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NEW GENOTYPES OF COTTON (*GOSSYPIUM HIRSUTUM*) BRED AT BRGV BUZĂU

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Abstract

Plant Genetic Resources Bank Buzău owns a rich collection of cotton (Gossypium hirsutum) varieties, predominantly ornamental cultivars, which are part of the institution's activity. Among these genotypes, two varieties with distinct phenotypic expressivity have been obtained as a result of intensive breeding work: L1, a cultivar with multiple uses, appreciated for its ornamental characteristics, and L2, which stood out for its earliness and high fiber production capacity. Biometric determinations revealed specific characteristics: L1 with an average height of 57 cm and mass/ boll 6.61 g and L2 with an average height of 109 cm and mass/ boll of 8.53 g. L1 was obtained from the crossing of the Romanian variety known as Brâncoveni and a local population from India and the L2 cultivar was based on the Adelin variety, one of the first cotton varieties in Romania. The present paper highlights the research carried out on the stabilization of two phenotypically distinct genotypes, to be approved and patented under the aegis of BRGV Buzau.

Key words*: biobanking, germplasm, phenotype, fiber, ornamental.*

INTRODUCTION

Cotton (*Gossypium* L.) belongs to the *Malvaceae* family and is one of the world's most important textile crops. It is grown in over 100 countries and meets about one third of global demand for textile fibers (Senchina et al., 2003). Management of cotton requires an understanding of the growth habit and responses of the plant to the environment (Whitaker et al., 2018). Throughout history, cotton has played a significant role in economic and social development, being used both in clothing and in other industries such as armaments and pulp (Townsend, 2020). In Romania, cotton cultivation began in 1868, initially on small areas of land in Moldova and Muntenia and later spread to the south of the country. Nowadays, cotton cultivation is facing challenges related to unfavorable climatic conditions, such as drought and high temperatures, which affect the quality and quantity of the fiber obtained (Majeed et al., 2021). Cotton is a perennial, semi-herbaceous species in its indigenous regions; however, in our country, it is grown as an annual crop. It develops a taproot that can extend up to 2 meters in depth, and the plant typically reaches a height ranging from 30 to 200 cm (Senchina et al., 2003).

The history of cotton cultivation is at least 3000 years old (Stewart et al. 2010). Cotton fiber quality is influenced by a complex interaction between genetics and environmental conditions (Bradow & Davidonis, 2000). Cotton is a crop that requires large auxiliary inputs and leaves a very low content of biomass residues on the soil after harvesting, compared to corn, wheat, and soybean (Trapalis et al., 2023).

Since the expression of different traits is often altered as breeding material due to environmental changes, cotton research must develop new, more productive cotton genotypes adapted to the current climate.

This paper presents research carried out at the BRGV Buzău on two cotton (*Gossypium hirsutum*) varieties: late-ripening L1 and early-ripening L2. The aim of the research was to evaluate the suitability of the two genotypes for introduction into cultivation in the S-E area under current climatic conditions.

MATERIALS AND METHODS

The research was carried out at BRGV Buzău, located in the SE part of Romania. The soil in this area (45°8′50″N 26°48′3″E/45.14722°N 26.80083°E), consist of loessosesoidal deposits, clays, marls and argillaceous marls (Soil map. Retrieved from http://geodim.meteoromania.ro/sia).

Groundwater is located at shallow depths ranging from 0.8 to 1 meter, although water in the surface crust layer tends to evaporate quickly, particularly during the heat of summer. The technology applied to the planted plots was that specific to cotton. In the fall, the soil was prepared through tillage to a depth of 30 cm. In spring, seedbed preparation was applied before sowing, which was carried out at the end of April, when the soil temperature reached 14°C. The sowing depth was 4-5 cm. The applied sowing pattern was 70 cm between rows and 40 cm plant spacing per row. Irrigation was carried out during the growing season as cotton is a species with low resistance to water stress.

The year 2024 was characterized by a pronounced drought during the peak period of plant growth and development, more precisely during the period of boll formation and maturation, with the phenomenon of heat stress. Phenological observations and biometric measurements were made according to UPOV (International Union for the Protection of New Varieties of Plants) and IPGRI (International Plant Genetic Resources Institute) requirements. The determinations were carried out in the experimental plots, on growing plants, and aimed at: measuring plant height, number of sympodial branches and number of capsules per plant. Capsule harvesting was staggered, from the base of the plants towards the top, as the ripening of the capsules is uneven and subject to genetic and climatic factors. Quality indices such as capsule weight, fiber/capsule mass and seed/capsule mass were determined, as well as biometric measurements of the seeds/capsule were made. Weighing was carried out in the Seed Quality Laboratory of BRGV Buzău using Kern five-decimal precision balance.

In order to valorize the production potential of the two lines, laboratory determinations of the physical quality assessment indices were carried out both at the fiber and seed level. In order to establish the vitality of the seeds as well as the vigor of the plants resulting from the harvested seeds, viability and germination determinations were made in Binder germination cabinets and the working parameters were monitored daily.

The methodology used was in accordance with ISTA (International Seed Testing Association) (2004) application standards. Viability testing was done to evaluate embryo structures and embryos in terms of their health, integrity and development, analyzing possible diseased, dry or immaturely developed seeds. For the viability test, from a 500 g seed homogenized sample, 200 seeds were extracted and divided into four replicates of 50 seeds each. These required prior preparation by removing, as far as possible, the felt brush, moistening in water for 18 hours, scarification by severing the integument at the distal end of the cotyledons, and longitudinally severing the seeds ¾ through the central axis. After preparation, the seeds were placed in a 1% concentration of 2,3,5-triphenyl tetrazolium (TTZ) solution in order to highlight viable tissues by staining for 3 hours. Evaluation was performed according to international standards (International Seed Testing Association, 2024).

Germination testing was carried out according to ISTA 2024 standards to determine the percentage of seeds capable of producing normal germination.

For the germination test, 200 seeds were placed equidistantly with sufficient space for growth, on two layers of blotting paper. The filter paper, with 80g/ml mass and 16.7 x 50.8 cm dimensions, was sterilized and moistened to 60% of the holding capacity for water. The pH of the water was neutral (7.28), as required for seed germination. The germination temperature was alternating 8 h, at 20°C and 16 h, at 30°C in the presence of light. The relative humidity inside the cabinet was 85%. Essential structures were subjected to evaluation when they were sufficiently well developed.

The genetic resources analyzed were two cotton varieties of the *Gossypium hirsutum* species. Variety L1 is characterized by late maturity, has ornamental importance and is ideal for commercialization of dried open caps stems andfor flower arrangements. It is also characterized by dark brown flowering stem, decorative, intensely coloured flowers (Figure 1 a) and very well encapsulated fibers, which do not deteriorate over a long period of time.

Variety L2 belongs to the early maturity group and is characterized by high fiber production, slightly dehiscent capsules that open wide, thus facilitating fiber harvesting (Figure 1 b).

a) b)

Figure 1. Cotton batches in forming phenophase:
a) L1 genotype; b) L2 genotype

In the full maturity phenophase, three successive measurements were made at 5-day intervals, and the mean value and standard deviation were calculated. Analysis of variance (Anova test) was performed to determine significant differences between the two genotypes.

RESULTS AND DISCUSSIONS

The vegetation time of the plants was 143 days and the developmental phenophase from the beginning of the flowering to the last boll harvest was of 85 days (Figure 2).
Both genotypes were characterized by an erect main stem, circular in cross-section. Plants formed at the base of the stem, 2-3 vegetative branches of flowers which were removed.

a) b)

Figure 2 a) Cotton flower; b) evolution of cotton flower after fertilization until boll maturation

The fruiting (sympodial) branches had a flower at each internode. Under 2024 weather conditions, cotton flowering lasted until the end of October (Figure 3).

Figure 3. Cotton bolls at different stages of ripening

The statistical analysis performed revealed significant differences between the two lines in plant height and number of seeds/capsule ($p \leq 0.001$) (Table 1), which can be attributed to genetic variation in their chromosomal map.

Table1. Biometric determinations on the two cotton lines L1 and L2

Geno type	pH (cm) Mean ± SD	NSB Mean ± SD	TNB Mean ± SD	NC/P Mean ± SD	MC (g) Mean ± SD	FL (cm) Mean ± SD	NS/C Mean ± SD
L1	62.8± 5.63	16.9± 2	29.6± 3.84	21.14 ±3.07	7.11± 0.93	3±0.3 8	18.4± 2.88
L2	109.2 ±2.77	21.8± 2.38	31.2± 1.92	25.8± 2.38	8.56± 0.20	3.56± 0.20	26.8± 2.38
p ≤ 0.001	<0.00 1	0.008	0.43	0.028	0.01	0.02	0.001

Legend: PH - plant height; NSB - Number of sympodial branches; TNB - Total number of branches; NC/P - Number of capsules/plant; MC - Mass of capsules; FL - Fiber length; NS/C - Number of seeds/ capsules.

The average length of the fibers extracted from the analyzed bolls showed that L2 was 3.5 cm compared to L1, which had a value of 2.7 cm. In terms of the number of seeds in the boll, L2 had an average of 26 seeds/boll compared to L1 of only 18 seeds/boll (Figure 4).

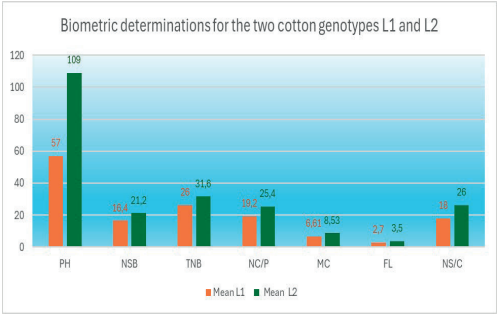


Figure 4. Graphical representation of the results of the biometric measurements on the two cotton lines L1 and L2

The analysis of the qualitative characteristics carried out aimed, in the first place, to identify the genotype with superior quality fiber, cotton being a technical plant intended for fiber production. The measurements performed on 100 bolls revealed that L2 has a 29.30% fiber/boll mass ratio compared to L1 with a ratio of 25% fiber/boll mass. L2 also showed a superior fiber quality in terms of organoleptic characteristics (Table 2).

Table 2. Qualitative determinations of cotton fibers obtained from batches L1 and L2

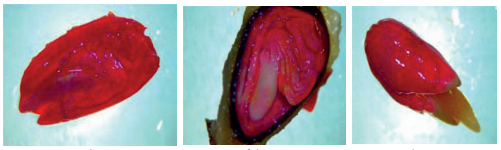
Determinations	L1 (late variant)	L2 (early variant)
Mass fiber/capsule %	25%	29.30 %
Thread smoothness	matte	silky
Colour	white yellowish	white, silky
Linter	short, gray	silky white, with adherence to the integument

By analyzing the two lines, the L2 variety was noted with an average seed weight of 5.1 g compared to 4.9 g recorded for L1. Qualitative characteristics such as seed shape and colour are identical in the two genotypes and the ratio of seed coat to seed mass was highest in L2 (22%) compared to L1 (21%). The maximum level of seed length was recorded in L2 cultivar, i.e 9.1 mm, the same for width, i.e 4.8 mm (Table 3).

Tabel 3. Biometric seed measurements of L1 and L2 cotton varieties

Determinations	L1 Mean/100 seeds	L2 Mean/100 seeds
Average mass/seed (g)	4.9	5.1
Shape	elongated oval	elongated oval
Colour	dark brown	dark brown
Mass ratio husk/seed	21%	22%
Seed coat thickness (mm)	0.25 mm	0.22 mm
Embryonic mass	80%	80%
Average length (mm)	8.5 mm	9.1 mm
Average width (mm)	4.2 mm	4.8 mm

Viability testing was carried out conforming to the assessment criteria (ISTA, 2024) according to which fully coloured seeds or seeds with small uncoloured areas of essential structures are considered viable and seeds showing uncoloured essential embryo structures or uncoloured essential areas of essential structures or uncoloured embryo as a whole are considered non-viable, according to the international assessment schemes (Tetrazolium Testing - International Seed Testing Association, 2003). After 3 hours of coloring, each seed was evaluated individually, L-2 showing a better uniformity between replications, the difference between minimum and maximum being only 4% (R1 and R3) (Figure 5).



a) b) c)

Figure 5. Viability assessment structures evaluated: a) viable embryos in section; b) viable embryos in tegument; c) non-viable embryo

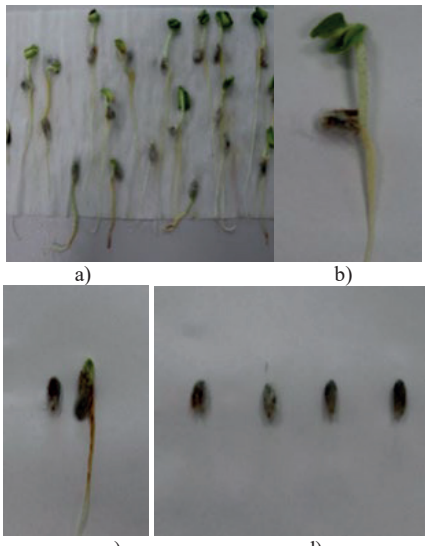
The uniformity of the coloration of the evaluated structure parts of each seed was also evident. Viability recorded a maximum value of 95% in L2 and 90% in L1. The non-viable seeds showed embryos totally uncoloured or with essential parts of the basic structures uncoloured. L1 revealed 10% non-viable seeds, while L2, 5% non-viable seeds (Table 4).

Table 4. Viability test evaluation in L1 and L2 cotton varieties

Replications	L1 (late variant)		L2 (early variant)	
	Viable seeds	Non-viable seeds	Viable seeds	Non-viable seeds
R1	46	4	48	2
R2	45	4	48	2
R3	48	4	46	4
R4	42	8	47	3
Total	180	20	189	11
%	90	10	95	5
Means	45.25	5	47.25	2.75
Variant calculation	6.25	4	0.9167	0.167

Following L1 evaluations, normal sprouts showed a development characteristic of the species for this time of evaluation (germination energy) with high vigour and good growth uniformity (Figure 6 a), a strong taproot, well

developed hypocotyl, characteristic cotyledons with a true leaf-like, crumpled appearance (Figure 6 b). The abnormal germs showed primary infection on all structures that did not allow their further development in normal plants (Figure 6 c) which led to the appearance of dead seeds (Figure 6 d).



a) b) c) d)

Figure 6. L1 germination evaluation:
a) germination layer detail; b) normal germ;
c) abnormal germ; d) dead seeds

It was found that L2 (the early variety) showed the maximum germination value of 94% and L1 (the late variety), 89% germination value. It was found to be a 3% abnormal germplasm for both genotypes, cotton showing a higher susceptibility to attack by seed-borne diseases. In case of dead seed evaluation, the maximum level was identified in L1, i.e 8% and 3%, in L2 (Table 5).

Table 5. Germination evaluation of the 2 cotton varieties L-1 and L-2

Replications	L1 (late variant)				L2 (early variant)			
	GN	GA	DS	FS	GN	GA	DS	FS
R1	45	2	3	0	47	1	2	0
R2	42	3	5	0	45	2	3	0
R3	46	1	3	0	48	1	1	0
R4	44	1	5	0	48	1	1	0
total	177	7	16	0	188	5	7	0
%	89	3	8	0	94	3	3	0
Mean	44.25	1.75	4.00	0	47.5	1.25	1.75	0

Legend: GN-normal germ; GA-abnormal germ; DS- dead seed; FS-fresh seed.

The analysis of the compatibility between viability and germination highlighted that L2 recorded an average value of 95%, while L1 recorded 90%. The graphical interpretation confirms the superior quality of the L2 genotype (Figure 7).

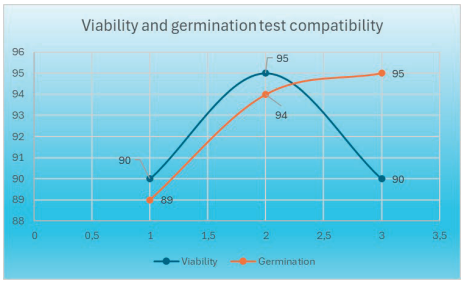


Figure 7. The graphical representation of the compatibility of the results from the two tests (viability and germination) for the two varieties L1 (late) - L2 (early)

CONCLUSIONS

The results obtained confirmed the significant phenotypic and technical differences between the two cultivars.

The breeding research conducted highlighted genotype L2 as having superior quantitative and qualitative characteristics compared to genotype L1. At the same time, the studies undertaken at BRGV Buzău confirmed the acclimatization potential of both genotypes, which can be successfully cultivated under the climatic conditions of the Buzău area.

L1 presents a great ornamental potential and can be cultivated for this purpose. The acclimatized and genetically stabilized genotypes will be submitted for homologation and patenting under the auspices of BRGV Buzău.

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