Biostimulants on Fruit Yield and Quality of Mango cv. Kent Grown in Semiarid

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Abstract

Mango (*Mangifera indica* L.) is the second most exported fruit of Brazil, which is especially grown in São Francisco Valley, a semi-arid region. One of the main cultivars exported is 'Kent' which has favorable physical-chemical characteristics, but irregular production and low fruit retention on the tree. Thus, the objective of the present study was to evaluate yield and postharvest quality of 'Kent' fruits as a function of foliar spray with biostimulants. The experiment followed a randomized complete block design with five treatments, four replications per treatment and four plants per replication evaluated in two consecutive years (2016 and 2017). The treatments consisted of foliar sprays with plant biostimulants, as follows: T1) Control (without biostimulants); T2) Biostimulant containing nutrients and L-α-amino acids; T3) Biostimulant containing nutrients and *Lithothamnium* algae extract; T4) Biostimulant containing nutrients and sucrose; and T5) Biostimulant containing nutrients, free amino acids and *Lithothamnium* algae extract. Biostimulants affected fruit yield and quality of 'Kent' fruits in distinct ways between seasons. The biostimulant T2 and T5, promoted the highest fruit yields, and maintained the postharvest quality of the fruits required by the European market.

Mango (*Mangifera indica* L.) is the second most exported fruit of Brazil, with 99 % of the national production coming from the Southeast and Northeast regions (IBGE, 2016). In the Northeast, the Pernambuco and Bahia states have 32,378 ha of mango trees, especially Petrolina and Juazeiro counties, located in São Francisco Valley, a semi-arid region (IBGE, 2016).

Among the cultivars grown in São Francisco Valley, 'Kent' has been outstanding in the international market, as it has a pleasant flavor, yellowish-green to purple-red color, high soluble solids concentration and reduced fiber content (Siddiq et al., 2017). Despite the favorable physical-chemical and sensory characteristics of the fruits, 'Kent' has irregular production due to difficult flowering management and low fruit retention (Ramírez and Davenport, 2010).

The fruit abscission in mango is a complex phenomenon, probably associated with the plant genotype (cultivar), competition for photoassimilates, lack of pollination and/or non-balanced fertilization, pests and diseases, high temperatures and nutritional and hormonal imbalances (Singh et al., 2005).

Biostimulant substances have been used in some studies to reduce fruit abscission and, consequently to increase mango fruit yield. Biostimulants have physiological effects similar to phytohormones, acting in physiological processes (Nardi et al., 2016).

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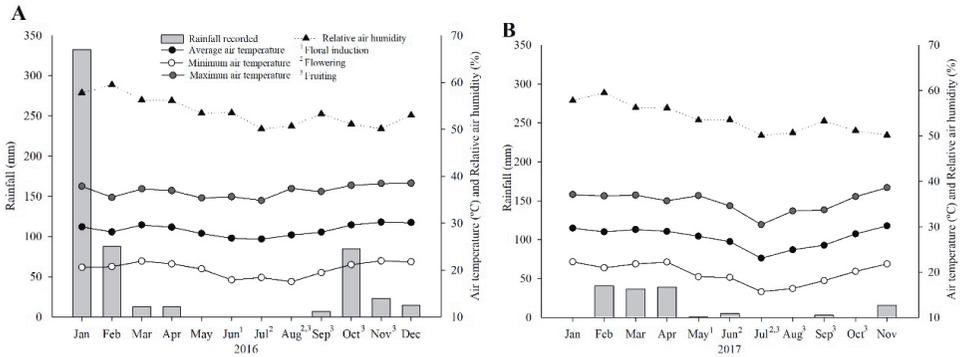* Corresponding author. E-mail: italo.cavalcante@univasf.edu.br

Fig. 1. Maximum, minimum and average air temperature, relative air humidity and rainfall recorded during the execution of the experiment. Petrolina, Pernambuco, Brazil (A − 2016; B − 2017).

Jarande et al. (2013) applied nutrients plus sucrose to 'Kesar', Khattab et al. (2016) studied the effect of a biostimulant containing calcium, boron and amino acids on different mango cultivars (but not 'Kent'), and Ahmed et al. (2015) demonstrated the benefits of algae extract associated with nitrogen fertilization on 'Taimour'.

Ebeed and Abd El-Migeed (2005) reported positive results on fruit characteristics and postharvest quality with the use of 10 % sucrose combined with 0.3 % potassium citrate for 'Fagri kalan' mango. However, the effects of biostimulants on the physical-chemical quality of mango fruits are scarce in the scientific literature, although they are crucial characteristics for the mango industry, since fruit postharvest quality depends on several factors such as plant genetics, climate and production practices performed in the orchard. In this sense, the objective of this study was to evaluate 'Kent' fruit yield and postharvest quality as a function of foliar spray with biostimulants.

Materials and Methods
Plant Material and Growing Conditions
Ten year-old 'Kent' mango (*Mangifera indica* L.) trees, with uniform size and vigor were used in this study. The experiment was performed in two consecutive years (2016 and 2017) in an experimental orchard located in Petrolina (09°09' S and 40º22' W; at an altitude of 365.5 m above sea level), Pernambuco, Brazil. The climate of this region is classified as Bsh (Köeppen), which corresponds to a semi-arid region.

During the experiment, climatic data were collected by a meteorological station (Fig. 1). Chemical characteristics of the soil before the experiment are in Table 1. The orchard nutritional status was also determined by leaf analysis before the experiment, as can be seen in Table 2. Leaves were collected in the middle part of the canopy, on 8 July 2016 and 10 June 2017. Leaves were chemically analyzed after they were washed and rinsed with distilled water and dried at 65 °C to a constant biomass following methodology

Table 1. Chemical soil characteristics (0-40 cm soil depth) in the experimental site before the experiment.

pH H₂O	O.M. g/100g	P mg/dm³	K⁺	Na⁺	Ca²⁺	Mg²⁺	Al³⁺	(H + Al)	S.B.	V %
			------------------- cmol$_c$/dm³ -------------------							
6.3	12	16	0.35	0.13	4.5	1.8	0	1.92	6.78	78

P, K and Na: Mehlich 1 (HCl + H₂SO₄); Ca, Mg and Al: KCl 1 M extractor. O.M.: organic matter; SB.: sum of bases; V: percent base saturation; mg: milligrams; cmol$_c$: centimole of charge; dm³: cubic decimeter.

Table 2. Leaf nutrient concentrations of mango cv. Kent before the treatments when leaves were sampled on 8 July 2016 and 10 June 2017.

Year	N	P	K	Ca	Mg	Mn	Fe	Zn	B
	------------ g/kg --------------					--------- mg/kg ---------			
2016	17.78	1.15	21.00	10.18	0.98	211.77	39.65	16.03	19.77
2017	13.49	1.54	8.90	15.60	1.50	171.78	41.28	15.08	97.65

N: Kjeldahl; P: spectrometry with yellow vanadate; K: Flame photometry; Mg, Ca, Fe, Zn and Mn: Spectrophotometry of atomic absorption; B: spectrometry with azometin-H

described by Malavolta et al. (1997).

The plants, spaced 5.0 m between the rows and 3.0 m between the plants, were daily irrigated (Micro sprinkler) with one emitter per plant, to provide about 60 L·h⁻¹ each, based on evapotranspiration registers recorded by a meteorological station and corrected according to the mango culture coefficient (Kc) defined by Genú and Pinto (2002). All management practices such as pruning, control of weeds, pests and diseases, plant growth regulator (Cultar®, Paclobutrazol) for gibberellin synthesis inhibition and dormancy break were performed following the instructions of Genú and Pinto (2002). The dose of Cultar® 250 SC (Syngenta Crop Protection, Paulinia, São Paulo, Brazil), equivalent to 2.25 g·m⁻¹ of active ingredient (a.i.) was applied each year at the rate of 9.0 mL per linear meter of tree canopy diameter diluted in 2 liters of water and applied as a soil drench once under the canopy dripline. Dormancy break included three foliar sprays with calcium nitrate (2.5%) at 90, 97 and 104 days after paclobutrazol application. Nutrients were applied through a fertirrigation system, according to plant demand (Genú and Pinto, 2002). Trees were manually pruned to synchronize vegetative flush events in the canopy.

Treatments and Experimental Design

The experiment was a randomized complete block design with five treatments, four replications per treatment and four plants per replication evaluated in two consecutive years (2016 and 2017). The treatments were defined considering the plant demands and physiological changes that occur during the pre-flowering and full flowering of mango in São Francisco Valley, properly described by Genú and Pinto (2002) and Cavalcante et al. (2016), which compositions are described in Table 3.

The treatments were applied in three phases: pre-flowering, beginning of flowering and full flowering (Fig. 2), according to the mango phenology described by Ramírez and Davenport (2010). The doses of each biostimulant followed the manufacturer recommendation but the total volume applied was 3.0 L of the mixture (biostimulant + water) per plant.

Evaluations and Statistical Analysis

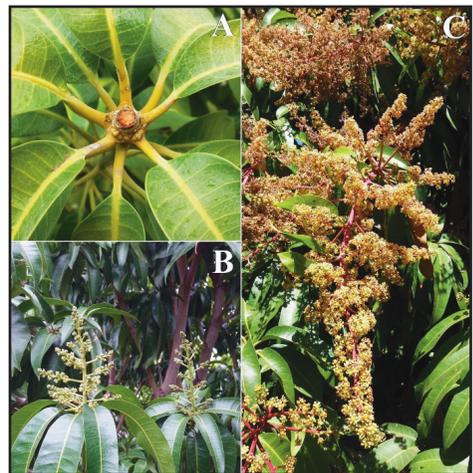Commercially mature fruits were manually harvested in a single day when they

Fig. 2. Plant stages when the treatments (biostimulants) were sprayed: pre-flowering on 10 June 2017 (A), beginning of flowering on 22 June 2017 (B), and full flowering on 12 July 2017 (C).

Table 3. Treatment description and composition of the biostimulants used in the experiment.

Treatment	Composition	Dose
T1	Without biostimulants spray	---
T2	Nitric nitrogen 10.0%, potassium oxide 5.0%, calcium oxide 7.15%, Magnesium oxide 1.2%, boron 0.1% and l-α amino acid 0.35%	3.0 mL·L^{-1}
T3	Organic nitrogen 5.0%, nitric nitrogen 1.2%, ammoniacal nitrogen 1.3% and amide nitrogen 2.5%. Raw materials: water, soybean pie, liquid sodium hydroxide, urea, ammonium nitrate, citric acid, *Lithothannium* seaweed extracts and sodium molybdate	1.0 mL·L^{-1}
T4	Calcium 8.0%, boron 2.0% and Sucrose	2.5 mL·L^{-1}
T5	Organic nitrogen 6.0%, Organic carbon of biologically origin 20.8% and total protein 31.0%. Raw materials: water, soybean pie, liquid sodium hydroxide, leonardite, *Lithothannium* seaweed extracts, citric acid, free amino acids	2.5 mL·L^{-1}

reached physiological maturity which was characterized by pulp color (yellow cream), following the fruit selection parameters recommended by the Brazilian Program for Horticulture Modernization (2004) for commercial farms. Production per plant was measured using a precision scale (0.5 grams of precision) for calculating fruit yield (t ha^{-1}).

After harvest, the fruits were stored for 23 days at 20°C until reaching the maturation stage 4 (Brazilian Program for Horticulture Modernization, 2004). Postharvest analyses were performed on 10 fruits per replication, as outlined by Zenebon et al. (2008) and included the usual parameters: length and width were measured with a digital paquimeter (0.01 mm–300 mm) and expressed in cm; fruit mass was measured using a precision balance (0.01 g precision) and expressed in g; fruit firmness (10^5·N/m^2) was measured using a fruit firmness tester on both fruit sides; soluble solids concentration (SS), expressed as percentage (%),

were measured with an Abbe® refractometer (Bausch and Lomb, Rochester, NY, USA); for the titratable acidity (TA), 20 g of macerated fruit pulp from a fruit was brought to a final volume of 100 mL by adding distilled water. A 20 mL sample was taken from the mixture, and three to four drops of phthalein were used as an indicator. This suspension was titrated with 0.1 N sodium hydroxide (NaOH). The results were expressed as g of citric acid/100 g pulp; SS/TA ratio; and number of fruits with the physiological disorder known as internal breakdown.

Statistical analyses included analysis of variance (ANOVA) by year and means were compared withTukey's test. All the calculations were performed using the ASSISTAT Statistical Program (ASSISTAT 7.7, UFCG, Campina Grande, Brazil) and terms were considered significant at $P \leq 0.05$.

Results and Discussion
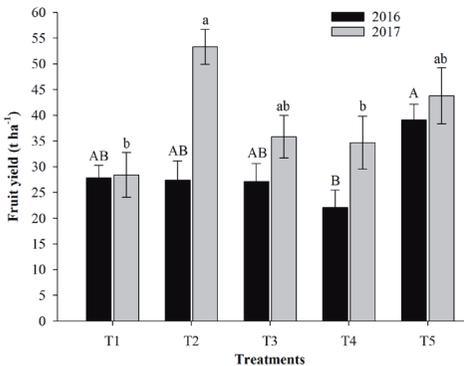
In the first season (2016), trees with T5

Fig. 3. Fruit yield of mango cv. Kent as influenced by biostimulants in two consecutive seasons (2016 and 2017) Bars with common capital letters (2016) and common lower case letters (2017) do not differ at the 5% level by Tukey's test. Error bars indicate standard error of the mean. Treatments description in Table 3.

had the highest yield (Fig. 3), 11.3 t ha^{-1} higher than the control treatment (T1). T4 reduced yield 21 % compared to T1, which was an unexpected result, considering the biostimulant composition. Non-treated plants (T1), plants sprayed with biostimulant containing water soluble nutrients and L-α-amino acids (T2), and biostimulant containing water soluble nutrients and *Lithothamnium* algae extract (T3) had similar yields (Fig. 3). In 2017, T2 had the highest yield and it was significantly greater than the control (T1). All treatments except the control produced higher yields in 2017 than in 2016. In the entire experiment the lowest fruit yield was 22 t ha^{-1}, recorded for T4 in 2016 and it was higher than the Brazilian average 16.1 t ha^{-1}, and even higher than yields reported for other countries, such as China (8.2 t ha^{-1}), India (7.3 t ha^{-1}), and Mexico (8.9 t ha^{-1}) (FAO, 2017), demonstrating the potential of mango trees in São Francisco Valley.

In 2016, only the width (FW) and fruit mass were affected by the treatments, while in 2017 all variables (fruit length, fruit width, fruit mass and fruit firmness) were affected by biostimulants (Fig. 4).

The average fruit length (FL) in 2016 was 108.74 mm (Fig. 4A). In 2017, the highest FL was recorded for T5 (125.48 mm), similar to T1, T3, and T4, but different from T2, and the difference between T5 and T2 was 7.38 mm (Fig. 4A).

Fruit width (FW) was affected by treatment in both 2016 and 2017 (Fig. 4B). In 2016 the biostimulants produced fruits with similar FW and an average value of 110.01 mm, 20 % higher than the control treatment (T1), which was only 91.47 mm. In 2017, the best treatments were T5 (110.34 mm) and T1 (106.95 mm), which was similar to the other treatments. Modesto et al. (2016) studied different mango cultivars and verified that for a given cultivar, fruit width can vary with the season due to the crop seasonality, plant's intrinsic factors, water availability, and temperature ranges. Plant biostimulants may be able to reduce these effects and standardize FW.

In 2017 FL and FW measurements were highest for T5 that is in agreement with Battacharyya (2015), who found similar results with the combination *Lithotamnium* seaweed extract and free amino acids. According to Aslam et al. (2010) the seaweed extract contains calcium, copper, manganese, zinc, iron, potassium, magnesium, and cobalt, essential nutrients for plant development that contributed to fruit growth.

The fruit mass was affected in both evaluation years (Fig. 4C). In 2016, T3 and T4 treatments were higher than the others, with averages of 543 and 533g, respectively; these averages are similar to 504 g (T5) and 449 g (T2), while fruits with lower mass were produced by the control treatment. In 2017, fruit mass was highest for T5 and it was 49 % higher than for trees treated with T3 (496.18 g), and 14 % higher than the control.

'Kent' fruits are traditionally produced with a focus on the export market, and therefore, the grade standards for those markets are considered for comparison purposes, especially the European Union, the main purchaser of Brazilian's mangoes
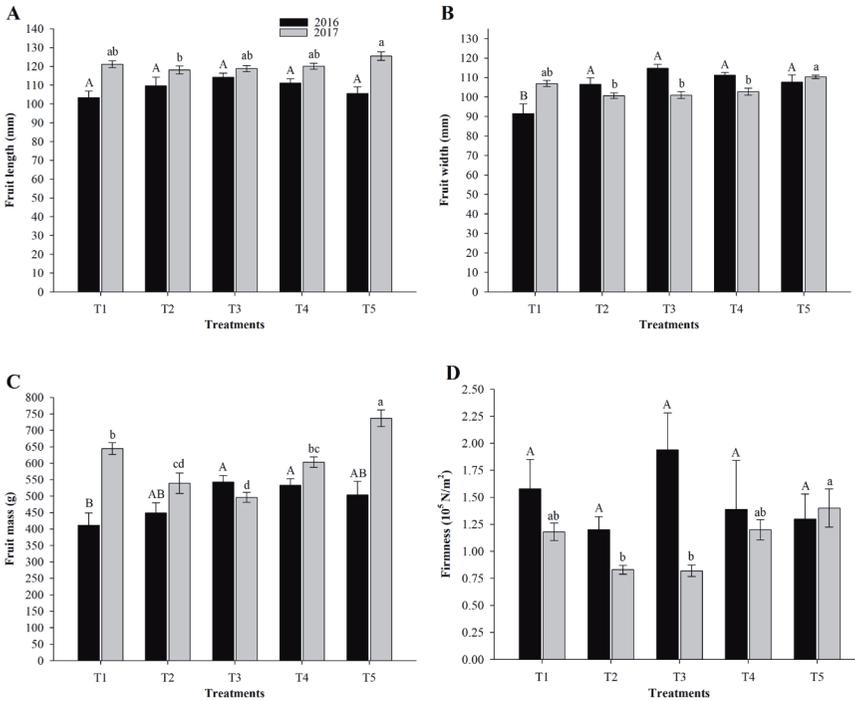
Fig. 4. Fruit length (A), width (B), fruit mass (C), and firmness of mango cv. Kent (D) as influenced by biostimulants in two consecutive seasons (2016 and 2017). Bars with common capital letters for 2016 and for lower case letters for 2017 do not differ at the 5% level, by Tukey's test. Error bars indicate standard error of the mean. Treatement descriptions in Table 3.

(Araújo and Garcia, 2012).

According to United Nations Economic Commission for Europe (UNECE, 2017) standard FFV-45 concerning marketing and commercial quality control of mangoes, the fruits for export must be at least 100 g and are classified by weight: size code A (100-350 g), size code B (351-550 g), size code C (551-800 g), and size code D (> 800 g). In 2016 all treatments produced 'size code B' fruits and in 2017 the fruits of T1, T4, and T5 were 'size code C', and T2 and T3 corresponded to 'size code B'.

According to the Normative Instruction of the Brazilian Department of Agriculture (Brazil, 2012), mango fruits are classified by caliber, which corresponds to the number of fruits in a commercial box. Traditionally 'Kent' is sold in boxes containing 4 kg of fruit; considering this criteria, the fruits produced in 2016 are classified as: T1 (10 caliber), T2 (9 caliber), T3 (7 caliber), T4 (7 caliber) and T5 (8 caliber); in 2017 the classification was:T1 (6 caliber), T2 (7 caliber), T3 (8 caliber), T4 (6 caliber) and T5 (5 caliber).

Exporting fruits for fresh consumption should be 6 to 10 calibers, while the 4 and 5 caliber fruits are usually commercialized for the fruit processing industry according to the classification adopted by fruit growers in the São Francisco Valley and exported to European Union.

European consumers prefer medium-sized fruits, with 8 and 9 calibers (Araújo and Garcia, 2012). Therefore, fruits produced by T2 and T5 in 2016 were suitable for the European market while in 2017, fruits from

T3 were the most desirable.

Fruit firmness was not affected by treatments in 2016 (Fig. 4D), with average values ranging between 1.20×10^5 and $1.94 \times 10^5 \cdot N/m^2$, and these fruits could be classified as 'soft ripe' according to Brecht scale (2017) that considers flesh firmness as an indication of fruit maturation. In the second harvest the T5 ($1.4 \times 10^5 \cdot N/m^2$) was firmer than T2 and T3 treatments, which were considered 'excessively mature' (flesh fruit firmness less than $0.89 \times 10^5 \cdot N/m^2$). T5 average was similar to T1 and T4 (1.18×10^5 and $1.20 \times 10^5 \cdot N/m^2$) and classified as 'soft ripe fruit' (Fig. 4D). Both classes recorded in 2016 and 2017 are recommended for fresh fruit consumption.

For the chemical fruit quality variables, treatments were only different for titratable acidity (TA) in 2016; and for TA, soluble solids (SS) and the SS/TA ratio in 2017 (Fig. 5).

In 2016, the SS were similar among all treatments (Fig. 5A), with an average of 18.38 %, value similar to those reported by Dick et al. (2009) (14.2 to 20 %) for 'Kent' fruits harvested at the same physiological stage of those evaluated in the present study. In 2017, the treatments affected SS and all values were lower than those recorded in 2016 (Fig. 5A). T1 had the highest SS (16.4 %), 53 % higher than the lowest value recorded for T3 (10.7 %), and the control was similar to the 16.8 % reported by Silva
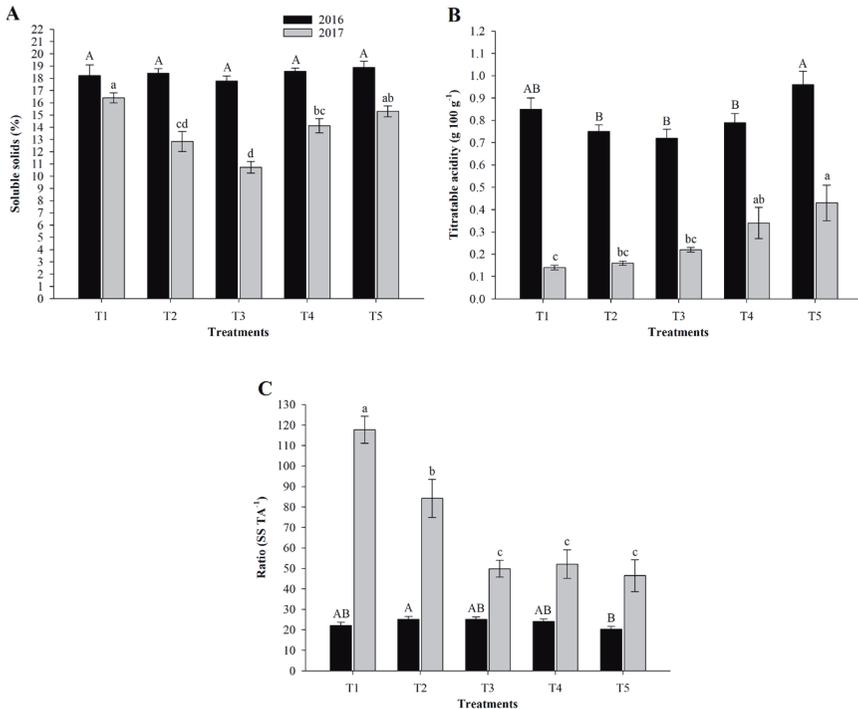

Fig. 5. Soluble solids (A), titratable acidity (B), and ratio (SS/AT) (C), of mango cv. Kent as influenced by biostimulants in two consecutive seasons (2016/2017). Bars with common capital letters for 2016 and lower case letter for 2017 do not differ at the 5% level, by Tukey's test. Error bars indicate standard error of the mean. Treatement descriptions in Table 3.

et al. (2012) for 'Kent' harvested at the ripe stage. In a survey of several mango farms in São Francisco Valley, 'Kent' averaged 12.2 % SS (Batista et al., 2015), showing that all treatments in 2016 and 2017 except for T3, had above average SS for fruits produced in this region.

Siller-Cepeda et al. (2009) studied characteristics of 'Kent' fruit following 12 days of storage at 20 °C. On day zero fruits had 4.1 % SS and firmness was $2.55 \times 10^5 \cdot N/m^2$, but after storage the values were 13.7 % SS and $18.1 \times 10^5 \cdot N/m^2$.

For all treatments, the fruit titratable acidity was higher in 2016 (Fig. 5B). These values are much higher than those reported by Silva et al. (2012) 0.27g of citric acid per 100g fruit. For good fruit palatability, low titratable acidity is desirable, which favors a high SS/TA ratio. Thus, in 2016 T2, T3, and T4 had the best SS/TA ratios because they had the lowest TA, while in 2017 the best values were produced by T1, T2, and T3.

The SS/TA ratio is an important fruit quality parameter and it is commonly used to determine the fruit palatability and maturation; the higher the SS/TA ratio, the better the flavor (Oldoni et al., 2018). In 2016 the average values were affected little by treatment and varied from 20.3 (T5) to 25.1 (T2), whereas in 2017 the control treatment (T1) had the highest SS/TA ratio (Fig. 5C). According to Costa et al. (2017), the relationship between SS and TA is often negative. In mango fruit, high SS/TA ratio is associated with the degradation of complex carbohydrates such as cellulose, pectins, and hemicellulose, to monosaccharides such as glucose and fructose (Tharanathan et al., 2006), which enhances fruit sweetness, but reduces the fruit firmness. Siller-Cepeda et al. (2009) evaluated 'Kent' fruits following 12 days of storage and reported a SS/TA ratio of 23.6, a value similar to that obtained in 2016. Silva et al. (2012) reported a SS/TA ratio of 62.2 for fruits harvested at the ripe stage, a value considered high but lower than fruit from T1 and T2 in 2017.

In both evaluation years, the incidence of the physiological disorder known as internal breakdown in fruits was not observed, despite the high nitrogen (17.78 and 13.49 g kg^{-1}), low calcium concentrations for the leaves (Table 2), and high N:Ca ratio (1.75 and 0.86, in 2016 and 2017, respectively).

The use of biostimulants affects fruit yield and quality of 'Kent' mango fruits, but results varied with season. When observing the striking differences in the climatic variables between the two seasons evaluated (Fig. 1), as well as in the biostimulants responses in these years, it is perceived that the biostimulant effect occurs in a differentiated way in relation to the prevailing climate during the application period.

The biostimulant containing water-soluble nutrients and L-α-amino acids (T2) promoted high fruit yields one year while the biostimulant containing water-soluble nutrients, free amino acids, and *Lithothamnium* seaweed extract (T5) promoted high yields in both years studied. Fruit size and fruit quality after storage of T2 and T5 plants, also met the standards required by the European market in both studied years.

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