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Journal of Enviromental Sciences and Natural Resources

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Knowledge Area

The works must be unpublished and refer to topics of agriculture-forest, pathology-sustainable, horticulture, fisheries and aquaculture, agricultural biotechnology and other topics related to Biotechnology and Agricultural Sciences.

Presentation of Content

As first article we present, *Influence of the LED (Ligth Emitted Diode) ligthing spectrum in bean (Phaseolus vulgaris) germination stage*, by BAUTISTA-RAMÍREZ Agustina, PÉREZ-JIMÉNEZ Genaro, MARTINEZ-RUIZ, Antonio and QUINTANAR-OLGUIN, Juan, with asdcription Instituto Nacional de Investigaciones Forestales Agrícolas y Pecuarias, as next article we present, *Content of anthocyanins of seed of Phaseolus vulgaris*, by MEX-ÁLVAREZ, Rafael Manuel de Jesús, GARMA-QUEN, Patricia Margarita, GUILLEN-MORALES, María Magali and NOVELO-PÉREZ, María Isabel, with asdcription Universidad Autónoma de Campeche, as next article we present, *Physicochemical analysis in Averrhoa carambola L., var. Golden star and Arkin, in two post-harvest periods*, by TEMORES-RAMÍREZ, Cynthia Guadalupe, GARCÍA-MARTÍNEZ, Miguel Ángel, MÉNDEZ-MORÁN, Lucila and ZAÑUDO-HERNÁNDEZ, Julia, with affiliation at the Universidad de Guadalajara, as the next article we present, Estudio comparativo de tres abonos verdes en la producción de biomasa y en algunas propiedades del suelo, by CRESPO-GONZÁLEZ, Marcos Rafael, ZARAZÚA-VILLASEÑOR, Patricia, GONZÁLEZ-EGUIARTE, Diego Raymundo and ZAMORA-NATERA, Juan Francisco, with affiliation at the Universidad de Guadalajara, as the next article we present, *Noninvasive thermographic evaluation of the thermal condition of piglets in the first month of life*, by RAMÍREZ-DE LA TORRE, Hugo, SANCHEZ-CHIPRES, David Román, MORENO-LLAMAS, Gabriel and JIMÉNEZ-CORDERO, Ángel Andrés, with affiliation at the Universidad de Guadalajara.

Content

Article **Journal of Environmental Sciences and Natural Resources**
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Influence of the LED (Ligth Emitted Diode) ligthing spectrum in bean (*Phaseolus vulgaris*) germination stage

Influencia del espectro de iluminación LED (Ligth Emitted Diode) en etapa de germinación de frijol (*Phaseolus vulgaris*)

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Abstract

Evaluate influence of photoperiod by LED lighting in the bean germination stage. Four bean genotypes were used (SE, X1, M1 and N1). A completely randomized blocks experimental design was used with factorial arrangement of treatment with five repetitions. A standard germination test was performed. The samples were placed in germination chambers at 25 ± 5 ºC with photoperiods under LED lights treatments (red, blue, purple) and a fluorescent light (control) for ten days, after that time the variables evaluated were germination percentage, dead seeds, normal and abnormal seedlings. The analysis of variance showed significant differences in all the variables evaluated for light interaction; likewise, the multiple comparison of means by Tukey for light interaction showed that the red spectrum has positive effects on the SE, X1 and N1 genotypes by increasing the germination percentage; the purple and blue LED`s for genotypes E, X1 and N1 increased the number of dead seeds; with blue light in genotypes M1 and N1 the percentage of normal seedlings increased. The red LED increase germination percentage to 15 %, whereas the use purple and blue lighting is recommended for the development of seedlings.

Light emitted diode, Normal seedlings, Seeds viability

Resumen

Se evaluó la influencia del fotoperiodo por iluminación LED en la etapa de germinación de frijol. Se utilizaron cuatro genotipos de frijol SE, X1, M1 y N1, con los espectros LED rojo, azul, morado y un testigo con luz flourescente. El diseño experimental fue de bloques completos al azar, con arreglo factorial de tratamientos con cinco repeticiones. Las muestras fueron colocadas en cámaras de germinación, a 25 ± 5 ºC con fotoperiodo bajo luces LED´s (roja, azul, morada) y una luz flourescente (testigo) durante 10 días, las variables evaluadas fueron porcentaje de germinación, semillas muertas, plántulas normales y anormales. El análisis de varianza mostró diferencias significativas en germinación, semillas muertas, plántulas normales y anormales en la interacción Genotipo * Luz. La comparación múltiple de medias por Tukey (α=0.05) en la interacción Genotipo * Luz mostró que el espectro rojo tiene efectos positivos en los genotipos SE, X1 y N1 al aumentar el porcentaje de germinación; los tratamientos de luz LED´s morado y azul para los genotipos SE, X1 y N1 incrementaron el número de semillas muertas; con la luz azul en los genotipos M1 y N1 se aumentó el porcentaje de plántulas normales. El tratamiento de luz LED rojo incrementa el porcentaje de la germinación en una 15 %, los tipos de iluminación morada y azul favorecen en el desarrollo de las plántulas de frijol.

Luz emitida por diodos, Plántulas normales, Viabilidad de semillas

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Introduction

Light is essential in the development of plants, the quality, quantity and intensity of illumination that they receive directly influences the photosynthesis process (Bures *et al*., 2018). The spectral composition of the different wavelengths is absorbed through proteins with photoreceptor functions such as phytochromes, cryptochromes and phototropins, which intervene during physiological processes such as germination, vegetative and generative growth (Bian *et al*., 2018).

Currently the use of LED lighting (Light Emitting Diode) has been proposed in the management of automated agricultural environments, with the aim of increasing the yield of crops in stations where outdoor climatic conditions do not allow it, these systems have been named cultivation indoors or indoors. LED lighting in indoor cultivation has shown advantages of use and management compared to high pressure sodium (HPS) and fluorescent lamps traditionally used, due to the fact that they require less electrical energy, do not release heat and increase the profitability of automated systems due to their long useful life (Lu *et al*., 2012).

Indoor cultivation with LED lighting is a technology in the process of being adopted in Mexico, despite being widely practiced in countries of the European Union and the Asian continent. The quality of the products obtained under this production system reports an increase in surface yield and high nutraceutical quality (Amaki *et al*., 2011; Xu *et al.* 2012). Keblawy (2017) defines these responses that increase the nutraceutical and stimulatory quality in processes caused by the artificial light source as photoblastic effects in a positive sense.

In Mexico, the consumption of beans represents an essential food due to the amount of protein and minerals that it contributes to the daily diet of human consumption. However, it has been sought to increase its yield through the modification of sowing dates, even in cultivation under intensive greenhouse systems (Doria, 2010; Bures et al., 2018), but little has been studied on its behavior in environments controlled lighting of light emitted by diodes (LED) in the early stages of the cultivation establishment.

Likewise, little has been documented on the specific responses to the quality and intensity of LED light in the optimization of physiological responses of the cultures put to artificial photoperiods. Therefore, the objective of this research was to study the influence of the photoperiod by LED lighting in red, blue and purple spectra in the germination of four bean genotypes.

Materials and method

The research was carried out in the laboratory of vegetable crops of the National Institute of Forestry, Agricultural and Livestock Research (INIFAP) Campo Experimental (CE) San Martinito (19 ° 14´y 19 ° 28´de N and 98 ° 29 '40´ O), municipality of Tlahuapan, Puebla. The bean genotypes SE, X1, N1 and M1 provided by INIFAP (Figure 1) were used.

The experimental design was a randomized complete blocks and the experiment consisted of standard germination tests using the method between paper (Sanitas ®) as recommended by the International Sees Testing Association (ISTA, 2016). The samples after sowing were distributed within a growth room under controlled conditions at 25 ± 5 ºC with LED lighting in the red, blue, purple spectrum and a fluorescent light (control), at an intensity of 62, 54, 25 and 100 lumens m^2 -1 (calculated with a STEREN® brand lux meter, Model: HER-408) respectively. The photoperiod was 16 hours of light and 8 hours of darkness for 10 days.

Genotype	Description				Image
	P (g)	A (cm)	DT (cm)	DE (cm)	
SE	2.99	0.41	1	0.56	
X1	3.25	0.43	1.01	0.53	
M1	2.38	0.29	0.75	0.44	
N1	2.86	0.62	1.08	0.73	
P = weight; A = area; DT: transverse diameter; DE = equatorial diameter.					

Figure 1 Morphometric characters of genotypes under study in viability tests with photoperiod of LED light.

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After the incubation time, the variables studied were germination percentage (G), dead seeds (SM), normal seedlings (PN) and abnormal seedlings (PA). Prior to the analysis of the variables, the assumptions of normality and homogeneity of variances were checked, after the analysis of variance (ANOVA) was carried out. In the case of the variables that showed significant differences, a multiple comparison of means was performed by Tukey ($p \leq 0.05$), all analyzes were performed with the statistical program SAS ver. 9.4 (Statical Analysis System, 2014).

Results and Discussion

The results of the analysis of variance showed significant effects ($P \leq 0.0001$) between genotypes for the variables G, SM and PA; and there were only significant differences for PN ($P \leq 0.05$). The results of the effect in light did not show statistical differences for any variable. The genotypes were significantly affected by the LED treatments, which showed interaction effects in all the variables (Table 1), a significance that agrees with that reported by Bello et al. (2016) in explants of Vanilla planifolia A.

FV	CM†					
	Mean	Gen	Light	Gen * Light	Error	R^2
G	63.54	10877.36 **	648.53 ns	775.14*	266.16	0.70
SM	35.87	10115.50**	475.19ns	737.51*	253.78	0.70
PN	27.99	1378.160*	284.78 ns	799.83*	249.70	0.43
PA	36.27	5218.97**	836.97 ns	892.52*	274.49	0.59

†CM: mean squares; R2: coefficient of determination; Gen: genotype; G: germination; SM: dead seeds; PN: normal seedlings; PA: abnormal seedlings; *: significant ($p \leq 0.005$); **: highly significant (≤ 0.0001); ns: not significant.

Table 1 Summary of the analysis of variance of the study variables of four bean genotypes and their interaction in germination by photoperiod with LED lighting.

The multiple comparison of means (Figure 1), showed that the X1 genotype presented a higher germination percentage (86.33) compared to the SE, X1 N1 genotypes (Figure 1). This may be related to the dormancy and dormancy of the seeds, which vary according to their genetics, environmental conditions regarding their storage conditions (Heslop and Schwarzacher, 2012) and collection.

In Figure 2 it is shown that the N1 genotype showed almost 40% PN compared to the other three genotypes. Doria (2010) points out that this characteristic shows a relationship with vigor, development and yield during the establishment of crops.

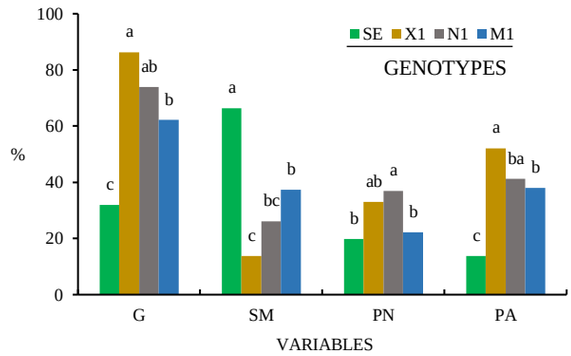

Figure 2 Multiple comparison of means in four genotypes of beans (*P. vulgaris*) in the variables G: germination; YE; dead seeds; PN: normal seedlings and PA: abnormal seedlings.

Although in the analysis of variance there were no significant differences, the multiple comparison of means between LED spectra showed differences only for the variable PA; where the red LED treatment showed malformation in the first true leaves, in addition to a low proliferation of capillary roots in the evaluated seedlings. Nevertheless; In the same red spectrum, although the Tukey test ($p \leq 0.05$), although it did not show differences for the grouping of means, a higher percentage was observed in germination compared to the control and the blue and purple LEDs. According to Valbuena *et al.* (2018), the presence of phytochromes inactivate endogenous processes when perceiving a nearby red-light source promoting the malformation of organs during the development of seedlings due to the specific effect based on the stimulation of germination and not on vegetative development in crops. (Figure 3).

Table 2 shows the multiple comparison of means of the Genotype * Light interaction and the effect on the behavior of the bean genotypes. Red lighting showed beneficial activity in genotypes SE, X1 and N1 by increasing the germination percentage compared to blue and purple light; In this regard, similar responses have been found by Paniagua et al. (2017) in lettuce germination studies where they obtained an increase when they were placed in an environment with a red spectrum LED. From the above, it can be mentioned that in the red spectrum of interaction with bean genotypes a photoblastic effect was observed in a positive sense, as described by Keblawy (2017).

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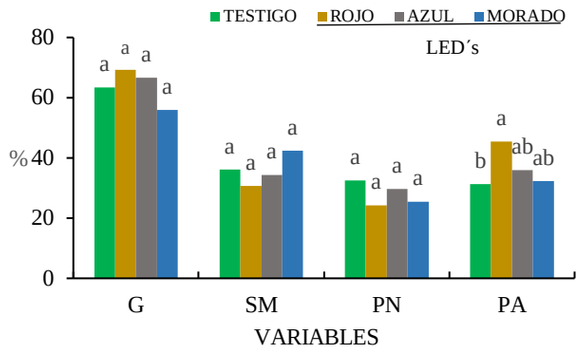

Figure 3 Multiple comparison of LED lighting means in the variables G: germination; YE; dead seeds; PN: normal seedlings and PA: abnormal seedlings.

The effects of the LED lights in the purple and blue spectrum for the genotypes SE, X1 and M1 showed a higher proportion of dead seeds between 3.33 and 18% with respect to the control. This can be attributed to what Bures et al. (2018), who state that purple light affects the viability of seeds by interrupting normal DNA synthesis. Likewise, it has been reported that blue illumination captured by cryptochromes and phototropins regulates stomatal opening and inhibits stem elongation, but does not stimulate germination (Chaves et al., 2011). However, the opposite effect has been reported by Nemahunguni et al. (2020) in Cleome gynandra seeds the increase in germination up to 35%.

Genotype	Witness	LED Red	Blue	Purple
		Germination (%)		
SE	36.67 de	42.66 b - e	24.66 e	23.40 e
X1	96.67 a	93.33 a	92.50 a	60.00 ae
M1	83.33 a	77.22 ab	75.55 ac	69.44 ad
N1	39.33 c e	67.33 ad	73.33 ad	68.66 ad
		Dead seeds (%)		
SE	63.33 a c	57.33 ad	75.33 a	69.33 ab
X1	3.33 e	6.67 e	7.50 e	40.00 ae
M1	16.66 e	22.78 ed	24.44 de	30.55 ce
N1	58.66 ad	32.66 be	26.66 ce	31.33 ce
		Normal seedlings (%)		
SE	25.33 a	18.66 a	12.66 a	22.66 a
X1	47.50 a	39.17 a	22.50 a	33.33 a
M1	44.44 a	16.66 a	47.22 a	30.00 a
N1	13.33 a	27.33 a	31.33 a	16.66 a
		Abnormal seedlings (%)		
SE	11.13 cd	24.00 dc	12.00 c	8.00 d
X1	46.16 ac	48.33 ac	70.00 a	26.66 bd
M1	38.89 ad	66.11 ba	28.33 bd	48.33 ac
N1	28.00 bd	40.00 ad	42.00 ad	41.80 ad
DMS	4.89	4.89	4.89	4.89

DMS *: minimum significant difference; Values with different letters between columns are statistically different ($p \leq 0.05$); G: germination percentage; SM Dead seeds; PN normal seedlings and PA abnormal seedlings.

Table 2 Multiple comparison of means of the study variables of four bean genotypes and their interaction in LED lighting in viability tests.

The variable PN did not show statistical differences (ANOVA); However, the effect observed in the blue light in the M1 genotype was higher than the control by 3.11%. Similar results were reported by Simlat et al. (2016), who mention that they obtained higher quality seedlings of Stevia rebaudiana. Previous studies by other authors on the effect of blue LED light, document responses on the stimulation of seedlings with greater vigor, increases the chlorophyll content, factors that are related to obtaining a higher percentage of normal seedlings (Bello et al., 2016).

Red light increased the number of abnormal seedlings in genotypes SE, M1 and N1 by almost 50% with respect to the control (Table 2). This indicates that the quantity, quality and intensity emitted in the red spectrum are not enough to promote good seedling development (Gonzalias and Ramirez, 2016); in this regard Okamoto et al. (1996) found that the spectrum in red LED light affected the morphology of Spinacia oleracea seedlings by causing malformations.

Conclusions

Treatment with red LED light increased germination by 15% in genotypes SE, X1 and N1, but did not show an effect on the development of normal bean seedlings. The blue spectrum showed a photoblastic effect in the development of morphological characteristics in obtaining normal seedlings, however, during germination, like the blue spectrum, it showed an increase in the presence of dead seeds. From the above, it is concluded that in viability tests for beans the LED spectrum stimulates seed viability with specific responses in red, blue and purple LEDs in germination and the presence of normal seedlings.

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Content of anthocyanins of seed of *Phaseolus vulgaris*

Contenido de antocianinas de la semilla de *Phaseolus vulgaris*

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Abstract

The common bean (*Phaseolus vulgaris* L) is an important food in Mexico that provides both macronutrients and bioactive substances that are beneficial for the health of its consumers, in particular it provides polyphenolic compounds such as anthocyanins that make it a functional food; the objective of the present work was to evaluate the content of polyphenols and their ferric iron reducing activity of beans grown in the Municipality of Hopelchen, Campeche (Mexico) to characterize their agronomic and alimentary value. The morphometric parameters of length, width, thickness, weight and volume were measured; An aqueous maceration of the bean seeds was carried out in an acid medium (0.1 M HCl) and the content of polyphenols (Folin Ciocalteu Method), anthocyanins (visible spectrophotometry) and determination of the Reducing Iron Power (FRAP, by the method of TPTZ) of the aqueous extracts. It was found that the concentration of anthocyanins correlates with the total content of polyphenols and with the reducing activity of the ferric ion, the aqueous extracts obtained had a content of total phenols of 413 mg EAG/100g of seed, of anthocyanins of 42 mg MGE/100 g of seed and an iron reducing power of 10.26 mol of Fe^{2+}/g of seed. The morphometric characteristics and the polyphenol content of the Hopelchén bean demonstrate that the harvested grain is of good quality and corresponds to the expected values for this food.

Functional food, Bean episperm, Phytotherapy, Polyphenols

Resumen

El frijol común (*Phaseolus vulgaris* L) es un alimento importante en México que aporta tanto macronutrientes como sustancias bioactivas que son benéficias para la salud de sus consumidores, en particular aporta compuestos polifenólicos como las antocianinas que lo configuran como un alimento funcional; el objetivo del presente trabajo fue evaluar el contendio de polifenoles y su actividad reductora de hierro férrico del frijol cultivado en el Municipio de Hopelchén, Campeche (México) para caracterizar su valor agronómico y alimentario. Se realizó una maceración acuosa de las semillas de frijol en medio ácido (HCl 0.1 M) y se determinó el contenido de polifenoles (Método de Folin Ciocalteu), antocianinas (espectrofotometría visible) y determinación del Poder Reductor de Hierro (FRAP, por el método de TPTZ) de los extractos acuosos. Se encontró que la concentración de antocianinas se correlaciona con el contenido total de polifenoles y con la actividad reductora del ión férrico, los extractos acuosos obtenidos presentaban un contenido de fenoles totales de 413 mg EAG/100 g de semilla, de antocianinas de 42 mg MGE/100 g de semilla y un poder reductor de hierro de 10.26 mol de Fe^{2+} / g de semilla. El contenido de polifenoles del frijol de Hopelchén demuestra que el grano cosechado es de buena calidad y corresponde a los valores esperados para este alimento.

Alimento funcional, Episperma de frijol, Fitoterapia, Polifenoles

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Introduction

The common bean (*Phaseolus vulgaris* L.) is an essential part of the Mesoamerican diet; Currently it is sown in traditional and modern production systems that lead to morphological, physiological and genetic changes in this crop; Beans are important as a nutritional element because they provide macro and micronutrients of interest for health and are a good source of macronutrients (protein and carbohydrates) and provide vitamins and minerals in relatively adequate amounts (Hoc *et al*., 2002. Camacho *et al*., 2007 Hernández-López *et al*., 2013. Puertas-Mejía *et al*., 2013. Lams *et al*., 2021).

The bean testa has various components to which antioxidant properties are attributed, in this group of secondary metabolites the polyphenolic compounds stand out, particularly anthocyanins, flavonoids and tannins that play an important role in the prevention and treatment of certain diseases. (Iniestra *et al*., 2005. Camacho *et al*., 2007. Puertas *et al*., 2016. Sánchez-Toledano *et al*., 2021).

Anthocyanins are very soluble in water and have bright colors, have an antioxidant activity that inhibits free radicals, preventing diseases such as cancer, atherosclerosis and inflammation, and they are also valued for their coloring power; For this reason, it is assumed that the bean contributes synergistically with its medicinal properties as an antioxidant, diuretic, anti-inflammatory, antitumor and antimicrobial and with a possible positive effect against some chronic diseases (Salinas-Moreno *et al*., 2005. Camacho *et al*., 2007. Puertas-Mejía *et al*., 2013).

However, information on the agronomic and food value of the common bean is scarce and scattered, despite its great ecological and economic importance. For this reason, there is a need to know the variation and phytochemical diversity of beans to define strategies for the rescue, conservation and use of native populations; since the macro and micronutrient content, in particular the anthocyanin concentration, varies according to the cultivation conditions and the sowing location (Cruz *et al*., 2009. Celis-Velázquez *et al*., 2010. Hernández-López *et al*., 2013 Barrios *et al*., 2014. Casasola *et al*., 2021).

The present investigation was carried out with the objective of knowing the anthocyanin concentration of common bean seeds collected in the Municipality of Hopelchén of the State of Campeche, in the Mexican southeast.

Methodology

Bean lots collected in the Hopelchén region, Campeche State (Mexico) were used; the seeds obtained were dried at room temperature and stored in plastic containers for approximately three months at 4 °C; for subsequent chemical analysis. The aqueous extracts of beans were obtained by static maceration, for this, 10 g of seed were deposited in 250 mL beakers and 100 mL of sterile distilled water was added, at intervals of 1, 2, 3, 4, 5, 6. At 7 and 8 h, aliquots of the supernatant were taken for the determination of total polyphenolic compounds by the Folin Ciocalteu method. The technique was carried out as follows: 100µL of the extract was added to 500µL of water in a test tube and then 100µL of the Folin-Ciocalteu reagent was added, it was left to react for 30 minutes and later 500□L of Na2CO3 was added. 20%, incubated at room temperature for 30 min. Finally, it was read in a spectrophotometer at 760 nm. A calibration curve was made with gallic acid to determine the concentration of polyphenols present in each extract.

For the quantification of anthocyanins, the absorption spectrum of anthocyanins from beans was first obtained in a Lambda XLS + ® spectrophotometer; Subsequently, the anthocyanins were extracted with 0.1 M HCl for ten replications, for this the weight of the bean seeds (1, 2, 3, 5 and 10 seeds) was recorded and they were deposited in conical tubes with a lid. to which 50 mL of the HCl solution was added, it was left to rest for 24 hours at room temperature and finally the solution was filtered to recover the supernatant. The absorbance of the acidic solution was measured at 540 nm and the anthocyanin concentration was estimated by the following formula:

$$Anthocyanins\left(\frac{mg}{L}\right) = A_{540nm} * 16.7 \qquad (1)$$

Where A_{540nm} is the absorbance of the acidic solution at 540 nm.

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Likewise, the acid extracts were determined antioxidant reducing power of the ferric ion (FRAP, for its acronym in English) using the reagent TPTZ (2,4,6-tripyridyl-S-triazine); First, the FRAP reagent was prepared by mixing 25 mL of 300 mM sodium acetate buffer, 2.5 mL of 10 mM TPTZ (2,4,6-tripyridyl-S-triazine) solution and 2.5 mL of 20 mM ferric chloride solution. Subsequently, 100 µL of the bean extract was added to 1000 µL of a freshly prepared solution of the FRAP reagent, it was mixed very well, it was left to react for 60 minutes and the ferrous-tripyridyltriazine complex (Fe^{2+}-TPTZ), formed by the reduction of the ferric-tripyridyl triazine complex (Fe^{3+}-TPTZ), at 590 nm in a spectrophotometer; A calibration curve was made with ferrous sulfate as a standard.

The statistical analysis of the data was carried out in Excel® and in the statistical software SPSS V25.0 ® of each morphometric variable, search in the bean grains to describe the population [graphs of descriptive statistics, mean (X), standard deviation (SD), maximum value (Max.) and minimum value (Min.)], subsequently a correlation matrix was made between the morphometric parameters obtained and a linear correlation analysis to obtain the Pearson coefficient and the equation of the line. For the determination of total polyphenols and Ferric ion reducing antioxidant activity (FRAP), a calibration curve was made by a linear correlation analysis and determination of the equation of the line, from which the values of the test sample were estimated; results are reported as the mean ± one standard deviation.

Results and discussions

Regarding the content of polyphenols in the bean seed, it is shown first, in Figure. 1, the kinetics of polyphenol extraction with pure water. A maximum of polyphenol extraction was reached at eight to 25 ºC, it is known that the extraction of polyphenolic compounds from beans increases with increasing temperature and using solvents such as methanol as well as using techniques such as microwave or ultrasound assisted extraction; The chemical characterization of the polyphenol content is important because the different varieties of *P. vulgaris* significantly appreciate their functional properties, especially the antioxidant activity (Puertas-Mejía *et al*., 2013. Puertas *et al*., 2016).

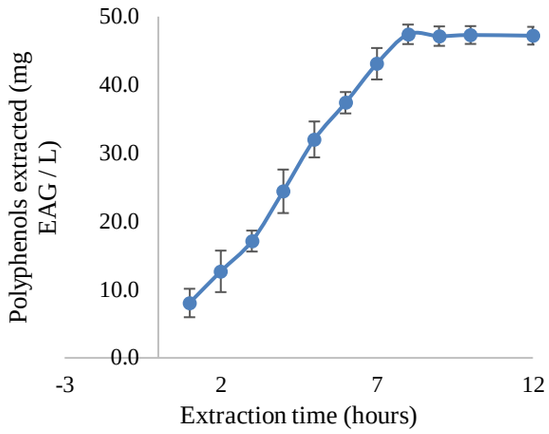

Figure 1 Kinetics of extraction of total polyphenols from bean seeds with distilled water (n = 10)
Source: Data obtained in the investigation

The content of total polyphenols and anthocyanins as well as the FRAP of the acidic aqueous extracts of the bean seed are reported in Table 1; the amount of total polyphenolic compounds extracted corresponds to the values reported for various bean varieties (Reynoso *et al*., 2007. Ombra *et al*., 2016. Puertas *et al*., 2016. Silva *et al*., 2018. Weidner *et al*., 2018). In the same way, the amount of anthocyanins determined in the bean samples analyzed coincide with those reported in the literature; Salinas-Moreno *et al*., (2005) determined that regardless of the racial origin, the black varieties had high contents of total anthocyanins in the whole grain with values between 37.7 and 71.6 mg / 100g; the content of polyphenols present in bean seeds depends on the geographic origin of the populations, environmental variations and genotype, so it is necessary to quantify the polyphenolic compounds in seeds of different varieties as this helps to determine which bean cultivars would provide the greatest benefits for its consumers (García-Díaz *et al*., 2016. Silva *et al*., 2018. Weidner *et al*., 2018).

Total polyphenols	413 ± 28 mg EAG / 100g of seed
Antocianinas	42±3 EMG/ 100 g de semilla
FRAP	10.26±0.58 de Fe^{2+}/g de semilla

Table 1. Polyphenol and anthocyanin content and ferric iron reducing activity of the acidic aqueous extracts of the Hopelchén bean seed. Results reported as X ± SD, EAG = gallic acid equivalent, EMG = malvidin glucoside equivalent, n=50
Source: Data obtained in the investigation

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The concentration of anthocyanins extracted with 0.1 M HCl increased linearly and proportionally to the amount of bean seeds used in the maceration (Figure 2), this allows us to assume that the system was not saturated and that the amount of anthocyanins extracted was a function of the quantity of seeds as a limiting factor (Table 2); Likewise, it is observed that there was a greater extraction of anthocyanins when HCl was used compared to the neutral aqueous extracts; since anthocyanins are highly soluble substances in water that contain in their structure a pyryllium ring in an acid medium, they are found in the form of salts that increase their solibility and facilitate their extraction (Astrid, 2008. Puertas-Mejía *et al*., 2013).

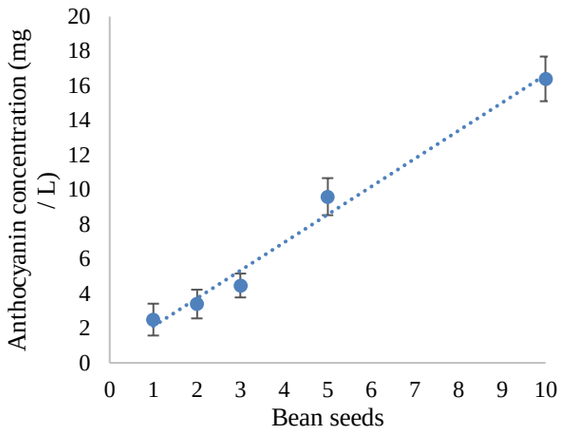

Figure 2 Anthocyanin concentration in 50 mL 0.1M HCl extract with 1,2,3,5,10 bean seeds (n = 10)

Number of seeds	mg of Anthocyanins / 100 g of seeds
1	42.7±4.6
2	42.5±4.1
3	41.9±3.5
5	44.2±4.2
10	41.0±3.4

Table 2 Anthocyanin content extracted from bean seeds with 0.1 M HCl. Results reported as X ± SD, no significant differences are observed (p <0.05), n = 10
Source: Data obtained in the investigation

With the values obtained by the determination of anthocyanins and the value of the FRAP, a correlation analysis was carried out that is shown in figure 3, it can be observed that the antioxidant activity (FRAP) is linearly correlated with the concentration of anthocyanins present in the extracts, that is, the anthocyanin content explains the antioxidant activity very well (R^2 = 0.9831).

This coincides with what is reported in the scientific literature that indicates that the therapeutic effects of anthocyanins are related to their antioxidant activity because they are effective to trap reactive oxygen species, inhibit lipoprotein oxidation and reduce metal ions (García-Díaz *et al*., 2016 Puertas *et al*., 2016. Weidner *et al*., 2018. Chí-Sánchez *et al*., 2021). One of the most used methods to determine the antioxidant activity of polyphenolic compounds is the FRAP; but it must be considered that in addition to polyphenols there are other compounds such as vitamins (C and E) and carotenoids that have antioxidant activity and may be present in extracts (Puertas *et al*., 2016. Silva *et al*., 2018. Weidner *et al*., 2018); in this case, the high correlation between FRAP and the amount of anthocyanins suggests that these are the main metabolites responsible for the antioxidant activity of the aqueous extracts of *P. vulgaris.*

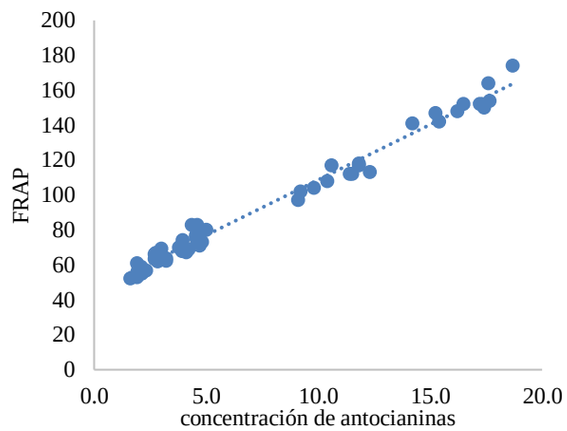

Figure 3 Correlation between anthocyanin concentration and FRAP
Source: Data obtained in the investigation

Conclusions

The morphometric characteristics and the polyphenol content of the beans from Hopelchén, Campeche (Mexico) show that the harvested grain is of good quality and corresponds to the expected values for this food; the antioxidant capacity of bean seeds correlates well with the amount of polyphenolic compounds, in particular with the anthocyanin content.

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Physicochemical analysis in *Averrhoa carambola* L., var. Golden star and Arkin, in two post-harvest periods

Análisis físico-químico en *Averrhoa carambola* L., var. Golden star y Arkin, en dos estadios post-cosecha

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Abstract

Golden Star (GS) and Arkin (Ar) are important varieties of carambola fruits cultivated in México. Fruits were collected in a plantation of Jalisco and their physicochemical characterization was performed either in fresh or lyophilized fruits in two post-harvest time-points: immediately after harvest (IH-0) and 10 days post-harvest (PH-10), at room temperature. In IH-0, in GS the content of starch, glucose and total reducing sugars (TRS) was higher, while fructose was reduced. At PH-10, the size of GS fruits decreased, whereas an increase in total soluble solids and acidity in Ar fruits contrasted with a reduced pH and TRS content. Non-structural carbohydrates (NSCs) increased, from IH-0 to PH-10 in both varieties. Pectinolytic activity was highest in GS and Ar at PH-10, as was amylolytic activity. However, both activities were higher in Ar. Lyophilization significantly decreased the protein and starch contents, particularly in PH-10 fruits, whereas NSCs increased considerably. These results indicated a contrasting post-harvest behavior between the two varieties. The reported findings could be used to improve post-harvest management of carambola fruits.

Carambola, lyophilization, post-harvest ripening

Resumen

Golden Star (GS) y Arkin (Ar) son variedades de carambola importantes en México. Se colectaron frutos en una plantación en Jalisco, México, y se caracterizaron fisicoquímicamente, tanto en fresco y liofilizado como en dos estados post-cosecha: corte inmediato (CI-0) y después del corte (DC-10) a temperatura ambiente. En CI-0, en GS el peso, el contenido de almidón, glucosa y azúcares reductores totales (ATR) fue mayor; pero menor de fructosa. En DC-10, GS disminuyó el tamaño y Ar incremento en sólidos solubles totales y acidez, con una disminución del pH y ATR. El contenido de carbohidratos no estructurales (CNE) aumentó, de CI-0 a DC-10 en ambas variedades. La actividad pectinolítica fue más alta en GS y Ar a DC-10, al igual que la actividad amilolítica. No obstante, ambas actividades fueron mayores en Ar. La liofilización disminuyó el contenido de proteína y almidón, principalmente en DC-10, mientras que los CNE aumentaron considerablemente. Estos resultados indicaron un comportamiento post-cosecha contrastante en estas variedades. Los hallazgos reportados podrían usarse para mejorar el manejo post-cosecha de la carambola.

Carambola, liofilización, madurez postcosecha

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Introduction

Carambola (*Averrhoa carambola* L.), an important member of the family Oxalidaceae, is a tropical shrub introduced in Mexico. The site of origin is not clearly defined. However, it is considered endemic to Indonesia (Nakasone and Paull, 1999; Orduz and Rangel, 2002) or Malaysia and Indonesia (Zeven and De Wet, 1982; Watson et al., 1988). It is distributed in Mexico, at tropical and sub-tropical regions, mainly in the states of Colima, Chiapas, Guerrero, Michoacán, Morelos, Nayarit, Sinaloa, Jalisco, Tabasco and Veracruz. Here, two introduced varieties are mostly cultivated: Golden Star and Arkin, (Romero-Gómezcaña, 2001; Aundón, 2006; Cruz and Garza, 2006). Having originated in Florida, USA, they both have a medium size and a color palette in which yellow and orange tones predominate. However, Arkin is characterized for having a mayor sugar content and less acidity than Golden Star (Campbell et al., 1985; Morton, 1987; Knight, 1989; Lamberts and Crane, 1990; Knight Jr. and Crane, 2002).

The fruit is very attractive because of its organoleptic, energetic and nutritional characteristics, which favor its traditional consumption. It can be consumed as fresh fruit or as processed as juice, although it may also be used to make fresh or fermented beverages, jelly, pastries, preserves, or dehydrated powders, among others. This flexibility confers a high economic potential to these fruits (Lim, 2012). Diverse medical properties are attributed to them, mainly related to antioxidant, hypoglycemic and anti-inflammatory properties (Dembitsky et al., 2011; Lim, 2012, Pantaleón-Velasco et al., 2014). For example, it is used in Western, Mexico as an empiric coadjutant for the treatment of Diabetes Mellitus type II (MD2). Their recognized hypoglycemic activity (Cazarolli et al., 2009; 2012), has great public health potential, considering that MD2 has become a serious worldwide health problem, mainly in developing countries, such as Mexico (Vázquez and Panduro, 2001; Olaiz-Fernández et al., 2006; Salcedo et al., 2008; Shamah-Levy et al., 2020).

Studies of the physicochemical characterization of carambola are scarce as well as nutritional information in the context of medical (Calzada, 1980, Pérez-Tello et al., 2001; Tello et al., 2002; Ding et al., 2007; Novillo, 2009).

Besides, the data available in this respect is highly variable, while most recent studies about this fruit, have only focused on the physiological analysis of the different maturity states of the carambola, without specifying the variety (Narain et al., 2001) or have concentrated on the Golden Star variety only (Siller-Cepeda et al., 2004). Respiratory and compositional data, of both fruit varieties, have been generated, but only in fruits harvested at physiological maturity (O'Hare, 1993). Nevertheless, studies designed to determine more precisely the post-harvest behavior of these fruits, are needed, mostly at room temperature conditions and including the two varieties with mayor commercial importance in Mexico and North America, such as Golden Star and Arkin. Therefore, one of the aims of this investigation was to generate physicochemical information of Golden star and Arkin carambola fresh fruits in two well defined post-harvest states: just after harvest, and ten days post-harvest, at room temperature. It is worth noting that in this investigation, the fruits analyzed were collected directly in a plantation located in the state of Jalisco, unlike many other studies, that have used fruit purchased in markets or convenience stores. Also, a physicochemical characterization of lyophilized fruits of these two varieties was performed in the two post-harvest periods mentioned. The latter based on findings that have reported that this drying process is capable of affecting the composition, and/or the biological activity of various plant tissues or plant-derived extracts, including the carambola fruit (Shofian et al., 2011). The information proportioned in this study contributes to a more defined knowledge of the post-harvest behavior at room temperature of these important Carambola fruit varieties and provides information regarding the effect that lyophilization has on the levels of important nutritional components. The data proportioned could serve as a guide for the correct post-harvest management of these varieties at room temperature. Also they suggest that the pointed differences between the studied varieties, their differing post-harvest storage behavior or the lyophilization effect, could have implications in their medical use, mainly as an antioxidant, hypoglycemic or anti-inflammatory agent.

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Materials and methods

Description of the study site, sample collection and preparation of plant material

For the collection of plant material, the phenology at maturation time, harvest and ingestion was considered. The fruits selected for analysis were those having a mature appearance, with the predominance of golden-yellow pulp color. A total of 120 carambola fruits, var. Golden Star and Arkin, were collected randomly in a commercial plantation established in the municipality of Cihuatlan, Jalisco, located at southwest end of the state (19° 22' 30" N, 104° 42' 30" W, at 13 m.a.s.l). The fruits were transported cold to the laboratory. Once in the laboratory, the fruit specimens chosen to determine the post-harvest effect were kept at room temperature during 10 days, before being stored at -20 °C. Thirty Golden Star and 30 Arkin fruits were used for each time of storage, i.e., immediately after harvest (IH-0) and after a 10-day post-harvest period (PH-10). Fruits were washed and blended in an electric blender (Taurus-Group, Barcelona, Spain). The crushed product was collected and stored at -20 °C, for their latter physicochemical analysis, which was performed after a maximum period of 10 days. Part of the triturated tissue was freeze-dried using a Labconco 77520 lyophilizer (Labconco, Kansas City, MO, USA).

Determination of size and weight of the fruit

The size of the collected fruits, along the longitudinal and radial axis, was measured using a millimetric Vernier caliper. The individual weight of each fruit (in g), was registered in an analytical balance (Ohaus, Parsippany, NJ, USA).

Determination of the Humidity content

For the determination of the humidity content, the fresh weight of the complete fruits was registered. Then, they were dried at a constant temperature of 130 °C during 48 h. The percentage of humidity was calculated by the method of Nollet (1996), taking into consideration the lost weight of the sample during drying.

Determination of pH, Total Soluble Solids content (TSS) and acidity (%)

The pH was measured immediately after crushing of the carambola fruit, using a manual pH-EC-TOS METER potentiometer (Hanna Instruments, Woonsocket, RI, USA). The Total Soluble Solids content (TSS) was determined using the method described by the Association of Official Analytical Chemistry (AOAC, 1999). Briefly, 0.3 g of carambola fruit tissues were weighed to prepare an aqueous extract (1:5 w/v), aliquots of which were placed in a manual electronic refractometer, (Atago Pocket Refractometer PAL-1 (0~53%/ 1000mL; Atago, Tokyo, Japan). The TSS were expressed as percentage in °Brix. The percentage of acidity was determined by the method described by the Association of Official Analytical Chemistry (AOAC, 1999), except that the acidity of the aqueous extracts was determined using a manual electronic refractometer, (Atago Pocket Acidity Meter: Citric Acid; Atago PAL ACID-1), range 1~40g/ 1000mL; Atago Tokyo, Japan).

Extraction and determination of the protein content

The protein content was determined using the Bradford method (1976), which is based on the interaction of the Comassie G250 blue pigment with proteins. Thus, aqueous extracts were prepared, using 20 mg of fresh and 50 mg of lyophilized tissues, due to the different characteristics of the samples tissue, in 400 µL of distilled water. These were stirred for 10 min. at 4 °C and centrifuged at 12,000 rpm for 10 min. Ten µL aliquots of the supernatant were added to 200 µL of the Bradford reactive and the color produced was read at 465 nm.

Determination of Starch content

For starch content determinations the Wright et al. (1998) and Geigenberger et al. (1998), methods were employed, with some modifications. To 200 mg of fresh tissue or 50 mg of lyophilized tissue, 500 µL of extraction buffer was added; the mix was stirred and centrifuged at 12000 rpm for 10 min. The supernatant was collected, and the pellet was extracted two more times, repeating the above steps.

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The combined supernatants were dried in a vacuum concentrator system Maxi-dry (Maxi-Dry Lyo; Heto-Holten, Denmark) during 24 h and suspended in 500 µL of Hepes buffer, pH 7.5 for the determination of soluble NSCs (see below). The starch-containing pellet was suspended in 500 µL of distilled water and incubated for 2 h in a water bath at 95 °C. Later, the tubes were covered with aluminum foil, and were allowed to cool for 5 min. Then, 500 µL of the reactive solution was added (0.8 mg α-amylase type VI-B from porcine pancreas [Sigma-Aldrich, St. Louis, MO, USA] and 0.34 mg de amyloglucosidase from *Aspergillus niger* [Sigma], in 50 mM Hepes buffer, pH 7.5) and incubated in a water bath at 55 °C during 24 h. The reactive mixture was centrifuged at 12000 rpm for 10 min. and aliquots of 10 to 15 µL were taken from the supernatant, which were placed in a 96-well microplate, adjusting the volume with 50 µL of distilled water. The plate was incubated at room temperature during 45 min and the absorbance was read at 460 nm. The content of starch is expressed in µmol eq of glucose/ g DW.

Extraction and determination of glucose, fructose and sucrose

The samples derived from the extraction of starch, were diluted 1: 20 v/ v and 5 to 10 µL aliquots were placed in a 96-well microplate. Then, 10 µL of extract and 200 µL of the reactive solution for sugars (0.1 mM NADP+, 0.2 mM ATP, glucose-6-phosphate-dehydrogenase [G6PDH, from yeast grade II, Roche Life Science, Indianapolis, IN, USA] in 50 mM of Hepes buffer, pH 7.5) were combined. The plate was stirred 10 seconds and 5 absorbance readings at 340 nm were realized (This is the blank absorbance). The glucose determination was performed adding 2 µL de hexokinase (Roche) after which the absorbance read at 340 nm each two min., till the absorbance value was constant. For the determination of fructose, the plate was removed from the lector and in each pool 2 µL of phosphoglucose-isomerase from yeast (Roche) was added and the absorbance read at 340 nm each two min., till the value of absorbance was constant. Finally, for the determination of sucrose, the plate was again removed from the lector and in each pool 4 µL of invertase was added (Invertase grade VII, from Bakers yeast, Sigma). Each of these non-structural carbohydrates was expressed in µmol/ g DW).

Determination of the amylolytic and polygalacturonase activity

Both activities were measured *in vitro* using lyophilized tissues from carambola fruits corresponding to the IH-0 and PH-10 post-harvest treatments, respectively. The amylolitic activity was measured according to the Bernfeld (1955), while the polygalacturonase activity was determined using the method described by Gross (1982). Both methods were modified for their use in microplates.

Results

Physical characterization of carambola fresh fruit and after storage at room temperature

Notable differences in the physical parameters determined in the two carambola varieties were detected. In fruits analyzed immediately after harvest (equivalent to the point of maturity, when the transition of color goes from green to yellow) or IH-0, the average weight of the Golden Star (GS) and Arkin (Ar) varieties were 91.93 g and 75.01 g, respectively. However, a notorious weight-loss occurred in GS fruits analyzed 10-days post-harvest at room temperature, or PH-10. This effect was not so pronounced in Ar fruits (Table 1). A similar tendency was observed in the fruit size measures made, although the differences were not so marked between the IH-0 and PH-10 treatments.

Compared to AR, the longitudinal axis at IH-0 was larger in GS fruits. However, and similar to the above parameter, longitudinal fruit size was considerably reduced in the GS variety at PH-10, but not in Ar fruits (Table 1). A similar pattern was observed when the radial axis the of the fruits of both varieties was measured at PH-10. Thus, this parameter was reduced significantly in GS, but not in Ar, fruits (Table 1). Conversely, no differences in the longitudinal and radial axes the of the fruits of both varieties subjected to IH-0 were detected. On average, the fruit of both varieties (at both times of harvest), presented a diameter of 4.94 cm, a longitude of 8.31 cm and a weight of 67.84 g. The data indicated on Table 1 also show that the humidity content in GS fruits remained constant between stages IH-0 and PH-10, whereas in Ar fruits it changed significantly.

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The activity of polygalacturonase enzymes, capable of softening the fruit cell walls, is shown in Graphic 1A. It indicates that a significant increase in polygalacturonase activity was similarly produced in GS and Ar fruits subjected to the PH-10 treatment. This finding discards the possibility that the differences observed in the physical integrity of the fruits of both varieties were related to loss of cell wall integrity factor. Nevertheless, a more extensive and detailed study of other cell-wall related enzymatic activities is suggested in future carambola fruit experiments. The latter, considering the obvious physical changes that the fruits of the GS and Ar varieties underwent at the different post-harvest times examined.

Chemical characterization in carambola fresh fruit and the effect of storage at room temperature

No significant differences in the pH of fresh fruits of the GS and Ar varieties was observed at IH-0. However, the pH was slightly reduced, by approximately 0.5 units, in fruits of both varieties subjected to PH-10 (Table 2). The acidity content remained stable in GS fruits, while it increased in PH-10 Ar fruits (Table 2), thereby coinciding with the pH values detected. The TSS concentration (in °Brix) was similar in both varieties of fruit at IH-0. However, at PH-10 the TSS increased significantly in Ar fruits, while they decreased slightly in GS fruits (Table 2).

Differences in protein content were also observed. Higher values were detected in Ar fruits at IH-0. However, a 21% decrease was produced at PH-10 while a contrary behavior was observed in GS fruits, where a 10% increase in protein concentration was observed (Table 2). Compared to Ar fruits, the average starch contents were significantly higher in GS fruits at IH-0. These were not modified by the PH-10 treatment (Table 2). The results displayed in Graphic 1 B suggest that the differences in starch levels observed between these two varieties could be associated with differences in amylolitic activity levels, which were lower in GS fruits, predominantly at PH-10. In addition, the PH-10 treatment also significantly affected the levels of soluble NSCs that, in general, tended to increase, except for sucrose, whose levels increased slightly in GS and not at all in Ar. Most noteworthy was the 1.7-fold increase in glucose levels observed in GS fruits after PH-10 (Table 2).

Effect of drying by lyophilization on the chemical composition of the carambola fruits in two states of maturity

In this work, the effect of lyophilization, or freeze-drying, on the chemical composition of GS and Ar fruits, at IH-0 and PH-10, was also studied. As shown in Table 3, this process had a drastic effect on all components analyzed, which was dependent on both the fruit variety and storage treatment. In GS fruits, the protein levels decreased markedly in lyophilized tissue predominantly at PH-10, where the reduction of protein content was almost of 75% lower in the lyophilized tissues. The lyophilization effect on the protein content in Ar fruits was only observed al PH-10, where a 40% reduction in the protein content of lyophilized tissues was observed (Table 3).

A drastic fall was also detected on the starch levels of, particularly in GS fruits and especially, at PH-10. A similar tendency was observed for Ar fruits (Table 3). In contrast, the freeze-drying process drastically increased the soluble NSCs content, which registered a ca. 10- to 15-fold increase of the levels detected in fresh fruit tissues. The effects on NSCs content observed depended on the NSC type, the fruit variety and the post-harvest treatment. In this regard, glucose and fructose content in Ar followed the same pattern observed in fresh tissue, where the highest values were detected at PH-10.

Discussion

Physical characterization of fresh fruits (IH-0) and after storage at room temperature (PH-10)

Carambola GS fruits retained their characteristic ovoid or ellipsoidal shape despite having undergone the most drastic changes in the axial axis as a result of the post-harvest period imposed (Daza et al., 1998; Cubillos and Isaza, 1999; González, 2000). This effect was probably due to the predominance of the axial longitude over the radial longitude, as observed in Table 1. However, it is important to mention, that the longitude of the GS fruits examined in the present study was superior to the one registered by other authors, where the axial longitude was between 6 to 12 cm, and the radial, from 3 to 6 cm (León, 2000).

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In addition, the longitudinal and radial axes reported by Palacios and Rodríguez (2001) were 7.46 cm and 4.56 cm, respectively similar to those found by Siller-Cepeda et al. (2004). The average diameter (4.9 cm), longitude (8.3 cm) and weight (67.84 g) of the GS and Ar fruits were similar to those reported by Narain et al. (2001), but contrary to the data presented by Pérez-Barraza et al. (2005).

The marked differences in fresh weight, in addition to the dimensional changes of the GS fruits that occurred at PH-10 could have been a direct consequence of their severe dehydration, considering that carambola fruits are known to suffer significant water losses when stored at room temperature. For example, Pérez-Tello et al. (2001) reported considerable water losses in fruits of the "Yau" variety, stored for 30 days in controlled environment chambers at 20 °C from day five on.

The maintenance of water content in carambola fruits is considered an important post-harvest quality factor, since dehydration can lead to a surface browning produced by the oxidation of the epidermal cells of the fruit (Nakasone and Paull, 1999). For this reason, they are normally covered with a wax coat or a polyethylene film, or are maintained under refrigeration. These precautions are designed to avoid, besides a drastic weight-loss, the generation of undesirable colors or their deformation, particularly due to degradation effects at the edges of the fruits (Vines and Grierson, 1966; Sanchez, 1990; Wiley, 1994; Thompson, 2006).

The drastic changes in weight and the water loss in Ar fruits, could be attributed to fluctuations in polygalacturonase activity levels. However, this factor was discarded, since the results showed a significant increase in polygalacturonase activity (Graphic 1A) in both GS and Ar fruits at PH-10. However, the negative effects observed coincided with similarly deleterious post-harvest damages reported in carambola fruits conserved at room temperature (Prado et al., 2005) and in other perishable fruits, like guayaba, (Singh and Chauhan, 1982; Patel, 2002; Rodeo et al., 2018) and grapefruit (Castro et al., 1999).

In this respect, it is pertinent to mention that polygalacturonases, related pectinolytic enzymes and other enzymes capable of degrading the cell wall (e.g. celluloses and hemicelluloses) were initially purified and analyzed in carambola fruits of an unspecified variety at an optimum maturity stage (Kwek and Ghazali; 1986; Ghazali and Leong, 1987) and in response to different storage conditions (Chin et al., 1999; Ali et al., 2004). A later study did not detect differences in polygalacturonidase (PG) activity in slices of carambola fruits of the Fwang Tung variety sampled at two different maturity stages (e.g. green-yellow and totally yellow) (Teixeira et al., 2005). However, another subsequent study found that PG activity increased considerably during the storage of fruit slices of the same variety, despite of the use of packages designed to maintain them in a controlled environment (Teixeira et al., 2007), similar to what was previously observed by Chin et al. (1999). In addition, Teixeira et al. (2012), reported the presence of soluble pectin derived from a greater activity of PG and pectoliase was identified as one of the components responsible of the reduced quality during storage of carambola fruit slices of an Israeli variety at its commercial maturity point. For the above reasons, it is pertinent to argue that a more thorough analysis of these crucial enzymes in the GS and Ar varieties should be contemplated in future experiments with these carambola fruits. In this context, the activity of these enzymes should be kept at a minimum, contrary to some other applications where active pectolytic enzymes are needed, such as in the clarification of carambola wine (*Averrhoa carambola*) at the start of the fermentation process or in the production of beer using other crops such as rice (Magadama-Ramírez, 2021). In this respect, the agronomic management of carambola fruit cultivation could be an important factor to be explored, considering a recent study that reported a considerably improved after-harvest life of carambola fruits resulting from the application of calcium hydroxide to the soil. This effect was attributed to their higher calcium levels, which was thought to contribute to the strengthening of their respective cell walls due to the modified structure and solubility of the pectin of the middle lamella that defines their firmness. The benefits of a greater firmness are associated to a minor water loss, a reduced respiration rate and lower susceptibility to infection by opportunistic microbes, (Prado et al., 2005).

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No change in pH values were registered in fresh carambola fruits. However, the pH in Ar fruits was found to be significantly reduced at PH-10. This tendency was not in accordance with the study of Narain et al. (2001), which found that the pH of carambola fruits at commercial maturity was higher and increased during the ripening process. Moreover, the pH values registered in GS and Ar fruits where similar to those reported in the green fruits analyzed in this study. On the other hand, the pH values recorded in the present study were similar to those reported in carambola fruits in Honduras by Novillo (2009), which had a pH that oscillated between 2.22 to 2.27, and in Malaysia by Ding et al. (2007) where the fruits had pH value within the 2.3 to 3.8 range.

Acidity was constant in GS fruits, contrary to Ar fruits in which this parameter increased after harvest. This behavior did not correspond to what has been generally observed during carambola fruit storage. For instance, a storage-related decrease in acidity was observed (Campbell et al., 1987) which, if pronounced, was found to negatively affect the sensorial perception of the fruit, making it tasteless (O'Hare, 1993). The reason(s) of the discrepancy between the acidity data of the present study and those normally reported for carambola fruit, remain(s) to be determined. However, the fact that the acidity levels in carambola fruits can widely vary between individuals is a point to ponder (Wilson et al., 1982; Campbell and Koch, 1989). Also, to consider is that the acidity levels are critically dependent of the harvest time stage, since the organic acid levels in the fruits usually increase in relation to the time spent attached to the tree (Wills et al., 1990; León, 2000). Furthermore, it should be mentioned that the acidity recorded in Ar fruits at PH-10 was an unexpected result, taking into account that this variety is considered to be sweet and to have a less acid character (Knight Jr. and Crane, 2002). In contrast, the slight reduction in acidity detected on GS fruits at PH-10, could be related, at least in part, with the loss by oxidation of vitamin C, (Lee and Kader, 2000), a phenomenon that also coincided with the natural cracking of the fruit.

The increased TSS levels in Ar fruits at PH-10 was comparable to findings in other studies (Daza et al., 1998; Tello et al., 2002; Siller-Cepeda et al., 2004; Pérez-Barraza et al., 2005), although the reported increment in TSS during ripening was accompanied by a reduction in acidity (Narain et al., 2001) or, curiously, was affected in oranges by previous application of chitosan (Zapata-Farroñan and Sunción-Guevara, 2021). In this sense, several authors have reported that TSS levels remain almost constant in carambola fruits during storage (Wan and Lam, 1984; Campbell and Koch, 1989; Neog and Mohan, 1991). However, similar to what was observed in this work, variations in the TSS content have been reported to depend on the variety and/or the time elapsed after the harvest (Wagner Jr. et al., 1975; Crane, 1993). It is important to emphasize that the TSS content is an indicative of the sugar content of the fruit and constitutes an important factor influencing the flavor of the juices, jellies, besides being an important quality control characteristic (Knee, 2002).

Storage at room temperature produced changes in the protein content of both GS and Ar fruits at the two storage periods examined. This behavior coincided with reports from Calzada (1980) and Pérez-Tello et al. (2001), in which the protein concentration of ca. 0.5 mg/ mL that was registered in carambola fresh fruits, decreased during maturation, possibly due to a concomitant reduction in the content of peroxidases, polyphenol oxidases and catalases (García et al., 2006). On the other hand, Patil et al. (2010) found, that the protein content in carambola fruits increased from 0.65% (in "young" fruits) to 0.85% (in "mature" fruits), a finding that suggested that the protein concentration could vary in relation to the time of harvest and time spent in storage. In relation to other fruit species, the protein content detected in carambola fruits analyzed in this study was slightly lower than those reported in guava (Soria et al., 2003; Athayde Uchôa-thomaz et al., 2014), apple (Bordeleau et al., 2002), pineapple (Krayn, 2006) and orange (Rodrigo and Zacarías, 2007). Notwithstanding, they were higher than those found in babaco (Araujo-Ramírez, 2021).

The high content of starch in GS fruit was probably to reflection of the reduced sweetness that these fruits have in comparison to Ar fruits. Increased starch levels have also been associated with a sandier texture.

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The NSCs levels were found not to be significantly affected at PH-10, except for glucose, in GS, where it underwent an approximately 25% reduction, while the sucrose levels in Ar fruits remained unchanged. The latter information regarding Ar fruits, was in agreement with the reported literature (Campbell et al., 1987). Conversely, the sucrose levels, in GS fruits at the PH-10 stage, which could have influenced the sweetness of the fruit, showed a very similar tendency to the one observed by Pérez-Tello et al. (2001) in carambola fruits stored at 20 °C during 30 days. In addition, the fructose levels in Ar fruits, that generally are 1.5 times higher than those in GS fruits, a difference that determines to some extent the difference in sweetness between both varieties (Campbell et al., 1987; Campbell and Koch, 1989), were significantly higher in Ar fruit at IH-0. On the other hand, this difference in NSCs (i.e., higher in GS fruits), was lost, mainly in terms of glucose, in GS fruits at PH-10. This stark modification suggests the possible activation of an active isomerization process in GS fruits during storage, which should be experimentally confirmed. In contrast, the possibility that the observed changes in glucose and fructose levels in both varieties were due to an increment in sucrolytic activity by invertases can be discarded considering that the sucrose levels in these fruits were increased or remained constant.

This scenario was contrary to the one suggested by other authors (Pérez-Tello et al., 2001). However, the results obtained in this study agreed with a proposal suggesting that the rise in glucose and fructose during the storage of carambola fruits may be due to the differential use of these sugars for respiration. Anyway, an aspect that remains to be defined is the origin of the increased levels of NSCs, except starch, observed in fruits of both varieties at PH-10. It is tempting to speculate that these generated by a process of gluconeogenesis from free amino acids, or inclusive fatty acids. This proposal is somewhat supported by a proteomic analysis of the maturation process of apple, in which an accumulation of enzymes involved in the gluconeogenic process were found (e.g., malic enzyme dependent of NADP+ and triose phosphate isomerase; Shi et al., 2014), similar to what was previously reported in the maturation process of tomato (Goodenough et al., 1985).

Effect of the drying by lyophilization in the chemical composition of carambola fruit in two states of maturation

The increase in the levels of NSCs, except starch, could be simply due to a process of concentration caused by the loss of great part of the moisture of the fruit that, as reported in Table 1, was higher than 90%. However, it may not to be discarded that during lyophilization, the hydrolysis of starch was activated, as supported by the drastically decrease in carambola fruit starch levels already mentioned, from which the rise in soluble NSCs could have been generated. This is a question raised by the results of this study that will require a posterior analysis to be validated or rejected.

However, the changes caused by freeze-drying observed in this study are in accordance with the findings of previous reports in which the lyophilization process was found to cause numerous changes in the composition of plant tissues of diverse type. Included among these, are the observed modification of total phenolic compounds content and antioxidant activity the levels in tropical fruits, including carambola (Shofian et al., 2011). Likewise, other investigations studied the effect of this process on the integrity of plant tissues (Chang et al., 2006) and found evidence suggesting that the observed changes occurring as a result of lyophilization, could be due to a perturbation of the integrity of the fruit cells, leading to a de-compartmentalization of enzymes, substrates and activators that promoted the degradation of certain phenolic compounds. In addition, studies realized with onions at different stages of long term storage, including the lyophilization, found that the levels of flavonoids could raise, decrease or remain unaltered depending on the type of post-harvest process they were summited to and also on pre-storage manipulation they received (Amarowicz et al., 2009; Pérez-Gregorio et al., 2011a; Pérez-Gregorio et al., 2011b).

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Variables	Variety			
	[1]IH-0		[1]PH-10	
	GS	Ar	GS	Ar
Fresh weight (g)	91.93 ± 1.11a	75.01 ± 2.64b	46.18 ± 1.50b	58.25±1.75a
Radial longitude (cm)	5.51 ± 0.31a	5.15 ± 0.09a	3.67 ± 0.07b	5.44 ± 0.13a
Axis longitude (cm)	9.62 ± 0.47a	8.46 ± 0.13a	6.38 ± 0.12b	8.81 ± 0.16a
Humidity (%)	93.50 ± 0.46a	93.40 ± 0.18a	93.60±0.26a	89.63±0.35b

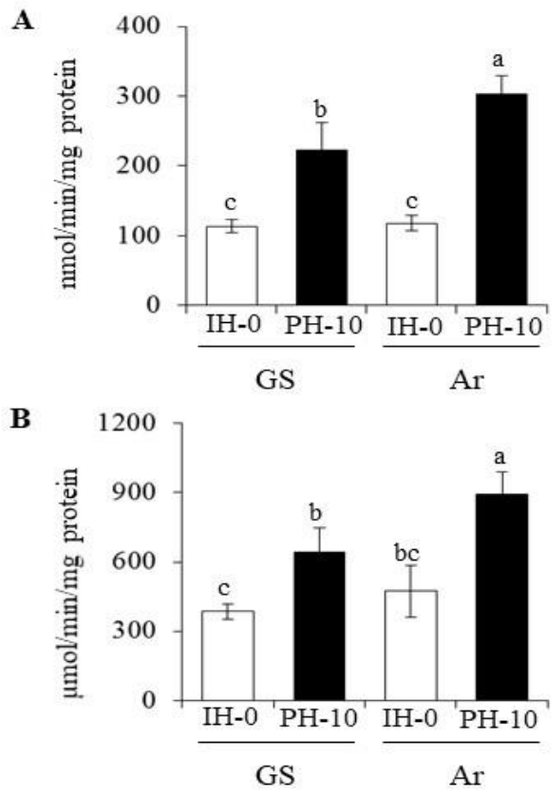
Table 1 Physical parameters (fresh weight, radial longitude and axis and percentage of humidity) in carambola (*Averrhoa carambola* L.) fresh fruits, varieties Golden Star (GS) and Arkin (Ar), immediately after harvest (cero days; IH-0) and after a 10-day post-harvest period (PH-10).[1] The data are means ± s.e. (n =30). Different letters within each column indicate statistically significant differences between the two varieties at $P < 0.05$ for the different measurement dates

Parameters	Variety			
	[1]GS		[1]Ar	
	IH-0	PH-10	IH-0	PH-10
pH	2.85 ± 0.62	2.32 ± 0.07	2.30 ± 0.03	1.91 ± 0.02
Acidity (%)	5.80 ± 0.95	4.53 ± 0.12	5.23 ± 0.11	6.46 ± 0.18
TSS (°Brix)	5.08 ± 0.77	4.29 ± 0.23	4.39 ± 0.10	5.64 ± 0.17
Protein (mg/ mL)	0.20 ± 0.01	0.22 ± 0.03	0.24 ± 0.02	0.19 ± 0.02
[2]Starch	4.76 ± 0.20	4.18 ± 0.10	3.32 ± 0.14	3.34 ± 0.1
Glucose (µmol/ g DW)	167.33 ± 6.08	175.80 ± 6.20	79.37 ± 2.99	133.90 ± 3.6
Fructose (µmol/ g DW)	87.85 ± 2.40	123.94 ± 8.90	101.73 ± 3.10	127.50 ± 3.2
Sucrose (µmol/ g DW)	15.81 ± 2.30	21.25 ± 1.10	15.12 ± 0.80	15.80 ± 0.8

Table 2 Chemical characterization in carambola (*Averrhoa carambola* L.) fruits, varieties Golden Star (GS) and Arkin (Ar), determined immediately after harvest (cero days; IH-0) and after a 10-day post-harvest period (PH-10). [1] The results are means ± s.e. (n = 30). [2] Starch levels are represented as µmol eq of glucose/g DW)

Parameters	Variety			
	[1]GS		[1]Ar	
	IH-0	PH-10	IH-0	PH-10
Protein (mg/ mL)	0.11 ± 0.01	0.06 ± 0.01	0.22 ± 0.01	0.12 ± 0.01
[2] Starch	0.35 ± 0.05	0.22 ± 0.02	1.00 ± 0.7	0.53 ± 0.04
Glucose (µmol g DW)	1160.70 ± 20.25	891.24 ± 27.02	924.22 ± 21.94	1120.29 ± 18.53
Fructose (µmol/ g DW)	740.96 ± 15.45	450.69 ± 38.50	672.98 ± 9.50	691.38 ± 7.91
Sucrose (µmol/ g DW)	97.36 ± 2.28	368.46 ± 30.58	73.11 ± 2.65	78.79 ± 2.42

Table 3 Chemical characterization lyophilized carambola (*Averrhoa carambola* L.) fruits, varieties Golden Star (GS) and Arkin (Ar), processed immediate after harvest (cero days; IH-0) or after a 10-days post-harvest period (PH-10). [1]Data are means ± s.e. (n = 30). [2] Reported in µmol equivalent of glucose/ g DW



Graphic 1 Levels of pectinolytic (A) and amilolytic (B) activity determined in carambola fruits, varieties Golden Star (GS) and Arkin (Ar), sampled at two states of maturity: IH-0, processed immediately after harvest at day 0 and representing fruits at physiological maturity, and PH-10, representing fruits harvested at physiological maturity and later stored at room temperature during 10 days. Each bar represents the means ± s.e. of measures performed using a total of 10 individual fruits. Different letters over the bars represent significantly different values ($P < 0.05$) determined by an ANOVA followed by a Tukey-Kramer test

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Conclusions

The following can be concluded based on the results obtained: i) the two more important commercial varieties of the carambola fruit, at least in the American continent, showed a contrasting behavior in terms of their physicochemical composition and in the way it was altered at two maturity stages. This finding reinforces reported data from previous studies in which a marked varietal effect is described regarding diverse aspects associated with post-harvest storage, such as browning by oxidation; ii) the divergent physicochemical characteristics of the varieties studied, that frequently differed from published data, as well as their contrasting post-harvest behavior, could have been affected by undetermined factors related to the cultivation site and/or to the agronomic practices employed for their production, and iii) the drying by lyophilization had a very marked effect on the protein and NSC contents of the carambola fruits. The effect was also dependent on the variety and on the maturity state of the tissue employed for the freeze-drying process. These aspects suggest that both short-term storage at room temperature, as well as drying by lyophilization, affect in a contrasting way, not only the physicochemical composition of the fruits but, very probably also, their biological activity. The latter aspect is important considering the wide potential for anthropogenic use offered by carambola fruits.

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Noninvasive thermographic evaluation of the thermal condition of piglets in the first month of life

Evaluación termográfica no invasiva de la condición térmica de lechones el primer mes de vida

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Abstract

In a commercial swine farm located in Zacoalco de Torres, Jalisco, we carried out an experiment to obtain information regarding environmental temperature effects on the performance of 13 litters of piglets during the first twenty eight days. It was used a Fluke® thermograph to obtain temperature images. Hypothesis was it is possible to establish the effect of heat sources trough thermographic images on the response of the litters. The aim was to evaluate the thermal condition of the piglets in the first twenty eight days of life and their performance until weaning. Piglets were F1 crosses (York x Landrace) x Pietrain. Treatments were minimum, maximum, average and central point temperatures. Variables registered were mortality, weaned piglets, weight of the weaned litters, piglets' individual weight and number of lactating days. Contribution is that temperatures in the first week have more influence on mortality, weaned piglets, weight of the weaned litters, piglets' individual weight and number of lactating days, than the ones in the first month.

Thermography, Piglets, Temperature

Resumen

La localidad del estudio fue una granja comercial situada en Zacoalco de Torres, Jalisco. El experimento se diseñó para obtener información sobre el efecto de la temperatura ambiental en el comportamiento de 13 camadas de lechones. Un termógrafo Fluke® obtuvo imágenes de las temperaturas. La hipótesis fue que es posible establecer el efecto del calor en los animales a través de imágenes termográficas. El objetivo fue evaluar la condición térmica de los lechones en los primeros 28 días y su desempeño hasta el destete. Los lechones fueron cruzas F1 de (York x Landrace) x Pietrain. Los tratamientos fueron temperaturas mínima, máxima, media y de punto central. Las variables fueron mortandad, lechones destetados, peso de lechones destetados, peso individual de lechones destetados y días de lactancia. La principal contribución es que las temperaturas los primeros siete días tuvieron mayor influencia en las variables estudiadas, que las temperaturas registradas durante el primer mes de vida.

Termografía, lechones, temperatura

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Introduction

Mexico pork meat production grew 2.2% in the last decade. In 2018, the production was 1.5 million tons, a 3.8% increase (SADER–SIAP, 2018). Pork meat consumption in 2017 was 2.11 million tons. Pork meat is an important source of protein for consumers. There are three categories of swine production systems in Jalisco: intensive, semi-intensive and extensive production. The three production types coexist in Jalisco, although the predominant one is the semi-intensive system. Largest farmers have a vertical integration as they practice artificial insemination, they have processing plants, places of slaughter and sales points. Genetic improvement done in the last two decades led to better productivity and higher feed to meat conversion. The intensive systems have the highest value added. High technology is used in every stage, as well as better racial genotypes, reproduction, nutrition and animal health (Montero-Lopez *et al.*, 2018). The use a non-invasive method to measure temperatures and its effect on the piglets' performance, may contribute to diminish inconvenient manipulation of the animals, and obtain valuable information.

Literature review

Neonatal mortality is frequent in piglets, as they do not have a hair cover, and their subcutaneal fat is scarce in the first days of life. Adequate management of piglets is crucial to reduce mortality in such a fragile stage. In the intrauterine life the piglet have a high and constant temperature. Outside, they do not find these conditions and loses heat due to its thermoregulation inability, hair shortage, and scarce subcutaneal tissue to diminish the heat flux from the blood vessels (Berthon *et al.*, 1994). When the piglets are born, have a narrow thermo neutrality (32ºC-35ºC) (Nielsen, 1997). If the environment is below the rank, they will use additional energy for heating, its growth will cease and they will consume their energy reserves rapidly, risking their lives. Since an evolutive point of view, 20% mortality is acceptable, but modern farms can reduce mortality to 5% (Arey, 1995). In the first hours after birth, piglets require an ideal microclimate to reach the mammaries. It is required to provide additional heat during the first five days, so they can have the necessary temperature (Curtis, 1974).

Hypothermia is the major cause of neonatal mortality in piglets (Tuchschereret *et al*., 2000; Edwards, 2002). Neonatal pigs with hypothermia are likely to die due to hunger, crushing or diseases (Pedersen *et al*., 2011). Rectal thermometry is a common assessment of the thermal condition of pigs, but this method could influence the pig performance, and its thermoregulation may be affected. (Kammersgaard *et al*., 2011). A non-invasive accurate method to evaluate the pigs' thermal condition, without individual handling of animals, is of great potential in research and in farm conditions. The base of infrared thermography is the infrared radiation measurement emitted from an object surface. Infrared rays are electromagnetic emissions between the visible radiation and the radio waves, so they are invisible to us, but perceptible in the form of heat through nerve receptors in the skin of the animal (Kammersgaard, *et al.*, 2013).

Thermography measures the corporal surface temperature, which exchanges heat in the surrounding area.

Problem approach

Piglets at birth have a very narrow thermal neutrality, with low critical temperature around 32^{O}-35^{O}C. If they use additional energy to keep warm, it could risk their lives. At birth, it is important to provide the piglets an adequate microclimate so they can reach the mammal glands without cold.

Justification

Survival until weaning is highly related with hypothermia between one or two hours after birth. Inside the uterus there are stable temperatures between 39-40^{O}C, but at birth piglets are in a colder environment. This transition leads to a corporal temperature drop in two to four ^{O}C. Piglets that cannot quickly overcome hypothermia, die because of it and of hunger. Studies on thermic biology allow for progress in the knowledge in the way temperature affects lactating piglets.

Hypothesis

Image thermographic analysis will make it possible to establish the heat sources effect in the response of the piglets the first seven days of life.

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General objective

Evaluate the thermic condition of piglets the first 30 days of life, using thermography.

Particular objectives

Analyze the thermographic images of the litters in the maternity area. Measuring the regulatory capacity of the litter. Relate it with the physiological response of the litter.

Materials and methods

Location

The study was carried out in December 2017 to January 2018, in a commercial farm in the Zacoalco de Torres County, state of Jalisco. The farm is a semi-technified one with 230 bellies.

Animals

It was considered 13 litters of (York x Landrace) x Pietrain crosses.

Treatments

Four types of temperatures around the litters during the first 28 days of life: maximum, minimum, average and midpoint.

Studied variables

Four variables were measured in the 13 litters: piglet mortality, number of weaned piglets, litter weight at weaning, and lactating days.

Experimental methodology

A Fluke® thermograph provided thermographic images of the environment around the piglets, from birth to the twenty-eighth day. Images were recorded in three daily periods. The Fluke® software was used for image analysis. Maximum, minimum, mean and mid-point temperatures were obtained from the upper section of the animals.

Experimental design

Randomized complete blocks. Main source of variation was the litters' temperatures. The means comparison method was Tukey procedure (Steel and Torrie, 1960).

Results

Temperatures

Table 1 shows result of statistical analysis of four different temperatures around or above the piglets. All four temperature measurements resulted significant ($p \leq 0.05$). Graph 1 contains four images related to the daily temperatures recorded in the 13 litters during the first 28 days. It can be noticed in the first week all temperatures are pretty close, but from the second week onwards, they split in a very strong way. Graph 2, also with four images, illustrates changes in temperatures throughout the first four weeks and its influence on the productive variables of 13 litters. Table 2 contains results of multiple regression analysis. It can be observed all determination coefficients are low; only piglet mortality and lactating days had $R^2 > 0.20$. This table also contains the summary of the regression equations calculated for the temperatures and the observations made on the litters.

Figure 1 Piglets' thermographic image at 7 days

Source of varia-tion	Ms Minimum tempera-ture	Ms mean tempera-ture
Litters	1384.35*	5003.47*
Days	116.06*	598.69*
Vc%	24.13	20.68
Source of varia-tion	**Ms Maximum tempera-ture**	**Ms Midpoint tempera-ture**
Litters	4585.60*	1642.47*
Days	465.52*	7.43*
Vc%	19.31	15.6

*Significant $p \leq 0.05$

Table 1 Environmental temperatures mean squares in litters during the first 28 days

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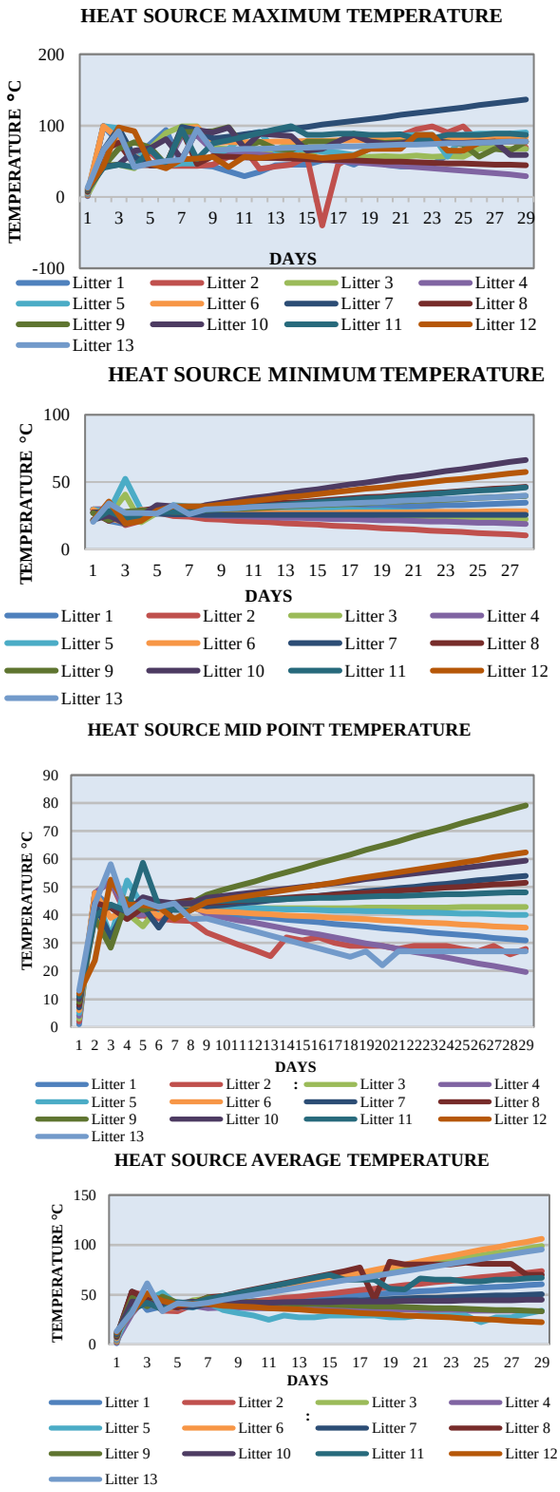
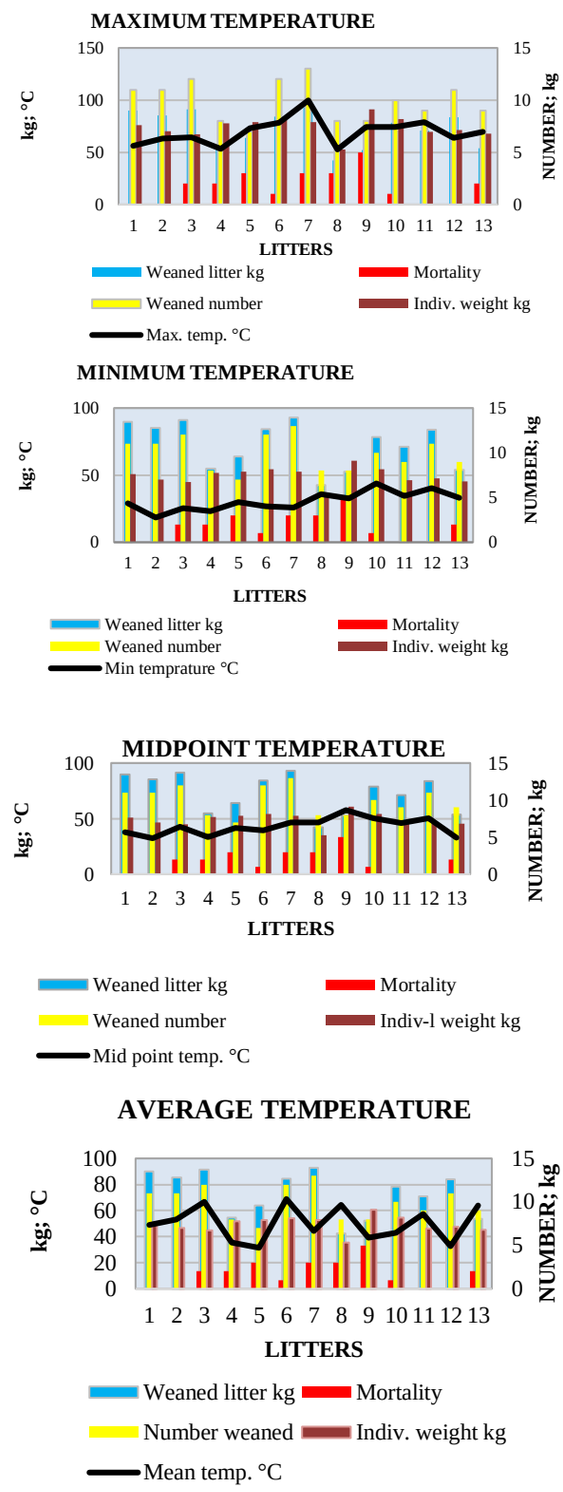


Graphic 1 Four images containing daily different measurements of temperatures recorded in 13 litters in the first 28 days



Graphic 2 Four images containing the influence of four different temperatures on 13 litters' production traits

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MULTIPLE REGRESSION EQUATIONS $\hat{y} = a+b1(x)+b2(x)+b3(x)+b4(x)$
Mortality number: $R^2 = 0.2332$
$\hat{y} = 0.7980-0.0349-0.0349-0.0615+0.0513$
Weaned litter weight: $R^2 = 0.0672$
$\hat{y} = 55.5328-0.0673+0.0101+0.0648-0.0460$
Weaned piglets' number: $R^2 = 0.0167$
$\hat{y} = 10.8418-0.0083+0.0270-0.0369+0.0002$
Lactating days: $R^2 = 0.2193$
$\hat{y} = 34.1419-0.0486+0.0101+0.0648-0.0460$
Individual weight of weaned piglets: $R^2 = 0.0167$
$\hat{y} = 2.3572+0.0100+0.0478-0.0162+0.0533$

Table 2 Multiple regression equations between four different temperatures and five traits

Performance of the litters from birth to weaning

In appendix table 4, there are five variables measured in 13 litters, and the ambient temperatures in which they lived 28 days after birth. Litters performance from birth to weaning are in table 3. Determination coefficients values (table 2) correspond to piglet mortality and lactating days and its relation with temperatures (0.233, and 0.219 respectively). In a previous report (Sánchez-Chipres *et al*., 2019) it was found a definite relation between minimum, maximum and midpoint temperatures during the first seven days after birth and data about piglet mortality, number of weaned piglets, weight of weaned litters, individual weight of weaned piglets and lactating days. Average temperature was the less relevant one in such report. The low R^2 for all traits obtained in the current work, may be due to temperatures separation as the piglets grew (Graphic 1). Besides, the average of the four temperatures showed a wide range of variation in the 28 period (Graphic 2).

LIT	IWW	LAD	LWW	MOR
1	7.9	30	93	3
12	6.7	36	91	2
11	7.6	30	90	0
2	7.0	30	85	0
13	8.1	30	84	1
9	7.2	31	84	0
5	8.2	29	79	1
3	7.0	30	71	0
10	7.9	31	64	3
8	7.8	34	55	2
6	6.8	30	54	2
7	9.1	34	52	5
4	5.3	34	42	3
Average	7.4	31.5	72.6	1.7
Criteria	$\geq$7.9	$\geq$30	$\geq$85	$\leq$1

Table 3 Productive traits of 13 litters

LIT=Litter; IWW=Individual weight at weaning; LAD=Lactating days; LWW=Litter weight at weaning; MOR=Mortality.

Better litters in the first month

Table 3 shows a comparison between litters in the first 28 days after birth. In an empirical manner, criteria used was the following: litter weaned weight $\geq$ 85 kg; mortality $\leq$ 1; weaned piglets $\geq$ 11; individual weight at weaning $\geq$ 7.9 kg; lactating days $\leq$ 30 days. With these criteria, the best litters were 1, 11, 2, and 13, as they had at least four out of the five mentioned traits.

Discussion

Temperature values recorded in this work with some influence on the piglets' performance were the average minimum temperature (26^{O}C) and the average mid-point temperature (41.4^{O}C). The piglets' thermal sensation depends on the temperature, humidity and air flux. There is a relation between these factors and the animal size, animal number, the isolation facilities, and the type of material in the bed (Mc Ginnis *et al*., 1981).

Reaching an ideal and constant temperature for the pigs in the facilities can be difficult, that is why it was defined a thermal neutrality zone, which is the adequate environmental temperature for keeping the normal body temperature in the pig (Herpin *et al*., 2002). The thermal neutrality zone is the basal metabolic rate, which is the minimum energy necessary to keep the animal alive, this way the animal maintain a constant heat loss.

There are two values in the neutral thermic tables, for piglets in the first week of life; the lowest value corresponds to the low critical temperature (30^{O}C), the minimum temperature that allows the greatest growth. Below it, the animal uses energy in fighting the cold. The upper value corresponds to the higher critical temperature (35^{O}C), this temperature allows the greatest growth, and above it, the animal decreases the feed intake (Lay *et al.*, 2002). As the animal growths, the thermoneutral zone becomes wider, and the animal is capable of withstanding more extreme temperatures.

The values found in this work allow us to recognize that litters were under temperatures below the low critical temperature. It is clear there is a positive relation between environmental temperature and heat production with the calostrum intake.

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This demonstrates that the piglets that cannot intake calostrum are unable to achieve thermo stability, which can affect their growth rate, reduce the vigor to compete for a tit and even to promote greater hair growth as an isolation to keep the adequate corporal temperature (Le Dividich y Noblet, 1981; Mota-Rojas *et al*., 2011). When reviewing the productive performance of the litters, minimum and maximum temperatures were highly related with the weight litter weaned weight, individual weight at weaning, and number of weaned piglets.

The use of thermography to determine heat source temperatures, contributes to minimizing the handling of the piglet, allowing the animal to devote its energy to finding its optimum temperature and its food; this makes possible a higher growth rate in grams day^{-1}. The non-invasive technique described, provides knowledge of the thermal condition for sows, and allows knowing if an excessive temperature will decrease milk production. The knowledge on the environmental condition of the females, will allow us to improve the efficiency of the biological behavior of sows and piglets.

Conclusion

The thermography used allowed us to verify the importance of the piglet microclimate. This methodology made possible to obtain useful information about temperatures in the maternity area. The heat source can provide a wide range of temperature areas in the beds. Some piglets had temperatures below the adequate range, which could influence their performance at weaning.

Appendix

LITTER NUMBER	MORT	WEAN WEIGHT kg	WEAN NUM	IND WEI kg	LAC DAYS	MEAN TEMP °C	MIN TEMP °C	MIDP TEMP °C	MAX TEMP °C
1	3	93	13	7.89	30	48.81	29.14	37.67	56.01
2	0	85.2	11	7.02	30	53.40	18.44	32.38	63.31
3	0	71	9	6.95	30	66.60	25.48	42.49	64.29
4	3	42.4	8	5.3	34	35.66	23.04	33.57	53.31
5	1	78.6	10	8.16	29	31.48	29.83	41.91	72.85
6	2	53.8	9	6.78	30	68.90	26.93	39.51	78.62
7	5	52.4	8	9.11	34	44.48	25.65	46.59	99.77
8	2	54.8	8	7.75	34	64.42	35.66	46.70	52.71
9	0	83.8	11	7.15	31	39.32	32.48	57.67	74.20
10	3	63.8	7	7.91	31	42.77	43.84	50.45	74.01
11	0	89.8	11	7.6	30	57.29	34.63	45.99	79.03
12	2	91.2	12	6.72	36	32.76	40.34	50.35	64.03
13	1	84.2	12	8.13	30	63.95	32.99	33.00	69.74
Average	1.69	72.6	9.9	7.42	1.5	49.99	30.65	42.94	69.38

Table 5 Traits of 13 litters recorded from birth to weaning and temperatures during the first 28 days

MORT = mortality; WEAN WEIGHT = weight litter weaned; WEAN NUM= number of weaned piglets; IND WEI = individual weight at weaning; LAC = lactating days; MEAN TEMP = mean temperature; MIN TEMP = minimum temperature; MIDP TEMP = mid-point temperature; MAX TEMP = maximum temperature

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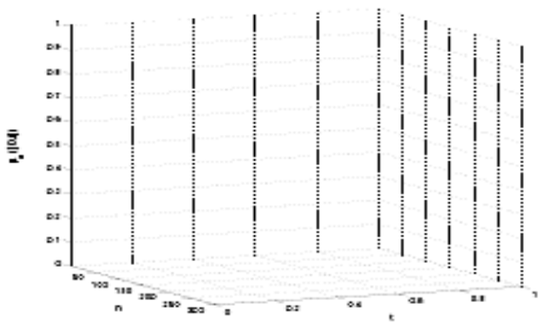

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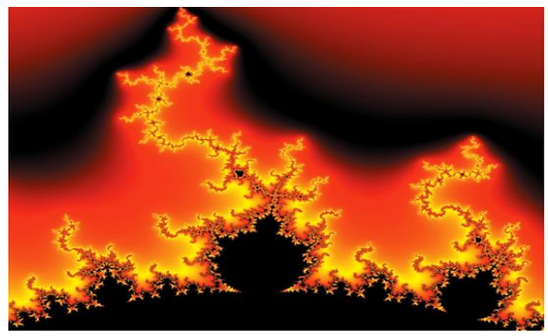

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