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Effects of Forchlorofenuron (CPPU) treatment on fruit properties in the fruit of common guava

ABSTRACT

The common guava is a small tree from *Myrtacaeae* family which is cultivated for its fruits. Researches have explored new methods to promote fruit yields and quality of crops. Application of Forchlorofenuron or CPPU (with Molecular Formula: $C_{12}H_{10}CIN_3O$) improves the fruit size as well as its quality, but there has not been an investigation evaluating its effects on common guava fruit under field conditions. This research was performed to study the effects of different doses of CPPU (0, 10, 20 and 40 mg L^{-1}) on common guava fruit size and quality characteristics under field conditions. Analysis of variance and LSD (least significant differences) mean compression indicated that total soluble solids, total acidity, ascorbic acid or vitamin C, fruit firmness, phenolics, 1,1-Diphenyl-2-picrylhydrazyl (DPPH), polygalacturonase, pectin methyl esterase and ethylene were significantly different in most traits and CPPU-40 produced high means. The principal components (PC) analysis explained 95% of the total variation and the first two principal components (PC1 and PC2) explained 78% and 17% of the total variation, respectively. According to biplot, CPPU-40 had the highest values for all of the measured traits except DPPH, ethylene and polygalacturonase. The most prominent relations by biplot were a strong positive correlation among phenolics, fruit firmness, total acidity, total soluble solids and ascorbic acid as indicated by the small obtuse angles between their vectors. The measured traits were grouped into two clusters and cutoff point verified via Wilks' lambda statistics. Cluster I consisted of three traits (ascorbic acid or vitamin C, fruit firmness and ethylene) while cluster II included total soluble solids. Findings of this study suggest that CPPU can be used as an effective growth regulator to improve the size and quality of common guava fruit.

Keywords: biochemical characteristics, fruit firmness, pectin methyl esterase

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INTRODUCTION

The common guava (*Psidium guajava* L.) is a small tree cultivated for its fruits and is native to the Central and South America, especially Brazil. It belongs to the family *Myrtacaeae*. Its medicinal effects have encouraged many researchers into investigation of its extract properties including antibacterial activity. Various parts of plant including stems and leaves have been used as liquor which can be obtained after slicing the plant and boiling it with lime juice or infused in water for several days. The common guava fruit is highly perishable and should be handled carefully during harvest and transportation while its processed by-products, leaves and fruits can be used to feed livestock.

The common guava can vary depending on many factors including physiological process and cultural practices. So it is possible to interfere with these factors by external application of growth regulators. The use of plant growth regulators is one way for stimulating cell division or enlargement, both of which promote final fruit size. Among the bio-regulators, CPPU has been the most effective one in increasing fruit properties. CPPU is a member of the synthetic cytokinin group with phenyl urea structure, with strong inhibitory effects on cytokinin oxidation (4). Despite the large number of reports on the effect of CPPU on production and fruit characteristics of many fruit crops, little is known about its influence on fruit properties of the common guava. The CPPU is an effective and well known plant growth regulator to improve the fruit size through stimulating cell division (3). Several investigations have been conducted on the potential roles and influences of the synthetic substituted phenylureas thidiazuron and CPPU on growth and ripping of different fruit trees.

Sugiyama and Yamaki (9) investigated the effect of CPPU on Japanese persimmon and found that the number of fruits after thinning did not differ among treatments while the yield of marketable fruits after hand-pollination was similar to that of after treatment. Antognozzi et al. (1) sprayed CPPU (20 ppm) on fruits of *Actinidia deliciosa* after full bloom which influenced the fruit growth soon after treatment; during fruit growth, soluble sugars were higher in treated fruits compared with the control ones and this shows that CPPU induced faster fruit ripening. Mousawinejad et al. (5) studied the effects of CPPU on tomato fruit and showed its significant effects on fruit mass, volume, density, length and width while its effects on the fruit biochemical characteristics such as sugar, titratable acids and vitamin C contents were not significant.

The aim of this study was to study the effects of CPPU application on fruit properties of the common guava during growth and ripening and understand how CPPU-induced modifications in fruit metabolism has an overall importance in improving the different aspects of fruit growth and quality in common guava.

MATERIALS AND METHODS

The experimental site was a uniform block of 15-year-old common guava trees located in Fekejur, Iran. Trees were planted in a light clay soil and irrigated by a permanent set of under-tree sprinklers. At the balloon stage of flower development, the trees were randomly assigned to chemical treatments (0, 10, 20 and 40 mg L^{-1}) at early fruit formation (1 cm diameter). Each experimental unit consisted of 20 uniform fruits which were transferred to the laboratory. Data were recorded according to randomized complete block design with four replications. For all treatments in each replication, three fruits were randomly selected to measure total soluble solids (TSS), total acidity (TA), ascorbic acid or vitamin C (Asc), fruit firmness (Firm), Phenolics, 1,1-Diphenyl-2-picrylhydrazyl (DPPH), polygalacturonase (PG), pectin methyl esterase (PME) and ethylene (C_2H_2).

The data were first tested for normality by Anderson and Darling normality test. Data from each trial were subjected to analysis of variance (ANOVA) using appropriate model. Comparisons of means were performed using least significant differences (LSD) test at 0.05 significance level.

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Principal components (PC) analysis was used (2) and plots of the first two principal components were drawn as this routine statistical analysis has been used by many authors (7). The variance was explained by principal components communality (variance explained by each parameter and correlation coefficients between parameters and components were determined) (8). Cluster analysis was conducted on the basis of dissimilarity measure as Euclidean distance and the clustering method was UPGMC (unweighted pair group method using centroids). The number of clusters was determined using multivariate ANOVA via Wilks' lambda statistics. The experimental data were statistically analyzed using Minitab 16 software.

RESULTS AND DISCUSSION

Mean values of the measured traits and their comparisons obtained by LSD method are presented in Table 1. According to the total soluble solids (TSS), CPPU-40 treatment had the highest TSS, while control treatment (CPPU-0) had the lowest TSS, and it ranged from 13.53 to 14.75 with an average of 14.15 (Table 1). Total acidity (TA) ranged from 2.32 to 2.83 with an average of 2.56, for which treatment control (CPPU-0) had the lowest value, while CPPU-40 treatment induced the highest TA (Table 1). The lowest ascorbic acid (Asc) was reported from control treatment (CPPU-0), while the highest Asc content was observed from CPPU-40 and CPPU-20 treatments (Table 1). In terms of fruit firmness (Firm), CPPU-40 resulted in the maximum Firm (69.8), while control treatment (CPPU-0) led to the minimum Firm (55.0). Phenolics ranged from 0.85 to 1.25 for which control treatment (CPPU-0) exhibited the highest Phenolics, while CPPU-40 induced the lowest Phenolics. Considering 1,1-Diphenyl-2-picrylhydrazyl (DPPH), CPPU-20 treatment had the maximum DPPH (0.63), whereas control treatment (CPPU-0) resulted in the minimum DPPH (0.59). In terms of the polygalacturonase (PG), CPPU-40 treatment showed the highest PG (1.02), while control treatment (CPPU-0) ended in the highest PG (1.12). CPPU-40 treatment resulted in the highest pectin methyl esterase (PME), while CPPU-10 showed the lowest PME, and it ranged from 2.89 to 3.05 with an average of 2.98 (Table 1). Ethylene (C_2H_2) ranged from 64.48 to 67.98 with an average of 65.53, for which control treatment (CPPU-0) had the highest C_2H_2, while CPPU-40 treatment had the lowest C_2H_2 (Table 1). In general, for most measured traits, CPPU-40 treatment showed high values and could be regarded as the most favourable treatment. Mousawinejad et al. (5) investigated the effects of CPPU on tomato fruit under field condition and found that fruit mass, volume, density, length and width were statistically affected by 20 mg L^{-1} concentration of CPPU. They suggested that CPPU application can be used as an effective growth regulator for improving the size and quality of tomato fruit.

The dataset principal components (PC) analysis explained 95% of the total variation. This high percentage reflects the simplicity of the relationships between the measured traits. The first two principal components (PC1 and PC2) explained

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Table 1. Mean comparison of four CPPU treatment via LSD procedure in common guava

Treat	TSS		TA		Asc		Firm		Phen		DPPH		PG		PME		C2H4	
CPPU-0	13.53	D	2.32	D	64.2	C	55.0	D	0.85	D	0.59	D	1.12	A	3.01	B	67.98	A
CPPU-10	13.95	C	2.45	C	66.9	B	59.8	C	0.90	C	0.61	C	1.08	C	2.89	D	64.88	B
CPPU-20	14.38	B	2.63	B	78.5	A	61.8	B	1.01	B	0.63	A	1.09	B	2.95	C	64.78	C
CPPU-40	14.75	A	2.83	A	79.1	A	69.8	A	1.25	A	0.62	B	1.02	D	3.05	A	64.48	D

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78% and 17% of the total variation, respectively (Table 2). The third principal component (PC3) explained only 5% of the total variation and so could be ignored from analysis. In the first principal component (PC1), some traits including total soluble solids (TSS), total acidity (TA), ascorbic acid or vitamin C (Asc), fruit firmness (Firm) and Phenolics, 1,1-Diphenyl-2-picrylhydrazyl (DPPH) gained more scores; while in the second principal component (PC2), pectin methyl esterase (PME) and ethylene (C_2H_2) showed high scores. polygalacturonase (PG) did not show any important role in PC analysis (Table 2). From communality aspect, only fruit firmness, phenolics and PME were detected as the most influencing traits in this dataset.

Table 2. Principal components (PC) loadings of common guava traits obtained for CPPU treatments

Variable	PC1	PC2	PC3	Comm.*
TSS	0.38	-0.03	-0.07	0.28
TA	0.38	0.06	-0.05	0.39
Asc	0.35	-0.05	-0.53	-0.23
Firm	0.37	0.10	0.24	0.71
Phenolics	0.36	0.24	0.08	0.68
DPPH	0.31	-0.40	-0.43	-0.52
PG	-0.34	-0.13	-0.57	-1.04
PME	0.13	0.75	-0.27	0.61
C2H4	-0.31	0.42	-0.27	-0.16
Eigenvalue	7.02	1.51	0.47	
Proportion	0.78	0.17	0.05	
Cumulative	0.78	0.95	1.00	

*Communality

In the biplot, a vector is drawn from the biplot origin to each marker of the treatment to facilitate the visualization of the relationships between the traits as well as treatments. According to Yan and Rajcan (11) and Rubio et al. (6), the basic structure among the traits can be captured by biplots. The vertex treatments in this investigation included all of the studied treatments (CPPU-0, CPPU-10, CPPU-20 and CPPU-40). These treatments were the best or the unsuitable

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treatments in some or all of the traits since they had the longest distance from the origin of biplot (Fig. 1). Therefore, it seems that CPPU-40 had the highest values for all of the measured traits except DPPH, C_2H_2 and PG (Fig. 1). The vertex treatment control (CPPU-0) was suitable for C_2H_2 and PG while the vertex treatment CPPU-20 was suitable for DPPH. The other vertex treatment (CPPU-10) was not suitable for the measured traits (Fig. 1). The vertex treatments in different seven sections of biplot were completely different (10) for the common guava production using these treatments.

The most prominent relations by this figure are: a strong positive correlation among phenolics, fruit firmness, total acidity, total soluble solids and ascorbic acid as indicated by the small obtuse angles between their vectors (r = cos 0 = +1). Also, a relatively moderate or weak positive correlation was observed between the mentioned traits with pectin methyl esterase as well as DPPH (Fig. 1). Such positive correlation was seen between C_2H_2 and polygalacturonase acid indicated by the small obtuse angles between their vectors (r = cos 0 = +1). The correlation between pectin methyl esterase and C_2H_2 as well as PME and DPPH was near zero (Fig. 1) as indicated by the near perpendicular vectors (r = cos 90 = 0). Also, the association between pectin methyl esterase and polygalacturonase was zero (Fig. 1). There was a negative correlation between DPPH and C_2H_2, and polyga-

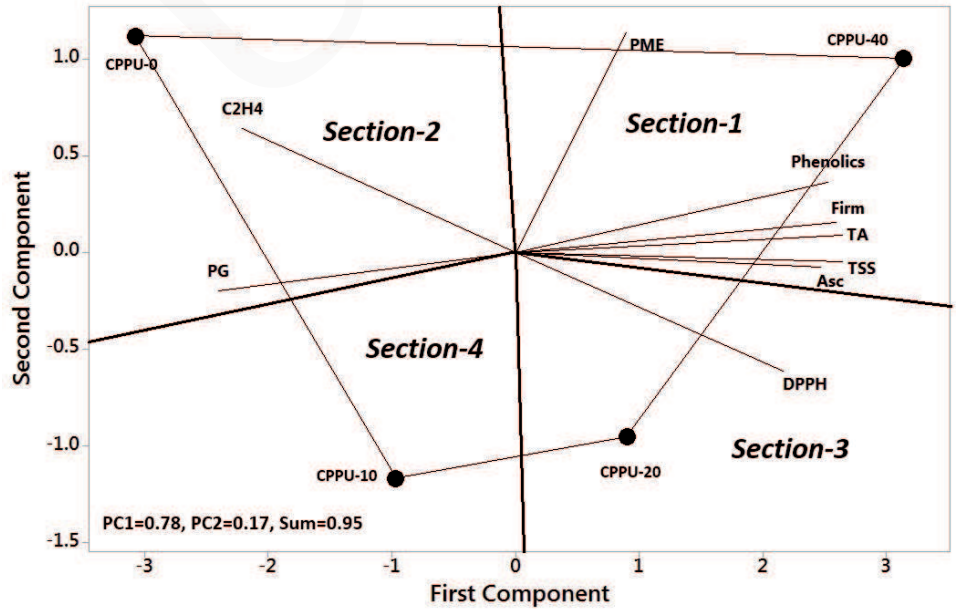

Fig. 1. Biplot of first two principal components showing relation of various common guava traits across four CPPU treatments.

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lacturonase and DPPH (Fig. 1) as their angles were approximately 180 degrees (r = cos 180 = -1). Such negative association was observed between polygalacturonase with phenolics, fruit firmness, total acidity, total soluble solids and ascorbic acid (Fig. 1). Most of above discrepancies of the biplot predictions and original data were expected, because the biplot accounted for high degree of total variation (95%). In minor cases, some of these descriptions can't be verified using original correlation coefficients, as the biplot accounted for <100% of the total variation.

The measured traits were grouped into two clusters based on UPGMC and cutoff point which is verified via Wilks' lambda statistics (Fig. 2). Cluster I involved three traits (ascorbic acid or vitamin C, fruit firmness, and ethylene) while cluster II included total soluble solids (TSS), total acidity (TA), Phenolics of phenols, 1,1-Diphenyl-2-picrylhydrazyl (DPPH), polygalacturonase (PG) and pectin methyl esterase (PME). Multivariate statistical methods, such as the PC analysis and clustering method showed the traits groupings as the best. In our analysis, we were able to determine clusters of accessions that were significantly different from each other for traits of interest.

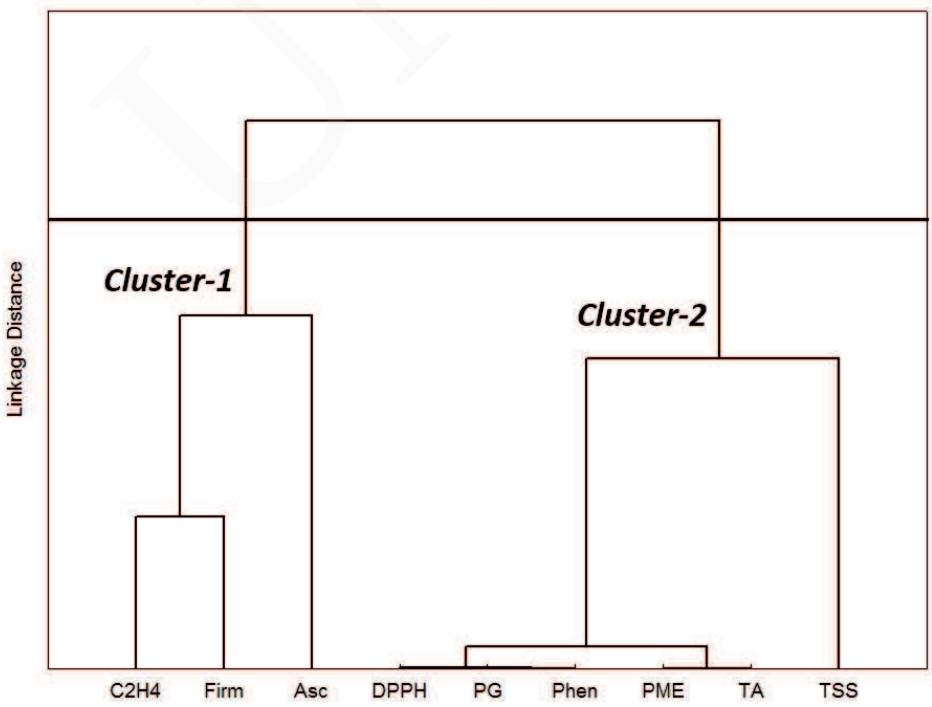

Fig. 2. Dendrogram of common guava traits at four CPPU treatments.

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CONCLUSION

This investigation demonstrated that CPPU-40 treatment had high potential of most of the traits. This treatment could be used as a commercial way to obtain new proper characteristics in the common guava. Also the biplot had some benefits including its easy use and interpretation, as well as the possibility to obtain other useful information and visualization pattern to identify suitable treatments.

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