

IN VITRO PHYTOCHEMICAL ANALYSIS AND ANTHELMINTIC ACTIVITY OF ACACIA NILOTICA WHOLE PLANT METHANOLIC AND AQUEOUS EXTRACT ON INDIAN ADULT EARTHWORM

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Abstract

Keywords: Anthelmintic activity, *Acacia nilotica*, *Pheretimaposthuma*, methanol extract, aqueous extract, phytochemical analysis

There are various beneficial effects of *Acacia nilotica* since the ancient time is being reported. But in the project work anthelmintic activity against *Pheretimaposthuma* (an Indian earthworms) is being studied by applying two different concentrations of plant methanolic extract. The result is then compared with standard drug Albendazole and control group. The pharmaceutical constituent of *Acacia nilotica* is being also studied here.

Introduction

There are number of deadly disease troubling humans every now and then. There are various medicinal plant is being used since the ancient time when there is no availability developed medical science *Acacia nilotica* is very much efficient against the various pathogens and having numerous therapeutic activity^[1]. Several diseases like, piles, tumor, wounds, parasitic infection is being cured by the root bark of the plant^[2]. Helminthiasis caused by various types of worm, which also may lead to burrowing our internal organs. Helminthic infections spread through the eggs and larvae of worms through contaminated food and is one of the most widespread infection of human, a huge population of the world is being infected by this infection. It is one of the most hazardous to human health. According to WHO the synthetic drugs used against this worm having various side effects^[3-6].

Recently helminthiasis has been a tedious thread to the world population, in order to overcome this problem, we have to think in a different way to produce efficient anti-helminthic agent from the ancient medicine^[7]. The population of tropical developing countries totally depends on developing herbal medicinal products as it very much precious to rural people and also contribute to global society finds of new drugs. Several plants showing the helminthic activity identified by regular investigation and research work. The potential is being tested various plants, including piper. Sp, ficusbenghalensis, *Alpinia nigra* etc^[8]. The aim and objective of this project work are to investigate the phytochemical and anthelmintic potent of different extracts of *Acacia nilotica* leaves and flower applied on Indian adult earthworm

Materials and Methods

Material

Against the most the common disease *Acacia nilotica* is being used by a number of villages and tribal people. Depending on that observation the *Acacia nilotica* is chosen for experimental tools. From Purba Medinipur district in the month of February, 2020 the sample is collected, (Latitude- 22°22'46" north, Longitude- 87°42'15" east, Altitude- 5.32 meters from mean sea level), west Bengal, India and it is available in throughout the year.

Experimental Worms

Various species of roundworm, parasite of human is very much physiologically similar and resemblance anatomically with the *Pheretima posthuma*. Different beaker with different concentration is taken and two similar size and shape in of earthworm place in it. After carefully observing the paralysis time and death Time of worm are noted down. The tasted result is being compared with the standard anthelmintic drug Albendazole (100 mg/ml).

Preparation of Extract

The powder of leaves and flower of *Acacianilotica* is made by drying under shade and crushed in electric blender, then subjected to Soxhlet extraction (Continuous hot extraction) by using methanol and water as solvent. Then by using rotary evaporator the methanolic and aqueous extract is concentrated and then checked the anthelmintic activity.

Albendazole Administration

During this study the result of various concentrations of methanol and aqueous plant extract and compared with the standard drug Albendazole (100 mg/ml).

Extract Administration

Two different concentration of extract i.e. methanolic and aqueous (50, 100 mg/l) was prepared. Albendazole was used as standard drug. The different concentration beaker was taken and earthworm of similar shape and size placed in the beaker^[9-10].

Experimental Design

According to standard method the anthelmintic activity would perform^[11]. Various species of roundworm, parasite of human similarities with *Pheretimaposthuma* in bath physiologically and anatomically. Several beakers with various concentrations was taken and place two earthworm with similar size and shape into each beaker. Then paralysis Time and death time being noted by carefully observing.

Phytochemical Analysis

According to the standard method the phytochemical constituents of *Acacia nilotica* is being carried out^[12]. To know which type of phytochemicals are present the following tests were done-

Alkaloid test

Crude extract it *has taken* and 2 ml Wagner's reagents is added in it. If a red - brown precipitate is formed, then alkaloids is present.

Flavonoid

A portion of crude extract is taken and 5 ml dilutes ammonium solution is poured into it. It is followed by the addition of concentrate H₂SO₄. Formation of yellow colour indicates presence of the flavonoid.

Glycosides

5 ml of extract is taken and 2ml of glacial acetic acid containing a drop of ferric chloride is added. Then 1 ml concentrate H₂SO₄ is added in it. A brown ring suggests the presence of glycosides.

Phenol

2 ml of distilled water is taken, with it few drops 10% or aq. ferric chloride is added. 1 ml of solvent extract is added in it. Formation of blue or green in colour indicates presence of phenol.

Terpenoid

5 ml of extract along with 2 ml of chloroform is taken. Carefully 3 ml of concentrate H₂SO₄ is added. Reddish brown coloration of the interface formation is indicating a positive result.

Anthraquinone

1 ml crude extract added taken. In it dilute NaOH was added. Blue, green or red coloration indicates the presence of anthraquinones.

Result and Discussion

A very much satisfactory result of anthelmintic activity is shown by the methanol extract and aqueous extract of *Acacia nilotica*. A different death and paralysis time is being shown with similar concentration

Albendazole was used to compare the result with the test sample concentration. But there are various side effect, possesses with practicing this drug. Investigation of modern research clearly certified the use and importance medicinal plant with having anthelmintic property. As expected, there is no positive result with control. But standard drug albendazole(100 mg/ml), methanol and aqueous plant extract (50 mg/ml) shows satisfactory result of paralysis and death time of each worm. Qualitative phytochemical activity of various extracts of the plant was shown in the table after proper analysis is made (Table-1), The potentiality of anthelmintic activity shown in (Table-2),Figure-1, Figure-2, this plant also have different phytoconstituents, which is shown in (Table-1).

Table 1: Preliminary phytochemical screening of Acacia nilotica

Sl.No	Phytochemical Constituents	Methanol	Acetone	Aqueous
1.	Alkaloids	+	+	-
2.	Flavonoids	+	+	-
3.	Glycosides	-	-	+
4.	Phenol	+	-	-
5.	Terpenoid	+	+	+
6.	Anthraquinones	+	+	+

+ = Positive, - = Negative

Table 2: Anthelmintic potency of methanolic and aqueous extract of Acacia nilotica

Extract	Concentration (mg/ml)	<i>Pheretimaposthuma</i>	
		Paralysis (P)	Death (D)
Control	-	-	-
Standard (Albendazole)	100 mg/ml	22min 33 sec	42min 28 sec
Methanolic extract	50 mg/ml	45 min 30 sec	80 min 21 sec
	100 mg/ml	20 min 15 sec	58 min 50 sec
Aqueous extract	50 mg/ml	54 min 12 sec	84 min 39 sec
	100 mg/ml	22 min 45 sec	39 min 50 sec

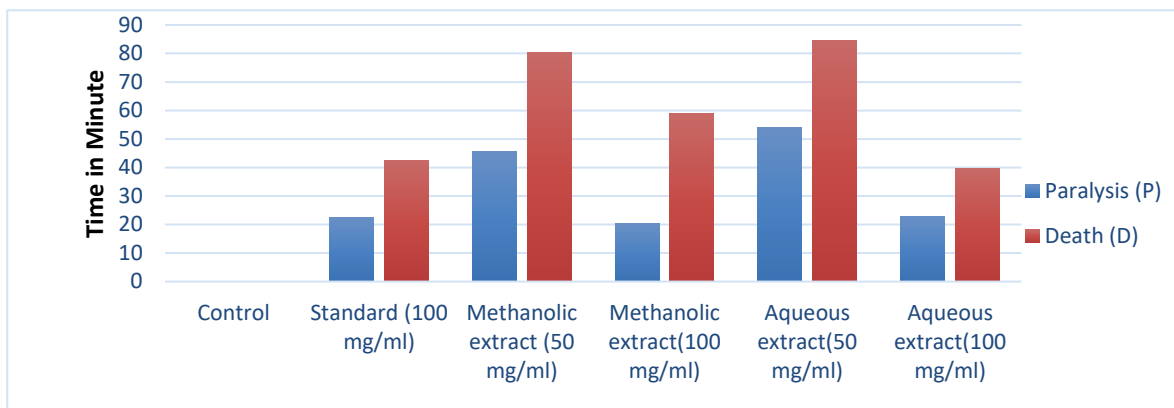


Figure 1: Anthelmintic activity of *Acacia nilotica* on *Pheretimaposthuma*.



(A) (B) (C) (D) (E) (F)
 Figure 2: Anthelmintic activity of *Acacia nilotica* leaves and flower on *Pheretimaposthuma* where (A) Control, (B) Standard Albendazole, (C) 50 mg/ml concentrations of Aqueous extract, (D) 100 mg/ml concentrations of Aqueous extract, (E) 50 mg/ml concentrations of Methanolic extract, and (F) 100 mg/ml concentrations of Methanolic extract.

Conclusion

After carefully observing all the results it is being conclude that: *Acacia nilotica* shows very potential phytoconstituents and anthelmintic activity against the Indian adult earth worm. At concentration of 100 mg/ml it shows very satisfactory results of both the death and paralysis time due to anthelmintic activity at the end of the experiment. The result of various concentrations of methanolic and aqueous extract of the plant can compare with the according the rules standard Albendazole and control. This plant will open a new branch for drug researchers for its various important activities without side effect.

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