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# INTEGRATION OF SPENT GRAIN INTO FOOD PRODUCTS

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**Abstract.** Spent grain is a component of grain that consists primarily of its shells and is a byproduct of the production of beer or ethyl alcohol. It is characterized by a diverse chemical composition, mainly composed of proteins and dietary fibers and, to a lesser extent, lipids, minerals, phenolic compounds, as well as B vitamins and vitamin E. In its native form, it is a product with a high moisture content, which makes it unsuitable for long-term storage. In this connection, several problems arise that worsen the economic and environmental situation at the global level. This article presents effective methods of processing spent grain to prevent negative consequences associated with its irrational use. Possibilities for the application of used spent grain in the food industry are also presented: bakery and pasta, confectionery, the meat and dairy industry, as well as the production of beverages. Thus, the integration of spent grain in the food composition will lead to solving the economic and environmental difficulties that have appeared in the last decades; it will allow the development of a functional food market, which will lead to the improvement of the quality of the population life by strengthening the trend of good nutrition and reducing the negative impact on the environment.

**Keywords:** spent grain, waste, biological value, bioprocessing, food products.

**Rezumat.** Borhotul este o componentă a cerealelor, care constă în principal din coaja acestora și este un subprodus al producției de bere sau etanol. Se caracterizează printr-o compoziție chimică diversă, alcătuită în principal din proteine și fibre alimentare și, într-o mai mică măsură, din lipide, minerale și compuși fenolici, precum și din vitaminele grupei B și vitamina E. În forma sa nativă, prezintă un produs cu un conținut ridicat de umiditate, ceea ce îl face nepotrivit pentru depozitarea pe termen lung. Acest fapt ridică o serie de probleme care agravează situația economică și de mediu la nivel global. În acest articol sunt prezentate metode eficiente de prelucrare a borhotului pentru a preveni consecințele negative asociate cu utilizarea lui irațională. De asemenea, sunt prezentate posibilități de aplicare ale borhotului în industria alimentară: panificație și paste făinoase, cofetărie, industria cărnii și a produselor lactate, precum și producția de băuturi. Astfel, integrarea borhotului în compoziția alimentelor va conduce la rezolvarea dificultăților economice și de mediu care au apărut în ultimele decenii; va permite dezvoltarea unei piețe de alimente funcționale, ceea ce va duce la îmbunătățirea calității vieții populației prin consolidarea tendinței de bună nutriție și reducerea impactului negativ asupra mediului.

**Cuvinte cheie:** *borhot, deșeuri, valoare biologică, bioprocesare, produse alimentare.* 

# 1. Introduction

Every year the world population increases, which, in turn, directly affects all spheres of human life, in particular, nutrition, both individual and whole social groups, as well as countries and continents. According to the World Bank, in the period from 2000 to 2022, the population growth varies from year to year in the range from 0.8 to 1.4% worldwide [1]. On this basis, not only the need for food but also the quantity of production at the industrial level is increasing. Along with the augmentation in population and annual volumes of produced and consumed products, there is also an increase in food waste, which has recently been considered a global environmental and economic problem that has affected the whole world and every part of it.

It is generally accepted that annually about 1/3 (30%) of the manufactured food products do not find their direct use, i.e., are lost, which is approximately more than one billion tonnes of products annually [2]. Also, if we consider the annual food waste per capita, in industrialized countries this figure is 95-115 kg/year, while in African and South Asian countries food waste per person is much lower (more than 10 times) and ranges from 6-11 kg/year [3]. In developing countries, the causes of food losses and waste are financial, technical, production, and organizational constraints, which are caused by the weak economies of the countries. At the same time, the listed constraints contribute to the aggravation of the current economic situation. In countries with developed economies and food industries, the cause of food waste is the human factor, i.e. consumers who buy excessive quantities of products daily, which are then thrown away, but this also affects the economic component of the countries.

In addition to the economic problem, it should be noted that unused food has a detrimental effect on the global ecological system. This entails a number of changes: climate restructuring, deterioration of air quality, water quality, and other vital components. These changes lead to a lower quality of life in general.

In addition, it should be noted that food loss and waste generation can occur at different stages of the production chain, which is based on the "farm to fork" strategy:

- In the fields, farms, and other components of agriculture.
- During the processing of raw materials into finished products.
- At the point of Retailing directly to the consumer or wholesale to intermediary organizations.
- At the point of storage and domestic use by consumers.

According to the Food and Agriculture Organization of the United Nations, the percentage of food waste at the mentioned sites in Europe is 23% for the agricultural sector,

17% is due to waste resulting from the processing of raw materials, 9% is due to losses at the point of sale and the highest percentage is due to consumer losses (52%) [4].

In this way, there is a need to find solutions to minimize all kinds of losses in the industrial process of food products, which is rapidly developing every year within the food industry. This is reflected in various developments related to zero-waste production, which implies the use of by-products of food processing, such as fruit and vegetable pomace, press cake and grist, spent grain, and other things that can be reused in food production.

The above is one side of the current situation in the food industry and the food market. The other side is represented by the increasing demand for healthy and improved products. This can be explained both by the above-mentioned problems and personal preferences, most of which are aimed at taking care of one's health through proper and, importantly, safe nutrition. It follows that a manufacturer interested in selling its goods will look for ways and methods that will allow it to produce appropriate food products without incurring material losses. One such solution may be the use of edible by-products in the technology of certain products, which will reduce production costs and, in some cases, increase the nutritional value of the product. As this trend grows, there is a high probability that over time the amount of food waste will be significantly lower. This fact will have a direct favorable impact on the ecological state of our planet and the health of the population, as well as on the economic performance of countries.

Referring to the points mentioned above, the main objective of this work was to choose a bibliographic analysis of waste-free technologies, namely the possibility of using spent grain in food products, which can contribute to the reduction of production waste and increase the biological value of the food into which it will be integrated.

#### 2. Definition and classification

Spent grain is a by-product of beer, ethanol, and other alcoholic beverages such as whisky production [5,6]. Spent grain is the components of grain consisting mainly of its shells which are not soluble [7]. To date, it is brewer's spent grain that has gained the most popularity, as evidenced by a significant number of scientific papers [5-7]. This type of waste is formed after malt mashing, i.e. at the stage of mixing milled malt with drinking water and soaking at certain temperatures, with further filtration. Therefore, the water-insoluble fraction is the brewer's spent grain, and the liquid part is wort, which is directed to further technological operations for beer production [7].

The spent grain produced at the initial stages of beer production accounts for 85% of the total amount of secondary products that are generated during all the technological stages of production of this beverage, which indicates a high level of losses and the corresponding economic and environmental problems associated with its utilization [7]. Other sources mention that when producing beer in quantities equal to 1000 t, the numerical value of solid waste generated varies from 137 to 173 t. However, solid brewing waste includes not only brewer's spent grain but also a certain amount of yeast, which at the end of its life activity has precipitated [8]. In any case, unused components account for approximately one-sixth of the product produced, reflecting significant technological losses that directly affect the economy of the enterprise, reducing its efficiency.

Thus, for example, in the period from 2020 to 2022, the production of beer from malt in the Republic of Moldova averaged 8372 thousand dal, distributed by year as follows: 2022-8418.3 thousand dal, 2021-8790.8 thousand dal and 2020-7901.1 thousand dal [9]. Based

on the above information, it can be assumed that the amount of solid waste averaged 14511.5 t for the mentioned years. The numerical values of solid waste and beer from 2020 to 2022 are displayed in Figure 2.

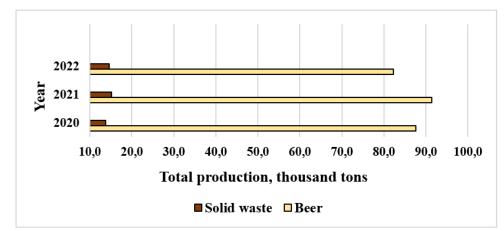


Figure 1. Production of beer and solid waste, thousand tons (2020-2022) [8,9].

The spent grain can also be obtained in whisky production at the end of the malt mashing step. Due to the expansion of the range and variety of raw materials used (different types of cereal crops and their mixtures) for beer making, the number of types of spent grain obtained is also increasing. Thus, it can be obtained from barley and rye, oats, maize, wheat, and rice [6].

In the case of the alcohol industry, the appropriately named spent grain is a secondary raw material or a waste product resulting from ethanol production. The production of alcoholic spent grain is similar to the above type. It is formed during the filtration of mash, which is a mixture of the initial grain raw materials with water, which has undergone the stages of cooking and saccharification. The spent grain obtained during alcohol production has a dense, homogeneous consistency with specific organoleptic indicators such as taste and smell, as well as color from dark yellow to brown. The percentage of spent grain obtained during ethanol production is also quite high. It varies from 60 to 65% of the original grain weight, which in physical units can be represented as 18-19.5 kg/day of alcohol produced [10].

# 3. Chemical composition

Referring to the above-mentioned, spent grain has interested researchers from different parts of the world, because, being a by-product of brewing and ethyl alcohol production, they are not used further in production, but represent production waste and have some negative consequences: a decrease in the output of finished products and, accordingly, the profit of producers and, no less importantly, affect the environmental situation of both individual countries and the world. It is stated that 70% is distributed for animal feed as an alternative to zero-waste production, 10% is processed into biogas and the remaining 20% is disposed of in landfills [11]. In addition, it was found that spent grain has a rich chemical composition of spent grain can vary depending on the quality and type of grain raw material used, the time of harvest, and the malting and mashing conditions. Nevertheless, spent grain contains a high amount of macro- and micronutrients such as proteins and, respectively, substitutable and essential amino acids in their composition, fats and fatty acids, polyphenolic compounds, vitamins and minerals, and at the same time dietary fiber [12]. For this reason,

new research work is increasingly being observed in the food and agricultural industries, medicine, and other fields to determine the potential of recycling this type of waste with maximum benefits for people and the environment.

Due to the structure of the raw material used in beer production and alcohol production, namely malt, the chemical composition of the resulting spent grain is due to its high biological value and it is common to consider this by-product as a lignocellulosic material. This is since the spent grain is predominantly the outer component of the grain (the shell), from which follows an increased content of dietary fibers such as cellulose (a polysaccharide of a linear structure consisting of glucose residues), hemicellulose and lignin (a phenolic polymer), which are insoluble dietary fibers. Their quantity can reach up to 50% of the total weight of the spent grain. Regarding hemicellulose, it should be mentioned that in spent grain it is contained in the form of arabinoxylans, non-starch biopolymers, which are present in all parts of cereals and consist of arabinose and xylose. In brewer's spent grain, the arabinoxylan content can reach up to 40 percent of the total dry matter. In addition, the spent grain has a high monosaccharide content in terms of dry matter, in particular glucose, xylose, and arabinose. In addition to fiber, spent grains are also rich in proteins, the content of which varies from 15 to 25%, and to a lesser extent in fats and residual endosperm starch [13,14]. The proteins of spent grain include about 30% of essential amino acids, which cannot be synthesized by the animal organism, but play an important role in the life support of these organisms, including the human organism. The most common essential amino acid included in the composition of spent grain is lysine. In addition to lysine, essential amino acids in spent grain include leucine and isoleucine, methionine, tryptophan, phenylalanine, and threonine, while non-essential amino acids include cysteine, glycine, proline, tyrosine, serine and others [7]. The most common lipids found in the composition of spent grain are triglycerides and their content varies from 55 to 67% (in the case of brewer's spent grain), the content of free fatty acids is inferior to triglycerides and is 18-30% (typical for brewer's spent grain). Free fatty acids include linoleic acid, which is an essential fatty acid ( $\omega$ -6), as well as oleic acid ( $\omega$ -9), stearic acid, and palmitic acid. In addition, lipid composition acts as a source of volatile compounds, including acetic acid, butyric and propionic acids [15]. Moreover, compounds of phenolic origin can be found in the composition of spent grain, which is most represented in the form of ferulic and p-Coumaric acids [16].

Other elements included in spent grain include vitamins and minerals-microelements required by the human body in milligrams and micrograms to support its vital functions. The minerals include silicon, calcium, cobalt, copper, iron, magnesium, phosphorus, potassium, selenium, sodium, sulfur, and manganese. The vitamin composition is mainly represented by water-soluble B vitamins (for example, biotin, pantothenic and folic acids, thiamin, riboflavin, and others) and fat-soluble tocopherols (vitamin E) [14].

#### 4. Biological value

Having considered the chemical composition of spent grain and its peculiarities, it is possible to assert the high biological importance of this by-product, which should not be ignored, but on the contrary to consider in more detail and determine the possible favorable effects on the health and life activity of the population. With its high fiber and protein content, as well as the presence of essential amino acids and fatty acids, vitamins, minerals, and phenolic components, spent grain can be analyzed as a potential ingredient in food products.

Hence a need to investigate the health benefits to the human body of both spent grain as a whole and its constituent components [11-14].

### 4.1 Dietary fiber

Dietary fiber is a class of substances in which polysaccharides, specifically carbohydrates with three or more monomers, occupy the major part. However, the exception is lignin, which refers to phenolic compounds. A distinctive feature of this class is that they are resistant to the influence of endogenous enzymes of the digestive tract, as a consequence of which they are not broken down and assimilated by the small intestine, but still fulfill their role affecting the organism [17].

The composition of spent grain consists largely of insoluble dietary fiber, which includes cellulose, hemicellulose, and lignin, and to a lesser extent, soluble fiber. A major part of soluble fiber is identified as  $\beta$ -glucans, which range from 4.7 to 13.4% of all carbohydrates in barley brewer's spent grain [15].

Research has been conducted for many years related to dietary fibers and their effects on the human body. They have the potential to reduce low-density cholesterol [18], to minimize the risk associated with diseases such as coronary heart disease, diabetes mellitus, and obesity, and also dietary fiber can improve intestinal peristalsis, which has a beneficial effect on the entire gastrointestinal tract [19, 20]. Regarding the differentiation of fiber into soluble and insoluble fiber, some differences have also been found in their effects on the body: soluble dietary fiber has immunomodulatory and anti-inflammatory effects, while insoluble fiber stimulates the intestine, being the main source of energy for its microbiota [21-23].

# 4.2 Proteins and amino acids

Proteins are biopolymers whose monomers are amino acids linked by peptide bonds. These substances play an integral part in the formation and repair of cells and tissues and are part of salts that are involved in maintaining osmotic balance. Also, amino acids form various complex protein molecules (glycoproteins, nucleoproteins, lipoproteins), hormones, enzymes, antibodies, and other protective components of the immune system, which allow our body to function normally [24].

Traditionally, the biological value of protein is determined based on its amino acid composition (qualitative and quantitative, as well as the ratio of essential to substituted amino acids), while taking into account individual human needs and the degree of digestibility and bioavailability of this type of protein by the body [25]. Despite the lack of precise data concerning the quality of spent grain proteins, we should not neglect the fact that in its composition the percentage of protein can reach 30%, which is not inferior to some types of food products of animal origin. It should also be added that brewer's spent grain from barley contains 7 essential amino acids (lysine and leucine have the highest proportion) and 11 non-essential amino acids in varying amounts, of which histidine and glutamic acid are predominant [7].

In turn, in addition to the fact that amino acids are part of proteins and fulfill a building function in the human body, the deficiency of each of them leads to a number of negative consequences that can be avoided with proper nutrition, which includes not only the necessary qualitative composition of amino acids but also a sufficient amount of them. For example, lysine has an antiviral effect, is able to reduce the level of triglycerides, and ensures the growth of bone tissue. Its deficiency contributes to a decrease in the number of red blood

cells and hemoglobin, provokes a lack of calcification of bones, and degenerative changes in muscles, liver, and lungs can be observed. Considering the fact that it is found in relatively small amounts in cereals and the daily requirement is high, its elevated content in brewer's spent grain may be one of the solutions to this problem [26].

### 4.3 Lipids and fatty acids

Lipids are organic high molecular-weight substances consisting of fatty acids and glycerol esters. One of the main functions of these nutrients is to saturate the body with energy, as well as the ability to reserve it. Other functions include plastic (they are part of cell membranes), thermal insulation, and regulatory functions (fats are precursors of steroid hormones), in addition to being carriers of fat-soluble vitamins [24].

The biological value of fats, as well as proteins, is directly related to their constituent parts, i.e. it is determined by the presence of essential fatty acids, both quantitatively and qualitatively, as well as their bioavailability [24].

Based on the lipid composition of spent grain, in which triglycerides and a smaller amount of free fatty acids predominate, it can be concluded that this product has a fairly high energy value directly due to neutral fats (triglycerides) and nutritional value due to free essential fatty acids ( $\omega$ -6), as well as monounsaturated fatty acids [15,24]. Several studies have shown that consumption of foods containing linoleic acid leads to a reduction in lowdensity lipoprotein levels, consequently minimizing the risks of cardiovascular disease [15,24,27]. In addition, linoleic acid can reduce the level of triglycerides in the blood consumed with food and restrain the occurrence of cardiac arrhythmias [27]. In addition to the above,  $\omega$ -6 fatty acids preserve the integrity of cell membranes, intensify the synthesis of hormone-like substances, reduce psycho-emotional stress, and strengthen the functional state of the dermis [28].

Despite the fact that oleic acid, which is a part of spent grain is not an essential fatty acid, it, as a monounsaturated acid, has a number of positive effects on the body: it has a preventive effect against heart attacks, regulates blood cholesterol and carbohydrate metabolism, improves memory, and has anti-inflammatory effects [28].

### 4.4 Phenolic compounds

Phenolic substances are the most widespread group of natural biologically active substances, the number of which exceeds 8000 compounds. They also belong to the main classes of secondary metabolites of plants [29]. From the chemical point of view, phenolic compounds are substances consisting of a benzene (aromatic) ring and one or more hydroxyl groups (-OH), in particular their derivatives [30]. This class of compounds is widely known for its biological properties, which include: antioxidant, anti-inflammatory, and antimicrobial abilities, can act as cardiovascular and neuroprotective agents, and helps to reduce the risk of cancer and diabetes [29].

Ferulic and *p*-coumaric acids have been identified as the main phenolic compounds in the composition of spent grain, which belong to the hydroxycinnamic acid group, formed from cinnamic acid, and, most often in a bound state [16,31]. Hydroxycinnamic acids have a simple chemical structure with a C6-C3 phenylpropanoid at the base and the carboxyl group located in the side chain [29].

In turn, hydroxycinnamic acids, as phenolic compounds, have all the above physiological properties and contribute to the quality of life. For example, these acids play

an important role in the prevention of coronary heart disease due to their ability to inhibit oxidative changes in low-density lipoproteins and total cholesterol [32].

So, the presence of hydroxycinnamic acids in the composition of spent grain also suggests an increased biological value of this industrial waste, which raises even more interest for further research on its integration into food products.

# 4.5 Vitamins and minerals

Micronutrients, which include vitamins and minerals, are essential substances for maintaining the normal functioning of all body systems, i.e. they are biologically important elements for humans [33]. Although a small number of micronutrients is sufficient to maintain the vital activity of the organism relative to the needs for proteins, fats, or carbohydrates, their deficiency can lead to serious disorders, some of which cannot be determined at the initial stages. For example, vitamin E deficiency can provoke the development of Bassen-Kornzweig syndrome (a disorder in which fats and fat-soluble vitamins from food cannot be absorbed), leading to ataxia and systemic pathology-mucoviscidosis. Alternatively, potassium deficiency is one of the causes of cardiac dysfunction [33].

Based on all of the above, it is appropriate to conclude that in terms of its functional features, spent grain is of great importance for the health and normal functioning of the human body, which is explained by the main components included in its composition. However, since this type of by-product is not recommended for human consumption in its native state, it is possible to ensure its maximum nutritional value through the production of certain food products - foods enriched with spent grain. In this case, the biological value will not only not be lost or reduced, but, on the contrary, may even be increased.

# 5. Spoilage and preservation methods

Despite the above-mentioned advantages of the composition of spent grain as a potential ingredient in the food industry and concerning the human body, the peculiarities of the chemical composition also generate several difficulties that can induce serious problems. In such cases, not only the quality of the waste but also its quantity plays a role. Thus, based on the previous information, malt processing generates a significant amount of spent grain as waste: per 100 kg of malt, approximately 100-130 kg of spent grain with a high water content (70-80%) are generated. Due to high moisture content during storage, the spent grains sour and lose their nutritional value [34]. In addition to high moisture content, spent grains are rich in polysaccharides and proteins, which makes them more vulnerable to microorganisms and becomes an attractive growth medium for them. In one study, spent grain was stored for thirty days at room temperature, and eight isolates of some fungal genera, including Aspergillus, Fusarium, Penicillium, Rhizopus, and Mucor, were found [7]. This fact hurts the safety and stability of the spent grain, as high concentrations of these fungi contribute to the accumulation of mycotoxins with strong toxicity. Also, despite its microbiological stability after production, the spent grain is subject to rapid multiplication of microaerophilic and anaerobic microorganisms, which reduces both the stability and safety of the waste. In addition to the reduced shelf life in native form, the transport of wet spent grain is also difficult and materially expensive [7]. Accordingly, when considering spent grain as a component of food products, their stabilization and optimization of storage conditions are necessary.

One of the directions of spent grain stabilization is the reduction of moisture content, which can be achieved by drying. In some countries, beer-producing companies use special

installations that allow to reduce the moisture level in the spent grain and this is done in two stages. In the first stage, the moisture level is reduced to 65% and below by pressing, while the second stage reduces the moisture content to below 10% [13]. The mechanical dewatering of the spent grain in the first stage can be done in two ways: using a press-screw separator or a hydrocyclone thickener [34].

As a second step, that is drying, several methods have been proposed: freeze-drying, drying oven, and superheated steam drying [7]. The analysis of the chemical composition of the spent grain after lyophilization application did not reveal significant changes, which may indicate that the nutritional value of the secondary product is preserved. However, freezedrying of the spent grain was not economically efficient. The drying oven also showed no changes in the chemical composition of the spent grain and is considered to be a fairly acceptable method of drying them, despite its energy intensity. It should be noted that the temperature in the chamber oven should not exceed 60 °C, as higher temperatures can lead to an unpleasant taste and a strong color change (darkening). There is also a risk of increasing the temperature of the grain as it approaches the exit of the drying chamber, which may contribute to the burning of the dried sample [7].

As an alternative method of drying, superheated steam has started to be used. By its specificity, it is more economical and effective, because this method uses less energy due to the circulation of steam in a closed circuit than in a drying oven, but the degree of extraction of organic compounds increases. In addition, the use of superheated steam reduces carbon emissions into the environment and minimizes the risk of explosion [13]. When using this method, attention should be paid to the rate at which the steam passes through the material and the temperature inside the unit, as excessively high temperatures (around 180 °C) have been shown to affect starch gelatinization [7, 13].

Other methods to extend the shelf life of spent grain include the following: cold preservation and preservation with food acids [7,13]. Freezing or cold preservation was found to be impractical because freezing requires large production areas and significant energy costs to maintain a certain sub-zero temperature. Also, changes in the quantitative content of arabinose have been found during freezing [7,13,14].

Solutions of lactic, acetic, formic, and benzoic acids were used for the preservation of spent grain with food acids. Increased efficiency of benzoic and methanoic acids application is highlighted.

Despite the positive results in increasing shelf life, the application of this technology may affect the demand for the product in which the acid-treated spent grain will be used. This is because there is currently a trend towards the consumption of products based on natural ingredients, with so-called "clean labeling" [7].

Summarizing the above, we can conclude that the most attractive method of extending the shelf life of spent grain is drying, which allows us to obtain a fairly stable product with minimal changes in composition and with acceptable energy costs. However, given that the drying process has several variations, it is necessary to choose the most appropriate method for a particular case in order to achieve the highest results from the process.

#### 6. Nutrient extraction

Based on the chemical composition analysis and the conclusions drawn earlier, it is necessary to consider ways of extracting those nutrients contained in the spent grain for their further use for various purposes, such as food fortification the creation of bioactive supplements, and so on. This will not only diversify the market but also allow to use of food industry waste, which will have a favorable impact on the environmental situation in the countries and will have a positive impact on the economic component of individual industries that generate this type of waste [34,35].

#### 6.1 Protein extraction

One of the peculiarities of spent grain at the time of its obtaining is its high moisture content (70-80%), which makes it necessary to perform an additional operation. This treatment includes the drying process and allows for prolonging the stability of the product, reduces its volume, and facilitate the processing of the material for the extraction of protein substances [34,35]. After the initial processing of the component used, a number of methods are applied to allow the extraction of protein products, of which alkaline extraction is the most widely used. In addition to drying, prior to the extraction process, the spent grain may be subjected to various pre-treatments to facilitate the release of protein from the spent grain structure. These can include degreasing, particle size reduction by grinding, enzymatic hydrolysis, and ultrasound. In addition, enzymatic hydrolysis and ultrasound can be applied as independent extraction methods [35].

Alkaline extraction is a sufficiently studied method for protein extraction not only from spent grain but also from other agricultural products. The essence of the method is that the alkaline medium provides solubilization of protein, thus changing its configuration, charge, and, accordingly, interaction with other substances [35]. One of the most used alkalis is sodium hydroxide in the obtaining of protein components. The main parameters of this method that determine its efficiency are the type and concentration of the alkali used, the extraction temperature, the ratio of solid and liquid phases, as well as the values of the isoelectric point at which protein precipitation occurs. By modifying the above conditions of the method, a rather wide range of protein extraction yields (18-82%) was obtained, and the values of its purity varied from 37 to 69% [35].

Since the composition of spent grain, in addition to large amounts of protein, also contains significant concentrations of hemicellulose, the method of extraction with the help of acids and reducing agents, such as sulfuric acid and sodium bisulfite, is used. This increases the solubility of proteins and facilitates their release by hydrolysis of the said polysaccharide and breaking disulfide bonds in proteins [38]. In addition to acids, this extraction method involves autoclaving the material at temperatures ranging from 121 to 130 °C and lasting 30-60 min, resulting in an accelerated process of decomposition of cell wall components. When acid extraction was used, the yield of protein substances was 63-90%, which is superior to the previous method, but the protein purity was reduced and was 24-39% [35].

Another method of protein extraction from the composition of spent grain is solvent extraction. Among the tested methods, the following two methods were considered specifically for spent grain: deep eutectic solvents (DES) and extraction in the presence of pressurized solvents. Deep eutectic solvents simplify the process of protein extraction due to the possibility of fractionating and dissolving lignin and starch, which are part of the material used [37]. When deep eutectic solvents are used, the heating parameters include a temperature of 80 °C for 4 h (low temperatures and long-time intervals), while if solvents are used in combination with pressure, the heating is carried out at higher temperatures (150 °C) and the duration is shortened accordingly. The proteins are then separated into a liquid fraction. The disadvantage of these methods may be the violation of functional characteristics of proteins due to the impact of high temperatures, as well as the need to analyze the

maximum permissible concentrations of each type of solvent and additional studies of their toxicity and impact on the human body. At the same time, the degree of protein extraction by these methods varies from 69-79% depending on the solvents used [35].

Hydrothermal and subcritical methods, which use water as a solvent, can be considered environmentally friendly and chemical-free methods of protein extraction from spent grain. In the case of hydrothermal extraction, high or low temperatures are applied at short and long-time intervals, respectively, to facilitate protein extraction. Subcritical extraction uses water brought to a temperature above 100 °C and not converted to vapor by a special pressurized system. According to the results of the analyses carried out, the protein yields were similar and, in some cases, higher than those of acid and alkaline extractions. For example, using hydrothermal protein release, 66% of the protein compounds were extracted and the protein purity was 53%. The treatment was carried out for 24 h at 60 °C [35, 38]. Subcritical extraction conditions allowed the colloidal dissolution of about 78% of proteins from the spent grain. These conditions implied a temperature regime of 180-185 °C maintained for 150 min [39].

Enzymatic extraction is designed to solubilize protein by breaking down carbohydrates and proteins in their matrix, resulting in partially degraded protein in the process of hydrolysis (protein hydrolysate). Carbohydrase and protease enzymes are used to perform the above actions. The yield of extracts in this method is quite wide and ranges from 31-86%, and protein purity is mostly equated to 40% [35].

It was also found that the use of ultrasound treatment had a positive effect on the amount of protein released. In one study, the yield increased from 46 to 86% after ultrasound treatment with a power of 250 W and a duration of 20 min at the time of alkaline extraction [40]. The use of microwave radiation during alkaline extraction also helps to increase the extraction rate of protein compounds. In addition, the use of microwaves during hydrothermal extraction provided an extract yield of about 90%. These values were achieved as a result of a ten-minute exposure at 110 °C and a maximum microwave power of 1800 W [35, 41].

#### 6.2 Extraction of carbohydrates

As for the extraction of proteins, so for the extraction of polysaccharides from spent grain, chemical and enzymatic extraction methods with or without primary treatment are mainly used, less often combined, as the full range of their action is not yet fully understood [7, 42]. To date, when choosing a method of extraction of components that will later become part of the technological chain for the production of food items, preference is given to enzymatic methods. This is due to a number of factors related to the safety of the extract obtained. Firstly, the use of enzymes allows for closer monitoring of the process and the nature of the components released. Secondly, it will allow for greater preservation of nutritional value and environmental friendliness, which will present food products containing extracts obtained by this method in a more favorable light to consumers. Thirdly, the enzymatic extraction process does not generate potentially toxic by-products, which also makes it more stable and environmentally friendly [43].

However, difficulty arises during extraction due to the complexity of the structure of the polymers comprising the spent grain, hence a complex of enzymes must be used to fully hydrolyze them. For example, the final degradation of hemicelluloses will require the use of several different enzymes, which are of the following types: xylanase,  $\theta$ -xylosidase, feruloyl esterase, acetyl esterase, glucuronidase, glucuronoyl esterase and  $\alpha$ -L-arabinofuranosidase [7].

In addition to studies on the extraction processes of components from spent grain, various methods are also being investigated to facilitate the extraction of these components. These include the use of microwave radiation or ultrasound, as well as extrusion processing [7]. Thus, one of the works considered the effect of physical and thermal primary treatments, relying on the improvement of extraction yield and efficiency. These treatments were represented by grinding and exposure to microwaves. Microwave treatment combined with acids and alkalis proved to be the most effective method. In this type of treatment at a temperature of 160 °C for 10 min and in the presence of sodium hydroxide, the polysaccharide yield was 49%, while the physical treatment by grinding did not significantly change the monosaccharide yield [44].

Ultrasound can also be used as a stand-alone method, which is used for the isolation of arabinoxylans. When comparing this method with alkaline extraction, it was observed that the use of ultrasound waves helps to shorten the process time and is consequently a less energy-intensive method [7].

### 6.3 Extraction of phenolic compounds

Phenolic compounds, being natural biologically active substances, are components that can significantly increase the functional value of products, which is currently one of the directions in the creation of innovative food items. Therefore, there is a need to search for new methods of their production and growing interest in the study of alternative sources of phenols and their derivatives. One of the solutions may be the extraction of phenolic compounds from spent grain, which is carried out by various methods, each of which has a number of its features. Modern developments are based on the use of advanced extraction methods: microwave radiation, hydrolysis in the presence of acids, saponification with sodium hydroxide, as well as liquid and solid-liquid extraction. Most often the isolation of phenolic acids from the composition of spent grain resorts to solid-liquid extraction, which is due to the convenience of its implementation, a significant degree of efficiency, and a wide range of applications [13]. The methods of obtaining phenolic compounds from spent grain by solidliquid extraction include extraction in a hot water bath using different solvents: methanol, acetone, ethanol, hexane, ethyl acetate, and direct water separately, as well as a mixture of methanol, ethanol, and acetone with water. From all the experiments carried out, the highest yield of phenols was found in the extract obtained using 60% acetone as solvent. At the same time, all the extracts obtained irrespective of the solvents used showed antioxidant activity to different degrees, which can be considered as a positive aspect of the method. Another method of solid-liquid extraction of phenolic components, as well as other constituents of spent grain, is enzymatic hydrolysis. As mentioned above, this method can preserve the bioactive capacity of the extracted components and at the same time will be more environmentally friendly compared to chemically extracted substances [13,45].

The next extraction method can be identified as high-intensity ultrasound, which improves the release of complex chemicals found in plants, particularly phenols, through cavitation [46]. This method uses high sound power at low frequencies, which favors compression and decompression of the liquid medium, from which follows the destruction of the cell wall, which in turn increases mass transfer [47]. Also, microwave technology can contribute to the efficiency of extraction of polyphenols from spent grain by heating the internal water molecules. The superheating of the molecules leads to the destruction of the cell wall of the sample, which helps to facilitate the release of the associated compounds [48].

Other methods by which phenolic compounds can be extracted from spent grain include high-pressure methods. In combination with pressure, different solvents are used, which differ in their physicochemical characteristics such as viscosity, density and dielectric constant. Also, by modifying the pressure and temperature parameters, it is possible to achieve different polarity of the solvent and, consequently, influence its ability to solvate ions or molecules of the soluble components [49]. The ohmic heating technology can also be considered as a method for extraction of phenolic compounds from spent grain. The method is based on the supply of electricity through the extraction medium, which is dispersed as heat due to the electrical resistance of its components, which promotes electroporation and electrical destruction of cell structures. This in turn promotes better mass transfer between sample and solvent [47].

In this way, among the current technologies for extraction of phenolic components, ultrasound and high-pressure fluid extraction are popular methods. However, methods such as microwave radiation extraction, ohmic heating and pulsed electric field are less well studied, hence there is an additional need to analyze them for further insights and data to draw appropriate conclusions on their feasibility and effectiveness for extraction of phenolic compounds [47].

#### 6.4 Lipid extraction

As mentioned above, spent grain has various chemical compositions, which also include free fatty acids in sufficient quantities [15]. In addition, fatty acids can be essential, i.e. substances necessary for the human body, which it cannot synthesize on its own, but must be obtained from food. This fact is important for the food industry, as the extraction of free essential fatty acids inherent in spent grain can be a good solution for food fortification and additional utilization of by-products.

In its essence, the extraction of lipid compounds is a method aimed at breaking ester bonds with other material components, such as proteins or phenolic substances. Consequently, it is predominantly a method to be performed in an integrated manner, i.e. to affect not only lipid fractions but also related substances [42]. At the same time, despite the complexity of the spent grain fraction matrix, one work attempted to extract fats by simple extraction. For this purpose, an alcohol solution with a volume fraction of 20 vol.% was used and the extraction was carried out for 24 h at room temperature. As a result, the authors were able to extract 5 fatty acids, of which 2 were essential fatty acids. These fatty acids included: palmitic, stearic, oleic, linoleic, and linolenic acids [50].

Extraction in the presence of a mixture consisting of chloroform and methanol in a ratio 2:1 was also applied. The spent grain was previously ground into fine particles of small size, and after the addition of the mixture, extraction was carried out for 30 s under conditions of high-speed homogenizer. According to the results of the experiment, the composition of fatty acids that were able to be released into the extract was presented in the following proportions: polyunsaturated fatty acids made up the majority of the extract (50-55%), followed by saturated acids in the amount of 25-30% and a smaller proportion was occupied by monounsaturated fatty acids (15-20%). The qualitative composition was represented by linoleic (50%), palmitic (25%), oleic (15%), linolenic (5%) and stearic (<5%) acids and traces of docosahexaenoic, eicosapentaenoic and arachidonic acids ( $\omega$ -3 and  $\omega$ -6) were also found [42,51].

An enzymatic method of lipid extraction from spent grain was also tested, in which the spent grain sample was fermented with *Bacillus subtilis* culture for two days at 37 °C. The data collected were analyzed in comparison with the extract obtained from unfermented spent grain, after which the authors found that the amount of palmitic and linolenic acids extracted was independent of the treatment under the influence of biological catalysts (in this case, microorganisms), while oleic and stearic acids were dependent (the yield of oleic fatty acid increased after biocatalytic treatment, while that of stearic acid decreased) [42,52].

Referring to the above-mentioned, it can be concluded that spent grain is a good material for efficient extraction of its macronutrients as well as phenolic compounds, which forms an additional area of application of this by-product. Expanding the scope of use will reduce the amount of utilized waste and consequently reduce the negative impact on the environment.

#### 7. Disposal methods and other applications

Given the volumes of spent grain produced in the beer or ethanol production process, there is increasing talk about the need to dispose of them without landfilling in huge quantities or to recycle them to minimize environmental pollution [53]. As discussed above, one alternative would be to use the spent grain as a material for extracting useful substances, but either way, some of the waste is left behind and eventually disposed of in landfills. Thus, today the most frequently used methods of utilization of spent grain are: disposal in landfills, use in its initial state in animal husbandry, production of various feed mixtures with high protein content using starter, preservation for extending shelf life by silage, drying or mechanical dehydration, as well as the use of spent grain as organic fertilizer and soil ameliorant [53].

For example, raw brewer's spent grain without additional treatment has been used for feeding both domestic animals and poultry for quite a long time, but as a rule, their purpose is narrowed down to feeding ruminants. However, as a result of special treatment, brewer's spent grain can become an excellent feed additive for other mammals and birds. Today there is a demand for a feed additive called "Probiocel", which is used as an additional source of nutrients for fattening piglets, broilers, and laying hens. This additive is produced by homogenizing brewer's spent grain with bran, digesting the resulting mixture with especially isolated Bacillus subtilis bacteria, which in turn partially process dietary fiber into digestible carbohydrates. Selenium is then added and after fermentation, the resulting mixture is dried to extend the shelf life of this supplement. In this form, it can be stored for a year. It also has a positive effect on the animal, both on its health and physiological features [54]. Despite the positive aspects, in some countries the feeding of spent grain to animals is being minimized due to some restrictions, for example, a reduction in the number of farms and people involved in animal husbandry, and stricter legislation that requires spent grain producers to have a certificate of quality and appropriate documentation that allows them to distribute spent grain as an alternative livestock nutritional supplement [16].

Another method of spent grain utilization is disposal to landfills or special polygons (about 20% of the amount of spent grain produced). However, this method is one of the least preferable, as it is associated with numerous environmental problems: air and water pollution, unpleasant odors, and associated health hazards [11,16]. It has been noted that the disposal of one tonne of brewer's spent grain releases an equivalent of 513 kg of  $CO_2$  into the atmosphere [55]. Such indicators are disappointing and even dangerous for the ecology of the

world and, therefore, require measures to eliminate this problem, which is caused by the amount of waste produced, its increased moisture and reduced shelf life, and costly conservation. Thus, it is necessary to consider the concept of bioprocessing of brewing and distillery by-products (spent grain) to ensure environmental and economic prosperity [16].

As an example of bioprocessing of spent grain, its application as a solid biofuel for energy generation can be considered, but for this purpose, the spent grain needs to be dried beforehand [16]. Also, in one study, bio-oil, hydrogen, and ethane were produced by pyrolysis of spent grain [56]. Another study reported the use of spent grain to produce bio-oil and bio-coal by hydrothermal liquefaction [57]. In addition, the spent grain can be modified by biological processes (e.g., anaerobic digestion or fermentation) into biogas and bioethanol appropriately [58, 59].

Also due to its high content of polysaccharides, proteins, vitamins, and minerals, spent grain can be an optimal medium for the cultivation of various microorganisms such as fungi and bacteria [16]. Experiments have been repeatedly carried out to grow different microorganisms using spent grain, aiming to produce a range of enzymes: amylase, cellulase, hemicellulase, protease, and enzymes that promote lignin degradation [60]. In addition to the production of enzymes, during the cultivation of microorganisms, spent grain can be transformed into the following chemical components: lactic and succinic acid, itaconate, and xylitol [16].

It can be added that spent grain can be used to produce building materials for wooden buildings, such as bricks, or the production of pulp and paper, due to its rich chemical composition, namely fibers [61]. In many studies, brewer's spent grain has been able to produce paper towels, business cards, and cellulose pulp properly [62,63]. In addition, the spent grain has been tested as an adsorbent for the removal of pollutants from wastewater, for example, synthetic dyes used in the textile and pulp and paper industries have been removed [64].

Thus, the problem of utilization of spent grain, as well as other by-products of production, is at the peak of discussions and search for solutions, since the increase in discarded waste is directly related to the worsening environmental situation in the world. This is why it is necessary to expand the use or utilization of spent grain, which will reduce the carbon dioxide emissions generated during the decomposition of spent grain when they are buried.

#### 8. Application in the food industry

For many reasons, it was proposed to study the properties of spent grain concerning food products in different areas: bakery, pasta and confectionery products, milk and dairy products, meat products, etc. This is explained by the chemical composition of spent grain, which allows it to be classified as a biologically valuable product but is not sufficiently acceptable for human consumption as an independent product in its native form, in terms of its taste, which creates the need for its integration into other food products, as auxiliary raw materials. Given the limited shelf life and the need for large spaces for its storage, various methods for its processing are also being developed to obtain optimal moisture levels to extend shelf life and further use in the food industry, as already mentioned earlier. Also, population growth and the corresponding expansion of the food industry sector are becoming compelling reasons to search for alternative technologies, which will make it possible to obtain highly nutritious products at fairly low prices and without harm to human health and the environment [6].

In this case, the purpose of this chapter and the work as a whole is to consider the currently known ways of the use of spent grain in various products suitable for human nutrition, with its technological properties and behavior at the time of processing of products into which spent grain were introduced.

# 8.1. Production of bakery, pasta, and confectionery products

The production of bread and other bakery products is one of the main areas of the food industry, which ensures the daily satisfaction of the needs of the majority of the population [66]. The variety of these products is becoming wider every year, which is associated with the dietary characteristics of consumers or their taste needs (for example, gluten intolerance or lack of desire to eat products with chemically derived additives). It follows that the addition of spent grain in the production of baked items can make them competitive and bring innovative food products with increased nutritional value to the market, which is a desirable aspect in the development of the food sector and, in particular, the bakery industry [67].

In one of the studies, two types of bread were produced using spent grain. In the first case, the spent grain was added directly to the dough; in the second case, at the initial stage, a sourdough was produced from the spent grain, which was subsequently added to the dough. In both cases, the spent grain content was 15%, and the samples were characterized by a high fiber content: 11.9% in the spent grain flour added to the dough and 12.1% in the sourdough flour. In terms of mineral composition, sourdough bread made from spent grain turned out to be more enriched compared to bread with the addition of flour from the same spent grain. For example, the calcium content in sourdough bread was 107.9 mg/100 g, while in bread without sourdough this figure was 98.9 mg /100 g, magnesium values were 12.7 mg/100 g and 11.6 mg/100 g, respectively, and potassium - 100.4 mg/100 g and 98.9 md/100 g. Bread acidity was also higher in the sample using sourdough (pH 5.3 versus 5.8) [65]. It was also noted that with an increase in the percentage of spent grain from 0 to 20% with its further use in the production of bread, the content of some mineral compounds also increases: the amount of calcium increased from 76.44 mg/100g to 150.93 mg/100 g, magnesium - from 87.12 mg/100 g to 176.81 mg/100 g and potassium - from 116.04 mg/100 g to 225.49 mg/100 q [66].

Another study analyzed bread made from spent grain starter through fermentation for eight days. The content of spent grain in the sourdough was 25, 50, 75% and the sourdough was exclusively based on spent grain (100%). Based on the results of the experiment, it was determined that sourdough bread with low concentrations of spent grain (25 and 50%) was distinguished by relatively high porosity, acidity, and corresponding moisture content when compared with a sample made from 100% sourdough. In addition, the bacteriostatic properties of the product were revealed due to the addition of spent grain, which is due to the presence of early signs of spoilage in the sample without the addition of spent grain (control sample). And the introduction of spent grain into bread made it possible to increase its shelf life by 1-2 days, due to a decrease in the activity of microorganisms that cause spoilage of bread [67].

However, it was found that replacing wheat flour with spent grain flour causes some technological deterioration: a decrease in gluten yield with a worsening of its quality and, as a consequence, a reduction in the sedimentation ability and stability of the dough along with

an increase in its softening. These negative aspects were justified by the duration of kneading the dough (longer than kneading wheat dough), the increased content of protein and dietary fiber, as well as the increased force to stretch the dough, which was increased. Based on this, the authors identified the optimal content of spent grain in bread made from wheat flour, at which the bread will not differ significantly in appearance, crust condition, and crumb properties. This can be achieved by adding 10% spent grain, however, in any case, color changes in the product are observed, which is associated with the characteristics of the raw materials used (from the usual light cream color, the sample moved to a brown color, the intensity of which directly depended on the concentration of the added spent grain: darker with increasing and lighter when decreasing) [6,68].

The following studies demonstrated the relationship between the amount of spent grain used and the physical properties of the resulting bread. Thus, the water absorption capacity of the product increased with increasing concentration of the introduced by-product: at 0% this value was equal to 58.40 mL/100 g, and at 20% the content was already 66.67 mL/100 g. Experts argued this by the composition of the raw materials, that is, the increased content of protein substances and non-starch polysaccharides, which are capable of absorbing large amounts of moisture [68]. High concentrations of proteins and dietary fiber also affected the duration of the kneading, increasing it several times (from 3.43 min to 17.57 min) [69]. Changes in the weight and volume of the finished product were also demonstrated: with an increase in the added spent grain, the weight of the bread increased from 127.58 g to 148.85 g, while the volume decreased from 2.92 cm $^{3}$ /g to 2.46 cm $^{3}$ /g. This is again due to the spent grain's richness in protein and fiber, which absorb large volumes of water, giving the product a firmer and more durable structure. According to organoleptic characteristics, the same trend is observed, that is a lower acceptability of the product with growing concentration of spent grain in it. This is justified by the darker color, malt aroma and, as well as crumb texture [6,66,69].

As for pasta products, the addition of spent grain is a good option to increase the nutritional value with minimal changes in functional properties, despite the concentration of 25% [6]. One study analyzed pasta recipes with the addition of two types of spent grain: einkorn and tritordeum. The resulting pasta had a higher content of protein and dietary fiber, in particular  $\beta$ -glucans, and some changes in antioxidant activity were also detected, with its increase. From an organoleptic point of view, these products turned out to be very acceptable [70].

Just like for the production of bread, when developing fortified pasta, two different fractions of spent grain were used: one containing 10-20% dietary fiber and the second containing 5-10% protein. Their quality was assessed based on a number of characteristics: relative chemical composition, optimal cooking time, sensory indicators. Thus, the addition of the protein fraction contributed to the production of pasta with a protein component content of about 18%, and fiber values exceeding 8%. In both cases, the color of the paste was significantly darker than the control sample. The optimal cooking time until the products were ready varied from 11 min to 13.5 min. In terms of the degree of elasticity when bitten, these pasta products were rated adequately [71].

Another similar study, based on the development of pasta by adding the protein and fiber fraction of spent grain products has been created with stronger gluten structure and binding characteristics, high hardness and elasticity, as well as dough viscosity, which is characterized by tensile strength, while glycemic index indicators were reduced. In addition, pasta products into which spent grain fiber was integrated showed lower cooking loss values (3.47%), which was due to the amount of spent grain used in their production, according to the authors [72].

As a new direction in the pasta industry, we can highlight the work of Romanian researchers who tested the use of flour from spent grain and spelled flour (a type of dinkel wheat), which currently has low demand in the production of these flour-based products. The spent grain used in the study is a by-product of whiskey production, which was subsequently dried at 50 °C for 24 h, then crushed and sifted to a particle size of less than 200 µm. The goal of the work of Romanian experts was to increase the nutritional value of pasta through the introduction of two new types of flour: spent grain and hulled wheat. When mixing the dough, different amounts of spent grain were used to determine the optimal concentration based on sensory and physicochemical quality indicators. In conclusion, the authors finalized that from all analyses performed, the most acceptable results were achieved in the case of the sample that was produced by adding 10% spent grain [73].

Regarding the introduction of spent grain into the cookie dough composition, work was carried out aimed at determining the relationship between the size of the introduced particles and the quality of the finished product. As a result, medium- and large-sized particles demonstrated higher quality indicators compared to the fine fraction. For example, the distribution coefficient of raw materials in the mass of cookies was higher, as well as the organoleptic characteristics were more acceptable [74].

In one of the works, in the production of biscuits, replacing part of the wheat flour with spent grain in various ratios (0, 10, 20, and 30%). Samples containing a 20% concentration of spent grain tended to reduce hydrolysis and glycemic index, as well as a decrease in total starch when compared to the reference sample. In addition, changes in the percentage of dough composition, reducing the amount of wheat flour and replacing it with spent grain, contributed to an increase in the nutritional value of the finished products such as a significant growth in the content of proteins and bioactive compounds (phenolic acids) [75].

Among other things, the mechanism of the effect of thermally and mechanically untreated raw spent grain on the quality of biscuits was analyzed. Wheat flour in the dough was replaced by 15, 25, and 50% brewer's spent grain, the purpose of which was to evaluate the effect of concentrations on the content of nutrients (protein and dietary fiber), microbiological stability, and the sensory characteristics of the finished product. The authors recommend using spent grain to increase the nutritional value of cookies but in conditions of maintaining optimal replacement percentages, which should not exceed 25%. This will preserve acceptable organoleptic characteristics without compromising microbiological stability [76].

In the technology for the production of sugar cookies, wheat flour was replaced with spent grain obtained as a by-product during the production of ethyl alcohol in different ratios, which varied from 5 % to 12%. At the same time, the density of the dough changed from 1.32 g/cm<sup>3</sup> to 1.26 g/cm<sup>3</sup>, and the humidity increased from 16.6% to 19%, since raw spent grain with a high moisture content was used. However, the authors note improvements in the quality indicators of the finished product: the density of the cookies decreased by 17.4%, the swelling capacity increased by 18.3% with a dosage of spent grain from ethanol production equal to 10%. In terms of physiological value, experimental examples are superior to control samples due to the content of essential amino acids represented by threonine, valine, isoleucine, leucine, and lysine, mineral compounds, such as calcium and phosphorus, as well as an increase in the concentrations of protein, fat and dietary fiber. Raising the fat content is a

positive aspect of this technology since it will reduce the amount of additional fat, and this, in turn, contributes to the production of a more affordable product [10].

In other work, spent grain flour was also used to partially replace wheat flour and increase the nutritional value of the finished muffins. Flour was added when forming the emulsion in various quantities: 5, 10, 15, and 20% of the total raw material content. The quality indicators of the finished products are represented by physicochemical (alkalinity, moisture content, weight loss of the product during baking, and specific volume) and sensory characteristics. Researchers recommend that to preserve organoleptic and physicochemical parameters, as well as to increase the nutritional value of muffins, a 15% concentration of brewer's spent grain flour should be used as the maximum amount [77].

Also, an attempt was made to introduce raw crushed brewer's spent grain into the recipe for gingerbread products in order to enrich the product. The quantitative content of spent grain, which replaced 1st grade wheat flour, was 5, 10, 15, 20 and 25%. Based on the results of the analysis, it was found that an increase in the dosage of spent grain leads to a gradual improvement in the firmness properties of the dough due to protein substances, which enhance the strength of the flour and result in greater firmness of the dough. From the point of view of sensory characteristics, the resulting products with the additive differed from the control sample without the addition of spent grain: they acquired a darker shade, malt flavor, and aroma. In terms of nutritional value, the experimental samples are superior to the control sample, since the addition of spent grain contributed to an increase in the content of protein, dietary fiber, mineral compounds (iron, zinc, manganese), and vitamins (mainly group B and vitamin E). From a microbiological point of view, no violations of requirements and standards were identified. As a result of the experiments, researchers recommend replacing 1st-grade wheat flour with crushed raw brewer's spent grain in an amount equal to 20% when producing gingerbread [78].

#### 8.2. Dairy food production

In the production technology of dairy products, in comparison with bakery and other flour products, cereal crops are quite rare, which makes the use of spent grain more difficult [79]. However, despite this fact, many studies have been carried out aimed at using spent grain in the production of yogurt and cheese, to obtain a more enriched food item and expand the range of dairy products.

Thus, spent grain in various proportions was used as a substitute for fermentation in the production of yogurt. At the same time, a significant decrease in syneresis of the product was noted, a reduction in the duration of fermentation with an increase in the viscosity of yogurt. Products with the addition of 5% and 10% spent grain were characterized as maximum quality yoghurt, taking into account acidity and the development of lactic acid bacteria during storage, as well as rheological properties. However, higher byproduct concentrations (15-20%) resulted in a more stable dispersion system with reduced liquid separation and flowability of the finished product. In conclusion, the authors recommend the use of 10% spent grain in yogurt as the optimal amount, which will maintain fluidity and other physicochemical indicators at a good level, but with further study of its sensory characteristics, in particular the taste of yogurt [79].

Spent grain was used as an additive in the production of yoghurt in percentages of 2, 4, 6, 8, and 10%, and yoghurt without added brewer's spent grain was chosen as a reference sample. With an increase in the concentration of applied spent grain, significant changes were

noted in the chemical composition, microbiological component, as well as in sensory characteristics. The sample with 10% added brewer's spent grain showed the highest quantitative contents of protein, fat, fiber, ash, and pH, but at the same time had the lowest values of titratable acidity, the number of lactic acid bacteria, and organoleptic parameters. The product remained organoleptically acceptable when the spent grain concentration was increased to a maximum of 8% (at 8% and 10% there were reduced sensory scores). Thus, researchers recommend the use of spent grains to fortify yogurt [80].

In another study, spent grain was used to add processed cheese to Karish cheese. Replacement was made out in the following sizes: 10, 20, 30, 40 and 50%. The analysis was carried out to evaluate the physicochemical, microbiological, and organoleptic characteristics of processed cheeses with the addition of various amounts of brewer's spent grain. The indicators were determined both for a freshly prepared product and during storage for 3 months, monthly. As in the previous experiment, significant changes were observed in the listed indicators with an increase in the percentage of spent grain in the processed cheese. The sample consisting of 50% brewer's spent grain was determined to be the sample with the highest dry matter content, pH value, and the best rheological properties, but relative to other samples it demonstrated lower titratable acidity and oil separation. Moreover, according to sensory analyses, all samples were assessed as acceptable, based on which the authors concluded that the use of brewer's spent grain is a good way to obtain a functional food product with beneficial properties for consumer health [81].

#### 8.3. Manufacture of meat and fish products

Today, products of animal origin, such as meat of animals, poultry, and fish, are characterized by frequent price increases, which in developing countries becomes a problem and leads to a reduction in consumption of this type of food. This in turn contributes to the development of various deficiencies and subsequent diseases caused by a lack of macro- and microelements present in meat (for example, iron deficiency anemia). Based on this, manufacturers are looking for ways to reduce the cost of meat products by replacing the components of the recipe with cheaper raw materials, which provide the necessary technological characteristics of the products. However, replacing raw materials (meat) with components of plant or synthetic origin leads to a decrease in the nutritional value and quality of manufactured products. Thus, the use of spent grain can become an alternative to currently available meat substitutes and at the same time maintain or increase the biological value of the product and reduce cost.

For example, in one of the works, the recipe for minced meat semi-finished products was optimized by introducing dry and frozen brewer's spent grain. The dry brewer's spent grain was used without preliminary hydration and substituted the bread according to the recipe in various proportions. Kyiv cutlets were chosen as a reference sample. The frozen spent grain was not subjected to defrosting or hydration and was introduced at the last stage of adding raw materials, based on the recipe. Three types of meat products were produced: Kyiv cutlets, rump steaks, and beefsteaks, in the compositions of which a certain type of raw material was replaced with frozen brewer's spent grain in different quantities. As in the case of dry spent grain, frozen spent grain served as a substitute for wheat bread in Kyiv cutlets. In rump steaks spent grain replaced hydrated soy protein and in beefsteaks-parts of beef and lard were substituted [82].

Based on the data obtained on sensory quality indicators and amino acid composition, experts recommend introducing into production activities samples with improved taste and external characteristics, as well as a higher amino acid balance, which is represented by the following experimentally obtained samples:

- Kyiv cutlets with a quantitative content of dried brewer's spent grain of 4%.
- Kyiv cutlets with frozen brewer's spent grain in the amount of 4%.
- Beefsteaks using frozen brewer's spent grain in an amount equal to 6.9% [82].

Next, an analysis was carried out of the chemical composition, nutritional value, and shelf life of the above-recommended semi-finished meat products with the addition of brewer's spent grain. Based on the results of the entire research work, the authors have not changed their recommendations for the use of brewer's spent grain in dried and frozen form. The percentage of dry and frozen spent grain equal to 4% in Kyiv cutlets, as well as frozen spent grain of 6.9% in beefsteaks, will increase the biological value of finished meat products, improve their organoleptic characteristics, increase product yield by reducing losses during heat treatment, and also minimize costs the listed semi-finished products [82].

The goal of the next work was to develop a new recipe and corresponding technology for the production of a meat product from chilled chicken using spent grain. Based on this, in further development, the dried spent grain was ground using a laboratory mill, followed by sifting to obtain flour. In this investigation, the most acceptable concentration of brewer's spent grain flour in a chicken meat and lard product was determined and was 3%. A similar product was chosen as a comparison sample, in which wheat flour took the place of spent grain flour. According to the results, the control sample was inferior to the product with the addition of spent grain according to several criteria: paler color, less pleasant taste, and weakly perceptible aroma, as well as a small amount of carbohydrates and dietary fiber [83].

In the production of fish items, attempts have also been made to integrate spent grain into their composition. Thus, in one of the works, to improve the functional, technological, and sensory properties of minced fish, it was proposed to use dry brewer's spent grain in molded fish products, as a raw material that can enrich the products with protein, carbohydrates, in particular dietary fiber, some minerals and vitamins, as well as lipids. During the study, it was noted that an increase in the concentration of brewer's spent grain in the minced fish system leads to an improvement in its water-holding capacity. When analyzing losses during heat treatment of minced meat, there was a tendency to reduce them with a growth in the percentage of spent grain, which increases the yield of the finished product. In terms of sensory characteristics, the highest scores were received by samples containing 3 and 4% dry spent grain. Tasters noted the presence in these samples of a color similar to pork meat, the absence of a fishy smell, and the appearance of a bready aroma. As for the finished products made from this type of minced meat, a regular, round shape with an even contour was noted, but in products with the addition of 8 and 10% spent grain, small cracks and inclusions of brewer's spent grain were observed. The color of the products, similar to the previous indicators, changed as the dosage of spent grain increased: 1% - unattractive appearance due to the gray color on the surface, but when cut, white meat was observed, 3 and 4% - yellowbrown crust with light meat on the cut, 5 and 6% - gray color on the section. In conclusion, the researchers noticed that the introduction of dried and ground brewer's spent grain into fish products has a beneficial effect on their functional and technological properties. The values of 3 and 4% by weight of the mixture were taken as the optimal percentage of spent grain in minced fish [84].

The general idea of all conducted works is to increase the nutritional value of food items while simultaneously maintaining all quality indicators and reducing the price of finished products. The authors of the mentioned studies demonstrated that the use of brewer's spent grain in the technology of these products can serve as an integral part of the food industry. It should be noted that the successful implementation of this direction requires additional research and development aimed at optimizing technology and creating new formulations, and it is also important to ensure high-quality control of spent grain to avoid adverse consequences associated with product safety. Consequently, the use of spent grain, in particular brewer's spent grain, in the production technology of food items is a promising direction for increasing the functional value of products while simultaneously reducing cost and maintaining quality indicators. Successful implementation of this area can ensure the development of the food industry and sustainable public health by improving the quality of nutrition.

#### 9. Conclusions

Spent grain, a byproduct of beer and alcohol production is rich in nutrients but poses storage and disposal challenges due to its high moisture and protein content. These factors lead to rapid spoilage and environmental issues, such as CO<sub>2</sub> emissions and unpleasant odors during inadequate storage. Effective storage methods include refrigeration, chemical treatment, and dehydration, with drying emerging as the most promising approach to extend shelf life and preserve nutritional value.

The disposal of spent grain primarily impacts the agricultural sector, as it is commonly used in livestock feed. However, declining farm numbers and regulatory changes are complicating this use, often resulting in landfill disposal, which raises environmental concerns. Bioprocessing spent grain for biofuels and food products offers a sustainable alternative, expanding the potential for functional foods that improve public health.

Particularly promising is the use of spent grain in flour products like bread, pasta, and pastries, where it can replace part of traditional flour, enhancing product quality and nutritional content. Additionally, spent grain can be incorporated into meat and dairy, products, improving their nutritional profiles and reducing costs, especially in economically disadvantaged regions.

Overall, spent grain presents a multifaceted solution to environmental, economic, and nutritional challenges. Continued research, educational initiatives, and international collaboration will be crucial in maximizing its potential, promoting sustainable development, and enhancing public health.

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