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Pharmacological Study of Chitraka Haritaki Avaleha & Shikhari Taila

Atara Achyuta [α], Manjusha R. [σ] & Nariya Mukesh [ρ]

Abstract- *Chitraka Haritaki Avaleha* is a *Leha Kalpana* (semisolid preparation of drugs, prepared with addition of jaggery & boiled with prescribed decoction) specifically indicated for oral use in treatment of nasal disorders in Ayurveda. *Shikhari Taila* is a formulated oil with the herbs having medicinal values. Use of *Shikhari Taila* for *Nasya* (one of the Panchakarma procedure mentioned in Ayurveda) in *Nasa Arsha* (Nasal polyposis) has been mentioned in Ayurvedic texts, but no work has been done at any of the PG centers of Ayurvedic Institutes. Both the drugs are having anti histaminic & anti inflammatory properties. Hence the present study was designed to ascertain whether it is possible to obtain experimental data to support the clinical study; and helps to prove the above theory, according to criteria of modern pharmacology too.

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I. Introduction

Pharmacology is an applied science. It forms the backbone of rational therapeutics, correct and skillful application of the drugs is impossible without a proper understanding of their basic Pharmacology. It is the science of drug action, has helped to elucidate many basic physiological and pathological mechanisms in health and disease. Various animal experimental models have been designed to study the effect of drugs on living organisms and isolated tissues. These give an insight about where and how a drug act, the mode of action of a drug, its effect on various body systems and probable adverse effects before administration of a drug. Therefore, the object of pharmacology is to provide such scientific data in animals as well as humans, which forms the basis of rational therapeutics.[i] Man occupies a supreme position among all the living creatures. Hence before administering drug to him it is desirable to experiment on other animals.

In the ancient Ayurvedic literature, lots of references are available regarding the testing of the drug and food on the animal for the safety of the mankind. The role of research in Ayurveda is not only to elucidate the principles of Ayurveda but also, to explain them in terms of modern parameters. A drug is accepted by modern science only if it has been proved safe by experimental studies. Drugs selected in the present study, i.e. *Chitraka Haritaki Avaleha* and *Shikhari Taila* are having anti histaminic & anti inflammatory properties.

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Hence the present study was designed to ascertain whether it is possible to obtain experimental data to support the clinical study; and helps to prove the above theory, according to criteria of modern pharmacology too. It was carried out with following aims & objects.

II. Plan of Study

Aims & Objectives

(1) To evaluate the Anti–inflammatory activity of *Chitraka Haritaki Avaleha* & *Shikhari Taila.*

(2) To evaluate the anti-histaminic activity of *Chitraka Haritaki Avaleha* & *Shikhari Taila.*

III. Materials and Methods

The animals for experimental study were obtained from animal house attached to pharmacology laboratory, IPGT and RA, Jamnagar, Gujarat, India. The experiment was carried out after obtaining permission from institutional animal ethics committee vide permission (IAEC/15/2013/38) as per CPCSEA guidelines. The dose for the experimental study was converted based on the body surface area ratio by referring table of Paget and Barnes (1969).

a) Test Drugs

i. *Source of Drug*

Both the formulations *Chitraka Haritaki Avaleha* and *Shikhari Taila* were prepared by the Gujarat Ayurved University Pharmacy for administration to the experimental animals. The *Avaleha* was taken and a stock solution was prepared freshly just prior to administration to animals by adding adequate quantity of water and used for the experimental purposes.

ii. *Test Drugs*

- *Chitraka Haritaki Avaleha.*
- *Shikhari Taila.*

b) Posology

i. *Dose Derivation*

Use of *Shikhari Taila* for *Nasya* in *Nasa Arsha* (Nasal polyposis) has been mentioned in *Chakradatta Nasarogaadhikara*.[ii] Dose for *Nasya* kept was 10 *Bindu*[iii] (drops) in each nostril for 6 sittings. *Chitraka Haritaki Avaleha* has been mentioned in *Chakradatta* [iv] & *Yogaratnakara* [v] in treatment of chronic rhinitis/sinusitis. Dose of *Chitraka Haritaki Avaleha* was 10 gms.[vi] twice a day for 3 months. Considering adult human dose of

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both samples, the dose for experimental study was calculated by extrapolating the human dose to animal dose based on the body Surface Area Ratio.

ii. *Dose calculation for Rat (in anti-inflammatory activity)*

The suitable rat dose was calculated by referring the table of Paget and Barnes (1969).

(1) *Test drug:* Chitraka Haritaki Avaleha

Rat dose = Adult human dose × 0.018 (conversion factor for rat weighing 200g)

= 20 g × 0.018

= 0.360 gm/ 200gm rat (360 mg/200gm rat)

= 1.8 gm/kg body weight

(2) *Test drug:* Shikhari Taila

Rat dose = 48 ml × 0.018 (conversion factor for rat weighing 200g)

= 0.864ml/ 200gm rat

= 4.3 ml/kg body weight

c) The Animals

i. *Animal Selection*

An overnight fasted Guinea pig was sacrificed to obtain fresh ileum for anti histaminic study & Charle's Foster albino rats of either sex weighing between 200 ± 35g were selected for anti inflammatory activity with the following conditions:

- *Husbandry conditions:* Standard husbandry conditions with ambient condition of temperature and relative humidity was maintained.
- *Diet:* Amrut brand rat pellet feed. Drinking water was given *ad libitum.*
- *Acclimatization period:* All the selected animals were kept under acclimatization for one week before experimentation.
- *Identification:* Animals were marked with saturated picric acid solution for proper identification.

ii. *Groups*

The rats were divided into three groups of six rats in each group;

- Group I (Water Control): Distilled water (10 ml/kg, po)
- Group II (Avaleha Group) 1.8 gm/kg, p.o.
- Group III (Taila Group) 4.3 ml/kg; p.o.

iii. *Route of drug administration*

The test drugs were administered through oral route for five days with the help of gastric catheter sleeved onto a syringe.

iv. *Statistical Analysis*

The data generated during the study was subjected to student 't' test Unpaired and Paired't' test used for assessing the significance of the results. Results within different groups at value of $P<0.05$ is considered as statistically significant.

d) Preparation

i. *Instruments Used*

Weighing balance, cotton, syringe, needle, catheters, centrifuge, refrigerator, plethysmograph, isolated organ bath, kymograph and other minor accessories.

ii. *Chemicals used*

- 1% carrageenan aqueous solution was used for anti-inflammatory study.
- Tyrode & Histamine solutions were used for anti histaminic study.

iii. *Experimental Model*

Experiment 1: Anti-inflammatory activity - Carrageenan induced paw oedema.

Experiment 2: Antihistaminic activity.

e) Anti-Inflammatory Activity

i. *Carrageenan induced paw oedema*

It is the basic test for screening anti-inflammatory effect. Carrageenan injection produces marked swelling of the paw and anti-inflammatory drugs are supposed to suppress this swelling. Method of Winter *et al.* (1962) was adopted to screen the anti-inflammatory activity of *Chitraka Haritaki Avaleha* and *Shikhari Taila* against carrageenan induced paw oedema in rats.

Rats were provided with food and tap water up to the start of the experiment. Initially left hind paw volumes up to the tibio-tarsal articulation were recorded by Using a Plethysmograph. The Plethysmograph employed, consists of 10 ml glass vessel (25 mm x 65 mm) fixed to 2 ml glass syringe through pressure tubing. About 5ml mercury was filled in the syringe and the mercury level was adjusted to zero mark on the micropipette. The space between the zero mark and the fixed mark of the glass vessel was filled with water and few drops of teepol. The initial level of fluid was adjusted and set at zero. The paw was immersed in water exactly up to the tibio-tarsal joint. The increased level of water in the glass vessel was adjusted to the prefixed mark by releasing the pressure of the connected syringe. The level where water and mercury interface in the micropipette was recorded as paw volume.

ii. *Procedure*

One hour after drug administration, oedema was produced by injecting 0.1 ml freshly prepared 1% carrageenan in sterile saline solution to the sub-plantar aponeurosis of the left hind limb. The rats were administered with the tap water in the dose of 2 ml/l00g body weight to ensure uniform hydration. This is supposed to minimize the variation in oedema formation. The paw volume is recorded at the interval of l hr, 2 hr, 3 hr and 6 hr.

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If the percentage of increase in paw volume is significantly less in test drugs administered groups in comparison to control group then the drugs were considered to possess anti- inflammatory activity.

f) Antihistaminic Activity

i. *Effect of test drug on the Guinea pig ileum (in vitro)*

There is increasing evidence that the airway epithelium may play an important role in airway inflammation, as disturbance of the epithelium, such as may occur on exposure to chemical, physical and immunological stimuli, can lead to the release of proinflammatory cytokines. There is an increased number of epithelial mast cells in nasal polyps. Total histamine levels in polyps are far higher than in other tissues (l00-1000 times that of plasma) .The release of histamine may be an important factor in causing plasma exudation. [vii] Because of this reason the test drugs were assessed for anti-histaminic property in isolated guinea pig ileum preparation.

ii. *Procedure*

This experiment was set-up following standard procedure. A Guinea Pig was sacrificed by cervical dislocation and a piece of ileum was excised out. It was set up in an isolated organ bath assembly following the standard procedure. The organ bath containing 40 ml of tyrode solution was maintained at 37 °C temperature and was aerated with oxygen. Tissue responses were recorded with an isotonic frontal writing lever system with 1:7 magnification and 500 mg initial tension on a smoked drum attached to a kymographic recording drum after 30 minutes of initial resting. Initially the dose responses were recorded with a standard spasmogenic drug i.e. histamine to select a dose producing sub-maximal response. Standard response was taken with histamine with a dose of 200 µg/ml of bath fluid. The effect of test drugs *per se* if any and the modulatory effect on the tissue response to histamine were recorded.

IV. Observation

a) Anti-Inflammatory Activity

Table 1: Anti-Inflamatory Effect of *Chitraka Haritaki Avaleha* & *Shikhari Taila* on Carrageenan Induced Paw Oedema in Albino Rats

Groups	Dose	% Increase in paw volume at different time interval after carrageen an injection					
		After 1 h	% change	After 3 h	% change	After 5 h	% change
Control	Q.S.	34.84 ± 4.27	--	78.54 ± 6.93		80.84 ± 7.75	--
Chitraka Haritaki Avaleha	1.8 gms/kg (360 mg/200gms)	22.33 ± 5.61	35.91↓	68.71 ± 7.67	12.51↓	64.99 ± 8.57	19.61↓
Shikhari Taila	4.32 ml/kg (0.864ml/ 200 gms)	29.612 ± 8.62 mine	15.01 ↓	83.88 ± 7.75	6.80	78.93 ± 8.06	2.36 ↓

Data: Mean ± SEM

***p> 0.01 in comparison to control group (Unpaired "t" test).

**P> 0.02. in comparison to control group (Unpaired "t" test).

Analysis from the above data reveals that *Chitraka Haritaki Avaleha* produced marked decrease (35.91%) in inflammation after one hour after carrageenan in comparison to control group. Shikhari Taila produced mild decrease (15.01%) in inflammation after one hour. Further *Chitraka Haritaki Avaleha* also produced mild decrease in inflammation after three hour (12.51%) and five hours (19.61%) of carrageenan in comparison to control group.

b) Antihistaminic Activity

i. *Effect of test drug on the Guinea pig ileum (in vitro)*

At the dose of 200 µg/ml of bath fluid were effective in inhibiting histamine (0.32µg/ml bath fluid) induced contraction of guinea pig ilium. *Shikhari Taila* produced almost 58% inhibition while *Chitraka Haritaki Avaleha* produced almost 32% inhibition of histamine induced ileum contraction. The kymographic recordings are provided in Fig- 01.

c) Compliance with Ethical standards

- *Funding:* This study was funded by Ministry of AYUSH. Grant in aid general of non- planned regular grant of IPGT & RA was provided for the present study.
- *Conflict of interest:* None.
- *Ethical approval:* The experiment was carried out after obtaining permission from institutional animal ethics committee vide permission (IAEC/15/2013/ 38) as per CPCSEA guidelines. The dose for the experimental study was converted based on the body surface area ratio by referring table of Paget and Barnes (1969).

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