

PROFILE OF PLANT SPECIES IN THE TROPICAL DRY FOREST OF TOLIMA (COLOMBIA) EXHIBITING ANTHELMINTIC ACTIVITY IN SHEEP

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Abstract

Phytotherapy is an area of growing scientific interest. Studies on anthelmintic bioactivity of plant species provide new alternatives to face problems such as gastrointestinal parasitism, resistance to synthetic chemical products, environmental impact, and residual activity in products of animal origin. To know the chemical profile of plants in the tropical dry forest (TDF) of Tolima with anthelmintic activity in sheep, a systematic study based on the document "The Tropical Dry Forest in Colombia", which includes 2569 plant species among native, naturalized, and exotic plants from Tolima, was conducted. By using the SCOPUS database, the academic Google search engine, and Microsoft Office 2010 Excel, each species was designated with the same criterion to identify those with anthelmintic activity and qualitative phytochemical content; then, the groups with the highest analysis were identified, and the species with simultaneous analysis were selected from the 3 chemical groups with the highest frequency, and the weight gain AND sheep search criteria was applied to the resulting group. 45 native, 6 naturalized, and 20 exotic species with anthelmintic activity were found. The Fabaceae (13 species), Amaranthaceae (4 species), Annonaceae, Apocynaceae, Euphorbiaceae, and Malvaceae (3 species each) families stand out. The three chemical groups with the high LEVELS OF PHYTOCHEMICALS were tannins, flavonoids, and alkaloids, and at a medium level saponins, steroids, and terpenoids. Simultaneous content of tannins, flavonoids and alkaloids was found in 19 native, 3 naturalized, and 12 exotic species, 5 of which present scientific report of use in sheep feed.

Keywords: bioactivity, internal parasites, secondary metabolites, plants.

Introduction

The use of regional vegetational resources for controlling or treating diseases in humans and animals has been increasing worldwide. This has been accompanied by research and the discovery of new molecules with biological activity against various diseases (carcinogenic, parasitic, bacterial, viral, fungal, metabolic disorders, etc.).

On the other hand, the context of farmed animal production presents environmental, economic, social, and commercial demands to develop proposals for obtaining safe products (meat, milk, and eggs) that are natural-environment friendly, and that generate economic benefits to producers and improve their life quality.

The starting point of the research on the use of vegetation resources for therapeutic purposes in animal health is based on documentary and ethnoveterinary studies to know both the scientific community findings on the composition and activity of plants under controlled conditions of observation and experimentation, as well as the aspects related to plant species, preparation, application, location, and origin of knowledge in the communities in the targeted region.

Once this initial part is approached, qualitative and quantitative phytochemical studies to identify and isolate fractions and biomolecules to, subsequently, develop the necessary biological tests to verify their activity can be initiated. The purpose of the present documentary study is to define the vegetal species of the tropical dry forest (TDF) of the department of Tolima with scientific anthelmintic activity in sheep.

Materials and Methods

The work was carried out in the TDF areas of the department of Tolima, and our bibliographic reference base, was Annex 1 of "The Tropical Dry Forest in Colombia", document, edited by Pizano and García which

includes 2569 plant species that include native, naturalized and exotic species.

Based on the species reported for the Tolima department, the SCOPUS database search criteria entered was the scientific name of the AND anthelmintic plant species restricted to abstracts only, with a data range from 2001 to 2017, and all kind of documents. Subsequently, to verify the results of the biological activity, the corresponding articles were reviewed.

For the group of plants with anthelmintic activity, the SCOPUS database and academic Google search criteria applied was AND phytochemical scientific name, which permitted to know the phytochemical qualitative analysis (reducing sugars, alkaloids, steroids, terpenoids, tannins, glycosides, carbohydrates, flavonoids, saponins, phenols, cardiac glycosides, and other metabolites).

By using Microsoft Office 2010 Excel, a filter was applied to select plants that, according to the report in the scientific literature, simultaneously contained the three secondary metabolites with the highest scoring peak. Finally, the list of plants resulting in the previous phases was searched on SCOPUS by plant's scientific name AND weight gain, restricted to abstracts only, with a data range of all years to this current year, and all kinds of documents criteria, and depending on the results, the word 'sheep' was also used, obtaining a list of plants reported to be used in food and anthelmintic activity.

Results and Discussion

Tropical Dry Forest Plants Associated with Anthelmintic Activity: The following are the native plants identified in the Tolima TDF with anthelmintic activity: *Trianthema portulacastrum* (Family: Aizoaceae); *Alternanthera sessilis*, *Amaranthus spinosus*, *Amaranthus viridis* and *Celosia argentea* (Family: Amaranthaceae); *Spondias mombin* (Family: Anacardiaceae); *Annona muricata*, *Annona reticulata* and *Annona squamosa* (Family:

Annonaceae); *Asclepias curassavica* (Family: Apocynaceae); *Pistia stratiotes* (Family: Araceae), *Bidens pilosa*, *Eclipta prostrata* (Family: Asteraceae); *Crescentia cujete* and *Tabebuia rosea* (Family: Bignoniaceae); *Bixa Orellana* (Family: Bixaceae); *Cordia alba* (Family: Boraginaceae); *Bursera graveolens* (Family: Burseraceae); *Evolvulus alsinoides* (Family: Convolvulaceae); *Euphorbia hirta*, *Euphorbia thymifolia* and *Hura crepitans* (Family: Euphorbiaceae); *Abrus precatorius*, *Acacia pennatula* and *Gliricidia sepium* (Family: Fabaceae); *Ocimum campechianum* (Family: Lamiaceae); *Spigelia anthelmia* (Family: Loganiaceae); *Sida acuta*, *Sida glomerata* and *Waltheria indica* (Family: Malvaceae); *Cissampelos pareira* (Family: Menispermaceae); *Ficus insípida* and *Ficus máxima* (Family: Moraceae); *Psidium guajava* (Family: Myrtaceae); *Oxalis corniculata* (Family: Oxalidaceae); *Argemone mexicana* (Family: Papaveraceae); *Phyllanthus niruri* (Family: Phyllanthaceae); *Scoparia dulcis* (Family: Plantaginaceae); *Portulaca oleracea* (Family: Portulacaceae); *Pityrogramma calomelanos* (Family: Pteridaceae), *Paullinia pinnata* and *Sapindus saponaria* (Family: Sapindaceae); *Manilkara zapota* (Family: Sapotaceae); *Cecropia peltata* (Family: Urticaceae) and *Lantana camara* (Family: Verbenaceae). In summary, 45 native plants with anthelmintic activity were found; the families with more than one plant were Amaranthaceae (4), Annonaceae (3), Euphorbiaceae (3), Fabaceae (3), Asteraceae (2), Bignoniaceae (2), Malvaceae (3), Moraceae (2) and Sapindaceae (2).

The following are the naturalized plants identified in the Tolima TDF with anthelmintic activity: *Calotropis procera* (Family: Apocynaceae); *Heliotropium indicum* (Family: Boraginaceae), *Momordica charantia* (Family: Cucurbitaceae); *Cyperus rotundus* (Family: Cyperaceae); *Senna occidentalis* (Family: Fabaceae) and *Cynodon dactylon* (Family: Poaceae). 6 naturalized plants, which belong to different families, with reports of anthelmintic activity were found.

Furthermore, the following are the exotic plants identified in the Tolima TDF with anthelmintic activity: *Anacardium occidentale*, *Mangifera indica* (Anacardiaceae); *Cascabela thevetia* (Family: Apocynaceae); *Cocos nucifera* (Family: Arecaceae); *Ananas comosus* (Family: Bromeliaceae); *Carica papaya* (Family: Caricaceae); *Bauhinia variegata*, *Caesalpinia pulcherrima*, *Cajanus cajan*, *Cassia fistula*, *Crotalaria retusa*, *Delonix regia*, *Leucaena leucocephala*, *Pithecellobium dulce* and *Tamarindus indica* (Family: Fabaceae); *Azadirachta indica* and *Melia azedarach* (Family: Meliaceae); *Mirabilis jalapa* (Family: Nyctaginaceae), *Cenchrus ciliaris* (Family: Poaceae) y *Antigonon leptopus* (Family: Polygalaceae). In summary, 20 plants were found and the families with the highest participation were Fabaceae (9), Anacardiaceae (2) and Meliaceae (2).

Main Chemical Groups in Vegetal Species: The systematic review about the content of secondary metabolites in native species shows that tannins, flavonoids, alkaloids, and phenols are the groups with the highest levels percentage in these vegetal species, while terpenoids, saponins and steroids are in a medium level (Edeoga *et al.*, 2005; Okoli *et al.*, 2009; Kumar *et al.*, 2010; Igwe *et al.*, 2010; Jayaveera *et al.*, 2010;

Valdés *et al.*, 2010; Jhade *et al.*, 2011; Bhalke & Chavan, 2011; Padhi *et al.*, 2011; Omogbai & Eze, 2011; Basma *et al.*, 2011; Olajuyigbe *et al.*, 2011; Nagarajan *et al.*, 2011; Savithramma *et al.*, 2011; Lor *et al.*, 2011; Antara, 2012; Accioly *et al.*, 2012; George *et al.*, 2012; Agrawal *et al.*, 2012; Kyei *et al.*, 2012; Akharaiyi *et al.*, 2012; Ahmed *et al.*, 2013; Souza *et al.*, 2013; Maobe *et al.*, 2013; John *et al.*, 2013; Joselin *et al.*, 2013; Deshmukh *et al.*, 2013; Gnanaraja *et al.*, 2013; Sinha, 2013; Sharmila *et al.*, 2013; Beltrán *et al.*, 2013; Naz & Bano, 2013; Kavitha *et al.*, 2014; Pizano & García, 2014; Asif *et al.*, 2014; Mondal *et al.*, 2014; Gavamukulya *et al.*, 2014; Jayanthi *et al.*, 2014; Yang, 2014; Bhuvaneshwari *et al.*, 2014; Hussain *et al.*, 2014; Atif *et al.*, 2014; Veni & Pushpanathan, 2014; Shanmugam *et al.*, 2014; Rajesh *et al.*, 2014; Jamkhande & Wattamwar, 2015; Saleem & Devi, 2015; Elufioye & Olaifa, 2015; Khanal *et al.*, 2015; Gopi, 2015; Adeyemi, 2016; Rajalakshmi *et al.*, 2016; Xuan & Khanh, 2016; Granja, *et al.*, 2016; Cruz & Cui-Lim, 2016; Kumar & Simon, 2016; Sukalingam, *et al.*, 2017; Kota, *et al.*, 2017; Rajeswari & Vijayashalini, 2017; Tang *et al.*, 2017; Ahuchaogu *et al.*, 2017; Agu & Okolie, 2017; Ma, *et al.*, 2017; Sri *et al.*, 2017; Ribeiro *et al.*, 2017; Paul *et al.*, 2017).

With regard to naturalized species, the systematic review on the content of secondary metabolites found that tannins, flavonoids, alkaloids, and phenols had the highest compositional percentage followed, at a medium level by steroids, saponins and terpenoids (Fig. 1) (Saidu *et al.*, 2011; Joshi & Kaur, 2013; Adeniyi *et al.*, 2013; Mohammad *et al.*, 2014; Chetia *et al.*, 2014; Abdillah *et al.*, 2015; Odeja *et al.*, 2015; Al-Snafi, 2016; Nandagoapalan *et al.*, 2016; Hassan *et al.*, 2017; Akindele *et al.*, 2017; Katiyar *et al.*, 2017; Kasarkar *et al.*, 2017).

In the analysis of anthelmintic activity of foreign plants, the systematization of the qualitative composition of the species shows that tannins, phenols, alkaloids, saponins and flavonoids are the metabolites with the highest analysis, followed at a medium level by terpenoids and steroids (Ademola *et al.*, 2005; Mali *et al.*, 2008; Bouzada *et al.*, 2009; Sivasankari *et al.*, 2010; Kumar *et al.*, 2010; Bhadoriya *et al.*, 2011; Antonisamy *et al.*, 2012; Kumar *et al.*, 2013; Susmitha *et al.*, 2013; Sultana *et al.*, 2013; Kalpana *et al.*, 2014; Doraiswamy *et al.*, 2014; Agrawal *et al.*, 2014; Olabinri *et al.*, 2014; Mustapha *et al.*, 2015; Koffi *et al.*, 2015; Lima *et al.*, 2015; Roopan, 2016; Monji *et al.*, 2016; Anju & Zachariah, 2016; Kousalya & Jayanthi, 2016; Modi *et al.*, 2016; Adebayo *et al.*, 2016; Chan *et al.*, 2017; Wilberforce & Olivia, 2017; Anadebe *et al.*, 2017; Sujatha *et al.*, 2017; Yadav *et al.*, 2017; Kumari, 2017; Yadima *et al.*, 2017; Cheenickal & Mendez, 2017; Hanani *et al.*, 2017; Arora *et al.*, 2017; Sravanthi *et al.*, 2017).

By jointly systematizing the content of metabolites in the three groups of plants, the metabolites with the highest levels were tannins, flavonoids, and alkaloids, while phenols, saponins, steroids and terpenoids are located at a medium level (Fig. 2). A series of studies are available for tannins, alkaloids, and flavonoids anthelmintic activity (Anthnasiadou *et al.*, 2001; Da Silva *et al.*, 2008; Kozan *et al.*, 2013). The mode of action that supports the anthelmintic effect of tannins has been discussed, but remains unidentified (Engström *et al.*, 2016). It has been suggested that the direct effect of plants is based on their interactions with vital egg proteins and larvae for biological function and development (Molan, 2014).

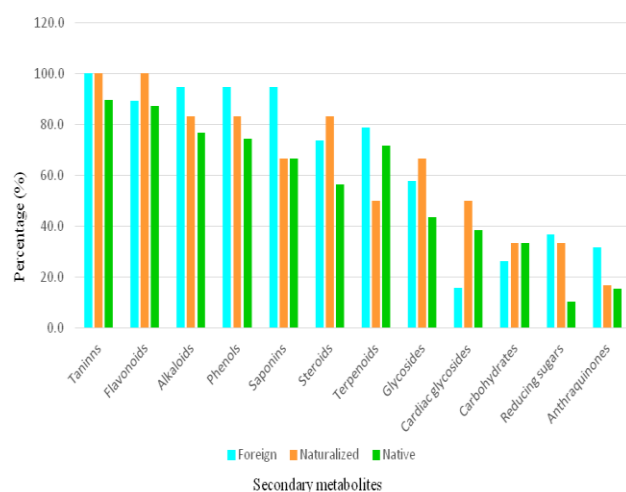


Fig. 1. Percentage content of metabolites in native, exotic, and naturalized plants with anthelmintic activity.

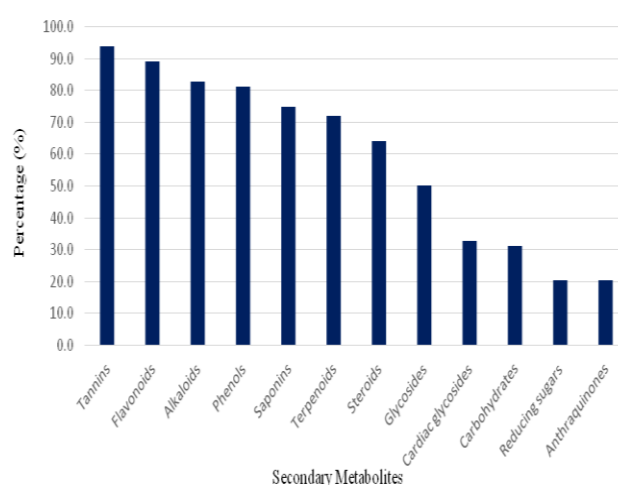


Fig. 2. Percentage consolidated content of metabolites in native, exotic, and naturalized plants with anthelmintic activity.

Table 1. Native plants with flavonoids, tannins, and alkaloids content.

Species	Family
<i>Trianthema portulacastrum</i>	Aizoaceae
<i>Amaranthus viridis</i>	Amaranthaceae
<i>Spondias mombin</i>	Anacardiaceae
<i>Annona muricata</i>	Annonaceae
<i>Annona reticulata</i>	Annonaceae
<i>Bidens pilosa</i>	Asteraceae
<i>Euphorbia thymifolia</i>	Euphorbiaceae
<i>Hura crepitans</i>	Euphorbiaceae
<i>Gliricidia sepium</i>	Fabaceae
<i>Spigelia anthelmia</i>	Loganiaceae
<i>Sida acuta</i>	Malvaceae
<i>Waltheria indica</i>	Malvaceae
<i>Psidium guajava</i>	Myrtaceae
<i>Oxalis corniculata</i>	Oxalidaceae
<i>Argemone mexicana</i>	Papaveraceae
<i>Phyllanthus niruri</i>	Phyllanthaceae
<i>Portulaca oleracea</i>	Portulacaceae
<i>Paullinia pinnata</i>	Sapindaceae
<i>Cecropia peltata</i>	Urticaceae

Table 2. Naturalized plants with flavonoids, tannins and alkaloids content.

Scientific name	Family
<i>Calotropis procera</i>	Apocynaceae
<i>Momordica charantia</i>	Cucurbitaceae
<i>Senna occidentalis</i>	Fabaceae

Table 3. Exotic plants with flavonoids, tannins, and alkaloids content.

Scientific name	Family
<i>Mangifera indica</i>	Anacardiaceae
<i>Cocos nucifera</i>	Arecaceae
<i>Ananas comosus</i>	Bromeliaceae
<i>Carica papaya</i>	Caricaceae
<i>Bauhinia variegata</i>	
<i>Cassia fistula</i>	
<i>Crotalaria retusa</i>	Fabaceae
<i>Leucaena leucocephala</i>	Fabaceae
<i>Pithecellobium dulce</i>	
<i>Tamarindus indica</i>	
<i>Melia azedarach</i>	Meliaceae
<i>Mirabilis jalapa</i>	Nyctaginaceae

In this regard, it has been reported that condensed tannins show anthelmintic activity. Inhibition of the larval viability of *Haemonchus contortus*, *Teladorsagia circumcincta* and *Trichostrongylus vitrinus* was observed when these species were subjected to the action of *Schinopsis spp.* whose extract contained 73% condensed tannins (Hoste *et al.*, 2006). The same extract reduced the burden of sheep eggs and worms with monospecific infection of *H. contortus* (Max *et al.*, 2007). Flavonoids belong to the largest group of secondary metabolites (> 6,000 identified) and widely distributed (Hoste *et al.*, 2006; Max *et al.*, 2007) found in almost all photosynthetic plants. A study (Kozan *et al.*, 2013) showed for the first time that flavonoid glycosides (luteolin-7- β -O-glucopyranoside and quercetin-3-O- β -glucopyranoside) of the acetic extract of *Vicia pannonica* Crantz. var. *Purpurascens* possess a significant *In vitro* anthelmintic activity against *Trichostrongylus sp.*, There is also evidence on the activity of luteolin and quercetin in combination with condensed tannins on the molt of L₃ larvae of *H. contortus* (Klongsiriwet *et al.*, 2015).

Regarding alkaloids, it has been reported that some isolated alkaloids were highly toxic to HL60 cells in tissue culture, such that those with a high RM (relative mobility) 50/IC (inhibitory concentration) 50 (selectivity index: SI > 100) were proposed as anthelmintic alkaloid molecules such as alocriptopine, dehydrocordalina and papaverine. However, the inhibition mechanism of larval motility is unclear so far (Hrckova & Velebny, 2012). Other researchers have found antihelminthic activity with molecules from this group such as dicentrin, isoquinoline alkaloid that showed biological activity *In vitro* in larvae of *H. contortus* (Ayers *et al.*, 2007). The extract of *Spigelia anthelmia* showed strong anthelmintic activity against *H. contortus* and the chemical study of this extract revealed as one of the main constituents of spigantine alkaloid (Morais *et al.*, 2002).

Finally, as a result of applying the filter to native plants with anthelmintic activity, which simultaneously contain flavonoids, tannins and alkaloids, a total of 19

plants were obtained, which are listed in (Table 1). For the naturalized plants, only 3 plants were obtained (Table 2) and for the exotic plants the same filter yielded 12 plants (Table 3).

Plants with anthelmintic activity used as food source:

A total of 5 plants were obtained of which 2 native (*Gliricidia sepium* and *Psidium guajava*), 1 naturalized (*Calotropis procera*), and 2 exotic (*Leucaena leucocephala* and sweet *Pithecellobium*) plants have been reported as being used in animal feeding activities. For the specific case of sheep, *Leucaena leucocephala*, *Gliricidia sepium*, *Calotropis procera* and sweet *Pithecellobium* species have also been evaluated as a food source (De Azevêdo *et al.*, 2010; Archimède *et al.*, 2010; Díaz *et al.*, 2013; Andrade *et al.*, 2014).

Conclusions

A considerable number of Tolima TDF plants, among native, naturalized and introduced plants, have anthelmintic activity. The families with the highest number of plants shown in this report are Fabacea, Amaranthaceae, Annonaceae and Euphorbiaceae.

The phytochemical profile of the plants with scientific anthelmintic activity shows that tannins, flavonoids, and alkaloids are secondary, predominant metabolites in these vegetal species.

Although antihelmintic activity of the three metabolite groups has been found, greater knowledge and understanding of the mechanisms of action involved is required.

The following are the plants with anthelmintic activity used in animal feed: *Gliricidia sepium*, *Psidium guajava*, *Calotropis procera*, *Leucaena leucocephala* and *Pithecellobium dulce*, constituting species with potential nutraceutical application in sheep.

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