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A Study on the Anti – Oxidative Stress Effect Spirulina Supplementation and the Improvement of Certain Biochemical Parameters in Female Rats with Alloxan – Induced Diabetes

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Abstract: This study aimed to evaluate the effects of Spirulina supplementation on oxidative and antioxidant status, as well as on key biochemical parameters—including functional markers and lipid profiles—in alloxan-induced diabetic rats. The study was conducted at Tikrit University over a period of 30 days, using female Wistar rats aged 12–13 weeks and weighing 135–200 g. Diabetes was induced in the animals using alloxan at a dose of 150 mg/kg of body weight. The animals were divided into four groups, with nine animals per group: Group 1 (healthy control group) and Group 2 (diabetic control group). Group 3: A group with induced diabetes treated with 20 ml of a Spirulina solution prepared from 10 g of Spirulina per 100 ml of water. Group 4: A group with induced diabetes treated with 20 ml of a Spirulina solution prepared from 15 g of Spirulina per 100 ml of water. Blood and tissue analyses revealed that the alloxan dose caused weight loss and elevated oxidative stress—driven by free radical production and the depletion and inhibition of antioxidants. This impacted biochemical parameters; specifically, it destroyed pancreatic beta cells and raised liver and kidney enzyme levels, resulting in tissue damage in these organs Compared to the groups that consumed Spirulina supplements, these groups showed weight improvement and a There was a significant decrease) $P < 0.01$ in blood glucose levels, insulin resistance, and glycated hemoglobin, as well as in liver and kidney enzyme levels, accompanied by a marked improvement in the functions of these organs. The results also revealed a significant increase ($p > 0.05$) in antioxidant levels—such as glutathione and glutathione peroxidase—and a reduction in malondialdehyde (a marker of oxidative stress).

Keywords Spirulina supplements, diabetes, oxidative stress, biochemical parameters, alloxan.

1. Introduction

Diabetes has recently become one of the most prevalent diseases. It results either from the inability of cellular receptors to utilize the hormone insulin to regulate blood sugar levels—leading to Type 2 diabetes—or from the failure of pancreatic beta cells to secrete insulin adequately—leading to Type 1 diabetes and gestational diabetes. Affecting all age groups—children, adolescents, and adults—it is projected to become the seventh leading cause of death by 2030 [1]. It is considered a non-communicable disease caused by a disorder in glucose metabolism [2]. The causes of diabetes vary among individuals, ranging from genetic factors and poor dietary habits to conditions such as pregnancy and its associated complications (gestational diabetes); these factors lead to health deterioration, disability, and death [3]. Slow wound healing, excessive thirst, and frequent urination are

considered key indicators of diabetes [4]. In addition to this, there are various other complications, such as diabetic nephropathy, fatty liver, and blurring vision [5]. The dietetic treatment is the ideal method of managing blood sugar levels and avoiding diabetes; choosing nutritious food items, including foods high in fiber, complex carbohydrates, and healthy fats, is an important dietary measure that helps in overcoming the seriousness of the condition due to their low glycemic index and antioxidants [6]. It is known that a high-fiber diet is the best option for a healthy lifestyle because it promotes a feeling of fullness and lowers glucose and inflammation due to the detoxification effect; however, although Metformin is the most common drug for diabetes treatment, it has many side effects, which led the scientists to look for natural substitutes like Spirulina, which are appreciated for their nutritional value and ability to increase the functioning of the endocrine system and immune system [7]. The Spirulina algae are believed to be an important source of food that plays nutritional and medicinal roles. The spirulina algae are some of the best dietary supplements due to the high content of essential minerals needed for the formation of the body such as essential amino acids, antioxidants, dietary fibers, iron, and vitamins (like thiamine, riboflavin, and niacin). Spirulina contains 65-70% proteins; *phycocyanin* is used as the major source of protein in Spirulina. These algae grow in fresh water or in saline lakes and even in artificially cultivated places; spirulina are so important that they have even been recommended to be grown in space for astronauts [8]. Spirulina algae are characterized by a high content of antioxidants that combat free radicals —which cause damage resulting from oxidative stress—restore the functional efficiency of internal organs such as the liver and kidneys, prevent hepatic fat accumulation, and promote satiety; this helps lower blood sugar levels, thereby playing a significant role in weight loss [9]. Additionally, the phycocyanin pigment found in Spirulina protects against diabetic nephropathy by dilating renal tubules, increasing bilirubin levels, inhibiting renal tissue deformation, and enhancing kidney function [10]. It also increases iron stores in mice during pregnancy and lactation, thereby helping to reduce the incidence of anemia [11]. Spirulina algae are considered safe (non-toxic) compounds; they reduce peroxide levels resulting from carbon tetrachloride-induced lipid peroxidation in mouse liver microsomes following the injection of phycocyanin [12], thereby preventing the progression of hepatic atrophy into liver fibrosis [13]. Its protective effect reduces oxidative stress in mice caused by overdoses of the antibiotic Tilmicosin (TIL) and protects the heart from damage [14]. Polyphenolic and carotenoid compounds in algal extracts have contributed to the treatment of diabetes due to their anti-inflammatory and antioxidant properties [15].

2. Methodology, Materials and Methods

Animals Female Wistar rats (n=36), weighing 135–200 g and aged 12–13 weeks, were used in this study. They were housed in cages within an animal facility under a 12-hour light/dark cycle for 30 days, with ad libitum access to water and standard feed. The animals were divided into four groups (9 animals per group):

- 1- Control group (healthy)
- 2- Control group (diseased)
- 3- Diseased group treated with 10 ml of prepared Spirulina solution (10 g Spirulina / 100 ml water)
- 4- Diseased group treated with 20 ml of prepared Spirulina solution (15 g Spirulina / 100 ml water);

note that the Spirulina supplements are from the Malaysian company DXN, which is authorized in Iraq

Induction of Diabetes in Rats

Alloxan was used to induce diabetes in female rats at a dosage of 155 mg/kg of body weight. The animals were fasted for one day prior to induction, and baseline blood glucose levels were measured. Alloxan—a chemical agent used experimentally to induce diabetes by destroying beta cells—was dissolved in a cold 0.9% physiological saline solution (concentration: 2–5 g/100 mL) and administered to the rats. Blood glucose levels were measured two days later; a level of 250 mg/dL or higher indicated the successful induction of diabetes. Additionally, glucose was administered during the experiment to prevent mortality.

Blood Analysis

The animals were fasted for a full day prior to dissection and blood collection. Blood glucose levels were measured, after which the animals were dissected and blood samples were collected and stored in specialized tubes for the required tests (blood glucose, glycated hemoglobin [HbA1c], kidney function, and liver function).

3. Results and Discussions

The results shown in Chart (1) indicate that the alloxan dose caused a significant decrease in body weight in the affected group (Group 2) compared to the control group (Group 1). In contrast, Groups 3 and 4, which consumed Spirulina, showed a clear difference and an improvement in body weight compared to Group 2.

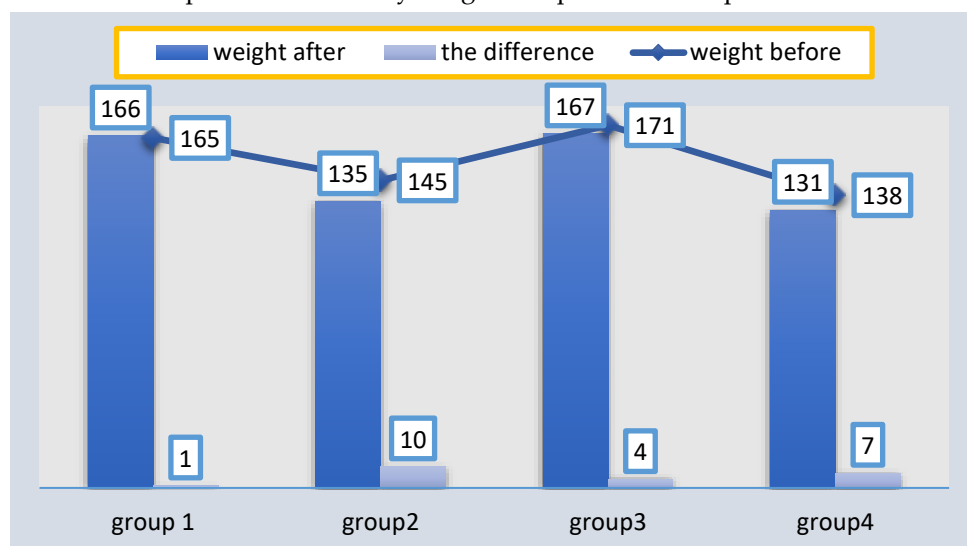


Figure 1. illustrates the change in body weight following the injury and the effect of Spirulina supplementation.

The results also showed elevated levels of blood glucose, glycated hemoglobin (HbA1c), and serum insulin in Group 2 compared to the healthy control group (Group 1); conversely, there was a significant decrease.

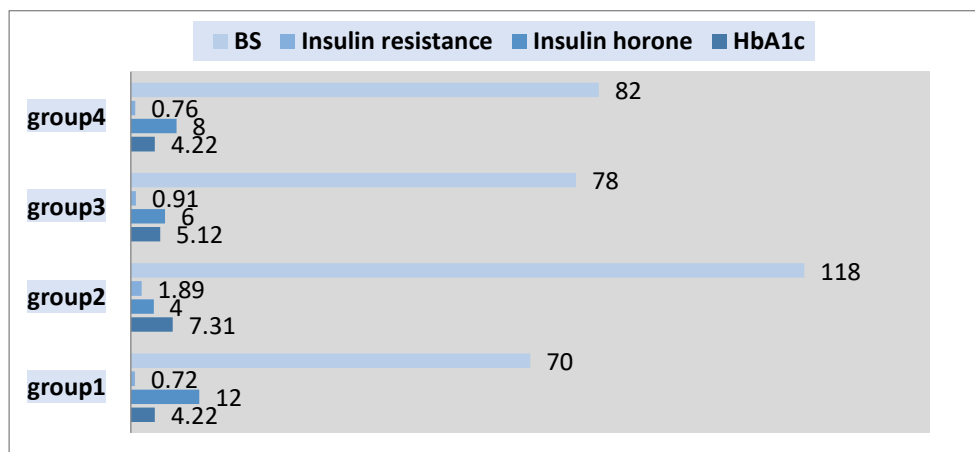


Figure 2. Effect of Spirulina supplementation on glucose and glycated hemoglobin (HbA1c) levels.

The above observations are in accordance with the results obtained by [16] who showed that in rats weighing 200-220 g, the induction of diabetes could be achieved via the use of two different dosages of streptozotocin (STZ), i.e., 45 and 60 mg/kg. The administration of the drug caused weight loss and increased blood glucose and HbA1c levels owing to the oxidative stress and dysfunction of organs. Also, according to [17], a dosage of 125 and 22 mg/kg of body weight of STZ was enough to cause the increase in blood glucose level by 0.9-6.4% in 24 hours after injection.

As seen from Chart (3), there were higher amounts of malondialdehyde (MDA) – an oxidative stress indicator – along with lower amounts of glutathione (GSH) and glutathione peroxidase (GPX) in the serum of the affected group (Group 2) than the healthy group (Group 3). On the other hand, the amount of MDA in the blood of groups 3 and 4, who took Spirulina, was lowered, while the amounts of GSH and GPX were raised than Group 2, due to the vitamins and antioxidant presence in Spirulina.

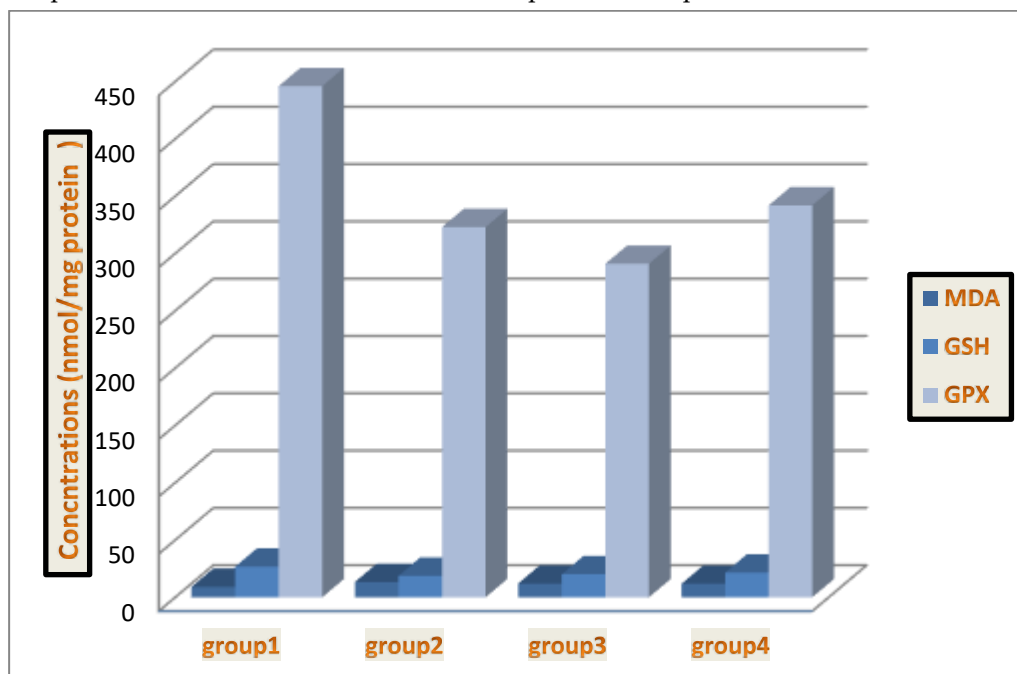


Figure 3. The effect of therapeutic Spirulina supplementation on increasing antioxidant levels.

This finding is consistent with the work done by [18], whose research showed the effectiveness of Spirulina supplementation in increasing blood plasma antioxidant activity

by 71% relative to a control group within a 7-week period of 5 mg per day. In a similar fashion, [19], examined the positive impact of Spirulina consumption in diabetes for a period of eight weeks; they concluded that Spirulina helped to achieve good health status through the elimination of free radicals, oxidative stress, and bad lipids as well as regulation of blood glucose levels. This is in agreement with the results obtained by [20], where it was observed that the consumption of Spirulina supplements with a dose of 128 mg/kg of body weight for 21-49 days facilitated the recovery process by increasing the antioxidants, resulting in the reduction of haloperidol-induced tardive dyskinesia in mice. In a similar study [21] examined the efficiency of Spirulina supplements (120 mg/kg) with a healthy diet in diabetic mice having diabetic nephropathy; the combination treatment caused increased activity of enzymes and non-enzymes antioxidants, which led to the normalization of free radicals and regulation of glucose levels.

As shown in figures (4) and (5), the findings indicated the negative influence of the alloxan dose on the functioning of kidneys and livers due to higher levels of inflammation brought about by free radicals in the serum of Group 2 as opposed to Group 1. In contrast, the findings from Groups 3 and 4 revealed that Spirulina administration was effective in improving biochemical measures and organ tissue structure through glucose management and antioxidant action.

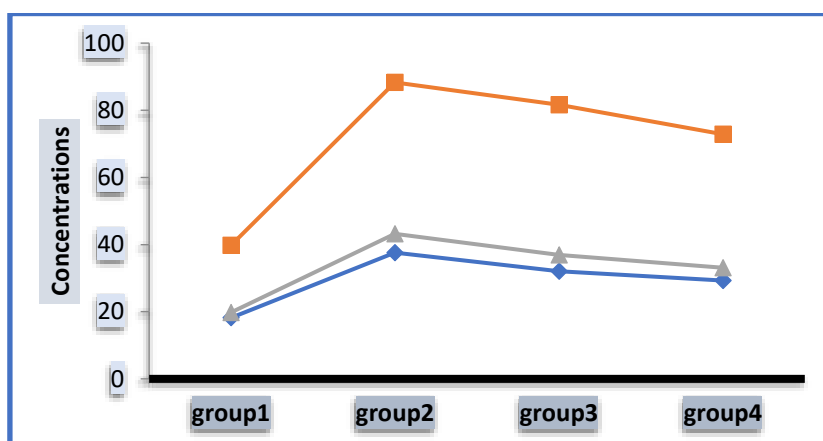


Figure 4. Effect of Spirulina supplementation on liver function in female diabetic rats

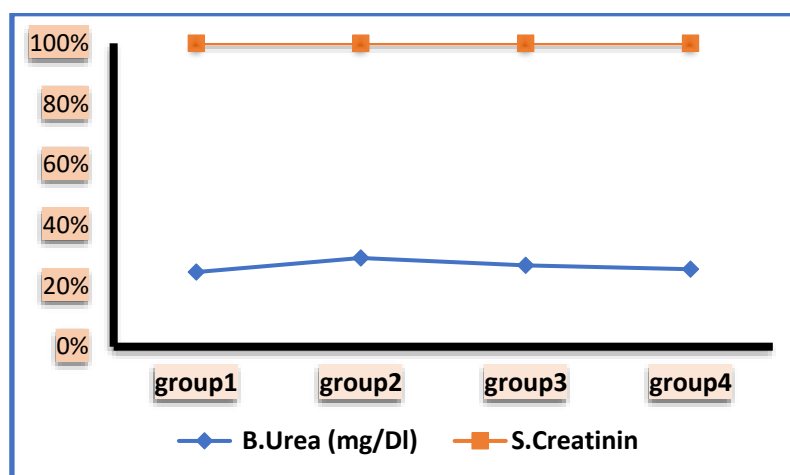


Figure 5. Effect of Spirulina supplementation on kidney function in female diabetic rats.

These findings are consistent with the results obtained by [22] concerning the effect of diabetes caused by alloxan on renal vasculature (glomeruli). This study revealed deposits in medullary interstitium as well as 70% thickening of the kidney wall which resulted in diabetic nephropathy because of poor filtration. Likewise, [23] in their study comparing Akita mice (model of type 1 diabetes) with the control group found a tenfold

increase in albumin secretion which was linked to nephrin secretion. The results coincide with those of [24], who verified that induction of diabetes with the use of streptozotocin leads to increase in the levels of blood glucose and creatinine and development of renal hypertrophy and dysfunction among affected rats. In turn, according to [25], the supplementation of *Spirulina platensis* at the dosage of 400 mg/kg for six weeks provided protection of kidney and liver tissues from the harmful effects; this was made possible due to decrease in blood glucose levels and oxidative stress resulting from excess of free radicals, owing to antioxidant nature of the supplement. In addition, [26] reported the effectiveness of *Spirulina*—a 25, 50, and 100 mg/kg dose given to mice for 10 days after the administration of alloxan—in enhancing certain biochemical parameters, mainly those associated with the pancreas and kidneys, through reducing urea, creatinine, and inflammation (TNF- α). Furthermore, [27] found that feeding rats with a 500 mg/kg dose of *Spirulina* for two months reduced blood glucose and inflammatory parameters, including MDA, TNF- α , and IL6, while suppressing protein expression in the MAPK (mitogen-activated protein kinase) pathway in liver tissues of STZ-induced diabetic rats. On the other hand, [28] stressed the role of *Spirulina* nanoparticles in restoring renal histological architecture in diabetic nephropathy of rats through their antioxidative and ROS-scavenging effects and effects on NRF2/HO-1 and NF- κ B pathways.

4. Conclusions

The current experiment showed that diabetes caused by alloxan administration induced significant metabolic and oxidative abnormalities in female Wistar rats. These included elevated levels of glucose, glycated hemoglobin, and oxidative stress along with decreased body weight and antioxidants in blood. Treatment with spirulina resulted in improvement in body weight and glycemic variables in comparison to the untreated diabetes group. Furthermore, there was enhancement in the level of antioxidants, as reflected by the increase in levels of glutathione and glutathione peroxidase and decrease in malondialdehyde, indicating the prevention of oxidative stress. Furthermore, the alterations in the biochemical functions of liver and kidney found in diabetic rats were reversed through *Spirulina* treatment. Such beneficial actions might be due to the antioxidant actions of *Spirulina*. Generally, the results indicate the possibility of *Spirulina* as a nutraceutical aid to enhance glucose metabolism, antioxidants, and certain biochemistry parameters of liver and kidney functions of alloxan-induced diabetic rats. Additional research with the use of standard dosages, larger number of animal groups, extended study periods, and extensive histopathology is suggested to determine the exact mechanism of action and dose dependency.

REFERENCES

- [1] O. Abu Al-Rub, "Diabetes: Symptoms, causes and treatment," 2023.
- [2] A. B. Kaiser, N. Zhang, and W. Van Der Pluijm, "Global prevalence of type 2 diabetes over the next ten years (2018–2028)," *Diabetes*, vol. 67, no. Suppl. 1, p. 202-LB, 2018.
- [3] F. F. Ferri, "Diabetes mellitus," *ClinicalKey*, 2022. [Online]. Available: <https://www.clinicalkey.com>. [Accessed: May 7, 2022].
- [4] A. B. Evert, A. Elson, and E. Nozi, "Nutrition therapy for adults with diabetes or prediabetes: A consensus report," *Diabetes Care*, 2019, doi: 10.2337/dci19-0014.
- [5] Y. Harika, "The best foods to lower blood sugar levels naturally," 2024.
- [6] F. Yasmin, "Foods that lower blood sugar," 2020.
- [7] S. Liao, "Is Spirulina good for you?" *Altibbi*, 2024. [Online]. Available: <http://www.altibbi.com>.

- [8] L. Burgess, "What are the benefits of spirulina?" *Altibbi*, 2024. [Online]. Available: <http://www.altibbi.com>.
- [9] Z. Jing *et al.*, "Phycocyanin and phycocyanobilin from *Spirulina platensis* protect against diabetic nephropathy by inhibiting oxidative stress," *Am. J. Physiol. Regul. Integr. Comp. Physiol.*, vol. 304, no. 2, pp. R110–R120, 2013, doi: 10.1152/ajpregu.00648.2011.
- [10] H. Thormar, "Patented non-antibiotic agents as animal feed additives," *Recent Pat. Food, Nutr. Agric.*, vol. 4, no. 2, pp. 155–168, 2012.
- [11] R. Kapoor and U. Mehta, "Supplementary effect of *Spirulina* on hematological status of rats during pregnancy and lactation," *Plant Foods Hum. Nutr.*, vol. 52, no. 4, pp. 315–324, 1998, doi: 10.1023/A:1008027408919.
- [12] V. B. Bhat and K. M. Madyastha, "C-Phycocyanin: A potent peroxyl radical scavenger in vivo and in vitro," *Biochem. Biophys. Res. Commun.*, vol. 275, no. 1, pp. 20–25, 2000.
- [13] N. Paniagua-Castro, G. Escalona-Cardoso, D. Hernández-Navarro, R. Pérez-Pastén, and G. Chamorro-Cevallos, "*Spirulina* (*Arthrospira*) protects against cadmium-induced teratogenic damage in mice," *J. Med. Food*, vol. 14, no. 4, pp. 398–404, 2011.
- [14] A. Ibrahim and M. Abdel-Daim, "Modulating effects of *Spirulina platensis* against tilmicosin-induced cardiotoxicity in mice," *Cell J.*, vol. 17, no. 1, pp. 137–144, 2015.
- [15] K. Skjånes, R. Aesoy, L. Herfindal, and H. Skomedal, "Bioactive peptides from microalgae: Focus on anti-cancer and immunomodulating activity," *Physiol. Plant.*, vol. 173, no. 2, pp. 612–623, 2021, doi: 10.1111/ppl.13472.
- [16] A. Ahmed, A. E. M. El-Mosallamy, N. Nabil, and N. M. E. Abo El-Nasr, "Dose dependent toxicity of streptozotocin inducing diabetic complications in male Sprague Dawley rats with metabolic biochemical histopathological effects," 2025.
- [17] S. R. Talbot, M. Heidari, M. W. W., A. Jörns, and O. Naujok, "Comparative analysis of dose response variability and severity in STZ-induced diabetes: Female vs. male NSG mice," *Sci. Rep.*, vol. 16, p. 8257, 2026.
- [18] M. S. Miranda, R. G. Cintra, S. B. Barros, and J. Mancini-Filho, "Antioxidant activity of the microalga *Spirulina maxima*," *Braz. J. Med. Biol. Res.*, vol. 31, no. 8, pp. 1075–1079, 1998, doi: 10.1590/S0100-879X1998000800007.
- [19] H. A. A. Rostami, M. Abdoljalal, M. Mohammed, R. Behdad, and M. Majid, "Effect of *Spirulina* on lipid profile, glucose and malondialdehyde levels in type 2 diabetic patients," *Braz. J. Pharm. Sci.*, vol. 58, p. e191140, 2022.
- [20] S. R. Thaakur and B. Jyothi, "Effect of *Spirulina maxima* on the haloperidol induced tardive dyskinesia and oxidative stress in rats," *J. Neural Transm.*, vol. 114, no. 9, pp. 1217–1221, 2007, doi: 10.1007/s00702-007-0744-2.
- [21] M. Gargouri, H. Harmel, A. Akrouti, X. Druart, C. Magné, and A. El Feki, "Effects of *Spirulina platensis* on lipid peroxidation, antioxidant defenses, and tissue damage in kidney of alloxan-induced diabetic rats," *Appl. Physiol. Nutr. Metab.*, vol. 43, no. 4, pp. 345–354, 2018, doi: 10.1139/apnm-2017-0461.
- [22] M. Parviz, E. Nasiri, and H. Sohrabi, "Early renal histological changes in alloxan-induced diabetic rats," *Int. J. Med. Cell.*, vol. 3, no. 1, 2014.
- [23] J. H. Chang *et al.*, "Diabetic kidney disease in FVB/NJ Akita mice: Temporal pattern of kidney injury and urinary nephrin excretion," 2012.
- [24] K.-M. Choi and H.-S. Yoo, "Amelioration of hyperglycemia-induced nephropathy by 3,3'-diindolylmethane in diabetic mice," *Molecules*, vol. 24, no. 24, p. 4474, 2019, doi: 10.3390/molecules24244474.
- [25] H. Ismail, A. Sakila, S. N. Nahida, W. Ummal, S. B. Abdus, A. Taskina, and R. Kazi, "Reno-pancreas protective effects of *Spirulina platensis* in alloxan induced diabetic rats," *Pak. J. Pharm. Sci.*, vol. 33, no. 4, 2020.
- [26] I. P. Joventino *et al.*, "The microalga *Spirulina platensis* presents anti-inflammatory action as well as hypoglycemic and hypolipidemic properties in diabetic rats," *J. Complement. Integr. Med.*, vol. 9, no. 1, 2012, doi: 10.1515/1553-3840.1534.

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- [27] K. M. Sadek, M. A. Lebda, S. M. Nasr, and M. Shoukry, "*Spirulina platensis* prevents hyperglycemia in rats by modulating gluconeogenesis and apoptosis via modification of oxidative stress and MAPK-pathways," *Biomed. Pharmacother.*, vol. 92, pp. 1085–1094, 2017, doi: 10.1016/j.biopha.2017.06.023.
- [28] A. Fayez *et al.*, "Exploring the NRF2/HO-1 and NF- κ B pathways: Spirulina nanoparticles as a novel approach to combat diabetic nephropathy," *ACS Omega*, 2024, doi: 10.1021/acsomega.4c02285.