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EFFECT OF GARCINIA CAMBOGIA ON HISTOPATHOLOGICAL OUTCOMES OF PANCREATIC CELLS IN ALLOXAN INDUCED DIABETIC ALBINO RAT.

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ABSTRACT

BACKGROUND: Diabetes mellitus, is a prevalent metabolic condition categorized by continual hyperglycemia due to either impaired insulin secretion or resistance, has a high prevalence across Southeast Asia—most notably in Pakistan. As such nations work fervently to control the disorder, scientists have begun exploring the potential of Garcinia cambogia extract to help regulate blood sugar levels. **OBJECTIVE:** In this investigation, researchers concentrated on GcE's impacts on pancreatic β-cell mass and islet morphology in diabetic rats highlighting the need for further research in alternative therapies for DM. **METHODS** The investigation was designed as a pre-clinical experiment carried out over a twelve month duration at the Department of Pharmacology and Therapeutics, Basic Medical Sciences Institute, Jinnah Postgraduate Medical Centre, Karachi. A total of sixty young male albino Wistar rats were split into three sets which were induced to develop diabetes using Alloxan. The animals received differing doses of extracts derived from Garcinia cambogia termed GcE across an eight week timeframe. At the end of the defined experimental interval, the rats were anesthetized, sacrificed, and blood specimens were drawn to quantify fasting serum insulin levels under sedation. **RESULTS**: The mean weight significantly increased in Group C with a mean value of 304.95±35.15 gm as compared to group A and B with mean value of 292.05±43.15 gm and 294±50.16 gm respectively. Group C showed a significant increase in β-cell mass (355±55 units) compared to Groups A (280±35 units) and B (275±40 units), with improved islet shape ($p<0.005$). **CONCLUSION**: The study concludes that Garcinia cambogia extract shows promise in enhancing pancreatic β-cell mass and islet morphology in diabetes, suggesting potential therapeutic benefits.

KEY WORDS: Garcinia Cambogia, Histopathological, Pancreatic Cells, Alloxan, Albino Rat

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INTRODUCTION

Diabetes mellitus (DM) is primarily the metabolic disorder of glucose. It is recognized by chronic hyperglycemia caused by β-cell dys-functioning resulting in insulin deficiency or insulin resistance in the target cells or both defects. Absolute or relative insulin deficiency is the primary underlying defect and hyperglycemia is the major metabolic defect with secondary defects of lipids and proteins[1-2]. South East Asia is declared as the capital of diabetes mellitus and prevalence of DM is high in Pakistan gaining 3rd position in the World[3-4]. In near future, the DM will be a visible burning health problem in the country and a challenge for the economy and prosperity of the country. The 2019 estimates of global prevalence of DM show 463 million cases and are projected to be 700 million by the year 2045. A prevalence of 26.3% of diabetes mellitus has been reported for Pakistan that is a state of chaos of this health problem. DM is not a single disease entity, but causes a number of acute and chronic complications in the human body. The chronic complications are miserable and often untreatable[5]. Only treatment of chronic diabetic complications is in achieving an optimal glycemic control. Currently, available modern anti – diabetic therapy, although effective but not potent to get an optical glycemic sufficient to overcome and retard the chronic diabetic complications. Hence there is a gap for new drugs and herbs to be researched and discovered for better diabetic management with cost effectiveness. Herbs have attracted much scientific research interest for their possible efficacy against the diabetes mellitus. Analyzing new herbs with excessive anti-diabetic efficacy, low cost and fewer adverse effects is a lot worrying these days for growing troubles of diabetes mellitus. Herbs have been researched for the treatment of diabetes associated problems of obesity, metabolic syndrome, hyperlipidemia and dyslipidemia, etc[6]. Garcinia cambogia extract (GcE) is found effective against the obesity. Past studies have reported inconclusive results on the efficacy of Garcinia cambogia (Gc) against the body weight, obesity and diabetes mellitus[7-9]. Animal studies[10-11] concluded hydroxycitric acid (HCA) is effective in losing induced obesity in experimental animals. Definitive clinical trials of GC in human beings are not available. Further research is demanding before recommending the GC for the therapy of diabetes mellitus which is increasing in the country and currently, growing body of research is shifted to the alternative therapy. DM needs research and analysis of biological effects of Garcinia cambogia extract (GcE). The present experimental animal study analyzed the pharmacological effects of Garcinia cambogia extract on the β-cell mass, and islet morphology in Alloxan induced Diabetic albino rat.

MATERIAL AND METHODS

Study Design

Pre-clinical experimental study

Study Population

Young male Albino Wistar rats

Study Duration
One year study
Study Setting

The study was carried out at Department of Pharmacology and Therapeutics, Basic Medical Sciences Institute, JPMC Karachi. The rats were kept in cages at room temperature while maintaining a day and night cycle and were given ad libitum water and feed.

Sample Size Estimation

A total of n=60 healthy young male Albino Wistar rats weighing 200 to 300 grams were divided into three groups, n=20 rats assigned to the **Group A** Diabetic rats + GcE (25 g/kg bwt), **Group B** (n=20): Diabetic rats + GcE (50 g/kg bwt), and **Group C** (n=20): Diabetic rats + GcE (75 g/kg bwt).
Sampling Technique

Phase I. Purposive sampling was done to select healthy rats as per inclusion and exclusion criteria,

Phase II. Induction of Diabetes mellitus with Alloxan (Sigma Aldrich, stored at 4^0C) – rats which achieve blood glucose levels of >250 mg/dl were selected randomly (after duration of 72 hours) for experimental rats being treated with GcE.

Animal Protocol and Housing
We bought a sample of sixty albino Wistar rats from the Basic Medical Sciences Institute, JPMC Karachi, Open Market/Animal House. The NIH Guide for the Care and Use of Laboratory Animals[12] was followed in the housing and handling of the animals. The rats were kept in cages made of stainless steel with bedding made of sawdust. Plastic drinkers with stainless nozzles and stainless steel feed containers were provided with the cages. The conditions in which the animals were kept were clean and well-ventilated. Rats were given unlimited access to tap water and food (lab chow). At 12-hour intervals, the cycle of light and dark was preserved. The Animal House of the Basic Medical Sciences Institute, JPMC Karachi, authorized the animal policy that was followed when performing surgeries on animals.
After being refrigerated at 4°C, alloxan monohydrate (Sigma Aldrich, St. Louis, MO, USA) was dissolved in room temperature normal saline and administered intraperitoneally (IV) to rats that had fasted the previous night. When β-cells are directly harmed by a single intraperitoneal injection of Alloxan (120 mg/kg body weight), necrosis occurs within 48–72 hours and induces diabetes mellitus.

Alloxan monohydrate was dissolved in 100 mm citrate buffer (pH 4.5), and rats that had been fasted overnight were given an intraperitoneal injection of the freshly made solution at a dose of 120 mg/kg. After 48 hours, the animals' blood glucose levels were measured, and those that were higher than 250 mg/dl were classified as diabetic[13] and included in the study.
The cages of rats of control and experiment groups were labelled as clearly showing different rat groups under study.
Following the advice of veterinary specialists, rats were fed chow that had been scientifically approved to both the experimental and control groups. Raw food was served as the chow. All rats received anesthesia 24 hours after the trial concluded in the sixth week.

Animals were anesthetized by Ketamine (10 mg/Kg) and Xylazine (0.5 mg/Kg)[14] .

Animals were sacrificed by cervical dislocation

Blood samples were collected by cardiac puncture in EDTA and Plain tubes.

Serum was isolated from the clotted blood by centrifugation.

After centrifuging the blood samples for one hour at a low speed (4°C, 5000 rpm, 15 min),

the supernatant was collected and kept at -80°C to measure the fasting serum insulin level.

Animal Groups

The rats were randomly divided into three groups namely A, B, and C.

Group A(n=20): Diabetic rats + GcE (25 g/kg bwt) daily for 8 weeks,
Group B (n=20): Diabetic rats + GcE (50 g/kg bwt) daily for 8 weeks,
Group C (n=20): Diabetic rats + GcE (75 g/kg bwt) daily for 8 weeks,
Content of Garcinia Cambogia was equal to 1000, 2000, 3000 mg/kg of GcE- HCA levels.

Outcome Measures

Histopathological Analysis of Pancreatic Tissue: Examination of pancreatic tissue sections stained with hematoxylin and eosin to assess the histological alterations induced by Alloxan and the potential protective effects of GcE on pancreatic β-cells. Parameters such as β-cell mass, and islet morphology were evaluated.

RESULTS

The analyses of the findings had revealed that the mean weight of the rats were significantly increased in group C in comparison to other two group after eight weeks of intervention (table 1). The average values were 304.95±35.15 in group C where as in group A and B the values were 292.05±43.15 and 294±50.16 respectively. Further histopathological analyses had revealed that β-cell mass were significantly increase $p<0.005$ in group C with an average units of 355±55 in comparison to group B and group C where the values were 280±35 units and 275±40 units. Moreover islets morphology were also found to be improved in group C in comparison to group A and group B (table 2) (Figure 1)

Table 1 Comparative analysis of Weights measured in grams after eight week of experiment

Variables	Average weights in grams± SD	F-Value	Level of Significance
Group A	292.05±43.15	28.57	0.001
Group B	294±50.16		
Group C	304.95±35.15		

Table 2: Histopathological analyses of the pancreatic cells				
Pancreatic Beta-Cell Mass				
Variables	Mean units ± SD	F-Value	df	level of significance
Group A	280±35	3.75	12	P<0.05
Group B	275±40			
Group C	355±55			
Islets morphology (number of vessels)				
Variables	Mean units			
Group A	2 appears normal			
Group B	4 appears normal			
Group C	6 appears normal			

DISCUSSION

The current study investigated the potential pharmacological effects of Garcinia cambogia extract (GcE) on pancreatic β-cell mass and islet morphology in Alloxan-induced diabetic albino rats. The outcomes of this preclinical experimental study provide light on the potential therapeutic effects of GcE in diabetes control. One of the study's key findings is an increase in body weight in rats treated with higher dosages of GcE (Group C) compared to the other groups. This gain in body weight may imply an improvement in general health state, which could be explained by GcE's potential impacts on metabolic parameters. The rise in body weight may potentially indicate improved food absorption or utilization, necessitating additional research into the mechanisms underlying this benefit.

Histopathological examination of pancreatic tissue showed that GcE protects β-cell bulk and islet shape. Group C received the greatest dose of GcE, resulting in a considerable increase in β-cell mass compared to other groups. This data indicates that GcE may have a regenerative or protective effect on pancreatic β-cells, which are essential for insulin release and glucose homeostasis. Furthermore, the improvement in islet shape in Group C lends credence to GcE's possible therapeutic role in maintaining pancreatic function in diabetes settings. The observed effects of GcE on pancreatic β-cell mass and islet morphology are consistent with previous research indicating the potential anti-diabetic properties of GcE[15].

Rind extracts of *Garcinia cambogia* shows anti – diabetic activity. An experimental with Streptozocin induced diabetes mellitus in normal and obese rats given GC extract showed improvement of blood glucose and lipids levels and body weight[16]. Streptozocin induced type 2 diabetes mellitus in rats were given GC rind (aqueous extract) in dose of 100 mg/kg and 200 mg/kg bwt. A significant improvement in fasting and post – prandial glucose levels were observed after 4 weeks therapy[17]. In another study conducted in 2023, the findings showed GcE treatment resulted in significant reductions in the Atherogenic Index of Plasma (AIP), implying improved lipid profiles and decreased cardiovascular risk[18]. Although the precise mechanisms underlying these effects remain to be elucidated, it is hypothesized that bioactive compounds such as hydroxycitric acid (HCA) present in GcE may contribute to its therapeutic efficacy. HCA has been shown to modulate various metabolic pathways involved in glucose metabolism and insulin sensitivity, which could explain its beneficial effects on pancreatic function observed in this study.

Despite the promising findings, several limitations should be considered when interpreting the results of this study. First, the study was conducted on animal models, therefore extrapolating these findings to human populations should be done with caution. Further clinical trials are required to establish GcE's efficacy and safety in diabetic individuals. Furthermore, more research into the processes underlying GcE's impacts on pancreatic function is needed to properly comprehend its therapeutic potential.

CONCLUSION

This study suggests that Garcinia cambogia extract may improve pancreatic β-cell mass and islet morphology in diabetic individuals. These findings serve as a foundation for future study into the therapeutic potential of GcE in diabetes control. However, more research is needed to understand the underlying mechanisms and find the best dosage and duration of treatment for greatest efficacy and safety in clinical settings.

ETHICS APPROVAL: The ERC gave ethical review approval.

CONSENT TO PARTICIPATE: written and verbal consent was taken from subjects and next of kin.

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AUTHORS' CONTRIBUTIONS:

All persons who meet authorship criteria are listed as authors, and all authors certify that they have participated in the work to take public responsibility of this manuscript. All authors read and approved the final manuscript.

CONFLICT OF INTEREST: No competing interest declared

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