COMPARISON BETWEEN THE EFFECTS OF EXTRACTS FROM THREE MEDICINAL PLANTS "AMMI VISNAGA, MYRISTICA FRAGRANS AND OLEA EUROPEA" ON STIMULATED GASTRIC SECRETION IN FASTING RABBITS.

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ABSTRACT

BACKGROUND: Peptic ulcer is mostly produced due to the over production of gastric acid. This study was undertaken to find out the effects of extract from the fruits of medicinal plants Ammi visnaga, seeds of Myristica fragrans and leaves of Olea europea (all of which contain documented natural Calcium channel blockers) on volume and acidity of Carbachol induced gastric section. Their effects were also compared to find out any difference in their efficacy.

OBJECTIVE: To find out the comparative effectiveness of three medicinal plants extracts for the treatment of peptic ulcer and disease associated with hyper gastric acidity conditions.

MATERIAL & METHODS: Forty rabbits of local breed, weighing 1-1.5kg were used. The animals were kept on fasting for 48 hours, divided into group A, B, C & D. pylorus of each animal was ligated after a midline incision. Each extracts 500 mg/kg body weight and Carbachol 600µg/kg body weight were administered intraperitoneally.

RESULTS: It was found that all the extracts reduced the volume, free and total acidity of gastric secretion, which were statistically highly significant when compared with Carbachol ($P < 0.001$). By observing the mean values of all the extracts, it was clear that Ammi visnaga was more effective than Myristica fragrans which was in turn more effective than Olea europea on all parameters included in study.

CONCLUSION: The three extracts therefore, can be used effectively in the treatment of peptic ulcer and all other diseases associated with hyper gastric acidity.

KEY WORDS: Extracts & gastric secretion

INTRODUCTION

Peptic ulcer is one of the most common ailments, with which a physician comes across in the clinical practice. Increased acid production from gastric mucosa is responsible for peptic ulceration in majority of the patients. Ulcers are not found in achlorhydric patients and almost always occur in patients with Zollinger- Ellison (Z.E) syndrome which is characterized by very high acid secretion.¹ Inhibition of over production of acid is a desirable therapeutic goal in the treatment of peptic ulcer. It has been

documented that 38 medicinal plants including fruits of Ammi visnaga, seeds of Myristica fragrans and leaves of Olea europea have natural calcium channel blocker². Khellin and visnagin were identified from Ammi visnaga fruit and were proved that all of them have calcium channel blocking mode of action³. Methanol extract from the fruit of Ammi visnaga showed significant calcium channel blocking activity⁴. In a study with the help of spectrophotometer and high performance liquid chromatography (HPLC) it was observed that Khellin and visnagin

are present in the fruits of *Ammi visnaga*⁵. In another study it was observed that extract from the seeds of *Myristica fragrans* showed significant calcium channel blocking activity⁶. Thirty percent ethanol extract from the leaves of *Olea europea* has significant calcium channel blocking activity⁷. The calcium channel blocking agents like Verapamil, nifedipine and diltiazem are commonly used in the treatment of hypertension, angina, myocardial infarction and supraventricular tachycardia⁸. Induction of hypercalcaemia through intravenous administration of calcium, is usually associated with increased gastric volume and acidity.^{9,10} The acid stimulating ability of calcium; is well known and there is extreme sensitivity to calcium in patients with Z.E. syndrome.^{11,12} Histamine release from peritoneal mast cells is critically dependent upon extracellular Ca^{++} concentration, so non-availability of Ca^{++} may cause reduced effects of histamine on acid production in the stomach. Calcium channel blockers have been mainly used in cardiovascular system as inhibitors of muscle contraction. In the stomach, motility and acid secretion have been shown to be dependent upon calcium ions. So this study was planned to evaluate the effects of extract from the fruits of *Ammi visnaga*, seeds of *Myristica fragrans* and leaves of *Olea europea* on the volume and acidity of Carbachol induced gastric secretion. Their effects were compared on these parameters.

MATERIALS AND METHODS

Forty rabbits of local breed were selected for the present study. Healthy animals of both sexes weighing 1-1.5kg were used in the study. All the animals were kept fasting for 48 hours with free availability of water before they were subjected to experimental procedure. The animals were divided into four groups each containing 10 animals. Group A was Carbachol treated, Group B was *Ammi visnaga* + Carbachol treated, Group C. was *Myristica fragrans* + Carbachol treated & group D was *Olea europea* + Carbachol treated. The operative procedure was the one adopted by Vischer et al. (1954)¹³. Animals were anaesthetized with ether, abdomen was opened

and pylorus was ligated with silk suture. Then abdominal wall was closed with suture clamps and intraperitoneal (I.P) injection of Carbachol 600 μg /Kg body weight were administered to group A, 500mg/Kg body weight of each extract to all the four groups & followed by Carbachol 600 μg /Kg body weight after 15 minutes to group B, C & D. The rabbits were deprived of water for four hours after administration of drugs. Then the rabbits were slaughtered, the thorax and abdomen were opened, oesophagus was ligated and the stomach was removed quickly. The contents of the stomach were collected. The volume of gastric juice was measured. Then the contents were centrifuged, filtered and subjected to titration for estimation of free and total acidity by the method described by Varley (1962)¹⁴. One ml of centrifuged and filtered gastric secretion was titrated against 0.1 N NaOH using Topfer's reagent as an indicator for determination of free acidity and 1% phenolphthalein as an indicator for combined acidity. The sum of the two titrations was total acidity. The data was analyzed statistically using student "t" test.

RESULTS

The volume, free acidity and total acidity of gastric secretion in group A (Carbachol treated group) was $28.7 \pm 0.650 \text{ ml}$, $6.39 \pm 0.408 \text{ m.Eq./dl}$ and $7.64 \pm 0.408 \text{ m.Eq./dl}$ respectively. Similarly the volume, free acidity and total acidity in group B (*Ammi visnaga* + Carbachol treated) was $13.8 \pm 0.578 \text{ ml}$, $2.41 \pm 0.216 \text{ m.Eq./dl}$ and $3.57 \pm 0.276 \text{ m.Eq./dl}$ respectively. The volume, free acidity and total acidity in group C (*Myristica fragrans* + Carbachol treated) was $15.3 \pm 0.597 \text{ ml}$, $2.9 \pm 0.331 \text{ m.Eq./dl}$ and $3.86 \pm 0.0426 \text{ m.Eq./dl}$ respectively. Similarly the volume, free acidity and total acidity in group D (*Olea europea* and Carbachol treated) was $16.5 \pm 0.763 \text{ ml}$, $3.15 \pm 0.375 \text{ m.Eq./dl}$ and $4.02 \pm 0.0353 \text{ m.Eq./dl}$ respectively. These reductions noticed in all the parameters were found to be statistically highly significant when compared with Carbachol group, $P < 0.001$ (Table 1).

Table 1

Comparison between the effects of extract from *Ammi visnaga*, *Myristica fragrans* and *Olea europea* on volume and acidity of Carbachol induced gastric secretion in fasting rabbits.

Drug	Volume of gastric secretion (ml)	Acidity(m.Eq./dl of gastric secretion)	
		Free	Total
Carbachol	28.7±0.65 0 (10)	6.39±0.408 (10)	7.64±0.408 (10)
Ammivisnaga + Carbachol	13.8 ±0.578 (10)	2.41±0.216 (10)	3.57±0.276 (10)
P. Values	<0.001	<0.001	<0.001
Myristica-fragrans +Carbachol	15.3±0.59 7 (10)	2.9±0.331 (10)	3.86±0.426 (10)
P. Values	<0.001	<0.001	<0.001
Olea europea + Carbachol	16.5±0.76 3	3.15±0.375	4.02±0.353
P value	<0.001	<0.001	<0.001

Each value represents mean of total observations. Figures in parenthesis indicate the number of animals in each group ± indicates standard error of mean. P. values when compared with Carbachol.

DISCUSSION

Acid secretion in the stomach is controlled at a variety of levels by neural, hormonal and paracrine mechanisms. When these regulatory mechanisms malfunction, acid and pepsin autodiagest the mucosa resulting in the ulceration of oesophagus, stomach and duodenum¹⁵.

Histamine, acetylcholine or Carbachol are potent secretagogues for the parietal cells of gastric mucosa leading to the production of HCl¹⁶.

Acetylcholine and gastrin act through calcium ions. Carbachol being a cholinomimetic drug increases free intracellular calcium ions which, in turn activate protein kinase by phosphorylation and lead to increased production of HCl. In this study we observed that all the three extracts reduced the volume free and total acidity. All these reductions were statistically highly significant when compared with the mean values in Carbachol treated group. This is due to the calcium channel blocking activity of natural calcium channel blockers present in these extracts. By comparing the mean values of all the parameters of these three extracts, it is clear that *Ammi visnaga* is more effective than *Myristica fragrans* which is in turn more effective than *Olea europea*.

Our study is in consistent with other workers who concluded that calcium channel blocker Verapamil significantly reduces gastric acid secretion^{17, 18}. The extracts containing natural calcium channel blockers inhibit the calcium ion influx, which may be responsible for the observed reductions in volume and acidity of gastric secretion. Besides, calcium channel blockers inhibit lipoxigenase pathway during metabolism of arachidonic acid. So leukotrienes, the injurious substance is not formed and all the arachidonic acid is metabolized through cyclooxygenase pathway. This will lead to the production of prostaglandin which couples with Gi protein, inhibits adenyl cyclase and thus decrease HCL production.¹⁹

Release of histamine from mast cells is critically dependent on external calcium ions, so by blocking calcium ions, can block histamine release which is a potent agent for HCL secretion²⁰.

Calcium channel blockers are also used in controlling contraction of cardiovascular smooth muscles²¹, allergic reaction²² and prevention of premature labor²³. All these actions are due to the calcium channels blocking activity.

CONCLUSION

It is concluded that these extracts may be beneficially used for the treatment of peptic ulcer & all other diseases which are due to hyper gastric acidity condition. It will also save a lot of national economy. Further studies in this regard for evaluation of these effects are suggested in human subjects.

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