

# Antihypertensive effect of aqueous extract of *Elaeocarpus ganitrus* Roxb. seeds in renal artery occluded hypertensive rats

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**Abstract:** Aqueous extract of *Elaeocarpus ganitrus* Roxb. seeds powder (Family: Elaeocarpaceae) was evaluated for its antihypertensive activity in renal artery occluded hypertensive rats. Male Wistar rats (180-200g) were pretreated with aqueous extract of *E. ganitrus* for 6 weeks. Hypertension was induced in animals by clamping the renal artery with renal bulldog clamp for 4 h. Ischemia of the kidneys causes elevation of blood pressure by activation of the renin-angiotensin system. Elevated blood pressure of the animals was significantly ( $p<0.05$ ) decreased by the aqueous extract of *E. ganitrus* at the dose levels of 25, 50 and 100mg/kg, *i.v.* Captopril, angiotensin converting enzyme inhibitor (ACE-I) at the dose of 1 mg/kg, *i.v.* showed significantly ( $p<0.05$ ) reduced in the elevated blood pressure. The antihypertensive activity of aqueous extract of *E. ganitrus* may be due to the action on rennin-angiotensin system.

**Key words:** *Elaeocarpus ganitrus*, antihypertensive activity, rennin-angiotensin system.

## Introduction

Cardiovascular diseases account for 12 million deaths, annually worldwide and are known to be number one group of 'killer disease'. Hypertension is one of the leading causes of disability, mortality, and morbidity along the populace. It is the most common chronic illness among the world faces.<sup>1,2</sup>

Hypertension is the most common cardiovascular diseases and constitutes a major factor for several cardiovascular pathologies including atherosclerosis, coronary artery disease, myocardium infarct, heart failure, renal insufficiency, stroke and dissecting aneurysm of aorta.<sup>3</sup>

An elevated arterial pressure is an important public health issue in developed countries. Although it is common, asymptomatic and readily detectable but it can often lead to lethal complication, if left untreated. Because of high incidence and morbidity, various drugs and regimes have

been advocated for the control of hypertension. Many new drugs have been introduced which may demonstrate better efficacy but possess side effects. Recently attention has been focused towards herbal and mineral preparations which are traditionally used as potential therapeutic agents in the prevention and management of cardiovascular diseases.<sup>4</sup>

*Elaeocarpus ganitrus* commonly known as 'Rudraksha' in Sanskrit and 'Rudraki' in Hindi is grown in the Assam and Himalayan region of India for its attractive fruit stones and medicinal properties.<sup>5</sup> It is used in folk medicine in the treatment of stress, anxiety, depression, palpitation, nerve pain, epilepsy, migraine, lack of concentration, asthma, hypertension, arthritis and liver diseases.<sup>6,7</sup> According to the Ayurvedic medical system, wearing of Rudraksha can have a positive effect on heart and nerves.

Ethanol extract of the fruit of *E. ganitrus* have been reported to exhibit sedative, hypnotic, tranquillizing, anticonvulsive, antiepileptic and antihypertensive properties.<sup>7, 8, 9</sup> The ethanolic extract of leaves of *E. ganitrus* yielded quercetin, gallic and ellagic acids, ( $\pm$ ) elaeocarpine, ( $\pm$ ) iso-elaecarpine<sup>10</sup> and rudrakine.<sup>11</sup> Though there were scanty reports available on its antihypertensive activity, its mechanism was still unknown. Hence, present work was undertaken to investigate its mechanism of action of antihypertensive effect of aqueous extract of *E. ganitrus* in renal artery induced hypertensive rats.

## Materials and method

### Plant Material and reagents

Seeds powder of *E. ganitrus* of batch number 013CS was supplied by the Rudraksha Research and Testing Laboratory Goregoan, Mumbai. A voucher specimen (2007/03/10) has been kept in our laboratory for future reference.

### Preparation of plant extract

The dried powdered seeds of *E. ganitrus* were kept for maceration with distilled water for 24 h. Extracts was filtered through vacuum filter and the filtrate was concentrated in vacuum evaporator. Dried extract

### Experimental animals

Male Wistar rats (180–200g) and Swiss albino mice (20–22g) of either sex were purchased from National Toxicology Centre (NTC), Pune. They were housed in polypropylene cages in a controlled room temperature  $22\pm 1^\circ\text{C}$  and relative humidity of 60–70%. They were kept under standard conditions of 12/12 h light and dark cycle. The animals were maintained with standard pellet diet (Chakan Mill, Pune) and water *ad libitum*. The animals were acclimatized to laboratory condition for seven days before commencement of experiment. All studies were carried out using 6 rats in each group. Ethical clearance was obtained from Institutional Animal Ethical Committee (IAEC).

### Chemicals and instrument

Urethane was purchased from Hi media Pvt. Ltd. India. Captopril (ACETEN, WOCKHARDT), 12.5mg tablet was procured from local market. All the chemicals used for the study were of analytical grade. Eight-channel recorder powerlab (AD Instruments) system was used for the measurement of blood pressure.

### Phytochemical evaluation

Aqueous extract of *E. ganitrus* was studied for its phytoconstituents such as alkaloids, steroid and/or triterpenoids and their glycosides, tannins, flavonoids and their glycosides, carbohydrates and cardiac glycosides using different phytochemical tests.<sup>12</sup>

### Acute oral toxicity study

Acute oral toxicity study of aqueous extract *E. ganitrus* was carried out in Swiss albino mice of either sex (20–22 g) according to OECD guidelines no 423. Extract at different doses up to 2000 mg/kg, *p.o.* was administered and animals were observed for behavioral changes, toxicity and mortality up to 48 h.<sup>13,14</sup>

### Antihypertensive activity<sup>15,16</sup>

Male wistar rats were divided into the 6 groups each group had six animals. Animals in normal control and negative control groups received distilled water. Aqueous extract of *E. ganitrus* was administered orally at the dose levels of 250, 500 and 1000mg/kg to the treatment groups for six week. At the end day of treatment, animals were anaesthetized by intraperitoneal injection of 1.25 gm/kg of Urethane. A small incision was given on the left side of peritoneal cavity of the animal to expose left kidney. The renal artery was occluded for the 4 h by using renal bulldog clamp. The jugular vein was cannulated for the administration of test drug. The carotid artery was cannulated to measure the blood pressure and connected to the blood pressure transducer of power lab eight channel recorder powerlab. After stabilization blood pressure, the renal bulldog clip was removed. Then 1/10<sup>th</sup> of the administered dose of the *E. ganitrus* aqueous extract, i.e. 25, 50, and 100 mg/kg was given respectively through jugular vein and mean arterial blood pressure (MABP) was measured at different time intervals (5,15,30,60 min). MABP of normal control groups were recorded without clamping the renal artery. Captopril 1mg/kg, *i.v.* was used as a standard. Changes in blood pressure of treated groups were compared with negative control.

### Statistical Analysis

The results are expressed as the mean $\pm$  SEM for each group. Statistical differences were evaluated using a one-way analysis of variance (ANOVA) followed by Dunnett's test. Results were considered to be statistically significant at  $p<0.05$ .

### Results

The phytochemical evaluation of aqueous extract of *E. ganitrus* revealed the presence of alkaloids, glycosides, steroids and flavonoids. Animals treated with aqueous extract *E. ganitrus* didn't showed any behavioral changes, toxic reaction or mortality. The extract was found to be safe at the dose of 2000mg/kg.

Removal of renal bulldog clip in the negative control group resulted in significant ( $p<0.05$ ) increase in MABP (Table 1).

Pretreatment of animals with *E. ganitrus* aqueous extract 25, 50 and 100 mg/kg *i.v.* showed significant decrease ( $p<0.05$ ) in the MABP at different time intervals (Table 1). Captopril (1mg/kg *i.v.*) produced significant reduction

in MABP. The hypotensive effect was maximum after 60 min.

### Discussion

Present study revealed the significant antihypertensive activity of aqueous extract of *E. ganitrus* in renal artery occluded hypertensive rats. The occlusion of renal artery upto 4 hour, leads to cause kidney ischemia. Ischemia of the kidneys causes elevation of blood pressure by activation of the renin-angiotensin system. The procedure can be used for acute and chronic hypertension. Acute renal hypertension can be induced in rats, by clamping the left renal artery for 4 h. After reopening of the vessel, accumulated renin is released into circulation.<sup>15</sup>

Renin acts on angiotensinogen (renin substrate), an  $\alpha_2$ -globulin to release the decapeptide angiotensin I. This decapeptide is cleaved by angiotensin converting enzyme (ACE) to yield the active angiotensin II (octapeptide) which is a potent vasoconstrictor leading to hypertension. Angiotensin II undergoes hydrolysis by an aminopeptidase to yield the heptapeptide angiotensin III which is also active. Further cleavage yields to peptides with little activity.<sup>17</sup>

The protease renin catalyzes the first and rate-limiting step in the formation of angiotensin II leading to acute

hypertension. The test is used to evaluate antihypertensive activities of drugs.<sup>15</sup>

The results showed that, intravenous injection of aqueous extract of *E. ganitrus* seeds significantly ( $p < 0.05$ ) decreased the elevated blood pressure in dose dependent manner. Captopril, ACE-I at the dose of 1 mg/kg, *i.v.* showed the significant ( $p < 0.05$ ) decrease in the elevated blood pressure. The extract differed from captopril in respect of the potency. After a sharp fall in MABP at 5 minute a stable baseline was observed whereas progressive decrease in MABP was shown by captopril. The drastic fall in MABP after 1 hour may precipitate reflex tachycardia and compensatory increase in sympathetic tone. The extract appears to be free from such hypertensive effect. The antihypertensive activity of aqueous extract of *E. ganitrus* may be resulted through the action on rennin-angiotensin system.

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**Table No.1: Effect of aqueous extract of *E. ganitrus* on renal artery-occluded hypertensive rats.**

Groups	Mean Arterial Blood Pressure (MABP) in mmHg at different time interval					
	Treatment (mg/kg)	MABP after removing clip	5 min	15 min	30 min	60 min
Normal control	Distilled water, <i>p.o.</i>	-----	83.33±5.92	85.83±4.89	84.83±6.31	86.83±5.89
Negative control	Distilled water, <i>p.o.</i>	127.45±6.35	117.16±4.57 <sup>@</sup>	119.83±2.89 <sup>@</sup>	106.33±4.85 <sup>@</sup>	103.33±6.19 <sup>@</sup>
<i>E. ganitrus</i>	25, <i>i.v.</i>	123.56±5.26	74.00±5.78*	75.33±4.44*	71.33±5.91*	69.00±4.81*
<i>E. ganitrus</i>	50, <i>i.v.</i>	115.76±6.53	71.66±4.35*	69.50±4.66*	67.50±4.86*	71.33±6.64*
<i>E. ganitrus</i>	100, <i>i.v.</i>	117.53±3.56	62.50±6.62*	62.50±7.59*	60.16±6.21*	60.50±6.97*
Captopril	1, <i>i.v.</i>	122.89±4.89	49.66±2.34*	39.16±4.08*	34.50±3.86*	26.83±3.50*

Values in the results are expressed as mean± SEM, (n=6), <sup>@</sup>  $p < 0.05$  significantly different in comparison with Normal control, \* $p < 0.05$  significantly different in comparison with Negative control

### References

1. Akinkigbe O., Current epidemiology of hypertension in Nigeria, In the achieves of Ibadan Medicine, Int J Med Sci., 2001, 1, 1.
2. Schutte A., Van Rooyen J., Huisman H., Krunger H., Malan N De J., Dietary risk markers that contribute to the etiology of hypertension in black South African children, The THUSA BAMMA study, J Hum Hypertens., 2003, 17, 29-35.
3. Oparil S., Treating multiple risk hypertensive populations, Am. J. Hypertens., 1999, 12, 121-129.
4. Bhatt J. D., Panchakshari U. D., Hemavathi K.G., Gulati O. D., Effect Of Abana, An Ayurvedic Preparation On Ethinyl Stradiolinduced Hypertension In Rats, Indian J Pharmacol., 1998, 30, 399-403.

5. Asolkar L. V., Kakkar R. R., and Chakre O. J., In Second Supplement to Glossary of Indian Medicinal Plants with Active Principles, Part I (1965–1981), PID & CSIR, New Delhi, India, 1992, 289.
6. Khare C. P., Encyclopedia of medicinal plants, Spring publication, New York, 2004, 198.
7. Dasgupta A., Agrawal S. S., and Basu D. K., Anticonvulsant activity of the mixed fatty acids of the *Elaeocarpus ganitrus* Roxb., Indian J of Phyiol. Pharmacology, 1984, 28, 245-286.
8. Bhattacharya S. K., Debnath, P. K., Pandey V. B. and Sanyal A. K., Pharmacological investigation on *Elaeocarpus ganitrus*, Planta Med., 1975, 23, 174–177.
9. Pandey V. B., and Bhattacharya S. K., Scientific appraisal of rudraksha (*Elaeocarpus ganitrus*): chemical and pharmacological studies, J. Res. Educ. Indian Med., 1985, Jan-June, 47–50.
10. Lal C., Tabulated phytochemical reports, Phytochemistry, 1975, 14, 2727–2728.
11. Ray A. B., Lal Chand and Pandey V. B., Rudrakine: A new alkaloid from *Elaeocarpus ganitrus*, Phytochemistry, 1979, 18, 700–701.
12. Farnsworth N. R., Biological and phytochemical screening of plants, J Pharm Sci., 1966, 55, 225-76.
13. Ghosh M. N., Toxicity studies, Fundamental of Experimental Pharmacology, Calcutta, India, Scientific book agency, 1984, 2, 153-158.
14. Shetty A. J., Shyamjith, Deepa, Alwar M. C., Acute toxicity studies and determination of median lethal dose, Current science, 2007, 93, 917-920.
15. Vogel G. H., Vogel W. H., (Eds.), Cardiovascular activity: Drug Discovery and Evaluation, Pharmacological Assays, 2<sup>nd</sup> Ed, Springer, USA, 1997, 172.
16. Ofem O. E., Eno A. E., Imoru J., Nkanu E., Unoh F., Ibu J. O., Effect of crude aqueous extract of *Viscum album* (mistletoe) in hypertensive rats, Indian J Pharmacol, 2007, 39 (1), 15-19.
17. Alfered G. G., Theodore W. R., Alan S. N., Palmer T., The pharmacological basis of Therapeutics, Cardiovascular Drugs, 1990, VIII<sup>th</sup> Ed, vol-I, 769-763.

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