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CHARACTERIZATION AND CLUSTERING OF SOME GUAVA GERMPLASM COLLECTIONS BASED ON LEAF AND FRUIT CHARACTERS

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ABSTRACT

Guava has several different accessions. Guava diversity needs to be studied and evaluated in order to determine the next steps in the guava breeding. The objective of this research was to characterize and cluster some guava germplasm collections. The study was conducted at Aripan and Subang experimental farm, Indonesian Tropical Fruit Research Institute from January 2012 to December 2012. Five fruits of each accession were randomly selected, sampled, and then characterized using UPOV guidelines. Obtained data were analyzed by NTSYS ver.2.1. The Similarity level of 19 accessions ranging from 70 – 90% or the genetic distance was between 0-20%. Dendogram obtained could be clustered into two different groups, namely group I (ARP9406, ARP9407, ARP8653, ARP8742, ARP10.2, JBT001, JBT002, ARP8740, JBT003andJBT004) and group II (ARP10.7, ARP10.6, ARP10.1, ARP10.12, ARP10.9, ARP10.11, ARP8744, ARP8741 and ARP8743). The result of this research can be used for guava breeding. Species diversity and genetic resources are very important to produce new varieties. This is expected to be highly valuable in the future.

Keywords: clustering, characterization, guava germ- plasm

INTRODUCTION

Guava (*Psidium guajava* L.) is an important tropical fruit crops. Guava has long been known to be used by the community for healing. The plant has been extensively studied in terms of pharmacological activity of its major components, and the result indicated potent anti-diarrheal, antihypertensive, hepatoprotective, antioxidant, antimicrobial, hypoglycemic and anti-mutagenic activities (Joseph, 2011). Owen *et al.* (2008) reported that the consumption of guava (*Psidium guajava* L.) and noni (*Morinda citrifolia* L.) may protect betel quid-chewing Papua New Guineans against diabetes. Guava leaves also have anti-oxidant (Daud *et al.,* 2011).

Guava has several different accessions so we need some activities to characterize them. Guava diversity needs to be studied and evaluated in order to determine the next steps in the guava breeding. Information about description and genetic distance are needed to get new hybrid. Species diversity and genetic resources are very important to get new varieties.

Morphological characterization is the easiest activity to be done because it is simple, inexpensive and useful to determine the relatedness between accessions. One of morphological characters easy to be observed is fruit. Fruit is noticeable without special tools. Fruit, an important component of production, is usually different in shape, weight and color.

The relatedness and genetic distance between accessions can be obtained through cluster analysis. Cluster analysis is grouping accessions which have the same characteristics in homogeneous categories of each stratum (Crossa *et al.,* 1995a, 1995b). The relatedness analysis based on morphological characters will be perfect when using the descriptions of the characters that have high heritability values and stability (Beer *et al*., 1993, Lamadji, 1998). Besides the morphological characterization, to determine the related-

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ness between accessions can also be done with biotechnology. Some countries such as Cuba have used microsatellites to characterize the guava (Infante *et al.* 2007). In India, to see the genetic diversity of guava derived from somatic embryo-genesis, SSR and ISSR markers are used (Rai *et al.,* 2012). Liu and Yang (2012) used ISSR markers to assess clonal fidelity of micro-propagated guava (*Psidium guajava* L.) plants.

The aim of the research was to characterize and accession the relatedness or grouping of guava. This research is expected to be used for selection of parents to get new superior varieties. This study is expected to be highly valuable in the future.

Table 1. Accessions list of guava germplasm collection at Aripan and Subang experimental farm

NO	Accession	Location
1	ARP9409	Aripan experimental farm
2	ARP9407	Aripan experimental farm
3	ARP8653	Aripan experimental farm
4	ARP10.2	Aripan experimental farm
5	ARP10.7	Aripan experimental farm
6	ARP10.6	Aripan experimental farm
7	ARP10.1	Aripan experimental farm
8	ARP10.12	Aripan experimental farm
9	ARP10.9	Aripan experimental farm
10	ARP10.11	Aripan experimental farm
11	ARP8740	Aripan experimental farm
12	ARP8742	Aripan experimental farm
13	ARP8744	Aripan experimental farm
14	ARP8741	Aripan experimental farm
15	ARP8743	Aripan experimental farm
16	JBT001	Subang experimental farm
17	JBT002	Subang experimental farm
18	JBT003	Subang experimental farm
19	JBT004	Subang experimental farm

MATERIALS AND METHODS

The study was conducted at Aripan and Subang experimental farm from January 2012 to December 2012. The plant materials were 19 germplasm accessions of guava (Table 1). Each accession was characterized in 5 pieces. The characterization was made based on qualitative and quantitative characters. Guidelines of UPOV (1987) were also applied. Qualitative characters include leaves (fully developed leaf, leaf curvature in cross section, leaf twisting, leaf curvature of midrib, leaf shape of base, leaf shape of tip, color of upper leaf, color of under leaf), and fruit (fruit shape, fruit shape at stalk end, fruit width of neck in relation to that of fruit, color of fruit skin, color of flesh). Quantitative characters cover leaf length (cm), leaf width (cm), fruit weight (g), fruit width (cm), fruit length (cm), petiole length (cm), diameter core (cm), flesh thickness (cm), total soluble solid (TSS) (° brix).

To make dendogram, results of quantitative data and qualitative characterization were grouped by category/ class and were converted into binary form. Binary data were analyzed with the program NTSYS version 2.1 with the final result of dendogram.

RESULTS AND DISCUSSION

Morphological characterization results showed that 19 accessions of germplasm collections guava in experimental farm at Aripan and Subang have morphological diversity (Appendix 1).

Leaf shape of some plants areobtrullate, obovate, oblong, and ovate. There was no difference in color of upper and lower surface of the leaf. In general, leaf is green with 11.96 – 15.02 cm in length and 5.39 to 10.34 cm in width. According to Mani *et al.* (2011), leaf shape of some *Psidium* species in India was ovate, lanceolate, oblong, elliptical to oblong and oval. Leaf length was about 4.2 - 13.3 cm, and 1.5 - 7.4 cm in width.

Fruit shape from 19 accessions observed had difference (Figure 1).There are round oval, oval, rounded, symmetry, rounded inverted cone, and ovate.

What becomes the main concern among consumers is fruit weight. Fruit size is a character which serves as the first attribute considered by consumers. In papaya, Chan *et al.* (1992) reported that papaya of a medium size is able to boost the market. Now the market preference is for small to medium size papayas because one fruit is enough for one person. In this research, the average of fruit weight ranged from 106.95 - 300.91 grams. Accessions with small weight involved ARP9406,

ARP9407, ARP8653, ARP10.2, ARP10.11, ARP8742, JBT001, JBT002, and JBT003, while the accessions with heavy weight comprised ARP10.8, ARP8741, and ARP8743. In Taiwan, there are some cultivars of guava. Taiwanese guava cv. has weights about 400-700 grams in average. Century guava cv. weighs about 200-400 grams. Pearl guava cv. weighs about 200-400 grams (Anonymous, 2011).

The color of fruit flesh varied for some accessions. Some accessions had red color, yellow, red purple, white, orange red. Red guava fruit has high content of vitamin C and beta carotene efficacious as anti-oxidant (Astawan 2013). However white guava has one special quality. Adnyana *et al.* (2004) stated that the ethanol extracts of white guava leaves showed stronger antibacterial activity than that of ethanol extracts of red guava leaves against *Escherichia coli*, *Shigella dysenteria*, *Shigella flexneri* and *Salmonella typhi*.

Total soluble solid (TSS) as a measurement tool was used to measure the level of sweetness. In this study, the values of TSS ranged from 7.8 – 11.03º brix. In general, there is an assumption that red guava fruit is sweeter than others. But in this research, it is known that red and white guava fruit almost had the same TSS values. So this assumption cannot always be said true. Accessions having low TSS values were ARP8743, and JBT004, while the accessions having high TSS values were ARP9406, ARP9407, ARP10.2, ARP10.7, ARP10.6, ARP10.8, ARP10.1, ARP10.12, ARP10.11, ARP8740, ARP8744 and JBT001.

Cluster analysis of 19 accession numbers were based on morphological characters of leaf and fruit (Figure 2.) It shows that 19 accessions of guava had 70-90% of similarity level or genetic distance of 0-20%. According to Cahyarini *et al*. (2004), the plants had the value of similarity level less than 0.60 or 60%, meaning that they had far genetic distance. In this research, accessions which had far genetic distance were ARP8744 with ARP8741 and JBT 003 with ARP 10.12. Both of these accessions had similarity distance on 0.59. Sharma *et al*. (2010) stated that the variation among genotypes for different morphological characters could be attributed to genetic differences of these genotypes.

The accessions showing slight morphological similarity were good to be used as parent for crossing by considering of theirs characters. The clustering of this cultivar is useful in breeding activity to get new variety. The new variety will be better if the parent has far distance genetic relationship. Tatineni *et al*. (1996) said that parent plants having far genetic distance had opportunity to get higher heterosis hybrids than their parents. In this research, ARP8744 with ARP8741 and JBT003 with ARP10.12 were suitable as parent plant because these accessions had small similarity level.

Dendogram was obtained by 2 different groups at 70% of the similarity level, namely group I (ARP9406, ARP9407, ARP8653, ARP8742, ARP10.2, JBT001, JBT002, ARP8740, JBT003 and JBT004). Group I has the similarities in the character of the leaf shape of tip (acute), Color of under leaf (green), short petiole length (1.2-2.28 cm), and thin flesh thick(0.98-3.17 cm). At group I, the accessions which have the high similarity value (89%) are ARP9406 with ARP9407.

Group I involved ARP10.7, ARP10.6, ARP10.1, ARP10.12, ARP10.9, ARP10.11, ARP8744, ARP8741 and ARP8743. Group II had the same character in leaf twisting (absent), leaf curvature of midrib (present), and color of upper leaf surface (green). In group I, the accession which had the high similarity value (89%) was ARP10.1 with ARP10.2.

This dendogram was based on quantitative and qualitative characters. It described the cluster of every accession. It can be used to choose accession to be material selection in breeding.

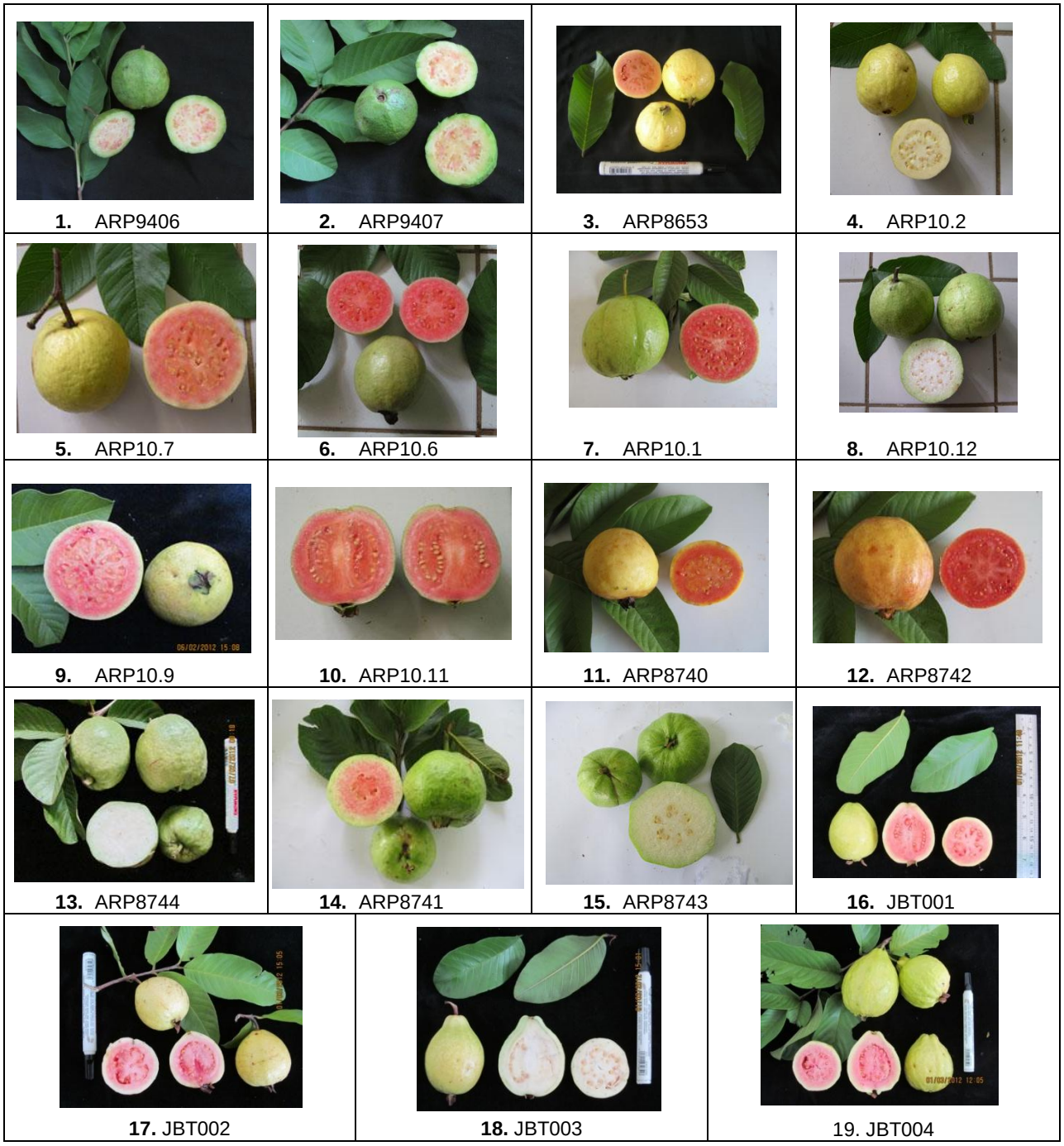

Figure 1. Fruit shape of nineteen accessions of guava

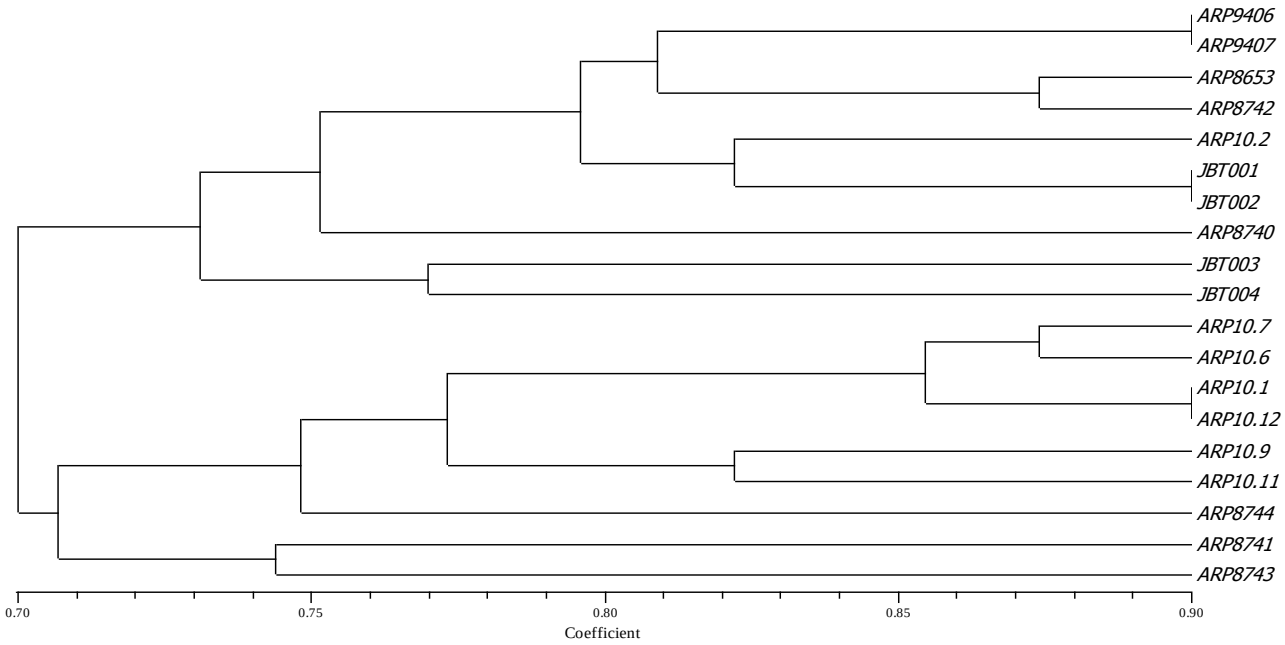

Figure 2. The dendogram of clustering some accessions of guava based on morphological leaf and fruit characters

CONCLUSIONS

Similarity level from 19 of guava accessions was 70 – 90% or genetic distance was 0 – 20%. All of accessions can be clustered into 2 clusters at 70% genetic similarity level. Accession which had the heaviest fruit weight was ARP8743 (300.91 grams) and accession which had the highest value of TSS was ARP10.1 (11.03°brix)

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Appendix 1. Morphological characterization of some guava germplasm collections at Aripan and Subang Experimental Farm

Accessions	ARP9406	ARP9407	ARP8653	ARP10.2	ARP10.7
Shape fully developed leaf	Obtrullate	Obtrullate	Obtrullate	Obovate	Oblong
Leaf curvature in cross section	Medium	Medium	Medium	Weak	Medium
Leaf twisting	Absent	Present	Absent	Absent	Absent
Leaf curvature of midrib	Absent	Present	Present	Absent	Absent
Leaf shape of base	Rounded	Rounded	Rounded	Cordate	Rounded
Leaf shape of tip	Obtuse	Obtuse	Obtuse	Obtuse	Apiculate
Leaf length (cm)	11.96	12.80	15.02	12.00	13.37
Leaf width (cm)	5.68	5.76	6.52	6.03	8.10
Color of upper leaf	Green	Green	Green	Green	Green
Color of under leaf	Green	Yellow green	Green	Green	Green
Fruit shape	Round oval	Round oval	Oval	Rounded	Rounded
Fruit shape at stalk end	Pointed	Pointed	Pointed	Rounded	Rounded
Fruit width of neck in relation to that of fruit	Very broad	Very broad	Very broad	Very broad	Very broad
Color of fruit skin	Green	Green	Yellow	Yellow	Yellow green
Color of flesh	Red	Red	Red	Yellow	Red purple
Fruit weight (gram)	115,82	148.43	162.04	118.31	191.09
Fruit width (cm)	6.32	6.29	6.50	5.92	6.86
Fruit length (cm)	6.45	6.23	7.07	6.40	7.11
Petiole length (cm)	1.57	1.67	1.84	1.55	2.21
Diameter core (cm)	4.26	4.28	4.30	3.74	4.43
Flesh thick (cm)	1.21	1.13	1.05	1.01	1.30
TSS (° brix)	11.00	10.60	9.20	10.20	9.86

Accessions	ARP10.6	ARP10.1	ARP10.12	ARP10.9	ARP10.11
Shape fully developed leaf	Obovate	Obovate	Obovate	Obovate	Obovate
Leaf curvature in cross section	Medium	Medium	Medium	Medium	Medium
Leaf twisting	Absent	Absent	Absent	Absent	Absent
Leaf curvature of midrib	Absent	Absent	Absent	Absent	Absent
Leaf shape of base	Rounded	Rounded	Rounded	Rounded	Rounded
Leaf shape of tip	Obtuse	Obtuse	Obtuse	Apiculate	Obtuse
Leaf length (cm)	14.82	13.02	14.39	11.97	12.03
Leaf width (cm)	8.08	6.30	6.05	5.39	7.94
Color ofupper leaf	Green	Green	Green	Green	Green
Color ofunder leaf	Green	Green	Green	Green	Green
Fruit shape	Round oval	Round	Round	Round	Round
Fruit shape at stalk end	Rounded	Rounded	Rounded	Broadly rounded	Rounded
Fruit width of neck in relation to that of fruit	Very broad	Very broad	Very broad	Very broad	Very broad
Color of fruit skin	Yellow green	Yellow green	Yellow green	Yellow green	Yellow green
Color of flesh	Red	White	White	Orange red	Red
Fruit weight (gram)	185.59	177.70	223.51	185.44	128.98
Fruit width (cm)	6.92	6.90	7.58	6.25	5.09
Fruit length (cm)	7.34	6.93	7.71	5.40	5.25
Petiole length (cm)	2,36	4.43	2.49	3.46	2.31
Diameter core (cm)	4.44	4.32	4.59	4.16	4.30
Flesh thick (cm)	1.20	1.21	1.04	1.37	0.98
TSS (° brix)	10.15	11.03	10.83	9.38	10.27

Accessions	ARP8740	ARP8742	ARP8744	ARP8741	ARP8743
Shape fully developed leaf	Obovate	Obovate	Ovate	Obovate	Obtrullate
Leaf curvature in cross section	Strong	Medium	Weak	Strong	Medium
Leaf twisting	Absent	Absent	Absent	Absent	Absent
Leaf curvature of midrib	Present	Absent	Absent	Absent	Absent
Leaf shape of base	Rounded	Rounded	Rounded	Rounded	Obtuse
Leaf shape of tip	Obtuse	Obtuse	Obtuse	Rounded	Obtuse
Leaf length (cm)	12.96	14.18	12.36	14.60	12.86
Leaf width (cm)	7.20	6.94	7.46	10.34	6.34
Color of upper leaf	Green	Green	Green	Green	Green
Color of under leaf	Yellow green	Green	Green	Green	Green
Fruit shape	Oval	Oval	Symmetry	Round	Round
Fruit shape at stalk end	Rounded	Rounded	Rounded	Broadly rounded	Broadly rounded
Fruit width of neck in relation to that of fruit	Very broad	Very broad	Very broad	Very broad	Very broad
Color of fruit skin	Yellow	Yellow	Yellow green	Yellow green	Yellow green
Color of flesh	Red	Red	White	Red	White
Fruit weight (gram)	227.08	158.39	175.82	287.54	300.91
Fruit width (cm)	7.33	6.06	7.56	8.40	8.93
Fruit length (cm)	7.31	5.58	8.29	7.27	6.65
Petiole length (cm)	1.25	1.97	2.26	4.31	1.50
Diameter core (cm)	4.03	4.30	7.56	2.67	3.89
Flesh thick (cm)	1.60	1.125	7.56	2.78	2.34
TSS (° brix)	10.10	9.00	10.02	8.80	7.10

Accessions	**JBT001**	**JBT002**	**JBT003**	**JBT004**
Shape fully developed leaf	Oblong	Oblong	Oblong	Oblong
Leaf curvature in cross section	Weak	Weak	Medium	Medium
Leaf twisting	Absent	Absent	Present	Absent
Leaf curvature of midrib	Absent	Absent	Absent	Absent
Leaf shape of base	Rounded	Rounded	Rounded	Rounded
Leaf shape of tip	Obtuse	Obtuse	Obtuse	Obtuse
Leaf length (cm)	12.45	12.96	13.15	15.04
Leaf width (cm)	5.82	5.51	7.07	7.42
Color of upper leaf	Green	Green	Green	Green
Color of under leaf	Green	Green	Green	Green
Fruit shape	Round oval	Rounded inverted cone	Ovate	Oval
Fruit shape at stalk end	Necked	Pointed	Necked	Necked
Fruit width of neck in relation to that of fruit	Broad	Broad	Medium	Medium
Color of fruit skin	Yellow	Yellow	Yellow	Yellow
Color of flesh	Red	Red	Yellow	Red
Fruit weight (gram)	122.64	106.95	139.82	215.008
Fruit width (cm)	5.93	5.74	5.96	7.14
Fruit length (cm)	6.69	6.32	7.54	8.60
Petiole length (cm)	1.48	1.67	1.29	1.20
Diameter core (cm)	3.20	3.69	3.99	4.41
Flesh thick (cm)	1.23	1.13	1.21	1.33
TSS (° brix)	10.75	9.71	9.13	7.80