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Assessment of '*Vipaka*' of *Drynaria Quercifolia*.Linn in Animal Models

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ABSTRACT: India is a country with a wide variety of flora. Although Ayurveda recognizes very *Dravya*as useful, we can only find a limited number of medicinal plants mentioned in Ayurvedic classical textbooks. *Acharyas* also mentioned we should identify and learn the uses of unknown medicinal plants from people who are residing near the forest. In the current era, that type of ethnomedical practice has to be scientifically evaluated. Also considering the increased demand and over-exploitation of the existing marketed medicines it is essential to add the extra pharmacopeial drugs to the ayurvedic pharmacopeia. For this, all the principles of the drug action put forward by our great acharyas have to be considered. For that detailed analysis shall be applied by using the well-established existing models and methodologies. Though Acharyas had mentioned Sapthapadarthas like Dravya, Rasa, etcas a medium for treatment, Vipakais considered as special among them because of its relation with the Agni. The development of the sapthadhatu and tridoshaarebased on this Agnipaka. After pakaof Dravyaonly the real action of it got cleared. ThusVipaka becomes responsible for several metabolic processes in the body. On this background, the present study was taken up for analysis of Vipaka of medicinal plant Drynariaquercifolia. Linn is of Polypodiaceae family, and which is traditionally used in peptic ulcer, jaundice, curing insanity, etc in different regions of India. The outcome of the study can be considered as preliminary evidence and will hopefully inspire more studies with different parameters for further validation.

KEYWORDS: *Drynariaquercifolia, Vipaka, Rasapanchaka*.

INTRODUCTION:

Due to the wide range of climate, topology, and habitat, the Flora of India is one of the richest in the world. There are estimated more than 50,000 species of plants, including a variety of endemics in India. The use of plants as a source of medicines has been an integral part of the tradition in India from the earliest times. There are more than 3000 Indian plant species are officially documented (Majid Husain - Geography of India 2014).

Though there is an innumerable number of plant species, many plantswere not recorded in classical ayurvedic works though they have medicinal properties. Ayurveda also emphasized the fact that any *dravya*in the world can be used as medicine if wisely used. *CharakaAcharya* explainedthat's it is difficult to explain all the herbal medicines in one textbook[1]. Acharyas also emphasized that the physician has to use his wisdom for knowing the properties of unexplained drugs[2].In the olden day's vaidyas use to gain knowledge about unknown plants through cowherds, shepherds, hermits, ethnic communities who are residing nearby the forest[3]. They were the best observers of nature and are familiar with the uses of the plants. Many of the undocumented herbs were utilized and practiced by the many ethnic groups. Many of such undocumented

herbs are not evaluated scientifically till date. In this modern era of evidence-based practice, proper scientific proof and its documentation are essential. Recently, herbal drug research is emphasizedin enlightening the medicinal properties of such undocumented herbs. But, it is very difficult to transform properties of such herbs into therapeutic application in Ayurveda, as Treatments in Ayurveda is based on Ayurvedic principles of drug actions i.e. *Rasa, Guna, Virya, Vipaka,* and *Prabhava*.

A drug was told to act by its potency. The potency of the drug implies all the qualities of the drug by which they act, viz*Rasa, Guna, Virya, Vipaka,* and *Prabhava*. *Rasa* and *Vipaka* indicate the chemical structure of the drug whereas *Guna*and *Virya* indicate the physico-pharmacological properties of the drug. Different drugs have different *Panchabhowthika* constitutions. The specific arrangement of the *bhoothas* will give rise to a particular *rasa*(taste) of the drug. Rasa has certain local and systemic actions. After the drug is digested and metabolizedit gets brokendown and reform its rasa qualities. It is called*Vipaka*. *Gunas* are the qualities that inherently reside in a *dravya* and *Virya*are the specific *gunas* (qualities) thatremain in the *dravya* even after the digestion and which is the cause for the actions done by the *dravya*. *Prabhava* is the specific action done by a *dravya* and which is also a differentiating factor that highlights one *dravya*from other *dravyas* which have similar qualities of taste, potency, and post-digestive changes. A drug is told to perform its general actions by its *Rasa* and *Guna* and certain specific actions by its *Vipaka* and *Virya*[4].

Thusthe outcome of the biotransformation of the *rasa* of a given *dravya* through the action of *jatharagni* is known as *Vipaka*[5]. Acharya *Susrutha* explains *Vipaka* is the mean cause for various pharmacological actions. He explains the *samyakpaka* of the *dravya* imparts good effects to the body whereas *mithyapaka* of the *dravya* imparts bad effects[6]. There flections of *Vipaka*can be seen from the *sthoola* to *sukshma* levels in the body.*BhadantaNagarjuna* explains the superiority of *Vipaka* as it is the *Nimittakarana* of the *Vridhi* and *Kshaya* of the *doshas* and *Dhatu*, the factor in which the therapeutic effect of the drug is dependent and it is a factor which is emphasized by all the classical textbooks. *Vipaka* helps in maintaining the healthy condition of the body[7].

Acharya Charaka explains *Vipaka*is inferred only from the actions it produces in the body. That is their action at the level of *Doshas, Dhatu,* and *Mala*. The changes in the substance may take place at the *Pachakagni* level, *Bhoothagni* level, and *Dhatwagni* level. Therefore the sphere of action of *Vipaka* extends from the GI tract to the dhatus. After ingestion of *Dravya*, any transformation of substance by any enzymatic or hormonal actions can be accounted to the vipaka. *AcharyaCharaka* explains 3 kinds of *Vipaka*, viz*Mathura, Amla,* and *Katu* in relation to*Tridosha*[8].He explains *Katuvipaka* promotes *Vata* and causes diminution of the *dhatus*and the suppression of feces and urine. While *Madhuravipaka* is promotive of *Kapha*and causative of increase of *dhatus*and elimination of feces and urine. The *Amlavipaka* promotes *Pitta*, diminishes the *dhatus*, and promotes the elimination of feces and urine[9].As the *Vipaka*cannot be assessed directly as taste, it has to be perceived from the action of the drug in Dhatu and Mala level for which specific controlled methods have to be followed. Currently,Ayurvedic scholars are following the available experimental method (Dhyani S.C., 2008) for evaluating the Vipaka of the drug, which is made as per Charakaacharya's explanations.

Drynariaquercifolia.Linn is one of the important medicinal plant which is used ethno-medically in several parts of India. It is called *Asvakathri*in Sanskrit, *Matilpanna* or *Marapannakizhangu*in Malayalam and *Oak leaf fern* in English. It is a fern of *Polypodiaceae* family and is found among rocks in crevices, shelves, or in the soil among boulders also epiphytic on tree trunks in open forests and rainforests throughout India. Its Rhizome is said to be having *Thiktha rasapradhana* and is effective in conditions like *Grahani, Sopha, Dustavrana, Suryavartha,*etc,and also used in conditions like phthisis, dyspepsia, typhoid fever, etc. In folklore practice it is used for conditions like peptic ulcer, jaundice, curing insanity, Postpartum hemorrhage condition,etc[10,11,12].

Considering all the above facts of *Aswakathri* (*Drynariaquercifolia*.Linn), To make it available in Ayurveda based on the ayurvedic principles, an experiment was carried out in the Wistar albino rats. In the study 12 Wistar albino rats were selected and divided into 2 groups. Group A – Control group, Group B – Trial group. Each rat was kept in separate metabolic cages provided with a constant amount of water and food per day. Assessment of Vipaka was done based on consumption of food, consumption of water, the quantity of fecal output, the quantity of urine output, and change in body weight (Dhyani S.C., 2008).

MATERIALS AND METHODS

Plant material:

Drynariaquercifolia (Linn.) J. Smith is collected from Pandalam, Pathanamthitta. It was authenticated by R. RAJAGOPAL. M.Sc. M.Phil Associate Professor (Retd.) Department of Botany S.N, College, Kollam.Fresh Rhizomes of *Drynariaquercifolia*collected from its natural habitat were cleaned by thorough washing with tap water to remove the dust particles. Then the wooly brown scales of the rhizomes were removed and the clean rhizomes and taken for Experimental study.

Preparation of Decoction of *Drynariaquercifolia*.Linn:

The decoction was be made by the conventional method of *kwatha* preparation. The required drug was boiled with 16 times water and reduced to 1/8th. The *kwatha*was sieved to remove all the solid drug particles and then used for the experimental study[13].

Experimental Animal:

Wistar albino rats weighing between 150 to 200 gms of either sex were used for the study. The animals were obtained from the animal house in the S.D.M Centre for Research in Ayurveda and Allied Sciences, Kuthpady, Udupi. 12 albino rats were selected and allotted to 2 groups of 6 rats each. Each rat was kept in separate metabolic cages.

Experimental procedure:

The experiment was conducted by observing the effect of the drug on the following factors:

1. Food consumption
2. Water intake
3. Urine output
4. Fecal output
5. Change in body weight

Method:

Twelve Wistar albino rats were selected for study and are divided into 2 groups of six rats each in each group (control group and trial group).

The study was performed in two phases

A. Preliminary phase: Duration – First 5 days

B. Experimental phase: Duration – 10 days

Preliminary phase:

The preliminary phase of the study was carried out prior to the experimental phase to understand and obtain baseline data about the normal amount of food consumption, water intake, urine, and fecal output of each rat.

In the preliminary phase, the initial weights of the rats were recorded and they are placed in separate metabolic cages. Metabolic cages have special arrangements for keeping food and water and this also prevents admixture of food with fecal matter. In these cages, urine is drained out and the fecal matter can be collected easily from the cages. The urine can be collected from the plastic glasses placed below the cage. In this phase, the drug was not administered.

Experimental phase:

In this phase, animals were administered the selected drug as per the calculated doses. All the previously mentioned parameters were recorded daily for 10 consecutive days.

- The rats of the control group received distilled water according to body weight during the experimental phase.
- The rats of the trial group received Rhizome decoction of *Drynariaquercifolia*.Linn according to body weight during the experimental phase.

Assessment of Parameters:

1. Method of estimation of the weight change: The difference between the body weight of animals before starting the experimental phase and after therapy indicates the actual weight change as a result of metabolic activity. Weight variation before and after treatment was calculated and variation of weight per 100gm was obtained (% change).

2. Method of estimation of food consumption: Each rat was provided a 200gm dry pellet/day to ensure maximum food consumption according to its capacity. The residual food was collected on the next day and it was weighed again. The total amount of food consumed by the animal in 24 hours was obtained by deduction of the remaining food from the given 200gms. This is the absolute value of food consumed in grams. This value was then calculated with the bodyweight of the animal and food consumed on gram/percentage of body weight per day was calculated. This was the relative value of food consumption.

3. Method of estimating the water intake: To each rat 200ml of tap water was supplied in bottles and after 24 hours the water remaining in each bottle was noted. The total amount of water intake in each rat was then calculated by deducing the remaining water in the feeding bottle from 200ml. The water intake in ml% of the body weight per day was then calculated. This was calculated as relative water intake.

4. Method of estimating the weight of fecal matter: The total amount of fecal matter passed by each rat was collected separately. It was then weighed and kept in the oven at 80^{o}c for 8 hours. Weights of dry fecal matter were then calculated. The fecal matter passed in gm% of body weight per day was then calculated. This was the relative weight of dry fecal matter.

5. Method of estimation of urine output: Daily urine (24 hours) which was collected in the plastic glass was measured in with help of a graduated test tube. The urine output in ml% of the body weight per day was then calculated. This was the relative urine output.

RESULTS:

Results obtained from the experiment are summarized in tables 1–15

Table – 01. Effect of *Aswakathri (Drynariaquercifolia* . Linn) on body weight of albino rats

Effect on Body Weight		
	Control - A	Trial - B
Mean ± SEM	-0.24 ± 0.9672	5.16 ± 1.563

Data : MEAN ±SEM

Table 1 shows there was increase in weight in therapeutic phase of the test drug group, when compared to the therapeutic phase of the control group, the observed increase was found to be statistically significant

Table – 02. Effect of *Aswakathri (Drynariaquercifolia* . Linn) on Relative food intake of albino rats

Effect on Relative Food Intake			
Groups	Preliminary phase	Therapeutic phase	% change
Control	5.04±0.63	4.03±0.40	--
Test drug	5.98±0.58	5.22±0.15*	29.53↑

Data : MEAN ±SEM

Table 2 shows there was increase in relative food intake in therapeutic phase of the test drug group, when compared to the therapeutic phase of the control group, the observed increase was found to be statistically significant.

Table – 03. Effect of *Aswakathri (Drynariaquercifolia* . Linn) on Absolute food intake of albino rats

Effect on Absolute Food Intake			
Groups	Preliminary phase	Therapeutic phase	% change
Control	18.23±1.75	14.80±1.07	--
Test drug	8.64±0.84	7.68±0.11**	48.11↓

Data : MEAN ±SEM, **P < 0.0001

Table 3 shows there was decrease in absolute food intake in therapeutic phase of the test drug group, when compared to the therapeutic phase of the control group, the observed decrease was found to be statistically extremely significant.

Table – 04.Effect of *Aswakathri (Drynariaquercifolia* . Linn) on Relative water intake of albino rats

Effect on Relative Water Intake			
Groups	Preliminary phase	Therapeutic phase	% change
Control	9.37±0.68	8.37±0.67	--
Test drug	12.99±1.19	12.48±1.60*	49.10↑

Data : MEAN ±SEM

Table 4 shows there was increase in relative water intake in therapeutic phase of the test drug group, when compared to the therapeutic phase of the control group, the observed increase was found to be statistically significant.

Table – 05. Effect of *Aswakathri (Drynariaquercifolia* . Linn) on Absolute water intake of albino rats

Effect on Absolute Water Intake			
Groups	Preliminary phase	Therapeutic phase	% change
Control	34.03±1.73	30.87±1.93	--
Test drug	18.8±1.81	18.37±2.22**	40.49↓

Data : MEAN ±SEM

Table 5 shows there was decrease in absolute water intake in therapeutic phase of the test drug group, when compared to the therapeutic phase of the control group, the observed decrease was found to be statistically very significant.

Table – 06. Effect of *Aswakathri (Drynariaquercifolia* . Linn) on Relative Urine output of albino rats

Effect on Relative Urine Output			
Groups	Preliminary phase	Therapeutic phase	% change
Control	1.05±0.37	0.74±0.18	--
Test drug	1.77±0.58	0.94±0.51 ##	27.03↑

Data : MEAN ±SEM

Table 6 shows there was increase in relative urine output in therapeutic phase of the test drug group, when compared to the therapeutic phase of the control group, the observed increase was found to be statistically not significant.

Data shows there was decrease in Relative urine output in therapeutic phase of the test drug group, when compared to the preliminary phase of the test group, the observed decrease was found to be statistically very significant.

Table – 07. Effect of *Aswakathri (Drynariaquercifolia* . Linn) on Absolute Urine output of albino rats

Effect on Absolute Urine Output			
Groups	Preliminary phase	Therapeutic phase	% change
Control	3.65±1.17	2.65±0.58	--
Test drug	2.57±0.82	1.37±0.72 ##	48.30↓

Data : MEAN ±SEM

Table 7 shows there was decrease in absolute urine output in therapeutic phase of the test drug group, when compared to the therapeutic phase of the control group, the observed decrease was found to be statistically not significant.

Data shows there was decrease in Absolute urine output in therapeutic phase of the test drug group, when compared to the preliminary phase of the test group, the observed decrease was found to be statistically very significant.

Table – 08.Effect of *Aswakathri (Drynariaquercifolia* . Linn) on Relative fecal output (wet) of albino rats

Effect on Relative Fecal Wet			
Groups	Preliminary phase	Therapeutic phase	% change
Control	1.40±0.09	1.14±0.08	--
Test drug	1.25±0.09	1.30±0.07	14.04↑

Data : MEAN ±SEM

Table 8 shows there was increase in relative fecal output wet in therapeutic phase of the test drug group, when compared to the therapeutic phase of the control group, the observed increase was found to be statistically not significant.

Table – 09.Effect of *Aswakathri (Drynariaquercifolia* . Linn) on Absolute fecal output (wet) of albino rats

Effect on Absolute Fecal Wet			
Groups	Preliminary phase	Therapeutic phase	% change
Control	5.19±0.46	4.25±0.33	--
Test drug	1.80±0.12	1.90±0.07**	55.29↓

Data : MEAN ±SEM , **P < 0.0001

Table 9 shows there was decrease in absolute fecal output wet in therapeutic phase of the test drug group, when compared to the therapeutic phase of the control group, the observed decrease was found to be statistically extremely significant

Table – 10.Effect of *Aswakathri (Drynariaquercifolia* . Linn) on Relative fecal output (Dry) of albino rats

Effect on Relative Fecal Dry			
Groups	Preliminary phase	Therapeutic phase	% change
Control	0.82±0.04	0.67±0.03	--
Test drug	0.90±0.07	0.89±0.03**	32.84↑

Data : MEAN ±SEM

Table 10 shows there was increase in relative fecal output dry in therapeutic phase of the test drug group, when compared to the therapeutic phase of the control group, the observed increase was found to be statistically very significant.

Table – 11.Effect of *Aswakathri (Drynariaquercifolia* . Linn) on Absolute fecal output (Dry) of albino rats

Absolute Fecal Dry			
Groups	Preliminary phase	Therapeutic phase	% change
Control	2.99±0.20	2.5±0.13	--
Test drug	1.29±0.10	1.31±0.04**	47.6↓

Data : MEAN ±SEM, **P < 0.0001

Table 11 shows there was decrease in absolute fecal output dry in therapeutic phase of the test drug group, when compared to the therapeutic phase of the control group, the observed decrease was found to be statistically extremely significant.

Table – 12.Effect of *Aswakathri (Drynariaquercifolia* . Linn) on Relative food conversion ratio of albino rats

Relative food conversion ratio			
Groups	Preliminary phase	Therapeutic phase	% change
Control	6.69 ± 1.19	6.16 ± 0.46	--
Test drug	7.07 ± 0.28	6.07 ± 0.14 #	1.46↓

Data : MEAN ±SEM

Table 12 shows there was decrease in Relative food conversion ratio in therapeutic phase of the test drug group, when compared to the therapeutic phase of the control group, the observed decrease was found to be statistically not significant

Data shows there was decrease in Relative food conversion ratio in therapeutic phase of the test drug group, when compared to the preliminary phase of the test group, the observed decrease was found to be statistically significant

Table – 13. Effect of *Aswakathri (Drynariaquercifolia* . Linn) on Absolute food conversion ratio of albino rats

Absolute food conversion ratio			
Groups	Preliminary phase	Therapeutic phase	% change
Control	6.69 ± 1.18	6.16 ± 0.46	--
Test drug	7.06 ± 0.28	6.07 ± 0.14 #	1.46↓

Data : MEAN ±SEM

Table 13 shows there was decrease in Absolute food conversion ratio in therapeutic phase of the test drug group, when compared to the therapeutic phase of the control group, the observed decrease was found to be statistically not significant

Data shows there was decrease in Absolute food conversion ratio in therapeutic phase of the test drug group, when compared to the preliminary phase of the test group, the observed decrease was found to be statistically significant

Table – 14. Effect of *Aswakathri (Drynariaquercifolia* . Linn) on Relative fecal water of albino rats

Relative fecal water			
Groups	Preliminary phase	Therapeutic phase	% change
Control	0.59 ± 0.08	0.47 ± 0.05	--
Test drug	0.36 ± 0.03	0.41 ± 0.06	12.76↓

Data : MEAN ±SEM

Table 14 shows there was decrease in Relative fecal water in therapeutic phase of the test drug group, when compared to the therapeutic phase of the control group, the observed decrease was found to be statistically not significant

Table – 15. Effect of *Aswakathri (Drynariaquercifolia* . Linn) on Absolute Fecal water of albino rats

Absolute fecal water			
Groups	Preliminary phase	Therapeutic phase	% change
Control	2.19 ± 0.33	1.75 ± 0.20	--
Test drug	0.51 ± 0.04	0.59 ± 0.07**	66.28↓

Data : MEAN ±SEM

Table 15 shows there was decrease in Absolute fecal water in therapeutic phase of the test drug group, when compared to the therapeutic phase of the control group, the observed decrease was found to be statistically extremely significant.

DISCUSSION ON VIPAKA STUDY

The outcome of the experimental study for *Vipaka* has been provided in the form of consolidated tables as follows for easy comparison and discussion.

Table. 16. Showing the Results of various parameters of *Vipaka* study in Wistar albino rats

Parameters of Test drug		
1	Relative Food intake	SI
2	Relative Water intake	SI
3	Relative Urine output	NSI
4	Relative Fecal wet	NSI
5	Relative Fecal dry	SI
6	Relative food conversion ratio	NSD
7	Relative fecal water	NSD
8	Body weight	SI

SI:– Significant increase, NSI:- Non-Significant
SD:- Significant decrease, NSD:- Non-Significant decrease

1. Effect on body weight: The rats of the trial group showed an increase in body weight after completion of the experimental phase as compared to the preliminary phase. Whereas the rats of the control group showed a decrease in the body weight after completion of the experimental phase as compared to the preliminary phase.
2. Effect of food consumption: There is a significant increase in the food consumption of the trial group in the therapeutic phase when compared with the food consumption of the control group in the therapeutic phase. Food consumption is taken as *AbhyavaranaShakthi*. *AbhyavaranaShakthi* is one of the factors to assess the *Agni* of an individual. Decreased *AbhyavaranaShakthi* indicates *Agnimandya* whereas increased A*bhyavarana Shakthi* indicates *Agnivridhi*. Here intake of Kashaya helped for the increase of *Agni*. *Agni vridhi* helps for *the dhathuparinama* and weight gain. In the present study also there is an increase in the bodyweight of the trial group, which may be to the increase of *Agni*. Hence it can be said that the *dravathva*of *pitta* is reduced here.
3. Effect on water intake: Water intake in the therapeutic phase of the trial group is significantly increased when compared to the therapeutic phase of the control group.
4. Effect on fecal output: Fecal output (dry and wet) was increased in the therapeutic phase of the trial group when compared with the therapeutic phase of the control group. In both cases wet and dry, there is an increase, but there is a highly significant increase in the fecal weight in the dry phase of the trial group, but only a non-significant increase in the fecal weight in the wet phase of the trial group. This indicates a greater percentage loss of liquid part in the stool of the trial group. Hence it can be said that the symptom of *pitta dravakshaya*.
5. Effect on urine output: Urine output was non significantly increased in the trial group during the therapeutic phase in comparison to the experimental phase of the control group.
6. Food conversion ratio: Here there is a non-significant decrease in the relative food conversion ratio of the trial drug in the therapeutic phase when compared with that of the control group. This parameter is considered important for ascertaining the *Pachana* property of the test drug. That is the enhancement of food assimilation. *Deepana* property is equated with food intake and *Pachana* is equated with Food conversion factor. The observed result shows that there is *Deepana*, which is increased food consumption but there is reduced *Pachana* property.

From the observations of the study, we found there increase in food intake, water intake, fecal output, urine output and there is an increase in the weight of the albino rats. Increased elimination of fecal and urine will contribute to either Madhura or AmlaVipaka. In Amlavipaka there is dhatukshaya. Here the Increased weight may be assumed due to an increase in the *dhatu* level. So based on the above criteria, here there is *mala vridhi* and *dhathuvridhi*, which are contributing to the *Madhuravipaka*. So rhizome decoction of the *Drynariaquercifolia* .Linn probably has *Madhuravipaka*.

CONCLUSION

A drug undergoes the final transformations in physicochemical nature after the process of the *vipaka*. The drug can only do its full effects after attaining *vipaka*. Thus to access the total effects done by a dravya we have to determine its *vipaka*. Considering this fact here, we have elaborated the *vipaka* study of an extra pharmacopeial drug *Drynariaquercifolia*.Linn.

From this preliminary assessment of *vipaka* with is achieved through the experimental study (Dhyani S.C., 2008),and with the available limited data, it may be concluded that the drug *Drynariaquercifolia*.Linn may possess *MadhuraVipaka*. These findings are to be taken as preliminary data. Based on these further clinical studies on human beings has to be conducted for amore accurate conclusion. Based on this study, it is suggested that studies that are more experimental in nature had to be conducted regarding the ayurvedic principles for getting the evidence-based data, especially in the case of drugs of extra pharmacopeial origin.

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