glyphosate is the inhibition of the enzyme 5-enolpyruvylshikimate-3-phosphate synthase (EPSP synthase) in the shikimate acid pathway. This inhibition blocks the synthesis of the aromatic amino acids phenylalanine, tyrosine, and tryptophan, leading to the death of susceptible plants (Pline et al. 2003). The commercial cotton transgenic event MON 88913, known as Roundup Ready® Flex (RF), was used in this study to confer tolerance to herbicide glyphosate in cotton germplasm adapted to North and Northeast growing areas.

BRS 368RF was obtained from a backcross breeding program, followed by pedigree selection method, to incorporate the commercial event MON 88913. Conventional cotton cultivar BRS 286 (CNPA Ita 90 x CNPA 7H) was used as the recurrent parent. BRS 286 has good fiber quality, earliness and adaptation to the Northeast Brazilian cerrado. The high-yielding cotton cultivar Sure Grow 125RF [Sure Grow 125\*3/Donor RR] (Bowman et al. 2006) was used as trait donor. However, Sure Grow 125RF is susceptible to cotton blue disease, caused by *Cotton leafroll dwarf virus*, and does not have good fiber quality. BRS 368RF was originated from a biparental cross followed by three backcrosses between BRS 286 and Sure Grow 125R (BRS 286/4\* Sure Grow 125R), carried out in 2007 and 2008. From the BC<sub>3</sub>F<sub>2</sub> generation, only plants homozygous for the RF gene were advanced to progeny rows tests.

A total of 50 progeny rows (BC<sub>3</sub>F<sub>2,3</sub>) were evaluated in the 2009/2010 season in Santa Helena de Goiás, GO. The progeny CNPA BA 2009 286 RF 50 was selected based on apparent yield potential, high volume instrument fiber properties, disease resistance, and overall plant conformation. During the subsequent season, the obtained seeds were grown and again screened for the presence of the RF gene. BRS 368RF was subsequently treated as a pure line, evaluated in four sites in the 2010/2011 season. Afterwards, eleven trials were carried out in the states of Bahia, Goiás, Mato Grosso do Sul, and Mato Grosso, in the 2011/2012 season, and two trials were performed in the state of Bahia, in the 2012/2013 season.

## CULTIVAR PERFORMANCE

BRS 368RF is a midseason to early maturity, similar to BRS 286. When grown in São Desidério, BA (lat 12° 57' S, long 45° 58' W, alt 794 m asl), plants are of medium size, reaching 110 to 120 cm, when 50 to 75g of the active ingredient of growth regulator (mepiquat chloride and chlormequat chloride) are applied. At these environmental conditions, the

**Table 1.** Means of total cottonseed yield (CY), lint percentage (LP), and lint yield (LY) of the cotton cultivars BRS 368RF and BRS 286 (control), in 17 field performance trials of cultivation value and use (VCU)

Municipalities/State	Season	BRS 368RF			BRS 286 (control)			CV*
		CY (kg ha <sup>-1</sup> )	LP (%)	LY (kg ha <sup>-1</sup> )	CY (kg ha <sup>-1</sup> )	LP (%)	LY (kg ha <sup>-1</sup> )	
Luís Eduardo Magalhães/BA	2010/11	2451.0	41.7	1022.1	2208.0	41.2	910	16.3
Santa Helena de Goiás/GO	2010/11	4148.8	40.9	1768.9	3726.3	39.4	1512.5	10.3
Santo Antônio de Goiás/GO	2010/11	5447.0	41.0	2232.9	5995.0	39.5	2362.5	11.0
Chapadão do Sul/MS	2010/11	2834.5	42.2	1198.3	2663.4	40.4	1077.7	13.3
São Desidério/BA	2011/12	3495.0	41.8	1447.5	3327.0	42.3	1402.5	16.7
Riachão das Neves/BA	2011/12	3547.5	42.8	1519.5	2494.5	43.0	1080.0	24.4
Correntina/BA	2011/12	3297.0	42.8	1414.5	3664.5	43.5	1587.0	17.8
Santa Helena de Goiás/GO	2011/12	2382.0	42.9	1024.5	2458.5	42.3	1044.0	12.4
Santo Antônio de Goiás/GO	2011/12	4681.5	40.4	1873.5	4926.0	43.4	2143.5	12.6
Cristalina/GO	2011/12	4473.0	43.8	1963.5	3829.5	44.6	1713.0	14.6
Montividiu/GO	2011/12	5545.2	42.2	2344.2	5497.2	42.7	2351.2	16.0
Chapadão do Sul/MS	2011/12	1870.3	45.2	844.6	1982.7	44.6	884.6	17.1
Sinop/MT	2011/12	2226.8	43.6	969.0	2234.3	42.2	946.8	12.7
Primavera do Leste/MT	2011/12	4998.0	42.2	2106.0	4854.0	45.0	2170.5	15.4
Campo Verde/MT	2011/12	5583.0	43.3	2419.5	3793.5	43.1	1641.0	23.5
Riachão das Neves/BA	2012/13	3765.5	48.3	1819.3	3074.7	45.9	1412.4	16.6
Luís Eduardo Magalhães/BA	2012/13	4659.1	43.8	2040.9	3794.9	42.8	1627.7	13.5
Mean		3847.4	42.9	1647.6	3560.3	42.7	1521.6	

\* Coefficient of variation (%) for cottonseed yield (kg ha<sup>-1</sup>)

**Table 2.** Traits of cultivar BRS 368RF and BRS 286 (control)

Traits	BRS 368RF	BRS 286	IAC 25
First flower (DAE) <sup>*</sup>	54	54	-
First open boll (DAE) <sup>*</sup>	115	115	-
Boll weight (g) <sup>a</sup>	6.0	6.0	-
Fiber length (UHML) (mm) <sup>a</sup>	29.1	29.9	-
Uniformity index (ML/UHML - %) <sup>a</sup>	84.5	84.8	-
Strength HVI (gf tex <sup>-1</sup> ) <sup>a</sup>	28.2	30.7	-
Micronaire reading <sup>a</sup>	4.1	4.2	-
Elongation (%) <sup>a</sup>	7.7	8.6	-
Reflectance – Rd (%) <sup>a</sup>	77.2	77.4	-
Yellowness (+ b) <sup>a</sup>	8.0	8.3	-
Short fiber index (%) <sup>a</sup>	7.0	5.7	-
Total seed oil (%) <sup>b</sup>	17.5	18.8	-
<i>Cotton leafroll dwarf virus</i> <sup>c</sup>	0.0	0.0	-
False mildew <sup>d</sup>	3.5	4.0	-
Bacterial blight <sup>d</sup>	1.0	1.0	-
Ramulosis <sup>e</sup>	86.3	-	33.5
Root-knot nematode <sup>f</sup>	2.1	-	1.3
Fusarium wilt <sup>g</sup>	18.3	-	9.6

<sup>\*</sup> Data recorded in São Desidério (lat 12° 57' 71" S, long 45° 58' 92" W, alt 794 m asl), BA, Brazil

<sup>a</sup> Means of 17 field trials in the 2010/2011, 2011/2012, and 2012/2013 seasons, in several environments (states of Goiás, Bahia, Mato Grosso, and Mato Grosso do Sul);

<sup>b</sup> Total seed oil (%) measured by nuclear magnetic resonance (NMR); <sup>c</sup> Incidence (%) of plants with cotton blue disease symptoms - data from two trials with no control of virus vector (*Aphis gossypii*), confirmed by SNP marker (Fang et al. 2010); <sup>d</sup> Disease severity (grades from 1 = resistant to 5 = highly susceptible) - data from two trials with no fungicide application. For bacterial blight, resistance was confirmed by using SNP marker (Xiao et al. 2010); <sup>e</sup> Amaral disease index calculated (Amaral 1969) - data from two trials artificially inoculated with *Colletotrichum gossypii* var. *cephalosporioides*, by using methods proposed by Oliveira et al. (2010); <sup>f</sup> Gallings index (Zhang et al. 2006); <sup>g</sup> Disease index (Machado et al. 2009).

first flower appears at about 54 days after emergence (DAE), and the first boll opens at about 110 to 120 DAE. Total harvest was performed at 150 to 160 DAE.

BRS 368RF plants are phenologically similar to BRS 286 plants, having trichome on the leaves and on the main stem. Also, BRS 368RF presents normal-shaped leaves and bracts (7 to 12 lobes). Leaves are glanded and nectaried. Generally, the first reproductive branch is inserted on the plant fifth node, and branches present oblique angle insertion. Flowers have petals, anthers, and pollen cream-colored. Full-size green bolls are longer than their width and are broader in the middle. Most of bolls have five locks, of which four occur occasionally. Lint and fuzz are white-colored. BRS 368RF are suitable for picker harvesting with open bolls resistant to shattering, but not storm proof.

Injury to BRS 368RF plants was not observed with any glyphosate application or combination of glyphosate applications past the 2-leaf stage. In addition, glyphosate applied topically to BRS 368RF cotton after the 2-leaf stage does not reduce lint yield, comparing to manual weeding (data not shown).

BRS 368RF is resistant to bacterial blight [caused by *Xanthomonas citri* subsp. *malvacearum*] and to cotton blue disease [caused by *Cotton leafroll dwarf virus*- CLRDV]; moderately susceptible to false mildew [caused by *Ramularia areola*], *Fusarium oxysporum* f. sp. *vasinfectum* - *Meloidogyne incognita* complex, and to *Rotylenchulus reniformis*; and susceptible to ramulosis [caused by *Colletotrichum gossypii* var. *cephalosporioides*]. Fungicide foliar sprays are necessary to avoid losses caused by false mildew. Resistances to bacterial blight and to blue disease were measured in field conditions and confirmed by genotyping a sample of seeds, considering the presence of molecular markers CIR 246, linked to the *B<sub>12</sub>* gene, which confers resistance to bacterial blight, and of the marker DC20027, linked to the *Cdb* gene, which confers resistance to cotton blue disease (Fang et al. 2010, Xiao et al. 2010).

The evaluation of 17 field performance trials of cultivation value and use (VCU) in central and northeastern Brazil in 2010/2011, 2011/2012, and 2012/2013 revealed that cultivar BRS 368RF produced 8.06% more cottonseed and 8.2% more lint yield than cultivar BRS 286. Lint percentage of cultivar BRS 368RF did not differ from that of BRS 286 in all the trials (Table 1). HVI measurements revealed desirable physical attributes of a medium fiber length cultivar: micronaire reading from 3.9 to 4.8; fiber length (UHML) from 27.6 to 31.2 mm; relative strength from 27.4 to 29.2 gf

tex<sup>1</sup>; reflectance from 71.5 to 80.5%; yellowness (+b) from 7.3 to 8.8; short fiber index from 5.1 to 8.7. Mean values of fiber quality obtained from 17 field trials are summarized in Table 2.

## SEED MAINTENANCE AND DISTRIBUTION

BRS 368RF is catalogued in the Ministry of Agriculture, Livestock and Food Supply under the number 30482. Basic seeds are produced by Embrapa Products and Markets (EMBRAPA SPM). The Bahia Foundation, working in partnership with Embrapa, is responsible for the certified seed.

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