Int. J. Ayur. Pharma Research, 2014; 2(2): 40-46 **ISSN: 2322 - 0910**

International Journal of Ayurveda and Pharma Research

Research Article

ANTIULCER ACTIVITY OF *AGNIMUKHA CHURNA*

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Received on: 21/03/2014 Revised on: 09/04/2014 Accepted on: 15/04/2014

ABSTRACT

Background: *Agnimukha Churna* is a popular Ayurvedic formulation, composed of herbs and spices, used for treatment of gastrointestinal disorders by traditional healers.

Objectives: In present study, we have to subject the pharmacognostic, phytochemical test and to evaluate the anti-ulcer activity of *Agnimukha Churna*.

Material and Methods: Study was carried out, by using two methods i.e., ethanol induced ulcers (5ml/kg, Absolute ethanol) and aspirin induced ulcers (200mg/kg) in rats pretreated with the doses of, Alcoholic Extract of *Churna* (AEC) (200mg/kg), Hydro-alcoholic Extract of *Churna* (HAEC) (200mg/kg), Ranitidine (20mg/kg). After 1 hour in ethanol induced method and 4 hour in aspirin induced method rat was sacrificed and stomach was removed for observation of ulcer scores, ulcer index and percentage of inhibition.

Results: The AEC and HAEC maintain the integrity of gastric mucosa by virtue of its effect on offensive and defensive gastric mucosal factors. AEC and HAEC significantly ($P<0.0001$) decreased ulcer index in ethanol induced and aspirin induced model.

Conclusions: Cyto-protective action may be the major mechanism responsible for the present study, which cause the generation of prostaglandins promoting inhibition of ulcer. Our study shows that *Agnimukha churna* and their fractions have the potential to be used as an anti-ulcer.

KEYWORDS: *Agnimukha Churna*, Antiulcer activity, Ethanol induced ulcer, Aspirin induced ulcer, Ulcer index.

INTRODUCTION

Peptic ulcer disease has been one of the leading causes of gastrointestinal surgery, with high morbidity and mortality rates. As the prevalence of this disease increase over time, one would expect peptic ulcers to have a significant global impact in the basic health and economic systems and in patient's life quality. They can occur most commonly (98-99%) in either duodenal or the stomach in the ration of 4:1.[1, 2]

Agnimukha Churna is an *Ayurvedic* compound prescribed for *Udavarta* (upward movement of gases), *Ajirna* (dyspepsia), *Gulma* (abdominal lump), *Phliharoga* (splenic diseases), *Arsa* (piles), *Sula* (pain), *Kasa* (cough), *Svasa* (asthma) and *Kshaya* (pthisis). *Agnimukha Churna* is also useful in peptic ulcer and bronchial asthma and used as carminative, digestive and appetizer.[3] It is composed of different herbs shown in Table 1.

Thus, the aim of present study is preliminary Phytochemical, Pharmacognostic investigation of *Agnimukha Churna* and their individual ingredients and comparative study of alcoholic extract of *Churna* and hydro-alcoholic extract of *Churna* against peptic ulcer.

Table 1 Formulation of *Agnimukha Churna*

S. No.	Ingredients	Quantity (gm)
1.	*Hingu* (*Ferula asfoetida* L.)	10
2.	*Vacha* (*Acorus calamus* L.)	20
3.	*Pippali* (*Piper longum* L.)	30

4.	*Sunthi* (*Zingiber officinale* Rosc.)	40
5.	*Ajowan* (*Trachyspermum ammi* (L.) Sprague)	50
6.	*Hritiki* (*Terminalia chebula* Retz. & Willd.	60
7.	*Chitrak* (*Plumbago indica* L.)	70
8.	*Kushtha* (*Saussurea lappa* C.B.CL.)	80

MATERIAL AND METHODS

Chemicals

Ethanol, Aspirin, Ranitidine, Sulphuric acid, Chloroform, Acetic acid, Dragendorff's reagent, Mayer's reagent, Wagner's reagent, Hager's reagent, Molisch reagent, Benedict's reagent, Ferric chloride were used in the study. All the chemicals or reagents used in the experimental study were procured from standard and reputed firms and are of analytical grade (EXLR) regularly used in the laboratory. All substances were prepared immediately before use.

Test drug

Agnimukha churna was prepared in house laboratory of the department, by following all aseptic measures.

Animals

Wistar Albino rats of either sex weighing between 150-200g were procured from the Department of pharmacology, Columbia Institute of Pharmacy, Raipur. The study was approved by Animal Ethical Committee (Ref. no. CIP/IAEC/2012-13/030). The animals were maintained under standard conditions proposed by CPCSEA. Animals were kept in spaceous cages and maintained under standard housing conditions of temperature (24-27°C) and humidity (60- 65%) with 12:12 light: dark cycles. They were acclimatized for seven days. Food was provided in the form of dry pellets and water ad libitum.

Dose selection and schedule

The fractions of different doses were selected and prepared in distilled water according to acute toxicity testing of OECD guidelines 420.[4]

Extraction of plant material

The powdered plant material (500g) was extracted using 90% ethanol (1.5L) for alcoholic extract of *Churna* and mixture of ethanol: water (60:40) was used for hot extraction of hydro-alcoholic extract. The extracts were evaporated using Rota flash evaporator under reduced pressure and low temperature and then on a water bath. The percentage yield, colour and consistency of the extract were recorded.

Standardization of individual plants and *Churna*

The standardization of *Churna* was done by WHO guidelines and determined moisture content, swelling index, foaming index, total ash, acid-insoluble ash and total extractive values.[5, 6]

Preliminary phytochemical screening of individual plants and *Churna*

Different phytoconstituents present in plants were determined by performing following tests for alkaloids, glycosides, carbohydrates, saponin, steroids, flavanoids, triterpenes, tannins and phenolic compounds. [7]

Experimental Procedure

Wistar Albino rats of either sex were divided into six groups, each group consists of six animals. All groups of animals received following treatments for 5 days: groups 1 (Normal) and 2 (Control) received vehicle 10 ml/kg, groups 3 and 4 were given Alcoholic Extract (AEC) (200mg/kg), Hydroalcoholic Extract (HAEC) (200mg/kg) respectively, and the group 5 (Standard) given reference drug Ranitidine at the dose of 20 mg/kg orally once daily for 15days. On the 5th day, 1h after final dose of treatment, the gastric ulcers were induced in rats by administering 96% ethanol (5ml/kg) and aspirin (200mg/kg) for ethanol induced and aspirin induced ulcers respectively. After 1h animals were sacrificed by cervical dislocation and stomach was incised along the greater curvature and examined for ulcers index. Percentage ulcer inhibition was calculated for each group on comparison with vehicle control group [8, 9].

Statistical Calculations

The data expressed as mean ± SEM. All statistical comparisons between the groups are made by means of one-way Analysis of Variance (ANOVA) with Bonferroni multiple comparison tests using Graph pad Prism 6 software. The values were considered significant at the levels of ****$p<0.0001$, ***$p<0.001$, **$p<0.01$.

RESULTS AND DISCUSSION

Pharmacognostic evaluation of individual plants and *Churna*

Bulk density and tapped density were performed to determine physical characteristics of crude drug and *Churna*. Quality test for crude drug powder and *Churna* was performed for foreign material, loss on drying at 105°C, ash value, acid insoluble value, water and alcohol extractive, swelling index and foaming index were found to be within standard range which is shown in Table 2.

Table 2: Standardization of Individual Plants and *Churna*

Parameters	*Hing*	*Vach*	*Pippali*	*Sonth*	*Ajowan*	*Hritika*	*Chitrak*	*Kushtha*
Foreign material (%)	1.19	0.25	1.38	Nil	1.83	Nil	1.53	1.42
Loss on Drying at 105°C (%)	0.15	0.11	0.07	0.14	0.08	0.10	0.15	0.11
Total ash (%)	9.2	7.2	6.6	5.6	8.6	2.4	7.8	4.8
Acid insoluble (%)	2.8	1.8	0.8	0.8	0.4	0.7	0.9	0.7
Water extractive value (%)	26.34	49.72	11.00	11.56	10.44	12.10	11.80	20.83
Alcohol extractive value (%)	13.52	26.75	14.28	11.83	13.52	37.02	22.01	12.52
Bulk density	0.6644	0.2631	0.5714	0.4651	0.5714	0.6250	0.3333	0.3968
Tap density	0.7142	0.3448	0.7142	0.5405	0.7142	0.4081	0.8695	0.3448
Swelling index	2	6	4	4	3	2	3.6	6.6
Foaming index	-	2.5	2	1.43	-	2	5	5

Preliminary Phytochemical test of individual plants and *Churna*

Active constituents present in the plant material are of vital importance as they are known to produce one or other definite pharmacological action. Phytochemical test performed using standard procedure with specific reagents, result of phytochemical investigation indicate the presence of carbohydrates, alkaloids, steroids, saponines, tannins and phenolic compounds which is shown in Table 3.

Table 3: Preliminary Phytochemical Screening

Test name	*Hing*	*Vach*	*Pippali*	*Sonth*	*Ajowan*	*Hritika*	*Chitrak*	*Kushtha*
Test for Alkaloids								
Dragendorff's Test	+	+	-	-	-	+	+	-
Mayer's Test	+	+	+	+	-	+	+	+
Wagner's Test	+	+	+	-	+	+	+	-
Test for Carbohydrates								
Molisch's test	+	-	-	-	+	+	+	-
Benedict's test	-	+	+	+	+	+	+	+
Test for Steroids								
Liebermann Buchard Test	-	-	+	-	+	+	-	+
Salkowski Test	-	-	+	-	+	+	-	+
Test for Tannins and phenolic compound								
Ferric chloride test	-	+	+	-	-	+	+	+
Acetic acid test	-	-	+	-	-	+	+	-
Test for saponin								
Froth formation test	-	+	+	+	-	+	+	+

+ Present, - Absent

Evaluation of Antiulcer activity in Ethanol induced gastric ulcer

In ethanol induced model ulcer index has been calculated by adding the total number of ulcers per stomach and the total severity of ulcers per stomach. This is shown in Table 4, Table 5, Figure 1 and Graph 1. Percentage inhibition of ulcer for AEC was 44.56 % which was higher than 27.01 % of HAEC but less than 56.14 % of standard groups. The alcoholic extract was found to be effective whose ulcer index was 0.316 at 200 mg/kg dose.

Table 4: Effect of drugs on ulcer index and percentage inhibition in Ethanol induced ulcer model

Groups	Treatment	Dose (mg/kg)	Ulcer Index	% inhibition
Normal control	Normal	-	0	100
Disease control	EtOH + vehicle	-	0.57	-
Standard	EtOH + Ranitidine	20	0.25	56.14
AEC	EtOH + AEC	200	0.316	44.56
HAEC	EtOH + HAEC	200	0.416	27.01

Table 5: Effect of drugs on mean ulcer index in Ethanol induced gastric ulcer

Groups	Treatment	Mean Ulcer index ± SEM
Control	Ethanol	3.000 ± 0.00
Standard	Ranitidine	0.75 ± 0.115****
AEC	AEC	1.083 ± 0.115****
HAEC	HAEC	1.583 ± 0.115****

*Mean ± SEM significant difference from control by one way ANOVA followed by Bonferroni's multiple comparison test, (n= 6) ****p<0.0001*

Evaluation of Antiulcer activity in aspirin induced gastric ulcer

In aspirin induced model ulcer index has been calculated by adding the total number of ulcers per stomach and the total severity of ulcers per stomach. This is shown in Table 6, Table 7, Figure 2 and Graph 2. Percentage inhibition of alcoholic extract for ulcer was 54.85% nearly same in ethanol induced ulcer where the percentage inhibition of hydro alcoholic extract in aspirin induced ulcer was 45.42 % which was greater than 27.01% of ethanol induced ulcer. Percentage inhibition of standard group was found to be 71.42% which was greater than ethanol induced ulcer of standard group. So, in both model the ulcer inhibition was high for alcoholic extract at 200mg/kg which can be further evaluated by changing dose to make it more effective.

Table 6: Effect of drugs on ulcer index and percentage inhibition in Aspirin induced ulcer model

Groups	Treatment	Dose (mg/kg)	Ulcer Index	% inhibition
Normal control	Normal	-	0	100
Disease control	Aspirin + vehicle	-	0.7	-
Standard	Aspirin + Ranitidine	20	0.20	71.42
AEC	Aspirin + AEC	200	0.316	54.85
HAEC	Aspirin + HAEC	200	0.382	45.42

Table 7: Effect of drugs on mean ulcer index in Aspirin induced gastric ulcer

Groups	Treatment	Mean Ulcer index ± SEM
Control	Aspirin	3.000 ± 0.00
Standard	Ranitidine	0.583 ± 0.083 ****
AEC	AEC	1.083 ± 0.083****
HAEC	HAEC	1.417 ± 0.083***

*Mean ± SEM significant difference from control by one way ANOVA followed by Bonferroni's multiple comparison test, (n= 6) ****p<0.0001, ***p<0.001, **p<0.01*

CONCLUSION

Present study clearly indicates that the oral administration of AEC and HAEC in Ethanol induced and Aspirin induced gastric ulcer in Albino rats displayed appreciable gastro protective activity as demonstrated by significant decrease in ulcer index and increased percent inhibition in both models. The antiulcer activity of *Agnimukha Churna* may be attributed to the presence of alkaloids, flavanoids, triterpenes and steroids in these fractions. Therefore the study supports the claims of traditional medicinal practitioners as an antiulcer remedy. It could also be a prospective substitute for the existing synthetic antiulcer drugs which are known to produce harmful adverse effects. The results demonstrated that extract produce anti-ulcerogenic effect possessing anti-secretory and cytoprotective activity. Further studies to isolate, identify and characterize the active principle(s) are in progress.

REFERENCES

1. Harold, Kalant, Grant, Denis M., Mitchel, Jane, In: Principles of Medical Pharmacology, seventh ed. Elsevier Canada Ltd., 2007; p. 557, 558, 559
2. Harsh Mohan. Text book of pathology. Sixth edition, jaypee publishers. 2009; 549-555
3. Anonymous, Vaidya Yoga Ratnavali (Formulary of Ayurvedic Medicines), The Indian Medical Practitioners' Co-operative Pharmacy & Stores Ltd, Chennai, 1994; 96-97.
4. The Organisation of Economic Cooperation and Development (OECD), 2001a The OECD Guidelines for testing of chemicals: 420 Acute Oral Toxicity, Paris, 1-7
5. Subramanyam CVS, Setty J Thimm, Laboratory manual of physical pharmaceutics, Vallabh publication, 1st edition 2002; pp 46-51.
6. Mukherjee Pulokh K. Quality control of herbal drugs- An approach to evaluation of botanicals; Bussiness horizons p'ceutical publishers, 1st edition 2002; pp 183-213.
7. Khandelwal KR, Kokate CK, Pawar AP, Gokhle SB. Preliminary phytochemical screening. Text book of Practical Pharmacognosy. 1st ed. Pune: Nirali prakashan; 1995; 149-156.
8. Gerhard Vogel H. Gastric function-Antiulcer activity. Text book of Drug Discovery and Evaluation: Pharmacological Assay. 2nd Ed. Germany: Springer; 2002; 867-872.
9. Kulkarni SK. Experiments on intact preparations. Hand book of Experimental Pharmacology. 3rd ed. Delhi: Vallabh Prakashan; 1997; 148-50.

Cite this article as:
S. Prakash Rao, Indu Amrit, Parag Jain, Vijay Singhl. Antiulcer Activity of Agnimukha Churna. Int. J. Ayur. Pharma Research 2014; 2 (2): 40-46.
Source of support: Nil, Conflict of interest: None Declared

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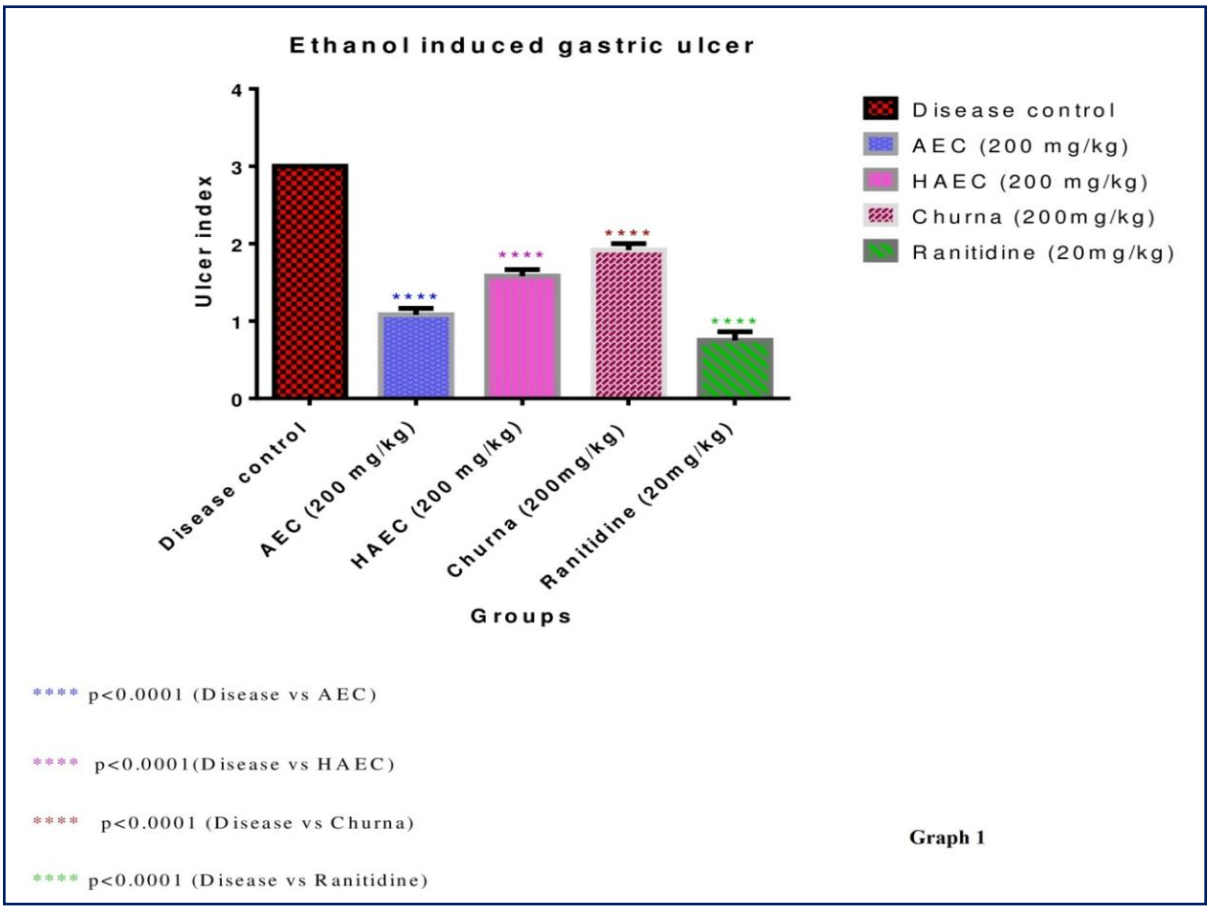
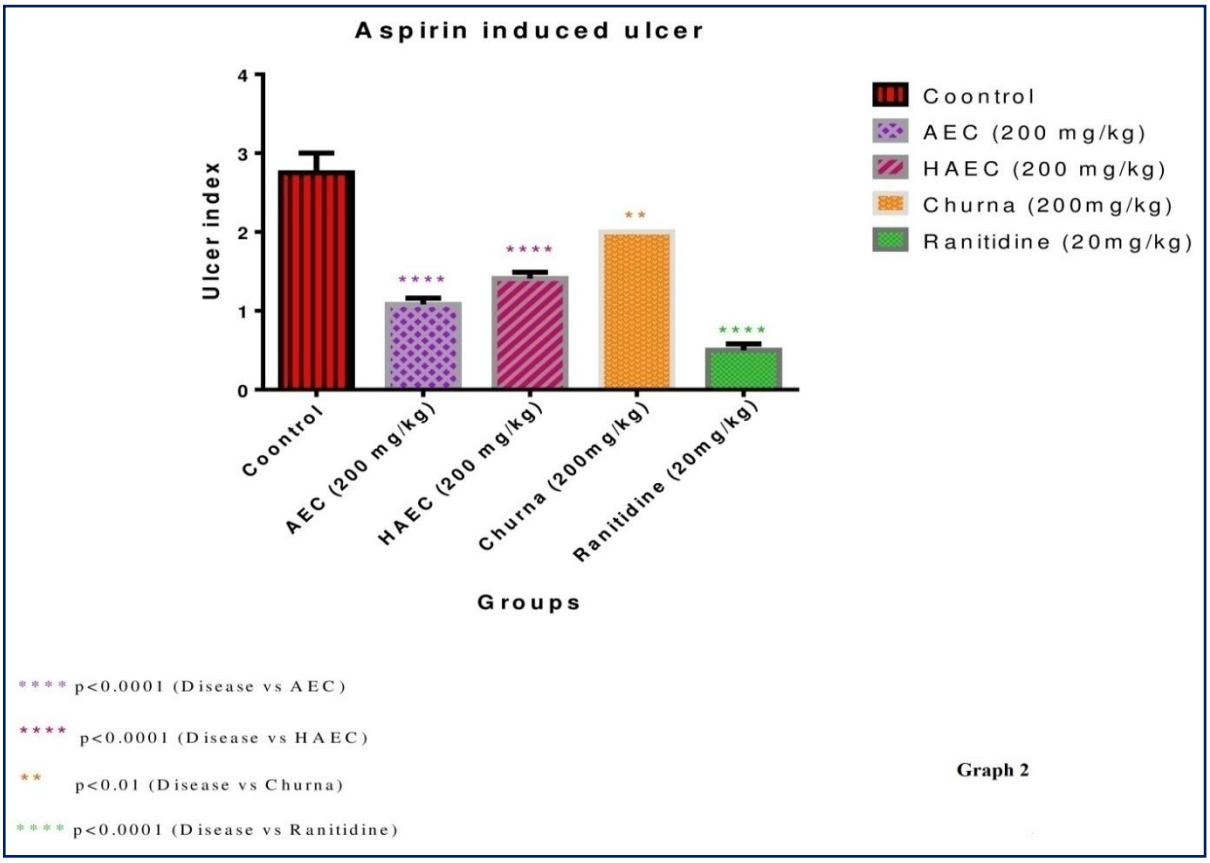
STUDY PHOTOGRAPHS

Figure 1: Evaluation of antiulcer activity in different groups of Ethanol induced gastric ulcer

Figure 2: Evaluation of antiulcer activity in different groups of Aspirin induced gastric ulcer



Graph 1: Effect of drugs on mean ulcer index in Ethanol induced gastric ulcer



Graph 2: Effect of drugs on mean ulcer index in Aspirin induced gastric ulcer