Anthelmintic Evaluation of Polyherbal Formulation: Krimikuthar Ras Tablet

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Abstract

Ayurveda has abundant collection of effective formulations against diseases. The polyherbal formulation contains *Embelia ribes*, *Ferula asafoetida, Holarrhena pubescens, Acorus calamus*. The present study deals with anthelmintic evaluation of ayurvedic formulation. Various concentrations of polyherbal formulation 50 mg/ml, 75 mg/ml and 100 mg/ml were taken. The results were compared with standard albendazole 50 mg/ml, 75 mg/ml and 100 mg/ml. The results were expressed in terms of time in minutes for the paralysis and time of death of the worms. Paralysis and death time were analyzed using one way ANOVA analysis using Graph Pad Prism 5.0 was used to conduct the statistical analysis, which included one-way analyses of variance (ANOVA) and Dunnett's Multiple Comparison test. The study indicates that polyherbal formulation shows more potent anthelmintic activity than standard reference.

Key words : Anthelmintic Evaluation, Krimikuthar Ras Tablet, Polyherbal formulation.

Nature has offered a vast storehouse of medicines to treat all of humanity's ills and disorders. Plants were utilized to make practically all medications in the past. For millennia, it has served as man's chemist. Bioactive compounds found in medicinal plants include tannins, alkaloids, carbohydrates,

terpenoids, steroids, and flavonoids, all of which have a physiological effect on the human body¹.

Helminthiasis :

Helminthiasis, or worm infection, is

one of the world's most common diseases. Helminthiasis affects over half of the world's population, and the disease is on the rise. Because of poor sanitation, poor family hygiene, hunger, and crowded living situations, it is not just limited to tropical and subtropical countries, but is also endemic in many locations².

Intestinal nematodes infect an estimated 2 billion individuals worldwide. Anthelmintic medications are used to treat parasitic infections caused by helminths. The demand for novel and effective anthelmintics is enormous, as the chemical medications currently used to control helminths are costly, and most of them lose their potency within 20 years due to resistance. Because of the strong link between these diseases and poverty, eradicating helminthiases is particularly difficult³. Metazoa, which includes roundworms (nematodes) and two forms of flatworms, flukes (trematodes) and tapeworms, are pathogenic for humans (cestodes). These physiologically distinct eukaryotes differ in terms of life cycle, bodily structure, development, physiology, host location, and chemotherapeutic susceptibility. Immature forms infect humans through the skin or gastrointestinal tract, developing into well-differentiated adult worms with distinct tissue distribution.

Helminthiasis is a macroparasitic disease of humans and animals in which a part of the body is infested with parasitic worms such as pinworm, roundworm or tapeworm. More than half of the population of the world suffers from infection of one or the other and majority of cattle's suffers from worm infections⁴. Anthelmintics are drugs that expel parasitic worms from the human body⁵. Treatment with an anthelmintic drug kills worms whose genotype renders them susceptible to the drug. Worms that are resistant survive and pass on their "resistance" genes. Resistant worms accumulate and finally treatment failure occurs⁶.

The parasitic helmintic infection is increasing death and morbidity all over the world. Intestinal nematodes (roundworms), trematodes (flukes), and cestodes are examples (tapeworms). Because it is a major source of environmental contamination and transmission, it is an unevenly distributed disease in lowincome nations that affects the worst and highest risk of morbidity. Due to the persistence of periodic emergence of resistance, albendazole, mebendazole, and praziquantel are the most regularly used anthelmintic medicines with broad spectrum activity and High Cure Rates⁷.

Selection of Herbs :

Krimikuthar Ras Tablet were prepared in laboratory by taking the same ingredients as mentioned above. Sodhana of *Embelia ribes, Ferula asafoetida, Holarrhena pubescens, Acorus calamus* were done as mentioned in Ayurvedic formulary. Dry granulation method was followed. Fine powder of each ingredient was passed through sieve no. 80 and compressed to form tablets using binding agent.

Selection of animals :

Adult Indian earthworms (Eudrilus eugeniae) and tapeworms (Haemonchus contortus) were used to test the activity. Earthworms were bought from local suppliers.

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The worms were authenticated by Zoology Department, RFNS Senior Science College, Akkalkuwa. The worms were placed in regular saline to offer them with the usual leaving circumstances they require. The earthworms of 3-5 cm in length and 0.1-0.2 cm in width were used for all the experimental protocol. Earthworms were chosen to examine anthelmintic activity because they were morphologically and physiologically similar to intestinal roundworms^{10,11}.

Preparation of samples :

To obtain different concentrations, required quantities of Krimikuthar Ras Tablet and a standard medication (Albendazole) were dissolved in DMSO and then diluted with distilled water. (50 mg/ml, 75 mg/ml, and 100 mg/ml).

In-vitro evaluation of anthelmintic activity^{6, 8, 9}

Adult Indian earthworms and tapeworms from the intestine of a goat were used to test the antihelmintic activity. Six groups of six worms of approximately identical size were given 10 ml of the necessary concentration. Individual worms were timed to see how long it took them to become paralysed and die. When there was no movement of any kind except when the worms were shaken strongly, the time of paralysis was recorded. After determining that worms did not move when shaken vigorously or dunked in heated water (50 °C), the time it took for them to die was recorded.

Statistical Analysis :

The results are presented as a mean standard error of the mean (SEM). Graph Pad Prism 5.0 was used to conduct the statistical analysis, which included one-way analyses of variance (ANOVA) and Dunnett's Multiple Comparison test. The statistical significance of P<0.05 was determined.

The evaluation of anthelmintic activity on earthworm revealed that the activity was dose dependent and inversely proportional to paralysis and death time. From the graph (Figure 1 and Figure 2), it is clearly seen that the herbal tablet is more potential than reference control. The activity of herbal tablet is more significant at 100mg/ml with significant level **p< 0.001.

	Concen-	Indian Earth Worm		Tape Worm	
Test Sample	tration	For	For	For	For
	(mg/ml)	Paralysis	Death	Paralysis	Death
Albendazole	50	50.64 ± 0.57	72.71 ± 0.74	41.26 ± 0.85	71.31 ± 0.53
	75	34.51 ± 0.35	50.72 ± 0.26	32.31 ± 0.64	62.11 ± 0.64
	100	22.43 ± 1.41	40.15 ± 0.42	20.15 ± 0.43	40.25 ± 0.31
Krimikuthar	50	33.27 ± 1.01	64.56 ± 0.29	31.46 ± 1.19	68.32 ± 0.64
Ras Tablet	75	25.33 ± 0.89	44.38 ± 1.15	25.66 ± 0.89	55.56 ± 1.08
	100	18.64 ± 0.49	34.89 ± 0.78	14.18 ± 0.78	37.17 ± 0.54

Table-1. Evaluation of Anthelmintic Activity

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Fig. 3. Graph of Paralysis time of Round Worm



Fig. 4. Graph of Death time of Round Worm

The evaluation of anthelmintic activity on round worm revealed that the activity was dose dependent and inversely proportional to paralysis and death time of worms. From the graph (Figure 3 and 4), it is clearly seen that the herbal tablet is more potential than reference control. The activity of herbal tablet is more significant at 100mg/ml with significant level **p< 0.001.

Adult Indian earthworms (Eudrilus eugeniae) and tapeworms were used to test the anthelmintic activity (Haemonchus contortus). The evaluation of anthelmintic activity on round worm revealed that the activity was dose dependent and inversely proportional to paralysis and death time of worms. From the graph (Figure 1 to 4), it is clearly seen that the herbal Krimikuthar Ras tablet shows significant activity as compared to reference control. The possible mechanism of the anthelmintic activity of polyherbal tablet cannot be explained on the basis of our present results. However, it may be due to its effect on inhibition of glucose uptake in the parasites and depletion of its glycogen synthesis. It may also have activated nicotinic cholinergic receptor in the worms resulting in either persistent depolarization or hyperpolarization.

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