ORIGINAL ARTICLE

EFFECTS OF '*OLEA EUROPEA*' EXTRACT ON VOLUME AND ACIDITY OF CARBACHOL INDUCED GASTRIC SECRETION, LIVER AND KIDNEY FUNCTION IN RABBITS

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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 leaves of medicinal plant Olea europea (which contains documented natural Calcium channel blocker) on volume and acidity of Carbachol induced gastric section. Its effects were also observed on liver and kidney function. **Methods:** Thirty rabbits of local breed, weighing 1–1.5 kg were used. The animals were kept on fasting for 48 hours, after that the pylorus of each animal was ligated. Carbachol 600 µg/kg body weight and extract from the leaves of Olea europea 500 mg/kg body weight were administered intraperitoneally to group A and B respectively. The extract was also administered in the same dose to group C for 45 days twice daily intraperitoneally to observe its effects on liver and kidney function. **Results**: The extract reduced the volume, free and total acidity of gastric secretion, which were statistically highly significant when compared with Carbachol (p<0.001). When the differences of means for both the liver and kidney function were compared with that of control group, all these were found statistically non-significant indicating that the extract has no adverse effects on these organs regarding all parameters included in study. **Conclusion**: Extract can be used effectively and safely in the treatment of hyper gastric acidity conditions and peptic ulcer after evaluation of its effects in human subjects.

Keywords: Medical plants, *Olea Europea*

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 (ZE) 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 leaves of Olea europea have natural calcium channel blocker.[2] Thirty percent ethanol extract from the leaves of Olea europea has significant calcium channel blocking activity[3].The calcium channel blocking agents like Verapamil, nifedipine and diltiazem are commonly used in the treatment of hypertension, angina, myocardial infarction and suprventricular tachyeardia.[4] Induction of hypercalcaemia through intravenous administration of calcium, is usually associated with increased gastric volume and acidity.[5,6] The acid stimulating ability of calcium is well known and there is extreme sensitivity to calcium in patients with Z-E syndrome.[7,8] 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 cardiovascular system as inhibitors of muscle contraction. In the stomach, motility and acid secretion have been shown to be dependent upon calcium ions.

This study was planned to evaluate the effects of extract from the leaves of Olea europea on the volume and acidity of Carbachol induced gastric secretion. Its effects were also compared on these parameters. The extract was also administered in the same dose for 45 days twice daily to observe its effects on liver and kidney function.

MATERIAL 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 Olea europea treated group for 45 days.

The operative procedure was the one adopted by Vischer *et al.*[9] 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 (IP) injection of Carbachol 600 µg/Kg body weight were administered to group A, 500 mg/Kg body weight of Olea europea to group B followed by Carbachol 600 µg/Kg body weight after 15 minutes to group B. The rabbits were deprived of

water for four hours after administration of drugs. Then the rabbits were sacrificed, abdomen was 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.[10] One ml of centrifuged and filtered gastric secretion was titrated against 0.1 N NaOH using Topfer's reagent for determination of free acidity and 1% phenolphthalein as an indicator for combined acidity. The sum of the two titrations was total acidity. In case of group C, blood samples were taken for estimation of liver function (serum bilirubin, SGPT, alkaline phosphatase) and kidney function (serum creatinine, blood urea) before start of treatment and then after forty five days treatment with extract 500 mg/Kg body weight twice daily. The data was analysed statistically using student's *t*-test.

RESULTS

The mean values of volume, free acidity and total acidity of gastric secretion in group A was 28.7±0.650 ml, 6.39±0.408 mEq/dl and 7.64±0.408 mEq/dl respectively. Similarly the mean values of volume, free acidity and total acidity in group B were 16.5±0.763 ml, 3.15±0.375 mEq/dl and 4.02±0.353 mEq/dl respectively. These reductions noticed in all the parameters were found to be highly significant when compared with Carbachol treated group (p<0.001) (Table-1).

The mean values for liver function and kidney function were found out. It was observed that the mean values for Serum creatinine before start of treatment and after treatment were 1.245±0.0 mg/dl and 1.230±0.01 mg/dl, blood urea 35.1±0.640 and 36.5±0.636 mg/dl, Serum creatinine 0.18±0.02 and 0.18±0.02 mg/dl, SGPT 36.6±0.6 and 37.3±0.6 IU/L, alkaline phosphatase 78.1±604 and 76.9±0.640 IU/L respectively. These changes were statistically non significant (p>0.5) (Table-2).

Table-1: Effect of *Olea eurepa* extract 500 mg/Kg body weight and Carbachol 600 µg/Kg body weight on volume and acidity of gastric secretion in fasting rabbits

Drugs	Volume of gastric secretion (ml)	Acidity (mEq/100 ml of gastric secretion)	
		Free	Total
Carbachol (A)	28.7±0.650 (10)	6.39±0.408 (10)	7.64±4.408 (10)
Olea europea + Carbachol (B)	16.5±0.763 (10)	3.15±0.375 (10)	4.02±0.353 (10)
p-Values	<0.001	<0.001	<0.001

Each value indicates mean of the total observations.
Figures in parenthesis indicate the number of animals in each group.
p-values between the means of group A and Group B

Table-2: Effect of *Olea europea* extract 500 mg/Kg body weight on kidney and liver functions in rabbits

Drug	Kidney function tests		Liver function tests		
	S. Creatinine Urea (mg/dl)	Blood (mg/dl)	S. Bilirubin (mg/dl)	SGPT (IU/L)	Alk: phosphatase (IU/L)
Control (before extract)	1.245±0.016 (10)	35.1±0.640 (10)	0.18±0.029 (10)	36.6±0.653 (10)	78.1±0.604 (10)
Olea europea	1.23±0.010 (10)	36.5±0.636 (10)	0.18±0.024 (10)	37.3±0.650 (10)	76.9±0.640
p-value	>0.5 NS	>0.5 NS	>0.5 NS	>0.5 NS	>0.5 NS

Each value indicates mean of the total observations. Figures in parenthesis indicate the number of animals in each group.
p-Values between the means before and after extract administration.

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 auto digest the mucosa resulting in the ulceration of oesophagus, stomach and duodenum.[11]

Acetylcholine and gastrin act through calcium ions. Carbachol, being a cholinomimetic drug increases free intracellular calcium ions which, intern activate protein kinase by phosphorylation and leads to increased production of HCl. In this study we observed that Olea europea 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 the extract.

Our study is in consistence with other study who concluded that calcium channel blocker Verapamil significantly reduces gastric acid secretion.[12] The extract containing natural calcium channel blockers inhibit the calcium influx, which may be responsible for the observed reductions in volume and acidity of gastric secretion. Besides, calcium channel blockers inhibit lipoxygenase 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 and inhibits adenyl cyclase and thus decrease HCl production.[13]

Release of histamine from mast cells is critically dependent on external calcium ions, so by blocking calcium ions can inhibit, histamine release which is a potent agent for HCl secretion.[14] When the changes in the mean values of all the parameters in group C were compared with that before treatment with extract and after injecting the extract 500 mg/kg body weight for 45 days, they were all found non-significant. This shows that the extract is safe for these organs.

CONCLUSION

It is concluded that Olea euporea extract may be used beneficially and safely in hyper gastric acid secretary conditions like gastritis, gastro-oesophageal reflux diseases and peptic ulcer. Further studies in this regard are suggested in human subjects.

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