Protective Effect of Methanolic Extract of Nardostachys Jatamansi Rhizomes on Aspirin plus Pylorus Ligation-Induced Gastric ulcer in Experimental Animals

Devarakonda Krishna Prasad[1], Bairam Ravindar[2], Vasam Mallikarjun[3], Cherukupally Srinivas[4], Shanmugarathinam Alagarsamy[5], Vasudha Bakshi[1], Kola Venu[2]

[1]School of Pharmacy, Anurag University, Hyderabad, Telangana, India.
[2]Department of Pharmacy, Srikrupa Institute of Pharmaceutical Sciences, Telangana, India.
[3]Department of Pharmacy, Chaitanya Deemed to be University, Telangana, India.
[4] Department of Pharmacy, Samskruthi College of Pharmacy, Telangana, India.
[5] Departments of Pharmacy, Anna University, Tiruchirappalli, India.

Abstract

Ulcer is the condition of stomach mucosa continuity. Peptic ulcer is caused because of imbalance between defensive factors and aggressive factors. The herbaceous crop of *Nardostachys jatamansi* DC (Valerianaceae family) is the herbal agent whose therapeutic herbal agent is its rhizome. The gastro protective activities of MENJ" methanolic extract of *Nardostachys jatamansi* rhizomes" utilizing aspirin plus pylorus ligation model should be evaluated. *Nardostachys jatamansi's* plant material has been gathered, dried under shade, powered and extracted with soxhlet soft methanol apparatus. The extract has been tested for flavonoids (Shinoda test), tannins (Gelatin test) and alkaloids (Mayer's and Wagner's test). The rats have been categorized into 4 groups; MENJ 100mg/kg and 200mg/kg, standard (Omeprazole 10mg/kg), control (0.3% CMC). Aspirin (200mg/kg) has been orally administered one time for five days. MENJ has been administered on 6th day for testing group as well as Omeprazole 30minute before pylorus ligation. The animals have been sacrificed after 4 hours, stomach has been excised and after that gastric juice has been gathered, centrifuged as well as biochemical studies such as ulcer and proteins index, total acidity, free acidity, gastric volume, and pH have been performed. A significant($p<0.001$)rise was seen in pH (4.46±0.37) where a significant decrease was seen in total proteins (283.42±15.49), total acidity (53.42±1.26), free acidity (29.46±0.85),gastric volume (2.92±0.25),and ulcer index (1.72±0.48) after comparing with control. In contrast with Omeprazole the test dosage of 200 mg/kg has also been found to have substantial effect ($p<0.01$). The current research shows the involvement of flavonoids, tannins and alkaloids as a gastro protective behaviour of MENJ.

Key words: *Nardostachys jatamansi*, Ulcer index, Aspirin plus Pylorus ligation, Gastro protective activity.

DOI Number: 10.14704/nq.2022.20.11.NQ66132 **NeuroQuantology 2022; 20(11): 1419-1424**

Introduction

Different herbal medicines are now generally accepted for different disorders. A plant can be regarded as a biosynthetic laboratory for a number of compounds including saponins, volatile oils, flavonoids, alkaloids, and glycosides which exert a physiological effect as well as for the chemical compounds, for example lipids, proteins, and carbohydrates, which are used as feed by humans. The therapeutic effects are normally caused by secondary metabolites (Mohod SM *et al*., 2011).*Nardostachys jatamansi* is a medicinal plant and its rhizome is the therapeutic herbicide of the Valerianaceae family. It has been documented that *Nardostachy Jatamansi* has various therapeutic activities such as cardio protective, hepatoprotective, antioxidant, antimicrobial, and anti-fungal properties. This is utilized in treatment of CNS and insomnia diseases. Inhibition activities of the plant were also recorded for the

spasmolytic, bronchodilator, and vasodilator as well as platelet aggregation. The key goal of present research was for evaluating Methanolic Extract of *Nardostachys Jatamansi* Rhizomes" gastro protective activity utilizing aspirin plus pylorus ligation model. In several cases, peptic ulcers etiology is not known, however, this is usually believed that these findings from an imbalance among the aggressive factors as well as the maintenance of mucosal integrity by the endogenous defense procedures. Various therapeutic agents are utilized in order to restore equilibrium by raising mucosal production, stabilize the surface epithelia cells or impeding prostaglandin synthesis to avoid gastric acid secretion or in order to improve mucosal protection mechanisms. Such secondary metabolites are very effectual as immune stimulating, anti-inflammatory, anti-ulcer, antineoplastic, and antioxidant agents (Paguigan ND *et al*., 2014; Repetto MG *et al*., 2002; Falcão HDS *et al*., 2008; Borrelli F *et al*., 2000; Reddy VP *et al*., 2012).

Materials and Methods

Plant Material

In the local region, *Nardostachys jatamansi* rhizomes have been collected and shaded at room temperatures and after that reduced in size to a fine powder form with the aid of a mixer grinder. The image of the *Nardostachys jatamansi* rhizomes has been showed in **Fig.1**

Fig 1 The image of the *Nardostachys jatamansi* rhizomes

Preparation of Methanolic Extract

The succus of the plant prepared from ~200g of fresh plant floors was dry and dissolved overnight in 100 mL of 100% methanol (MEOH). Extracts (50 mL), evaporated to dryness, were subsequently conveyed to the clean vessels and dimethyl sulfoxide (DMSO) was redissolved to achieve a final concentration of around 10mg/mL (Kumar AK., *et al* 2010 Krishna Prasad D., *et al* 2015).

Experimental animals

For experimental purposes, both albino rats (Wistar strain) of 150gm-200gm weight and Albino mice of either sex between 16-25g have been obtained from the breeder. Since acquisition, all animals have been acclimatized in normal manufacturing conditions of 26 ± 20C for seven days at room temperature and have preserved the light-/dark period at 12:12 h with relative humidity 45-55%. The animals have been fed Amrut Laboratories synthetic regular diet (Pranava Agro Industries Ltd. Sangli.). Water was permitted *ad libitum* and strict hygienic conditions have been maintained.

Chemicals and drugs

The chemicals used for anti-arthritic study were - Distilled water [Mysore petro chemicals, Raichur, India], Aspirin [Dr. Reddy's laboratories, Hyderabad] Anesthetic ether (Sigma Solvents and Pharmaceuticals-Mumbai. All the chemicals as well as drugs used were of Pharmaceutical grade.

Determination of acute oral toxicity (LD_{50})

In female albino mice (16-25g), an analysis of acute oral toxicity [8] of S. *indica* fruit extracts was found to be performed in normal husbandry conditions. The animals were

eISSN 1303-5150

rapidly fasting 4 hours before the evaluation and CPCSEA procedure for acute toxicity tests was taken up and down (OECD guidance No. 425). During the 48-hour study cycle, animals were treated with individual dosages and observed for their mortality (short term toxicity). The doses for the next animals have been assessed based on the short-term extract profile. Long-term toxicity was found for all animals (7 days). The LD_{50} studies of the test extracts were conducted up to the maximum dose level of 2000mg/kg body wt. $1/20^{th}$, $1/10^{th}$ and $1/5^{th}$ doses of the LD_{50} dose of the individual extracts have been chosen for the research as low, medium and high doses (Venu K *et al*., 2019). The protocol was approved by "Institutional Animal Ethical Committee with CPCSEA Number"– VIPS/IAEC/2017/14.

Aspirin plus pylorus ligation induced gastric ulcer in rats

Rats have been categorized into 4 groups; MENJ 100mg/kg and 200mg/kg, standard (Omeprazole 10mg/kg), control (0.3% CMC). Aspirin (200mg/kg) has been orally administered one time for five days. MENJ has been administered on 6th day for testing group and Omeprazole 30minute before pylorus ligation. The animals have been sacrificed after 4 hours, stomach has been excised and after that gastric juice has been gathered, centrifuged as well as biochemical studies such as ulcer and proteins index, total acidity, free acidity, gastric volume, and pH have been performed (Shay H *et al*., 1945; Ganguly AK *et al*., 1973; Kulkarni SK *et al*., 1999;).

Data analysis

All three models displayed the obtained values as a mean ± SD for seven species, and were subject to statistical analysis with the help of a one way ANOVA and the Dunnett's-'t' -test for determining important discrepancies between classes. P<0.05*, 0.01**and 0.001*** has been found substantial.

Results and Discussion

Study of Gastro Protective Effect on Aspirin Plus Pylorus Ligation Method.

Different herbal remedies are also generally known for different diseases. A plant can be considered not just for chemical compounds but a biosynthetic laboratory for example lipids, proteins, and carbohydrates, which are used as food by man but also for a multitude of compounds for example saponins, volatile oils, flavonoids, alkaloids, and glycosides that exert a physiological effect. The medicinal impact compounds are typically the secondary metabolites.

The plant content can be phytochemically preliminary tested for different components of the plant. Herbal medicines and their derivatives were used in several countries to cure different illnesses as an alternative to allopathic medicines. While herbal medicines are usually utilized in several ailments treatment, very few scientific studies have been conducted on herbs to gather information on their effectiveness and protection. Initial safety test analysis of the drug. Acute toxicity provides us with the basis for the definition and labelling. It also includes initial information on a substance's mechanism of toxic action, from which a dosage of a new drug can be calculated for animal experiments and dose determinations.

Flavonoids are suspected to increase the number of mucosal prostaglandins, decrease histamines through inhibition of histidine decarboxylase, from mast cells, inhibit the production of Helicobacter pylori, function as free radical scavengers and inhibit H+/K+-ATPase (Repetto MG *et al*., 2002; Sharath SS *et al*., 2015) Saponins can cause defensive factors of the mucous membrane, and tannins may, for example, decrease the permeability of the outermost mucosa layer through chemical irritation. Moreover,

alkaloid and terpenoids compounds also have high gastric ulcers activity. (Klein-Júnior LC *et al*., 2012; Mitra P et al., 2014).

The cytoprotective effects of *Nardostachys* are demonstrated by reduction of ulcer index and overall acidity, pepsin and protein in the gastric fluid, as well as by increased mucin content. The *Nardostachys jatamansi* was reported with anti-ulcer activity due to the incidence of tannins, saponins, alkaloids. In aspirin plus pylorous ligation induced gastric ulcer the Methanolic extract of *Nardostachys jatamansi rhizomes* where a significantly ($p<0.001$) rise was seen in pH (4.05±0.22) where a significant decrease was seen in total proteins (312.64±6.00), total acidity (59.12±1.79), free acidity (31.96±2.31), gastric volume (3.14±0.14), and ulcer index (2.12±0.35) after comparing with control. In contrast with Omeprazole, the 200 mg/kg test dosage also demonstrated a meaningful effect ($p<0.01$). The detailed results of % of protection were given in tabulated when compared with control as well as standard drug i.e. **Table 1** and their graphs were shown in **Figure 2 and 3.**

Table 1: Effect of Methanolic extracts of *Nardostachys jatamansi rhizomes* on Various Parameters

Treatment and Dose	Gastric volume (ml)	pH	Ulcer Index (UI)	Free Acidity (meq/l/ 100g)	Total Acidity (meq/l/ 100g)	Total proteins (µg/ml)	% of Protection
Control (0.3% w/v CMC)	6.17 ± 0.28	2.46 ±0.15	3.71±0.32	44.53±1.92	78.64 ±2.12	463.62±5.42	---
Omeprazole (10mg/kg)	2.43±0.32**	4.62±0.16**	1.21±0.38**	23.62±1.26**	39.83±1.77**	258.72±3.34**	67.38
MENJ (100mg/kg)	3.14 ± 0.14*	4.05±0.22*	2.12±0.35*	31.96 ±2.31*	59.12 ± 1.79*	312.64 ± 6.00*	47.16
MENJ (200mg/kg)	2.92±0.26**	4.33±0.48**#	1.86±0.21**#	28.23±1.51**#	47.29±1.46**#	287.53±2.21**	52.49

All the test group results were compared with standard group as well as control group (Omeprazole). The significant * shows $p<0.05$ Vs control group; # indicates $p<0.05$ Vs Standard. The findings were evaluated statistically with the one-way *ANOVA* and the t-test of Dennett.

Fig: 2 Effect of MENJ Rhizome Extract on Aspirin plus Pylorus Ligated Model

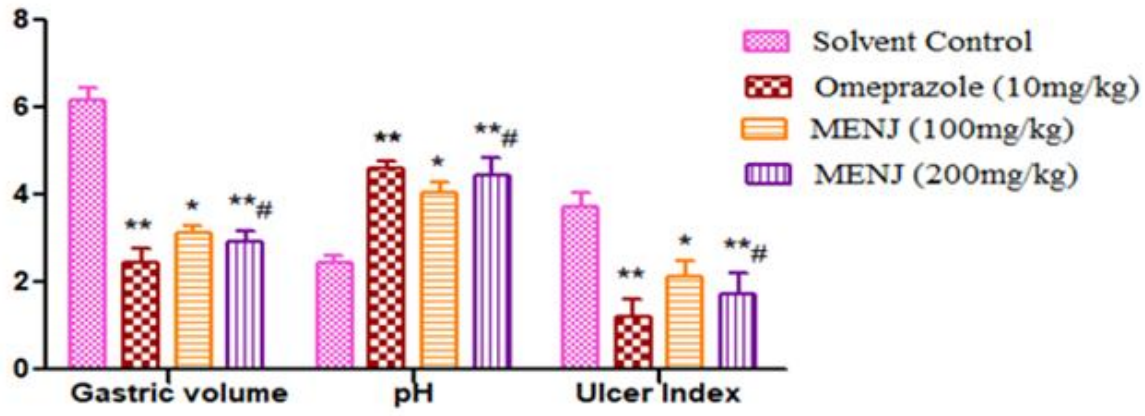
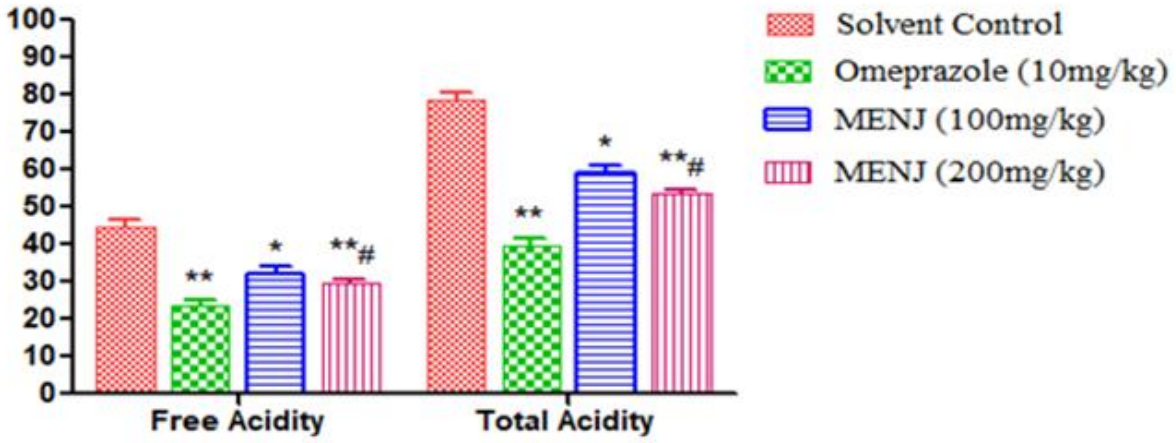


Fig: 3 Effect of MENJ Rhizome Extract on Aspirin plus Pylorus Ligated Model



Conclusion

Preliminary phytochemical investigations presented the existence of alkaloids, flavonoids and terpenoids, therefore the *Nardostachysjatamansi rhizomes'* antiulcer activity in this experimental model may be because of above mentioned chemicals. The findings indicate that methanolic extract of *Nardostachysjatamansi rhizomes* produced anti-secretory antiulcerogenic effects; it may be through proton pump inhibition mechanism. This particular research shows that *Nardostachysjatamansi rhizomes* extract has a potential anti-ulcer activity due to presence of phytochemical constituents like alkaloids and flavonoids. Moreover research is important for isolating the active action molecule.

Author contribution

All contributors participated in the research, data gathering, creating or updating the article, granted official approval of the published version, and agreed to be liable for all components of the project.

Declaration of competing interest

The authors report no conflicts of interest in this work.

Corresponding author:
Dr. Kola Venu
Email ID:venupharmacology@gmail.com

References

Borrelli F, Izzo AA. The plant kingdom as a source of anti-ulcer remedies. *Phytother Res*. 2000; 14(8):581–591.

Falcão HDS, Leite JA, Barbosa-Filho JM, *et al*. Gastric and duodenal antiulcer activity of alkaloids: a review. *Molecules* 2008;13(12):3198–3223.

Ganguly AK, Bhatnagar OP. Effect of bilateral adrenal anatomy on production of restraint ulcers in the stomach of albino rats. *Can J Physiol Pharmacol* 1973;51:748-50.

Krishna Prasad D and Nagaraj Sriharsha S. Evaluation of Anxiolytic activity of leaf extracts of *Nelumbo nucifera* in laboratory rodents". *International Journal of Pharmacy and Biological Sciences* 2015; 5(4): 24-30.

Kumar A. K., Ramachandra S. S. and Narsu L. Pharmacognostic and phytochemical investigations of roots of *Hibiscus micranthus* Linn. *Research Journal of pharmaceutical, Biological and Chemical Sciences* 2010; 1(4): 324–337.

Klein-Júnior LC, Santin JR, Niero R, de Andrade SF, Cechinel-Filho V. The therapeutic lead potential of metabolites obtained from natural sources for the treatment of peptic ulcer. *Phytochem Rev.* 2012; 11(4):567–616.

Kulkarni SK. Handbook of Experimental Pharmacology,. 3rd ed. New Delhi: Vallabh Prakashan; 1999. p. 148-50.

Mitra P, Ghosh T, Mitra PK. Anti gastric ulcer activity of *Amaranthus spinosus* Linn. leaves in aspirin induced gastric ulcer in rats and the underlying mechanism. *SMU Med J*. 2014; 1(2):313–328.

Mohod SM, Bodhankar SL. Evaluation of antiulcer activity of methanolic extract of leaves of *Madhuca indica* J.F Gmel in rats. *Pharmacologyonline* 2011; 3:203–213.

Paguigan ND, Castillo DH, Chichioco-Hernandez CL. Anti-ulcer activity of leguminosae plants. *Arq Gastroenterol*. 2014;51(1):64–67

Reddy VP, Sudheshna G, Afsar SK, et al. Evaluation of anti-ulcer activity of *Citrullus colocynthis* fruit against pylorus ligation induced ulcers in male Wistar rats. *Int J Pharm Sci. 2012;*4(2):446–451.

Repetto MG, Llesuy SF. Antioxidant properties of natural compounds used in popular medicine for gastric ulcers. *Braz J Med Biol Res*. 2002;35(5):523–534

Sharath SS, Preethy J, Kumar GS, et al. Screening for anti-ulcer activity of *Convolvulus pluricaulis* using pyloric ligation method in Wister rats. *Int J Pharm Sci Res.* 2015;6(1):89–99

Shay H, Komarov SA, Fels SE, Meraze D, Gruenstein M, Siplet H. A simple method for the uniform production of gastric ulceration. *Gastroenterology* 1945; 5:43- 61.

Venu K, Mondal P, Rajesh BRC, Arjun Goje, Gummadevelly Sandeep, Shaik Chanbasha, Bhookya Padmaja. Evaluation of Nootropic activity of Ethanolic and Aqueous Root Extracts of *Adhatoda vasica nees* in rodents. *Int. J. Pharm. Investigation*.2019;9(4):150-156.