International Journal of Research Studies on Environment, Earth, and Allied Sciences (IJRSEAS)
| *Volume-3 Issue-2* | *April-2026* | *ISSN: 3066-0637* | *Website:* https://eesrd.us

Manuscript ID:
IJRSEAS-2026-030220

OPEN ACCESS

Quick Response Code:

Website: https://eesrd.us

DOI: 10.5281/zenodo.19676787

DOI Link:
https://doi.org/10.5281/zenodo.19676787

Volume: 3

Issue: 2

Pp. 103-108

Month: April

Year: 2026

E-ISSN: 3066-0637

Submitted: 07 Mar. 2026

Revised: 16 Mar. 2026

Accepted: 08 Apr. 2026

Published: 30 Apr. 2026

Address for correspondence:
T. A. Gangavane
P.G. Department of Zoology, Sadguru Gadge Maharaj College, Karad Masur Rd, Ashtavinayak Colony, Vidyanagar, Karad, Maharashtra (India)
Email:
digitaltejaswita8@gmail.com

How to cite this article:
Gangavane, T. A., & Bodare, R. D. (2026). Influence of Aging on Pancreatic Protein Homeostasis in Alloxan-Induced Diabetic Mice and Therapeutic Potential of Selected Ethanolic Plant Extracts. International Journal of Research Studies on Environment, Earth, and Allied Sciences, 3(2), 103–108.
https://doi.org/10.5281/zenodo.19676787

Influence of Aging on Pancreatic Protein Homeostasis in Alloxan-Induced Diabetic Mice and Therapeutic Potential of Selected Ethanolic Plant Extracts

T. A. Gangavane[1], Dr. R. D. Bodare[2]
[1]P.G. Department of Zoology, Sadguru Gadge Maharaj College, Karad Masur Rd, Ashtavinayak Colony, Vidyanagar, Karad, Maharashtra (India)
[2]Department of Zoology Prof. Dr. N. D. Patil Mahavidyalaya, Malkapur- Perid, Tal. Shahuwadi, Dist. Kolhapur Maharashtra (India)

Abstract

Diabetes mellitus disrupts protein metabolism through enhanced proteolysis, impaired insulin-mediated anabolic signaling, and oxidative stress. Aging further intensifies these disturbances by reducing cellular repair capacity and protein synthesis efficiency. The present study investigated age-related changes in pancreatic protein content in adult and aged type 2 diabetic mice and evaluated the protective effects of selected ethanolic plant extracts. Experimental diabetes significantly reduced pancreatic protein levels in both age groups, with a more pronounced decline observed in aged diabetic mice. Treatment with ethanolic extracts of Mangifera indica, Aegle marmelos, *and* Cynodon dactylon *significantly improved pancreatic protein content, although recovery was greater in adult mice compared to aged animals. These findings indicate that aging increases pancreatic vulnerability to diabetic damage and influences tissue recovery potential. The study suggests that preservation of pancreatic protein integrity may be an important mechanism underlying the protective role of selected medicinal plant extracts in age-associated diabetic conditions.*

Keywords: *Type 2 diabetes, Aging, Pancreatic protein, Herbal extracts, Oxidative stress.*

Introduction

Diabetes mellitus is a metabolic disorder characterized by chronic hyperglycemia resulting from impaired insulin secretion or action. Besides disturbances in carbohydrate metabolism, diabetes significantly alters protein homeostasis through enhanced proteolysis, reduced protein synthesis, and oxidative stress–mediated damage [1]. These alterations contribute to progressive tissue dysfunction, particularly in metabolically active organs such as the pancreas [2].

The pancreas plays a central role in glucose regulation, and prolonged hyperglycemia can compromise its structural and functional integrity [3]. Estimation of total pancreatic protein content serves as a useful biochemical indicator of tissue status under diabetic conditions [4].

Aging further influences metabolic efficiency and cellular resilience. Decline in protein synthesis, mitochondrial function, and antioxidant defense with advancing age increases susceptibility to oxidative damage and delays tissue recovery [5]. Despite extensive studies on diabetes-induced biochemical changes, limited data are available regarding age-related differences in pancreatic protein alterations and their modulation by medicinal plants [6].

Mangifera indica, *Aegle marmelos*, and *Cynodon dactylon* are traditionally recognized for their antidiabetic and antioxidant properties [7]. Therefore, the present study aimed to evaluate age-dependent changes in pancreatic protein content in diabetic mice and to assess the protective potential of selected ethanolic plant extracts [8].

Materials and Methods

Chemicals

All chemicals and reagents used in the present investigation were of analytical grade. Alloxan monohydrate was procured from a certified biochemical supplier. Reagents required for protein estimation were used as per the standard Lowry's method protocol [9].

Plant Samples

Fresh plant materials of *Mangifera indica* (leaves), *Cynodon dactylon* (leaves), and *Aegle marmelos* (leaves) were collected from the botanical garden SGM college, Karad. The collected samples were authenticated in Department of Botany, SGM college, Karad, thoroughly washed with distilled water to remove adhering impurities.

The collected plant materials were thoroughly washed, shade-dried, and mechanically pulverized to obtain a coarse powder. The powdered samples were subjected to extraction using analytical-grade ethanol as the solvent by employing the Soxhlet extraction method. Approximately measured quantities of plant powder were placed in a thimble and extracted continuously for 48–72 hours to ensure complete recovery of ethanol-soluble phytoconstituents [10].

Following extraction, the solvent containing dissolved bioactive compounds was filtered through Whatman No. 1 filter paper to eliminate insoluble residues. The filtrate was then concentrated under reduced pressure using a rotary evaporator at a controlled temperature (40–50°C) to prevent thermal degradation of active constituents [11]. The resulting semi-solid crude extract was weighed to determine percentage yield and subsequently transferred into sterile, airtight containers. The extracts were stored at 4°C until further pharmacological evaluation [12].

Animals

Healthy Swiss albino mice (*Mus musculus* Linn.) were selected for the present study. The animals were procured from Rajarambapu College of Pharmacy, Kasegaon, Taluka Walwa, District Sangli – 415404 (CPCSEA Registration No. 1209/PO/Re/S/09/CPCSEA.; dated 16 March 2019).

Adult mice weighing between 35–40 ± 2 g body weight were included in the experiment. The animals were provided with a standard pellet diet (Amrut mice feed, Pranav Agro Industries Pvt. Ltd., Sangli) and had free access to drinking water throughout the study period [13].

All mice were housed in clean polypropylene cages under controlled laboratory conditions. Environmental parameters were maintained at a temperature of 24–25°C with relative humidity ranging from 35–60%, and a 12-hour light/dark cycle was followed [14]. The animals were monitored regularly to ensure proper health and acclimatization before initiation of the experimental procedures [15].

Experimental Design

Table: Allocation of Experimental Groups and Treatment Regimen

Group No.	Study Category	Description of Intervention
I	Normal Control	Male mice belonging to adult (4–5 months) and aged (20–24 months) categories were maintained under standard laboratory conditions. These animals were provided with a routine pellet diet and administered distilled water (0.5 mL per animal per day) orally during the study period. No diabetogenic agent was given to this group.
II	Diabetic Control	Adult and aged male mice were subjected to overnight fasting prior to induction of diabetes. Experimental diabetes was established by intravenous administration of Alloxan monohydrate at a dose of 150 mg/kg body weight, freshly dissolved in acetate buffer. Animals exhibiting sustained hyperglycemia were designated as diabetic controls.
III	EEMI-Treated Group	Diabetic mice received ethanolic extract of *Mangifera indica* leaves at a dosage of 200 mg/kg body weight. The extract was administered orally once daily for 20 successive days.
IV	EEAM-Treated Group	Alloxan-induced diabetic mice were treated with ethanolic extract of *Aegle marmelos* leaves at 200 mg/kg body weight per day via oral route for a duration of 20 days.
V	EECD-Treated Group	Diabetic animals were administered ethanolic extract of *Cynodon dactylon* at a dose of 200 mg/kg body weight orally for 20 consecutive days following confirmation of diabetes.

The experimental animals were randomly allocated into five groups, each consisting of six mice (n = 6).

Induction of Type 2 Diabetes

Experimental diabetes was induced using a standard diabetogenic protocol. Animals exhibiting sustained hyperglycemia were considered diabetic and included in the study [16].

Treatment Schedule

Diabetic mice were treated with ethanolic extracts of *Mangifera indica* (EEMI), *Aegle marmelos* (EEAM), and *Cynodon dactylon* (EECD). Separate treatment groups were maintained for adult and aged mice along with their respective controls [17].

Estimation of Pancreatic Protein Content

Pancreatic tissue was excised, homogenized in appropriate buffer, and total protein content was estimated using Lowry's method. Protein concentration was expressed as micrograms per milligram (µg/mg) of tissue.

Statistical Analysis

Results were expressed as Mean ± SD (n = 6). Statistical significance was determined using appropriate analysis of variance followed by post hoc comparison. A value of $P < 0.01$ was considered statistically significant, while $P < 0.05$ was considered marginally significant.

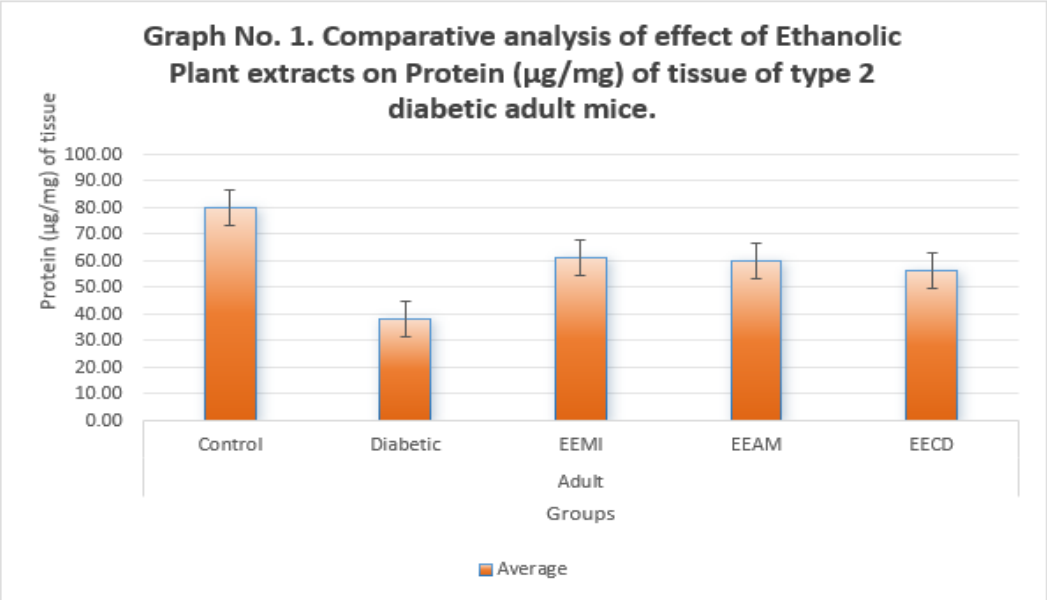
Results

Effect of Diabetes and Plant Extracts on Pancreatic Protein Content in Adult Mice

Induction of type 2 diabetes resulted in a pronounced reduction in tissue protein concentration in adult mice. The untreated control group exhibited a mean protein level of 80.25 ± 0.2739 µg/mg tissue**,** whereas diabetic animals showed a marked decline to **38.12 ± 15.5639 µg/mg tissue**, demonstrating significant metabolic deterioration following alloxan administration (P < 0.01).

Oral administration of ethanolic plant extracts substantially improved tissue protein levels when compared with the diabetic group. Treatment with *Mangifera indica* extract (EEMI) increased protein concentration to 61.13 ± 0.8847 µg/mg tissue. Similarly, *Aegle marmelos* extract (EEAM) elevated protein levels to 59.83 ± 0.2160 µg/mg **tissue**, while *Cynodon dactylon* extract (EECD) restored protein content to 56.33 ± 0.2733 µg/mg tissue**.**

Statistical analysis revealed that the reduction observed in the diabetic group was highly significant relative to control (P < 0.01). All treatment groups demonstrated significant improvement when compared with diabetic animals (P < 0.05).



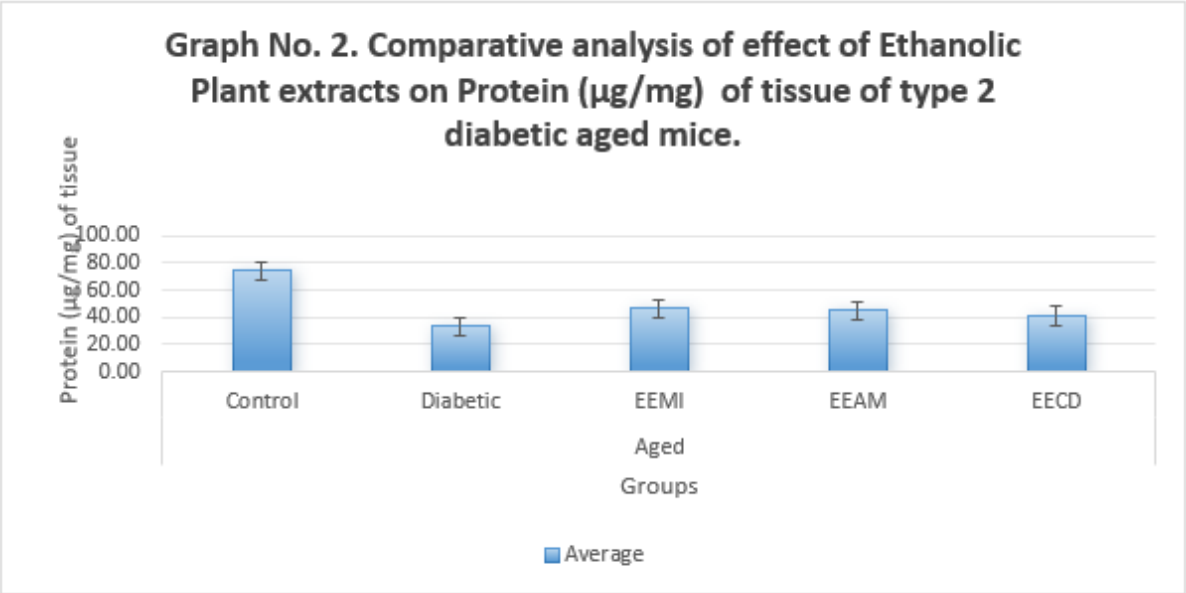
The findings indicate that diabetes-induced protein depletion was considerably reversed following plant extract supplementation, with EEMI exhibiting the most pronounced restorative effect among the treated groups.

Effect of Diabetes and Plant Extracts on Pancreatic Protein Content in Aged Mice

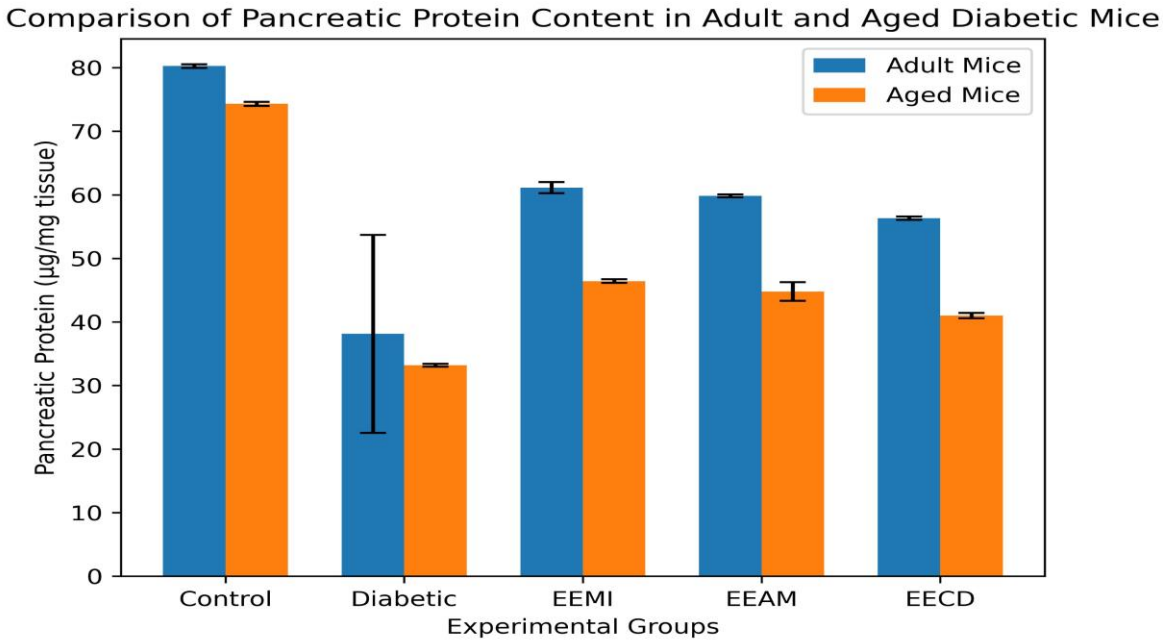
A similar but more severe decline in tissue protein concentration was observed in aged diabetic mice. The control aged group showed a protein level of **74.30 ± 0.3162 µg/mg tissue**, while diabetic aged animals displayed a reduced level of **33.18 ± 0.2317 µg/mg tissue**, confirming significant protein loss under hyperglycemic conditions (P < 0.01).

Following treatment, partial recovery of protein content was observed. Mice treated with EEMI showed protein levels of **46.42 ± 0.2858 µg/mg tissue**, whereas EEAM treatment resulted in **44.78 ± 1.4580 µg/mg tissue**. Administration of EECD improved protein concentration to **41.00 ± 0.4099 µg/mg tissue**.

Comparison between groups demonstrated that EEMI and EEAM treatments significantly improved protein levels relative to diabetic aged mice (P < 0.01), while EECD exhibited a moderate yet statistically meaningful improvement (P < 0.05).



Comparative Analysis of Adult and Aged Mice



The comparative graphical representation clearly demonstrates age-dependent differences in pancreatic protein content across all experimental groups. Under normal conditions, aged mice exhibited slightly lower baseline protein levels compared to adult controls. Induction of diabetes resulted in a significant reduction in pancreatic protein concentration in both age groups; however, the decline was more pronounced in aged diabetic mice, indicating greater susceptibility to hyperglycemia-induced tissue damage.

Following treatment with ethanolic plant extracts, restoration of protein content was observed in both adult and aged diabetic mice. Nevertheless, the magnitude of recovery was comparatively higher in adult animals across all treatment groups. Among the extracts, *Mangifera indica* showed the most substantial improvement in protein levels in both age categories.

Overall, the graph highlights that aging exacerbates diabetes-induced pancreatic protein depletion and reduces the extent of biochemical recovery following therapeutic intervention.

Discussion

Alteration in tissue protein content is a well-recognized biochemical consequence of chronic hyperglycemia [1]. In the present investigation, induction of type 2 diabetes using alloxan resulted in a significant decline in tissue

protein levels in both adult and aged mice.[2] The marked reduction observed in diabetic animals may be attributed to enhanced protein catabolism, impaired protein synthesis, and increased oxidative stress associated with persistent hyperglycemia.

Alloxan is known to generate reactive oxygen species, which can damage cellular macromolecules, including structural and functional proteins. Oxidative modification of proteins may lead to fragmentation, denaturation, and enhanced degradation, thereby reducing total measurable protein content in tissues.[3] Additionally, insulin deficiency or resistance promotes increased proteolysis and reduced anabolic activity, further contributing to protein depletion.

Administration of ethanolic extracts of *Mangifera indica*, *Aegle marmelos*, and *Cynodon dactylon* significantly improved tissue protein concentration when compared with untreated diabetic groups [4]. The restoration of protein levels suggests that these plant extracts may exert protective effects through multiple mechanisms, including antioxidant activity, improvement of glycemic control, and modulation of metabolic pathways involved in protein synthesis [5].

Among the treated groups, the extract of *Mangifera indica* demonstrated comparatively greater efficacy in restoring protein levels in both adult and aged mice. This enhanced effect may be associated with the presence of bioactive phytoconstituents such as flavonoids, phenolic compounds, and other antioxidant molecules that help reduce oxidative stress and stabilize cellular proteins [9].

Although all extracts showed beneficial effects in aged mice, the magnitude of recovery was comparatively lower than in adult animals [7]. This difference may be related to age-associated decline in metabolic efficiency, reduced cellular repair mechanisms, and increased baseline oxidative stress in older animals. Aging is known to impair antioxidant defense systems, which may limit the responsiveness to therapeutic interventions [14].

Overall, the findings indicate that ethanolic extracts of the selected medicinal plants possess the ability to mitigate diabetes-induced protein depletion. The improvement in tissue protein content supports their potential role in protecting against metabolic and oxidative damage associated with type 2 diabetes.

Conclusion

The present investigation demonstrates that experimental diabetes leads to a pronounced reduction in pancreatic protein levels, and this depletion is further intensified under aging conditions. Administration of ethanolic extracts of *Mangifera indica*, *Aegle marmelos*, and *Cynodon dactylon* significantly improved tissue protein concentration in both adult and aged diabetic mice, with comparatively greater restoration observed in adult animals.

The protective effect of these plant extracts may be attributed to their potential antioxidant and metabolic regulatory properties, which help counteract diabetes-associated protein degradation. Overall, the findings support the therapeutic potential of selected medicinal plants in mitigating diabetes-induced biochemical alterations, particularly in age-related metabolic impairment.

Acknowledgment

I express my sincere gratitude to my research guide, Dr. R.D. Bodare, for his constant guidance, valuable suggestions, and encouragement throughout the course of this study.
His expertise and insightful comments greatly contributed to the successful completion of this research work.

Financial support and sponsorship

Nil.

Conflicts of interest

The authors declare that there are no conflicts of interest regarding the publication of this paper.

References

1. Aggarwal N, Shishu. A review of recent investigations on medicinal herbs possessing anti-diabetic properties. J Nutr Disord Ther. 2011;1(102):1–2.
2. American Diabetes Association. Diabetes vital statistics. Alexandria (VA): American Diabetes Association; 2001.
3. Arika WM, Nyamai DW, Agyirifo DS, Ngugi MP, Njagi ENM. In vivo antidiabetic effect of aqueous leaf extract of *Azadirachta indica* A. Juss in alloxan-induced diabetic mice. J Diabetic Complications Med. 2016;1(2):1–6.
4. Basu R, Rhinesmith SE. Insulin resistance and aging. Diabetes Spectr. 2004;17(3):159–167. doi:10.2337/diaspect.17.3.159
5. Bauer JM, Bosy-Westphal A. Sarcopenia and muscle function in the elderly: Implications for nutrition. Nutrition. 2009;25(1):1–6. doi:10.1016/j.nut.2008.10.006
6. **Bhatti R, Sharma S, Singh J, Singh A, Ishar MPS.** Effect of *Aegle marmelos* (L.) Correa on alloxan-induced early-stage diabetic nephropathy in rats. **Indian J Exp Biol.** 2013;51(8):650–655.
7. Chavarria AR, Motta M. Diabetes and aging: The biological link between type 2 diabetes and aging. Curr Diabetes Rev. 2018;14(5):331–338.

8. El-Shenawy NS, Abdel-Nabi IM. Hypoglycemic effect of *Cleome droserifolia* ethanolic leaf extract in experimental diabetes and its effect on non-enzymatic antioxidants, glycogen, thyroid hormone, and insulin levels. Diabetol Croat. 2006;35(1):15–22.
9. Lowry OH, Rosebrough NJ, Farr AL, Randall RJ. Protein measurement with the Folin phenol reagent. *J Biol Chem.* 1951;193:265–275.
10. Ifeanyichukwu I, Chika E, Emmanuel N, Anthonia O, Ngozi A, Agabus N. Medicinal efficacy of methanol and ethanol crude extracts of *Mangifera indica* leaf. J Microbiol Res. 2014;4(5):180–182.
11. Joseph B, Jini D. Insight into the hypoglycaemic effect of traditional Indian herbs used in the treatment of diabetes. Res J Med Plant. 2011;5(4):352–376.
12. Kandwal MK, Sharma ML. *Cynodon dactylon* (L.) Pers.: A self-treatment grass for dogs. Curr Sci. 2011;101(5):619–621.
13. Komatsu M, Takei M, Ishii H, Sato Y. Glucose-stimulated insulin secretion: A newer perspective. J Diabetes Investig. 2013;4(6):511–516. doi:10.1111/jdi.12094
14. Lenzen S. The mechanisms of alloxan- and streptozotocin-induced diabetes. Diabetologia. 2008;51(2):216–226.
15. Madhuri AS, Mohanvelu R. Evaluation of antidiabetic activity of aqueous extract of *Mangifera indica* leaves in alloxan-induced diabetic rats. Biomed Pharmacol J. 2017;10(2):1029–1035.
16. Szkudelski T. The mechanism of alloxan and streptozotocin action in beta cells of the rat pancreas. Physiol Res. 2001;50(6):537–546.
17. Walvekar MV, Pol SB, Deshmukh VM. Modulatory effect of fenugreek-loaded PLGA nanoparticles on lipofuscinogenesis in pancreas of alloxan-induced diabetic mice. Int J Pharm Clin Res. 2016;8(1):22–25.