COMPARISON BETWEEN THE EFFECTS OF EXTRACT FROM MEDICINAL PLANT "*OLEA EUROPEA*" AND MYRISTICA FRAGRANS ON THE VOLUME AND ACIDITY OF CARBACHOL INDUCED 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 extracts from medicinal plants *Olea europea* and *Myristica fragrans* (Both of them contains documented natural Calcium channel blocker) on volume and acidity of Carbachol induced gastric section.

METHODS: Thirty rabbits of local breed, weighing 1-1.5kg were used. The animals were kept on fasting for 48 hours. After general anesthesia, pylorus of each animal was ligated. Both the extracts 500 mg/kg each and Carbachol 600µg/kg body weight after 15 minutes were administered intraperitoneally.

RESULTS : When the means of all the parameters were compared with that of carbachol, it was found that the extracts reduced the volume, free and total acidity of gastric secretion which were statistically highly significant ($P < 0.001$). When we compared the mean values of volume, free and total acidity for the two extracts, it was observed that these differences in all the three parameters between groups B and C were found to be non-significant ($P > 0.5$).

CONCLUSION: The extracts can be used effectively and safely in the treatment of hyper acidity conditions and peptic ulcer after evaluation its effects in human being.

KEY WORDS: *Olea europea*, *Myristica fragrans* & gastric secretion.

INTRODUCTION

Common disease in gastrointestinal tract is Peptic ulcer. Increased acid production from gastric mucosa is responsible for peptic ulceration in majority of the patients. It is a common experience that Ulcers are not found in case of achlorhydria. It almost always occurs in patients with Zollinger-Ellison (Z.E) syndrome which is characterized by very high acid secretion¹. 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 *Olea europea* and *Myristica fragrans* have natural calcium channel blockers². Thirty percent ethanol extract from the leaves of *Olea europea* has significant calcium channel blocking activity³. In a study it was observed that extract from the seeds of *Myristica fragrans* showed significant calcium channel blocking activity⁴. The calcium channel blocking agents like Verapamil, nifedipine and diltiazem

are commonly used in the treatment of hypertension, angina, myocardial infarction and supraventricular tachycardia⁵. Induction of hypercalcaemia through intravenous administration of calcium is usually associated with increased gastric volume and acidity^{6,7}. The acid stimulating ability of calcium is well known and there is extreme sensitivity to calcium in patients with Z-E syndrome^{8,9}. Calcium channel blocker Verapamil may interfere with H^+K^+ATP Ase due to its high affinity for the site H^+K^+ATP Ase system which is accessible from luminal side of the stomach¹⁰. Histamine release from peritoneal mast cells is critically dependent upon extra cellular 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 CVS 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 leaves of *Olea europea* and seeds of *Myristica fragrans* on the volume and acidity of Carbachol induced gastric secretion. Their effects were also compared on these parameters.

MATERIALS AND METHODS

Thirty rabbits of local breed were selected for the present study. Healthy animals of both sexes weighing 1-1.5 kg 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 3 groups each containing 10 animals. Group A was Carbachol treated, Group B was *Olea europea*+ Carbachol treated and Group C was *Myristica fragrans*+Carbachol treated.

The operative procedure was the one adopted by Vischer et al (1954)¹¹. Animals were anaesthetized with ether, abdomen was opened by a mid line incision and pylorus was ligated with silk suture. Then abdominal wall was closed with suture clamps and intraperitoneal (I.P) injection of Carbachol 600µg/Kg body weight were administered to group A, 500mg/Kg body weight of extracts *Olea europea* & *Myristica fragrans* to group B and C respectively. It was followed by Carbachol 600 µg/ Kg body weight after 15 minutes to group B and C. The rabbits were deprived of water for four hours after administration of drugs. Then the rabbits were slaughtered, thorax and abdomen were opened, oesophagus was ligated and the stomach was removed quickly. The contents of the stomach were collected by incision through greater curvature. 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)¹². 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 for combined acidity. The sum of the two titrations was total acidity. The

formula of $N_1 V_1 = N_2 V_2$ was used. The data was analyzed statistically using student "t" test.

RESULTS

The volume, free acidity and total acidity of gastric secretion in group A was 28.7 ± 0.650 ml, 6.39 ± 0.408 m.Eq./dl and 7.64 ± 0.408 m.Eq./dl respectively. Similarly volume, free and total acidity in group B was 16.5 ± 0.763 ml, 3.15 ± 0.375 m.Eq./dl and 4.02 ± 0.353 m.Eq./dl respectively. Likewise the volume, free acidity and total acidity in group C were 15.3 ± 0.597 ml, 2.9 ± 0.331 m.Eq./dl and 3.86 ± 0.426 m.Eq./dl respectively.

These reductions noticed in all parameters were found to be highly significant when compared with Carbachol ($P < 0.001$). All these changes are shown in Table 1.

Drugs	Volume of gastric secretion (ml)	Acidity (m.Eq/dl ml of gastric secretion)	
		Free	Total
Carbachol	28.7 ± 0.650 (10)	6.39 ± 0.408 (10)	7.64 ± 0.408 (10)
<i>Olea europea</i> + Carbachol	16.5 ± 0.763 (10)	3.15 ± 0.375 (10)	4.02 ± 0.353 (10)
P Values	<0.001	<0.001	<0.001
<i>Myristica fragrans</i> +Carbachol	15.3 ± 0.597 (10)	2.9 ± 0.331 (10)	3.86 ± 0.426 (10)
P Values	<0.001	<0.001	<0.001

When we compared the mean values of volume, free and total acidity for the two extracts, it was observed that these differences in all the three parameters between groups B and C were found to be nonsignificant ($P > 0.5$). All these changes are shown in Table 2.

Table 1

Effects of extracts from *Olea europea* and *Myristica fragrans* on the volume and acidity of gastric secretion induced by Carbachol in fasting rabbits. Carbachol was injected 600 µg/kg body

weight, *Olea europea* and *Myristica fragrans* 500 mg/kg body weight each intraperitoneally.

Each value indicates mean of the total observation.

Figures in parenthesis indicate the number of animals in each group.

± Indicates standard error of mean

P Value between Carbachol, *Olea europea* + Carbachol and *Myristica fragrans* + Carbachol

Table 2

Comparison between the effects of extracts from *Olea europea* and *Myristica fragrans* on the volume and acidity of gastric secretion induced by Carbachol in fasting rabbits. Carbachol was injected 600 µg/kg body weight, *Olea europea* & *Myristica fragrans* 500 mg/kg each intraperitoneally.

Drugs	Volume of gastric secretion (ml)	Acidity (mEq /dl ml of gastric secretion)	
		Free	Total
<i>Olea europea</i> + Carbachol	16.5±0.763 (10)	3.15±0.37 5 (10)	4.02±0.353 (10)
<i>Myristica fragrans</i> + Carbachol	13.12±0.32 6 (10)	2.04±0.15 0 (10)	3.21±0.408 (10)
P Values	P<0.05	P<0.05	P<0.05

Each value indicates mean of the total observation.

Figures in parenthesis indicate the number of animals in each group.

± Indicates standard error

P Value between *Olea europea* and *Myristica fragrans*.

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

autodiagete the mucosa resulting in the ulceration of esophagus, stomach and duodenum¹³. Histamine, acetylcholine or Carbachol are potent secretagogues for the parietal cells of gastric mucosa leading to the production of HCl¹⁴. 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 *Olea europea* and *myristica fragrans* reduced the volume free and total acidity. This is due to the calcium channel blocking activity of natural calcium channel blocker present in the extract. All these reductions were statistically highly significant when compared with the mean values in Carbachol treated group. When we compared the mean values of volume, free and total acidity for the two extracts, it was observed that these differences in all the three parameters were found to be nonsignificant indicating that they are equally effective.

Our study is in consistent with other workers who concluded that calcium channel blocker verapamil significantly reduces gastric acid secretion^{15,16}. It is due to the fact that Verapamil, a well known calcium channel blocker inhibits the calcium influx, which may be responsible for the observed reductions in volume and acidity of gastric secretion. Besides, Verapamil inhibits lipoxygenase pathway during metabolism of arachidonic acid. So leukotrienes, the injurious substances are not formed and all the arachidonic acid is metabolized through cyclooxygenase pathway. This will lead to the production of prostaglandin which couples with G_i protein, inhibits adenyl cyclase and thus decrease HCl production¹⁷. Release of histamine from mast cells is critically dependent on extra cellular calcium ions, so calcium channel blocker will block calcium ions influx which in turn will inhibit histamine release which is a potent agent for HCl secretion¹⁸.

Verapamil is also used in controlling contraction of cardiovascular smooth muscles¹⁹, allergic reaction²⁰ and prevention of premature labor²¹.

All of these actions are due to the calcium channels blocking activity as the extract also contains calcium channel blocker, so it can also be used for the treatment of the above mentioned diseases and peptic ulcer.

CONCLUSION

It is concluded that the extracts may be beneficially used as a single drug therapy in patients having peptic ulcer alone or concurrent with angina, myocardial infarction, bronchial asthma and prevention of premature labor. So it will 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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