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Tratamientos de almacenamiento y vida en florero en tres variedades de *Heliconia*

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TRATAMIENTOS DE ALMACENAMIENTO Y VIDA EN FLORERO EN TRES VARIEDADES DE *Heliconia*

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RESUMEN

El objetivo de este estudio fue evaluar el efecto de dos tratamientos postcosecha con dos niveles: almacenamiento en frío, a 12 y 16 ºC, y recubrimiento con quitosano al 1.0 y 1.5 % sin almacenamiento en tres variedades de *Heliconia* (*H. psittacorum* L. f. cv. Trópica, *H. bihai* (L.) L. cv. Halloween y Lobster Claw). La hipótesis fue que el quitosano mantiene la calidad en los tallos florales, cuando se almacenan a temperatura ambiente, por la promoción de mecanismos de protección y sus propiedades antimicrobianas, igualando o superando la vida en florero de los tratamientos con frío. El diseño en medidas repetidas desbalanceado se utilizó para investigar la interacción tratamiento x tiempo en las variables no destructivas (vida en florero, pérdida de peso, luminosidad, cromaticidad y matiz) y el diseño completamente al azar para las variables destructivas (antocianinas, flavonoides totales y azúcares totales) y se aplicaron pruebas de comparaciones de medias. Las evaluaciones se hicieron en los días 1, 5, 10 y final de la vida en florero. Cada tratamiento estuvo formado por seis repeticiones y un tallo floral como unidad experimental. El quitosano mostró efecto positivo en la vida en florero en todos los cultivares. *H. psittacorum* tuvo la vida mayor en florero (23.1 y 21 d con 1.0 y 1.5 % de quitosano, en comparación con 12.1 d del control), le siguió *H. bihai* cv. Halloween (19.8, 16.6 y 9.6 d) y *H. bihai* cv. Lobster Claw (16.8 y 12.1 d); en esta variedad, el almacenamiento a 16 ºC tuvo vida en florero similar al tratamiento con quitosano al 1.0 % (15.3 d). La cubierta con quitosano extendió la vida en florero en las tres variedades; pero, dependió de la variedad.

Palabras clave: calidad, longevidad, ornamentales tropicales, quitosano

STORAGE TREATMENTS AND VASE LIFE OF THREE CULTIVARS OF *Heliconia*

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ABSTRACT

The objective of this study was to evaluate the effect of two postharvest treatments with two levels: cold storage at 12 and 16 ºC and chitosan coating at 1.0 % and 1.5 % without storage in three varieties of Heliconia (*H. psittacorum* L. f. cv. Trópica, *H. bihai* (L.) L. cv. Halloween y *H. bihai* (L.) L. cv. Lobster Claw). The hypothesis was that chitosan preserve the quality of flower stems stored at room temperature compared with cold treatments. An unbalanced repeated measures design was used to investigate the treatment x time interaction in non-destructive variables (vase life, weight loss, luminosity, chromaticity and hue). A completely random design was used for the destructive variables (anthocyanins, total flavonoids, and total sugars) and a Tukey tests. Evaluations were made on days 1, 5, 10 and end of vase life. Each treatment consisted of six replicates and one floral stem as an experimental unit. Chitosan showed a positive effect on vase life in all cultivars. *H. psittacorum* had the longest vase life (23.1 and 21 d with 1.0 and 1.5% chitosan, compared to 12.1 d of control), followed by *H. bihai* cv. Halloween (19.8, 16.6 and 9.6 d) and H. bihai cv. Lobster Claw (16.8 and 12.1 d); in this variety, the stored at 16 ºC had a vase life similar to chitosan at 1.0% (15.3 d). The chitosan coating extended the life in vase in the three varieties; but, depended on the variety.

Key words: quality, longevity, tropical ornamental, chitosan coating

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CONTENIDO

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INTRODUCCIÓN GENERAL

La floricultura es una actividad económica con potencial de crecimiento. En 2013 las exportaciones globales de flores de corte, follaje, plantas en maceta y bulbos representaron 20.6 billones de dólares, y representó un crecimiento del 142 % en un período de 12 años (Rijswick, 2015). De estos productos, las flores de corte son el grupo de mayor demanda para su comercialización, su expansión depende de la producción y de los avances en la logística y manejo, implementados para su transporte a lugares estratégicos para comercializarse. El principal exportador de flores de corte a nivel global es EE.UU., con 80 % del total, le sigue el Reino Unido, con ganancias de 37 mdd (Madrid y Lovell, 2007).

Sin embargo, para promover el crecimiento del sector es necesario introducir productos novedosos que presenten características distintas a las flores de corte tradicionales. Al respecto, las ornamentales tropicales son una alternativa potencial para enriquecer el sector. Entre ellas están las heliconias, junto con las alpinias, antorchas y anturios por su belleza, formas diversas y coloridas, durabilidad, disponibilidad estacional y vida extensa en florero (Criley y Paull, 1993a; Jerez-Mompié, 2007).

El género *Heliconia* está conformado por alrededor de 250 especies y México alberga 16 de ellas, todas con potencial ornamental (Kress et al., 1999). Además, los requerimientos climáticos para el establecimiento a gran escala de estas especies se encuentran en México. Las heliconias requieren alrededor de 32 ºC / 20 ºC (día-noche), irradiancias moderadas (entre 250 y 710 µmol m^{-2} s^{-1}) (Catley y

Brooking, 1996). El mercado para las heliconias en Hawaii en la década de 1970 registró ganancias superiores a los 11.5 mdd (Akamine, 1976).

Sin embargo, en el nivel global no existen métodos estandarizados que promuevan la conservación de la calidad postcosecha en recorridos por largas distancias o períodos largos de tiempo. Esto representa una limitante para la promoción a gran escala de su comercialización, pues implica pérdidas postcosecha significativas. Las pérdidas postcosecha en el sector hortícola se han estimado que pueden ser entre 20 y 40 % del total de la producción (Kasso y Bekele, 2016). Ellas se deben principalmente al manejo inapropiado de los productos, el desarrollo de microorganismos, senescencia acelerada que resultan en un decremento de la calidad y vida en anaquel (Olayemi et al., 2010).

El almacenamiento con temperaturas bajas es una técnica que se utiliza comúnmente para mantener o extender la vida en florero de flores de corte. Las temperaturas frías disminuyen los procesos metabólicos relacionados con la senescencia en algunas especies (Matsumoto y Ikoma, 2016) y aumentan la vida en florero; pero, en las especies tropicales pueden ocasionar lesiones por frío debido a su alta sensibilidad (Costa et al., 2011b, 2011a).

Existen recomendaciones diversas para el almacenamiento refrigerado de heliconias. Broschat y Donselman (1988) y Criley y Paull, (1993) recomiendan temperaturas de almacenamiento superiores a 13 ºC, Jaroenkit y Paull (2003) sugieren que el almacenamiento a 10 ºC es suficiente para mantener la calidad y vida en florero adecuadas. Costa et al. (2011a) sugieren que *H. bihai* cv. Lobster Claw se almacene a 19 ºC y se evite el almacenarla a menos de 12 ºC, pues se promueven lesiones por frío. Para los cultivares “Golden Torch”, “Andromeda” y “St.

Vincent Red" se ha demostrado que el almacenamiento a 23 ºC permite alcanzar vida en florero de 14 a 15 d, el almacenamiento de *H. psittacorum* cv. Trópica a 15 ºC por 10 días permitió una vida en florero de 6 días (Bañuelos-Hernández et al., 2016) y *H. marginata* almacenamiento a 25 ºC mostró una vida en florero de 8 días.

Entre las prácticas diversas para extender la vida en florero y mantener la calidad de las inflorescencias se señala el uso de soluciones pulso, conservantes y nutritivas, y aplicación de ceras para evitar la desecación (de La Riva Morales, 2011; Jaroenkit y Paull, 2003b). Sin embargo, en estas especies se ha comprobado que las soluciones conservantes no mejoran ni extienden la vida en florero (Broschat y Donselman, 1983). Las soluciones antitranspirantes pueden aumentar la vida postcosecha (Broschat y Donselman, 1987). La respuesta ineficiente de las soluciones preservativas está asociada al desarrollo vascular escaso de la base del tallo floral, que ocasiona absorción deficiente de las soluciones (Criley and Paull, 1993a).

Al respecto, numerosas investigaciones se han enfocado en buscar alternativas para mejorar la calidad postcosecha de los productos hortícolas. Una línea de investigación se ha dirigido al uso de biopelículas. Entre ellas, las preparadas con quitosano han mostrado impacto significativo en la manutención y extensión de la vida de anaquel en productos hortícolas. El quitosano es un polisacárido orgánico, biodegradable e inocuo, obtenido principalmente del exoesqueleto de los crustáceos. Sus propiedades se basan en su naturaleza policatiónica, la cual permite su acción floculante, humectante y quelante (Bautista-Baños et al., 2005) y el impacto de su respuesta depende de la concentración que se utilice, su peso molecular y pH de la solución (Fei Liu et al., 2001a; Liu et al., 2006).

El tratamiento con quitosano de frutos de kiwi, manzanas, peras, fresas, grosellas y litchi mostraron inhibición de *Botrytis cinerea* inoculado (Du et al., 1997; El Ghaouth et al., 1991a; Li y Yu, 2001; Zhang y Quantick, 1997). Esta propiedad antifúngica parece que es producto de la inducción de la quitinasa por los tejidos vegetales; esta enzima cataliza la degradación de las paredes celulares fúngicas (Hirano y Nagao, 1989; Lin y Zhao, 2007). Además, la efectividad del quitosano se ha demostrado en el retraso de procesos asociados a la maduración y la disminución de tasas respiratorias en frutas y vegetales (Vargas et al., 2006), reducción en la pérdida de biomasa y coloración en calabaza, pimiento y tomates (El Ghaouth et al., 1991b). La conservación de la coloración está asociada a la capacidad de inhibición de la actividad peroxidasa, lo cual promueve el oscurecimiento de los frutos (Zhang y Quantick, 1997). Chien et al. (2007b) reportaron la efectividad del recubrimiento de quitosano aplicado a mango sobre la preservación de la calidad y la extensión de la vida de anaquel del fruto.

En flores de corte los reportes sobre la efectividad del quitosano son escasos. La literatura disponible muestra que sus efectos podrían ser positivos. Es el caso de quitooligosacarido, que es un derivado del quitosano, aplicado a rosas que incrementó 54 % la vida en florero, (18.1 días) en comparación con el control (11.7 días) (Jing y Li, 2015).

De acuerdo con la literatura, el intervalo de temperatura de almacenamiento para las especies y variedades de heliconias es amplio; por lo tanto, cada especie y variedad con potencial económico debe estudiarse para identificar los intervalos de temperatura adecuados para su almacenamiento. Además, los resultados con quitosano son promisorios para mantener la calidad y extender la vida de anaquel

en productos hortícolas; pero, es necesario investigar su aplicación a los ornamentales tropicales para encontrar alternativas que promuevan la ampliación de la vida en florero de estas especies.

Por lo tanto, los objetivos de esta investigación fueron:

- Identificar temperaturas de almacenamiento adecuadas para los tallos florales de *H. psittacorum* L. f. cv. Trópica, *H. bihai* (L.) L. cv. Halloween y *H. bihai* (L.) L. cv. Halloween.
- Determinar el efecto del recubrimiento de quitosano en los tallos florales de *H. psittacorum* L. f. cv. Trópica, *H. bihai* (L.) L. cv. Halloween y *H. bihai* (L.) L. cv. Halloween sobre la vida en florero.
- Comparar ambas prácticas postcosecha para determinar el o los tratamientos que presenten ventajas mayores para la preservación de las variables fisiológicas evaluadas.

Las hipótesis planteadas fueron:

- El quitosano conserva las características de calidad en los tallos florales, aunque se almacenen a temperatura ambiente, debido a sus propiedades antimicrobianas y la promoción de mecanismos de protección de los tejidos, por lo que puede igualar o superar la vida en florero de los tratamientos a bajas temperaturas.
- Cada variedad presenta una temperatura óptima de almacenamiento.

1.1 LITERATURA CITADA

Akamine, E.K., 1976. Postharvest handling of tropical ornamental crops. HortScience 11, 125.

Bañuelos-Hernández, K.P., García-Nava, J.R., Leyva-Ovalle, O.R., Peña-Valdivia, C.B., Ybarra-Moncada, M.C., 2016. Flowering stem storage of *Heliconia psittacorum* L. f. cv. Trópica. Postharvest Biol. Technol. 112, 159–169. doi:10.1016/j.postharvbio.2015.10.006

Bautista-Baños, S., Hernández-Lauzardo, A.N., del Valle, M.V., Bosquez-Molina, E., Sánchez-Domínguez, D., 2005. Quitosano: una alternativa natural para reducir microorganismos postcosecha y mantener la vida de anaquel de productos hortofrutícolas. Rev. Iberoam. Tecnol. Postcosecha 7, 1–6.

Broschat, T., Donselman, H., 1983. Production and postharvest culture of *Heliconia psittacorum* flowers in Souht Florida. Proc Fla State Hort Soc 96, 272–273.

Broschat, T.K., Donselman, H., 1987. Tropical cut flower research at the University of Floridas`s Ft. Lauderdale Research and EducationCenter. Bul Heliconia Soc Intern 2, 5–6.

Broschat, T.K. (University of F., Donselman, H., 1988. Production and postharvest culture of red ginger in south Florida. Proc. Annu. Meet. Fla. State Hortic. Soc. USA 101, 326–327.

Catley, J.L., Brooking, I.R., 1996. Temperature and light influence growth and flower production in *Heliconia Golden* Torch'. HortScience 31, 213–217.

Chien, P., Sheu, F., Yang, F.-H., 2007. Effects of edible chitosan coating on quality and shelf life of sliced mango fruit. J. Food Eng. 78, 225–229. doi:10.1016/j.jfoodeng.2005.09.022

Costa, A.S., Nogueira, L.C., dos Santos, V.F., Finger, F.L., Camara, T.R., Loges, V., Willadino, L., 2011a. Characterization of symptoms of senescence and chilling injury on inflorescences of *Heliconia bihai* (L.) cv. Lobster Claw and cv. Halloween. Postharvest Biol. Technol. 59, 103–109. doi:10.1016/j.postharvbio.2010.08.015

Costa, A.S., Nogueira, L.C., Santos, V.F. dos, Camara, T.R., Loges, V., Willadino, L., 2011b. Storage of cut *Heliconia bihai* (L.) cv. Lobster Claw flowers at low temperatures. Rev. Bras. Eng. Agríc. E Ambient. 15, 966–972. doi:10.1590/S1415-43662011000900013

Costa, A.S., Nogueira, L.C., Santos, V.F. dos, Camara, T.R., Loges, V., Willadino, L., 2011c. Storage of cut *Heliconia bihai* (L.) cv. Lobster Claw flowers at low temperatures. Rev. Bras. Eng. Agríc. E Ambient. 15, 966–972.

Criley, R.A., Paull, R.E., 1993a. Review: Postharvest handling of bold tropical cut flowers- *Anthurium*, *Alpinia purpurata*, *Heliconia*, and *Strelitzia*. Acta Hortic., New Floricultural Crops 337, 201–212.

Criley, R.A., Paull, R.E., 1993b. Review: Postharvest handling of bold tropical cut flowers- *Anthurium, Alpinia purpurata, Heliconia*, and *Strelitzia*. Acta Hortic. 337, 201–212.

de La Riva Morales, F., 2011. Poscosecha de flores de corte y medio ambiente. Idesia Arica 29, 125–130.

Du, J., Gemma, H., Iwahori, S., 1997. Effects of chitosan coating on the storage of peach, japanese pear, and kiwifruit. J. Jpn. Soc. Hortic. Sci. 66, 15–22. doi:10.2503/jjshs.66.15

El Ghaouth, A., Arul, J., Ponnampalam, R., Boulet, M., 1991a. Chitosan coating effect on storability and quality of fresh strawberries. J. Food Sci. 56, 1618–1620. doi:10.1111/j.1365-2621.1991.tb08655.x

El Ghaouth, A., Arul, J., Ponnampalam, R., Boulet, M., 1991b. Use of chitosan coating to reduce water loss and maintain quality of cucumber and bell pepper fruits. J. Food Process. Preserv. 15, 359–368. doi:10.1111/j.1745-4549.1991.tb00178.x

Fei Liu, X., Lin Guan, Y., Zhi Yang, D., Li, Z., De Yao, K., 2001. Antibacterial action of chitosan and carboxymethylated chitosan. J. Appl. Polym. Sci. 79, 1324–1335. doi:10.1002/1097-4628(20010214)79:7<1324::AID-APP210>3.0.CO;2-L

Hirano, S., Nagao, N., 1989. Effects of chitosan, pectic acid, lysozyme, and chitinase on the growth of several phytopathogens. Agric. Biol. Chem. 53, 3065–3066. doi:10.1080/00021369.1989.10869777

Jaroenkit, T., Paull, R.E., 2003a. Postharvest handling of heliconia, red ginger, and bird-of-paradise. HortTechnology 13, 259–266.

Jaroenkit, T., Paull, R.E., 2003b. Postharvest Handling of heliconia, red ginger, and bird-of-paradise. HortTechnology 13, 259–266.

Jerez-Mompié, E., 2007. El cultivo de las heliconias. Cultiv. Trop. 28, 29–35.

Kasso, M., Bekele, A., 2016. Post-harvest loss and quality deterioration of horticultural crops in Dire Dawa Region, Ethiopia. J. Saudi Soc. Agric. Sci. doi:10.1016/j.jssas.2016.01.005

Kress, W.J., Betancur, J., Echeverry, B., 1999. Heliconias: llamaradas de la selva colombiana. Cristina Uribe Ediciones.

Li, H., Yu, T., 2001. Effect of chitosan on incidence of brown rot, quality and physiological attributes of postharvest peach fruit. J. Sci. Food Agric. 81, 269–274. doi:10.1002/1097-0010(20010115)81:2<269::AID-JSFA806>3.0.CO;2-F

Lin, D., Zhao, Y., 2007. Innovations in the development and application of edible coatings for fresh and minimally processed fruits and vegetables. Compr. Rev. Food Sci. Food Saf. 6, 60–75. doi:10.1111/j.1541-4337.2007.00018.x

Liu, N., Chen, X.-G., Park, H.-J., Liu, C.-G., Liu, C.-S., Meng, X.-H., Yu, L.-J., 2006. Effect of MW and concentration of chitosan on antibacterial activity of *Escherichia coli.* Carbohydr. Polym. 64, 60–65. doi:10.1016/j.carbpol.2005.10.028

Madrid, G., Lovell, T., 2007. Working with flowers in Colombia: The “lucky chance”? Womens Stud. Int. Forum 30, 217–227. doi:10.1016/j.wsif.2007.03.003

Matsumoto, H., Ikoma, Y., 2016. Effect of postharvest temperature on the muscat flavor and aroma volatile content in the berries of “Shine Muscat” (*Vitis labruscana* Baily×V. vinifera L.). Postharvest Biol. Technol. 112, 256–265. doi:10.1016/j.postharvbio.2015.09.004

Olayemi, F.F., Adegbola, J.A., Bamishaiye, E.I., Daura, A.M., 2010. Assessment of post-harvest challenges of small scale farm holders of tomatoes, bell and hot

pepper in some local government areas of Kano State, Nigeria. Bayero J. Pure Appl. Sci. 3, 39–42. doi:10.4314/bajopas.v3i2.63217

Rijswick Cindy, 2015. World Floriculture Map 2015 (No. 475), Food & Agribusiness Research and Advisory. Rabobank Industry.

Vargas, M., Albors, A., Chiralt, A., González-Martínez, C., 2006. Quality of cold-stored strawberries as affected by chitosan–oleic acid edible coatings. Postharvest Biol. Technol. 41, 164–171. doi:10.1016/j.postharvbio.2006.03.016

Zhang, D., Quantick, P.C., 1997. Effects of chitosan coating on enzymatic browning and decay during postharvest storage of litchi (*Litchi chinensis* Sonn.) fruit. Postharvest Biol. Technol. 12, 195–202.

2. CHITOSAN COATING EFFECT ON VASE LIFE OF FLOWERING STEMS OF *Heliconia bihai* (L.) L. cv. Halloween

CHITOSAN COATING EFFECT ON VASE LIFE OF FLOWERING STEMS OF *Heliconia bihai* (L.) L. cv. Halloween

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Keywords: biopolymer, longevity, quality, tropical ornamental, vase life

2.1 Abstract

Tropical ornamentals such as heliconias play a key role in the growth of the floriculture market due to their bright coloured and attractively shaped inflorescences. *Heliconia bihai* is one of the most popular of the Heliconias in the global market. New postharvest handling techniques to increase distribution of the cut flowers without compromising flower stem quality as measured by vase life are needed. The effects of chitosan coating on vase life of flower stems of *Heliconia*

bihai (L.) L. Halloween stored under laboratory conditions has been studied. Flowering stem fresh weight loss, bract colour (L*, C* and hº), concentration of anthocyanin, total flavonoids and total sugars, percentage of absolute integrity of cell membranes (PAI) of stem, peduncle and bracts, and duration of vase life were evaluated on day 1, 5, 10 and the final day of vase life. Concentrations of 1.0 and 1.5 % of chitosan, extended the vase life by 10.3 and 7 d more than the control, respectively. An internal cell damage gradient from the base to the apex based on ion release was observed and the visual quality of inflorescences decreased over time in all treatments, but at different rates.

2.2 Introduction

Heliconias flowers are one of the ornamental tropical flowers with great economic potential due to the variety in shapes and colours, beauty and strength, *Heliconia bihai* is one of the most commonly traded species worldwide (Jerez, 2007). Several studies have focused on investigating postharvest practices that improve the vase life of this species; Broschat and Donselman (1983) showed that preservative solutions such as 8-hydroxyquinoline sulphate, sucrose, citrate, silver thiosulfate, dithiothreitol, and benzyladenine in *Heliconia* did not improve the vase life. These authors also suggested that antitranspirants and waxes are a limited alternative due to a failure to completely coat the bract surface. In contrast, Ka-ipo et al. (1989) found that the dipping of *H. psittacorum* cv. Parakeet in Wilt – Pruf® or wax showed a 36 % increase of vase life. Leyva et al. (2011) found that a hydrophilic polymer in combination with the addition of preservative solutions (citric acid, sucrose and

Crystal clear®) was effective in the storage of *H. psittacorum x H. spathocircinata* Aristeguieta var. Golden Torch Adrián and *Rosa* spp. However the use of preserving solutions did not extend the vase life.

The use of chitosan and its derivatives to improve postharvest life of several horticultural commodities due to its fungicidal effects and elicitation of defence mechanism in plant tissues has been proposed, (Terry and Joyce, 2004). Chitosan is a biopolymer produced by deacetylation of chitin (Lodhi et al. 2014). Hong-juan and Huan-qing (2015) reported the effectiveness of the coating of cut roses with chitooligosaccharide (a derivate of chitosan) with increase of vase life by 6.4 d compared with the control treatment. However, there are no reports about the effects of chitosan on the vase life of heliconias flowers. Our objective was to assess the effects of chitosan coating on the vase life of flower stems of *H. bihai* (L.) L. cv. Halloween.

2.3 Materials and methods

2.3.1 Plant material

Flower stems of *H. bihai* (L.) L. cv. Halloween were obtained from a commercial plantation at 780 masl, at Campo Grande, Veracruz, Mexico (18º49´30.0´´N and 97º00´54.5´´W). Harvests were made between June and July of 2015. The inflorescences had four to six open bracts and 0.8 m long stems without leaves. The flower stems were cut at 8:00 am at an inclination at 45 º, following farm practices.

The whole stems were sprinkled with water and 0.2 m of the stems were immersed in water for 4 h during transport to the laboratory.

The stems were randomly distributed into three treatments; for each treatment four evaluations were made (day 1, 5, 10 and final day of vase life). Each evaluation time was composed for six replicates. The treatments assessed were 1.0 and 1.5 % chitosan, and the control treatment sprinkled with distilled water.

To prepare 2 L of 1.0 and 1.5 % chitosan solution, 20 or 30 g of chitosan (low molecular weight, Sigma-Aldrich) were added in 1.8 L of distilled water plus 200 mL of glacial acetic acid to dissolve the chitosan. The pH of the solution was adjusted to 5. The flower stems were covered with the solution using a natural bristle brush and allowed to dry after coating for one hour, then the flower stems of each treatment were placed individually into a bucket containing 800 mL distilled water and stored at room temperature (26 ºC ± 4 and 40 ± 5 % RH).

2.3.2 Variables evaluated:

2.3.2.1 Fresh weight loss

The stem flower fresh weight was recorded with a digital balance (Precisa, XB 2200C®). The results were expressed as a percentage of fresh weight loss with respect to the original.

2.3.2.2 Colorimetry (L, C*, hº)*

The colour of basal and apical (first and last) bracts was evaluated by analysing digital images (Samsung®, 12 Mp) with the app ColorPixLab (Colegio de Postgraduados) in MATLAB® Version 7.10.0. The colour space CIE L*C*hº (luminosity, chroma and hue angle, respectively) was used for the determinations. The photographs were taken at the same position, time of day (10 am) and distance (0.3 m). Based on the reading taken by the CIE L*a*b* model, chroma (C*) and hue (hº) were calculated using the following equations (Aular et al*.,* 2002):

$$C^* = (a^{*2} + b^{*2})^{1/2}$$
$$h^\circ = Tan^{-1}\left(\frac{b^*}{a^*}\right)$$

2.3.2.3 Percentage of absolute integrity of cell membrane (PAI) and electrical conductivity

The percentage of absolute integrity of cell membrane (PAI) was assessed according to Costa et al. (2011a) and Maki et al. (2010). Tissue disc samples (1 g) were obtained with a hollow punch from the basal stem, peduncle, basal and apical bract. The electrical conductivity was evaluated in fresh and frozen tissues of all treatments. The measurements were made with a conductivity meter (Horiba B-173®) every 30 min over 2 h, the values were expressed as μS cm^{-1} g^{-1} of fresh weight; PAI was calculated with the following equation:

$$PAI\ (\%) = \left(1 - \frac{FC}{TC}\right) * 100$$

Where: FC is the free conductivity and TC the total conductivity (frozen tissue) (Azevedo et al., 2008).

2.3.2.4 Anthocyanin

Anthocyanin in the basal bracts was evaluated according to Rahmani et al. (2015), Salinas et al. (2003), and Krizek et al. (1993); 1 g of lyophilized ground sample was extracted in 5 mL 1.0 % HCl-methanol solvent (1:99, v:v). The samples were centrifuged at 10,000 g and kept in the dark overnight. The absorbance was measured in a spectrophotometer (Genesys 10 UV, Thermo Electron Corporation®) at 520 nm in an aliquot of 4 mL of the filtered extract. Anthocyanin concentration was calculated using an extinction coefficient of 33,000 mol^{-1} cm^{-1}. The anthocyanin concentration was expressed as μM g^{-1} of dry weight tissue.

2.3.2.5 Total flavonoids

Flavonoid concentrations in the basal bracts were measured according to Rahmani et al. (2015), Barrón et al. (2011), and Krizek et al. (1993). Lyophilized bracts samples (1 g) were extracted in 3 mL 1 % acetic acid-ethanol solvent (1:99, v:v), centrifuged at 10,000 g, shake and heated at 80 ºC in water bath for 10 min. The absorbance was measured at 415 nm for the quantification of the concentration. A calibration curve of catechin (0.1 mg L^{-1}) was made in concentrations of 0 to 100 μL with intervals of 20 μL. The concentration of total flavonoids was expressed as mg of catechin equivalent per kg of bract dry weight.

2.3.2.6 Total sugars

Samples of 1 g of fresh tissue of the basal part of each stem, peduncle, and basal and apical bracts were frozen in liquid nitrogen and stored at -20 ºC until evaluation. Subsequently, the samples were ground and boiled in ethanol (80 ºC) for 5 min, mixed and filtered to obtain the alcoholic extract.

Total sugar content was calculated using the anthrone method on 0.5 mL of alcoholic extract (Whitman et al., 1971). The absorbance was measured at 620 nm. The sugar concentration was estimated from a standard curve of glucose (0 to 100 μg mL^{-1}).

2.3.2.7 Vase life

The visual quality of inflorescences was rated using a 1 – 4 scale, where grade 1 was considered as the worst quality and 4 as the best condition (Costa et al. 2011b). The end of vase life was considered when more than 50 % of flowering stems had reached grade 1.

2.3.3 Experimental design and statistical analysis

An unbalanced repeated measures design (RDM) was used to analyse the effects of chitosan on fresh weight loss and changes in bract colour for four evaluations (1, 5, 10 and final day of vase life). A Tukey test ($\alpha = 0.05$) comparison was established

beforehand among treatments (day 1 and final) when the interaction of treatments with time was significant.

A completely random design (CRD) with an analysis of variance and Tukey test (α=0.05) was used to analyse PAI, anthocyanin, total flavonoids, total sugars, electric conductivity, and duration of vase life.

Tissue from basal stem, peduncle, basal and apical bracts was used to evaluate PAI and total sugars. Basal and apical bracts were evaluated to determine changes in colour (L*, chroma and hue) and anthocyanin and total flavonoids were evaluated with tissue from basal and apical bracts. SAS® software (version 9.3) was used for the statistical analysis.

2.4 Results

2.4.1 Vase life

Differences among treatments for vase life duration were found (Figure 2.1). Longest vase life (19.9 and 16.6 d) was recorded in stems treated with chitosan 1.0 and 1.5 %, respectively. There were no significant differences between chitosan treatments; however, the treatment with 1.0 % chitosan increased the vase life 3.3 days more than 1.5 % chitosan treatment, and 10.3 d more than the control.

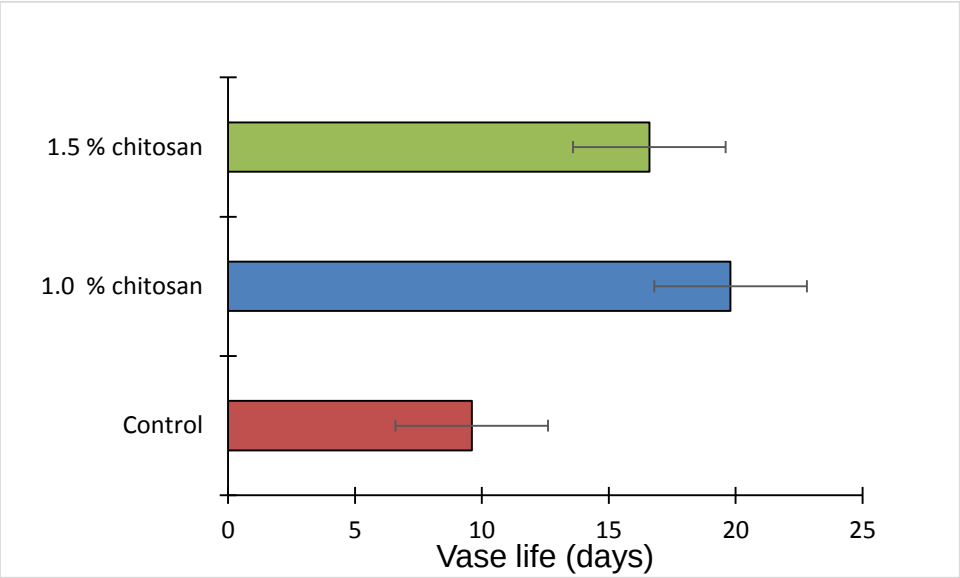


Figure 2.1. Vase life of stem flowers of *Heliconia bihai* (L.) L. cv. Halloween. Each bar represents the mean of six observations ± SE.

2.4.2 Fresh weight loss

The interaction between treatments and time was significant. All treatments showed progressive fresh weight loss until day 5 (Figure 2.2 and 2.3) and after that it stayed constant. At the end of vase life, treatments with chitosan showed a small loss of fresh weight loss (average of 9.3 %), close to one half that of control loss (17.5 %). The control treatment evaluation was completed on day 10 because more than 50 % of the stems were classified as grade 1 according to the scale of visual quality.

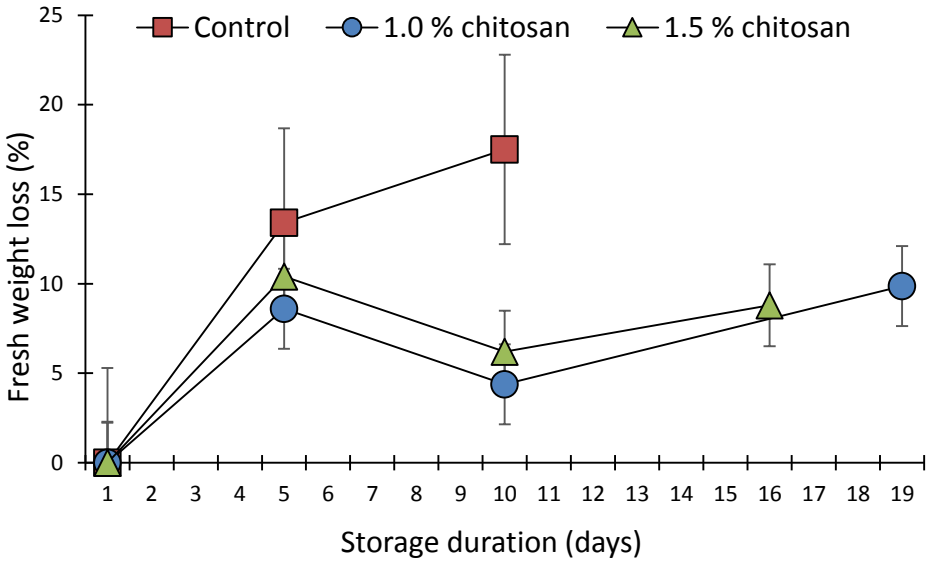


Figure 2.2. Percentage of fresh weight loss of stem flowers of *Heliconia bihai* (L.) L. cv. Halloween during the vase life period. Each bar represents the mean of six observations ± SE.

Figure 2.3. Flowers stem of *Heliconia bihai* (L.) L. cv. Halloween according to time and treatment. A= Initial state of inflorescences (day 1); B= chitosan treatment 1.0 % at final vase life (19.9 d); C= chitosan treatment 1.5 % at final vase life (16.6 d); D= control treatment at final day vase life (9.6 d).

2.4.3 Colorimetry

2.4.3.1 Luminosity (L)*

The interaction treatment x time was significant for basal bract and apical bract, and as consequence the Tukey test was applied to compare between the 1 d and the final day of vase life in the same treatment.

The luminosity of basal bracts on days 1, 5 and 19 was higher than day 10, but not between day 1 and final day of evaluation. The 1.5 % chitosan treatment showed the greatest L* loss on day 10, but the L* did not show differences among vase life days (Figure 2.4-A).

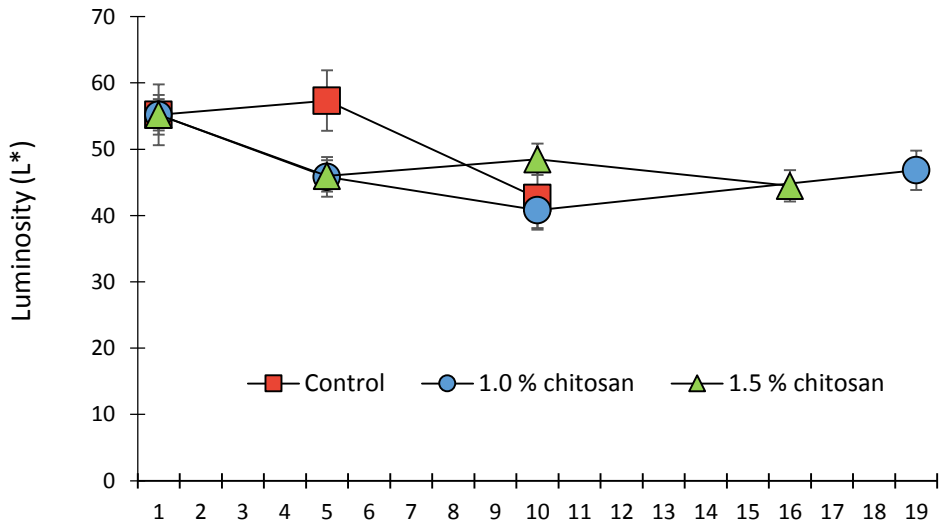
For apical bracts, L* values did not differ over time for the initial L* brightness was maintained until the end of vase life. Control had the lowest L* on day 10 with a loss of 15.3 % in comparison with chitosan treatment 1.0 % (10 % loss) (Figure 2.4-D).

2.4.3.2 Chroma (C)*

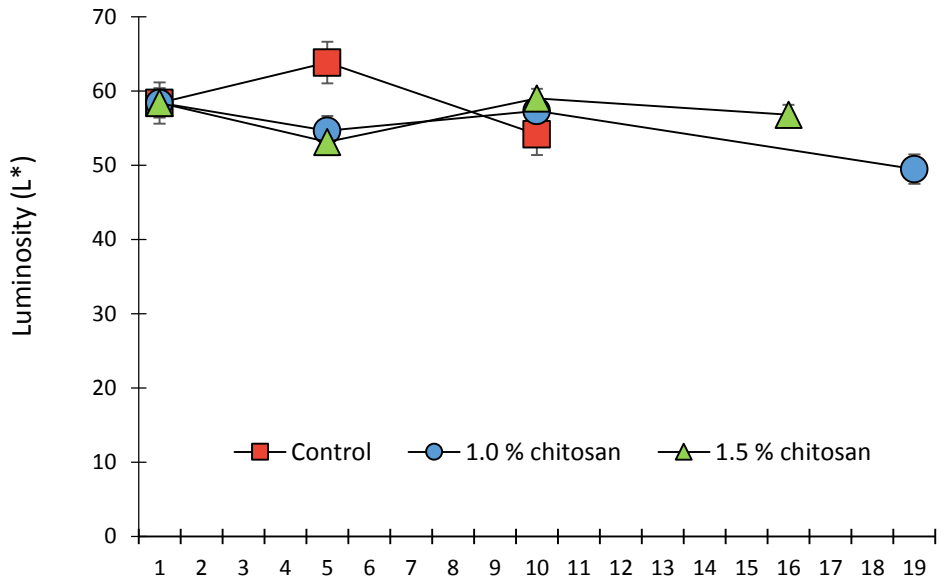
For both, basal and apical bracts the interaction treatment x time was significant. On basal bracts in the 1.0 % chitosan treatment best retained the chroma values during the experiment, while for the 1.5 % chitosan treatment a slight loss was evident on day 5 (Figure 2.4-B). For apical bracts, chitosan treatments showed an increase from day 1 to day 5, and subsequently presented a loss until the end of the experiment (Figure 2.4-E). In both structures the control treatment showed an accelerated decrease during the study.

2.4.3.3 Hue (hº)

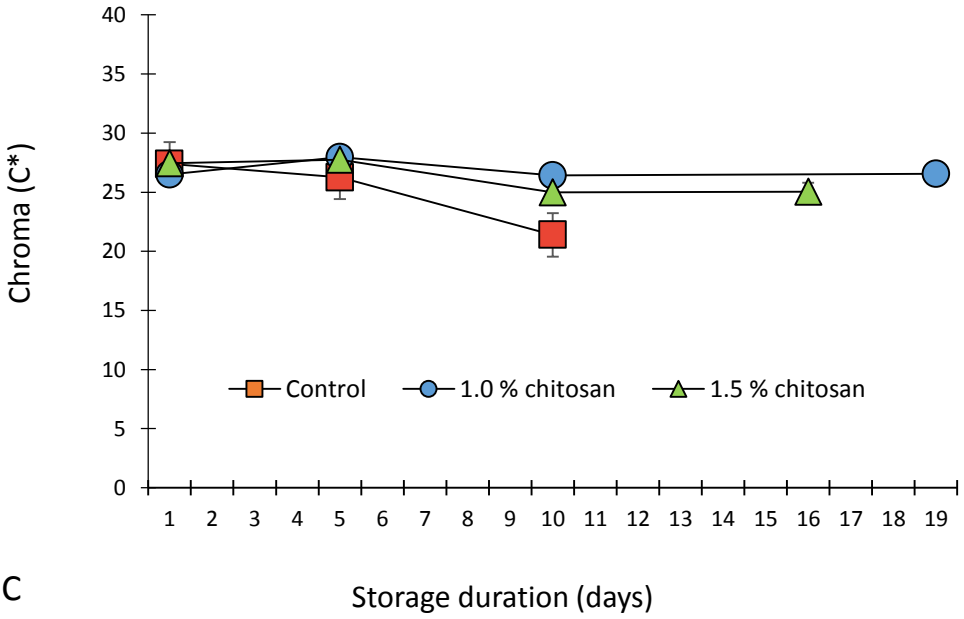
The interaction of treatment x time was significant for both basal and apical bracts. However, the Tukey test did not detect changes in this attribute of any of the treatments evaluated between day 1 and the end of the evaluations. The hue of the basal bracts increased in stems treated with chitosan 1.5 % from the beginning to day 5, but was not affected by any other treatment (Figure 2.4-C). The hº of the apical bracts increased after chitosan treatment on day 1 and then declined (Figure 2.4-F). In contrast, the control presented a slight decline from the start to final, but with hue loss values lower than those of chitosan treatments.



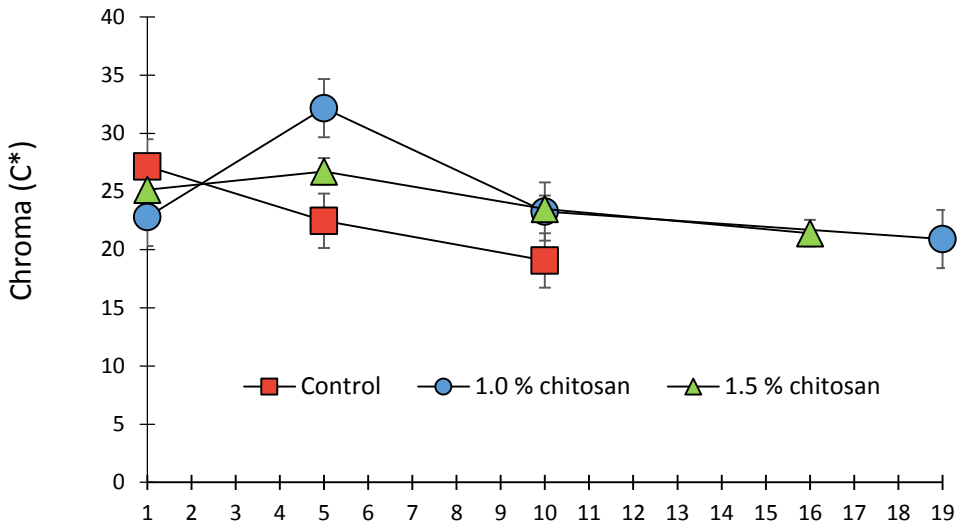
A


Luminosity (L*)
70
60
50
40
30
20
10
0
Control
1.0 % chitosan
1.5 % chitosan
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19
Storage duration (days)

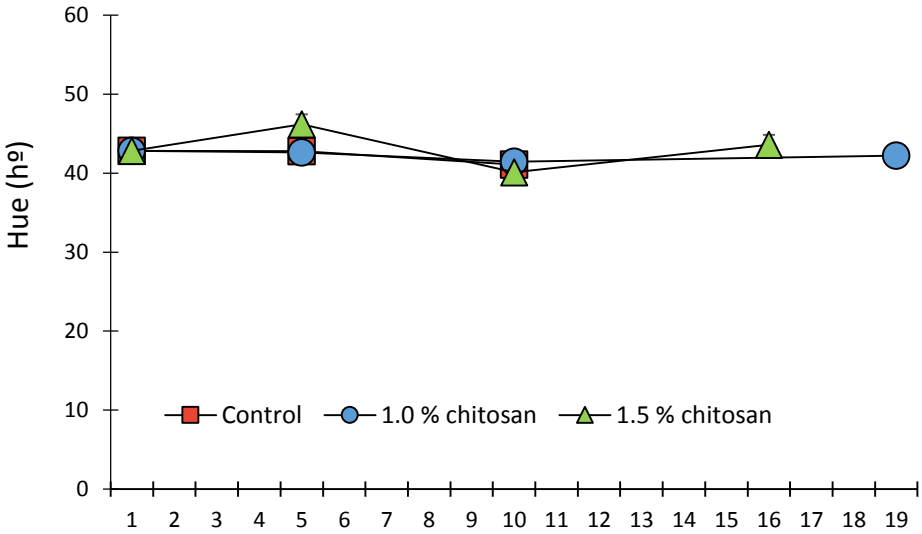
B


Chroma (C*)
40
35
30
25
20
15
10
5
0
Control
1.0 % chitosan
1.5 % chitosan
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19
Storage duration (days)

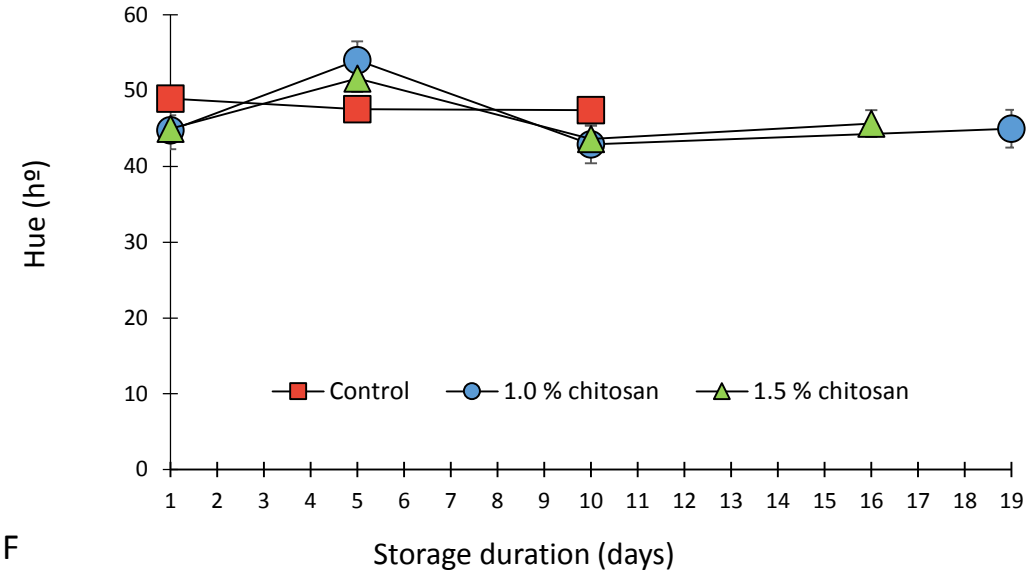
C


Chroma (C*)
40
35
30
25
20
15
10
5
0
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19
Control
1.0 % chitosan
1.5 % chitosan
Storage duration (days)

D


Hue (hº)
60
50
40
30
20
10
0
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19
Control
1.0 % chitosan
1.5 % chitosan
Storage duration (days)

E



F

Figure 2.4. Colour progress in bracts over duration of vase life of fresh stem flowers of *Heliconia bihai* (L.) L. cv. Halloween. Chitosan 1.0 %, chitosan 1.5 %, and the control. Each bar represents the mean of six observations ± SE. A: luminosity (L*) on basal bracts (BB), B: Chroma on BB, C: Hue on BB, D: luminosity (L*) on apical bracts (AB), E: Chroma on AB, F= Hue on AB.

2.4.4 Percentage of absolute integrity of cell membranes (PAI) and electrical conductivity

The differences in PAI between treatments were significant for the basal and apical bracts at final day of vase life (Figure 2.5), chitosan treatments showed a higher PAI in comparison with the control. Electrical conductivity from day 5 showed a gradual increase (Figure 2.6). The peduncle showed the lowest electrolyte leakage during day 10 compared with chitosan treatments. PAI for the basal part of stem and peduncle did not differ from day 1 to the final day of storage. These results suggest that chitosan may allow to the young tissues to remain intact for longer in this species or that the impermeable chitosan did not allow the same ion leakage.

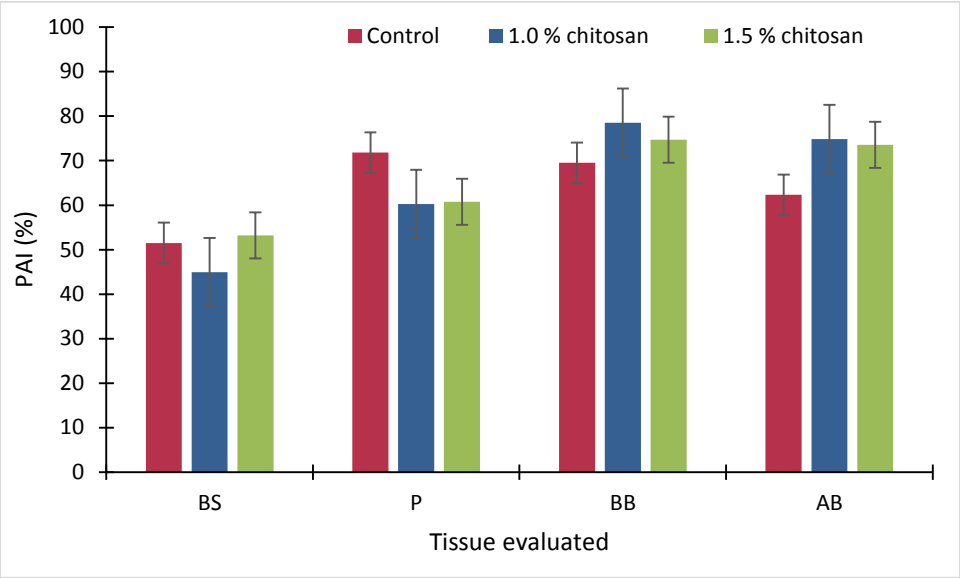
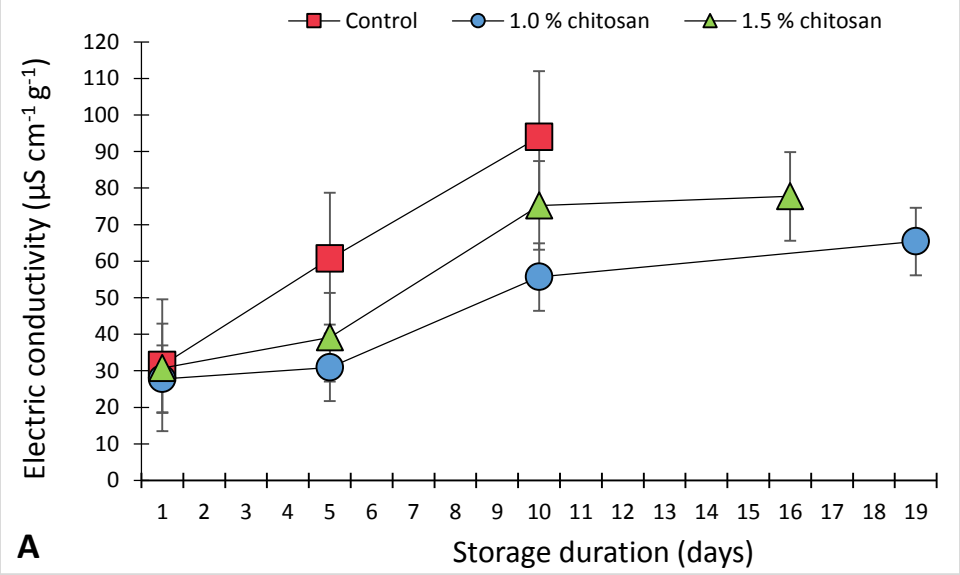


Figure 2.5. Percentage of absolute integrity of cell membranes (PAI) of fresh flower stems of *Heliconia bihai* (L.) L. cv. Halloween at the end of vase life. Each bar represents the mean of six observations ± SE. BS: basal part of stem, P: peduncle, BB: basal bract and AB: apical bract.



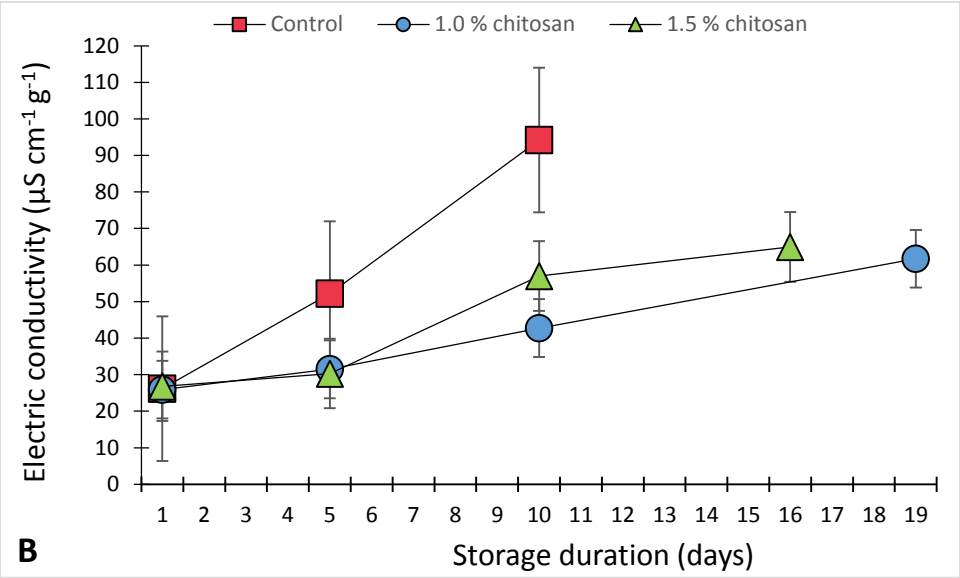


Figure 2.6. Electrical conductivity of fresh stem flowers of *Heliconia bihai* (L.) L. cv. Halloween during the vase life period. A = basal bract (BB), B = apical bract (AB). Each bar represents the mean of six observations ± SE.

2.4.5 Anthocyanin

The anthocyanin concentration in the basal bract differed between chitosan treatments and the control. The control had the greater concentration at the beginning of the experiment, and on day 5 (Figure 2.7). Nonetheless, at day 10 the anthocyanin concentration decreased in the control. Chitosan treatments showed a progressive increase from day 1 to day 10. Even though the control had the highest concentration at the beginning, it showed the greatest decrease over time; from the beginning to the final day (17 %) in contrast to the 1.0 and 1.5 % chitosan treatments, which had an increase of 22.5 and 26 %, respectively by the end of vase life.

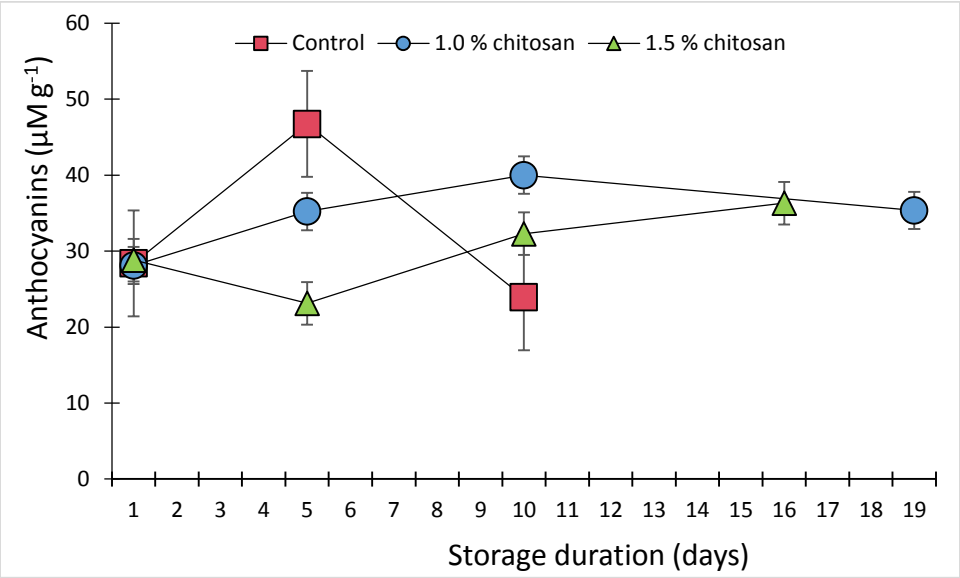


Figure 2.7. Anthocyanin concentration of the basal bract (BB) of flower stems of *Heliconia bihai* (L.) L. cv. Halloween over the vase life period. Each bar represents the mean of six observations ± SE.

2.4.6 Flavonoids

Chitosan treatments had a greater total flavonoids concentration in the flowers at the end of vase life than on day 1. Stem flowers coated with chitosan 1.5 % had the highest increase of flavonoid concentration from day 1 to 5 (46 %); corresponding to 19.1 mg kg^{-1} of catechin equivalent. Significant differences on flavonoids from day 1 to 10 were found on the control, showing a decrease of 27.6 % with respect to day 1, (Figure 2.8).

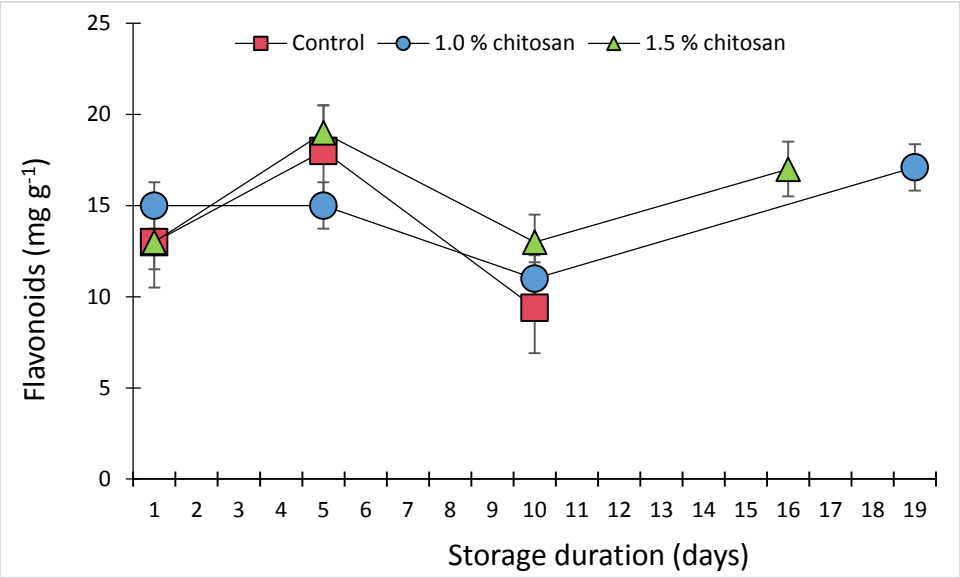


Figure 2.8. Total flavonoid concentration in the basal bract (BB) of stem flowers of *Heliconia bihai* (L.) L. cv. Halloween over the vase life period. Each bar represents the mean of six observations ± SE. The results are expressed as mg of catechin equivalent per gram.

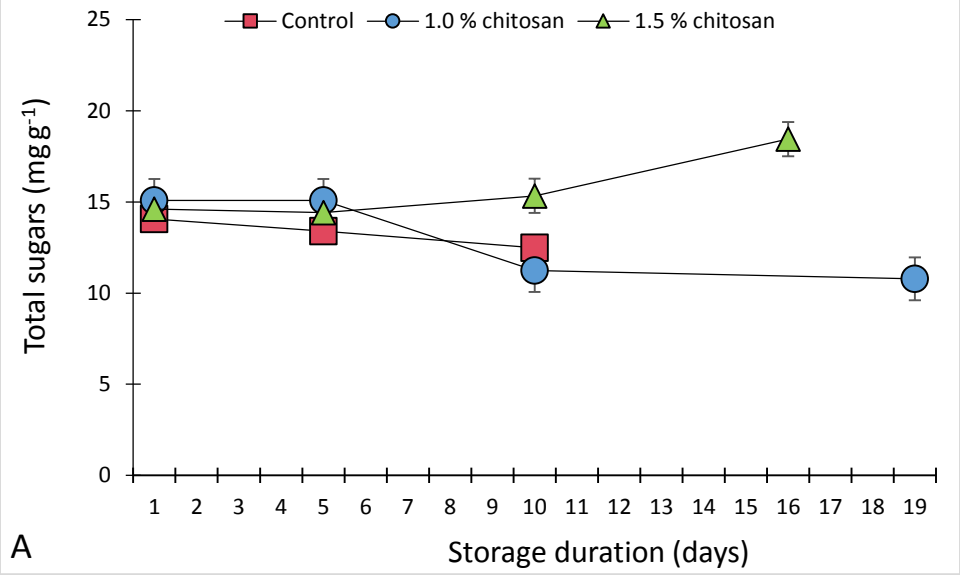
2.4.7 Total sugars

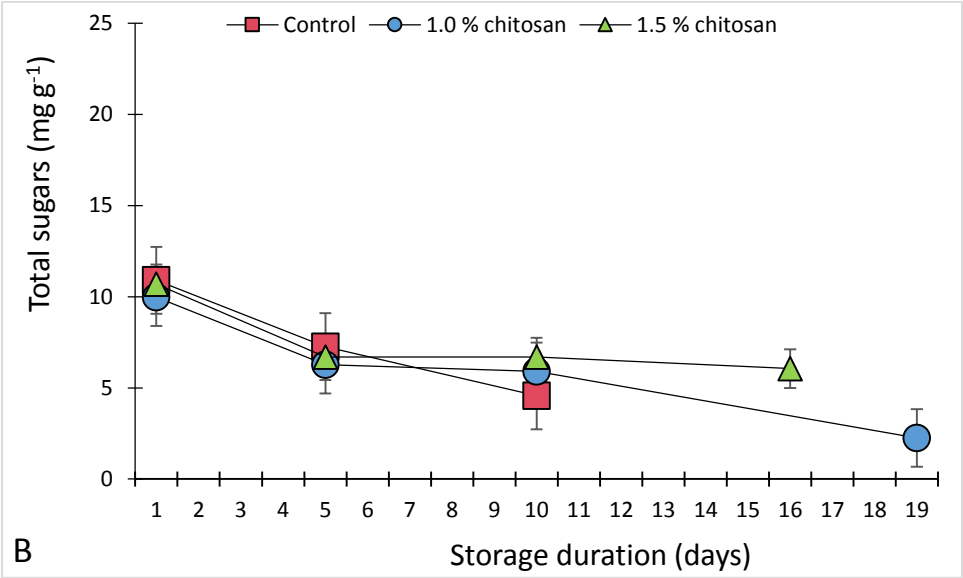
Differences were found in total sugars concentration of the basal stem between the beginning and end of the experiment (Figure 2.9-A), chitosan 1.0 % and control treatments showing a decline from the beginning to the end of the experiment, in contrast, chitosan 1.5 % increased from day 5 to the final of the vase life.

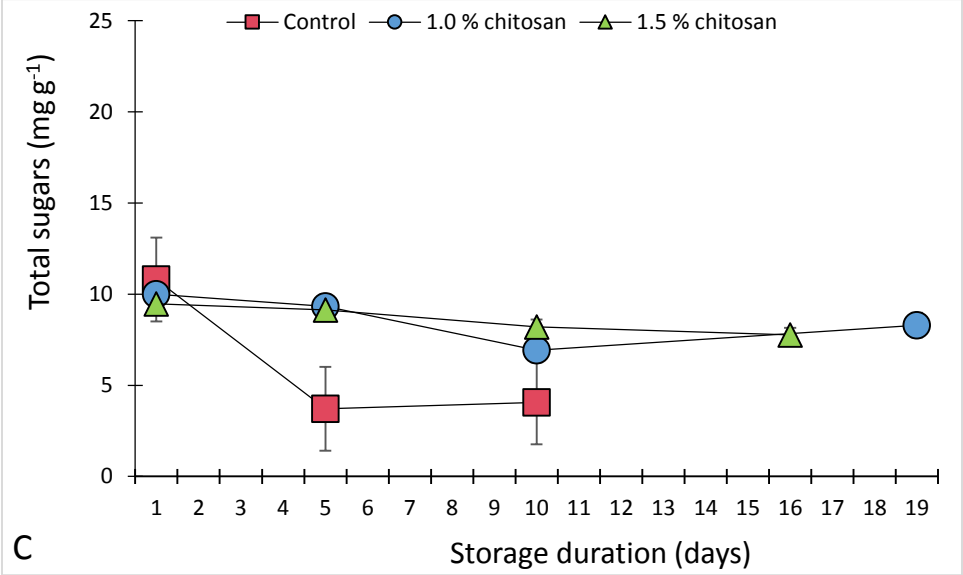
Total sugar concentration in the peduncle (Figure 2.9-B) differed between treatments and evaluation times. All treatments had high levels of sugar concentration on day 1 with a subsequent decrease until the final sample day, except chitosan 1.5 % treatment, which remained constant from day 5 to the end.

In the basal bracts chitosan treatments had slight decreases in sugar concentration from start to finish, in comparison with control that had an accelerated decline from day 5, and remained constant until the final. At the end of vase life, chitosan treatments maintained better levels of sugar concentration than control (Figure 2.9-C).

In apical bracts chitosan treatment 1.0 % had a slight decrease until day 10, and a subsequent increase at the final sample in contrast to chitosan treatment 1.5 % that had an increase on day 5, and a later decrease to the end of vase life. Control had the greatest loss from day 1 to the end of the experiment (Figure 2.9-D).




Control
1.0 % chitosan
1.5 % chitosan
Total sugars (mg g-1)
25
20
15
10
5
0
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19
Storage duration (days)
B


Control
1.0 % chitosan
1.5 % chitosan
Total sugars (mg g-1)
25
20
15
10
5
0
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19
Storage duration (days)
C

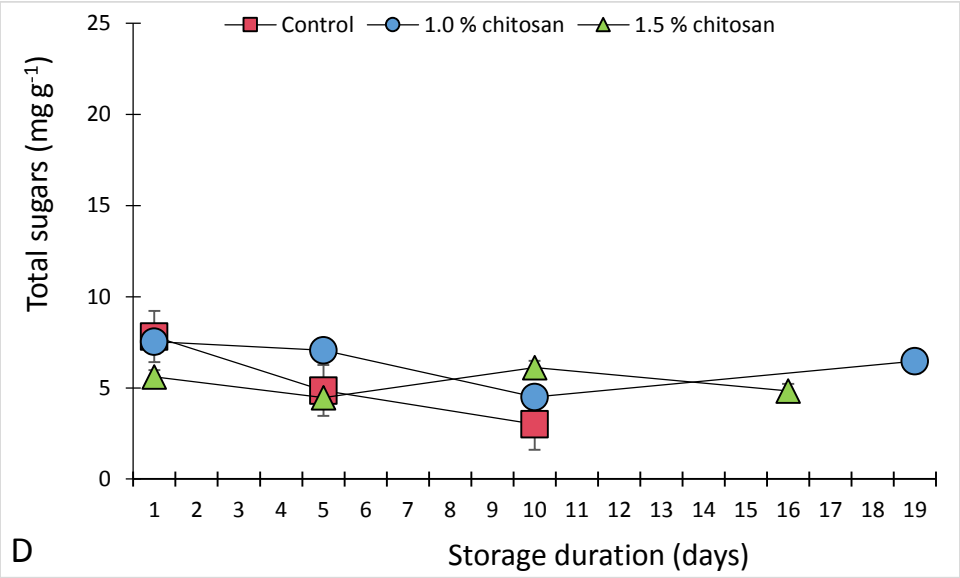


Figure 2.9. Total sugar concentrations of fresh stem flowers of *Heliconia bihai* (L.) L. cv. Halloween over the vase life period. A = basal stem (BS), B = peduncle (P), C = basal bract (BB), D = apical bract (AB). Each bar represents the mean of six observations ± SE.

2.5 Discussion

Chitosan positively affected quality of *Heliconias bihai* (L.) L. cv. Halloween flower stems during vase life. Beneficial properties of chitosan are related to its polycationic nature, chain length, inhibitory synthesis of certain fungal enzymes, production of phenolic compounds, and formation of structural barriers (Freepons, 1991; Benhamou et al., 1994; El Ghaouth, et al., 1997; Bhaskara et al., 2000)

Chitosan seemed to confer stability to the cell membrane and avoided flowering stems electrolyte leakage. As a consequence, the symptoms of senescence like

necrosis and loss of turgor were uniform across the flower stems treated with chitosan. In contrast, senescence of the control flower stems was heterogeneous.

The low loss of fresh weight in stems coated with chitosan may be a result of a semipermeable cover formed by the biopolymer; diminishing transpiration and delays ripening as a result of the decrease of production of CO_2 and/or ethylene (Bautista-Baños et al*.,* 2005).

Hong-juan and Huan-qing (2015) reported the positive effect of chitooligosaccharide on vase life duration of cv. Red France roses treated stems had higher fresh weight compared with the control, and extended vase life by 6.4 d compared to the control, while in our study chitosan treatments increased vase life duration by 10.3 d (1.0 % chitosan) and 7 d (1.5 % chitosan) in comparison with the control, which may be due to the concentrations used. It has been found that the type of response is in function of the molecular weight (directly related to its antimicrobial activity), the concentrations (that affect the gas permeability) used and/or the pH of the solution (Liu et al*.,* 2006; Fei et al*.,* 2001). Studies on postharvest of longan fruit showed that chitosan concentrations of 2 % kept percentage of weight loss lower than the control (Jiang and Li, 2001).

In our study, a basipetal damage gradient was observed in the flowers stems. Basal and apical bracts treated with 1.0 and 1.5 % chitosan had the highest PAI; these structures also retained the highest levels of anthocyanin, total flavonoids and total sugars, which may be an indication that young tissues use chitosan more efficiently than older tissues. These results suggest that chitosan had positive effect in these organs. As they are the younger tissues this could be important in the

response of chitosan. Less membrane damage was related to a better maintenance of sugars, anthocyanins and flavonoids.

Loss of PAI results in a non-selective ion release as well as loss of soluble sugars and amino acids. Costa et al. (2011a) found that the PAI evaluated on the bracts, was constant in their control treatment (an average of 83.4 %) during 8 d of storage of *Heliconia* flower stems at 24 ºC with 66 % RH. In contrast, in our study, the control (26 °C ± 4 and 40 % ± 5 RH) had a lower PAI after 10 d of vase life (69 and 52 % in basal and apical bracts, respectively). The differences in temperature and RH between experiments could be the cause of the differences in the values of PAI.

Chroma is a measure of the colour intensity. Pale, weak or dark colours have low chroma values. The floral stems of the control treatment showed a significant loss of chroma in the basal bracts compared to treatments with chitosan. Similarly, there was loss of hue in the apical and basal bracts. The variability of hue across treatments could be a result of the limited synthesis of anthocyanins, resulting in an increase in the flower colour value (Tourjee et al., 1993).

Costa et al. (2011a) showed the effect of storage temperature on the inflorescence colouration of *H. bihai* cv. Halloween. They observed a decrease in L * values as a function of temperature with values between 58.7 and 49.9. In our study the values ranged between 63.8 and 40.8 (data not presented).

Studies by Jiang et al. (2005) and Ducamp et al. (2008) support the idea that chitosan can improve the colouration, maintain the quality and increase shelf life in litchi fruit by the blocking of polyphenol oxidase activity. The intensity of red colouration is closely related to the concentration of anthocyanins. In the present study, the control had the largest decrease in L*, chroma, hue (except in the apical

bracts), and anthocyanins, showing that colour is closely related with the anthocyanins and possibly with others pigments such as carotenoids and betalains, responsible for granting bracts their colour.

Ducamp et al. (2008) reported on the effect of 0.75 % chitosan and citric acid on retention of a red colouration of litchi fruit in the cultivars red Kwai-Guiwei and Wai chee-Huaizhi. Both cultivars showed a better conservation of anthocyanin after 21 d of storage in comparison with the control. Nevertheless, significant differences between cultivars treated with chitosan were found; cv. Wai chee-Huaizhi lost 75 % of cyaniding-3-rutinoside and 70 % of cyaniding-3-glucoside, while cv. Kwai-Guiwei lost 25 % and 45 %, respectively.

Chitosan stimulates the synthesis of phenolic compounds, including flavonoids (Bautista et al., 2005). At the same time, secondary metabolites such as flavonoids, anthocyanins, betalains and carotenoids play a key role in the flower pigmentation (Tanaka et al., 2008). In our study, from the beginning to day 10 there was an increase in the production of these compounds in the chitosan treatments, while control had a marked decrease from day 5.

A positive correlation of PAI in apical bracts with anthocyanins ($r = 0.5$), total flavonoids ($r = 0.68$) and sugars ($r = 0.53$) in the bracts was observed. Bracts with less damage (high PAI value) preserved colour and anthocyanin better, and increased sugar throughout the duration of vase life.

The strong electron density of chitosan found by Benhamou et al. (1994) suggests that it could be enriched with phenolic compounds, especially with phenols containing *O*-dihydroxy groups. It appears that chitosan treatments trigger either the

novo synthesis of phenolic compounds, or the polymerization of pre-existing free, soluble phenols, or both.

In our study, chitosan may have an effect on the availability of sugars in cells of the floral bracts. Some discolouration seems to indicate that a large fraction of the soluble carbohydrates are unavailable for respiration, at the time of wilting. A small shift in vacuolar pH is adequate to cause a change of the co-pigmentation of anthocyanins, which are located in the vacuole (Paulin, 1971; Asen et al*.,* 1971). Even though there are adequate levels of available carbohydrates, the mitochondria may not be able to use or transport them from the cytosol (van Doorn, 1999).

Our results show a lower loss of sugars in bracts treated with chitosan (average loss of 2.25 mg kg^{-1} of fresh weight in contrast with 4.8 mg kg^{-1} of the control, from the beginning of the experiment to the end), especially in the apical bracts, which are consider as the youngest tissues in the flowering stems. This fact can be explained because these organs have a high metabolic rate, and therefore compete with other structures (peduncle and base of stem, in this case) for photosynthates (Ho, 1988).

2.6 Conclusions

Chitosan treatments improved quality and vase life of fresh flower stems in comparison with untreated flowers stems. Chitosan at both 1.0 and 1.5 % were equally effective protecting fresh flower stems. Young bract tissue (apical) showed less cell damage than the older bracts and the peduncle. The literature shows that chitosan functions in different ways, depending on species, and even cultivar. The

results found in this study represent a breakthrough in research on the use of chitosan in the postharvest treatment of tropical ornamentals, providing the precedents for future studies focused on the specific mechanisms of chitosan action on cellular systems.

2.7 Acknowledgments

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2.8 References

Asen, S., Norris, K.H., Stewart, R.N. 1971. Effect of pH and concentration of the anthocyanin-flavanol co-pigment complex on the colour of Better Times roses. J. Amer. Soc. Hort. Sci. 96:770 – 773.

Aular, J. Ruggiero, C., Durigan, J. 2002. Relación entre el color de la cáscara y las características del fruto y jugo de la parchita maracuyá. Bioagro. 14(1):47 -51.

Azevedo, I.G., Oliveira, J.G., da Silva, M.G., Pereira, T., Corrêa, S.F., Vargas, H., Façanha, A.R., 2008. P-type H^+-ATPases activity, membrane integrity, and apoplastic pH during papaya fruit ripening. Postharvest Biol. Technol. 48, 242 – 247.

Barrón, Y.R., García, M.A., Soto, H.R., Colinas, L.T. Geoffrey, K. 2011. Flavonoides y actividad antioxidante de *Calia secundiflora* (Ort.) Yakovlev. Rev. Fitotec. Mex. 34(3):151 – 157.

Bautista, B.S., Hernández, L.A.N., Velázquez del Valle, M.G., Bosquez, M.E., Sánchez, D.D. 2005. Quitosano: Una alternativa natural para reducir microorganismos postcosecha y mantener la vida de anaquel de productos hotofrutícolas. Revista Iberoamericana de Tecnología Postcosecha. 7(1):1 – 6.

Benhamou, N., Lafontaine, P.J., Nicole, M. 1994. Induction of systemic resistance to *Fusarium* crown and root rot in tomato plants by seed treatment with chitosan. Phytopathology. 84(12):1432 - 1444.

Bhaskara, R.B.M.V., Belkacemi, K., Corcuff, F.C. Arul, J., Angers, P. 2000. Effect of pre-harvest chitosan sprays on post-harvest infection by *Botrytis cinerea* and quality of strawberry fruit. Postharvest Biol. Technol. 20:39 - 51.

Broschat, T.K., Donselman, H.M., 1983. Production and postharvest culture of *Heliconia psittacorum* flowers in south Florida. Proc. Fla. State Hortic. Soc. 96:272 – 273.

Costa, A.S., Nogueira, L.C., dos Santos, V.F., Finger, F.L., Camara, T.R., Loges, V., Willadino, L., 2011a. Characterization of symptoms of senescence and chilling injury on inflorescences of *Heliconia bihai* (L.) cv. Lobster Claw and cv. Halloween. Postharvest Biol. Technol. 59:103 – 109.

Costa, A.S., Nogueira, L.C., dos Santos, V.F., Camara, T.R., Loges, V., Willadino, L., 2011b. Storage of cut *Heliconia bihai* (L.) cv. Lobster Claw flowers at low temperatures. Rev. Bras. Eng. Agric. E: Ambient. 15:966 – 972.

Ducamp, C.M.N., Ramarson, H., Lebrun, M., Self, G., Reynes, M. 2008. Effect of citric acid and chitosan on maintaining red colouration of litchi fruit pericarp. Postharvest Biol. Technol. 49:241 – 246.

El Ghaouth, A., Arul, J., Wilson, C., Benhamou, N. 1997. Biochemical and cytochemical aspects of the interactions of chitosan and *Botrytis cinerea* in bell pepper fruit. Postharvest Biol. Technol. 12:183 – 194.

Fei, L.X., Lin, G.Y., Zhi, Y.D., Li, Z., De, Y.K. 2001. Antibacterial action of chitosan and carboxymethylated chitosan. J. Appl. Polym. Sci., 79:1324 – 1335.

Freepons, D. 1991. Chitosan, Does it have a place in agriculture? Proc. Plant Growth Regulators Soc. Amer. 11 - 19.

Ho, L.C. 1988. Metabolism and compartmentation of imported sugars in sink organs in relation to sink strength. Annu. Rev. Plant. Physiol. Plant. Mol. Biol. 39:355 – 378.

Hong-juan, J., Huan-qing, L. 2015. Chitooligosaccharide prolongs vase life of cut roses by decreasing reactive oxygen species. Kor. J. Hort. Sci. Technol. 33(3):383 – 389.

Jerez, E. 2007. El cultivo de las Heliconias. Cultivos Tropicales. 28(1):29 - 35.

Jiang, Y., Li, Y., 2001. Effects of chitosan coating on postharvest life and quality of longan fruit. Food Chem. 73, 139–143.

Jiang, Y., Li, J., Jiang, W. 2005. Effects of chitosan coating on shelf life of cold-stored litchi fruit at ambient temperature. LWT Food Sci. Technol. 38: 757 – 761.

Ka-ipo, R., Sakai, W.S., Furutani, S.C., Collins, M. 1989. Effect of postharvest treatment with antitranspirants on the shelf-life of *Heliconia psittacorum* cv. Parakeet cut flowers. Bul. Heliconia Soc. Intl. 4:13 – 14.

Krizek, D.T., Kramer, G.E., Upadhyaya, A., Mirecki, R.M. 1993. UV-B response of cucumber seedling grown under metal halide and high pressure sodium/deluxe lamps. Physiol. Plantarum. 88:350 – 358.

Leyva, O.O.R., Rodríguez, A.Y., Herrera, C.J.A., Galindo, T.M.E., Murguía, G.J. 2011. Polímero hidrofílico combinado con soluciones preservadoras en la vida de florero de tallos florales de rosa y heliconia. Tropical and Subtropical Agroecosystems. 13:551 – 559.

Liu, N., Chen, X.G., Park, H.J., Liu, C.G., Liu, C.S., Meng, X.H., Yu, L.J. 2006. Effect of MW and concentration of chitosan on antibacterial activity of *Escherichia coli*. Carbohyd. polym. 64(1):60 - 65.

Lodhi, G., Kim, Y.S., Hwang, J.W., Kim, S.K., Jeon, Y.J., Je, J.Y., Ahn, C.B., Moon, S.H., Jeon, B.T., Park, P.J., 2014. Chitooligosaccharide and its derivatives: preparation and biological applications. BioMed Res. Int. Vol. 2014, Article ID 654913, 13 pages, 2014.

Maki, D.G., Peña, V.C.B., Gutiérrez, E.J.A., Arévalo, G.M.L., Gaytán, A.A. 2010. Curvatura geotrópica, daño en membranas celulares y tasa respiratoria en tallos florales de *Antirrhinum majus* L., cv. Potomac White Ivory, tratados con AOA y TIBA. Rev. Fac. Agron. 27:574 – 593.

Paulin, A. 1971. Influence de la composition de la solution mutritive sur la teneur en divers acides aminés libres et en ammoniac des pétales de fleurs coupées. Ann. Techn. Agric. 20:283 – 303.

Rahmani, I., Ahmadi, N., Ghanati, F., Sadeghi, M. 2015. Effects of salicylic acid applied pre- or post-transport on post-harvest characteristics and antioxidant enzyme activity of gladiolus cut flower spikes. New Zeal. J. Crop Hort. 43:4, 294-305,

Salinas, M.Y., Martínez, B.F., Soto, H.M., Ortega, P.R., Arellano, V.J., 2003. Efecto de la nixtamalización sobre las antocianinas del grano de maíces pigmentados. Agrociencia. 37(6):617 - 628.

Tanaka, Y., Sasaki, N., Ohmiya, A. 2008. Biosynthesis of plants pigments: anthocyanins, betalains and carotenoids. Plant J. 54:733 – 749.

Terry, L.A., Joyce, D.C. 2004. Elicitors of induced disease resistance in postharvest horticultural crops: a brief review. Postharvest Biol. Technol. 32:1 – 13.

Tourjee, R.K., Harding, J., Byrne, T. 1993. Calorimetric analysis of Gerbera flowers. HortScience, 28(7):735 - 737.

van Doorn, W.G. 1999. Role of soluble carbohydrate in flower senescence: A survey. Acta Hort. 543:179 – 183.

Witham, F.H., Blaydes, D.F., Devlin, R.M. 1971. Experiments in Plant Physiology. Van Nostrand Reinhold Company. New York, USA. 245 p.

3. COLD STORAGE AND CHITOSAN COATING EFFECTS ON VASE LIFE OF FLOWERING STEMS OF *Heliconia psittacorum* L. f. cv. Trópica

COLD STORAGE AND CHITOSAN COATING EFFECTS ON VASE LIFE OF FLOWERING STEMS OF *Heliconia psittacorum* L. f. cv. Trópica

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3.1 Abstract

Alternative postharvest techniques to extend the vase life of tropical ornamental species are a crucial factor for the growth of the flower market. *Heliconia psittacorum* L. f. cv. Trópica stands out due to their great demand. Two cold storage conditions (12 °C and 16 °C), two chitosan concentrations (1.0 % and 1.5%) without cold storage and a control maintained at room temperature (25 ± 2 ºC and 32 ± 5 % RH) were evaluated and compared. Changes in fresh weight, bract colour (luminosity, chroma and hue), anthocyanins, total flavonoids and total sugars were evaluated. 1.0 and 1.5 % chitosan treatment had the total postharvest longevity (TPL) of 23.1

and 21 days, it was 91 and 75 % higher than the control (12 days); cold storage at 16 °C (17 days) and 12 °C (16 days) also increased TPL, but to a lesser extent. In general, chitosan treatments delayed colour changes in floral bracts; and along with 16 °C storage treatment preserved luminosity, chroma, total flavonoids and total sugars. No significant differences were found in anthocyanin content among the treatments at the end of vase life. The use of chitosan as a coating for tropical ornamentals is an alternative to extend vase life, surpassing the results with traditional methods such as cold storage.

3.2 Introduction

The floriculture is important due to the high economic earnings per unit of harvest o cultivated area and the sale of new species is promoted to increase the market (Sosa, 2013). The genus *Heliconia* has outstanding ornamental characteristics, among which are great variety of shapes, colours, and sizes, coupled with the beauty of its flowers and its exoticism.

Heliconias are used as garden plants or as cut flowers. The bracts that protect the real flowers are jazzy with intense and exuberant colour, this characteristic favours their acceptance by the consumer (de Castro et al., 2007).

Inadequate techniques during storage negatively affect vase life and significant postharvest stem losses. The stems respiration and transpiration continue after harvest together with pathogen attacks, resulting in deterioration of quality and decrease of vase life (Akkerman et al., 2010).

Chitosan is a polysaccharide formed by poly-β-(1→4)N-acetyl-D-glucosamine units; it exhibits positive responses in controlling pre and postharvest diseases of agricultural products (Bautista-Baños et al., 2006), extending the shelf life of horticultural commodities (Liu et al., 2016).

Chitosan-based films and coating has a positive effects on maintaining quality during postharvest storage (Kerch, 2015). Studies performed by Chien et al. (2007b) showed the positive effects of chitosan coating on water loss, which was correlated with an increase of ascorbic acid during shelf life of sliced mango fruit. It has been suggested an induction of antioxidant activity by chitosan, the increase of superoxide dismutase and catalase activities, resulting in the inhibition of superoxide free radical production in guava fruits (Hong et al., 2012). Chitosan coating can delay the increase of polyphenol oxidase activity and suppress peroxidase activity in *Luffa cylindrica* during storage (Han et al., 2014),

The aim of this research was to investigate the effects of chitosan coating and comparing them with cold postharvest storage over the vase life of floral stems of *H. psittacorum* L. f. cv. Trópica.

3.3 Material and methods

3.3.1 Plant material

Flower stems were obtained from a commercial plantation at 780 m. a. s. l., at Campo Grande, Veracruz, Mexico (18º49´30.0´´ N and 97º00´54.5´´ W). Harvests were made between April and May of 2014 for cold storage experiment and between June and July of 2015 for chitosan experiment. Inflorescences of *H. psittacorum* L.

f. cv. Trópica had three to four open bracts and 80 cm long stems without leaves. The flower stems were cut at 8:00 am, with an inclination at 45º as a following farm practices. The whole stems were sprinkled with water and 20 cm of the stems were immersed in water for 4 h during transport to the laboratory at the Colegio de Postgraduados, Mexico.

3.3.2 Storage experiment

The flowering stems were placed individually into a container with 800 mL distiller water into a controlled environment chamber for storage over 10 days at 12 and 16 ºC, and the vase life was registered. Stems at room temperature (25 ºC ± 2 and 32 ± 5 % RH) were the control treatment. Each treatment included six experimental units and four times of evaluation (day 1, 5, 10 and final day).

3.3.3. Variables responses evaluated

3.3.3.1 Vase life

The visual quality of inflorescences was rated using a 1 – 4 visual scale, where 1 was the worst quality and 4 the best one (Costa et al., 2011b). A control treatment was set up with flowering stems without any previous chitosan treatment and maintained at room temperature as chitosan treatments. The end of vase life was considered when more than 50 % of flowering stems had reached grade 1. Total postharvest longevity was considered for the storage simulation period (10 days), plus post-storage vase life.

3.3.3.2 Fresh weight loss

The fresh weight (FW) of stem flower was recorded with a digital balance (Precisa, XB 2200C®). The results were expressed as a percentage of fresh weight loss with respect the original fresh weight.

3.3.3.3 Changes in bract colour (Luminosity, Chroma y Hue)

The colour of basal and apical (first and last) bracts were evaluated analysing digital images (Samsung®, 12 Mp) with the app ColorPixLab (Colegio de Postgraduados) in MATLAB® Version 7.10.0 using the colour system CIE L*C*hº. The photographs were taken at the same position, time of day (10 am) and distance (0.3 m). The parameters assessed were: luminosity (L= 0-100 units), green to red axis (a*= -128 to 127 units) and blue to yellow axis (b*= -128 to 127 units). a* and b* were used to calculate chroma (C*) and hue (hº) through the following equation (Aular et al., 2002):

$$C^* = (a^{*2} + b^{*2})^{1/2}$$

$$h^{\underline{o}} = Tan^{-1}\left(\frac{b*}{a*}\right)$$

Where C^* is chroma, hº is hue, a^* is red to green axis (128 to 127), b^* is yellow to blue axis (128 to 127), and y Tan^{-1} is the inverse tangent.

3.3.3.4 Anthocyanins

The anthocyanin content was evaluated according to Rahmani et al. (2015) and Salinas et al. (2003); 0.025 g of lyophilized ground sample of basal and apical bracts

were extracted in 5 mL of methanol with HCl (1 %). The samples were centrifuged at 10,000 g and kept in the dark overnight. The absorbance was measured in a spectrophotometer (Genesys 10 UV, Thermo Electron Corporation®), at 520 nm, in an aliquot of 4 mL of the filtered extract. Anthocyanin concentration was calculated using an extinction coefficient of 33,000 mol cm^{-1}. The anthocyanin content was expressed as mM g^{-1} of dry weight (DW).

3.3.3.5 Total flavonoids

Total flavonoids in the basal bracts were measured according to Rahmani et al. (2015) and Barrón-Yánez et al. (2011). Bracts samples (0.02 g) were extracted in 3 mL of ethanol containing acetic acid (1 %), centrifuged at 10,000 g and heated at 80 ºC in water bath for 10 min. The absorbance was measured at 415 nm for the concentration quantification. A calibration curve of catechin - CA (0.1 mg mL^{-1}) was made, including 0 to 100 µL. The content of total flavonoids was expressed as mg CA g^{-1} DW.

3.3.3.6 Total sugars

Samples of 5 g of fresh tissue of the basal region of stem, peduncle, basal and apical bract were used to quantify soluble total sugars. Samples were frozen in liquid nitrogen and stored at –20 ºC until evaluation. Subsequently, the samples were ground and boiled in ethanol (80 ºC) for 5 minutes, mixed and filtered to obtain the alcoholic extract.

For sugar determination anthrone method was used in 0.5 mL of alcoholic extract. The absorbance was registered at 620 nm. The sugar concentration was estimated using a standard curve of glucose (0 to 100 μg mL^{-1}) in 1 g of FW.

3.3.4 Chitosan experiment

The stems were randomly distributed into three treatments. For each treatment four evaluation times were made (day 1, 5, 10 and final day). The treatments assessed were chitosan concentration of 1 and 1.5 %, and a control sprayed with distiller water. To prepare 2 L of 1 and 1.5 % chitosan solution, 20 or 30 g of chitosan (low molecular weight, Sigma-Aldrich) were added in 1.8 L of distiller water plus 200 mL of glacial acetic acid to dissolve the chitosan. The pH of the solution was adjusted to 5. Stems were covered with the solution using a natural bristle brush and allowed to dry after coating for one hour, then the flower stems were placed into a bucket containing 800 mL distilled water and stored at room temperature (25 ºC ± 2 and 32 ± 5 % RH).

The variables responses evaluated on the storage experiment were the same in this experiment, and were assessed in the same way that the experiment previously aforementioned.

3.3.5 Experimental design and statistical analysis

In both experiments an unbalanced repeated measures design (RDM) was used to analyse the effects of storage and chitosan treatments on fresh weight loss and changes in bract colour for four evaluations (day 1, 5, 10 and final). A comparison of

Tukey test (α=0.05) was established beforehand among treatments when the interaction of treatments with time was significant.

A completely random design (CRD) with and analysis of variance and Tukey test (α=0.05) was used to analyse anthocyanin, total flavonoids, total sugars, vase life, and total postharvest longevity. The software SAS®, version 9.3 was used for the statistical analysis.

3.4 Results

3.4.1 Vase life

Control treatment showed the shorter vase life (2.1 d). In contrast, 1.0 % chitosan treatment had the longest vase life, reaching more than six times longer than the control; 1.5 % chitosan treatment showed more than five times longer than the control, and cold stored treatments at 16 ºC and 12 ºC had more than three and three times longer vase life than the control Table 3.1, Figure 3.1). These results represent the actual time that floral stems can maintain post-harvest quality in response to treatment (Table 3.1).

Table 3.1. Vase life (VL) and total postharvest longevity (TPL) in flower stems of *Heliconia psittacorum* L. f. cv. Trópica after 10 days of storage. VL values are the

means of six replicates. Different letters on one side of the variables indicate statistical differences Tukey test α = 0.05.

Treatment	VL (days)		TPL (days)	
Control	2.1	e	12.1	e
1.0 % chitosan	13.1	a	23.1	a
1.5 % chitosan	11.3	b	21.3	b
12 ºC	5.8	d	15.8	d
16 ºC	7.3	c	17.3	c

Figure 3.1. Flowers stem of *Heliconia psittacorum* L. f. cv. Trópica according to time and treatment. A: Initial state of inflorescences (day 1), B: 1.0 % chitosan treatment at final vase life (23.1 d), C: 1.5 % chitosan treatment at final vase life (21.3 d), D: storage treatment at 12 ºC (15.8 d), E: storage treatment at 16 ºC (17.3 d) and F: control treatment at final day vase life (12.1 d).

3.4.2 Fresh weight loss

The interaction between treatments and time was significant. All treatments (except 1.0 and 1.5 % chitosan treatments) showed a significant weight loss from the start

of the experiment to day 5 (Figure 3.2). During this period, treatments with chitosan had minor weight loss, treatments under cold storage had intermediate weight loss and in the control treatment, which showed the greatest loss of weight, this represented 11 % of the original weight. After 10 days, chitosan and storage treatments showed a similar loss weight. At the end of the experiment the 1.5 % chitosan and 16 ºC treatments presented the lowest percentage of weight loss, followed by 1.0 % chitosan and 12 ºC treatments, and the control had the greatest weight loss throughout the experiment.

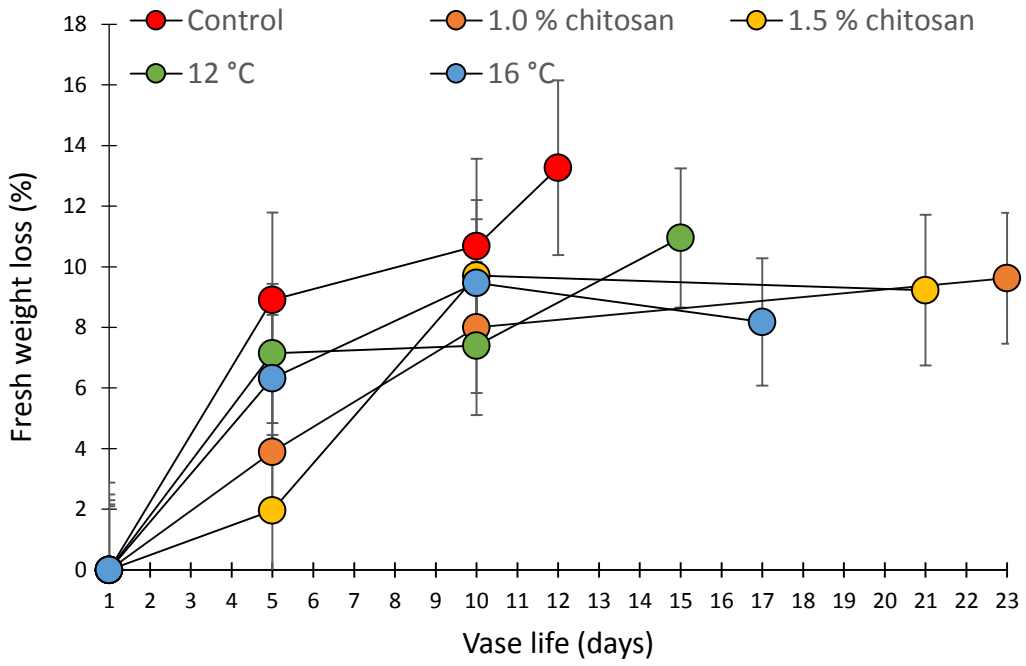


Figure 3.2. Percentage of fresh weight loss of stem flowers of *Heliconia psittacorum* L. f. cv. Trópica during the vase life period. Each value represents the mean of six observations ± SE.

3.4.3 Changes in bract colour (Luminosity, Chroma y Hue)

3.4.3.1 Luminosity

1.0 % chitosan treatment remained unchanged from day 1 until the end of vase life. The remaining treatments had a significant loss from the beginning of the experiment, being the control that had the greatest decrease (Figure 3.3-A). At day 10, apical bracts of all treatments showed a decrease on the levels of luminosity, the exception was 1.0 % chitosan treatment, which showed an increase from day 10 until the end of the study. At the end of the study the cold stored treatments and 1.0 % chitosan treatment had the highest luminosity (Figure 3.3-B).

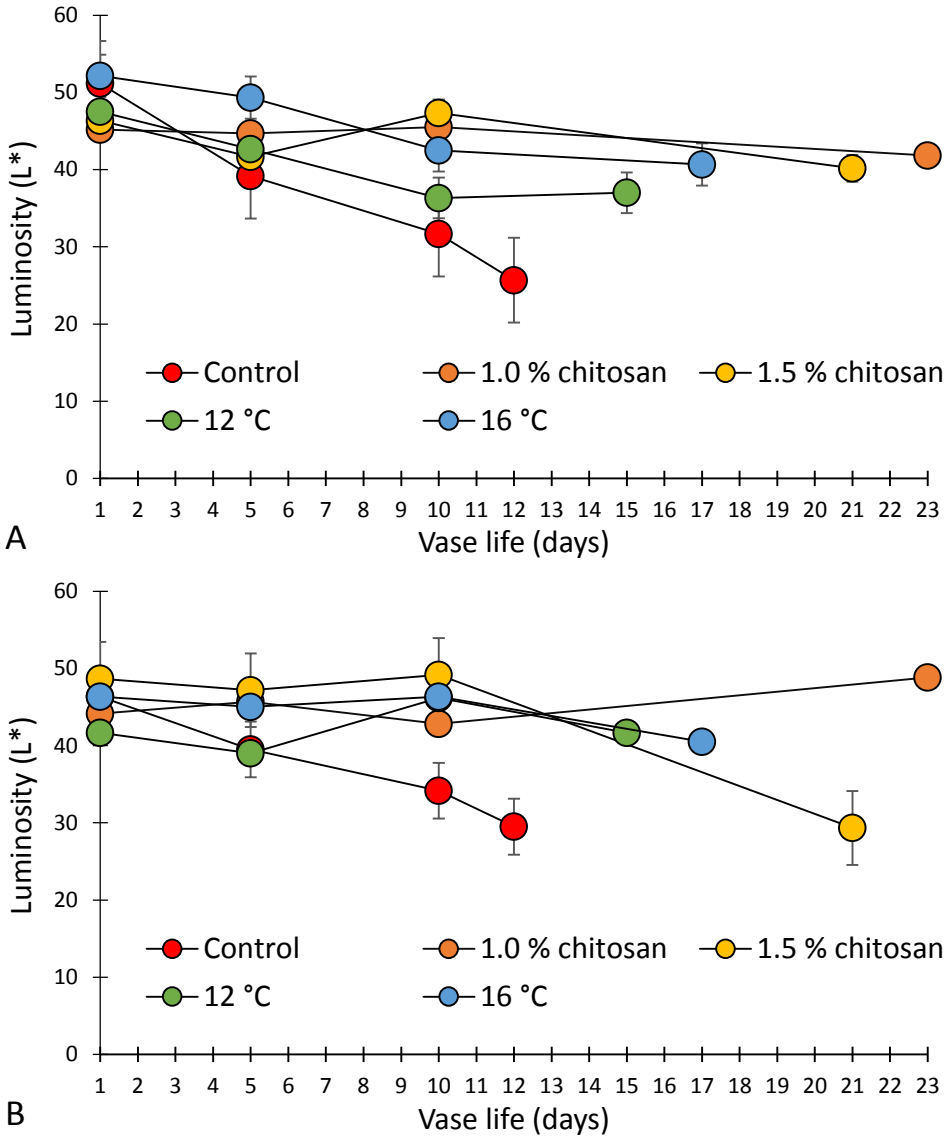


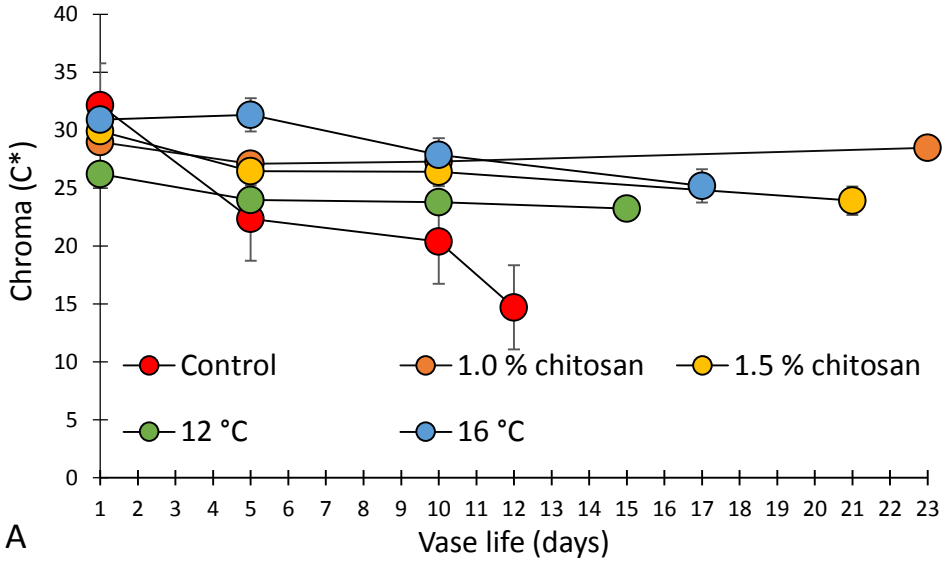
Figure 3.3. Luminosity, values are the average of six observations ± SE of flower stems of *Heliconia psittacorum* L. f. cv. Trópica during the vase life period (n = 6). A: basal bracts and B: apical bracts.

3.4.3.2 Chroma (C)*

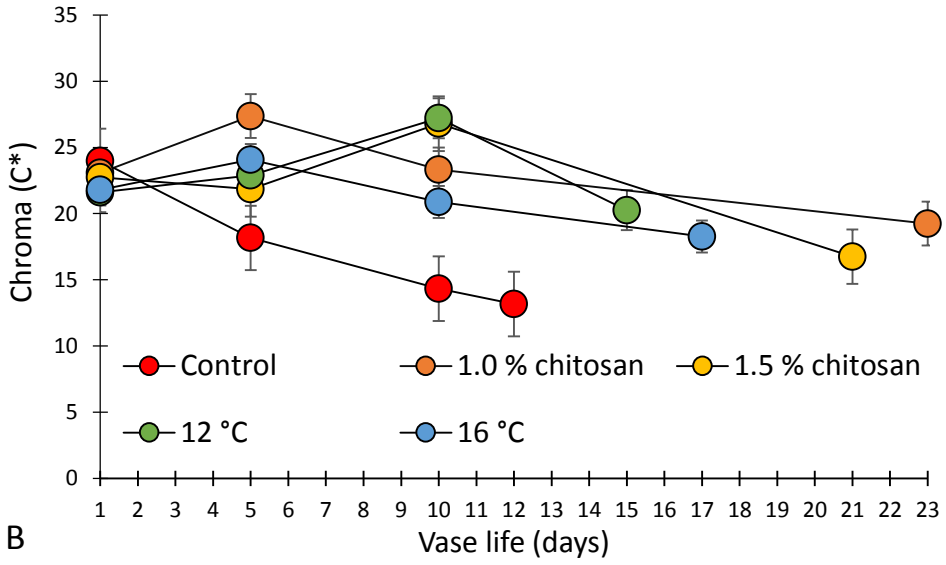
Basal and apical bracts had a significant interaction between treatments and time on chroma. On basal bracts cold stored treatments and 1.0 % chitosan treatment did

not change chroma values. At the end of vase life the 1.0 % chitosan and 16 ºC treatments showed the highest chroma values, 1.5 % chitosan and 12 ºC treatments had intermediate values, and the control showed the lowest chroma, which represented a decrease of 55 % of the initial chroma (Figure 3.4-A).

At the end of the study on apical bracts there were no significant differences on chroma among treatments (Figure 3.4-B).



A



B

Figure 3.4. Chroma, values are the average of six observations ± SE of stem flowers of *Heliconia psittacorum* L. f. cv. Trópica during the vase life period (n = 6). A: basal bracts and B: apical bracts.

3.4.3.3 Hue (hº)

The interaction between treatments and time was significant on basal and apical bracts for hue. On basal bracts 1.0 % chitosan treatment showed the highest values of hue throughout the experiment (except on day 10), followed by 1.5 % chitosan and 16 ºC treatments, which showed constant values during the evaluation days (Figure 3.5-A).

Apical bracts of 1.5 % chitosan treatment had a constant increase from the beginning to the final of the study. Chitosan treatments and the control showed the highest values in hue, in comparison with the cold storage treatments (Figure 3.5-B).

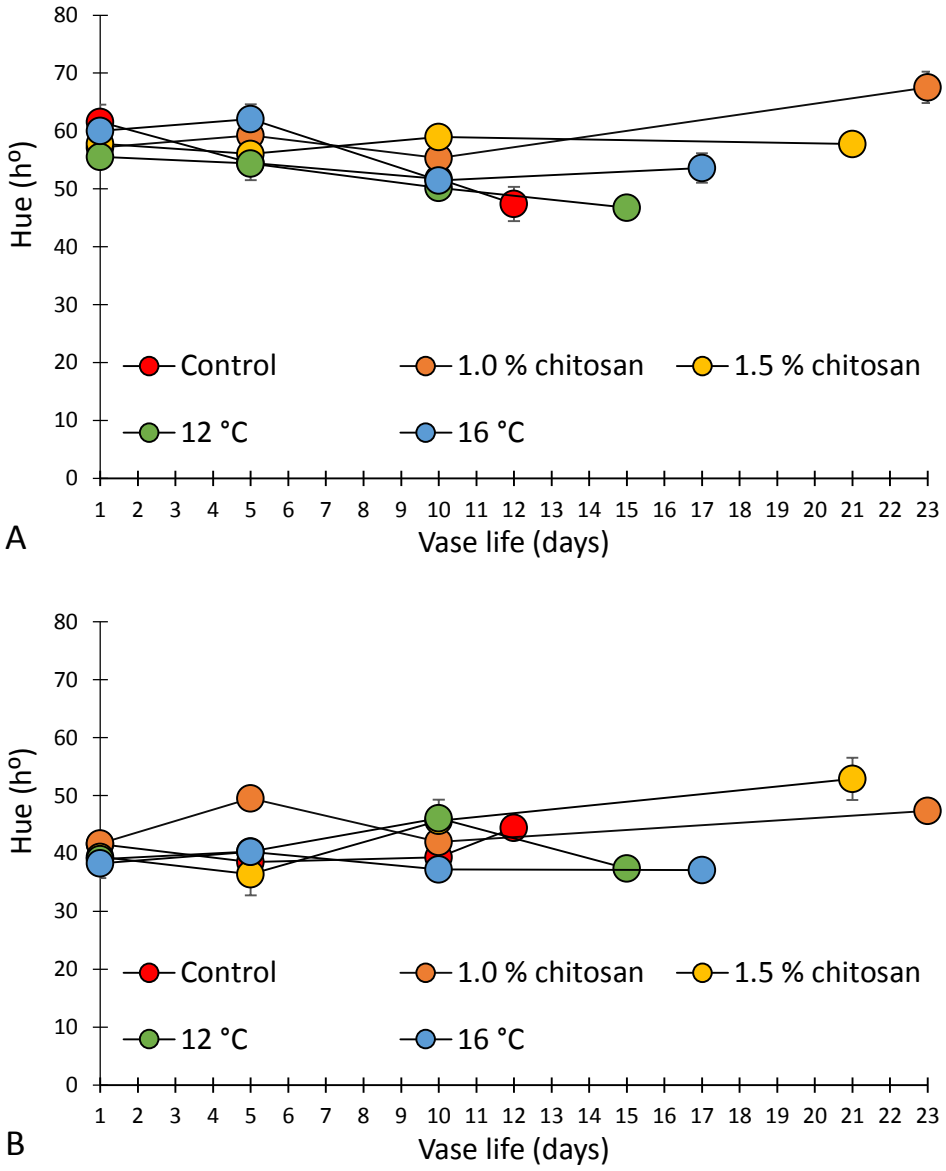


Figure 3.5. Hue, values are the average of six observations ± SE of stem flowers of *Heliconia psittacorum* L. f. cv. Trópica during the vase life period (n = 6). A: basal bracts and B: apical bracts.

3.4.4 Anthocyanin

No significant differences were found in the concentration of anthocyanins for the evaluated treatments at the end of vase life. (Figure 3.6).

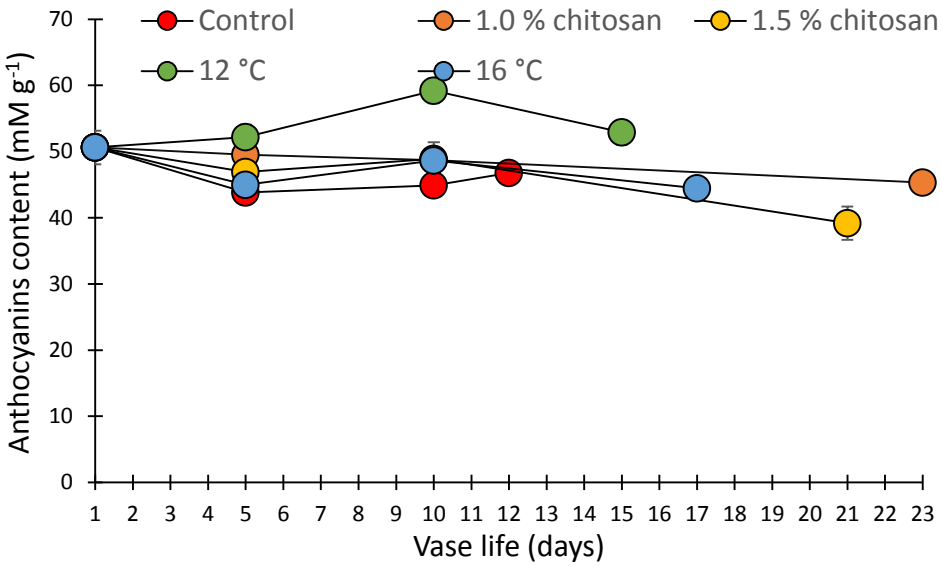


Figure 3.6. Concentration of anthocyanins, values are the average of six observations ± SE on stem flowers of *Heliconia psittacorum* L. f. cv. Trópica during the vase life period (n =6). A: basal bracts and B: apical bracts.

3.4.5 Total flavonoids

Statistical differences were found in the total flavonoid content among the treatments at the end of vase life. 12 °C treatment had the higher amount of total flavonoids at the end of vase life (Figure 3.7).

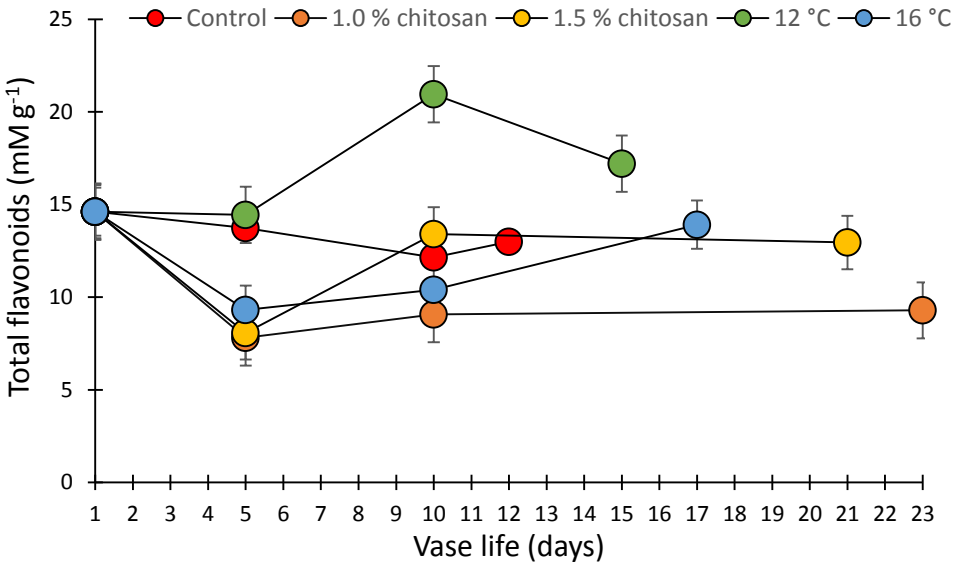


Figure 3.7. Concentration of total flavonoids, values are the average of six observations ± SE on stem flowers of *Heliconia psittacorum* L. f. cv. Trópica during the vase life period (n = 6). A: basal bracts and B: apical bracts.

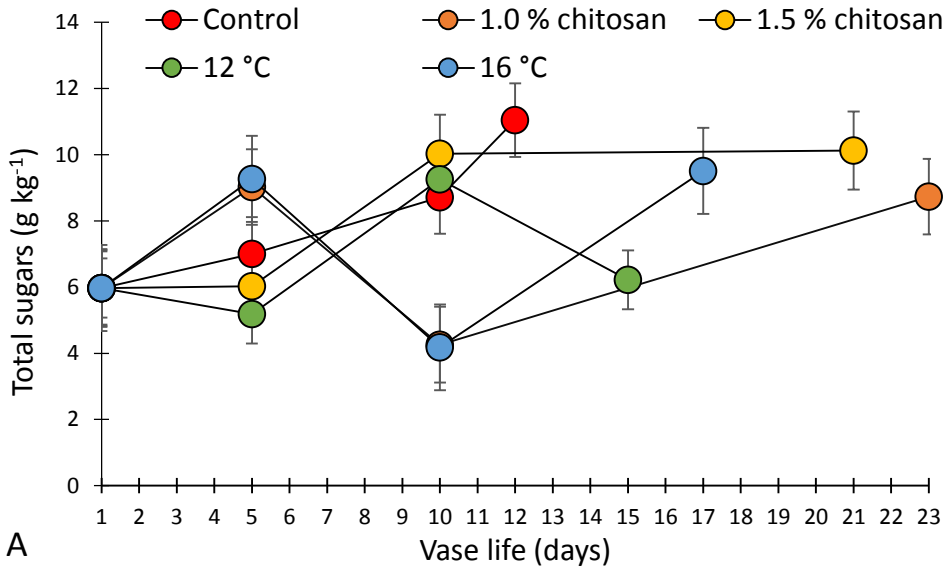
3.4.6 Total sugars

Significant differences were found on total sugars content in the stem base among treatments at the end of the vase life (Figure 3.8-A).

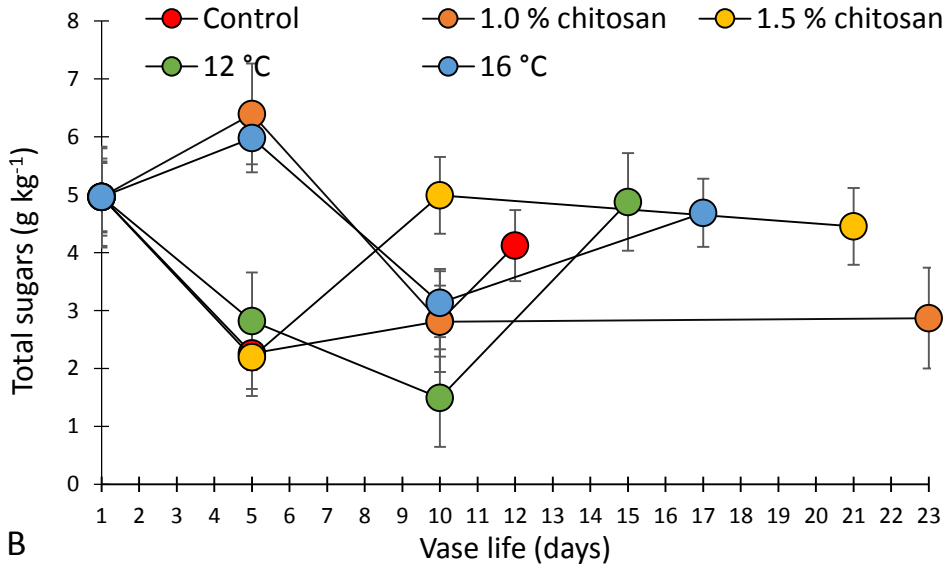
On peduncle and basal bracts of all treatments no significant differences were found in the total sugars content at the end of the experiment (Figure 3.8-B). On basal bracts, at the end of the vase life 1.0 % chitosan, both cold storage treatments and the control had the highest concentrations of sugars, compared to 1.5 % chitosan treatment (Figure 3.8-C).

On apical bracts 12 ºC treatment and the control had a decrease from the beginning of the experiment to day 5, with a subsequent increase until day 12 (control) and day 10 (12 ºC treatment). 1.5 % chitosan treatment presented constant

levels from day 1 to 10, however, as of this day had a significant decrease (Figure 3.8-D).



A



B

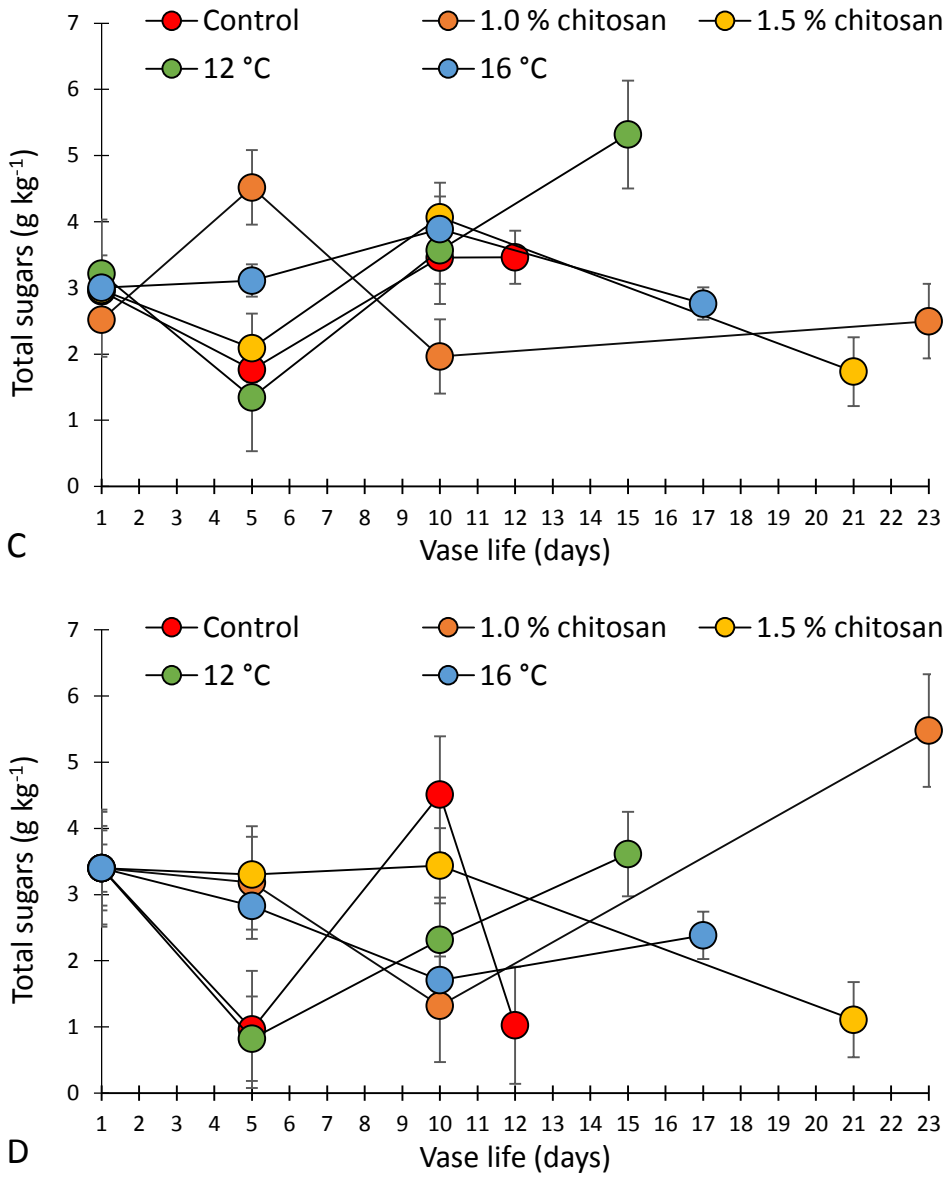


Figure 3.8. Total sugars, values are the average of six observations ± SE on stem flowers of *Heliconia psittacorum* L. f. cv. Trópica during the vase life period (n = 6). A: basal stem, B: peduncle, C: basal bracts and D: apical bracts.

3.5 Discussion

This study showed the chitosan effect on the vase life of the flower stems of *Heliconia psittacorum* L. f. cv. Trópica. Chitosan treatments had the longest vase life, cold storage treatments had intermediate vase life, presenting increases of 90.9, 76.0, 30.5, and 42.9 % in 1.0 % and 1.5 % chitosan treatments, and 12 and 16 ºC storage treatments, respectively, in comparison with the control treatment. This result can be explained by the formation of the coating, and certain characteristics of chitosan, such as molecular weight and pH of chitosan solution (Fei Liu et al., 2001). It has been demonstrated that chitosan coatings applied in fruits and vegetables extend the storage life because they inhibit the growth of microorganisms (Chien et al., 2007a). Liu et al. (2006) showed that chitosan at 200 ppm eliminated all bacteria, as *Escherichia coli*, possibly as effect of flocculation.

The 1.0 % chitosan treatment showed the greatest positive effects on the conservation of the physiological variables, such as vase life, total postharvest longevity, luminosity, chroma, hue and total sugars. Minor effect was obtained with the 1.5 % chitosan treatment, but it was also positive in the physiological variables. 1.0 % chitosan treatment equalled the results of cold storage treatment at 16 °C, except for vase life. This suggests that chitosan can increase the activities of some defence-related enzymes, such as phenylalanine ammonia-lyase, chitinase and β-1-3-glucanase (Zhang et al., 2011). Moreover, chitosan can activate pathogenesis-related gene functions, such as chitinases, chitosanase, β-glucanases, lignin, and callose (Kauss et al., 1989; Mauch et al., 1984; Benhamou and Thériault, 1992; Notsu et al., 1994).

Chitosan at 1.5 % delayed the loss of water expressed as fresh weight. The delay in water loss prevents premature deterioration by shrivelling (Burdon et al., 1994). Citrus fruits treated with chitosan (0.2 %, 0.05 % and 0.1 %) during post-harvest maintained firmness and water content (Chien et al., 2007a).

Fruit browning is the oxidation of phenolic compounds (Macheix and Fleuriet, 1990). This could be involved in breakdown of anthocyanins; it has been suggested that peroxidase may be involved in the oxidation of phenolics, glutathione and ascorbic acid, promoting colour changes in the products (Zhang and Quantick, 1997). Additionally, tissue browning in flowers may be due to cellular breakdown leading to mixing of browning-related enzymes and substrates, resulting in the enzymatic oxidation in the presence of oxygen (He and Luo, 2007).

The browning effect was evaluated by measuring L* and Hue angle. In this study, the decrease of the chroma values indicates a change to less vivid colours, and the hue angle the darkening of the bract tissue, which is characteristic of the oxidative browning reactions typical of senescence (Vargas et al., 2006). Chitosan coated stems evidencing the ability to reduce the brownish effect in comparison with the others treatments. The positive effect of the chitosan coating has been demonstrated in the prevention of browning of banana, litchi and breadfruit (No et al., 2007; Win et al., 2007). Studies by Zhang and Quantick (1997) showed the positive effect of chitosan coating on litchi fruits delaying changes of anthocyanins, flavonoids, total phenolics, and an increase of PPO activity.

The control treatment had the greatest loss in the parameters of colour. This can be explained because it was the treatment exposed to the highest

temperature as chitosan treatments were. The rate of enzyme-catalysed reaction is controlled to a great extent by temperature. The stored stems reduced their kinetic energy of the reactant molecules, resulting in a decrease in both mobility and collisions needed to establish enzyme-substrates complexes (He and Luo, 2007). However, in this study the protective effect of chitosan was evident, as both treatments with chitosan as well as the control were stored at the same room temperature, showing different results over vase life.

Coating semipermeable films can present a delay in senescence symptoms, as ripening, by the modification of the endogenous CO_2, O_2, and ethylene levels of fruits and flowers (El Ghaouth et al., 1991).

The trend toward decreasing anthocyanin content towards the end of the study may be due to the desiccation of the floral bracts, promoting the vacuoles breakdown, anthocyanins leakage and destroying the compartmentation of browning-related enzymes and their substrates (Underhill and Critchley, 1994).

The highest concentration of chitosan used in this experiment (1.5 %) seems to have a negative effect on the conservation of total sugar levels in comparison with the other treatments, which had higher concentrations of total sugars. Due to this conservation, some treatments showed a supply of respiratory substrates, maintain an adequate water balance, a delay in the increase in mRNA abundance of a number of senescence-associated genes (Hoeberichts et al., 2007; Ichimura et al., 2000).

Storage temperature for tropical cut flowers has a more or less broad range, since each species has an optimal temperature for storage. For heliconias temperatures higher than 10 °C are suggested to maintain good quality (Jaroenkit and Paull, 2003), however, Criley and Paull (1993) suggest not storing

less than 13 °C. In this study, the best results - in relation to the treatments under storage- in conservation of quality parameters and vase life were obtained at 16 °C, which coincides with the recommendations made by the last authors. The shortest vase life of stems stored at 12 °C can be explained because they were removed from refrigeration and placed at higher temperatures (Costa et al., 2011a).

The short vase life of the control compared to chitosan treatments (treatments exposed to the same temperature during the study) can be explained by the high respiratory rate of the stems. The respiration can be considered as a metabolic process for the oxidative breakage of organic substrates into simple molecules such as CO_2 and H_2O with the production of energy.

3.6 Conclusions

The 1.0 % chitosan treatment had a positive effect on the total postharvest longevity and vase life compared to all other treatments. Considering that both treatments were stored under similar conditions. In turn, the concentrations of chitosan evaluated had different effects on the variables evaluated, suggesting that depending on the expected response, a specific concentration should be chosen. For *H. psittacorum* to use storage temperatures close to 16 °C is recommended, avoiding the use of lower temperatures. 1.0 % chitosan treatment and 16 °C storage treatment had similar responses to maintain physiological characteristics, being able to use any of these treatments to assure a vase life that exceeds 15 days. The results found in this study mark the precedents for the realization of new investigations that focus the efforts in the understanding of the mechanisms of action through which the chitosan allows to extend the vase life

in the tropical ornamental ones, representing an economical and eco-friendly alternative with promising results.

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3.8 References

Akkerman, R., Farahani, P., Grunow, M., 2010. Quality, safety and sustainability in food distribution: a review of quantitative operations management approaches and challenges. Spectr. 32, 863–904. doi:10.1007/s00291-010-0223-2

Aular, J., Ruggerio, C., Durigan, 2002. Relación entre el color de la cáscara y las características del fruto y jugo de la parchita maracuyá. Bioagro 14, 47–51.

Barrón-Yánez, R.M., García-Mateos, M. del R., Soto-Hernández, M.R., Colinas-León, T., Kite, G., 2011. Flavonoides y actividad antioxidante de Calia secundiflora (Ort.) Yakovlev. Rev. Fitotec. Mex. 34, 151–157.

Bautista-Baños, S., Hernández-Lauzardo, A.N., Velázquez-del Valle, M.G., Hernández-López, M., Ait Barka, E., Bosquez-Molina, E., Wilson, C.L., 2006. Chitosan as a potential natural compound to control pre and postharvest diseases of horticultural commodities. Crop Prot. 25, 108–118. doi:10.1016/j.cropro.2005.03.010

Benhamou, N., Thériault, G., 1992. Treatment with chitosan enhances resistance of tomato plants to the crown and root rot pathogen Fusarium oxysporum f. sp. radicis-lycopersici. Physiol. Mol. Plant Pathol. 41, 33–52. doi:10.1016/0885-5765(92)90047-Y

Burdon, J.N., Dori, S., Lomaniec, E., Marinansky, R., Pesis, E., 1994. The post-harvest ripening of water stressed banana fruits. J. Hortic. Sci. 69, 799–804. doi:10.1080/14620316.1994.11516515

Chien, P., Sheu, F., Lin, H.-R., 2007a. Coating citrus (Murcott tangor) fruit with low molecular weight chitosan increases postharvest quality and shelf life. Food Chem. 100, 1160–1164. doi:10.1016/j.foodchem.2005.10.068

Chien, P., Sheu, F., Yang, F.-H., 2007b. Effects of edible chitosan coating on quality and shelf life of sliced mango fruit. J. Food Eng. 78, 225–229. doi:10.1016/j.jfoodeng.2005.09.022

Costa, A.S., Nogueira, L.C., dos Santos, V.F., Finger, F.L., Camara, T.R., Loges, V., Willadino, L., 2011a. Characterization of symptoms of senescence and chilling injury on inflorescences of Heliconia bihai (L.) cv. Lobster Claw and cv. Halloween. Postharvest Biol. Technol. 59, 103–109. doi:10.1016/j.postharvbio.2010.08.015

Costa, A.S., Nogueira, L.C., Santos, V.F. dos, Camara, T.R., Loges, V., Willadino, L., 2011b. Storage of cut Heliconia bihai (L.) cv. Lobster Claw flowers at low temperatures. Rev. Bras. Eng. Agríc. E Ambient. 15, 966–972.

Criley, R.A., Paull, R.E., 1993. Review: Postharvest handling of bold tropical cut flowers- Anthurium, Alpinia purpurata, Heliconia, and Strelitzia. Acta Hortic., New Floricultural Crops 337, 201–212.

de Castro, C.E.F., May, A., Gonçalves, C., 2007. Espécies de helicônia como flores de corte. Ornam. Hortic. 12, 87–96.

El Ghaouth, A., Arul, J., Ponnampalam, R., Boulet, M., 1991. Chitosan coating effect on storability and quality of fresh strawberries. J. Food Sci. 56, 1618–1620. doi:10.1111/j.1365-2621.1991.tb08655.x

Fei Liu, X., Lin Guan, Y., Zhi Yang, D., Li, Z., De Yao, K., 2001. Antibacterial action of chitosan and carboxymethylated chitosan. J. Appl. Polym. Sci.

79, 1324–1335. doi:10.1002/1097-4628(20010214)79:7<1324::AID-APP210>3.0.CO;2-L

Han, C., Zuo, J., Wang, Q., Xu, L., Zhai, B., Wang, Z., Dong, H., Gao, L., 2014. Effects of chitosan coating on postharvest quality and shelf life of sponge gourd (Luffa cylindrica) during storage. Sci. Hortic. 166, 1–8. doi:10.1016/j.scienta.2013.09.007

He, Q., Luo, Y., 2007. Enzymatic browning and its control in fresh-cut produce. Stewart Postharvest Rev. 3, 1–7.

Hoeberichts, F.A., van Doorn, W.G., Vorst, O., Hall, R.D., van Wordragen, M.F., 2007. Sucrose prevents up-regulation of senescence-associated genes in carnation petals. J. Exp. Bot. 58, 2873–2885. doi:10.1093/jxb/erm076

Hong, K., Xie, J., Zhang, L., Sun, D., Gong, D., 2012. Effects of chitosan coating on postharvest life and quality of guava (Psidium guajava L.) fruit during cold storage. Sci. Hortic. 144, 172–178. doi:10.1016/j.scienta.2012.07.002

Ichimura, K., Kohata, K., Goto, R., 2000. Soluble carbohydrates in Delphinium and their influence on sepal abscission in cut flowers. Physiol. Plant. 108, 307–313. doi:10.1034/j.1399-3054.2000.108003307.x

Jaroenkit, T., Paull, R.E., 2003. Postharvest handling of Heliconia, red Ginger, and Bird-of-Paradise. HortTechnology 13, 259–266.

Kauss, H., Jeblick, W., Domard, A., 1989. The degrees of polymerization and N-acetylation of chitosan determine its ability to elicit callose formation in suspension cells and protoplasts of Catharanthus roseus. Planta 178, 385–392. doi:10.1007/BF00391866

Kerch, G., 2015. Chitosan films and coatings prevent losses of fresh fruit nutritional quality: A review. Trends Food Sci. Technol. 46, 159–166. doi:10.1016/j.tifs.2015.10.010

Liu, N., Chen, X.-G., Park, H.-J., Liu, C.-G., Liu, C.-S., Meng, X.-H., Yu, L.-J., 2006. Effect of MW and concentration of chitosan on antibacterial activity of Escherichia coli. Carbohydr. Polym. 64, 60–65. doi:10.1016/j.carbpol.2005.10.028

Liu, Y., Wisniewski, M., Kennedy, J.F., Jiang, Y., Tang, J., Liu, J., 2016. Chitosan and oligochitosan enhance ginger (Zingiber officinale Roscoe) resistance to rhizome rot caused by Fusarium oxysporum in storage. Carbohydr. Polym. 151, 474–479. doi:10.1016/j.carbpol.2016.05.103

Macheix, J.-J., Fleuriet, A., 1990. Fruit Phenolics. CRC Press.

Mauch, F., Hadwiger, L.A., Boller, T., 1984. Ethylene: Symptom, Not Signal for the Induction of Chitinase and beta-1,3-Glucanase in Pea Pods by Pathogens and Elicitors. Plant Physiol. 76, 607–611.

No, H.K., Meyers, S.P., Prinyawiwatkul, W., Xu, Z., 2007. Applications of Chitosan for Improvement of Quality and Shelf Life of Foods: A Review. J. Food Sci. 72, 87–100. doi:10.1111/j.1750-3841.2007.00383.x

Notsu, S., Saito, N., Kosaki, H., Inui, H., Hirano, S., 1994. Stimulation of phenylalanine ammonia-lyase activity and lignification in rice callus treated with chitin, chitosan, and their derivatives. Biosci. Biotechnol. Biochem. 58, 552–553. doi:10.1271/bbb.58.552

Rahmani, I., Ahmadi, N., Ghanati, F., Sadeghi, M., 2015. Effects of salicylic acid applied pre- or post-transport on post-harvest characteristics and

antioxidant enzyme activity of gladiolus cut flower spikes. N. Z. J. Crop Hortic. Sci. 43, 294–305. doi:10.1080/01140671.2015.1096799

Salinas, Y., Martínez, F., Soto, M., Ortega, R., Arellano, J.L., 2003. Efecto de la nixtamalización sobre las antocianinas del grano de maíces pigmentados. Agrociencia 37, 617–628.

Sosa Rodríguez, F.M., 2013. Cultivo del género Heliconia. Cultiv. Trop. 34, 24–32.

Underhill, S., Critchley, C., 1994. Anthocyanin decolorisation and its role in lychee pericarp browning. Aust. J. Exp. Agric. 34, 115–122. doi:10.1071/ea9940115

Vargas, M., Albors, A., Chiralt, A., González-Martínez, C., 2006. Quality of cold-stored strawberries as affected by chitosan–oleic acid edible coatings. Postharvest Biol. Technol. 41, 164–171. doi:10.1016/j.postharvbio.2006.03.016

Win, N.K.K., Jitareerat, P., Kanlayanarat, S., Sangchote, S., 2007. Effects of cinnamon extract, chitosan coating, hot water treatment and their combinations on crown rot disease and quality of banana fruit. Postharvest Biol. Technol. 45, 333–340. doi:10.1016/j.postharvbio.2007.01.020

Zhang, D., Quantick, P.C., 1997. Effects of chitosan coating on enzymatic browning and decay during postharvest storage of litchi (Litchi chinensis Sonn.) fruit. Postharvest Biol. Technol. 12, 195–202.

Zhang, H., Li, R., Liu, W., 2011. Effects of Chitin and Its Derivative Chitosan on Postharvest Decay of Fruits: A Review. Int. J. Mol. Sci. 12, 917–934. doi:10.3390/ijms12020917

4. COMPARISON OF PRACTICAL METHODS FOR POSTHARVEST PRESERVATION OF STEMS OF *Heliconia bihai* (L.) L. cv. Lobster Claw

COMPARISON OF PRACTICAL METHODS FOR POSTHARVEST PRESERVATION OF STEMS OF *Heliconia bihai* (L.). L. cv. Lobster Claw

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Keywords: chitosan coating, longevity, quality, storage, tropical ornamental, vase life

4.1 Abstract

Two post-harvest methods were compared to evaluate vase life of *Heliconia bihai* (L.) flower stems. Typical storage methods at 12 ºC and 16 ºC and chitosan coating treatments at 1.0 and 1.5 % at laboratory temperature (26 ºC ± 4 and 40 ± 5 % RH) were investigated. The effects of both methods on vase life (VL), total postharvest longevity (TPL), weight loss, colour changes (L*, C*, hº), anthocyanins, flavonoids, and total sugar concentration were evaluated. Treatments of 1.0 % chitosan and 16 ºC storage treatment had the best results on vase life. 1.0 % chitosan treatment had a TPL of 16.8 days (6.8 days of VL),

whereas at 16 ºC stored treatment had TPL of 15.3 days (5.3 days of VL), representing 4.7 and 3.2 days more than the control.

4.2 Introduction

It has been estimated that postharvest losses of horticultural products can reach between 20 % and 40 % of total production (Kasso and Bekele, 2016). They are mostly caused by attack of microorganisms, natural senescence, extreme temperatures, drought and improper postharvest handling, resulting in a quality deterioration, and a subsequent decrease in product shelf life (Olayemi et al., 2010). Pre and postharvest alternatives such as storage treatments, applications of wax, pulse solutions, coatings, among others, have been created to mitigate postharvest problems.

Temperature has been found to influence different biological process as respiration rate, water loss, and physical damage in cut flowers (Celikel and Reid, 2002). In general, low temperature storage is used to reduce the metabolic process that involves the senescence and maintain the quality (Matsumoto and Ikoma, 2016). Tropical ornamental plants exhibit high sensitivity at low temperatures, developing chilling injuries (Costa et al., 2011a, 2011b).

Each species and variety has an optimum temperature range for postharvest storage. Costa et al. (2011b) reported chilling injury in stems of *H. bihai* stored at 12 ºC, early senescence at 25 ºC, and an adequate vase life at 19 ºC (6.2 days), while stems of *H. psittacorum* cv. Tropica stored at 15 ºC had 6 days of vase life (Bañuelos-Hernández et al., 2016). In contrast, Broschat and Donselman (1983) recommended storage temperatures close to 23 ºC for the colour preservation of floral bracts for the cultivars Golden Torch, Andromeda, and St. Vincent Red.

Ribeiro et al. (2010) stored inflorescences of *H. marginata* at 25 ºC, and had a vase life of 8 days.

Anon (1991) recommended for the storage of Heliconias not less than 13 ºC, and should not be placed in cold storage. Others recommendations are to store tropical cut flowers are as follow > 10 ºC for heliconias, > 12 ºC for red ginger, and > 8 ºC for bird of paradise, coupled with a relative humidity higher than 90 % (Broschat and Donselman, 1983, Broschat and Donselman, 1988; Halevy et al., 1978; Jaroenkit and Paull, 2003).

Protective coatings and packaging has shown potential for extending postharvest life of fruit and vegetables. An alternative is to use organic coatings due to their environmental friendliness and effectiveness (Durango et al., 2011).

Actions that minimize postharvest losses are essential to promote the growth of the ornamental market. Therefore, this study was carried out to evaluate the vase life of chitosan coated stems of *Heliconia bihai* (L.) L. cv. Lobster Claw, under laboratory conditions (26 ºC ± 4 and 40 ± 5 % RH), as postharvest treatment and to compare it with refrigerated storage stems.

4.3 Materials and methods

4.3.1 Plant material

Flower stems of *H. bihai* cv. Lobster claw were obtained from a commercial plantation at Campo Grande, Veracruz, Mexico (18º49´30.0´´ N, 97º00´54.5´´ W and at 780 m. a. s. l.). Harvests were made between June and July of 2015. The stems were 0.8 m in length, did not have leaves, and had one inflorescence with four to six open bracts. The flower stems were harvested at 8:00 am and the stem

was cut at 45 º inclination, as a usual farm practices. Besides, the whole stems were sprayed with water and 0.2 m of the stems were immersed in water for 4 h during transportation to the laboratory.

The stems were randomly distributed into five treatments; each, were sampled at day 1, 5, 10 and at the end day of vase life. Each sample included six replicates and one flower stem represent a repetition. The treatments assessed were 1.0 and 1.5 % chitosan and the control treatment sprayed with distilled water at 26 ºC ± 4 and 40 ± 5 % RH, and storage in controlled environment chamber at 12 ± 0.5 ºC and 16 ± 0.5 ºC during 10 days.

To prepare 2 L of 1.0 or 1.5 % chitosan solution, 20 or 30 g of chitosan (low molecular weight, Sigma-Aldrich) were added in 1.8 L of distilled water with 200 mL of glacial acetic acid. The pH of the solution was adjusted to 5. The flower stems were covered with the chitosan solution using a natural bristle brush and allowed to dry after coating for one hour, then the flower stems of each treatment were placed individually into a bucket containing 800 mL distilled water.

4.3.2 Variables evaluated:

4.3.2.1 Vase life

The quality of inflorescences was rated using a 1 – 4 visual scale, where 1 was the worst quality and 4 the best condition (Costa et al., 2011b). The end of vase life was when more than 50 % of flowering stems had reached grade 1. The total postharvest longevity was considered as the storage simulation period (10 days) plus post storage vase life.

4.3.2.2 Fresh weight loss

The fresh weight of flower stems were recorded with a digital balance (Precisa, XB 2200C®). The results were expressed as a percentage of fresh weight loss with respect to the initial weight.

4.3.2.3 Changes in colour (L, C*, hº)*

The colour of basal and apical bracts was measured using digital images (Samsung®, 12 Mp) and the app ColorPixLab (Colegio de Postgraduados) in MATLAB® Version 7.10.0. The CIE colour space L*C*hº (luminosity, chroma and hue angle, respectively) was used for this determinations. The digital images were taken at the same flower stems position, time of day (10 a.m.) and distance (0.3 m). Based on the reading taken by the CIEL*a*b* model, chroma (C*) and hue (hº) were calculated using the following equations (Aular et al., 2002):

$$C^* = (a^{*2} + b^{*2})^{1/2}$$

$$h° = Tan^{-1}\left(\frac{b*}{a*}\right)$$

Where C^* is chroma, hº is hue, a^* is red to green axis (128 to 127), b^* is yellow to blue axis (128 to 127), and y Tan^{-1} is the inverse tangent.

4.3.2.4 Anthocyanin

Anthocyanin in the basal bracts was evaluated according to Rahmani et al. (2015) and Salinas et al. (2003). 1 g of lyophilized ground sample was extracted in 5 mL of methanol with 1.0 % (v/v) HCl in water. The samples were centrifuged at

10,000 g and kept in the dark overnight. The absorbance was measured in a spectrophotometer (Genesys 10 UV, Thermo Electron Corporation®) at 520 nm in an aliquot of 4 mL of the filtered extract. Anthocyanin concentration was calculated using an extinction coefficient of 33,000 mol^{-1} cm^{-1}. The anthocyanin concentration was expressed as μM g^{-1}.

4.3.2.5 Total flavonoids

Flavonoid concentrations in the basal bracts were measured according to Rahmani et al. (2015) and Barrón-Yánez et al. (2011). 1 g of lyophilized bracts tissue was extracted in 3 mL ethanol containing 1.0 % acetic acid, centrifuged at 10,000 g, shaked and heated at 80 ºC in water bath for 10 min. The absorbance was measured at 415 nm. Sample concentration was obtained using a calibration curve of catechin (0.1 mg L^{-1}). It was made in 0 to 100 μL concentrations and 20 μL intervals. The concentration of total flavonoids was expressed as mg of catechin equivalent per kg of bract dry weight.

4.3.2.6 Total sugars

Samples of 1 g of fresh tissue of the base of stem, peduncle, and basal and apical bracts were frozen in liquid nitrogen and stored at -20 ºC until evaluation. Subsequently, the samples were ground and boiled in ethanol (80 ºC) for 5 min, mixed and filtered to obtain the alcoholic extract.

Total sugar content was calculated using the anthrone method on 0.5 mL of alcoholic extract. The absorbance was measured at 620 nm. The sugar concentration was estimated from a standard curve of glucose (0 to 100 μg mL^{-1}).

4.3.3 Experimental design and statistical analysis

An unbalanced repeated measures design (RDM) was used to analyse the effects of chitosan on fresh weight loss and changes in bract colour for four evaluations (1, 5, 10 and vase life). A Tukey test (α = 0.05) comparison was established beforehand among treatments (day 1 and final) when the interaction of treatments with time was significant. A completely random design with an analysis of variance and Tukey test (α=0.05) was used to analyse anthocyanin, total flavonoids, total sugars, and duration of vase life. SAS® software (version 9.3) was used for the statistical analysis.

4.4 Results

4.4.1 Vase life

The vase life was statistically different among the treatments (Table 1). 1.0 % chitosan treatment, and 16 ºC and 12 ºC storage treatments had the longest vase life, they were followed by 1.5 % chitosan and control treatments. 1.0 %chitosan treatment increased the vase life 4.7 days (3.2 times) more than the control, while storage at 16 ºC and 12 ºC increased the vases life 3.2 and 3 days (53 and 43 %, respectively) more than the control. There were differences between cold treatments and 1 % chitosan were no significant, nor was total postharvest longevity (Table 4.1).

Table 4.1. Vase life (VL) and total postharvest longevity (TPL) in flower stems of *Heliconia bihai* (L.) L. cv. Lobster Claw after 10 days of storage. VL values are

the means of six replicates. Different letters on one side of the variables indicate statistical differences Tukey test $\alpha = 0.05$.

Treatment	VL (days)		TPL (days)	
Control	2.1	c	12.1	c
1.0 % chitosan	6.8	a	16.8	a
1.5 % chitosan	4.5	b	14.5	b
12 ºC	5.1	a	15.1	a
16 ºC	5.3	a	15.3	a

4.4.2 Fresh weight loss

The interaction between treatments and time was significant in the fresh weight loss. After 5 days of evaluation a gradient of this variable was observed. Both cold storage treatments showed the lowest lost weight, they were followed by 1.0 % chitosan, 1.5 % chitosan treatments, and the control, with no statistical differences. Fresh weight loss continued increasing in cold storage treatments and there were no significant differences among treatments on day 10.

Although vase life differed between treatments, fresh weight loss did not show statistical differences between them (Figure 4.1). In general the control presented the greatest values and variation in the weight loss; but, treatment stored at 12 ºC presented greater changes among 1 to 15 days.

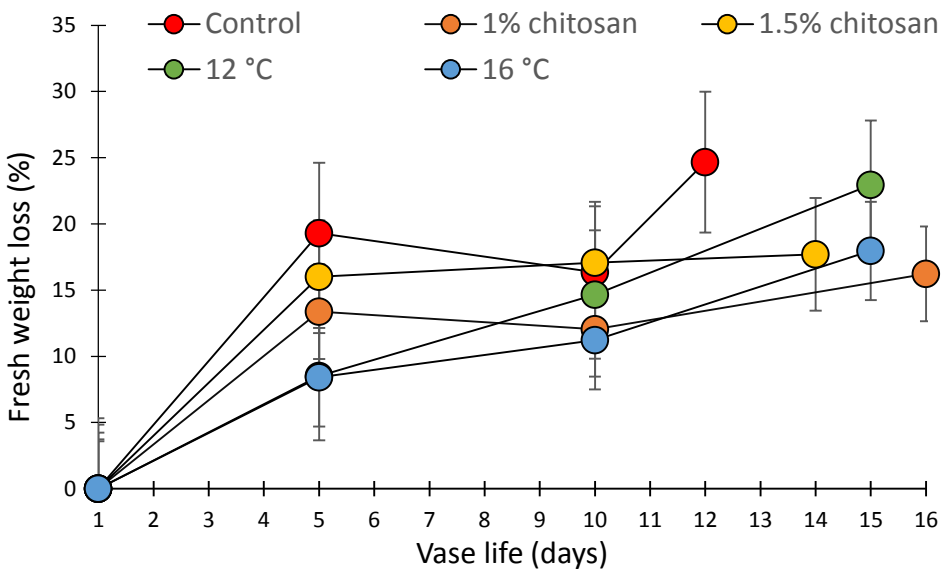


Figure 4.1. Fresh weight loss, values are the average of six observations ± SE of flower stems of *Heliconia bihai* (L.) L. cv. Lobster Claw during the vase life period (n = 6).

4.4.3 Changes in colour (L*, C*, hº)

4.4.3.1 Luminosity (L)*

The interaction treatment x time was significant for luminosity of basal and apical bract. There were no significant differences on bracts luminosity among treatments at the end of vase life, however on day 5 the treatment stored at 12 °C and the control presented the highest values of luminosity (Figure 4.2, Figure 4.3-A).

Apical bracts there were no significant differences among treatments on day 10 and final day of vase life (Figure 4.2, Figure 4.3-B). On day 5, the 12 ºC stored treatment maintained the highest values, in contrast to 1.5 % chitosan treatment.

The luminosity showed to be higher in the apical bracts than in the basal ones, in turn, there was a greater fluctuation between the values in the basal bracts.

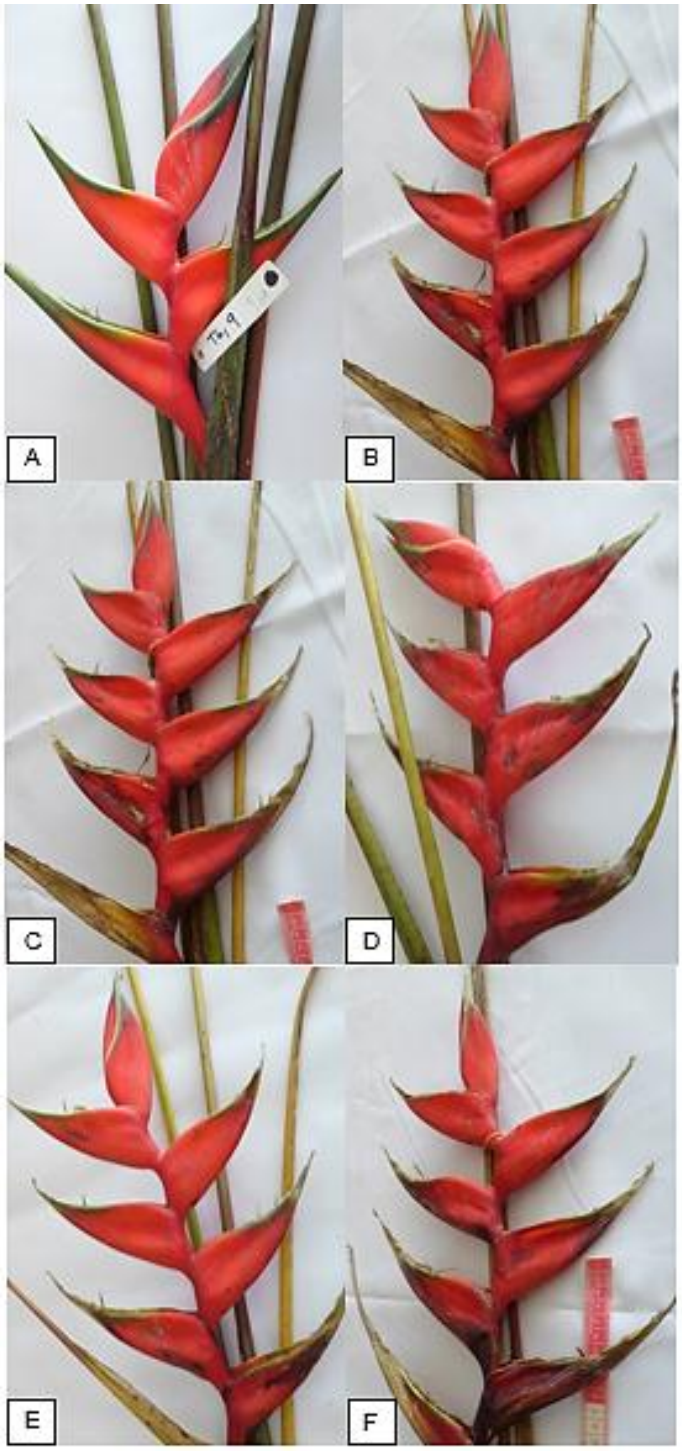


Figure 4.2 Flower stems of *Heliconia bihai* (L.) L. cv. Lobster Claw according to time and treatment. A: initial state of inflorescences (day 1), B: chitosan treatment 1.0 % at final vase life (16.8 d), C: chitosan treatment 1.5 % at final vase life (14.5 d), D: storage treatment at 12 ºC (15.1 d), E: storage treatment at 16 ºC (15.3 d) and F: control treatment at the end vase life (12.1 d).

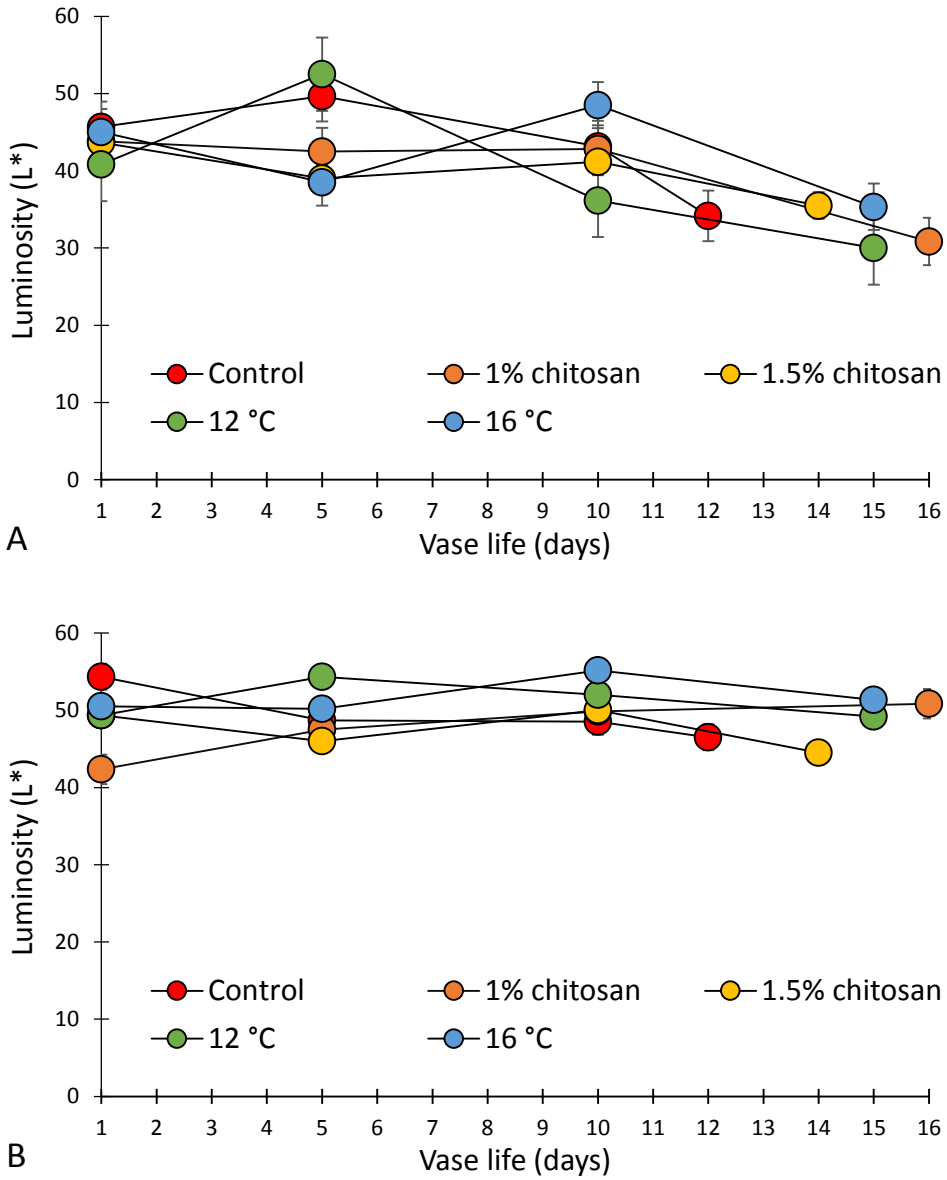


Figure 4.3. Luminosity, values are the average of six observations ± SE of flower stems of *Heliconia bihai* (L.) L. cv. Lobster Claw during the vase life period. A: basal bracts, B: apical bracts (n = 6).

4.4.3.2 Chroma (C)*

The interaction treatment x time was significant for basal and apical bract. On basal bracts there were differences on day 5. There were no significant differences among treatments in the later days (Figure 4.2, Figure 4.4-A). Only

treatment stored at 12 ºC had significant differences in each evaluation time (from day 5).

Similar results were observed on apical bracts, only significant differences on the chroma values were observed at day 5, at that moment 1.0 % chitosan treatment had the highest chroma value. In the following days the differences were no significant (Figure 4.2, Figure 4.4-B). The exception was the 1.0 % chitosan and storage at 16 ºC had differences on chroma on the days of evaluations. 1.0 % chitosan treatment had the highest value at day 5, while 16 ºC storage treatments presented the lowest chroma levels at the end of the vase life, in comparison with the other days.

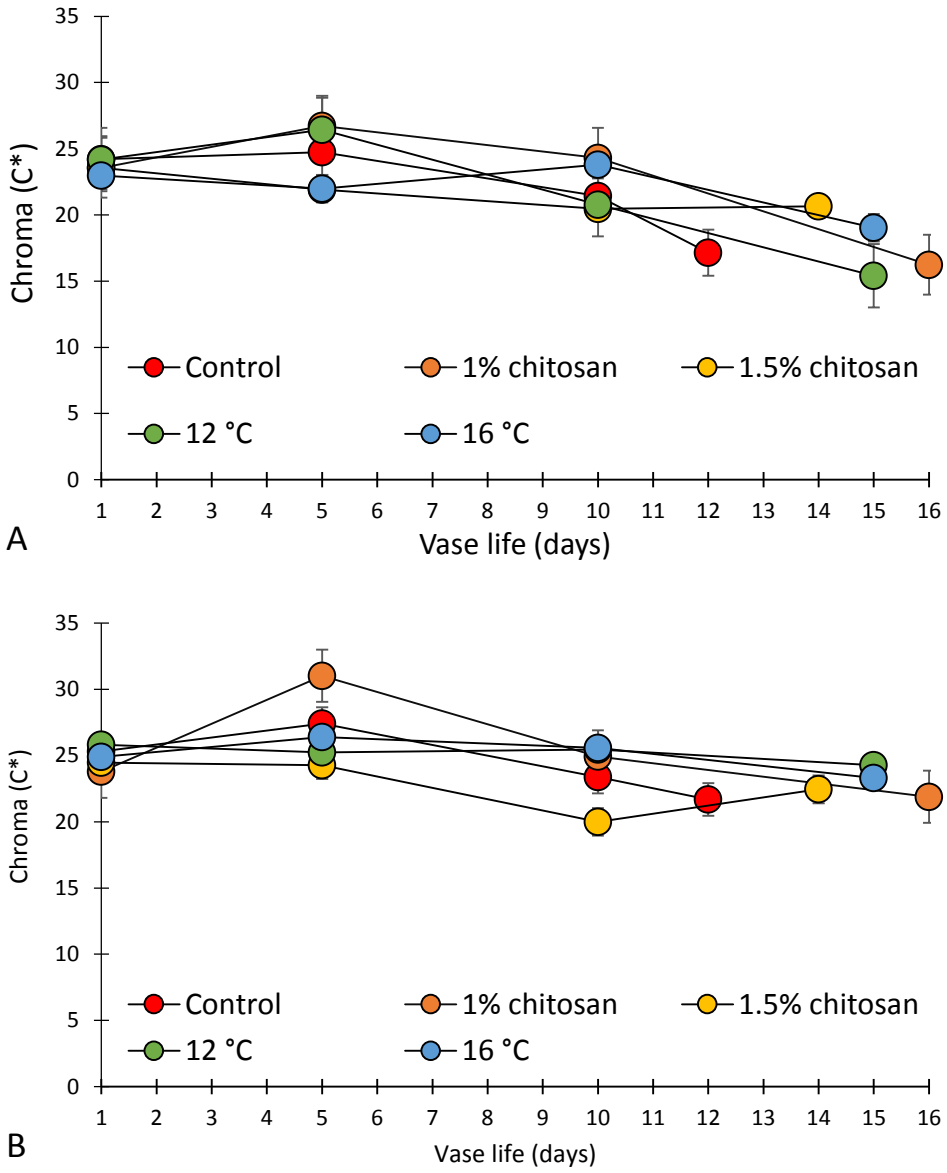


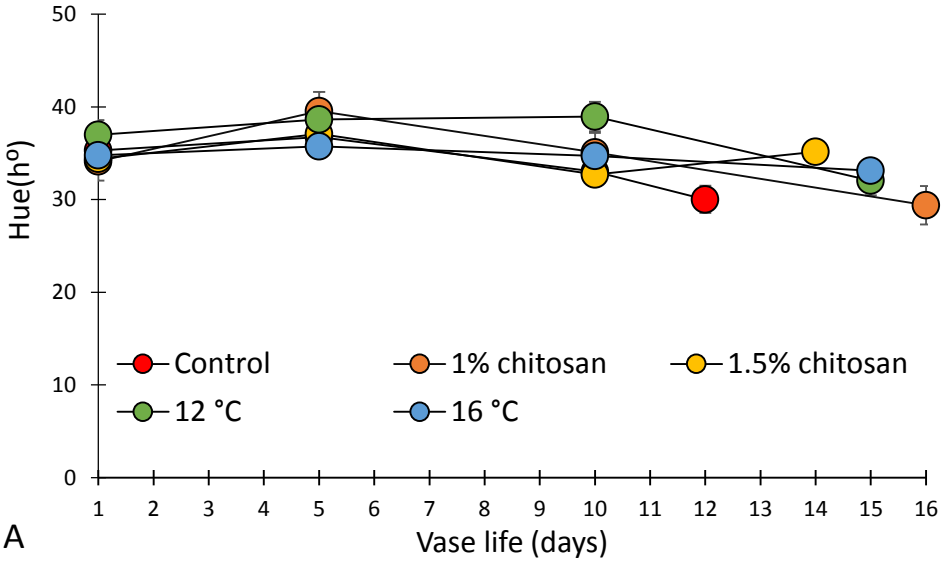
Figure 4.4. Chroma, values are the average of six observations ± SE of flower stems of *Heliconia bihai* (L.) L. cv. Lobster Claw during the vase life. A: basal bracts, B: apical bracts (n = 6).

4.4.3.3 Hue (hº)

On basal and apical bracts the interaction treatment x time was significant for hue. On basal bracts, only on day 8 significant differences were observed; when the treatment stored at 12 ºC had the higher hue levels, in comparison with the others treatments. Throughout the study, the 1.0 % chitosan treatment had the

greatest variations in this parameter, while the treatment stored at 12 ºC did not have changes during the experiment (Figure 4.2, Figure 4.5-A).

On apical bract differences on Hue were found on day 5 and 10. After 5 days of evaluation, 1.0 % chitosan treatment and the control had the higher levels of hue, followed by 1.5 % chitosan treatment, while on day 10, the 1.5 % chitosan treatment had the largest decrease in this parameter (Figure 4.2, Figure 4.5-B). 1.5 %, chitosan treatment, 16 ºC stored treatment and the control did not show significances differences among the days of evaluation; 1.0 % chitosan and 16 ºC stored treatments showed the largest increase on day 5, and day 10, respectively.



A

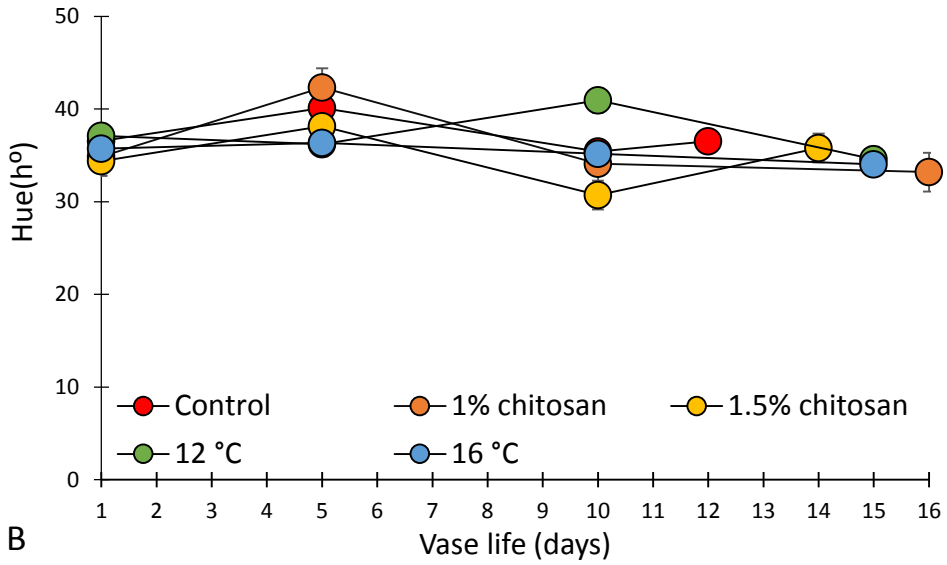


Figure 4.5. Hue, values are the average of six observations ± SE of flower stems of *Heliconia bihai* (L.) L. cv. Lobster Claw during the vase life period. A: basal bracts, B: apical bracts (n = 6).

4.4.4 Anthocyanins

The differences in the anthocyanins concentrations among treatments were significant on day 10 and the final day of vase life. On day 10, the storage treatments had the highest concentrations of anthocyanins, followed by the chitosan treatments, while on the end of vase life, the 16 ºC stored treatment preserved better the concentrations of anthocyanins, followed by chitosan treatments and the 12 ºC stored treatment. The control presented the greatest loss of these pigments (Figure 4.6). With exception of the stems treated with chitosan at 1.0 %, all treatments showed an increase in anthocyanins concentration as the experiment proceeded, being the end of vase life, the day that they presented the highest levels.

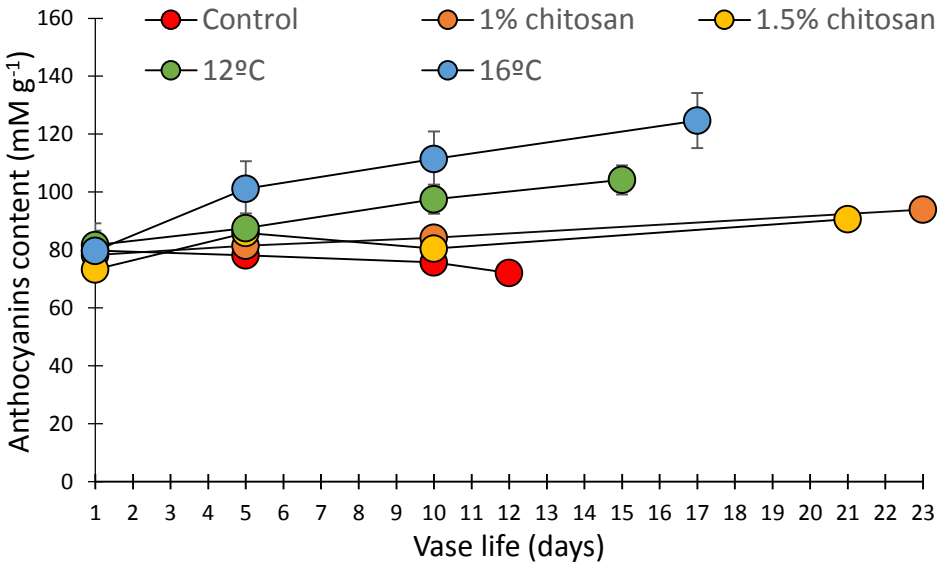


Figure 4.6. Concentration of anthocyanins, values are the average of six observations ± SE on flower stems of *Heliconia bihai* (L.) L. cv. Lobster Claw during the vase life period (n = 6).

4.4.5 Total flavonoids

The total flavonoids in the basal bract differed among the treatments. The wide variation was observed on day 5, when the control and treatment stored at 12 ºC had up to 1.6 and twice flavonoids content than the others treatments. Independent of treatment, the content of total flavonoids decreases, on average 32 %, at the end of the vase life respect of the start time evaluation, the exception was the 16 ºC stored treatment, which slightly increased its content. (Figure 4.7).

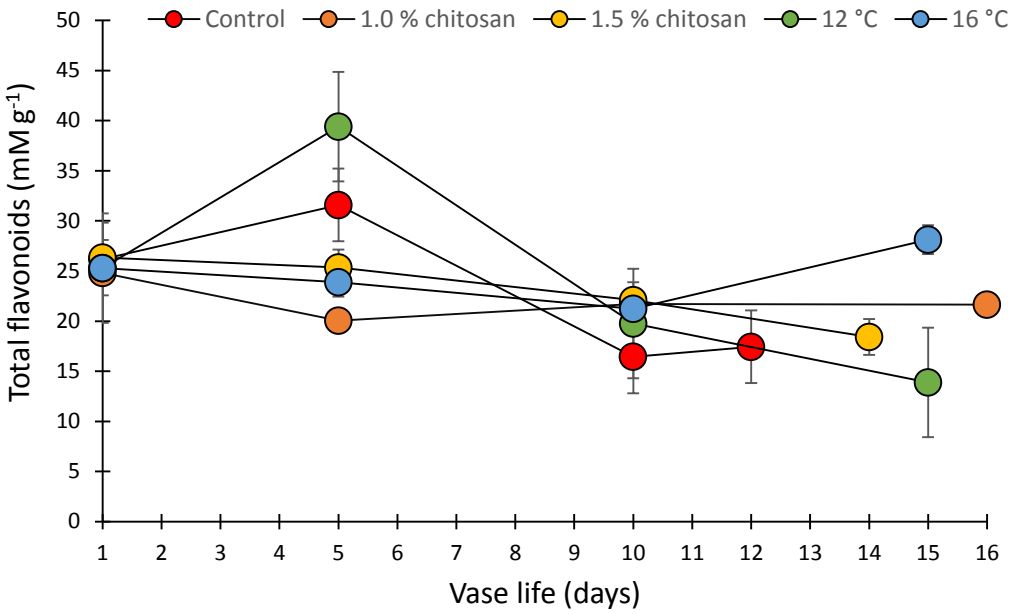


Figure 4.7. Concentration of total flavonoids, values are the average of six observations ± SE on flower stems of *Heliconia bihai* (L.) L. cv. Lobster Claw during the vase life period (n = 6).

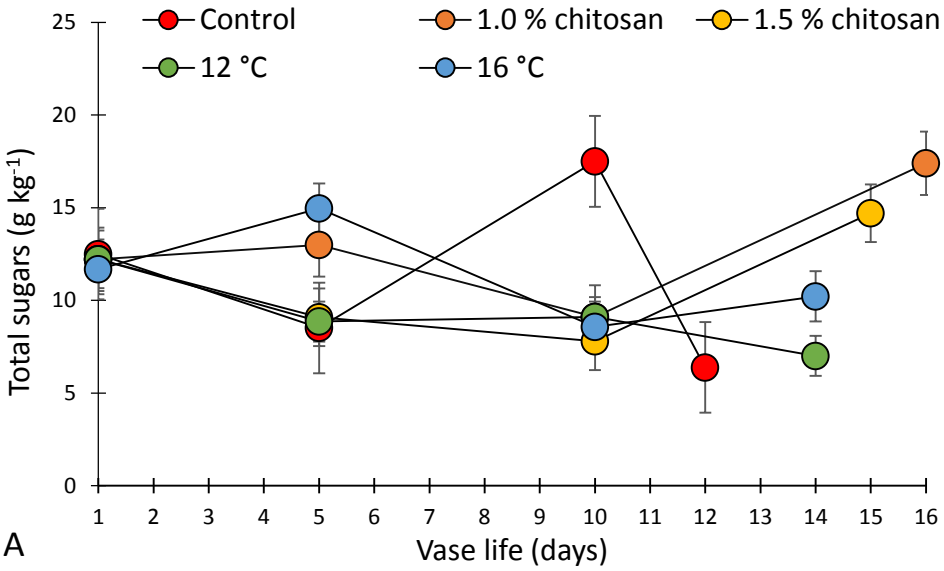
4.4.6 Total sugars

The differences in total sugars between treatments were significant for base of the stem, peduncle, and basal and apical bracts. The tissue on base of stem presented differences among the days evaluated, on day 5, 1.0 % chitosan treatment and 16 ºC stored treatment had the highest levels of total sugars; on day 10 the control had the highest concentration, and at the end of the vase life the chitosan treatments showed the lowest loss of sugars, in comparison with the other treatments. Throughout the study, 1.5 % chitosan treatment had the greatest variation in the concentrations of total sugars. At the end of vase life only both chitosan treatments increased total soluble sugars content at the base of stem (Figure 4.8-A).

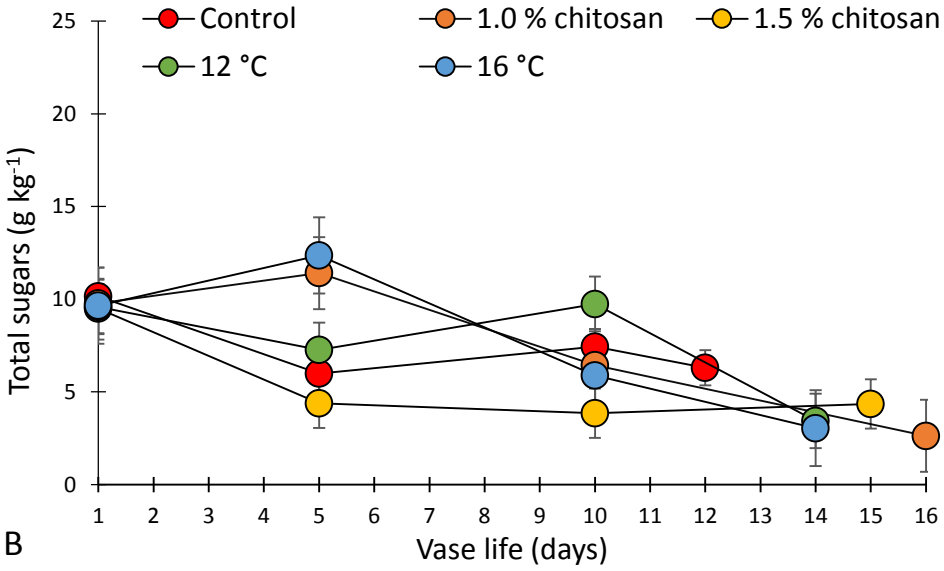
The general trend of total soluble sugars along the time up to peduncle showed significant differences only during day 5 of evaluation; where the 16 ºC stored treatment and 1.0 % chitosan treatment presented the highest concentrations (Figure 4.8-B). Throughout the evaluation, the treatments that showed greater fluctuations in the content of sugars were 1.0 % chitosan and the 12 ºC stored treatment, presented a significant reduction as the evaluation days elapsed, in contrast to the control that did not showed changes.

Total sugar content was different on basal bracts significant on day 5 and 8. Throughout the study, 1.5 % chitosan treatment and control did not show variations, while 1.0 % chitosan treatment had the greatest changes on the sugars concentration, reaching its highest concentration on day 5 (Figure 4.8-C).

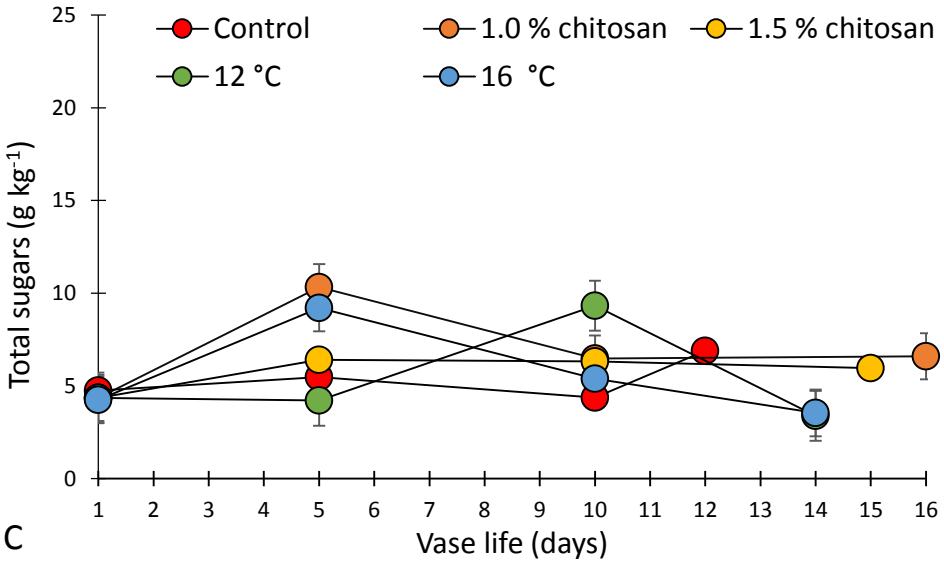
The apical bracts presented some significant differences between the days of evaluations. On day 5, the 16 ºC stored treatment and 1.0 % chitosan treatment had the highest levels of total sugars (50 % respect of start time; on day 10, all treatments did not change the sugar content as the start of the study, with the exception of 1.5 % chitosan treatment, that presented the highest decrease. At the end of vase life, again the 16 ºC stored treatment and 1.0 % chitosan treatment had the highest concentrations (Figure 4.8-D). Throughout the study, the 16 ºC stored treatment and 1.0 % chitosan treatment showed a tendency towards the increase of the concentrations of sugars as they passed the evaluation days.


Control
1.0 % chitosan
1.5 % chitosan
12 °C
16 °C
Total sugars (g kg-1)
Vase life (days)

A


Control
1.0 % chitosan
1.5 % chitosan
12 °C
16 °C
Total sugars (g kg-1)
Vase life (days)

B


Control
1.0 % chitosan
1.5 % chitosan
12 °C
16 °C
Total sugars (g kg-1)
Vase life (days)

C

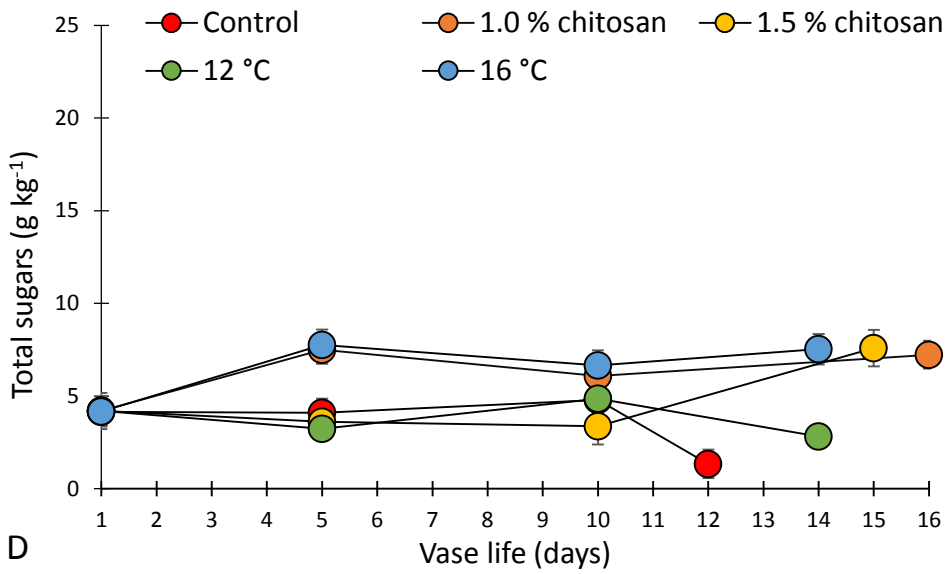


Figure 4.8. Total sugar content, values are the average of six observations ± SE of stem flowers of *Heliconia bihai* (L.) L. cv. Lobster Claw during the vase life period. A: base of stem, B: peduncle, C: basal bracts, D: apical bracts (n = 6).

4.5 Discussion

Storage at 16 ºC of the flower stems had a vase life time similar to 1.0 % chitosan treatment, which was maintained from the first day at room temperature (26 ºC). Therefore, a positive effect of chitosan on the post-harvest floral stems preservation was evident.

The organic coatings have recently been used for preservation on postharvest products (Durango et al., 2011). Among coatings, chitosan has attracted recent special attention due to its bactericidal and fungicidal activity; it is caused by its ability to bind to water molecules and inactivate microbial enzymes (Vásconez et al., 2009). Furthermore, the coatings modify the tissue materials exchange with the atmosphere surrounding it, resulting in the creation of a modified atmosphere, which acts as a barrier to gas exchange, mainly O_2, CO_2 and ethylene, reducing the respiration rate and delaying the senescence (Awad et al., 2017). The

chitosan coating can allow to maintain quality attributes and improving appearance during transportation, storage and shelf life (Amarante and Banks, 2000).

In this study both cold storage temperatures did not show differences on vase life. These results are different to those found by Costa et al. (2011b); these researchers observed that temperatures around to 12 ºC should not be used for more than 4 days in the cv. Lobster Claw, because the stems will present chilling injuries. In contrast, in our study the stems were stored for 10 days and its total postharvest life was 15.1 days. This represents 5 days more than those shown by these authors. These results suggest that transport over long distances could be possible, even if the flower stems have to be stored several days. This represents an advantage for the commercialization of species such as *Heliconia,* to new markets far away from growing sites.

Temperature plays a critical role on the life of cut flowers; generally, low temperatures delay senescence by reducing some tissue processes such as reserves exhaustion, respiration, ethylene production, or the excessive loss of water, even microorganism invasion, resulting all in the maintenance of the quality of the cut flowers (Nowak and Duncan, 1990).

The natural process of senescence can be accelerated or delayed, according to the management techniques during processing. Physiological and biochemical changes involve darkening, degradation and oxidation of pigments, increase of respiratory rate, loss of water, wilting, among others. An excellent alternative to avoid undesirable changes in the quality of horticultural products includes coatings of chitosan; but, research has been focused on the fruits conservation, leaving aside the effect it might have on cut flowers.

In our study, 1.0 % chitosan treatment had the lowest fresh weight loss at the end of vase life; therefore, we assume that this treatment conserved the water balance in the stems. We suggest this coat may retard the water loss due to the chitosan coating acts as a gas barrier of flower stems. Similarly El Ghaouth et al. (1991a) showed that 1.0 % chitosan coating on bell pepper and cucumber, at 13 ºC and 20 ºC, decreased water loss, delaying senescence most likely due to its ability to alleviate water stress. Similar results were obtained in strawberry, banana, and mango, which allowed to conclude that chitosan coatings reduce water loss due to respiration and transpiration during storage (Khosh et al., 2016; Kittur et al., 2001; Ribeiro et al., 2007). Additionally, these results showed that higher concentration of chitosan, as 1.5 %, did not prevent weight loss as 1.0 % concentration, suggesting that higher concentrations tended to form denser coating, resulting in less permeable coverage.

Similar results were obtained in papaya “Maradol”, stored at 5 ºC, and coated with 0.01 and 0.02 g mL^{-1} chitosan (González-Aguilar et al., 2009).

In the present study no differences on the conservation of the coloration of floral bracts between treatments at the end of vase life were found. This results contrasted with those found in fresh frozen strawberries, where chitosan coating with acetate delayed the colour change (Han et al., 2004); but, in litchi fruit the chitosan film decreased browning by reducing polyphenol oxidase and peroxidase activities (Ducamp-Collin et al., 2008) and in banana the chitosan coating delayed peel colour changes, possibly as a result of increase of carotenoids, delaying the degradation of chlorophylls or both (Awad et al., 2017).

The stability of anthocyanins can contrast with the flavonoids degradation (Rojas-Graü et al., 2009). Anthocyanins are phenolic compounds responsible to

produce colours, ranging from orange to red, with various shades of blue and purple, of a large variety of cut flowers. In this study, cold storage treatments preserved these pigments better at the end of vase life; in contrast, chitosan was unable to maintain the parameters L*, C*, and h°. This can be explained because the chitosan treatments and the control were maintained to a high temperature during the study. This result may indicate that chitosan could be more efficient at cold temperature. Temperature can affect these pigments concentration, because anthocyanin content is dependent of enzymatic activity of the anthocyanin biosynthetic pathway catalized by phenylalamine ammonia lyase and UDP-glucose: flavonoid-3-O-glucosyltransferase (Gil et al., 1995). Özdemir and Gökmen (2017) reported an increase in anthocyanin concentration during cold storage in pomegranates. In contrast, Tezotto-Uliana et al. (2014) suggested that the lower concentrations of anthocyanins presented in the chitosan treatments may be due to the activity of peroxidase and polyphenol oxidase are influenced by the presence of O_2, which is blocked by the chitosan coating, resulting in the metabolism inhibition. Studies by El Ghaouth et al. (1991b) showed than 1.0 % chitosan coting on strawberries decelerated the anthocyanins synthesis, and the response was opposite when the highest concentration (1.5 %) was used.

In our study, the total flavonoids content fluctuated throughout the study, but at the end of vase life in 1.0 % chitosan coated and 16 ºC stored treatments showed a higher concentration of these compounds. These results suggested the possible chitosan protective effect on the cellular structure, delaying the phenol concentrations decrease during senescence due to maintaining cellular integrity. Naczk and Shahidi (2004) reported that the flavonoid levels can be affected by

postharvest conditions, as temperature. Additionally, the decrease in total phenolics may be result of the enzymatic reactions occurring during the senescence of the plants (Ayaz, 2001).

The results in the present study showed the positive effect over the content of total sugars in 1.0 % chitosan treatment and the storage at 16 °C treatment on the apical bracts. This result may be an indication that chitosan acts as a protection to consume sugars at high temperatures by decreasing respiration. High temperatures increase respiration rates and the water consumption of in cut flowers (Adachi et al., 2000). This confirms the results regarding weight loss, where 1.0 % chitosan treatment resulted in the less weight loss, suggesting that the maintenance of carbohydrates contribute to the maintenance of water uptake and retarding the senescence.

The conservation of adequate levels of sugars during postharvest of cut flowers is crucial to achieving an extensive vase life, due to carbohydrates are an important source of energy and structural components, besides being metabolism regulatory molecules, and as regulatory osmotic elements (Kumar et al., 2008); additionally, it has been found that sugars are actively involved in delaying the increase in mRNA abundance of a senescence-associated genes (Hoeberichts et al., 2007).

4.6 Conclusions

Both 1.0 % chitosan and 16 ºC storage treatment showed the best results of vase life and TPL. The results of this research are evidence that chitosan may be used in post-harvest of floral stems; however, deeper investigations should be

performed to identify the exact mechanisms under which chitosan exerts its effects.

4.7 Acknowledgments

To the Consejo Nacional de Ciencia y Tecnología (CONACyT) for the scholarship to the project "Storage treatments and vase life of three varieties of *Heliconia*". Also a special thanks to Dr. José Andrés Herrera Corredor and M.C. Mirna López Espíndola for lending the colour analysis program.

4.8 References

Adachi, M., Kawabata, S., Sakiyama, R., 2000. Effects of temperature and stem length on changes in carbohydrate content in summer-grown cut chrysanthemums during development and senescence. Postharvest Biol. Technol. 20, 63–70. doi:10.1016/S0925-5214(00)00106-X

Amarante, C., Banks, N.H., 2000. Postharvest Physiology and Quality of Coated Fruits and Vegetables, in: Janick, J. (Ed.), Horticultural Reviews. John Wiley & Sons, Inc., pp. 161–238. doi:10.1002/9780470650806.ch4

Anon., 1991. Care of tropicals. Flor Rev 182, 35–38.

Aular, J., Ruggerio, C., Durigan, 2002. Relación entre el color de la cáscara y las características del fruto y jugo de la parchita maracuyá. Bioagro 14, 47–51.

Awad, M.A., Al-Qurashi, A.D., Mohamed, S.A., El-Shishtawy, R.M., Ali, M.A., 2017. Postharvest chitosan, gallic acid and chitosan gallate treatments effects on shelf life quality, antioxidant compounds, free radical scavenging capacity and enzymes activities of “Sukkari” bananas. J. Food Sci. Technol. 54, 447–457. doi:10.1007/s13197-016-2481-8

Ayaz, F.A., 2001. Changes in phenolic acids of cherry laurel [Laurocerasus officinalis Oxygemmis] fruit during maturation. Acta Biol Cracov Bot 43, 23–26.

Bañuelos-Hernández, K.P., García-Nava, J.R., Leyva-Ovalle, O.R., Peña-Valdivia, C.B., Ybarra-Moncada, M.C., 2016. Flowering stem storage of Heliconia psittacorumL. f. cv. Trópica. Postharvest Biol. Technol. 112, 159–169. doi:10.1016/j.postharvbio.2015.10.006

Barrón-Yánez, R.M., García-Mateos, M. del R., Soto-Hernández, M.R., Colinas-León, T., Kite, G., 2011. Flavonoides y actividad antioxidante de Calia secundiflora (Ort.) Yakovlev. Rev. Fitotec. Mex. 34, 151–157.

Broschat, T., Donselman, H., 1983. Production and postharvest culture of Heliconia psittacorum flowers in Souht Florida. Proc Fla State Hort Soc 96, 272–273.

Broschat, T.K. (University of F., Donselman, H., 1988. Production and postharvest culture of red ginger in south Florida. Proc. Annu. Meet. Fla. State Hortic. Soc. USA 101, 326–327.

Celikel, F.G., Reid, M.S., 2002. Storage temperature affects the quality of cut flowers from the Asteraceae. HortScience Publ. Am. Soc. Hortic. Sci. 37, 148–150.

Costa, A.S., Nogueira, L.C., dos Santos, V.F., Finger, F.L., Camara, T.R., Loges, V., Willadino, L., 2011a. Characterization of symptoms of senescence and chilling injury on inflorescences of Heliconia bihai (L.) cv. Lobster Claw and cv. Halloween. Postharvest Biol. Technol. 59, 103–109. doi:10.1016/j.postharvbio.2010.08.015

Costa, A.S., Nogueira, L.C., Santos, V.F. dos, Camara, T.R., Loges, V., Willadino, L., 2011b. Storage of cut Heliconia bihai (L.) cv. Lobster Claw flowers at low temperatures. Rev. Bras. Eng. Agríc. E Ambient. 15, 966–972.

Ducamp-Collin, M.-N., Ramarson, H., Lebrun, M., Self, G., Reynes, M., 2008. Effect of citric acid and chitosan on maintaining red colouration of litchi fruit pericarp. Postharvest Biol. Technol. 49, 241–246. doi:10.1016/j.postharvbio.2008.01.009

Durango, A.M., Soares, N.D.F., Arteaga, M.R., 2011. Edible films and coatings as biodogradable active packaging in the preservation of food products. Biotecnol. En El Sect. Agropecu. Agroindustrial 9, 112–118.

El Ghaouth, A., Arul, J., Ponnampalam, R., Boulet, M., 1991a. Use of chitosan coating to reduce water loss and maintain quality of cucumber and bell pepper fruits. J. Food Process. Preserv. 15, 359–368. doi:10.1111/j.1745-4549.1991.tb00178.x

El Ghaouth, A., Arul, J., Ponnampalam, R., Boulet, M., 1991b. Chitosan coating effect on storability and quality of fresh strawberries. J. Food Sci. 56, 1618–1620. doi:10.1111/j.1365-2621.1991.tb08655.x

González-Aguilar, G.A., Valenzuela-Soto, E., Lizardi-Mendoza, J., Goycoolea, F., Martínez-Téllez, M.A., Villegas-Ochoa, M.A., Monroy-García, I.N., Ayala-Zavala, J.F., 2009. Effect of chitosan coating in preventing deterioration and preserving the quality of fresh-cut papaya “Maradol.” J. Sci. Food Agric. 89, 15–23. doi:10.1002/jsfa.3405

Halevy, A.H., Kofranek, A.M., Besemer, S.T., 1978. Postharvest handling methods for bird of paradise flowers (Strelitzia reginae Ait). J Amer Soc Hort Sci 103, 165–169.

Han, C., Zhao, Y., Leonard, S.W., Traber, M.G., 2004. Edible coatings to improve storability and enhance nutritional value of fresh and frozen strawberries (Fragaria × ananassa) and raspberries (Rubus ideaus). Postharvest Biol. Technol. 33, 67–78. doi:10.1016/j.postharvbio.2004.01.008

Hoeberichts, F.A., van Doorn, W.G., Vorst, O., Hall, R.D., van Wordragen, M.F., 2007. Sucrose prevents up-regulation of senescence-associated genes in carnation petals. J. Exp. Bot. 58, 2873–2885. doi:10.1093/jxb/erm076

Jaroenkit, T., Paull, R.E., 2003. Postharvest handling of Heliconia, red Ginger, and Bird-of-Paradise. HortTechnology 13, 259–266.

Kasso, M., Bekele, A., 2016. Post-harvest loss and quality deterioration of horticultural crops in Dire Dawa Region, Ethiopia. J. Saudi Soc. Agric. Sci. doi:10.1016/j.jssas.2016.01.005

Khosh, A.S., Badii, F., Hashemi, M., Ardakani, A.Y., Maftoonazad, N., Gorji, A.M., 2016. Effect of nanochitosan based coating on climacteric behavior and postharvest shelf-life extension of apple cv. Golab Kohanz. LWT - Food Sci. Technol. 70, 33–40. doi:10.1016/j.lwt.2016.02.002

Kittur, F., Saroja, N., Habibunnisa, Tharanathan, R., 2001. Polysaccharide-based composite coating formulations for shelf-life extension of fresh banana and mango. Eur. Food Res. Technol. 213, 306–311. doi:10.1007/s002170100363

Kumar, N., Srivastava, G.C., Dixit, K., 2008. Flower bud opening and senescence in roses (Rosa hybrida L.). Plant Growth Regul. 55, 81. doi:10.1007/s10725-008-9263-x

Naczk, M., Shahidi, F., 2004. Extraction and analysis of phenolics in food. J. Chromatogr. A, Food Science 1054, 95–111. doi:10.1016/j.chroma.2004.08.059

Nowak, J., Duncan, A.A., 1990. Postharvest Handling and Storage of Cut Flowers, Florist Greens, and Potted Plants. Timber Press.

Olayemi, F.F., Adegbola, J.A., Bamishaiye, E.I., Daura, A.M., 2010. Assessment of post-harvest challenges of small scale farm holders of tomotoes, bell and hot pepper in some local government areas of Kano State, Nigeria. Bayero J. Pure Appl. Sci. 3, 39–42. doi:10.4314/bajopas.v3i2.63217

Özdemir, K.S., Gökmen, V., 2017. Extending the shelf-life of pomegranate arils with chitosan-ascorbic acid coating. LWT - Food Sci. Technol. 76, Part A, 172–180. doi:10.1016/j.lwt.2016.10.057

Rahmani, I., Ahmadi, N., Ghanati, F., Sadeghi, M., 2015. Effects of salicylic acid applied pre- or post-transport on post-harvest characteristics and antioxidant enzyme activity of gladiolus cut flower spikes. N. Z. J. Crop Hortic. Sci. 43, 294–305. doi:10.1080/01140671.2015.1096799

Ribeiro, C., Vicente, A.A., Teixeira, J.A., Miranda, C., 2007. Optimization of edible coating composition to retard strawberry fruit senescence. Postharvest Biol. Technol. 44, 63–70. doi:10.1016/j.postharvbio.2006.11.015

Ribeiro, W.S., Carneiro, G.G., Almeida, E.I.B., Lucena, H.H., Barbosa, J.A., 2010. Pós-colheita e conservação de inflorescências de Heliconia marginata x heliconia bihai (Heliconia rauliana) submetidas a soluções de manutenção. Agropecuária Téc. 31, 70–74.

Rojas-Graü, M.A., Oms-Oliu, G., Soliva-Fortuny, R., Martín-Belloso, O., 2009. The use of packaging techniques to maintain freshness in fresh-cut fruits and vegetables: a review. Int. J. Food Sci. Technol. 44, 875–889. doi:10.1111/j.1365-2621.2009.01911.x

Salinas, Y., Martínez, F., Soto, M., Ortega, R., Arellano, J.L., 2003. Efecto de la nixtamalización sobre las antocianinas del grano de maíces pigmentados. Agrociencia 37, 617–628.

Tezotto-Uliana, J.V., Fargoni, G.P., Geerdink, G.M., Kluge, R.A., 2014. Chitosan applications pre- or postharvest prolong raspberry shelf-life quality. Postharvest Biol. Technol. 91, 72–77. doi:10.1016/j.postharvbio.2013.12.023

Vásconez, M.B., Flores, S.K., Campos, C.A., Alvarado, J., Gerschenson, L.N., 2009. Antimicrobial activity and physical properties of chitosan–tapioca starch based edible films and coatings. Food Res. Int. 42, 762–769. doi:10.1016/j.foodres.2009.02.026

5. CONCLUSIONES GENERALES

El quitosano es una alternativa para mantener la calidad y extender la vida en florero de las inflorescencias de *Heliconia*, aunque los tallos florales tratados con este biopolímero sean almacenados a temperatura ambiente, pues superó al control a los tratamientos almacenados en refrigeración.

No obstante, el efecto del quitosano fue diferente entre las especies y las variedades. Esta diferencia en la respuesta puede atribuirse a sus diferencias genéticas, y/o a las concentraciones del polímero utilizadas, por lo cual, las investigaciones deberán realizarse en cada especie. El quitosano al 1.0 % mostró el efecto mayor en extensión en la vida en florero de los tallos de *H psittacorum* cv. Trópica, pero también incrementó significativamente la de *H. bihai* cv. Halloween y *H. bihai* cv. Lobster Claw. Estos resultados indican que el quitosano en concentraciones pequeñas presentan mejor efecto en la longevidad en estas especies y variedades; a la vez, *H. psittacorum* mostró que tiene resistencia mayor a la manipulación postcosecha.

En general, ninguno de los tratamientos conservó la coloración (luminosidad, cromaticidad ni el matiz) de las inflorescencias al final de la evaluación. Pero, la combinación de quitosano y refrigeración podría mantener la coloración de las brácteas florales por períodos largos de tiempo.

En *H. bihai* cv. Lobster Claw el quitosano al 1.0 % y el almacenamiento a 12 y 16 ºC mostraron la vida mayor en florero. Los efectos mejores se observaron en los tallos fueron tratados con quitosano al 1.0 % y los almacenados a 16 ºC, lo que permite concluir que el quitosano es una opción para sustituir el almacenamiento refrigerado para esta variedad en sitios donde el almacenamiento postcosecha refrigerado no exista.

Al respecto, las temperaturas de refrigeración no ocasionaron lesiones por frío en las variedades evaluadas; sin embargo, el almacenamiento a 16 ºC mostró la mayor vida en florero. Por lo que, intervalos cercanos a esta temperatura son recomendados para el almacenamiento de especies tropicales, como las heliconias. También, la variedad Losbter Claw presenta sensibilidad mayor a la temperatura baja en comparación con las otras variedades evaluadas.

De acuerdo con los resultados, se propone que el daño en los tejidos de heliconias presenta un gradiente con dirección basipétala; por lo tanto, los tejidos más jóvenes (brácteas florales apicales) mantienen su funcionamiento y constitución celular por tiempo mayor.

En conclusión, tanto el tratamiento con quitosano como el frío presentaron ventajas en la vida en florero y calidad de los tallos florales de las variedades evaluadas; sin embargo, el quitosano es una alternativa potencial novedosa para usar en postcosecha en ornamentales de origen tropical. Por lo tanto, esta investigación sienta los precedentes para desarrollar estudios enfocados en el uso del quitosano en flores de corte y para dilucidar los mecanismos de protección celular, molecular y proteómico promovidos por el biopolímero y del efecto sinérgico del quitosano y otros métodos naturales de control biológico, que contrarrestan la actividad de patógenos.