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The comparative studies of phytochemical screening of *Piliostigma thonningii* root and leaves extract

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ABSTRACT

Nature was and will continue to be the source and lead for all medicinal drugs. The present study evaluates with the qualitative analysis of Aqueous, Chloroform, Ethyl acetate, Methanolic and N-Hexane extracts of various parts of Piliostigma thonningii. These parts (Leaves and root) of Piliostigma thonningii were extracted by cold percolation method using organic solvents such as Aqueous, Chloroform, Ethyl acetate, Methanol and Hexane. The various parts' extracts of the plant reveal alkaloids, flavonoids, glycosides, reducing sugars, resins, saponnins and tannins. P. thonningii is known as" Kargo or kalgo" in Hausa, is a tree which has many uses as its various parts are employed in traditional medicine, and modern medical research has found that it has many beneficial properties too.

Keywords: *Piliostigma thonningii*, qualitative analysis, Kargo, N-Hexane

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INTRODUCTION

Medicinal plants play a significant role in providing primary health care services to rural dwellers and are used by about 80% of the marginal communities around the world [9, 14, and 16]. Each medicinal plant species has its own nutrient composition besides having pharmacologically important phytochemicals. These nutrients are essential for the physiological functions of human body.[19] Such nutrients and biochemicals like carbohydrates, fats and proteins play an important role in satisfying human needs for energy and life processes [13]. Phytochemicals'studies have attracted the attention of plant scientists due to the development of new and sophisticated techniques. These techniques play a significant role in giving the solution to systematic problems on one hand and in the search for additional resources of raw materials for pharmaceutical industry on the other hand. Plant synthesizes a wide variety of chemical compounds, which can be sorted by their chemical class, biosynthetic origin and functional groups into primary & secondary metabolites. Knowledge of the chemical constituents of plants is desirable, not only for the discovery of therapeutic agents, but also because such information be of value in disclosing new resources of such chemical substances. [10]

Piliostigma thonningii schum is an underexplored leguminous plant that belongs to the family of *caesalpiniacea*, a family that comprises of trees, shrubs or very rarely scramblers, distributed in Africa and Asia in open woodlands and savannah regions that are moist as well as wooded grasslands in low to medium altitudes [6, 11]. The fruit is hairy, hard and flattish pod, which turns rusty brown, woody, twisted and splits at ripening and usually persistent on the tree between June and September [3, 4]. *P. thionningii* has been reported in literature to have age-long folkloric use in traditional medicine, especially in the treatment of malaria fever, wounds, ulcers, gastric/heart pain, gingivitis and as an antipyretic. According to the traditional healers in Doila, this plant is called child remedy as it is mainly used as a remedy for children except its use against arthritis, headache, hemorrhoids and backache [1, 3].

MATERIALS AND METHODS

Plant Materials

Fresh Leaves and root of *P.thoninngii* were collected from the outskirt of Mashi town in Mashi Local Government area of Katsina State, Nigeria. The plant materials were collected and air dried. These plants were identified by Prof. B. S. Aliyu of the Biological Science Department, Bayero University, Kano, Nigeria, where the voucher numbers of the plants were submitted.

Preparation of Extracts

The Leaves and root-bark of *P.thoninngii* were air-dried and pounded into fine powder. The powder of these parts of the plant (200 g) were percolated with 750 ml of methanol at room temperature for two weeks, and then filtered. Maceration of the concentrates were carried out using different solvents in other of their polarity, starting with N-Hexane, then Chloroform, Ethyl acetate, Methanol while ending with Water.

Chemicals and drugs

All the chemicals and solvents were standard and of analytical grade.

Phytochemical Screening:

The fractions of various solvents of the Leaves and root of *P.thonningii* were subjected to preliminary phytochemical screening, to identify the secondary metabolites present. The methods of analysis employed were those described by Brian and Turner (1975) [3].

Alkaloids:

A quantity (3 ml) of concentrated extract was taken into a test tube and 1 ml HCl was added the mixture was heated gently for 20 min cooled and filter, the filtrate was used for following test.

a) Wagner test: Filtrate was treated with Wagner's reagent; formation of brown reddish precipitate indicates presence of alkaloids.

b) Hager's test: Filtrate was treated with Hager's reagent, presence of alkaloids confirmed by the yellow colored precipitate.

Cardial Glycosides:

Keller-Killani Test: Plant extract treated with 2 ml glacial acetic acid containing a drop of $FeCl_3$. A brown colour ring indicates the presence of positive test.

Flavonoids:

Shinoda test: The presence of flavonoids was estimated by Shinoda. The extracts were treated with few drops of concentrated HCl and magnesium ribbon. The appearance of pink or tomato red colour within few minutes indicated the presence of flavonoids.

Alkaline reagent test: The extracts were treated with few drops of diluted sodium hydroxide (NaOH) separately. Formation of intense yellow color which turned colorless on addition of few drops of diluted HCl indicated presence of flavonoids.

Reducing Sugars:

5ml of the extracts was diluted with distilled water. Fehling solutions A & B were added and the mixture warmed. The brick red precipitate at the bottom of the test tube indicates reducing sugars.

Resins:

10ml of extracts was obtained in a test-tube, the same amount of cupper acetate solution was added and the mixture was shaken vigorously and allowed to separate, a green colour indicates the presence of resin.

Saponin:

5 ml extract was mixed with 20 ml of distilled water then agitated in graduated cylinder for 15 min formation of foam indicates Saponin.

Steroid:

1ml extract was dissolved in 10 ml of chloroform & equal volume of concentrated H_2SO_4 acid was added from the side of test tube .The upper layer turns red and H_2SO_4 layer showed yellow with green fluorescence .This indicates the presence of steroid.[18]

Tannin:
2ml extract was added to 1% lead acetate a yellowish precipitate indicates the presence of tannins.

Table 1: Physical Characteristics of the Various Fractions Leaves and root bark of *P. thoninngii*

Piliostigma thoninngii (Parts)	Solvents	Texture	Colour
Roots	N-Hexane	Solid	Violet-red
	Chloroform	Solid	Violet-red
	Ethylacetate	Solid	Deep-red
	Methanol	Solid	Deep-red
	Water	Solid	Reddish
Leaves	N-Hexane	Oily	Dark green
	Chloroform	Oily	Dark green
	Ethylacetate	Oily	Green
	Methanol	Oily	Green
	Water	Oily	Green

Table 2: Phytochemical Analysis of Leaves extract of *P. thoninngii* Linn

S/N	SECONDARY METABOLITES	A.L.E	C.L.E	E.L.E	M.L.E	H.L.E
1	ALKALOIDS					
	a. Wagner's reagent	+	-	+	+	+
	b. Hager's reagent	+	-	+	+	+
2	FLAVONOID					
	a. Alkaline reagent test	+	+	-	+	-
	b. NH_4OH test	+	+	-	+	-
3	REDUCING SUGAR					
	a. Fehling Solution A&B	NT	NT	NT	NT	NT
4	TANNINS					
	a. Lead Acetate test	+	+	-	+	-
	b. $FeCl_3$	+	+	-	+	-
5	CARDIAC GLYCOSIDES	-	+	+	+	+
6	SAPONINS	+	+	-	+	-
7	STEROIDS	-	-	-	-	-
8	RESINS	-	-	-	-	-

A.L.E.-Aqueous Leaves Extract, C.L.E.-Chloroform Leaves Extract, E.L.E.-Ethyl acetate Leaves Extract, H.L.E- Hexane Leaves Extract, M.L.E-Methanol Leaves Extract
Note: (+) = Present, (-) = Absent

Table 3: Phytochemical Analysis of Root extract of *P. thonningii* Linn

S/N	SECONDARY METABOLITES	A.R.E	C.R.E	E.R.E	M.R.E	H.R.E
1	ALKALOIDS					
	a. Wagner's reagent	+	-	+	+	+
	b. Hager's reagent	+	-	+	+	+
2	FLAVONOID					
	a. Alkaline reagent test	+	+	-	+	-
	b. NH_4OH test	+	+	-	+	-
3	REDUCING SUGAR	NT	NT	NT	NT	NT
	a. Fehling Solution A&B					
4	TANNINS					
	a. Lead Acetate test	+	+	-	+	-
	b. $FeCl_3$	+	+	-	+	-
5	CARDIAC GLYCOSIDES	-	+	+	+	+
6	SAPONINS	+	+	-	+	-
7	STEROIDS	-	-	-	-	-
8	RESINS	-	+	-	-	-

A.R.E.-Aqueous Root Extract, C.R.E.-Chloroform Root Extract, E.R.E.-Ethylacetate Root Extract, H.R.E-Hexane Root Extract, M.R.E- Methanol Root Extract
Note: (+) = Present, (-) = Absent, NT- Not tested.

RESULTS AND DISCUSSION

Table 2 shows the result of Phytochemical screening of the Leaves of *Piliostigma_thonningii* Linn. Preliminary Phytochemical investigation of the Aqueous, Chloroform, Ethylacetate, Methanol and Hexane extracts of the Leaves of the plant *Piliostigma thonningii* Linn were compared. Alkaloids were present in all the solvents' extracts except Chloroform extract, Cardiac glycosides were evident in Chloroform, Ethyl acetate, N-Hexane, Methanol extracts of the Leaves of the plant except the aqueous extract. It also shows that Tannins were present in Aqueous, Chloroform and Methanol soluble extracts only. Cardiac Glycosides were only evident in Chloroform and Hexane extracts whereas Resins and Steroids were absent in all the compared extracts of the plants.

Table 3 shows the result of Phytochemical screening of extract of root bark of *Piliostigma thonningii* Linn. Preliminary Phytochemical investigation of Aqueous, Chloroform, Ethyl acetate, Methanol and Hexane extracts of the root bark of the plant were compared. Reducing sugars were not tested for, Alkaloids were present in all the solvents' extracts compared except in the Chloroform root bark extract of *P. thonningi whic*h it is evidently absent. Cardiac Glycosides were present in all the solvents' extracts compared except in Aqueous root bark of the plant which it is glaringly absent. Tannins, Saponins and Flavonoids were present in all but absent in Ethyl acetate and methanol extracts of *P. thonningi.* Resins were observed only in Chloroform extract of the plant's root bark but Steroid was evidently absent in all the compared Solvents' extracts of *P. thonningi's* root bark.

Present study deals with qualitative analysis of Leaves and root bark extracts of *P. thonningi* Linn, These parts reveal Tannins, Alkaloids, Saponins and Cardiac glycosides which has been earlier confirmed by Mustapha A.J, *et al* [12] and the root rich in Tannins have been validated by Tshisikhave M.P, *et al.* [17] On the basis of these data researcher can easily isolated particular metabolite from the Leaves and root extract quantitatively.

CONCLUSION

It is concluded that in the present study, the root of *P. thonningii* Linn contains Resins in Chloroform extract whereas the Leaves of the same plant reveals no presence of it. Both parts of the plant reveal that Methanolic extract contains more number of phytochemicals than the remaining organic solvents' extracts whereas no presence of steroid was observed in the Leaves and root of *Piliostigma thonningii* Linn.

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