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Growth of aloe vera (*Aloe barbadensis* Miller) basal shoots in companion planting systems

Crecimiento de retoños basales de aloe vera (*Aloe barbadensis* Miller) en sistemas de producción asociados

Jacobo Robledo¹, Jessica Valencia¹, and William A. Hincapié²

ABSTRACT

Aloe vera (*Aloe barbadensis*) is an important plant to cosmetics, pharmaceuticals, and food industry worldwide. In Colombia its cultivation has grown even when technical crop management is unknown. This study evaluated the growth of three aloe basal shoots weights ranges in two companion planting systems and monoculture (control). A completely randomized split plot design was used. Main plots were: aloe monoculture (AMN), common bean companion planting (CBCP), and giant taro companion planting (GTCP). Treatments were weight ranges from 50 to 150 g (LWe), 151 to 250 g (MW), and 251 to 350 g (HW). Data were analyzed using ANOVA, Duncan multiple range test ($P \leq 0.05$), and linear regressions. Variables evaluated were total height (TH), number of leaves (NOL), length (LL), width (LW), and leaf thickness (LT). In CBCP, GTCP, and MW variable LL predicted GH. Models fitted to HW and AMN were not representative ($R^2 < 0.64$). CBCP obtained the highest values in NOL (17.8), TH (56.2 cm), LL (40.2 cm), and LW (5.8 cm). LWe and MW basal shoots reached non-significant differences one year after planting in any variable ($P > 0.05$). Companion planting promotes predictability of aloe growth and CBCP associated with HW are a promising alternative to aloe cultivation.

Key words: medicinal plants, asexual reproduction, cropping system, crop physiology.

RESUMEN

Aloe vera (*Aloe barbadensis*) es una planta importante para la industria cosmética, farmacéutica y alimenticia en el mundo. En Colombia su cultivo ha crecido incluso cuando su manejo técnico es desconocido. Este estudio evaluó el crecimiento de tres rangos de pesos de brotes basales en dos sistemas asociados y monocultivo (control). Se utilizó un diseño de parcelas divididas al azar. La parcela principal fue monocultivo de aloe (MNA), asociación con frijol (CAF) y asociación con boniato (CAB). Los tratamientos fueron rangos de 50 a 150 g (PB), 151 a 250 g (PM) y 251 a 350 g (PA). Se realizó ANAVA, prueba de Duncan ($P \leq 0.05$) y regresiones lineales. Las variables evaluadas fueron altura total (AT), número de hojas (NH), longitud (LH), ancho (AH) y grosor de la hoja (GH). En CAF, CAB, y PM la variable LH predijo AH. Los modelos realizados con PA y MNA no fueron representativos ($R^2 < 0.64$). CAF obtuvo los valores más altos en NH (17,8), AT (56,2 cm), LH (40,2 cm) y AH (5,8 cm). Los brotes basales de PB y PM no alcanzaron diferencias significativas un año después de la siembra en todas las variables ($P > 0,05$). Los cultivos asociados promueven la previsibilidad del crecimiento del aloe y CAF en conjunto con PA es una alternativa prometedora para el cultivo del aloe.

Palabras clave: plantas medicinales, reproducción asexual, sistema de cultivo, fisiología del cultivo.

Introduction

Aloe vera (*Aloe barbadensis* Miller) is a succulent plant native of Northern Africa and resistant to drought, it belongs to Xanthorrhoeaceae family of the order Asparagales (Baruah *et al.*, 2016). The family Xanthorrhoeaceae is compounded by more than 250 species of plants. However, only two species acquire commercial importance and *Aloe barbadensis* is one of them (Manvitha and Bidya, 2014). The first reports of the cultivation and use of aloe vera in folk medicine date back as long ago as 1,500 B.C. (Hasanuzzaman *et al.*, 2008). Nowadays, this plant is industrially

processed in a wide range of food, healthcare, and cosmetics products due to its nutraceutical qualities (Javed and Atta-ur, 2014). Among the therapeutic properties of aloe vera must be consider its laxative effect (Hamman, 2008), wound healing qualities (Bozzi *et al.*, 2007; Baruah *et al.*, 2016), immunomodulatory action (Mulay *et al.*, 2013), anti-inflammatory activity (Davis *et al.*, 1989), and anti-viral potential (Choonhakarn *et al.*, 2008).

In Colombia the cultivation of *aloe vera* represents an economic potential due to the deficit on supply of products elaborated with the gel of this plant and the relatively

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high adaptability of aloe vera to different environments (SIOC, 2016; Manvitha and Bidya, 2014). In 2014 Colombia achieved the highest amount of aloe vera yield and planted area with 6,000 kg (44% of this yield was obtained in the Department of Cundinamarca) and 185 ha, respectively (Agronet, 2017). Despite this, the technical management of aloe vera cultivation varies. For instance, there are reports that claim a time between 7 to 36 months as the recommended time since planting to first harvest (Manvitha and Bidya, 2014; Biswas, 2010; Nilanjana-Das and Chattopadhyay, 2004; Figueredo and Morales, 2010; Alegbejo, 2012) and values from 10 cm to 20 cm as the optimal height of aloe vera basal shoots for plant propagation (Nilanjana-Das and Chattopadhyay, 2004; Alegbejo, 2012; Díaz, 2013). These variances directly affect the cost of aloe vera plantations and demonstrate the necessity to obtain suitable parameters to promote economically sustainable development and basic management practices based on local technical advances.

High yields and productivity must be maintained in modern sustainable agriculture throughout environmental-accepted practices (Tilman *et al.*, 2002). Companion planting systems are a promising alternative that develops two or more vegetal species at the same time in the same field. The species involved in companion planting systems could finish their productive cycle simultaneously or separately. Companion planting offers advantages that increase the field use and crop yield by optimizing resources such as water, radiation, nutrients (decreasing external farm inputs), and cash flow (Rodríguez *et al.*, 2008; Leihner, 1983). One of the most important companion planting systems in Latin America is the common bean (*Phaseolus vulgaris* L.) (Lithourgidis *et al.*, 2011). This vegetal species is recognized due to its high nutritional qualities for humans and animals, environmental adaptability and short productive cycle (Popelka *et al.*, 2004). In addition, giant taro (*Alocasia macrorrhiza*) is an emergent plantation in Colombia adapted to local conditions that demonstrates nutritional potential for feeding broilers and fish (López *et al.*, 2012; Poot-López *et al.*, 2012).

The interest in applying sustainable strategies in aloe vera production is increasing. However, in Colombia the cultivation of aloe vera lacks suitable information about its accurate establishment and development justified under its relatively easy management. For that reason, the present study aims to determinate the effect of different weights of basal shoots and companion planting systems on the growth and development of aloe vera cultivation in the department of Caldas, Colombia.

Materials y methods

Location

This study was conducted from September 2014 to December 2015 at Montelindo farm of Caldas University located at 5°05'10.2" N and 75°41'20.0" W in the municipality of Santagueda, Caldas, Colombia. With an altitude of 1,010 m a.s.l, average annual precipitation of 2,100 mm, average relative humidity of 76%, average temperature of 23.5°C, annual solar brilliance of 2010 h, and sandy loam soils with a slope lower than 3%.

Material for plant propagation

The basal shoots used in this study were obtained from a commercial crop of 18 months in an excellent phytosanitary condition located at 5°01'16.3" N and 75°31'57.1" W in the Buenavista district of the municipality of Manizales, Colombia with an average altitude of 1,780 m a.s.l. The extraction of the basal shoots was performed during the morning, retiring the emergent plant from the base of the mother plant manually. Afterwards, the basal shoots were cleaned, weighed, and classified into three groups of: (i) 50 g to 150 g (low weight = LWe), (ii) 151 g to 250 g (medium weight = MW), and (iii) 251 g to 350 g (high weight = HW). Cleaned and weighed basal shoots were transported into plastic baskets of 60×40×18 cm to Montelindo farm.

Cropping systems

Three cropping systems were used in this study: i) *Phaseolus vulgaris* var. ICA Quimbaya (common bean) as planting companion of aloe vera (CBCP), ii) giant taro (*Alocasia macrorrhiza*) as planting companion of aloe vera (GTCP), and iii) aloe vera monoculture cropping system (AMN) used as control. AMN (control cropping system) was planted in double rows with 0.50 m between plants, 0.50 m between linear rows and 1 m between double rows (26,666 plants/ha). CBCP and GTCP were respectively planted between the double rows of MN reaching a planting density of 19,047 plants/ha (0.35 m between plants and 1.5 m between aloe vera double rows) and 4,443 plants/ha (1.5 m between plants and 1.5 m between aloe vera double rows) (Fig. 1). The first cycle of common bean in CBCP was performed 15 d after the establishment of the aloe vera cultivation and the second cycle six months later. Giant taro plants were already established (80 cm average height) when the aloe vera basal shoots were planted.

Experimental design and data analysis

A completely randomized split plot design with ten replications and one plant as experimental unity was used. The main plot was composed of three levels equivalent to the

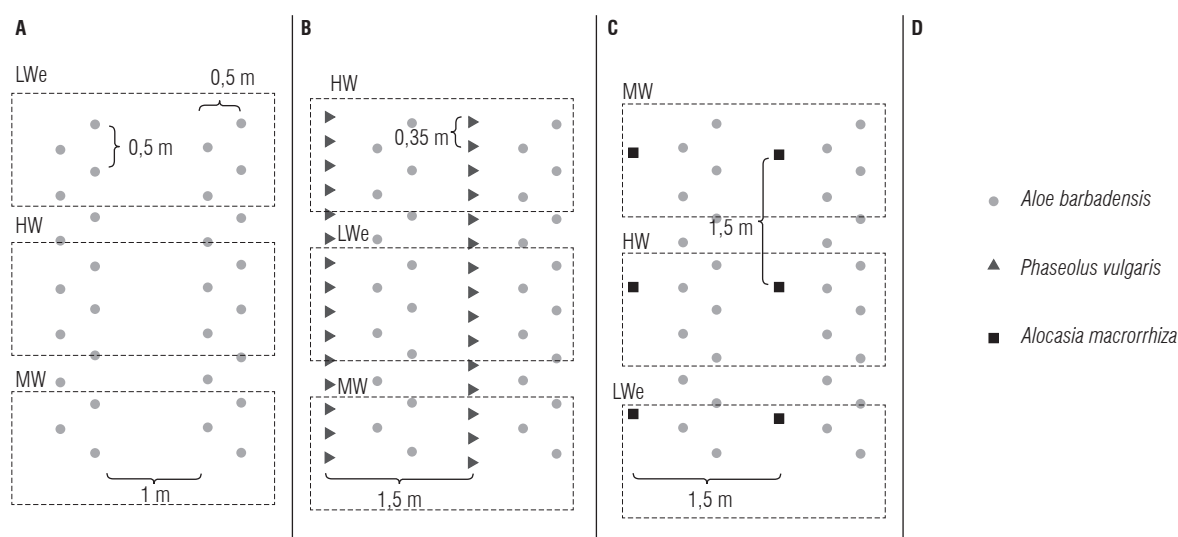


FIGURE 1. Spatial arrangement of main plots, treatments and planting distances. A) Aloe vera monoculture or control cropping system (AMN); B) Common bean (*Phaseolus vulgaris*) companion planting (CBCP); C) Giant taro (*Alocasia macrorrhiza*) companion planting (GTCP); D) plant representation. LWe: low weight (50-150 g), MW: medium weight (151-250 g), HW: high weight (250-350 g).

cropping system described in the *Cropping system* section (AMN, CBCP, and GTCP) and treatments were composed of three levels, equivalent to the basal shoot weight ranges described in the *Plant material* section (LWe, MW, HW) (Fig.1). Total number of observations were composed of 90 plants. Data were analyzed using analysis of variance (ANOVA) Duncan multiple range test ($P \leq 0.05$) and linear regression models (models with R value greater than 0.8 were considered representative for this study) through Agricolae library of R language software (R Development Core Team, 2010).

Variables

Five morphometric variables of aloe vera plant and leaf were evaluated 6 months and 12 months after planting: total height (TH), total number of leaves (NOL), leaf length (LL), leaf width (LW), and leaf thickness (LT). TH was the perpendicular distance from the base of the plant to the end of the apex of the longest leaf. LL, LW, and LT were respectively calculated with the length, width, and thickness of leaves number one, four, and eight of each plant using ruler

and digital calibrator, respectively. Leaves were counted from the most external leaf with excellent phytosanitary condition (first leaf) to the most internal leaf (last leaf) following the natural architecture of the plant.

Results and discussion

Cropping systems

Values in all the variables evaluated in the plots with the different cropping systems (main plots) increase both six and twelve months after planting. This fact allows claiming that each cropping system and their particular microclimate properties stimulated growth and generation of new vegetative structures in the cultivation and thus modified the morphometric qualities (Tab. 1 and 2). These qualities, also called phenotype, are determined by the summation of the genotype of the plant and the environmental condition (Coleman *et al.*, 1994). However, in commercial crops of CAM (Crassulacean acid metabolism) plants propagated from basal shoot, genetic variability decreases and phenotype is linked principally to the environmental conditions

TABLE 1. Variables evaluated in the cropping systems through time.

Cropping system	Six months after planting					Twelve months after planting				
	NOL	TH (cm)	LL (cm)	LW (cm)	LT (cm)	NOL	TH (cm)	LL (cm)	LW (cm)	LT (cm)
CBCP	14.1 a	44.8 ab	34.2 a	4.2 a	1.1 b	17.8 a	56.2 a	40.2 a	5.8 a	1.3 a
AMN	11.6 b	47.4 a	33.5 a	4.2 a	1.4 a	14.2 b	53.0 a	36.3 b	4.1 b	1.1 b
GTCP	12.3 b	41.7 b	28.1 b	3.3 b	1.0 b	15.0 b	46.3 b	36.5 b	4.2 b	1.4 a
P value	<0.001	<0.01	<0.001	<0.001	<0.001	<0.001	<0.05	<0.05	<0.001	<0.01

Means followed with the same letter do not differ according to Duncan Test at 5%.

TH: total height; NOL: number of leaves; LL: leaf length; LW: leaf width; LT: leaf thickness.

AMN: aloe vera monoculture or control cropping system; CBCP: common bean (*Phaseolus vulgaris*) companion planting; GTCP: giant taro (*Alocasia macrorrhiza*) companion planting.

and its possible disturbances such as caused by the planting companion (Bartholomew *et al.*, 2002).

Plants of the cropping systems showed significant differences ($P \leq 0.05$) in the total number of leaves (NOL), total height (TH), leaf length (LL), leaf width (LW), and leaf thickness (LT) at six months and at twelve months after planting. Common bean planting companion (CBCP) obtained the highest average in NOL six and twelve months after planting (Tab. 1). Duncan multiple range test defined two groups for the NOL variables: **A** for CBCP and **B** for AMN and GTCP. In this cropping system TH fitted into a linear model with LL and LW twelve months after planting with a coefficient of determination (R^2) above 0.67 (Fig. 2G and H). Linear behavior was also observed within LL as explicative variable of LW ($R^2 = 0.74$) (Fig. 2I). Averages of TH, LL, and LW in CBCP reached the highest values compared to the other cropping systems one year after planting and support the clear relations between these variables (Tab. 1).

CBCP, distinctly to GTCP and AMN (control cropping system), gives to the aloe vera plants variable shadow levels related to the height of the common bean plant. Physiologically, the length of aerial plant structures raise due to the increase of the sensitivity to auxins directly affected by the availability of light. Insofar solar radiation decreases, plant tissues promote anatomic changes (Basuk and Maynard, 1987). When a plant is continuously exposed to far red light (700-800 nm) synthesis of carbohydrates moves towards the stem instead of other structures such as roots (Bastías and Corelli-Grappadelli, 2012). Moderate shadow level (30% shadow) favors carbon retention in the leaves without affecting the glucose content decreasing the development of roots and basal shoots in aloe vera crops (Kawather *et al.*, 2001) while high light intensity and water deficit let to the photoinhibition of photosynthesis (Hazrati *et al.*, 2016). Páez *et al.* (2000) report respectively values of total number of leaves and total height of 16 leaves and 34 cm in aloe vera under field conditions, 21 leaves and 47 in aloe vera under moderate shadow, and 14 leaves and 41 cm in aloe vera under deep shadow. These results could be comparable with those obtained in this study.

Soil fertility and nutrient availability during the experimental phase were limited by the presence and absence of common bean crop residues and the ability of this crop to fix nitrogen from air as a result of a symbiotic relationship between roots and *Rhizobium* bacteria (Beck and Roughley, 1987). In aloe vera crops, nitrogen is an indispensable nutrient due to its importance in the conformation of photosynthetically active pigments that affect

development and yield (Olfati *et al.*, 2015). According to Trejo *et al.* (2008) the concentration of pigments and nitrogen fertilization are correlated. In aloe vera plantations cow manure and organic matter are equivalent to mineral fertilization with urea (Hasanuzzaman *et al.*, 2008). However, mineral nitrogen fertilization increases length, number and fresh weight of leaves (Egbuchua and Enujeke, 2015). As reported by Olfati *et al.* (2015) nitric sources of nitrogen associated with ammonium in low concentration promote growth of aloe vera. Nitrogen generates higher yield, content of gel, chlorophyll and aloin in the leaf (Hazrati *et al.*, 2012).

Plants that have grown under the cropping system GTCP presented the lowest values in TH, LL, LW, and LT six months after planting. Twelve months after planting GTCP achieved higher nominal values in NOL, LL, LW, and LT than those acquired by AMN (control cropping system). However, according to the Duncan test, only LT presented a differential behavior among these variables. These regular averages were confirmed with the linear models fitted to variables of GTCP which obtained the higher coefficient of determination among cropping systems ($R^2 > 0.70$). The linear models fitted to NOL, TH, and LL obtained the greatest R^2 values correlated to TH, LL, and LW, respectively (Fig. 2 A, B, D, and E). NOL and LL also fitted into a linear model as predictive variables of LT (Fig. 2 C and F). Nevertheless, data of LT do not support a tendency in any cropping system among time because the average of this variable fluctuates (Tab. 1). Different irrigation regimens have shown responses into the growth and yield of aloe vera affecting variables such as stomata resistance, plant and leaf growth (Rodríguez-García *et al.*, 2007). Although aloe vera is not a demanding plant in term of water (Sánchez-Machado *et al.*, 2017), according to Genet and Van Schooten (1992) leaf thickness directly depends on soil hydric availability at certain stages and Hazrati *et al.* (2017) report a maximum leaf fresh weight when plants were irrigated after depleting 40% of the field capacity. For that reason, if a cropping system improves soil humidity and prevents evapotranspiration it could directly influence the leaf thickness of aloe vera plants. However, Baruah *et al.* (2016) claim that chemical composition of aloe vera leaf can vary depending on abiotic factors such as annual season rainfall, temperature, incident solar radiation, harvest date, climate, land and cultivation methods.

Response of TH, LL, and LW evaluated in AMN (control cropping system) six months after planting reported a similar behavior to plants on CBCP (non-significant differences were reported between this two cropping systems)

TABLE 2. Variables evaluated in the basal shoot weight ranges through time.

Basal shoot	Six months after planting					Twelve months after planting				
	NOL	TH (cm)	LL (cm)	LW (cm)	LT (cm)	NOL	TH (cm)	LL (cm)	LW (cm)	LT (cm)
LWe	11.4 b	38.8 b	28.6 b	3.5 b	1.0 b	15.1 b	51.1 a	37.2 b	4.6 a	1.2 b
MW	13.3 a	46.6 a	32.9 a	4.0 a	1.2 a	15.2 b	50.4 a	36.5 b	4.6 a	1.3 b
HW	14.1 a	47.5 a	34.7 a	4.3 a	1.2 a	18.0 a	58.3 a	41.1 a	5.3 a	1.4 a
P value	<0.001	<0.001	<0.001	<0.001	<0.001	<0.01	ns	< 0.05	ns	< 0.01

Means follow with the same letter do not differ according to Duncan Test at 5%.

TH: total height; NOL: number of leaves; LL: leaf length; LW: leaf width; LT: leaf thickness; LWe: low weight (50-150 g); MW: medium weight (151-250 g); HW: high weight (251-350 g).

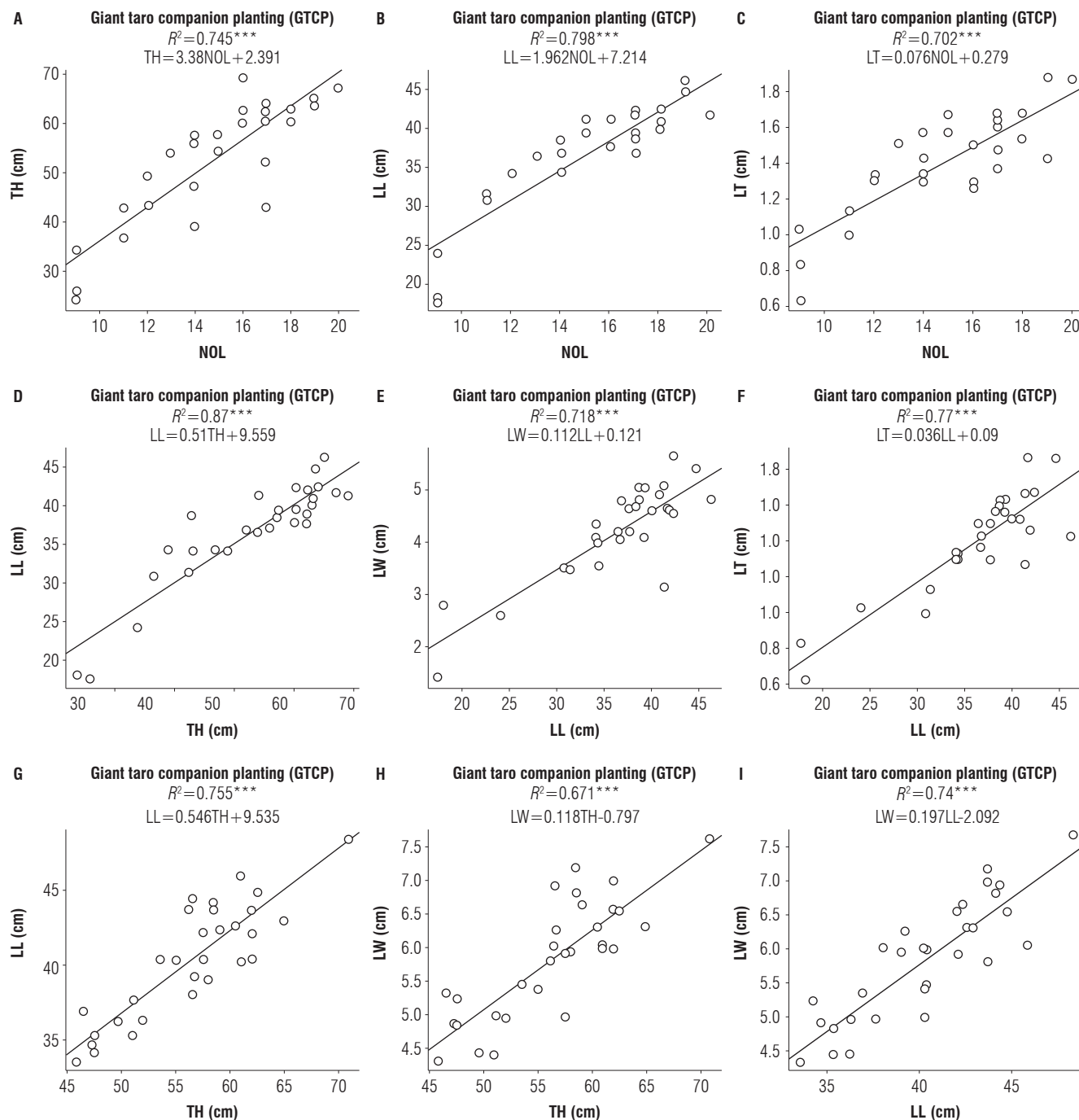


FIGURE 2. Linear model of variables evaluated in the cropping systems with a coefficient of determination (R^2) higher than 0.64. LL: leaves length; TH: total height; NOL: number of leaves; LW: leaves width; LT: leaves thickness; $^{***}P \leq 0.001$.

(Tab. 1). One year after planting, only TH continued this tendency and LL and LW were surpassed by the CBCP values. Coefficients of determination of linear models fitted among AMN (control cropping system) variables were not representative ($R^2 < 0.64$). Nonetheless, averages obtained by AMN (control cropping system) are comparable with those reported by Paez *et al.* (2000) in cropping systems under field conditions.

Basal shoots weight ranges

Six months after planting medium weight (MW) and high weight (HW) basal shoots exhibited a statistically similar response (non-significant differences were obtained among MW and HW) in NOL, TH, LL, LW, and LT. During this period of time low weight (LW) basal shoots reported the lowest averages ($P \leq 0.05$) in the variables previously mentioned being AT the one that reached the highest

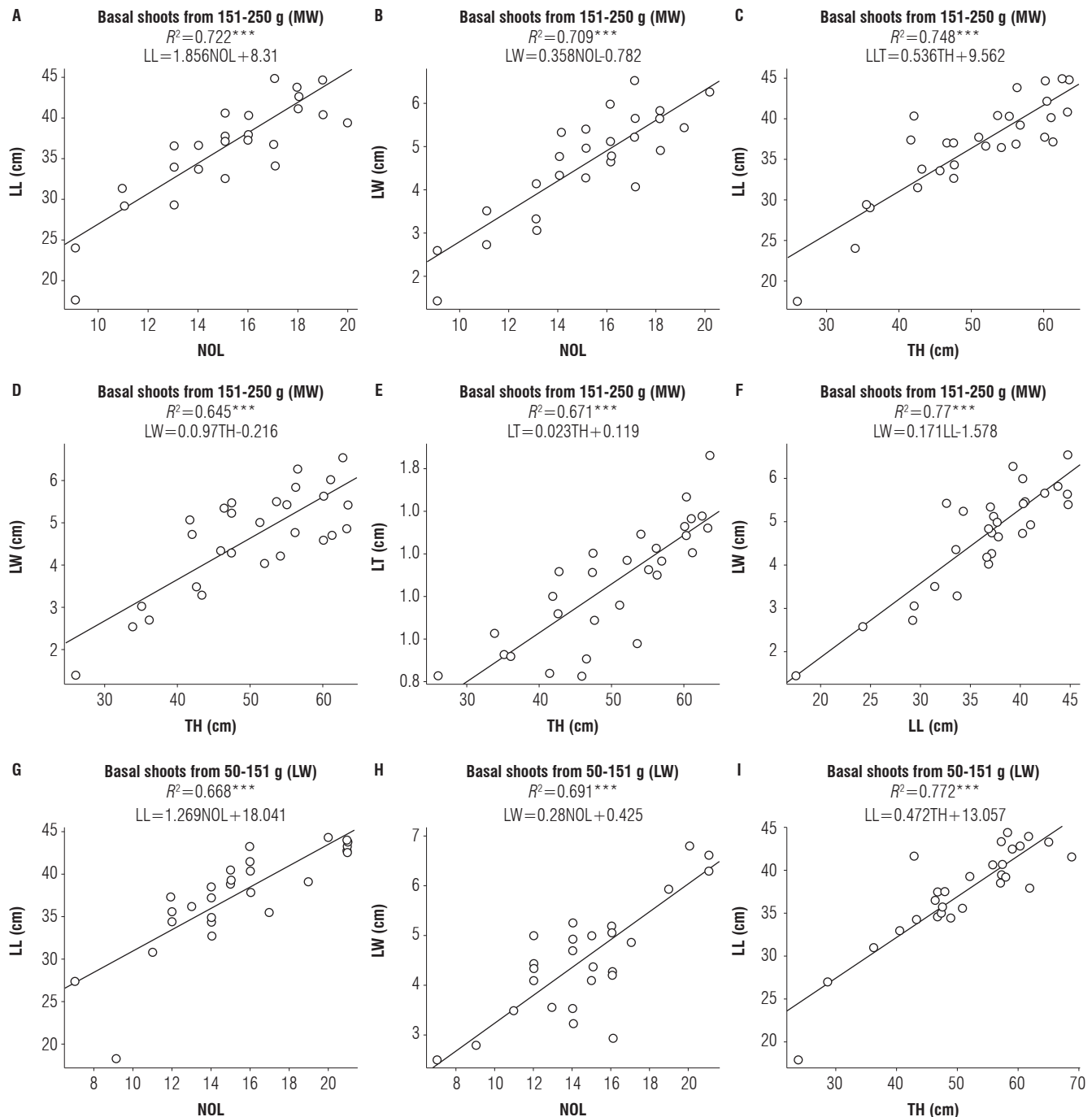


FIGURE 3. Linear model of variables evaluated in the basal shoot weight ranges with a coefficient of determination (R^2) higher than 0.64. LL: leaves length; TH: total height; NOL: number of leaves; LW: leaves width; LT: leaves thickness; $^{***}P \leq 0.001$.

difference in comparison with HW (8.7 cm). However, twelve months after planting basal shoots, they reported significant differences ($P \leq 0.05$) and HW acquired the highest values in NOL, LL, and LW. Otherwise, LWe and MW expressed phenotypic similar qualities in NOL, LL, LW, and LT (Tab. 2). TH and LW presented a comparable development (non-significant differences were found) in all ranges of basal shoots (LWe, MW, and HW) one year after planting. This tendency empirically allows observing no variation through basal shoots from a certain cropping system.

Linear models fitted to growth variables in LWe permitted to observe a directly proportional relation between NOL and TH as predictive variables of LL, LW, and LT (Fig. 3 A-E). This behavior was also observed between LL and LW in MW ($R^2 = 0.77$) (Fig. 3 F). In addition, linear models applied to LWe variables demonstrated a linear correlation in NOL and LL ($R^2 = 0.67$), NOL and LW ($R^2 = 0.69$), and TH and LL ($R^2 = 0.77$) (Fig. 3 G-I). Coefficient of determination (R^2) obtained in HW basal shoots was not representative. These results justified the lack of significant differences between TH and LW (Tab. 2) and suggest a high variability in TH and LL in spite of the fact that these variables reached the highest values in HW.

In CAM plants different from aloe vera with a similar agronomic management such as pineapple (*Ananas comosus*) and maguey (*Agave* spp.) the use of basal shoots from mother plants selected by their health and yield is the best commercial option for plant propagation due to crop homogeneity, genetic stability and cost (Brenes-Gamboa, 2011; Arizaga and Ezcurra, 2002). According to Bhandari *et al.* (2010) seed resulting from sexual crosses is ineffective and slow in commercial crops of aloe vera. In pineapple crops basal shoots with higher weight achieve, in certain time, bigger plants and fruits of those obtained in the lower weight basal shoots. For instance, the time lasting from planting to harvest is 15 months using basal shoots, 20 months using axillary shoots, and 24 months using collar roots (Office of the Gene Technology Regulator, 2008). These facts coincide with the results presented in aloe vera basal shoots because as the weight of basal shoots increases, most of the morphometric variables of the plant also increase as well.

Conclusions

Initial weight of aloe vera basal shoots does affect the performance of morphometric variables related to plant growth. Common bean planting companion (CBCP) and

basal shoots with an initial weight from 251 to 350 g (HW) evidenced promising results for its implementation as a sustainable alternative based on magnitude, variance, and predictability of commercially important morphometric growth qualities. Basal shoots from 50 to 250 g (LWe and MW) presented a similar performance one year after planting, thus this study claims that using basal shoots within this weight ranges does not make any difference in the growth of aloe vera cultivation. Although planting companion systems improve the homogeneity in the growth of aloe vera crops compared with monoculture, new studies are needed in order to determinate the cost of each cropping system.

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