

“YASINI” - A NEW HIGH YIELDING BLACK SHANK DISEASE RESISTANT CHEWING TOBACCO VARIETY FOR TAMIL NADU

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Yasini (BSR-1) is a high yielding and Black Shank disease resistant chewing tobacco variety developed through back cross breeding ((VR-2 Beinhart1000-1) VR-2) at ICAR-CTRI Research Station, Vedasandur for Chewing tobacco farmers Tamil Nadu, more particularly for coastal area. It was tested in the name of BSR-1 in All India Coordinated Research Project on Tobacco, it consistently performed better in the Research station as well as in the black shank sick fields in Vedaranyam areas of coastal chewing tobacco belt (Hot spot area) in replicated yield trials and recorded 80% higher yield over check variety VR-2 (check -1) and >22% over Kaviri (check-2). In agronomic trials, it recorded 5.8% higher leaf yield over Kaviri. In on-farm trials conducted for three years, the Yasini recorded 30% higher yield over VR-2 and 19% over Kaviri. Yasini produces broad ovate leaves, dark green, sessile with moderately puckered, margins slightly wavy, tip acute, base cuneate, auricle prominent, leaf number vary from 24-25 with around 10 economic leaves, spangling medium, leaf length 70-75 cm and width 40-45 cm as compared to its parent VR-2 that has plants open, slightly drooping lamina, lanceolate, green leaves with medium auricle. By cultivating this variety, Tamil Nadu chewing tobacco farmers will raise a healthy crop without application of fungicide for black shank disease and thus cutting down the cost on chemical fungicides thereby reducing production costs and increasing net returns to the growers besides being environment friendly.

INTRODUCTION

Tobacco (*Nicotiana tabacum* L.) is one of the most important commercial crops of India. It is the major contributor for the Indian economy and earning foreign exchange. This crop was introduced to India from its originating places, South and

North America through European traders (Sierrobet *al.*, 2014). Tobacco can be classified broadly into two categories, 'smoke' and 'smokeless'. Chewing tobacco comes under smokeless category and are mainly cultivated in Tamil Nadu, West Bengal, Bihar, Assam and Uttar Pradesh. In Tamil Nadu, chewing tobacco is being cultivated as commercial crop for more than 10 decades in Dindigul, Karur, Erode, Coimbatore, Thirunelveli, Namakal, Salem, Vedaranyam, and Nagapattinam. The other tobaccos cultivated in Tamil Nadu are cigar and cheroot that are cultivated in smaller areas.

Black shank (*Phytophthora parasitica*) disease is a devastating root and crown rot disease of all types of tobacco, causing yield losses up to 100%. Black shank occurs sporadically in every type of tobacco and causes more damage, to tobacco grown under high rainfall or irrigated conditions in light soils of Karnataka, Andhra Pradesh, Gujarat and some parts of Tamil Nadu. This disease can be dealt with two ways one by plant production & protection technologies and another by developing high yielding resistant varieties (Sullivan *et al.*, 2005). Sun-cured chewing tobacco is grown in about 2000 acres in Vedaranyam tract on the coastal belt of Tamil Nadu. VR 2 is the only popular variety being grown in this area. To overcome these issues, breeding programme for development of a superior chewing tobacco (Sun-cured type) with resistance to black shank disease has been launched during 1983-8 resistance to black shank disease. In this article the progress made in the project and a superior selection, BSR 1 developed, tested and released as new variety, "YASINI" is being reported.

Key words: Yasini, Black Shank, Chewing Tobacco and yield

MATERIALS AND METHODS

In order to develop a high yielding and Black Shank disease resistant chewing tobacco variety, the popular chewing tobacco cultivar VR-2 was crossed with a promising tobacco line Beinhart1000-1 and the resultant segregating materials in the next generations were handled through Back-cross breeding method (Fig 1). Among the advanced breeding lines, a promising chewing line, BSR-1 was identified based on yield and morphology. Further, the BSR-1 was tested All India Coordinated Research Project on Tobacco, CTRI, Rajahmundry, black shank sick fields in Vedaranyam was evaluated. In view of its yield superiority over the check, BSR-1 in replicated trials, it was advanced for testing under bulk trial for four years (2006-2009) through multi location trials under All India network Project on Tobacco (AINPT). In order to assess the response of BSR-1 to agronomic variables a trial was conducted with different levels of nitrogen (0, 100, 150, 200 and 250 kg N). Observations on morphological characters and yield parameters were recorded. Cured leaf samples were analyzed for chemical leaf quality parameters *viz.*, nicotine (%), reducing sugars (%) and chlorides (%). The data recorded was used for standard statistical procedures as per Gomez and Gomez, 1984).

DNA extraction and PCR

Genomic DNA was isolated from 30 days old seedlings by CTAB method, purified DNA was quantified by spectrophotometer (Nanodrop) and analyzed in agarose gel electrophoresis. The DNA isolated was amplified SSR markers from the list published by Bindler *et al.*, (2007) in thermal cycler (Eppendorf). For each PCR reaction 50ng of DNA was taken as template. The PCR was performed in 25µL final volume containing 1xPCR buffer (10 mM Tris-HCl pH 8.3, 50 mM KCl, 1.5 mM MgCl₂, 0.2 mM dNTPs, 10 pmol of each forward and reverse primers and 1U of Taq polymerase PCR programme consisted of Initial denaturation at 94°C for 5 minutes, 34 cycles of consisting of denaturation at 94°C for 1 minute, annealing at 55°C for 1 minute, extension 72°C for 1 minute followed by final extension at 72°C for 10 minutes. The PCR products were analyzed QIAxcel Advanced (Qiagen) capillary system with QIAxcel ScreenGel Software.

BACKGROUND

Yasini (Tested as BSR-1) is a high yielding and Black Shank disease resistant chewing tobacco variety developed through back cross breeding ((VR-2 Beinhart1000-1) VR-2) at ICAR-CTRI Research Station, Veda sandur for Chewing tobacco farmers Tamil Nadu, more particularly for coastal areas. The crosses were attempted during 1983-84 (total 5 crosses were made using VR 2 as female and Bhagyalakshmi, F2-7-1, CKV4, Beinhart, and Javiz as male) and F₁s were raised during 1984-85 at CTRI Research Station Veda sandur and farmers field (M/s Swasthic Company Farm) at Vedaranyam. The details of the crosses and progressive generation advancement are given in the Fig 1. (ICAR-CTRI, 2022).

A total of 1000 F₂ population of VR2 Beinhart were grown during 1985-86 at M/s Swasthic company in the blank shank sick field. During this season there was two cyclones in this area and plants withstand after two cyclones were selected and resistance to black shank was artificially tested at CTRI, Rajahmundry and natural condition at Vedaranyam. In 1986-87, the seeds of F₂ selections of VR-2 Beinhart1000-1 collected at Swastick Farm, Vedaranyam were artificially screened for black shank resistance at CTRI Rajahmundry. A total of 16 selections (6 plants in each selection) were screened and 3 selections (Selection-4, Selection-6 and Selection 14) showed 0 infection and these lines were again backcrossed with recurrent parent, VR 2 and progenies were tested under natural black shank sick filed at Vedaranyam. In F₃, the resistant to black shank disease were identified and again grown at Vedaranyam and repeated back crosses with parent VR 2 was made till BC₆ and selections were made after that for yield and resistance to black shank disease. Among the derivatives, a promising line coded as BSR-1 was tested in replicated trials and multilocation trials under All India network Project on Tobacco (AINPT) during 2006-09. Simultaneously agronomic evaluation trials, bulk trials, pre-release trials and screening for resistance to major pest and diseases were also taken up.

Based on the performance of BSR-1, it was identified for release during VIIIth group meeting of

Stage	Cross/Selection	Year
Crossing	VR 2 Beinheart	1983-84
F ₁	F ₁ -11	1984-85
F ₂	F ₂ -6	1985-86
F ₃	F ₃ -5-1	1986-87
BC ₁ -12	F ₃ -5-3 #2 X VR.2	1987-88
BC ₁ -13	F ₃ -5-3 #3X VR.2	1987-88
BC ₂ -27	BC ₁ -12#1X VR.2	1988-89
BC ₂ -28	BC ₁ -14#1X VR.2	1988-89
BC ₂ -29	BC ₁ -14#2X VR.2	1988-89
BC ₂ -30	BC ₁ -15#2X VR.2	1988-89
BC ₃ -20	BC ₂ -29#1X VR.2	1989-90
BC ₃ -21	BC ₂ -30#1X VR.2	1989-90
BC ₃ -S1	BC ₂ -27#1X VR.2	1989-90
BC ₃ -S2	BC ₂ -29#1X VR.2	1989-90
BC ₃ -S3	BC ₂ -30#1X VR.2	1989-90
BC ₄ -17	BC ₃ -20#1X VR.2	1990-91
BC ₄ -18	BC ₃ -20#2X VR.2	1990-91
BC ₄ -19	BC ₃ -31#1X VR.2	1990-91
BC ₅ -10	BC ₄ -17#2X VR.2	1991-92
BC ₆ -1	BC ₅ -10#4X VR.2	1992-93
BC ₅ -10	BC ₅ -S07	1995-96
BC ₅ -11	BC ₅ -S08	1995-96
BC ₅ -12	BC ₅ -S09	1995-96
BC ₅ -13	BC ₅ -S11	1995-96
BC ₅ -14	BC ₅ -S12	1995-96
BC ₅ -S1	BC ₅ -S10	1996-97
BC ₅ -S2	BC ₅ -S11	1996-97
BC ₅ -S3	BC ₅ -S12	1996-97
BC ₅ -S4	BC ₅ -S13	1996-97
BC ₅ -S5	BC ₅ -S14	1996-97
BC ₅ -S1	BC ₅ -S1#1	1997-98
BC ₅ -S2	BC ₅ -S4#4	1997-98
BC ₅ -S3	BC ₅ -S5#5	1997-98

Fig. 1: Pedigree of the Yasini (BSR-1)

AINPT held during 27-28 September 2021, recommended for its release. This was further discussed in the Tobacco Variety Release Committee of Tamil Nadu on May 2023 and recommended to release in the name of Yasini to chewing tobacco areas of Tamil Nadu.

VARIETY DESCRIPTION

The varietal description of Yasini is given in the table 1 and fig 2. Yasini produces broad ovate leaves, dark green, sessile with moderately puckered, margins slightly wavy, tip acute, base cuneate, auricle prominent, leaf number vary from 24-25 with around 10 economic leaves, spangling medium, leaf length 70-75 cm and width 40-45 cm as compared to its parent VR-2 that has plants open, slightly drooping lamina, lanceolate, green leaves with medium auricle.(Prasad Rao, 1999).Further DNA amplification profile with 12 SSR markers revealed that PT40035 marker has specific amplification at ~190 bp in BSR1 (Fig. 2) whereas other chewing tobacco varieties VR-2, Abirami and Kaviri has shown differential banding pattern.

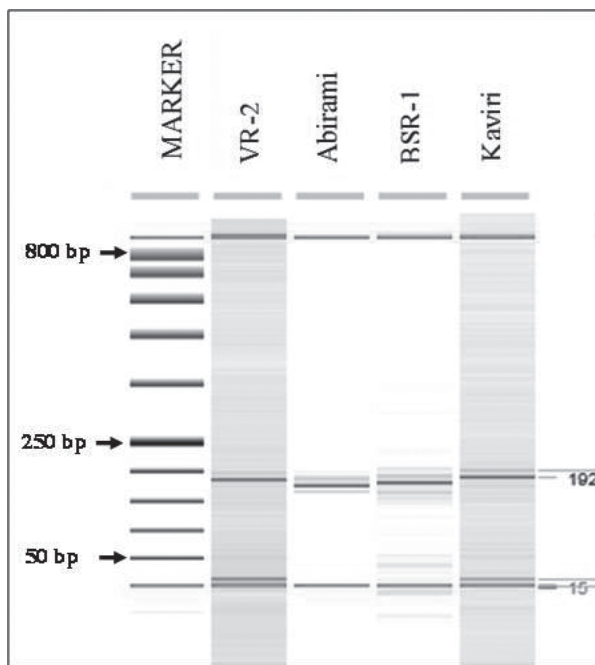


Fig.2: PCR amplification profile of BSR1 along with other Chewing tobacco varieties VR-2, Abirami and Kaviri using SSR marker PT40035



Fig. 3. Photo of BSR-1 (Yasini) in comparison with its parent VR 2 and Check variety, Kaviri.

Table 1: Description of the Chewing tobacco proposed variety Yasini (BSR-1)

Traits	Description
Name of the variety	: Yasini (BSR-1)
Species	: <i>Nicotiana tabacum</i> L
Type	: Sun-cured
Use	: Chewing
Plant habit	: Open
Height upto crow foot	: 1.4 to 1.5 m
Internodal length	: Medium (5.8cm)
Stem colour	: Green
Leaf character	:
a) Shape	: Broad ovate
b) Surface	: Moderately puckered
c) Margin	: Slightly wavy
d) Tip	: Acute
e) Base	: Cuneate
f) Auricle development	: Prominent
g) Colour	: Dark green
h) Angle of insertion	: 45°
i) Stalk	: Sessile
j) Midrib and venation	: Prominent
Size Length(cm)	: 70 to 75 cm
Width (cm)	: 40 to 45cm
Maturity	: Medium (115 to 120 days)
Total leaf numbers	: 24 to 25
Economic leaf number	: 8 to 10 leaves
Suckering habit	:
Without topping	: Low
On topping	: Heavy
Spangling	: Medium
No. of days taken to flowering	: 60-65 days
Inflorescence	:
Panicle	: Semi open and branched
Peduncle	: Thick and sparsely pubescent
Flower colour	: Pink
Corolla shape	: Funnel
Capsule shape	: Elliptic
Seed colour, size and yield per plant (gms)	: Brown, 10,000 to 10,500 per gram 15 to 20 gms
Rooting habit (Mention type of soil)	: Deep Sandy soil
Reaction to diseases	: Susceptible to TMV, Leaf curl and resistant to black shank.
Pests	: Susceptible to tobacco leaf eating caterpillar <i>Spodoptera litura</i> and aphid attack (Comparable to parent VR 2 and check Kaviri)
Growing season	: Rabi (1 st week of October – 28 th February)
Yield data kg/ha	: Green leaf Cured leaf Whole leaf 15,000 3391 75-80%
Quality characteristicsa)	:
a) Chemical	: Nicotine : 5.0% Chlorides : 4.2%
b) Physical	: Cured leaf heavy bodied, dark brown elastic with whitish incrustation, sweet aroma and highly pungent.

YIELD PERFORMANCE

Bulk and Pre-release yield Trials

In replicated three bulk, three pre-release and two agronomic trials were conducted during 2006 to 2009, BSR-1 found significantly superior mean leaf yield compared to two check varieties VR 2 and Kaviri (Table 2). In replicated bulk trials, Yasini recorded 3687 leaf yield which is 80 and 22.2% higher yield than the check varieties VR 2 and Kaviri, respectively. In location testing trials Yasini had 3391 kg leaf yield (average of three trials) which was 29.9% higher than VR 2 and 19.1% higher than Kaviri. In agronomic yield trial conducted with check Kaviri, it recorded 5.8% higher than Kaviri (Table 2). The increased yield advantage in Yasini as compared to check varieties may be due to the

desirable recombination of genes from its parents and backcross progeny, the major yield advanced might have realised because of many characters like dark green leaves, leaf length and width, total number of economic leaves, etc. besides the resistance to black shank resistance.

Agronomic Adaptability Trials

The total leaf yield during 2011-12, 2012-13 and pooled data didn't show significant difference between the varieties *viz.* BSR-1 and Kaviri. There were no significant differences among the varieties for total leaf yield during 2011-12 and 2012-13 (Table 3). The response of nitrogen was up to 150 kg N/ha with respect to total leaf yield. The pooled total yield recorded was 2577 kg/ha. The yield with 150 kg N/ha and 200 kg N/ha was comparable.

Table 2: Summary yield data of replicated bulk and pre-release testing trials of BSR-1 (Yasini).

S. Item No.	Year of testing	No. of trials	Mean leaf yield Kg/ha (Percent increase over Check)		
			BSR-1	Check I, VR-2	Check 2, Kaviri
a. Replicated yield trials	2006 - 2009	3	3687	2045 (80.0%)	3016 (22.2%)
b. Location testing trials	2006 - 2009	3	3391	2610 (29.9%)	2847 (19.1%)
c. Agronomic evaluation	2011-2013	2	2624	-	2479 (5.8%)

Frequency in the top group: Top in 8 trials.

Table 3: Adaptability to changes in agronomic conditions for Yasini

Item		BSR-1	Check Kaviri
Sowing date Experiments	Yield kg/ha under the recommended Sowing date	3687	3016
	Percentage gain or Loss when sown under	Early	Only one planting season
	Loss when sown under	Normal	commences in first week of
	Recommended dose	Late Fertiliser	of October Yield kg/ha
Experiments	under recommended dose		
	Percentage gain or Loss when under	F0 Nil	
	Other doses	F1 100 kg	2410
		F2 150 kg	2577 - 6.9% increase over F1
Irrigation Experiments Wherever applicable	Yield kg/ha with adequate irrigation	F3 200 kg Adequate	2600 - 0.8% increase over F2
	Percentage gain or loss	Irrigation level I	
		Irrigation level II	Not applicable
		Irrigation level III	

The existing recommended N dose of 150 kg N/ha recommended for the coastal areas for achieving the higher yield (AINPT, Annual report 2017-2018).

Testing against major disease and pests

The testing of Yasini for black shank and other major diseases, Tobacco mosaic virus (TMV) and tobacco leaf curl (TLC) and major pests (aphid and caterpillar) were done artificially and field conditions and results are presented in Tables 4a

and 4b. Under artificial condition for both TMV and leaf curl virus proved that it is mostly like control varieties (VR-2 and Kaviri). (Hoque Reza *et al.* 2019).

Evaluation for quality parameters

Chemical quality characteristics viz., nicotine, reducing sugars and chlorides are comparable to check varieties (Prasad Rao, 1999) (Table 5a and 5b).

Table 4a: Reaction of BSR 1 (Yasini) to major diseases at Vedasandur conditions

Diseases		Year	BSR-1	VR-2	Kaviri
Tobacco Mosaic Virus	Artificial	2006-09	Susceptible	Susceptible	Susceptible
	Natural	2006-2007	5.4	6.8	4.5
	Natural	2007-2008	3.6	8.2	5.2
	Natural	2008-2009	2.0	5.0	4.0
		Mean		3.7	6.7
Tobacco leaf curl	Artificial	2006-09	Susceptible	Susceptible	Susceptible
	Natural	2006-2007	2.0	4.6	3.0
		2007-2008	1.2	3.6	2.0
		2008-2009	-	2.0	1.8
		Mean		1.6	3.4
Black shank	Artificial	2006-2009	Resistant	Susceptible	Susceptible
	Natural	2006-2007	Nil	60.5	12.8
		2007-2008	Nil	56.0	13.0
		2008-2009	Nil-	45.3.	13.3
		Mean		Nil	50.6

Table 4b: Reaction of Yasini to insect pests at Vedaranyam condition

Insect Pests	Condition	Year	% of damaged/affected plants		
			BSR-1	VR-2	Kaviri
Tobacco leaf eating caterpillar	Artificial	2006-2009	Susceptible	Susceptible	Susceptible
	Natural	2006-2007	-	48.0	38.0
		2007-2008	1.2	64.8	45.0
		2008-2009	1.6	56.8	52.5
		Mean		1.4	56.5
Aphid	Artificial	2006-2009	Susceptible	Susceptible	
	Natural	2006-2007	4.0	6.0	2.0
		2007-2008	-	-	-
		2008-2009	1.6	3.2	4.0
		Mean		2.8	4.6

Table 5(a): Data on quality characteristics -Physical quality / chewability of BSR-1

Quality attributes	Year	Trader's opinion			Consumer's preference		
		BSR-1	VR-2	Kaviri	BSR-1	VR-2	Kaviri
Body (10)	2007-2008	8.0	7.0	8.0	8.5	7.5	8.2
	2008-2009	8.5	7.0	8.5	8.5	7.5	8.2
	Mean	8.3	7.0	8.3	8.5	7.5	8.1
Aroma (10)	2007-2008	8.2	8.0	7.5	8.2	8.0	8.0
	2008-2009	8.2	7.0	7.5	8.2	8.0	8.0
	Mean	8.2	7.5	7.5	8.2	8.0	8.0
Incrustation (10)	2007-2008	6.0	6.0	6.0	6.0	6.0	6.0
	2008-2009	6.0	7.0	7.0	6.0	6.0	6.0
	Mean	6.0	6.5	6.5	6.0	6.0	6.0
Taste (10)	2007-2008	8.4	8.0	7.0	8.4	7.4	8.2
	2008-2009	8.2	7.0	7.2	8.4	7.4	8.2
	Mean	8.3	7.5	7.1	8.4	7.4	8.2
Pungency (10)	2007-2008	6.4	6.0	7.0	6.4	6.5	6.5
	2008-2009	6.8	6.0	7.0	6.8	6.5	6.5
	Mean	6.6	6.0	7.0	6.6	6.5	6.5
Saliva secretion (10)	2007-2008	7.8	6.0	6.0	7.8	7.2	7.0
	2008-2009	7.8	6.0	6.0	7.8	7.2	7.0
	Mean	7.8	6.0	6.0	7.8	7.2	7.0
Duration of) pungency (10)	2007-2008	6.0	7.0	7.5	6.0	7.0	7.5
	2008-2009	6.4	6.0	6.0	6.4	7.0	7.5
	Mean	6.2	6.5	6.8	6.2	7.0	7.5
Stiffness in the mouth(10)	2007-2008	6.2	6.0	7.0	6.2	6.2	6.2
	2008-2009	6.4	6.0	7.2	6.4	6.2	6.2
	Mean	6.3	6.0	7.1	6.3	6.1	6.2
Mean score out of 80		57.7	53.0	56.3	58.0	55.7	57.5

Table 5(b): Data on quality characteristics (Chemical quality characters-2008-09)

S.No.	Quality characteristics	BSR-1	VR-2	Kaviri
1.	% Nicotine	5.0	5.2	5.1
2	% Chlorides	4.2	4.3	3.8

Table 6(a): Replicated evaluation (2006-2007 to 2008-2009)at CTRI RS, Vedasandur

Selection / variety	Total leaf (kg/ha) pooled
BSR-1	3687*
VR-2 (Check-1)	2045
Kaviri (Check-2)	3016
CD at 5%	311
CV%	12.7

In the replicated evaluation BSR-1 recorded 80 % increased yield over the VR-2 and 22 % increased yield over Kaviri (Table 6a). In the bulk plot testing at Ayakaranpulam, BSR-1 recorded a mean yield of 29.9 and 19.1 % with VR-2 and Kaviri, respectively (Table6b).



Fig. 4: Field view of Yasini at Periakuthagai, Ayakaranpulam-I, Vedaranyam taluk.

The total leaf yield during 2011-12, 2012-13 and pooled data didn't showed significant difference between the varieties *viz.* BSR-1 and Kaviri. How ever there was a yield increase with the variety BSR-1 over Kaviri (Table 7). The response of nitrogen was up to 150 kg N/ha with respect to total leaf yield. The pooled total yield recorded was 2577 kg/ha. The yield with 150 kg N/ha and 200 kg N/ha was comparable. The existing recommended N dose of 150 kg N/ha recommended for the coastal areas is sufficient to get higher yield.

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Table 6(b): Bulk plot testing from 2006-2007 to 2008-2009 at Ayakaranpulam(Mean)

S.No	Variety	Cured leaf yield (kg/ha)	% over Check1	% over Check-2
1	BSR-1	3391	29.9	19.1
2	VR-2 (Check-1)	2610	-	-
3	Kaviri (Check-2)	2847	-	-

Table 7: Adaptability to changes in agronomic conditions

Treatments	Total leaf yield (kg/ha)		
	2011-12	2012-13	Pooled
Varieties :			
BSR-1	2676	2624	2650
Kaviri	2505	2492	2479
S.Em	25.1	17.9	12.0
C.Dat 5%	NS	NS	NS
N Levels			
100 kg/ha	2395	2402	2410
150 kg/ha	2637	2607	2577
200 kg/ha	2741	2650	2600
S.Em	42.2	32.0	37.0
C.Dat 5%	127.6	103.6	113.2

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