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Pre-treatments used for the recovery of brewer's spent grain- a minireview

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Abstract

Lignocellulosic biomass, of which brewer's spent grain (BSG) is a part, has a varied composition and requires various pre-treatments to extract compounds with added value. Pre-treatments bring several advantages: they open the structure of the cell wall, decrease the particle size, reduce cellulose crystallinity, and improve digestibility. Solvent extraction methods, diluted acid methods, alkaline, supercritical carbon dioxide extraction, microwave-assisted extractions, or ultrasonic extractions have as their main objective the change of the crystallinity of lignocellulosic biomass and the detoxification of inhibitors to improve hydrolysis and general saccharification. Pre-treatments are essential strategies that disrupt the structure of the BSG and help remove lignin, thus facilitating the exposure of polysaccharides for optimal extraction efficiency. The complexity and heterogeneity of biomass decide the extraction method used. Efficient, economically feasible, simple extraction methods, from which no inhibitors and no corrosive materials are produced, are taking into account.

Keywords: brewer's spent grain, valorisation, extraction methods

1. Introduction

Beer is one of the most consumed beverages in the world, in 2018, 1.94 milliard hl of beer were produced globally [1]. Brewer's spent grain is the most abundant by-product of this industry, generating 20 kg per hectolitre of produced beer [2-4]. Globally, there have been 38.8 million tons of wet BSG, which is an environmental problem that needs to be solved. Brewer's spent grain is the solid residue left after filtration of the beer wort, being the insoluble part of the wort [2,5-7] and is composed mainly of polysaccharides, lignin, lipids, and a small fraction of proteins [8].

Until recently, food waste was of no interest, being used only for animal feed or composting, but current trends have come to the attention of researchers because BSG represents a cheap and valuable raw material. BSG consists of the layer of peel, pericarp, and seeds, with residual amounts of endosperm and aleurone. BSG has 80% moisture, sweet taste, malt smell, and can be considered lignocellulosic material [9-11] being characterized by large amounts of fibres (up to 70%), including cellulose, hemicelluloses, lignin, and protein content of 25-30% [12-14].

In traditional brewing, which employs a lauter tun, the BSG plays an important role as it forms the bed through which the mash is filtered to separate the wort [4] obtained after the saccharification of the malted cereal grains [15].

2. Chemical composition of BSG

BSG is a little-used by-product due to its high moisture content, which makes it difficult to transport and store and makes it an unstable product, conducive to microbial growth. [4,16]. The chemical composition of BSG varies depending on several factors: such as the cereal variety, time of harvesting, type of hops added, the malting and mashing regime, and whether adjuncts were employed during brewing; however, within-brewery sampling has shown BSG to be quite homogenous substance [4]. BSG is a lignocellulosic material, the major constituents of which are fibres (hemicelluloses and cellulose), protein, and lignin [10,17-19].

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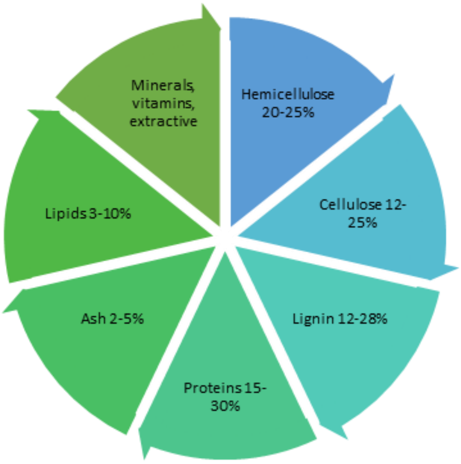

Figure 1. Chemical composition of BSG

Fibres are the components with the greatest interest in health intake (arabinoxylans and β-glucans), but also the phenolic components (hydroxycinnamic acid) [4]. Hemicelluloses, consisting mainly of arabinoxylan (AX) can be present in up to 40% of the dry weight of BSG (AX is the main non-cellulosic polysaccharide present in cereals and herbs). Another important polysaccharide in BSG is cellulose, and among monosaccharides are found xylose, glucose, arabinose, and traces of rhamnose and galactose [4].

Proteins are found around 20% of the dry mass, and the most abundant are hordein, glutelin, globulin, and albumin. Of the protein content, 30% are essential amino acids, the most present being lysine [4].

Lipids are found in a low percentage, of which triglycerides have a higher percentage (approximately 55%), followed by 30% fatty acids, according to Mussatto & Roberto [10].

BSG also contains phosphorus (6000 mg/kg), calcium (3600 mg/kg) and magnesium (1900 mg/kg) [4,9], iron, copper, potassium [13], and the vitamins present are biotin (0.1 ppm), niacin (44 ppm), choline (1800 ppm), folic acid (0.2 ppm), pantothenic acid (8.5 ppm), riboflavin (1.5 ppm), thiamine (0.7 ppm) and pyridoxine (0.7 ppm) [9,11,14]. In addition to all these components are extractives, made up of waxes, gums, resins, tannins, essential oils, and other cytoplasmic components [14].

3. Possible applications of BSG

Usually, brewer's spent grain is used for feeding animals, especially cattle, but due to its high fibres and protein content it is used to feed fish, pigs, or birds or to produce dog rewards [11,13,20]. Alternatives are being sought to obtain added-value products. Due to high moisture (approximately 80%), BSG is not suitable for long storage, the shelf life is 7-10 days.

Due to biologically active compounds with physiological roles in the body, foods fortified with BSG are considered functional foods, foods that offer health benefits, used as an adjunct in a balanced diet [11].

The bakery industry is the field where most studies on the incorporation of BSG [9, 21-23] have been carried out, but other studies have also been carried out for meat products. BSG can be added to frankfurters sausages to produce low-fat products [11]. A maximum of 15% BSG can be added to the frankfurters. The addition of BSG to fruit juices and smoothies has a beneficial effect on increasing phenolic content and antioxidant activity [4].

BSG was successfully used to obtain a fermented beverage rich in phenolic compounds. That beverage has a term of validity regarding the bioactive components for 15 days [24].

Another application of BSG is the production of polymers. The fact that BSG without pre-treatment resulted in a high amount of volatile fatty acids (91.3 mg chemical oxygen demand $CODl^{-1}h^{-1}$ volumetric productivity and a concentration of 24.9 gl^{-1} after 16 days HRT (hydraulic retention times) reduces the costs and complexity of the process and opens an opportunity in harnessing BSG in the production of polyhydroxyalkanoate (PHA) [25].

Xylan is the main component of hemicelluloses present in the walls of plant cells. Cellulose is composed of a chain of glucose molecules with hydrogen bonds between different layers of polysaccharides, giving a crystalline conformation.

Hemicelluloses, consisting mainly of xylan, are the target component for XOS production. Xilo-oligosaccharides (XOS) are mainly produced by hydrolysis of xylan and its prebiotic effects have been demonstrated. BSG contains hemicelluloses in a percentage of 16.5%, composed mainly of xylan (10.3%), arabinan (5.1%), and a small group of acetyl groups (1.1%) [26].

A novelty for the food industry is food containing symbiotics, both probiotic bifidobacteria and prebiotic oligosaccharides, enriched products both in terms of physicochemical properties and in terms of health benefits. Non-digestible oligosaccharides have potential as food ingredients, improving the quality of many foods in terms of flavour and physicochemical characteristics [27].

Sawdust is commonly used in the production of bricks and can be replaced with BSG due to the increase in the porosity of the bricks without changing their colour or quality [14]. The high amount of fibrous material combined with the reduced amount of ash makes BSG suitable for obtaining bricks [14, 28].

BSG has also been successfully used as an adsorbent of orange acid dye 7 in wastewater, with an adsorption capacity of 30.5 mg of orange acid 7 per gram BSG at 30°C [13] but also for adsorption of Congo red dye, used in the textile industry and stationery. As a very toxic dye to human health, methods of removal are sought as effectively as possible using BSG as an adsorbent material. For this purpose, pre-treatment with 0.1 mol/L sulphuric acid, 0.1 mol/L sodium hydroxide at 121°C was used for 20 minutes and white-rot fungus *Coriolus versicolor* at 28°C for 25 days, sodium hydroxide treatment was found to be the most effective [29].

BSG has a high carbon content and volatility, retains its physical properties when used as a bio-adsorbent at temperatures above 50°C, has roughness and cracks, has a low specific area of 6.79 m^2/g, an almost neutral loading surface, making it suitable for treating water containing dyes. The maximum amount of methylene blue was 284.75mg/g (optimal pH 11.00) and 26.18 mg/g for tartrazine yellow (optimal pH 2.00) when BSG was used as a bio-adsorbent [30].

BSG is a generous source of hydroxycinnamic acids (HCA) [9] which accumulate in cell walls, is considered the most important source of antioxidants in cereals, both in free form and is the bound form [28]. Phenolic compounds are considered natural antioxidants associated with some chronic diseases, such as cardiovascular disease, neurodegenerative diabetes, and cancer [9].

Natural cellulose-rich fibres, including BSG, can be used as an alternative to produce organic polyurethane, which has proven to be the most promising fillers. Different BSG polyurethane rations and ground tire rubber slots have been studied by Formela et al. with improvements in physicomechanical properties (apparent density increases by 37%, compression resistance by 50%) and thermal stability [31].

Protein hydrolysates in BSG have had good rheological results, protein changes by enzymatic or chemical means bringing improvements to certain functional properties (water/oil retention capacity, emulsion properties and foam expansion, turbidity) [32].

BSG can also be used for obtaining paper-serves-cards, producing coal-inferior in terms of combustion properties, production of resins, an antifoaming agent in beer [13,14], production of bacteriocin using *Lactococcus lactis* and *Enterococcus mundtii*, antimicrobial activity against *Lysteria monocytogenes* as a bioindicator. Lignocellulosic material is subjected to treatment, usually hydrolysis, to solubilize in the constituent monomers. The sugars formed are subject to microbial fermentation as precursors of added-value compounds (e.g. bacteriocins) or enzymes [33].

Ndayishimiye et al. conducted a study on the encapsulation of oils recovered from BSG by an eco-friendly technique of saturated gas solutions (CO_2 supercritical), aiming to obtain a product with improved physical properties and with oxidative stability [34].

Pyrolyized BSG is used to obtain biochar, which is a porous material with stable physical and chemical properties and a high content of C. It is used to increase the amount of C in the soil, to reduce nutrient leakage, for soil amending, as a nutrient supplier for plant growth, and for improving soil characteristics. Biochar has a large amount of ash, stable aromatic C structures, low bulk density, and moderate cation exchange capacity, which makes it suitable for this use [35].

The submerged fermentation of BSG using *Aspergillus niger* and *Saccharomyces cerevisiae* can produce citric acid in amounts of 0.512% and 0.312% respectively [36-38].

4. Extraction methods of added value compounds from BSG

It is necessary to develop existing extraction techniques and to find new techniques and possible applications for BSG to use this by-product as a value-added resource, given the number of bioactive compounds available with a wider range of applications [2]. Extraction methods are always developed and optimized for achieving higher extraction yields of bioactive compounds [2]. Lignocellulosic materials are opposed to hydrolysis, which is why lignocellulosic biomass undergoes pre-treatments that make it accessible [39], and then cellulose and hemicelluloses are subjected to hydrolysis with their transformation into monomers (sugars) that can be used by microorganisms [40]. Pre-treatments are expensive and have a significant impact on the environment, and so far no pre-treatment with conversion factor 100 has been found, the most used being with acidic solutions (cheap and effective) using low concentrations of acids (<5%), but high temperatures (120-210°C) and pressure < 10 atm, or concentrated acids (<30%) and temperatures <100°C and atmospheric pressure [41]. The selection of the pre-treatment method to use will depend on type of compound to extract.

Autohydrolysis is a pre-treatment used in the extraction of hemicelluloses without the use of chemicals, an interesting alternative from this point of view, being an environmentally friendly method [2,42]. Hydrothermal treatment, also called autohydrolysis, uses high-temperature water (over 200°C), but no chemical solvents are used, which makes it an eco-friendly method, and the resulting products can be used in the food industry [2,43]. Other advantages are that it does not raise problems of corrosiveness of equipment, it is a simple and economical operation [43] and there is a 90% conversion of cellulose into glucose [44]. The lignocellulosic material is immersed in water at high temperature and pressure, penetrating the cell structure, cellulose hydrates, depolymerizes hemicelluloses in mono- and oligomers [43]. Although in principle this pre-treatment was used to obtain bioethanol, it is currently also used for converting some agri-food by-products into food ingredients.

The operation principle of autohydrolysis assumes that lignocellulosic matter is subjected to liquid water at high pressure and temperature so that the structure of cells can be penetrated, cellulose is hydrated, and cellulose depolymerized in monomers and oligomers. A percentage of 40-60% of lignocellulosic biomass is dissolved as a result of this process. Hemicelluloses are depolymerized into soluble products, while solids are enriched in cellulose and lignin [43].

Pre-treatments with diluted acids help deconstruct the cell wall of plants. Although it is a low-cost method, it can lead to the production of acetic acid and formic acid, as well as other inhibitors enzymatic fermentation and fermentation such as 5-hydroxymethylfurfural [2]. Pre-treatments with diluted acids can be carried out in two situations: at a temperature above 180°C for a short period or at lower temperatures (120°C) for a retention period of 30-90 minutes [44]. The heterocyclic ether bonds between sugar monomers in the polymer chains formed by hemicelluloses and cellulose are broken by the protons released by the acid. When the above bonds break, some compounds are released (xylose, arabinose, and glucose mainly) [2]. The greatest degradation power of hemicelluloses is sulphuric acid among the other inorganic acids used: hydrochloric, nitric, and phosphoric acid. Sulphuric acid pretreats also have several drawbacks: it is expensive, requires treatment at high temperatures, it is very corrosive and dangerous, large amounts of inhibitors are formed that can subsequently affect fermentation [44]. Pre-treatments with diluted acids are used to obtain bioethanol from agricultural waste [2,45,46].

On the other hand, alkaline pre-treatments have a low cost and result in celluloses-rich biomass, improving the biodegradability of raw material by removing lignin and increasing porosity. Alkaline reagents disrupt the cell wall by dissolving lignin and hemicelluloses, thus improving cellulosic enzymes access to cellulose and decreasing the likelihood of non-productive enzymes to bind to lignin [2]. Alkaline pre-treatment can be performed at room temperature and in times that can vary from a few seconds to days, being more effective on agricultural residues than on wood materials [47]. The suitable chemicals for this method are natrium hydroxide, potassium hydroxide, calcium hydroxide, and ammonium hydroxide. $Ca(OH)_2$ helps to remove amorphous substances, such as lignin, which increases the crystalline index, it helps to increase the internal surface of cellulose, decreases the degree of polymerization of lignin and is low-cost [47]. Mussatto et al. used alkaline pre-treatment to obtain ferulic and p-coumaric acid from brewer's spent grain [48]. Active mechanisms during alkaline pre-treatment involve the saponification of the ester bonds linking lignin hemicelluloses [49]. Among the advantages of this method is noted the decrease of the crystalline index, resulting in a more glucose yield, does not require temperatures, the method is carried out at room temperature, causes swelling and rapid dissolution of lignin [44]. Wilkinson et al. performed an alkaline treatment on BSG, as follows: BSG and alkaline reagent were mixed to obtain the necessary solid load and brought to pH 11.5 using NaOH 40% (weight/volume), then the samples were placed on the water bath (50-100°C) for a time between 1-12 hours [50]. After pre-treatment the samples were brought to pH 7.0 with glacial acetic acid and centrifuged at 5000 rpm for 10 min, drying the solid residue overnight at 60°C. Xiao et al. approached alkaline pre-treatment differently: lignocellulosic matter is soaked in an aqueous solution of NaOH 1 M in ratio 1:100 g/mL, and then shaken slightly for 18 hours at 30°C. The residue is washed and filtered with 95% water and ethanol until the filtrate is neutral, then dried in an oven at 60°C for 16 hours. Solubilized hemicelluloses have been isolated by precipitating filtrates and drying them in the air [51].

Another method that does not require expensive equipment is enzymatic pre-treatment, but solutions are being sought to improve the costs of the enzymes used, which impedes to the use of this method. Oxidative and hydrolytic enzymes that are not inhibited by the final products are used [52]. Enzymatic hydrolysis results in fermentable sugars from cellulosic materials [2].

An alternative and ecological method to conventional extractions with organic solvents is the method of supercritical CO_2 extract (SCE-CO_2), which uses CO_2 in the supercritical condition when both temperature and pressure are equal to or exceed the critical point of 31°C and 73 atm [28] providing the ideal conditions for extracting compounds with a high degree of recovery in a short time [53]. The supercritical fluid has characteristics of both gases and liquids [28]. This method involves loading the material into an extraction vessel (attached with temperature controllers and pressure valves both at the induction and exit), the existence of a pump that provides the desired pressure. Dissolved compounds and fluid are directed to separators, where the fluid's solvation power can be adjusted by lowering the pressure or increasing the temperature. In the lower part of the separators, there is a valve where the product is collected and the fluid is regenerated and recycled further [28,34]. SFE (supercritical fluid extraction) helps to obtain clean extracts, free from the certain gradation of labile or easily oxidized compounds that can form due to exposure to high temperatures or oxygen, as emerging from traditional extraction methods [54].

Supercritical carbon dioxide extraction is a fast, selective method, which does not result in residual solvents, where carbon dioxide is used as a solvent for extraction. As advantages, carbon dioxide is cheap, has good solvation power when in supercritical condition, and is considered by the FDA (Food and Drug Administration) as a safe product. The main disadvantage of this method is the lower polarity of carbon dioxide, which can be solved with the help of co-solvents (e.g. ethanol) [55], but also the high costs involved [54]. In their study Spinelli et al. optimized the method of extracting bioactive compounds from BSG, the best extraction conditions being 35 MPa for pressure, 40°C for temperature, 240 min for time, and 60% ethanol v/v as co-solvent for obtaining 0.35±0.01 mg GAE/g BSG phenolic compounds and 0.22±0.01 mg QE/g BSG flavonoids [55]. Ferrentino et al. compared the BSG oil extraction method using the supercritical fluid method with the Soxhlet method using hexane as a solvent [56]. Higher content of phenolic compounds resulted from hexane extraction, as well as antioxidant activity, as well as a higher stability of oils. The disadvantage of the hexane method is that traces are possible to remain in the final product, which is not desirable because it seriously affects human health. On the other hand, carbon dioxide extraction offers the advantage of lower extraction time, of 1 hour compared to 6 hours in the Soxhlet method also of lower temperatures.

The same carbon dioxide extraction technique is used by Ndayishimiye et al. later encapsulating oils recovered from BSG for use in the food, cosmetics, or pharmaceutical industries [34].

Solvent extraction is a common method of extraction, and the extract obtained can be used in the food industry, is an alternative to synthetic antioxidants [2]. BSG contains a significant number of phenolic compounds that can be extracted by solid-liquid extractive methods, where the solvent is a very important factor for extraction yield. When the extract is used in foods, the solvents used are very important, regulated from the perspective of food safety (according to good manufacturing practices is the mixture acetone: water or ethanol: water). Methanol is a very effective extraction solvent, but because of its toxic nature, it limits the use of the extract in the food industry or the pharmaceutical industry [57].

Microwave-assisted extraction uses microwave energy to heat the solvent in contact with the sample in a shorter time than other extraction methods, making it faster and consuming less energy, without resulting in residual substances as a result of this process. Microwave-assisted extraction (MAE) is an efficient, promising, and very fast technique because the movement of polar particles generates rapid heating [58]. Coelho et al. performed the extraction of arabinoxylans (AX) and arabinoxylo-oligosaccharides (AXOS) by microwave-assisted extraction at 210°C for 2 minutes obtaining 43% of AX and AXOS from BSG [59]. The solvent used in microwave-assisted extraction is an important factor in the extraction yield, as well as the amount of water present in the solvent. Microwave-assed gravitational hydro diffusion (MHG) is a new technique for extracting bioactive compounds from plants, combining microwave heating with gravity, with the whole process taking place at atmospheric pressure, without the involvement of water or other solvents [**Error! Reference source not found.**]. Small volumes of solvents are used, the mixture is heated in a short time (15-30 min), being an advantageous method compared to traditional methods [60].

Ultrasonic extractions require less time, using less solvent and lower extraction temperatures compared to alternative extraction methods, as small dimensions of the matter analyzed are produced, thus enriching hydrolysis [44]. All the advantages mentioned result in higher extraction yields and higher purity polysaccharides [2]. Direct sonication with horn-type devices or indirect sonication in a cleaning type device can be used [44]. High-intensity ultrasounds cause pressure variations that form microscopic bubbles that collapse immediately after their formation, generating a shear force (shear forces) that results in temperature rise. Ultrasound waves and the resulting cavitations break the cell walls, and their contents will be released into the extraction medium. It generates a mechanical effect similar to a milling process, which is due to micro jets and micro streaming [44].

Reis et al. conducted a study in which they used the ultrasonic extraction method compared to the alkaline extraction method, in which the extraction time was reduced from 7 hours to 25 minutes, during which 60% of arabinoxylans from BSG were recovered [62]. Ultrafiltration is a method of separation of compounds that depends on several factors: properties of the feed, molecular weight membranes cut off (MWCO), transmembrane pressure, and cross-flow rate [2]. Two membranes of MWCO were used (5 and 30 kDa) and the protein content was 20.09 ± 1.40% for the 5 kDa membrane and 15.98 ± 0.58% for 30 kDa.

Electric-field based technologies may be differentiated according to several factors: electrical flow, impulse application, electric field power, or heat extension, with the advantage of reusing by-products [2].

Another method of pre-treatment is ammonia expansion fibres (AFEX), where cellulosic biomass is treated at temperatures between 70-200°C and pressures between 0.7-27 MPa in the presence of ammonia as a solvent. The advantages of this method is the low presence of inhibitors and the growth of the accessible surface by delignification simultaneously with the solubilization of a part of hemicelluloses, but among the disadvantages are the high cost of a large amount of ammonia and inefficiency in high lignin content materials [44].

The steam explosion together with acid or alkaline solvents at high pressures (0.69-4.83 MPa), temperatures between 160-260°C for 20-30 minutes facilitates hydrolysis of cellulose. The disadvantages of these methods of pre-treatment are the generation of toxic compounds and partial degradation of hemicelluloses [44]. Ionic liquid extracts are considered organic and can be easily used [44]. Outeiriño et al. used a cholinium-based ionic liquid that contains glycine as anion to delignify BSG biomass [15].

Several pre-treatments can be used to obtain different brewer's spent grain compounds, e.g. Yu et al. use an ultrasonic-assisted ecological enzyme process in their.

5. Conclusions

Due to the increasing demands in fields such as food production, cosmetics, and chemical, capitalization and use of BSG is an interesting research topic. BSG is a valuable resource for industrial operation due to its low cost, year-round availability and high volume. The chosen extraction methods are imposed according to the value-added compounds targeted, the impact they have on the environment, and last, but not least, the costs. To reach zero waste and an integrated circular economy it is necessary to develop and optimize these methods to obtain high extraction yields, as well as valuable compounds with low costs.

Compliance with Ethics Requirements. Authors declare that they respect the journal's ethics requirements. Authors declare that they have no conflict of interest and all procedures involving human or animal subjects (if exist) respect the specific regulation and standards.

References

1. https://www.statista.com/statistics/270275/worldwide-beer-production/ (accesed on 15 september 2020)
2. Bonifácio-Lopes, T., Teixeira, J. A.; Pintado, M., Current extraction techniques towards bioactive compounds from brewer's spent grain–A review, *Critical Reviews in Food Science and Nutrition* **2019**, 1-12, https://doi.org/10.1080/10408398.2019.1655632
3. Fărcaş, A.C.; Socaci, S.A.; Dulf, F.V.; Tofană, M.; Mudura, E.; Diaconeasa, Z., Volatile profile, fatty acids composition and total phenolics content of brewers' spent grain by-product with potential use in the development of new functional foods, *J Cereal Sci*. **2015,** *64*, 34-42, http://dx.doi.org/10.1016/j.jcs.2015.04.003
4. Lynch, K.M.; Steffen, E.J.; Arendt, E.K., Brewers' spent grain: a review with an emphasis on food and health, *J Inst Brew*. **2016**, *122*, 553-568, DOI 10.1002/jib.363
5. del Rio, J. C.; Prinsen P.; Gutierrez A., Chemical composition of lipids in brewer's spent grain: A promising source of valuable phytochemicals, *Journal of Cereal Science* **2013**, 58(2),248–54, doi:10.1016/j.jcs.2013.07.001
6. Pires, E. J.; Ruiz, H. A.; Teixeira, J. A.; Vicente, A. A., A new approach on brewer's spent grains treatment and potential use as lignocellulosic yeast cells carriers, *Journal of Agricultural and Food Chemistry* **2012** *60*(23), 5994–9. doi: 10.1021/jf300299m
7. Vieira, E.; Rocha, M. A. M.; Coelho, E.; Pinho, O.; Saraiva, J. A.; Ferreira, I. M. P.L. V. O.; Coimbra, M. A., Valuation of brewer's spent grain using a fully recyclable integrated process for extraction of proteins and arabinoxylans, *Industrial Crops and Products* **2014,** *52*, 136–143, doi: 10.1016/j.indcrop.2013.10.012
8. Ravindran, R.; Jaiswal, S.; Abu-Ghannam, N., Jaiswal, AK., A comparative analysis of pretreatment strategies on the properties and hydrolysis of brewers' spent grain, *Bioresour Technol*. **2018**, *248*, 272-279, doi:10.1016/j.biortech.2017.06.039
9. Ikram, S.; Huang, L.Y.; Zhang, H.; Wang, J.; Yin, M., Composition and Nutrient Value Proposition of Brewers Spent Grain, *J. Food Sci.* **2017**, *82*, 2232–2242, doi: 10.1111/1750-3841.13794
10. Mussatto, S.I.; Roberto, I.C., Acid hydrolysis and fermentation of brewer's spent grain to produce xylitol, *J. Sci. Food Agric.* **2005**, *85*, 2453–2460, DOI: 10.1002/jsfa.2276
11. McCarthy, A.L.; O'Callaghan, Y.C.; Piggott, C.O.; FitzGerald, R.J.; O'Brien, N.M., Brewers' spent grain; Bioactivity of phenolic component, its role in animal nutrition and potential for incorporation in functional foods: A review, *Proc. Nutr. Soc.* **2013**, *72*, 117–125, doi:10.1017/S0029665112002820
12. Roth, M.; Jekle, M.; Becker, T., Opportunities for upcycling cereal byproducts with special focus on Distiller's grains, *Trends Food Sci. Technol.* **2019**, *91*, 282–293, https://doi.org/10.1016/j.tifs.2019.07.041
13. Mussatto, S.I., Brewer's spent grain: A valuable feedstock for industrial applications, *J. Sci. Food Agric.* **2014**, 94, 1264–1275, DOI 10.1002/jsfa.6486
14. Mussatto, S.I.; Dragone, G.; Roberto, I.C., Brewers' spent grain: Generation, characteristics and potential applications, *J. Cereal Sci.* **2006**, *43*, 1–14, doi:10.1016/j.jcs.2005.06.001
15. Outeiriño, D.; Costa-Trigo, I.; Paz, A.; Deive, F.J.; Rodríguez, A.; Domínguez, J.M., Biorefining brewery spent grain polysaccharides through biotuning of ionic liquids, *Carbohydr. Polym*. **2019**, *203*, 265–274, https://doi.org/10.1016/j.carbpol.2018.09
16. Chetrariu, A.; Dabija, A., Brewer's Spent Grains: Possibilities of Valorization, a Review, *Applied Sciences* **2020,** *10*(16), 5619, doi:10.3390/app10165619
17. Plaza, P.E.; Gallego-Morales, L.J.; Peñuela-Vásquez, M.; Lucas, S.; García-Cubero M.T.; Coca, M., Biobutanol production from brewer's spent grain hydrolysates by Clostridium beijerinckii, *Bioresour Technol.* **2017**, *244*, 166-174, doi:10.1016/j.biortech.2017.07.139
18. López-Linares, J.C.; García-Cubero, M.T., Lucas, S.; González-Benito, G.; Coca, M., Microwave assisted hydrothermal as greener pretreatment of brewer's spent grains for biobutanol production, *Chem Eng J.* **2019**, *368*, 1045-1055, doi:10.1016/j.cej.2019.03.032
19. da Silva, E.G.; Borges, A.S.; Maione, N.R.; Castiglioni, G.L.; Suarez, C.A.G.; Montano, I.D.C., Fermentation of hemicellulose liquor from Brewer's spent grain using *Scheffersomyces stipitis* and *Pachysolen tannophilus* for production of 2G ethanol and xylitol, *Biofuels, Bioprod. Bioref.* **2019**, 1-11, doi:10.1002/bbb.2072
20. Jayant, M.; Hassan, M.A.; Srivastava, P.P.; Meena, D.K.; Kumar, P.; Kumar, A.; Wagde, M.S., Brewer's spent grains (BSGs) as feedstuff for striped catfish, *Pangasianodon hypophthalmus* fingerlings: An approach to transform waste into wealth, *J. Clean. Prod.* **2018**, *199*, 716–722, https://doi.org/10.1016/j.jclepro.2018.07.213

21. Ktenioudaki, A.; Alvarez-Jubete, L.; Smyth, T.J.; Kilcawley, K.; Rai, D.K.; Gallagher, E., Application of bioprocessing techniques (sourdough fermentation and technological aids) for brewer's spent grain breads. *Food Res Int*. **2015**, *73*, 107-116, doi:10.1016/j.foodres.2015.03.008
22. Stojceska, V.; Ainsworth, P., The effect of different enzymes on the quality of high-fibre enriched brewer's spent grain breads, *Food Chem*. **2008**, *110*(4), 865-872, doi:10.1016/j.foodchem.2008.02.074
23. Martins, Z.E.; Pinho, O.; Ferreira, I.M.P.L.V.O., Food industry by-products used as functional ingredients of bakery products, *Trends Food Sci Technol*. **2017**, *67*, 106-128, doi:10.1016/j.tifs.2017.07.003
24. Gupta, S.; Jaiswal, A.K.; Abu-Ghannam, N., Optimization of fermentation conditions for the utilization of brewing waste to develop a nutraceutical rich liquid product, *Ind. Crops Prod*. **2013**, *44*, 272–282, http://dx.doi.org/10.1016/j.indcrop.2012.11.015
25. Ribau, M.; Guarda, E.C.; Freitas, E.B.; Galinha, C.F.; Duque, A.F.; Reis, M.A.M., Valorization of raw brewers 's spent grain through the production of volatile fatty acids, *N Biotechnol*. **2020**, *57*, 4-10, doi:10.1016/j.nbt.2020.01.007
26. Amorim, C.; Silvério, S.C.; Rodrigues, L.R., One-step process for producing prebiotic arabino-xylooligosaccharides from brewer's spent grain employing Trichoderma species. *Food Chem*. **2019**, *270*, 86-94, doi:10.1016/j.foodchem.2018.07.080
27. Mussatto, S.I.; Mancilha, I.M., Non-digestible oligosaccharides: A review, *Carbohydr Polym*. **2007**, 68(3), 587-597, doi:10.1016/j.carbpol.2006.12.011
28. Guido, L.F.; Moreira, M.M., Techniques for Extraction of Brewer's Spent Grain Polyphenols: a Review, *Food Bioprocess Technol*. **2017**, *10*(7), 1192-1209, doi:10.1007/s11947-017-1913-4
29. Wu, J.; Zhang, Z.; Xu, J.; Lu, X.; Wang, C.; Xu, H.; Yuan, H.; Zhang, J., Brewer's Grains with Different Pretreatments Used as Bio-Adsorbents for the Removal of Congo Red Dye from Aqueous Solution, *BioResources* **2020**, *15*(3), 6928-6940
30. Araújo, T.P.; Tavares, F.D.O.; Tait, D.; Barros, M.A.S.D., Biosorption mechanisms of cationic and anionic dyes in a low-cost residue from brewer's spent grain, *Environ Technol*. **2020**, 1-32, doi:10.1080/09593330.2020.1718217
31. Formela, K.; Zedler, Ł.; Hejnaa, A.; Zedlera, L.; Przybysza, M.; Rylb, J.; Reza Saebc, M., Structural, thermal and physico-mechanical properties of polyurethane/brewers' spent grain composite foams modified with ground tire rubber, *Ind Crops Prod*. **2017**, *108*, 844-852, doi:10.1016/j.indcrop.2017.07.047
32. Kotlar, C.E.; Ponce, A.G.; Roura, S.I., Improvement of functional and antimicrobial properties of brewery byproduct hydrolysed enzymatically, *LWT-Food Sci Technol*. **2013**, *50*(2), 378-385, doi:10.1016/j.lwt.2012.09.005
33. Paz, A.; da Silva Sabo, S.; Vallejo, M.; Marguet, E.; Pinheiro de Souza Oliveira, R.; Domínguez, J.M., Using brewer's spent grain to formulate culture media for the production of bacteriocins using *Patagonian strains*, *Lwt*. **2018**, *96*, 166-174, doi:10.1016/j.lwt.2018.05.027
34. Ndayishimiye, J.; Ferrentino, G.; Nabil, H.; Scampicchio, M., Encapsulation of Oils Recovered from brewer's Spent Grain by Particles from Gas Saturated Solutions Technique, *Food Bioprocess Technol*. **2020**, *13*(2), 256-264, doi:10.1007/s11947-019-02392-x
35. Amoriello, T.; Fiorentino, S.; Vecchiarelli, V.; Pagano, M., Evaluation of Spent Grain Biochar Impact on Hop (*Humulus lupulus L.*) Growth by Multivariate Image Analysis, *Applied Scinces* **2020**, *10*, 533; doi:10.3390/app10020533
36. Rachwał, K.; Waśko, A.; Gustaw, K.; Polak-Berecka, M., Utilization of brewery wastes in food industry, *PeerJ*. **2020**, 1-28, doi:10.7717/peerj.9427
37. Atere, V.A., Citric acid production from brewers spent grain by *Aspergillus niger* and *Saccharomyces cerevisiae, International Journal of Research in BioSciences* **2013**, *2*(1), 30-36
38. Dabija, A.; Tulbure, M., *Capitalization of by-products in the beer industry*, PIM Publishing House, Iasi. **2010**
39. Yamakawa, C.K.; Qin, F.; Mussatto, S.I., Advances and opportunities in biomass conversion technologies and biorefineries for the development of a bio-based economy, *Biomass and Bioenergy* **2018**, *119*, 54-60, doi:10.1016/j.biombioe.2018.09.007
40. Mussatto, S.I.; Roberto, I.C., Kinetic behavior of *Candida guilliermondii* yeast during xylitol production from highly concentrated hydrolysate, *Process Biochem*. **2004,** *39*(11), 1433-1439, doi:10.1016/S0032-9592(03)00261-9
41. Mussatto, S.I., *Biomass Pretreatment With Acids.* In: *Biomass Fractionation Technologies for a Lignocellulosic Feedstock Based Biorefinery*, Elsevier Inc., Amsterdam, The Netherlands, **2016**, pp. 169-185, doi:10.1016/B978-0-12-802323-5.00008-6
42. Carvalheiro, F.; Garrote, G.; Parajo, J.C.; Pereira, H.; Girio, F.M., Kinetic Modeling of Brewery 's Spent Grain Autohydrolysis, *Biotechnol. Prog.* **2005**, *21*, 233−243, doi: 10.1021/bp049764z

43. Ruiz, H.A.; Rodrıguez-Jasso, R.M.; Fernandes, B.D.; Vicente, A.A.; Teixeira, A.J., Hydrothermal processing, as an alternative for upgrading agriculture residues and marine biomass according to the biorefinery concept : A review, *Renewable and Sustainable Energy Reviews* **2013**, *21*, 35-51, doi:10.1016/j.rser.2012.11.069
44. Bhatt, S.M.; Shilpa, Lignocellulosic feedstock conversion, inhibitor detoxification and cellulosic hydrolysis – a review, *Biofuels* **2015**, DOI: 10.1080/17597269.2014.1003702
45. Rajan, K.; Carrier, D.J., Effect of dilute acid pretreatment conditions and washing on the production of inhibitors and on recovery of sugars during wheat straw enzymatic hydrolysis, *Biomass and Bioenergy* **2014**, *62*, 222–7, doi: 10.1016/j.biombioe.2014.01.013.
46. Mikulski, D.; Klosowski, G., Efficiency of dilute sulfuric acid pretreatment of distillery stillage in the production of cellulosic ethanol, *Bioresource Technology* **2018**, *268*, 424–33, doi: 10.1016/j.biortech.2018.08.005.
47. Alvira, P.; Tomás-Pejó, E.; Ballesteros, M.; Negro, M.J., Pretreatment technologies for an efficient bioethanol production process based on enzymatic hydrolysis: A review, *Bioresource Technology* **2010**, *101*, 4851–4861, doi:10.1016/j.biortech.2009.11.093
48. Mussatto, S.I.; Dragone, G.; Roberto, I.C., Ferulic and p-coumaric acids extraction by alkaline hydrolysis of brewer’s spent grain. *Ind Crops Prod*. **2007**, *25*(2), 231-237, doi:10.1016/j.indcrop.2006.11.001
49. Carvalheiro, F.; Duarte, L.C.; Gírio, F.M., Hemicellulose biorefineries: a review on biomass pretreatments. *J. Sci. Ind. Res*. **2008**, *67*, 849–864
50. Wilkinson, S.; Smart, K.A.; Cook, D.J., Optimisation of alkaline reagent based chemical pre-treatment of Brewers spent grains for bioethanol production, *Ind Crops Prod*. **2014**, *62*, 219-227, doi:10.1016/j.indcrop.2014.08.036
51. Xiao, B.; Sun, X.F.; Sun, R., Chemical, structural and thermal characterizations of alkali-soluble lignins and hemicelluloses, and cellulose from maize stems, rye straw, and rice straw, *Polymer Degradation and Stability* **2001**, *74*, 307-319
52. Zhang, Y.; Kang, X.; Wang, Z.; Kong, X.; Li, L.; Sun, Y.; Zhu, S.; Feng, S.; Luo, X.; Lv. P., Enhancement of the energy yield from microalgae via enzymatic pretreatment and anaerobic co-digestion, *Energy* **2018,** *164*, 400–7, doi: 10.1016/j.energy.2018.08.124.
53. Kitryte, V.; Šaduikis, A.; Venskutonis, P.R., Assessment of antioxidant capacity of brewer’s spent grain and its supercritical carbon dioxide extract as sources of valuable dietary ingredients, *J Food Eng*. **2015**, *167*, 18-24, doi:10.1016/j.jfoodeng.2014.12.005
54. Maróstica Jr, M.R.; Leite, A.V.; Romanelli, Dragano, N.R.V., Supercritical Fluid Extraction and Stabilization of Phenolic Compounds From Natural Sources – Review (Supercritical Extraction and Stabilization of Phenolic Compounds), *The Open Chemical Engineering Journal*, **2010**, *4*, 51-60
55. Spinelli, S.; Conte, A.; Lecce, L.; Padalino, L.; Del Nobile, M.A., Supercritical carbon dioxide extraction of brewer’s spent grain. *J Supercrit Fluids*. **2016**, *107*, 69-74, doi:10.1016/j.supflu.2015.08.017
56. Ferrentino, G.; Ndayishimiye, J.; Haman, N.; Scampicchio, M., Functional Activity of Oils from Brewer’s Spent Grain Extracted by Supercritical Carbon Dioxide, *Food Bioprocess Technol*. **2019**, *12*(5), 789-798, doi:10.1007/s11947-019-02249-3
57. Meneses, N.G.T.; Martins, S.; Teixeira, J.A.; Mussatto, S.I., Influence of extraction solvents on the recovery of antioxidant phenolic compounds from brewer’s spent grains, *Sep Purif Technol*. **2013**, *108*, 152-158, doi:10.1016/j.seppur.2013.02.015
58. Aguilar-reynosa, A.; Romaní, A.; Rodríguez-jasso, R.M.; Aguilar, C.N.; Garrote, G.; Ruiz, H.A., Microwave heating processing as alternative of pretreatment in second- generation biorefinery : An overview, *Energy Convers Manag*. **2017**, *136*, 50-65, doi:10.1016/j.enconman.2017.01.004
59. Coelho, E.; Rocha, M.A.M.; Saraiva, J.A.; Coimbra, M.A., Microwave superheated water and dilute alkali extraction of brewer’s spent grain arabinoxylans and arabinoxylo-oligosaccharides, *Carbohydr Polym*. **2014**, *99*, 415-422, doi:10.1016/j.carbpol.2013.09.003
60. Asofiei, I. Nonconventional Techniques for the Separation of Valuable Compounds from Plants. Ph.D. Thesis,Politehnica University, Bucharest, Romania, 2017
61. Steiner, J.; Procopio, S.; Becker, T., Brewer’s spent grain: Source of value-added polysaccharides for the food industry in reference to the health claims, *European Food Research and Technology* **2015**, *241*(3), 303–15, doi: 10.1007/s00217-015-2461-7
62. Reis, S.F.; Coelho, E.; Coimbra, M.A.; Abu-Ghannam, N., Improved efficiency of brewer’s spent grain arabinoxylans by ultrasound-assisted extraction. *Ultrasonics Sonochemistry* **2015,** *24*, 155–64, doi:10.1016/j.ultsonch.2014.10.010.
63. Yu, D.; Sun, Y.; Wang, W.; O”Keefe, S.F.; Neilson, A.P.; Feng, H.; Wang, Z.; Huang, H., Recovery of protein hydrolysates from brewer ’ s spent grain using enzyme and ultrasonication, *International Journal of Food Science and Technology* **2020**, *55*, 357–368, doi:10.1111/ijfs.14314