

Phytochemical Analysis of Three Common Medicinal Plants (*Gliricidia sepium*, *Melothria pendula*, and *Pithecellobium dulce*) in the Philippines

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Abstract

Original Research Article

Three common medicinal plants in the Philippines, *Gliricidia sepium* (madre de cacao), *Melothria pendula* (wild pipino), and *Pithecellobium dulce* (Camachile), were studied to determine their phytochemical components. Healthy, matured leaves of which were collected from Piat, Cagayan, thoroughly washed, air-dried, and then subjected for ethanolic extraction. Phytochemical analysis showed that *G. sepium* contains coumarins, saponins, steroids (cardiac glycosides), tannins, and terpenoids. *M. pendula*, on the other hand, possesses alkaloids, anthraquinones, quinones, saponins, steroids, tannins, terpenoids, and xanthoproteins. *P. dulce*, meanwhile, has alkaloids, anthraquinones, coumarins, quinones, saponins, steroids, tannins, terpenoids, and xanthoproteins. These secondary metabolites present on the leaf extract of the aforementioned plants have various pharmacological properties such as analgesic, anti-diarrheal, anti-inflammatory, and antimicrobial. This is the reason that these plants were widely used in the local community for the treatment and immediate remedy for common ailments such as fever, loose bowel, and simple wounds.

Keywords: *Gliricidia sepium*; *Melothria pendula*; *Pithecellobium dulce*; Phytochemical analysis; Philippines.

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INTRODUCTION

Traditional medicine is the sum of the knowledge, skill, and practices based on the theories, beliefs, and experiences indigenous to different cultures, used in the maintenance of health as well as in the prevention, diagnosis, improvement or treatment of physical and mental illness. The World Health Organization (WHO) recognizes traditional medicine as one of the resources of primary health care services that could contribute to improved health outcomes, including those in the Millennium Development Goals [1].

Natural products have been the backbone of traditional system of healing throughout the globe. Plants especially those with ethnopharmacological uses have been the primary sources of medicine for early drug discovery [2]. In a work of Mandal et al., 2005 as mentioned in a study of Vaghasiya et al., 2011, extraction and characterization of several active phytocompounds from these green factories have given birth to some high activity profile drugs [3].

The present review considers the background of interest in the use of plants and their extracts as

alternative performance enhancers, and the non-nutrient bioactive compounds of plants. Secondary metabolites have shown to possess various biological effects, which provide the scientific base for the use of herbs in the traditional medicine in many ancient communities [4]. Various studies have shown that many plants are rich source of antioxidants such as vitamins A, C, E, and phenolic compounds such as flavonoids, tannins, and lignins, found in plants [5]. Phytochemicals are bioactive non-nutrient plant compounds that have protective or disease preventive property [6].

Many plants in the Philippines are known for their folkloric uses in various health conditions, which include *Gliricidia sepium*, *Melothria pendula*, and *Pithecellobium dulce*. Plant based antimicrobials has enormous therapeutic potential as they can serve the purpose with lesser side effects that are often associated with synthetic antimicrobials. The phytochemical analysis of the plants is very important commercially and has great interest in pharmaceutical companies for the production of the new drugs for curing of various diseases [7]. The study aimed to determine the phytochemical components of the three plants as with

adequate antibacterial efficiency can be used for the treatment of bacterial infections.

MATERIALS AND METHODS

MATERIALS

This study made use of the following materials in the laboratory: beakers, conical flasks, filter paper discs, glass test tubes, and pulverizer. It also made use of assorted chemical reagents such as acetic anhydride, chloroform, concentrated sulfuric acid (H₂SO₄), diluted ammonia (NH₃) solution, Dragendroff's reagent, ethyl alcohol (80% concentration), lead acetate (1% concentration), nitric acid (HNO₃), sodium hydroxide (NaOH), and distilled water as the diluent.

Equipment and Apparatus

This study utilized the use of the following equipment and apparatus in the laboratory: digital weighing scale, incubator, platform shaker, and sample concentrator.

Procedure

Collection of plant materials

The leaves of *G. sepium* (madre de cacao), *M. pendula* (wild pipino), and *P. dulce* (Camachile) were harvested from a farm in Piat, Cagayan. The leaves which were green in color, healthy, matured, and were located near the stem were chosen for the extraction procedure.

Drying of Plant Samples

The collected leaves of the medicinal plants were thoroughly washed with distilled water to remove any impurity present from their surfaces. They were then subjected to natural drying (air-drying) at room temperature for two weeks.

Ethanolic Extraction

Twenty grams of the air-dried leaves of the medicinal plants were pulverized separately. The pulverized leaves were then soaked in 80% ethyl alcohol. The solid particles were removed and the resulting solution was filtered using a cheese cloth and a sterile gauze pad. The solution was placed in an incubator with 300 cps for two hours.

Residual ethyl alcohol in the solution was made to evaporate by immersing the solution in a water bath for four hours at +80C until a negative result is obtained. A negative result can be confirmed by conducting a flame test on the solution by using a sample concentrator.

Phytochemical Analysis

Test for Alkaloids: To 2 ml of extract, 2 ml of concentrated hydrochloric acid was added. Then 1 ml of Dragendroff's reagent were added. Presence of orange or red precipitate produced immediately indicates the presence of alkaloids.

Test for Combined Anthraquinones: To 5 ml of the extract, hydrolyzed it with concentrated sulfuric acid extracted with benzene, then add 1 ml of ammonia. A rose-pink coloration suggests the presence of anthraquinones.

Test for Coumarins: To 2 ml of the extract, add 3 ml of 10% sodium hydroxide. Observe for the formation of a yellow color that indicates the presence of coumarins.

Test for Flavonoids: To 1 ml of the extract, add a few drops of diluted sodium hydroxide. An intense yellow color was produced that turns colorless upon the addition of a few drops of diluted acid. This indicates the presence of flavonoids.

Test for Quinones: About 1 ml of the extract was added with 2 ml diluted sodium hydroxide. The formation of a blue green or red coloration indicates the presence of quinones.

Test for Saponins: 2 ml of extract and 2 ml of distilled water were added together and shaken in a graduated cylinder for 15 minutes. It resulted in the formation of 1 cm layer of foam that indicated the presence of saponins.

Test for Steroids: About 1 ml of the extract was dissolved in 10 ml chloroform and an equal volume of concentrated sulfuric acid was then added through the sides of the test tube. The upper layer turns red while the sulfuric acid layer shows a yellow with green fluorescence which indicates the presence of steroids.

Test for Tannins: To 2ml of extract, a few drops of 1% lead acetate was added. The formation of yellow precipitates indicates the presence of tannins.

Test for Terpenoids: About 2 ml of the extract was treated with 2 ml acetic anhydride. A few drops of concentrated sulfuric acid were then added to the solution and the formation of blue green rings was observed that indicates the presence of terpenoids.

Test for Xanthoproteins. To 1 ml of the extract, add a few drops of concentrated nitric acid and ammonia solution. The formation of reddish orange precipitates indicates the presence of xanthoproteins.

RESULT AND DISCUSSION

Table-1: Results of the Phytochemical Screening of the Leaf Ethanolic Extracts of *Gliricidia sepium* (madrede cacao), *Melothria pendula* (Wild pipino) & *Pithecellobium dulce* (Camachile)

Secondary Metabolite	Ethanolic Extracts		
	<i>G. sepium</i>	<i>M. pendula</i>	<i>P. dulce</i>
Alkaloids	-	+	+
Carotenoids	-	-	-
Combined Anthraquinones	-	++	-
Coumarins	+	-	++
Flavonoids	-	-	-
Quinones	-	++	-
Saponins	+++	++	++
Steroids	+	+	++
Tannins	+++	+	++
Terpenoids	+++	+	++
Xanthoproteins	-	+	+

The qualitative phytochemical screening of the ethanolic leaf extracts of *G. sepium*, *M. pendula*, and *P. dulce* exhibits the presence of various secondary metabolites such as saponins, steroids, tannins, and terpenoids and the absence of carotenoids and flavonoids. *M. pendula* and *P. dulce* exhibits the presence of alkaloids and xanthoproteins while *G. sepium* exhibits the absence of these metabolites. *M. pendula*, on the other hand, exhibits the presence of anthraquinones and quinones while *G. sepium* and *P. dulce* do not exhibit these two metabolites.

Many literatures claim that the secondary metabolites contain several pharmacological activities which are responsible why many plants containing these metabolites are widely used in various parts of the world, including the Philippines, for medicinal purposes. *G. sepium*, *M. pendula*, and *P. dulce* are traditionally used in the locality for the treatment, or at least as a first-aid regimen for various kinds of ailments.

In the study of Tatengco & Jacinto 2015 (as cited in Abdulaziz *et al.*, 2019), the decoction and poultice preparation of the *G. sepium* leaves are used in the Philippines for the treatment of pruritic ailments and skin infections and as an antipyretic for the relief of fever. It has an antibacterial property against *Bacillus subtilis* and tends to inhibit the growth of the fungus *Candida albicans* [8]. Akharaiyi 2012 (as mentioned in Abdulaziz *et al.*, 2019) further mentions that it is antibacterial against *Bacillus cereus*, *Enterococcus faecium*, *Escherichia coli*, *Klebsiella pneumoniae*, *Proteus mirabilis*, *Pseudomonas auroginosa*, *Serratia marcescens*, *Staphylococcus aureus*, and *S. typhimurium* [8].

Not much is known about *M. pendula*. However, Arseniuk describes that the folkloric uses of the leaves of *M. pendula* include as a purgative. *M. pendula* leaf decoction is also used to clean wounds and burns as well as serve as a diuretic. Meanwhile, the

whole part of the plant when crushed is generally used as an anti-inflammatory [9].

On the other hand, folkloric uses of the leaves of *P. dulce* include the treatment for leprosy, intestinal disorders, peptic ulcers, toothache, earache, emollient, abortifacient, and larvicidal. When used as a plaster, the leaves can relieve pain due to venereal sores and also act as anticonvulsants, and when taken with salt, can cure indigestion based on the study of Sunarjono & Coronel [10]. It is also used as an anti-helminthic and for lowering the blood cholesterol levels (hypolipidemic activity) [11].

Basically, all parts of *P. dulce* possess pharmacologically active substances and so are used for various medicinal purposes. According to the studies of Sukantha *et al.*, [12], Megala & Geetha [13], Manna *et al.*, [14], & Mule *et al.*, [15], as mentioned by Geeta *et al.*, all parts of the plants are used as an antidiabetic, anti-inflammatory, antibacterial, anti-dysentery, anti-mycobacterial, anti-convulsant, anti-ulcer, anti-diarrheal, anti-fungal, and anti-oxidative [16].

Alkaloids

Alkaloids demonstrate various pharmacological activities such as analgesia, as a local anesthesia, hypotensive properties, and muscle relaxation [17]. This information may explain why *M. pendula* is widely traditionally employed for the treatment of inflammation in the Philippines including the treatment of burns and wounds while *P. dulce* is used to treat ailments like convulsions, diabetes, inflammatory conditions, and intestinal disorders (anti-diarrheal, anti-dysentery, and indigestion).

Coumarins

Coumarins are found in many plants. One of the most important biological activities of this secondary metabolite is anti-inflammatory [18]. This information explains why *P. dulce* is commonly used for the

treatment of infections caused by certain microorganisms, for intestinal disorders, peptic disorders, and other inflammatory conditions.

Quinones and Combined Anthraquinones

Quinones particularly anthraquinones display a variety of biological and pharmacologic activities. Among of these are its anti-bacterial (especially on the acid-fast bacillus *Mycobacterium*) and anti-fungal properties. Anthraquinones are being divided into two types namely, alizarin and emodine. The emodine type is known to have antibacterial properties [19]. The presence of this metabolite may explain why the decoction of *M. pendula* leaves are primarily used to dress burns and wounds and the crushed parts used as an anti-inflammatory agent owing to its ability to inhibit the growth of both bacteria and mycotic organisms.

Saponins

Saponins tend to limit the inflammatory process, help in decreasing hypercholesterolemia, fights off certain fungi (antifungal), parasites (anti-protozoan), and viruses (anti-viral), and aids in treating for typhoid, hemorrhoids, and impetigo [8]. This may explain the antimicrobial activities of *G. sepium* and *P. dulce* and the activity of *P. dulce* against intestinal disorders like diarrhea, dysentery, indigestion, and peptic ulcer as well as against aches, convulsions, and even diabetes. This may also explain the anti-inflammatory property of *M. pendulum* and why it is used to wash wounds and burns to avoid further infection.

Steroids

Steroids display several biological and pharmacological activities. Some of these are antibacterial, anti-fungal, and anti-inflammatory [20]. The presence of this metabolite may explain why *G. sepium* is used to treat bacterial and fungal infections in the local community; *M. pendula* for burns, wounds, and other inflammatory conditions; and *P. dulce* for bacterial, fungal, and viral infections and inflammatory conditions. The steroids like cardenolides, cucurbitacins, saponinins (steroid saponins), and steroidal alkaloids are attributed to produce these kinds of pharmacological actions [20].

Tannins

Tannins are an example of a phenolic compound known to exhibit a lowering activity on blood pressure and tend to decrease the serum lipid concentrations [21]. This explains the role of *P. dulce* in the local community as an alternative treatment for diabetes, which is a disorder characterized by an increased blood glucose that is directly associated with hypertension. However, there are no reports that *G. sepium* and *M. pendula*, both containing the same metabolite, are being used for the same folkloric purpose.

Terpenoids

Terpenoids possess anti-bacterial activity as well as anti-viral, anti-spasmodic, and anti-diabetic properties aside from its immune-modulatory properties, among many others [21]. This may be the reason why *G. sepium* is used as an anti-microbial, *M. pendula* to treat wounds, burns, and inflammation, while *P. dulce* is used as an anti-microbial, in treating convulsions, diabetes, pains (earaches and toothaches), and several intestinal disorders like diarrhea or dysentery and indigestion.

Xanthoproteins

Xanthoproteins are known to have anti-bacterial activity more importantly on Gram-negative bacteria like *Escherichia coli* than Gram-positive bacteria like *Staphylococcus aureus*. These secondary metabolites also possess varying antifungal activity against *Aspergillus niger*, *Candida albicans*, and *Epidermophyton floccosum* [22]. Aside from this, these secondary metabolites are also active against other organisms like *Micrococcus luteus*, *Pseudomonas aeruginosa*, and *Salmonella abony* as well as fungi like *Aspergillus niger*, *Candida albicans*, *C. krusei*, *Mucor* spp., and *Rhizopus oryzae* [23]. This probably explains why the presence of these secondary metabolites in *M. pendula* and *P. dulce* make these two plants widely considered by the community folks as an antibiotic treatment to bacterial and fungal infections.

CONCLUSION

The three medicinal plants found in the Philippines and are locally used in the community for the treatment of various ailments can be attributed to the presence of various biologically and pharmacologically active secondary metabolites found in their leaves.

G. sepium is traditionally used in the country as a regimen against bacterial and fungal infections. The anti-bacterial, anti-fungal, and anti-inflammatory properties of *G. sepium* may be due to the presence of active metabolites such as coumarins, saponins, steroids, and terpenoids.

M. pendula is also used in the local community as a purgative and for dressing of burns and wounds. The plant is generally used as an anti-inflammatory. The medicinal values of this plant owes to the fact that it exhibits alkaloids, quinones and anthraquinones, saponins, steroids, terpenoids, and xanthoproteins.

Lastly, *P. dulce* is widely used in folkloric medicine in the country as an anti-bacterial, anti-fungal, anti-viral, and anti-inflammatory. It is also used to treat diabetes, intestinal disorders like diarrhea, dysentery, and even peptic ulcer and is also recommended as an anti-convulsant. The medicinal uses of this plant can be attributed to the presence of active substances like alkaloids, coumarins, quinones and anthraquinones, saponins, steroids, tannins, terpenoids, and xanthoproteins.

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