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DISSERTATION

RESEARCH AND DEVELOPMENT OF TECHNOLOGY FOR THE PRODUCTION OF YOGHURT ENRICHED WITH CEREAL β-GLUCAN

Specialty 181 - Food Technology

Field of study –18 "Production and Technology"

Submitted for a scientific degree of Doctor of Philosophy. The dissertation contains the results of own research. The use of ideas, results and texts of other author have references to the relevant source Qu Xiaoqing **Qu Xiaoqing**

Scientific supervisor: Nazarenko Yuliya Valentynivna,
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ANNOTATION

Qu Xiaoqing Research and development of technology for the production of yoghurt enriched with cereal β-glucan – Quality scientific work as a manuscript.

Dissertation for the degree of Doctor of Philosophy in the specialty 181 - «Food technology» - Sumy National Agrarian University, Sumy, 2022.

The dissertation is devoted to the development of an improved industrial technology for the production of Yogurt with functional properties due to the addition of dietary fibers. An urgent issue is the creation of fermented milk products with functional properties that contain all the nutrients vital for the body and have increased storage ability without the addition of preservatives.

Yogurts contain all biologically active components necessary for the functioning of the body, except for ballast substances (dietary fibers). The introduction of dietary fibers into the composition of yogurts significantly increases their nutritional value and gives them functional properties. Dietary fibers are not absorbed by the body, but they play an important role in the digestion process, increasing intestinal peristalsis. It is known that dietary fibers affect the metabolism of cholesterol in the body, in particular, they stimulate the production and excretion of bile acids, which contain cholesterol. Products containing ballast substances should be included in the daily diet and can be used in therapeutic and preventive nutrition.

The application of the oat β-glucan is offered as a source of dietary fibers. The effect of added 0.3% oat β-glucan in yogurt on protein digestion was evaluated using an in vitro gastrointestinal model. It was found that during the digestion phase in the stomach, the amount of soluble proteins and peptides increases up to 25% for the control Yogurt without additives and up to 40% for the yogurt with β-glucan. The presence of 0.3% β-glucan promotes the hydrolysis of product proteins in the gastric digestion phase. Compared with control Yogurt samples (without additives), inhibition of cholesterol solubility of β-glucan Yogurt showed no differences after transbuccal digestion, but was significantly higher after gastrointestinal digestion (21.3% for gastric and 22.7% for intestinal digestion).

The research showed that oat β-glucan has water-holding capacity that allows it to maintain the product's optimal viscosity, texture and acidity during the entire storage period (21 days).

It was established that the addition of β-glucan has a positive effect on the content of live probiotic bacteria in yogurt during 21 days of storage. Probiotics help stimulate gastric juices and natural enzymes necessary for proper digestion, reduce the number and severity of side effects of antibiotics, promote the breakdown of bile acid salts and normalize lipid metabolism. The presence of probiotic microorganisms in yogurt during the entire storage period gives it functional properties.

Adding of oat β-glucan also improves the organoleptic and physicochemical properties of the product. The addition of oat β-glucans to Yogurt showed that the physical and textural properties of Yogurt can be significantly modified by the phase separation between β-glucan and milk proteins.

Therefore, the purpose of the dissertation is the development and scientific justification of the technology of a high-quality fermented milk drink - Yogurt with the addition of oat β-glucan as a source of dietary fiber. The effect of the additive on the organoleptic, physicochemical, and rheological properties of the product was investigated in the dissertation. Also its digestibility and storage ability was studied. The therapeutic and preventive properties of the product have been confirmed by the method of medical and biological research.

An improved industrial technology for the production of Yogurts with oat β-glucan has been developed by reducing the duration of fermentation by 16 minutes.

The introduction substantiates the relevance of the topic of the dissertation, reflects its connection with scientific programs and topics, defines and substantiates the purpose and tasks of the research, object, subject and main methods, indicates the novelty and practical significance of the obtained work results, determines the personal contribution of the candidate in the conducted research, approbation of the results, and publications on the topic of the dissertation, the structure and scope of the work are given.

The first chapter analyzes and summarizes the data of the scientific and technical literature on the effect of oat β-glucan on the properties of fermented milk drinks, sources

of dietary fiber acceptable for use in the production of food products. It is shown that oat β-glucan is a promising raw material for the production of yogurts with functional properties.

In the second section, a general program of scientific research is developed, in which the main directions of the dissertation work are reflected, the materials and the subject of research are given. The section describes the methods and indicates the equipment that was used in conducting of experimental research. The experimental part of the work was carried out in laboratory conditions at the Department of Technology and Food Safety of the Sumy National Agrarian University and in the laboratories of the School of Food Sciences of the Henan Institute of Science and Technology (China). The industrial approval of the developed product was carried out on the basis of the BRANCH "Sumy Dairy Plant".

The standard and generally accepted research methods were used in the study of physico-chemical, microbiological, structural-mechanical and organoleptic indicators, microstructure and nutritional and biological value. The results of the experiments were processed using the methods of mathematical statistics.

In the third section, the effect of adding oat β-glucan on the quality of Yogurt, its viscosity, sensory indicators and physicochemical properties is investigated. Oat β-glucan was added to Yogurt of the set-type as a functional ingredient. It was found that the optimal amount to add is 0.3% of oat β-glucan. The introduction of the additive contributes to the improvement of the technology by reducing the fermentation time of yogurt by 16 minutes.

The acidity of set-type Yogurt with oat β-glucan was slightly higher. The addition of OG has a slight effect on chromaticity. The result of microstructure showed that the addition of oat β-glucan destroyed the three-dimensional network structure of Yogurt, and some spherical aggregate particles could be clearly observed at 0.3% OG.

The addition of oat β-glucan increased the total number of lactic acid bacteria in the coagulated yogurt, and had a little effect on the fatty acid and amino acid composition of set-type yogurt. SPME-GC/MS analysis confirmed that the addition of oat β-glucan was helpful to increase the types of volatile flavor compounds in the set-type Yogurt.

The storage experiment results showed that 0.3% oat β-glucan Yogurt has better water-holding capacity. The acidity value and pH reached their maximum values at 7 d of storage, and no significant (p<0.05) changes were observed after 7 d. Interestingly, the viscosity values increased throughout storage. The addition of 0.3% oat β-glucan has a protective effect on probiotic microflora in Yogurt, which confirms functional properties of the developed product. The textural characteristics of Yogurt were affected by the addition of 0.3% oat β-glucan, leading to decreased adhesiveness, but enhanced hardness and gumminess, throughout storage. The sensory results indicated that 0.3% oat β-glucan yogurt had the highest acceptability value of 86.49 at 14 d of storage.

The fourth section studied the effect of OG on in vitro digestion characteristics of set-type yogurt ,the antioxidant activity of set-type yogurt during digestion , and the effects of OG set-type Yogurt on blood lipid metabolism in mice with a high fat diet .

In the simulated in vitro digestion experiment, the effects of added 0.3% oat β-glucan in Yogurts on Yogurt protein digestion were evaluated using an in vitro gastrointestinal model. The amount of soluble proteins and peptides increased throughout digestion. Large spherical vesicles were formed for both control Yogurt and 0.3% oat β-glucan Yogurt after gastric digestion. 0.3% oat β-glucan Yogurt after digestion had higher antioxidant activity and higher inhibition of cholesterol solubility. 0.3% oat β- glucan Yogurt after digestion had higher antioxidant activity and higher inhibition of cholesterol solubility.

Through DPPH -1,1-diphenyl-2-picrylhydrazyl radical and ABTS 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid assays, the antioxidant activity of yogurt was mainly produced during the gastric digestion phase, and 0.3% oat β- glucan could further improve the antioxidant activity of yogurt by promoting the beneficial enzymatic hydrolysis. And, compared with DPPH assay, ABTS assay is more suitable for evaluating the antioxidant activity of set-type Yogurt during digestion.

In biomedical experiments, the effect of oat β- glucan set-type Yogurt on blood lipid metabolism in mice with high-fat diet was mainly determined Lee's index, blood lipids TG, TC, HDL, LDL, AST, ALT, liver weight and liver sections were studied. The experiment of effects of oat β-glucan set-type yogurt on blood lipid metabolism in mice

with high fat diet provided evidence that 0.3% oat β- glucan set-type Yogurt can be an alternative and a complementary treatment to therapy of obesity-related complications.

The fifth section organized the recipe, nutritional value of the product and the technological scheme for the production of Yogurt with addition of oat β- glucan. The production process of OG set-type yogurt is described, the energy loss is calculated, and the energy balance in the production process is carried out. The loss rate of materials is calculated by taking the production of 20 t oat β- glucan set-type Yogurt per day as an example. According to statistics and evaluation of the pilot production, the social and economic benefits of scientific and technological development and the implementation achievements in actual production are listed.

The addition of oat β-glucan can effectively enhance the functional characteristics of fermented yogurt. Yogurt fortified 0.3% oat β-glucan could be an innovative healthy dairy for enhancing oat β- glucan consumption. OG set-type Yogurt products will bring more health benefits to consumers. Long-term and appropriate consumption will help consumers' health, reduce medical and health expenditure, and thus bring profound social and economic benefits.

Key words: Yogurt; fermented drinks; fermented milk products; vegetable raw materials; oat β-glucan; functional product; therapeutic and preventive purpose; proteins; digestion in vitro; antioxidant activity; solubility of cholesterol; rheological characteristics; viscosity; medical nutrition.

LIST OF THE APPLICANT'S PUBLICATIONS

SCOPUS / Web of Science publications

1. Qu X, Nazarenko Y, Yang W, Nie Y, Zhang Y, Li B. (2021). Effect of Oat β-Glucan on the Rheological Characteristics and Microstructure of Set-Type Yogurt. *Molecules*, 26(16): 4752. https://doi.org/10.3390/molecules26164752. (*The applicant participated in research, analysis of the results and writing the article*).
2. Qu X, Li B, Yang W, Nazarenko Y. (2022). Effect of oat β-glucan on in vitro digestion characteristics of set- type yogurt. *Acta Innovations*, №43: 5-14. https://doi.org/10.32933/ActaInnovations.43.1. (*The applicant participated in research, analysis of the results and writing the article*).
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4. Qu Xiaoqing, Y. Nazarenko, Li Bo. (2019). Promising areas of use of soybeanpeptide in the production of yogurt. *Special collections of Kharkiv State University of Food Technology and Trade*, 55–63. DOI: 10.5281/zenodo.3263496. (*The applicant participated in research, analysis of the results and writing the article*).
5. Qu Xiaoqing, Y. Nazarenko, Li Bo. (2021). The use of oat beta-glucans in the dairy industry: an overview and assessment of opportunities in yogurt technology. *Научный взгляд в будуще*, 1(19-01), 108–113. https://doi.org/10.30888/2415-7538.2020-19-01-013. (*The applicant participated in research, analysis of the results and writing the article*).
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характеристики йогурту, збагаченого вівсяним β-глюканом. *Науковий журнал «Тваринництво та технології харчових продуктів»,* 12(3), 96-104. http://dx.doi.org/10.31548/animal2021.03.009. (*The applicant participated in research, analysis of the results and writing the article*).

7. Xiaoqing Qu, Nazarenko Ju., Bo Li (2021). The physiological function and application of cereal β-glucan in the fermented dairy. *Прогресивні техніка та технології харчових виробництв ресторанного господарства і торгівлі*, Вип. 1(33). С. 62-76. DOI: 10.5281/zenodo.5036077. (*The applicant participated in research, analysis of the results and writing the article*).

Conference papers

8. Xiaoqing Qu, Yuliya Nazarenko, Bo Li. (2021). Physicochemical properties of cereal β-glucan and its application in food industry.information and innovative technologies in hotel and restaurant business, tourism and design. Collection of abstracts of reports of the II International Scientific and Practical Internet Conference, 6 (December 1-2, 2021, DNIPRO–OPOLE).
9. Xiaoqing Qu, Yuliya Nazarenko, Bo Li. (2021). Application of protein-polysaccharide compound system in functional yogurt. Information and innovative technologies in hotel and restaurant business, tourism and design. Collection of abstracts of reports of the II International Scientific and Practical Internet Conference, 8-9 (December 1-2, 2021, DNIPRO–OPOLE). (*PhD participant in carrying out of experimental research, processing of results, and writing the article*).
10. QU Xiaoqing, Yuliya Nazarenko, LI Bo. (2021). Characteristics of Prebiotics of Cereal B-Glucan and Its Application in Fermented Milk. The 9th China diary science and technology conference. Collection of abstracts of reports, 187 (July 22-24, 2021, Hohhot city, Inner Mongolia) (*PhD participant in carrying out of experimental research, processing of results, and writing the article*).
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22-24, 2021, Hohhot city, Inner Mongolia) (*PhD participant in carrying out of experimental research, processing of results, and writing the article*).

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TABLE OF CONTENTS

THE LIST OF SYMBOLS

OG -Oat β-glucan

GI -Glycemic index

CHD -Coronary heart disease

WHC -Water-holding capacity

DWS -Diffusing wave spectroscopy method

MVI -Macroscopic viscosity index

EI -Elasticity index

SLB -Solid–liquid balance

SEM -Scanning electron microscope

TPA -Texture profile analysis

PDI -Polydispersity

ABTS -2, 2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid

DPPH -1,1-Diphenyl-2-picrylhydrazyl radical

ALT -Alanine aminotransferase

AST - Aspartate aminotransferase

TG -Triglyceride

TC - Total cholesterol

HDL -High-density lipoprotein cholesterol

LDL -Low-density lipoprotein cholesterol

SPME-GC/MS -Solid Phase Micro-extraction- Gas Chromatography/Mass Spectrometry

INTRODUCTION

The number of patients suffering from chronic diseases, such as hypertension, diabetes, dyslipidemia and overweight/obesity caused by unhealthy lifestyle is increasing worldwide. It is known that nutrition takes an important role in the forming of human's health. Because of this more and more developers and food producers work at creating food products with certain functional and therapeutic and preventive properties.

Eating food rich in fiber is one of the important methods of dietary treatment. Cereal β-glucan has a variety of physiological effects which can effectively prevent metabolic syndrome, type II diabetes and other chronic diseases.

Relevance of the topic. A topical issue is the creation of functional fermented milk products, which are the product of everyday consumption and contain all nutrients vital for the body and have an increased storage capacity without adding of preservatives.

Fermented milk drinks, including yogurts, contain all bioactive components, except for ballast substances (dietary fibers) necessary for functioning of the human's body. The introduction of dietary fibers into the composition of Yogurts significantly increases their nutritional value and gives them functional properties. The dietary fibers are not absorbed by the body, but they play an important role in the digestion process, increasing intestinal peristalsis. It is known that dietary fibers affect the metabolism of cholesterol in the body, in particular, they stimulate the production and excretion of bile acids, which contain cholesterol. The products containing ballast substances should be included in the daily diet and can be used in therapeutic and preventive nutrition.

The use of oat β-glucan, which has a lot of useful functional properties that have been proved by many researchers, is offered as a source of dietary fiber. However, there are no experimental studies on the use of powdered oat β-glucan in the production of yogurts. The impact of this additive on organoleptic, physicochemical, microbiological and other indicators of the product quality, as well as changes in the traditional technological process of Yogurt production, should be investigated.

Connection of work with scientific programs, plans, themes. The dissertation was developed within the research work theme plan of Sumy National Agricultural

University of Ukraine, on the subject of state budget research of the Department of Technology and Food Safety 0119U101237 "Innovative technological solutions in the production of food products".

The purpose and objectives of the work. *The purpose of the dissertation* is the development and scientific justification of the technology of a high-quality fermented milk –Yogurt with the addition of oat β-glucan as a source of dietary fiber.

To achieve this, the main tasks were formulated:

-to justify the choice of oat β-glucan as a physiologically active additive for effective use in the technological process of Yogurt production;

- to investigate the effect of adding oat β-glucan on the chemical composition of the finished product, and based on these studies to calculate the optimal formulation of Yogurt with the addition of oat β-glucan;

- to investigate the effect of oat β-glucan on the process of homogenization and fermentation of Yogurt, to determine its organoleptic, physicochemical, and rheological properties;

- to evaluate the nutritional value of the finished product and develop a technological scheme for its production in industrial conditions;

- to investigate the conformity of Yogurts with oat β-glucan to the requirements of existing standards in Ukraine and China according to the main indicators (organoleptic, physicochemical, microbiological);

- to evaluate the digestion and assimilation of Yogurt protein in vitro by simulating the physiological characteristics of animal digestion and adapting the digestion environment and the system of digestive enzymes similar to those found in the animal body;

- evaluate the functional properties of the product;

- calculate the energy efficiency of yogurt production with oat β-glucan;

- to carry out industrial testing of the developed technology of an innovative fermented milk –Yogurt with the addition of oat β-glucan.

The research object – technology for production of fermented milk- set-type Yogurt with adding OG.

The research subject – whole milk; cream; skimmed milk; oat β-glucan; model samples of Yogurts with and without additives; homogenized normalized mixture with the addition of oat β-glucan; bacterial starters of direct application; fermented Yogurt with the addition of oat β-glucan.

Research methods include a complex of traditional and modern methods of sensory assessment, physics, chemistry, microbiology, simulated in vitro digestion, biomedical experiments, experimental planning, and experimental data processing.

Scientific novelty of the obtained results. The expediency of using oat β-glucan in the production of Yogurts by the tank method has been scientifically substantiated and experimentally proven.

The results of experimental studies established a positive effect of oat β-glucan on the fermentation process in the production of Yogurt. An increase in the number of probiotic microflora and a reduction in the duration of the fermentation process have been established. It was shown that the addition of oat β-glucan leads to an increase in viscosity and a decrease in adhesiveness, while the hardness and stickiness of the structure of the finished product is preserved throughout the entire storage period. Sensory results showed that yogurt with the addition of 0.3% oat β-glucan had the highest acceptability value.

The effect of oat β-glucan on the in vitro digestion characteristics of Yogurt and its effect on blood lipid metabolism in mice with a high-fat diet has been proven. It was established that the amount of soluble proteins and peptides increased during digestion. The therapeutic and preventive properties of the product have been confirmed by the method of medical and biological research, which makes it possible to recommend Yogurt with the addition of oat β-glucan for diabetic nutrition.

A yogurt recipe and industrial technology for the production of Yogurts with oat β-glucan have been developed. A positive effect of the addition of oat β-glucan on the viscosity of the finished product and the stability of the consistency during a long period of storage was established, without the addition of additional thickening ingredients.

The cost of the product was calculated, and its economic efficiency was proved. According to statistical data and evaluation of research and industrial production, socio-

economic advantages of scientific and technical development and introduction of development into real production are listed.

The practical significance of the results obtained. On the basis of experimental research, industrial technology for production of OG Yogurt was improved. The technology was tested under real production conditions at the Sumy Dairy Plant branch of the Aromat SE and at the enterprise in China (Tianjin Huaming Dairy Co., Ltd. and You Dian Ai (Chuzhou) Health Technology Co., Ltd.).

When the developed technologies are implemented in the industry, additional profit is obtained.

Personal contribution of the applicant includes: analysis of the state of the problem, development of a research program, organization, execution and generalization of analytical and experimental studies, analysis and generalization of the obtained data in the form of conclusions, preparation of research materials for publications, preparation of a dissertation, conducting industrial testing.

The dissertation work was carried out with the methodological and scientific support of Ph.D., Assoc. Yu.V. Nazarenko.

The personal contribution of the dissertation student is documented by scientific works.

Approbation of dissertation results. The dissertation research results are introduced at: the 11th International Student and Graduate Scientific Conference on Food Production Equipment and Technology (Belarus, Mogilev, April 18-19, 2019); the II International Scientific and Practical Internet Conference on (DNIPRO-OPOLE, December 1-2,2021); The 9th China diary science and technology conference, Collection of abstracts of reports (Hohhot city, Inner Mongolia, July 22-24, 2021).

Publications. Published 12 scientific papers, of which 4 were published in Ukrainian professional journals and scientific proceedings, 3 were published in the foreign publications included to the international scientometric databases and 5 theses of reports at scientific, scientific-practical and international conferences.

Structure and scope of the dissertation. The dissertation consists of an introduction, 5 sections, conclusions, a list of used literary sources and appendices. The

main content of the work is represented on 148 pages, including 31 tables, 44 figures, appendices (per page). The list of used bibliographic sources contains 214 names (on 20 pages).

SECTION 1 RESEARCH OF PROPERTIES AND DIRECTIONS OF APPLICATION OF CEREAL β-GLUCAN

1.1 Influence of the diet on rate of diseases

With the rapid development of the modern food industry, the proportion of health food in people's diet is increasing sharply. Meanwhile, due to the accelerated pace of life, and people's physical exercise is reduced, which increases the risk of people suffering from chronic diseases, such as hypertension, diabetes, dyslipidemia and overweight/obesity. Nutrition intervention can prevent and control these chronic diseases. Among them, adding dietary fiber with multiple health functions is one of the important ways of nutritional intervention. The incidence rate of diseases caused by unhealthy lifestyles is increasing all over the world. The WHO reported that the global proportion of overweight adults has reached 1.8 billion, of which more than 600 million adults were affected by obesity.

Many diseases, such as coronary heart disease (CHD), stroke, insulin resistance, type II diabetes, hypertension and metabolic syndrome, are associated with overweight [1]. From 1980 to 2014, the number of diabetes cases increased fourfold in the world (from 108 million to 422 million). It is estimated that by 2045, there will be as many as 700 million adults with diabetes, and 90% of them will be patients with type II diabetes [2; 3]. Nutrition therapy combined with physical exercise plays an extremely key role in the prevention and treatment of lifestyle diseases.

Among them, the intake of food rich in fiber is one of the important nutritional therapies [4]. Cereal β-glucan has a variety of physiological effects, which can effectively prevent metabolic syndrome, type II diabetes and other chronic diseases [5; 6], and has attracted more and more attention of researchers in various countries.

1.2 Research on the peculiarities of the development of functional fermented milk products

Yogurt is a traditional food loved by people all over the world and is part of people's diet.

The functional Yogurt industry was born as consumers increasingly demand healthy food [7]. Compared with traditional yogurt, functional Yogurt is a type of dairy product that adds functional active substances to the traditional yogurt system to give yogurt more functions. Currently, functional factors that can be used to yogurt include minerals (calcium, iron) [8], vitamins [9], omega-3 fatty acids, conjugated linoleic acid, phytosterols [10], etc.

Some other functional factors, such as biologically active plant extracts: tea polyphenols [11], curcumin [12], carotenoids [13], anthocyanin [14; 15; 16], etc., can also be added to yogurt. However, these functional factors are often prone to oxidation or difficult to dissolve. How to add functional factors into yogurt and maintain their biological activity are an urgent problem to be solved in the dairy industry. At the same time, due to differences in structure and properties, functional factors, when they exist in the yogurt system, may lead to changes in the physic and chemical and rheological properties.

At present, due to the frequent occurrence of lifestyle diseases, adding polysaccharides to yogurt can not only enhance the health effects of yogurt, but also increase the dietary fiber in the dietary structure.

1.3 Application of dietary fibers during production of Yogurts

Yogurt is the most popular dairy product in the world [17]. Yogurt is rich in essential amino acids, vitamins and minerals, but does not contain fiber. At present, when dietary fiber is added to Yogurt as a functional component and nutritional food, it can achieve a variety of functions. When adding grape pomace as antioxidant dietary fiber to Yogurt and salad dressing, nutritional value and storage resistance are improved [18].

The study of the effect of adding passion fruit fiber to probiotic Yogurt showed that its rheological properties, microstructure, and sensory characteristics improved [19]. The results showed that the apparent viscosity of yogurt made by Lactobacillus fermentation increased. The micrograph of freeze dried yogurt was produced by SEM. Compared with the control sample, it is denser [19]. The effects of pumpkin fiber (PF) on the

physicochemical, microbial, rheological and microstructure of low-fat yogurt samples during storage were studied [20].

The results showed that the Yogurt with PF had lower synthesis rate and higher viscosity structure than the control. SEM images showed that more filamentous structures and dense reticular structures were observed in the samples added PF, and these structures increased with the increase of PF level. The results showed that PF improved the physical quality and texture properties of half fat yogurt [20].

It is generally believed that active intake of dietary fiber has a positive impact on the overall state of the human body. The application of dietary fiber in a variety of foods has attracted the attention of food professionals. Previous studies have shown that dietary fiber can change the basic quality parameters of yogurt, such as physicochemical, texture, microorganism, nutrition, functional and sensory properties. As these changes have both advantages and disadvantages, it is necessary to select the appropriate fiber at a precise level. It is a hot topic to select the appropriate kind and amount of dietary fiber added to Yogurt. The β-glucans has been widely concerned because of its structure and functional properties. Barley and oats, especially OG, have attracted much attention due to their high content of soluble non starch polysaccharides (fibrous substances).

1.4 Structural characteristics and sources of β-glucan

Glucan is a kind of carbohydrate polymerized from D-glucopyranose monomer [21]. According to the different glycosidic bond, it can be divided into α-glucan and β-glucan, which is an important part of cereal organisms. β-glucan is a kind of water-soluble / water-insoluble non starch homopolysaccharide, which can be linked by β-1,3, β-1,4, β-1,6 glycosidic bonds, and its structure is branched or circular. β-glucan widely exists in plants, fungi and bacteria, especially in cereals and fungi [22].

The cereal sources of β-glucan are barley [23], oat, wheat, etc., with different contents, such as barley 2.5%-11.3%, 2.2%-7.8%, wheat 0.2%-1.2% [23]. The structure of cereal β-glucan is generally composed of β-1,3 and β-1,4 glycosidic bonds without branched chain structure, as shown in Fig. 1.1 [22]. Barley, oat and wheat have similar structure of β-glucan, which are composed of β-1, 3 and β-1,4 glycosidic bonds, but the

chain length and the proportion of β- β-1,4 glycosidic bond and β-1,3 glycosidic bond are different.

In addition, the ratio of cellotriose (DP3) to cellotriose (DP4) determines the diversity of cereal β-glucan, and the molar ratio of the two is known as the structural fingerprint of cereal β-glucan, which is an important structural feature of cereal β-glucan [24]. In edible fungi, yeast and bacteria, β-glucan is an important component of cell wall.

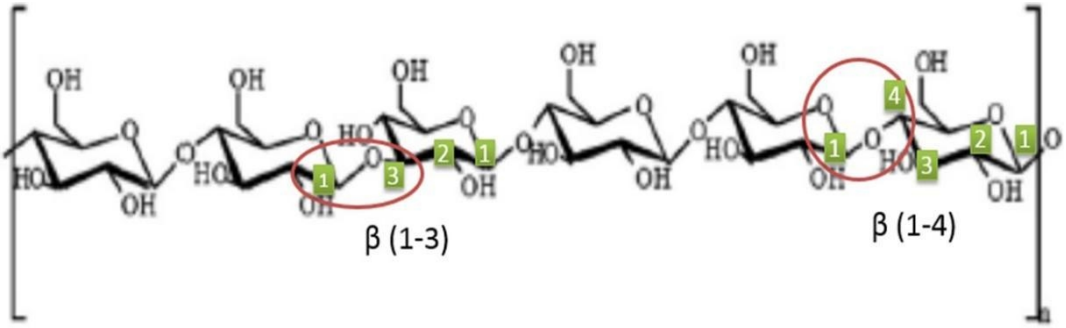


Fig. 1.1 Chemical structure of β-glucan from cereals.

Source: Adapted from Zhu, Du, & Xu [22].

It is mainly composed of β-1,3 glycosidic bond and β-1,6 glycosidic bond alone or / and mixed, and most of them have branched chain structure. At present, the main sources of edible fungi studied are *Pleurotus ostreatus* [25], *Coprinus comatus* and *Agaricus bisporus* [26], the main source of fungi is yeast [27], and the main source of bacteria is *Lactobacillus brevis in* beer [28].

1.5 β-glucan effect on human body

1.5.1 Regulating blood sugar

Because of its strong hydration ability and high viscosity, β-glucan can delay intestinal absorption of carbohydrate and reduce exogenous blood glucose [29]. β-glucan can protect and improve the functional characteristics of islet β cells, so that insulin can maintain stable secretion and achieve the purpose of regulating blood glucose; in addition, β-glucan can improve glucose transport of L6 muscle cells, It significantly reduced the blood glucose of diabetic rats [30, 31]. OG can significantly reduce glycemic index (GI). It has been considered that OG can significantly reduce the GI value of food [32; 33]. Kim and others reported that with the increase of OG content, the GI value of food showed a downward trend; after adding β-glucan, the GI value of starch showed a downward trend with the increase of β-glucan molecular weight and viscosity [34].

OG can effectively reduce the body's fasting blood glucose [35], improve the body's postprandial blood glucose. Previous studies have shown that bedtime snacks containing 1.8g OG can significantly reduce postprandial blood glucose in children with type II diabetes [36]. Dietary supplementation of 6% OG can reduce postprandial blood glucose of experimental pigs [37].

1.5.2 Regulating cholesterol

β-glucan can reduce the levels of total cholesterol and low-density lipoprotein cholesterol in blood, increase the level of free fatty acids in blood, regulate lipid metabolism disorder, and restore the level of triglyceride in blood to normal [38]. Wolever et al. found that OG with high, medium and low molecular weight has different ability to reduce serum low-density lipoprotein cholesterol, but the treatment effect was not affected by age and gender [39]. The research of some scientists showed that β-glucan from different sources can reduce cholesterol and blood glucose [40; 41].

1.5.3 Regulating immunity

β-glucan has the function of regulating immunity [42; 43] and can stimulate antibacterial activity. β-glucan can regulate the expression level of genes related to immune cells, and has the effect of anti-tumor [44] and anti-cancer [45] .

1.5.4 Other physiological functions

β-glucan can reduce fat obesity [46], enhance gastrointestinal peristalsis [47], maintain intestinal health [48], anti-inflammatory [49], and effectively prevent metabolic syndrome and other chronic diseases [50]. In addition, β-glucan can relieve stress [51], relieve pain[52], and resist oxidation.

β-glucan has been certified as health food by the food and Drug Administration (FDA) and the European Food Safety Authority (EFSA). It is recommended to ingest at least 0.75g β-glucan per day, and 3g β-glucan can achieve health function (EFSA).

1.6 Physicochemical properties of β-glucan and its application in food industry

1.6.1 Physicochemical properties of β-glucan

The physicochemical properties of β-glucan mainly include hydration (water holding and swelling), rheological properties, foaming properties and emulsifying properties. Rheological properties include viscosity, gelation, shear thinning, and viscoelasticity, and so on. The difference of their properties is mainly related to the structure and concentration of β-glucan.

Compared with the cereal derived β-glucan, microbial derived β-glucan has stronger swelling ability, but lower hydration ability, foaming ability, foaming stability and fat binding ability. For example, the high viscosity and poor water solubility of β-glucan in yeast limit its application in industry [53]. By dissolving in water, cereal β-glucan can form a highly viscous solution in the gastrointestinal tract [34]. OG has good hydration capacity, with swelling capacity of 12.1~15.1 g / g [54;55]. High viscosity barley β-glucan has water holding capacity of about 2.9 g / g [56]. It has been reported that cereal β-glucan has higher bioactivity due to its better water solubility [57].

The viscosity of β-glucan solution is affected by its source, molecular weight, temperature, concentration and solvent. The structure of β-glucan is the most important factor. Mikkelsen et al. found that the viscosity of OG was 100 times higher than that of barley β-glucan at the same concentration [58]. This is mainly because its molecular chain contains a high proportion of β - 1, 4 / β - 1, 3 and DP4 / DP3 [59]. Agbenorhevi et al. studied the rheological properties and microstructure of OG with different molecular weight, and considered that the rheological behavior highly depended on the molecular weight and the concentration of β-glucan [60]. The higher the molecular weight of OG, the higher the viscosity of OG. The concentration is also an important factor affecting the viscosity of OG solution. OG solution can produce higher viscosity at lower concentration.

When the concentration is above 2g / L, the apparent viscosity decreases with the increase of shear rate, which shows the typical characteristics of pseudoplastic fluid [61]. According to the research of Autio, Myllymaki et al. [62], OG showed better rheological properties when the concentration was lower than 1%. The gelation of cereal β-glucan is one of its important processing characteristics [63]. It plays a decisive role in the texture of food. Its texture characteristics can be controlled by molecular weight fraction or

concentration [33]. In practical application, it is necessary to select appropriate molecular weight and concentration of β-glucan to meet the industrial requirements.

1.6.2 Application of β-glucan in food industry

β-glucan products on the markets of various countries are mainly extracted from cereal, yeast and other raw materials. In the actual industrial processing and application, considering the availability of β-glucan sources and the differences in its processing characteristics and physiological functions, cereal β-glucan, especially OG, has important application value, which has attracted the close attention of researchers all over the world [64].

The global market value of β-glucan in 2016 was US $307.8 million, according to the survey data of MRI. Market and market forecast: by 2022, the global β-glucan market will reach 476.5 million US dollars, showing great development potential [65]. β-glucan has become a hot spot in food science and life science due to its availability of raw materials, processing characteristics and various physiological functions. As an important functional food ingredient, it is widely used in biscuits, bread [66], cake [67], meat [68], dairy [69; 70] and other foods. In addition to affecting the sensory properties of food, cereal β-glucan can interact with fat, protein, starch and other substances in food through its water holding, oil holding and emulsifying properties.

1.7 Structural and functional properties of OG

OG, cell wall polysaccharides of cereal grains, is recognized worldwide as functional bioactive ingredients. Health benefits of oat-based food products are attributed to the OG. It is a linear homopolysaccharide of consecutively linked (1 /4)-β-D-glucosyl residues in oligomeric segments that are separated by single (1/3)-bonds along the polymer chain backbone; i.e., its structure consists mostly of cellotriosyl and cellotetraosyl oligomers interlinked via β-(1/3) glycosidic linkages. Furthermore, OG has all the health benefits of soluble dietary fibers, including the reduction of blood serum cholesterol and the regulation of post-prandial blood glucose levels.

Both the US Food and Drug Administration (US FDA) and the European Food Safety Authority (EFSA) have authorized health claims, according to which OG

consumption leads to the reduction of blood plasma cholesterol concentrations and heart disease risk with a daily consumption of 3g of β-glucan (a minimum of 0.75 g per serving) originating from oat-formulated products that include oat bran, rolled oats and whole oat flour. Fortification of foods with OG is of great interest. Many foods, such as pasta, tea cakes, muffins, bread, and beverages have been fortified with OG [71; 72; 73; 74].

The potential use of the neutral polysaccharide in food products has been proposed based on its rheological characteristics that can provide desirable textural properties to food products, such as viscosity enhancement and gelling properties. The rheological characteristics appear to be related to the concentration, molecular weights, and structural features of these indigestible polysaccharides [75].

1.8 Application of protein-polysaccharide compound system in functional Yogurt

Proteins and polysaccharides are two important natural functional food materials. Their composite systems are widely used in the fields of food, medicine and cosmetics to improve product texture and enhance system stability. At the same time, protein-polysaccharide complexes is an important type of delivery system, which can enhance the solubility of many functional active ingredients, improve food processing factors and external environments, such as heat, irradiation, shearing, etc., to interfere with the activity of functional factors and destruction, maximizing their bioavailability. Therefore, it can be used as a delivery system for the development of functional Yogurt.

The application of protein-polysaccharide complex system in functional Yogurt includes the following aspects:

1. Enhance the taste and texture of Yogurt**.** The protein-polysaccharide complex system can change the taste and texture characteristics of food; taking water-soluble OG as an example, OG has a fat substitute and viscosity-increasing effect in yogurt, which can improve the quality of nonfat yogurt texture and mouthfeel [76].
2. Help improve the quality of Yogurt**.**

Certain polysaccharides are also prebiotics. Adding to Yogurt can help probiotics proliferate and increase vitality during yogurt fermentation, as well as maintain the number of probiotics during storage [77].

3. Extend the shelf life of Yogurt. The protein-polysaccharide complexation can change the functional properties of protein, such as stability, water retention and many other properties, and help extend the shelf life of Yogurt [78].

4. As a function delivery system. Protein-polysaccharide complex system can be used as a carrier protection system. When carrying and protecting active ingredients such as phenols, carotenoids, vitamins, fatty acids, etc. [79], it can not only improve their solubility, but also resist adverse external environmental effects during storage and processing.

The research of protein-polysaccharide complex system has been a research hotspot in recent years, and it has great application potential in the solubilization, protection and improvement of bioavailability of nutrients. Further development of the protein-polysaccharide complex system that can be used in the yogurt system can provide a new solution for the development of "clean label" functional yogurt, and provide new ideas for adding yogurt with embedded functional bioactive ingredients.

1.9 Application of OG during production of Yogurts

Yogurts are particularly popular among the consumers. Yogurt is a high protein healthy food, its consumption is a sign of healthy diet and lifestyle [7]. Many researchers tried to add oat and barley β-glucan to the fermented dairy products.

Oat and barley β-glucan are mainly used as polysaccharide/dietary fiber to influence the texture of Yogurt, and have the prebiotic properties. There has been an increasing interest in fortification of yogurt with different kinds of ingredients to further improve added nutritional benefits to health. Several studies have been reported on the influence of some ingredients like proteins, polysaccharides and metal ions [80; 81].

Among these functional foods, much attention is paid especially to the probiotic products and food containing OG. On the other hand, incorporation of cereal β-glucans into milk or milk protein derived gels [82], through acidification with glucono-d-lactone

(GDL) or by bacteria fermentation, showed that their physical and textural properties can be largely modified due to phase separation between β-glucans and milk proteins [83]. The changes in physical and textural properties seem to be governed by the concentration and the structural features of the added polysaccharide. On the other hand, gut-health benefits associated with probiotics can be complemented by the heart-health benefits of β-glucan in a yogurt system to provide a doubly healthful product.

1.9.1 Effect of cereal β-glucan on texture and structure of Yogurt

Current studies have proved that in an appropriate range, cereal β-glucan can improve the texture characteristics of yogurt, such as affecting the network structure of protein in Yogurt, reducing the separation of whey, improving the water holding capacity and viscosity value of yogurt, etc. The specific addition amount and application methods are shown in Table 1.1. by means of comparison the results obtained by different researchers.

Table 1.1 Application of cereal β-glucan in fermented dairy products

Source	Brand, concentration	Amount	Application Method	Technological function	Main results	References
1	2	3	4	5	6	7
Barley β-glucan	Differing in molecular size 40×10^3 Da-250×10^3 Da	0.5%–2.0%	Different concentrations of fresh β - glucan solution were prepared, and different levels of skimmed milk were added. After sterilization, the dispersion was acidified at 42 ºC for 18 h.	High concentration of β-glucan reduces serum separation of milk gel	Using high b-glucan concentration (1.5% and 2.0%, w/w) in milk gels containing low skim milk levels (up to 12%, w/w) or b-glucans having structural features that promote high viscosity or a secondary gel network formation by the polysaccharide, one can reduce significantly serum separation of the milk gels compared to control formulations (free of β-glucans).	[75]

Table 1.1. is continued

1	2	3	4	5	6	7
Cereal β-glucan	β-glucan composite / fat replacer	0.25%-1%	Milk was mixed with β-glucan complex and skimmed milk powder at 5% (w / w) level, homogenized, cooled, added with starter, fermented and refrigerated.	The use of β-glucan hydrocolloidal composite in non-fat yogurt affects the whey separation and viscosity of fat free yogurt.	The best results were obtained by addition of the composite at a level of 0.25% or 0.50% for the manufacture of non-fat Yogurt.	[82]
Oat β-glucan	β-glucans were extracted from oats	1%,2%	1%, and 2% levels of β - glucan were added to milk, homogenized, sterilized, probiotics added, filled, fermented and stored.	β-glucan had significantly ($P< 0.05$) increased effects on viscosity, Water-Holding Capacity (WHC) and the viability of probiotic bacteria .	Results showed that the optimum conditions to obtain the synbiotic yogurt made by camel milk were defined as: adding 2% β-glucan (prebiotic agent) to milk with 1.9% fat content inoculated to 0.5% probiotic bacteria with a storage time of 7 ds.	[84]

Table 1.1. Is continued

1	2	3	4	5	6	7
Barley β-glucan	Commercial food grade β-glucan,75%	0.25%-1.0%	Weighed amount of b-glucan 0.25%–1.0%) was added gradually in buffalo skim- med milk at 45 ºC, sterilized, probiotics added, filled, sealed and stored	Enhanced the functional properties, provided functionality of missing fat and improved physical, rheological and textural properties of the final product.	Syneresis and viscosity was positively affected with the addition of β-glucan till 0.5% level and higher concentration caused destabilization of the product.	[76]
Barley β-glucan	91.52%	0.5%-2.0%	Milk containing 4.5% fat and 2% skimmed milk powder were homogenized, heated for 5 minutes, β-glucan was	Improved the rheological, physicochemical and sensory properties of the yogurt,improved the nutritional values	Addition of b-glucan significantly (< 0.05) improved whey separation (syneresis), viscosity, texture profile and sensory characteristics during storage.	[69]

Table 1.1. is continued

1	2	3	4	5	6	7
			dispersed into 100 mL of hot milk at 60 ºC, stirred continuously for 10 minutes, and then introduced into the rest of the milk used for yogurt preparation, stirred for 2 min. Sugar (12%) was added to the milk, mixed for 5 min, then pasteurized at 80 ºC for 5 minutes, cooled to 45 ºC for 5 min, starter was added, fermented and stored.	by acting as a prebiotic in probiotic Yogurt.		

Table 1.1 is continued

1	2	3	4	5	6	7
Oat β-glucan	86%	0.10%, 0.15%, 0.20%	The β - glucan powder was added into the milk matrix at the levels of 0.10%, 0.15% and 0.20%, heated at 95 ºC for 10 minutes for pasteurization, distributed into the pre-sterilized glass container and cooled to 4 ºC for one day. The starter was added and fermented at 40 ºC until the final pH value is 4.9 ± 0.1.	Better sensory evaluation results, lower syneresis, and better water holding capacity.	Low-fat yogurt enriched with β-Glucan at 0.10% level demonstrated superior quality, less whey separation, good textural properties and lower syneresis.	[85]

The End of the Table 1.1

1	2	3	4	5	6	7
Oat β-glucan	86%	0.15%	The milk was heated to 30 ºC, standard β - glucan was added gradually, heated to 60 ºC, the mixture was stirred continuously at 600 r/min for 20 min in a magnetic stirrer, then cooled to the fermentation temperature of 40 – 42 ºC, the starter was added, fermented and stored.	Improved the texture of yogurt, shortened the fermentation time and improved the function of probiotics.	After adding β-glucan, the dehydration rate and water holding rate of yogurt increased from 23.6% and 44.1% to 17.06% and 49.6%, the viscosity of yogurt increased by 2.3 mPa · s, the fermentation time shortened by 30 min, and the total number of microorganisms (probiotics) increased by $2 \cdot 10^7$cfu/g.	[86]

1.9.2 Prebiotic properties of β-glucan

In addition to improve the texture of Yogurt, the cereal β-glucan has been shown to have a prebiotic properties. In the gastrointestinal tract, cereal β-glucan is act as a substrate for microbial fermentation, it selectively stimulating the growth and activity of a small number of beneficial bacteria [87; 88; 89]. It was adopted the way of gavage the mice with OG and barley β-glucan for 6 weeks and analyzed the intestinal health indicators, who found that the number of *Lactobacillus* and *Bifidobacterium* in the intestinal tract of mice were increased during the administration of cereal β-glucan, and the intestinal health promotion effect of OG was better than that of barley β-glucan[90].

Gee et al. studied the effect of barley β-glucan on the growth of two yogurt starter cultures (SC_S) composed of *Streptococcus thermophilus*, *Lactobacillus delbrueckii* and *subsp.* [91]. *Bulgaricus*, and through monitoring the fermentation efficiency of SC_S with pH value, who found that barley β-glucan had not adversed effect on the growth of SC_S [91]. Ibrahim et al. studied the effects of adding OG and/or *Bifidobacterium* producing exopolysaccharides (EPS) on the physical properties, fermentation time and sensory standard of low-fat yogurt (fat content 1.5%) [86], the results showed that adding OG and EPS producing *Bifidobacterium* to low-fat Yogurt could improve the physical and sensory properties of Yogurt and increased the activity of probiotics. It was found that 0.44% β-glucan (concentrated or freeze-dried) had a protective effect of the *Bifidobacterium* under cold storage [92].

Some scholars have conducted in vitro digestion experiments and medical nutrition experiments on the application of cereal β-glucan in Yogurt. It was studied the bioavailability of peptides and amino acids (AA) in acidic milk gel (or yogurt) containing denatured corn starch, pectin and 75.6% pure barley β-glucan, the results showed that the different polysaccharides added at the end of digestion had not effect on the bioavailability of protein [93].

It was studied that the addition of 0.6% OG had not effected on the fermentation time of fermented products (yogurt, Kefir and fermented milk drinks), and significantly increased the viscosity of the products. The medical nutrition experiment showed that after 21 d of consumption of fermented products containing β-glucan, the total

cholesterol and low density lipoprotein in the blood were significantly decreased, while the high density lipoprotein (HDL) was significantly increased, and eating fermented milk beverage based on buttermilk skimmed milk and rich in OG was beneficial to human health [94].

In a word, the cereal β-glucan has a good solubility, high viscosity, thickening and other processing characteristics, which can improve the texture structure of Yogurt, also, has prebiotic characteristics. Therefore, it has a good application prospect in the production of functional Yogurt.

Previous studies suggested a prebiotic action of cereal β-glucans on some Lactobacillus species [95; 96]. Some studies indicated that incorporation of low levels (up to 0.5%) of oat and barley β-glucans into fermented milk products have no adverse effects on growth of two yogurt starter cultures (SCs) each consisting of *Streptococcus thermophilus* and *Lactobacillus delbrueckii subsp. bulgaricus* and even improved the probiotic viability and stability of some bifidobacteria strains during cold storage [91].

OG can be used on the development of cereal-based functional dairy products (such as Yogurt) with sufficient viability and acceptable sensory characteristics. It was studied the growth and metabolic activity of probiotic organisms in Yogurt and reported that addition of OG resulted in improving probiotic viability and stability [97]. There is an assumption that that OG imparted a protective effect on bifidobacteria strains (for example, *B. longum*) in Yogurt under prolonged cold storage at 4ºC [92]. It was investigated the effects of using dietary fiber barley and OG as a prebiotic on the viability of *Bifidobacterium bifidum* in probiotic yogurt and properties of Yogurt during storage [98]. It was found that the survival of *B. bifidum* was within biotherapeutic level (> 7 log cfu/g) due to the prebiotic effect of barley and oat based β-glucan. The addition of β-glucan to yogurt significantly affected physicochemical properties of yogurts, including pH, titratable acidity (LA %), whey separation, color (L*, a*, b* values) and sensorial properties.

In order to meet the nutrient guidelines, adequate amounts of OG should be added to Yogurt to have added nutritional benefits. It was found that 0.5% β-glucans addition improved serum retention and viscoelastic nature of yogurt [99]. It was

reported the effect of OG on the fermentation of set-style Yogurt. It was found that OG could be added to yogurt up to 0.3%, which meets the FDA guidelines of 0.75 g β-glucan/serving for a health claim, resulting in Yogurts with quality characteristics similar to the control Yogurt [100]. However, higher contents of OG retarded the fermentation process with noticeable difference in the characteristics of the Yogurt. As observed by phase-contrast microscope, it was found that OG formed aggregates with casein micelle and did not form phase-separated domains.

It was studied the impact of cereal β-glucans (1.4%) on acidification, gelation kinetics and microbiological properties of milk products fermented by yogurt starter culture bacteria and/or probiotic strains [75]. Incorporation of purified OG to milk resulted in phase separation between the added polysaccharide and milk proteins and altered the entire gelation process. Fortification of milk with 1.4% OG retarded the protein aggregation and acidification processes, leading to the formation of significantly weaker gels compared to control formulations. Furthermore, fortification of milk with OG led to a liquid-like structure at the end of fermentation (pH=4.6) at 36 ºC. Moreover, the probiotic strain of *L. paracasei* exhibited good compatibility with the Yogurt starter culture and OG addition enhanced the viability of the probiotic strain in the milk fermented products during cold storage (4ºC).

Conclusions to Section 1

Through a review of literature sources and informative data, the following conclusions were made:

1.Research related to the development of food products for therapeutic and preventive purposes is relevant because the number of people suffering from a number of diseases caused by improper nutrition is constantly increasing in the world.

2. Cereal β-glucan is an available raw material with good physicochemical properties, but its effect on dairy raw materials and the quality of the finished product should be investigated.

3. It has been shown that cereal β-glucan has good solubility, high viscosity, thickening and other processing properties, which can not only improve the texture

structure of yogurt, but also have prebiotic properties. Therefore, it has good performance in the production of functional Yogurt application prospects.

4. OG can be used as a functional food ingredient to improve the nutritional value and physical-chemical, therapeutic-preventive properties of yogurt.

5. There are no studies on the use of oat β-glucan in the production of fermented milk products and their impact on the biological and nutritional value of the finished product, the properties of in vitro digestion, which limits its use in the production of Yogurt.

6. There is no technology for the production of Yogurts using oat β-glucan, and a balanced formulation taking into account the probiotic properties of the supplement. It is necessary to develop an industrial technology for the production of Yogurts with oat β-glucan.

7. The application of OG in fermented dairy products can not only the range of functional dairy products, contribute to the sustainable development of β-glucan production, but also increase the diversification of the global dairy industry. It is worth investigating the potential value of using oat beta-glucans in Yogurt, the energy efficiency of production, and the product's compliance with existing quality and safety requirements.

SECTION 2 OBJECTS, MATERIALS AND METHODS OF RESEARCH

The main goal of this work is to develop and scientifically justify the technology of a high-quality fermented milk drink - Yogurt with the adding of oat β-glucan as a source of dietary fiber.

To achieve this, the main tasks were formulated:

- to guide the selection of oat β-glucan as a physiologically active additive for effective use in the technological process of yogurt production;

- to investigate the effect of adding oat β-glucan on the chemical composition of the finished product, and based on these studies to calculate the optimal formulation of yogurt with the addition of oat β-glucan;

- to study the effect of adding oat β-glucan on the process of homogenization and fermentation of yogurt, to determine its organoleptic, physicochemical, rheological and antioxidant properties;

- to evaluate the nutritional value of the finished product and develop a technological scheme for its production in industrial conditions;

- to investigate the conformity of Yogurts with oat β-glucan to the requirements of existing standards in Ukraine and China according to the main indicators (organoleptic, physicochemical, microbiological);

- to evaluate the digestion and assimilation of yogurt protein in vitro by simulating the physiological characteristics of animal digestion and adapting the digestive environment and digestive enzyme system similar to those found in the animal body;

- to evaluate functional properties of the product;

- to calculate the energy efficiency of Yogurt production with oat β-glucan;

- to carry out industrial testing of the developed technology of an innovative fermented milk drink - Yogurt with the addition of oat β-glucan.

2.1 Object and subjects of research

The subject of this research is raw material, which was used for the production of Yogurt experiment samples:

- pure cow milk was purchased from Yili Industrial Group Co. Ltd (Neimenggu, China);

- OG (95% purity) were purchased from Zhongkang Food Co., (Guangzhou, China);

-starters: *Streptococcus thermophilus* and *Lactobacillus bulgaricus (Lactobacillus dechellii Bulgarian subspecies)* (viable bacteria count was about 1×10^9cfu/g) were purchased from Danisco (China) Co., Ltd, (Shanghai, China);

- model samples of yogurts with and without additives;

-homogenized normalized mixture with the addition of oat β-glucan.

The research objects are the technology for production of a fermented drink - Yogurt with the addition of oat β-glucan.

Research methods include a complex of traditional and modern methods of sensory assessment, physics, chemistry, microbiology, simulated in vitro digestion, biomedical experiments, experimental planning, and experimental data processing.

2.2 Research methods

2.2.1 Sample preparation

The various amounts of OG (0.1%, 0.2%, 0.3%, 0.4% and 0.5%; W/W) was added to pure milk, respectively. After stirring, the milk was sterilized at 95 °C for 5 min, and then cooled to 43 °C, added the starters (containing *Streptococcus thermophilus* and *Lactobacillus denderi Bulgarian subspecies*), fermented at 43 °C for 5 h, and stored at 4 °C for 24 h.

Milk, according to the corresponding quality standard, was purified and heated to a temperature of 40-45ºC. A milk base enriched with oat β-glucan in various amounts (0.1%, 0.2%, 0.3%, 0.4% and 0.5%) was prepared on the basis of skim milk. After thorough mixing for 15-20 minutes, the enriched milk base was heated to a temperature of 70-75ºC and homogenized (18-20 mPa). The mode of homogenization was agreed upon by a number of experimental studies. Pasteurization of the homogenized milk mixture was carried out at a temperature of 90-95 oC for 10-15 minutes. The pasteurized mixture was stirred for 5 min, cooled to 40ºC. Starter (*Streptococcus*

thermophilus and *Lactobacillus denderi Bulgarian subspecies*) was added to the cooled milk, mixed for 20-30 minutes for effective fermentation. Fermentation was for 2.5-3 hours, while the acidity of the fermented sample was 70-80ºT. The fermented product was cooled and stored at 4ºC for 21 days.

2.2.2 Sensory evaluation

10 volunteers (5 males and 5 females), who had experience in sensory evaluation of fermented milk, were selected to conduct sensory evaluation of set-type Yogurt with different supplemental of OG. Sensory score criteria are represented in the Table 2.1.

Table 2.1 Sensory score criteria of set-type Yogurt

Characteristic	Scoring criteria	Score
1	2	3
Color	Uniform color, milky white or milky yellow	7-10
	Different colors	4-6
Structural state	Good coagulation, fine and uniform structure, no whey precipitation	25-30
	Good coagulation, fine and uniform structure, a small amount of whey precipitation	15-24
	Poor coagulation, different structure and serious whey precipitation	0-14
Texture	The taste is smooth and delicate, with thickness and viscosity	25-30
	The taste is smooth and delicate, with thickness and viscosity	15-24
	The taste is not smooth, delicate and astringent	0-14
	The unique fermented and milk flavor of yogurt, with strong flavor	25-30
	The fermented and milk flavor of yogurt is light, and the flavor is general	15-24
	Loss of flavor of fermented milk and abnormal odor	0-14

2.2.3 Determination of WHC

To assess the ability of the clot to retain moisture, samples of Yogurts (30 g) were placed in a test tube and centrifuged at 4000 rpm for 25 min at 4°C and the volume of the obtained serum was measured in ml.

The calculation formula is as follows:

$$\text{WHC (\%)} = \frac{W_2 - W_0}{W_1 - W_0} \times 100, \qquad (2.1)$$

Where W_0 – mass of the test tube, g;

W_1 – quantity of Yogurt, g;

W_2 – quantity of separated whey, g.

2.2.4 The method of research of oat beta-glucan on the rheological characteristics of yogurt

According to the methods [101; 102], the rheological characteristics of set-type Yogurt was measured by optical microrheometer (RHEOLASER, Formulaction Company, France). The changes of MVI, EI and SLB with time during the gel process were observed at 42 °C. The micro-rheological data of the system were analyzed by software of RHEOLASER master optical micro-rheological analyzer.

2.2.5 Research of microstructure

According to the method [103], OG samples stored for 24 h after post-ripening were evenly and thinly coated on the inner wall of the petri dish. After being frozen in liquid nitrogen, the samples were quickly put into a vacuum freeze dryer for drying treatment, and then the samples were prepared by scanning electron microscope (OXFORD INCA250, Shanghai Oxford Instrument Technology Co. Ltd., China). In the studied samples the images were observed and collected under 10kV voltage and 500 ~ 3 000 times magnification.

2.2.6 Study of free amino acids content

Fermented milk not only produces lactic acid, but also degrades protein to produce amino acids and other beneficial components. Amino acids affect the texture, nutrition and flavor of fermented milk. There have been many researches on the application of automatic amino acid analyzer in milk and dairy products. The analyzer method has the advantages of fast analysis time, wide application range and high sensitivity. It is widely used in the determination and analysis of amino acid content in modern food.

In this experiment, the s-433 (d) amino acid analyzer of Sykam Company in Germany was used to analyze the yogurt added with OG. With reference to the experimental methods [104; 105] and slight modification, the effects of OG before and after the addition of OG and different amounts of OG on the amino acid composition of Yogurt were determined.

The pretreatment of Yogurt samples is slightly modified according to the method [106]. Take the yogurt samples to be tested, add 5% sulfosalicylic acid, mix them evenly, centrifuge them, put them into the film, and put them into the sample bottle for standby. Chromatographic conditions: analytical column LCA K06/Na (4.6 mm×150 mm, 3 μm), column temperature 57.0 °C, reaction column temperature 74 °C, mobile phase flow rate 0.45 mL/min, sample injection volume 20 μL. UV detector: the absorbance of proline is measured at 440nm, and that of other amino acids is measured at 570nm. To ensure the reliability of the experimental results, the experiment was repeated for 3 times.

The free amino acid content is calculated by formula:

$$\text{Amino acid content} = \frac{5\times10^{-5}\times C_1\times V\times F\times M}{m}, мг/г \quad , \qquad (2.2)$$

Where: *Ci* is the mass concentration of free amino acid / (ng/mL);

V is sample volume /mL; F is the dilution multiple;

M is the molar mass of amino acid / (g/mol);

M is the quality of yogurt /mg.

2.2.7 Study of fatty acids content

The gas chromatograph model GC-2030AF, specification HS-20/GC-2030/ FID, ECD, PFPD, used to determine the fatty acid composition of Yogurt is manufactured by Shimadzu Company of Japan. The fatty acids were methylated first and then analyzed by GC.

Fatty acid methyl esterification was carried out according to the methods[107; 108]. About 0.15 g of milk was taken fat extracted from OG Yogurt and put it into a 20mL plugged test tube. 2 mL of 0.5 mol/L sodium hydroxide methanol solution was added and kept the temperature at 65 ^{o}C for 30 min. 2 mL of boron trifluoride ether methanol solution (volume ratio 1:3) was added and continued heating for 30 min (shake fully). After cooling to room temperature, 2 mL of n-hexane was added to shake, let it stand for layering, added saturated sodium chloride solution to lift the organic phase, 1.5 mL of the upper organic phase was taken, a small amount of anhydrous sodium sulfate was added to dry, centrifuged at 10000 r/min for 5 min, and then the membrane was passed for GC analysis.

GC condition: CP SIL 88 (100 m × 0.25 mm ×0.20 μm) Chromatographic column; Carrier gas: high purity helium; Column flow: 1.2 mL/min; Column temperature: the initial temperature shall be 145 ^{o}C for 10 min, the temperature shall be increased to 185 ^{o}C at the rate of 2 ^{o}C/min for 15 min, and the temperature shall be increased to 215 ^{o}C at the rate of 2 ^{o}C /min for 15 min; Injection port temperature 250 ^{o}C; Split ratio 15: 1; Injection volume: 0.5 μL.

2.2.8 Study of volatile substances content

Volatile substances in OG Yogurt were determined by headspace solid-phase microextraction combined with GC/MS analysis according mainly to the method documented [109] with some modifications. In brief, 5 g sample was placed into a sealed sample bottle, and then transferred to a magnetic stirring heating table operated at 60 ℃ for 10 min. Subsequently, an activated extraction head (75 μm, CAR/PDMA) was inserted into its upper void space, which was 1.5 cm above the sample. After heating at 50 ℃ for 1h, gas-solid and gas-liquid equilibrium were reached, and then

the extraction head was inserted into the GC inlet and suffered from thermal desorption at 230 ℃ for 3 min. GC was equipped with a HP-5 column (30 m×0.25 mm×0.25 μm), and the following temperature profile was used: starting at 25 ℃ and maintaining for 3 min, heating to 130 ℃ at a rate of 2 ℃/min and holding for 1 min, and then ramping up to 210 ℃ at a rate of 5.5 ℃/min and lasting for 3 min. MS was equipped with an electron ionization source working under electron energy of 70 eV, and interface temperature and ion source temperature were both 230 ℃. Mass scanning was in the range of 33 to 260 U. Each sample was tested for 3 times. The compounds were identified mainly by searching the NIST library. Semi-quantitative analysis was carried out by the peak area normalization method.

2.2.9 Study of physical and chemical indicators of the product

2.2.9.1 Determination of the mass fraction of fats

The set-type Yogurt with different OG contents is frozen dry for 72 h, weighy 5g of fully mixed sample, accurate to 0.001g, all moved into the filter cartridge. The filter paper cartridge is put into the extraction cartridge of Soxhlet extractor, connected the receiver bottle that has been dried to constant weight, petroleum ether is added from the upper end of the extractor condenser pipe to two-thirds of the volume in the bottle, and heated it on a water bath to make the petroleum ether continuously reflux for extraction for 10 h.

The receiving bottle is removed and fat is collected. It is steam dry in a water bath, and then dry at 100 ± 5°C for 1 hour. It is placed in a dryer and cool for 0.5 hours before weighing. It is tepeated with the upper operation until the weight is constant (until the difference between the two weights does not exceed 2 mg).

The fat content in the sample is calculated according to the formula:

$$X = (m_1-m_0) / m_2 \times 100 \qquad (2.3)$$

X - the content of fat in the sample, g/100 g;

m_1 - the content of the receiving bottle and the fat after constant weight, g;

m_0- the mass of the receiving bottle, g;

m_2 - the mass of the sample, g.

2.2.9.2 Determination of the mass fraction of dry solids

5 g of OG type yogurt is frozen dry for 72 hours, placed in a drying box at 100°C ± 2°C for 3 hours, removed and placed in a desiccator for cooling for 0.5 hours, weighed, and placed in a drying cabinet at 100 ± 2 °C for 3 hours. It is dried for 1 hour, removed, cooled and weighed until the difference in mass between the two portions does not exceed 1.0 mg. Calculate the total solids content of the yogurt, denoted as m.

According to method 2.2.9.1, mark the fat content in Yogurt OG set as m1.

The content of skimmed milk powder in the sample is calculated according to the following formula:

$$XNFT = m - m_1 \quad (2.4)$$

XNFT - the content of non-fat milk solids in yogurt, g/100g;

m - the content of total solids in yogurt, g/100g;

m_1 - the content of fat in yogurt, g/100g.

2.2.9.3 Determination of the mass fraction of protein

The yogurt samples are frozen dry for 72 h, and weighed 1.0g of fully mixed solid sample, accurate to 0 001 g, put it into the digestive tube, and then 0,4G copper sulfate, 6G potassium sulfate and 20mL sulfuric acid are added. They are placed in an automatic digestion furnace (FOSS company). When the temperature of the digestion furnace reaches 420 °C, continue digestion for 1H. At this time, the liquid in the digestion tube is green and transparent. It is taken out and 50mL of water is added after cooling.

The process of automatic liquid addition, distillation, titration and recording of titration data is realized on the FOSS analytical AB (adding sodium hydroxide solution, hydrochloric acid or sulfuric acid standard solution and boric acid solution containing mixed indicator a or B before use).

The protein content of Yogurt is calculated based on the results of the experiment.

2.2.9.4 Determination of acidity and pH

Determination of acidity was performed by an acidimeter (PB-10, Seidolis instruments, Germany): set-type Yogurt samples were taken every 1 h during the fermentation process, and the yogurt samples were determined after stored 24 h. 10 g Yogurt samples were put into 250 mL triangle bottle, and 20 mL distilled water was added to dilute and mixed, 0.5% phenolphthalein was added as an indicator, and titrated with 0.1mol/L NaOH standard solution until it was slightly red. Do not fade within 30s is the end. Consuming 0.1 M of NaOH is equal to 1 °T.

All the pH values were monitored after adding starter bacterial cultures, using a digital pH meter (Thermol Scientific Inc., USA).

2.2.9.5 Determination of color

The CR-400 colorimeter was used to analyze the chromaticity of yogurt samples. Use the blackboard to calibrate the lowest reflectance value, and use the white board to calibrate the highest reflectance value. The values of L*, a*, b* values can be read out directly and calculated ΔE value according to the formula:

$$\Delta E = \sqrt{L^{*2} + a^{*2} + b^{*2}} \qquad (2.5)$$

ΔE is the size of the total color difference, the smaller the value, the smaller the color difference;

L* - brightness value;

a* - reddening value;

b* - value of yellowness

2.2.10 Research of microbiological indicators

2.2.10.1 Microbial indicators

Determination of *E. scherichia coli.* 25 g of Yogurt is weighed, put it into a sterile homogenizing cup containing 225 mL of phosphate buffer or normal saline, and homogenized it at 8000 r/min~10000 r/min for 1 min~2 min to make a 1:10 Sample homogenate.

2 to 3 suitable serial dilutions are selected, and inoculated 2 sterile plates for each dilution, each with 1 mL. At the same time, 1 mL of normal saline was added to a sterile plate as a control.

Immediately pour 15 mL ~ 20 mL of crystal violet neutral red bile salt agar (VRBA) melted and thermostated to 46 °C into each plate. The plate is rotated carefully, the medium and the sample solution thoroughly are mixed, and after the agar solidifies, 3 mL~ 4 mL VRBA are added to cover the surface of the plate. The plate is flipped and placed at 36 °C ± 1 °C for 18 h~24 h.

The plate with the colony number between 15 CFU ~ 150 CFU is selected, and the typical coliform colonies appearing on the plate respectively are counted. The typical colony is purple-red, with a red bile salt precipitation ring around the colony, and the colony diameter is 0.5 mm or more.

The proportion of test tubes that were finally confirmed to be coliform-positive was multiplied by the number of plate colonies counted, and then multiplied by the dilution factor, which was the number of coliforms per g sample.

Determination of *Staphylococcus aureus*. The preparation before sample inoculation is the same as the determination of *E. scherichia coli*. 2 to 3 sample homogenates with appropriate dilution are selected, while performing 10 fold incremental dilution, pipette 1 mL of sample homogenate for each dilution and added to the Baird-Parker plate, the plate is let stand for 10 min, and then the plate is flipped, cultured at 36 °C±1 °C for 24 h~48 h. The plate is selected with typical *Staphylococcus aureus* colonies, and the total number of colonies is between 20 CFU ~ 200 CFU, and count the typical number of colonies.

The number of *Staphylococcus aureus* colonies was calculated according to the dilution ratio.

Determination of *Salmonella*. The preparation before sample inoculation is the same as the determination of *E. scherichia coli*. The cultured sample mixture is shaked gently, 1 mL is removed, transferred to 10 mL of enrichment solution, and incubated at 42 °C ± 1 °C for 18 h to 24 h. At the same time, 1 mL of cultured samples is taken,

inoculated again in 10 mL of enrichment solution, and cultured at 36 °C ± 1 °C for 18 h to 24 h.

A loop of the enrichment solution with a 3 mm diameter inoculation loop is taken, streaken it on an HE agar plate, and incubated at 36 °C ± 1 °C for 18 h ~ 24 h.

Through biochemical tests and serological identification, it was reported that Salmonella was detected or not detected in the 25g (mL) sample.

Determination of *Yeast*

The preparation before sample inoculation is the same as the determination of *E. scherichia coli*. 2 ~ 3 sample homogenates of appropriate concentration are selected, and at the same time of 10-fold incremental dilution, 1 mL of sample homogenate is drawn into 2 sterile plates for each dilution, and a blank control is made. 20 ~ 25 mL of potato dextrose agar is poured at 46 °C into the plate in time, and after the agar is mixed evenly and solidified, it is placed in a 28°C ± 1°C incubator for cultivation, and the results of the cultivation to the 5th day are observed and recorded.

2.2.10.2 Quantity of lactic acid bacteria

1 g of Yogurt with 9 mL of 0.9% (w/v) NaCl was mixed and diluted to a concentration of 10^4, 10^5, and 10^6, and then 1 mL of each dilution was inoculated onto plates containing the MRS agar. Bacteria were counted by the pour plate technique. The plates in duplicates were incubated anaerobically at 37 ℃ for 72 h, and then colonies were counted [92; 110; 84].

2.2.11 Determination of viscosity

Viscosity of Yogurt was measured at 4 ℃ with a spindle (No. 3) rotation of 1500 r/min using the digital display rotating viscometer (NDJ-8S, Shanghai Yueping Scientific Instrument Co., Ltd. (Shanghai, China). The readings were recorded at 20th second of the measurement. The measurements were made in triplicate.

2.2.12 Texture Profile Analysis (TPA) parameters

The experiments were performed by a texture analyzer (TA-XT.plus Texture Analyzer; Stable Micro Systems, Godalming, UK). The Yogurt samples were placed

in 25 mL beakers, and the TPA test mode and P/0.5 probe were used to determine the gel structure of the fermented milk. The parameters are as follows: the speed is divided into 2.0 mm / s before measurement, 1.0 mm / s during measurement and 2.0 mm / s after measurement. Test samples shall be made in triplicate.

2.2.13 In vitro digestion

Buccal, gastric and duodenal digestion stages were performed sequentially the methods described by [111; 112] with some minor modifications. Amylase (1000–3000 U/mg protein), pepsin from porcine stomach mucosa (1: 60,000), pancreatin from porcine pancreas (8 × USP) and sodium deoxycholate, cholesterol, oleic acid, phosphatidylcholine, and bile from bovine were obtained from Sigma Aldrich (St. Louis, MO, USA). All other chemicals were of analytical grade.

Buccal stage: simulated salivary fluid at pH 7.0 was added to yogurt samples in a ratio 1:1 (w/v) under gentle stirring using a kitchen blender for 2 min at 37 °C. Human α–amylase was added as a part of the salivary fluid to reach a desired concentration (75 U/mL). Gastric stage: simulated gastric fluid (pH 3.0) was added to tubes in a 1:1 (v/v) ratio including 2 mL pepsin. The pH of yogurt samples was adjusted to 2.0–2.5 with 2.5 M HCl. Then, the sample solutions were mixed thoroughly and incubated at 37 °C for 30 min by a shaking incubator. Further analyses were conducted for the supernatant.

Duodenal stage: After gastric stage, simulated intestinal fluid containing 2 mL of porcine pancreatin and 1 mL of bile acid mixture (pH 6.0 or 7.0) was added in 1:1 (v/v) ratio to tubes containing the gastric chyme. The pH of sample solutions was adjusted to 6.5–7.0 with 4 M NaOH. The sample solutions were incubated at 37 °C for 90 min by a shaking incubator. After the incubation, the sample solutions were centrifuged (10,000×g, 10 min). Further analyses were conducted for the supernatant.

2.2.14 Total soluble protein content of in vitro digested samples of Yogurt

The contents of proteins in undigested Yogurt samples and in supernatants from digested Yogurt samples after centrifugation were determined with the bicinchoninic acid protein assay kit (Pierce Company). The content of soluble protein in digested

yogurt samples was expressed as percentage (%) of total protein in undigested Yogurt samples.

2.2.15 Optical microscopy

The microstructure of control Yogurt and digested Yogurts was observed by optical microscopy (Axio Vert.A1, Carl Zeiss), according to the previous works [113]. Yogurt samples were put between glass slides and immediately observed at a magnification of 100× at room temperature. All experiments were performed in triplicate.

2.2.16 Particle size and size distribution

The particle sizes of control Yogurt and digested Yogurts were measured by dynamic light scattering using a Zetasizer Nano ZS90 (Malvern Instruments Ltd, Worcestershire, U.K.). Particle size was obtained by the Stokes-Einstein equation. The polydispersity index (PDI), representing the distribution of particle size, was also reported. Before measurement, all samples were diluted by 1:5 (v/v) with deionized water at the corresponding pH values and then equilibrated for 2 min inside the instrument at 25 ^{o}C. Data were collected over at least 20 sequential readings. All experiments were performed in triplicate.

2.2.17 Antioxidant activities

The 2, 2-diphenyl-1-picryhydrazyl (DPPH) radical-scavenging activity of yogurts was determined by the method [114] with minor modifications, and evaluated using an ELX800 Microplate Reader (Bio-Tek, Bedfordshire, UK). 2 mL of yogurt samples was added into 2 mL of 0.1 mM DPPH solution (90% methanol). After vortexed igorously, the the mixtures were allowed to keep in the dark for 30 min at room temperature. Methanol was used instead of yogurt samples for the control measurements. The scavenging capacity was determined as follows:

$$Scavenging\ Activity\ (\%) = 100 \times \frac{A_{DPPH} - A_S}{A_{DPPH}}, \qquad (2.6)$$

Where A_S is the absorbance of the measured Yogurt samples, and A_{DPPH} is the absorbance of the blank samples.

The 2, 2′-azino-bis-3-ethylbenzthiazoline-6-sulphonic acid (ABTS) assay was determined according to the method proposed by [115] with minor modifications. ABTS radical cation was produced by mixing ABTS with 2.45 mM aqueous potassium persulfate and then leaving them in the dark for 15 h at room temperature. The ABTS stock solution was diluted with phosphate buffered solution (pH 7.4) to make its absorbance value of 0.70 ± 0.02 at 734 nm. 0.2 mL of Yogurt samples was mixed with 3.8 mL of the prediluted ABTS solution, allowing it to stand for 6 min at room temperature before measurement. 0.2 mL water was used instead of yogurts for the control sample.

2.2.18 In vitro cholesterol micelle assay

Cholesterol micelles were prepared following the two methods described by [116; 117] with some minor modifications. An emulsion at pH 7.4, mainly containing 0.5 mM cholesterol, 10 mM sodium taurocholate, 1 mM oleic acid, 1 mM cholesterol, 132 mM NaCl, and 15 mM sodium phosphate buffer, was prepared. And then, the emulsion was treated with ultrasonic energy (400 W, 20 kHz, 20 min), and incubated at 37 ℃ overnight. Yogurt samples were mixed with the emulsion and the obtained mixtures were incubated at 37 ℃ for 24 h. Afterwards, the mixtures were centrifuged at 8000 g for 30 min and the supernatants were collected. Cholesterol contents in the supernatants were determined by a total cholesterol kit. Micelle cholesterol uptake inhibition was calculated according to the formula used by [118]:

$$\text{Inhibition Capacity (\%)} = (1 - C_1/C_0) \times 100\%, \quad (2.7)$$

Where C_0 is the cholesterol concentration in the micelle,

C_1 is the cholesterol concentration with peptides.

2.2.19 Experimental animals

The experiments were performed as approved by the Institutional Animal Care and Use Committee of Henan Institute of Science and Technology, and in accordance with the Animals (Scientific Procedures) Act 1986 (amended 2013). Sixty male C57BL/6 mice of 8 weeks old were obtained from Experimental Animal Tech Co. of Weitonglihua (Beijing, China). SPF mouse-breeding cages were used with an average of 5 SPF mice placed in each cage (22-25 ^{o}C, 50-55% humidity, and 12 h light-dark cycle).

After 1 week of acclimatization, the experimental mice were randomly divided into four groups and then received different treatments, according to the method of Lasker et al. (2019) with minor modifications [119].

(a) Control (Group 1), received normal water and normal food for eight weeks.

(b) HF (Group 2), received the high-fat diet for eight weeks.

(c) HF+0.0% OG Yogurt (Group 3), received the high-fat diet and 0.0% OG yogurt (5%, w/w) for eight weeks.

(d) HF+0.3% OG Yogurt (Group 4), received the high-fat diet and 0.3% OG yogurt (5%, w/w) for eight weeks.

The normal food was composed of wheat, wheat bran, and rice polishing and fish meal. The high-fat diet was composed of 1% cholesterol, 20% lard, 4% sucrose and 75% normal food.

The body weight and length were recorded once a week.

Sample Harvesting from Mice. At week 9, all mice were sacrificed (under anesthesia by pentobarbital; 90mg/kg). The blood samples was collected from the angular vein into tubes, and centrifuged at 8000 r/min (15 min; 4 ^{o}C) to separate the plasma. Separated plasma was transferred to 1,5 mL Eppendorf tubes and stored at -80 ^{o}C.

2.2.20 Mice Lee’s index and liver wet weight

The weight of the mice was weighed, the length of the mice (the distance from the tip of the nose to the anus) was measured accurately, and their Lee's index was calculated.

$$Lee\ index\ = \frac{\sqrt[3]{body\ weight} \times 10^3}{Body\ \ length} \qquad (2.8)$$

body weight, g;

body length, cm

After mice were sacrificed and plasma was collected, livers were quickly removed and weighed, and then stored in neutral buffered formalin for further analysis.

2.2.21 Determination of Serum Biochemical Indices of blood fat.

Chemicals.Triglyceride (TG), cholesterol (TC), high-density lipoprotein cholesterol (HDL) and low-density lipoprotein cholesterol (LDL) assay kits were procured from Nanjing JianCheng Bioengineering Institute (Nanjing, China). All other chemicals were of analytical grade.

The levels of TC, TG, HDL, and LDL were detected by kit (Nanjing JianCheng Bioengineering Institute, Nanjing, China), according to the manufacturer's recommended protocols.

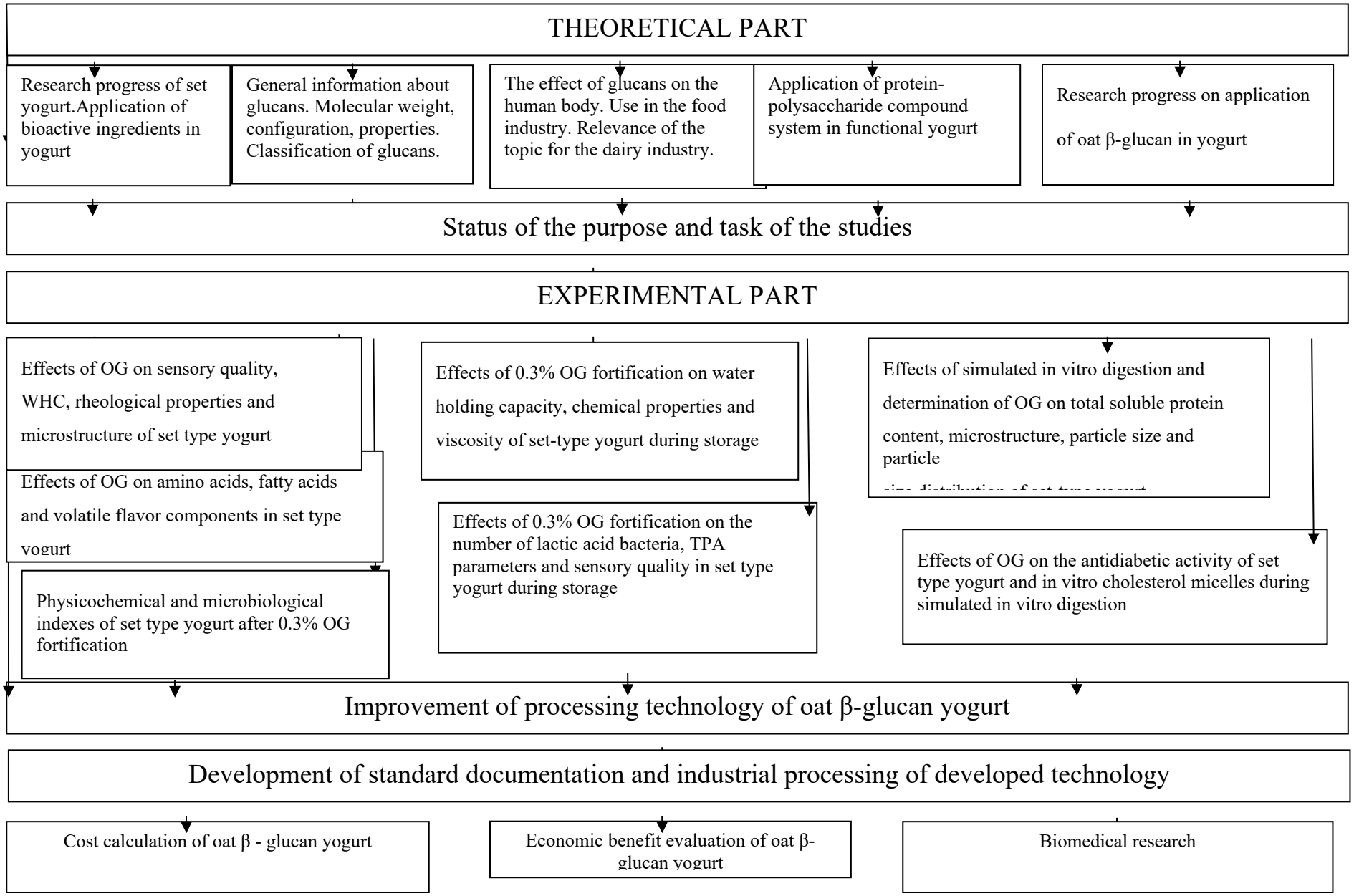
2.2.22 Determination of Alanine aminotransferase (ALT), aspartate aminotransferase (AST).

Chemicals. Alanine aminotransferase (ALT), aspartate aminotransferase (AST) assay kits were procured from Nanjing JianCheng Bioengineering Institute (Nanjing, China). All other chemicals were of analytical grade.The ALT and ALP activities were detected by kit (NanJing JianCheng Bioengineering Institute, Nanjing, China), according to the manufacturer's recommended protocols.

2.2.23 Liver histological analysis

The liver tissues fixed in 10% formalin were embedded in the paraffin. Sections with 5μm thickness were prepared and stained with hematoxylin and eosin to visualize the architecture of hepatic tissue. Then, stained sections were observed under an optical microscope (Eclipse E100, Nikon, Tokyo, Japan).

2.3 Research program



2.4 Statistical processing of research results

All experiments were conducted in triplicate, and the results are expressed as the mean ± standard deviation. SPSS version 17.0 (SPSS Inc., Chicago, IL, USA) was used for all statistical evaluations, and OriginPro 8.6.0 (Originlab, Northampton, Massachusetts, USA) was used for the construction of the graphs. The sensory evaluation and yogurt WHC were analyzed by an independent sample t-test of SPSS 17.0 software. One-way ANOVA was used to analyze the titration acidity of yogurt, LSD and Dunnett's T3 test were applied for multiple comparisons and differences were considered to be statistically significant at $P< 0.05$.

Conclusions to Section 2

1. Methodological approaches adopted in the dissertation work that include theoretical research, laboratory experiment, biomedical experiment, an experiment with pc use, practical testing, subordinate various methods of research of the only goal decision set in the dissertation work scientific problem.

2. The object of research is the technology for production of fermented drink-Yogurt with adding OG.

3. The selected set of methods allows you to comprehensively characterize the nutritional value, physicochemical, functional, technological and medical biological characteristics of new types of OG set-type Yogurt product.

4. The experiments carried out on animals allowed to confirm therapeutic-preventive properties of Yogurt with the addition of oat β-glucan.

SECTION 3 THE EFFECT OF OAT β-GLUCAN ON THE QUALITY INDICATORS AND STORAGE OF YOGURT

3.1 Effects of adding OG on the quality of Yogurt

3.1.1 Sensory evaluation of Yogurt with adding of β-glucan

The organoleptic indicators of Yogurt play an important role in its quality, as most of the consumers choose the product based on its taste-aromatic properties and appearance.

Based on the sensory evaluation results, it was established that when the OG concentration increased from 0 to 0.3%, the sensory scores gradually increased.

The sensory evaluation results of yogurt with adding of β-glucan are shown in Table 3.1.

Table 3.1 Sensory evaluation of set-type Yogurt

OG addition (%)	Characteristic index				
	Color	Structural state	Consistency	Flavor	Score
0	8.12±0.12^{a}	26.05±0.36^{b}	24.28±0.22^{e}	23.70±0.46^{d}	82.15±0.61^{d}
0.1	8.60±0.28^{b}	25.60±0.47^{c}	24.35±0.32^{d}	24.70±0.52^{a}	83.25±0.77^{b}
0.2	8.65±0.19^{b}	25.40±0.72^{d}	24.60±0.41^{c}	24.60±0.36^{b}	83.25±0.53^{b}
0.3	8.71±0.62^{a}	26.78±0.43^{a}	24.97±0.69^{a}	24.67±0.29^{ab}	85.12±0.78^{a}
0.4	8.33±0.32^{c}	25.56±0.61^{cd}	24.67±0.49^{c}	24.67±0.36^{ab}	83.22±0.64^{b}
0.5	8.15±0.22^{d}	25.20±0.78^{e}	24.80±0.32^{b}	24.50±0.49^{c}	82.65±0.76^{c}

Note: All data are expressed as the mean ± standard deviation (n =3),the angle a-d indicates whether there is a difference, the same indicates no difference, and the different indicates difference.

When the OG concentration increased up to 0.3%, the set-type Yogurt had the highest sensory score (about 85). And when OG concentration increased, the sensory scores continuously decreased, reaching the lowest sensory scores (about 82) at 0.5% OG concentration.

It was established that the color and flavor of OG Yogurt were significantly different from those without OG ($p< 0.05$), but the consistency of yogurt was not significantly different ($p> 0.05$) when the amount of OG was less than or equal to 0.4%. In general, the sensory evaluation showed that the addition of β-glucan in the amount of 0.3% had a positive effect on the organoleptic characteristics of the finished product.

3.1.2 Effect of β-glucan on water-holding capacity of Yogurts

Addition of oat β-glucan to yogurt had a positive effect on its water-holding capacity.

The results of the study are presented in Fig. 3.1.

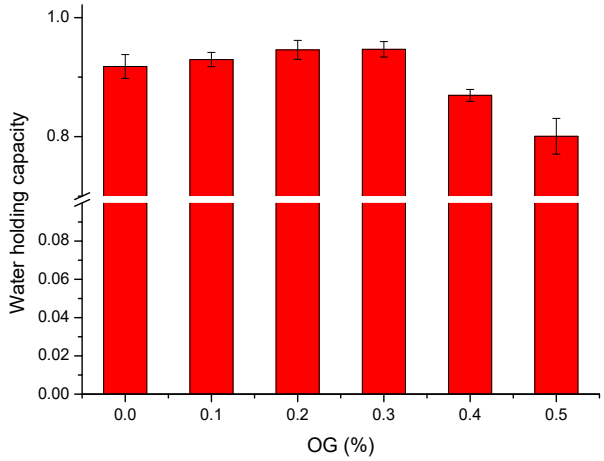


Fig. 3.1 Comparison of WHC of set-type Yogurt with different OG concentrations (0, 0.1%, 0.2%, 0.3%, 0.4%, and 0.5%).

As can be seen from Fig. 3.1, when 0.3% of OG was added to Yogurt, the Yogurt had the highest WHC of 94.67%. And then, the WHC continuously decreased, reaching the lowest WHC (about 80%), when OG concentration was increased up to 0.5%.

It is known that WHC can affect the taste and consistency of set-type yogurt. The better the WHC of yogurt, the more bound water in its gel structure, the better the taste [120; 121]. The results showed that adding the proper amount of OG could enhance the water stability of set-type Yogurt. The reason may be that the OG has a strong water-holding ability, and the interaction between OG and casein can effectively intercept water, prevent the precipitation of whey, and enhance the gel structure of the set-type Yogurt system [122]. However, excessive OG might hinder the interactions of OG with casein, destroy the formation of casein-OG mesh structure, and reduce the WHC of Yogurt.

3.1.3 Effect of oat β-glucan on rheological characteristics of Yogurt

In this section, diffusing wave spectroscopy (DWS) method was used to monitor the macroscopic viscosity index (MVI), elasticity index (EI) and solid–liquid balance (SLB) values of set-type yogurt with and without the OG during the fermentation and post-ripening (Fig. 3.2, Fig. 3.3, and Fig. 3.4).

The results showed that the viscosity change of Yogurt with OG was similar to that of Yogurt without OG. After the stable system was formed, the viscosity of yogurt with OG was higher than that of Yogurt without OG. According to the changes of EI values and SLB values, the addition of 0.3% OG increased the liquid behavior of yogurt, and shortened the gel point.

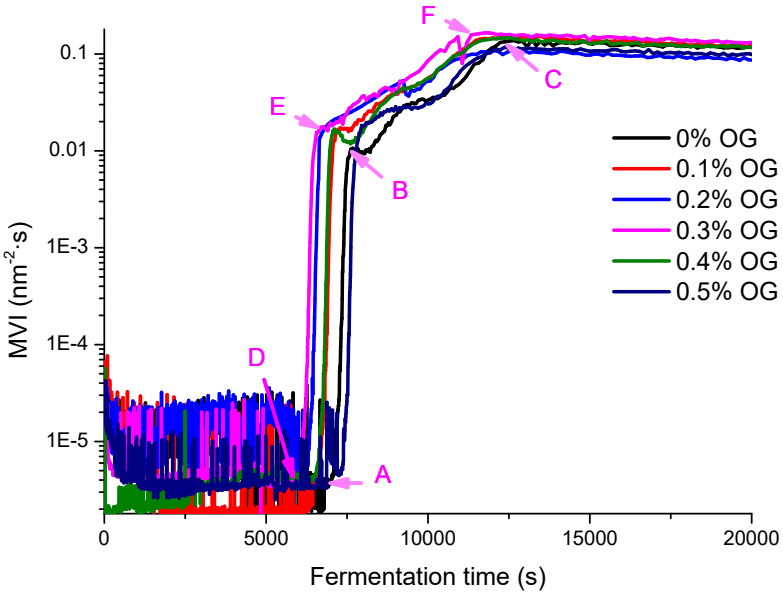


Fig. 3.2 The MVI value curve of set-type Yogurt with different OG concentrations during fermentation.

Macroscopic viscosity index can directly reflect the viscosity characteristics of the system. It can be seen from Fig. 3.2 as a function of fermentation time, the fermentation process of the set-type Yogurt is a multi-stage process. Before fermentation, the MVI for the set-type Yogurt without OG was 0.4×10^{-5} at 117.6 min (point A), which was called the initial stagnation stage with low viscosity. And then, the fermentation of the set-type yogurt started and the fermentation process entered the rapid viscosity change stage. The viscosity increased rapidly. At 128.3 min (point B), the MVI was 0.01, 2500 times compared to that of the MVI before fermentation. This is because fermentation led to a gradual decrease in the pH and an increase in the acidity. At same time, casein particles aggregated to form a gel network structure, which led to an increase in the viscosity of yogurt [123]. At 215.4 min (point C), the set-type yogurt enters the high viscosity stage. The fermentation process ended and a stable gel system was formed. At this stage, the MVI increased to the maximum (point C; 0.13), which was 3250 times and 1.3 times compared to that of the initial stagnation stage and the high viscosity stage, separately.

For the set-type Yogurt with OG, although the viscosity change was similar to that of the set-type yogurt without OG, the fermentation time was changed significantly. As shown in Fig. 3.2, when the OG concentration increased from 0 to 0.3%, the fermentation time gradually shortened. At 0.3% OG concentration, the fermentation start time and end time was shortened by 15.1 min (point D; 102.5 min) and 12 min (point F; 203.4 min), respectively. In addition, at point F, the MVI is 0.025 higher than that of the point C, indicating that the OG increased the viscosity of the set-type Yogurt.

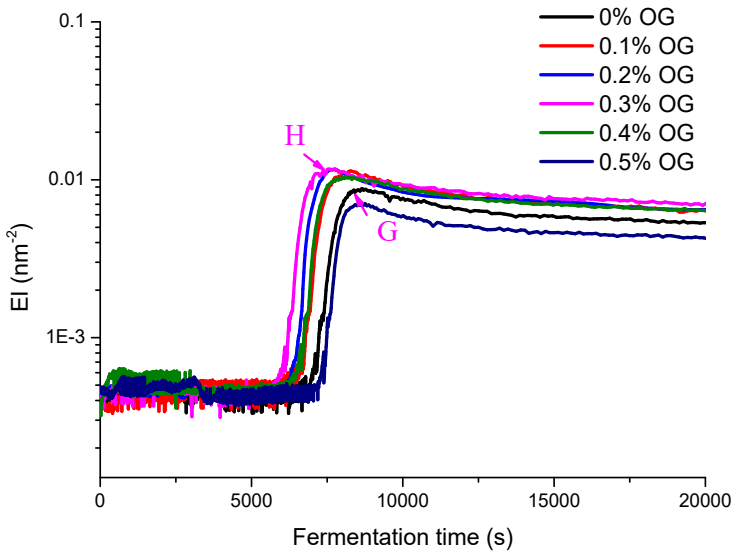


Fig. 3.3 The EI value curve of set-type Yogurt with different OG concentrations during fermentation.

According to WHC, sensory evaluation, and rheological characteristics results, the optimal addition amount of OG is 0.3%.

As a function of time, EI can directly reflect the elastic characteristics of the set-type Yogurt. As can be seen from Fig. 3.3, when the set-type yogurt was in the initial stagnation stage, the EI value remained unchanged. As the fermentation process entered the rapid viscosity change stage, the EI value was increased. When the fermentation time was 145 min (G point), the Yogurt system without adding OG had the maximum EI value was 0.0085. When the fermentation time was 129 min (H point) and 0.3% OG was added, the EI value reached the maximum value (0.0117). It is

generally believed that the gelation rate of Yogurt is mainly affected by the temperature and the lactic acid bacteria type. However, the gel point of the set-type Yogurt with 0.3% OG was shortened by 16 min compared with that of without OG. So, as a functional food ingredient, OG not only enhanced the nutritional and functional properties of set-type Yogurt, but also shortened the fermentation time, thus increasing the production efficiency.

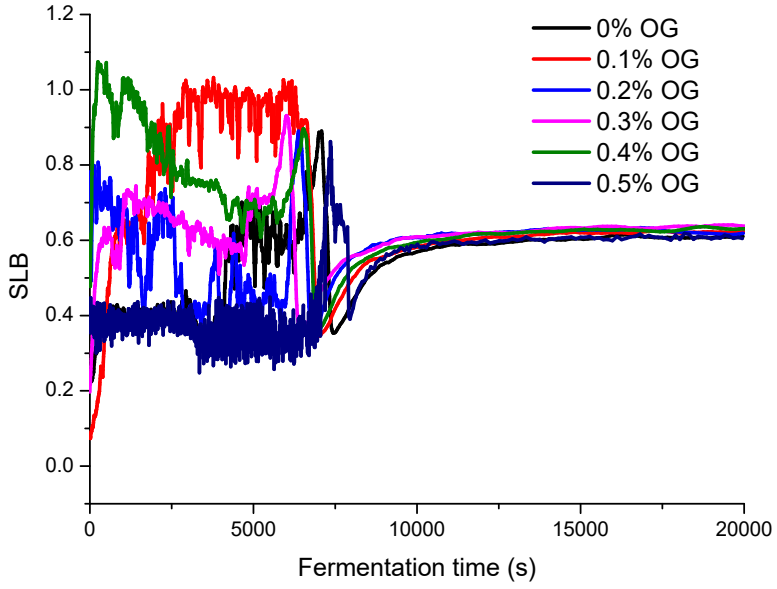


Fig. 3.4 The SLB value curve of set-type Yogurt with different OG concentrations during fermentation.

During the fermentation stage, the gel structure began to form. In the post-ripening stage, the gel structure was further changed, forming a "mature" yogurt system. Hemar et al. had successfully monitored the processing of fermented milk using the DWS method [124].

The value of SLB directly reflects the solid or liquid properties of the product [92]. SLB value is between 0 and 0.5, indicating that the system tends to be solid. A range of 0.5 to 1.0 indicates that the system tends to be liquid. As can be seen from Fig. 4.4, as a function of fermentation time, the SLB value of sample without OG was increased rapidly from 0.4 to 0.86 (118 min), and then decreased rapidly to 0.34 (124 min). At 180 min, the SLB value stabilized around 0.60, indicating that the set-type

yogurt was stable. According to the change of SLB value, the yogurt fermentation process went through the change process of "solid → liquid → solid → liquid". After the fermentation, the system was more inclined to be liquid. Interestingly, the SLB value of sample with 0.3% OG yogurt was 0.63, which was 0.03 higher than that of yogurt without OG, indicating that 0.3% OG addition increased the liquid behavior of the set-type yogurt.

Thus, the addition of oat β-glucan affects the change in the parameters of the technological process, namely, it helps to reduce the duration of fermentation by 1.5 hours due to the displacement of the gelation point.

3.1.4 Yogurt structure research results

As it is shown in Fig. 3.5 (a), a clear three-dimensional network structure is observed for yogurt without additives. When adding oat β-glucan, the structure of Yogurt changed significantly depending on its concentration. The addition of 0.1% OG partly destroyed the three-dimensional network of casein in the set-type Yogurt (fig. 3.5 (b)). while in Yogurt supplemented with 0.3% oat β-glucan, most of the three-dimensional casein network was destroyed and some spherical aggregate particles could be clearly observed (Fig. 3.5(c)).

In general, the three-dimensional network structure of Yogurt is considered to be formed by the aggregation of casein. As shown in Fig. 3.5 (a), the casein aggregates were clearly seen in the set-type Yogurt without OG. No lactic acid bacteria were observed (Fig. 3.5 (a)).

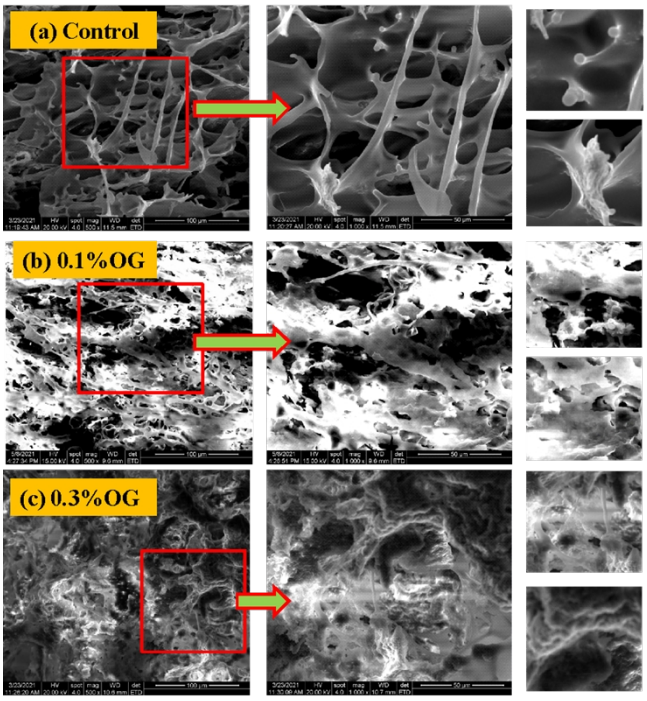


Fig. 3.5 Structure of Yogurt before and after adding OG; (a) - without OG addition; (b) - with 0.1% OG addition; © - with 0.3% OG addition.

The reason is possible that the lactic acid bacteria were damaged during vacuum freeze drying, which was used in the sample pre-treatment process. However, there were some high-brightness aggregates that adhered to the casein network. We hypothesized that these aggregates were the whey and exopolysaccharides [125].

The addition of 0.1% OG partly destroyed the three-dimensional network of casein in the set-type yogurt, suggesting that OG can affect the structure of Yogurt by its interaction with casein. When the OG concentration was 0.3%, the interaction of OG and casein in the set-type yogurt was more obvious, which is supported by the observation of the spherical structures. These structures were similar to the microstructures of the complex formed by OG and lactoferrin, previously reported by Yang et al. [123].

It is obvious that due to the addition of oat β-glucan, the interaction between casein particles was broken, and therefore three-dimensional aggregates cannot be formed during fermentation. These are probably aggregates formed by casein particles that aggregate on the oat β-glucan molecular chain during the Yogurt fermentation process. Oat β-glucan is an unbranched flexible polysaccharide polymer consisting of

linear chains of β-D-glucopyranosyl units. Some researchers have reported that there is an interaction between oat β-glucan and proteins such as lactoferrin, soy protein isolate, gliadin, and whey protein. Therefore, there are some protein bonds in its sugar chain. When oat β-glucan is added to Yogurt, casein can attach to the corresponding regions of the oat β-glucan chain and form protein clusters. During yogurt fermentation, these casein proteins are able to aggregate further, forming spherical structures.

3.1.5 Amino acid composition yogurt research results

Amino acids participate in metabolism, protein synthesis, and are part of nucleic acids. The biological role of amino acids is to maintain the pH of cell juice at a constant level. Some amino acids are involved in the biosynthesis of glycogen and hormones. The increased content of amino acids in the product gives it additional functional properties.

According to the results of the study of the amino acid composition in yogurt, 17 types of free amino acids were found, including 8 types of essential amino acids. In general, the content of essential amino acids in yogurt increased after the addition of oat β-glucan. The Yogurt group added with 0.2-0.4% OG was significantly higher than the control group ($P<0.05$). In the amino acid pattern recommended by FAO/WHO, the human AASs are Ile 4.0, Leu 7.0, Lys 5.5, Met+Cys 3.5, Phe+Tyr 6.0, Thr 4.0, Val 5.0. It can be seen from the figure 3.6 that the AAS of Ile, Lys, Phe+Tyr Thr and Val were higher than the standard value of the pattern spectrum, and the group added 0.3% OG was the highest. However, Met+Cys did not reach the standard value of the amino acid pattern spectrum. The closer it is to the amino acid pattern, the more balanced the nutritional value is. Therefore, it can be seen that the nutritional value of the experimental samples is not balanced, but the score of lysine, one of the main limiting amino acids in the human body, is higher, and the 0.2% and 0.3% groups are the highest.

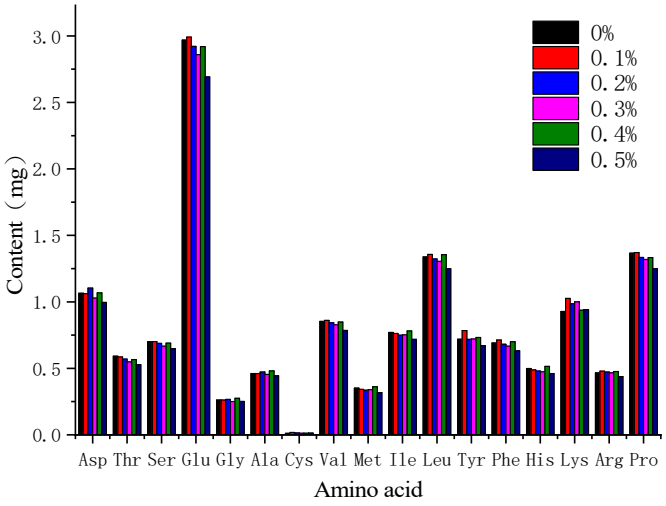


Fig. 3.6 Effects of different addition ratios of OG in set-type Yogurt on its amino acid composition

For control Yogurt, the amino acid composition shows that the dominant total AA is Glu (participation 7.0 %), followed by Pro (3.25 %), Leu (3.19 %), Asp (2.54 %), Lys (2.21 %), Val (2.03 %), Ile (1.83 %), Tyr (1.71 %), Ser (1.67 %), Phe (1.65 %), Thr (1.41 %), His (1.19 %), Arg (1.11 %), Ala (1.10 %). Met (0.84 %), Gly (0.63 %), Cys (0.03 %) are the lowest. But, the differences in total AA content between control yogurt and Yogurt fortified with OG are less noticeable ($P > 0.05$), although there are some differences.

The addition of OG did not change the type of amino acids, but affected the content of amino acids. Under the influence of OG, the content of most amino acids decreased, Gly, Ala, Tyr and Arg had little effect, but Lys increased.

In the amino acid model recommended by FAO / who, the AAS of human body are ile 4.0, Leu 7.0, Lys 5.5, Met+Cys 3.5, Phe+Tyr 6.0, thr 4.0 and Val 5.0 respectively. The results show that the AAS of ile, Lys, Phe+Tyr, thr and Val are higher than the standard value of the model spectrum, and the AAS of 0.3% OG group is the highest. The overall AA score was consistent with the recommended amino acid pattern. Thus, Yogurt with the addition of oat β-glucan can be considered a product for therapeutic and preventive purposes.

3.1.6 Fat-acid composition Yogurt research results

The area normalization method is used to calculate the relative percentage of fatty acids in general quantitative analysis. The peak areas of fatty acid methyl esters were determined by mass spectrometry of fatty acid methyl esters mixed standard solution. 18 kinds of fatty acid methyl ester standards were fully scanned by gas chromatography-mass spectrometry, and the mass spectra of the samples were retrieved by using the standard spectrum library.

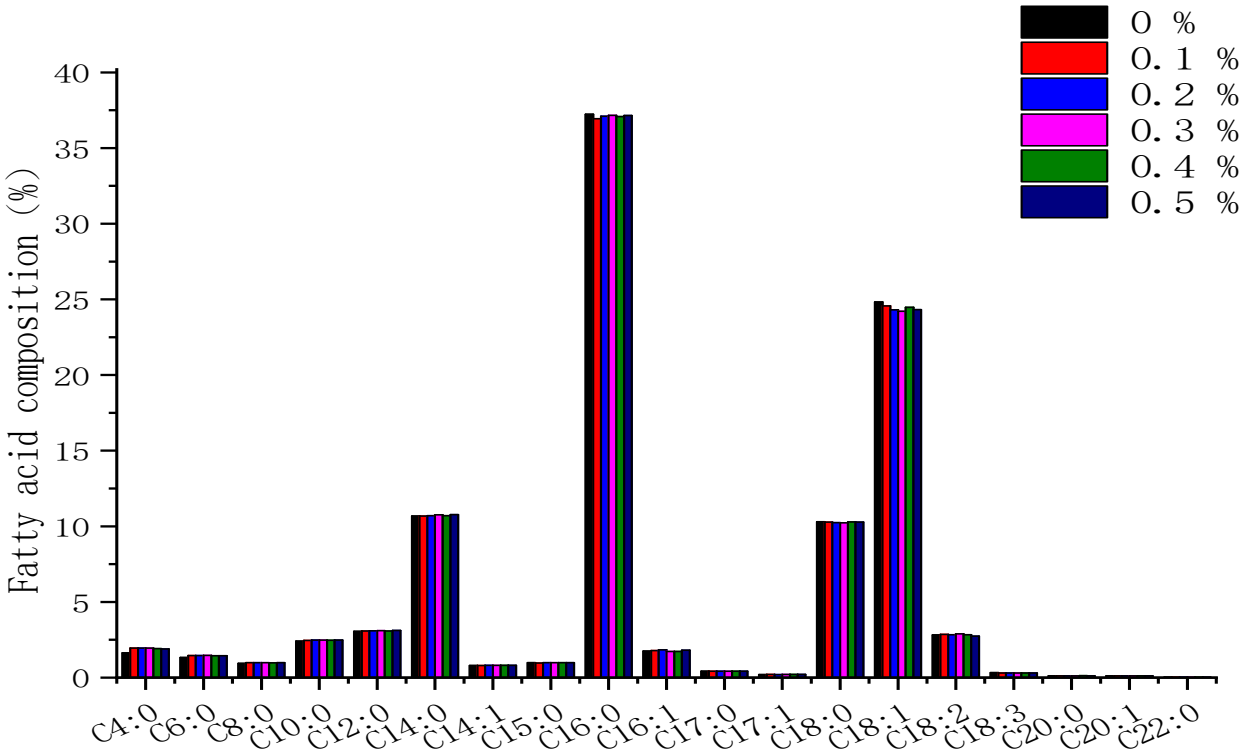


Fig. 3.7 Fatty acids content in Yogurts with different β-glucan content

Combined with the comparison of the chromatographic retention time of the mixed standard solution with fatty acid methyl ester, the fatty acid components in the fermented milk without OG and the fermented milk samples with different OG additions were determined. From Fig. 3.7. it can be seen that the concentration of β-glucan in yogurt does not affect the content of fatty acids in it.

For a detailed analysis, a comparison of Yogurts without additives and with a β-glucan content of 0.3% was carried out. The results of the study are presented in Fig. 3.8, 3, 9.

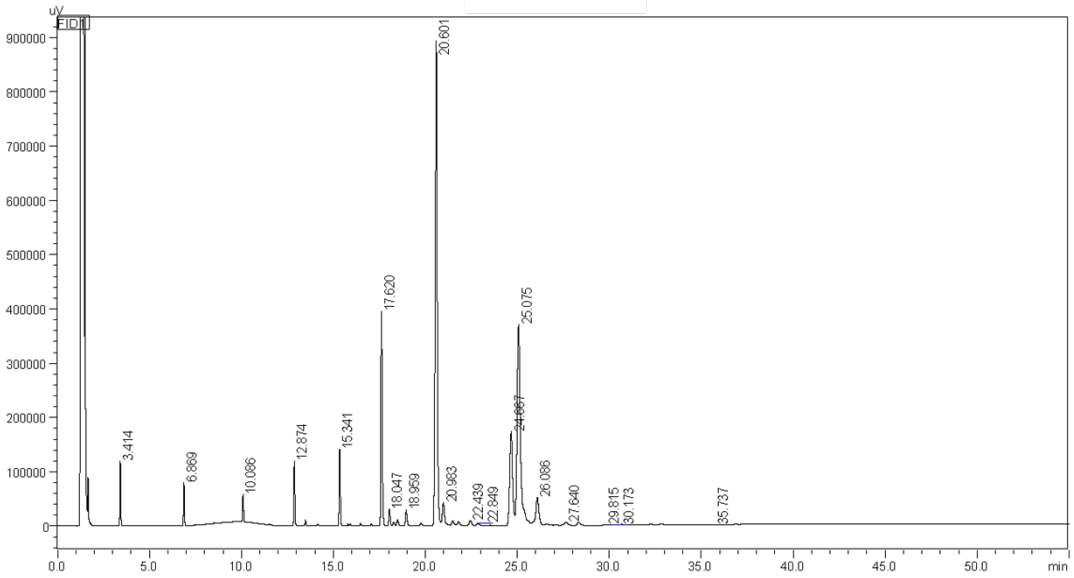


Fig. 3.8 Analysis of fatty acid profile of control set-type Yogurt without additives

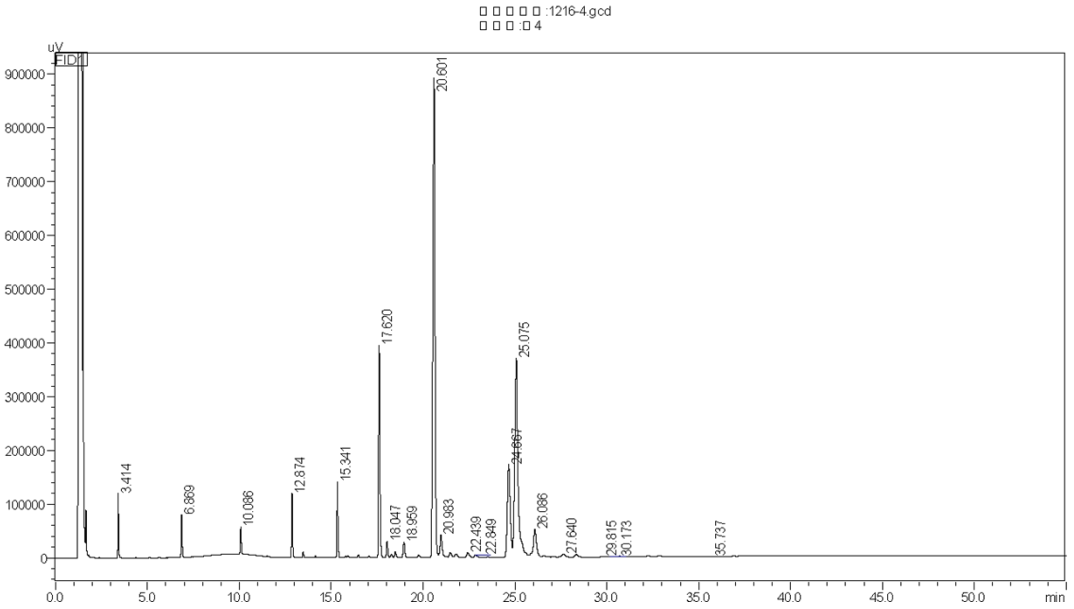


Fig. 3.9 Analysis of fatty acid profile of 0.3% OG set-type Yogurt

The presence of oat β-glucan affected the fatty acid content of yogurt (although the change was insignificant).

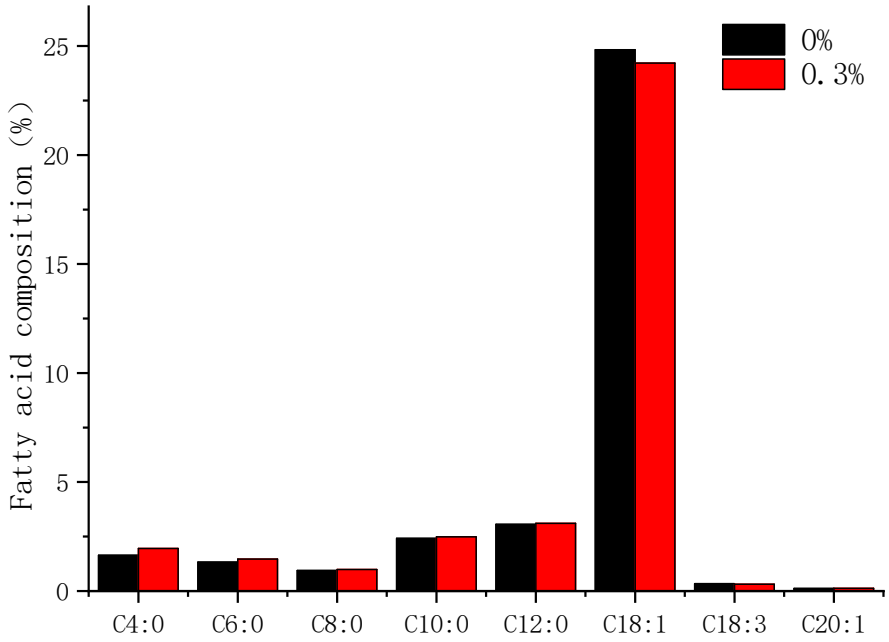


Fig. 3.10 Fatty acids with changes in control Yogurt and 0.3% OG set-type yogurt.

The content of C18:1 and C18:3 decreased; C4:0, c6:0, c8:0, c10:0 and c20:0 increased slightly. This may be due to the fact that OG is easy to combine with fat.

3.1.7 Determination of volatile substances in Yogurt

By using SPME-GC-MS to determine the volatile substances in solidified Yogurt, it can be seen from the table that OG fortification increases the types of volatile substances in solidified yogurt and helps Yogurt form a richer flavor.

The unique flavor of fermented milk comes from lactic acid produced by starter and many aromatic compounds naturally existing in milk and produced during fermentation. A variety of volatile compounds have been identified in Yogurt, which come from different chemical groups, such as acids, alcohols, ketones, esters, aldehydes, etc. [125]. However, not all flavor substances have an effect on the flavor, mainly because the concentration of each flavor substance is different. Only when the concentration exceeds the flavor threshold of fermented milk, it will play a role, thus affecting the main flavor of fermented milk.

Table 3.2 Comparison of volatile flavor components identified in 0% and 0.3% OG set-type Yogurt

Volatile substances	RT		Area Pct		Ref	
	Control	OG Yogurt	Control	OG Yogurt	Control	OG Yogurt
1	2	3	4	5	6	7
Acetic acid	2.828	-	0.9831	-	263	-
Silanediol, dimethyl-	3.7572	3.4617	19.2818	15.1409	2399	2399
Acetoin	3.9884	3.693	19.2818	1.6322	2006	2006
Cyclotrisiloxan, hexamethyl	6.4848	6.4934	19.2818	2.8191	79619	79617
2-Heptanone	8.8741	8.8656	19.2818	4.8197	7431	7426
Oxime-, methoxy-phenyl-_	10.1288	9.9832	19.2818	2.2157	24837	24837
Cyclotetrasiloxan, octamethyl-	14.3551	14.3551	19.2818	1.8293	141483	141483
2-Nonanone	18.5086	18.5129	19.2818	1.7752	19938	19938
Cyclopentasiloxa,decamethyl-	22.0112	22.0198	19.2818	1.0734	196316	196318
Butylated Hydroxytoluene	37.2893	37.2979	19.2818	3.3874	77555	77554
1,2-Benzenedicarboxylic acid	48.9191		19.2818		125896	

The End of the Table 3.2

1	2	3	4	5	6	7
Hexanedioic acid	54.3358	54.3401	19.2818	48.8771	196965	196965
(R)-(-)-2-Amino-1-propanol	-	1.8432	-	3.1826	-	903
Propane, 2-(ethenyloxy)-	-	1.9802	-	2.8503	-	1794
Decane, 2,2-dimethyl-	-	13.3574	-	1.4832	-	38346
Heptane, 2,2,4,6,6-pentamethyl-	-	15.1387	-	1.1686	-	38359
Decane, 2,3,8-trimethyl-	-	15.5241	-	1.972	-	48895
Hexane, 2,2,5-trimethyl-	-	16.4918	-	1.8929	-	12722
Octane, 2,6-dimethyl-	-	17.057	-	1.0366	-	19181
Decane, 3,7-dimethyl-	-	17.5965	-	1.0116	-	38338
Sulfurous acid, butyl	-	18.2089	-	0.8959	-	200447
Phthalic acid, isobutyl nonyl ester	-	48.9234	-	0.9362	-	182720
Hexanedioic acid, bis(2-ethylhexyl) ester	-	54.3401	-	48.8771	-	196965

In this study, the addition of 0.3% OG did not reduce the volatile flavor substances of set-type Yogurt (12 kinds of control Yogurt), but increased 11 kinds of volatile flavor substances, which helped to enhance the flavor of yogurt.

3.1.8 Results of analysis of physical and chemical indicators of Yogurt

3.1.8.1 Mass fraction of fat in Yogurt

Mass fraction of fat in the frozen Yogurt was studied. The results are shown in

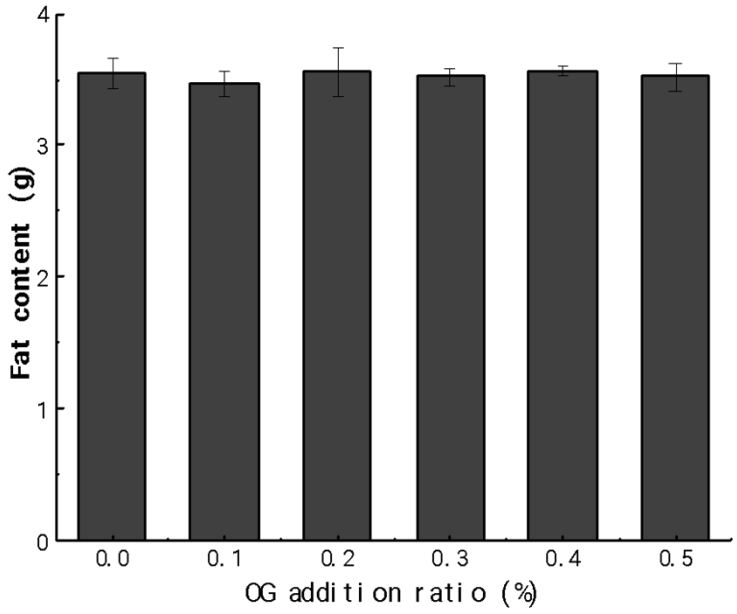


Fig. 3.11 Mass fraction of fat in Yogurt with adding of different ratio of oat β-glucan (g/100mg)

The fat content of the raw pure milk used in this study is 3.8g/100mL. Compared with the raw pure milk, the fat content of the coagulated Yogurt measured after the addition of OG decreases slightly. The analysis results show that the first reason is that the combination of OG with milk fat and milk protein makes it difficult to extract milk fat, and the second reason is that there may be small errors in the experiment.

3.1.8.2 Mass fraction of nonfat milk solids

Yogurt differs from other fermented milk drinks in its increased content of dry substances. The introduction of dietary fibers such as β-glucan affects their share. The results of the study of the mass fraction of nonfat milk solids in Yogurt are shown in fig. 3.12

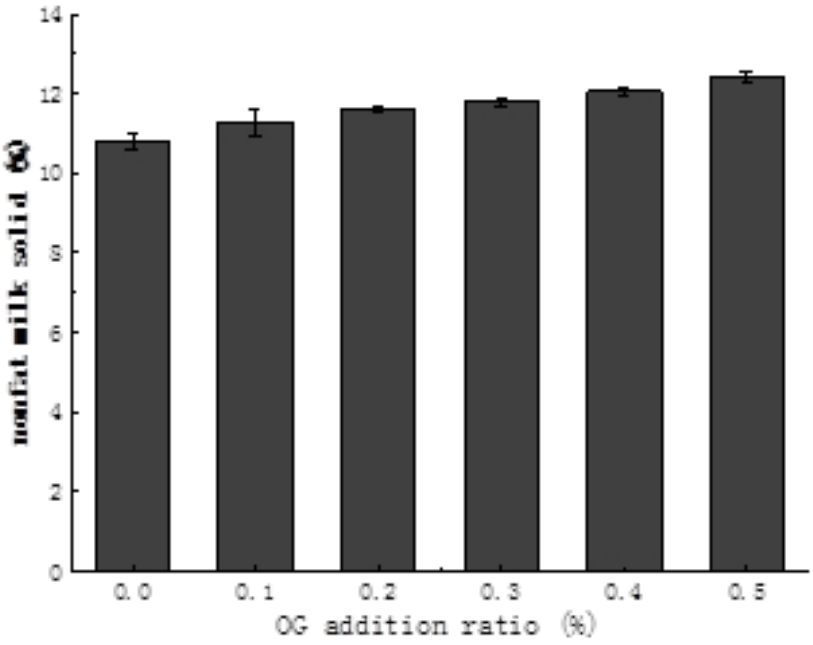


Fig. 3.12 Mass fraction of the nonfat milk solids content in set-type Yogurt with different OG addition

Mass fraction of the nonfat milk solids content of set-type Yogurt measured after the addition of OG increased slightly compared with set-type yogurt. The reason is that due to the increased amount of dietary fiber, the proteins of the oat β-glucan and the set type of Yogurt combine to form aggregates. With an increase in the content of dietary fibers, there is a better interaction between proteins and dietary fibers, as a result of which the mass fraction of nonfat milk solids in Yogurt increases.

3.1.8.3 Mass fraction of protein in Yogurt

Proteins determine the nutritional and biological value of the finished product, so their content is of great importance. Although proteins account for only 1/4 of body weight in the human body, and the daily need for these compounds in an adult is about 70-80 g, the value of proteins is great.

The complexity and diversity of protein macromolecules provided this group of substances, compared to other organic compounds, with the following functions: structural, catalytic, transport, protective, reserve, energy, reducing. Protein-containing products are often used in therapeutic and preventive nutrition. This is due to the fact that they perform a large number of functions in the body.

The measured values of the mass fraction of protein in Yogurt are presented in Fig.3.13.

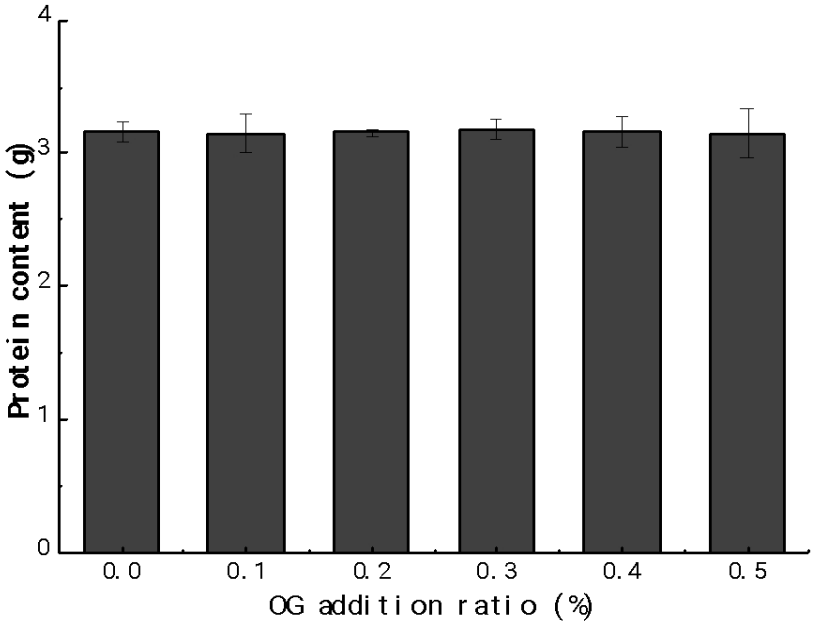


Fig. 3.13 Mass fraction of protein in set-type Yogurt with different OG addition, g/ml

After adding oat β-glucan, the protein content of Yogurt increases. This is because oat β-glucan contains a protein that can increase the protein content of yogurt. However, due to the formation of partial aggregates between OG and milk protein at high temperature (during sterilization), the aggregates are closely bound, which may affect the determination of protein.

The final determination results showed that the addition of OG had little effect on the protein content of fermented Yogurt, and the protein content met the requirements of the standard.

3.1.8.4. Acidity and pH of Yogurt

As can be seen from Fig. 3.14, the acidity of the set-type Yogurt samples increased with the fermentation time. When the OG concentration increased from 0 to 0.4%, the acidity of set-type yogurt gradually increased. At OG concentrations of 0.3% and 0.4%, the set-type yogurt had the highest acidity (about 84). But, at 0.5% OG concentration, the acidity decreased to 82, equal to that at OG concentration of 0,2%

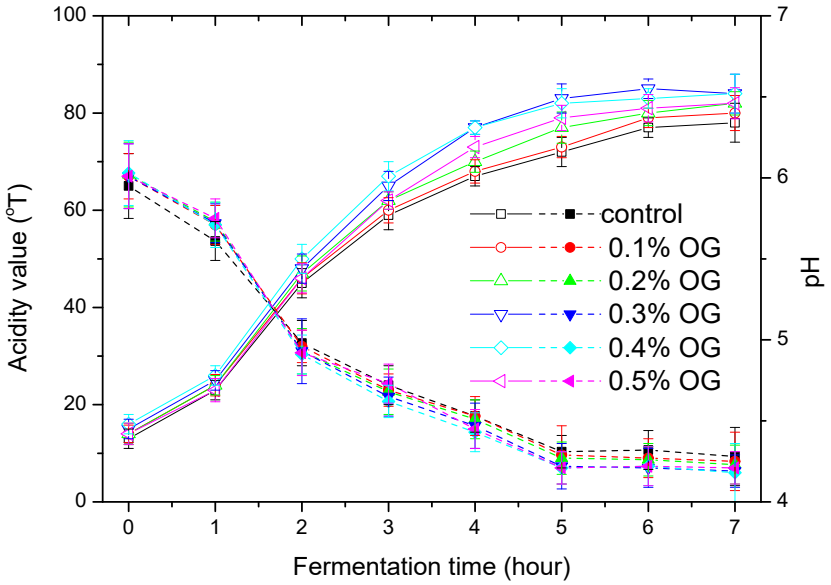


Fig. 3.14 Changes of titrated acidity and pH during fermentation of set-type Yogurt with and without OG addition

An opposite trend was observed for pH values, and reached the minimum (about 4.18) at 0.3% and 0.4% OG. All the pH values ranged from 4.18 to 4.28, which are within the normal ranges for set-type yogurts.

The acidity of set-type Yogurt with OG was slightly higher than that of without OG, and the pH was slightly lower. Thus, by adding oat β-glucan, you can reduce the fermentation time of the product, reducing the energy consumption of the process. The production technology of Yogurt with β-glucan is improved by reducing the duration of fermentation by 1.5 hours. This was because OG, as a prebiotic, can help the Lactobacillus to produce acid during the fermentation process.

This was because OG, as a prebiotic, can help the Lactobacillus to produce acid during the fermentation process. It is known that prebiotics selectively stimulate the growth and activity of the protective microflora of the human intestine and thus, improve its health. Yogurt enriched with oat β-glucan has prebiotic properties and can be used for nutrition of people with digestive system disorders.

3.1.8.5 Results of Yogurt color research

It can be seen from table 3.3 that the chromaticity L* shows a downward trend with the increase of OG content, and there is a significant difference between yogurt L* value with different OG content and the blank control group ($p< 0.05$).

Table 3.3 Color of control sample and 0.3% OG set-type Yogurt

L^*	a^*	b^*	ΔE	OG ratio (g/100g)
96.66 ± 0.454^{a}	-1.12 ± 0.024^{a}	3.9 ± 0.059^{c}	96.76 ± 0.449^{a}	0
96.3 ± 0.39^{a}	-1.16 ± 0.035^{a}	4.26 ± 0.105^{b}	96.24 ± 0.279^{bc}	0.10%
95.95 ± 0.102^{a}	-1.14 ± 0.007^{a}	4.88 ± 0.091^{a}	95.55 ± 0.162^{ab}	0.20%
95.87 ± 0.162^{a}	-1.15 ± 0.01^{a}	4.32 ± 0.121^{b}	95.9 ± 0.059^{abc}	0.30%
94.89 ± 0.349^{a}	-1.15 ± 0.009^{a}	4.97 ± 0.017^{a}	95.04 ± 0.364^{cd}	0.40%
94.3 ± 0.035^{b}	-1.45 ± 0.009^{a}	4.68 ± 0.102^{a}	94.5 ± 0.046^{d}	0.50%

Note: the angle a-d indicates whether there is a difference, the same indicates no difference, and the different indicates difference.

The yellowness value b* and redness value a* changed in varying degrees with the increase of OG content. ΔE decreased significantly with the increase of OG content Δ E value was lower than that of the control group. With the increase of OG content, the brightness value L* of yogurt becomes smaller and the color becomes darker; Redness value a* fades; the yellowness value b* becomes smaller and the color becomes lighter; chroma ΔE becomes smaller and darker. It can be seen from the above that the addition of OG with different content has different effects on the chromaticity, and the chromaticity value decreases with the increase of the addition amount.

High quality Yogurt should be milky white or slightly yellowish with an uniform color. As the color of OG itself is white with a little light yellow, the addition of OG may have a slight impact on the chromaticity of solidified yogurt. According to the test results, the addition of OG with different content had different effects on the chromaticity, and the chromaticity value decreased with the increase of the addition.

3.1.9 Results of microbiological research of Yogurt

3.1.9.1 The results of microbiological indicators of Yogurt

The analysis and treatment of 0,3% OG set-type Yogurt samples complied with GB 4789.3-2016, GB 4789.10-2016, GB 4789.4-2016, GB 4789.15-2016. At the same time the content of *E. scherichia coli*, *Staphylococcus aureus, Salmonella, Yeast, Mould* was determined. The results are shown in Table 3.4.

Table 3.4 The results of microbiological study of Yogurts with the addition of 0.3% oat β-glucan.

Test items	Microbial quantity	Test method
E. scherichia coli	Not detected	GB 4789.3-2016
Staphylococcus aureus	Not detected	GB 4789.10-2016
Salmonella	Not detected	GB 4789.4-2016
Yeast	Not detected	GB 4789.15-2016
Mould	Not detected	GB 4789.15-2016

Yogurt, obtained according to the improved technology as regards to the microbial indicators, does not differ from Yogurts produced according to the classical technology having fixed fermentation duration.

3.1.9.2 Quantity of lactic acid bacteria

According to the current standards in Ukraine and China, the content of lactic acid bacteria, which are the characteristic microflora of all fermented milk products, must be determined in Yogurts. The total number of lactic acid bacteria in control yogurt and yogurt with addition of 0,3% oat β-glucan within 24 hours after cooling was (2,39±0,1) × 10^7 CFU/ml and (3,45±0,3)×10^7 CFU/ml, that meets the requirements of the standards.

The results showed that the total number of lactic acid bacteria in yogurt with addition of 0,3% oat β-glucan was higher, than in control Yogurt without addition of

oat β-glucan. This is due to the fact that dietary fibers of β-glucan are probiotics, which are a good environment for the development of lactic acid microflora. These bacteria are among the few microorganisms and are considered to be safe for nutrition due to their prevalence in food and a role in healthy flora of the digestive tract. The increased content of “live” lactic acid microflora in Yogurt with addition of 0,3% oat β-glucan gives reasons to affirm that this product can be used for therapeutic and preventive nutrition.

3.2 Changes of the major quality indicators of Yogurt enriched with 0,3% OG during storage

3.2.1 Sensory evaluation results

Based on the represented research, addition of 0,3% oat β-glucan reduced fermentation time of set-type Yogurt, having reached the highest sensory score 148]. As a functional food ingredient, 0,3% oat β-glucan is important for enhancing the nutraceutical quality and physical characteristics of Yogurt. In addition, 0,3% oat β-glucan shows its effect on the growth of the starter culture. However, it is important to study the effect of 0,3% oat β-glucan on Yogurt quality, its sensory evaluation and physical characteristics during storage. The study was carried out for 21 days of storage, at the same time, organoleptic, physical and chemical and microbiological indicators of the finished product quality were investigated. The Yogurt sensory indicators are shown in Table 3.5.

Table 3.5. Sensory indicators of control Yogurt and Yogurt with addition of 0,3% oat β-glucan during storage.

Variables	Days	Yogurt samples	
		Control Yogurt	0.3% OG Yogurt
Color	1	8.12 ± 0.24^{f}	8.71 ± 0.46^{a}
	7	8.13 ± 0.34^{f}	8.68 ± 0.32^{b}
	14	8.18 ± 0.28^{de}	8.51 ± 0.48^{c}
	21	8.20 ± 0.14^{d}	8.50 ± 0.28^{c}
Structural state	1	26.05 ± 0.47^{h}	26.78 ± 0.63^{g}
	7	27.05 ± 0.12^{e}	27.32 ± 0.76^{c}
	14	27.02 ± 0.32^{f}	28.08 ± 0.92^{a}
	21	27.08 ± 0.28^{d}	27.98 ± 0.88^{b}
Texture	1	24.28 ± 0.39^{g}	24.97 ± 0.79^{e}
	7	24.92 ± 0.22^{e}	25.12 ± 0.82^{c}
	14	25.04 ± 0.28^{d}	25.98 ± 0.70^{a}
	21	24.88 ± 0.40^{f}	25.88 ± 0.70^{b}
Flavor	1	23.7±0.90c	24.67 ± 0.47^{a}
	7	23.1 ± 0.70^{d}	24.02 ± 0.32^{b}
	14	23.0 ± 1.20^{e}	24.12 ± 0.22^{b}
	21	23.1 ± 1.20^{d}	23.88 ± 0.28^{bc}

Note: the angle a-f indicates whether there is a difference, the same indicates no difference, and the different indicates difference.

All yogurts had mean sensory scores above 80, indicating the superiority of both the control Yogurt and Yogurt with the addition of 0,3% oat β-glucan throughout the storage period. The results showed that there was a statistically significant ($p<0,05$) difference in sensory characteristics between control Yogurt and Yogurt with the addition of 0,3% oat β-glucan. The control Yogurt had the highest acceptability value equal to 83,26 at 21 days of storage, while Yogurt with adding of 0,3% oat β-glucan had the highest acceptability value equal to 86,49 at 14 days of storage. It is obvious that adding 0,3% oat β-glucan had really a positive effect on yogurt acceptability, regardless of storage time.

3.2.2 The results of physical and chemical indicators change during storage of Yogurt enriched with 0,3% OG

As regards to the water-holding capacity values, a significant decrease was observed for the control Yogurt, while water-holding capacity value was not changed almost for Yogurt enriched with 0,3% OG (fig. 3.15(a)). This can be attributed to the hydration properties of oat β-glucan which affect the shelf life of the food product by preventing texture loss and avoiding the problem of syneresis.

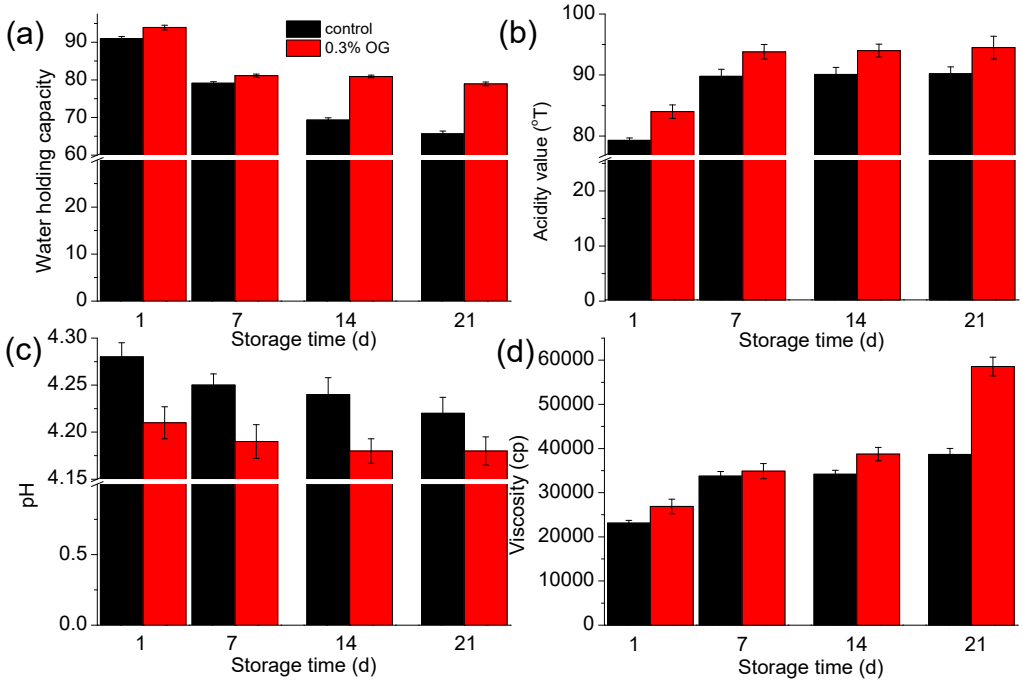


Fig. 3.15 Change in physical and chemical indicators of Yogurt quality during storage:a - water-holding capacity; b - acidity value; c – pH; d - viscosity.

As can be seen from Fig. 3.15(b), the acidity of both control Yogurt and 0.3% OG Yogurt significantly increased to 93.8±1.2 during 7 d of storage. And then (7-21 d of storage), no significant ($p<0,05$) changes were observed in acidity values. An opposite trend was observed for pH values and reached the minimum (about 4.21 for control yogurt, about 4.18±0.13 for 0.3% OG Yogurt) (Fig. 3.15(c)).

The decrease in pH during storage period might be mainly due to the utilization of OG by viable probiotic bacteria and production of lactic acid. In addition, small amounts of CO_2 and formic acid from lactose may also lead to the decrease in pH [127; 128]. All pH values ranged from 4.18 to 4.28, which are within the normal ranges for set-type yogurts.

Fig. 3.15(d) shows the changes in viscosity values during 21 d of storage. Significant differences were noted between control yogurt and 0.3% OG Yogurt. In general, the viscosity values increased throughout storage in concentrated and non-fat plain yogurt throughout storage [129; 69].

The increasing viscosity during storage could be due to the protein rearrangement and protein–protein contact. 0.3% OG Yogurt had higher viscosity than the control Yogurt, and the highest values were 58560±2120 cp at 21 d for 0.3% OG Yogurt. This change could be attributed to the fact that OG has the ability to entrap water within the product. On the other hand, the addition of 0.3% OG could improve the protein rearrangement and protein–protein.

For water holding capacity values, a significant decrease was observed for control Yogurt, while a slight decrease was observed for 0.3% OG Yogurt. This can be attributed to the hydration properties of OG, which have an impact on shelf life of food product, by preventing texture loss and avoiding syneresis problem. Thus, the optimal shelf life of yogurt with addition of oat β-glucan, which preserves its optimal physical and chemical properties, is 21 days. It is worth noting that the duration of storage of yogurts produced according to classical technology, without use of artificial stabilizers, is 7-14 days. It means that addition of oat β-glucan contributes to the improvement of the existing technology.

3.2.3 Total lactic acid bacteria quantity during storage of Yogurt

The content of lactic acid bacteria was counted by determining of colonies quantity. Live and active probiotic bacteria are considered to be beneficial for human health [130; 131]. OG is considered to be a prebiotic and may contribute to the activity of probiotic bacteria. So, the viability of probiotic bacteria in yogurts was checked. As shown in Fig. 3.16, during the whole storage period, the content of probiotics decreased, which was only (0.63±0.05) $\times 10^{7}$ cfu/mL at 21 d.

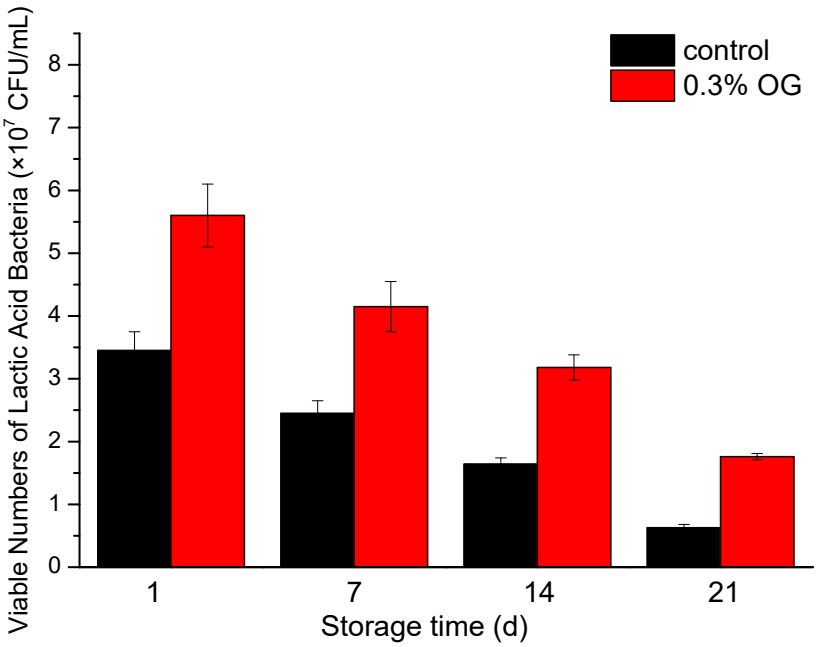


Fig. 3.16 The viability of probiotic bacteria of Yogurts during the storage of yogurts

However, 0.3% OG yogurt contained significantly more living probiotic bacteria compared to the control one ($p<0,05$), throughout the whole cold storage period. The viability of probiotic bacteria of 0.3% OG Yogurt at 14 d ((3.18±0.2) ×10 ^{7} cfu/mL) was only slightly lower than that of control yogurt at 1 d ((3.45±0.3) ×10 ^{7} cfu/mL). Similar results have also been reported [84]. This fully demonstrates that the addition of OG has a protective effect on probiotics in Yogurt. The survival of yogurt microbiota in 0.3% OG Yogurt will be beneficial for human health.

Thus, therapeutic-preventive properties of Yogurt with addition of 0.3% OG are maintained within 21 days of storage.

3.2.4 Texture parameters research results during storage of yogurt

The textural parameters of yogurt are important for its consumer properties, as they can simulate their breakdown occurring in the mouth. Results of texture profile analysis (TPA) of yogurts, including hardness (N), consistency, cohesiveness, and springiness (%) are summarized in Table 3.6.

Table 3.6 TPA results in control Yogurt and Yogurt with adding of 0.3% OG

Test	Days	Yogurt samples	
		Control Yogurt	0.3% OG Yogurt
1	2	3	4
Hardness	1	21.14±0.21^{g}	22.85±0.26^{f}
	7	24.77±0.14^{e}	28.36±0.04^{d}
	14	28.76±0.32^{d}	35.99±0.21^{c}
	21	40.99±0.22^{b}	50.45±0.16^{a}
Adhesiveness	1	-8.23±0.03^{d}	-9.15±0.06^{c}
	7	-9.18±0.12^{c}	-10.21±0.15^{b}
	14	-9.25±0.20^{c}	-11.37±0.07^{a}
	21	-10.39±0.06^{b}	-11.85±0.22^{a}
Gumminess	1	9.26±0.31^{f}	13.78±0.19^{d}
	7	12.79±0.36^{e}	13.81±0.53^{d}
	14	14.71±0.17^{c}	19.82±0.25^{b}
	21	21.54±0.29^{a}	13.78±0.33^{d}
Cohesiveness	1	0.43±0.03^{c}	0.43±0.06^{c}
	7	0.41±0.05^{cd}	0.44±0.02^{c}
	14	0.51±0.08^{ab}	0.57±0.03^{a}
	21	0.53±0.03^{ab}	0.59±0.06^{a}
Chewinness	1	8.75±0.33^{g}	9.46±0.47^{f}
	7	11.39±0.29^{e}	12.87±0.66^{d}
	14	14.06±0.23^{c}	13.77±0.54^{cd}
	21	20.72±0.43^{a}	18.99±0.38^{b}
Springiness	1	0.94±0.03^{c}	0.92±0.06^{e}
	7	0.95±0.07^{b}	0.93±0.03^{d}
	14	0.95±0.06^{b}	0.95±0.05^{b}
	21	0.96±0.07^{a}	0.97±0.04^{a}

Note: the angle a-g indicates whether there is a difference, the same indicates no difference, and the different indicates difference

For both the control Yogurt and Yogurt with mass concentration of 0,3% oat β-glucan, the hardness of the Yogurts was improved, while adhesiveness and gumminess values decreased during storage. The addition of 0,3% oat β-glucan affected the textural characteristics of Yogurts. The evaluation index of Yogurt is mainly the hardness. The hardness of 0.3% OG Yogurt was higher than that of control Yogurt, especially after 21 d of storage (50.45±0.16 for 0.3% OG Yogurt, 40.99±0.22 for control Yogurt). The higher hardness values could be attributed to ability of the OG to entrap water, and maintain structure within the product [69; 132].

In contrast, the adhesiveness of 0.3% OG Yogurt was lower than that of control Yogurt, especially after 21 d of storage (-11.85±0.22 for 0.3% OG Yogurt, -10.39±0.06 for control Yogurt). The decrease in adhesiveness may be attributed to that OG can reduce adherence of Yogurt with teeth during chewing. 0.3% OG Yogurt had a slight increase in gumminess as compared to control Yogurt. And, during storage, an increase in gumminess was observed, which might be due to increased hardness of the Yogurts. A similar behavior was found for cohesiveness, which can be attributed to the fact that OG has the ability to form new gel structures with casein, effectively intercepting and entrapping water within the yogurt. No significant differences were obtained for springiness values between control Yogurt and 0.3% OG Yogurt. Thus, the results of textural profile tests showed appropriate hardness, less gumminess, less adhesiveness and less destruction in 0.3% OG Yogurt, giving firm and creamy texture that was near to creamy mouth feel of full fat yogurt, improving the mouth feel characteristics thereby enhanced sensory appeal for the product in which incorporated.

Conclusion to Section 3

1. OG can be used as a functional ingredient during producing of yogurts. When 0.3% OG was added, the highest score of sensory evaluation was about 85, the set-type Yogurt had the highest WHC of 94.67%. The acidity of set-type yogurt with OG was slightly higher. The addition of OG has a slight effect on chromaticity.

2. Rheological evaluation results showed that the fermentation process went through the changes as follows: solid → liquid → solid → liquid. The addition of 0.3% OG decreased the fermentation time of set-type yogurt by about 16 minutes. The technology for production of Yogurt with OG is improved by reducing the duration of fermentation.

3. The results of microstructure study showed that the addition of OG destroyed the three-dimensional network structure of Yogurt, and some spherical aggregate particles could be clearly observed when OG was added. That is, the structure, which is characteristic for finished Yogurt, is formed almost at the beginning of the fermentation process.

4. The addition of OG had a little effect on the fatty acid and amino acid composition of set-type Yogurt. SPME-GC/MS analysis confirmed that the addition of OG was helpful to increase the types of volatile flavor compounds in the set-type Yogurt.

5. Storage experiments showed that the acidity value and pH reached their maximum values at 7 d of storage, and no significant ($p<0,05$) changes were observed after 7 d. The addition of 0.3% OG has a protective effect on probiotics in Yogurt. The textural characteristics of Yogurt were affected by the addition of 0.3% OG, leading to decreased adhesiveness, but enhanced hardness and gumminess, throughout storage. The sensory evaluation results indicated that 0.3% OG Yogurt had the highest acceptability value of 86.49 at 21 d of storage.

6. The addition of oat β-glucan contributes to the improvement of the existing technology of Yogurt production due to the intensification of the process, improvement of the main physicochemical indicators of the quality of the product during fermentation.

SECTION 4 THE EFFECT OF OG ON THE IN VITRO DIGESTIVE PROPERTIES OF SET-TYPE YOGURT AND BLOOD LIPID METABOLISM IN MICE

4.1 The indicators affecting biological functions of Yogurt

Yogurt is a kind of dairy product fermented by symbiotic cultures of lactic acid bacteria, thermophilic lactic acid microorganisms and bifidobacteria. It is popular among consumers due to its high nutritional and biological value, which is formed due to the metabolic activity of these microorganisms and the chemical composition of normalized mixture enriched with oat beta-glucan during the fermentation process.

It is considered to affect some regulatory systems (such as glucose and lipid metabolism), lower blood pressure, promote insulin secretion, and maintain body weight, etc [133; 134; 135]. However, these nutritional and biological functions are closely connected with its digestion process. The bioactive peptides, existing in the amino acid sequence of protein, can be released and activated only through enzymatic hydrolysis during the digestion process. Especially, some branched-chain amino acids, which can influence several postprandial metabolic responses, are present in digested dairy products [93].

OG is an important soluble dietary fiber, consisting of linear chains of β-D-glucopyranosyl units linked with (1→3) and (1→4) linkages [136]. It has many biological activities, such as enhancing antioxidant activity, reducing blood lipid, preventing cardiovascular diseases, regulating gastrointestinal environment and cholesterol level in the body [31; 39; 48; 137]. OG is also well-known for its thickening, stabilizing, emulsifying and gelling properties to maintain the stability of ingredients [138].

More importantly, it has been found that OG has prebiotic properties and could selectively enhance activity and raise growth of probiotic bacteria (such as lactobacilli and bifidobacteria). So, OG can be used as a texturizer, fat replacer, and prebiotic in enhancing the physical characteristics and nutraceutical qualities of Yogurt [84; 85;

139]. According to our previous study, the addition of 0.3% (w/w) OG could maximize the quality characteristics of set-type Yogurt, and shorten the fermentation time [140].

Due to the addition of OG, the digestion characteristics (e.g., the degree of hydrolysis) of set-type Yogurt and the structural and functional properties of proteins or peptides after digestion (e.g., molecular weight, charge and hydrophobicity, etc.) may be changed. However, there are relatively few studies on the effect of 0.3% OG on the in vitro digestion characteristics of set-type Yogurt, which will limit the application of this type of yogurt.

In vitro digestion models have been designed to study the structural changes, digestibility/degradation, and digestion characteristics of food components under simulated gastrointestinal conditions [141]. Through these models, the digestion characteristics of food systems, such as plant-, dairy-, and emulsion-based foods, has been successfully studied.

The effect of 0.3% OG on the in vitro digestion characteristics of set-type Yogurt by an in vitro gastrointestinal (GI) model was studied. The proportion of yogurt soluble proteins and peptides after digestion was measured. The microstructural morphology and particle size of Yogurts after digestion were characterized by optical microscopy and dynamic light scattering, respectively. And, their antioxidant activities and inhibition of cholesterol solubilisation into micelles were also evaluated.

4.2 Effect of OG on in vitro digestion characteristics of Yogurt

4.2.1 In vitro protein digestibility

Fig. 4.1 shows the proportion of yogurt soluble proteins and peptides after the buccal, gastric and duodenal digestion phases. On the whole, the amount of soluble proteins and peptides increased during digestion. In particular, the amount increased significantly during gastric and duodenal digestion phase.

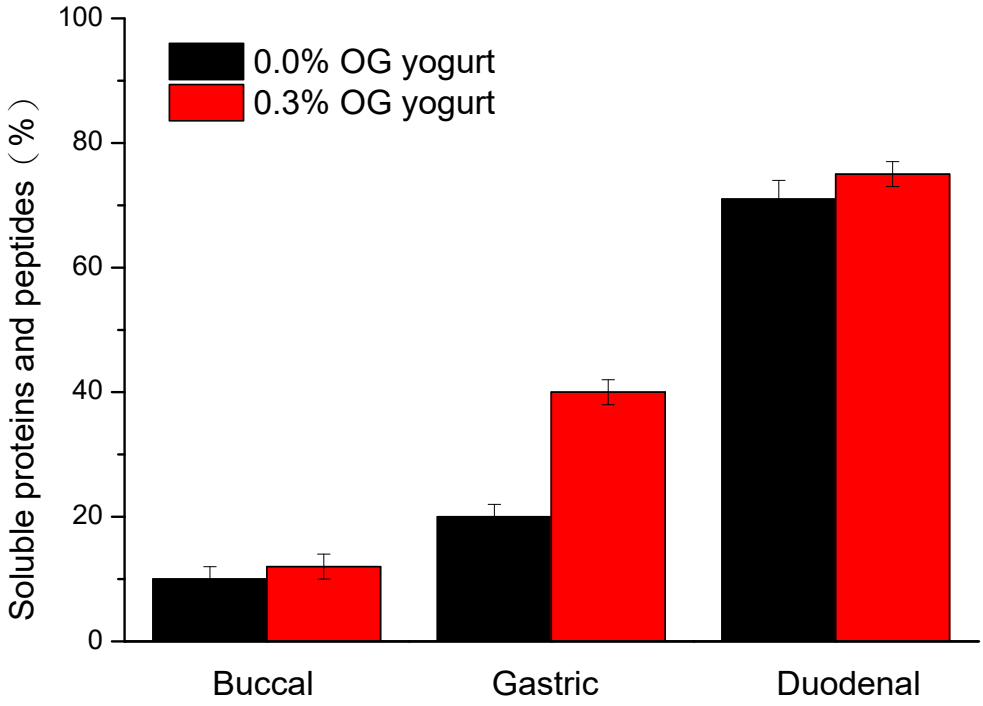


Fig. 4.1 Soluble proteins and peptides (%) after the buccal, gastric and duodenal digestion phases.

The amount of soluble proteins and peptides increased slightly in the course of buccal digestion phase. But, after the simulated gastric digestion, there was a significant ($p < 0,05$) increase (25% for control Yogurt, 40% for 0.3% OG Yogurt) of the soluble proteins.

This result is agreed with the study by Rinaldi [93], who reported that due to the presence of OG, Yogurts exhibited faster proteolysis, thus leading to the lower release behavior of large peptides while higher percentage of free amino acids. After the simulated duodenal digestion, the soluble proteins and peptides were slightly higher for 0.3% OG Yogurt than for the control Yogurt. Thus, OG addition does influence the in vitro protein bioaccessibility in Yogurt, especially after the gastric phase. It was reported that some polysaccharides, such as gum arabic, low-methylated pectin, and xylan, could inhibit β-lactoglobulin digestibility, due to the formation of protein-polysaccharide complexes [142]. The difference might be attributed to the different physicochemical characteristics of polysaccharides. This also may indicate that OG is

more suitable for use as a functional food ingredient in enhancing the nutraceutical quality of yogurt compared with other polysaccharides.

4.2.2 Microstructure and particle size during digestion process

To gain more structural insights, the microstructural morphologies of particles for control Yogurt and 0.3% OG yogurt after buccal, gastric and duodenal digestion were observed by optical microscopy (Fig. 4.2).

Buccal digestion has little effect on the structure of yogurts. After buccal digestion, the microstructure of control yogurt showed a clear three-dimensional protein network structure (Fig. 4.2 (a1)). The microstructure of 0.3% OG Yogurt also showed a denser three-dimensional network structure (Fig. 4.2 (b1)).

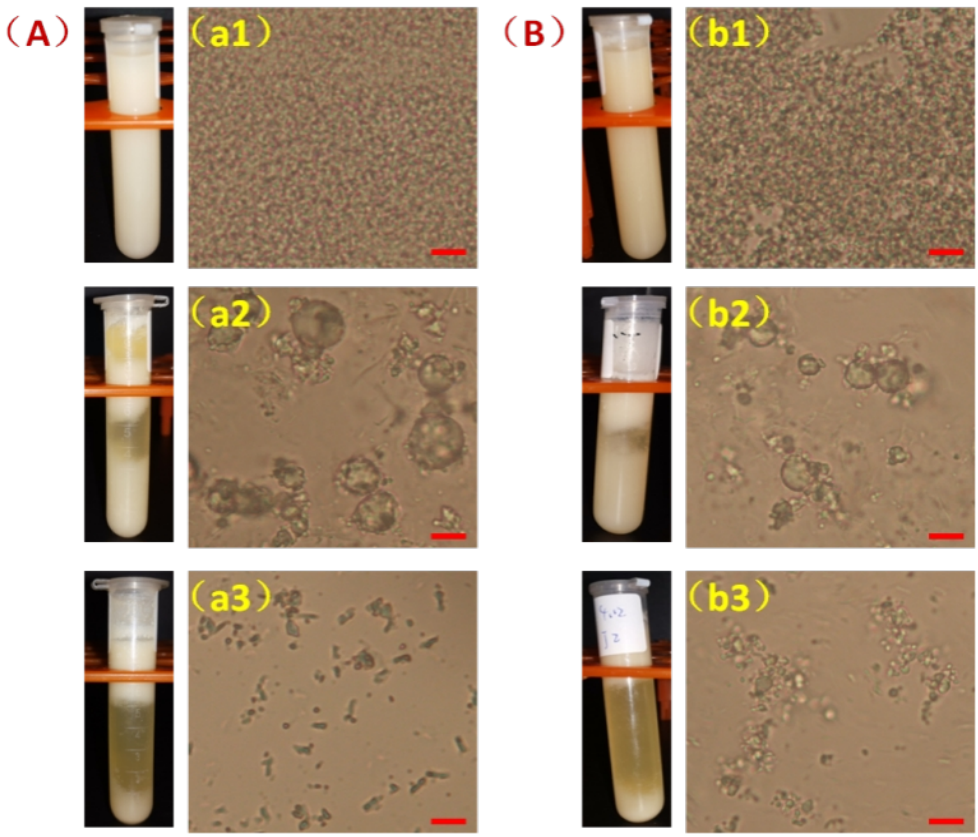


Fig. 4.2 The microstructure of Yogurts after digestion. (A) control Yogurt; (B) 0.3% OG Yogurt. (a1, b1) after buccal digestion; (a2, b2) after gastric digestion; (a3, b3) after duodenal digestion.

After buccal digestion, the microstructure of control Yogurt showed a clear three-dimensional protein network structure (Fig. 4.2 (a1)). In general, during fermentation, casein aggregates form a three-dimensional network in yogurt [143].

The microstructure of 0.3% OG yogurt also showed a denser three-dimensional network structure (Fig. 4.2(b1)). This could be due to the network structure formed by OG or the complexes dominated by OG-casein interactions, in good agreement with in our previous work (studied by scanning electron microscopy) [140].

After gastric digestion, large spherical vesicles were formed for both control Yogurt and 0.3% OG Yogurt (Fig. 4.2 (a2, b2)). In general, the main role of pepsin is to enzymatically hydrolyze proteins into large peptides. These spherical vesicles should be protein aggregation caused by gastrointestinal digestion. Interestingly, the particle sizes of spherical vesicles for 0.3% OG yogurt were smaller than those for control yogurt. As far as we know, the smaller particle sizes of spherical vesicles were observed for the first time by optical microscopy.

This indicated clearly that the presence of 0.3% OG caused a fast enzymatic hydrolysis, leading to a significant ($p<0,05$) increase in the proportion of low-molecular-mass peptides. An earlier investigation even pointed out that after gastric digestion, intact dairy proteins remained in the control yogurt whereas less in Yogurts enriched in pectin/OG, as measured by sodium dodecyl sulfate polyacrylamide gel electrophoresis analysis [93].

After duodenal digestion, spherical vesicles disappeared and some small flake structure occurred instead for control Yogurt (Fig. 4.2 (a3)). This indicates complete digestion of control Yogurt. It means that change of technological parameters in classical technology of yogurt production, due to the addition of oat β-glucan, does not have any negative effect on its ability to be absorbed by the body.

4.2.3 Particle size and PDI of Yogurts

It can be seen from Table 4.1, for control Yogurt, the average particle size was 7.2 μm with PDI of 0.73 after buccal digestion. This indicated that particles were

aggregated with each other. After the gastric digestion, its particle size was decreased to 5.3μm, and PDI also decreased to around 0.63. After the intestinal digestion, its particle size was about 1.2 μm, with a low PDI of 0.31.This directly indicated that Yogurt was completely digested. Thus, this product can be included in the diet of patients having digestive system disorders.

The study results of the effect of particle size on the polydispersity of yogurts are presented in table 4.1.

Table 4.1. The study results of the effect of particle size on the polydispersity of Yogurts after different digestion phases.

Digestion phases	Samples	Average particle size (d/nm)	Polydispersity (PDI)
Buccal digestion	Control Yogurt	7211 ± 45^{b}	0.73 ± 0.09^{b}
	0,3% OG Yogurt	8327 ± 52^{a}	0.82 ± 0.11^{a}
Gastric digestion	Control Yogurt	5319 ± 122^{c}	0.63 ± 0.08^{c}
	0,3% OG Yogurt	3427 ± 53^{d}	0.53 ± 0.04^{d}
Intestinal digestion	Control Yogurt	1253 ± 22^{f}	0.31 ± 0.02^{e}
	0,3 OG Yogurt	1503 ± 42^{e}	0.39 ± 0.05^{e}

Note: the angle a-f indicates whether there is a difference, the same indicates no difference, and the different indicates difference

Previous studies of some researchers showed [144] that some compounds, such as short chain fatty acids, are easier to hydrolyse by lipases under the role of bile salts. In comparison, there were still some spherical particles with small particle sizes in 0.3% OG Yogurt, and they were connected with each other (Fig. 3.13 (b3)).

It is obvious, these connected spherical particles were related to the presence of OG, as OG can interact with proteins and peptides and resist hydrolysis by lipases to some extent [145].

This may play an important role in stabilizing and reinforcing the functional properties of these peptides. In fact, OG has been used in the food, cosmetic and pharmaceutical fields to deliver bioactive compounds [146].

It can be seen from Table 4.1, for control yogurt, the average particle size was as high as 7.2 μm with PDI of 0.73±0.09 after buccal digestion. This indicated that particles were aggregated with each other. After the gastric digestion, its particle size was decreased to 5.3μm, and PDI also decreased to around 0.63±0.08. After the intestinal digestion, its particle size was about 1.2 μm, with a low PDI of 0.31±0.02. This directly indicated that Yogurt was completely digested.

Differently, the particle size after buccal digestion was higher for 0.3% OG Yogurt than for the control yogurt. This was obviously related to the addition of OG. But, after the gastric digestion, the particle size was decreased to 3.4μm, and PDI also decreased to around 0.53±0.04.

This again indicated that 0.3% OG addition caused an increase in the proportion of peptides during digestion, as observed by optical microscopy. The fast protein digestion for yogurts with OG could indicate a phase separation phenomena between OG and protein. We hypothesize that in gastric solution, digestion conditions favor the phase separation, forming a "micro-reactor" among OG, Yogurt proteins, and enzymes. The similar result was also obtained by other researchers [93].

The enzymes and yogurt proteins are in close contact in the micro-reactor, thus facilitating the hydrolysis, leading to small particle sizes. After the intestinal digestion, the particle size was slightly higher (about 1.5μm) than that of control Yogurt, with a higher PDI of 0.39±0.05. This may be due to the undigested OG.

4.2.4 Antioxidant activities of yogurt with OG

Yogurt is an important source of food derived protein. In the digestion process, yogurt can release some functional active substances from milk protein, especially some bioactive peptides with good antioxidant properties.

In order to evaluate the antioxidant activity of functional foods, DPPH assay and ABTS assay are often used. Two methods were used to evaluate the antioxidant activity of set-type yogurt throughout digestion.

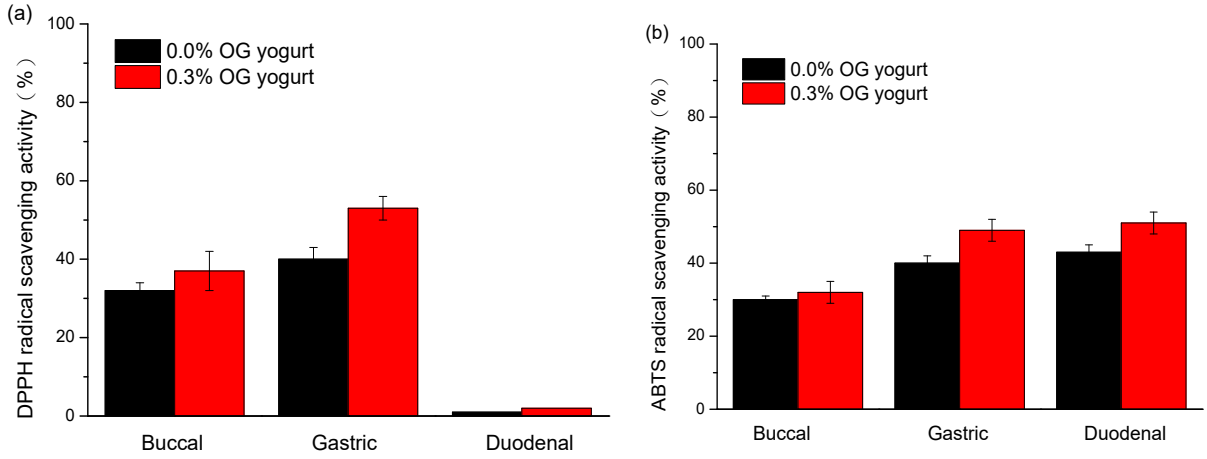


Fig. 4.3 DPPH (a) and ABTS (b) radical scavenging capacity of Yogurts after the buccal, gastric and duodenal digestion phases.

Fig. 4.3 (a) presents the DPPH radical scavenging assay results of control Yogurt and 0.3% OG Yogurt throughout digestion. Both control yogurt and 0.3% OG Yogurt showed certain DPPH radical scavenging activity after buccal digestion, moreover, 0.3% OG Yogurt had stronger antioxidant capacity. For the DPPH radical scavenging activity of control yogurt, it was increased by 25% after the gastric digestion compared with that after buccal digestion.

The results of ABTS assay were similar to DPPH assay (Fig. 4.3(b)). After buccal digestion, both control yogurt and 0.3% OG Yogurt showed ABTS radical scavenging capacity.

For the DPPH assay, as shown in Fig. 4.3 (a), after buccal digestion, both control Yogurt and 0.3% OG Yogurt showed certain DPPH radical scavenging activity. Moreover, 0.3% OG yogurt had stronger antioxidant capacity, which can be attributed to the antioxidant activity of OG. It has been reported that OG inhibits significantly the fat oxidation of low-fat beef patties [147].

After the gastric digestion, it was increased by 25% compared with that after buccal digestion, indicating that some active components were produced during gastric

digestion. This result was similar to other report [31]. Interestingly, 0.3% OG Yogurt exhibited higher DPPH scavenging ability (43%) than the control. The result clearly indicated that the presence of OG promoted the yogurt protein to produce more antioxidant components, giving enhanced antioxidant properties.

After the intestinal digestion, the DPPH radical scavenging activity of yogurts can not be detected. In the study, DPPH was dissolved in methanol, which is suitable for the determination of hydrophilic compounds, not suitable for lipophilic compounds [148; 149]. It was assumed that the lipophilic compound after the intestinal digestion may interfere with the determination. Detailed reasons need further study.

The results of ABTS assay were similar to DPPH assay for buccal and gastric digestion (Fig. 4.3(b)). In comparison, the ABTS radical scavenging capacity of yogurts can be detected after the intestinal digestion. It was further improved (43% for control Yogurt and 59% for 0.3% OG Yogurt). Clearly, the antioxidant activity of Yogurt was further improved.

4.2.5 Effect of yogurt with OG on in vitro cholesterol micelles

Dietary cholesterol needs to be digested by various enzymes under salivary and gastrointestinal conditions to form micellar solution with triglycerides, phospholipids and bile acids before it can be transported into intestinal mucosal cells. The cholesterol lowering effect was evaluated by an in vitro cholesterol micelle model.

For control Yogurt, the inhibition of cholesterol solubilisation into micelles increased gradually (15.7% for buccal digestion, 17.2% for gastric digestion, and 19.1% for intestinal digestion) (Fig. 4.4). Clearly, this could be related to the released bioaccessible peptides and amino acids.

Interestingly, compared with that of control Yogurt, the inhibition of cholesterol solubility of 0.3% OG Yogurt showed no differences after buccal digestion but significantly ((P<0,05) increased after gastrointestinal digestion (21.3% for gastric and 22.7% for intestinal digestion).

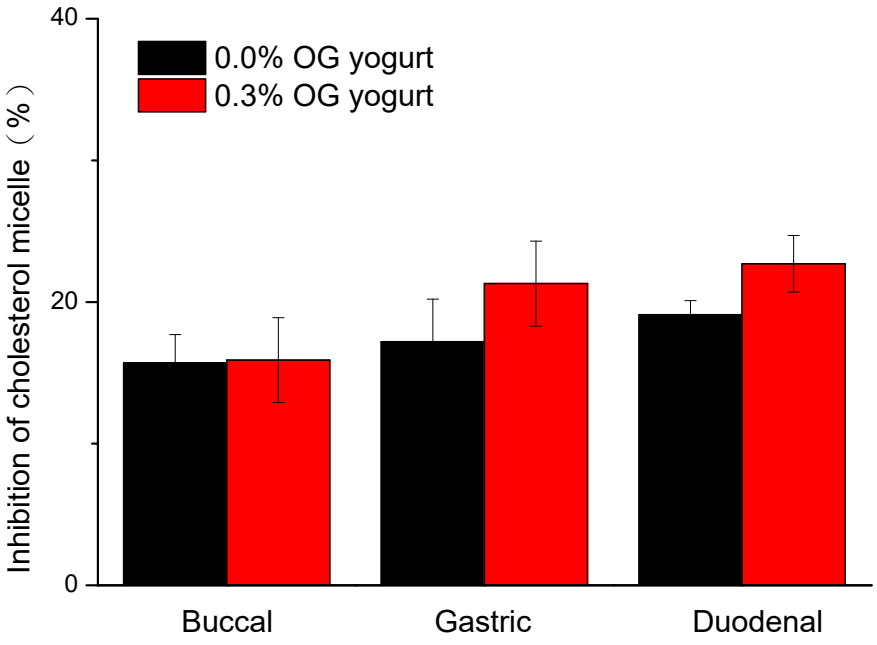


Fig. 4.4 Inhibition percent of the micellar cholesterol solubilisation in yourts after the buccal, gastric and duodenal digestion phases.

In most developed countries and a few developing countries, cardiovascular diseases are considered to be the first leading cause of death and morbidity, and a major contributor to greatly reduced quality of life [150; 151]. Prevalent cases of total cardiovascular diseases nearly doubled from 271 million in 1990 to 523 million in 2019, and the number of cardiovascular diseases deaths increased steadily from 12.1 million in 2020 [151]. The risk of cardiovascular diseases can be reduced 2%-3% by every 1% decrease of serum total cholesterol. Dietary cholesterol needs to be digested by various enzymes under salivary and gastrointestinal conditions to form micellar solution with triglycerides, phospholipids and bile acids before it can be transported into intestinal mucosal cells [152]. So, the cholesterol lowering effect was evaluated by an in vitro cholesterol micelle model [153].

For control Yogurt, the inhibition of cholesterol solubilisation into micelles increased gradually throughout digestion (15.7% for buccal digestion, 17.2% for gastric digestion, and 19.1% for intestinal digestion) (Fig.3.15). Clearly, this could be related to the released bioaccessible peptides and amino acids.

Compared with that of control Yogurt, the inhibition of cholesterol solubility of

0.3% OG yogurt showed no differences after buccal digestion but significantly ($p<0,05$) increased after gastrointestinal digestion (21.3% for gastric digestion and 22.7% for intestinal digestion). These can be related to the presence of OG. On the one hand, OG could influence the type and conformation of amino acids present in peptides, facilitating the production of more hydrophobic amino acids. It has been reported that peptides with more hydrophobic residues can compete with cholesterol molecules through rearrangements [153]. On the other hand, OG could compete with cholesterol to enter the micelle solution and reduce the cholesterol solubility.

Nonetheless, for OG-fortified Yogurt, its property of inhibition of cholesterol solubilisation into micelles may not be solely due to the two reasons above. Recent studies reveal that the gut microbiota plays a significant role in human lowering cholesterol ([154; 155; 156]. Importantly, OG has the ability to modulate the human gut microbiota [157; 158]. So, further studies are necessary to evaluate the effect of specific interactions between digested yogurt components and the human gut microbiota on the inhibition of cholesterol solubilisation into micelles.

However, the obtained results confirm the therapeutic properties of yogurts with oat β-glucan against diseases caused by increased cholesterol levels.

4.3 Effects of OG set-type Yogurt on blood lipid metabolism in mice with high fat diet

4.3.1 Effect of adding 0.3% OG into yogurt on the body weight and the wet weight of the liver in mice

Due to the changes in lifestyle and dietary habits, the global incidence of obesity caused by a high-fat diet have significantly increased in recent years, which has become an increasingly serious health problem. Obesity is not only body weight gain, but more importantly, it can cause many serious diseases. For example, visceral obesity has a critical role in the development of cardiovascular disease, which is one of the leading causes of death and premature mortality [159]. Obesity can change the structure of the cardiovascular system (for example, left ventricular hypertrophy and left atrial

enlargement) [160]. Hypertension is closely associated with vascular disease mortality and cardiovascular disease [161]. Previous studies have demonstrated that hypertension showed a clear and strong relationship with obesity [162; 163; 164].

It has been proved that the consumption of Yogurt can ameliorate the high-fat diet-induced metabolic syndrome and oxidative stress [119]. According to our previous study, the yogurt fortified with 0.3% OG has desirable quality and sensory properties. OG is a well-known soluble healthy dietary fiber, especially due to its hypolipidemic activity. So, the 0.3% OG yogurt could be used as a functional dairy product to prevent obesity more effectively.

Body weight gain is an important parameter, by which it can be evaluated the effect of a high-fat diet on the prevalence of obesity and control its treatment [119].

Table 4.2 Body weights and liver weights of mice being fed for nine weeks

Item	Initial body weight (g)	Final body weight (g)	Liver weight (g)
Control sample	20.2 ± 1.0^{a}	27.6 ± 1.4^{d}	1.03 ± 0.08^{a}
HF	20.4 ± 0.8^{a}	35.5 ± 2.3^{a}	1.36 ± 0.04^{b}
HF+0.0% OG Yogurt	20.4 ± 1.2^{a}	33.8 ± 1.2^{b}	1.21 ± 0.04^{b}
HF+0.3% OG Yogurt	20.0 ± 1.2^{a}	29.1 ± 1.8^{c}	1.09 ± 0.06^{a}

Note: the angle a-d indicates whether there is a difference, the same indicates no difference, and the different indicates difference

As shown in Table 4.2, high-fat diet-fed mice showed a significant increase ($p<0,05$) in body weight gain compared with the control mouse. This result is reasonable as the diet rich in fat could cause a high weight gain [165]. Interestingly, the final body weight of the high-fat diet-fed mice was slightly decreased from 35.5±2.3g to 27.6±1.4 g for 0.0% OG Yogurt, while significantly decreased from 35.5±2.3 g to 29.1±1.8 for 0.3% OG Yogurt. Clearly, the addition of OG in Yogurt contributed to reducing the weight gain rate.

This is because the presence of OG can give a feeling of satiety and thus, reduce the food intake amount, resulting in a decrease in weight [166]. Similar results were reported by Yang et al. [167], who found that OG-fortified set-type yogurt decreased the rate of weight gain of mice fed on the diet rich in fat.

The mouse wet weight of the liver is an important parameter for understanding of diet-induced obesity. As shown in Table 4.2, the wet weight of the liver was significantly higher in high-fat diet-fed mice (1.36±0.04 g) than in the control mice (1.03±0.08 g) ($p<0,05$). This is due to the existence of the fatty depots in the liver. Yogurt decreased the wet weight of the liver in high-fat diet-fed mice, especially the 0.3% OG Yogurt ($p<0,05$). This indicated that OG could inhibit body fat accumulation in mice fed a high fat diet.

Yogurt, enriched with oat β-glucan, can be used as the basis of a diet for people who are overweight.

4.3.2 Effect of adding 0.3% OG into yogurt on the lipid profiles

Obesity is related to atherogenic dyslipidemia [168]. The level of total cholesterol (TC), triglyceride levels (TG), HDL, and LDL in the plasma of high-fat diet-fed mice were measured to study the lipid-lowering activity of 0.3% OG yogurt.

As shown in Fig. 4.5, high-fat diet-fed mice showed a significant increase ($p<0,05$) in the TC, TG and LDL levels compared with the control mice. The high levels of TC, TG and LDL are directly responsible for the occurrence of obesity and other disorders [169; 170]. Oral supplementation yogurt significantly reduced these levels in high-fat diet-fed mice ($p<0,05$), especially for 0.3% OG yogurt. An inverse result was observed for HDL.

Clearly, OG has a significant effect on bringing the levels of TC, TG, HDL, and LDL within normal limits. Some researchers believed that the lipid-lowering and cholesterol-lowering properties of OG may related to its high viscosity, which could form a viscous layer in the small intestine, inhibiting intestinal uptake of cholesterol and reabsorption of bile acids [171; 172].

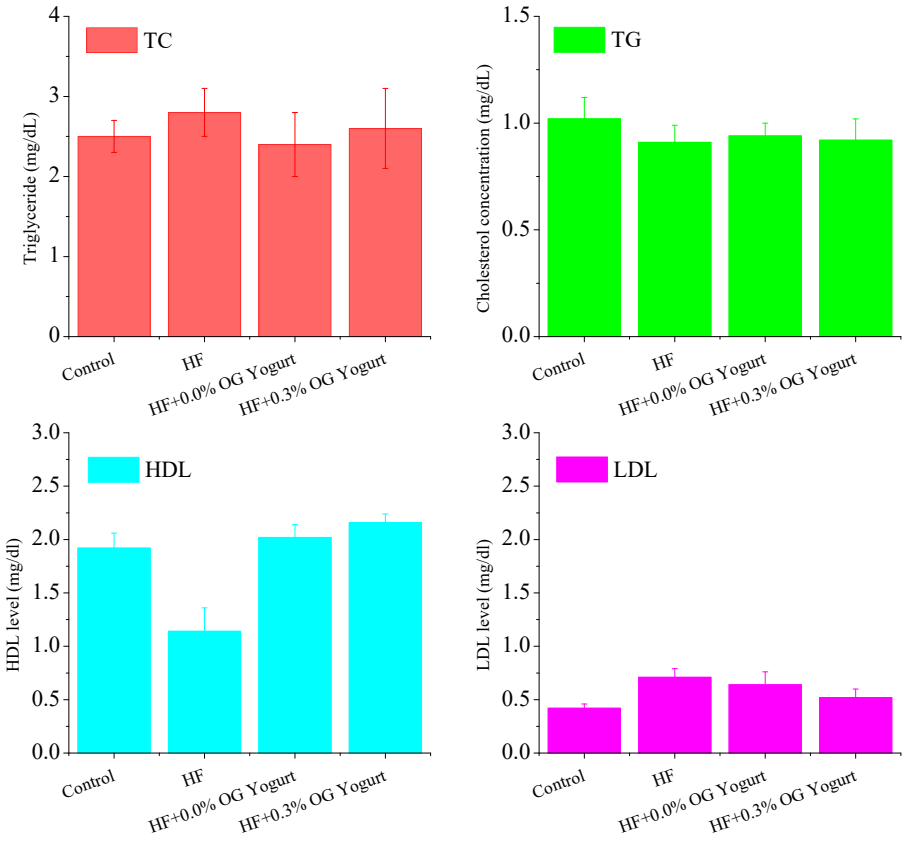


Fig. 4.5 Effect of adding 0.3% OG into Yogurt on the lipid profiles

It was also pointed [29] that OG can exert its anti-lipid effect by activating the AMPK signaling pathway to regulate its downstream effectors such as acetyl-CoA carboxylase, fatty acid synthase, and sterol regulatory element-binding protein. The obtained results testify about functional properties of enriched yogurt.

4.3.3 Effects of 0.3% OG Yogurt on ALT and AST activities in blood serum of mice

Steatosis and hepatic damage may be developed at the high-fat diet-fed animals, due to the fat deposition in the liver. Serum ALT and AST activities of liver function can be increased when hepatic damage occurs [173].

As shown in Fig. 4.6 both the ALT and AST activities were significantly increased ($p<0,05$) in mice compared with the control mice, which may be due to the damage from fatty depots in the liver.

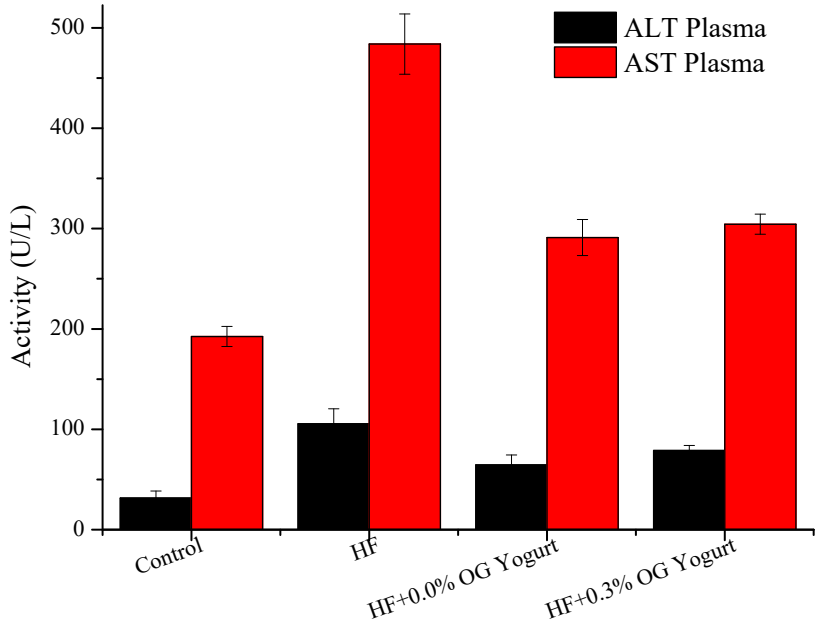


Fig. 4.6 Effects of 0.3% OG yogurt on alanine aminotransferase (ALT) and aspartate aminotransferase (AST) activities in blood serum of mice.

Interestingly, oral supplementation with 0.0% OG yogurt only decreased the ALT and AST activities in high-fat diet-fed mice to some extent, while oral supplementation with 0.3% OG Yogurt completely normalized the activities of these liver enzymes.

4.3.4 Histological assessment results of the liver in mice

As shown in Fig. 4.7, the hepatic tissue of the control group revealed a normal architecture of hepatocytes, without appearing of lipid/fat deposition, whereas high-fat diet-fed groups showed degenerative changes in hepatocytes along with lipid/fat droplet deposition.

Mathieu et al. [174] indicated that excessive accumulation of fat in the adipose tissue leads to macrophage infiltration and elevated production of proinflammatory cytokines, which contribute to the development of atherosclerosis.

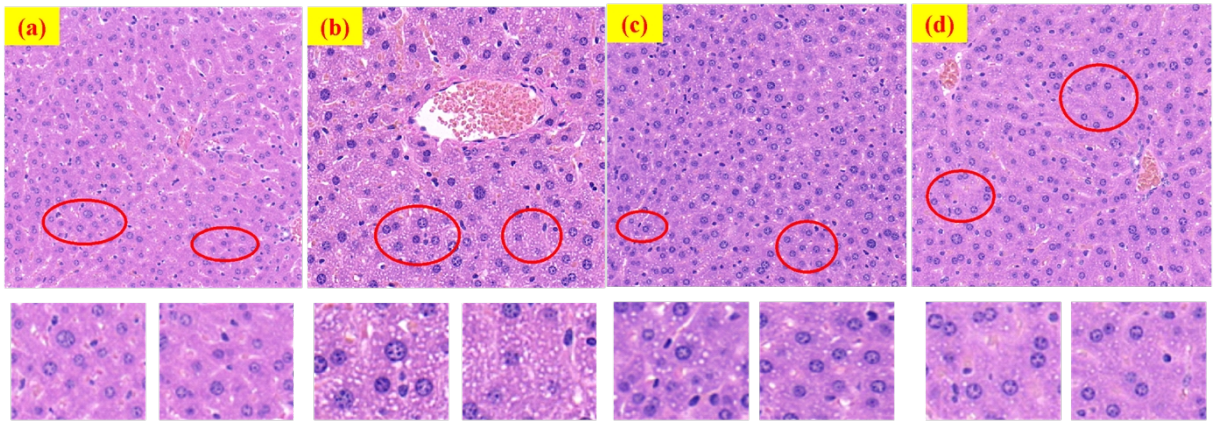


Fig. 4.7 Effect of 0.3% OG Yogurt on liver infammation in high-fat diet-fed mice. (a) Control; (b) HF; (c) HF+0.0% OG Yogurt; (d) HF+0.3% OG Yogurt

High-fat diet-fed mice that received the 0.0% OG Yogurt supplement displayed less fat/lipid deposition, and high-fat diet-fed mice that received the 0.3% OG Yogurt supplement showed a similar pattern to the control group, indicating the hepatoprotective effect.

Conclusion to Section 4

1. Compared with DPPH assay, ABTS assay is more suitable for evaluating the antioxidant activity of set-type yogurt during digestion. The antioxidant activity of Yogurt was mainly produced during the gastric digestion phase, and 0.3% OG could further improve the antioxidant activity of Yogurt by promoting the beneficial enzymatic hydrolysis.

2. The addition of OG can effectively enhance the functional characteristics of fermented Yogurt. The amount of soluble proteins and peptides increased throughout digestion. Large spherical vesicles were formed for both control yogurt and 0.3% OG Yogurt after gastric digestion. 0.3% OG Yogurt after digestion had higher inhibition of cholesterol solubility.

3.In biomedical experiments, the effect of OG set-type Yogurt on blood lipid metabolism in mice with high-fat diet was mainly determined by Lee's index, blood lipids TG, TC, HDL, LDL, AST, ALT, liver weight and liver sections were studied.

4. The experiment of effects of OG set-type yogurt on blood lipid metabolism in mice with high fat diet confirmed that 0.3% that set-type Yogurt can be an alternative and a complementary treatment to therapy of obesity-related complications.

5. Yogurt with addition of oat β-glucan, produced according to the improved technology with shorter duration of fermentation, is well digested and can be used as a product for medical and preventive purposes.

SECTION 5 QUALITY AND SOCIO-ECONOMIC EFFECTIVE EVALUATION OF OG SET-TYPE YOGURT PRODUCT

5.1 The development of the technology for production of Yogurts with addition of oat β-glucan

Taking into account the results of previous studies and physicochemical properties of the raw materials, it is suggested using the tank method for production of Yogurt with addition of oat β-glucan under industrial conditions. The peculiarity of this method is that fermentation, fermenting and ripening of the container is carried out in one tank. At the same time, fillers can be added both before and after fermentation.

The research showed that oat β-glucan has a positive effect on the fermentation process, that's why it is suggested adding before beginning of the fermentation. We have already developed a yogurt recipe with addition of oat β-glucan (Table 5.1.).

Table 5.1. Yogurt recipe with addition of oat β-glucan

Raw material	Quantity of raw material, %
Cow milk	99,7
Oat β-glucan	0,3
Bacterial culture	20 DCU/100L
Total	100

The technological scheme for production of Yogurt with β-glucan is shown in Fig.5.1. According to this scheme, milk, which complies with all quality requirements, is purified and cooled to prevent the development of microorganisms. After ripening, milk is heated to the temperature of separation. Normalized mixture is purified, homogenized under pressure 18-20 MPa.

	Приймання і оцінка якості молока, резервування – Acceptance, milk quality evaluation, reservation	*Вівсяний β-глюкан- Oat β-glucan*
	Підігрів до t 40-45°C – Heating up to t 40-45°C	Якісна оцінка, зберігання β-глюкану- Quality evaluation, β-glucan storage
Вершки- cream	Сепарування молока- Milk separation	Приготування збагаченої молочної основи – Preparation of enriched milk base
Охолодження t= 4-6°C- Cooling down t= 4-6°C	*Знежирене молоко- Skimmed milk*	
Резервування - Reservation	Підігрів до t 70-75°C - Heating up to t 40-45°C	Перемішування 15-20хв – Mixing 15-20 min.
Зберігання не більше 6 год – Storage for not more than 6 hours	Гомогенізація 18-20 МПа- Homogenization 18-20 MPa	
	Пастеризація суміші t=90-95°C, 10-15хв – Mixture pasteurization t=90-95°C, 10-15min.	
	Перемішування 5хв- Mixing 5 min.	
	Охолодження t=40 °C – Cooling down t=40 °C	
	Перемішування 20 - 30хв- Mixing 20-30 min.	Внесення закваски і заквашування – Introduction of the starter and fermenting
	Сквашування 2,5-3 год, К=70-80°C- Fermentation 2,5-3 hrs, K=70-80°C-	
	Охолодження, 20-25 °C – Cooling down, t=20-25 °C	
	Фасування - Packaging	
	Зберігання t=4-6°C, 21 доба – Storage t=4-6°C for 21 hours	

Fig.5.1. The technological scheme for production of Yogurt with β-glucan

The mode of homogenization is one of the main factors of the kinetic stability of milk-fat suspensions, including the enriched milk base, and the choice of optimal process parameters (temperature and pressure) is important for providing the stability of the relevant technological parameters of raw materials during storage and technological processing. The homogenization pressure varied from 10 to 20 MPa, temperature - from 55 to 75°C. One stage homogenization of the combined milk mixture was used during conducting of research.

Increasing the homogenization pressure of the combined milk mixture from 10 to 20 MPa contributes to enhance the process efficiency, which is evidenced by the increase in the degree of homogenization by 7-7.8%, depending on the process temperature. This is due to the fact that when the homogenization pressure is increased, the speed of movement of the mixture in the valve gap increases, which contributes to the formation of suspension particles with a diameter of 1,0-1,5 μm. At this value of the radius of the fat balls, the electric forces of repulsion exceed the van der Waals forces of attraction, and such balls do not form clusters. When the homogenization pressure is further increased to 21 MPa, the degree of homogenization decreases by 13-14.1%, which is explained by the increased fat content in the enriched milk mixture compared with milk.

An increase in the homogenization temperature from 50 to 65 °C has a more significant effect on the increase in the degree of homogenization than an increase in the temperature from 65 to 75 °C. This is explained by the fact that when the homogenization temperature increases, the fat balls in the valve gap of the homogenizer are pulled into thinner "threads" and form new balls with a diameter of up to 1.0 μm. The highest efficiency of the process is observed at a temperature of 72-75 °C; the degree of homogenization is about 81%. In this way, the optimal mode of homogenization according to the new improved technology was established: the homogenization pressure is 18-20 MPa and the temperature is 70-75 °C. The

homogenized milk mixture enriched with oat β-glucan is sent for pasteurization. The mixture is pasteurized for 10-15 minutes at a temperature of 90-95°C. After pasteurization, the mixture is thoroughly mixed and cooled to the fermentation temperature (40°C). The recommended equipment and technological scheme of yogurt production is presented in Figure 5.2.

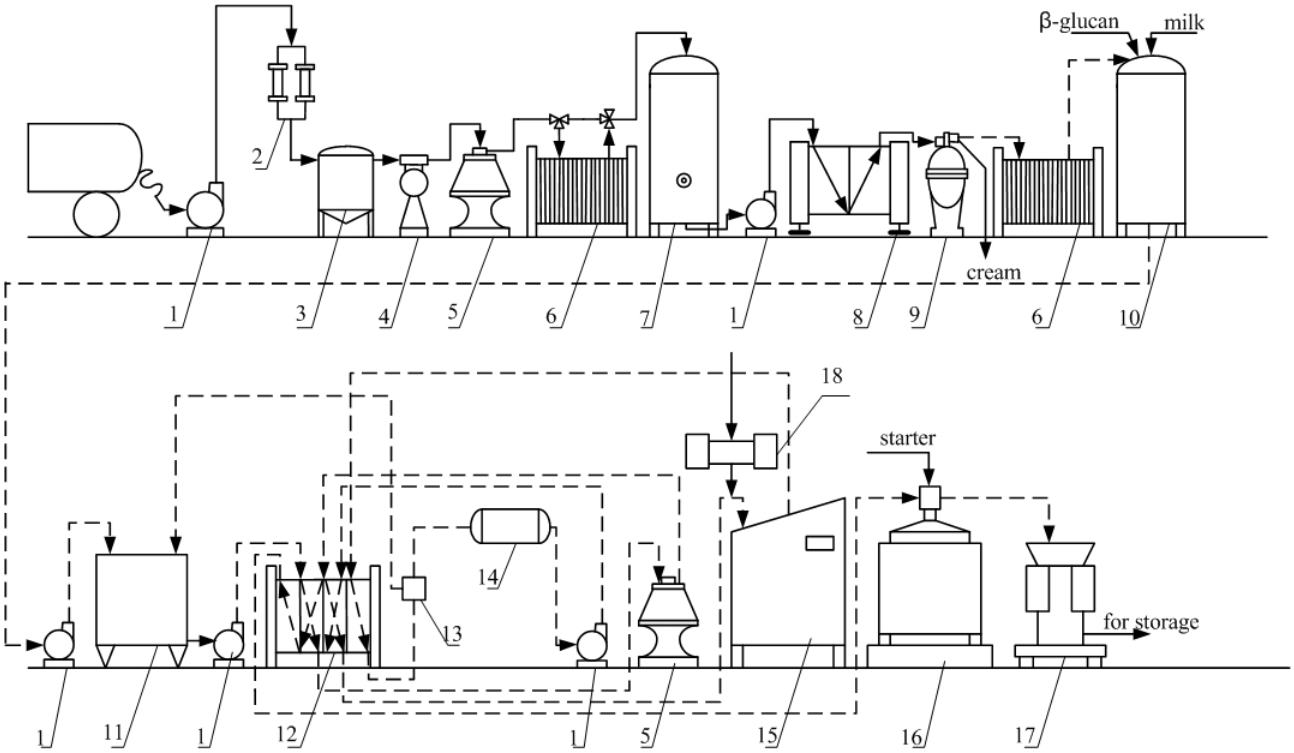


Fig. 5.2. Equipment and technology scheme for the production of yogurts with β-glucan: 1– milk pump; 2 – milk filter; 3 – air separator; 4 – milk-counter; 5 – milk separator; 6 – plate cooler; 7 – receiving tank; 8 – plate heater; 9 – cream separator; 10 – tank for normalization; 11 – equalizing tank; 12 – plate pasteurizing cooling plant; 13 – thermal sensor; 14 – container holder; 15 – homogenizer; 16 – starter; 17 – filling machine; 18 – injector

For fermentation, starter cultures are used, which include *Lactobacillus delbrueckii subsp. bulgaricus, Streptococcus salivarius subsp. thermophilus*. After adding the starter, the mixture is stirred for 20-30 minutes to evenly distribute the starter culture in the volume of the milk mixture. Fermentation is carried out at a temperature of 40-45°C for 2.5-3 hours until the formation of a clot with an acidity of 70-80°T. The duration of fermentation is shortened due to the acceleration of lactic acid

fermentation when β-glucan is introduced. After it is ready, the clot is cooled to a temperature of 20-25°C. The thoroughly mixed product is packaged in a container, further cooled to 4-6°C and sent to storage. The recommended shelf life of yogurt with oat β-glucan is 21 days, which is higher than the typical production technology of natural Yogurts (7-14 days).

5.2 The results on research of chemical composition, sensory, physicochemical and microbiological indexes of OG set-type Yogurt

The chemical composition, sensory, physicochemical and microbiological quality indicators of the IPS1 and IPS2 experimental samples obtained under the industrial production conditions of Youdianai (Chuzhou) Health Technology Co., Ltd. and Tianjin Huaming Dairy Co., Ltd. were determined, after packaging and storage at 21 day compared with a laboratory-made sample (control sample). The results of the study are listed in Table 5.2.

The data presented (Table 5.2) show that the physicochemical, sensory and microbiological quality indicators of the control sample and the samples obtained under industrial production conditions after packaging and after 21 d of storage fully meet all the requirements for Yogurt products. The main components of the chemical composition of the samples meet yogurt standard requirements.

Table 5.2 Quality indexes of Yogurts, produced under industrial conditions

Item indicators	Samples					
	Control sample		IPS1		IPS2	
	After packaging	After 21 d of storage	After packaging	After 21 d of storage	After packaging	After 21 d of storage
1	2	3	4	5	6	7
Color	Creamy, the color of the whole product is uniform		Creamy, the color of the whole product is uniform		Creamy, the color of the whole product is uniform	

The continued of the Table 5.2

1	2	3	4	5	6	7
Taste and flavor	Clean, yogurt flavor, no other flavor and smell	The taste of yogurt with obvious sour taste	Clean, yogurt flavor, no other flavor and smell	The taste of yogurt with obvious sour taste	Clean, yogurt flavor, no other flavor and smell	The taste of yogurt with obvious sour taste
Consistency and appearance	Good coagulation, fine and uniform structure, deployable		Good coagulation, fine and uniform structure, deployable		Good coagulation, fine and uniform structure, deployable	
Mass fraction of fat, g/100g	3.56±0.21	3.54±0.11	3.56±0.37	3.55±0.25	3.56±0.41	3.54±0.27
Mass fraction of non-fat milk solids in yogurts,g/100g	11.30±0.48	11.72±0.47	11.65±0.22	11.81±0.25	11.39±0.18	11.79±0.41
Mass fraction of protein, g/100g	3.16±0.18	3.16±0.14	3.16±0.11	3.16±0.12	3.16±0.14	3.16±0.13
Acidity, °T	84±0.09	93.8±0.06	84±0.13	93.8±0.11	84±0.07	93.8±0.09
Lactic acid bacteria, q-ty (cfu/mL)	(3.15±0.3) $\times 10^7$	(0.63±0.03) $\times 10^7$	(3.22±0.04) $\times 10^7$	(0.71±0.05) $\times 10^7$	(3.23±0.2) $\times 10^7$	(0.72±0.02) $\times 10^7$
E. scherichia coli	Not detected	Not detected	Not detected	Not detected	Not detected	Not detected

The End of the Table 5.2

1	2	3	4	5	6	7
Staphylococcus aureus	Not detected	Not detected	Not detected	Not detected	Not detected	Not detected
Salmonella	Not detected	Not detected	Not detected	Not detected	Not detected	Not detected
Yeast	Not detected	Not detected	Not detected	Not detected	Not detected	Not detected
Mould	Not detected	Not detected	Not detected	Not detected	Not detected	Not detected

The research results of the quality indicators of the experimental samples of IPS1 and IPS2 produced under industrial conditions were correlated with the results under laboratory conditions, indicating that the selection of processing parameters of dairy raw materials in the production process was correct. It should be noted that the quality index values of the produced samples all meet the requirements of ДСТУ 4343:2004 and GB 19302-2010 (Chinese National Food Safety Standard Fermented milk).

The analysis of industrial samples showed that oat β-glucan does not have any negative effect on production process and storage that gives reasons to conclude about the possibility of producing this product under industrial conditions.

5.3 Energy value calculation for Yogurts with oat β-glucan

In order to calculate energy value the following formula is used:

$$E = k_p \times (M_p + M_c) + k_p \times M_f, \qquad (5.1)$$

where E - energy value,kcal;

M_p - mass fraction of protein, g/100 g of product;

M_c - mass fraction of carbohydrates, g/100 g of product;

M_f - mass fraction of fat , g/100 g of product;

k_p = 4 – coefficient of energy value of 1g of protein or 1g of carbohydrates in the product, kcal/g;

k_f = 9 - coefficient of energy value of 1g of fat in the product, kcal/g.

For control sample, energy value is the following :

$E = 4 \times (3,16 + 4,58) + 9 \times 3,56 = 30,96 + 32,04 = 63$ kcal

The energy value of IPS1 is:

$E = 4 \times (3,16 + 4,93) + 9 \times 3,56 = 32,36 + 32,04 = 64,4$ kcal

The energy value of IPS2 is:

$E = 4 \times (3,16 + 4,67) + 9 \times 3,56 = 31,3 + 32,04 = 63,36$ kcal

The introduction of the additive contributes to increase the energy value of finished product by increasing the amount of carbohydrates.

5.4 Calculation of material costs for production of Yogurt with oat β-glucan

The calculation is performed for production of 20 tons of set-type OG Yogurt. This technological process is calculated for 10 h production per day.

Table 5.3 Main data on production of OG set-type Yogurt

Project	Name	Percentage, %	Project	Name	Percentage (%)
1	2	3	4	5	6
Fixed indicator	Raw material	98	Yogurt production loss ratio	Cooling loss	5.0
	Raw milk fat mass fraction	3.6		Fermentation loss	1.6

The End of the Table 5.2

1	2	3	4	5	6
	Product fat mass fraction	3.5		Filtration loss	1.6
Raw material ratio	Raw milk	99.7		Bottling loss	3.0
	OG	0.3			

Material calculation：

Raw milk weight: M'=20 t

Effective amount of raw milk :

$$Mraw=20t\times 98\%=19.60\ t \quad (5.1)$$

Total mass after ingredients:

$$Mtotal=19.60\div 99.7\%=19.66\ t \quad (5.2)$$

After each loss process, remainder:

After filtering:

$$M_1=(19.66-0.0005)\times 98.4\%=19.34\ t \quad (5.3)$$

After cooling down:

$$M_2=19.34\times 95\%=18.37\ t \quad (5.4)$$

After filling:

$$M_3=18.37\times 97\%=17.82\ t \quad (5.5)$$

After fermentation:

$$M_4=17.82\times 98.4\%=17.54\ t \quad (5.6)$$

Thus, the actual output of OG yogurt is 17,54 t.

5.5 Energy balance in the production of OG set-type yogurt

5.5.1 Energy consumption of ice water

The milk collection system collects 20 tons of milk every day. According to the raw milk temperature of 15ºC, ice water needs to be used to cool the raw milk to a storage temperature of 4 ºC. The ice water is usually used under the condition of 2 ºC water inlet, and the temperature rise does not exceed 12 °C, that is, 14 ºC. The specific heat of raw milk is 3.99KJ/Kg when the ice water is refluxed at 4 ºC, so the daily consumption of ice water in the milk collection system is calculated as follows:

$$Q=MC\Delta T \qquad (5.7)$$

$$Q_{\text{ice water}} = Q_{\text{fresh milk}}$$

Hence: $20000\times3.99\times(15-4)=M_{\text{ice water}}\times4.2\times(14-2)$

Amount of ice water: $M_{\text{ice water}} = 17417$ L

The daily production of milk is calculated at 20 tons, and the mixing amount usually accounts for 1/3 of the total material volume. Therefore, the mixing system has 7 tons of materials that need to be heated and cooled every day. According to the mixing temperature of 70 ºC, cooling tower water and ice are required. Water cools the mixture to a temporary storage temperature of 15 ºC, and the cooling tower water is 35 ºC in summer. According to the design of the heat exchanger, the outlet temperature difference is 2 ºC. The temperature section that needs to be cooled by ice water is 37°C to 15 ºC. Ice water is usually used that is supplied at 2 ºC, and the temperature rise does not exceed 12°C, that is, the ice water is refluxed at 14 ºC. The specific heat of the mixture is calculated according to the specific heat of milk. Therefore, the daily consumption of ice water in the mixing system is calculated as follows (formula 5.7):

$Q_{\text{ice water}} = Q_{\text{mixed material}}$

$7000\times3.99\times(37-15)=M_{\text{ice water}}\times4.2\times(14-2)$

$M_{\text{ice water}} = 12192$ L

The daily production of solidified yogurt is calculated at 20 tons. According to the different equipment capacity, the consumption of ice water is:

3t/h Pasteurizer: 4950 L/h, 10 hours a day.

The daily production of set-type yogurt is calculated according to 20 tons, according to the fermentation temperature of 42 ºC, the yogurt needs to be cooled with ice water to the temporary storage temperature of 20 ºC, and the ice water is usually used under the condition of 2 ºC water intake, but in order to ensure product quality, so use the water which is refluxed at 18 ºC, the specific heat of yogurt is calculated according to the specific heat of milk, so the daily consumption of ice water in the fermentation system is calculated as follows:

$$20000\times3.99\times(42-20)=M_{\text{ice water}}\times4.2\times(18-2)$$

$$M_{\text{ice water}}=26125\ \text{L}$$

The filling system consumes 1140 L/h of ice water, working 10 hours a day, the daily consumption of ice water is 11400 L,

That is, the daily cooling capacity of the factory is:

$$Q_{\text{cooling capacity}}=11400\times4.2\times(14-2)=574560\ \text{KJ}$$

5.5.2 Energy consumption of steam

The materials in the mixing system need to be heated and cooled. According to the mixing temperature of 55 ºC, it is necessary to use steam to raise the raw milk from 4 ºC. The mixing temperature is 55 ºC, the steam is usually used with saturated steam of 3 Bar, and the latent heat of steam is 2200 KJ/kg.

The specific heat of the mixed material is calculated according to the specific heat of milk, regardless of the system energy loss, so the daily steam consumption of the mixing system is calculated as follows:

$$Q_{\text{steam}}=Q_{\text{mixed material}}$$

$20000\times3.99\times(55-4)=M_{steam}\times2200$

$M_{steam}=1850$ kg

The milk sterilization temperature is 95 ºC, and the raw milk temperature is 4 ºC, which is calculated according to the heat recovery efficiency of the equipment 90%. Therefore, the daily steam consumption of the pre-sterilization pasteurizer is:

$M_{steam}=20000\times3,99\times(95-4)\times(1-90\%)/2200=330$ kg

Steam consumption per ton of product = 12.4 kg

The sterilization temperature for the production of set-type yogurt is 95 ºC, and the feed temperature is 10 ºC. According to the calculation of 20 tons, the heat recovery efficiency of the equipment is 92%. The daily steam consumption of the yogurt pasteurizer is:

$M_{steam}=20000\times3.99\times(95-10)\times(1-92\%)/2200=247$ kg

Steam consumption per ton of product = 12.4 kg

The daily production of solidified yogurt is calculated according to 20 tons. According to the fermentation temperature of 42 ºC, the yogurt is heated to the canning temperature of 43 ºC. It is necessary to use steam to heat the hot water, and the hot water to heat the yogurt. The energy transfer efficiency is 98%, so the calculation of the daily consumption of fermentation steam is as follows:

$M_{steam}=20000\times3.99\times(43-10)/2200/98\%=1220$ kg

The cleaning medium needs to be heated for daily cleaning. According to the design data of the tubular heat exchanger for cleaning, the peak steam consumption of each cleaning pressure line is 300 kg/h, 6 Bar, saturated steam. Since there are a total of 6 pressure lines, but basically no steam is consumed at the same time, it can be calculated according to 70%, that is, the average consumption of steam is 1260 kg steam.

5.6 Calculation of product cost

5.6.1 Mixing system energy calculation

In this design (see appendix D), the ingredients of the mixing system will use partial mixing, and the final mixing method will only use 1/3 of the raw materials for heating and cooling, and avoid using all materials for heating and cooling, so 4064 kg of ice water is saved every day, save 617 kg of steam per day.

The cooling capacity per ton of ice water is: Q=1000×4.2×(14-2)=50400 KJ，requires 14 KWH of power. Without considering the power conversion efficiency, 57 KWH is required to produce 4.064 tons of ice water, and the electricity price is 0.09 $/KWH, that is, saving 57×0.09=5.13 $/d，annual saving 0.617×18×365=$4053.69.

Therefore, the use of this process saves energy costs of $0.812 per ton of product, and the annual direct energy cost savings is about $5926.04.

5.6.2 Fermentation system energy calculation

In this study, 0.3% OG was added to the set-type Yogurt product. The results showed that OG could increase the fermentation rate of lactic acid bacteria and shorten the fermentation time by 16 min (total fermentation time was 5 h). The amount of steam that can be saved per day in the fermentation stage is: 1220÷18.75×2=130 kg.

Steam is calculated at $18 per ton, the energy cost per ton of product is $0.117, and the annual saving is: 0.13×18×365=$854.1.

5.7 Socio-economic effectiveness of scientific and technical developments and implement the results of work in practical production

To apply the technology of OG set-type Yogurt as food innovation. According to this technology, pilot production has been carried out in 2 factories in China (You Dian Ai (Chuzhou) Health Technology Co. LTD; Tianjin Huaming Dairy Co. LTD) .

We have already counted and evaluated, the socio-economic effectiveness of scientific and technical developments and implement the results of work in practical production as follows:

Firstly, to adopt appropriate production technology and rationally arrange production equipment, producing 1 ton of product can reduce 0.5 ton ice water and 31 kg steam compared with the traditional processing, the cost of production in the mixing system and fermentation system can be reduced by $0.93 per ton.

Secondly, OG contributes to increasing the fermentation rate of lactic acid bacteria and to shorten the fermentation time by 1,5 h, it is beneficial to reduce the consumption of fossil energy, water and carbon dioxide emissions, thus, reduce the greenhouse effect and waste water, and, as well as production is beneficial environmental protection.

Thirdly, based on data from 2 factories, it takes 5%~6% less time to produce a ton of product, resulting in higher productivity, increased workshop and equipment utilization, and reduced workshop and equipment depreciation by $1.00~$2.00 per ton. On other hand, labor costs are reduced by $6.00~$8.00 per ton.

Fourthly, due to the physiological effects of OG, OG set-type Yogurt products will bring more health benefits to consumers. Long-term and appropriate consumption will help consumers' health, reduce medical and health expenditures, and thus, bring profound social and economic benefits.

Finally, according to the above data, OG set-type Yogurt is an innovative and functional food. Reasonable process and technology can reduce production costs and bring production benefits to enterprises, while functional food also brings social benefits.

Conclusion to Section 5

1. This section organized the recipe and production process of OG set-type Yogurt, and listed the main parameters and equipment descriptions required for the

production of OG set-type Yogurt under industrial conditions, as a reference for the production of the product.

2. The chemical composition, physicochemical (mass fraction of non-fat dry solids, proteins, fats), organoleptic and microbial indicators, and the number of lactic acid bacteria of the OG set-type Yogurt samples produced under industrial conditions were studied, and it was established that the product met the requirements of the “Food Safety Law”. It was established that the fortification of OG in set-type yogurt increased the nutritional value of yogurt.

3. The calculations of energy consumption based on the production processes were performed on the condition that 20 t of milk per day is processed. The calculation results showed that an appropriate production process and a reasonable choice of the equipment can reduce production costs.

4. The technology has been applied to food innovation, and the product of OG set-type Yogurt has been produced in two factories in China. According to statistics and evaluation, the social and economic benefits of scientific and technological development and the implementation achievements in actual production are listed.

CONCLUSIONS

1. On the basis of theoretical generalization and experimental research, it is suitable and possible to use OG as a physiologically active additive that can provide the product with additional functional properties has been proven. It is shown the ability to reduce the glycemic index, the level of glucose and cholesterol level in the blood, to improve the transport of glucose by muscle cells, to stimulate antibacterial activity, to reduce obesity, to enhance peristalsis of the gastrointestinal tract, and to maintain intestinal health.

2. The effect of adding OG on chemical composition of finished product (mass fraction of dry solids, proteins and fats) was studied. Based on these studies, a Yogurt recipe with the addition of OG was proposed. It was established that the optimal dosage of oat β-glucan is 0.3%.

3. It was established that the rheological and antioxidant properties of yogurts are improved due to the addition of β-glucan. A positive effect on the textural characteristics of Yogurt, a decrease in its adhesiveness, was observed. The addition of oat β-glucan in the amount of 0.3% leads to an increase in the water-holding capacity of Yogurt to 94.67%. The recommended shelf life of yogurts is 21 days, while the product has an appropriate texture and consistency.

4. The addition of 0.3% oat β-glucan has been shown to have a protective effect on probiotics in Yogurt. A biomedical experiment confirmed that the addition of 0.3% oat β-glucan increases the serum activity of liver ALT and AST. Yogurt with the addition of 0.3% oat β-glucan has functional properties. The product can be an alternative and adjunct to the treatment of obesity-related complications.

5. A series of experimental studies determined the optimal mode of homogenization of the enriched milk base. It is shown that increasing the homogenization pressure of the combined milk mixture from 10 to 20 MPa helps to increase the efficiency of the process, which is evidenced by the increase in the degree of homogenization by 7-7.8%, depending on the process temperature. The highest

efficiency of the homogenization process is observed at a temperature of 72-75 °C, while the degree of homogenization is about 81%.

6. The effect of oat β-glucan on the organoleptic, physicochemical, and microbiological indicators of Yogurt was studied. Yogurt with oat β-glucan meets the requirements of the standard in all respects. When the concentration of oat β-glucan was increased to 0.3%, the yogurt had the highest sensory evaluation score (about 85).

7. A technological scheme for the production of Yogurts with oat β-glucan has been developed, and optimal modes of the main technological processes have been proposed. According to the improved technology, the duration of fermentation is reduced by 16 minutes due to the prebiotic effect of the additive.

8.Energy consumption for production is calculated under the condition of processing 20 tons of milk per day. The results of the calculations showed that a suitable production process and a reasonable choice of equipment can reduce the cost of production.

9. Industrial testing of the developed technology was carried out. It was established that the energy value of Yogurt with additives is higher than Yogurt without additives due to the increase in the amount of carbohydrates.

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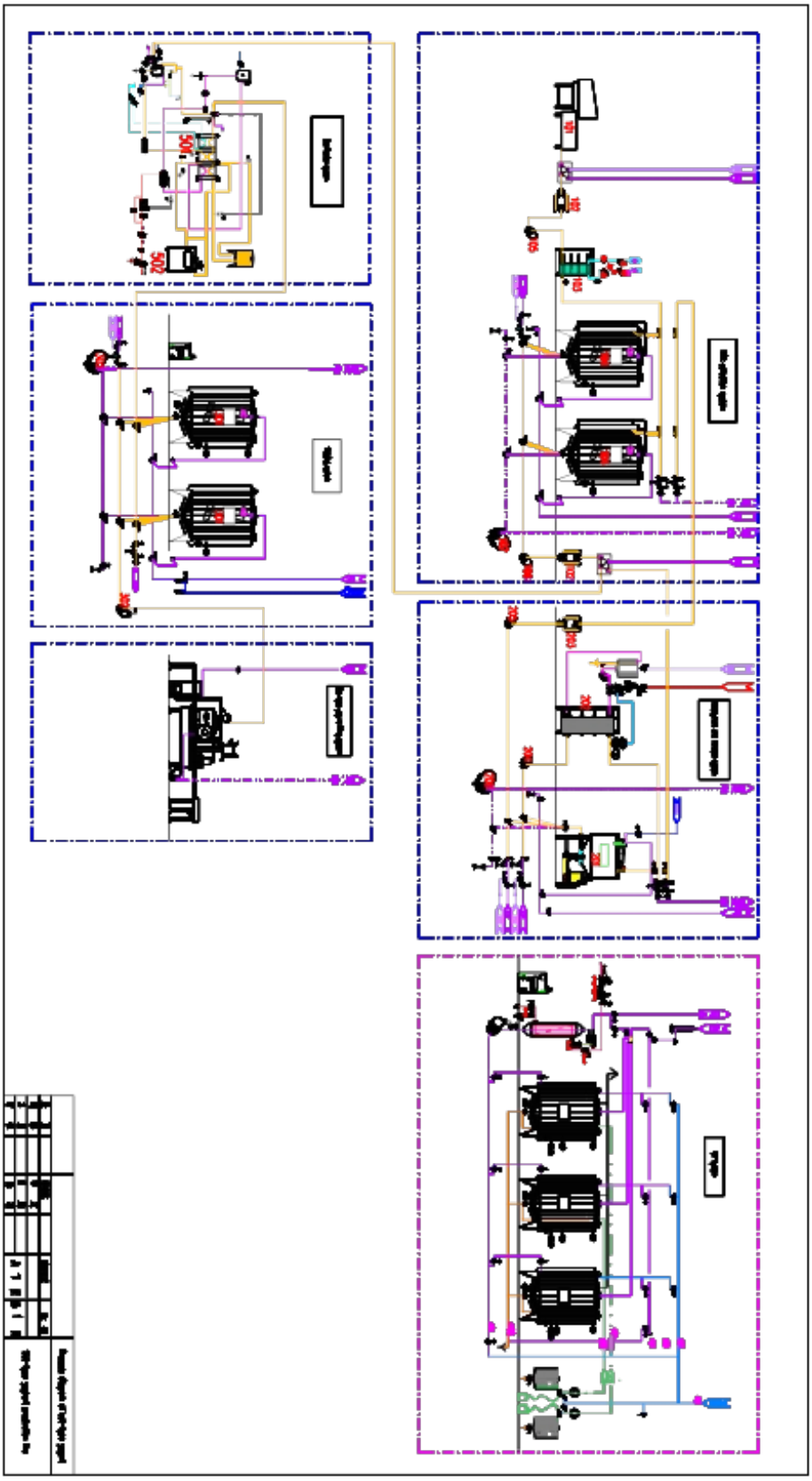
APPENDICES

Appendices A OG set-type yogurt production line



Appendices B Hazard analysis of OG set-type yogurt production

Product Description: frozen yogurt, single cup Storage and sales method: 2 ℃ -6 ℃ cold storage and cold chain system sales Intended use and consumers: ready to eat products, general public					
Machining steps	**Potential hazards**	**Whether the hazard is significant**	**Judgment basis for potential hazards**	**preventive measure**	**Whether it is a key control point?**
Purchase acceptance of raw and auxiliary materials	Biological: bacteria, pathogenic bacteria	Yes	Raw materials are infected with miscellaneous bacteria, and dairy cows are sick	Control according to hygienic standard operating procedures	Yes
	Chemical: pesticides, antibiotics	Yes	Pesticide and veterinary drug residues	Control according to hygienic standard operating procedures	
	Physical: feed, insects, etc	No	Raw material pollution	Filtration separation can be removed	
Filter	Biological: bacteria, pathogenic bacteria	Yes	Operator hygiene, equipment pollution	Control of equipment, personal hygiene and hygienic standard operating procedures	No
	Chemical: None	No			
	Physical: mechanical wear	No	Improper operation and mechanical wear	Operate correctly and regularly repair and inspect the equipment	
Cold storage	Biological: bacteria, pathogenic bacteria	Yes	The temperature is too high, causing excessive bacterial reproduction	Control temperature at 4-6 ℃	Yes

	Chemical: None	No			
	Physical: None	No			
Standardization	Biological: bacteria, pathogenic bacteria	Yes	Incomplete disinfection of the instrument and carrying of raw materials	Subsequent sterilization can control hazards	No
	Chemical: None	No			
	Physical: mechanical wear	No	Improper operation and mechanical wear	Operate correctly and regularly repair and inspect the equipment	
mixed ingredients	Biological: microbial	Yes	Infected miscellaneous bacteria	Controlled by hygienic standard operating procedures	No
	Chemical: heavy metal pollution	Yes	Heavy metal pollution	Controlled by hygienic standard operating procedures	
	Physical: hair, etc.	No	Accidental falling during processing	Controlled by hygienic standard operating procedures	
Preheat homogenization	Biological: bacteria, pathogenic bacteria	Yes	The machine is not thoroughly disinfected, and the raw material carries bacteria	Subsequent sterilization can control hazards	No
	Chemical: None	No			
	Physical: mechanical wear	No	Improper operation and mechanical wear	Correct operation and regular repair and inspection of equipment shall be controlled by hygienic standard operating procedures	

Sterilization	Biological: bacteria, pathogenic bacteria	Yes	Unqualified sterilization	Control sterilization conditions (95 ℃, 5min), re sterilization	Yes
	Chemical: None	No			
	Physical: mechanical wear	No	Improper operation and mechanical wear	Correct operation and regular repair and inspection of equipment shall be controlled by hygienic standard operating procedures	
Cooling	Biological: microbial	Yes	Incomplete equipment disinfection	Controlled by hygienic standard operating procedures	No
	Chemical: None	No			
	Physical: None	No			
Inoculation	Biological: microbial	Yes	Introduction of miscellaneous bacteria and insufficient vitality of strains	Select and cultivate excellent strains	Yes
	Chemical: None	No			
	Physical: None	No			
Filling	Biological: microbial	Yes	Incomplete disinfection of packaging machinery	The mechanical cleanliness is controlled by hygienic standard operating procedures and aseptic filling is adopted	Yes
	Chemical: hazardous substances, cleaning agents	No	Cleaning machine residual detergent, packaging materials containing harmful substances	Mechanical cleaning is controlled by hygienic standard operating procedures, and packaging materials are correctly selected	

	Physical: mechanical wear	No	Improper operation and mechanical wear	Correct operation and regular repair and inspection of equipment shall be controlled by hygienic standard operating procedures	
Seal	Biological: microbial	Yes	Improper operation introduction	Controlled by hygienic standard operating procedures	No
	Chemical: None	No			
	Physical: None	No			
Fermentation	Biological: microbial	Yes	Sterilization can only be commercial, not really sterile.	A small amount of bacteria is harmless, which is controlled by hygienic standard operating procedures	No
	Chemical: None	No			
	Physical: None	No			
Cold storage	Biological: microbial	Yes	The temperature is too high, causing microbial reproduction and deterioration	Control temperature at 4 ℃	Yes
	Chemical: None	No			
	Physical: None	No			

Appendices C.1 Certificate of implementation of research results

Application Testify

Application Enterprise	You Dian Ai (Chuzhou) Health Technology Co., LTD
Postal address	398 East Wuhu Road, Chuzhou City, Anhui Province, China
Starting and Ending Time	Sep., 10, 2021--Now
Contact Person	Cai Manyi (+086 18937383791)

Our enterprise has trial produced set-type yogurt with 0.3% oat β- glucan. The recommended classification of the product is functional yoghurt. The product is characterized by the addition of prebiotics and the number of viable bacteria in yoghurt ≥3.2×10^7 CFU/ mL, which is conducive to health, is prepared as an alternative product of the company.

You Dian Ai (Chuzhou) Health Technology Co., LTD

Dec.,22, 2021

Appendices C.2 Certificate of implementation of research results

Application Testify

Application Enterprise	Tianjin Huaming Dairy Co., LTD
Postal address	150 Gongmao Dajie, Xuguantun, Wuqing District, Tianjin, China
Starting and Ending Time	June, 8, 2020 - Now
Contact Person	Zhang Dexin (+086 16650507636)

Our enterprise has already added 0.3% oat β-glucan to set-type yogurt in a pilot test. Under the same conditions, the fermentation time was shortened to 16 min, and the sensory score of the obtained product exceeded 85; The addition of 0.3% oat β-glucan improves the nutritional value of the set-type and prolongs the shelf life.

Tianjin Huaming Dairy Co., LTD

July, 20, 2020

«ЗАТВЕРДЖУЮ»
Проректор з наукової роботи
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«ЗАТВЕРДЖУЮ»
Директор
Філії «Сумський молочний завод» ДП «Аромат»
Бєлік Л.В.
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АКТ

ПРОМИСЛОВОЇ АПРОБАЦІЇ ТЕХНОЛОГІЇ ВИРОБНИЦТВА ЙОГУРТУ З ДОДАВАННЯМ ВІВСЯНОГО БЕТТА-ГЛЮКАНУ

Ми, представники від підприємства філії «Сумський молочний завод» ДП «Аромат», що нижче підписалися:
директор – Бєлік Л.В.;
головний технолог – Белик В.О. ;

і представники Сумського національного аграрного університету:
доцент кафедри технологій та безпечності харчових продуктів Сумського НАУ, - к. т. н., Назаренко Ю.В.;
аспірант кафедри технологій та безпечності харчових продуктів Сумського НАУ - Qu Xiaoqing;

склали цей акт у тому, що з 03.04.2022 по 05.04.2022 на устаткуванні підприємства було проведено виробництво дослідної партії йогурту з додаванням вівсяного бетта-глюкану.

1. Підстава щодо роботи.
Тимчасова технологічна картка на проведення дослідних робіт з виробництва йогурту з додаванням вівсяного бетта-глюкану.

2. Цілі вироблення дослідної партії:

1.1 Відпрацювання оптимальних режимів технологічних операцій, що становлять процес виробництва йогурту з додаванням вівсяного бетта-глюкану.

2.1 Підтвердження можливості використання існуючого серійного обладнання з цієї технології.

2. Призначення продукту.
Продукт призначений для споживання, як альтернатива для традиційного йогурту.

3. Рецептура та норми витрат на 100 кг готового продукту наведені у додатку.

4. Контроль технологичного процессу.

В ході проведення випробувань температуру контролювали місцевими термометрами, активну кислотність вимірювали за допомогою pH-метра П-201 з електродом Інгольд.

5. Висновки та рекомендації.

1.1 У процесі проведення випробувань для отримання вищезгаданого продукту використовували серійне обладнання, таким чином, технологія виробництва йогурту з додаванням вівсяного бетта-глюкану може бути реалізована на вітчизняних підприємствах.

2.1 Відпрацьовано режими технологічних операцій, що становлять процес виробництва йогурту з додаванням вівсяного бетта-глюкану: температура пастеризації (95 ± 2) °С з витримкою 15 сек ÷ 3 хв; температура сквашування (42 ÷ 44) °С; термін сквашування 8-9 годин.

В результаті проведених випробувань отримано йогурт з додаванням вівсяного бетта-глюкану в кількості трьох партій по 100 кг.

Від підприємства філії «Сумський молочний завод» ДП «Аромат»:

Директор _____ Белік Л.В.

Головний технолог _____

Від Сумського національного аграрного університету:

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Production process and operation key points of produce OG set-type yogurt

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1. Objective：

Standardize product production and ensure product quality.

2. Application scope:

It is suitable for the OG set-type yogurt products of Tianjin Huaming Dairy Co. LTD.

3. Product formula:

Fresh milk 4985 kg, Oat β-glucan 15 kg, Strain YO-MIX300 1000DCU. Therein, fresh milk fat ≥ 3.2%, Oat β- Glucan (purity) ≥ 95.0%, whole milk solid ≥ 11.5.

4. Process content:

4.1 Technological process:

Raw milk receive → filter → cooling, storage → dissolve OG→ static hydration → homogenization* → adding starter* → stirring → filling → packing → labeling → fermentation* → warehousing.

Note: * indicates the key process

4.2 Production process points

4.2.1 Raw material reception:

The quality of raw milk is higher than that of general dairy raw milk. In addition to passing the acceptance inspection according to the regulations, the following requirements must also be met: (1) Raw milk color is milky white or slightly yellow, the smell is fragrant, and there is no peculiar smell. (2) The total milk solids shall not be less than 11.5%. (3) Bacteria-contaminated milk, milk containing antibiotics or

fungicides shall not be used. (4) Do not use the milk of cows suffering from mastitis, otherwise it will affect the flavor of yogurt and the gel strength of protein.

4.2.2 Purification of raw milk

Raw milk may contain cellular substances (such as epithelial cells and leukocytes, etc.), some of which are inherent in cow's milk, and some are brought into milk by contamination of cow's milk during the milking process, and are brought into raw milk at the same time There are also pollutants such as straw, leaves, hair, dirt and seeds. In order to ensure a good final product quality, the first step in the production of yogurt is to remove these contaminants. There are many ways to remove these impurities, one of the most widely used is filtration. The specially designed centrifuge (ie. the milk purifier) used to remove the white blood cells and other visible foreign bodies in the milk.

4.2.3 Chopping Standardization of raw milk

The composition of milk produced by different types or different places of milk is very different. Even if it is the milk produced by the same cow, its chemical composition will also be affected by other unavoidable factors (such as different lactation periods) during lactation. There are fluctuations in stage, age of the cow, interval and efficiency of milking, seasonal and environmental temperature changes, the way the cow is raised, the nutrition of the cow, the disease the cow suffers from, such as mastitis, etc.). To compensate for these inherent fluctuations in milk composition, raw milk must be standardized or fortified with specific ingredients before it can be used as an ingredient to make yogurt. This standardization process includes the standardization of fat and non-fat milk solids, especially the protein content will be directly related to the viscosity and texture of the product, so the standardization of non-fat milk solids is very important. The ultimate purpose of

standardization of raw milk is to correct the chemical composition of milk within the scope permitted by food regulations and according to the quality characteristics of the desired lactic acid finished product, so as to ensure that the quality of each batch of products is stable and consistent.

4.2.4 Ingredients

This product is a set yogurt with OG added. OG is added before fermentation. First, the consistency between batches of OG raw materials must be ensured. Secondly, OG should be added after the fresh milk has been heated up, stirred evenly, homogenized, and fully hydrated at rest.

4.2.5 Sterilization and cooling

The purpose of sterilization is to kill most of the miscellaneous bacteria and all pathogenic bacteria in the raw milk, to ensure the normal growth and reproduction of *lactic acid bacteria*; to passivate the natural inhibitor that has an inhibitory effect on fermentation bacteria in the raw milk; to make the whey protein in the milk Denaturation to improve tissue state, increase viscosity and prevent precipitation of finished whey.

The sterilization method is to heat the raw milk to 95 ℃ for 5 min; 85 ℃ for 30 min; 95 ℃ for 5 min to 10 min; 118 ℃ to 135 ℃ for 3 s to 5 s. After sterilization, the base liquid is cooled to about 45 ℃.

4.2.6 Inoculate

The sterilized milk should be cooled to about 45℃ immediately in order to inoculate the starter. The amount of inoculum depends on the activity of the strains, the fermentation method, and the arrangement of the production time and the ratio of the mixed strains. The production starter is *Streptococcus thermophilus* and

Lactobacillus bulgaricus, the ratio is 1:1 or 2:1, and the general inoculation amount is 200 DCU/T (operated in a sterile room). The added starter should be stirred into a uniform and delicate state under aseptic operating conditions in advance, and there should be no large clots, so as not to affect the quality of the finished product. Stir for 5 min after inoculation, so that the starter is evenly distributed in the milk.

4.2.7 Filling

Use sterile glass bottles or sterile cup, use automatic aseptic filling and semi-automatic filling. Keep the upper part of the product as small as possible and do not get the packaging material wet. Avoid air pollution and keep the room sterile. When filling in plastic bottles, the caps or plate cover should be tightly closed to avoid contamination by mold and yeast.

4.2.8 Labeling, packing

Check whether the printing date is correct and clear, and whether there is ghosting, less code, and broken code. Conduct sealing inspection on the finished products. Take 3-5 cups for inspection. If there is any slight leakage, stop immediately and adjust.

4.2.9 Fermentation

After filling and capping , it is quickly sent to the fermentation room, and it is fermented at 41~43℃ for 5 h. The fermentation can be terminated when it reaches a solidified state, that is, when the acidity reaches 70 °T~75 °T, the heating is stopped, the pH value is lower than 4.6, and a small amount water marks appears on the surface, milk thickened. Fermentation should pay attention to avoid vibration, otherwise it will affect the state of the organization; the fermentation temperature

should be constant to avoid high and low; master the fermentation time to prevent insufficient or excessive acidity and whey precipitation.

4.2.10 Cool down

Cooling can quickly and effectively inhibit the growth of lactic acid bacteria, reduce enzyme activity, prevent excessive acid production, and prolong the shelf life of yogurt. There are two cooling methods: (1) direct cooling method. To the end of fermentation,immediately put the yogurt into the 2 ℃~ 6 ℃ cold storage (or cut off the power immediately). (2) Pre-cooling method. At the end of fermentation, the temperature is gradually decreased in stages, that is, 42 ℃~ 45 ℃→35 ℃~ 38 ℃→19 ℃~ 20 ℃→10 ℃~20 ℃→5 ℃, 5 ℃ is the lower limit of mold and yeast growth temperature. During the cooling process, handle with care to prevent vibration. Yogurt is very sensitive to mechanical shock, and once the tissue state is destroyed, it is difficult to restore.

4.2.11 refrigeration and warehousing

Yogurt must be stored at 2 ℃ ~ 6 ℃ for 12 h, which can promote the production of aromatic substances and increase the viscosity of yogurt products. Set-type yogurt products are gelatinous, white and opaque, smooth and soft, and have a unique flavor of fermented milk. The packaged products should be stored in a warehouse at 0~4 ℃ with a shelf life of 21 days.

5. End of production

At the end of production, first of all, all operators should carefully check the equipment at all times until the end of production, then stop the machine, count the

output of the shift, turn off the power, wipe the equipment with alcohol, and disinfect the workshop.

6. CIP cleaning

After production, connect the material pipe with the CIP circuit, and the CIP personnel shall clean it. During the cleaning process, special personnel shall be assigned to check the cleaning effect, and coordinate with the CIP personnel to eliminate abnormal conditions in time.

7. Production records

Carefully fill in the "operation record form", "random inspection record form"and various statements. It is required that the items are complete, the data are true, accurate and clear, there is no alteration, no contamination and loss, and timely transfer.

8. If the production equipment fails, it should be indicated in the column of "equipment operation status".