

Flowering and fruiting of *Ananas comosus* L. Merr. in two cultivation systems under subtropical conditions

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Pineapple production in the subtropics is limited by low temperatures. Frost affects the growth and development of the plants, being the main factor that compromises production. One way to mitigate this problem is the use of plastic covers (greenhouses), which could, however, negatively affect the productivity. We studied the flowering and fruiting of pineapple (*Ananas comosus* L. Merr.) in two cropping systems (greenhouse and field) in a subtropical region in the northeast of Argentina. Two experimental batches, with plastic covers and uncovered, divided into eight plots, were set up and phenological crop monitoring was performed. The experimental design was completely random. At the time of harvest, physical and chemical variables, such as length of fruit length with crown, fruit length without crown, equatorial diameter, density, firmness, "Brix and acidity values, were analysed to represent fruit quality. The results showed significant differences between the cultivation systems studied. Cultivation type had a marked effect on fruit quality; fruits produced in the greenhouse had a higher weight and size than those produced in the field; however, the chemical variables related to flavour did not differ significantly between the two cultivation types.

Keywords: field, fruit, greenhouse, pineapple, quality

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La producción de piña (*Ananas comosus L. Merr*) en el subtrópico encuentra como principal factor limitante a las bajas temperaturas, ya que afectan el crecimiento y desarrollo de la planta. Una posibilidad concreta para controlar esta dificultad es el uso de coberturas plásticas o invernáculos, el cual podrían incidir en el cultivo y su productividad. El objetivo de este trabajo fue evaluar el comportamiento de la floración y fructificación de la piña en dos sistemas de cultivo en regiones subtropicales del nordeste argentino. Se establecieron dos lotes experimentales, con cobertura plástica y sin cobertura, divididos cada uno en ocho parcelas. El diseño experimental fue completamente al azar. Se realizó el seguimiento fenológico del cultivo y para determinar la calidad del fruto se analizaron variables físicas y químicas: longitud del fruto con y sin corona, diámetro ecuatorial, densidad, firmeza, °Brix y acidez. Los resultados mostraron diferencias significativas entre los sistemas de cultivo estudiados, en donde el sistema de cultivo tuvo un marcado efecto a favor de las frutas producidas por plantas bajo cobertura plástica presentaron un peso y tamaño superior a las producidas sin cobertura; sin embargo las variables químicas relacionadas con el sabor en ambos sistemas no fueron afectadas significativamente.

Palabras clave: calidad, campo, fruta, invernáculo, piña

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INTRODUCTION

Pineapple (*Ananas comosus* L. Merr.) is cultivated in more than 60 tropical and subtropical countries. The fruits are in great demand in developed countries, and pineapple cultivation has a great economic importance. Globally, pineapple is the third most produced tropical fruit after banana and citrics (Botella & Smith, 2008). From an economic point of view, it is the most important species from the Bromeliaceae family. The "Smooth Cayenne" cultivar is the dominant fresh fruit for export (Loeillet et al. 2011) and the most widely grown cultivar in the world (Chan et al. 2003).

The pineapple flowering process involves a transition from the vegetative to the reproductive stage, when the differentiation of the flowers takes place and the vegetative growth stops (Py, 1969). To obtain a quality fruit and a uniform production, it is necessary to apply floral induction techniques to standardize fruiting and fruit ripening (Cunha, 1993; Maruthasalam et al., 2010). The environment has a considerable effect on the morphological characteristics of plants and even generates physiological modifications, which has also been observed for the Bromeliaceae (Martin, 1994).

The main limiting factor for the cultivation of pineapple in subtropical regions is temperature. Although the plant resists mild and short frosts down to -3°C, its growth is delayed by low temperatures and stops, depending on the cultivar, at temperatures between 10 and 16°C. According to Cunha (1999), the ideal temperature for the growth and development of roots and leaves is between 22 and 32°C; at predominantly low temperatures, growth is reduced.

In the South American subtropics, the conditions for pineapple cultivation are favourable; however, it is necessary to consider the possible occurrences of frost. This calls for the need to develop appropriate technologies to adjust the pineapple crops to the specific conditions of the region.

One possibility is the use of greenhouses; however, environmental conditions generated by the very presence of plastic could affect the growth and development of the plants as well as the phenological phases and the quality of the fruit.

Information on the phenology of pineapple is important for the distinction between growth stages, problem identification in the development of cultivation, adoption of agronomic practices and studies of the phases of the cycle and, especially, to guide the management of the crop to the optimum harvest moment (Kist, 2011).

In this context, the aim of this work was to study the flowering and fruiting of the pineapple (*Ananas comosus* L. Merr.) in two cropping systems, greenhouse and field conditions, in the Argentinian north-eastern subtropical region.

MATERIALS AND METHODS

The experiments were carried out between September 2013 and January 2015 at the Estación Experimental de la Facultad de Ciencias Agrarias de la Universidad Nacional del Nordeste, Corrientes – Argentina, 27° 28' 27" S., 58° 47' 00" W; 70 meters above sea level.

The soil at the experimental site has been classified as Udipsamentselven mixt, hyperthermic, belonging to the Ensenada Grande series. Based on its natural low fertility and susceptibility to erosion, it is located in Subclass II and III. The relief of the area is gently undulating, with slopes of 1 to 1.5%. The climate is subtropical or mesothermal, with an average annual rainfall of 1.300 mm and an average annual temperature of 21,6°C; it has a well-defined frost-free period of 340 to 360 days. Average annual temperature is around 21,5°C; the average temperature of the coldest month (July) ranges between 16 and 13°C and the average temperature of the warmest month (January) is between 27 and 26°C; annual variation is scarce. In summer, absolute maximum temperatures reach 42,5 to 46,5°C; although frost is rare, absolute minimum winter temperatures are -1 to 5,5°C (Escobar,

The rainfall pattern is characterised by abundant and frequent rainfall, exceeding 1.500 mm per year. The main feature of this system is the seasonal irregularity in rainfall distribution, with autumn being the rainiest and winter the driest season.

In this study, we used the pineapple cultivar "Smooth Cayenne". Two experimental batches (treatments), one under field conditions and other under plastic cover (greenhouse), were established and drip-irrigated. The experimental design was completely random. The plots consisted of two sowing beds with a density equivalent to 74.000 plants per hectare; bed distance from centre to centre was 1,80 m, with a length of 8,40 m. In each bed, there were four rows of plants, with a spacing of 30 cm between plants and rows; the two central rows were considered as useful plots (96 plants) to avoid edge effects.

The greenhouse was 9 m wide, 21 m long and 2,5 m high; maximum ceiling length was 4 m. It consisted of a metal structure with a front door; the plastic sheet was 150 µm thick.

Both cultivation systems were fertilised to maintain the ratio 1N: $0.5\ P_20_5$: 3 K_2O , applying a total of 150 kg ha nitrogen and 75 kg ha phosphorus in the form of P_20_5 and 450 kg ha; potassium was applied as K_2O . The doses were divided into three equal parts and applied at planting (September 2013), in summer (December 2013) and in late autumn (May 2014) in each year.

Floral induction consisted of the application of Etephon (2-cloroetilfosfonic acid) in Ethrel ® solution (13 mL), urea (40 g) and CaOH (11 g), dissolved in 20 L of water, pH 8,2–8,5. Each plant received 50 mL on two occasions at an interval of seven days. The measurements and determinations made as follows:

Following the phenological stages of the crop: in both cultivation systems and after the floral induction, weekly observations on all plants of each batch were made. Inflorescence display inside the rosette leaves was considered as the beginning of the flowering state. For each observation period, we calculated the degree days, following the methodology proposed by Carvalho et al. (2005):

Degree days (DD) = days observed (mean temperature -15.8°C), where we considered15.8°C as base temperature of the crop.

Fruits growth curves: measures of length and diameter (in cm) of each developing fruit were taken weakly.

Fruit quality analysis: at harvest time, when fruits showed 25% of yellow cover, physicochemical determinations were made from the half portions of 48 fruits per treatment that were chosen randomly, analysing the following parameters:

Size: fruit length with and without crown, superior, middle and lower diameters (in cm);

Fresh weight: the fruits of each treatment were weighed. Crown fresh weight was determined separately. Proportion between fresh weight and crown weight was also determined.

Number of single fruits or fruitlets: the number of individual fruits harvested was counted.

Fruit density: the density of the fruits was determined by the following relationship: fruit weight (in g) divided by the volume of water displaced by the fruit (g mL⁻¹).

Firmness: evaluated by measuring the force required to penetrate a 11-mm diameter probe to a depth of 1 cm, using a peeled fruit and a penetrometer (Kg cm⁻²).

Total soluble solids: the concentration of soluble solids was analysed using a manual refractometer (° Brix). Acidity: was determined potentiometrically by titrating

10 ml of pineapple juice filtrate with NaOH 0.1N until pH 8.1, expressed in grams of citric acid per litre of juice. In the experimental lots, we measured maximum and minimum temperatures and ambient relative humidity. In the greenhouse, measurements were taken with a digital thermohydrometer and in the field, data were

In the greenhouse, measurements were taken with a digital thermohygrometer and in the field, data were obtained from a meteorological station from 2013 to 2015.

Prior to comparing the measured variables, normality of the data (Shapiro-Wills test) and homogeneity of variance were tested. Analysis of variance (ANOVA) and mean comparisons were performed using Tukey's test (p <0.05). Statistical analyses were carried out using the software INFOSTAT (Di Rienzo et al., 2016).

RESULTS

Tables 1 and 2 show the phenological stages of pineapple plants in both systems; mean monthly temperatures, monthly relative humidity and degree days accumulated during the months of observation.

Table 1. Phenological tracking of pineapple plants under greenhouse conditions (Ph. stage): Phenological stage, (Aver.temp.): Average temperature, (Aug.): August, (Sept.): September, (Octob.): October, (Novem): November, (Decem.): December, (Floral Diff.): Floral differentiation, (Floral Initi.): Floral initiation.

Month	Ph. stage	Grade Days	Aver. Temp. (∘C)
May	Floral	151,9	25,6
June	-	125,1	24,1
July	-	99,2	22,2
Aug.	Floral Diff.	125,9	23,9
Sept.	Floral Init.	150,3	25,8
Octob.	Fruit Growth	192,5	28,2
Novem.	Fruit Growth	233,6	31,4
Decem.	Fruit Growth	174,8	31
Decem.	Harvest	1253,605 GD in total	-

Table 2. Phenological tracking of pineapple plants under field conditions. (Ph. stage): Phenological stage, (Aver.temp.): Average temperature, (Aug.): August, (Sept.): September, (Octob.): October, (Novem): November, (Decem.): December, (Jan.): January, (Floral Diff.): Floral differentiation, (Floral Initi.): Floral initiation.

Months	Ph. Stage	Grade Days	Aver. Temp. (°C)
May	Floral Induction	69,2	20,2
June	-	4,5	16,1
July	-	0	13,2
Aug.	-	21,5	17,1
Sept.	Floral Diff.	61,9	19,9
Octob.	Floral initi.	102,8	22,4
Novem.	Fruit growth	155,9	26,1
Decem.	Fruit growth	168,4	26,6
Decem.	Fruit growth	85,3	28,9
Jan.	Harvest	669,9 GD in total	-

Plants from both treatments had 570 days of cultivation from planting to the time they received the flowering inducer (May 2013). In plants cultivated in the greenhouse, floral differentiation occurred in the middle of August (90 days after induction), while field-cultivated plants showed floral differentiation 101 days after induction (Table 1). In the greenhouse, the average maximum temperatures recorded during this period were between 34 and 37°C, while in the field system, temperatures oscillated between 20 and 27°C until the beginning of flowering. Average minimum temperatures during this period in the greenhouse were also higher than those in the field (Table 3). In the greenhouse, there were 502° DD from floral induction to floral differentiation, whereas in the field treatment, there were only 95° DD (Tables 2 and 3).

The flowering in the plots cultivated in the greenhouse was homogeneous, since 80% of these plants flowered simultaneously. In contrast, in the open-field plots, the

flowering was more staggered, since only 50% of the plants flowered simultaneously. This behaviour was maintained in the later stages, so that the plots cultivated in the field presented a high heterogeneity in the occurrence of the phenological stages. In the field treatment, the relatively low temperatures prior to flowering and at the flowering stage delayed the following phenological stages (Table 1).

The total degree days accumulated in the period between floral differentiation and fruit growth were 877,25 and 569,05 for the greenhouse and field plants, respectively.

The degree days accumulated from floral induction to harvest time were 1253,60 for fruits obtained from greenhouse cultivation and 669,91 for field cultivars.

As can be seen in Figure 1a and b, the fruits obtained from greenhouse cultivation had a growth period of 68 days between October and December, whereas the growth period for fruits cultivated in the field was 82 days (from October to January).

Table 3. Climate data from January 2014 to January 2015. (Max.Field): Maximum temperature in the field, (Min.Field): Minimum temperature in the field, (Max.Greenh.): Maximum temperature in the greenhouse, (Min. Greenh.): Minimum temperature in the greenhouse.

	Temperature(∘C)				Humidity (%)			
Month	Max. Field	Min. Field	Max. Greenh.	Min. Greenh.	Max. Field	Min. Field	Max. Greenh.	Min. Greenh.
January	36,4	21,5	38,5	23,1	88,4	34,7	91,7	42,9
February	36,2	21,1	36,2	22,5	89,6	40,1	85,8	45,5
March	32,7	18,6	37,6	19,1	92,1	36,1	79,6	28,4
April	28,1	16,7	38,6	17,0	94,3	37,5	96,7	35,0
May	27,1	13,5	37,0	14,2	95,1	48,1	97,2	45,5
June	22,0	10,2	36,6	11,7	91,6	42,9	98,4	48,8
July	20,4	6,1	36,3	8,2	87,7	34,7	96,4	36,5
August	25,8	8,6	35,0	12,9	89,6	36,3	89,4	38,0
September	29,1	10,8	37,8	13,8	98,2	36,6	91,2	36,0
October	30,5	14,4	40,4	16,0	93,3	40,2	94,3	46,5
November	32,1	20,3	40,1	22,6	93,6	40,6	94,0	46,3
December	33,1	20,2	38,4	23,6	94,5	43,5	89,3	42,3
January	36,4	21,5	38,5	23,1	88,4	34,7	91,7	42,9

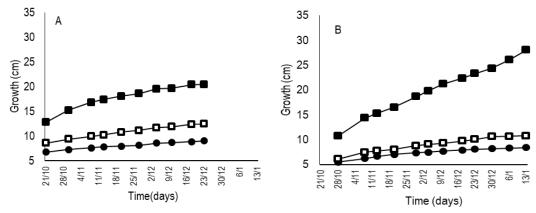


Figure 1. Growth curves of pineapples fruits cultivated under A: Greenhouse and B: Field conditions. (■) Length with crown, (□) Length without crown and (•) equatorial diameter.

The fruits cultivated in the greenhouse had initial values of 12,77 cm (length with crown), 8,5 cm (length without crown) and 6,49 cm (diameter) and final values of 20,36, 10,88 and 9,37 cm, respectively, which meant an increase of 60,22, 46,12 and 46,38%, respectively. However, for fruits cultivated in the field, the increase of growth was 177,91% for length with crown, 80,39% for length without crown and 58,58% for fruit diameter, with initial values of 10,73, 6,07 and 5,48 cm, respectively, and final values of 30,92, 10,26 and 8,92 cm, respectively.

Table 4 shows the final fruit size at harvest; the variables length of the fruit with and without crown in the two cropping systems were 20,63 cm and 10,88 cm and 30,92 cm 10,26 cm for the fruits produced in the greenhouse and in the field, respectively. Significant differences were only observed in the length of the fruit with the crown; at harvest, fruits cultivated in the field presented a higher final length than those grown in the greenhouse.

Under greenhouse conditions, the relation between the length of the crown and the length of the fruit was 0,89. In contrast, for fruits cultivated in the field, this ratio was 1,98, indicating a greater development of the crown in respect of the fruit. On the one hand, the crown weight/fruit weight ratio (Table 5) showed values of 13%, on the other hand, fruits obtained from the field system showed a ratio of 27%. Average crown weight differed significantly between greenhouse and field cultivation, with values of 96.81 and 191,68 g, respectively, and crown length values of 9,75 and 20,66 cm, respectively.

Fruit diameter in both culture systems was

characteristic of the variety (cylindrical form). No significant differences were found in basal and average diameter.

Fresh fruit weight and firmness were not significantly different between the two cultivation systems. The average fruit weight was 700,74 and 761,67 g for the field and greenhouse, respectively. A significant difference between the two systems was observed for fresh fruit weight without crown (Table 5). For fruits cultivated in the greenhouse, fresh fruit weight without crown was, on average, 662 g compared to 507,64 g for fruits cultivated in the field.

Fruit density values (Table 5) showed the particularity that although the fruits did not show differences in terms of total fresh weight, fruit density values were higher for fruits cultivated in the field, evidencing some differences in fruit composition. The lower amount of water in the fruits from field cultivation may be due to the higher temperatures recorded in this treatment and the low relative humidity, causing water loss.

Although the length of the fruit without crown and the equatorial diameter of the fruits from the greenhouse were not above the values of those from the field, the quantity of individual fruits was greater in the greenhouse cultivation than in the field cultivation.

Table 6 presents the results of the chemical analyses of fruit quality. The analysed variables did not present significant differences between the two cultivation systems. The values of total soluble solids (°Brix) oscillated between 12,99 and 14,31. Titratable acidity values were 0,49 to 0,58 g of citric acid 100 g⁻¹ pulp. The fruit ratio of both cropping systems ranged from 26,25 to 30,73.

Table 4. Pineapple fruit size measured at the harvest in the two cultivation systems. (G): Greenhouse, (F): Field. Different letters within the columns indicate significant differences (p<0,05).

Cultivation system	Fruit length with crown (cm)	Fruit length without crown (cm)	Crown length (cm)	Crown/fruit ratio	Superior diameter (cm)	Equatorial diameter (cm)	Inferior diameter (cm)
G	20,63 (a)	10,88 (a)	9,71 (a)	0,89 (a)	8,23 (b)	9,37 (a)	9,17 (a)
F	30,92 (b)	10,26 (a)	20,33 (b)	1,98(b)	7,63 (a)	8,92 (a)	8,67 (a)

Table 5. Physical parameters of pineapple fruits at the time of harvest in the two cultivation systems. (G): Greenhouse, (F): Field. Different letters within the columns indicate significant differences (p<0,05).

Cultivation system	Total fresh weight (g)	Fruit weight (g)	Crown weight (g)	Crown (%)	Density (g/ml)	Firmness (kg/cm ²)	N° of individual fruits
G	761,67 (a)	662 (b)	96,81 (a)	13 (a)	1,04 (a)	2,05 (a)	93,56 (b)
F	700,74 (a)	507,74 (a)	191,68 (b)	27 (b)	1,35 (b)	2,08 (a)	79,42 (a)

Table 6. Chemical parameters of pineapple fruits measured at the time of harvest in the two cultivation systems. (G): Greenhouse, (F): Field. Different letters within the columns indicate significant differences (p<0,05).

Cultivation system	°Brix	Acidity	Ratio
G	12,99 (a)	0,52 (a)	26,25 (a)
F	14,31 (a)	0,50 (a)	30,73 (a)

DISCUSSION

In both cultivation systems, the time between induction and floral differentiation was lower than that mentioned by Carvalho et al. (2005), who stated that in the same cultivar in the state of Paraná, Brazil, this period was 107 days. In relation to the degree day model, the conditions generated in the greenhouse reached the conditions of the tropical environments mentioned in Carvalho et al. (2005), where similar values were found for this period. In contrast, in the field treatment, the accumulation of degree days until flowering was below the values found by Carvalho et al. (2005) and Kist et al. (2011). The 11 days gap for floral differentiation between the two cropping systems indicates not only a response to the degree accumulation model, but also a relation to the low winter radiation, coinciding with the findings of Rainha et al. (2016).

There is a strong correlation between mean monthly temperatures and the response of the pineapple plants to the passage of the flowering stages, which proves that temperature is the variable responsible for the duration of the flowering cycle in subtropical regions (Rainha et al.2016).

The values accumulated in the period between floral differentiation and fruit growth were below those mentioned by Kist et al. (2011) in Mato Grosso, Brazil, who reported 1.349,5 degree days for this phenological period for the cultivar Cayenne Lisa, obtaining an earlier harvest in this tropical zone with a greater accumulation of degree days than in our zone.

The values calculated from floral induction to harvest time for the greenhouse cultivation are similar to those found for the same cultivar in the province of Paraná, Brazil, with 1.389,3 degree days in the same phenological period (Carvalho et al., 2005).

The fruit sizes obtained at harvest in both cropping systems were lower than the crop's potential, according to Pereira et al.(2009), who obtained fruit lengths with and without crown of 35,4 to 43,2 cm and 15,8 to 20,3 cm, respectively. This difference is due to the contribution of the crown to the fruit length, obtaining a relation of fruit length with crown/fruit length without crown of 1,89 in the greenhouse, whereas in those fruits cultivated in the field, this relation was of 3,01, which indicates a strong development of the crown under field conditions.

Montero Calderón (2005) states that the length of the crown should be 1 to 1,5 the length of the fruit. In our study, the fruits cultivated in the greenhouse showed a value of 0,89. The crown weight/fruit weight ratio (Table 5) showed values of 13% for the fruits grown in the greenhouse, which is in accordance with Rebolledo Martínez et al. (2006) for "Smooth Cayenne". The same authors also found that the crown weight/fruit weight ratio of this cultivar increases in favour of the crown at high planting densities.

Fresh weight and fruit firmness of fruits from both cropping systems were below the values expected for the Smooth Cayenne, which, according to Rebolledo Martínez et al. (2006), should range between 1.500 and 3.500g. In terms of fruit weight without crown, several authors indicate that a larger plant size at the time of floral induction allows the harvest of larger fruits (Hotegni et al., 2014). This result corresponds to the

fact that the plants grown in the greenhouse showed greater vegetative development at the moment of floral induction (Gonzalez Leguizamón et al., 2013), which agrees with the results of a study from Mexico, where the largest fruits were found in the plants with the highest leaf area index values (Rebolledo Martinez et al.,2006). This also corroborates the findings of Bartholomew and Malézieux (1994), who mention that the photosynthetic efficiency at the time of floral induction significantly influences the yield.

The significant differences between the two cropping systems in terms of fruit density indicate that the fruits from the field cultivation had a higher density and, consequently, a more compact texture (see also Wisdom et al., 2009) and less water compared to fruits grown under greenhouse conditions.

After flower induction, pineapple plants show an increase of the width of the apex,which bears the florets (Wee and Rao, 1979). The number of individual fruits is associated with the number of flowers or florets that were developed during flowering (Py, 1969, Hotegni et al., 2014). In our study, optimum environmental conditions in terms of temperature and humidity were found in the greenhouse, which resulted in a greater number of fruits compared to the field treatment. Besides, there is a high correlation between fruit weight and number of individual fruits for this variety (Rebolledo Martinez et al., 2006).

Internal fruit quality was within the ranges mentioned by Manica (2000), who cites values between 10.9 and 18.8° Brix. However, other authors state that this variety contains between 13.31-14 °Brix (Reinhardt and Medina. 1992: Rebolledo Martínez 2006). Titratable acidity values were as expected for this variety and similar to those reported by Manica (2000), with 0.49 to 0.58 g of citric acid 100 g⁻¹ pulp, while Reinhardt and Medina (1992) indicate values between 0.61 to 0.65 g of citric acid 100 g⁻¹ pulp. The fruit ratio values of fruits from both cropping systems ranged from 26.25 to 30.73 and are higher than those reported by Da Silva Berilli (2011), who calculated ratios between 19.12 and 28.46.

CONCLUSIONS

The floral differentiation in the greenhouse and field cropping systems under subtropical conditions did not respond to the degree accumulation model.

Cultivation systems affected the physical and fruit size variables, markedly affecting the crown/fruit ratio under field cultivation conditions.

Based on our analyses, the different environmental conditions in the greenhouse and in the field did not result in differences in fruit quality. However, they did affect the variables total fresh weight, fruit weight, density, number of individual fruits and fruit size.

The two cultivation conditions had no significant impact on the chemical fruit parameters. The quality of the fruits produced in both cropping systems was within the chemical standards of °Brix and acidity required by the market.

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