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CHANGES IN PHYSIOLOGICAL AND BIOCHEMICAL INDICATORS DURING THE GROWTH AND DEVELOPMENT OF BANANA FRUIT (*Musa paradisiaca* L.) GROWN IN VINH PHUC PROVINCE

Le Van Trong[1] and Nguyen Nhu Khanh[2]
[1]*Faculty of Natural Sciences, Hong Duc University*
[2]*Faculty of Biology, Hanoi National University of Education*

Abstract. The banana fruit reached its maximum size in terms of length and diameter at 18 weeks of age. The content of chlorophyll a in banana peel reached the highest value at 14 weeks old, chlorophyll b content reached the highest value at 12 weeks old and decreased rapidly. The content of carotenoid was low from fruit formation and then increased rapidly until the fruit is fully ripening. The vitamin C content and total organic acid content reached a maximum at 16 weeks old and then decreased until the fruit is fully ripened, the reducing sugar content increased continuously and reached a maximum at 18 weeks old and then decreased slightly. The starch content and pectin content reached a maximum when the fruit was 14 weeks old and then gradually decreased. The tannin content decreased from fruit formation until fruit ripening. Through the research process, we found that banana achieves the best quality to harvest when fruits are 18 weeks old. The results of this study provide important data for the harvesting and storage of banana fruit.

Keywords: banana fruit, biochemical indicators, physiological indicators, ripening.

1. Introduction

Banana (*Musa paradisiaca* L.) is an edible fruit - botanically a berry produced by several kinds of large herbaceous flowering plants in the genus *Musa*. The scientific names of most cultivated bananas are *Musa acuminata*, *Musa balbisiana*, and *Musa* × *paradisiaca* for the hybrid *Musa acuminata* × *M. balbisiana*, depending on their genomic constitution. The old scientific name for this hybrid, *Musa sapientum*, is no longer used.

Bananas are the developing world's fourth most important food crop in terms of the gross value of production. The crop is grown in more than 100 countries throughout the tropics and sub-tropics, with an annual world production of around 98 million tons, of which around a third is produced in each of the African, Asia-Pacific, and Latin American

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Contact: Le Van Trong, e-mail address: levantrong@hdu.edu.vn

and Caribbean regions (Frison and Sharrock, 1999) [1]. In many countries, bananas are more than just a food crop. They provide an important source of fiber and among other uses, can be fermented to produce alcohol. Bananas are rich in antioxidants, magnesium, and vitamin C. The other nutrients found in the fruit are vitamin B6, protein, dietary fiber, riboflavin, niacin, iron, etc. There have been many studies on the physiological and biochemical changes in fruits at different stages of development. Rajkumar *et al.* (2012) [2] studied banana fruit quality and maturity stages using hyperspectral imaging. Maduwanthi and Marapana (2017) [3] concluded that softening of the texture, yellowing of peel, reduction of astringency, and increase of sweetness are major organoleptic changes that can be noted in banana ripening. These changes occur as a result of a series of biochemical changes in the peel and flesh of banana fruit.

In Vietnam, bananas are grown relatively popular with many new varieties for high and stable yield. However, the harvesting and preservation of bananas have not had a scientific basis but based on the experience of gardeners, this makes the majority of bananas in the market not yet ensure quality, affecting the health of consumers. Therefore, we conducted fruit sampling, analyzing the physiological and biochemical indicators of bananas from formation to fruit ripening. Thereby finding out the physiological ripening time of bananas to help consumers use and preserve bananas better.

2. Content

2.1. Materials and methods

**** Materials***

The banana was harvested in Vinh Phuc, Vietnam (21°21′49″N and 105°32′54″E) from January to July in 2011 and followed the mixed sampling method. Analytical experiments of physiological and biochemical criteria were analyzed at the Department of Plant Physiology and Application, Hanoi National University of Education.

**** Sample collection method***

Samples were collected according to the mixed sampling method. Samples were collected in the morning, then refrigerated and transferred to the laboratory. Part of the sample is used to immediately analyze indicators of pigments content, enzymes, vitamin C. The rest of the sample is stored at -80^{O}C to analyze other indicators.

**** Determination of length and diameter fruit***

The length and diameter of the fruit were measured by Palme calipers and accurate to mm.

**** Determination of pigment content in the peel [4]***

Chlorophyll content is calculated by the formula: C_a (mg/L) = 9.784 × E_{662} - 0.990 × E_{644}. C_b (mg/L) = 21.426 × E_{644}-4.650 × $E_{662.}$ Carotenoid content is calculated by the formula: $C_{carotenoid}$(mg/l) = 4.695 × $E_{440.5}$ - 0.268 × $C_{(a+b)}$. Then the pigment content per 1 g of fresh fruit peel is calculated by the formula: $A = \frac{C \times V}{P \times 1000}$ where E_{662}, E_{644} and $E_{440.5}$ are the results of measuring chlorophyll color at wavelengths of 662 nm, 644 nm and 440.5 nm; C_a, C_b, C_{a+b}, $C_{carotenoid}$ are respectively chlorophyll content a, b, a + b and carotenoids; A is the content of the pigment in 1g of fresh fruit peel; V is the volume of pigment extract (10 mL); P is the sample mass (g).

**** Determination of reducing sugar content, starch [5]***

Reducing sugar content is calculated by the formula: $X = \frac{a \times V_1 \times 100}{V \times b \times 1000}$

The starch content is calculated by the formula: $Y = \frac{a \times V_1 \times 100 \times 0.9}{V_2 \times b}$, where X is the reducing sugar content (%); Y is the content of starch (%); a is the weight (mg) of glucose obtained; b is the weight of the analyzed sample (g); V is the volume of the diluted sample solution (mL); V_1 is the volume of the analyzed sample solution (mL); V_2 is the volume of diluted sample solution (mL); *100* is the conversion factor to %; *1000* is the coefficient converting g to mg; *0.9* is the coefficient of converting glucose into starch.

**** Determination of pectin content by calcium pectate precipitation method [5]***

The amount of pectin taken for saponification (B) is calculated by the formula $B = \frac{W \times V_2}{V_1}$ Where: W is the weight of pectin introduced into the solution (g); V_1 is the volume of the initial pectin solution (mL); V_2 is the volume of pectin solution taken for saponification (mL).

The content of pectin (P) is calculated by the formula: $P = \frac{A \times 0.92 \times 100}{B}$, where: A is the amount of the calcium pectate precipitate (g); B is the amount of pectin taken for saponification (g); *0.92* is the transfer coefficient except for the calcium content of the precipitate.

**** Determination of total organic acid content [6]***

Total organic acid content is calculated by the formula: $X = \frac{a \times V_1 \times 100}{V_2 \times P}$, where X is the amount of total organic acid present in the extract; P is the amount of analytical sample (g); V_1 is the total volume of extract (mL); V_2 is the volume to be titrated (mL); a is the amount of 0.1N NaOH titration (mL).

**** Determination of tannin content by Leventhal method [6]***

The tannin content is calculated by the formula $X(\%) = \frac{(a-b) \times V \times k \times 100\%}{V_f \times g}$ where X is the tannin content (%); a is the volume of $KMnO_4$ used for titration in the flask (mL); b is the volume of $KMnO_4$ used for titration in the control vessel (mL); V is the total volume of extract (mL); V_f is the volume of the analyzed extract (mL); g is the weight of the analyzed sample (g); k is the tannin coefficient.

**** Determination of vitamin C content [7]***

Vitamin C content is calculated by the formula: $X = \frac{V \times V_1 \times 0.00088 \times 100}{V_2 \times P}$, where X is the content of vitamin C in the materials (%); V is the volume of diluted sample solution (mL); V_1 is the volume of 0.01N I_2 solution (mL); V_2 is the volume of analyzed solution

(mL); *P* is the weight of the sample (g); *0.00088* is the weight (g) of vitamin C which was equivalent to 1 mL of 0.01N I_2.

**** Statistical analysis***

The results are expressed as mean values and standard deviation (SD). The results were subjected to an analysis of variance. Data were compared according to Tukey's test using IRRISTAT software (version 5.0) for Windows computers.

2.2. Results and discussion

2.2.1. Changes in the length and diameter of banana fruit

The length and diameter of the fruit increased according to the growth and development of the fruit. From the period of 2 weeks to 19 weeks of age, the length of fruit increased from 10.565 cm to 17.470 cm, while the diameter of fruit increased from 1.125 cm to 4.340 cm. In particular, a sharp increase in fruit size was observed during the period from 2 weeks to 17 weeks of age. This is due to an increase in both the number and size of cells in the banana fruit. From 17 to 18 weeks old, the fruit reached the maximum value of the cultivar in the study condition. After 18 weeks of age, fruit size increased very slowly and almost unchanged (Figure 1).

Table 1. Length and diameter of banana fruit

Age of fruit development	**Length *(cm)***	**Diameter *(cm)***
2 weeks	$10.565^{g} \pm 0.212$	$1.125^{d} \pm 0.069$
6 weeks	$11.580^{f} \pm 0.321$	$1.630^{d} \pm 0.132$
10 weeks	$13.275^{e} \pm 0.436$	$2.850^{c} \pm 0.095$
12 weeks	$14.155^{d} \pm 0.137$	$3.420^{b} \pm 0.215$
14 weeks	$15.100^{c} \pm 0.218$	$3.760^{b} \pm 0.154$
16 weeks	$16.550^{b} \pm 0.097$	$4.250^{a} \pm 0.078$
17 weeks	$17.450^{a} \pm 0.327$	$4.305^{a} \pm 0.215$
18 weeks	$17.465^{a} \pm 0.086$	$4.320^{a} \pm 0.314$
19 weeks	$17.470^{a} \pm 0.158$	$4.340^{a} \pm 0.213$

Note: In the same data column, values with similar letters represent non-significant differences, values with different letters represent significant differences ($P \leq 0.05$) according to the Tukey test.

During the study, we observed that at 18 weeks of age, banana peel began to appear in the first yellow spot. Therefore, it can be said that the time of 18 weeks of age is the physiological maturity stage of banana.

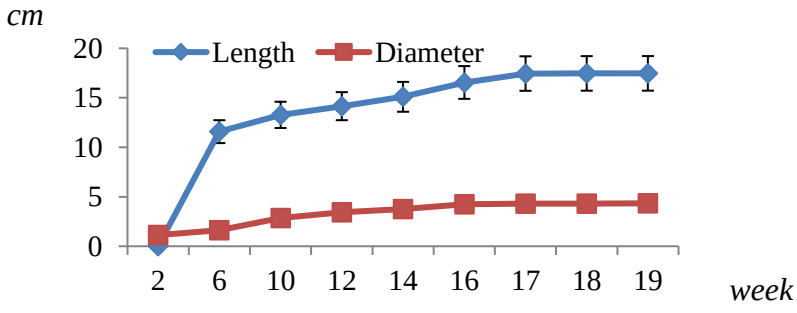

Figure 1. Changes in length and diameter during the growth and development of banana fruit

2.2.2. Changes in the pigment content of the banana peel

The data from Table 2 showed that, in the first week, the content of chlorophyll in banana peel was low. The content of chlorophyll *a* was 0.074 mg/g fresh peel, chlorophyll *b* was 0.031 mg/g fresh peel and total chlorophyll was 0.105 mg/g fresh peel at 2 weeks old. The content of chlorophyll *a* in banana peel reached the highest value at 14 weeks old (Chlorophyll *a* was 0.094 mg/g fresh peel), the content of chlorophyll *b* reached the highest value at 12 weeks old and decreased after 12 weeks.

After 14 weeks old, the content of chlorophyll gradually decreased and decreased rapidly at 18 and 19 weeks old. Chlorophyll content decreased become absent in ripe fruit [8-10] and the peel color changed from green to yellow during the ripening of banana fruit.

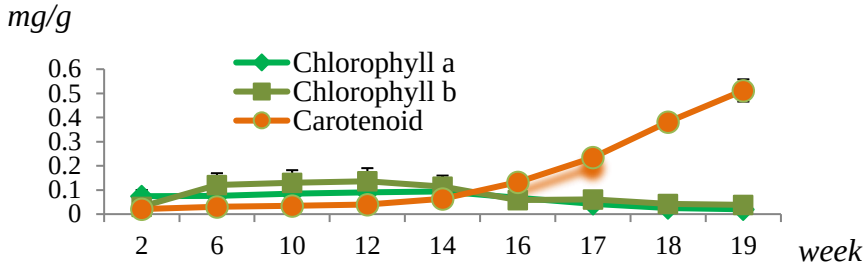

Figure 2. Changes in chlorophyll and carotenoid content during the growth and development of banana fruit

Table 2. Content of pigment systems in the banana peel

Age of fruit development	Chlorophyll a *(mg/g fresh peel)*	Chlorophyll b *(mg/g fresh peel)*	Carotenoid content *(mg/g fresh peel)*
2 weeks	$0.074^{b} \pm 0.002$	$0.031^{d} \pm 0.004$	$0.021^{f} \pm 0.003$
6 weeks	$0.076^{b} \pm 0.004$	$0.121^{b} \pm 0.002$	$0.030^{f} \pm 0.002$
10 weeks	$0.085^{a} \pm 0.003$	$0.130^{a} \pm 0.004$	$0.034^{f} \pm 0.001$
12 weeks	$0.090^{a} \pm 0.001$	$0.136^{a} \pm 0.003$	$0.040^{f} \pm 0.004$
14 weeks	$0.094^{a} \pm 0.004$	$0.114^{b} \pm 0.001$	$0.064^{e} \pm 0.003$
16 weeks	$0.068^{c} \pm 0.002$	$0.058^{c} \pm 0.003$	$0.132^{d} \pm 0.001$
17 weeks	$0.042^{d} \pm 0.001$	$0.061^{c} \pm 0.001$	$0.234^{c} \pm 0.001$
18 weeks	$0.025^{d} \pm 0.002$	$0.042^{d} \pm 0.004$	$0.381^{b} \pm 0.003$
19 weeks	$0.020^{d} \pm 0.001$	$0.038^{d} \pm 0.002$	$0.512^{a} \pm 0.001$

Note: In the same data column, values with similar letters represent non-significant differences, values with different letters represent significant differences ($P \leq 0.05$) according to the Tukey test.

Carotenoid content in banana peel increased with the age of fruit development. In the first week of bananas, low carotenoid content reached 0.021 mg/g of fresh peel at 2 weeks old. From 2 to 14 weeks old, the content of carotenoid increased slowly, then increased rapidly according to the ripening of the fruit. At 19 weeks old, the content of

the carotenoid reached 0.512 mg/g of the fresh peel. Thus, it can be seen that the reduction of chlorophyll content along with the increase of the carotenoid content of fruit development.

2.2.3. Changes in reducing sugar content and starch content

The results in Table 3 showed that the content of reducing sugar in the early period of bananas fruit (2 weeks) was relatively low, reached 0.765% weight of fresh fruit.

Table 3. Content of reducing sugar and starch in banana fruit

Age of fruit development	**Reducing sugar content** ***(% weight of fresh fruit)***	**Starch content** ***(% weight of fresh fruit)***
2 weeks	$0.765^{e} \pm 0.120$	$3.241^{e} \pm 0.076$
6 weeks	$0.843^{e} \pm 0.171$	$8.517^{d} \pm 0.098$
10 weeks	$1.025^{c} \pm 0.217$	$17.213^{b} \pm 0.154$
12 weeks	$1.156^{c} \pm 0.240$	$23.314^{a} \pm 0.221$
14 weeks	$1.212^{c} \pm 0.650$	$24.716^{a} \pm 0.314$
16 weeks	$2.589^{b} \pm 0.238$	$22.765^{a} \pm 0.120$
17 weeks	$3.468^{b} \pm 0.431$	$20.327^{a} \pm 0.154$
18 weeks	$4.098^{a} \pm 0.765$	$16.431^{b} \pm 0.255$
19 weeks	$3.865^{a} \pm 0.129$	$12.723^{c} \pm 0.128$

Note: In the same data column, values with similar letters represent non-significant differences, values with different letters represent significant differences ($P \leq 0.05$) according to the Tukey test.

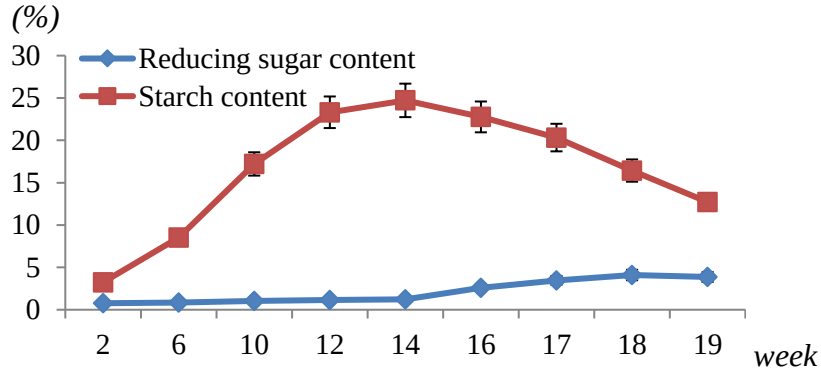

Figure 3. Changes in reducing sugar and starch content during the growth and development of banana fruit

From 2 to 14 weeks old, the content of reducing sugar increased slowly and reached 1.212% when the fruit was 14 weeks old. In the fruit period from 14 to 18 weeks old, the content of reducing sugar increased rapidly and reached 4.098% when the fruit was 18 weeks old. At 19 weeks old, the content of reducing sugar decreased to 3.865% weight of fresh fruit so the quality of the fruit decreased.

When the fruit has just formed, low starch content only reached 3.241% weight of fresh fruit flesh (2 weeks old). After that, saccharose from leaves and peels is

transferred into the fruit to provide materials for the synthesis of starch, so the starch content in the fruit increased gradually. The highest starch content was 24.716% at 14 weeks old. After 14 weeks of old, the content of starch in the fruit decreased.

2.2.4. Changes in total organic acid content and vitamin C content

The data in Table 4 showed that at the stage when fruit starts to the formation, the accumulation of large organic matter amounted to 15.425 mg/100g fresh fruit.

Table 4. Content of total organic acid and vitamin C in banana fruit

Age of fruit development	Total organic acid content *(mg/100g fresh fruit)*	Vitamin C content *(mg/100g fresh fruit)*
2 weeks	$15.425^{c} \pm 0.043$	$17.607^{c} \pm 0.035$
6 weeks	$18.750^{b} \pm 0.069$	$35.230^{b} \pm 0.046$
10 weeks	$22.523^{a} \pm 0.046$	$39.600^{b} \pm 0.124$
12 weeks	$23.250^{a} \pm 0,052$	$44.420^{a} \pm 0.096$
14 weeks	$23.750^{a} \pm 0,124$	$45.802^{a} \pm 0.047$
16 weeks	$24.538^{a} \pm 0,097$	$46.610^{a} \pm 0.035$
17 weeks	$18.750^{b} \pm 0,086$	$39.400^{b} \pm 0.162$
18 weeks	$17.756^{b} \pm 0.047$	$36.142^{b} \pm 0.085$
19 weeks	$17.250^{b} \pm 0.080$	$34.037^{b} \pm 0.045$

Note: In the same data column, values with similar letters represent non-significant differences, values with different letters represent significant differences ($P \leq 0.05$) according to the Tukey test.

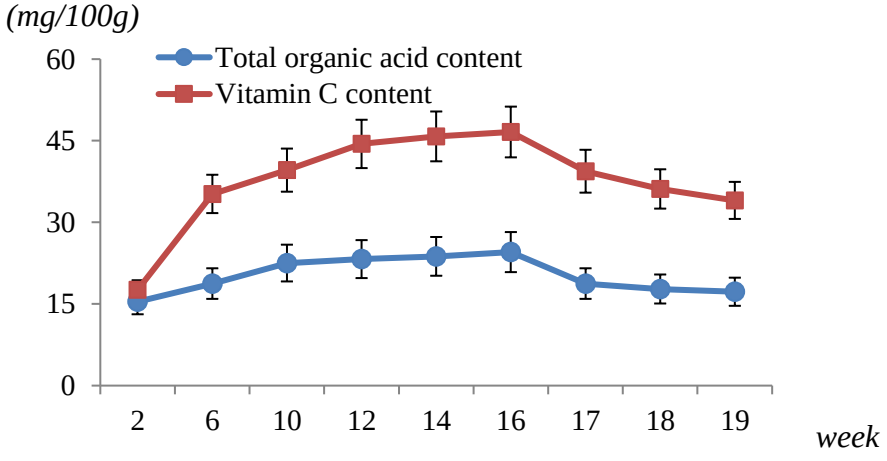

Figure 4. Changes in total organic acid and vitamin C content during the growth and development of banana fruit

In the fruiting period from 2 to 16 weeks old, the total organic acid content increased gradually and reached the highest value of 24.538 mg/100g fresh fruit at 16 weeks old. This is because in the fruit, protein exchange processes, hydrocarbon exchange, lipids take place strongly, creating intermediate products such as amino

acids, xetoaxit, etc., increasing the content of organic acids. In the fruiting period from 16 to 19 weeks old, organic acid content decreased due to organic acid used in respiration to provide energy for starch synthesis processes [11].

The content of vitamin C from 2 to 16 weeks old increased rapidly, this is a period of strong flesh fruit development and the accumulation of vitamin C along with other nutrients in the fruit, the highest value reached 46.610 mg/100g fresh fruit on the 16th week, after 16 weeks, vitamin C content decreased.

2.2.5. Changes in pectin content and tannin content

The results of the data in Table 5 showed that the content of pectin in bananas increased slightly from young fruit to 14 weeks old. At this time, pectin content did not increase much (from 3.259% to 4.849% of dry weight). This is because at this stage, the number of cells changes is small but the size and mass of the cells increase rapidly, so the content of pectin that makes up the intercellular binder also increases to ensure the bonding of cells together [12].

Table 5. Content of pectin and tannin in banana fruit

Age of fruit development	**Pectin content *(% dry weight)***	**Tannin content *(% dry weight)***
2 weeks	$3.259^{c} \pm 0.009$	$3.608^{a} \pm 0.005$
6 weeks	$3.435^{b} \pm 0.023$	$3.354^{a} \pm 0.014$
10 weeks	$3.914^{b} \pm 0.002$	$2.659^{b} \pm 0.018$
12 weeks	$4.542^{a} \pm 0.007$	$2.328^{b} \pm 0.009$
14 weeks	$4.849^{a} \pm 0.013$	$2.195^{b} \pm 0.021$
16 weeks	$3.839^{b} \pm 0.025$	$1.546^{c} \pm 0.016$
17 weeks	$3.074^{c} \pm 0.037$	$1.397^{c} \pm 0.013$
18 weeks	$2.467^{d} \pm 0.004$	$0.964^{d} \pm 0.008$
19 weeks	$2.031^{d} \pm 0.015$	$0.531^{d} \pm 0.014$

Note: In the same data column, values with similar letters represent non-significant differences, values with different letters represent significant differences ($P \leq 0.05$) according to the Tukey test.

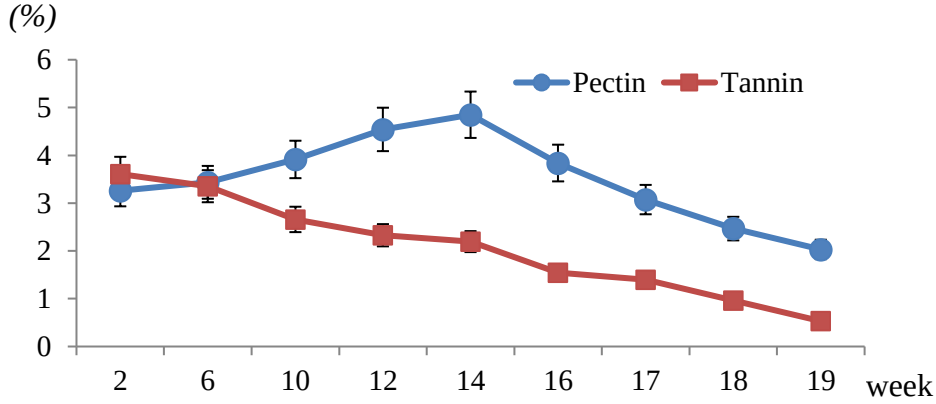

Figure 5. Changes in pectin and tannin content during the growth and development of banana fruit

Pectin content decreased sharply in the period from 14 to 19 weeks old (from 4.849% to only 2.031%). This is a period of ripe fruit, a strong decrease in pectin content during this period due to the increase in the activity of protopectinase enzyme that has dissolved pectin. Pectic enzymes are related to the softening of fruit along with the increase in soluble pectin [13].

Tannin in banana fruit had a relatively high content from 2 weeks old (reached 3.608%). The content of tannin is high in the early period of making banana fruit acrid and pungent. The tannin content in bananas gradually decreased with age and rapidly decreased in the period of 14 to 19 weeks old (Figure 5). In the period of fruit maturity from 18 to 19 weeks old, tannin content decreased to only 0.531% at 19 weeks old, makes banana ripen soft, not acrid.

3. Conclusions

The length and the diameter of the banana fruit reached the maximum size at 18 weeks old. The content of chlorophyll a in banana peel reached the highest value at 14 weeks old and decreased rapidly to 19 weeks old, the content of chlorophyll b reached the highest value at 12 weeks old and decreased to 19 weeks old. In contrast, low carotenoid content from fruit formation to 14 weeks old, then increased rapidly until the fruit is fully ripening.

The starch content increased gradually from the beginning and reached the maximum when the fruit was 14 weeks old, then gradually decreased. Reducing sugar content increased continuously and reached maximum at 18 weeks old, then decreased slightly. Total organic acid content increased slightly from the beginning to 16 weeks old. From 16 to 19 weeks old, total organic acid content decreased sharply. The vitamin C content increased continuously and reached a maximum of 18 weeks old, then decreased slightly.

The content of pectin in banana fruit had a relatively high content at 14 weeks old and decreased to 19 weeks old. The tannin content in bananas decreased gradually according to the ripening of the fruit.

Throughout the study, we found that bananas reached physical maturity at 18 weeks of age, at which time the fruit size is almost unchanged and bananas fruit achieved the best quality. Therefore, this is the time to harvest the most appropriate. If harvested earlier or later, the quality of the banana fruit will be significantly reduced.

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