I. GENERAL PROJECT INFORMATION

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RESEARCH PROJECT TITLED

EFFECT OF A DIET LOW IN ADVANCED GLYCATION END PRODUCTS ON LIVER FUNCTION AND THE DEGREE OF STEATOSIS IN ADULTS WITH METABOLIC LIVER STEATOSIS

JUNE 17th, 2026.

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II. PROJECT

ABSTRACT

Background: Reduced insulin sensitivity and impaired glucose and lipid metabolism promote the formation of Advanced Glycation End Products (AGEs), which contribute to liver damage. AGEs exert their effect by binding to AGE receptors (RAGE). The AGE/RAGE interaction induces the activation of various intracellular pathways and subsequent cytokine synthesis. RAGE and sRAGE, which is a soluble receptor, are the most relevant AGE receptors in Metabolic Liver Steatosis (MASLD). The AGE-RAGE interaction contributes to fat accumulation in the liver and may be a key factor in the progression of steatosis to a more severe stage of MASLD.

Objective: To analyze the effects of a diet low in advanced glycation end products on liver function and the degree of steatosis in adults with MASLD.

Materials and Methods: A longitudinal, comparative clinical study will be conducted, including 48 participants with high AGE intake (>8,000 KU) and MASLD with steatosis of any degree. Participants will be divided into two groups and sequentially assigned to either the AGE-restricted diet group or the usual diet group for 12 weeks. Data on weight, height, waist, hip, and abdominal circumference, and blood pressure will be collected. Fasting blood samples will be taken to determine serum levels of glucose, insulin, triglycerides, total cholesterol, HDL, LDL, AGEs, glycated hemoglobin, and liver function tests. Statistical analysis: The data will be analyzed using descriptive statistics; qualitative variables will be reported as frequencies or percentages, and quantitative variables will be reported as means and standard deviations. The Kolmogorov-Smirnov (K-S) test with Yates' correction will be used to assess the normality of the numerical data. If the normality criterion is met, the data will be analyzed using a one-way ANOVA, crossing diet types (treatment) with serum levels of biochemical parameters for liver function and lipid profile. To determine specific differences in follow-up across different time points of data collection, a repeated-measures ANOVA will be used. If the normality criterion is not met, the chi-square test will be used. To assess the effect of diet type (treatment) on the type of metabolic hepatic steatosis (mild, moderate, and severe), the chi-square test will be used. All statistical tests will be performed using SPSS version 29, with a significance level of $p < 0.05$.

BACKGROUND

Advanced Glycation End Products

Advanced Glycation End Products (AGEs) are the result of the Maillard reaction and occur when reducing sugars react non-enzymatically, primarily with amino groups belonging to proteins, as well as with lipids and nucleic acids. AGEs are classified as endogenous and exogenous (1).

AGEs are part of physiological metabolism and normal aging, so they are generated even in the absence of hyperglycemia. Furthermore, their levels can increase under conditions of elevated oxidative stress (2).

AGEs are present in raw foods of animal origin, and cooking causes the formation of new AGEs in these foods. In particular, grilling, pan-frying, roasting, searing, and deep-frying promote and accelerate the formation of new AGEs. A wide variety of foods in the modern diet are subjected to cooking or heat treatment for reasons of safety and convenience, as well as to improve flavor, color, and appearance (3).

AGEs can also be derived from highly processed foods (heated to over 160°C) or dry-heated foods, which have a bioavailability of 10% when ingested. There are different types of AGEs, such as N-6-carboxymethyl-lysine (CML), pentosidine, N-6-carboxyethyl-lysine, (MGO)-lysine dimer, and glyoxal-lysine dimer, found in both serum and liver in humans and animals with Metabolic Liver Steatosis (MASLD) (4).

AGEs, by binding to their AGE receptors (RAGE), can promote oxidative stress, the inflammatory response, and endothelial dysfunction. However, other groups of cell-surface AGE receptors with functions opposite to RAGE, including AGE-R1, R2, and R3, and macrophage scavenger receptors are involved in AGE homeostasis (5).

RAGE is associated with intracellular signal transduction, and soluble RAGE (sRAGE) lacks the transmembrane and cytoplasmic domains, acting as a competitive inhibitor of AGE-RAGE binding (6).

Elevated AGEs activate RAGE, which induces the activation of nuclear factor kappa-B (NFκB) in activated B cells, inflammation, and nicotinamide adenine dinucleotide phosphate (NADPH) oxidase. This further increases oxidation and suppresses the protective survival systems AGE-R1 and silent information regulator of gene pairing type 2 homolog 1 (SIRT1). Elevated AGEs induce insulin resistance (IR) through the oxidative activation of protein kinase C (PKC), which phosphorylates the serine residues regulating IRS1 in insulin-sensitive tissues. Exogenous dietary AGEs (dAGEs) activate cellular stress responses and induce IR independently of obesity (7).

The biological significance of glycation has been studied with growing interest in recent years, largely because this mechanism has been linked to various diseases, such as diabetes, obesity, metabolic syndrome, and MASLD, among others (8).

Metabolic Liver Steatosis

It is a clinical-pathological condition characterized by the abnormal accumulation of lipids in hepatocytes in the absence of excessive alcohol consumption (9). The natural history of Metabolic Liver Steatosis (MASLD) ranges from simple steatosis, which is a slowly progressive disorder, to Non-Alcoholic Steatohepatitis (NASH), fibrosis, and even cirrhosis (10, 11). Most studies indicate that MASLD can develop very early in life, and there is a direct relationship with the degree of obesity (12).

The “double-hit” theory was proposed to explain the progression from simple steatosis to NASH and cirrhosis (13). The “first hit” is hepatic steatosis, and the second is hepatic inflammation (14). Hepatic steatosis is potentially reversible and does not necessarily lead to permanent liver damage. However, it is postulated that it sensitizes the liver to the second hit. Although less common, the second hit involves inflammation associated with oxidative stress or endogenous toxins from fructose metabolites (14).

Hepatic steatosis is closely linked to nutrition, including obesity, and possibly to the consumption of high-fructose corn syrup and certain types of fats (15).

MASLD and Insulin Resistance

IR is accompanied by elevated insulin levels which, in the presence of increased lipolysis and high dietary fat intake, promote hepatic triglyceride synthesis. For this reason, NHL has been associated with IR (16). Therefore, when insulin reaches the target tissue—in this case, the liver—the excess of free fatty acids causes resistance due to the excessive availability of substrate (17).

IR has been linked to the pathogenesis of NAFLD (18). The mechanism underlying this is still unknown (15). Experimental evidence suggests that both IR and MASLD are the result of a chronic inflammatory state (19).

MASLD is associated with peripheral and hepatic IR (20). The relationship between insulin sensitivity and MASLD appears to be, in part, driven by abdominal fat content (21).

AGEs and Insulin Resistance

The binding of AGEs to RAGE activates mitogen-activated protein kinases (MAPKs) and c-Jun N-terminal protein kinase (JNK). The JNK/MAPK pathway is strongly associated with insulin resistance. The critical role of the JNK1 isoform in the development of MASLD and its progression to EHNA has been established using animal models (22).

Litwinowicz et al. report that a low-AGE diet was associated with weight loss and a positive impact on markers of insulin resistance (22).

AGEs and MASLD

Reduced insulin sensitivity and impaired glucose and lipid metabolism promote the formation of AGEs, which contributes to liver damage. AGEs exert their

effects by binding to AGE receptors (RAGE). The AGE/RAGE interaction induces the activation of various intracellular pathways and the subsequent synthesis of cytokines. The increase in these biological processes leads to greater inflammation, reactive oxygen species (ROS), and insulin resistance (IR), as well as the production of AGEs and, in the case of liver fibrosis (23).

RAGE and sRAGE, which is a soluble receptor, are the most relevant AGE receptors in MASLD. The AGE-RAGE interaction contributes to fat accumulation in the liver and may be a key factor in the progression of steatosis to a more severe stage of MASLD (4).

Endogenous and exogenous AGEs are primarily expressed in Kupffer cells and hepatic stellate cells, where AGEs bind to RAGE (4, 24) and activate stress-activated protein kinase/c-Jun N-terminal kinase and the Janus kinase/signal transducer and activator of transcription pathways along with NFκB. NFκB induces the expression of cytokines and inflammatory factors such as transforming growth factor beta 1 (TGF-β1), monocyte chemotactic protein-1, tumor necrosis factor-α, and interleukins (4). Furthermore, NFκB activation generates ROS, which increases RAGE expression. In hepatic stellate cells, AGE compounds can trigger transdifferentiation into myofibroblasts, which also stimulates the formation of TGF-β1, monocyte chemotactic protein-1, and alpha-smooth muscle actin (α-SMA), among other compounds, promoting inflammation and liver fibrosis (4).

AGEs may be associated with MASLD due to their role in promoting oxidative stress, inflammation, and fibrosis (4).

On the other hand, the hypothesis regarding the involvement of AGEs in MASLD is based on the toxic AGE (TAGE) theory, which focuses on the role of AGEs derived from glyceraldehyde and acetaldehyde, which are produced in the liver through fructolysis, glycolysis, and alcoholysis (22).

Low-AGE Diet

There are short-term clinical trials in which the amount of dietary AGEs has been manipulated to study their effects in patients with chronic diseases (kidney disease, diabetes, or both) (25).

The concept of a low-AGE diet is based on the fact that foods with similar nutrient content can produce different amounts of AGEs depending on the cooking method (25). The highest production of CML occurs when food is roasted or fried (25).

AGE production can be limited by cooking at low temperatures, in the presence of moisture, and for short periods of time. Therefore, one should opt for cooking methods such as steaming, boiling, and simmering (25).

One strategy to reduce AGE formation is to marinate foods—especially those rich in protein and fat, such as meats—in acidic solutions like vinegar and citrus juices such as lemon or orange (25).

Additionally, the material used for cooking should be taken into account. Some metals, such as iron and copper, catalyze the formation of AGEs through the Fenton reaction. This reaction contributes to creating an oxidizing environment (through the formation of reactive oxygen species), which promotes the formation of carbonyl compounds, which in turn increase the production of AGEs (25).

Liver function tests

Patients with MASLD are initially diagnosed based on moderate elevations (1.5 to 4 times the normal range) in serum alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP), and gamma-glutamyl transferase (GGT). As the severity of the disease increases, there is a rise in AST, as well as in the AST/ALT ratio (26).

According to a report by Bernal-Reyes R et al., in a Mexican population with risk factors for developing MASLD, screening using liver function tests (AST, ALT, ALP, and GGT) is recommended as a valuable strategy for the early diagnosis of hepatic steatosis (27).

Use of Ultrasound in MASLD

For the differential diagnosis of MASLD, clinical features should be considered first, followed by noninvasive tests, and finally, liver biopsy (13).

The gold standard for diagnosing MASLD is liver histology (biopsy) (10, 28); however, due to its non-invasive nature (29, 30), low cost, safety, and high availability, ultrasound is the most commonly used imaging method (29); furthermore, ultrasound has demonstrated a sensitivity of 70% and a specificity of 100% compared to liver biopsy (31).

Khov et al. consider ultrasound to be a useful diagnostic tool for detecting MASLD. It is an accurate and reliable method for reducing the need for a liver biopsy in the appropriate clinical context (30).

Carrillo et al. mention that a 10% reduction in body weight will positively influence ultrasound findings of steatosis in patients with MASLD (32).

Parada et al. conducted a study in a Mexican population in which patients with some degree of hepatic steatosis were supplemented with omega-3 polyunsaturated fatty acids for 8 weeks. They found that the effect of supplementation was statistically significant in reducing the degree of hepatic steatosis (33).

A previous study by our group evaluated the effects of 8-week L-citrulline supplementation in obese adolescents and observed changes in hepatic steatosis (34). It is also important to note that the AGE-RAGE interaction contributes to fat accumulation in the liver and may be a key factor in the progression of steatosis to a more severe stage of MASLD (4), and dietary AGEs play a significant role in the metabolic pool of AGEs and in the AGE-RAGE interaction.

JUSTIFICATION

Poor dietary habits have been linked to excessive consumption of AGEs, high-fat foods, larger portion sizes, and excessive intake of simple sugars. These habits are considered a potential risk factor that may contribute to the development of dyslipidemia, MASLD, type 2 diabetes, insulin resistance, kidney damage, and hypertension.

The link between AGEs, liver function tests, and MASLD requires further investigation. While there is experimental evidence of the metabolically adverse effects of AGE intake, it has not been demonstrated that a low-AGE diet improves the health status of adults with MASLD. Given that MASLD has been linked to IR, this parameter will be used to examine the effect of a low-AGE diet. This would support a recommendation to improve dietary habits in adulthood and contribute to the prevention of liver damage, thereby reducing healthcare costs associated with metabolic diseases.

For these reasons, there is interest in studying this topic, and consequently, the following research question is posed.

STATEMENT OF THE PROBLEM

Could a low-AGE diet improve liver function and reduce the degree of steatosis in adults with Metabolic Liver Steatosis?

HYPOTHESIS

Ha. A low-AGE diet improves liver function and reduces the degree of steatosis in adults with Metabolic Liver Steatosis.

Ho. A low-AGE diet does not improve liver function and the degree of steatosis in adults with Metabolic Liver Steatosis

OBJECTIVES

GENERAL OBJECTIVE

To analyze the effects of a diet low in advanced glycation end products on liver function and the degree of steatosis in adults with Metabolic Liver Steatosis

SPECIFIC OBJECTIVES

1.- Identify adults aged 18 to 55 with Metabolic Liver Steatosis.

2.- Quantify dietary AGE intake in the study population.

3.- Compare the effects of a low-AGE diet and a usual diet on liver function and the degree of steatosis in the study population.

4.- Compare the effects of a low-AGE diet and a usual diet on lipid profile, glycated hemoglobin, and insulin resistance in the study population.

5.- To quantify plasma AGE levels in participants on an AGE-restricted diet and a habitual diet.

6.- To compare the effects of a low-AGE diet and a habitual diet on the degree of Metabolic Liver Steatosis using ultrasound.

METHODOLOGY (Materials and Methods)

STUDY DESIGN

A clinical, longitudinal, and comparative study will be conducted.

SAMPLE SIZE CALCULATION

The sample size was determined using the “Detect Disease” formula (random sampling and perfect diagnosis), with the statistical software Working in Epidemiology Win Epi. Formula:

$$n = \left(1 - (1 - NC)^{\frac{1}{d}}\right) \cdot \left(N - \frac{d-1}{2}\right)$$

n = required sample size.

N = population size.

d = expected number of affected individuals in the population.

NC = confidence level expressed as a proportion (=1-α)

Assuming a 95% confidence level and an expected minimum prevalence of metabolic hepatic steatosis of 14.4% (35), this results in n = 20 participants per group. Allowing for a 20% attrition rate, this yields n = 24 participants per group, for a final sample size of 48 participants.

The 48 participants will be divided into two groups, sequentially assigned to either the AGE-restricted diet group or the usual diet group (no intervention) for 12 weeks. Using snowball sampling, sequential assignment will be carried out by assigning a number in order of recruitment; participants recruited with an odd number will be assigned to the control group, and those recruited with an even number to the experimental group (36).

Participants will go to the mutual aid center at the Celaya–Salvatierra Campus of the University of Guanajuato, where they will be asked to sign an informed consent form, agreeing to participate in the study and to comply with the following:

Inclusion criteria:

1. Men and women aged 18 to 55.

2. High AGE intake > 8,000 KU.

3. MASLD of any severity.

4. Not currently undergoing nutritional treatment.

5. Sedentary lifestyle.

6.- No history of alcoholism.

7.- Not taking any medications.

8.- No comorbidities.

Exclusion criteria:

1.- Men and women currently undergoing treatment for metabolic diseases.

2.- Men and women with cardiovascular and/or coronary diseases.

3.- Pregnant women.

4.- Men and women undergoing hormone therapy.

5.- Men and women taking dietary supplements.

6.- Men and women who refuse to sign the informed consent form.

7.- Those in whom space-occupying masses (tumors) are detected during the ultrasound.

Elimination criteria:

1. Failure to adhere to the dietary regimen by at least 80%.

2. Become pregnant during the course of the study.

3. Develop any metabolic disease other than MASLD (such as diabetes mellitus, hypertension, or metabolic syndrome, among others) during the study.

4. Begin taking medication during the study.

ESTIMATION OF AGE CONSUMPTION AND ASSESSMENT OF NUTRITIONAL STATUS

A survey will be conducted among the population aged 18 to 55 to identify individuals with AGE intake exceeding 8,000 KU using 24-hour dietary recalls (two on weekdays and one on the weekend) in conjunction with the tables by Uribarri J et al. (3) to quantify dietary AGE intake. Once patients with high AGE and MASLD intake have been identified, they will be assigned to one of the two groups.

Nutritional status will be assessed based on anthropometric, dietary, and biochemical parameters.

Anthropometry

Anthropometric parameters will be assessed at baseline and at the end of the intervention at 12 weeks (see Annex 1). Weight, height, waist circumference, hip circumference, abdominal circumference, and blood pressure will be measured. Height, waist circumference, hip circumference, and abdominal circumference will be measured according to the technique described by Lohman et al. (37). The interpretation of waist, hip, and abdominal circumference will be based on the World Health Organization (WHO) (38), and measurements will be taken using a Seca® fiberglass measuring tape. For weight, a Seca® digital scale with 100-gram precision will be used, and for height, a Seca® height rod with 1-centimeter precision will be used. BMI will be calculated using the Quetelet formula. BMI interpretation will be based on WHO guidelines (38), and blood pressure will be measured using the technique described in NOM-030-SSA2-2009 (39).

AGE-Restricted Diet

To follow the AGE-restricted diet, only the method of food preparation will be specified, as reported by Uribarri et al. (3) and Luévano et al. (25).

To reduce AGE intake, follow these recommendations (25):

- Preferably consume vegetables raw, steamed, boiled, or stewed.

- Cook at low temperatures (low heat) and for short periods of time.

- Choose preparations that include water or moisture.

- Marinate meats with lemon or vinegar 1 hour before cooking; do not use soy sauce, barbecue sauce, or sweet sauces for marinating.

- Do not reheat protein-rich foods such as fish, meat, or chicken in the oven, toaster, or microwave. Reheat foods by steaming.

Diet adherence will be assessed using 24-hour food recalls over 3 days (two on weekdays and one on the weekend) (see Annex 2) during the 12-week period. Questionnaires will be collected every 15 days. Subsequently, macronutrient

quantification will be performed using The Food Processor® software and the tables by Uribarri et al. to determine AGE intake.

Based on the above, the percentage of adherence to the diet will be determined to be at least 80%.

The standardization of food records will follow the methods described by Luévano et al. (40) and Rodríguez et al. (41), both of whom conducted the intervention in a Mexican population.

Usual Diet

Participants assigned to this group will follow their usual diet, and to assess their adherence, 24-hour dietary recalls will be collected every 15 days for 3 days (two on weekdays and one on the weekend). Macronutrient quantification will be performed using The Food Processor® software and the tables by Uribarri et al. to determine AGE intake.

BLOOD SAMPLING

Venous blood samples will be collected from both groups after a 12-hour fast at baseline and at the end of the intervention at 12 weeks. Serum will be obtained from the blood samples and divided into aliquots; one aliquot will be stored at 2–8°C for the analysis of glucose, triglycerides (TG), cholesterol, HDL, and LDL within 2 hours, while another will be stored at -20°C for subsequent testing of insulin, glycated hemoglobin, ALT, AST, ALP, GGT, and AGEs (42, 43, 44).

EVALUATION OF BIOCHEMICAL PARAMETERS AND LIVER FUNCTION TESTS

Enzymatic spectrophotometric and ELISA assays will be used to measure serum levels of glucose, insulin, glycated hemoglobin, triglycerides, cholesterol, HDL, LDL, ALT, AST, ALP, GGT, and AGEs.

HOMA-IR will be calculated according to Mathew et al.: (Basal Insulin (μU/mL) × Basal Glucose (mg/dL)/405) (45).

Measurements of glucose, TG, total cholesterol, HDL, LDL, and glycated hemoglobin will be performed using enzymatic spectrophotometric assays. Liver function tests for ALT and AST will be performed by ELISA. Meanwhile, insulin will be determined by chemiluminescence using the ALPCO STELLUX Diabetes ELISA Kit, for subsequent calculation of HOMA-IR.

For the quantification of AGEs, the analytical biochemical ELISA technique using spectrophotometry will be employed. For this procedure, N(6)-

Carboxymethyllysine (CML) and the MBS2883946 competitive ELISA kit will be used, following the guidelines of the manufacturer MyBioSource.ing. In summary: The MBS2883946 ELISA kit is based on CML antibody-antigen interactions (immunoabsorption) and an HRP colorimetric detection system to detect CML antigenic targets in samples. The ELISA kit is designed to detect native, non-recombinant CML.

EVALUATION OF MASLD

Use of Ultrasound in MASLD

The ultrasound evaluation will be performed by [redacted] (in the event of a discrepancy in the ultrasound interpretation, a third opinion will be requested from [redacted] and will be performed after a 12-hour fast in the Department of Nursing and Obstetrics using a portable Honda MT 2000 unit with a linear transducer via the standardized technique. These will include a section at the level of the left hepatic lobe, a section at the level of the portal bifurcation, at the level of the hepatic veins, and within the hepatic lobe. Figure 1.

The ultrasound criteria evaluated will be:

a) Increased echogenicity compared to the kidney.

b) Uninvolved areas, defined as specific zones of the liver without fatty infiltration.

c) Visualization of the portal vessel wall and diaphragm.

d) Sound attenuation.

e) Hepatomegaly

Different degrees of fatty infiltration will be considered, as summarized in Table 1.

1.- Mild when increased echogenicity and hepatomegaly are observed.

2.- Moderate when sound attenuation is present.

3.- Severe when the portal vein wall and diaphragm are not visualized (44).

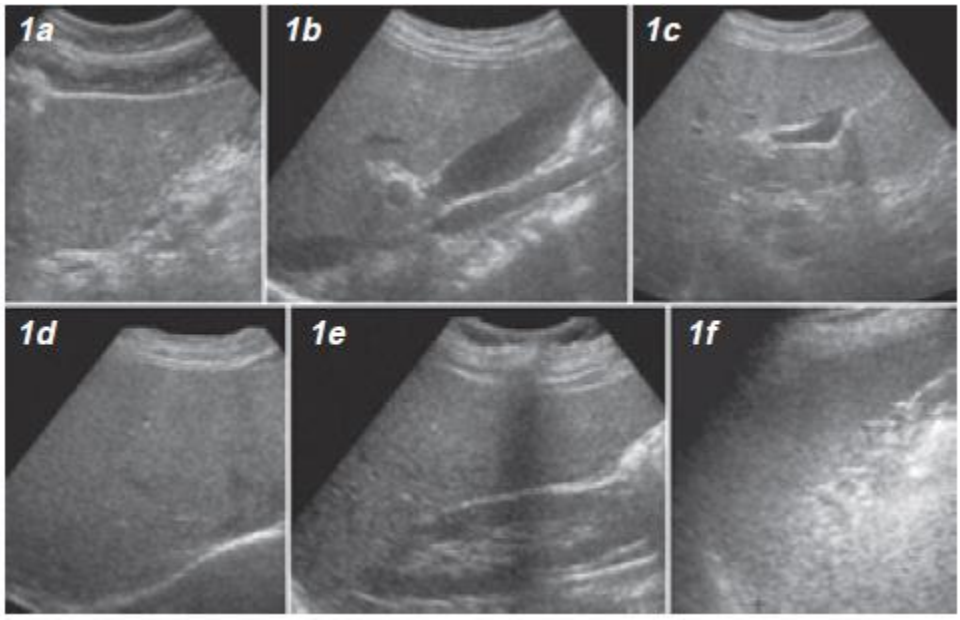

Figure 1. Assessment of Fatty Infiltration. Sections a through f used for the assessment of fatty infiltration. a: left hepatic lobe, b: gallbladder bed, c: portal bifurcation, d: hepatic veins, e: right lobe including the kidney, f: spleen (46).

Table 1. Ultrasonographic characteristics of MASLD

Ultrasonographic assessment of steatosis severity (46).

Grades	**Echogenicity (increase)**	**Sound (attenuation)**
Mild	Mild	No
Moderate	Moderate	Yes
Severe	Severe	Yes

ETHICAL CONSIDERATIONS

This study complies with the provisions of the Declaration of Helsinki (47). The confidentiality of participants' data will be protected in accordance with Article 16 of the REGULATIONS of the General Health Law (46). In addition, participants will be informed of the nature of the study, its benefits, and possible risks—which are minimal (Article 17, Category II, REGULATIONS of the General Health Law) (48)—

both verbally and in writing via an information letter (see Annex 3) and an informed consent letter (see Annex 4).

This research project was approved by the Research Committee of the Division of Health Sciences, under agreement CLDCS2025-O1-4.3.1. Furthermore, it will be submitted for review by the Research Ethics Committee of the University of Guanajuato (CEPIUG), and once approved by the CEPIUG, it will be registered on ClinicalTrials.gov.

STATISTICAL ANALYSIS

The data will be analyzed using descriptive statistics; qualitative variables will be reported as frequencies or percentages, and quantitative variables will be reported as means and standard deviations. The Kolmogorov-Smirnov (K-S) test with Yates' correction will be used to assess the normality of the numerical data. If the normality criterion is met, the data will be analyzed using a one-way ANOVA, crossing diet types (treatment) with serum levels of biochemical parameters for liver function and lipid profile. If normality is not met, the Kruskal-Wallis test will be used.

A circular flow diagram will be used to visualize and explore the flows of qualitative variables. This is a new method for visualizing and exploring data flow, which improves the ability to graphically assess patterns and trends, in this case, between the severity of fatty liver before and after treatments.

To determine specific differences in the follow-up of data collected at different time points, a repeated-measures ANOVA will be used. If the normality criterion is not met, the chi-square test will be used. To assess the effect of diet type (treatment) on the severity of metabolic hepatic steatosis (mild, moderate, and severe), the chi-square test will be used. All statistical analyses will be performed using SPSS version 29, with a significance level of $p < 0.05$.

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ANNEXES

ANNEX 1. VARIABLE DATA SUMMARY SHEET

Date: ____/____/____
month day year

NAME: _____

Baseline (initial)

Weight = _____

Height = _____

BMI = _____

Waist Circumference = _____

Hip Circumference = _____

Abdominal Circumference = _____

Waist-to-hip ratio = _____

Blood Pressure = _____

Date: ____/____/____
month day year

NAME: _____

Final (at 12 weeks)

Weight = _____

Height = _____

BMI = _____

Waist Circumference = _____

Hip Circumference = _____

Abdominal Circumference = _____

Waist-to-hip ratio = _____

Blood Pressure = _____

ANNEX 2. 24-HOUR REMINDER

Date: ____/____/____
month day year

NAME: _____

TIME	FOOD AND QUANTITY	FOOD PREPARATION METHOD
BREAKFAST		
MORNING SNACK		
LUNCH		
AFTERNOON SNACK		
DINNER		

UNIVERSIDAD DE GUANAJUATO

ANNEX 3. INFORMATION LETTER

UNIVERSITY OF GUANAJUATO
DEPARTMENT OF NURSING AND OBSTETRICS
DEPARTMENT OF MEDICAL SCIENCES
INFORMATION LETTER

Research study titled: “Effect of a Diet Low in Advanced Glycation End Products on Liver Function and the Degree of Steatosis in Adults with Metabolic Liver Steatosis,” with the objective of: Analyzing the effects of a diet low in advanced glycation end products on liver function and the degree of steatosis in adults with metabolic fatty liver disease.

You are cordially invited to participate in this study. Below, you will find information about what it entails.

We are a group of researchers working in the Department of Nursing and Obstetrics and the Department of Medical Sciences at the University of Guanajuato, and we are conducting the following research.

During the study, 12 ml of blood will be drawn using a syringe. The blood sample will be analyzed to determine the levels of glucose (sugar) and fats present. Additionally, you will undergo an ultrasound to assess the condition of your liver, be asked about your general health and dietary habits, and have your weight, height, waist, hip, and abdominal measurements taken, as well as your blood pressure. Finally, a nutritionist will prescribe a diet, which will only indicate how to prepare the food and will be followed for 12 weeks.

Diet adherence will be monitored using food questionnaires, which will be collected every 15 days over the 12-week period.

At 12 weeks, you will be scheduled for a follow-up to repeat the evaluations described above.

It is important to note that blood samples will be collected by [redacted] a surgeon with 26 years of experience.

Your participation in this study is entirely voluntary. You may choose to participate or not. Whether you choose to participate or not, all the services you receive at this institution will continue. If you choose to participate, you may change your mind later and withdraw from the study.

If you participate in this study, you will receive the following benefits: you will undergo a health checkup, including blood tests (your lab results will be provided to you), you will have the opportunity to make lifestyle changes, and you will improve your eating habits. On the other hand, the risk is minimal and consists of the possibility of a bruise appearing at the site of the blood draw; however, the researchers will monitor you closely and address any discomfort you may experience.

The blood samples, ultrasound scans, and data from this study will be used exclusively for the research described here and/or, if applicable, in subsequent studies.

Once the blood sample has been processed, you will decide whether or not to authorize its use in other future studies conducted by the same research group.

You will not incur any expenses related to your participation (e.g., meals, blood samples, ultrasound, etc.), nor will you receive financial compensation for participating in this study.

If you have any questions about the research, you may contact any of the following members of the research team:

[redacted]

Department of Nursing and Obstetrics

Email: [redacted]

[redacted]

Department of Medical Sciences

Email: [redacted]

ANNEX 4. INFORMED CONSENT LETTER

UNIVERSIDAD DE GUANAJUATO

UNIVERSITY OF GUANAJUATO
DEPARTMENT OF NURSING AND OBSTETRICS
DEPARTMENT OF MEDICAL SCIENCES
INFORMED CONSENT LETTER

I _____ I agree to participate in the research project titled: “Effect of a Diet Low in Advanced Glycation End Products on Liver Function and the Degree of Steatosis in Adults with Metabolic Liver Steatosis,” the objective of which is: To analyze the effects of a diet low in advanced glycation end products on liver function and the degree of steatosis in adults with metabolic fatty liver disease.

When sugar binds to proteins in the body, substances called Advanced Glycation End Products (AGEs) are formed. These harmful substances attach to the liver, causing fat to accumulate. Over time, this excessive accumulation of fat in the liver leads to a condition called fatty liver or Metabolic Liver Steatosis.

Poor eating habits can cause AGEs to accumulate in your body. This could be a risk factor for developing fatty liver disease, although scientists are still studying the exact relationship between AGEs and this condition.

To participate in this study, you must be a man and/or woman between the ages of 18 and 55, consume excessive amounts of AGEs, have been diagnosed with fatty liver via ultrasound, not be on a dietary regimen, be sedentary (not exercise), not consume alcohol or medications, and have no underlying medical conditions.

Participation in the project involves completing a medical history form and questionnaires that will include questions about my diet. In addition, my weight, height, waist, hip, and abdominal measurements will be taken, and my blood pressure will be checked. A 12-ml blood sample will be drawn, and I will undergo a liver ultrasound to assess whether I have fatty liver disease. All of the above will be performed twice (at the start and at 12 weeks).

The estimated time required for the procedure described above will be approximately 30 to 45 minutes.

As mentioned earlier, a blood sample will be taken to evaluate certain biochemical parameters. All evaluations will be performed by highly trained and experienced staff. We kindly ask you to authorize the blood draw, or if you do not agree, to decline the blood draw.

_____ I consent to having blood samples taken from me.

_____ I do not consent to having blood samples taken from me.

If you agree that your sample may be used in other projects within our research group, we kindly ask you to give your consent, or, if you prefer, to decline this option.

_____ I consent to this sample being used in future studies by the same research group.

_____ I do not consent to this sample being used in future studies by the same research group.

This project was submitted for approval to the Research Ethics Committee of the University of Guanajuato (CEPIUG). Participants may contact the Ethics Committee to address any questions regarding the project. If I have any questions about the ethical aspects or rights outlined in this protocol, I may contact:

[redacted]

Secretary of the Research Ethics Committee at the University of Guanajuato.

Email: cepiug@ugto.mx

If you have any questions about the research, please feel free to contact any of the following members of the research team:

[redacted]

Department of Nursing and Obstetrics

Email: [redacted]

[redacted]

Department of Medical Sciences

Email: [redacted]

I have been provided with all the necessary information about the research project and the reason for my participation. Furthermore, all my questions have been answered, and the potential risks and benefits of my participation have been explained to me. I have also been informed that the principal investigator is responsible for safeguarding my health and well-being in the event of any unexpected effects or side effects resulting from the intervention during the course of the research project. The principal investigator has assured me that I will not be identified in any presentations or publications resulting from this study, and that data related to my privacy will be handled confidentially.

I voluntarily consent to participate in this research and understand that I have the right to withdraw at any time, without this affecting me in any way.

Therefore, I hereby give my consent to participate in the project described above.

Participant's Name: _____
Participant's Signature: _____

Date of Acceptance: _____

Name and Signature of the Project Manager: _____

Name of Witness 1: _____
Signature of Witness 1: _____
Address of Witness 1: _____

Name of Witness 2: _____
Signature of Witness 2: _____
Address of Witness 2: _____