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Ameliorative effect of phytoconstituents of *Momordica charantia* on body weight in Cerulein induced acute pancreatitis in Swiss albino mice

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Abstract

Premature activation of the intracellular digestive enzymes (proenzymes) within the pancreas, in turn lead to the autodigestion of the pancreas due to colocalization with pancreatic hydrolases such as cathepsin-B. Cerulein induced Acute Pancreatitis model is one among the highly used animal experimental models because of its high reproducibility and being economical in nature. The present study was conducted on Swiss albino mice in which Acute Pancreatitis was induced by cerulein and the ameliorative effect by *Momordica charantia* fruit extract was observed. Phytoconstituents in *Momordica charantia* are responsible for the antioxidant and anti-inflammatory activities thereby help in ameliorating the changes induced by oxidative stress.

Keywords: Acute pancreatitis, Cerulein, *Momordica charantia*

Introduction

Acute pancreatitis (AP) an inflammatory condition of the pancreas which appears at a rapid pace and cause severe inflammation of the pancreas. AP remains to be the 3rd most common cause of immediate hospital admission under gastrointestinal diseases and is 6th in position for the non-malignant cause of death, and it even acts as a major contribution factor for the development of chronic pancreatitis and pancreatic cancer (Peery *et al.*, 2019) ^[1].

The pathogenesis of AP starts with the activation of intra-pancreatic acinar cell zymogen leading to conversion of inactive trypsinogen to active trypsin which cause dysregulated digestive enzyme secretion causing the accumulation of vacuoles and simultaneously triggering the inflammatory pathways which terminally end up in apoptosis and necrotic acinar cell death (Gukovskaya *et al.*, 2019) ^[2]. The manifestation of the disease is a multi-step process which starts with local limited inflammation and takes its turn towards systemic inflammation by release of diverse inflammatory mediators such as cytokines, reactive oxygen species (ROS), chemokines, leukocyte adhesion molecules, lipids and gaseous mediators (Granger and Remick, 2005) ^[3].

Cholecystokinin (CCK) analogue, cerulein is well known for induction of AP with high reproducibility. The supramaximal doses of cerulein result in dysregulated enzyme production, formation of vacuoles in the cytoplasm, edema formation and inflammatory cell infiltration into the pancreas (Lee *et al.*, 2022) ^[4].

Momordica charantia Lam. (MC) is commonly called a bitter melon of the Cucurbitaceae family and is well known for its medicinal value and nutritional properties (Polito *et al.*, 2016) ^[5]. The main chemical constituents include heteropolysaccharides, proteins and peptides (Schrot *et al.*, 2015) ^[6], terpenoids, saponins, flavonoids, phenolic compounds, essential oils, fatty acids, amino acids and sterols (Dandawate *et al.*, 2016) ^[7]. Phenolic compounds present in the pericarp, stem and leaves serve as good antioxidants by directly acting on the ROS and even inducing the endogenous defence mechanisms (Yoshime *et al.*, 2016) ^[8]. The glycosidic compounds from the fruit grouped as cucurbitine-type triterpenoids (cucurbitacins) are known to have anti-inflammatory and anti-diabetic properties (Rios *et al.*, 2005) ^[9].

Materials and Methods

The present experimental study was conducted on male *Swiss albino* mice of uniform age (about 6-8 weeks), which were procured from Jeeva Life Sciences, Hyderabad. Sterile mice feed in the form of pellets was purchased from M/s. Jeeva Life sciences, Hyderabad. All the drugs used in the study were purchased locally in Hyderabad. Fruit samples of *Momordica charantia* were thoroughly washed with tap water to remove any waste and dust particles. The excess water was removed by pressing with a paper towel gently and allowed to dry under shade for 3-4 weeks until constant weight is achieved. Once the samples were dried they were ground to semi powder form using a commercial grinder and were stored in an airtight polythene bags in the refrigerator at 4 °C for further analysis. Later, ethanolic extracts were prepared following the established protocol.

A total of 40 male *Swiss albino* mice of 6-8 weeks of age, weighing between 22-30 g. were obtained from Jeeva Life sciences, Hyderabad. The animals were kept in polypropylene cages and maintained with 12 hrs. dark/light cycle under hygienic conditions having ambient temperature (22–24 °C) for acclimatization (1 week) prior to the beginning of the experiment. Animals were placed on commercial standard pellet feed and provided water *ad libitum* throughout the experiment. The experimental protocol was approved by the Institutional Animal Ethics Committee, College of Veterinary Science, Rajendranagar, Hyderabad. All the groups were maintained as per the following treatment schedule for 7 days. The mice were randomly divided into 5 groups consisting of 6 mice in each group with the following schedule.

Experimental Design

Group-1	Normal control (NC)/Sham- normal saline for 7 days.
Group-2	<i>Momordica charantia</i> fruit extract control (MCFE)/ per se @ 400 mg/kg b.wt. through oral route.
Group-3	Cerulein control (CC)/Positive control/Disease control (DC) received 6 consecutive hourly intraperitoneal injections of cerulein @ 50 µg/kg b.wt. on 7 th day.
Group-4	<i>Momordica charantia</i> fruit extract (MCFE) low dose @ 200 mg/kg b.wt. + cerulein (Pre-treatment with <i>Momordica charantia</i> fruit extract (MCFE) low dose @ 200 mg/kg b.wt. + 6 consecutive hourly intraperitoneal injections of cerulein @ 50 µg/kg b.wt. on 7 th day).
Group-5	<i>Momordica charantia</i> fruit extract (MCFE) high dose @ 400 mg/kg b.wt. + cerulein Pre-treatment (Pre-treat with <i>Momordica charantia</i> fruit extract (MCFE) high dose @ 400 mg/kg + 6 consecutive b.wt. + 6 hourly intraperitoneal injections of cerulein @ 50 µg/kg b.wt. on 7 th day).

Mice body weights were measured and recorded on the 7th day.

Statistical analysis

The results of the experiment are presented as mean ± SEM (standard error of the mean) where multiple determinants of were considered and comparison among groups was analyzed using one-way analysis of variance (ANOVA) followed by Tukey's Multiple comparison test using GraphPad prism scientific software version 5.0. P- value < 0.05 was considered as significant.

Results and Discussion

Average body weight

The average body weight (g) in group 3 (27.83±1.38) was not significantly different from group 1 (28.00±1.69). Similar results were noticed in treatment groups 4 and 5 (27.00±0.55 & 28.80±1.16 respectively). Further, no significant difference was observed in the average body weights of mice among groups 1 and 2 (Table 1).

Table 1: Average body weight (g) in different groups of mice

Group	Day 7
1. Normal Control (NC)	28.00±1.69
2. Momordica Control	29.67±1.12
3. Cerulein Control (CC)	27.83±1.38
4. Cerulein + Momordica Low Dose	27.00±0.55
5. Cerulein + Momordica High Dose	28.80±1.16

Values are Mean ± SE (n=6); One-way ANOVA with Tukey's Multiple Comparison Test (GraphPad Prism version 5.0). Mean ±SE without any superscripts were non-significant.

Discussion

The average body weights (g) of mice were not significantly altered among the groups.

This could be assigned to the reduced expression of inflammatory proteins and also could be because of activation of PPARα and PPARγ activators which are known for maintaining the blood lipids and also exhibit anti-inflammatory activity through inhibition of cytokine production by activation of PPARγ (Ciou *et al.*, 2014) [10].

Conclusion

It is evident from the investigation that presence of terpenoids, saponins, flavonoids, phenolic compounds, essential oils, fatty acids, amino acids and sterols, cucurbitine-type triterpenoids (cucurbitacins) are known to have anti-inflammatory and anti-diabetic properties (Rios *et al.*, 2005) [9] that might be attributed for maintenance of the body weight in *Momordica charantia* treated mice. Further, investigations are required to delineate the actual role of these phytoconstituents at molecular level.

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