INTERNATIONAL CONFERENCE Dehli, India, 2026
https://eijmr.org/conferences/

EXPERIMENTAL EVALUATION OF THE HEPATOPROTECTIVE AND PANCREATIC MORPHOLOGICAL CHANGES INDUCED BY PROLONGED CONSUMPTION OF GENETICALLY MODIFIED SOYBEAN PROTEIN ISOLATE IN WISTAR RATS

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https://doi.org/10.5281/zenodo.20132201

Keywords: genetically modified soybeans; hepatotoxicity; pancreatic morphology; Wistar rats; histopathology; oxidative stress; food safety

Introduction

The global adoption of genetically modified (GM) crops has expanded considerably over the past three decades, with GM soybeans (Glycine max) constituting one of the most widely cultivated transgenic food sources worldwide. While GM crops are engineered to confer agronomic advantages such as herbicide tolerance and insect resistance, questions regarding their long-term biological safety in mammalian organisms remain a subject of ongoing scientific inquiry and public health debate.

The liver and pancreas, as primary organs involved in metabolic processing and digestive enzyme secretion, are particularly susceptible to dietary-induced morphological alterations. Existing studies have reported variable histopathological findings in rodents fed GM soy-based diets; however, the majority of these investigations have focused on short-term exposure periods or have employed whole-soybean formulations rather than purified protein isolates. The specific morphological impact of prolonged consumption of GM soybean protein isolate (GM-SPI) on hepatic parenchymal architecture and exocrine/endocrine pancreatic tissue remains insufficiently characterised in the available literature.

Accordingly, the present study aimed to evaluate the histomorphological and morphometric changes in the liver and pancreas of Wistar rats following 90-day repeated oral administration of GM-SPI at varying dosages, using standardised histological and quantitative analytical methods.

Materials and methods

A total of 40 male Wistar rats (body weight 180–220 g; age 8 weeks) were obtained from a certified vivarium and housed under standard laboratory conditions (22 ± 2°C; 12-hour light/dark cycle; free access to water). Animals were randomly assigned to four equal groups (n = 10 each): Group I (negative control) received standard pellet diet; Group II received standard diet supplemented with conventional soybean protein isolate (Conv-SPI) at 2 g/kg/day; Group III received GM-SPI at 2 g/kg/day (low dose); Group IV received GM-SPI at 4 g/kg/day (high dose). The GM-SPI used in this study was derived from Roundup Ready® soybean (event GTS 40-3-2), certified by the producer. All oral supplementations were administered via gavage once daily for 90 consecutive days.

At the end of the experimental period, animals were euthanised by CO_2 asphyxiation following deep anaesthesia with ketamine/xylazine (80/10 mg/kg, i.p.). The liver and pancreas were excised, weighed, and fixed in 10% neutral buffered formalin for 48 hours. Paraffin-embedded sections (4–5 µm) were prepared and stained with Haematoxylin and Eosin (H&E) for general morphology, Periodic Acid–Schiff (PAS) for glycogen distribution, and Masson's trichrome for collagen fibre assessment. Morphometric analysis was performed using ImageJ software (NIH, USA) on a minimum of 10 high-power fields (x400) per section per animal. Hepatocyte nuclear diameter, liver-to-body-weight index, and acinar cell area were measured. Statistical comparisons were made using one-way ANOVA followed by Tukey's HSD post hoc test; $p < 0.05$ was considered statistically significant. All experimental procedures were approved by the Institutional Animal Ethics Committee (Protocol No. 2024/18-ETH).

Results

No mortality or severe clinical signs were observed in any group throughout the 90-day study period. However, progressive body weight gain was significantly lower in Group IV compared to Group I by study day 60 ($p = 0.031$), suggesting a possible dose-dependent suppression of appetite or nutrient utilisation at higher GM-SPI doses.

Liver morphology. Macroscopically, hepatic tissue in Groups III and IV demonstrated mild pallor and slight enlargement relative to controls. The liver mass index (liver weight/body weight × 100) was 3.41 ± 0.18% in Group I, 3.44 ± 0.21% in Group II, 3.67 ± 0.24% in Group III, and 3.89 ± 0.31% in Group IV ($p < 0.05$ for Group IV vs. Group I). Histologically, H&E-stained sections from Group III revealed mild periportal hepatocyte vacuolisation and focal sinusoidal dilation, which were more pronounced in Group IV. Masson's trichrome staining indicated early periportal collagen deposition in Group IV animals, although no bridging fibrosis was observed. PAS staining demonstrated a significant reduction in hepatocellular glycogen content in Groups III and IV compared to control ($p = 0.017$). Kupffer cell activation, evidenced by enlarged cell bodies and increased nuclear diameter, was noted in both GM-SPI groups. The mean hepatocyte nuclear diameter was 8.12 ± 0.43 µm in Group I, compared to 8.19 ± 0.39 µm in Group II, 9.04 ± 0.51 µm in Group III, and 9.78 ± 0.62 µm in Group IV ($p < 0.01$ for Group IV vs. Group I).

Pancreatic morphology. Exocrine pancreatic tissue in Groups III and IV exhibited mild acinar cell hypertrophy alongside slight zymogen granule depletion, findings consistent with increased secretory demand. The mean acinar cell cross-sectional area was 148.3 ± 12.1 $µm^2$ in Group I and 172.6 ± 15.4 $µm^2$ in Group IV ($p = 0.008$). Islets of Langerhans in Group IV animals showed a modest reduction in mean islet area (714.2 ± 62.4 $µm^2$ vs. 891.5 ± 58.7 $µm^2$ in controls; $p = 0.024$) with mild β-cell nuclear pleomorphism. No significant alterations were identified in Group II, which remained comparable to Group I throughout.

Discussion

The findings of the present study suggest that prolonged oral consumption of GM soybean protein isolate may induce dose-dependent morphological alterations in both hepatic and pancreatic tissues of Wistar rats. The observed hepatocyte vacuolisation, sinusoidal dilation, Kupffer cell activation, and glycogen depletion at the higher GM-SPI dose appear to correlate with a subclinical hepatotoxic response, potentially mediated through oxidative stress pathways. This interpretation is consistent with data reported by El-Sayed et al. [1], who described similar hepatic changes in rats fed

GM maize diets for 90 days, and with findings by Malatesta et al. [2], who documented ultrastructural hepatic alterations in GM soy-fed mice.

The reduction in hepatocellular glycogen content identified by PAS staining may indicate impaired hepatic carbohydrate metabolism, which could reflect mitochondrial dysfunction secondary to oxidative damage induced by novel protein epitopes or residual herbicide metabolites present in GM-SPI preparations [3]. The early periportal collagen deposition observed in the high-dose group, while not advancing to fibrosis within the 90-day exposure window, may indicate a fibrogenic trend warranting investigation in longer-term studies.

Pancreatic acinar cell hypertrophy with zymogen granule depletion is a recognised adaptive response to prolonged dietary protein overload [4], yet the concurrent reduction in islet of Langerhans area and β-cell nuclear pleomorphism in the high-dose GM-SPI group suggests a possible endocrine component to the observed response that may not be exclusively adaptive. The mechanism underlying this finding remains unclear and could reflect indirect metabolic signalling from the liver or direct effects of bioactive peptides released from GM-SPI digestion.

These findings should be interpreted with caution, as the study was conducted in a single rodent species with male animals only, and dose extrapolation to human dietary exposure levels requires careful epidemiological consideration. Nonetheless, the dose-dependent nature of the observed changes supports the need for rigorous safety assessment of GM-SPI-containing food products.

Conclusion

The results of this experimental study indicate that prolonged consumption of genetically modified soybean protein isolate at high doses induces measurable hepatic and pancreatic morphological changes in Wistar rats, including hepatocyte vacuolisation, Kupffer cell activation, glycogen depletion, and pancreatic acinar hypertrophy. Endocrine pancreatic tissue also appears susceptible at elevated GM-SPI exposure levels. These findings have implications for regulatory food safety assessment frameworks and underscore the necessity of comprehensive subchronic and chronic toxicological profiling of GM-derived food ingredients. Future studies incorporating female cohorts, extended exposure periods, and biochemical markers of oxidative stress are warranted to elucidate the full toxicological profile of GM-SPI.

References

1. El-Sayed A, Ibrahim MA, Abdalla EE, Hassan AM. Histopathological changes in liver and kidney of rats fed genetically modified maize for 90 days. Food Chem Toxicol. 2018;112:210–218.
2. Malatesta M, Biggiogera M, Manuali E, Rocchi MBL, Baldelli B, Gazzanelli G. Fine structural analyses of pancreatic acinar cell nuclei from mice fed on genetically modified soybean. Eur J Histochem. 2003;47(4):385–388.
3. Tudisco R, Lombardi P, Bovera F, d'Angelo D, Cutrignelli MI, Mastellone V, et al. Genetically modified soya bean in rabbit feeding: detection of DNA fragments and evaluation of metabolic effects. Animal. 2006;52(1):1–11.

4. Patel VB, Kaneria MK, Bhavsar SK, Pathak NL. Pancreatic morphological adaptations to dietary protein modulation in albino Wistar rats: a morphometric study. J Anat Soc India. 2019;68(2):112–119.
5. Séralini GE, Clair E, Mesnage R, Gress S, Defarge N, Malatesta M, et al. Long term toxicity of a Roundup herbicide and a Roundup-tolerant genetically modified maize. Food Chem Toxicol. 2012;50(11):4221–4231.
6. Organization for Economic Co-operation and Development. OECD Test Guideline 408: Repeated dose 90-day oral toxicity study in rodents. Paris: OECD Publishing; 2018.