
**EFFECTS OF PACLOBUTRAZOL AND KNO₃ OVER FLOWERING
AND FRUIT QUALITY IN TWO CULTIVARS OF MANGO MANILA**

Andrés Rebolledo-Martínez, Ana Lid del Ángel-Pérez and José Rey Moreno

SUMMARY

Different doses of Paclobutrazol (PBZ) combined with KNO₃ were evaluated in order to determine their effect on flowering and fruit quality in two mango clones. The experiment was carried out at the INIFAP's Cotaxtla Experimental Station in Veracruz, Mexico. A 14-year old orchard of Manila Cotaxtla 1 and Manila Cotaxtla 2 clones planted in 8×8m plots without irrigation was used. The experimental design was of random blocks in split plots. The eight treatments applied were: a) 0.0, b) 0.5, c) 1.0, d) 1.5, e) 2.0g a.i. PBZ per meter of canopy diameter, all combined with the application of 2% KNO₃, f) 4% KNO₃ without PBZ, g) 1.5g. a.i. PBZ, without KNO₃, and h) control with neither product added. PBZ was applied on the soil and KNO₃

in two applications on the foliage. The variables considered were number of panicles, fruit weight, fruit weight loss, total soluble solids (TSS), titratable acidity (TA) and firmness. PBZ and KNO₃ application induced a larger number of advanced panicles in relation to the control, with higher values observed as PBZ dose increased. However, in total flowering there was no significant difference between treatments ($p < 0.05$). PBZ did not have negative effects in the quality of fruit of neither cultivar. The treatments with higher doses of PBZ had higher content of TSS, less TA, less firmness and greater weight loss. Both cultivars were different in TA of the ripe fruits.

Introduction

Mexico produces 1.5×10^6 tons of fresh mango, placing the country in the fourth place worldwide after India, China and Thailand, which produce 10.8, 3.6 and 1.7×10^6 tons, respectively (FAO, 2005). This illustrates the

relevance of this crop for the Mexican economy. There are about 175000ha planted in Mexico and some 25000 of them are located in the Veracruz state, representing 14% of the national total and making this state the second largest mango producer in the country, even though most of the state's crop is made up of

the Manila cultivar (SAGARPA, 2005).

One of the main problems that mango producers in Mexico face is that the cropping season is too short because 70% of the fruit is harvested in May and June. Consequently, fruit prices suddenly drop and determine a low profitability. One strategy followed

is to produce off season by using growth regulators such as Paclobutrazol (PBZ; IUPAC name: (2RS, 3RS)-1-(4-chlorophenyl)-4,4-dimethyl-2-(1H-1,2,4-triazol-1-yl) pentan-3-ol), in the group of triazols. This compound induces an early and intense flowering, diminishing vegetative growth and reducing the extension of buds

KEYWORDS / *Mangifera indica* L. / Growth Regulation / Flowering Induction / Forced Flowering / PBZ /

Received: 11/07/2006. Modified: 06/02/2008. Accepted: 06/04/2008.

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EFECTO DEL PACLOBUTRAZOL Y KNO₃ SOBRE LA FLORACIÓN Y CALIDAD DE FRUTO EN DOS CULTIVARES DE MANGO MANILA

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RESUMEN

Se evaluaron diferentes dosis de paclobutrazol (PBZ) combinadas con KNO₃ para determinar su efecto sobre floración y calidad de fruta en los clones de mango Manila Cotaxtla-1 y Manila Cotaxtla-2. El experimento fue realizado en el Campo Experimental Cotaxtla del INIFAP, en Veracruz, México. Ambos clones, de 14 años de edad, fueron plantados a 8x8m, sin riego. El diseño experimental utilizado fue de bloques al azar en parcelas divididas. Se aplicaron ocho tratamientos: a) 0,0; b) 0,5; c) 1,0; d) 1,5; y e) 2,0g i.a. por metro de diámetro de copa de PBZ, todos junto con aplicación de KNO₃ al 2%; f) 4% KNO₃ sin PBZ; g) 1,5g i.a. PBZ sin KNO₃; y h) testigo sin PBZ ni KNO₃. El PBZ fue aplicado al suelo y el KNO₃ al follaje en

dos aplicaciones. Las variables consideradas fueron número de panículas, peso de fruto, pérdida de peso del fruto, sólidos solubles totales (SST), acidez titulable (AT) y firmeza. La aplicación de PBZ y KNO₃ promovió mayor número de panículas adelantadas con relación al testigo, con mayores valores al incrementar la dosis de PBZ; aunque en floración total no hubo diferencia significativa entre tratamientos ($p < 0,05$). Los niveles de PBZ no tuvieron efecto negativo en la calidad de fruto en ambos cultivares. A mayor dosis de PBZ, hubo mayor contenido de SST, menor AT, menor firmeza y mayor pérdida de peso. Los cultivares mostraron diferencias en AT de los frutos maduros para consumo.

EFEITO DO PACLOBUTRAZOL E KNO₃ SOBRE A FLORAÇÃO E QUALIDADE DE FRUTO EM DOIS CULTIVARES DE MANGA MANILA

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RESUMO

Avaliaram-se diferentes doses de paclobutrazol (PBZ) combinadas com KNO₃ para determinar seu efeito sobre floração e qualidade de fruta nos clones de manga Manila Cotaxtla-1 e Manila Cotaxtla-2. O experimento foi realizado no Campo Experimental Cotaxtla do INIFAP, em Veracruz, México. Ambos os clones, de 14 anos de idade, foram plantados a 8x8m, sem regar. O desenho experimental utilizado foi de blocos aleatórios em parcelas divididas. Aplicaram-se oito tratamentos: a) 0,0; b) 0,5; c) 1,0; d) 1,5; e) 2,0g i.a. por metro de diâmetro de copa de PBZ, todos junto com aplicação de KNO₃ a 2%; f) 4% KNO₃ sem PBZ; g) 1,5g i.a. PBZ sem KNO₃; e h) testemunha sem PBZ nem KNO₃. O PBZ foi aplicado ao solo e o KNO₃

na folhagem em duas aplicações. As variáveis consideradas foram; número de panículas, peso do fruto, perda de peso do fruto, sólidos solúveis totais (SST), acidez titulável (AT) e firmeza. A aplicação de PBZ e KNO₃ promoveu maior número de panículas adelantadas em relação à testemunha, com maiores valores ao incrementar a dose de PBZ; embora em floração total não houvesse diferença significativa entre tratamentos ($p < 0,05$). Os níveis de PBZ não tiveram efeito negativo na qualidade do fruto em ambos os cultivares. Com maior dose de PBZ, houve maior conteúdo de SST, menor AT, menor firmeza e maior perda de peso. Os cultivares mostraram diferenças em AT dos frutos maduros para consumo.

(Burondkar y Gunjate, 1991; Litz, 1997; Singh, 1998), allowing for ripening and the initiation of apical buds inflorescence (Pérez *et al.*, 2000).

Oosthuysen and Jacobs (1997) tested the effect of two PBZ concentrations (0.25g i.a. and 2.5g i.a.) on fruit retention and size, as well as yield and profitability per mango tree of the cultivars Sensation and Tommy Atkins. They found a differential response between cultivars to PBZ, 'Tommy Atkins' having a greater reduction in buds growth than 'Sensation'. Furthermore, the number of developing inflorescences increased. On the contrary, there was a reduction in size, length between shoots, diameter growth and leaf size, but there was an

increased yield per tree.

Sergent *et al.* (1997) found that the application of PBZ at doses of 1.0 and 1.5g i.a. per meter of canopy diameter increased production, allowing for regular crops in two consecutive production cycles. PBZ doses in 'Tommy Atkins' mango had a direct effect on fruit weight (less weight at higher doses), yield and new budding. Moreover, the onset of flowering was also affected by the doses of the product (Oosthuysen and Jacobs, 1997).

In Nayarit, México, early flowering was not observed in 'Tommy Atkins' mango as a result of spraying 2% ammonium nitrate (NH₄NO₃), but the use of PBZ at a dose of 20g per tree induced

flowering 28 to 35 days earlier; furthermore, vegetative growth was suppressed during summer and fall, resulting in flower bud initiation and early flowering (Pérez *et al.*, 2000). NH₄NO₃ as well as ethrel were ineffective in stimulating flowering in mango 'Tommy Atkins' (Pérez *et al.*, 2000). In Veracruz, Mexico, PBZ was applied in July at 1g a.i. per meter of canopy diameter combined with two foliar sprays of KNO₃ at 2% in October, and the flowering of mango 'Manila' took place 80 days ahead from the regular natural flowering; resulting in a selling price 15 times higher, as compared to the regular crop value for the season (Rebolledo and Del Ángel, 2004).

Mosqueda (1989) reported that KNO₃ was effective in stimulating the emergence of mango inflorescences more than 30 days in advance. Its application before normal flowering increased by 50% the number of buds showing inflorescence, as well as the yield when compared to the control group. Salazar *et al.* (2000) advanced by 22 and 44 days, respectively, the harvest of mango 'Ataulfo' and 'Manila', as a result of applying NH₄NO₃; however, there was no flowering response in mango 'Tommy Atkins'. Oosthuysen and Jacobs (1997) tested one and two applications of KNO₃ at 2 and 4% on mango 'Tommy Atkins', 'Kent', 'Heidi', 'Sensation', 'Irwin' and 'Keitt'. All cultivars showed a

slight reduction in fruit weight associated to an increase in fruit retention and increasing yield in all the cultivars. There were no significant differences in fruit quality.

Most studies on mango forced production only considered monoembryonic cultivars, while research on polyembryonic materials such as 'Manila' mango is scarce. The present study aims to evaluate the effect of PBZ, applied either alone or combined with KNO₃, on flowering, yield, and fruit quality of two clones of mango under the rainfed conditions of an agrosystem in central Veracruz, Mexico.

Material and Methods

Experimental site and vegetative material

The experiment was carried out at the Cotaxtla Research Station of the Instituto Nacional de Investigaciones Forestales, Agrícolas y Pecuarias (INIFAP) located in the Municipality of Medellín, Veracruz, Mexico, at 18°16'N and 96°16'W, 40masl. The soil is clayish (Vertisol Pelico). A 14 year-old orchard composed of the Manila 'Cotaxtla 1' and 'Cotaxtla 2' mango clones trees, grafted on mango Manila and planted every 8x8m was used, maintained under rainfed conditions and managed according to the recommendations given by Mosqueda *et al.* (1996). These clones were obtained in 1992 from a selection of 36 manila mango trees from Veracruz State originally planted in 1957. Of the total mango production in Veracruz 90% is manila cultivar (De los Santos y Mosqueda, 1992).

Experimental design and treatments

The experimental design was that of unbalanced random blocks in split plots. There were five blocks for the selection 'Cotaxtla 1' and four for 'Cotaxtla 2'. A large plot was used for selection and a small plot for growth regula-

tor (doses of PBZ and KNO₃) application. The mango tree was considered as the experimental unit. The treatments were: a) 0.0, b) 0.5, c) 1.0, d) 1.5, e) 2.0g a.i. of PBZ per meter of canopy diameter, all combined with the application of 2% KNO₃ (20g.l⁻¹), f) 4% KNO₃ (40g.l⁻¹) without PBZ, g) 1.5g a.i. PBZ, without KNO₃, and h) control with neither compound added. The PBZ solutions were applied on the soil in four parts distributed under the canopy and the KNO₃ to the foliage in two applications.

Application of treatments

PBZ diluted in 2 liters of water was applied to soil in the dripping area of four ditches 10x40x20cm each on July 30, 2003, according to Subhadarabandu *et al.* (1999), Protacio *et al.* (2000), Shinde *et al.* (2000), Singh (2000) and Singh and Ram (2000). A foliar spray of KNO₃ was applied on October 22 and November 2, as described by Mosqueda, *et al.*, 1996.

Variables

Counting flower panicles was done on Dec 7, when the first floral panicles in anthesis were observed, and again

at the end of flowering, on Jan 29, when 100% of panicles showed anthesis. The tree canopy was divided into four longitudinal parts and counting was carried out on each of them. Fruit quality was evaluated by its physiological ripeness using two fruits per experimental unit. Three ripe fruits per tree were harvested and left for 8 days at room temperature (19 ±3°C) in a well ventilated and shaded place until the fruit reached its optimal point for human consumption. Weight loss was determined using a digital scale on three fruits from every experimental unit. Total soluble solids (TSS) were determined using a digital refractometer (Serie Palette, ATAGO, Japan) from a 2ml juice sample obtained from both faces of each fruit. Titratable acidity (TA) was obtained by NaOH 0.1N (AOAC, 1980), and firmness (F) was measured by applying a universal penetrometer on both sides of two mango fruits after sectioning the fruit epicarp with a knife.

Statistical analysis

In order to homogenize variances the data were transformed using the root square $\sqrt{(Y_i + 1)}$. Statistical analysis and comparisons of means

(Tukey) were performed using SAS (SAS Institute, Cary, NC, USA).

Results and Discussion

Flowering

There were no significant differences by clone in the number of panicles (P=0.28) and their interactions. However, treatments in the small plot were significantly different (P≤0.05) only in the first counting, performed on Dec 7 (Table I). Results show that PBZ treatments combined with KNO₃ were statistically different from the others, but not among them; that is, low doses exhibited the same levels of early flowering than high doses, with percentages ranging between 5 and 14% of the total of panicles; but higher percentages occurred at higher doses. Neither PBZ applied alone nor KNO₃ alone had a significant effect on flowering. The total number of panicles at the end of the period of floral organogenesis was not affected by treatments (Table I). PBZ had a significant effect on early flowering when combined with KNO₃. Conversely, PBZ used alone did not have any effect.

In addition, there was a tendency in both clones to-

TABLE I
NUMBER OF PANICLES AT THE BEGINNING AND END OF NATURAL FLOWERING IN TWO CLONES OF 'MANILA' MANGO TREATED WITH DIFFERENT DOSES OF PBZ

Treatments	Number of panicles					
	7/12/03			29/01/04		
	Clon 1 ^y	Clon 2 ^x	Media	Clon 1 ^y	Clon 2 ^x	Media
0.0g i.a. PBZ + 20g.l ⁻¹ KNO ₃	53.7 abc ^z	26.8 a	40.2 ab	1438.3 a	1155.1 a	1690.0 a
0.5g i.a. PBZ + 20g.l ⁻¹ KNO ₃	100.7 ab	30.2 a	65.5 ab	1337.0 a	1647.9 a	1296.7 a
1.0g i.a. PBZ + 20g.l ⁻¹ KNO ₃	100.1 ab	88.8 a	94.5 ab	1467.2 a	1351.6 a	1492.5 a
1.5g i.a. PBZ + 20g.l ⁻¹ KNO ₃	149.8 a	152.6 a	151.2 a	1040.8 a	1327.7 a	1409.4 a
2.0g i.a. PBZ + 20g.l ⁻¹ KNO ₃	202.8 a	142.7 a	172.8 a	1261.8 a	1273.6 a	1184.3 a
0.0g i.a. PBZ + 40g.l ⁻¹ KNO ₃	12.4 bc	24.6 a	18.5 bc	1563.9 a	1176.0 a	1267.7 a
1.5g i.a. PBZ + 0g.l ⁻¹ KNO ₃	7.0 c	3.8 a	5.4 c	1284.2 a	1539.9 a	1370.0 a
Control	0.2 c	0.0 a	0.1 c	1504.1 a	1875.9 a	1412.0 a
CV (%)	44	78	58	11	18	5
Clon	ns ^y	ns ^y	ns	ns	ns	ns
Doses PBZ-KNO ₃	**	ns	**	ns	ns	ns
Interaction	ns	ns	ns	ns	ns	ns

^y Manila Cotaxtla 1, ^x Manila Cotaxtla 2, ^z values with the same letter by column are equal by Tukey test at P≤0.05, ns: not significant, *: significant at P≤0.05, **: significant at P≤0.01.

wards an earlier flowering with the higher doses of PBZ and KNO₃, which coincides with the findings of Pérez *et al.* (2000), Burondkar and Gunjate (1991) and Cárdenas and Rojas (2003). Medina and Núñez (1997) indicated that for mango 'Haden' and 'Tommy Atkins', the combination of PBZ with sprayed ammonium nitrate was the most effective treatment for an early production of panicles, compared to their application alone. Even though PBZ induced a higher number of panicles at the onset of flowering, the total number was not greater in those treatments with PBZ, as pointed out by Oosthuysen and Jacobs (1997), but differing from Burondkar and Gunjate (1991) and Cárdenas and Rojas (2003), who obtained a higher number of panicles after treating with PBZ. The fact that the control group was similar to PBZ treatments might have resulted from the fact that the trees were on an "on" period of alternance, and because of having more reserves after having spent a year "off", they had more mature vegetative buds, thus differentiating more vegetative buds to floral ones.

Fruit weight and weight loss

There were no significant differences between selections ($P=0.09$), the factors under study and their interactions ($P\leq 0.05$). The same occurred for weight loss; that was, PBZ application did not have an effect on these variables, even under rainfed conditions (Figure 1).

This lack of significant differences in fruit weight among treatments was also found by Singh and Dhillon (1992) for the cultivar 'Dusheri' and by Oosthuysen, (1997) in 'Tommy Atkins' and 'Heidi', but not in the cultivar 'Alphonso' by Burondkar and Gunjate (1991). Medina and Núñez (1997) for 'Tommy Atkins' and Oosthuysen (1997) for 'Kent' reported that as PBZ dose increases fruit weight decreases; they interpret this fact as due to fruit overcrowd-

ing resulting in more competition for the available resources. Contradicting results have been reported by Oosthuysen and Jacobs (1997) for mango 'Sensation', since fruit weight increased as PBZ concentration did; however, for 'Tommy Atkins' the opposite effect was found.

Weight loss findings in this study are not similar to those in apple by Khurshid *et al.* (1997), who found a positive relationship between an increase in PBZ dose and weight loss. Conversely, the control and a higher KNO₃ treatment (40g·l⁻¹) showed lower weight losses, meaning that probably PBZ exerted some influence on the permeability of pericarp cells. It is worth mentioning that values obtained at 2.0g a.i. PBZ showed less weight loss in 'Manila Cotaxtla 1' than in 'Manila Cotaxtla 2' pointing to a differential response between both clones to such a treatment.

Total soluble solids

There were no significant differences in TSS of physiologically ripe fruits among clones, PBZ doses or interactions, although a positive trend ($P=0.06$) in the relationship in PBZ and KNO₃ doses and TSS increase was observed, indicating that both products increase the fruit total sugar content when applied together, but not when applied alone (Figure 2a).

TSS in ripe fruits for consumption increased as a result of the ripening process. Performance was similar to that obtained for physiological ripeness, since there were no statistical differences among clones, doses ($P=0.16$) and interactions ($P=0.3$). Likewise, the two highest doses of PBZ and KNO₃ exhibited fruits with numerically greater TSS content, even though not statistically different from the others (Figure 2b).

When a condition of physiological ripeness exists, TSS content shows a trend to be slightly greater under PBZ and

KNO₃ treatments (Singh and Dhillon, 1992), with values ranging from 12 to 14°Bx. In general terms, fruit ripeness is coupled with an increase in TSS content. However, Oosthuysen (1997) did not find changes in TSS content, pH, and flavor of ripe fruits of 'Tommy Atkins', 'Heidi', and 'Kent' cultivars treated with KNO₃. In this study, TSS content of ripe fruits for consumption ranged from 15.5 to 17.5°Bx, about 4°Bx more than for the physiological ripeness condition. The observed trend of a greater TSS content in the treatments with PBZ and KNO₃ uphold the hypothesis that PBZ and KNO₃ treatments lead to an accumulation of a greater amount of reserve substances.

Titrateable acidity (TA)

TA analysis of physiologically mature fruits did not result in significant differences between clones or in their interactions. However, a difference ($P=0.052$) in doses was detected. Individual doses of PBZ and KNO₃ resulted in higher citric acid content in the fruits. Conversely, the two products combined determined a decrease in the acidity of the fruits (Figure 2c). A significant difference ($P=0.01$) between the clones was detected in TA of fruits ripe for consumption, even though none was found for PBZ doses and the interactions. 'Manila Cotaxtla 1' showed a statistically greater concentration of citric acid, which is indicative

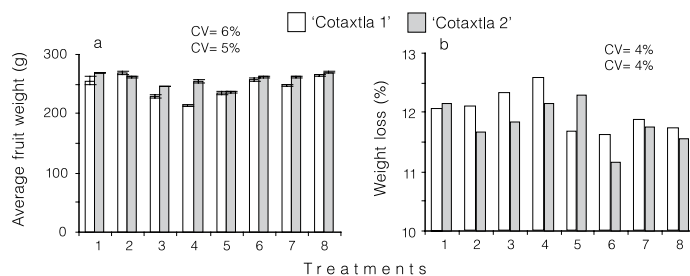


Figure 1. a: weight of fruits in physiological ripeness and, b: weight loss for each mango clone with the following doses of PBZ and KNO₃. 1: 0.0g a.i. PBZ + 20g·l⁻¹ KNO₃, 2: 0.5g a.i. PBZ + 20g·l⁻¹ KNO₃, 3: 1.0g a.i. PBZ + 20g·l⁻¹ KNO₃, 4: 1.5g a.i. PBZ + 20g·l⁻¹ KNO₃, 5: 2.0g a.i. PBZ + 20g·l⁻¹ KNO₃, 6: 0.0g a.i. PBZ + 40g·l⁻¹ KNO₃, 7: 1.5g a.i. PBZ + 0g·l⁻¹ KNO₃, 8: control. There were no statistical differences between treatments and clones ($P\leq 0.05$).

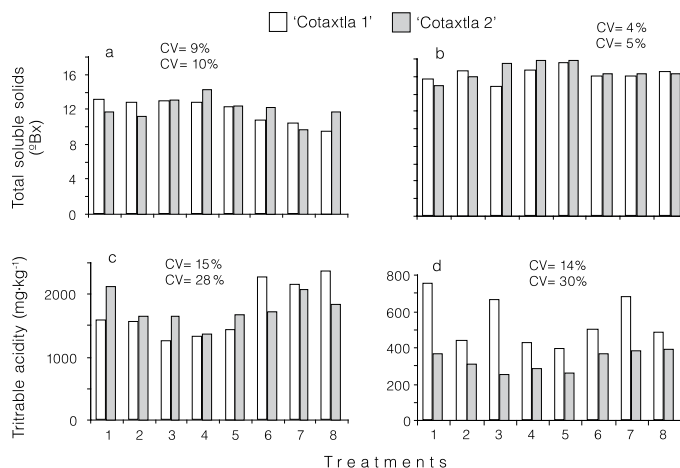


Figure 2. a: TSS at physiological ripeness, b: TSS at ripeness for consumption, c: AT at physiological ripeness, and d: AT at ripeness for consumption, for each of the mango clones with the following doses of PBZ and KNO₃. 1: 0.0g a.i. PBZ + 20g·l⁻¹ KNO₃, 2: 0.5g a.i. PBZ + 20g·l⁻¹ KNO₃, 3: 1.0g a.i. PBZ + 20g·l⁻¹ KNO₃, 4: 1.5g a.i. PBZ + 20g·l⁻¹ KNO₃, 5: 2.0g a.i. PBZ + 20g·l⁻¹ KNO₃, 6: 0.0g a.i. PBZ + 40g·l⁻¹ KNO₃, 7: 1.5g a.i. PBZ + 0g·l⁻¹ KNO₃, 8: control. There were no statistical differences between treatments and clones ($P\leq 0.05$) except for d, where there were different between clones ($P\leq 0.01$).

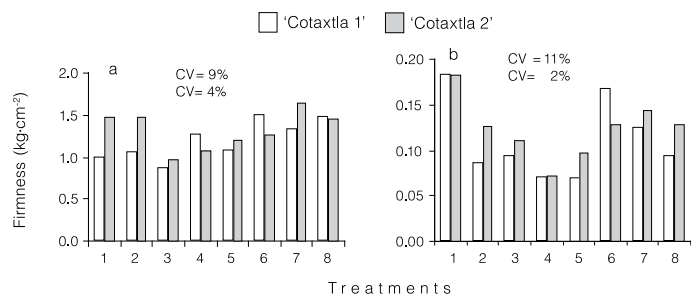


Figure 3. a: texture of fruit at physiological ripeness, and b: texture of fruit at ripeness for consumption for mango clones with the following doses of PBZ and KNO₃, 1: 0.0g a.i. PBZ + 20g·l⁻¹ KNO₃, 2: 0.5g a.i. PBZ + 20g·l⁻¹ KNO₃, 3: 1.0g a.i. PBZ + 20g·l⁻¹ KNO₃, 4: 1.5g a.i. PBZ + 20g·l⁻¹ KNO₃, 5: 2.0g a.i. PBZ + 20g·l⁻¹ KNO₃, 6: 0.0g a.i. PBZ + 40g·l⁻¹ KNO₃, 7: 1.5g a.i. PBZ + 0g·l⁻¹ KNO₃, 8: control. There were no statistical differences between treatments and clones (P≤0.05).

of a slower ripening process resulting from fewer of these acids being consumed during respiration (Figure 2d).

For the fruits at physiological ripeness, PBZ and KNO₃ treatments showed the lowest amount of organic acids, with 1200 to 1600mg·kg⁻¹ of citric monohydrated acid.

Acidity content of fruits ripe for consumption markedly diminished as a result of the ripening process, with fruits treated with PBZ and KNO₃ showing the lowest values. However, Singh and Dhillon (1992) reported that PBZ treatments increased the acidity content of fruits in comparison with the control group. It is noteworthy that 'Manila Cotaxtla 1' was statistically different from 'Manila Cotaxtla 2', showing a higher content of citric acid in all the treatments.

Firmness

There were no significant differences in the firmness of physiologically mature fruits due to clones, PBZ doses (P=0.17) or interactions (Figure 3a). In fruits ripe for consumption, something similar happened because neither clone, nor PBZ doses nor interactions showed significant differences. In fruits ripe for consumption, the loss of firmness related to physiological ripeness was >50% in all the treatments.

As PBZ doses increased a trend (P=0.13) was observed toward less fruit firmness (Fig-

ure 3b), which also seemed to be associated with those treatments exhibiting the highest TSS content, the highest weight loss and the lowest TA, and hence, less substrate available for respiration. Khurshid *et al.* (1997) found an increase in the firmness of apples treated with PBZ. On the other hand, treatments based only on KNO₃ (20 and 40g·l⁻¹) were noteworthy for inducing fruit firmness in both periods of evaluation.

Conclusions

The application of PBZ and KNO₃ allowed flowering 51 days before the regular flowering time of non-treated plants and the highest doses of PBZ and KNO₃ employed produced a greater number of advanced panicles. In contrast, PBZ individually applied had no effect. The only difference between de 'Manila' clones 'Cotaxtla 1' and 'Cotaxtla 2' was in the titratable acidity of ripe fruits. Moreover, PBZ levels assayed had no negative effect on the fruit quality of either clone, even though at higher PBZ doses there was less TSS and TA content, less firmness and a greater weight loss. Results show that the application of this product is possible under natural rainfed conditions, without leading to negative effects in fruit quality. Furthermore, fruits are produced according to the approved standards, allowing produc-

ers to obtain better prices for their crops.

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