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Fernanda Rojas Salas
School of Pharmacy, University of Costa Rica, San José, Costa Rica

Felipe González Young
School of Pharmacy, University of Costa Rica, San José, Costa Rica

María Fernanda Sánchez-Romero
School of Pharmacy, University of Costa Rica, San José, Costa Rica

Sofía Staikidis Méndez
School of Pharmacy, University of Costa Rica, San José, Costa Rica

German L Madrigal Redondo
Laboratory of Phytomarcology, Pharmaceuthical and Cosmetic Technology (LAFITEC) Institute of Pharmaceutical Research (INIFAR), Faculty of Pharmacy, University of Costa Rica, San José, Costa Rica

Corresponding Author:
Fernanda Rojas Salas
School of Pharmacy, University of Costa Rica, San José, Costa Rica

Ethnic study of traditional use of *Cuayote* (*Gonolobus taylorianus*) in Costa Rica

Fernanda Rojas Salas, Felipe González Young, María Fernanda Sánchez-Romero, Sofía Staikidis Méndez and German L Madrigal Redondo

Abstract
The *cuayote* (*Gonolobus taylorianus*) is a very unknown, endemic plant from the Central America zone. In Costa Rica, two species of the genus Apocynaceae, *Gonolobus edulis* and *Gonolobus taylorianus* are known by the popular name of *cuayote*. The fruit of both species is traditionally used for food purposes. A medicinal use of the plant and the fruit have also been reported for the treatment of warts and prostate. The fruit characterization revealed, through a phytochemical screening, the presence of important compounds such as flavonoids, steroids, triterpenes, carbohydrates and cardiotonic glycosides.

Keywords: *Cuayote*, *gonolobus taylorianus*, traditional use, ethnobotany, secondary

Introduction
The use of natural sources as medicines has been used since ancient times" (Jácome-Roca, 2017) [6] (Organización Mundial de la Salud, 2013) [11]. Natural products, such as extracts and derivatives of plants, contain secondary metabolites that provide multiple opportunities for the development of new drugs.

It is a very pertinent challenge to increase the scientific knowledge that one has regarding natural products, especially those that are of pharmaceutical interest. It is well known that the different conditions that human beings have had to face throughout history have been both diverse and changing. Due to this, the discovery of new treatments and drugs must evolve in parallel to the species.

To know the content of secondary metabolites of plants and, especially, of endemic species, allow us to have a renewable natural source of these. These compounds have a potential pharmacological value" (Payo *et al*., 1996) [12]. Obtaining metabolites from nature represents an efficient way to discover new molecules and expand the treatments available throughout the globe.

"The tropical areas of the neotropic American continent, where Costa Rica is located, harbor a greater diversity of species and ecosystems and a wider range of interactions, compared to the other tropical regions of the world" (Felice *et al*., 2011) [14]. Therefore, it is wise to say that there are many species that have not yet been explored, and that could be really useful in the field of pharmacognosy and pharmacology.

The species Gonolobus *edulis*, is native to the Central American region. In Costa Rica it is known as "*cuayote*" (Chízmar Fernandez, 2009) [3]. The research conducted on this species, in the field of Pharmacognosy is null, however it has been popularly used by the Costa Rican population.

On the other hand, the native species *Gonolobus taylorianus* is distributed from Guatemala to Costa Rica (Stevens & Montiel, 2002) [15].

This article aims to perform a validation of the traditional use of the specimen known as *Cuayote* in Costa Rica.

Methodology
The investigation was carried out in the Greater Metropolitan Area of Costa Rica. It is important to mention that multiple people recognize two species with the same name: *Gonolobus edulis* and *Gonolobus taylorianus*. Therefore, it was decided to work with *Gonolobus taylorianus*, which showed greater fruit availability during the period when this research was taking place.

In the first part of the investigation, an instrument was applied to 21 people who were working at municipal farmers' markets. The inclusion criteria were based on the following aspects: to be over 30 years old, to have lived in the Greater Metropolitan Area of Costa Rica, to have prior knowledge of the Cuayote fruit (whether to have used it or have known someone who has already use it before). With the instrument, it was sought to know the traditional use given by different people to the fruit of the plant known as Cuayote. The instrument used was of a simple type and included six questions; two related to personal data, two about the identification of the fruit (one of them showed the image of the fruit of *Gonolobus taylorianus*) and the last two on use and preparation. The questions used were:

1. Are you over 30 years old?
2. Do you live within the Greater Metropolitan Area?
3. Do you know what the fruit of the image is?
4. Have you heard about the *cuayote*?
5. For what purpose is the fruit of the *cuayote* used?
6. How is it prepared according to its use?

The second part of the investigation was related to the determination of secondary metabolites, through a phytochemical screening. To carry out this phase, it was necessary to obtain fruits from the *cuayote* plant.

Due to the availability of samples, *Gonolobus taylorianus* was used. The samples obtained were collected in San Antonio de Escazú at the coordinates 9.893140, -84.127853.

Following the collection, the identification was properly carried out by the biologist from the University of Costa Rica, Carlos Morales Sánchez.

Once the sample was identified, a procedure is done to prepare the product for its subsequent drying by lyophilization. In a subsequent step, grinding was performed.

The sample previously prepared was used to perform the phytochemical screening. In this phase, tests were carried out to witness the existence of metabolites in the extract. The metabolites to identify were:

1. Desoxy carbohydrates, through the Keller-Kiliani test.
2. Flavonoids, using the Shinoda and Wilson tests.
3. Tannins, using the ferric chloride test.
4. Carbohydrates, using the Molish and Benedict tests.
5. Steroids and triterpenes. Using the Libermann-Burchard and Salkowski tests.
6. Cardiotonic glycosides, using the Kedde and Baljet tests.

Results and discussion

The results shown were updated during the interviews, to determine the ethno botanical use of the *Cuayote* (*Gonolobus taylorianus*). Each graph shows the answers to the key questions.

The difficulty for many to differentiate between the two species is explained by the fact that the morphology of both species is similar. According to Fernández *et al*. (2008) [5], the same common name to different species of the genus *Asclepias* is given in some indigenous communities and in different regions of Mexico since they are not differentiated at a glance because both their morphology and the uses attributed to them are the same. For example *A. albicans* and *A. subulata* have the same medicinal uses and properties. In the case of Costa Rica, a similar case occurs with the species of the *Gonolobus* genus.

The results obtained during the identification of the *cuayote* fruit are presented below (see graph 1). It is evident that 67% of the interviewees correctly identified the fruit of the *cuayote*, while the remaining 33% of the interviewees were not able to determine the identity of the fruit.

This last percentage of people who did not identify the fruit is certainly low; however, it must be taken into account that the people interviewed were older than 30 years, and they have carried out work activities in municipal and farmers' markets.

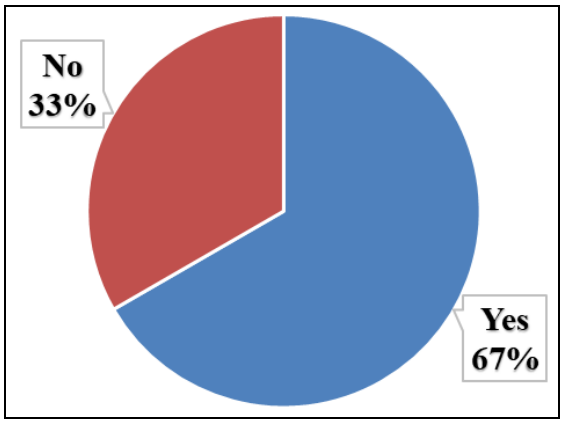

Fig 1: Cuayote identification

Own interview source conducted between Oct 11th and Oct 24th of 2017 at the pharmacognosy laboratory (FA-0229) of the University of Costa Rica.

Differently, in relation to the uses given to the fruit of the *cuayote* (see graph 2), 84% of the interviewees indicated that it is used as food and 16% of the interviewees stated that the fruit is used for medicinal purposes.

In 2012, the Costa Rican Ministry of Health, together with the Food and Agriculture Organization of the United Nations, published a document called (15 Underutilized Food of Great Value for Costa Rica). The same was carried out thanks to the Millennium Development Goals Achievement Fund and sought to provide information on different endemic species of Costa Rica, usable to achieve food sovereignty. In the document, the species *Gonolobus edulis*, known as *cuayote*, was mentioned. However, the document did not contain a nutritional description or analysis of the fruit.

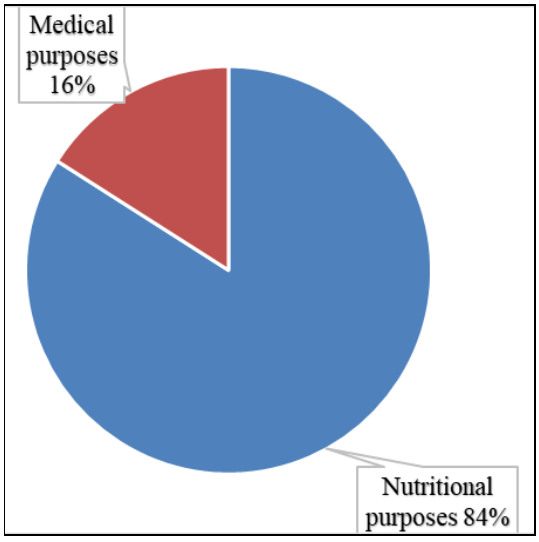

Fig 2: Uses of *cuayote*

Own interview source conducted between Oct 11th and Oct 24th of 2017 at the pharmacognosy laboratory (FA-0229) of the University of Costa Rica.

Regarding the nutritional uses of the *cuayote* fruit, see (graph 3). 52% of the interviewees mentioned that they use it as

honey, 32% of the interviewees use it as mincemeat, 8% of the interviewees consume the seeds, 4% of the interviewees use it to make soda and 4% of the interviewees eat it roasted.
The interviewees indicated some ways of making food based on the fruit of the *cuayote*. For the *picadillo*, the fruit is peeled, left in water releasing until the milk is released and cut into slices. For honey the indicated procedures are as follows:

- Peel the fruit, chop into segments, sugar and water are used. Optional: add fig leaves and syrup.
- Using *tapa dulce*. Remove the shell, boil the fruit, remove the seed and cook with honey.

Picadillo: hash
Tapa dulce*:* sugarcane
They also reported that the seeds are consumed raw, and the fruit could be ingested without being cooked, by just opening it.

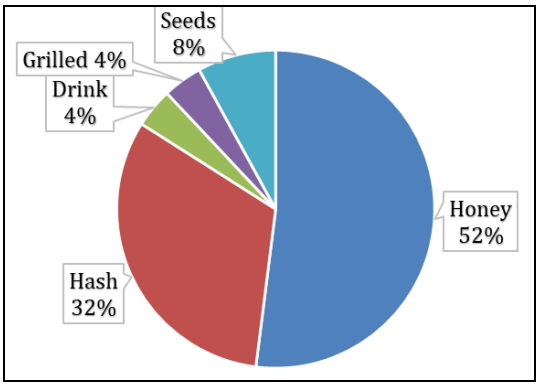

Fig 3: Culinary uses of coyote

Own interview source conducted between Oct 11th and Oct 24th of 2017 at the pharmacognosy laboratory (FA-0229) of the University of Costa Rica.
Regarding the interviewees who mentioned that they used coyote for medicinal purposes (see graph 4), information was obtained on the uses and methods of preparing the product. 50% of the interviewees indicated that it is used to eliminate warts, while the other 50% of the interviewees mentioned that it is used to prevent or treat prostate problems. They also pointed out that the preparation of the remedies varies according to their use, for the elimination of warts, the latex of the coyote fruit is used directly on the affected area and to prevent or treat prostate problems, the fruit is boiled.

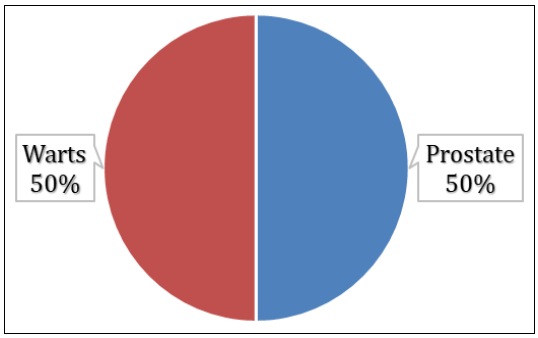

Fig 4: Medical applications of cuayote

Own interview source conducted between Oct 11th and Oct 24th of 2017 at the pharmacognosy laboratory (FA-0229) of the University of Costa Rica.
The results obtained in the graph are interesting, since the food uses and the how it is prepared match with the use of some specific examples of the Apocynaceae family in Mexico. According to Juárez *et al*. (2007) [7], even though the family is made up of toxic plants, some have been included as a condiment for regional dishes; the green fruits of *Marsdenia* and *Gonolobus* are eaten roasted; in the central depression of Chiapas, those of *Gonolobus* are preserved in syrup and their seeds, boiled or roasted, are eaten like popcorn.
From the grinding of the fruit, a total of 26.1 g of sample was obtained (Table 1). The obtained sample was macerated in methanol: ethyl ether with an 8: 2 ratio and for each gram of sample, 10 mL of the solvent was used.

Table I: Determination of the mass of the ground sample of *Gonolobus taylorianus.*

Sample	Container weight (g)	Container and sample weight (g)	Simple weight (g)
1	146,9	162,3	15,4
2	147,1	157,8	10,7
Total	__	__	26,1

Own source of grinding from the sample executed on Oct 9th, 2017. Pharmacognosy laboratory (FA-0229) of the University of Costa Rica. Pharmacognosy Laboratory (FA-0229), UCR.

Table 2: Results obtained during the phytochemical screening of the different extracts of the Gonolobus tayloranius.

Phase	Test	Compound	Result
Aqueous	Keller-Killiani	Desoxy carbohydrates	-
	Shinoda	Flavonoids	+
	Wilson		+
	Ferric chloride	Tanines	-
	Molish	Carbohydrates	+
	Benedict		+
Hexane	Liebermann-Burchard	Steroides y triterpenes	+
	Salkowski		+
Ethereal	Keller-Killiani	Desoxy carbohydrates	-
	Shinoda	Flavonoids	-
	Wilson		+
	Kedde	Cardiotonic glycosides	-
	Baljet		+

Own source of phytochemical screening executed on Nov 1st and Nov 2nd, 2017. Pharmacognosy laboratory (FA-0229) of the University of Costa Rica.
Regarding the phytochemical analysis (Table 2), the presence of the following metabolites was achieved qualitatively: Flavonoids, carbohydrates, steroids and triterpenes, and cardiotonic glycosides. The studied species belongs to the genus Asclepias L. (Apocynaceae, Asclepiadoideae). This information was provided by the botanist Carlos Morales Sánchez. The genus Asclepias, which is included within the Apocynaceae family, it is an American genus with around 150 species (Juárez Jaimes *et al*., 2007) [7].
Due to the nutritional importance and the various health benefits of these components, it is of interest that the fruit has them; flavonoids are known to have antioxidant activity, ability to fix free radicals, prevent coronary heart disease, be anti-inflammatory, hepatoprotective, among others (Kumar & Pandey, 2013) [10] (Primo Yúfera, 1995) [13].
Steroides and triterpenes usually have pharmacological action such as analgesic activity and anticancer properties and cardiotonic glycosides with anticancer activity and in cardiac muscle for cardiac diseases; however, it is not possible to indicate what effect the fruit has in particular, since it is unknown the amount of triterpenes and specific compounds these metabolites have (Akhtar *et al*., 2018) [1] (Cabildo

Miranda, 2011) [2] (Khundmiri, 2014) [8] (Koolman & Röhm, 2004) [9] (Sharapin *et al*., 2000) [14].

Conclusions and Recommendations

The analysis carried out generates knowledge to show that the Cuayote (*Gonolobus taylorianus*) is used by the Costa Rican population in a traditional way, as food and as a remedy for the treatment of warts and prostate-related problems.

Through phytochemical studies, it was elucidated that there are different metabolites such as flavonoids, carbohydrates, steroids and triterpenes, and cardiotonic glycosides. All these metabolites are of interest for drug development, so it would be a positive fact to carry out a more in-depth analysis and determine the metabolites, their chemical structures, and what biological activity they possess.

Since the population uses the fruit mainly for food purposes, it would be of interest to carry out a nutritional analysis in the future, so that useful information can be made available to people, which can help guarantee food sovereignty.

Acknowledgement

This research work was carried out with the inputs and support of the professors and staff in charge of the pharmacognosy laboratory of the School of Pharmacy from the University of Costa Rica. Professor Carlos Morales Sánchez, biologist from the University of Costa Rica, is thanked for identifying the *cuayote* fruit. Dr. Jonathan Parra is also thanked for guiding the research and being of help with Dr. Nien Tzu Weng in carrying it out. Finally, Deyanira Salas is thanked for contributing her knowledge regarding not commercially known fruits, that have great value for Costa Rican roots due to their traditional use.

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