Pharmaceutical Sciences & Analytical Research Journal
ISSN: 2640-6659

Academic Strive
Researcher's perception is our reality

*Research Article* *Volume 6 Issue 3*

# Study of High-Fructose Diet Induced Metabolic Syndrome: An Experimental Study on Rats

**Chetehouna S[1,2], Derouiche S[3,4]*, Boulaares I[3,4] and Reggami Y[2,5]**

[1]Department of Microbiology and Biochemistry, University of M'sil, Algeria
[2]Laboratory of Biology: Applications in Health and Environment, University of M'sila, Algeria
[3]Department of Cellular and Molecular Biology, El Oued University, Algeria
[4]Laboratory of Biodiversity and application of biotechnology in the agricultural field, El Oued University, Algeria
[5]Department of Natural and Life Sciences, University in Skikda, Algeria

***Corresponding author:** Samir Derouiche, Department of Cellular and Molecular Biology, Laboratory of Biodiversity and Application of Biotechnology in the Agricultural Field, Faculty of Natural Sciences and Life, University of El-Oued, El-Oued 39000, Algeria, Email: dersamebio@gmail.com

**Received Date:** July 03, 2024; **Published Date:** July 25, 2024

## Abstract

Metabolic syndrome (MetS) is a combination of metabolic disorders that co-occur, among them dyslipidemia, insulin resistance, hyperglycemia, oxidative stress, and pro-inflammatory states. It further increases the risk of developing different chronic diseases, which can even lead to death, as reported by international health institutions. This study assessed the impact of a high-fructose diet (HFD) on the development of metabolic syndrome in rats. Twelve male rats were randomly split into two groups: six rats in each group; the control group; and the HFD group (35%). Both groups had assessments of their growth parameters and biological markers. The obtained results demonstrated that HFD significantly increased the levels of liver glycogen, lipid profile, and transaminase enzymes activities when compared to control group. Furthermore, compared to the control group, the HFD group showed a significant increase in malondialdehyde (MDA) level and a significant decrease in reduced glutathione (GSH) level, glutathione peroxidase (GPx), and superoxide dismutase (SOD) activities. Moreover, histological analysis revealed a significant alteration of hepatocytes architecture and appearance of lipid droplet accumulation in the hepatic tissue of the HFD group. In summary, fructose is a potent inducer of metabolic syndrome in rats by affecting many bodily metabolisms.

**Keywords:** Metabolic Syndrome; High Fructose Diet; Oxidative Stress; Biochemical Markers

## Abbreviations

MetS: Metabolic Syndrome; HFD: High-Fructose Diet; MDA: Malondialdehyde; GPx: Glutathione Peroxidase; SOD: Superoxide Dismutase; ROS: Reactive Oxygen Species; EDTA: Ethylenediaminetetraacetic Acid; CRSTRA: Center of Scientific and Technical Research on Arid Regions; GOT: Glutamate Oxaloacetate Transaminase; GPT: Glutamate Pyruvate Transaminase; TG: Triglycerides; TC: Total Cholesterol; HDL-C: High-Density Lipoprotein Cholesterol;

VLDL-C: Very Low Density Lipoprotein Cholesterol; TBS: Tris Buffer Solution; BSA: Bovine Serum Albumin; NAFLD: Non-Alcoholic Fatty Liver Disease.

## Introduction

Metabolic syndrome (MetS) is known as a complex condition involving interrelated variables such as insulin resistance, atherosclerotic disease, dyslipidemia, endothelial dysfunction, low testosterone levels, and low-grade inflammation [1]. MetS raises the risk of cardiovascular problems, nonalcoholic fatty liver disease, diabetes mellitus, high blood pressure, and potentially death [2]. The link between MetS and the aforementioned related disorders is the intracellular redox imbalance and oxidative stress brought on by the persistent inflammatory conditions that distinguish MetS; the increase in oxidant species production in MetS has been identified as a major underlying mechanism for mitochondrial dysfunction, accumulation of lipid and protein oxidation products, and impairment of the antioxidant systems [3]. A high-calorie diet that includes high carbohydrates, high fat, high fructose, and other foods can induce the metabolic syndrome in animal models, according to experimental studies. This can lead the animals to acquire different features of the metabolic syndrome, such as insulin resistance, hyperglycemia, lipid disorders, hypertension, pro-inflammatory, and oxidative stress states [4]. In recent decades, high fructose intake has been identified as a possible cause of MetS in statistical investigations. Fructose is a glucose isomer that is frequently utilized in modern industries [5]. Simple ketose monosaccharide, fructose, penetrates enterocytes via the small intestine's brush surface and is immediately absorbed by the glucose transporter (GLUT 5). GLUT2, which is located in the basolateral pole of the enterocytes, then transports it from the enterocytes to the blood [6]. The largest amount of fructose is expected to be absorbed and processed in the liver's cells through its portal circulation when it enters the blood stream. As a result, direct or indirect metabolites from the liver's metabolism of fructose contribute to at least some of the load of fructose metabolism on extrahepatic organs [7]. The synthesis of dihydroxyacetone phosphate, which is used to produce glycerol—a prerequisite for lipogenesis activities-and glyceraldehyde-which joins the glycolytic or gluconeogenic route-is catalyzed by fructokinase and aldolase B in the fructolysis pathway [8]. Triose kinase is also necessary for the metabolism of fructose since it causes the liver to store more fat [9]. Unlike glucose, whose metabolism is carefully regulated by phosphofructokinase, fructose metabolism is not restricted, meaning that fructolysis has limitless potential. A large amount of substrate is generated and used in multiple metabolic pathways (the processes of glycolysis, glucose production, glycogenesis, and aerobic phosphorylation) that correspond with cellular needs since there is a lack of regulation [10]. Increased synthesis and secretion of extremely light-density lipoproteins results from elevated fructose levels stimulating the synthesis of malonyl CoA, which inhibits fatty acid oxidation and promotes triglyceride accumulation in hepatocytes [11]. Fructose also increases insulin resistance, which modifies lipid profiles in the liver and plasma and increasing diacylglycerol [7]. Further, the infinite fructolysis requires increased ATP synthesis for phosphofructokinase to phosphorylate fructose, which raises the creation of reactive species by further activating oxidative phosphorylation. In both hepatic and extrahepatic organs, they can interfere with the insulin signaling pathway [7]. Oxidative stress is known to be a major factor in the inflammatory response and to be a promoter of inflammation. Reactive oxygen species (ROS) are produced in significant quantities by mitochondria as a result of the electron transport chain's operation. By increasing the level of oxidative stress next to the mitochondrial DNA, this raises the chance of DNA damage. If the processes governing DNA repair were disturbed, stress levels would rise, and insulin resistance may result [12]. In addition to changing transcription factors and stimulating the generation of proinflammatory cytokines, oxidative stress also causes metabolic issues such as lipid abnormalities and impaired glucose tolerance, which ultimately result in MetS [13]. This study aimed to evaluate the harmful effect of high fructose diet for inducing metabolic syndrome in albino wistar rats.

## Materials and Methods

### Collection of Plant Sample

Sonchus maritimus was obtained in November from Djamaa town in El-Oued state, Algeria, and their taxonomy was confirmed by Pr. Halis Youcef, an expert in botany, at the Center of Scientific and Technical Research on Arid Regions (CRSTRA), Touggourt state, Algeria. After being thoroughly rinsed with distilled water, the leaves were let dry at the ambient temperature. They were crushed into powder and kept at ambient temperature until needed.

### Preparation of *Sonchus maritimus* Leaves Extract

Ten grams of dry *Sonchus maritimus* powder were mixed with one hundred milliliters of distilled water to produce the aqueous extract. After filtering through filter paper, the mixture was macerated for 24 hours at ambient temperature and then dried on a stove at 50°C [14].

### Animals

Twelve male albino Wistar rats, weighing 173.08± 3.48 g and aged 7–8 weeks, were acquired from the Pasteur National Institute of Algiers. The rats were kept in plastic cages at the Echahid Hamma Lakhdar-El Oued University in Algeria's Animal House of Natural and Life Sciences Faculty.

Standard conditions were maintained for the animals, with a temperature of 25 ± 2°C and a 12/12 h light/dark cycle. The animals were accustomed to this environment throughout the trials by being fed a normal diet and having unlimited access to water. The Ethics Committee of the Department of Cellular and Molecular Biology, Faculty of Natural Sciences and Life, University of El-Oued, Algeria, cited (06 EC/DCMB/FNSL/EU2021), ensured that all experimental protocols followed international norms.

### Experimental Design

After two weeks of acclimatization, the rats were haphazardly divided into two groups of 6 as following:
**Group 01 (Control):** control group received standard diet;
**Group 02 (HFD):** experimental group received high-fructose diet group;

For 13 weeks, the experimental rats were fed a diet containing 35% fructose. The rats were weighed once a week and measurements were taken of their water and food.

### Sacrifice, Blood Sampling and Tissue Collection

After a 12-hour fast and at the end of the experiment, the animals were put to death while under a mild anesthesia caused by inhaling chloroform (94%) into their mouths. Blood samples were taken during the decapitation process and placed into numbered ethylenediaminetetraacetic acid (EDTA) tubes for each rat. The plasma was then separated by centrifugation at 1500 rpm for 10 minutes, and the samples were kept at -20°C until the biochemical parameters and lipid profile were evaluated. Each rat›s fasting blood glucose was measured using a glucometer (Vital Chek®, China). The liver was thoroughly removed, cleaned in 0.9% sodium chloride (NaCl), and ws weighed before being frozen at -20°C to prepare homogenates for measuring oxidative stress, glycogen, protein, and lipid peroxidation.

### Plasma Biochemical Parameters

The plasma's glutamate oxaloacetate transaminase (GOT), glutamate pyruvate transaminase (GPT), and lipid profile, which includes triglycerides (TG), total cholesterol (TC), and high-density lipoprotein cholesterol (HDL-C), were measured employing commercial reagent kits (Bio Lab, France, and Spin, Spain) using Mindray BS-200, China. While glycosylated hemoglobin was measured utilizing the same reagent kits but using Medcoon, Germany.

The following equation was used to determine the levels of very low density lipoprotein cholesterol (VLDL-C) and low density lipoprotein cholesterol (LDL-C) [15]:
VLDL-C = Triglyceride / 5LDL-C = TC – VLDL-C - HDL-C

### Preparation of Homogenate and Tissue Biochemical Parameters

Each part of the rat's liver tissue was homogenized in a cold Tris buffer solution (TBS, pH = 7.4). The obtained homogenate was subsequently centrifuged for 15 minutes at 4°C at 5000 rpm, and the supernatant was utilized to measure oxidative stress markers and lipid peroxidation. The homogenate's protein content was quantified utilizing the Bradford method, with bovine serum albumin (BSA) serving as the standard [16]. Using the anthrone reagent, liver glycogen was calculated using the Duvhåteau and Florkin technique [17].

### Oxidative Stress Markers

The homogenates were subjected to estimate the MDA and GSH levels according to the procedures described by Yagi [18]; Weckbecker, et al. [19], respectively. The methodologies provided by Flohe, et al. [20]; Beauchamp, et al. [21] were used to measure the GPx (EC 1.11.1.9) and SOD (EC 1.11.1.9) activities in the homogenates, respectively.

### Histological Analysis

After the sacrifice, each rat's liver was removed and fixed for 48 hours in a solution containing 10% formaldehyde and phosphate buffer with a pH of 7.6, dried in ethanol gradations, cleaned with toluene, and then placed into paraffin blocks. The submerged samples were cut into 5µm-thick pieces using a rotator microtome; subsequently, they were stained with hematoxylin-eosin. An optical microscope (Optika B-293, Italy) with a camera (Optika C-B5, Italy) was used to do the histological examination. Optika, an image processing program, was used to analyze photomicrographs.

### Statistical Analysis

The results are presented as mean ± SEM (mean ± standard error of mean), and the data analysis based on the comparison of two means was conducted using the Student's T test. The curves and tests were performed with the assistance of the MINITAB software (Version 19) and EXCEL (Version 2019).

## Results and Discussion

### Growth Parameters

The initial body weights of the rats in both groups were statistically identical. The final body weight, food consumption, and water intake of the experimental group showed a highly significant reduction ($P < 0.001$) when compared to the control rats. Also, there was a significant increase ($P < 0.001$) in relative liver weight in the HFD group in comparison to the control (Table 1).

According to earlier research findings, mice that were given a high-fructose diet in drinking water for a 10-week period lost their final body weight and consumed less food and liquids overall [22]. A rise in metabolic needs opposes the maintenance of a steady body weight, whereas variations in cellular energy use are linked to the maintenance of either a decreased or raised body weight [23]. It was established that rats on an HFD developed insulin resistance without becoming obese [24]. Moreover, fructose supplementation has been shown to reduce food consumption in the past [25]. Additionally, the infinite fructolysis requires increased ATP synthesis for phosphofructokinase to phosphorylate fructose, which raises the creation of reactive species by further activating oxidative phosphorylation. These may interfere with the insulin signaling system in the liver and extrahepatic tissues, which could be responsible for the rats' decreased final body weight [7]. Necrosis may be the cause of the increased relative liver weight, according the histology section [26]. Also other study confirmed the damage that induced by the high fructose diet in the liver [27].

| Parameters | Control | HFD |
|---|---|---|
| | (n=6) | (n=6) |
| Initial body weight (g) | 179.17±4.72 | 173.33±6.38 |
| Final Body Weight (g) | 217.50±4.91 | 127.77±5.41*** |
| Relative liver Weight (g/100g b.w) | 2.1776±0.0178 | 3.200±0.133*** |
| Food intake (g/rat/day) | 9.500±0.0990 | 5.160±0.0317*** |
| Water intake (ml/rat/day) | 20.815±0.004 | 8.250±0.205*** |

**Source:** Values are expressed on (mean ± SEM): * P<0.05, **P<0.01, ***P<0.001: comparison with control group.
**Table 1:** Growth parameters of control and HFD groups.

## Biochemical Parameters

Table 2 shows a significant alteration in the biochemical markers of the HFD group. Specifically, our findings revealed a highly significant (P<0.001) rise in glycated hemoglobin and blood glucose, transaminase activities (GPT and GOT activities), and liver protein levels. The lipid profile was also significantly increased (P<0.05), including cholesterol, triglyceride, VLDL-C, and LDL-C levels, compared to the control, while a significant decrease was observed in HDL-C and liver glycogen levels.

Other studies confirm the high-fructose diet's ability to elevate blood glucose levels and modify lipid profile [28,29]. Increased blood glucose levels in fructose-fed animals [30] as a result of insulin resistance [31] have been linked to an increase in the quantity of glycosylated hemoglobin in the blood. The reduction of liver glycogen level is associated with resistance to insulin and glucose tolerance; also, fructose cannot boost the secretion of insulin from the beta cells of the pancreas due to the fructose transporter, GLUT 5, which is not extremely abundant in the described cells [32]. Fructose consumption causes insulin resistance-a condition in which cells do not respond to insulin as intended-insulin is an efficient lipoprotein lipase activator that stimulates the breakdown of triglycerides in lipoproteins (VLDL, chylomicrons) and improves LDL clearance via the LDL-receptor pathway. Accordingly, the state of insulin resistance is linked to the majority of lipid abnormalities [33,34]. Hepatocytes contain the enzyme HMG-CoA reductase, which controls the rate at which cholesterol is synthesized [35]. Studies have shown that when rats were given 20% fructose in their drinking water over a 12-week period, HMG-CoA reductase synthesis and activity were increased. This led to an excess of endogenous cholesterol being created [36]. The fundamental processes by which high fructose induces dyslipidemia are poorly understood [37]. Overconsumption of fructose has been connected to the buildup of fat in the liver, which can result in non-alcoholic fatty liver disease (NAFLD), which harms the liver and irritates hypatocytes in addition to oxidative stress [38]. Through a number of methods, oxidative stress may change the structure of macromolecules, including proteins, and even cause the death of cells. Among them, it can react with lipid peroxidation or interact with pro-oxidants to add a carbonyl group to protein molecules [39,40]. Proteases often cannot break down the huge masses of carbonylated proteins that develop in hepatocytes [41].

| Parameters | Control | HFD |
|---|---|---|
| | (n=6) | (n=6) |
| Blood Glucose (mg/dL) | 113.50±1.16 | 153.67±3.69*** |
| Glycosylated Hemoglobin (%) | 6.9±0.0516 | 7.5±0.0258*** |

| | | |
|---|---|---|
| Liver glycogen (mg/g of tissue) | 4.868±0.290 | 1.845±0.164*** |
| Triglyceride (g/L) | 1.6067±0.0429 | 2.04±0.0459*** |
| Total cholesterol (g/L) | 0.6667±0.0166 | 0.9825±0.0210*** |
| HDL-Cholesterol (g/L) | 0.216±0.0015 | 0.1867±0.0107* |
| LDL-Cholesterol (g/L) | 0.1785±0.0119 | 0.4473±0.0400*** |
| VLDL-Cholesterol (g/L) | 0.321±0.008 | 0.408±0.00918*** |
| GOT activity (IU/L) | 116.56±3.62 | 193.2±21.5* |
| GPT activity (IU/L) | 46.05±1.98 | 66.58±3.19*** |
| Protein of liver (mg/g of tissue) | 4.130±0.133 | 6.725±0.150*** |

**Source:** Values are expressed on (mean ± SEM): * P<0.05, **P<0.01, ***P<0.001: comparison with control group.
**Table 2:** Biochemical markers of control and HFD groups.

## Oxidative Stress Parameters

According to our results, the HFD group's liver MDA level increased significantly (P<0.001) in comparison to the control group. Additionally, relative to the control group, the comparison showed that the high-fructose diet feeding resulted in a very significant (P<0.001) decrease in enzymatic antioxidants, such as SOD and GPx activities, and no enzymatic antioxidants, such as GSH and GPx/GSH rate, in the liver homogenates of the HFD rats (Table 3).

MetS increases the generation of reactive species, which modifies the antioxidant balance and permits the cells to hold onto more reactive species [42]. Consuming too much fructose causes the antioxidant defense system to become unbalanced and increases the generation of free radicals in different tissues [43,44]. Other research has validated our results, showing that increased liver MDA is a critical indicator of oxidative damage to the liver cells [45]. Lipid susceptibility to peroxidation and oxidative damage increases when there are deficiencies in the functioning of the enzymatic antioxidant system as well, such as superoxide dismutase (SOD) and glutathione peroxidase (GPx), together with a decrease in total glutathione. These changes could happen before oxidative stress manifests itself [32]. According to Montesano, et al. [46] hepatic fat accumulation results in a reduction of SOD, a major and fundamental antioxidant defense line.

| Parameters | Control (n=6) | HFD (n=6) |
|---|---|---|
| MDA (nmol/mg of prot) | 6.006±0.620 | 11.258±0.338*** |
| GSH (nmol/mg of prot) | 1.9879±0.0193 | 1.1164±0.0548*** |
| SOD activity (mUI/mg of prot) | 2.09±0. 37 | 0.76±0.04*** |
| GPx activity (µmol/mg of prot) | 42.249±0.769 | 26.161±0.496*** |
| (GPx /GSH) x$10^3$ | 23.223±0.178 | 18.518±0.054*** |

**Source:** Values are expressed on (mean ± SEM): * P<0.05, **P<0.01, ***P<0.001: comparison with control group.
**Table 3:** Lipid peroxidation and antioxidant markers of liver in control and HFD groups.

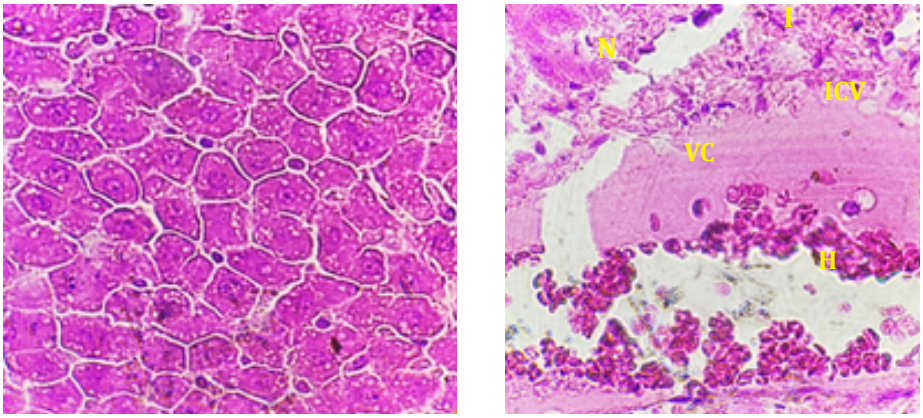
## Histopathology Study

The histological analysis of the liver tissue section of the control group was shown in Figures 1 and table 4, which showed normal hepatocyte structure. In contrast, the HFD group showed significant alterations, such as necrosis, hemorrhage, inflammatory cell infiltration, and cytoplasm vacuolization with a number of intracytoplasmic vesicles (lipid droplet vacuoles).

The histological modification of the liver caused by a high fructose intake was verified by Mirzaei, et al. [47] who confirmed that a high fructose intake helps to boost the creation of free radicals in the tissues, which causes inflammation and the development of vacuolar degradation of cytoplasm in liver tissue. As previously mentioned, hepatocytes have disseminated intracytoplasmic vacuoles, which are lipid droplets in the cytosol that develop as a result of fructose-induced lipid dysfunction. These histologic

alterations in the structure of liver cells have been linked to increased steatosis and damage, which are related to the infiltration of inflammatory cells and the activation of macrophage marker expression [48]. Due to increased mitochondrial oxidative activity brought on by excessive lipid inflow into the liver, ROS generation advances and inflammatory cytokine production increases, ultimately resulting in cell necrosis [49].

**Control Group HFD Group**




**Source:** I: inflammation; N: necrosis; VC: vacuolar cytoplasm; H: hemorrhage; ICV: intracytoplasmic vacuole.

**Figure 1:** Histological photomicrographs of liver section in control and HFD groups with hematoxylin and eosin (H&E), at magnification ×400.

| Parameters | Control | HFD |
|---|---|---|
| Inflammation | - | +++ |
| Necrosis | - | +++ |
| Hemorrhage | - | +++ |
| Cytoplasmic vacuolization | - | +++ |
| Intracytoplasmic vacuoles | - | ++ |

**Source:** None (-) ; Moderate (+) ; Severe (++) ; Very Severe (+++).

**Table 4:** Grading of histological changes in liver and brain sections of control and HFD groups.

## Conclusion

A high-fructose diet can induce metabolic syndrome in Albino Wistar rats through the observed harmful effect of providing hyperlipidemia, the hyperglycemia effect, which is followed by an oxidative stress state and histological alteration of liver tissue.

## References

1. Gorbachinsky I, Akpinar H, Assimos DG (2022) Metabolic syndrome and urological diseases. Rev Urol 12(4): 157-180.

2. Shen WC, Sun ZJ, Chou CY, Tsung CY, Hwa LF, et al. (2022) Association of simple renal cysts with metabolic syndrome in adults. Front Public Heal 10: 1-8.

3. Vona R, Gambardella L, Cittadini C, Straface E, Pietraforte D (2019) Biomarkers of oxidative stress in metabolic syndrome and associated diseases. Oxid Med Cell Longev, pp: 1-19.

4. Lasker S, Rahman MM, Parvez F, Mushfera Z, Pintu M, et al. (2019) High-fat diet-induced metabolic syndrome and oxidative stress in obese rats are ameliorated by yogurt supplementation. Sci Rep 9(1): 1-15.

5. Semnani AZ, Khan TA, Blanco MS, Russell JS, Lawrence AL, et al. (2020) Association of Major Food Sources of Fructose-Containing Sugars with Incident Metabolic Syndrome: A Systematic Review and Meta-analysis. JAMA Netw Open 3(7): 1-15.

6. Jones HF, Butler RN, Brooks DA (2011) Intestinal fructose transport and malabsorption in humans. Am J Physiol - Gastrointest Liver Physiol 300(2): 202-206.

7. Pan Y, Kong LD (2018) High fructose diet-induced metabolic syndrome: Pathophysiological mechanism and treatment by traditional Chinese medicine. Pharmacol Res 130: 438-450.

8. Tappy L, Rosset R (2017) Fructose Metabolism from a Functional Perspective: Implications for Athletes. Sport

Med 47(S1): 23-32.

9. Liu L, Li T, Liao Y, Wang Y, Gao Y, et al. (2020) Triose Kinase Controls the Lipogenic Potential of Fructose and Dietary Tolerance. Cell Metab 32(4): 605-618.

10. Chetehouna S, Derouiche S, Reggami Y (2024) *In Vitro* Antioxidant and Antidiabetic properties of leaves aqueous extract of *Sonchus maritimus*. Int J Chem Biochem Sci 25(19): 1-8.

11. Coronati M, Baratta F, Pastori D, Ferro D, Angelico F, et al. (2022) Added Fructose in Non-Alcoholic Fatty Liver Disease and in Metabolic Syndrome : A Narrative Review. Nutrients 14(6): 1-10.

12. Ziolkowska S, Binienda A, Jablkowski M, Szemraj J, Czarny P (2021) The interplay between insulin resistance, inflammation, oxidative stress, base excision repair and metabolic syndrome in nonalcoholic fatty liver disease. Int J Mol Sci 22(20): 1-27.

13. Lee H, Jose PA (2021) Coordinated Contribution of NADPH Oxidase- and Mitochondria-Derived Reactive Oxygen Species in Metabolic Syndrome and Its Implication in Renal Dysfunction. Front Pharmacol 12: 1-18.

14. Derouiche S, Abbas K, Djermoune M (2017) Polysaccharides and Ascorbic Acid Content and the Effect of Aqueous Extract of *Portulaca Oleracea* in High-Fat Diet-Induced Obesity, Dyslipidemia and Liver Damage in Albino Wistar Rats. Alger J Arid Environ 7(2): 16-26.

15. Friedewald WT, Levy RI, Fredrickson DS (1972) Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. Clin Chem 18(6): 499-502.

16. Bradford MM (1976) A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. Anal Biochem 72(1): 248-254.

17. Duvhateau G, Florkin M (1959) On Insect Trehalosemia and Its Significance. Arch Int Physiol Biochim 67(2): 306-314.

18. Yagi K (1976) A simple fluorometric assay for lipoperoxide in blood plasma. Biochem Med 15(2): 212-216.

19. Weckbecker G, Cory JG (1988) Ribonucleotide reductase activity and growth of glutathione-depleted mouse leukemia L1210 cells *in vitro*. Cancer Lett 40(3): 257-264.

20. Flohe L, Gunzler WA (1984) Assays of glutathione peroxidase. Methods Enzymol 105: 114-120.

21. Beauchamp C, Fridovich I (1971) Superoxide dismutase: Improved assays and an assay applicable to acrylamide gels. Anal Biochem 44(1): 276-287.

22. Akar F, Uludag O, Aydin A, Yasin AA, Sehri E, et al. (2012) High-fructose corn syrup causes vascular dysfunction associated with metabolic disturbance in rats: Protective effect of resveratrol. Food Chem Toxicol 50(6): 2135-2141.

23. Chetehouna S, Derouiche S, Reggami Y, Boulaares I (2024) *Sonchus maritimus* extracts-loaded niosomes bioconjugated by linoleic acid enhance high fructose diet-induced hepatic encephalopathy in albino Wistar rats. Arch Razi Inst 79(1): 189-200.

24. Bugga P, Mohammed SA, Alam MJ, Jahangir A, Parmeshwar K, et al. (2022) Empagliflozin prohibits high-fructose diet-induced cardiac dysfunction in rats via attenuation of mitochondria-driven oxidative stress. Life Sci 307: 120862.

25. Gancheva S, Zhelyazkova SM, Galunska B, Chervenkov T (2015) Experimental models of metabolic syndrome in rats. Scr Sci Medica 47(2): 14-21.

26. Derouiche S, Chetehouna S, Atoussi W (2022) The effects of aqueous leaf extract of Portulaca oleracea on haemato-biochemical and histopathological changes induced by sub-chronic aluminium toxicity in male wistar rats. Pharmacol Res - Mod Chinese Med 4: 100101.

27. Lu Y, Wu Y, Chen X, Yang X, Xiao H (2021) Water extract of shepherd's purse prevents high-fructose induced-liver injury by regulating glucolipid metabolism and gut microbiota. Food Chem 342: 128536.

28. Feyisa TO, Melka DS, Menon M, Labisso WL, Habte ML (2019) Investigation of the effect of coffee on body weight, serum glucose, uric acid and lipid profile levels in male albino Wistar rats feeding on high-fructose diet. Lab Anim Res 35(1): 29.

29. Said MA, Nafeh NY, Abdallah HA (2021) Spexin alleviates hypertension, hyperuricaemia, dyslipidemia and insulin resistance in high fructose diet induced metabolic syndrome in rats via enhancing PPAR-ɣ and AMPK and inhibiting IL-6 and TNF-α. Arch Physiol Biochem 129(5): 1111-1116.

30. Chetehouna S, Derouiche S, Boulaares I, Reggamie Y (2024) Phytochemical profile, anti-inflammatory analysis and cytotoxic activity of SmE-SeNPs against

breast (MCF-7) cancer cells. Biocatalysis and Agricultural Biotechnology 57: 103122.

31. Hadzhibozheva P, Pashova-stoyanova L, Tsokeva Z, Maria G, Krasimira N, et al. (2023) Appetite–regulating hormones in rats with fructose-induced metabolic changes. Pharmacia 70(1): 1-7.

32. Chetehouna S, Atoussi O, Boulaares I, Guemari IY, Derouiche S (2020) The effect of Chronic Tobacco smoking on Atherogenic index and Cardiovascular diseases risk in El-Oued (Algeria) men. Asian J Res Chem 13(6): 489-493.

33. Kuefner MS (2021) Secretory Phospholipase A2s in Insulin Resistance and Metabolism. Front Endocrinol (Lausanne) 12: 1-8.

34. Shang R, Rodrigues B (2021) Lipoprotein lipase and its delivery of fatty acids to the heart. Biomolecules 11(7): 1-11.

35. Hasimun P, Sulaeman A, Mulyani Y, Islami WN, Lubis FAT (2019) Antihyperlipidemic Activity and HMG CoA Reductase Inhibition of Ethanolic Extract of Zingiber Cassumunar Roxb in Fructose-Induced Hyperlipidemic Wistar Rats. J Pharm Sci Res 11(5): 1897-1901.

36. Kumar SR, Ramli ESM, Nasir NAA, Ismail NM, Fahami NAM (2021) Methanolic extract of piper sarmentosum attenuates obesity and hyperlipidemia in fructose-induced metabolic syndrome rats. Molecules 26(13): 1-14.

37. Ichigo Y, Takeshita A, Hibino M, Nakagawa T, Hayakawa T, et al. (2019) High-fructose diet-induced hypertriglyceridemia is associated with enhanced hepatic expression of ACAT2 in Rats. Physiol Res 68(6): 1021-1026.

38. Boulaares I, Derouiche S, Niemann J (2024) *Ocimum basilicum* L.: A Systematic Review on Pharmacological Actions and Molecular Docking Studies for Anticancer Properties. J Biochem Technol 15(1): 12-18.

39. Bachi A, Dalle DI, Scaloni A (2013) Redox Proteomics : Chemical Principles, Methodological Approaches and Biological / Biomedical Promises. Chem Rev 113(1): 596-698.

40. Chetehouna S, Derouiche S, Atoussi O (2024) Identification of New Alkaloids from Algerian Purslane by HPLC-QTOF-MS and Beneficial Effect of Purslane Enriched with Zinc on Experimental Alzheimer Disease in Rats. Curr Trends Biotech Pharm 18(1): 1595-1607.

41. Nair D, Nedungadi D, Mishra N, Nair BG, Nair SS (2021) Identification of carbonylated proteins from monocytic cells under diabetes-induced stress conditions. Biomed Chromatogr 35(6): e5065.

42. Mehta R, Sonavane M, Migaud ME, Gassman NR (2021) Exogenous exposure to dihydroxyacetone mimics high fructose induced oxidative stress and mitochondrial dysfunction. Environ Mol Mutagen 62(3): 185-202.

43. Shi YS, Li CB, Li XY, Jiao W, Yang L, et al. (2018) Fisetin Attenuates Metabolic Dysfunction in Mice Challenged with a High-Fructose Diet. J Agric Food Chem 66(31): 8291-8298.

44. Atoussi O, Sara C, Boulaares I, Guemari IY, Derouiche S (2021) Analysis of Blood Pressure, Lipid Profile and Hematological Biomarkers in men Addicted to Tobacco Chewing. Research Journal of Pharmacology and Pharmacodynamics 13(1): 1-4.

45. Wang G, Zhang Y, Zhang R, Jianlong P, Dengfei Q, et al. (2020) The protective effects of walnut green husk polysaccharide on liver injury, vascular endothelial dysfunction and disorder of gut microbiota in high fructose-induced mice. Int J Biol Macromol 162: 92-106.

46. Montesano A, Senesi P, Vacante F, Mollica G, Benedini S, et al. (2020) l-Carnitine counteracts in vitro fructose-induced hepatic steatosis through targeting oxidative stress markers. J Endocrinol Invest 43(4): 493-503.

47. Harrell CS, Zainaldin C, McFarlane D, Hyer MM, Stein D, et al. (2018) High-fructose diet during adolescent development increases neuroinflammation and depressive-like behavior without exacerbating outcomes after stroke. Brain Behav Immun 73: 340-351.

48. Schmidt NH, Svendsen P, Albarran JJ, Moestrup SK, Bentzon JF (2021) High-fructose feeding does not induce steatosis or non-alcoholic fatty liver disease in pigs. Sci Rep 11(1): 1-10.

49. Iskender H, Yenice G, Terim Kapakin KA, Eda D, Cigdem S, et al. (2022) Effects of high fructose diet on lipid metabolism and the hepatic NF-κB/ SIRT-1 pathway. Biotech Histochem 97(1): 30-38.