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EVALUATION OF *IN-VITRO* ANTHELMENTIC ACTIVITY OF *BETA VULGARIS*

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ABSTRACT: Plant products are of increasing interest in the search for new drugs and medicines in the treatment of diseases; and the discovery of new alternatives to existing commercially available drugs is an important area of this research due to pathogen resistance. The present investigation focused on the screening of anthelmintic potential of pet-ether, benzene, ethanol and aqueous extracts of the plant beet root (*Beta vulgaris*). The crude extract of the *Beta vulgaris* with pet ether, benzene, ethanol and aqueous were screened for anthelmintic activity against Indian adult earthworm *Pheretima posthuma*. The *in-vitro* anthelmintic activity was performed by using standard procedures. The comparison of the paralysis time and death time in different extracts with respect to the standard showed that the ethanol extract had most significant activity than petroleum ether; benzene and aqueous extract. The phyto-chemical constituents like flavanoids and alkaloids may be responsible for the activity. Finally our study conclude that *Beta vulgaris* has significant anti-helminthic activity and potential to develop as useful and safe alternative to the other existing drugs which are having more side-effects and resistance. The ethanolic extracts exhibited highest activity against Indian adult earthworms whereas the other extracts exhibited positive activity. The results obtained in the present study suggest that the ethanol extract of *Beta vulgaris* revealed scope to develop a novel broad spectrum of anthelmintic medicinal plants.

INTRODUCTION: Helminthes are a polyphyletic group of eukaryotic parasites. They are worm-like organisms living in and feeding on living hosts, receiving nourishment and protection while disrupting their hosts' nutrient absorption, causing weakness and disease. Those that live inside the digestive tract are called intestinal parasites. They can live inside humans and other animals. Helminthology is the study of parasitic worms and their effects on their hosts. The word helminthes comes from Greek hélmin, a kind of worm [1].

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Helminthe is a polyphyletic group of morpho-logically similar organisms, consisting of members of the following taxa: monogeneans, cestodes (tapeworms), nematodes (roundworms), and trematodes (flukes) [2].

Infection by intestinal parasitic worms (geohelminths) is widespread throughout the world, affecting hundreds of millions of people. Children are particularly susceptible and typically have the largest number of worms. Three of the most common kinds of worms are roundworm (*Ascarislum bricoides*), whipworm (*Trichuris trichiura*) and hookworm (*Ancylostoma duodenale* and *Necator americanus*). These worms live in the intestines and their numbers build up through repeated infection. It is possible to be infected with more than one kind of worm [3].

Anthelmintics are drugs that act locally to expel worms from the gastro-intestinal tract or systemically to eradicate adult helminths or developmental stages that invade organs and tissues. Parasitic diseases cause ruthless morbidity affecting principally population in endemic areas. The gastro-intestinal helminthes becomes resistant to currently available anthelmintic drugs therefore there is a foremost problem in treatment of helminthes diseases. Hence there is an increasing demand towards natural anthelmintics [4].

Beta vulgaris (beet) is a plant in the Chenopodiaceae family which is now included in Amaranthaceae family. It has numerous cultivated varieties, the best known of which is the root vegetable known as the beetroot or garden beet [5].

Beta vulgaris has been used for centuries as a traditional natural coloring agent in dairy and meat products in many cuisines. Medicinally, the roots and leaves of the *B. vulgaris* have been employed as a folk remedy to treat a wide variety of ailments including immune system Stimulation, liver and kidney diseases. It is also useful to prevent some kinds of cancers.

B. vulgaris seeds have cooling and diaphoretic effects, while the roots have a good nutrient effect. The present study was designed to evaluate the anthelmintic activity of *Beta vulgaris* using adult Indian earth worms [6].

The assay was performed *in-vitro* using adult earthworm (*Pheretima posthuma)* owing to its anatomical and physiological resemblance with the intestinal roundworm parasites of human beings for preliminary evaluation of anthelmintic activity of *Beta vulgaris*.

MATERIALS AND METHODS:

Chemicals: The Following solvents were collected from the drug store of CMR COLLEGE OF PHARMACY. Pet ether, Benzene, Ethanol, Aqueous

Plant Materials: The plant material of *Beta Vulgaris* (6 kgs) were purchased from the local market at suchitra in Feb, 2014.

Preparation of Extract: Commercially available beetroot was used for the extraction process. The vegetable material of *Beta vulgaris*root was washed thoroughly and cut into thin slices and kept for shade drying for 4-5 days. Then the dried *Beta vulgaris* was made into powder form by grinding. The powder obtained is then passed through the sieve number 22 to obtain a fine powder [7].

Maceration: In this process, the whole or coarsely powdered crude drug is placed in a stoppered container with the following solvents pet ether, benzene, ethanol and water and allowed to stand for 5 days at room temperature with frequent agitation. The mixture then is strained, the marc (the damp solid material) is pressed, and the combined liquids are clarified by filtration or decantation after standing. The filtrate obtained is allowed to concentrate on heating mantle at 20 °C. The dried extracts were stored in well closed containers until further study [8].

Phytochemical Screening: Ethanolic extract was treated with various reagents which showed the presence of various phytochemical constitutents. The results are shown in **Table 1.**

WORMS: Adult earthworms (*Pheretima posthuma*) were used to evaluate anthelmintic activity *in-vitro.* Earthworms were collected at bio-fertilizer near kompally area and washed with water to remove all the soil matter. The average size of earthworm was 15-16cms

FIG. 1: *PHERETHIMA POSTHUMA*

Standard Drug: Albendazole (20mg) was used as standard drug during the experimental protocol [9].

Anthelmintic Activity: The anthelmintic assay was carried out as per the method of (Satish B. Koslge *et al*). The assay was performed *in-vitro* using adult earthworm (*Pheretima posthuma)* owing to its anatomical and physiological

resemblance with the intestinal roundworm parasites of human beings for preliminary evaluation anthelmintic activity [10].

Test samples of the extracts was prepared at the concentration 100mg/ml in distilled water and worms *i.e. Pheretima posthuma,* approximately equal size (15-16) were placed in each nine cm Petri dish containing 25 ml of above test solution of extracts [11]. Albendazole (20mg/ml) was used as reference standard and distilled water as control. All the test solution and standard drug solution were prepared freshly before starting the experiments.

Observations were made for the time taken for paralysis was noted when no movement of any sort could be observed except when the worms were shaken vigorously. Time for death of worms were recorded after ascertaining that worms did not moved when shaken vigorously the results were shown in **Table 3** and expressed as a mean± SEM [12].

RESULTS AND DISCUSSION:

TABLE 1: RESULTS OF PHYTOCHEMICAL SCREENING

Extract constituents	Pet. Ether	Benzene	Ethanol	Aqueous
Alkaloids	-	+	+	-
Carbohydrates	-	-	+	+
Glycosides	-	+	+	+
Tannin	-	-	+	+
Phenolics	-	-	+	+
Flavonoids	-	+	+	+
Proteins & amino acids	-	-	-	+
Saponins	-	-	-	-
Acidic compounds	-	-	-	-
Mucilage	+	-	-	-
Resins	-	-	-	-
Lipids/fats	+	-	-	-

(Absent - , present +).

TABLE 2: *IN-VITRO* ANTHELMENTIC ACTIVITY OF *BETA VULGARIS*

Control	Time of Paralysis (Min)	Time of Death (Min)
Albendazole 20 mg/ml	22.50 ± 0.846	54 ± 1.90
Pet. Ether extract 100 mg/ml	17.33±0.666	73 ± 0.31
Benzene extract 100 mg/ml	43.50±0.500	66 ±0.80
Ethanol extract 100mg /ml	7.33±0.421	11 ±0.40
Aqueous extract 100mg/ml	9.66±0.557	72 ±0.56

The values were expressed as Mean ± SEM, n =6

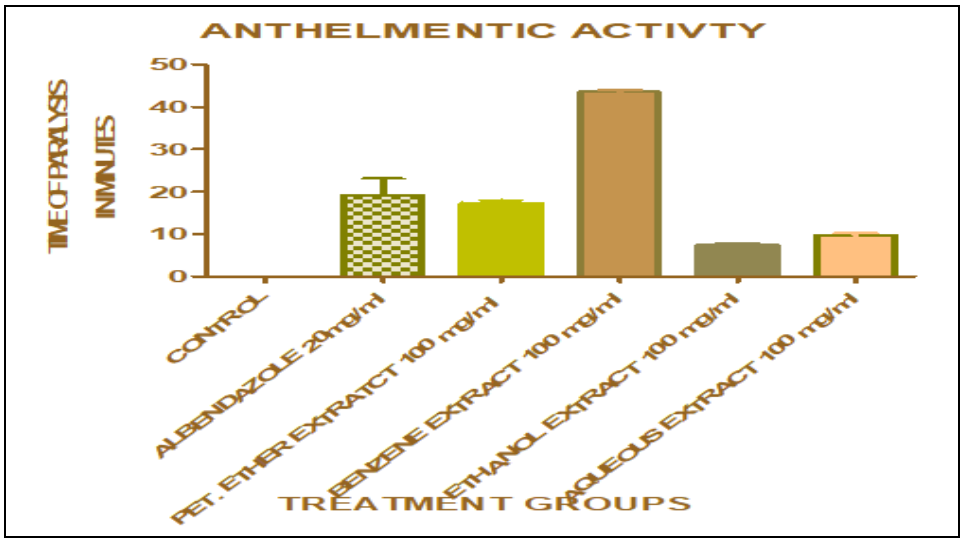

FIG. 2: *IN-VITRO* ANTHELMENTIC ACTIVITY OF *BETA VULGARIS* IN TERMS OF PARALYSISIN MINUTES

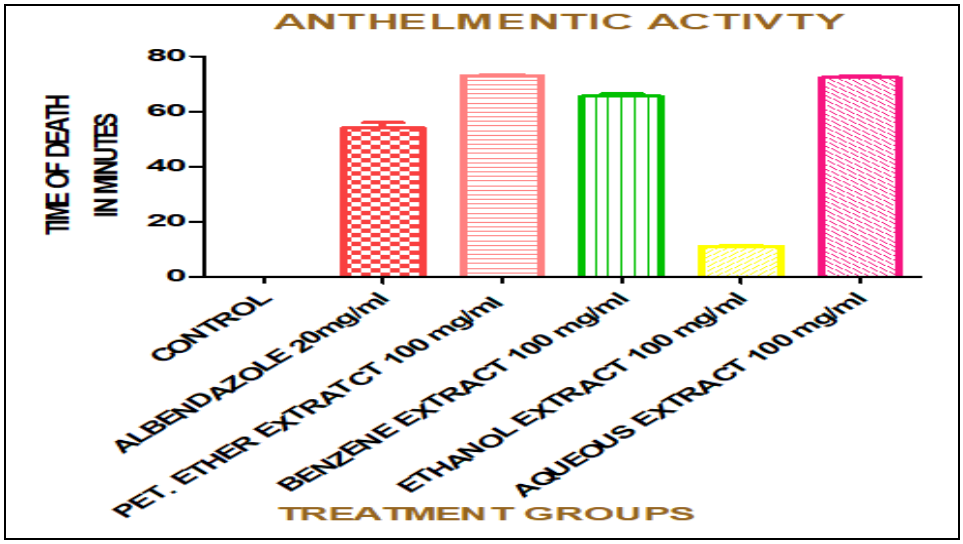

FIG. 3: *IN-VITRO* ANTHELMENTIC ACTIVITY OF *BETA VULGARIS* IN TERMS OF DEATH TIME IN MINUTES

Evaluation of *In-vitro* Anthelmentic Activity of Evaluation of Various Extracts of *Beta Vulgaris:*

FIG. 4: ALBENDAZOLE

FIG. 5: PETETHER

FIG. 6: BENZENE EXTRACTION

FIG. 7: ETHANOLICEXTRACTION

FIG. 8: AQUEOUS EXTRACTION

The phytochemical screening revealed the presence of phytoconstituents such as alkaloids, carbohydrates, flavonoids, tannins, phenolics in the extract of *Beta vulgaries* shown in the **Table 1** by performing the preliminary phytochemical screening. And the following tests were performed for alkaloids –Mayer's test, carbohydrates and glycosides-molisch's test, phenolic compounds and tannins- ferric chloride test, proteins and amino acids-millon's test or biuret test, fixed oils and fats-saponification test, saponins-foam test, gums and mucilage-alcoholic, precipitation and molisch's test [13]. Anti-helminthic effect of *Beta vulgaries* extracts was studied. All of the data obtained from the experimental groups have been compared to Albendazole group. The data was analyzed statistically by one way ANOVA followed by Dunnett test using graph pad prism 5.0 Software [14]. Values are expressed as mean ± SEM (n=6). Where, *** ($p<0.001$) vs., ** ($p<0.01$) vs. Albendazole control, * ($p<0.1$) vs. Albendazole control### ($p<0.001$) vs. normal control.

The results of anthelmintic activity are shown in **Table 2**. In the present study it was observed that all the extracts have shown positive response to certain degree of anthelmintic activity. Whereas ethanol extract of plant shown significant activity as compared to standard.

CONCLUSION: *In-vitro* anthelmintic activity was performed and the paralysis time and death time were recorded. Statistical evaluation of the data was performed by one-way ANOVA. The results were expressed as mean ± SEM using Graph Pad 5 (n = 6). The results show that for the 20 mg/ml concentration of albendazole showed the activity for paralysis time (22.50 ± 0.846 min) death time (54 ± 1.90 min). The Petroleum ether extract of *Beta vulgaris* showed paralysis time (17.33 ± 0.666min) death time of (73 ± 0.31min).

The benzene extract of *Beta vulgaris* showed paralysis time (43.50±0.500min) death time of (66 ±0.80min). The ethanol extract of *Beta vulgaris* showed paralysis time (7.33±0.421min) death time of (11 ±0.40min). The aqueous extract of *Beta vulgaris* showed paralysis time (9.66±0.557) death time of (72 ±0.56). The study revealed that ethanol extracts had significant activity compared with the standard albendazole (20 mg/ml).

The comparison of the paralysis time and death time in different extracts with respect to the standard showed that the ethanol extract had most significant activity than petroleum ether, benzene and aqueous extract. The phyto-chemical constituents like flavanoids and alkaloids may be responsible for the activity. Finally our study conclude that *Beta vulgaris* has significant anti-helminthic activity and potential to develop as useful and safe alternative to the other existing drugs which are having more side-effects and resistance.

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REFERENCES:

1. Maizels Rick M and Yazdanbakhsh M: Immune regulation by helminth parasites: cellular and molecular mechanisms. Nature Reviews Immunology 2003; 3: 733-744.
2. Borkow G & Bentwich Z: Eradication of helminthic infections may be essential for successful vaccination against HIV and tuberculosis". Bulletin of the World Health Organization 2000; 78(11): 1368-1369.
3. Musa Suleiman MD, Mamman MD, Yusuf Otaru Aliu and Joseph Ologunja Ajanusi: Anthelmintic activity of the crude methanol extract of *Xylopia aethiopica* against *Nippostrongylus brasiliensis* in rats". Veterinarskiarhiv 2005; 75 (6): 487-495.
4. Danquah CA, Koffuor GA, Annan K and Ketor EC: The Anthelmintic Activity of *Vernonia amygdalina* (Asteraceae) and *Alstonia boonei* De Wild (Apocynaceae). Journal of Medical and Biomedical Sciences 2012; 1(1): 21-27.
5. Marwa M. Dlim, Sami G. Alsabri, Sofian S. Mohamed, Abdulmottaleb E. Zetrini, Asma AH. Sale, Abdurazag A and Auziand Salah B. Mohamed: Use of *Beta vulgaris* as natural coloring agent for foods and cosmetics in Libya". Journal of Chemical and Pharmaceutical Research 2013; 5(11): 340-345.
6. Aura S, Marta S, Anicuţa S and Tănase D: Betanine extraction from *Beta vulgaris* experimental research and statistical modeling; U.P.B. Sci. Bull Series B 2011; 73(1): 145-156.
7. Canadanovic-Brunet JM and Savatovic SS: Antioxidant and Antimicrobial activities of beet root pomace extracts. Czech Journal of Food Sciences 2011; 29(6): 575-585.
8. Satish B. Kosalge, Ravindra and Fursule: Investigation of *in-vitro* anthelmintic activity of *Thespesia lampas* CAV); Asian Journal of Pharmaceutical and Clinical Research 2009; 2(2): 69-71.
9. Baskar Lakshmanan, Papiya Mitra Mazumder, D. Sasmal, S. Ganguly and Simon Santosh Jena: "*In–vitro* anthelmentic activity of some 1-substituted Imidazole derivatives. Acta Parasitologica Globalis 2011; 2(1) 01-05.
10. Sangh Partap, Saurabh Kumar, Amit Kumar, Neeraj K. Sharma and Jha KK: *In-vitro* Anthelmintic activity of *Luffa cylindrical* leaves in indian adult earthworms. Journal of Pharmacognosy and Phytochemistry 2012; 1(2): 27-30.
11. Dinesh Kumar Mehta, Rina Das and Anil Bhandari: *In-vitro* anthelmintic activity of seeds of *Zanthoxylum armatum* DC. against *Pheretima posthuma*. International Journal of Green Pharmacy 2012; 6(1): 26-28.
12. Dhawal Dorwal: Anthelmintic activity of Methanolic and Ethanolic Leaf Extract of *Calendula officinalis*; International Journal of Research in Pharmaceutical and Biomedical Sciences 2012; 3(2): 831-832.
13. Dhunmati K, Jaison D, Kousalya M, Mohammad yaseen A, Swetha S and Kuppuram G: Evalution of Anthelmintic activity of ethanolic root extract of *Zizyphus oenoplia*. International Journal of Pharmacy and Pharmaceutical Sciences 2012; 4: 597-599.

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