

Genetic progress in cotton dry matter partitioning in Argentina

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ABSTRACT

The Argentinian cotton (*Gossypium hirsutum* L.) has exhibited a rate of genetic progress of 3.24 kg ha⁻¹ y⁻¹, resulting in an increase of 194.4 kg ha⁻¹ of lint from 1965 to the present day. This improvement has been achieved alongside enhanced seed oil content without compromising lint quality. While previous studies have explored trends in cotton lint yield, lint quality, and other agronomic traits over time, no research in Argentina have focused specifically on genetic progress concerning aboveground biomass, morphological attributes, and mapping-related variables. This study aimed to: i) quantify and compare the levels of aboveground biomass, morphological and mapping-related variables in varieties utilized over the past 50 years in Argentina, and ii) estimate the genetic progress in these variables. Over a two-year period, 20 cotton cultivars were assessed across four different environments through different sowing dates. Several parameters were evaluated, including 11 biomass-related variables, six morphological-related factors, and six mapping-related traits at three different moment of cotton cycle: 50 (M1), 100 (M2), and 150 (M3) days after emergence. Significant differences were observed across genotypes, environments, and their interactions. Our findings revealed that variables such as leaf biomass (M2), reproductive branch biomass (M2, M3), first reproductive branch node (M1, M2), first reproductive branch height (M1, M2, M3), and global retention (M2) exhibited either increases or decreases on the rate of genetic progress. This study presents the first report on aboveground biomass values, morphological variables, and mapping-related parameters for Argentinian cotton. The information obtained from the work provides valuable insights into the dynamics of cotton biomass and related variables, contributing to the understanding of genetic improvement in Argentinian cotton cultivation.

1. Introduction

Cotton (*Gossypium hirsutum* L.), derived from the Arabic terms "quṭn", "kutum", or "gutum", refers to the plant that produces spinnable fibers in its seeds (Lee, 1984). As the world's most important natural textile fiber, cotton contributes significantly to the global textile industry, which boasts an annual economic impact exceeding \$600 billion (Khan et al., 2020). Known as "white gold" in certain regions due to its immense economic value (Iqbal et al., 2013), is grown on approximately

32.2 million hectares of land worldwide with fluctuations between 28 and 36 million hectares in recent years. In Argentina, the planted area reached a historical record of 1.13 million hectares in 1998; however, it has since stabilized to approximately 0.5 million hectares (Scarpin et al., 2022).

Genetic improvement, as described by Wakelyn and Chaudhry (2010), is a process of human-directed plant evolution that leads to significantly higher crop yields. Cotton breeding began with initial selection efforts in the United States around 1850, with the introduction of

Abbreviations: LY, lint yield; G, genotype; E, environmental; G*E, genotype-by-environmental interaction; S, season; SD, sowing date; DAE, days after emergence; YOR, year of release; St, stem; Rb, reproductive branches; Sq, squares; Le, leaves; Vb, vegetative branches; Gb, green bolls; Bu, bur; FS, fiber and seed; TDW, total dry weight; VDW, vegetative dry weight; RDW, reproductive dry weight; R/V, reproductive/vegetative ratio; RA, reproductive allocation; HI, harvest index; GR, global retention; GR1P, global retention in 1st position; GR2P, global retention in 2nd position; He, height; HR, height per nodes ratio; NN, number of nodes; N1RP, node number of the 1st reproductive branch; H1RP, height of 1st reproductive branch; NVB, number of vegetative branches.

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seeds from regions such as the West Indies, Mexico, Brazil, and Peru, where cotton had already been cultivated (Giband et al., 2010). In Argentina, cotton breeding began in 1923, initially focusing on evaluating a wide range of introduced varieties, primarily from the U.S., to identify those best suited to local environmental conditions. This breeding program has driven significant genetic advancements in Argentinian cotton production, with yields increasing annually by $3.24 \text{ kg ha}^{-1} \text{ y}^{-1}$ since 1965 (Scarpin et al., 2022). This increase has been accompanied by improvements in seed cotton yield, lint percentage, boll density per unit area, and lint per seed. However, no significant increases were recorded in other yield components or in any of the fiber and seed quality parameters analyzed, except for an increase in oil content (Scarpin et al., 2023).

Cotton is a perennial crop that displays a shrub-like, indeterminate growth habit before human domestication (Hutchinson et al., 1947). Through selective breeding and genetic modification, cotton has evolved into a crop with a more determinate growth habit, which prioritizes reproductive development over vegetative production compared to its ancestral species (Wells, 2016). The allocation of dry matter between vegetative and reproductive structures can be evaluated using parameters such as the reproductive/vegetative dry matter ratio (R/V), reproductive allocation (RA), and harvest index (HI). Modern cotton varieties, for example, exhibit higher R/V ratios, which have been linked to yield improvements in the U.S (Wells and Meredith Jr., 1984b). The concept of RA, introduced by Bazzaz et al. (1987) and later adapted for cotton by Sadras et al. (1997), is defined as the ratio of reproductive dry matter (i.e., fiber and seed) to total dry matter produced at physiological maturity. Meanwhile, HI, a widely used crop index, represents the proportion of fiber to total dry matter at physiological maturity and can be influenced by the fiber percentage in different varieties (Shorter et al., 1991). Unlike RA, HI is affected by the percentage of fiber obtained in the different varieties.

Cotton plant mapping is a method of quantifying the number of reproductive structures retained and comparing these indices. This is a useful technique for the morphological characterization and evaluation of the flowering patterns of plants. Local mapping of fruiting and/or vegetative structures can aid in diagnosis and making management decisions (Raphael et al., 2019). This method is an effective technique for the different fruiting patterns of different cultivars as well as their susceptibility to pests and management strategies (Constable, 1991). The main output is the percentage of global retention (GR) of reproductive structures analyzing their distribution throughout the plant, as these parameters have a significant impact on final yield (Paytas, 2010).

Few studies have examined variables such as: aboveground biomass, morphological and mapping-related, considering different years of release of cultivars. Wells and Meredith Jr. (1984c) attributed the US yield increase from 1905 to 1978 to a shift in the proportion of R/V dry matter in modern cultivars. Additionally, according to Meredith Jr. and Wells (1989), no differences were found in the total dry matter between cultivars, although modern varieties had a greater amount of reproductive dry matter than obsolete varieties. In contrast, Yang et al. (2019) reported differences in the R/V ratio at two moments for modern varieties compared with obsolete ones in China. Similarly, Conaty and Constable (2020) discovered significant genetic advancements in the total dry matter and HI in a study on various Australian cultivars, highlighting that fiber percentage played a fundamental role in HI improvement. Regarding the variables associated with ecophysiological mappings, multiple authors have used these indices (Guinn et al., 1981; Constable, 1991; Bange et al., 2008), nonetheless Kilby et al. (2012) reported genetic progress related to global retention on cotton cultivars.

Based on scientific evidence collected, our hypothesis was that modern cultivars possess a higher ability to retain reproductive organs, resulting in a greater relationship between the weight of reproductive and vegetative structures. Therefore, the aims of this study were to: i) quantify and compare the levels of aboveground biomass, morphological variables and mapping-related parameters in varieties utilized over

the past 50 years in Argentina, and ii) estimate the genetic progress in these variables.

2. Materials and methods

2.1. Plant material and experimental design

Field experiments were carried out during the summer growing seasons of 2017/18 (S1) and 2018/19 (S2) at INTA Reconquista, located in Santa Fe Province ($29^{\circ}15'54''\text{S}$; $59^{\circ}44'33''\text{W}$, 50 m alt), Argentina. The soil type was silt loam Aquertic Argiudoll (USDA, soil taxonomy) belonging to the Reconquista series (Giorgi et al., 2011), with the following characteristics: soil organic matter content of 1.86 %; available P of 23.6 mg kg^{-1} ; inorganic of 68.9 mg kg^{-1} ; and available K 234.6 mg kg^{-1} . The climate is transitional subtropical-subhumid climate, with approximately 70 % of annual rainfall occurring between October to March (Zuil, 2011).

To create four distinct environmental conditions (E) two sowing dates - an early (SD I) and a late (SD II)- were implemented across two seasons (S1 and S2). In each E, 20 varieties of upland cotton grown in Argentina since 1965 were evaluated (Table 1). The cultivars used in this study were selected not only on their release date but also on their relevance to INTA cotton breeding program and its popularity among farmers (Table 1) (Delssin, 2003). Also, two cultivars still under development, whose release date is scheduled for 2024 and 2025 by the cotton breeding program of INTA, were included in the experiment. Seeds were provided by the INTA cotton germplasm bank, which is located at Saenz Peña, Chaco.

The experimental design for each E was a randomized complete block with four replicates. Each plot consisted of four 8-m long rows with 0.52 m row spacing. Seeds were manually sown at a rate of approximately 192,000 plants ha^{-1} , which is the usual population used by farmers in Argentina. In each E, two separate applications of fertilizer were carried out: at sowing time and at the early vegetative stage with 100 kg ha^{-1} of diammonium phosphate (N-P-K:18-46-0) and 100 kg ha^{-1} of urea (N-P-K: 46-0-0), respectively. Crop management included supplementary irrigation and pest control as needed. Meteorological data from sowing to maturity were recorded using the INTA Reconquista weather station, located 250 m from the experimental plots. The environmental conditions presented during the different S and

Table 1

List of cultivars evaluated in the experiment indicating year of release (YOR). Note: This table was previously published in a work from the same author, nonetheless it is presented again, since it contains basic information to understand the work (Scarpin et al., 2022). * Represents INTA's pre-commercial cotton and + Represents varieties currently available for sowing in Argentina.

N°	Cultivar	Year of release
1	Stoneville 508	1965 ¹
2	Deltapine 16	1967 ¹
3	Toba II SP	1969 ³
4	Chaco 510	1980 ³
5	Porá	1982 ³
6	Deltapine 50	1984 ²
7	Guazuncho 2	1989 ³
8	Chaco 520	1994 ³
9	Guazuncho 2000 RR ⁺	2001 ⁴
10	Guazuncho 3	2004 ⁴
11	Oro Blanco 2	2004 ⁴
12	Poraité	2008 ⁴
13	NuOpal ⁺	2009 ⁴
14	Deltapine 402	2010 ⁴
15	Deltapine 1238 ⁺	2014 ⁴
16	Guazuncho 4 ⁺	2019 ⁴
17	Guarani ⁺	2019 ⁴
18	Porá 3 ⁺	2019 ⁴
19	SP 41255*	2024*
20	SP 6565*	2025*

SD were presented in Scarpin et al. (2022) and Scarpin et al. (2023).

2.2. Biomass measurements

Above-ground biomass samples were obtained at three different moments (M) after cutting 0.25 m² of plants per plot at 50 (M1) and 100 (M2) days after emergence (DAE) and 0.5 m² of plants per plot at 150 (M3) DAE. Moments were selected since they matched with the phenological stages of the first visible open white flower (M1), first open boll (M2), and physiological maturity (M3), respectively. All plants from each plot were partitioned into vegetative (VDW) and reproductive biomass (RDW). VDW included biomass from stems (St), leaves (Le), vegetative branches (Vb), and reproductive branches (Rb). RDW included biomass from squares (Sq), green bolls (Gb), fibers and seeds (FS), and burs (Bu). All biomass components were weighted following the drying of plants in an air-forced oven at 65 °C for a minimum of one week, with daily weighing starting on the seventh day to ensure the dry weight was accurately determined. Calculations were performed to express the dry weights per m². The reproductive/vegetative dry matter ratio (R/V) was calculated by dividing RDW/VDW for each plot at each moment. The reproductive allocation (RA) and the harvest index (HI) were calculated considering data from M3 and lint yield (LY) values from Scarpin et al. (2022) by dividing FS/TDW (total dry matter) and lint (without seed)/TDW, respectively.

2.3. Morphological measurements

Morphological variables were recorded prior to biomass partition at the different moments: M1, M2, and M3. These variables were measured at each moment as follows:

- Height (H): measured from the base of the stem to the top of the apex.
- Number of nodes (NN): the number of nodes present on the stem of each plant sampled.
- Average distance between nodes (HR): obtained by dividing the recorded H by NN.
- Number of vegetative branches (NVB): mean of number of vegetative branches per plant present at each M.
- Node of insertion of the 1st reproductive branch (N1RB): node number where the 1st reproductive branch was recorded.
- Height of 1st reproductive branch (H1RB): height from the soil to the 1st reproductive branch was recorded.

2.4. Plant mapping measurements

Plant mapping was performed simultaneously with the morphological measurements on all the plants collected from the field at each moment. We recorded the presence of square (S), flower (F), green boll (B), open boll (C), or missing positions (A) in each reproductive position, as described in Fig. 1 (Guthrie and Kerby, 1993). Using these data, the flowering dynamics of the different varieties were determined by calculating the number of structures and global retention (GR) at each moment as explained in Fig. 1.

2.5. Statistical analysis

In this study, genotype (G), sowing date (SD), sampling moment (M), and all interactions between these factors were considered fixed effects; season or year (S), block within each season (B), plot number within season and block were considered random effects. A repeated-measures ANOVA analysis with a symmetrical error correlation model and heteroscedasticity among M were performed on all measured variables using the restricted maximum likelihood method (REML) with InfoStat software (Di Rienzo et al., 2011). Each model selection criteria was based on the Akaike Information Criterion (AIC). Similarly, to calculate genetic progress, an ANOVA was conducted on each of the variables

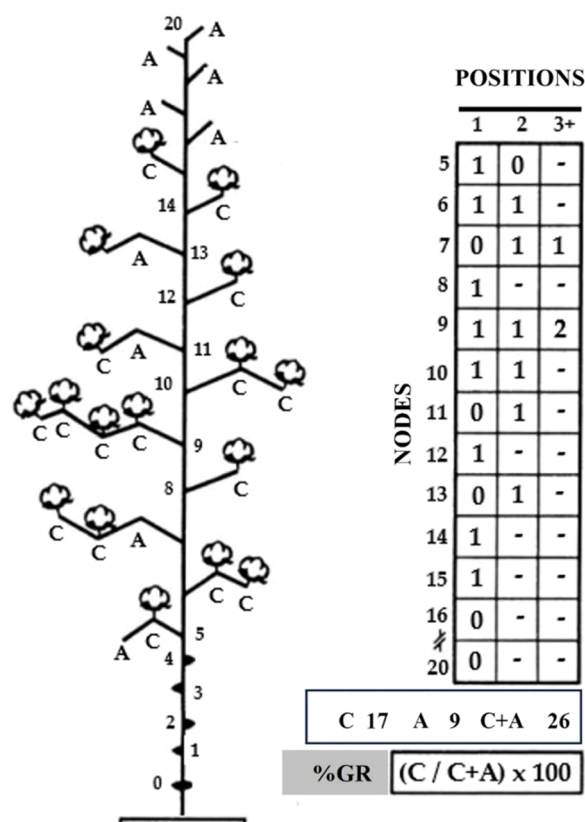


Fig. 1. Illustrative scheme for mapping cotton plants. Each reproductive structure was recorded based on its position within each node. References: C: open boll; A: missing position; %GR: percentage of global retention as the ratio of the total number of reproductive structures retained by the total number of reproductive positions generated per plant. This scheme was adapted from Guthrie and Kerby (1993).

partitioned by M. In this case, the fixed effects were the year of release (YOR), SD, and their interaction (YOR*SD), while the random effects were S and B within S. Following this, a simple linear regression analysis was conducted to assess the associations between the variables analyzed and YOR. When a significant interaction between YOR and SD was detected in ANOVA, the results were analyzed based on individual SD.

3. Results

3.1. Climatic data

The environmental conditions presented during the different S and SD were previously described in Scarpin et al., (2022) and Scarpin et al., (2023). In summary, S1 had the lowest historical rainfall value between October and February (300.1 mm), whereas S2 had the highest historical rainfall value in January (473.1 mm). These differences in rainfall induced changes in temperature and daily global solar radiation registered for each environment.

3.2. Analysis of variance and genetic progress for above-ground biomass

ANOVA results exhibited significant differences among genotypes and a strong interaction between SD and M for most biomass variables, except for vegetative branches (Vb), leaves (Le), and reproductive dry weight (RDW) (Table 2). Early sowing dates (SDI) showed higher biomass values than late sowing dates (SDII) for all the variables and moments measured, except for RDW and fiber and seeds (FS) at M3 and squares (Sq) at M2. SDI resulted in 58 %, 66 %, and 32 % higher total dry weight (TDW) at M1, M2, and M3, respectively, compared to SDII. In

Table 2

Statistical analysis (ANOVA), mean values and analysis of genetic progress for the sowing dates (SD), moments (M), genotypes (G), and SD*M interaction evaluated in the experiment. TDW: total dry weight, VDW: vegetative dry weight, St: stem, Vb: vegetative branches, Le: leaves, RDW: reproductive dry weight, Rb: reproductive branches, Sq: squares, Gb: green bolls, FS: fiber and seed, Bu: bur. M1, M2, and M3 measurement moments were at 50, 100, and 150 days after emergence. * $p < 0.05$, ** $p < 0.01$, and *** $p < 0.001$. LSD: least significant difference for M*SD interaction.

Sowing Date	Moment	TDW	VDW	St	Vb	Le	Rb	RDW	Sq (M1 & M2)	Gb	FS (M3)	Bu (M3)
SDI	-	1008.5	423.7	214.4	23.1	112.7	73.5	584.8	11.2	154.1	310.7	108.8
SDII	-	825.2	225.8	95.1	13.5	77.3	39.9	599.4	11.6	110.3	353.2	124.3
-	M1	334.0	301.2	95.7	15.7	135.4	54.4	32.8	22.0	10.8	-	-
-	M2	763.3	377.0	171.7	18.3	120.7	66.3	386.3	0.8	385.5	-	-
-	M3	743.2	296.0	197.0	21.0	29.0	49.0	447.2	-	0.2	331.6	116.0
SDI	M1	399.4	358.9	124.7	18.8	150.9	64.5	40.5	22.4	18.1	-	-
	M2	920.7	491.8	242.3	23.7	139.6	86.2	428.9	0.1	428.8	-	-
	M3	849.8	411.4	274.9	25.3	44.8	66.4	438.4	-	4.5	309.1	124.8
SDII	M1	252.4	230.1	65.0	10.0	116.2	38.9	22.3	20.8	1.5	-	-
	M2	555.3	243.6	98.7	9.9	95.0	40.0	311.7	1.7	310.0	-	-
	M3	641.6	178.6	120.4	15.6	13.6	29.0	463.0	-	0.0	355.0	108.0
<i>Source of variation</i>												
Moment (M)		***	***	***	***	***	***	***	***	***	-	-
Genotype (G)		ns	***	***	***	***	***	ns	***	ns	*	***
Sowing Date (SD)		***	***	***	***	***	***	***	***	***	***	***
M * SD		***	***	***	ns	ns	***	ns	**	***	-	-
G * SD		ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
M * G		ns	***	***	ns	ns	ns	ns	***	*	-	-
M * G * SD		ns	ns	*	ns	ns	ns	ns	ns	ns	-	-
L.S.D (M*SD)		40.8	21.2	10.4	4.1	9.1	4.4	29.8	1.6	23.0	18.6	6.7
<i>Genetic progress</i>												
p -value M1 (r^2)		ns	ns	ns	ns	**	***	ns	ns	ns	-	-
p -value M2 (r^2)		ns	ns	ns	ns	ns	**	ns	ns	ns	-	-
p -value M3 (r^2)		ns	ns	ns	ns	ns	ns	ns	-	ns	ns	ns

addition. M2 exhibited the highest TDW across both sowing dates. At M1, TDW was predominantly composed of VDW, with leaves (Le) and stems (St) as major contributors, while Sq were the primary components of RDW (Table 2). From M2 to M3, RDW was the main component, with green bolls (Gb) and FS being the main components of RDW at M2 and M3, respectively (Table 2).

Most biomass variables showed no significant genetic progress, except for leaf biomass (Le) at M1 and reproductive branch biomass (Rb) at M1 and M2 (Fig. 2). Both variables showed significant decrease over the years, with a rate of $0.28 \text{ g m}^{-2} \text{ y}^{-1}$ [LE (M1) = $677.7 - 0.28 \cdot \text{YOR}$; $p = 0.02$; $R^2 = 0.33$] for Le at M1 and a rate of 0.18 and $0.21 \text{ g m}^{-2} \text{ y}^{-1}$ for Rb=M1 [Rb (M1) = $404.5 - 0.18 \cdot \text{YOR}$; $p = 0.004$; $R^2 = 0.43$] and M2 [Rb (M2) = $485.4 - 0.21 \cdot \text{YOR}$; $p = 0.01$; $R^2 = 0.33$], respectively (Fig. 2). In addition, although TDW, VDW, and RDW showed no significant genetic progress rates, they displayed a tendency toward negative values at M1 and M2, transitioning to positive or stable trends by M3 (Fig. 3).

3.3. Analysis of variance and genetic progress for morphological-related variables

Morphological variables measured during the experiment exhibit no differences among varieties, nonetheless they showed significant interactions between SD and M, except for number of vegetative branches per plant (Table 3). Node of the first reproductive branch (N1RB) and its height (H1RB) showed significant genetic progress across the year of released explored, presenting an increase of 0.01 node y^{-1} for N1RB at both M1 ($-4.5 + 0.01 \cdot \text{YOR}$; $p = 0.02$; $R^2 = 0.24$) and M2 (N1RB = $-7.0 + 0.01 \cdot \text{YOR}$; $p = 0.03$; $R^2 = 0.27$), respectively. Similarly, H1RB showed annual increases of 0.02 , 0.03 , and 0.04 cm y^{-1} at M1 (H1RB = $-28.9 + 0.02 \cdot \text{YOR}$; $p = 0.03$; $R^2 = 0.21$), M2 (H1RB = $-49.5 + 0.03 \cdot \text{YOR}$; $p = 0.005$; $R^2 = 0.41$), and M3 (H1RB = $-49.3 + 0.04 \cdot \text{YOR}$; $p < 0.001$; $R^2 = 0.56$), respectively (Fig. 4).

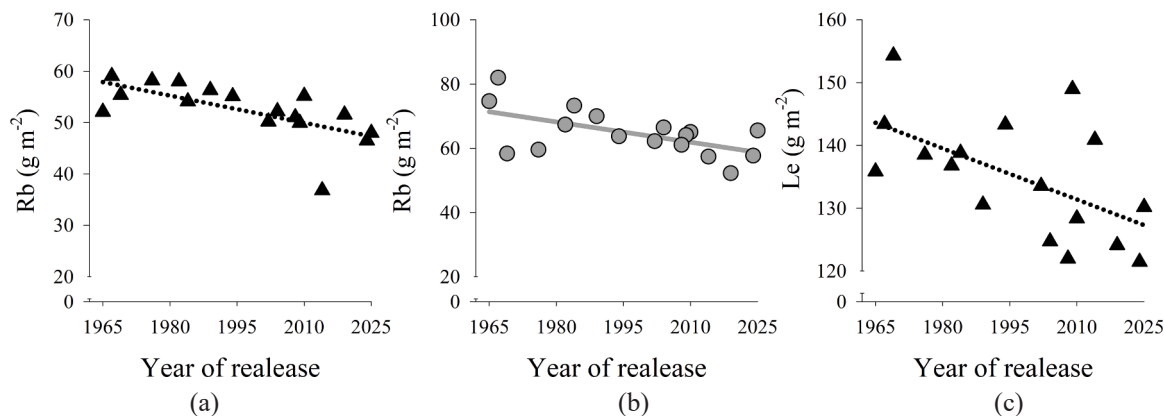


Fig. 2. Rate of genetic progress for reproductive branches biomass (Rb – a and b) and leaves biomass Le – c). Black triangle represented data from 50 days after emergence (DAE, M1), whereas gray circle represent data from 100 DAE (M2). Rates were estimated using cultivars from 1965 to present, evaluated in four different environmental conditions. Linear regressions were significant in all cases ($p < 0.05$) with black dotted and grey solid lines representing rate of genetic progress at M1 and M2, respectively. The points for 2004 and 2019 are an average of two and three genotypes sharing the year of release, respectively.

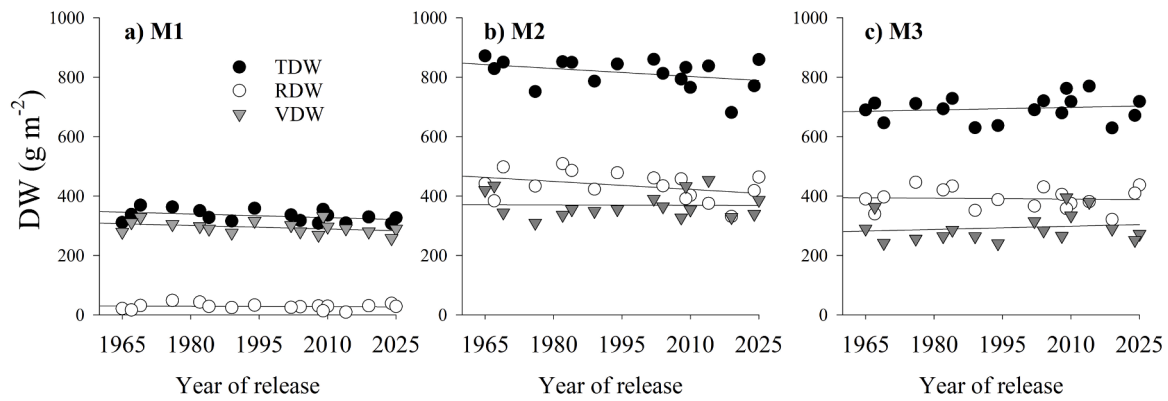


Fig 3. Rate of genetic progress for total dry weight (TDW), reproductive dry weight (RDW), and vegetative dry weight (VDW), represented as black circles, white circles, and gray triangles, respectively. Rates were estimated using cultivars from 1965 to present, evaluated in four different environmental conditions. Linear regressions were non-significant in all cases ($p > 0.05$). The points for 2004 and 2019 are an average of two and three genotypes sharing the year of re.

Table 3

Statistical analysis (ANOVA), mean values, and analysis of genetic progress for the sowing dates (SD), moments (M), and genotypes (G) evaluated in the experiment. He: height, NN: number of nodes, HR: height per nodes ratio, NVB: number of vegetative branches, N1RP: node number of the 1st reproductive branch, H1RP: height of 1st reproductive branch. M1, M2, and M3 measurement moments were at 50, 100, and 150 days after emergence. * $p < 0.05$, ** $p < 0.01$, and *** $p < 0.001$. LSD: least significant difference for M*SD interaction.

Morphological measurements							
Sowing Date	Moment	H (cm)	NN	HR (cm node ⁻¹)	NVB (n pl ⁻¹)	N1RB	H1RB (cm)
SDI	-	65.2	17.0	3.8	1.8	6.9	21.3
SDII	-	91.4	16.0	5.6	1.6	5.7	24.4
-	M1	69.6	14.5	4.7	1.5	6.2	22.3
-	M2	82.4	17.6	4.7	2.3	6.2	23.0
-	M3	83.0	17.6	4.8	1.2	6.4	23.3
SDI	M1	54.8	14.7	3.8	1.6	6.9	21.4
	M2	67.8	18.0	3.8	2.4	6.9	21.4
	M3	70.6	18.1	4.0	1.3	6.8	21.2
SDII	M1	80.4	14.4	5.7	1.5	5.5	23.4
	M2	92.5	17.1	5.6	2.1	5.7	24.9
	M3	91.0	16.4	5.7	1.1	6.0	25.7
Source of variation							
Moment (M)		***	***	***	***	**	***
Genotype (G)		ns	ns	ns	ns	ns	ns
Sowing Date (SD)		***	***	***	***	***	***
M * SD		***	***	*	ns	***	***
G * SD		ns	ns	ns	ns	ns	ns
M * G		ns	ns	ns	ns	ns	ns
M * G * SD		ns	ns	ns	ns	ns	ns
L.S.D (M*SD)		2.6	0.4	0.1	0.2	0.2	0.9
Genetic progress							
p-value M1 (r^2)		ns	ns	ns	ns	*	*
p-value M2 (r^2)		ns	ns	ns	ns	*	**
p-value M3 (r^2)		ns	ns	ns	ns	ns	***

3.4. Analysis of variance and genetic progress for mapping-related variables

Mapping variables and reproductive/vegetative ratio (R/V) demonstrated significant interactions between SD and M, and R/V also showed interaction between M and G (Table 4). Late sowing date (SDII) recorded the highest global retention (GR) values at M1 whereas, early sowing date (SDI) recorded higher GR than SDII at M2 and M3. In addition, GR values for the first position (GR1P) exceeded those for the second position (GR2P) across all environments and moments. R/V showed higher values in SDII than SDI, except at M1 where SDI had higher values. Reproductive allocation (RA) and harvest index (HI) were consistently higher in SDI compared to SDII, with differences among genotypes.

Among all the variables, only GR showed a significant increase of $0.02 \% y^{-1}$ at M2 ($GR = -26.8 + 0.026 * YOR$; $p = 0.04$; $R^2 = 0.25$) (Fig. 5).

4. Discussion

We have recently demonstrated that LY presented a rate of genetic progress of $3.24 kg ha^{-1} y^{-1}$, which represents an increase of $0.47 \% y^{-1}$ resulting in an increase of $194.4 kg ha^{-1}$ of lint during the period analyzed in Argentina. Yield improvements in modern varieties were linked to increases in lint percentage, seed cotton yield, boll number, and lint per seed (Scarpin et al., 2022). Additionally, we reported that oil content in the seed increased in Argentine cotton varieties released from 1965 to the present (Scarpin et al., 2023), and any of the main lint quality parameters have shown a decrease despite the increase in LY. We also evaluated several correlations between LY and its components providing guidance for future breeding efforts on INTA germplasm lines. This study expands upon previous work by reporting the first comprehensive analysis of biomass, morphological, and mapping-related variables for Argentinian cotton genotypes across different sowing dates and crop cycle stages.

4.1. Analysis of the variance

Total dry weight (TDW) exhibited a consistent pattern across all four environments, increasing from M1 to M2 before declining at M3. This decrease aligns with the findings of Meredith Jr. and Wells (1989), who attributed this to leaf senescence and organ abscission. Previous research on cotton cultivation reported no significant differences among varieties, describing TDW values of 161, 963, and $944 g m^{-2}$ at moments corresponding to M1, M2, and M3, respectively (Wells and Meredith Jr., 1984a). In contrast, Yang et al. (2019), Conaty and Constable (2020), and Singh et al. (2023), reported differences in TDW at M3, with values of 1625, 2228, and $852.3 g m^{-2}$ recorded for Chinese, Australian, and from the US varieties, respectively. This difference could be associated to the length of the cycle of the genotypes used in the countries above-mentioned, since the growing season is longer than in Argentina, possible allowing a higher plant development.

At M1, TDW primarily consisted of vegetative structures, with leaves and stem accounting for 90 % of TDW. At M2, reproductive dry weight (RDW) became dominant, driven mainly by green bolls (Gb) and fiber and seeds (FS) at M2 and M3, respectively. Vegetative dry weight (VDW) at M2 and M3 was mainly composed of stems, while leaf biomass decreased significantly from M1 to M3 due to senescence and leaf drop. Few studies have examined the composition of dry matter at different sampling moments in cotton genetic progress studies, nonetheless, Meredith and Wells (1989) reported similar trends, with vegetative structures as the main components of TDW during M1 and M2.

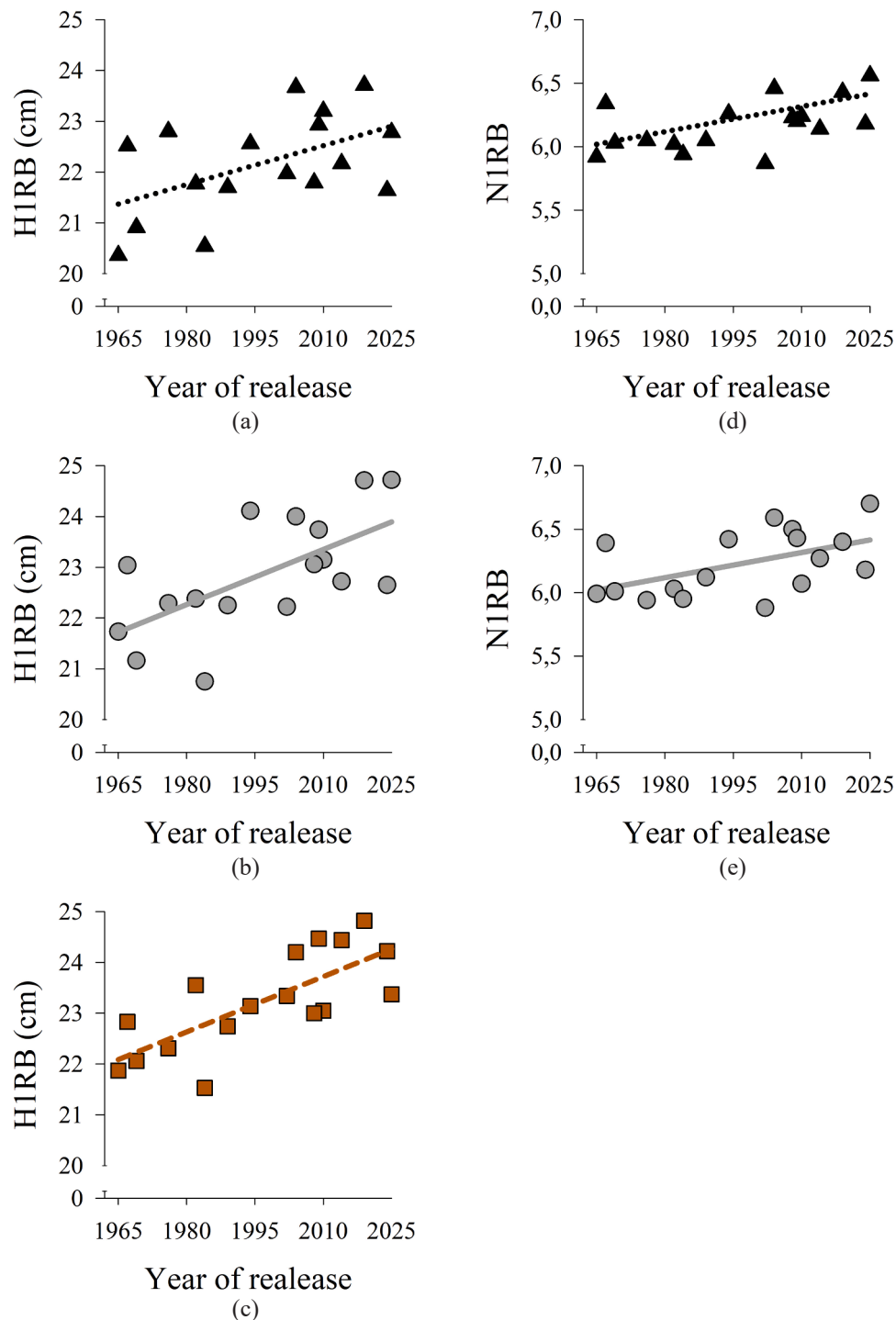


Fig. 4. Rate of genetic progress for: height of 1st reproductive branch (H1RB) (a, b, and c) and node number of the 1st reproductive branch (N1RB) (d and e). Black triangles represented data from 50 days after emergence (DAE, M1) (a and d), gray circles represent data from 100 DAE (M2) (b and e), and orange squares represent data from 150 DAE (M3) (c). Rates were estimated using cultivars from 1965 to present, evaluated in four different environmental conditions. Linear regressions were significant in all cases ($p < 0.05$) with black dotted, grey solid, and orange dashed lines representing rate of genetic progress at M1, M2, and M3 respectively. The points for 2004 and 2019 are an average of two and three genotypes sharing the year of release, respectively.

Morphological analysis revealed significant differences influenced by moments (M), sowing date (SD), and their interaction. In this work, some novel measurements were included, such as number of the first reproductive branch (N1RB) and its height (H1RB), in addition to common morphological variables for the crop such as height (H), number of nodes (NN), and height to node ratio (HR). These metrics, particularly those linked to earliness (Shakeel et al., 2011; Song et al., 2012), offer valuable insights into production location within the plant,

with practical and physiological implications (Constable, 1991). Different morphological variables, such as stem diameter (Guzman et al., 2021) and the angle of reproductive branches insertion (Mondino, 2000), have been used in previous cotton studies. However, few studies on genetic progress in cotton have analyzed morphological variables alongside lint yield or fiber quality parameters. Wells and Meredith Jr. (1984a) found differences in NN, HR, and N1RB, with lower H values at M1 but higher values at M2 and M3 than ours, suggesting the impact of

Table 4
Statistical analysis (ANOVA), mean values, and analysis of genetic progress for the sowing dates (SD), moments (M), and genotypes (G) evaluated in the experiment. GR: global retention, GR1P: global retention in 1st position, GR2P: global retention in 2nd position, R/V: reproductive/vegetative ratio, RA: reproductive allocation, HI: harvest index. M1, M2, and M3 measurement moments were at 50, 100, and 150 days after emergence. **p* < 0.05, ***p* < 0.01, and *** *p* < 0.001. LSD: least significant difference for M*SD interaction.

Retention measurement							
Sowing Date	Moment	GR (%)	GR1P (%)	GR2P (%)	R/V	AR (M3)	HI (M3)
SDI	-	42.2	48.4	34.6	0.71	0.37	0.20
SDI	-	38.4	53.9	35.3	1.05	0.53	0.27
-	M1	69.6	78.1	65.0	0.10	-	-
-	M2	25.4	37.6	20.4	1.16	-	-
-	M3	25.9	37.7	19.5	1.39	0.45	0.24
SDI	M1	68.5	70.7	61.5	0.11	-	-
SDI	M2	29.6	38.2	21.0	0.95	-	-
SDI	M3	29.0	36.1	20.9	1.09	0.37	0.20
SDII	M1	72.2	85.8	67.6	0.09	-	-
SDII	M2	22.3	37.0	19.4	1.34	-	-
SDII	M3	24.2	39.4	17.2	1.72	0.53	0.27
Source of variation							
Moment (M)		***	***	***	***	-	-
Genotype (G)		ns	ns	ns	***	***	***
Sowing Date (SD)		*	***	ns	***	***	***
M * SD		***	***	*	***	-	-
G * SD		ns	ns	ns	ns	ns	ns
M * G		ns	ns	ns	***	-	-
M * G * SD		ns	ns	ns	ns	-	-
L.S.D (M*SD)		3.0	2.9	3.9	0.02	0.02	0.01
Genetic progress							
p -value M1 (r ²)		ns	ns	ns	ns	-	-
p -value M2 (r ²)		*	ns	ns	ns	-	-
p -value M3 (r ²)		ns	ns	ns	ns	ns	ns

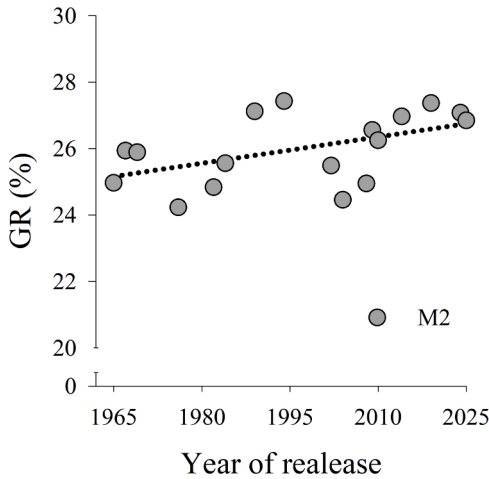


Fig. 5. Rate of genetic progress for global retention (GR) from data recorded 100 days after emergence (DAE, M2). Rates were estimated using cultivars from 1965 to present, evaluated in four different environmental conditions. The linear regression was significant (*p* < 0.05) with gray solid line representing rate of genetic progress at M2. The points for 2004 and 2019 are an average of two and three genotypes sharing the year of release, respectively.

longer growing conditions. (Meredith and Wells, 1989). Also, Guzman et al. (2021) reported 159 cm of H at M3 for varieties from Venezuela, suggesting that differences in H could be attributed not only to genetics but also to the spacing between rows (Roche and Bange, 2022), since we carried out our experiments using narrow rows (52 cm), while the cited authors carried out their assay using “conventional” spacing (90 cm).

Global retention (GR) decreased after M1 as the crop cycle progressed, with an average of 25.9 % at M3 across all environments. This

important reduction was attributed to the shedding of reproductive structures, which could be caused by physiological, environmental, or biotic factors. In terms of environmental differences, SDI had higher values, except for M1, where SDII presented higher GR than SDI. Genetic progress studies on plant retention variables remain limited, however Kilby et al. (2012) reported GR values (22 %–38 %) similar to our findings among six Australian cultivars. Additionally, the reproductive-to-vegetative dry weight (R/V) ratio increased across stages, consistent with earlier studies (Wells and Meredith Jr., 1984b, Meredith Jr. and Wells, 1989). Nonetheless, higher R/V values have been reported in recent studies suggest scope for improvement in Argentinian breeding programs (Yang et al., (2019), Conaty and Constable, (2020), Singh et al. (2023)).

4.2. Genetic progress

This study showed that among all the variables analyzed, only Le (at M2), Rb (at M2 and M3), N1RB (at M1 and M2), H1RB, and GR (at M2) exhibited significant increases or decreases depending on the YOR. The estimated TDW showed a non-significant decline with YOR, decreasing by 41 and 89 g m⁻² at M1 and M2, respectively, over the analyzed period. Interestingly, while TDW exhibited non-significant declines with YOR at M1 and M2, a positive trend emerged at M3, indicating a higher rate of biomass accumulation in modern cultivars later in the season. Wells and Meredith Jr. (1984a) reported no increases or decreases in TDW across different years and sowing dates in the US, determining that modern cultivars exhibited negative correlations between VDW and YOR at M2 and M3. They suggested that modern cultivars complete the transition from vegetative to reproductive stages earlier than obsolete cultivars (Wells and Meredith Jr., 1984a). Positive trends in TDW at M3 were reported by Yang et al. (2019) in China and Conaty and Constable (2020) in Australia, associating this increase with improvements in LY. These authors explained that cultivars with higher biomass allowed to maintain a greater number of bolls per unit area. An increase in TDW can be achieved by improving certain parameters involved in its determination, such as the rate of radiation received, cycle length, radiation use efficiency, or radiation interception efficiency (De la Fuente et al., 2023). To increase the TDW of modern cultivars, the options would be either to lengthen the cycle or increase physiological parameters such as carbon assimilation (by increasing the photosynthetic rate or decreasing the consumption of carbohydrates by respiration), radiation use efficiency, or radiation interception efficiency. In this sense, recent studies have shown differences up to 53 % in radiation use efficiency among genotypes (Virk et al., 2023). Cotton crop cycle is limited by environmental conditions in Argentina, so long-cycle crops may not adapt well in all regions of the country. Additionally, negative trend change observed in this study for TDW between M2 and M3 in modern cultivars could suggest higher carbon fixation during the middle and late cycle stages in the modern cultivars, agreeing with results from Yang et al. (2019). However, further studies on carbon assimilation levels during different stages are needed to confirm this assumption for Argentinian genotypes.

No significant progress was detected in indices related to TDW, VDW, RDW, suggesting that modern and obsolete cultivars allocated similar proportions of TDW to reproductive organs. The non-significant increase in harvest index (HI) and reproductive allocation (RA) indicates that modern cultivars favor lint over seed weight, potentially impacting seed size and seedling vigor (Snider et al., 2016). Although ginning turnout has increased in modern Argentine varieties (Scarpin et al., 2022), it has not been enough to significantly increase the HI term, which directly relates lint to TDW. These findings do not align with the observed increase in the HI parameter without a corresponding increase in RA (Sadras et al., 1997; Conaty and Constable, 2020). Investigating these trade-offs and optimizing resource allocation within bolls may enhance breeding outcomes for cotton in Argentina.

Among morphological variables, positive genetic progress rates were

observed in H1RB and N1RB at different moments. Mauney, 1986 suggested that N1RB is determined not only by genetics but also by environmental factors such as temperature and solar radiation. In addition, positive trends were reported in H, NN, and HN, indicating that modern cultivars tend to have more height and length between nodes, as well as more space from the soil surface. These features suggest strong environmental stability and high heritability, indicating higher earliness in modern cultivars (lower placement of the first reproductive branch). Unlike the increased square and flower in modern genotypes count by Wells and Meredith (1984b), our experiment indicated that modern cultivars prioritize retaining existing reproductive structures rather than producing more initially. This translates to a higher final number of bolls per area at harvest, as previously reported (Scarpin et al., 2022). The increase in GR in modern cultivars may be attributed to the availability of assimilates, allowing for greater retention of reproductive structures. Further studies are needed to confirm this assumption. Despite the lack of similar studies on genetic progress in these morphological variables, their potential value in breeding programs should not be underestimated. The resource-intensive nature and laborious measurement techniques highlight the need for more efficient and faster methods to measure these valuable traits.

5. Conclusion

The present study represents the first to quantify aboveground biomass, morphological, and mapping related variables of cotton in Argentina with the aim of assessing their genetic progress. Based on the experiments and cultivars used in the present study we can conclude that recent cotton breeding in Argentina has affected the levels of leaves biomass (at M2), reproductive branches biomass (at M2 and M3), node of the first reproductive branch (at M1 and M2), height of first reproductive branch (at M1, M2, and M3), and global retention (at M2) showing this variables either increases or decreases on the rate of genetic progress. Our findings support the hypothesis that modern cultivars demonstrate a greater capacity for retaining reproductive organs. However, they do not exhibit a stronger relationship between reproductive and vegetative biomass allocation. The limited reporting of aboveground biomass, morphological, and mapping-related variables in Argentinian cotton is likely due to the labor-intensive and time-consuming nature of these measurements. Nevertheless, the differences observed across environments and cultivars underscore their importance in breeding programs. Future studies should integrate these variables into broader evaluations of lint yield, yield components, and fiber and seed quality traits. This approach will strengthen the foundation for genetic improvement strategies in cotton, not only in Argentina but also in other regions with similar environmental and genetic conditions.

CRedit authorship contribution statement

Pablo N. Dileo: Methodology, Formal analysis, Conceptualization. **H. Martin Winkler:** Visualization, Methodology, Conceptualization. **Gonzalo J Scarpin:** Writing – review & editing, Writing – original draft, Visualization, Validation, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. **Marcelo J. Paytas:** Writing – review & editing, Supervision, Project administration, Investigation, Funding acquisition, Conceptualization. **Carlos Acuña:** Writing – review & editing, Supervision. **Robertino J Muchut:** Methodology, Conceptualization. **Roxana A Roeschlin:** Methodology. **Fernando G Lorenzini:** Writing – review & editing, Methodology, Investigation, Conceptualization. **Antonela E Cereijo:** Writing – review & editing, Methodology, Conceptualization.

Declaration of Competing Interest

The authors declare that there is no conflict of interest.

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Data availability

Data will be made available on request.

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