Evolution of maturation indexes of guava (*Psidium guajava* L.) and their relation with light scattering during postharvest

A. Padilla-Tena[1], B. Yahuaca-Juárez[1], J.A. Huerta-Ruelas [2], C.J. Cortés-Penagos[1], R. Salgado-Garciglia[3] y J. Ponce-Saavedra[4]

1 Facultad de Químico Farmacobiología, Universidad Michoacana de San Nicolás de Hidalgo. **2** Centro de Investigación de Ciencia Aplicada y Tecnología Avanzada, Instituto Politécnico Nacional Unidad Querétaro. **3** Instituto de Investigaciones Químico Biológicas, Universidad Michoacana de San Nicolás de Hidalgo. **4** Facultad de Biología, Universidad Michoacana de San Nicolás de Hidalgo. ibqadrianapad@hotmail.com

RESUMEN:

Para determinar la firmeza, ácido ascórbico y contenido de carotenoides totales en guayaba se emplean métodos destructivos, por lo que se requiere de implementar métodos no destructivos que permitan determinar la calidad del fruto en cada etapa de su maduración. El objetivo de este proyecto fue implementar la técnica de esparcimiento de luz para evaluar los cambios ocurridos en guayaba durante su maduración. Se trabajó con guayabas cultivar "media china" en estado de maduración fisiológica, con almacenamiento por 6 días a 20±5°C y humedad relativa del 90-95%. Se usó la técnica de esparcimiento de luz empleando un láser de 635nm, en la evaluación fisicoquímica se determinó firmeza mediante la resistencia a la compresión; ácido ascórbico por titulación volumétrica con 2-6-diclorofenol indofenol y carotenoides totales por espectrofotometría. Los resultados indican que la intensidad de la luz esparcida aumenta conforme avanza la maduración del fruto, relacionándose con una disminución en la firmeza de 56.1N a 14.8N, y un incremento en los carotenoides totales de a 86.1 mg/kg. El ácido ascórbico no presenta una tendencia clara. El esparcimiento de luz puede considerarse una alternativa no destructiva para detectar cambios en los frutos durante su maduración y predecir su comportamiento biológico.

Palabras clave: esparcimiento de luz, guayaba, maduración, *Psidium guajava*, poscosecha, técnica no destructiva

ABSTRACT:

In order to determine firmness, ascorbic acid and total carotenoid content in guava, destructive methods are used. Therefore, it is necessary to implement non-destructive methods to determine the quality of the fruit at each stage of ripening. The objective of this project was to implement the technique of light scattering to evaluate the changes that occurred in guava during its maturity. We worked with guavas "media china" in physiological maturation state, with storage for 6 days at 20 ± 5°C and relative humidity of 90-95%. The light scattering technique was used using as a laser of 635nm, in the physicochemical evaluation the firmness was determined by the resistance to the compression; ascorbic acid by volumetric titration with 2-6 dichlorophenol indophenol and total carotenoids by spectrophotometry. The results indicate that the intensity of light scattering increases as fruit maturation progresses, related to a decrease in firmness from 56.1N to 14.8N, and an increase in total carotenoids from 32.7 to 86.1 mg / kg. Ascorbic acid does not show a clear trend. Light scattering can be considered a non-destructive alternative to detect changes in fruits during maturation and to predict their biological behaviour.

Palabras clave: guajava, light scattering, maturity, non-destructive technique, postharvest, *Psidium guajava*

INTRODUCTION

Guava (*Psidium guajava* L.) belongs to the family *Myrtaceae*; it grows well in tropical and subtropical regions. It is currently one of the fruits that has acquired great importance, the production of guava is carried out in several countries, where Mexico stands out, the state of Michoacan being the main producer with a production of the order of 148,300 tons (Servicio de Información Agroalimentaria y Pesquera, 2017). This fruit is very versatile in terms of its different uses, it is mainly consumed in fresh (80-85 %) and the rest is industrialized in products such as juices, jams, etc. (Ramírez-Méndez et al., 2010;González-Gaona et al., 2002).

Fresh fruits play a significant role in human nutrition, especially as sources of vitamins (Vitamin C or ascorbic acid, Vitamin A, Vitamin B6, thiamine, niacin). Other constituents that may lower risk of cancer and other diseases include flavonoids, carotenoids and polyphenols (Kader, 2013). Regarding vitamin C, this is an important and essential nutrient, the lack of this vitamin leads to scurvy and on the other hand, the carotenoid pigments are fundamental in the feeding, since they are the precursors of vitamin A; both compounds are also considered natural antioxidants used in formulations of food products in order to improve its shelf life. During the postharvest stage the ascorbic acid content is decreasing by various factors, by the aerobic degradation pathway is related to the presence of oxygen and on the other hand the anaerobic pathway by temperature conditions and low relative humidity during storage (Castro et al., 2004) and the carotenoids increase exponentially as the maturation process progresses (González Cárdenas, 2010).

For the determination of these parameters of maturity traditionally they are evaluated by analytical methods in which the destruction of some fruits is required, require the use of equipment and chemical reagents that are expensive and the time of analysis is prolonged (Gregorio, 2014). Currently, new researches have been developed that are evaluating new techniques that are not destructive and that allow us to predict the ripening indices to determine the quality of the fruits, among these techniques we can find those that are based on optical properties (Gregorio, 2014; Lu and Peng, 2006; Nassif et al., 2012; Nicolaï et al., 2007; Wang and Li, 2013). Therefore, the objective of the present research was to establish the optical technique of light scattering to evaluate the changes of firmness, ascorbic acid and carotenoids in the fruit of guava during postharvest.

MATERIALS Y METHODS

Guava fruits (*Psidium guajava* L.) Cv. media china were used in physiological maturation stage with cream colored pulp, these were obtained at the local market of Morelia, Michoacán, where 30 fruits were selected and stored at 20 ± 5 °C and relative humidity of 90-95 % for a period of 6 days of storage for analysis.

- Fruits selection

The fruits were selected according to the colour of the epicarp (green) and subsequent a fast test of firmness using a compression distance of 1 mm (Maldonado, 2011).

- Physicochemical analysis

a. Firmness

It was performed by means of the compressive strength with the use of a TA-XT2 texturometer, a stainless steel flat plate of compression of 75 mm in diameter, with a compression distance of 3 mm and a displacement speed of 1 mm / S, expressing the results in Newtons (Maldonado, 2011).

b. Ascorbic acid (Vitamin C)

It was determined by the volumetric titration method with 2-6-dichlorophenol-indophenol, according to AOAC, 1988.

c. Total Carotenoids

The total extraction of carotenoids was carried out by means of the modified method of Minguez-Mosquera and Garrido-Fernández, 1989, where 0.5g of sample of guava pulp was added 10 ml of N, N-dimethylformamide saturated with $MgCO_3$ (0.2 mg of $MgCO_3$ in 500 ml of N, N-dimethylformamide). Shake for 5 min in a vortex and filter in a Whatman No. 42 filter paper. The solid residue was collected and the extraction operation was performed three times on the same sample. The extracts were combined and transferred to a separatory funnel in which 7 mL of hexane had previously been placed. After shaking for 1 min, it was left to settle approximately 10 min, until there was complete phase separation. The upper hexane layer, with light yellow colour, it is the layer of interest. In a graduated test tube recover the carotenes extract and perform the absorbance at 470 nm. Perform a previous calibration with hexane 100 %.

Calculation:

$$Total\ carotenoids\ \left(\frac{mg}{kg}\right) = \frac{A * (470) *\ 10^{6}}{200 * 100 * VC} \qquad \text{Ec. (1)}$$

VC =Volume carotene extract

- Optical characterization

For the optical characterization the laser light scattering technique was used, using as a light source a 635 nm He-Ne laser with emission power of 3 mV and as detector an array of 16 silicon photodiodes. The same experimental unit and point of incidence was used for the monitoring.

- Statistical Analysis

For the statistical analysis, a linear regression model was used where the light intensity (mV) and the firmness (N), ascorbic acid and total carotenoids were considered as a variable response with a confidence level of 95 % ($\alpha = 0.05$), using the statistical package JMP version 6.0.

RESULTS AND DISCUSSION

Table I shows the values obtained from the physicochemical characterization of the guava fruit (firmness, ascorbic acid and total carotenoids) in six days of storage and different stages of maturity. The firmness oscillates between 14.8 and 56.1 N and a tendency to decrease during the ripening process can be observed. Comparing the values with those obtained by Mercado-Silva et al., 1998 of guava (*Psidium guajava* L.) cultivar media china in three stages of ripening: green, yellow-green and yellow with values of 35.9 to 7.1 N in spring-summer crop and 39.1 At 11.8 N autumn-winter, a decrease is also observed in firmness and the values present a difference due to the fact that the firmness is linked to the structural changes of the biological material, specifically to the concentration of water, the turgidity of the cells and The composition, where the cell wall of the fruit changes, becoming increasingly soft as it matures (Dussán-Sarria et al., 2008).

The trend towards ascorbic acid varies greatly and decreases during postharvest as stated by Solarte et al., 2010, which states that the ascorbic acid content does not present a clear trend regarding the ripening stage. Values between 156.1 and 227.3 mg / 100 g of sample were found. The decrease in this can be associated with some of the organic acids are essential components in the cycle of the tricarboxylic acids of respiration, they are an available energy source that can be used when other reserves are depleted and the increase of sugars (Solarte et al., 2010), on the contrary the increase of Vitamin C, Srivastava and Narasimhan, 1967 reported that some components obtained from the hydrolysis of the pectins used to synthesize the ascorbic acid known as vitamin C.

Tabla I. Physicochemical characterization of the guava fruit (firmness, ascorbic acid and total carotenoids)

Storge day	Firmness (N)	Ascorbic acid (mg /100 g of sample)	Total Carotenoids (mg / kg fruit)
D1	56.1 ± 4.0	198.5 ± 17.5	32.7 ± 8.1
D2	28.8 ± 6.9	213.3 ± 30.5	57.8 ± 5.6
D3	17.5 ± 3.7	182.6 ± 20.5	72.9 ± 12.6
D4	16.4 ± 4.1	227.3 ± 29.3	75.7 ± 9.7
D5	16.7 ± 6.1	156.1 ± 30.6	78.3 ± 15.2
D6	14.8 ± 2.7	218.6 ± 25.3	86.1 ± 9.9

On the other hand, according to the values obtained in reference to the concentration of the total carotenoids there is an increase as the maturation process progresses from 32.7 to 86.4 mg/kg of fruit. The color of the guavas is due to the presence of pigments like chlorophyll, that is degraded during the maturation which causes the loss of the green color which gives rise to the biosynthesis of new pigments like the carotenoids that are responsible for yellow and red colorations. The main carotenes are b-carotene and lycopene (González et al., 2011; Solarte Cruz, 2013). As reported by Espinal Ruiz, 2010 in guava ICA I (*Psidium guajava* L.) in three stages of maturation there is an increase of total carotenoid content is in the green state 95.42 µg/g of fruit, pint 97.40 µg/g of fruit, mature 244.86 µg/g of fruit, and a decrease in the state senescent 238.51 µg/g of fruit.

The results obtained from the optical characterization show how the intensity of the scattering light of the fruit increases progressively from 2 to 25 mV during the process the ripening process. The average results during the six days of storage are shown in Fig. 1.

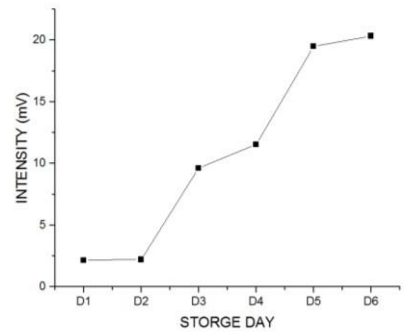

Figure 1. Intensity of light scattering as guava ripening

Three different patterns of light scattering were obtained for three ripening stages: yellow, green, yellow and green (Fig. 2), where it can be observed that there is an increase in the area under the curve during postharvest.

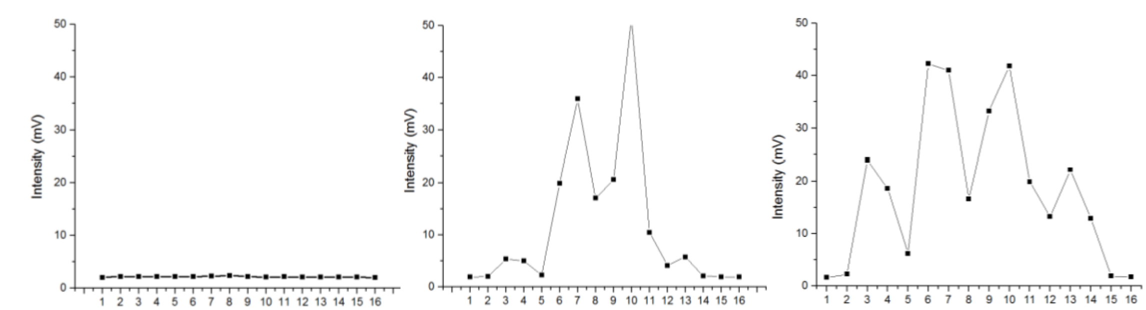

Figure 2. Patterns of scattering of light technique of ripening stages: a) green, b) green-yellow and c) yellow.

The interaction of light with matter can present two important phenomena: the absorption of light which is related to the concentration of certain components such as sugars, water and pigments, and on the other hand, the light scattering which offers a composition, density, cellular structures, intracellular and extracellular junctions (Lu and Peng, 2006).

Statistically significant correlations were found in the statistical analysis. The intensity of light scattered and the firmness presented a correlation coefficient of r = -0.73 and an adjustment of $r^2adj = 0.53$, which indicates an inversely proportional correlation, since the firmness of the fruit decreases as the ripening process progresses the scattered light intensity shows an increase (Fig. 3). The predictive formula Ec. (2) obtained was:

Ec. (2)

Regarding the concentration of ascorbic acid, a statistically significant correlation wasn´t obtained with the intensity of light scattering (r=-0.25) as it didn´t present a clear behaviour during its maturation process (Fig. 4).

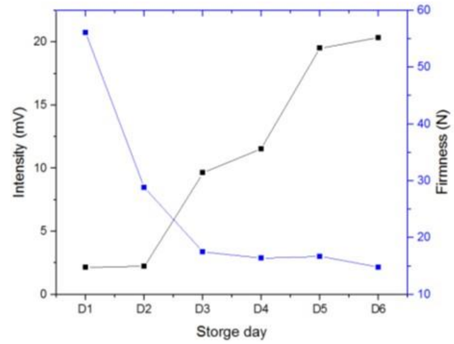

Figure 3. Changes in the intensity of scattered light and firmness during postharvest storage

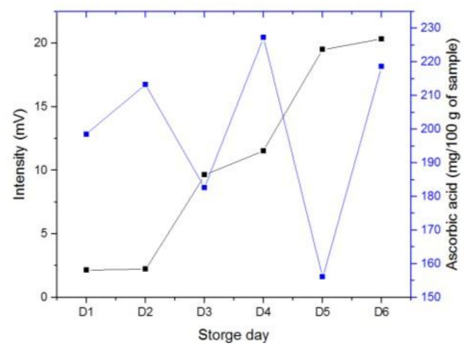

Figure 4. Behavior of ascorbic acid and light scattering as maturation progresses

Finally, a directly proportional correlation with the total carotenoid content of r = 0.85 and r^2 = 0.66 (α=0.05) was obtained, observing that in relation to the maturation process the total carotenoids show an increase as well as the intensity of the light scattered by the optical technique (Fig. 5), obtaining a predictive formula Ec. (3) to be able to predict its behaviour.

$$Total\ carotenoids\ (mg/kg) = 44.713549 + 2.0669142\ Intensity(mV)$$

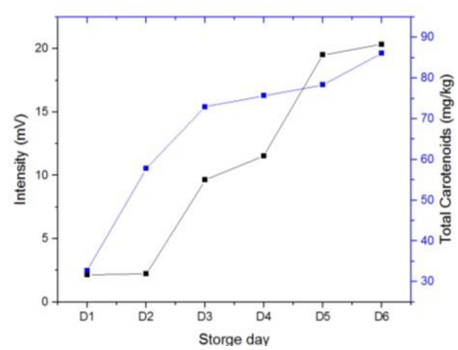

Figure 5. Change in total carotenoid content in relation to the intensity of light scattering during storage.

CONCLUSION

The technique of laser light scattering can be considered as a non-destructive alternative to predict the changes that present the parameters of firmness and content of total carotenoids of the guava fruit during its maturation process according to the results obtained, however for the determination of Ascorbic acid can´t be used for its prediction since there is no correlation with the intensity of light scattered.

BIBLIOGRAPHY

Castro, I., Teixeira, J. A., Salengke, S., Sastry, S. K., & Vicente, A. A. (2004). Ohmic heating of strawberry products: Electrical conductivity measurements and ascorbic acid degradation kinetics. *Innovative Food Science and Emerging Technologies*, *5*(1), 27–36. http://doi.org/10.1016/j.ifset.2003.11.001

Dussán-Sarria, S., Honório, S. L., & Matias, M. D. L. (2008). Mechanical resistance, respiratory rate and ethylene production of persimmon "Fuyu" during storage. *Revista Brasileira de Engenharia Agrícola E Ambiental*, *12*(5), 498–502. http://doi.org/10.1590/S1415-43662008000500009

Espinal Ruiz, M. (2010). Antioxidant capacity and softening of guava Palmira ICA I *(Psidium guajava)*. Universidad Nacional de Colombia, Bogotá, Colombia.

González-Gaona, E., Padilla-Ramírez, J. S., Reyes-Muro, L., Perales de la Cruz, M. A., & Esquivel-Villagrana, F. (2002). Guava cultivation in Mexico. Pabellón, Aguascalientes, México: Instituto Nacional de Investigaciones Forestales, Agrícolas y Pecuarias. Centro de Investigación Regional Norte Centro.

González, I. A., Osorio, C., Meléndez-Martínez, A. J., González-Miret, M. L., & Heredia, F. J. (2011). Application of tristimulus colorimetry to evaluate colour changes during the ripening of Colombian guava (Psidium guajava L .) varieties with different carotenoid pattern. *International Journal of Food Science & Technology*, *46*(2001), 840–848. http://doi.org/10.1111/j.1365-2621.2011.02569.x

González Cárdenas, I. A. (2010). Chemical characterization of the color of different varieties of guava *(Psidium guajava L.)* colombian. *Tesis*. Universidad Nacional de Colombia.

Gregorio, M. P. (2014). Optical methods for establishing cherry maturity. Universidad Zaragoza.

Kader, A. A. (2013). Postharvest Technology of Horticultural Crops - An Overview from Farm to Fork. *Journal of Applied Sciences and Technology*, *1*(1), 1–8.

Lu, R., & Peng, Y. (2006). Hyperspectral scattering for assessing peach fruit firmness. *Biosystems Engineering*, *93*(2), 161–171. http://doi.org/10.1016/j.biosystemseng.2005.11.004

Maldonado, N. E. (2011). Methods for determining maturation in guava varieties *(Psidium guajava L.)*. Universidad Michoacana de San Nicolás de Hidalgo.

Mercado-Silva, E., Benito-Bautista, P., & García-Velasco, M. de los A. (1998). Fruit development, harvest index and ripening changes of guavas produced in central Mexico. *Postharvest Biology and Technology*, *13*(2), 143–150. http://doi.org/10.1016/S0925-5214(98)00003-9

Minguez-Mosquera, M. I., & Garrido-Fernández, J. (1989). Chlorophyll and carotenoid presence in olive fruit (Olea europaea). *Journal of Agricultural and Food Chemestry*, *37*(1), 1–7. http://doi.org/10.1021/jo00245a001

Nassif, R., Pellen, F., Magné, C., Le Jeune, B., Le Brun, G., & Abboud, M. (2012). Scattering through fruits during ripening: laser speckle technique correlated to biochemical and fluorescence measurements. *Optics Express*, *20*(21), 23887. http://doi.org/10.1364/OE.20.023887

Nicolaï, B., Beullens, K., Bobelyn, E., Peirs, A., Saeys, W., Theron, K., & Lammertyn, J. (2007). Nondestructive measurement of fruit and vegetable quality by means of NIR spectroscopy: A review. *Postharvest Biology and Technology*, *46*, 99–118. Retrieved from http://www.siap.gob.mx/cierre-de-la-produccion-agricola-por-estado/

Ramírez-Méndez, R., Quijada, O., Camacho, R., & Burgos, M. E. (2010). Physicochemical quality of guava fruits (*Psidium guajava* L.) cultivated in three localities of the Zulia state, Venezuela. *Boletín Del Cento de Investigaciones Biológicas*, *44*(3), 285–296.

Servicio de Información Agroalimentaria y Pesquera. (2017). Guava, queen of vitamin C. Retrieved March 1, 2017, from http://www.gob.mx/siap/articulos/guayaba-reina-de-la-vitamina-c?idiom=es

Solarte, M. E., Hernández, M. S., Morales, A. L., Fernández-Trujillo, J. P., & Melgarejo, L. M. (2010). Physiological and biochemical characterization of guava fruit during maturation. In *DEVELOPMENT OF FUNCTIONAL PROMOTIONAL PRODUCTS FROM GUAYABA (Psidium guajava L.) FOR STRENGTHENING THE PRODUCTIVE CHAIN* (pp. 1–37). Cartagena, Colombia: Universidad Nacional de Colombia.

Solarte Cruz, M. E. (2013). Echophysiological aspects and bioactibe compounds of guava (*Psidium guajava* L.) in the province of Velez, Santander-Colombia. Universidad Nacional de Colombia.

Srivastava, H. C., & Narasimhan, P. (1967). Physiological Studies During the Growth and Development of Different Varieties of Guavas (Psidium Guajava L.). *Journal of Horticultural Science*, *42*(1), 97–104. http://doi.org/10.1080/00221589.1967.11514197

Wang, W., & Li, C. (2013). Measurement of the light absorption and scattering properties of onion skin and flesh at 633nm. *Postharvest Biology and Technology*, *86*, 494–501. http://doi.org/10.1016/j.postharvbio.2013.07.032