

Dose Dependent Anti- Inflammatory Effect of Valeriana Wallichii in 0.1 ML Of 1% Carrageenan Induced Hind Paw Edema In Male Albino Rats

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Abstract:

Introduction: Inflammation is the complex biological response of vascular tissues to harmful stimuli, such as pathogens, damaged cells, or irritants. The importance of inflammation in progression of casual disease is one of the factors for use of anti-inflammatory drugs in clinical medicine. The main aim of this study is to assess anti-inflammatory properties of herbal plants. In this context, the present study was carried out to evaluate the dose dependent anti-inflammatory effect of Valeriana wallichii in rats using plethysmograph.

Objective: To study the dose related anti-inflammatory effect of Valeriana wallichii by using plethysmograph.

Methodology: A Randomized controlled trail was conducted in the Dept. of Pharmacology, Dr.Pinnamaneni Siddhartha Institute of Medical Sciences and Research Foundation (Dr.PSIMS & RF), Chinoutapalli, Krishna District, Andhra Pradesh with the institutional ethical committee clearance. All male albino wistar rats weighing between 250-300gm were selected for the study and rats were randomly divided into 4 groups (Control, Test-1, Test-2, test-3). Control group rats were administred 0.2ml of normal saline, whereas, test-1, test-2 and test-3 group rats were administered Valeriana wallichii in a dose of 40 mg/kg, 80 mg/kg, 120 mg/kg BW respectively, as single oral dose half an hour before injecting 0.1ml of 1% carrageenan to the sub-plantar region of hind paw and the paw edema of each rat was measured at 3hours.

Conclusion: Valeriana wallichii was found to be having anti-inflammatory property at dose of 40mg/kg and the anti-inflammatory property of Valeriana wallichii is increased with increasing dose. However the above preclinical experiments only give us an idea about the anti-inflammatory activity of Valeriana wallichii and large scale clinical trials are necessary for final assessment.

I. Introduction

Valeriana wallichii is a perennial herb that is widely distributed in various temperate regions of globe, including Asia, Europe, and North America. It is one of the most widely used herbal medicines in the world. Valerian root has been used in Europe as a mild sedative for many centuries and was also used to treat epilepsy in the 17th century and was regarded a good option for treating convulsions by some European physicians during 18th and 19th centuries, but it was later abandoned due to the uncertain chemical composition of various preparations, as well as its disagreeable odor and taste [1].

Indian valerian is an erect, perennial plant that grows to a height of 4 feet with pinnate, divided and heart-shaped leaves. Pink or white flowers are found in clusters on the leaf top. The roots are a hairy and spindly mass that is collected in the autumn from two-year old plants. The rhizomes are greenish-brown in color and hard and tough internally. The herb is cultivated in Belgium, England, Eastern Europe, France, Germany, the Netherlands, the Russian Federation and the United States of America. The medicinal plant is inhabitant to the Himalayas in Nagar, Minapin Glacier and Bultora Glacier in India. The herb is known as gilgiti valerian in Hindi and, mushkbala and rishawala in Urdu [2]. Inflammation is the complex biological response of vascular tissues to harmful stimuli, such as pathogens, damaged cells, or irritants. Inflammation is a complex reaction in tissues that consists mainly of response of blood vessels and leukocytes. It is a series of host responses directed as protective attempt by the organism to remove the injurious stimuli as well as initiate the healing process for the tissue [3]. The organisms require that they eliminate foreign invaders, such as infectious pathogens, and damaged tissues. These functions are mediated by a series of host responses called inflammation. Inflammation plays key role in maximum pathological conditions or diseases in the practice of medicine. The term inflammation is derived from the Latin word "inflammare" meaning to burn. Inflammation is a protective response, designed to get rid the organism of both the initial cause of cell injury (e.g. microbes, toxins) & the consequences of such injury (e.g. necrotic cells and tissues). The vascular and cellular reactions of inflammation are triggered by soluble factors that are produced by various cells (or) derived from plasma proteins and are

generated (or) activated in response to the inflammatory stimulus. The clinical features of inflammation were described in an Egyptian papyrus dated as early as around 3000 BC, The first four cardinal signs of inflammation were first enunciated by Celsus, a Roman writer and include Rubor (Redness), Tumor (Swelling), Calor (Heat), Dolor (Pain)^[4] The fifth sign; Functiolaesa (loss of function) was added by Rudolf Virchow in the 19th century. These signs are more prominent in acute inflammation than in chronic inflammation. Inflammation may contribute to a variety of diseases that are not thought to be primarily due to abnormal host responses. For instance, chronic inflammation may play role in atherosclerosis, type-2 diabetes, degenerative disorders like Alzheimer disease, cancer. In recognition of the wide-ranging harmful consequences of inflammation, the lay press has rather melodramatically referred to it as “the silent killer”. The importance of inflammation in progression of casual disease is one of factor for use of anti-inflammatory drugs in clinical medicine. More research on anti-inflammatory activity is done on allopathic drugs than on herbal drugs even though the herbal drugs are claimed to have less adverse effects.

Valeriana wallichii is useful in Ayurvedic medicine as an analeptic, antispasmodic, carminative, sedative, muscle relaxant, analgesic and also possess anti-inflammatory property^[4]. Terpenoids of Valeriana wallichii inhibit in vitro formation of PGE₁ and have suppressive effect on iNOS and COX-2 activity. Moreover, natural terpenoids are reported to be natural inhibitors of NF-κB signaling just like aspirin and other NSAIDs^[5].

Hence, the present study was carried out to evaluate the dose dependent anti-inflammatory effect of Valeriana wallichii by using plethysmograph.

II. Materials & Methods

A Randomized controlled trail was conducted in the Dept. of Pharmacology, Dr.Pinnamaneni Siddhartha Institute of Medical Sciences and Research Foundation (Dr.PSIMS), Chinoutapalli, Krishna District, Andhra Pradesh with the institutional ethical committee clearance.

All male albino wistar rats weighing between 250-300gm were selected for the study and rats were randomly divided into 4 groups (Control, Test-1, Test-2, Test-3) and each group contains 6 rats. A mark is made at the ankle joint (tibio-tarsal joint) of each rat and Initial paw edema of each rat was measured before giving drug carrageenan^[6] by using plethysmograph^[7]. Control group rats were administered 0.2 ml of normal saline, orally half an hour before injecting 0.1ml of 1% carrageenan to the sub-plantar region of the hind paw and the paw edema of each rat was measured at 3 hours.

Test-1 group rats were administered 40mg/kg BW of Valeriana wallichii as single oral dose half an hour before injecting 0.1ml of 1% carrageenan to the sub-plantar region of hind paw and the paw edema of each rat is measured at 3hours. Test-2 group rats were administered 80mg/kg BW of Valeriana wallichii as single oral dose half an hour before injecting 0.1ml of 1% carrageenan to the sub-plantar region of hind paw and the paw edema of each rat is measured at 3hours. Test-3 group rats were administered 120mg/kg BW of Valeriana wallichii as single oral dose half an hour before injecting 0.1ml of 1% carrageenan to the sub-plantar region of hind paw and the paw edema of each rat is measured at 3hours.

Chemicals and Solutions: Carrageenan, Double distilled water, Normal saline, Valeriana wallichii (Tagara capsules)

Animals:- Male Albino wistar rats weighing about 250-300gm.

Equipments:- Plethysmograph (MKM)^[8], Mercury, Insulin syringes, Tuberculin syringes, Infant feeding tube, Hypodermic syringes, Measuring jar, Glass beakers, Animal weighing balance, Animal cages, Cotton, Spirit, Stopwatch, Glass rod, Disposable needles.

Carrageenan induced paw oedema model

Carrageenan is a widely used irritant or inflammogen to study the acute and sub acute phases of inflammation in rats. Chemically, it is a sulphated polysaccharide obtained from sea weed (rhodophyceae). The experimental tissue injury caused by this irritant initiates a cascade of inflammatory events leading to formation of exudates. The inflammation induced by it is biphasic in nature. The first phase is attributed to the release of histamine, 5-hydroxy tryptamine (serotonin) and kinin while the second phase is related to the release of prostaglandins. The well-recognized method of winter et al, 1962^[9] is followed.

A 1% w/v suspension of carrageenan is prepared freshly in normal saline and injected into sub planter region of left hind paw (usually 0.1ml in rats). In control group animals only vehicle is injected. Test drug is usually administered intraperitoneally (or) orally, according to body weight, half an hour before the carrageenan challenge. In this study test drug was given orally. A mark is made at the ankle joint (tibio-tarsal joint) of each rat. Paw edema volume up to the ankle joint is measured in drug treated and untreated groups at 0 and at 3hours following carrageenan challenge by using mercury Plethysmograph filled with mercury and % of reduction in

edema is calculated Data was collected, compiled and analyzed using SPSS-V19. Statistical tools applied were means, SD, Percentages and t-Test.

III. Results And Discussion

Table-1: Volume of paw edema in (ml) in four groups at 0 and 3hrs

Group	Drug	Dose	Paw edema in ml		Paired t-value	P-value	Inference
			At 0 Hrs Mean ± SD	At 3 Hrs Mean ± SD			
Control	Normal Saline	0.2ml	0.42±0.08	1.22±0.16	21.91	<0.01	HS
Test-1	Valeriana wallichii	40 mg/kg	0.38±0.08	1.02±0.04	19	<0.01	HS
Test-2	Valeriana wallichii	80 mg/kg	0.4±0.06	0.92±0.12	16.81	<0.01	HS
Test-3	Valeriana wallichii	120 mg/kg	0.4±0.06	0.82±0.04	13.56	<0.01	HS

HS-Highly Significant

In case of normal saline, the volume of paw edema at 0 Hrs was 0.42 ml and increased to 1.22 ml after 3 Hrs. In case of Valeriana wallichii 40 mg/kg, the volume of paw edema at 0 Hrs was 0.38 ml and increased to 1.02 ml after 3 Hrs. In case of Valeriana wallichii 80 mg/kg, the volume of paw edema at 0 Hrs was 0.4 ml and increased to 0.92 ml after 3 Hrs. In case of Valeriana wallichii 120 mg/kg, the volume of paw edema at 0 Hrs was 0.4 ml and increased to 0.82 ml after 3 Hrs.

Table-2: Increase in mean paw edema volume after 3 hrs

Group	Mean	SD
Normal saline	0.80	0.09
Valeriana wallichii 40 mg/kg	0.63	0.08
Valeriana wallichii 80 mg/kg	0.52	0.08
Valeriana wallichii 120 mg/kg	0.42	0.08

Increase in mean paw edema volume after 3 Hrs was 0.80 ml in case of normal saline, 0.63 ml with Valeriana wallichii 40 mg/kg, 0.52 ml with Valeriana wallichii 80 mg/kg, and 0.42ml with Valeriana wallichii 120 mg/kg.

Table-3: Comparison between the groups regarding paw edema volume inhibition.

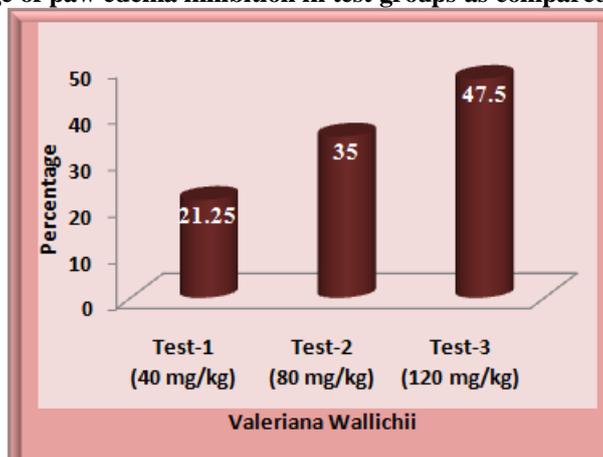
Comparison	% Decrease	Independent t-value	P-value	Inference
Control vs Test-1	21.25	2.42	<0.01	HS
Control vs Test-2	35	4.6	<0.01	HS
Control vs Test-3	47.5	6.3	<0.01	HS

HS-Highly Significant

The percentage inhibition of paw edema in rats treated with Valeriana wallichii 40 mg/kg was 21.25% in comparison with normal saline. The percentage inhibition of paw edema in rats treated with Valeriana wallichii 80 mg/kg was 35% in comparison with normal saline. The percentage inhibition of paw edema in rats treated with Valeriana wallichii 120 mg/kg was 47.5% in comparison with normal saline.

Hence it shows that anti-inflammatory effect of Valeriana wallichii is more with increasing doses of 40 mg/kg, 80 mg/kg, 120 mg/kg BW respectively

Fig-3: Percentage of paw edema inhibition in test groups as compared to control group.



IV. Summary And Conclusion

Valeriana wallichii was found to be having anti-inflammatory property at dose of 40mg/kg. Anti-inflammatory property of Valeriana wallichii was increased with increasing dose, which was statistically highly significant. The percentage decrease of edema in rats treated with Valeriana wallichii 120 mg/kg was 47.5%, whereas it is only 35% and 21.2% in rats treated with Valeriana wallichii 80 mg/kg and 40 mg/kg respectively. However the above preclinical experiments only give us an idea about the anti-inflammatory activity but large scale clinical trials are necessary for final assessment.

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