



OPEN ACCESS

EDITED BY

Hayriye Sebnem Harsa,
Izmir Institute of Technology, Türkiye

REVIEWED BY

Muhammad Rizwan Tariq,
University of the Punjab, Pakistan
Amr Adel Elkelish,
Suez Canal University, Egypt
Shayma Thyab Gddoa Al-Sahlanly,
University of Basrah, Iraq

*CORRESPONDENCE

Enrique Raya-Álvarez
✉ enriraya@ugr.es
Ehab Kotb Elmahallawy
✉ sa2elele@uco.es

RECEIVED 27 October 2023

ACCEPTED 10 June 2024

PUBLISHED 16 July 2024

CITATION

Moselhy SN, Al-Nashwi AA, Raya-Álvarez E, Abu Zaid FO, Shalaby HST, El-Khadragy MF, Shahein MR, Hafiz AA, Aljehani AA, Agil A and Elmahallawy EK (2024) Physicochemical, microbiological, and sensory properties of healthy juices containing aloe vera gel and probiotics and their antidiabetic effects on albino rats. *Front. Nutr.* 11:1328548. doi: 10.3389/fnut.2024.1328548

COPYRIGHT

© 2024 Moselhy, Al-Nashwi, Raya-Álvarez, Abu Zaid, Shalaby, El-Khadragy, Shahein, Hafiz, Aljehani, Agil and Elmahallawy. This is an open-access article distributed under the terms of the [Creative Commons Attribution License \(CC BY\)](https://creativecommons.org/licenses/by/4.0/). The use, distribution or reproduction in other forums is permitted, provided the original author(s) and the copyright owner(s) are credited and that the original publication in this journal is cited, in accordance with accepted academic practice. No use, distribution or reproduction is permitted which does not comply with these terms.

Physicochemical, microbiological, and sensory properties of healthy juices containing aloe vera gel and probiotics and their antidiabetic effects on albino rats

Sara Naiim Moselhy¹, Ahmed Aladdin Al-Nashwi¹, Enrique Raya-Álvarez^{2*}, Fouad Omar Abu Zaid³, Hanan Said Tawfik Shalaby¹, Manal F. El-Khadragy⁴, Magdy Ramadan Shahein⁵, Amin A. Hafiz⁶, Abeer A. Aljehani⁷, Ahmad Agil⁸ and Ehab Kotb Elmahallawy^{9,10*}

¹Food Science Department, Faculty of Agriculture, Zagazig University, Zagazig, Egypt, ²Rheumatology Department, Hospital Universitario San Cecilio, Granada, Spain, ³Agri- Industrialization Unit, Plant Production Department, Desert Research Center, Cairo, Egypt, ⁴Department of Biology, College of Science, Princess Nourah Bint Abdulrahman University, Riyadh, Saudi Arabia, ⁵Department of Food Science and Technology, Faculty of Agriculture, Tanta University, Tanta, Egypt, ⁶Department of Clinical Nutrition, Faculty of Applied Medical Sciences, Umm Al-Qura University, Makkah, Saudi Arabia, ⁷Department of Food and Nutrition, Faculty of Human Sciences and Design, King Abdulaziz University, Jeddah, Saudi Arabia, ⁸Department of Pharmacology, Biohealth Institute Granada (IBs Granada) and Neuroscience Institute, School of Medicine, University of Granada, Granada, Spain, ⁹Departamento de Sanidad Animal, Grupo de Investigación en Sanidad Animal y Zoonosis, Facultad de Veterinaria, Universidad de Córdoba, Córdoba, Spain, ¹⁰Department of Zoonoses, Faculty of Veterinary Medicine, Sohag University, Sohag, Egypt

The consumption of fruit and vegetable juices is widely recognized as a healthy choice across all age groups. Orange, carrot, and aloe vera are renowned for their functional properties and health benefits. In this study, we investigated the potential incorporation of aloe vera gel into blended orange and carrot juices. We also evaluated the resulting mixed probiotic juices (chemical, microbiological, and sensory aspects) during a 14-day storage period at refrigerator temperature. The chemical composition and phytochemical structure of aloe vera gel were examined, followed by an assessment of the biological effects of these healthy juices on diabetic albino rats. The results indicated improvements in total soluble solids, reducing sugars, and total sugars with increasing storage duration. Furthermore, the study demonstrated that incorporating aloe vera into the natural mixed juices enhanced their phytochemical quality. The treatment supplemented with aloe vera gel gave the highest total content of phenolic and flavonoid substances, which were 310 mg of GAE/100 g and 175 mg of quercetin/100 g, respectively. Probiotic strains (*Bifidobacterium animalis* subsp *lactis* Bb12, *Lactiplantibacillus plantarum* 299V, and *Lactobacillus acidophilus* L10) exhibited good viable cell counts in orange and mixed orange and carrot probiotics juices with viable counts of 7.42–8.07 log CFU/mL. Regarding sensory attributes, the study found that increasing the ratio of orange juice improved the taste while increasing the ratio of carrot juice enhanced the color in juice mixtures. Incorporation of aloe vera into mixed natural juices also enhanced the reduction of blood glucose, triglyceride, cholesterol, LDL, creatinine, ALT, AST, and urea levels while increasing total protein and HDL

levels in diabetic rats. Based on these findings, oranges, carrots, and aloe vera offer the potential to produce new, flavorful, nutritious, and appealing juices. Moreover, this study determined that a functional juice with favorable sensory properties can be created by blending 75% orange juice, 20% carrot juice, and 5% aloe vera gel. Additionally, aloe vera demonstrated greater efficacy as an antidiabetic agent in rats. Further research is suggested to explore the potential advantages of aloe vera gel and probiotic juices in mitigating diabetes and other metabolic syndromes.

KEYWORDS

juice, probiotic, diabetes, functional properties, sensory attributes

1 Introduction

Food components and functional foods play a crucial role in improving various health conditions beyond their nutritional value (1–12). This dietary choice encompasses various nutritional supplements, whole grains, vegetables, fruits, and beverages, all selected by individuals to enhance their health. Among others, beverages that reduce blood glucose by mixing fruit juices with low-sugar elements (vegetables) can be very beneficial through special biological and physical properties (13). Consuming vegetables and fruits is a substantial portion of a healthy diet, amongst other various food elements, and they are a superb provenance of bioactive ingredients (14). Orange and carrot juice can supply vitamins, polyphenols, and carotenoids, making them effective in preventing diabetes, heart disease, and cancer. Orange juice is made by squeezing fresh oranges and then undergoing a dehydration process. Oranges (*Citrus sinensis*) are rich in zinc, pectin, iron, manganese, chlorine, potassium, phosphorus, folic acid, and sodium (15). They have an enormous content of dietary fiber and are considered an excellent origin of bioactive ingredients such as carotenoids and phenolics (16–18). Oranges also have antioxidants, vitamins, and minerals. Vitamin C is necessary to protect the body from free radicals within cells (16, 19). More importantly, oranges are available in quantities throughout the year at an affordable price (20), and a glass of orange juice contains 112 g calories (21, 22). Carrots (*Daucus carota*) are rich in antioxidants (phenolic, flavonoids) and β -carotene (23, 24). They contain vitamin A, which is necessary for eye health and vision and reduces the risk of macular degeneration and cataracts that cause vision loss (25). Carrot juice is a ratable origin of carotenoids, vitamins, and minerals (21). Juice mixture is deemed a method that alleviates the nutritional quality of the juice product and also sensory characteristics (21). Prior research (26) has found that blending carrot juice with various fruits and vegetables improves overall nutritional characteristics and acceptability. Blending two or more fruits to create beverages offers a promising approach for developing new products with beneficial effects on sensory attributes, nutrition, and health (23).

Aloe vera (*Aloe barbadensis* Miller) is a perennial succulent shrub belonging to the family of *Asphodelaceae* (*Liliaceae*) (27–29). It is grown and cultivated in dry regions across the globe. Multiple studies have indicated that aloe vera can be an alternative to certain medications because it contains bioactive

ingredients such as anthraquinones, anthrones, alkaloids, and others. These components contribute to its potent antidiabetic and anti-inflammatory properties (30, 31). Furthermore, aloe vera is an effective agent in combating most diseases (32) and can decrease blood glucose in diabetic patients (33). There are many vitamins, minerals, and antioxidants in aloe vera gel (34), which is used to enhance the nutritional quality of some juices and can be consumed because of its beneficial influence on health (35). Developing blended juices presents a viable option for enhancing the taste of aloe vera gel. Blending aloe vera gel with orange juice aims to enhance its physicochemical, microbial, and organoleptic properties (36). A previous study (37) evidenced that aloe vera gel has medicinal effects and contains bioactive polysaccharides, including mannose and glucose, known as “glucomannans.” Additionally, aloe vera juice contains significant amounts of xylose, arabinose, galactose, rhamnose, and numerous vitamins such as C, E, A, B1, B2, and niacin. However, aloe vera juice has low levels of magnesium, calcium, selenium, sodium, zinc, potassium, and manganese. In previous literature (38), fruit juices supplemented with probiotics have been considered functional drinks fortified with calcium and vitamins. Another study (39) revealed that fruit beverages might be a suitable medium for adding probiotics. Probiotics are defined as microorganisms that have a beneficial effect on health when taken in convenient doses (40).

The incorporation of functional foods to promote microbial balance and gut health has emerged as an innovative approach to reducing the risk of chronic illnesses. Extensive evidence suggests that supplementing food with probiotics can enhance health by modulating gut bacteria. Although numerous bacterial species are recognized as probiotics, *Lactobacillus* and *Bifidobacterium* spp. are the most extensively studied and have been incorporated into various food matrices to create a diverse range of functional products (41, 42). In this regard, *Lactiplantibacillus plantarum* (*L. plantarum*) is a microorganism commonly used in food fermentation technology and is generally considered safe. It also has applications in producing probiotic foods, including the commercially available *L. plantarum* 299v strain. This facultative heterofermentative lactic acid bacterium (LAB) exhibits remarkable resilience to conditions typically fatal to LAB, such as high acidity and ethanol concentrations. Notably, *L. plantarum* possesses unique characteristics, including its adaptability to various fermentation processes and metabolic versatility. Its ability to thrive in diverse environments is likely due to its relatively large

genome size, averaging 3.3 Mb, which is one of the largest among the *Lactobacillus* genera. It should be noted that *L. plantarum* has been isolated from numerous food sources, including cereals, meats, dairy products, vegetables, fruits, beverages, and human and mammalian niches (43, 44).

Probiotics, also known as “good” bacteria, include *Bifidobacterium animalis* subsp. *lactis* (*B. lactis*), which produces lactic and acetic acids. These beneficial bacteria, such as *B. lactis*, play a role in food digestion and nutrient absorption, and defense against harmful organisms that can cause illness. *B. lactis*, a subspecies of *B. animalis*, is commonly found in probiotic supplements and the human gut. It is utilized to address various health issues such as respiratory tract infections, constipation, irritable bowel syndrome (IBS), and colic in newborns. Additionally, it is used to manage conditions such as diarrhea, hay fever, dental cavities, and many others, although scientific evidence supporting its efficacy in some cases is limited. For instance, it is worth noting that there is insufficient data to support the use of *B. lactis* for treating COVID-19.

Sometimes, *Bifidobacterium animalis* subsp. *lactis* is labeled as *Bifidobacterium lactis* or *B. lactis* on product packaging. It is crucial to differentiate *B. lactis* from other probiotics and fermented food products, such as yogurt, kefir, and fermented milk, as they are not interchangeable (45, 46). First identified in 1900, *Lactobacillus acidophilus* (*L. acidophilus*) is a rod-shaped, homofermentative, Gram-positive anaerobic bacterium commonly found in the human body, notably in the mouth, vagina, and digestive system, as well as in various fermented foods such as yogurt and milk. *L. acidophilus* thrives best in acidic environments with pH levels below 5.0, and its optimal growth temperature is 37°C. Certain commercial strains of *L. acidophilus* are utilized in dairy production due to their potent probiotic properties.

The genome sequencing of *L. acidophilus* has been completed, and it is known to inhibit the growth of harmful bacteria through antagonistic mechanisms (47, 48). Numerous previous studies have indicated that probiotics serve as a dietary supplement to support intestinal health and prevent gastrointestinal infections (49–51). Several previous works have investigated the impact of incorporating fermented plants or juices fermented by probiotic bacteria. In this context, a previous study (52) found that adding date juice to bio-yogurt made with a probiotic starter (*Bifidobacterium longum*, *L. acidophilus*, and *Streptococcus thermophilus*) affected its microbiological and physicochemical properties, with the viability of probiotic bacteria declining over a 21-day period; however, the bio-yogurt containing 10% date juice retained a higher count of probiotic bacteria compared to other samples. Another study (53) assessed the bacterial viability of different types of fermented onions utilizing probiotic starters (*Lactobacillus acidophilus* (LA-5), *Bifidobacterium bifidum* (BB-12), and *Streptococcus thermophilus*) after a 24-h fermentation at 37°C and subsequent 28-day refrigeration. The findings revealed microorganism viability levels at Log 7.79 and 7.57 CFU/g.

Further, the literature also suggests that providing probiotic-fortified juice to individuals with lactose intolerance who cannot consume dairy products would be highly beneficial and practical. However, there is currently limited information available regarding the effects of fortifying healthy juices with aloe vera gel and probiotics. This includes understanding their impact on various

physicochemical and organoleptic properties and their potential antidiabetic effects on albino rats. Therefore, this study aimed to blend aloe vera gel with orange and carrot juices to enhance their functional, physicochemical, and organoleptic properties while also investigating its potential antidiabetic effects on albino rats.

2 Materials and methods

2.1 Plant sources and phytochemicals composition of aloe vera gel

Fresh aloe vera leaves (*Aloe barbadensis* Miller) between 50 and 70 cm in length were acquired from the desert of Matrouh governorate, Egypt. The area is located 240 km (150 mi) west of Alexandria and 222 km from Sallum, along the main highway that connects the Nile Delta to the Libyan border. Oranges (*Citrus sinensis*), especially an assortment of baladi oranges at a full maturity phase, and carrots (*Daucus carota* L) were brought from the local supermarket on the tenth day of Ramadan, in Egypt. The chemical and phytochemical composition of aloe vera gel was determined as reported elsewhere (54).

2.2 Microorganisms and their maintenance

The Phytone-Yeast Trypticase (TPY) medium contains (per liter) 10 g of trypticase (BBL), 5 g of phytone (BBL), 5 g of glucose, 2.5 g of yeast extract (Difco), 1 ml of Tween 80, 0.5 g of L-cysteine HCl, 2 g of K₂HPO₄, 0.5 g of MgCl₂ 6H₂O, 0.25 g of ZnSO₄ 7H₂O, and 0.15 g of CaCl₂. The TPY medium was combined with agar-agar at a concentration of 15 g/l to create TPY agar. The medium had a pH level of about 6.0. In 1 L of Beeren's agaridium, there were 44 g of Columbia agar (Oxoid CM331), 5 g of glucose, 0.5 g of L-cysteine HCl, 5 g of agar-agar, and 5 mL of propionic acid. Propionic acid was added following sterilization of the medium, and 1N NaOH was used to bring the pH to 5.0. De Man–Rogosa–Sharpe (MRS) agar, which was prepared based on a recipe described elsewhere (55). The probiotic strains (*Bifidobacterium animalis* subsp. *lactis* Bb12, *Lactiplantibacillus plantarum* 299V, and *Lactobacillus acidophilus* L10) were provided by Chr. Hansen A/S (Hørsholm, Denmark).

2.3 Determination of probiotic viability during storage

The vitality of the probiotic *L. plantarum* in various juice samples was assessed using spread plating and serial dilution methods. Viable cell counts were performed during 2 weeks of chilled storage on days 1, 7, and 14. Juice samples were serially diluted up to 10⁻⁷ using 0.1% (w/v) sterile peptone water, and 100 μL of an appropriate dilution was spread in triplicate on MRS agar plates. Viable counts were determined after 48 h of anaerobic incubation at 37°C. In contrast, *B. lactis* and *L. acidophilus* required anaerobic conditions for growth, and this environment was created using the Bugbox anaerobic chamber (Ruskin Technology, USA). *B. lactis* was cultured on TPY agar, while *L. acidophilus* was cultured

on MRS-sorbitol. Both were incubated at 37°C for 48–96 h (56). The colony-forming units were counted during fermentation using the plate counting method (57). Results for probiotic counts in juice samples were expressed as log CFU per mL.

2.4 Preparation of natural juices and phytochemicals composition of aloe vera gel

Oranges and carrots were chosen and washed carefully in tap water. Then, the fruits were cut into slices, and the juice was extracted by hand reamer (vegetable juicer). The extracted juice was then filtered directly and used for blending. All juices were pasteurized at 85°C for 10 min, and subsequently, pasteurized juices were warm-filled in sterilized and cleaned glass bottles (125 ml). The fruit juice was stored in anaerobic conditions at 37°C using an Anaerobe Jar + GasPak System (OXOID) or a Bugbox anaerobic chamber after being injected with *Bifidobacterium* or *Lactobacillus* bacteria. All samples were incubated with a 24-h-old probiotic culture (>105 CFU/mL) at 30°C for 72 h. *Bifidobacteria* and *Lactobacilli* colony-forming unit (cfu) counts were performed on samples at regular intervals. Additionally, the pH was determined. The juice had 1% of the fructo-oligosaccharide Raftiline added as a prebiotic during 0, 16, and 24 h of fermentation and preserved for 21 d at refrigerator temperature. Aloe vera gel was obtained by cutting leaves perpendicularly and mingled in a juice mixer to make it homogenized and smooth. Then, it was filtered through muslin cloth and stored at refrigerator temperature until use.

2.5 Preparation of juice treatments and its blends

In this study, six treatments were made of orange juice, carrot juice, and their mixtures, and the treatments were as follows:

- T1: 100% nature orange juice
- T2: 75% orange juice with 25% carrot juice
- T3: 95% orange juice with 5% aloe vera gel
- T4: 75% orange juice, 20% carrot juice and 5% aloe vera gel
- T5: 100% orange juice and probiotics
- T6: 75% orange juice, 25% carrot juice and probiotics.

All juice treatments were packaged in 200 ml sterile glass bottles and stored at refrigerator temperature until use.

2.6 Method of analysis

All juice blends were estimated chemically and microbiologically at zero time, then after 7 d of storage at refrigerator temperature. Sensory evaluation was carried out only at zero time. Changes in total and reducing sugars were evaluated (58). A total of 278 µL of diluted samples were put into 2 mL Eppendorf tubes. Following an extraction solvent,

278 µL of 5% aqueous phenol solution was added to the tubes. Thousand microliter of sulfuric acid was carefully added to each tube after a brief vortex agitation, adhering to all chemical safety requirements and utilizing the appropriate protective practices. Then, using a Power Wave XS 201595 spectrophotometer (BioTek Inc., Winooski, VT, USA) outfitted with a plater reader (Biotek KcJunior), absorbance was recorded at 480 nm after 30 min. The calibration curve was made using galacturonic acid (GalA) (G212598%; Sigma-Aldrich, St. Louis, MO, USA). The data were given in units of g kg⁻¹DM. The 3,5-dinitrosalicylic acid technique was used to determine the amount of reducing sugar. GalA (0–5 mg mL⁻¹) in methanol was used to create the calibration curve. At 540, absorbance was measured. The value of non-reducing sugars was calculated by deducting the reducing sugars from the total sugars. The changes in the brix value of all total soluble solids (TSS) in the juices were recorded by a digital refractometer (DR 6000, A. Kruss Optronic GmbH, Hamburg, and Germany). The changes in the pH value were measured by the glass electrode of a digital pH meter (Model Mettler Toledo, Switzerland), as described elsewhere (58). The color was measured by using the Hunterlab Colorflex (HunterLab, Reston, Va., US), as reported by Stinco et al. (59).

Total polyphenol content (TPC) was specified using the Folin-Ciocalteu system at 765 nm (UV-Vis spectrophotometer, Jenway, Staffordshire, UK), as illustrated by Gao et al. (60). The total phenol content was used as gallic acid equivalent in mg/L. In a test tube, 1.5 mL of Folin-Ciocalteu reagent and 0.2 mL of the juice were combined. 1.5 mL of a 6% sodium carbonate solution was added to the mixture after it had been incubated at room temperature for 5 min. The mixture was again incubated at room temperature for 90 min. A quartz cuvette was used to measure the blue color's absorption at 725 nm. The following gallic acid standards were created: 0.75 mg of gallic acid was dissolved in 5 mL of distilled water. Different amounts (0, 0.1, 0.2, and 0.3) of the gallic acid standard were pipetted into four marked test tubes. To finish the quantities to 0.3 mL, water was added. Later, 2.25 mL of Folin-Ciocalteu reagent was added, and the mixture was allowed to sit for 5 min at room temperature. The mixture was then re-incubated for 90 min at room temperature with 2.25 mL of a 6% sodium carbonate solution. The total phenolic content of the samples was calculated and expressed as milligrams of gallic acid equivalents (GAE) per 100 mL of sample. A standard curve was plotted using the absorbance of the resulting blue color, which was read using a quartz cuvette at 725 nm. The flavonoid content was determined spectrophotometrically, as mentioned elsewhere (61), which was then read at 510 nm. The value of total flavonoid was accurately determined in triplicate and expressed as quercetin equivalent in mg/L. AlCl₃ test was used to assess the flavonoid content. In this step, a total of 300 µL of 5% NaNO₂ was added to an aliquot of 500 µL of sample. Six hundred microliter of 10% after 5 min. After another 5 min, AlCl₃ was added, and then 2 mL of 1 M NaOH was added to the mixture. Using a UV-Vis Agilent 8453 spectrophotometer (Agilent Technologies, Italy), the absorbance was measured at = 510 nm. The data were given as mg/100 mL of quercetin equivalents.

The sensory evaluation of natural mixed juices was assessed by 13 members of the staff of the Agri-Industrialization Unit, Desert Research Center (Cairo), and 15 members of the staff of the Food Science Department, Faculty of Agriculture, Zagazig University. A

team of panelists was asked to estimate color, taste, odor, textures, and overall acceptability using a 10-point scale as described (62). The panelists were instructed to wash their mouths with low-sodium spring water during the sensory evaluation session, and they were encouraged to write down any criticisms of the tested products. Plain and treated juices were presented in plastic cups coded with three-digit random codes. Each cup contained 100 mL of juice samples freshly removed from the refrigerator. The sensory evaluation was conducted using a comparative test with fresh juices as a reference sample. The data were collected in specially designed ballots.

2.7 Biological experimental design

This study was conducted with the approval of the Institutional Animal Care and Research Unit, Zagazig University, Egypt (Approval no. ZU-IACUC/2/F/339/2022). A total of 30 male albino rats, each weighing 130–135 g, were purchased from the Agricultural Research Center of Giza, Giza, Egypt, and housed in wire cages in a 25°C environment. The rats were housed in solo stainless-steel cages in healthy laboratory conditions in the biology laboratory of the Faculty of Agriculture, Zagazig University. Rats were fed on a basal diet for 7 days (adaptation interval). The specially prepared (63) basal diet contained 12.5% casein, 10% corn oil, 4% salt mixture, 5% fiber, 1% vitamin mixture, 0.3% DL-methionine, 0.2% choline chloride, and completed to 100% by adding corn starch. Animals also had access to water throughout the duration of the experiment. The animals were divided into five groups. After the adaptation duration (7 days), the first group continued feeding on the basal diet and was considered a negative control (G1). The other four groups were diabetic rats and fed on a basal diet. The rats in the second group (G2) were fed a basal diet and considered a positive control. All rats except the control (negative and positive) received juices percent 100 mg/Kg body weight (64). The three other groups were allowed to be fed on juices as follows: G3: diabetic rats fed on 75% fresh orange juice and 25% fresh carrot juice; G4: diabetic rats fed on 75% orange juice, 20% carrot juice, and 5% aloe vera gel; and G5: diabetic rats fed on probiotic juice (75% orange juice, 25% carrot juice). The original treatments were a 75% orange juice mixture + a 25% carrot juice mixture, as it was the best treatment in terms of sensory acceptance by the arbitrators. The other two treatments were chosen to study the effect of supplementing the aforementioned original treatments with 5% aloe vera, compared to supplementation with probiotic bacteria with a known therapeutic effect. All groups were left throughout the duration of the experiment (45 days), and the amount of diet consummation was registered every day to compute the food intake, while body weight was recorded every week. The food efficiency ratio (FER) was calculated using the equation $FER = \text{body weight gain/food consumed "45 days"}$ (65).

2.8 Induction of diabetes

To induce diabetes, male albino rats received a single intraperitoneal injection of alloxan monohydrate at a dose of 150

TABLE 1 Chemical and phytochemical composition of aloe vera gel.

Chemical constituents (g/100 g)	
Moisture	96.6 ± 2.4
Fat	0.05 ± 0.01
Protein	0.16 ± 0.03
Crude fiber	0.20 ± 0.02
Ash	0.26 ± 0.05
Phytochemicals	
Total phenol content (mg of GAE/100 g)	12.40 ± 0.74
Total flavonoid content (mg of quercetin/100 g)	98.80 ± 1.7

mg/kg body weight, following the described protocol (66). Rats were allowed 5% glucose solution to overcome severe hypoglycemia (67). Fasting blood glucose levels in rats were measured after 2 days. Rats were inspected for diabetes through glycosuria with a blood glucose level of 200–310 mg/dL 3 d following experimental induction. Blood samples were collected from the tip of the tail after 3 and 6 weeks of feeding and analyzed using the enzymatic kit method (68).

2.9 Biochemical analysis of blood samples

At the conclusion of the study period (45 days), rats underwent overnight fasting and were euthanized under complete anesthesia through intraperitoneal injection of ketamine (90 mg/kg) and xylazine (5 mg/kg). Following the careful separation of the abdominal skin from the thoracic cavity, blood was drawn from the posterior vena cava and placed into a serum separator tube. The sera were obtained by centrifuging the collected blood at 3,000 rpm for 10 min. Subsequently, the serum samples were stored at –20°C until further analysis. Estimation of Triglycerides, LDL, and total cholesterol was performed (68–71). Glucose was measured (72), ALT and AST were determined as mentioned by Reitman and Frankel (73), and the total protein was determined using the methods described earlier (74). Uric acid and creatinine were measured using the methods mentioned elsewhere (75)¹.

2.10 Statistical examination

A statistical analysis was chosen to determine the impact of the treatment. All evaluations were approved in triplicate, and the data was described as mean. Significant differences ($p < 0.05$) were designed using Duncan multiple range tests (76). The mean values and their standard errors (SEM) are presented, and GraphPad Prism V5.0 software (GraphPad, San Diego, CA, USA) was employed for data visualization. A significance threshold of $p < 0.05$ was used for all statistical analyses.

¹ Standard A, Sample A. Creatinine. Jaffé. Colorimetric–Kinetic. Ref.: Jaff-005.

3 Results and discussion

3.1 Chemical and phytochemicals composition of aloe vera gel

According to the present study, the amounts of moisture, ash, fat, protein, and crude fiber were 96.6, 0.26, 0.05, 0.16, and 0.20 (g/100 g), shown in Table 1. These values are within the scope of the research described elsewhere (54). Aloe vera gel contains 12.40 mg of GAE/100 mL of phenol and 98.80 mg of quercetin/100 mL of flavonoid. A previous study (77) estimated the phenol and flavonoid concentration to be 9.71 and 100.87 mg/lit extract.

3.2 Effect of aloe vera and probiotic on reducing, non-reducing, and total sugar contents of juices during storage interval

Table 2 depicts the effect of fortification of aloe vera and probiotic juices during the storage interval on the total sugar contents of the final product. It was noticed that the contents of non-reducing sugars decreased in all treatments after 7 days of storage, and then it increased again after 15 days of storage. On the contrary, the reducing sugar content increased for all treatments after 7 days of storage, and then it decreased again after 15 days of storage. The total sugar content increased during the storage periods in all treatments. It was also noted that the content of reducing, non-reducing, and total sugars decreased in the treatments containing probiotics compared to the rest of the treatments during all storage periods because of the activity of probiotic bacteria and their metabolites. Different lactic acid bacteria strains displayed their distinctive sugar metabolizing features and common traits for using sugars. Similar findings were noticed by Kelebek and Selli (78), who revealed that LAB increased the amount of glucose by producing a significant amount of lactic acid in the juice, creating a low pH environment, and accelerating the rate at which sucrose is hydrolyzed into glucose and fructose. Meanwhile, a previous study (79) observed that cell strain, fermentation substrate, and other parameters were strongly correlated with the consumption of sugar by microorganisms. These outcomes provided additional confirmation of the soluble sugars' metabolic properties during the digestion of the LAB. It appears that the diminution in non-reducing sugar is accompanied by an increase in reducing sugar in juices (80, 81). Results in Table 2 revealed an obvious increase in the reducing sugars of all the mixed juices due to the long storage interval, and this rise reached its extreme values at the end of the storage period at room temperature for the storage interval (81). It has been suggested that this increase was attributed to the change of sucrose in reducing sugars (glucose and fructose), which might be caused by the acidity of the juice; the longer storage interval, and the high temperature during the storage interval. In our study, there was an increase in the orange ratio, which led to an increase in reducing sugar content for all studied treatments, consistent with some previous works (82). Concerning the total sugars, the previously presented data illustrated approximately the same observation of reducing sugars. However, increasing the storage phase led to a marked rise in the

TABLE 2 Effect of aloe vera and probiotics on the total sugar content of juices during storage interval.

Treatments	Non-reducing sugars						Reducing sugars			Total sugars		
	Storage interval			Storage interval			Storage interval			Storage interval		
	First day	7 d	14 d	First day	7 d	14 d	First day	7 d	14 d	First day	7 d	14 d
Control juices	T1 2.15 ± 0.01 ^c	1.33 ± 0.01 ^e	2.14 ± 0.01 ^b	5.7 ± 0.01 ^c	6.72 ± 0.01 ^b	6.32 ± 0.01 ^d	7.85 ± 0.01 ^b	8.04 ± 0.01 ^f	8.44 ± 0.01 ^a	7.85 ± 0.01 ^b	8.04 ± 0.01 ^f	8.44 ± 0.01 ^a
Aloe vera juices	T2 2.04 ± 0.01 ^d	2.3 ± 0.01 ^a	2.23 ± 0.01 ^a	5.3 ± 0.06 ^d	5.43 ± 0.01 ^f	6.12 ± 0.01 ^e	7.24 ± 0.01 ^f	7.72 ± 0.01 ^f	8.34 ± 0.01 ^c	7.24 ± 0.01 ^f	7.72 ± 0.01 ^f	8.34 ± 0.01 ^c
	T3 2.03 ± 0.01 ^d	1.83 ± 0.01 ^b	2.06 ± 0.01 ^c	5.81 ± 0.01 ^b	6.22 ± 0.01 ^e	6.57 ± 0.01 ^c	7.84 ± 0.01 ^c	8.05 ± 0.01 ^f	8.64 ± 0.01 ^a	7.84 ± 0.01 ^c	8.05 ± 0.01 ^f	8.64 ± 0.01 ^a
Probiotic juices	T4 1.67 ± 0.01 ^e	1.56 ± 0.01 ^b	1.49 ± 0.01 ^e	6.33 ± 0.01 ^a	6.95 ± 0.01 ^a	7.22 ± 0.01 ^a	7.99 ± 0.01 ^a	8.51 ± 0.01 ^f	8.71 ± 0.06 ^d	6.33 ± 0.01 ^a	7.99 ± 0.01 ^a	8.71 ± 0.06 ^d
	T5 2.33 ± 0.01 ^a	1.9 ± 0.06 ^c	1.78 ± 0.01 ^d	4.11 ± 0.01 ^f	4.44 ± 0.01 ^b	4.8 ± 0.06 ^b	6.43 ± 0.01 ^f	6.34 ± 0.01 ^f	6.68 ± 0.01 ^c	4.11 ± 0.01 ^f	4.44 ± 0.01 ^b	6.68 ± 0.01 ^c
T6	2.23 ± 0.01 ^b	2.08 ± 0.01 ^b	1.92 ± 0.01 ^b	4.24 ± 0.01 ^e	4.44 ± 0.01 ^b	4.94 ± 0.01 ^f	6.47 ± 0.01 ^b	6.52 ± 0.01 ^f	6.86 ± 0.01 ^f	4.24 ± 0.01 ^e	6.52 ± 0.01 ^f	6.86 ± 0.01 ^f
Sig.	**	**	**	**	**	**	**	**	**	**	**	**

^{a-c} Means in the same column with the same classification with different superscripts differ significantly ($p < 0.05$); and ^{**} $p < 0.01$. T1, 100% mature orange juice; T2, 75% mature orange juice, 25% carrot juice; T3, 95% orange juice and 5% aloe vera gel; T4, 75% orange juice, 20% carrot juice, and 5% aloe vera gel; T5, 100% orange juice and probiotics T6, 75% orange juice, 25% carrot juice fermented with probiotics.

TABLE 3 Effect of aloe vera and probiotics on total soluble solids content in juices during the storage interval.

Treatments		Total soluble solids (°Brix)		
		Storage interval		
		First day	7 days	14 days
Control juices	T1	14.2 ± 0.06 ^a	14.3 ± 0.06 ^{ab}	14.6 ± 0.06 ^b
	T2	13.5 ± 0.06 ^c	13.7 ± 0.06 ^c	14.1 ± 0.06 ^c
Aloe vera juices	T3	14.1 ± 0.06 ^a	14.4 ± 0.06 ^a	14.8 ± 0.06 ^a
	T5	13.8 ± 0.06 ^b	14.2 ± 0.06 ^b	14.5 ± 0.06 ^b
Probiotic juices	T5	13.4 ± 0.06 ^c	13.8 ± 0.06 ^c	13.34 ± 0.01 ^d
	T6	12.2 ± 0.06 ^d	12.5 ± 0.06 ^d	12.85 ± 0.01 ^e
Sig.		**	**	**

^{a-c}Means in the same column with the same classification with different superscripts differ significantly ($p < 0.05$); and ^{**} $p < 0.01$.

total sugar content of all studied juice blends, where this increment reached the maximum values at the end of the storage period. The marked increase in total sugars might be attributed to the hydrolysis of polysaccharides such as starch, cellulose, and pectin.

3.3 Effect of aloe vera and probiotic on the total soluble solids contents of juices during storage period

Table 3 shows the total soluble solids (TSS) in aloe vera and probiotic juices during the storage interval. The data demonstrated a considerable rise in the TSS percentage for the control juice without additions in all storage intervals as compared with the other sample juice. The TSS decreased in juice-probiotic samples after 14 days, especially 75% orange juice, 25% carrot juice, and probiotics, due to sugar consumption. These present findings concur with several previous works (83, 84) that interpreted an increase of total soluble solids (TSS) due to the elevated amount of organic acid and polysaccharides subsidized with orange juice. Similarly, previous studies (36, 85) observed that the elevated total soluble solid values have a major role in preserving the taste and flavor of juice mixtures throughout the storage interval. Furthermore, another study (86) demonstrated that values might have been lower if TSS values were not modified to 12°Brix.

3.4 Effect of aloe vera and probiotics on the pH values of juices

The effect of aloe vera and probiotic juices on pH values during storage is shown in Figure 1. A decrease was observed in the pH values over the storage period. By the end of the storage period, the pH value of the control samples was 5.4 and 5.7. At the same time, it was 4.61 and 4.73 for the aloe vera gel-supported treatments, while the probiotic-supported treatments were 3.6 and

4.1, respectively. In the probiotic juice, there was a slight increase in the acidity values, corresponding to a slight decrease in the pH values. The present results are inconsistent with a previous study (87) that recorded no modulation in pH values of the various pasteurized juices of oranges and carrots at 4°C in the refrigerator and 10°C during the storage interval. One study (83) pointed out that three different formulations were created by blending carrot-orange juice, and the pH values decreased in all treatments during storage. Another study (36) pointed out that fresh orange-aloe vera juice treatments revealed no modulation in pH values during storage. A previous study (88) showed that probiotics reduced the pH of the juices during 10 h of incubation, either with or without encapsulated strains, due to the rise in acidity, which is consistent with our present findings (Figure 1). However, there were tenuous rises in pH values between the control and probiotic juices and a reduction during the storage from zero time (4.05) to 90 days (3.99). This reduction in pH may be due to the utilization of sugars present in juice by the probiotics to produce organic acids. Similarly, adding *Lactobacillus acidophilus* to carrot juice decreased the pH values of the resulting juice, as reported elsewhere (89).

3.5 Effect of aloe vera and probiotic on the color of juices during the storage interval

It should be stressed that color is one of the main indicators that show the changes in foods during the storage interval and significantly impacts consumer approval (90). As depicted in our study (Figure 2), control treatments experienced higher values of lightness L^* (47.39 and 46.67), as compared to T6 (38.49). In relation to variance, a^* and b^* values increased when the natural juices were subsidized with aloe vera gel. Another research project (91) illustrated that orange is excellent for the immune system, the heart, eyes, and cells. The data established that the aloe-gel addition ratio influences the taste and color. A previous study (92) indicated that the color estimate value decreases when the storage interval is increased. Another study (93) illustrated the color difference and concluded that the color was preferable at room temperature in the control treatment. However, the treated groups scored the least value at refrigerator temperature. Another study (94) indicated that the color variation in aloe vera gel at more than 4°C revealed a brown color at 25°C, including caramel anthraquinone oxidation, Maillard reaction, and ascorbic acid hydrolysis. On the other hand, color contrast in orange-carrot subsidized with aloe vera was decreased more than in the control sample at the end of the storage interval.

3.6 Effect of aloe vera and probiotic on the antioxidant content of juice during storage interval

The results of total phenol and flavonoid contents of aloe vera and probiotic juice during storage are illustrated in Table 4. It could be noted that control juices (without adding) on the first day and after 14 days had the highest level of total flavonoids and phenolics compared to the probiotic juices. The fortification

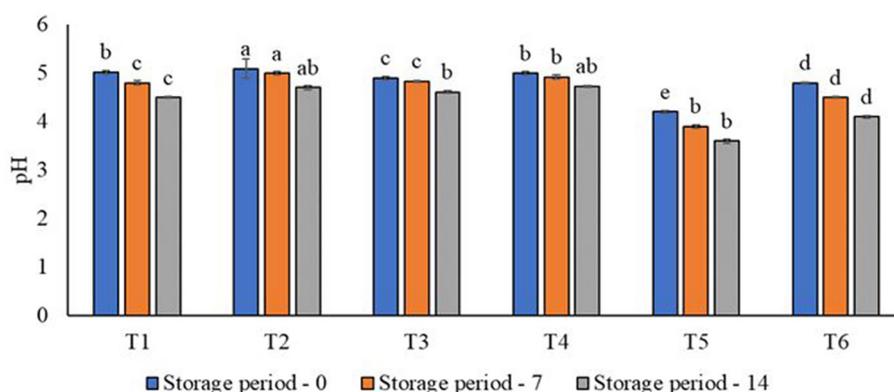


FIGURE 1

Effect of aloe vera and probiotic juice on the pH values during storage. T1: 100% natural orange juice; T2: 75% orange juice, 25% carrot juice; T3: 95% orange juice and 5% aloe vera gel; T4: 75% orange juice, 20% carrot juice, and 5% aloe vera gel; T5: 100% orange juice and probiotics; T6: 75% orange juice, 25% carrot juice fermented with probiotics. Each bar carrying different letters (a, b, c, d) is significantly different ($p < 0.05$).

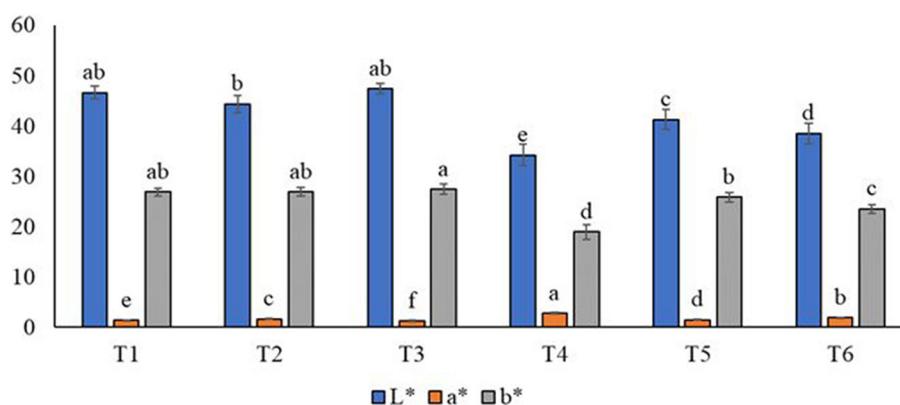


FIGURE 2

Effect of aloe vera and probiotics on color values of juice. T1: 100% nature orange juice; T2: 75% orange juice, 25% carrot juice; T3: 95% orange juice and 5% aloe vera gel; T4: 75% orange juice, 20% carrot juice and 5% aloe vera gel; T5: 100% orange juice and probiotics; T6: 75% orange juice, 25% carrot juice fermented with probiotics. Each bar carrying different letters (a, b, c, d, e, f) is significantly different ($p < 0.05$).

of control juices with aloe vera gel increased the content of total phenols and total flavonoids compared to control juices. The rise in the antioxidant activity might have resulted from the hypothesis that polyphenols undergo polymerization reactions (95). A previous research study (96) revealed the rise in antioxidant activity, which is commonly attributed to Maillard interactions. In our study, the flavonoids in treatment 4 (mixed natural juice with aloe vera) were higher than in treatment 6 (mixed natural juice with probiotics), which was 180 mg/100 mL and 117.08 mg/100 mL, respectively. Polymerization reactions and the formation of new compounds led to unstable phenomena in flavonoids and phenols, including an increase and decrease during the storage interval. Our results are consistent with a previous study (97) that showed that most polyphenol compounds are found in aloe vera gel. The present findings are consistent with the study, which revealed that the flavonoid compounds of berry juices decreased during storage (98). Our study is in line with previous work (94), which documented an increase in the percentage of total phenol contents (TPC) and total flavonoid contents (TFC)

of aloe vera juice at 25°C during storage for 30 days compared to storing it at 4°C. Other previous studies (36, 99) have also documented a prominent reduction in reducing the antioxidants of various fruits and vegetables at refrigeration temperature during storage intervals.

3.7 Viable counts in the fermented orange juices and their mixtures

Cell viability is considered an important factor for the evaluation of functional products. Briefly, the three tested LAB strains revealed ~ 7.0 log CFU/mL under the same suitable conditions before addition into juices. All three LAB strains exhibited good viable cell counts in orange juices at pH 6.6, 37°C for 48 h, with viable counts of 7.42–8.07 log CFU/mL (Table 5). The result showed that orange juice, as a lactic fermentation substrate, could be beneficial for the growth of LAB, whose concentration was always higher than the minimum to maintain a healthy life (7.0

TABLE 4 Effect of total polyphenols (TPC) and flavonoids (TFC) content on aloe vera and probiotic juices during storage interval.

Treatments		TPC mg/100 mL			TFC mg/100 mL		
		Storage interval (days)			Storage interval (days)		
		First day	7 day	14 days	First day	7 day	14 days
Control juices	T1	330 ± 0.0 ^a	340 ± 0.0 ^a	300 ± 0.0 ^b	200 ± 0.0 ^a	190 ± 0.0 ^a	175 ± 0.0 ^a
	T2	280 ± 0.0 ^e	305 ± 0.0 ^d	320 ± 0.0 ^a	140 ± 0.0 ^d	130 ± 0.0 ^d	115 ± 0.0 ^f
Aloe vera juices	T3	310 ± 0.0 ^b	310 ± 0.0 ^c	270 ± 0.0 ^d	190 ± 0.0 ^b	175 ± 0.0 ^b	170 ± 0.0 ^b
	T4	300 ± 0.0 ^c	320 ± 0.0 ^b	250 ± 0.0 ^e	180 ± 0.0 ^c	165 ± 0.0 ^c	160 ± 0.0 ^c
Probiotic juices	T5	226.02 ± 0.01 ^f	219.5 ± 0.06 ^f	222.7 ± 0.06 ^f	112.12 ± 0.01 ^h	108.67 ± 0.01 ^h	110.3 ± 0.06 ^g
	T6	212.1 ± 0.06 ^g	209.87 ± 0.01 ^g	210.8 ± 0.06 ^g	117.08 ± 0.01 ^g	113.97 ± 0.01 ^g	115.53 ± 0.01 ^e
Sig.		**	**	**	**	**	**

^{a-c}Means in the same column with the same classification with different superscripts differ significantly ($p < 0.05$); and ^{**} $p < 0.01$.

TABLE 5 Viable counts in the fermented orange juices during storage.

Strains (log CFU/mL)	Storage period (day)	Probiotic juices	
		T5	T6
<i>L. acidophilus</i>	1	7.54 ± 0.02 ^a	7.86 ± 0.08 ^a
	7	7.20 ± 0.11 ^b	7.48 ± 0.06 ^b
	14	6.88 ± 0.04 ^c	7.02 ± 0.02 ^c
<i>L. plantarum</i>	1	7.66 ± 0.24 ^a	7.94 ± 0.14 ^a
	7	7.30 ± 0.18 ^b	7.58 ± 0.30 ^b
	14	7.12 ± 0.12 ^c	7.26 ± 0.16 ^c
<i>B. lactis</i>	1	7.96 ± 0.18 ^a	8.02 ± 0.30 ^a
	7	7.72 ± 0.20 ^b	7.88 ± 0.14 ^b
	14	7.58 ± 0.26 ^c	7.70 ± 0.24 ^c

^{a-c}Means in the same column with the same classification with different superscripts differ significantly ($p < 0.05$).

log CFU/mL (100). Our results agree with data reported by Quan et al. (101), who found that orange juice could be beneficial for the growth of LAB. Also, the fortification of orange juice with carrot juice enhanced the viability of LAB strains compared with orange juice, possibly due to the lower acidity of carrot juice. Carrot juice also contains prebiotics that might improve the viability of LAB bacteria (102). The viability of LAB strains decreased as the storage period progressed in two treatments.

3.8 Effect of aloe vera and probiotic on the sensory evaluation of juice during storage

The sensory properties of aloe vera and probiotic juice during the storage interval are illustrated in Table 6. The presented results showed that all samples were changed significantly in color, taste, odor, texture, and overall acceptability. The data in Table 6 indicates that the highest color score (9.5) was recorded for T2 and T4, followed by T6, which recorded a color score of (9.4). The results of the color parameter indicated that the carrot percent in natural mixed juices led to improved juice color compared with orange.

From the same table, it could be observed that carrot percent led to a noticeable negative effect on the taste of the obtained natural mixed juices. Where the highest test scores (9.4) were recorded for T4, T5, and T6. Concerning flavor results, which are shown in Table 6, T5 had the highest flavor score (9.5), followed by T1 and T6 (9.4), while T2, T3, and T5 had the lowest flavor score (9.3). Regarding texture results presented in Table 6, all treatments had a slight effect on juice texture, where the scored values ranged between 9.3 and 9.5. In relation to overall acceptability results, T3, T4, and T6 have the highest overall acceptability (9.5), followed by T2 and T5 (9.4), while the lowest values were recorded for T1 and T3 (9.3). As depicted in Table 6, orange had a noticeable positive effect on the taste and flavor of the obtained natural mix. On the contrary, carrots had a noticeable positive effect on the color and textures of the obtained natural mixed. A previous study (36) demonstrated that adding aloe vera gel at the rate of 5% might be helpful in ameliorating taste, flavor, and overall acceptance of samples. During storage, there was a good sensory evaluation for aloe vera juice-orange juice for 45 days, which declined when the storage period reached 90 days. Another work (103) proved that the convenient mixing of vegetable and fruit juices modifies acidity and saccharides, besides promoting microbial inhibition, taste, bioactive component preservation, and odor of the final juice product. Another previous study (86), considering the overall acceptability of samples fortified with probiotics, concluded that there were no significant variances in sensual characteristics of freshly prepared probiotics juice samples after 10 h of incubation, while orange juice without probiotics cleared the lowest acceptance.

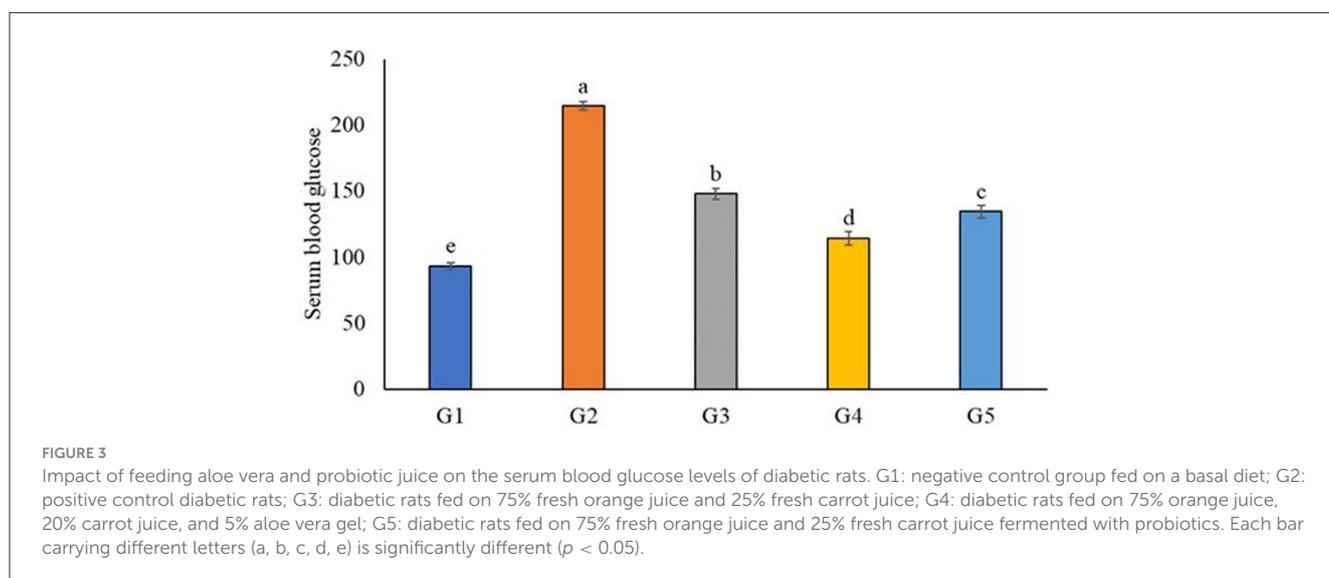
3.9 Impact of feeding aloe vera and probiotic juice on blood glucose level of diabetic rats

Figure 3 shows the blood glucose level of diabetic rats that fed on functional natural juice subsidized with aloe vera and probiotics vs. the negative (-ve) and positive (+ve) control groups. In our study, the lowest glucose level reduction was in G4, which represented diabetic rats fed on 75% orange juice, 20% carrot juice, and 5% aloe vera (G4), and the blood glucose level was 114.27

TABLE 6 Effect of aloe vera and probiotics on the sensory evaluation of juice.

Treatments		Color	Taste	Odor	Textures	Overall acceptability
Control juices	T1	9.3 ± 0.06 ^{bc}	9.3 ± 0.06 ^a	9.4 ± 0.06 ^{ab}	9.3 ± 0.06	9.4 ± 0.06 ^a
	T2	9.5 ± 0.06 ^a	9 ± 0.06 ^b	9.3 ± 0.06 ^{bc}	9.4 ± 0.06	9.1 ± 0.06 ^b
Aloe vera juices	T3	9.2 ± 0.06 ^c	9 ± 0.06 ^b	9.3 ± 0.06 ^{bc}	9.3 ± 0.06	9.5 ± 0.06 ^a
	T4	9.5 ± 0.06 ^a	9.4 ± 0.06 ^a	9.3 ± 0.06 ^{bc}	9.5 ± 0.06	9.5 ± 0.06 ^a
Probiotic juices	T5	9.3 ± 0.06 ^{bc}	9.4 ± 0.06 ^a	9.5 ± 0.06 ^a	9.4 ± 0.06	9.4 ± 0.06 ^a
	T6	9.4 ± 0.06 ^{ab}	9.4 ± 0.06 ^a	9.4 ± 0.06 ^{ab}	9.5 ± 0.06	9.5 ± 0.06 ^a
Sig.		**	**	**	NS	**

^{a-c}Means in the same column with different superscripts differ significantly ($p < 0.05$); NS, not significant; and ^{**} $p < 0.01$.



g/dl. This was followed by G5, which was fed 75% orange juice, 25% carrot juice, and probiotics, and it had a blood glucose level of 134.63 g/dl. Similarly, a previous study (104) reported that the aloe vera gel reduced the blood glucose level of diabetic animals. These results concur with previous studies (33, 105–107), which confirmed that aloe vera significantly helps lower glucose sugar. A previous work (108) illustrated that probiotics were an active factor in reducing blood glucose levels and triglyceride in rats compared to the control group.

3.10 Body weights of diabetic albino rats feeding on aloe vera and probiotic juices

Table 7 depicts the feeding effect of diabetic rats on functional natural juice supplemented with aloe vera and probiotics on body weight gain (BWG), food intake (FI), and feed efficiency ratio (FER). As shown, the positive control group (-ve) had a higher rate of BWG ($81 \pm 0.58a$), food intake ($31 \pm 0.58a$), and feed efficiency ratio ($0.06 \pm 0.01a$) when compared to the other groups. Meanwhile, diabetic rats fed functional natural juice supplemented with aloe vera and probiotics showed a significant decrease in these parameters compared to the positive control group, but this decrease was gradual. Furthermore, diabetic rats fed on functional

TABLE 7 Body weight of diabetic rats feeding on aloe vera and probiotic juices.

Groups	Body weight gain (g 60 day)	Food intake (g day)	Feed efficiency ratio
G1 (-ve)	81 ± 0.58^a	31 ± 0.58^a	0.06 ± 0.01^a
G2 (+ve)	71 ± 0.58^b	28 ± 0.58^b	0.06 ± 0.01^b
G3	48 ± 0.58^c	29.1 ± 0.06^b	0.04 ± 0.01^c
G4	27 ± 0.58^e	29.3 ± 0.06^b	0.03 ± 0.01^e
G5	38 ± 0.58^d	28 ± 0.58^b	0.03 ± 0.01^d
Sig.	**	**	**

^{a-c}Means in the same column with the same classification with different superscripts differ significantly ($p < 0.05$); and ^{**} $p < 0.01$. -ve, negative control group fed on a basal diet; +ve, positive control diabetic rats; G3, diabetic rats fed on 75% fresh orange juice and 25% fresh carrot juice; G4, diabetic rats fed on 75% orange juice, 20% carrot juice, and 5% aloe vera gel; G5, diabetic rats fed on 75% fresh orange juice and 25% fresh carrot juice fermented with probiotics.

natural juice supplemented with aloe vera and probiotics displayed significantly reduced body weight gain, food intake, and feed efficiency ratio compared to rats fed on a basal diet. A previous study (104) showed the oral administration of aloe vera juice decreased rats' body weight at the end of the experiment. It was

TABLE 8 Effect of aloe vera and probiotics juices on serum lipids profile diabetic rats.

Groups	Triglycerides (mg/dl)	Total cholesterol (mg/dl)	HDL (mg/dl)	LDL (mg/dl)
G1	83.70 ± 3.40 ^d	80.60 ± 3.35 ^d	40.50 ± 1.70 ^a	24.98 ± 1.64 ^e
G2	132.30 ± 2.66 ^a	100.20 ± 4.70 ^a	28.60 ± 1.40 ^d	45.14 ± 2.02 ^a
G3	108.20 ± 2.80 ^b	93.50 ± 2.90 ^b	30.50 ± 1.70 ^b	41.54 ± 2.60 ^b
G4	95.80 ± 2.70 ^b	87.90 ± 2.70 ^b	35.40 ± 1.50 ^c	33.34 ± 1.20 ^d
G5	102.30 ± 2.30 ^c	91.90 ± 2.50 ^c	32.60 ± 1.60 ^c	38.84 ± 1.94 ^c
Sig.	**	**	**	**

^{a-c}Means in the same column with the same classification with different superscripts differ significantly ($p < 0.05$); and ^{**} $p < 0.01$. -ve, negative control group fed on a basal diet; +ve, positive control diabetic rats; G3, diabetic rats fed on 75% fresh orange juice and 25% fresh carrot juice; G4, diabetic rats fed on 75% orange juice, 20% carrot juice, and 5% aloe vera gel; G5, diabetic rats fed on 75% fresh orange juice and 25% fresh carrot juice fermented with probiotics.

TABLE 9 Effect of aloe vera and probiotic juices on the Kidney and liver functions of diabetic rats.

Groups	Kidney functions		Liver functions		
	Creatinine (mg/dl)	Urea (mg/dl)	Total protein (g/dl)	ALT (U/L)	AST (U/L)
G1	0.52 ± 0.01 ^e	19.53 ± 0.01 ^d	5.44 ± 0.01 ^a	37.25 ± 0.01 ^e	27.15 ± 0.01 ^c
G2	0.94 ± 0.01 ^a	31.53 ± 0.01 ^a	3.75 ± 0.01 ^e	67.96 ± 0.01 ^a	54.99 ± 0.01 ^a
G3	0.80 ± 0.01 ^b	29.3 ± 0.06 ^b	3.84 ± 0.01 ^d	53.22 ± 0.01 ^c	45.44 ± 0.01 ^b
G4	0.73 ± 0.01 ^c	24.5 ± 0.01 ^c	4.64 ± 0.01 ^c	50.25 ± 0.01 ^b	22.46 ± 0.01 ^d
G5	0.58 ± 0.01 ^d	14.5 ± 0.01 ^e	5.37 ± 0.01 ^b	41.46 ± 0.01 ^d	22.5 ± 0.06 ^d
Sig.	**	**	**	**	**

^{a-c}Means in the same column with the same classification with different superscripts differ significantly ($p < 0.05$); and ^{**} $p < 0.01$. -ve, negative control group, fed on a basal diet; +ve, positive control diabetic rats; G3, diabetic rats fed on 75% fresh orange juice and 25% fresh carrot juice; G4, diabetic rats fed on 75% orange juice, 20% carrot juice and 5% aloe vera gel; G5, diabetic rats fed on 75% fresh orange juice and 25% fresh carrot juice fermented with probiotics.

found that the group orally given aloe vera juice gained the highest value of BWG, followed by the negative (healthy) control group, while the positive control group with diabetes decreased in weight due to type 2 diabetes. These findings are consistent with the main characteristics of diabetes, which include thirst, frequent eating, urinating, hyperglycemia, weight loss, and low insulin levels (109). The increase in the weight of animals treated with aloe vera juice could be attributed to the presence of magnesium, as it plays a major function in stabilizing lipid membranes, reproduction, and metabolic processes (110, 111).

3.11 Effect of aloe vera and probiotic juices on serum lipids profile diabetic rats

The results presented in Table 8 show that triglyceride and total cholesterol levels of the positive control group were significantly higher than the other experimental groups during the experimental intervals compared with the negative control group. In recent years, functional foods have become popular and are considered a curative and preventive agent for some chronic diseases such as diabetes. As shown in Table 8, feeding diabetic rats on aloe vera and probiotic juices caused a significant improvement in lipid profile. Our study illustrated that diabetic rats (G2 and G3) had the highest levels of TG, TC, LDL, and VLDL compared to the groups that feed on functional natural juices supplemented with aloe vera and probiotics (G4 and G5). Also, it could be observed that in groups G4 and G5, the value of HDL (35.40 and 32.60 mg/dl, respectively) was increased compared to groups G2 and G3. A previous study

(34) noticed that all liver enzymes, blood cholesterol, HDL (high-density lipoprotein) cholesterol, and LDL (low-density lipoprotein) cholesterol reverted to almost normal levels because of the aloe vera gel. Feeding rats on aloe vera gel did not show any important alteration in total protein and albumin when compared with the control group.

3.12 The kidney and liver functions of diabetic albino rats

Table 9 shows the impact of aloe vera and probiotic juices on the kidney and liver functions of diabetic rats. Concerning kidney function, feeding diabetic rats on aloe vera and probiotic juices (G4 and G5) showed a significant reduction in serum creatinine and urea levels compared with G2 and G3. Furthermore, the positive control group (+ve) illustrated an important rise in ALT and AST enzyme compared with the other groups. Similarly, a previous study (104) illustrated the impact of oral management of aloe vera and probiotic juice on the significant increase in blood urea level of the animals of the group given aloe vera juice compared to the control groups. The same finding was noticed in the significant value of creatinine for the animals of the group given aloe vera juice. A previous report (104) pointed out the influence of oral administration of aloe vera juice on the significant values of liver enzymes for animals of the group given aloe vera juice, whereas GOT significantly decreased in the treated group (46.33 mg/dl) when compared to the infected group (58.33 mg/dl).

4 Conclusions

The study demonstrated the possibility of using aloe vera gel and probiotics in orange and carrot juices, in addition to evaluating the functional element of the juices (chemically, microbiologically, and sensory) during 14 days of storage at refrigerator temperature. The present study also revealed the improvement in the physicochemical, microbiological, and sensory properties of healthy juices containing aloe vera gel and probiotics. This was reflected by the improvement of total soluble solids, reducing sugars and total sugars, and the phytochemical quality with good and acceptable organoleptic properties. Moreover, the study confirmed that adding aloe vera to the natural mixed juices improved sensory attributes. The study revealed that increasing the orange juice ratio improved the taste, whereas increasing the carrot ratio improved the color. Given the abovementioned results, oranges, carrots, and aloe vera can be used to produce new, tasteful, and healthy juices. In addition, this research concluded that aloe vera juice is more efficacious as an antidiabetic agent in diabetic rats. Collectively, the study pointed out that using orange and carrot juice enriched with aloe vera might be promising for treating diabetes, while orange and carrot juice fortified with probiotics could also help overcome lactose intolerance. Further research on the mechanistic pathways underlying these antidiabetic effects of healthy juices containing aloe vera gel and probiotics should be studied.

Data availability statement

The original contributions presented in the study are included in the article/supplementary material, further inquiries can be directed to the corresponding authors.

Ethics statement

This study was conducted with the approval of the approval of the institutional animal care and research Unit, Zagazig University (Institutional Review Board Number ZU-IACUC/2/F/339/2022). The study was conducted in accordance with the local legislation and institutional requirements.

Author contributions

SM: Conceptualization, Data curation, Formal analysis, Investigation, Methodology, Project administration, Validation,

Visualization, Writing – original draft, Writing – review & editing. AA-N: Formal analysis, Investigation, Methodology, Software, Supervision, Validation, Writing – original draft. ER-Á: Funding acquisition, Project administration, Resources, Software, Visualization, Writing – review & editing. FA: Data curation, Investigation, Software, Supervision, Validation, Visualization, Writing – original draft. HS: Formal analysis, Investigation, Software, Supervision, Validation, Visualization, Writing – original draft. ME-K: Data curation, Funding acquisition, Resources, Software, Validation, Writing – review & editing. MS: Data curation, Funding acquisition, Resources, Software, Validation, Visualization, Writing – review & editing. AH: Funding acquisition, Resources, Software, Writing – review & editing. AAA: Data curation, Funding acquisition, Resources, Software, Validation, Visualization, Writing – review & editing. AA: Data curation, Funding acquisition, Resources, Software, Validation, Visualization, Writing – review & editing. EE: Data curation, Formal analysis, Funding acquisition, Investigation, Resources, Software, Supervision, Validation, Visualization, Writing – original draft, Writing – review & editing.

Funding

The author(s) declare that financial support was received for the research, authorship, and/or publication of this article. This study was supported by Princess Nourah bint Abdulrahman University Researchers Supporting Project number (PNURSP2024R23), Princess Nourah bint Abdulrahman University, Riyadh, Saudi Arabia.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

References

- John R, Singla A. Functional foods: components, health benefits, challenges, and major projects. *DRC Sustain Fut.* (2021) 2:61–72. doi: 10.37281/DRCSF/2.1.7
- Atwaa ESH, Shahein MR, El-Sattar ESA, Hijazy HHA, Albrakati A, Elmahallawy EK. Bioactivity, physicochemical and sensory properties of probiotic yoghurt made from whole milk powder reconstituted in aqueous fennel extract. *Fermentation.* (2022) 8:52. doi: 10.3390/fermentation8020052
- Atwaa ESH, Shahein MR, Radwan HA, Mohammed NS, Aloraini MA, Albezrah NKA, et al. Antimicrobial activity of some plant extracts and their applications in homemade tomato paste and pasteurized cow milk as natural preservatives. *Fermentation.* (2022) 8:428. doi: 10.3390/fermentation8090428
- Shahein MR, Atwaa ESH, Alrashdi BM, Ramadan MF, Abd El-Sattar ES, Siam AAH, et al. Effect of fermented camel milk containing pumpkin seed milk on the

- oxidative stress induced by carbon tetrachloride in experimental rats. *Fermentation*. (2022) 8:223. doi: 10.3390/fermentation8050223
5. Shahein MR, Atwaa ESH, Elkot WF, Hijazy HHA, Kassab RB, Alblihed MA, et al. The impact of date syrup on the physicochemical, microbiological, and sensory properties, and antioxidant activity of bio-fermented camel milk. *Fermentation*. (2022) 8:192. doi: 10.3390/fermentation8050192
 6. Shahein MR, Atwaa ESH, El-Zahar KM, Elmaadawy AA, Hijazy HHA, Sitohy MZ, et al. Remedial action of yoghurt enriched with watermelon seed milk on renal injured hyperuricemic rats. *Fermentation*. (2022) 8:41. doi: 10.3390/fermentation8020041
 7. Shahein MR, Atwaa ESH, Radwan HA, Elmeligy AA, Hafiz AA, Albrakati A, et al. Production of a yogurt drink enriched with golden berry (*Physalispubescens* L) Juice and its therapeutic effect on hepatitis in rats. *Fermentation*. (2022) 8:112. doi: 10.3390/fermentation8030112
 8. Shahein MR, Atwaa E-SH, Babalghith AO, Alrashdi BM, Radwan HA, Umair M, et al. Impact of incorporating the aqueous extract of hawthorn (*C. oxyanatha*) leaves on yogurt properties and its therapeutic effects against oxidative stress induced by carbon tetrachloride in rats. *Fermentation*. (2022) 8:200. doi: 10.3390/fermentation8050200
 9. Shahein MR, Elkot WF, Albezrah NKA, Abdel-Hafez LJM, Alharbi MA, Massoud D, et al. Production of a yogurt drink enriched with golden berry (*Physalispubescens* L) Juice and its therapeutic effect on hepatitis in rats. *Fermentation*. (2022) 8:390. doi: 10.3390/fermentation8080390
 10. Swelam S, Zommara MA, Abd El-Aziz AE-AM, Elgammal NA, Baty RS, Elmahallawy EK. Insights into Chufa milk frozen yoghurt as cheap functional frozen yoghurt with high nutritional value. *Fermentation*. (2021) 7:255. doi: 10.3390/fermentation7040255
 11. Shahein MR, Raya-Álvarez E, Hassan MA, Hashim MA, Dahran N, El-Khadragy MF, et al. Assessment of the physicochemical and sensory characteristics of fermented camel milk fortified with cordia myxa and its biological effects against oxidative stress and hyperlipidemia in rats. *Front Nutr*. (2023) 10:1130224. doi: 10.3389/fnut.2023.1130224
 12. Zommara MA, Bedeer EG, Elmahallawy EK, Hafiz AA, Albrakati A, Swelam S. Preliminary studies of bio-fortification of yoghurt with chromium. *Fermentation*. (2022) 8:727. doi: 10.3390/fermentation8120727
 13. Putnik P, Pavlič B, Šojić B, Zavadlav S, Žuntar I, Kao L, et al. Innovative hurdle technologies for the preservation of functional fruit juices. *Foods*. (2020) 9:699. doi: 10.3390/foods9060699
 14. Gonçalves D, Ferreira P, Baldwin E, Cesar T. Health benefits of orange juice and citrus flavonoids. In: Ye X, editor. *Phytochemicals in Citrus-Applications in Functional Foods*. Boca Raton, FL: CRC Press (2018). 299–324. doi: 10.1201/9781315369068-10
 15. Czech A, Zarycka E, Yanovych D, Zasadna Z, Grzegorzczak I, Klys S. Mineral content of the pulp and peel of various citrus fruit cultivars. *Biol Trace Elem Res*. (2020) 193:555–63. doi: 10.1007/s12011-019-01727-1
 16. de Moraes Crizel T, Jablonski A, de Oliveira Rios A, Rech R, Flóres SH. Dietary fiber from orange byproducts as a potential fat replacer. *LWT Food Sci Technol*. (2013) 53:9–14. doi: 10.1016/j.lwt.2013.02.002
 17. Alemdar S, Agaoglu S. Investigation of *in vitro* antimicrobial activity of aloe vera juice. *J Anim Vet Adv*. (2009) 8:99–102.
 18. Saini RK, Ranjit A, Sharma K, Prasad P, Shang X, Gowda KGM, et al. Bioactive compounds of citrus fruits: a review of composition and health benefits of carotenoids, flavonoids, limonoids, and terpenes. *Antioxidants*. (2022) 11:239. doi: 10.3390/antiox11020239
 19. Bayraktar E, Kocapıçak Ö, Mehmetoglu Ü, Parlaktuna M, Mehmetoglu T. Recovery of amino acids from reverse micellar solution by gas hydrate. *Chem Eng Res Design*. (2008) 86:209–13. doi: 10.1016/j.cherd.2007.12.001
 20. Romero-Lopez MR, Osorio-Diaz P, Bello-Perez LA, Tovar J, Bernardino-Nicanor A. Fiber concentrate from orange (*Citrus sinensis* L) bagasse: characterization and application as bakery product ingredient. *Int J Mol Sci*. (2011) 12:2174–86. doi: 10.3390/ijms12042174
 21. Karangwa E, Hayat K, Rao L, Nshimiyimana DS, Foh MB Li L, et al. Improving blended carrot-orange juice quality by the addition of cyclodextrins during enzymatic clarification. *Food Bioprocess Technol*. (2012) 5:2612–7. doi: 10.1007/s11947-011-0557-z
 22. Aydogan Ö, Bayraktar E, Parlaktuna M, Mehmetoglu T, Mehmetoglu Ü. Production of L-aspartic acid by biotransformation and recovery using reverse micelle and gas hydrate methods. *Biocatal Biotransform*. (2007) 25:365–72. doi: 10.1080/10242420701510395
 23. Chantaro P, Devahastin S, Chiewchan N. Production of antioxidant high dietary fiber powder from carrot peels. *LWT Food Sci Technol*. (2008) 41:1987–94. doi: 10.1016/j.lwt.2007.11.013
 24. Boadi NO, Badu M, Kortei NK, Saah SA, Annor B, Mensah MB, et al. Nutritional composition and antioxidant properties of three varieties of carrot (*Daucus carota*). *Sci Afr*. (2021) 12:e00801. doi: 10.1016/j.sciaf.2021.e00801
 25. Ahmad T, Cawood M, Iqbal Q, Ariño A, Batool A, Tariq RMS, et al. Phytochemicals in *Daucus carota* and their health benefits. *Foods*. (2019) 8:424. doi: 10.3390/foods8090424
 26. Alina PG, Camelia V. Evolution of antioxidant capacity of blend juice made from beetroot, carrot and celery during refrigerated storage. *Annals of the University Dunarea de Jos of Galati*. (2013) 37:93–9.
 27. Tiwari M, Upadhayay M. The medicinal plant components and applications (aloe vera). *J Med Plants Stud*. (2018) 6:89–95.
 28. Saleem M. Aloe Barbadensis Miller: a comprehensive review. *Pak J Sci*. (2021) 73:388. doi: 10.57041/pjs.v73i4.388
 29. Goswami A, Malik A, Malik M, Chaudhary S. Medicinal benefits of aloe vera (*Aloe barbadensis*). *Ann Horticul*. (2020) 13:160–6. doi: 10.5958/0976-4623.2020.00028.6
 30. Kumar R, Singh AK, Gupta A, Bishayee A, Pandey AK. Therapeutic potential of aloe vera—a miracle gift of nature. *Phytomedicine*. (2019) 60:152996. doi: 10.1016/j.phymed.2019.152996
 31. Haghani F, Arabnezhad M-R, Mohammadi S, Ghaffarian-Bahraman A. Aloe vera and streptozotocin-induced diabetes mellitus. *Rev Bras Farmacogn*. (2022) 32:1–14. doi: 10.1007/s43450-022-00231-3
 32. Gao Y, Kuok KI, Jin Y, Wang R. Biomedical applications of aloe vera. *Crit Rev Food Sci Nutr*. (2019) 59:S244–56. doi: 10.1080/10408398.2018.1496320
 33. Budiastutik I, Subagio HW, Kartasurya MI, Widjanarko B, Kartini A, Soegiyanto SS. The Effect of aloe vera on fasting blood glucose levels in pre-diabetes and type 2 diabetes mellitus: a systematic review and meta-analysis. *J Pharm Pharmacogn Res*. (2022) 10:737–47. doi: 10.56499/jppres22.1378_10.4.737
 34. Anwar DA, Eid HR, Ghadir A. Nutritional and antioxidant properties of mango juice and aloe vera gel and their effect on diabetic rats. *Lett Appl NanoBioScience Appl*. (2020) 9:1602–14. doi: 10.33263/LIANB594.16021614
 35. Hamid GH, El-Kholany EA, Nahla EA. Evaluation of aloe vera gel as antioxidant and antimicrobial ingredients in orange-carrot blend nectars. *Middle East J*. (2014) 3:1122–34.
 36. Kausar T, Shamim F, Gorski FI, Ainee A. 23 Preparation and quality evaluation of ready to serve beverage (RTs) from orange juice and aloe vera gel during storage. *Pure Appl Biol*. (2020) 9:219–28. doi: 10.19045/bspab.2020.90026
 37. Ahlawat KS, Khatkar BS. Processing, food applications and safety of aloe vera products: a review. *J Food Sci Technol*. (2011) 48:525–33. doi: 10.1007/s13197-011-0229-z
 38. Sheehan VM, Ross P, Fitzgerald GF. Assessing the acid tolerance and the technological robustness of probiotic cultures for fortification in fruit juices. *Innovat Food Sci Emerg Technol*. (2007) 8:279–84. doi: 10.1016/j.ifset.2007.01.007
 39. Mattila-Sandholm T, Myllärinen P, Crittenden R, Mogensen G, Fondén R, Saarela M. Technological challenges for future probiotic foods. *Int Dairy J*. (2002) 12:173–82. doi: 10.1016/S0958-6946(01)00099-1
 40. Savaiano DA, Hutkins RW. Yogurt, cultured fermented milk, and health: a systematic review. *Nutr Rev*. (2021) 79:599–614. doi: 10.1093/nutrit/nuaa013
 41. Loo YT, Howell K, Chan M, Zhang P, Ng K. Modulation of the human gut microbiota by phenolics and phenolic fiber-rich foods. *Compr Rev Food Sci Food Saf*. (2020) 19:1268–98. doi: 10.1111/1541-4337.12563
 42. Leonard W, Liang A, Ranadheera CS, Fang Z, Zhang P. Fruit juices as a carrier of probiotics to modulate gut phenolics and microbiota. *Food Funct*. (2022) 13:10333–46. doi: 10.1039/D2FO01851A
 43. Szutowaska J. Functional properties of lactic acid bacteria in fermented fruit and vegetable juices: a systematic literature review. *Eur Food Res Technol*. (2020) 246:357–72. doi: 10.1007/s00217-019-03425-7
 44. Wang Q, Sun Q, Wang J, Qiu X, Qi R, Huang J. *Lactobacillus plantarum* 299v changes mirna expression in the intestines of piglets and leads to downregulation of litaif by regulating Ssc-Mir-450a. *Probiotics Antimicrob Proteins*. (2021) 13:1093–105. doi: 10.1007/s12602-021-09743-1
 45. Jungersen M, Wind A, Johansen E, Christensen JE, Stuer-Lauridsen B, Eskesen D. The science behind the probiotic strain *Bifidobacterium animalis* Subsp. Lactis Bb-12[®]. *Microorganisms*. (2014) 2:92–110. doi: 10.3390/microorganisms2020092
 46. Cheng J, Laitila A, Ouwehand AC. *Bifidobacterium animalis* Subsp. Lactis Hn019 effects on gut health: a review. *Front Nutr*. (2021) 8:790561. doi: 10.3389/fnut.2021.790561
 47. Tahmourespour A, Kermanshahi RK. The effect of a probiotic strain (*Lactobacillus acidophilus*) on the plaque formation of oral streptococci. *Bosnian J Basic Med Sci*. (2011) 11:37. doi: 10.17305/bjbm.2011.2621
 48. Huang Z, Zhou X, Stanton C, Ross RP, Zhao J, Zhang H, et al. Comparative genomics and specific functional characteristics analysis of *Lactobacillus acidophilus*. *Microorganisms*. (2021) 9:1992. doi: 10.3390/microorganisms9091992
 49. Parvez S, Malik KA, Ah Kang S, Kim HY. Probiotics and their fermented food products are beneficial for health. *J Appl Microbiol*. (2006) 100:1171–85. doi: 10.1111/j.1365-2672.2006.02963.x
 50. Nomoto K. Prevention of infections by probiotics. *J Biosci Bioeng*. (2005) 100:583–92. doi: 10.1263/jbb.100.583
 51. Shanahan F. Probiotics in inflammatory Bowel disease—therapeutic rationale and role. *Adv Drug Deliv Rev*. (2004) 56:809–18. doi: 10.1016/j.addr.2003.11.003

52. Al-Sahlany STG, Khassaf WH, Niamah AK, Abd Al-Manhel AJ. Date juice addition to bio-yogurt: the effects on physicochemical and microbiological properties during storage, as well as blood parameters *in vivo*. *J Saudi Soc Agric Sci*. (2023) 22:71–7. doi: 10.1016/j.jssas.2022.06.005
53. Al-Sahlany ST, Niamah AK. Bacterial viability, antioxidant stability, antimutagenicity and sensory properties of onion types fermentation by using probiotic starter during storage. *Nutr Food Sci*. (2022) 52:901–16. doi: 10.1108/NFS-07-2021-0204
54. Khursheed T, Imran M, Khalil AA, Malik M, Raza A, Naz S, et al. Determination of proximate composition and antioxidant potential of aloe vera gel with the development and sensory evaluation of aloe vera based drinks. *Asian J Allied Health Sci*. 4:14–20.
55. Man D, Rogosa, M, Sharpe M. A medium for the cultivation of lactobacilli. *J Appl Bact*. (1960) 23:130–5. doi: 10.1111/j.1365-2672.1960.tb00188.x
56. Nguyen BT, Bujna E, Fekete N, Tran ATM, Rezessy-Szabo JM, Prasad R, et al. Probiotic beverage from pineapple juice fermented with *Lactobacillus* and *Bifidobacterium* strains. *Front Nutr*. (2019) 6:54. doi: 10.3389/fnut.2019.00054
57. Bujna E, Farkas NA, Tran AM, Dam MS, Nguyen QD. Lactic acid fermentation of apricot juice by mono-and mixed cultures of probiotic *Lactobacillus* and *Bifidobacterium* strains. *Food Sci Biotechnol*. (2018) 27:547–54. doi: 10.1007/s10068-017-0269-x
58. AOAC G. *Official Methods of Analysis of Aoac International*. Rockville, Md: Aoac International, ISBN: 978-0-935584-87-5. (2016).
59. Stinco CM, Fernández-Vázquez R, Escudero-Gilete ML, Heredia FJ, Meléndez-Martínez AJ, Vicario IM. Effect of orange juice's processing on the color, particle size, and bioaccessibility of carotenoids. *J Agric Food Chem*. (2012) 60:1447–55. doi: 10.1021/jf2043949
60. Gao X, Ohlander M, Jeppsson N, Björk, Trajkovski V. Changes in antioxidant effects and their relationship to phytonutrients in fruits of Sea Buckthorn (*Hippophae rhamnoides* L) during maturation. *J Agric Food Chem*. (2000) 48:1485–90. doi: 10.1021/jf991072g
61. Yoo KM, Lee CH, Lee H, Moon B, Lee CY. Relative antioxidant and cytoprotective activities of common herbs. *Food Chem*. (2008) 106:929–36. doi: 10.1016/j.foodchem.2007.07.006
62. Iwe M. *Handbook of Sensory Methods and Analysis*. Enugu: Re-Joint Communications Services Ltd. (2010). p. 75–78.
63. Reeves PG. Purified diets for laboratory rodents: final report of the American institute of nutrition ad hoc writing committee on the reformulation of the AIN-76a rodent diet. *J Nutr*. (1993) 123:1939–51. doi: 10.1093/jn/123.11.1939
64. Kumar S, Shachi K, Prasad N, Dubey N, Dubey U. Anti-diabetic, haematinic and anti-cholesterolemic effects of carrot (*Daucus carota* Linn) juice metabolites to cure alloxan monohydrate induced type-1 diabetes in albino rats. *J Diabetes Metab Disord Control*. (2020) 7:37–40. doi: 10.15406/jdmcd.2020.07.00197
65. Proll J, Petzke KJ, Ezeagu IE, Metges CC. Low nutritional quality of unconventional tropical crop seeds in rats. *J Nutr*. (1998) 128:2014–22. doi: 10.1093/jn/128.11.2014
66. Jelodar Gholamali A, Maleki M, Motadayen M, Sirus S. Effect of Fenugreek, onion and garlic on blood glucose and histopathology of pancreas of alloxan-induced diabetic rats. *Indian J Med Sci*. (2005) 59:64–9. doi: 10.4103/0019-5359.13905
67. Ravi K, Ramachandran B, Subramanian S. Protective effect of *Eugenia jambolana* seed kernel on tissue antioxidants in streptozotocin-induced diabetic rats. *Biol Pharm Bull*. (2004) 27:1212–7. doi: 10.1248/bpb.27.1212
68. Young D. *Effects of Disease on Clinical Lab. Tests*. 4th ed. Washington, DC: AACC (2001).
69. Stein EA, Myers GL. Lipids, lipoproteins and apolipoproteins. *Fund. Clin Chem*. (1987) 3:478–9.
70. Lopes-Virella MF, Stone P, Ellis S, Colwell JA. Cholesterol determination in high-density lipoproteins separated by three different methods. *Clin Chem*. (1977) 23:882–4. doi: 10.1093/clinchem/23.5.882
71. Friedewald WT, Levy RI, Fredrickson DS. Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. *Clin Chem*. (1972) 18:499–502. doi: 10.1093/clinchem/18.6.499
72. Trinder P. Glucose & triglycerides enzymatic colorimetric methods. *J Ann Clin Biochem*. (1969) 6:24–7. doi: 10.1177/000456326900600108
73. Reitman S, Frankel S. A colorimetric method for the determination of serum glutamic oxalacetic and glutamic pyruvic transaminases. *Am J Clin Pathol*. (1957) 28:56–63. doi: 10.1093/ajcp/28.1.56
74. Young DS, Friedman RB. *Effects of Disease on Clinical Laboratory Tests*. 4th ed. Washington, DC: AACC (2001).
75. Marsh WH, Fingerhut B, Miller H. Automated and manual direct methods for the determination of blood urea. *Clin Chem*. (1965) 11:624–7. doi: 10.1093/clinchem/11.6.624
76. Steel R, Torri J. *Principles and Procedures of Statistical Biometrical Approaches*. 2nd ed. New York, NY; London: McGraw-Hill Book Company (1980).
77. Kamble S, Gatade A, Sharma A, Sahoo A. Physico-chemical composition and mineral content of aloe vera (*Aloe barbadensis miller*) gel. *Int J Multidiscip Educ Res*. (2022) 11:73.
78. Kelebek H, Selli S. Determination of volatile, phenolic, organic acid and sugar components in a Turkish Cv. Dortyol (*Citrus sinensis* L Osbeck) orange juice. *J Sci Food Agric*. (2011) 91:1855–62. doi: 10.1002/jsfa.4396
79. Hashemi SMB, Khaneghah AM, Barba FJ, Nemati Z, Shokofte SS, Alizadeh F. Fermented sweet lemon juice (*Citrus limetta*) using *Lactobacillus plantarum* Ls5: chemical composition, antioxidant and antibacterial activities. *J Funct Foods*. (2017) 38:409–14. doi: 10.1016/j.jff.2017.09.040
80. Mehmood Z, Zeb A, Ayub M, Bibi N, Badshah A, Ihsanullah I. Effect of pasteurization and chemical preservatives on the quality and shelf stability of apple juice. *Am J Food Technol*. (2008) 3:147–53. doi: 10.3923/ajft.2008.147.153
81. Wisal S, Ullah J, Zeb A, Khan MZ. Effect of refrigeration temperature, sugar concentrations and different chemicals preservatives on the storage stability of strawberry juice. *Int J Eng Technol*. (2013) 13:160–8.
82. Pareek S, Paliwal R, Mukherjee S. Effect of juice extraction methods and processing temperature-time on juice quality of Nagpur Mandarin (*Citrus reticulata* Blanco) during storage. *J Food Sci Technol*. (2011) 48:197–203. doi: 10.1007/s13197-010-0154-6
83. Pokhrel PR, Boulet C, Yildiz S, Sablani S, Tang J, Barbosa-Cánovas GV. Effect of high hydrostatic pressure on microbial inactivation and quality changes in carrot-orange juice blends at varying Ph. *LWT*. (2022) 159:113219. doi: 10.1016/j.lwt.2022.113219
84. Yadav RB, Yadav BS, Kalia N. Development and storage studies on whey-based banana herbal (*Mentha arvensis*) beverage. *Am J Food Technol*. (2010) 5:121–9. doi: 10.3923/ajft.2010.121.129
85. Reddy AH, Chikkasubbanna V. Standardization of recipe and storage behaviour of lime blended amla squash. *Asian J Horticult*. (2008) 3:203–7.
86. Deshpande H, Katke S, Poshadri A. Influence of probiotics on physico-chemical and organoleptic characteristics of sweet orange juice. *J Environ Biol*. (2022) 43:170–6. doi: 10.22438/jeb/43/1/MRN-1940
87. Rodrigo D, Arranz J, Koch S, Frigola A, Rodrigo M, Esteve M, et al. Physicochemical characteristics and quality of refrigerated spanish orange-carrot juices and influence of storage conditions. *J Food Sci*. (2003) 68:2111–6. doi: 10.1111/j.1365-2621.2003.tb07028.x
88. Afzaal M, Saeed F, Hussain S, Mohamed AA, Alamri MS, Ahmad A, et al. Survival and storage stability of encapsulated probiotic under simulated digestion conditions and on dried apple snacks. *Food Sci Nutr*. (2020) 8:5392–401. doi: 10.1002/fsn3.1815
89. Shigematsu E, Dorta C, Rodrigues FJ, Cedran MF, Giannoni JA, Oshiiwa M, et al. Edible coating with probiotic as a quality factor for minimally processed carrots. *J Food Sci Technol*. (2018) 55:3712–20. doi: 10.1007/s13197-018-3301-0
90. Esteve M, Frigola A, Rodrigo C, Rodrigo D. Effect of storage period under variable conditions on the chemical and physical composition and colour of spanish refrigerated orange juices. *Food Chem Toxicol*. (2005) 43:1413–22. doi: 10.1016/j.fct.2005.03.016
91. Suriati L, Mangku IGP, Datri LK, Hidalgo HA, Red J, editors. Effect of aloe-gel and bignay fruit proportions to acidity, total dissolved solids, and color of aloe-bignay beverage. In: *WICSTH 2021: Proceedings of the 1st Warmadewa International Conference on Science, Technology and Humanity, WICSTH 2021, 7-8 September 2021, Denpasar, Bali, Indonesia*. European Alliance for Innovation (2022).
92. Islam S, Koly S. A review on phytochemical and pharmacological potentials of *Antidesma bunius*. *J Anal Pharm Res*. (2018) 7:602–4. doi: 10.15406/japlr.2018.07.00289
93. Genovese D, Elustondo M, Lozano J. Color and cloud stabilization in cloudy apple juice by steam heating during crushing. *J Food Sci*. (1997) 62:1171–5. doi: 10.1111/j.1365-2621.1997.tb12238.x
94. Saberian H, Hamidi-Esfahani Z, Abbasi S. Effect of pasteurization and storage on bioactive components of aloe vera gel. *Nutr Food Sci*. (2013) 43:175–83. doi: 10.1108/00346651311313553
95. Morales-De La Peña M, Salvia-Trujillo L, Rojas-Graü M, Martín-Belloso O. Changes on phenolic and carotenoid composition of high intensity pulsed electric field and thermally treated fruit juice-soymilk beverages during refrigerated storage. *Food Chem*. (2011) 129:982–90. doi: 10.1016/j.foodchem.2011.05.058
96. Klimczak I, Małecka M, Szlachta M, Gliszczynska-Swigło A. Effect of storage on the content of polyphenols, vitamin c and the antioxidant activity of orange juices. *J Food Comp Anal*. (2007) 20:313–22. doi: 10.1016/j.jfca.2006.02.012
97. Nejatizadeh-Barandozi F. Antibacterial activities and antioxidant capacity of aloe vera. *Org Med Chem Lett*. (2013) 3:1–8. doi: 10.1186/2191-2858-3-5
98. Szymanowska U, Baraniak B, Gawlik-Dzik U. Changes in the level and antioxidant activity of polyphenols during storage of enzymatically treated raspberry juices and syrups. *Acta Scientiarum Polonorum Technologia Alimentaria*. (2017) 16:269–82. doi: 10.17306/J.AFS.2017.0491
99. Gorski FI, Kausar T, Murtaza MA. 27. Evaluation of antibacterial and antioxidant activity of aloe vera (*Aloe barbadensis* Miller) gel powder using different solvents. *Pure Appl Biol*. (2019) 8:1265–70. doi: 10.19045/bspab.2019.80068

100. Peng W, Meng D, Yue T, Wang Z, Gao Z. Effect of the apple cultivar on cloudy apple juice fermented by a mixture of *Lactobacillus acidophilus*, *Lactobacillus plantarum*, and *Lactobacillus fermentum*. *Food Chem.* (2021) 340:127922. doi: 10.1016/j.foodchem.2020.127922
101. Quan Q, Liu W, Guo J, Ye M, Zhang J. Effect of six lactic acid bacteria strains on physicochemical characteristics, antioxidant activities and sensory properties of fermented orange juices. *Foods.* (2022) 11:1920. doi: 10.3390/foods11131920
102. Malik M, Bora J, Sharma V. growth studies of potentially probiotic lactic acid bacteria (*Lactobacillus plantarum*, *Lactobacillus acidophilus*, and *Lactobacillus casei*) in carrot and beetroot juice substrates. *J Food Process Preserv.* (2019) 43:e14214. doi: 10.1111/jfpp.14214
103. Bhardwaj RL, Pandey S. Juice blends—a way of utilization of under-utilized fruits, vegetables, and spices: a review. *Crit Rev Food Sci Nutr.* (2011) 51:563–70. doi: 10.1080/10408391003710654
104. Khleef AA, Saeed SK. Evaluation of the therapeutic efficacy of aloe vera juice on weight, liver and renal function in laboratory rats induced with diabetes. *Evaluation.* (2022) 140.
105. Tanaka M, Misawa E, Ito Y, Habara N, Nomaguchi K, Yamada M, et al. Identification of five phytosterols from aloe vera gel as anti-diabetic compounds. *Biol Pharm Bull.* (2006) 29:1418–22. doi: 10.1248/bpb.29.1418
106. Rajasekaran S, Sivagnanam K, Subramanian S. Modulatory effects of aloe vera leaf gel extract on oxidative stress in rats treated with streptozotocin. *J Pharm Pharmacol.* (2005) 57:241–6. doi: 10.1211/0022357055416
107. Fazlani T, Memon M-U-R, Baloch J, Soomro S, Soomro J, Khan A, et al. Effect of aloe vera and metformin on diabetic albino rats. *Pure Appl Biol.* (2020) 9:2122–7. doi: 10.19045/bspab.2020.90226
108. Almada-Érix CN, Almada CN, Cabral L, Barros de Medeiros VP, Roquette AR, Santos-Junior VA, et al. Orange juice and yogurt carrying probiotic bacillus coagulans Gbi-30 6086: impact of intake on wistar male rats health parameters and gut bacterial diversity. *Front Microbiol.* (2021) 12:623951. doi: 10.3389/fmicb.2021.623951
109. Ghadge AA, Kuvalekar AA. Controversy of oral hypoglycemic agents in type 2 diabetes mellitus: novel move towards combination therapies. *Diabetes Metab Syndr.* (2017) 11:S5–S13. doi: 10.1016/j.dsx.2016.08.009
110. Yang W, Lee JY, Nowotny M. Making and breaking nucleic acids: two-Mg²⁺-ion catalysis and substrate specificity. *Mol Cell.* (2006) 22:5–13. doi: 10.1016/j.molcel.2006.03.013
111. Payandeh J, Pfoh R, Pai EF. The structure and regulation of magnesium selective ion channels. *Biochim Biophys Acta Biomembranes.* (2013) 1828:2778–92. doi: 10.1016/j.bbamem.2013.08.002