

Phenology and environment in the presence of secondary metabolites in *Psidium guajava* L.

Fenología y ambiente en la presencia de metabolitos secundarios en *Psidium guajava* L.

Fenologia e ambiente na ocorrência de metabólitos secundários em *Psidium guajava* L.

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Rev. Fac. Agron. (LUZ). 2023, 40(4): e2340Spl04

ISSN 2477-9407

DOI: [https://doi.org/10.47280/RevFacAgron\(LUZ\).v40.supl.04](https://doi.org/10.47280/RevFacAgron(LUZ).v40.supl.04)

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Received: 09-10-2023

Accepted: 22-11-2023

Published: 06-12-2023

Crop production

Associate editor: Dr. Jorge Vilchez-Perozo  

University of Zulia, Faculty of Agronomy
Bolivarian Republic of Venezuela

Keywords:

Phenology
Environment
Secondary metabolites

Abstract

Guava (*Psidium guajava* L.) is one of the most important fruit trees in Venezuela due to the acceptance of its fresh and processed fruit with relevant sensory and nutritional characteristics. The establishment of the crop in the producing areas has been the result of the initiative of leading farmers in the country. Due to the potential of guava, the present review aimed to describe the phenology, environment, and presence of secondary metabolites in *P. guajava*. The search for information on *P. guajava* included several key words such as phenology, flowering, fruiting, and secondary metabolism. Sixty-six references were selected from 130 results, including research articles, reviews, and books published between 1991 and 2023. Secondary metabolite biosynthesis is a dynamic process that depends on numerous factors associated with the plant and the environment. The content of phenols and flavonoids in guava can help characterize its production, agroindustrial, and pharmaceutical importance, be a tool for cultivar selection, and anticipate the content of other secondary metabolites to identify plants that differ in their production.



Resumen

El guayabo (*Psidium guajava* L.) se ubica entre los frutales más relevantes en Venezuela por la aceptación de su fruta fresca y procesada con características sensoriales y nutricionales relevantes. El establecimiento del cultivo en las zonas productoras se ha realizado por la iniciativa de agricultores líderes del país. Debido al potencial del guayabo la presente revisión tuvo como objetivo describir la fenología, el ambiente y la presencia de metabolitos secundarios en *P. guajava*. La búsqueda de información sobre *P. guajava* incluyó varias palabras clave como fenología, floración, fructificación y metabolismo secundario. Se seleccionaron 66 referencias de 130 resultados, incluyendo artículos de investigación, revisiones y libros publicados entre 1991 y 2023. La biosíntesis de metabolitos secundarios es un proceso dinámico que depende de numerosos factores asociados a la planta y al ambiente. El contenido de fenoles y flavonoides en guayabo puede ayudar a caracterizar la producción e importancia agroindustrial y farmacéutica, ser una herramienta para la selección de cultivares y anticipar el contenido de otros metabolitos secundarios para identificar plantas que se diferencien en la producción de ellos.

Palabras clave: fenología, ambiente, metabolitos secundarios.

Resumo

A goiaba (*Psidium guajava* L.) é uma das árvores frutíferas mais importantes na Venezuela devido à aceitação de seus frutos frescos e processados com características sensoriais e nutricionais relevantes. O estabelecimento da cultura nas áreas produtoras tem sido efetuado por iniciativa dos principais agricultores do país. Devido ao potencial da goiaba, a presente revisão teve como objetivo descrever a fenologia, o ambiente e a presença de metabólitos secundários em *P. guajava*. A busca de informações sobre *P. guajava* incluiu várias palavras-chave, como fenologia, floração, frutificação e metabolismo secundário. Foram selecionadas sessenta e seis referências de 130 resultados, incluindo artigos de investigação, revisões e livros publicados entre 1991 e 2023. A biossíntese de metabólitos secundários é um processo dinâmico que depende de numerosos factores associados à planta e ao ambiente. O conteúdo de fenóis e flavonóides na goiaba pode ajudar a caracterizar a produção e a importância agroindustrial e farmacêutica, ser uma ferramenta para a seleção de cultivares e antecipar o conteúdo de outros metabólitos secundários para identificar plantas que diferem na sua produção.

Palavras-chave: fenologia, ambiente, metabólitos secundários.

Introduction

Guava (*Psidium guajava* L.; family: Myrtaceae) cultivation in Venezuela faces challenges due to oversupply and high costs derived from the excess of chemical inputs for pest, disease and weed management. This causes a decrease in market prices and profitability, which hinders the international positioning of the crop. Guava fruit has excellent values in minerals, vitamins (Vijaya *et al.*, 2020) and secondary metabolites such as phenols and flavonoids (Pérez-Pérez *et al.*, 2014; 2019; 2020) generating biotechnological interest for its antioxidant, anti-inflammatory, antibacterial, antihyperlipidemic, cardioprotective, antimutagenic (Naseer *et al.*, 2018), antifungal

(Liu *et al.*, 2018), antidiabetic, hepatoprotective (Li *et al.*, 2021) and larvicidal (Mendes *et al.*, 2017) properties.

The leaves contain phenols (Pérez-Pérez *et al.*, 2014) and flavonoids (Pérez-Pérez *et al.*, 2014; Rivero-Maldonado *et al.*, 2013), and its essential oil is rich in phenolic compounds from the tannin and flavonoid groups (Jassal and Kaushal, 2019). Secondary metabolite biosynthesis is a dynamic process influenced by factors such as plant type and ontogenetic stage (Verma and Shukla, 2015). The variety of compounds in plants supports phenotypic plasticity (Kaplan *et al.*, 2008).

The aforementioned allows describing the relevance of *P. guajava* plants in the present review, in correspondence with phenology, the relationships between the environment and phenology, phenological phases and the presence of secondary metabolites and between the environment and the presence of secondary metabolites, this based on Verma and Shukla (2015), who consider that the biosynthesis of secondary metabolites is a dynamic process that depends mainly on numerous factors associated with the plant itself and with the environment. Within the first group of factors, the most important are the type of plant and the stage of its ontogenetic development. The objective of this review was to describe the phenology, the environment and the presence of secondary metabolites in *P. guajava*.

Methods

The search words or phrase were “*Psidium guajava* L.”, “phenology”, “flowering”, “fruiting”, “vegetative sprouting”, “secondary metabolites”, “phenolic compounds”, “flavonoids”, “antioxidant capacity”, “biotic stress” and abiotic stress”. Scientific literature was searched using the web search engine Scholar Google, Scopus, Web of Science, Taylor and Francis, Springer Link, PubMed and Science Direct. For the review 66 references were selected from the 130 resulting from the search; those included research articles, review articles and books published between 1991 and 2023.

Discussion

Phenology of *P. guajava*

Phenology included flowering, fruiting, budding and leaf fall on a temporal scale to identify the vegetative and reproductive processes of the species (Biondi *et al.*, 2007). These vary among genotype, edaphoclimatic conditions and crop management (Fisher and Orduz-Rodríguez, 2012). The adult guava plant has been described as a semi-deciduous tree, because after harvesting it undergoes a phenomenon of exhaustion or lethargy with the presence of yellowing and the fall of most of the leaves, which continues during the dry period, resuming the growth of new branches and bud regrowth with the onset of the rainy season (Gómez, 1995).

This phenological development was evidenced in the municipality of Mara, Zulia state, Venezuela (Marín *et al.*, 2000); the study showed that it was mainly due to the interaction of age and/or genetic potential with the environment. Pérez-Pérez (2023) affirmed the occurrence of the phenological phases of fruiting, vegetative sprouting and flowering, the first being the most frequent, followed by vegetative sprouting, a behavior that corresponded with that reported by Esparza *et al.* (1993) and Marín *et al.* (2004). In turn, Marín *et al.* (2000) identified fruit set in all plants throughout the phenological stages of vegetative and reproductive sprouting, during the evaluation of *P. guajava* Criolla roja, Cubana and Montalbán cultivars grafted on Cas

(*P. friedrichsthalianum*). They also pointed out that the differences in phenological behavior were affected by the interaction between canopy/pattern and environment, reporting that flowering time varied among the materials from 15 to 45 days.

Marín *et al.* (2004) evaluated organic amendments in the recovery of guava trees growing in a field infested with nematodes of the genus *Meloidogyne*, in the state of Zulia, Venezuela. The study showed that during the evaluation period, the plants that received organic amendments registered continuous vegetative and reproductive structures, with the highest fruiting occurring during the month of July. According to Esparza *et al.* (1993), this behavior is due to the physiological response of the plant to the degree of soil moisture that promotes the onset of the flowering phase, which is directly associated with the fruiting potential, finding a greater supply of fruit in the months from June to August (50.6 %) and from November to January (31.1 %) remaining production.

Tong *et al.* (1991) documented guava production in Mara, Venezuela, in two seasons, each influenced by the bimodal rainfall of the area, with significant flower emergence. However, in *P. guajava* the phenological phases overlap (Pérez-Pérez *et al.*, 2023), due to the continuity of guava production during the year (Marín *et al.*, 2000). The growth of vegetative buds during the flowering and vegetative sprouting phase (Pérez-Pérez *et al.*, 2023) has been associated with the preparation of vegetative organs, mainly leaves, which will guarantee the production of photoassimilates, such as carbohydrates, that will support the development of reproductive structures, including fruits. In general, the guava plant takes about 100 to 150 days from flowering to harvest (Singh, 2011).

In this regard, Pawar and Rana (2019) have pointed out that the balance between vegetative and reproductive growth is an important aspect to improve the yield and quality of fruit trees, based on the source-sink relationship, the authors stated that profuse flowering and fruiting create a high demand for the limited source of carbohydrates, which affects fruit set and fruit development, hence the importance of knowing the regulation of carbon distribution in fruit plants in the short and long term, given the perennial nature of the trees.

Relationship between environment and phenology of *P. guajava*

The growth cycle depends on the genotype of the plant (Salazar *et al.*, 2006) and the environment. Hence, it is essential to evaluate the climatic circumstances that favor plant development (Ferreira *et al.*, 2019). When grown in different environments, identical genotypes can present multiple developmental stages. Hence, it is essential to understand the phenological cycles of crops in order to define agronomic management strategies in accordance with the phenological stages of the plant (Salazar *et al.*, 2006).

Another aspect to consider, during flowering and fruiting of guava, is that environmental conditions and nutrition play an important role in their determination. For efficient flowering and fruiting, sunlight is necessary; however, periods of water stress are critical to improve flowering. Often under favorable conditions, many of the flowers emerging from the leaf axils of young shoots (solitary or in groups of two or three), generate fruit on trees (Shivpoojan *et al.*, 2018). In this regard, Mendoza *et al.* (2017) noted that the reproductive phase of plants was promoted by precipitation by 73.4 %, compared to temperature (19.3 %) followed by solar radiation or photoperiod (3.2 %), results obtained in their study on agrometeorological influence on the reproductive phase.

In the case of tropical conditions, phenological changes are defined by periods of drought and rain, manifesting physiological changes with increased growth, swelling and bud detachment, increased stem diameter, floral initiation and differentiation, initiation of fruit set and fruit ripening (Quirós *et al.*, 2009; Salazar *et al.*, 2006), with the difference that guava can produce fruit throughout the year with maximum and minimum production peaks, depending on climatic conditions (Marín *et al.*, 2000; 2004; Pérez-Pérez *et al.*, 2023).

Marín *et al.* (2000), in their study on the behavior of guava types grafted on *P. friedrichsthalianum* Berg-Niendenzu, reported vegetative and reproductive sprouting with the presence of physiologically mature fruit on all plants. This is due to the behavior of this species, which maintains the three phenological stages active on the plant throughout the year (Marín *et al.*, 2004), with a variation in the intensity of occurrence, responding to the climate in the area.

Pérez-Pérez *et al.* (2023) studied the environmental impact on guava flower, fruit and vegetative bud production in Mara, Zulia, Venezuela. These researchers emphasized the highest production of vegetative buds in October-November, the subsequent decrease and the maximum in may. Precipitation affected the vegetative budburst phase (da Fontoura *et al.*, 2021) between October and February, while absence in March affected it, similar to Marín *et al.* (2000). The researchers did not identify any significant relationship with temperature, humidity, evaporation and insolation (Mendoza *et al.*, 2017).

Flower production alternates with fruit production. The higher or lower occurrence of fruit corresponds to the lower or higher flower production that occurred 90 days earlier (Pérez-Pérez *et al.*, 2023; Singh, 2011), as a response to the higher amount of water available to the plants due to the occurrence of rainfall (Marín *et al.*, 2000).

Relationship between the phenological phases of *P. guajava* and the presence of secondary metabolites.

In the course of evolution, plants have developed the ability to produce an enormous amount of phenolic secondary metabolites, which are not necessary in the primary processes of growth and development, but are of vital importance for their interaction with the environment, for their defense mechanisms (Cheynier *et al.*, 2013) and perpetuate their species (Lustre, 2022; Mamani and Filippone, 2018; Rioja Soto, 2020).

A large number of secondary molecules are biosynthesized from primary metabolites and accumulate in plant cells (Böttger *et al.*, 2018; Narayani *et al.*, 2017; Rejeb *et al.*, 2014), whose production could be induced under *in vitro* conditions when cell cultures by using biotic, abiotic elicitors and signaling molecules and are usually increased by specific stress effect (Piñol *et al.*, 2013).

The accumulation and concentration of polyphenolic compounds varies greatly in different plant parts and organs, closely related to their function in the life cycle and growth phase (Verma and Shukla, 2015). For example, during the fruiting phase of guava, leaves have the highest amount of phenols, compared to vegetative shoots and flowers that contain less (Pérez-Pérez *et al.*, 2023).

Phenols can modulate essential physiological processes, such as transcriptional regulation and signal transduction. Some interesting effects of phenols in plants are also those associated with growth hormone (auxin). An additional role has been observed for flavonoids in the functional development of pollen. Finally, anthocyanins represent a class of flavonoids that provide the orange, red and blue/purple colors to many plant tissues. According to co-evolution theory, red is a signal of tree status to insects migrating to (or moving between) trees in autumn (Cheynier, 2013).

Phenols are synthesized in leaves (Vranová *et al.*, 2012), participate in their development (Pawar and Rana, 2019) and from there are transported to other tissues and organs. Leaves have higher concentrations of these compounds than the rest of the plant (Vranová *et al.*, 2012). However, they are not uniformly found throughout the plant and are often restricted to particular organs and to certain cells and tissues within that organ (Vranová *et al.*, 2012).

The leaf is the main organ where photosynthesis takes place and provides carbohydrates that are diverted to the reproductive structures (Fotirić *et al.*, 2020; Pawar and Rana, 2019) and to the root apices and stems of the plant (Pawar and Rana, 2019). The fate of carbohydrates formed during photosynthesis, part of which will be used in the structural makeup of the plant (primary metabolism) (Azad *et al.*, 2020) and others in secondary metabolism, will have a major influence on the quantity and quality of phenolic compounds produced (Saltveit, 2017).

Flavonoids play important roles in plant biology, responding to light and controlling the levels of growth-regulating auxins, plant differentiation and in virtually every interaction a plant establishes with its environment (Vicente and Boscaiu, 2018). Among these, anthocyanins (Peñarrieta *et al.*, 2014), act in the development of petal color to attract pollinators (Zhang *et al.*, 2014).

Flavonoids (anthocyanins, flavones, flavonols and isoflavones) are the most abundant phenolic compounds, have an important role in plant growth, as well as in the defense mechanism against microorganisms and insects (Shen *et al.*, 2022) and are abundant in woody species (Baskar *et al.*, 2018). In the leaf, they are found in high concentration as catechin and epicatechin (Pérez-Pérez *et al.*, 2020). The highest flavonoid content is found in the flowering phase, followed by the vegetative sprouting and fruiting phase (Pérez-Pérez *et al.*, 2023), due to the competition for carbon skeletons between flavonoid synthesis and the synthesis of compounds involved in fruit cell division (Li *et al.*, 2021).

Guava leaf extracts contain phenolic compounds such as gallic acid, catechin, chlorogenic acid, caffeic acid, epicatechin, rutin, quercetin, kaempferol and luteolin, crucial for antioxidant activity, not only because of their ability to donate hydrogen, but also because of their stable intermediate radicals, which prevent oxidation (Morais-Braga *et al.*, 2017).

Determinations carried out in different geographical locations have reported compounds such as flavonoids, glycosides and aglycones (Wang *et al.*, 2017). As asserted by Camarena-Tello *et al.* (2018), because phenolic compounds are considered the main group of secondary metabolite present in plants (Tolić *et al.*, 2017).

The guava plant directs its secondary metabolism towards the production of metabolites at the level of various plant organs such as the leaf (Azad *et al.*, 2020), which are being demanded by the organs involved in the phenological phase in which the plant is found (Verma and Shukla, 2015). Thus, higher flower production was associated with higher production of flavonoids determined in the leaf (Pérez-Pérez *et al.*, 2023). For Coutinho (2013), secondary metabolites represent a chemical interface between plants and the environment, therefore, their synthesis is often affected by environmental conditions.

Effect of the environment on the presence of secondary metabolites

Plants produce secondary metabolites during growth, which serve a variety of cellular essential for physiological processes and stress response signaling (Isah, 2019). The type and concentration of these molecules depend on the species, genotype, developmental stage,

plant tissue or organ, and developmental environment (Isah, 2019; Speed *et al.*, 2015). Genetic characteristics influence intrapopulation variability (Speed *et al.*, 2015), with phenotypic character being quantified (Arbona *et al.*, 2013), influenced by genetic and environmental factors (Turner *et al.*, 2016).

Environmental factors

Environmental factors, including biotic and abiotic stresses, influence plant secondary metabolite synthesis. Factors such as nutrient supply, air temperature, light and water can affect their concentration (Arbona *et al.*, 2013; Isah, 2019; Speed *et al.*, 2015), to counteract stress (Arbona *et al.*, 2013; Lustre, 2022; Mamani and Filippone, 2018). Then environmental factors are decisive in the biosynthesis of secondary metabolites (Yang *et al.*, 2018).

Appiah *et al.* (2022) suggest that factors such as development, seasonality, precipitation, temperature and altitude are correlated and can influence secondary metabolism. According to Huyskens-Keil *et al.* (2020), high temperatures and plant age can increase the accumulation of phenolic compounds, while light enhances the biosynthesis of these compounds. Both biotic and abiotic stresses stimulate carbon fluxes from primary to secondary metabolic pathways, as noted by Lattanzio (2013).

Biotic factors

Plants are physically attacked by many biological agents such as fungi, viruses, bacteria, nematodes, among others, which causes stress in plants known as biotic stress. According to Jan *et al.* (2021), plants show resistance against these pathogens through secondary metabolites. Some secondary metabolites have antimicrobial activities that function as a plant defensive system against these pathogens (Xu *et al.*, 2022). The requirement of high concentrations of secondary metabolites, by plants, during defense against pathogens causes their biosynthesis to be rapidly triggered.

Abiotic factors

During ontogeny, plants interact with the surrounding environment and come into contact with different abiotic components such as water, light, temperature, soil, and chemicals (minerals/fertilizers) (Jan *et al.*, 2021; Xu *et al.*, 2022). Plants require these components for their development and survival in adequate amounts. However, more or less of these abiotic components cause stress to plants and ultimately lead to variations in the production or accumulation of secondary metabolites during different developmental stages of the plant life cycle, depending on their need (Verma and Shukla, 2015).

Secondary metabolites play an important role in plant adaptation to the environment and in overcoming stress conditions (Lattanzio, 2013) and consequently plant adaptation to a given area (Pant *et al.*, 2021).

According to Coutinho (2013) and Sampaio *et al.* (2016) temporal and spatial variations, total content, as well as proportions of secondary metabolites in plants occur at different levels (seasonal and daily, intraplant, inter- and intraspecific) and despite the existence of genetic control, expression may undergo modifications resulting from the interaction of biochemical, physiological, ecological and evolutionary processes. They represent a chemical interface between plants and the surrounding environment, so their synthesis is frequently affected by environmental conditions (Chiveu *et al.*, 2019).

In a recent investigation Pérez-Pérez *et al.* (2023) obtained higher concentrations of flavonoids in the fruiting phase in the absence of precipitation, as well as high temperatures and insolation in the study area. Espinosa-Leal *et al.* (2021) and Piasecka *et al.* (2017) indicated that plant exposure to drought promoted increased production of

several classes of secondary metabolites including terpenes, complex phenols, and alkaloids during *in vitro* and *in vivo* growth through the induction of ionic or osmotic stress.

Conclusion

Guava is a plant rich in secondary metabolites, particularly phenolic compounds, such as phenols and flavonoids, with biological anti-inflammatory, antimicrobial and antioxidant properties. Scientific papers on the phenology of *P. guajava* were found less frequently than expected. However, it is a characteristic that deserves attention because it allows the development of agronomic management techniques to support crop production. Scientific studies have shown that *P. guajava* is a highly productive species of secondary metabolites under stress conditions. The production of secondary metabolites of the plant could be a useful indicator for the characterization of its production of agroindustrial and pharmaceutical importance and, therefore, would constitute a useful tool in the selection process.

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