Uniwersytet Przyrodniczy we Wrocławiu
Wydział Biotechnologii i Nauk o Żywności
Dyscyplina naukowa: Technologia żywności i żywienia

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Modyfikacja składu chemicznego młóta browarnianego w celu poprawy aktywności antyoksydacyjnej oraz właściwości techno-funkcjonalnych

Modification of brewers' spent grain chemical composition to improve their techno-functionality and antioxidant capabilities

Rozprawa doktorska

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Wrocław 2023

Acknowledgments

I would like to express my gratitude to my supervisors
Assoc. Prof. Malgorzata Korzeniowska, DSc. and Prof. Baoru Yang, PhD
for the opportunities and challenges for scientific development.

I would like to thank
my colleagues, PhD students, for their motivation
and encouragement during the study.

I would like to thank
my beloved wife for the support in the last year of my study.

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Abstract

Brewers' spent grain (BSG) has been continuously evaluated as a food and nutraceutical ingredient for its health benefits. BSG contains dietary fiber, phenolic compounds, protein, fatty acids and minerals thus enhancing the bioactivity of BSG-added food products and its nutritional value. To mention a few, BSG possesses antimicrobial activities, DNA-protection activity, anti-mutagenic, inhibit of lipid peroxidation as well as maintaining colon health and gut microbiota. BSG is a complex matrix which is mainly dominated by insoluble dietary fiber (IDF). In general, polysaccharides is the main body of BSG in which several important compounds are attached such as phenolic compounds and fatty acids; meanwhile protein is mainly entrapped in the cell wall of BSG which is covered by polysaccharides. The complexity of BSG inhibits the bioavailability of such important compounds. From the perspective of BSG as a food ingredient, BSG improved the nutritional value of BSG-added food products. However, it diminished the techno-processing properties such as textural formation and expansion index thus decreasing the food production efficiency. This phenomenon occurred due to the high amount of IDF which is capable of binding high amounts of water thus disrupting the textural formation of food products. Aiming to improve the quality and quantity of important compounds from BSG, several techniques have been developed. BSG is a complex material, and it consists of dietary fiber as a main compound to which several important compounds are attached such as phenolic compounds, fatty acids, amino acids, and minerals. Among those reported methods, thermal treatment is one of the highly used in improving the phenolic compounds and dietary fiber extraction. Furthermore, protein extraction is in second place as the most studied compound from BSG after phenolic extracts.

The study aimed to evaluate the impact of thermal treatment and enzymatic protein extraction on the chemical composition, functionality, and antioxidant properties of BSG. The study was conducted in 2 different stages including thermal exposure and enzymatic protein extraction on chemical compounds, antioxidant activity and techno-functionality of BSG. Thermal

treatment was carried out by treating the BSG using autoclave and water bath at different temperature and time exposure. Protein extraction was done on 3 different enzyme incubations and the impact on polyphenolic and antioxidant activity of extracted protein and BSG residue was studied.

Preliminary study revealed that the improvement in polyphenolic compounds and antioxidant activity of autoclaved BSG is related to the degradation of dietary fiber composition of BSG. Autoclave and water bath heating treatments decreased the amount of saturated fatty acids and increased the amount of polyunsaturated fatty acids. Moreover, thermal treatment fluctuated the antioxidant activities and volatile compounds of BSG, in addition to the alteration in techno-functional properties of BSG. Most of the autoclave treatment increased polyphenolic compounds and antioxidant activity of treated BSG while water-bath treatment lowered the antioxidant properties (FRAP and ABTS) and total amount of phenolic acids of treated BSG. The study showed that after separation of protein fraction, BSG residues had a lower total polyphenolic content, ABTS and FRAP value compared to that in BSG protein. However, BSG residues and protein possessed the same level of ORAC value.

To conclude, different treatments of BSG fluctuated its chemical composition, thus its biological properties as well as techno-functional properties. For instance, the level of ABTS and FRAP was discovered to be the lowest at water-bath treatment while the highest was observed in multi-dried autoclaved BSG. The highest flavan-3-ols was identified at autoclaved fresh BSG while the lowest was obtained at multi-dried autoclaved BSG. Finding the most efficient treatment would be depending on the specific target. In case of specific bioactive compounds extraction such as phenolic, fatty acids, dietary fiber and/or others optimization of techniques can be expected. From the perspective of techno-processing in food development, modification of functionality such as rehydration properties and oil holding capacity as well as modification in odor perception are approachable to increase the processing efficiency and final products acceptability.

Streszczenie

Młóto browarniane (BSG) jest ciągle uważane za składnik żywności i nutraceutyk z uwagi na swoje korzyści zdrowotne. BSG zawiera błonnik pokarmowy, związki fenolowe, białko, kwasy tłuszczowe i związki mineralne, dzięki czemu zwiększa bioaktywność i wartość odżywczą produktów spożywczych uzyskiwanych z ich dodatkiem. BSG ma działanie przeciwdrobnoustrojowe, osłaniające DNA oraz działa antymutagennie, hamuje peroksydację lipidów, a także utrzymuje w dobrym stanie zdrowotnym okrężnicę i pobudza mikroflorę jelitową. BSG to złożona matryca, w której dominuje głównie nierozpuszczalny błonnik pokarmowy (IDF). Polisacharydy z przyłączonymi do ich cząsteczek m.in. związkami fenolowymi lub kwasami tłuszczowymi są podstawowymi składnikami BSG; tymczasem białko jest głównie uwięzione w ścianie komórkowej BSG, która jest pokryta polisacharydami. Złożoność BSG ogranicza biodostępność tych ważnych składników pokarmowych. BSG dodane do żywności poprawia ich wartość odżywczą, jednakże zmniejsza właściwości technologiczne i przetwórcze, takie jak tworzenie tekstury i wskaźnik ekspansji, zmniejszając w ten sposób wydajność produktów. Jest to związane z obecnością dużej ilości IDF, który jest zdolny do wiązania dużych ilości wody, zakłócając w ten sposób tworzenie tekstury produktów spożywczych. W celu poprawy jakości i zwiększenia dostępności ważnych związków chemicznych zawartych w BSG, opracowano kilka technologii. Jedną z najczęściej stosowanych metod modyfikacji BSG celem uwolnienia większej ilości związków fenolowych i ekstrakcji błonnika pokarmowego jest obróbka termiczna. Drugą najczęściej stosowaną metodą jest ekstrakcja białek z BSG.

Celem niniejszej pracy była ocena wpływu obróbki termicznej i enzymatycznej ekstrakcji białek na skład chemiczny, funkcjonalność i właściwości przeciwutleniające BSG. Eksperyment przeprowadzono w 2 odrębnych etapach, tj. obróbkę termiczną oraz enzymatyczną ekstrakcję białek, celem określenia ich wpływu na skład chemiczny, aktywność przeciwutleniającą i właściwości techno-funkcjonalne BSG. Obróbkę termiczną przeprowadzono przez traktowanie BSG w autoklawie oraz w łaźni wodnej w różnych temperaturach i czasie ekspozycji. Ekstrakcję

białka przeprowadzono w 3 różnych inkubacjach enzymatycznych i zbadano jej wpływ na aktywność polifenolową i przeciwutleniającą wyekstrahowanego białka oraz pozostałości BSG.

Wstępne badania wykazały, że zwiększenie ilości związków polifenolowych i aktywności przeciwutleniającej autoklawowanego BSG jest związana z degradacją błonnika pokarmowego BSG. Obróbka termiczna w autoklawie i łaźni wodnej zmniejszyła ilość nasyconych kwasów tłuszczowych, a zwiększyła ilość wielonienasyconych kwasów tłuszczowych. Ponadto, obróbka termiczna wpłynęła na aktywność przeciwutleniającą i zawartość lotnych związków w BSG, nie zmieniając istotnie właściwości techno-funkcjonalnych BSG. Większość wariantów obróbki w autoklawie zwiększała zawartość związków polifenolowych i aktywność przeciwutleniającą traktowanego BSG, podczas gdy obróbka w kąpieli wodnej obniżała właściwości przeciwutleniające (FRAP i ABTS) i całkowitą ilość kwasów fenolowych w BSG. Badania wykazały, że po oddzieleniu frakcji białkowej reszty BSG charakteryzowały się niższą zawartością polifenoli ogółem, aktywnością do zmiatania rodnika ABTS oraz redukcji żelaza FRAP w porównaniu z białkiem natywnym BSG. Jednakże, pozostałość po ekstrakcyjna BSG i pozyskane białko miały te same wartości ORAC.

Podsumowując, zastosowane w badaniach różne techniki modyfikacji BSG wpływały na jego skład chemiczny, a tym samym właściwości biologiczne, jak również właściwości techno-funkcjonalne. Wykazano, że zdolność do zmiatania rodnika ABTS oraz redukcji żelaza FRAP była najniższa podczas obróbki w kąpieli wodnej, podczas gdy najwyższe wartości obu wyróżników analizowano w BSG poddanym wielokrotnie autoklawowaniu. Najwyższą zawartość flawan-3-oli stwierdzono w autoklawowanym świeżym BSG, a najniższą w wielokrotnie autoklawowanym BSG. Opracowanie najskuteczniejszej metody obróbki zależy od wyznaczonego konkretnego celu. W przypadku ekstrakcji określonych związków bioaktywnych, takich jak kwasy fenolowe, kwasy tłuszczowe, błonnik pokarmowy i/lub inne, można dokonać optymalizacji zastosowanych technik. W procesie tworzenia nowych produktów żywnościowych modyfikacja cech funkcjonalnych BSG, takich jak zdolność do wchłaniania i utrzymywania wody czy oleju, a także

wpływ na profil związków zapachowych są możliwe w celu zwiększenia wydajności produkcji, jak również akceptowalności produktów końcowych.

1. Introduction

Brewers' spent grain (BSG) is a main byproduct of the brewery industry. BSG stands for about 40% of beer production waste. Every 1 L of beer production generates approximately 0.2 kg of BSG (Mussatto et al., 2006). In 2021, European countries produced an estimated 359 million hectoliters of beer, which consequently generated almost 7.2 million tons of BSG. Poland is in the third place for the highest beer production in Europe after Germany and the United Kingdom. By this, Poland contributes to the high amount of BSG (Conway, 2022).

BSG contains several bioactive compounds including phenolic compounds, fatty acids, amino acids, and other minor compounds. Those substances are responsible for several health benefits. To mention a few, phenolic compounds, protein and protein isolates, fatty acids, and dietary fiber from BSG are responsible for several antioxidant properties, including enzymatic and non-enzymatic antioxidants (Barbosa-Pereira et al., 2014; Crowley et al., 2017; Kumari et al., 2019; McCarthy et al., 2014; Socaci et al., 2018; Verni et al., 2020). The abundance of nutritional value of BSG has been considered for its high potential as a food ingredient on several food products such as bread, cookies, and pasta (Cappa & Alamprese, 2017; Fărcaş et al., 2015; Heredia-Sandoval et al., 2020; Nocente et al., 2019).

As mentioned previously, the incorporation of BSG in food processing has been extensively evaluated. Moreover, nutritional-related extraction process, health benefits and potential application of BSG has also been reported in excellent literature studies (Bonifácio-Lopes et al., 2020; Lynch et al., 2016; Mussatto et al., 2006; Wen et al., 2019; Xiros & Christakopoulos, 2012). However, the negative issues associated with food processing and final products acceptability of BSG-added food has never been reported. The important mechanism of nutritional-related compounds released from BSG matrix due to the treatments and extraction process has not also been issued. Therefore, the literature review on underlining the major issue caused by the addition of BSG in food processing was seemingly important in relation to the

possibility of improving the functionality and nutritional value of BSG as a food ingredient. The result of the literature review is presented in publication 1:

Naibaho J. & Korzeniowska M. (2021). **Brewers' spent grain in food systems: Processing and final products quality as a function of fiber modification treatment**. *Journal of Food Science* (2021), *86*(5), 1532–1551. https://doi.org/10.1111/1750-3841.15714. (Publication 1).

According to the literature review, there are two main current trends in terms of valorizing BSG including the evaluation of BSG as a food ingredient and evaluation of several techniques in improving the extraction efficiency of important compounds from BSG.

Besides the fact that BSG enhances the nutritional value of food products (dough and bread processing, extrusion process, baked snacks, pasta, and yogurt), the addition of BSG affected the whole process from mixing to the final food products, as well as, its acceptability. Due to the high water binding capability of BSG, it disrupted the network formation of food products by regulating the rheological behavior of the mixtures during the mixing process as well as disrupting the gluten network, protein stability and starch gelatinisation (Heredia-Sandoval et al., 2020; Nascimento et al., 2017; Nocente et al., 2019; Steinmacher et al., 2012; Torbica et al., 2019). At a certain level, BSG also reduced the sensory acceptability due to the harder texture and color modification of food products. This phenomenon is mainly due to the high amount of insoluble dietary fiber, particularly arabinoxylan, which is capable of adsorbing water up to 10 times of its amount (Steiner et al., 2015). By this, the modification of dietary fiber composition on BSG could be expected to improve its appeal, not only from nutritional value, but also in food techno-processing and food acceptability. Aiming to improve the food acceptability of BSG-added foods, fermentation was able to increase the solubility of cell walls thus enhancing the quality of sourdough (Aprodu et al., 2017; Ktenioudaki et al., 2015; Magabane, 2017; Torbica et al., 2019). Enzymatic treatment improved the solubility of arabinoxylans and consequently enhanced the specific volume of BSG-added bread (Ktenioudaki et al., 2015; Steinmacher et al., 2012). In addition to that, the modification of screw speed and temperature on extrusion of BSG has been reported (Steinmacher et al., 2012; Torbica et al., 2019).

Several treatments, which had been done on BSG, were summarized in publication 1 (Table 5). In general, physical methods are the most common techniques used in BSG treatments followed by enzymatic treatments and combinations of physical and enzymatic methods. Only a small amount of study used alkaline and acids in treating BSG. This phenomenon might relate to the sustainable goal developments, which are promoted by the United Nations. Currently, green processing seems to be a future global target for its benefits to the environment. Physical treatment is one of the most common and most developed methods considered as green processing including thermal exposures, pulsed electric field, ultrasound, and particle degradation. Steam explosion and microwave superheated are two of thermal exposures, which have improved the solubility of arabinoxylans from BSG (Coelho et al., 2014; Kemppainen et al., 2016). Ultrasound has been utilized to enhance protein and arabinoxylans recovery (Reis et al., 2015; Yu et al., 2020).

Dietary fiber plays an important role in BSG. Brewery spent grains are dominated by insoluble dietary fiber (IDF), which is a main component of polysaccharides in the cell wall of BSG. The majority of important nutrition-related compounds, including phenolic compounds and fatty acids are attached to its moieties; protein and amino acids are mainly covered in the vacuole cell wall of BSG. Therefore, improvement of yield extraction from BSG, which had been obtained by several techniques might be a consequence of dietary fiber degradation. Therefore, the applied methods in enhancing the extract recovery from BSG is seemingly important, not only for extraction efficiency, but also for improving the performance of BSG in food processing. Therefore, further research was carried out in investigating the impact of certain treatments in chemical compositions, techno-functionality, and antioxidant activities of BSG.

Among those reported methods in publication 1, thermal treatment is one of the highly used in improving the phenolic compounds and dietary fiber extraction. After polyphenolic compounds, protein is the second most studied compound from BSG. Therefore, the study was designed to evaluate the impact of thermal treatment and protein extraction on chemical composition, functionality, and antioxidant activities of BSG.

The race is to find low cost, practically simple, nutritionally benefits and efficient methods. Autoclave and water-bath instruments are commonly found and used, and they are easy to operate. In addition to that, autoclave and water-bath are economically low-cost methods. Being considered as a simple technique, autoclave treatment has been reported for its ability to modify the dietary fiber composition of soybean curd residues, increasing the resistant starch in rice grains, and increasing the solubility-related properties and stability of the colloidal suspension (S. Li et al., 2019; Nawaz et al., 2020; Zheng et al., 2020). Water-bath alters the fresh flavor and bitter taste of fish, improves the microwave-ultrasound capability in increasing the strength and water-holding capacity of gels, as well as, the capability of ultrasound-assisted treatments to facilitate the Maillard reaction (Jung et al., 2020; Ye et al., 2022; H. Zhang et al., 2015). Considering their benefits they seemed to be promising in improving the BSG properties.

Therefore, the first stage of the study was conducted to evaluate the influence of autoclave treatment and water-bath heating on chemical composition, techno-functional properties, and biological properties of BSG. Preliminary study was carried out to confirm the hypothesis emphasized in publication 1, applied treatments modify the dietary fiber composition and consequently increase the nutritional-related compounds.

The second stage of the study aimed to evaluate the impact of different enzymatic protein extraction on polyphenolic compounds and antioxidant properties of BSG compared to obtained protein. As mentioned earlier, protein is covered in the vacuole cell wall of BSG. Degradation of polysaccharides, as a main body of chemical structure of BSG obviously will also affect the protein release thus consequently the release of polyphenolic compounds from polysaccharides moieties.

2. Purpose and scope of the research

The study aimed to modify the chemical composition, techno-functionality, and antioxidant activities of BSG by several techniques including physical treatments, such as autoclave heating and water-bath heating, as well as, enzymatic treatments. The main hypothesis was that different applied methods could alter the chemical compositions and biological properties differently due to the different stability of specific compounds to certain exposure conditions.

The research was designed in several phases with different research hypotheses according to their purposes.

- Preliminary study aimed to confirm that the improvement of phenolic compounds and antioxidant properties is linked to the dietary fiber composition of treated BSG.
- Thermal treatment, such as autoclave and water-bath heating, were designed to evaluate the impact of thermal level, water ratio, and time exposures on physico-chemical and biological properties of BSG. It was hypothesized that different levels of temperature, water ratio and time exposures significantly alter chemical composition of BSG including volatile composition, fatty acid profile, and polyphenolic compounds, as well as, techno-functionality, such as water holding and oil holding capacity of BSG. Moreover, the amount of flavan-3-ols, phenolic acids and antioxidant capabilities (FRAP and ABTS) is highly correlated to the temperature and time exposures during the thermal treatment.

Final study was conducted in the evaluation of the protein extraction process with and without enzymes that could alter the phenolic compounds and antioxidant capabilities of BSG residues after protein separation. As protein is entrapped in the vacuole cell of BSG, the incubation process disrupted the vacuole cell of BSG differently depending on the presence of enzymes. It was hypothesized that incorporation of enzymes improved the release of phenolic compounds from polysaccharides matrices as a result of enzymes attacking the polysaccharide to release the protein.

3. Materials and Methods

3.1 Materials

BSG was collected from a local brewery, beer-light producer, in Wroclaw Poland and stored at freeze temperature for the study. All chemicals used were analytical grade.

3.2 Research design

In general, the study was designed at several stages including literature review and further investigation related to the trend from literature review. Research scheme is depicted in Figure 1 with 6 manuscripts included in the thesis listed in Table 1.

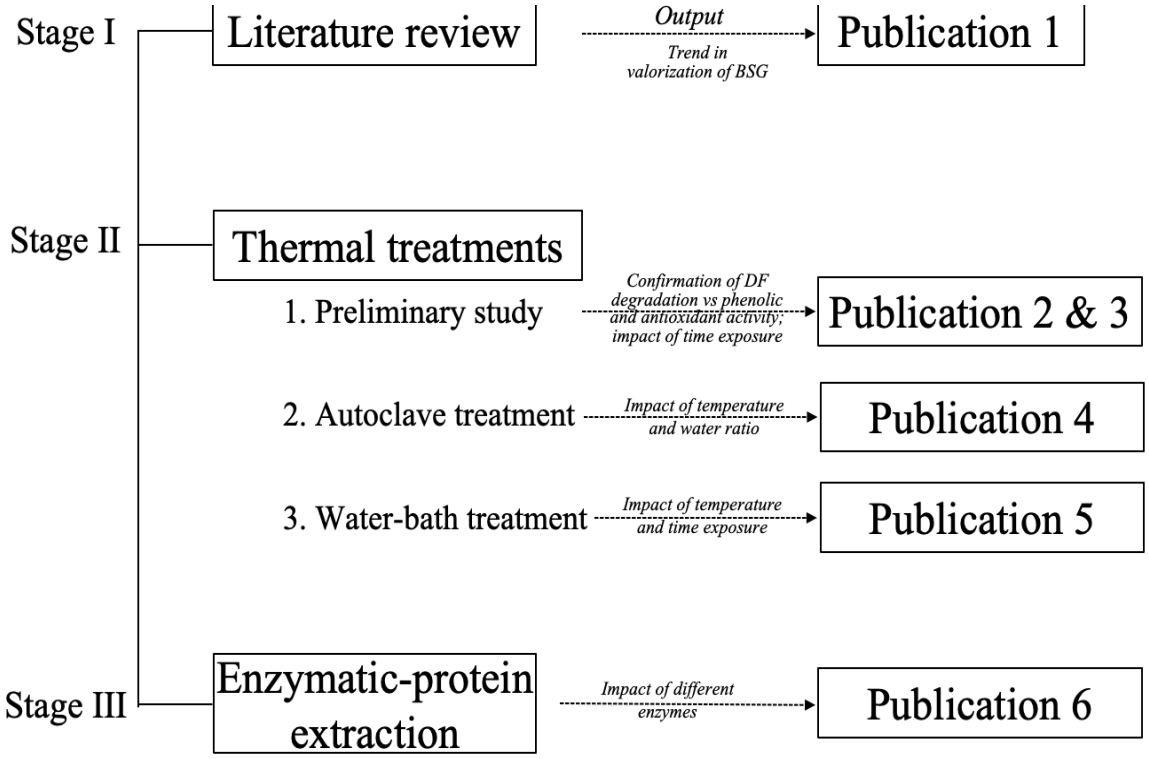

Figure 1. Research scheme

3.3 Methodology

According to the literature review, several treatments have been applied to improve the yield extracts from BSG. There are several target compounds from BSG including polyphenolic compounds, fatty acids and proteins, in addition to dietary fiber. Therefore, the impact of thermal exposure on chemical compounds of BSG such as polyphenolic compounds, fatty acids, and volatile compounds were evaluated. Furthermore, antioxidant activity as well as techno-functionality such as water holding and oil holding capacity were also investigated. It was

highlighted that the increase in yield extraction of targeted compounds might have been linked to the dietary fiber degradation. Therefore, a preliminary study was carried out to confirm the relation of dietary fiber degradation and polyphenolic compounds, antioxidant activity and techno-functional properties of BSG as an impact of autoclave treatment.

Preliminary study was conducted on autoclave treatment investigating different levels of time exposures (9, 12, and 15 min) and thermal elevation (90, 100, 110, and 130 C) using dried BSG. Considering the treatment efficiency, further investigation was carried out using fresh BSG to avoid multi-drying and multi-milling steps of BSG during the sample preparation. After the preliminary study, the impact of autoclave treatment at different water ratios (1:1 and 1:2) and temperature (90, 110, and 130 C) was conducted for 12 min on fresh milled BSG. The study was then compared to water-bath heating treatment at lower temperature levels (80, 90, and 100 C) and longer time exposure (15, 30, and 60 min). Finally, the last stage of the study was to evaluate the impact of protein extraction using different enzymes on polyphenolic compounds and antioxidant properties of methanolic extract from extracted proteins and BSG residues.

Table 1. List of publications included in the thesis

No	Authors	Title	Source	Impact factor	MES Point
Literature review					
1	Naibaho J., & Korzeniowska M.	Brewers' spent grain in food systems: Processing and final products quality as a function of fiber modification treatment	*Journal of Food Science* (2021), *86*(5), 1532–1551. https://doi.org/10.1111/1750-3841.15714	3.16	70
Preliminary study					
2	Naibaho J., Korzeniowska M., Wojdyło A., Figiel A., Yang B., Laaksonen O., Foste M., Vilu R., & Viiard E.	Fiber modification of brewers' spent grain by autoclave treatment to improve its properties as a functional food ingredient	*LWT – Food Science and Technology (2021)*, *149*, 111877. https://doi.org/10.1016/j.lwt.2021.111877	4.95	100
3	Naibaho J., Wojdyło A., Korzeniowska M., Laaksonen O., Föste M., Kütt M.-L., & Yang B.	Antioxidant activities and polyphenolic identification by UPLC-MS/MS of autoclaved brewers' spent grain	*LWT – Food Science and Technology (2022)*, *163*, 113612. https://doi.org/10.1016/j.lwt.2022.113612	6.05	100
Main study					
4	Naibaho J., Bobak L., Pudlo A., Wojdylo A., Andayani S. N., Pangestika L. M. W., Korzeniowska M., & Yang B.	Chemical compositions, antioxidant activities and techno-functionality of spent grain treated by autoclave treatment: evaluation of water and temperature levels	*International Journal of Food Science and Technology (2022),* 16042. https://doi.org/10.1111/ijfs.16042	3.61	70
5	Naibaho J., Pudlo A., Bobak L., Wojdylo A., Lopez A. A., Pangestika L. M. W., Andayani S. N., Korzeniowska M., & Yang B.	Conventional water bath heating on undried brewers' spent grain: techno-functionality, fatty acids, volatiles, polyphenolic profile and antioxidant properties	*Food Bioscience (2023),* 102523. https://doi.org/10.1016/j.fbio.2023.102523	5.32	70
6	Naibaho, J., Korzeniowska, M., Wojdyło, A., Ayunda, HM., Foste, M., & Yang, B.	Techno-functional properties of protein from protease-treated brewers' spent grain (BSG) and investigation of antioxidant activity of extracted proteins and BSG residues	*Journal of Cereal Science (2022), 107, 103524.* https://doi.org/10.1016/j.jcs.2022.103524	4.07	140
			TOTAL	**27.16**	**550**

4. Results and discussion

4.1 The impact of thermal treatments on BSG

4.1.1 Preliminary study: fiber modification by autoclave heating treatment and its impact on polyphenolic compounds and antioxidant activities of BSG

Autoclave treatment has been applied to disrupt hemicellulose and cellulose as well as reducing the molecular weight which consequently increased the amount of soluble dietary fiber on soybean curd residue (S. Li et al., 2019). It also broke down the peptide-fiber bonds thus intensifying the solubility-related properties and stability of colloidal suspension (Nawaz et al., 2020). Considering the benefits of autoclave in the degradation of dietary fiber bonds, autoclave heating was carried out to confirm the ability of autoclave to modify dietary fiber of BSG. The study was done by implementing various temperatures from 90 ℃ to 130 ℃ at different time exposures 9, 12 and 15 min on dewatered dried BSG with a ratio 1:3 (BSG:water). The investigation was done on dietary fiber composition (IDF and SDF) in addition to the chemical surface properties and proximate composition. Moreover, physical properties of BSG were investigated including water activity, swelling capacity, water holding capacity, oil holding capacity and color properties. The dietary fiber modification on BSG was expected to affect the amount of polyphenolic compounds and its antioxidant capacity. The results of this preliminary study are presented in publication 2 and 3:

Naibaho, J., Korzeniowska, M., Wojdyło, A., Figiel, A., Yang, B., Laaksonen, O., Foste, M., Vilu, R., & Viiard, E. (2021). Fiber modification of brewers' spent grain by autoclave treatment to improve its properties as a functional food ingredient. *LWT – Food Science and Technology, (149) 111877*. https://doi.org/10.1016/j.lwt.2021.111877. (Publication 2).

Naibaho, J., Wojdyło, A., Korzeniowska, M., Laaksonen, O., Föste, M., Kütt, M.-L., & Yang, B. (2022). Antioxidant activities and polyphenolic identification by UPLC-MS/MS of autoclaved brewers' spent grain. *LWT – Food Science and Technology, 163*, *113612*. https://doi.org/10.1016/j.lwt.2022.113612. (Publication 3).

The majority of the treatments increased the amount of SDF and decreased the IDF. Autoclaved BSG contained SDF at a range of 5% – 11% which increased from a range of 3.9% – 8.2%; while IDF declined from a range of 44% – 45% to a range of 37% – 48%. Compared to untreated BSG in several studies, the current study obtained a higher level of SDF. Previous studies

reported the amount of SDF in BSG is at a range of 1% – 10% (Angioloni & Collar, 2011; Nocente et al., 2019). This phenomenon demonstrated the degradation of IDF and conversion to SDF which occurred as an impact of depolymerization, debranching, and/or de-esterification of the polysaccharides due to the thermal exposures as previously observed in other studies (Coelho et al., 2014; B. Li et al., 2019; S. Li et al., 2019). The ratio between SDF and IDF seems to be an important factor in the functional properties of BSG since SDF regulates soft textures while IDF regulates harder textures. Therefore, the impact of autoclave on BSG dietary fiber composition was performed at a degradation level (%) which was calculated by the difference of SDF/IDF in autoclaved and untreated BSG. The results revealed that autoclave treatments increased the SDF/IDF value from a range of 0.09 – 0.23 to a range of 0.15 – 0.37. Therefore, most of the autoclave treatments degraded the dietary fiber composition of BSG up to 87% depending on the treatments, although a slight decline in SDF/IDF was observed in some treatments.

Polysaccharides degradation leads to several alterations including in chemical composition, techno-functionality, and its biological properties. At this stage, FTIR was performed to investigate the chemical surface characteristics of BSG. The results (Figure 1) identified 7 differences in chemical surface characteristic of BSG representing the modification in cellulose and hemicellulose, amine, amide, aromatic hydrocarbon, and aliphatic ethers. The results highlighted the significant impact on the amount of extractable fat content from BSG matrix. Autoclave treatments increased the amount of extractable fat content of BSG from a range of 10% – 11% to a range of 15% – 20% depending on the temperature. The improvement of extractable fat content due to the thermal exposures on BSG has also been observed previously (Kemppainen et al., 2016).

Dietary fiber composition (SDF and IDF) plays an important role in techno-functionality of BSG as a food ingredient. As reported that BSG disrupted food matrix formation due to its excessive capability in binding water. Current study confirmed the ability of autoclave treatments in reducing the water holding and oil holding capability of BSG (Figure 2). Water holding capacity

significantly ($p<0.05$) declined from a range of 3.7 g/g – 3.9 g/g to a range of 2.9 g/g – 3.3 g/g; oil holding capacity declined from a range of 1.9 g/g – 2.1 g/g to a range of 0.9 g/g – 1.6 g/g. By this, the impact of BSG in disrupting the food matrix will be diminished. In other words, BSG might have better performance in food processing. Moreover, autoclaved BSG possessed a higher swelling capacity which might be due to the increasing in SDF and declining in IDF. Swelling capacity significantly ($p<0.05$) increased from 1.7 ml/g to a maximum of 2.2 ml/g. The rise in swelling capacity might be due to the increase of SDF. The positive impact of SDF in regulating textural behavior has also been investigated. It was reported that SDF tends to form softer textures and better gelling and foaming properties while IDF forms rigid texture thus intensifies the hardness of food products (Föste et al., 2020).

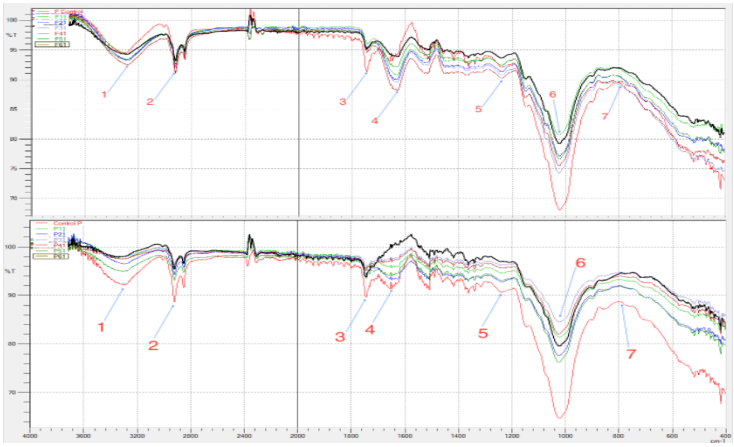

Figure 1. Modification of chemical surface characteristics of autoclaved spent grain (modified from publication 2).

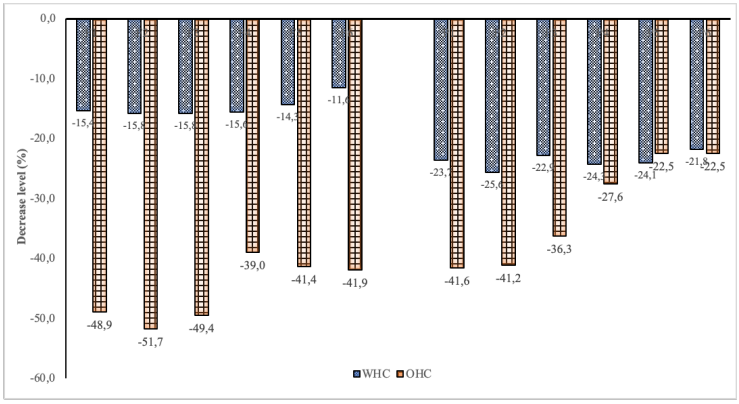

Figure 2. The declining in water holding (WHC) and oil holding capacity (OHC) of autoclaved spent grain (modified from publication 2).

Phenolic compounds and its antioxidant properties of autoclaved BSG were hypothesized to be altered by dietary fiber degradation. It was observed that the weakening bond between lignin

and phenolic groups occurred due to thermal degradation of IDF thus increasing the lignin solubility (Gil-López et al., 2019). Therefore, this phenomenon might have allowed the release of phenolic compounds from polysaccharides moieties thus enhancing the antioxidant activities.

Phenolic compounds of autoclaved BSG were predicted to be influenced by dietary fiber degradation due to the thermal exposures. Therefore, the investigation on the amount of flavan-3-ols and phenolic acids as well as its tentative identification were conducted. Moreover, antioxidant activity of autoclaved BSG was carried out for ORAC (oxygen radical absorbance capacity), ABTS (2,2′-Azinobis-(3-ethylbenzthiazoline-6-sulfonic acid) and FRAP (ferric-reducing antioxidant power).

In general, autoclave treatments dramatically dropped the total amount of flavan-3-ols from 825 mg/kg in untreated BSG to a range of 21 – 48 mg/kg in autoclaved BSG, which is about up to 97% declining. Among those autoclaved BSG, temperature level seemed to significantly ($p<0.05$) impact the amount of total flavan-3-ols. Lower temperatures (90 ℃ and 100 ℃) generated an amount of 21 – 25 mg/kg while at higher temperatures (110 ℃ and 130 ℃) contained an amount of 38 – 48 mg/kg flavan-3-ols. There was a fluctuation in the total amount of phenolic acids as an impact of autoclave treatment. At the temperature level of 90 ℃ and 100 ℃, the amount of phenolic acid dropped from a range of 100 – 105 mg/kg to 30 – 39 mg/kg, which is about 70% decreasing from the initial amount. In contrast, there was an increase of phenolic acids to a range of 344 – 426 mg/kg at 110 ℃ and 1057 – 1168 mg/kg at 130 ℃, which are about 300% and 1000% rising respectively. These findings revealed that the degradation of dietary fiber on autoclaved BSG consequently altered the amount of polyphenolic content as hypothesized. The difference impact of autoclave treatments on flavan-3-ols and phenolic acids suggests the difference in binding stability between phenolic compounds and polysaccharides to thermal exposures. Functional groups of lignin consist of guaiacyl and syringic which similarly bind in all lignin (Ohra-aho et al., 2016), therefore the fluctuation in flavan-3-ols and phenolic acids might be due to the differences in binding resistance to thermal exposures. The increase in phenolic acids due

to thermal exposure has been observed previously on BSG, as an impact of the disruption of cell vacuoles and/or cleave of covalent bonds (Rahman et al., 2021).

Tentative identification of polyphenolic compounds on autoclaved BSG shows that benzoic acid and (+)-catechin were absent at 90 ℃ and 100 ℃ while they were present at 110 ℃ and 130 ℃ autoclaved BSG. It has been identified that hydroxybenzoic acid is the most abundant phenolic acid in BSG (Bonifácio-Lopes et al., 2022). Therefore, the absence of benzoic acid might be responsible for the lower amount of phenolic acids at 90 °C and 100 ℃ as described above. Depolymerization could also occur in phenolic compounds which leads to the conversion into elementary units. For instance, conversion of caffeic acid into ferulic acid (Wojdyło et al., 2014)and derivatization of ferulic acid into 4-vinylguaiacol (Zago et al., 2022) thus improving the antioxidant properties. Therefore, a discharging of certain compounds does not always decline its biological properties.

Although autoclave treatments at 90 ℃ – 100 ℃ declined the number of phenolic acids and dismissed benzoic acid and (+)-catechin, as well as autoclave at 90 ℃ – 130 ℃ declined the amount of flavan-3-ols, it increased the antioxidant activities for ORAC, ABTS and FRAP. Autoclaved BSG contained a higher ORAC value at a range of 4.4 – 6.3 mmol Trolox/100 g which increased from a range of 1.7 – 2.6 mmol Trolox/100 g in untreated BSG; the ABTS value of autoclaved BSG increased from 0.08 – 0.11 mmol Trolox/100 g to 0.15 – 0.84 mmol Trolox/100 g while FRAP value increased from 0.1 – 0.2 mmol Trolox/100 g to a range of 0.4 – 0.5 mmol Trolox/100 g. The improvement in antioxidant activities of autoclaved BSG revealed a benefit impact as a consequence of dietary fiber modification.

The alteration in the amount of polyphenolic compounds, discharging and formation of phenolic compounds as well as increasing and decreasing of antioxidant activity due to thermal exposures has also been reported by several studies. Roasting process was emphasized to reduce the amount of hydroxycinnamic acid content (McCarthy et al., 2013); microwave treatment enhances the antioxidant activity of BSG although the bound phenolic compounds remain stable

(Budaraju et al., 2018). Caffeic acid was formed at >100 °C treated BSG and sinapic acid was present at 160 °C oven heated BSG (Rahman et al., 2021).

The capability of autoclave treatment as a simple and low-cost method in the depolymerization of BSG matrix consequently modified the chemical composition of BSG as well as its techno-processing properties. However, during the experiment, it was observed that using a dried BSG in such an experiment requires more water and energy to re-dry the treated materials. It was also noticed by the reviewer of the published paper. Therefore, further studies were investigated using fresh wet materials thus saving water, time, and energy. The result has clearly shown that the improvement in polyphenolic content and antioxidant activities are linked to dietary fiber modification. The impact of thermal treatment on volatile compounds and fatty acids composition of BSG have never been evaluated. In addition to polyphenolic content, antioxidant activities and techno-functionality, further investigations are conducted on other important parameters including fatty acids composition and volatile compounds, instead of evaluation on dietary fiber degradation.

4.1.2 Autoclave heating treatment on fresh undried-BSG

After preliminary experiments, autoclave treatment was re-designed on fresh-undried BSG. At this stage, fresh BSG contains 69 – 75% moisture content. In the preliminary study, it was observed visually that the amount of water added is important to generate a viscous mixture. Moreover, time exposures did not give a regular trend on BSG properties. For instance, at 90 ℃ autoclaved BSG, the amount of flavan-3-ols was higher at 12 min, at 100 ℃ was higher at 15 min treatment while at 110 and 130 ℃ it was higher at 15 min and 9 min, respectively. Therefore, autoclave treatment was carried out at 12 min as a medium exposure at different water ratios (BSG:water; 1:1 and 1:2; *w:w*) and temperatures (90, 110, and 130 ℃). The results of the study have been published as presented in publication 4:

> Naibaho J., Bobak L., Pudlo A., Wojdylo A., Andayani S. N., Pangestika L. M. W., Korzeniowska M., & Yang B. (2022). Chemical compositions, antioxidant activities and techno-functionality of spent grain treated by autoclave treatment: evaluation of water and temperature levels. *International Journal of Food Science and Technology, 16042.* https://doi.org/10.1111/ijfs.16042. (Publication 4).

In general, there was no clear effect of water ratio on the amount of fatty acids. The results show that autoclave heating decreased the number of saturated fatty acids (SFA) from 45% to 24 – 29% and increased the amount of polyunsaturated fatty acids (PUFA) from 35% to 54 – 71%. All treatments discharged the presence of C17:0; however, the majority of the treatments formed C18:0, C20 and C20:1. Autoclave heating at 130 °C with water ratio 1:1 eliminated the majority of fatty acids. Although in total the fatty acid of BSG is dominated by unsaturated fatty acids (UFA), C16:0 was the highest fatty acid in untreated BSG while autoclaved BSG contained C18:2 (n-6) as the highest fatty acid compound. Autoclave heating might have facilitated the re-arrangement and or depolymerization of SFA into UFA. This phenomenon occurred due to the increasing mass transfer from BSG matrix due to the transesterification (Mallen & Najdanovic-Visak, 2018).

The profile of volatile compounds was evaluated in order to investigate the impact of water ratio (1:1 and 1:2; *w:w*) at 110 °C autoclave treatment for 12 min. The results showed that

autoclave heating declined the number of ketones, alcohols, furans and inclined the amount of fatty acids and aldehydes. This phenomenon was followed by the elimination and formation of several volatile compounds on autoclaved BSG. To mention a few, (E)-2-hexenal and 2-methyl-3-octanone were discharged, while octanal and (E, E)-2,4-decadienal were formed. (E)-2-hexenal is a green leaf volatile compound which acts as anti-fungi and insects for its unpleasant odor (Kunishima et al., 2016) and 2-methyl-3-octanone is responsible for meat-related odor perception (Xia et al., 2020). Different water ratios influenced the elimination and formation of several volatile compounds. The water ratio at 1:1 formed dodecanal while at 1:2 ratio formed (E, E)-2,4-dodecadienal. These compounds had never been reported in BSG. had never been identified in BSG. In addition, several alcohol compounds which are responsible for fatty odor perception were removed and some compounds responsible for fruity and herbal perception were formed. The elimination and formation of those compounds are needed to be investigated further in terms of their significance in general odor acceptability of BSG and BSG-added food products. The actual odor perception can be assessed by the amount of specific compounds to its odor activity value.

The difference in the amount of water ratio had no significant ($p>0.05$) impact on the amount of flavan-3-ols and total polyphenolic compounds. However, the amount of flavan-3-ols and total polyphenols content increased as the temperature increased. Total flavan-3-ols increased from 123 mg/kg to 217 – 997 mg/kg, phenolic acid increased from 44 mg/kg to 70 – 167 mg/kg, and total polyphenolic content increased from 167 mg/kg to 326 – 1163 mg/kg. This phenomenon might be due to the thermal degradation on the dietary fiber composition which allows the release of polyphenolic from BSG matrix, as identified in the preliminary study in previous section. In addition, the increase of the amount of phenolic acid on BSG has also been reported in previous studies (Budaraju et al., 2018; Martín-García et al., 2020) which intensified up to 1.7 – 2.7 folds (Martín-García et al., 2020). The increase in temperature level also increased the antioxidant activities (FRAP and ABTS). However, the difference in water ratio had no significant ($p<0.05$) influence on the antioxidant capabilities. FRAP value increased from 0.7 mmol Trolox/kg to 0.5 –

2.9 mmol Trolox/kg and ABTS value increased from 1.1 mmol Trolox/kg to 0.9 – 3.3 mmol Trolox/kg. In general, the value of polyphenolic content and antioxidant activity in this study are lower compared to that in autoclaved dried BSG. This might be due to the multi-drying process in the preliminary study. High temperature is able to discharge ester-linked ferulic acid from polysaccharides functional groups (Sibhatu et al., 2021) and/or other important phenolic compounds thus improving the antioxidant capabilities. This phenomenon might have increased the bioavailability of phenolic compounds as antioxidants. This is linked to a previous study which reported that antioxidant activity had no correlation with bound phenolic acid (Budaraju et al., 2018).

The majority of the treatment declined the oil holding capacity of autoclaved BSG. This result is aligned with the preliminary study in the previous section. However, autoclave heating increased the water holding capacity of treated BSG from 2.9 g/g to a range of 3.3 – 4.1 g/g regardless of the water ratio and this result is in contrast with the results in the preliminary study. The modification in water holding and oil holding capacity of BSG is influenced by the properties of dietary fiber as observed in rice grain (Zheng et al., 2020). Water holding capacity is highly dependent on the availability of arabinoxylans (Steiner et al., 2015).

Autoclave treatment affects the chemical composition, techno-functionality, and antioxidant capabilities of BSG depending on several factors including the handling materials such as drying process, as well as temperature levels, time exposures and water addition during the treatment.

4.1.3 Water-bath treatment

In the autoclave treatment, water ratio seemed to only influence the volatile composition which is related to the odor perception. The most influential factor on alteration of chemical compounds, functionality, and antioxidant activities of BSG is temperature levels and time exposures. Therefore, water-bath treatment was carried out evaluating different temperature levels and time exposures. Water-bath was used at 40 and 90 ℃ for 60 and 30 min respectively in heating treatment of low-sodium surimi (Ye et al., 2022) and 80 ℃ for 1 – 8h in heating treatment of chitose-fructose Maillard reaction product and modification of taste in seafood (H. Zhang et al., 2015; R. Zhang et al., 2018). Current study was conducted at 80, 90, and 100 ± 1 ℃ for 15, 30, and 60 min with water ratio 1:1 (BSG:water; *w:w*). The impact of water-bath heating on fatty acids, volatile compounds, polyphenolic content, antioxidant activities and techno-functionality of BSG is presented in publication 5:

> Naibaho J., Pudlo A., Bobak L., Wojdylo A., Lopez A. A., Pangestika L. M. W., Andayani S. N., Korzeniowska M., & Yang B. (2023). Conventional water bath heating on undried brewers' spent grain: techno-functionality, fatty acids, volatiles, polyphenolic profile and antioxidant properties. Food Bioscience.

As autoclave treatment, water-bath treatment also decreased the amount of SFA and increased the number of PUFA. The amount of SFA declined from 45% to 26 – 28% while PUFA inclined from 35% to 53 – 55%. Water-bath treatment also eliminated the presence of C17:0 and formed C18:0 and C20:1 as it occurred in autoclaved BSG. Interestingly, several fatty acids which were formed including C13:0, C17:0, C20:2, C22:1 and C24:1 have been identified in dried and lyophilised BSG (Fărcaş et al., 2015). The increase of UFA in BSG has also been identified due to the solid-state fermentation (Tan et al., 2019).

The impact of water-bath treatment on volatile compounds of BSG revealed that the amount of aldehydes and furan increased as the temperature and time exposure increased. However, it declined the amount of ketones, alcohols, alkenes, and other minor groups. As observed in autoclave treatment, several volatile compounds were dismissed and at the same time

some other compounds were formed due to the water-bath treatment. Further investigation is needed in order to investigate the impact of thermal treatment on odor perception of BSG.

Water-bath treatment decreased the amount of phenolic acid from 44 mg/kg to a range of 9 – 27 mg/kg. The amount of flavan-3-ols fluctuated from 123 mg/kg to a range of 108 – 151 mg/kg while total polyphenolic content fluctuated from 167 mg/kg to 118 – 187 mg/kg. The amount of flavan-3-ols, phenolic acid, and total polyphenolic content on BSG treated with water-bath heating were lower compared to that in BSG treated with autoclave treatment. Compared to autoclave treatment in the previous section, autoclaved BSG at 90 ℃ contained phenolic acids at 69 – 118 mg/kg, flavan-3-ols at 216 – 257 mg/kg, and total polyphenolic content at 326 – 335 mg/kg. This result demonstrates that the amount of polyphenolic compounds is not only influenced by different temperature and time exposures but also the pressure which was applied during the autoclave treatment. However, this explanation might not be applicable for antioxidant activities. A wide fluctuation in antioxidant activities of BSG was also observed in the current study. Most of the treatment declined the antioxidant activities of BSG. Water-bath treatment fluctuated the FRAP value from 0.7 mmol Trolox/kg to 0.2 – 0.8 mmol Trolox/kg and fluctuated the ABTS value from 1.1 mmol Trolox/kg to 0.7 – 1.3 mmol Trolox/kg. Compared to autoclave treatment, BSG treated at 90 ℃ possessed FRAP value at 0.5 – 0.7 mmol Trolox/kg and ABTS value at 0.9 mmol Trolox/kg. By this, the majority of BSG treated with water-bath treatment had the same level of ABTS value as in autoclaved BSG at 90 ℃ while some of BSGs treated with water-bath had the same level of FRAP value.

Water-bath treatment significantly ($p<0.05$) increased the water holding and oil holding capacity of BSG regardless of temperature levels and time exposures. Water holding capacity increased from 2.9 g/g to 3.4 – 3.8 g/g and oil holding capacity increased from 2.05 g/g to 2.09 – 2.16 g/g. Compared to the previous section, water-bath heating generated the same water holding capacity as it was in autoclaved BSG. However, water-bath treatment generated a higher oil holding capacity of BSG.

The results showed that, autoclave and water-bath modified the chemical constituent of BSG differently, particularly in fatty acid composition and polyphenolic compounds. Consequently, this might influence the biological properties of BSG including FRAP and ABTS antioxidant activity. The impact of thermal treatments on the odor perception of BSG still needs to be evaluated further. From a techno-processing or functionality point of view, autoclave and water-bath heating treatment generated different functionality of BSG which might influence the application of BSG in food processing.

4.2 The impact of protein extraction on polyphenolic compounds and antioxidant properties of BSG and obtained protein

According to the literature review in publication 1, protein is in the second place of the most evaluated compounds from BSG after polyphenolic compounds. It is widely accepted that protein is mainly covered in the vacuole cell of BSG matrix. Therefore, polysaccharides degradation including dietary fiber occurs due to the protein extraction and consequently might lead to decomposition of chemical compounds and alteration of its antioxidant properties. As protein is protected in the vacuole cell of BSG, the incubation process disrupted the vacuole cell of BSG differently depending on the presence of enzymes. BSG was incubated with three different incubation conditions with the addition of 0.5% Protamex, combination of 0.5% Protamex and 0.1% Flavourzyme, and control without enzyme treatment (Kriisa et al., 2022). The study mainly aimed to investigate the influence of protein extraction on antioxidant properties, total amount of total polyphenolic compounds and tentative identification of polyphenolic compounds in BSG residue as a byproduct of protein extraction as well as in extracted protein. The results of the study are presented in Publication 6:

Naibaho, J., Korzeniowska, M., Wojdyło, A., Ayunda, HM., Foste, M., & Yang, B. (2022). Techno-functional properties of protein from protease-treated brewers' spent grain (BSG) and investigation of antioxidant activity of extracted proteins and BSG residues. *Journal of Cereal Science, 107, 103524.* https://doi.org/10.1016/j.jcs.2022.103524.

The results show that the amount of phenolic acid was discovered to be higher in protein extracts than in BSG sediments. BSG proteins contained phenolic acids at a range of 65 – 123 mg/kg while the sediments at 17 – 26 mg/kg. By this, protein extraction can increase the release rate of phenolic acid from BSG matrix to BSG protein. Comparing the total amount of phenolic acid in sediment and protein, protein extraction without enzyme generated a higher total amount of phenolic acid. Incubation with Protamex and Flavourzyme seemed to inhibit the release of phenolic acid from BSG matrix. Total phenolic acids in BSG residues and protein extracts in control treatment was at 140 mg/kg while in 0.5% Protamex and Protamex/Flavourzyme treatment was at 92 mg/kg and 90 mg/kg respectively. As mentioned in previous section, original untreated

BSG contained 44 mg/kg of phenolic acid. This result demonstrated that protein extraction increased the availability of phenolic acids which was released from BSG matrix to the protein fraction. The amount of flavan-3-ols in BSG protein and sediments seemed to be depending on the enzymatic incubation during the protein extraction. Incubation process without enzymes generated a higher flavan-3-ols in protein fraction while incubation with Protamex and Flavourzyme generated a higher flavan-3-ols in BSG sediments. However, the total amount of flavan-3-ols in BSG protein and sediment is higher than that in the untreated BSG. Untreated BSG contained 123 mg/kg flavan-3-ols while the protein extraction process generated total flavan-3-ols in protein and residue at a range of 204 – 419 mg/kg.

Tentative identification of polyphenolic compounds revealed that several compounds including syringic acid, sinapic acid, di-ferulic acid and (+)-catechin were removed from BSG sediments to BSG protein fraction. The main phenolic compounds in BSG are dominated by sinapic acid, p-coumaric acid, ferulic acid and caffeic acids as well as their derivatives (Sibhatu et al., 2021). The study showed that caffeic acid was not present in both BSG proteins and sediments. This phenomenon might be due to the conversion of caffeic acid into ferulic acid (McCarthy et al., 2013). Conversion of several phenolic compounds could occur due to the instability of certain compounds to certain circumstances. For instance, ohmic heating significantly facilitates the release of hydroxybenzoic acids from BSG matrix (Bonifácio-Lopes et al., 2022), caffeic acid was absent in <100 ℃ oven treatment but it was present in >100 ℃ treatment, and sinapic acid was present at >160 ℃ oven heating (Rahman et al., 2021).

The study demonstrated that protein fraction possessed a higher antioxidant activity (ABTS and FRAP) compared to that in BSG sediments. BSG proteins had the ABTS level at 41.5 – 55.5 mmol Trolox/kg while BSG sediments at a range of 3.2 – 6.3 mmol Trolox/kg. FRAP value of BSG protein fractions ranged from 4.4 to 5.3 mmol Trolox/kg while the sediments ranged from 1.7 to 3.4 mmol Trolox/kg. Compared to the untreated BSG which possessed ABTS 1.1 mmol Trolox/kg and FRAP 0.7 mmol Trolox/kg, protein extraction generated a higher ABTS and FRAP

value both in proteins and the sediments. A high antioxidant activity in BSG protein might be due to the availability of protein in addition to the presence of phenolic compounds. A higher level of antioxidant activity in BSG sediments compared to the original BSG revealed that protein extraction might have increased the bioavailability of phenolic compounds in BSG matrix as antioxidant. As hypothesized that protein extraction processes have an influence on the release of phenolic compounds from BSG matrix thus improving the bioavailability of phenolic compounds both in protein fraction and BSG sediments.

5. Conclusion

The study demonstrated that several treatments could be applied in modifying the chemical composition, such as polyphenolic compounds, fatty acid, and volatile compounds, techno-functionality, including water holding capacity and oil holding capacity, as well as, antioxidant properties of BSG. The modification in those properties is mainly due to the degradation of polysaccharides, as a main body of chemical structure of BSG. A conversion of insoluble dietary fiber into soluble dietary fiber was observed due to the high temperature exposure on BSG. Different treatments on BSG including autoclave heating, water-bath heating and protein extraction have fluctuated the chemical composition of BSG, particularly polyphenolic compositions, thus its antioxidant activities. This phenomenon occurs along with the elimination and formation of certain polyphenolic compounds. The impact of studied treatments on chemical composition of BSG revealed the different stability of the chemical composition of BSG to certain exposure conditions.

Multi-drying steps might have intensified the number of polyphenolic compounds due to the degradation of dietary fiber composition. However, it requires higher energy and excessive water use. In addition, there might be a negative impact on protein functionality due to the excessive thermal exposures. Therefore, the treatment on BSG can be applied on fresh undried BSG. It is worth mentioning that autoclave treatment affects the chemical composition, techno-functionality, and antioxidant capabilities of BSG depending on several factors, including the handling materials, such as drying process, as well as, temperature levels, time exposures and water addition during the treatment. Autoclave treatment and water-bath heating decreased the amount of saturated fatty acids and increased the amount of polyunsaturated fatty acids. Thermal treatment fluctuated the amount of volatile compounds of BSG along with discharging and forming certain compounds. Autoclave treatment generated a higher level of total flavan-3-ols, phenolic acid and total polyphenolic content compared to that in water-bath treatment, suggesting that pressure in autoclave treatment might have facilitate the release of certain compounds from BSG

matrix. Protein extraction from BSG increased the release of flavan-3-ols, phenolic acids and total phenolic acids from BSG thus increasing the availability of those compounds in BSG protein and residues as antioxidants.

Finding the most efficient treatment would be depending on the specific target. In case of specific bioactive compounds extraction such as phenolic, fatty acids, dietary fiber and/or others optimization of techniques can be expected. From the perspective of techno-processing in food development, modification of functionality such as rehydration properties and oil holding capacity as well as modification in odor perception are approachable in order to increase the processing efficiency and final products acceptability. The conducted studies have the limitation, therefore, several investigations are seemingly important to be continued including the significant impact on odor profile, quantification of specific phenolic compounds, as well as the impact of treated BSG in food processing efficiency, food products acceptability and nutritional value.

6. Bibliography

Angioloni, A., & Collar, C. (2011). Physicochemical and nutritional properties of reduced-caloric density high-fibre breads. *LWT - Food Science and Technology*, *44*(3), 747–758. https://doi.org/10.1016/j.lwt.2010.09.008

Aprodu, I., Simion, A. B., & Banu, I. (2017). Valorisation of the Brewers' Spent Grain Through Sourdough Bread Making. *International Journal of Food Engineering*, *13*(10). https://doi.org/10.1515/ijfe-2017-0195

Barbosa-Pereira, L., Bilbao, A., Vilches, P., Angulo, I., LLuis, J., Fité, B., Paseiro-Losada, P., & Cruz, J. M. (2014). Brewery waste as a potential source of phenolic compounds: Optimisation of the extraction process and evaluation of antioxidant and antimicrobial activities. *Food Chemistry*, *145*, 191–197. https://doi.org/10.1016/j.foodchem.2013.08.033

Bonifácio-Lopes, T., Teixeira, J. A., & Pintado, M. (2020). Current extraction techniques towards bioactive compounds from brewer's spent grain – A review. *Critical Reviews in Food Science and Nutrition*, *60*(16), 2730–2741. https://doi.org/10.1080/10408398.2019.1655632

Bonifácio-Lopes, T., Vilas-Boas, A., Machado, M., Costa, E. M., Silva, S., Pereira, R. N., Campos, D., Teixeira, J. A., & Pintado, M. (2022). Exploring the bioactive potential of brewers spent grain ohmic extracts. *Innovative Food Science & Emerging Technologies*, *76*, 102943. https://doi.org/10.1016/j.ifset.2022.102943

Budaraju, S., Mallikarjunan, K., Annor, G., Schoenfuss, T., & Raun, R. (2018). Effect of pre-treatments on the antioxidant potential of phenolic extracts from barley malt rootlets. *Food Chemistry*, *266*, 31–37. https://doi.org/10.1016/j.foodchem.2018.05.110

Cappa, C., & Alamprese, C. (2017). Brewer's spent grain valorization in fiber-enriched fresh egg pasta production: Modelling and optimization study. *LWT - Food Science and Technology*, *82*, 464–470. https://doi.org/10.1016/j.lwt.2017.04.068

Coelho, E., Rocha, M. A. M., Saraiva, J. A., & Coimbra, M. A. (2014). Microwave superheated water and dilute alkali extraction of brewers' spent grain arabinoxylans and arabinoxylo-oligosaccharides. *Carbohydrate Polymers*, *99*, 415–422. https://doi.org/10.1016/j.carbpol.2013.09.003

Conway, J. (2022). *Volume of beer produced in Europe in 2020*. Statista. https://www.statista.com/statistics/444575/european-beer-production-by-country/

Crowley, D., O'Callaghan, Y., McCarthy, A. L., Connolly, A., Fitzgerald, R. J., & O'Brien, N. M. (2017). Aqueous and enzyme-extracted phenolic compounds from brewers' spent grain (BSG): Assessment of their antioxidant potential: CROWLEY et al. *Journal of Food Biochemistry*, *41*(3), e12370. https://doi.org/10.1111/jfbc.12370

Fărcaş, A. C., Socaci, S. A., Dulf, F. V., Tofană, M., Mudura, E., & Diaconeasa, Z. (2015). 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. *Journal of Cereal Science*, *64*, 34–42. https://doi.org/10.1016/j.jcs.2015.04.003

Föste, M., Verheyen, C., Jekle, M., & Becker, T. (2020). Fibres of milling and fruit processing by-products in gluten-free bread making: A review of hydration properties, dough formation and quality-improving strategies. *Food Chemistry*, *306*, 125451. https://doi.org/10.1016/j.foodchem.2019.125451

Gil-López, D. I. L., Lois-Correa, J. A., Sánchez-Pardo, M. E., Domínguez-Crespo, M. A., Torres-Huerta, A. M., Rodríguez-Salazar, A. E., & Orta-Guzmán, V. N. (2019). Production of dietary fibers from sugarcane bagasse and sugarcane tops using microwave-assisted alkaline treatments. *Industrial Crops and Products*, *135*, 159–169. https://doi.org/10.1016/j.indcrop.2019.04.042

Heredia-Sandoval, N. G., Granados-Nevárez, M. del C., Calderón de la Barca, A. M., Vásquez-Lara, F., Malunga, L. N., Apea-Bah, F. B., Beta, T., & Islas-Rubio, A. R. (2020). Phenolic Acids, Antioxidant Capacity, and Estimated Glycemic Index of

Cookies Added with Brewer's Spent Grain. *Plant Foods for Human Nutrition*, *75*(1), 41–47. https://doi.org/10.1007/s11130-019-00783-1

Jung, H., Moon, J. H., Park, J. W., & Yoon, W. B. (2020). Texture of surimi-canned corn mixed gels with conventional water bath cooking and ohmic heating. *Food Bioscience*, *35*, 100580. https://doi.org/10.1016/j.fbio.2020.100580

Kemppainen, K., Rommi, K., Holopainen, U., & Kruus, K. (2016). Steam explosion of Brewer's spent grain improves enzymatic digestibility of carbohydrates and affects solubility and stability of proteins. *Applied Biochemistry and Biotechnology*, *180*(1), 94–108. https://doi.org/10.1007/s12010-016-2085-9

Kriisa, M., Taivosalo, A., Föste, M., Kütt, M.-L., Viirma, M., Priidik, R., Korzeniowska, M., Tian, Y., Laaksonen, O., Yang, B., & Vilu, R. (2022). Effect of enzyme-assisted hydrolysis on brewer's spent grain protein solubilization – peptide composition and sensory properties. *Applied Food Research*, *2*(1), 100108. https://doi.org/10.1016/j.afres.2022.100108

Ktenioudaki, A., Alvarez-Jubete, L., Smyth, T. J., Kilcawley, K., Rai, D. K., & Gallagher, E. (2015). Application of bioprocessing techniques (sourdough fermentation and technological aids) for brewer's spent grain breads. *Food Research International*, *73*, 107–116. https://doi.org/10.1016/j.foodres.2015.03.008

Kumari, B., Tiwari, B. K., Walsh, D., Griffin, T. P., Islam, N., Lyng, J. G., Brunton, N. P., & Rai, D. K. (2019). Impact of pulsed electric field pre-treatment on nutritional and polyphenolic contents and bioactivities of light and dark brewer's spent grains. *Innovative Food Science & Emerging Technologies*, *54*, 200–210. https://doi.org/10.1016/j.ifset.2019.04.012

Kunishima, M., Yamauchi, Y., Mizutani, M., Kuse, M., Takikawa, H., & Sugimoto, Y. (2016). Identification of (Z)-3:(E)-2-Hexenal Isomerases Essential to the Production

of the Leaf Aldehyde in Plants. *Journal of Biological Chemistry*, *291*(27), 14023–14033. https://doi.org/10.1074/jbc.M116.726687

Li, B., Yang, W., Nie, Y., Kang, F., Goff, H. D., & Cui, S. W. (2019). Effect of steam explosion on dietary fiber, polysaccharide, protein and physicochemical properties of okara. *Food Hydrocolloids*, *94*, 48–56. https://doi.org/10.1016/j.foodhyd.2019.02.042

Li, S., Chen, G., Qiang, S., Tang, D., Chen, Y., Zhang, Z., Lei, Z., & Chen, Y. (2019). Intensifying soluble dietary fiber production and properties of soybean curd residue via autoclaving treatment. *Bioresource Technology Reports*, *7*, 100203. https://doi.org/10.1016/j.biteb.2019.100203

Lynch, K. M., Steffen, E. J., & Arendt, E. K. (2016). Brewers' spent grain: A review with an emphasis on food and health. *Journal of the Institute of Brewing*, *122*(4), 553–568. https://doi.org/10.1002/jib.363

Magabane, I. E. (2017). *Technologies for improving the quality of bread doughs made with barley spent grain and sorghum*. University of Pretoria.

Mallen, E., & Najdanovic-Visak, V. (2018). Brewers' spent grains: Drying kinetics and biodiesel production. *Bioresource Technology Reports*, *1*, 16–23. https://doi.org/10.1016/j.biteb.2018.01.005

Martín-García, B., Tylewicz, U., Verardo, V., Pasini, F., Gómez-Caravaca, A. M., Caboni, M. F., & Dalla Rosa, M. (2020). Pulsed electric field (PEF) as pre-treatment to improve the phenolic compounds recovery from brewers' spent grains. *Innovative Food Science & Emerging Technologies*, *64*, 102402. https://doi.org/10.1016/j.ifset.2020.102402

McCarthy, A. L., O'Callaghan, Y. C., Connolly, A., Piggott, C. O., FitzGerald, R. J., & O'Brien, N. M. (2014). Phenolic-enriched fractions from brewers' spent grain possess cellular antioxidant and immunomodulatory effects in cell culture model systems:

Anti-inflammatory effects of brewers' spent grain phenolic extracts. *Journal of the Science of Food and Agriculture*, *94*(7), 1373–1379. https://doi.org/10.1002/jsfa.6421

McCarthy, A. L., O'Callaghan, Y. C., Neugart, S., Piggott, C. O., Connolly, A., Jansen, M. A. K., Krumbein, A., Schreiner, M., FitzGerald, R. J., & O'Brien, N. M. (2013). The hydroxycinnamic acid content of barley and brewers' spent grain (BSG) and the potential to incorporate phenolic extracts of BSG as antioxidants into fruit beverages. *Food Chemistry*, *141*(3), 2567–2574. https://doi.org/10.1016/j.foodchem.2013.05.048

Mussatto, S. I., Dragone, G., & Roberto, I. C. (2006). Brewers' spent grain: Generation, characteristics and potential applications. *Journal of Cereal Science*, *43*(1), 1–14. https://doi.org/10.1016/j.jcs.2005.06.001

Nascimento, T. A., Calado, V., & Carvalho, C. W. P. (2017). Effect of Brewer's spent grain and temperature on physical properties of expanded extrudates from rice. *LWT - Food Science and Technology*, *79*, 145–151. https://doi.org/10.1016/j.lwt.2017.01.035

Nawaz, A., Li, E., Irshad, S., Hhm, H., Liu, J., Shahbaz, H. M., Ahmed, W., & Regenstein, J. M. (2020). Improved effect of autoclave processing on size reduction, chemical structure, nutritional, mechanical and in vitro digestibility properties of fish bone powder. *Advanced Powder Technology*, *31*(6), 2513–2520. https://doi.org/10.1016/j.apt.2020.04.015

Nocente, F., Taddei, F., Galassi, E., & Gazza, L. (2019). Upcycling of brewers' spent grain by production of dry pasta with higher nutritional potential. *LWT*, *114*, 108421. https://doi.org/10.1016/j.lwt.2019.108421

Ohra-aho, T., Niemi, P., Aura, A.-M., Orlandi, M., Poutanen, K., Buchert, J., & Tamminen, T. (2016). Structure of Brewer's Spent Grain Lignin and Its Interactions with Gut Microbiota in Vitro. *Journal of Agricultural and Food Chemistry*, *64*(4), 812–820. https://doi.org/10.1021/acs.jafc.5b05535

Rahman, Md. J., Malunga, L. N., Eskin, M., Eck, P., Thandapilly, S. J., & Thiyam-Hollander, U. (2021). Valorization of Heat-Treated Brewers' Spent Grain Through the Identification of Bioactive Phenolics by UPLC-PDA and Evaluation of Their Antioxidant Activities. *Frontiers in Nutrition*, *8*, 634519. https://doi.org/10.3389/fnut.2021.634519

Reis, S. F., Coelho, E., Coimbra, M. A., & Abu-Ghannam, N. (2015). Improved efficiency of brewer's spent grain arabinoxylans by ultrasound-assisted extraction. *Ultrasonics Sonochemistry*, *24*, 155–164. https://doi.org/10.1016/j.ultsonch.2014.10.010

Sibhatu, H. K., Anuradha Jabasingh, S., Yimam, A., & Ahmed, S. (2021). Ferulic acid production from brewery spent grains, an agro-industrial waste. *LWT*, *135*, 110009. https://doi.org/10.1016/j.lwt.2020.110009

Socaci, S. A., Fărcaş, A. C., Diaconeasa, Z. M., Vodnar, D. C., Rusu, B., & Tofană, M. (2018). Influence of the extraction solvent on phenolic content, antioxidant, antimicrobial and antimutagenic activities of brewers' spent grain. *Journal of Cereal Science*, *80*, 180–187. https://doi.org/10.1016/j.jcs.2018.03.006

Steiner, J., Procopio, S., & Becker, T. (2015). Brewer's spent grain: Source of value-added polysaccharides for the food industry in reference to the health claims. *European Food Research and Technology*, *241*(3), 303–315. https://doi.org/10.1007/s00217-015-2461-7

Steinmacher, N. C., Honna, F. A., Gasparetto, A. V., Anibal, D., & Grossmann, M. V. E. (2012). Bioconversion of brewer's spent grains by reactive extrusion and their application in bread-making. *LWT - Food Science and Technology*, *46*(2), 542–547. https://doi.org/10.1016/j.lwt.2011.11.011

Tan, Y. X., Mok, W. K., Lee, J., Kim, J., & Chen, W. N. (2019). Solid State Fermentation of Brewers' Spent Grains for Improved Nutritional Profile Using Bacillus subtilis WX-17. *Fermentation*, *5*(3), 52. https://doi.org/10.3390/fermentation5030052

Torbica, A., Škrobot, D., Janić Hajnal, E., Belović, M., & Zhang, N. (2019). Sensory and physico-chemical properties of wholegrain wheat bread prepared with selected food by-products. *LWT*, *114*, 108414. https://doi.org/10.1016/j.lwt.2019.108414

Verni, M., Pontonio, E., Krona, A., Jacob, S., Pinto, D., Rinaldi, F., Verardo, V., Díaz-de-Cerio, E., Coda, R., & Rizzello, C. G. (2020). Bioprocessing of Brewers' Spent Grain Enhances Its Antioxidant Activity: Characterization of Phenolic Compounds and Bioactive Peptides. *Frontiers in Microbiology*, *11*, 1831. https://doi.org/10.3389/fmicb.2020.01831

Wen, C., Zhang, J., Duan, Y., Zhang, H., & Ma, H. (2019). A Mini-Review on Brewer's Spent Grain Protein: Isolation, Physicochemical Properties, Application of Protein, and Functional Properties of Hydrolysates. *Journal of Food Science*, *84*(12), 3330–3340. https://doi.org/10.1111/1750-3841.14906

Wojdyło, A., Figiel, A., Lech, K., Nowicka, P., & Oszmiański, J. (2014). Effect of Convective and Vacuum–Microwave Drying on the Bioactive Compounds, Color, and Antioxidant Capacity of Sour Cherries. *Food and Bioprocess Technology*, *7*(3), 829–841. https://doi.org/10.1007/s11947-013-1130-8

Xia, Q., Feng, T., Lou, X., Wang, Y., Sun, Y., Pan, D., & Cao, J. (2020). Headspace fingerprinting approach to identify the major pathway influencing volatile patterns of vinasse-cured duck processed by high pressure, as well as its impact on physicochemical and sensory attributes. *International Journal of Food Science & Technology*, *55*(2), 669–680. https://doi.org/10.1111/ijfs.14321

Xiros, C., & Christakopoulos, P. (2012). Biotechnological Potential of Brewers Spent Grain and its Recent Applications. *Waste and Biomass Valorization*, *3*(2), 213–232. https://doi.org/10.1007/s12649-012-9108-8

Ye, Y., Liu, X., Bai, W., Zhao, W., Zhang, Y., Dong, H., & Pan, Z. (2022). Effect of microwave-ultrasonic combination treatment on heating-induced gel properties of

low-sodium tilapia surimi during gel setting stage and comparative analysis. *LWT*, *161*, 113386. https://doi.org/10.1016/j.lwt.2022.113386

Yu, D., Sun, Y., Wang, W., O'Keefe, S. F., Neilson, A. P., Feng, H., Wang, Z., & Huang, H. (2020). Recovery of protein hydrolysates from brewer's spent grain using enzyme and ultrasonication. *International Journal of Food Science & Technology*, *55*(1), 357–368. https://doi.org/10.1111/ijfs.14314

Zago, E., Tillier, C., De Leener, G., Nandasiri, R., Delporte, C., Bernaerts, K. V., & Shavandi, A. (2022). Sustainable production of low molecular weight phenolic compounds from Belgian Brewers' spent grain. *Bioresource Technology Reports*, *17*, 100964. https://doi.org/10.1016/j.biteb.2022.100964

Zhang, H., Yang, J., & Zhao, Y. (2015). High intensity ultrasound assisted heating to improve solubility, antioxidant and antibacterial properties of chitosan-fructose Maillard reaction products. *LWT - Food Science and Technology*, *60*(1), 253–262. https://doi.org/10.1016/j.lwt.2014.07.050

Zhang, R., Qiu, W., Zhang, M., Ho Row, K., Cheng, Y., & Jin, Y. (2018). Effects of different heating methods on the contents of nucleotides and related compounds in minced Pacific white shrimp and Antarctic krill. *LWT*, *87*, 142–150. https://doi.org/10.1016/j.lwt.2017.08.078

Zheng, Y., Wei, Z., Zhang, R., Deng, Y., Tang, X., Zhang, Y., Liu, G., Liu, L., Wang, J., Liao, N., & Zhang, M. (2020). Optimization of the autoclave preparation process for improving resistant starch content in rice grains. *Food Science & Nutrition*, *8*(5), 2383–2394. https://doi.org/10.1002/fsn3.1528

7. Attachment

Publication 1

Received: 27 October 2020 | Revised: 1 March 2021 | Accepted: 11 March 2021

DOI: 10.1111/1750-3841.15714

CONCISE REVIEWS & HYPOTHESES IN FOOD SCIENCE

Journal of Food Science WILEY

Brewers' spent grain in food systems: Processing and final products quality as a function of fiber modification treatment

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Abstract: The nutritional properties of brewers' spent grain (BSG) have been widely studied, considering its potential as a healthy food ingredient. Because of its fiber composition (amount and ratio), however, adding BSG into the food matrix to bring about changes in physical properties has been believed to impact negatively on the acceptability of the final products' properties, particularly color and texture. Fiber modification can enhance the quality of fiber and can be applied to BSG. Although it appears challenging, modifying fiber composition requires further study, particularly if the acceptability of the final products is to be improved. Furthermore, the level of fiber degradation during the modification treatment needs to be examined to meet the increased demand for BSG in final food products. This concise synthesis provides a new perspective for increasing the use of BSG as a food ingredient that is characterized by high nutrition and acceptability.

KEYWORDS
agro-industrial byproduct, brewers' spent grain, dietary fiber, fiber modification, physical properties

1 | INTRODUCTION: VALORIZATION AS A TREND

Valorization of agro-industrial byproducts (ABPs) is becoming increasingly popular due to their effects on waste and environmental harm reduction, economic growth, and enhancement of human welfare, particularly when they are used as a global food ingredient (Morone et al., 2019). Several ABPs have been studied for their properties and have been reported to contain bioactive compounds and could thus potentially be used as food ingredients (Ačkar et al., 2018; Aggelopoulos et al., 2014; Bora et al., 2019; Grasso, 2020; Prandi et al., 2019; Spinelli et al., 2019).

Brewers' spent grain (BSG), as the main byproduct of the brewery industry, accounts for approximately 40% of overall beer manufacturing waste (Garcia-Garcia et al., 2019). In the past nine years, about 1.9 billion hectoliters of beer have been produced annually globally, led by China and followed by the United States and Brazil (Conway, 2019), that is, about 0.76 billion hectoliters of BSG are produced annually. In European countries, it is stated that about 3.4 million tons of BSG are generated annually (Nigam, 2017). Currently, BSG is used as an animal feed for ruminants and fish and as an organic fertilizer (Lao et al., 2020; Skendi et al., 2018). From a biotechnological perspective, BSG has been widely studied for enzyme production, value-added products, and functional protein and

fat production (Xiros & Christakopoulos, 2012). Another utilization of BSG is for the production of construction bricks and as a biosorbent for metal (Aliyu & Bala, 2011).

BSG contains abundant nutritional compounds. Thus, it has been extensively explored for its potential in several fields, such as extracted bioactive compounds and protein (Carciochi et al., 2018; Cian et al., 2018; Connolly et al., 2019; Socaci et al., 2018; Stefanello et al., 2018; Zuorro et al., 2019). The use of BSG as a food ingredient, and in a wider range of food applications and treatments, has been studied by numerous researchers as shown in Table 2. The current study collected and reviewed original research papers that examined and used BSG in food production and were published during the 10-year period from 2010 to 2020. A total of 38 articles were collected from a Scopus and Google Scholar search. In this review study, utilization of distillers' spent grain (DSG), a cereal byproduct from bioethanol production, is included. BSG and DSG are cereal byproducts that have the same impact in terms of their valorization as food ingredients (Roth, Schuster, et al., 2016). The applications of BSG and DSG in food products include bread, extrudate products, cookies, snacks and crackers, pasta, composite flour, dough, yogurt, and cheese (Abd EL-Moneim et al., 2015; Abd El-Moneim et al., 2018; Heredia-Sandoval et al., 2020; Nocente et al., 2019; Sobukola et al., 2013; Torbica et al., 2019).

The sensory property of food products is an important factor. The use of BSG in food processing is known to have positive health effects and is expected to increase the food's appeal, and a consumer survey placed sensory properties as the second most important factor after health issues (Combest & Warren, 2019; Crofton & Scannell, 2020). The maximum addition of BSG as a substitutive ingredient that is considered to be effective is between 20% and 25%, depending on the products and treatments. Interestingly, a more positive result from the addition of 30% of BSG to cereal-breakfast extrudates (Thorvaldsson, 2020) and the highest sensory value for the addition of 50% of BSG in cheese block production (Abd El-Moneim et al., 2018) were noted, compared to lower additions, as discussed in Section 3.

Most of the published papers underline the limitations of BSG. In general, the addition of BSG to food processing is limited because sensory properties, including taste, texture, color, and aroma of the modified products, have been found to decline (Nascimento et al., 2017; Żelaziński et al., 2018), and changes have also been reported in mechanical properties (Ktenioudaki, O'Shea, et al., 2013). The most recent updates regarding BSG relate to functional properties, including the functionality of dietary fiber (DF) from BSG in bread making (Stojceska, 2011), functional ingredients of BSG (Ikram et al., 2017; Stojceska, 2019; Xiros & Christakopoulos, 2012), opportunities to use BSG as a food ingredient (Aliyu & Bala, 2011; Lynch et al., 2016; Rachwał et al., 2020; Roth et al., 2019; Steiner et al., 2015), and bioactive compound extraction (Bonifácio-Lopes et al., 2020; Guido & Moreira, 2017; Wen et al., 2019). To the best of the authors' knowledge, the impact of BSG as a substitute ingredient during food processing and its impact on the quality of the final food products has not been closely examined.

The aim of this review is to present the main issues of the incorporation of BSG in food processing, such as those relating to mechanical and processing technology properties, textural properties, color, and acceptability, in addition to the developed methods that have been applied to improve the quality of final products. Moreover, the possibility of fiber modification treatment of BSG, its impact on physical properties and final product quality, and the challenges to increasing the use of BSG in food systems are presented.

2 | NUTRITION AND BIOLOGICAL ACTIVITY OF BSG

Dried BSG contains approximately 5% to 8% moisture content, 2.7% to 5% ash, 14.5% to 30% protein nitrogen, 0.43% to 2.17% starch, and extracted fat ranging from 8% to 34.82% (Balogun et al., 2017; Cooray & Chen, 2018; Nocente et al., 2019). BSG contains minerals such as phosphorus, calcium, sulfur, magnesium, and potassium (Meneses et al., 2013). The amounts of those compositions can differ due to the type of beer processing, handling, or treatment during storage and drying methods of the spent grain (Meneses et al., 2013). The biological activity of BSG has been widely studied as shown in Table 1. From the number of observed papers, the most well-known biological properties of BSG are phenolic compounds followed by isolate protein. However, other compounds, such as fatty acids, soluble DF (SDF), ß-glucan, and melanoidins, have also been reported (Table 1).

The major phenolic compounds in BSG include protocatechuic, caffeic, *p*-coumaric and ferulic acids, catechin, and derivatives (Barbosa-Pereira et al., 2014; McCarthy et al., 2013; Moreira et al., 2013; Sibhatu et al., 2021). The extract compositions of BSG and its biological activity vary depending on several factors, such as the roasting and brewing process (McCarthy et al., 2013), solvents used, and extraction methods (Barbosa-Pereira et al., 2014; Bonifácio-Lopes et al., 2020; Meneses et al., 2013). Interestingly, the notably high antioxidant activity of BSG phenolic extract is similar to that of butylated hydroxyanisole and higher than that of butylated hydroxytoluene (Barbosa-Pereira et al., 2014).

The main compounds of BSG protein are hordein, glutelin, globulin, and albumin (Wen et al., 2019). It is reported that BSG noticeably has a similar number

TABLE 1 Biological activities of brewers' spent grain (BSG)

Compounds	Biological activity	References
Phenolic compounds	1. Antioxidant activities	(Barbosa-Pereira et al., 2014; Crowley et al., 2017; Kumari et al., 2019; McCarthy et al., 2014; Moreira et al., 2013; Socaci et al., 2018; Verni et al., 2020)
	Total Phenolic Content (TPC), di(phenyl)-(2;4;6-trinitrophenyl)iminoazanium (DPPH), ferric-reducing antioxidant power (FRAP), 2,2'-Azinobis-(3-Ethylbenzthiazolin-6-Sulfonic Acid) (ABTS)	
	Superoxide dismutase (SOD), catalase (CAT), Glutathione (GSH)	
	Lipid peroxidation	
	Deoxyribose scavenging activity	
	2. Antimicrobial activities	(Barbosa-Pereira et al., 2014; Ibarruri et al., 2019; Kumari et al., 2019; Socaci et al., 2018)
	Antibacterial against gram-positive and negative	
	Antifungal	
	3. DNA-protective and antimutagenic activities	(Crowley et al., 2017; McCarthy et al., 2012; Socaci et al., 2018)
	4. Immunomodulatory effects	(Kumari et al., 2019; McCarthy et al., 2013)
Protein and isolate	1. Antioxidant activities	(Kitrytė et al., 2015; McCarthy et al., 2013; Verni et al., 2020)
	TPC, DPPH, FRAP, ABTS, Oxygen Radical Absorbance Capacity (ORAC)	
	SOD and comet assay	
	Lipid peroxidation	
	2. Anti-inflammatory	(Cian et al., 2018; McCarthy et al., 2013)
	3. Immunomodulatory	(Crowley et al., 2017; McCarthy et al., 2013)
	4. Angiotensin-converting enzyme inhibitory	(Connolly et al., 2015)
	5. Modulation of glycemic response	(Connolly et al., 2017)
	6. Antithrombotic and blood coagulation	(Cian et al., 2018)
	7. Dipeptidyl peptidase IV inhibitory	(Cermeño et al., 2019)
	8. Protective ability against oxidative stress	(E. F. Vieira et al., 2017)
Fatty acids	Antioxidant DPPH and TPC	(Parekh et al., 2017)
Crude dietary fiber (DF)	Antioxidant DPPH, hydroxyl radical, and superoxide radical	(Fu et al., 2011)
SDF	1. Enzymatic antioxidant activities (SOD, CAT, and glutathione peroxidase)	(Zhang et al., 2018)
	2. Maintain intestinal cholesterol efflux	
ß-glucan	Maintains colon health	(Zielke et al., 2017)
Lignin	Maintain gut microbiota	(Ohra-aho et al., 2016)
Arabinoxylans (AXs)	Prebiotic potential	(Reis et al., 2014)
Melanoidins	1. Antioxidant (TPC, DPPH, FRAP, TBRAS)	(Patrignani & González-Forte, 2020; Piggott et al., 2014)
	2. Metal-chelating	

of amino acids as barley (Cermeño et al., 2019). The most abundant amino acids identified included glutamine, proline, and leucine, whereas the lowest were cysteine, methionine, lysine, and threonine (Connolly et al., 2013). Hordein accounts for 60% of the total protein and is recognized for its high glutamine content (Cian et al., 2018). The protein of BSG has low solubility at pH 6 and reaches the maximum solubility at pH 8 to 9 (E. Vieira et al., 2016). Therefore, the degree of solubility impacts the yield and the quality of the protein, in addition to the amino acids (Cian et al., 2018). However, enzymatic treatment and alkaline extraction methods have been reported to generate a higher solubility of BSG protein, in addition to higher foaming and emulsifying ability (E. Vieira et al., 2016; Wen et al., 2019). Enzymes are also able to decrease the molecular weight of the BSG protein and thus increase its digestibility (Wen et al., 2019). Non-enzymatic protein was observed to be more soluble when initially treated with carbohydrase, suggesting that the protein of BSG is located in aleurone cells that remain stable after mashing and store

the protein (Niemi et al., 2013). Additionally, hydrothermal pretreatment was studied to increase the effectiveness of protein extraction from BSG (Qin et al., 2018). Currently, according to a previous review (Wen et al., 2019), the structural information of BSG protein is needed to be studied in future research.

The lipid content of BSG is a source of volatile compounds, such as propionic, acetic, and butyric acids (Ribau Teixeira et al., 2020), fatty acids including palmitic, linoleic, oleic, and stearic acid (Fărcaş et al., 2015; Parekh et al., 2017; Patel et al., 2018; Tan et al., 2019), and tocotrienols (Bohnsack et al., 2011). Triglyceride was discovered as the most abundant in BSG lipid, accounting for about 55% to 67% followed by free fatty acids, accounting for about 18% to 30% (Fărcaş et al., 2015). Lipid from BSG is a valuable source for phytochemical production (del Río et al., 2013).

BSG provides 51% to 53% of total DF and approximately 58.2% and 1.3% of insoluble DF (IDF) and SDF, respectively (Nocente et al., 2019). The major components of IDF from BSG include cellulose 28.7%, hemicellulose 17.5%, and lignin 16.9% per 100 g BSG (Sibhatu et al., 2021). Arabinoxylans (AXs) are a major component of hemicellulose, consisting of arabinose and xylose. The ratio of arabinose:xylose (Arb:Xyl) impacts the extractability of AXs and their functional properties (Severini et al., 2015). ß-glucan is the major part of SDF from BSG, accounting for 4.7% to 13.4% of the total carbohydrates (Verni et al., 2019; E. Vieira et al., 2014).

3 | SPENT GRAIN PROPERTIES IN FOOD PRODUCTS AND PROCESSING

The impacts of the addition of BSG to food products or ingredients are shown in Table 2. In general, BSG improves the nutritional value of the end products. However, it also changes the processing technological, mechanical, and physicochemical properties, including visual appearances such as color and compact texture, in addition to acceptability. The impact of BSG on food production and the quality of final food products is discussed below.

3.1 | BSG impact on technological processing and mechanical properties

3.1.1 | Dough and bread making

A composite generated from a combination of three different byproducts—amaranth, BSG, and apple pomace—was noted to be a good source of energy due to the high amount of carbohydrates and fulfillment of required micronutrients (Awolu et al., 2016). From the rheological properties of the generated composites, such as pasting and functional properties, it is possible to obtain a desired composite mix for food production purposes (Awolu et al., 2016). However, the impact on food products varies depending on the types of food.

The impact of BSG on bread making can be identified from the first step of the process until the final product is obtained. Mixing to homogenize ingredients and allowing interaction with water promote hydration. However, the addition of BSG increases the amount of DF, which contains high hydroxyl groups, thus leading to an increase in the ability to absorb water. As a result, the water availability in the dough formation is diminished, disrupting the network formation and gelatinized starch (Ktenioudaki, O'Shea, et al., 2013; Magabane, 2017; Roth, Döring, et al., 2016; Steinmacher et al., 2012; Torbica et al., 2019; Waters et al., 2012). This phenomenon directly affects several factors as described below.

Mixing BSG into the ingredients intensifies the solidity and reduces the tendency to flow. It is shown by the storage modulus (*G'*) and loss modulus (*G"*), which are determined by frequency sweeps. The increase in both *G'* and *G"* has been reported due to the addition of BSG, showing a change in structural properties of the dough and describing a lower tendency to flow from elastic-like to solid-like behavior (Ktenioudaki, O'Shea, et al., 2013). Furthermore, the addition of BSG increases the mixing tolerance index and mechanical stress needed (Steinmacher et al., 2012; Waters et al., 2012). A decrease in Tan ð was also observed as an indicator of increasing dough elasticity (Ktenioudaki, O'Shea, et al., 2013).

The change in rheological properties of dough as an impact of BSG substitution also has been observed with Mixolab analysis, which showed a disruption of gluten network and protein stability. These properties include: Maximum consistency (C1) and minimum consistency (C2), which describe the weakening of protein based on mechanical work and temperature; starch gelatinization (C3); stability of starch gel formed (C4); and starch retrogradation (C5) (Ktenioudaki et al., 2015; Magabane, 2017; Torbica et al., 2019). Addition of BSG increases water adsorption (WA) in order to achieve C1, thus increasing the water holding capacity (WHC; Aprodu et al., 2017). The increase in WA occurs due to the presence of gluten, whereas the increase in WHC is due to the presence of DF (Waters et al., 2012). The rise in the amount of BSG in the dough increases the level of C2, C3, and C3-C2, which describes the gelatinization range. BSG has a low starch content combined with a high fiber content, thus lowering the gelatinization of the starch (Ktenioudaki, O'Shea, et al., 2013). The rise in C2 and C3 indicates weakened gluten properties in the dough (Aprodu et al., 2017; Ktenioudaki et al., 2015; Magabane, 2017). However, an

TABLE 2 Incorporation of spent grain in food ingredients

Food products/treatment	Main finding
Dough and bread making	
BSG 5%–25% in wheat bread making (Haruna et al., 2011)	The changes in physical properties are reported; the nutritional properties such as fat, mineral, crude fiber, and protein are increased but the hedonic sensory results for aroma, color, taste, texture, and overall acceptability are decreased
BSG (5%–20%) addition in bread making (Fărcaş et al., 2015)	BSG increases the nutritional value such as DF, protein, fat, and mineral content. Maximum addition is 10% with the attribution of volatile compounds
The impact of BSG 10%–20% on aroma and sensory properties (Roth, Schuster, et al., 2016)	BSG addition increases the crumb firmness, sour flavor, off odor, and off taste, and decreases the overall acceptability including look and texture, mouthfeel, odor, and taste
The addition of 10%–20% on thermomechanical properties in sourdough bread making (Aprodu et al., 2017)	The increase in BSG increases water absorption, dough development time (DDT), starch gelatinization, and crumb hardness; decreases the stability of starch gel, starch retrogradation, cooking setback, and specific volume of the bread
BSG combined with corn grifts (45:55) for bread making (Torbica et al., 2019)	The addition of byproducts increases the DF and lowers the sensory acceptability
Enzymatic treatment of BSG for bread making (Steinmacher et al., 2012)	The enzymatic treatment increases the water-soluble AXs. BSG increases the water absorption, mix tolerances index, and DDT, and decreases the time stability
BSG (15%–20%) addition for wheat bread making with technological process modification (Magabane, 2017)	Particle-size reduction, protein enrichment on BSG, sponge and dough, dough sheeting, flour pregelatinization, and sourdough fermentation are involved in order to improve the quality of the bread
Addition of BSG with fermentation as a pretreatment (Gmoser et al., 2020)	Shows promising outcomes due to the enrichment of nutrition and improves the textural properties
Addition of fermented BSG 5%–20% in bread making (Waters et al., 2012)	Increase in BSG increases water absorption, hardness, and mechanical stress and decreases DDT and specific volume
Bioprocessing techniques (sourdough fermentation and technological aids) for improving bread quality containing 15% BSG (Ktenioudaki et al., 2015)	Bioprocessing techniques improve the technological quality of the BSG bread. Sensory properties are lower, compared to commercial bread
Incorporation of distillers' spent grain (DSG ; 5%–30%) in cornbread making (Liu et al., 2011)	The texture of the bread significantly changes; 30% addition of DSG shows a decline in textural quality and batter elasticity
The impact of 5%–20% unground distillers' grains on the wheat dough and bread quality (Roth, Döring, et al., 2016)	Increase in water absorption, dough softening, and bread firmness and decrease in loaf volume, stability time, maximum dough height, maximum gaseous release, and springiness
Combination of BSG 15%–35% and apple pomace on dough rheological properties (Ktenioudaki, O'Shea, et al., 2013)	The changes in the structural properties are identified by the rise of biaxial extensional viscosity and storage modulus and the decreasing of strain hardening index and uniaxial extensibility
Extruded products	
Extrusion of BSG 5%–15% with maize flour (Jozinović et al., 2015)	BSG increases soluble DF (SDF), insoluble DF, resistant starch, water solubility index, and water absorption index. Extrusion damages the starch and increases the SDF
Addition of BSG 5%–15% and modification of treatment: Barrel temperature and screw speed (Sobukola et al., 2013)	Maximum level addition is 9.58% with a temperature of 110°C and 121.47 rpm screw speed
The addition of 10%–40% BSG to extruded snack for bioactivity (Reis & Abu-Ghannam, 2014)	Increases the phenolic content four-fold, antioxidant activity (DPPH and FRAP), and AXs content and decreases the glycemic index

(Continues)

TABLE 2 (Continued)

Food products/treatment	Main finding
BSG addition of 30% to extruded cereal foods (Żelaziński et al., 2018)	The addition of BSG > 30% is possible in appropriate processing conditions but lowers the expansion, increases the hardness, and darkens the color.
Addition of 10%–30% BSG to the extrusion for batters' production (Żelaziński et al., 2017)	The increase in BSG increases the hardness of the final product and apparent density of extrudates and reduces grindability
Addition of 5%–15% BSG, beet pulp, and apple pomace with pectin to improve the quality of corn snacks (Ačkar et al., 2018)	Byproduct addition decreases the expansion ratio and fracturability and increases the bulk density and hardness. Changes in color, water absorption, and water solubility index are also observed
Combination of six malt types of BSG as an extrusion material (Ivanova et al., 2017)	The optimization of the mixture of BSG is obtained based on three major parameters: Total phenolic content and antioxidant activity of DPPH and FRAP
Combination of BSG, whey protein isolate (WPI), barley starch, and waxy corn starch (Kirjoranta et al., 2016)	Snacks containing barley and BSG have lower expansion and higher hardness and the addition of WPI enhances the expansion
Combination of BSG 15%–30% with rice to produce snacks with a variability of temperature (Nascimento et al., 2017)	Increased BSG increases the DF and apparent density but has a negative impact on the volumetric expansion index and reduces specific mechanical energy
Fermentation of BSG and utilization in cereal extrudates (Thorvaldsson, 2020)	The addition of 30% fermented BSG to extruded cereals results in a pleasant taste and texture
Mixing of rice and BSG in extruded flour (Nascimento et al., 2017)	The flow behavior characteristic is influenced by BSG amount and processing temperatures
Valorization of spent grain for extruded snack development (Woomer, 2018)	Spent grain decreases the expansion level and porosity of extruded products; increases hardness, bulk density, and dark intensity.
Cookies, baked snack, biscuits, crackers	
Incorporation of BSG 10%–25% in baked snacks (Ktenioudaki, Crofton, et al., 2013)	10% addition of BSG results in a high crispiness index, low crispiness work, and a positive impact on the crispiness of the snacks, and lowers the quality of texture and crumbliness of the structure
Use of BSG in cookie making combined with wheat and sweet potato flour (Okpala & Ofoedu, 2018)	BSG addition reduces the bulk density and water absorption capacity and increases the emulsion capacity, oil absorption, and protein, fiber, and ash content. Maximum acceptance on 3%–6% addition
Cookie production using 3%–15% of BSG (Ajanaku et al., 2011)	Sensory evaluation shows that 6% is the maximum acceptability, and spread ratio test shows that the maximum formulation is 3%
Cookie production using 15%–50% of BSG (Petrovic et al., 2017)	25% addition of BSG generates the best sensory characteristics of cookies for their appearance, hardness, grittiness, and flavor
Elaboration of BSG 10%–30% in cookie making as a substitution for wheat flour (Heredia-Sandoval et al., 2020)	Increase in BSG increases the protein content and bioactive compounds, and it is suggested that 20% substitution is the best in terms of hydrolysis and glycemic index
Combination of BSG 10%–30% with cocoyam in biscuit making with a different level of mixing and baking times (Odeseye et al., 2020)	Nutritional composition and spread ratio are significantly increased
Combination of BSG 20%–40% with sprouted pigeon pea and unripe plantain in cracker production (Uchegbu, 2016)	The addition of BSG does not change the taste and reduces the overall acceptability; crispiness, color, and texture are also reduced but still in an acceptable range
Combination of BSG 20%–40% with sprouted pigeon pea and unripe plantain in cracker production (Uchegbu & Ishiwu, 2016)	It is observed that the crackers produced has a significant effect on hypoglycemic condition in diabetic rats
Others (pasta, composite flour, yogurt, cheese)	

(Continues)

TABLE 2 (Continued)

Food products/treatment	Main finding
Additional level of BSG (5%–20%) to improve semolina dry pasta (Nocente et al., 2019)	Improvement of nutritional properties with a slight decrease in sensory characteristics, firmness, yellow index, and optimal cooking time
Addition of BSG 3%–25% combined with the addition of egg for structural improvement (Cappa & Alamprese, 2017)	BSG addition significantly lowers the average breaking strain, which is improved by egg addition
Composite flour production by combining wheat, amaranth seed, BSG, and apple pomace (Awolu et al., 2016)	The best formulation is obtained based on proximate analysis and minerals for further analysis of antioxidant, functional, and rheological properties
Substitution of cow's milk by 2%–10% BSG in yogurt making (Abd EL-Moneim et al., 2015)	Significant changes in the chemical, microbial, and sensory evaluation are obtained by increasing BSG.
Incorporation of BSG 10%–50% in the production of cheese blocks (Abd El-Moneim et al., 2018)	50% addition has the highest value of several properties including sensory evaluation, whereas it has the lowest titratable acidity and oil separation

interesting phenomenon was reported that BSG addition had no effect on the Mixolab analysis, including C2 and C3, in addition to C4 and C5, in the dough after extrusion with corn. It is suggested that because of the high protein content it noticeably has similar properties to that of barley protein (Torbica et al., 2019). Furthermore, IDF dilutes the dough gluten and consequently expands the C2 level (Ktenioudaki et al., 2015). As previously mentioned, BSG increase in C4 and C5 is due to the low level of free water availability which, in turn, occurs because free water is bound to the hydroxyl groups in DF of BSG (Ktenioudaki et al., 2015; Magabane, 2017) and as a result of the initially applied torque during mixing (Ktenioudaki et al., 2015). However, a contradiction was reported in C4 and C5, which decreased with the addition of BSG because of the weakened effect of the fiber from BSG (Aprodu et al., 2017). Furthermore, it was noted that the addition of BSG generally reduced pasting properties, including peak viscosity, holding strength, breakdown, final viscosity, and setback (Ktenioudaki, O'Shea, et al., 2013).

A different phenomenon in dough development time (DDT) and dough stability (DS) due to the addition of BSG has been reported. It is noticeably dependent on DF composition, molecular weight, and solubility (Aprodu et al., 2017). The addition of BSG simultaneously increased DDT and DS (Ktenioudaki, O'Shea, et al., 2013). In contrast, an excessive addition of BSG (20%) in bread making decreases DDT, and the maximum DDT was obtained with the addition of 15% BSG (Aprodu et al., 2017). A contrasting result in DS was also observed in which it decreased, while DDT increased due to the addition of BSG (Magabane, 2017; Steinmacher et al., 2012; Torbica et al., 2019). Sponge and dough treatment raises DS but lowers the extensibility and prevents the weakening effect of BSG on dough viscoelastic properties by restoring the dough strength (Magabane, 2017).

Specific loaf volume of bread is most commonly found to be diminished due to the presence of BSG in bread making (Aprodu et al., 2017; Haruna et al., 2011; Magabane, 2017; Steinmacher et al., 2012; Torbica et al., 2019; Waters et al., 2012). A further impact of the addition of BSG is an increase in the WHC and disruption of the network formation of gluten and gelatinized starch (Magabane, 2017; Steinmacher et al., 2012; Torbica et al., 2019; Waters et al., 2012). This phenomenon inhibits the production of Carbondioxide (CO_2) during fermentation of the bread and leads to reduced loaf volume (Roth, Döring, et al., 2016). Moreover, the composition and properties of AXs in BSG impact the volume formation of the bread. Compared to wheat bran and endosperm, the ratio of Arb:Xyl in BSG is very low (Aprodu et al., 2017). High loaf volumes can be observed by the values of biaxial existential viscosity, which are inversely proportional to each other. The substitution of BSG increases the biaxial existential viscosity, thus decreasing the loaf volume (Ktenioudaki, O'Shea, et al., 2013).

3.1.2 | Extrusion process

There are several factors that are usually evaluated in the extrusion process, namely, temperature, moisture content, specific mechanical energy (SME), and velocity, in addition to screw configuration, such as the speed and type of the screw (Reis & Abu-Ghannam, 2014; Sobukola et al., 2013; Woomer, 2018). These processing technologies change the properties of the ingredients and their interaction due to the exposure to thermal and mechanical treatment. A lower moisture content increases the SME and radial expansion ratio and decreases the hardness; higher screw speed increases the SME and the hardness level, and decreases the radial expansion ratio; a higher temperature

raises the radial expansion and decreases the bulk density (Woomer, 2018). The involvement of BSG in the extrusion process requires moisture content below 20% due to the impact on radial expansion and hardness of the final product (Woomer, 2018). However, moisture content of 65% was studied in the reactive extrusion of BSG as an ingredient in bread making (Steinmacher et al., 2012).

The application of extrusion in the processing of BSG as a food ingredient can be categorized into three different types of food production including extrusion of BSG as texturizing agent (Żelaziński et al., 2017), namely, food products such as snacks and breakfast cereals (Ačkar et al., 2018; Kirjoranta et al., 2016; Nascimento et al., 2017; Reis & Abu-Ghannam, 2014; Thorvaldsson, 2020; Woomer, 2018; Żelaziński et al., 2018), bread (Steinmacher et al., 2012; Torbica et al., 2019), and pasta (Sobukola et al., 2013), in addition to BSG alone as a food ingredient (Ivanova et al., 2017).

Extrusion reduces IDF and increases SD (Jozinović et al., 2015), and increases protein availability, minerals, and fat (Reis & Abu-Ghannam, 2014; Sobukola et al., 2013). The recovery of chemical compounds and bioactive compounds of BSG in food extrudates was discovered to be lowered due to the polymerization between polysaccharides and other constituents, as an impact of thermal exposure (Reis & Abu-Ghannam, 2014). However, a composition design was developed to optimize the bioavailability of bioactive compounds of BSG in extrudate products (Ivanova et al., 2017).

As previously stated, DF is abundant in BSG and contains hydroxyl groups that have a high ability to bind water. Thus, DF affects the ability of extruded products to absorb water, their water solubility, and their rheological properties, such as density, elasticity, and viscosity, in addition to volume expansion. Extrusion damages the starch granules, thus more water is bound and the volume of soluble materials is increased (Sobukola et al., 2013). Therefore, the addition of BSG to extrudate products increases the water absorption index and water solubility index (WSI; Ačkar et al., 2018; Jozinović et al., 2015; Sobukola et al., 2013; Żelaziński et al., 2018). Moreover, the presence of protein in BSG, and its lower moisture content, contributes to a higher WSI in extrudate products because low moisture content induces the resisting forces in the matrix and leads to the mechanical degradation of the starch (Żelaziński et al., 2018).

There is a variety of different levels of temperature during the extrusion of BSG from 100 to 170°C. An increase in extrusion temperature contributes to the gelatinization and strengthening of the structure, which consequently leads to an increase in bulk density (Sobukola et al., 2013). However, BSG utilization in extrusion diminishes the bulk density (Sobukola et al., 2013) and increases the apparent density of the extrudate (Nascimento et al., 2017; Żelaziński et al., 2017). Furthermore, the addition of BSG influences the flow behavior of the extrudates because it enhances the viscosity (Nascimento et al., 2017).

The reduction in the expansion index has been observed due to the addition of BSG in the extrudate processing (Ačkar et al., 2018; Kirjoranta et al., 2016; Nascimento et al., 2017; Sobukola et al., 2013; Żelaziński et al., 2018). A higher amount of DF in BSG is reported to be responsible for this phenomenon (Ačkar et al., 2018). It has been noted that fiber binds water more strongly than does starch, reduces water loss, and consequently decreases product expansion (Ačkar et al., 2018). The extrusion ratio of BSG:corn (45:55) as a substitution ingredient in bread making was observed to have no impact on DDT (Torbica et al., 2019), taking into account that a treatment can be developed to improve the properties of BSG in the food matrix.

However, extrusion processes can be modified in appropriate ways, such as screw speed and temperature, to generate high levels of expansion in the products. Increasing the temperature in the extrusion process can improve expansion (Żelaziński et al., 2018), whereas a contradictory result has also been obtained (Ačkar et al., 2018). Noticeably, IDF can break the walls formed during mixing, releasing the entrapped air, and promoting expansion (Kirjoranta et al., 2016). Hence, the transformation of IDF into SDF appears to be a promising means of improving BSG-enhanced extrudate products.

3.1.3 | Cookies, baked snacks, and crackers

A negative effect of the incorporation of BSG in biscuits is a reduction in the spread ratio, which results from the increase in dough viscosity. However, baking time can enhance the spread ratio due to mechanical effects during the mixing process, which allow the formation of a protein network (Odeseye et al., 2020). In cookie making, a contradictory result was obtained in terms of the effect of BSG substitution on the spread ratio of the cookies (Ajanaku et al., 2011; Heredia-Sandoval et al., 2020). It appears that this phenomenon is dependent on the amount of BSG added. A lower quantity of added BSG increases the diameter, whereas the diameter is reduced with a greater addition (Uchegbu, 2016). The same phenomenon was also investigated for thickness and width. Additionally, BSG also reduces the bulk density and water absorption in cookie production and increases emulsion and oil absorption capacities (Okpala & Ofoedu, 2018), DS, and DDT (Heredia-Sandoval et al., 2020).

TABLE 3 The improvement of nutritional value due to the addition of BSG in food products

Nutritional improvement	Food products	References
Fat, mineral, protein and DF	Bread	(Fărcaş et al., 2015; Haruna et al., 2011; Ktenioudaki et al., 2015; Liu et al., 2011; Magabane, 2017; Torbica et al., 2019)
	Extruded products	(Reis & Abu-Ghannam, 2014; Sobukola et al., 2013)
	Baked snacks, biscuits, and crackers	(Heredia-Sandoval et al., 2020; Ktenioudaki, Crofton, et al., 2013; Odeseye et al., 2020; Okpala & Ofoedu, 2018; Petrovic et al., 2017)
	Pasta	(Cappa & Alamprese, 2017; Nocente et al., 2019)
	Yogurts	(Abd EL-Moneim et al., 2015)
Antioxidant and phenolic compounds	Bread	(Aprodu et al., 2017)
	Extruded products	(Reis & Abu-Ghannam, 2014; Sobukola et al., 2013)
	Yogurt	(Abd EL-Moneim et al., 2015)
Antihyperglycemic	Cookies	(Uchegbu & Ishiwu, 2016)
Decrease hydrolysis and glycemic indexes	Cookies	(Heredia-Sandoval et al., 2020)
ß-glucan and resistant starch	Pasta	(Cappa & Alamprese, 2017; Nocente et al., 2019)

3.1.4 | Pasta

Based on a modeling study, BSG has a strong impact on pasta processing, both before and after cooking (Cappa & Alamprese, 2017). BSG weakens the protein functionality and increases the matter loss due to an excess swelling of starch granules during cooking. This prevents the recovery of rolling deformation, and a lower elasticity (breaking strain) is obtained (Cappa & Alamprese, 2017). Cooking time was found to be reduced with the substitution of BSG due to the poor gluten matrix formed as a result of the decline in water absorption (Nocente et al., 2019). An increment in the diameter was predicted due to the rough surface that results from large particles of BSG (Nocente et al., 2019). In contrast, a decrease in diameter/thickness was also reported (Cappa & Alamprese, 2017). The possibility of obtaining desirable pasta with the inclusion of BSG was suggested, considering minimal matter loss, maximum breaking strain, and minimum Young's modulus (Cappa & Alamprese, 2017).

3.1.5 | **Dairy products: Cheese** blocks and yogurt

The impact of BSG on yogurt processing in terms of techno-mechanical properties is not reported. However, BSG significantly affects the rheological behavior of cheese blocks (Abd El-Moneim et al., 2018). As mentioned previously, BSG disrupts network formation and consequently results in a more compact texture. Therefore, it increases hardness, adhesiveness, cohesiveness, gumminess, and chewiness (Abd El-Moneim et al., 2018). Moreover, an increase in meltability and a decrease in oil separation have been observed (Abd El-Moneim et al., 2018).

3.2 | Impact of BSGs on nutritional value and chemical composition

The enhancement of the quality of food products due to the addition of BSG is shown in Table 3. It was mentioned previously that BSG enhances nutritional value and can potentially be used as a food ingredient and for nutraceutical purposes. Thus, the addition of BSG to food products improves health benefits due to changes in chemical composition and biological activity. Exceptionally, the addition of BSG in cheese making lowered protein content and had no impact on fat and ash content (Abd El-Moneim et al., 2018); the combination of BSG with other byproducts has been reported for lowering protein and fat content in cookies (Uchegbu & Ishiwu, 2016).

The ability of BSG to increase the pH value and lower the titratable acidity in cheese blocks has been observed (Abd El-Moneim et al., 2018). It has been found that processing treatments can be modified to improve the nutritional aspects of BSG in food production, for example, fermentation. Fermentation of sourdough in bread making has been found to enhance the antioxidant activity of bread (Aprodu et al., 2017), and solid-state fermentation and a reduction in particle size have been observed to increase bread's protein content (Gmoser et al., 2020; Magabane, 2017). Fermentation of BSG is observed to be able to increase the solubility of cell walls (Magabane, 2017), which might increase the bioavailability of several bioactive compounds.

TABLE 4 Maximum addition of spent grain in food products regarding the sensory evaluation

Food products	Maximum addition (%)
Bread (Fărcaş et al., 2015)	10
Cookies (Petrovic et al., 2017)	25
Pasta (Nocente et al., 2019)	10
Yogurt (Abd EL-Moneim et al., 2015)	6
Cheese (Abd El-Moneim et al., 2018)	50

3.3 | Impact of BSGs on sensory profile and acceptability

The sensory acceptability of BSG-enhanced food products varies depending on the specific processing involved and ingredients used. The maximum accepted levels of substitution of BSG in food are shown in Table 4. In general, the addition of BSG in small amounts yields a positive impact on food acceptability, whereas higher additions decrease the desirability. In contrast, a higher level of BSG incorporation in cheese blocks generates a high acceptability value (Abd El-Moneim et al., 2018).

3.3.1 | Bread products

The acceptability of bread enhanced by BSG has been studied using a hedonic scale (Fărcaş et al., 2015; Haruna et al., 2011) and rank tests (Steinmacher et al., 2012; Torbica et al., 2019) involving untrained panels and a maximum addition of BSG to bread varying from 10% to 15%. The addition of 10% BSG generated a higher acceptability than control bread (Fărcaş et al., 2015), whereas the addition of 15% had lower acceptability compared to the control (Haruna et al., 2011) and commercial bread (Ktenioudaki et al., 2015). Trained panelists defined the main difference between wheat bread and BSG-enhanced bread as the aroma profile of the produced bread: BSG induced the response of a single aroma and overall aroma impression by panelists (Roth, Schuster, et al., 2016). Fermentation of BSG produced lactic acid and generated bread with an acidulous aroma (Waters et al., 2012). As a consequence, participants perceived a sour smell and taste due to a 10% addition of DSG, in addition to an off-odor and off-flavor (Roth, Schuster, et al., 2016). However, the panelists were unable to define this impact. The acidulous aroma might be generated from the production of organic acids during fermentation (Roth, Schuster, et al., 2016).

As shown in Table 4, the maximum addition accepted by panels was 10%; the addition of 15% to 20% was a critical factor for the generation of a pleasant aroma (Roth, Schuster, et al., 2016; Waters et al., 2012). The factors with the greatest influence in the acceptability of BSG-enhanced bread are taste and texture (Fărcaş et al., 2015). BSG has been identified for its bitterness due to polyphenolic compounds, which mask the sweetness of bread (Waters et al., 2012). Furthermore, a slightly finer distribution of pores and denser crumb structure for bread with higher DSG has been expressed (Roth, Döring, et al., 2016).

3.3.2 | Extrusion

Extruded BSG has been applied in several food types, with the acceptability of extruded products varying depending on the food type studied. Extrusion of BSG was found to enhance the acceptability of bread to the same level as that of wheat bread (Torbica et al., 2019). A contrasting study found there was no significant difference in the acceptability of BSG-enhanced bread with and without extrusion because the mix of ingredients has a significant impact on the extrusion products (Steinmacher et al., 2012). Extrusion for breakfast cereal-like production was developed using fermented and non-fermented BSG. It was noted that panelists preferred the commercial breakfast cereal due to the appearance of the final product. However, a comparison of all of the treatments concluded that 30% fermented BSG was preferred by the panelists because of the crunchiness, rich scent, and taste (Thorvaldsson, 2020). BSG in extruded products had a negative impact on smell and aftertaste (Ačkar et al., 2018). This might be a reason for the minimum addition of BSG in extrusion products. The sensory score was reported to be significantly related to the volume expansion: The greater the volume expansion, the higher the acceptability (Ačkar et al., 2018). Hence, improving the physical properties of BSG-enhanced products is important to improve their acceptability.

3.3.3 | Cookies

The most influential factors in general acceptability were found to be taste and texture (Ktenioudaki, Crofton, et al., 2013) because the incorporation of BSG in cookie production induced a bitter taste and brittle texture (Heredia-Sandoval et al., 2020). According to the overall acceptability score, the maximum substitution that was able to be tolerated by panelists was found to be 6% (Ajanaku et al., 2011; Okpala & Ofoedu, 2018) and 10% (Ktenioudaki, Crofton, et al., 2013). Interestingly, the addition of 20% BSG in cookies was still accepted by panelists. This might be because of the inclusion of other byproducts, in addition to the properties of BSG itself, thus increasing the acceptability (Heredia-Sandoval et al., 2020; Petrovic et al.,

2017; Uchegbu, 2016). The impact of BSG on the aroma of food products has been investigated by a discriminatory test, which reported that BSG intensified the aroma (Ktenioudaki, Crofton, et al., 2013). BSG has been reported to contain specific odor compounds, such as 2-butyl-1-octanol, 3-methyl-butanal, 2-heptane, butanal, benzene, and 2,3-butanedione, which clearly affect the aroma of products (Ktenioudaki, Crofton, et al., 2013). It is notable that odor-active compounds are the result of the breakdown of proteins, fat, and sugars. Additionally, a compact texture hinders the release of odor compounds; thus, a large addition of BSG has a strong impact on the aroma (Ktenioudaki, Crofton, et al., 2013).

3.3.4 | Pasta

The addition of BSG to pasta affects its acceptability, which is described through BSG's impact on color and stickiness, and general hedonic acceptability. From the perspective of processing technology, a 6.2% addition of BSG yields high acceptability of the pasta (Cappa & Alamprese, 2017). However, from the perspective of panelists, a 10% addition of BSG during pasta making was acceptable for its sensory evaluation, including physical appearance and hedonic acceptability (Nocente et al., 2019). It was noted that the presence of fiber in BSG increased the stickiness of pasta products (Nocente et al., 2019).

3.3.5 | Dairy-based products

A 50% substitution of BSG in cheese blocks received the highest score for acceptability due to its desirability in terms of color, flavor, texture, and overall acceptability (Abd El-Moneim et al., 2018). BSG has a uniquely pleasant aroma, and a predominantly brown color, which might contribute to higher scores in sensory evaluation. However, in yogurt products, the maximum addition of BSG that was found to be acceptable was only 6% (Abd EL-Moneim et al., 2015).

3.4 | Impact of BSG on product color

Color is an important parameter in food products. BSG has a dark brown color, which significantly darkens food products such as bread (Liu et al., 2011; Waters et al., 2012), extrusion products (Ačkar et al., 2018; Thorvaldsson, 2020; Żelaziński et al., 2018), cookies (Ajanaku et al., 2011; Heredia-Sandoval et al., 2020; Petrovic et al., 2017), and pasta (Nocente et al., 2019). To a point, the unique dark brown color of BSG has a beneficial impact on the color of the food product, which is assumed to be healthy. No results are available for color analysis of dairy products enhanced with BSG. However, the effect of the color change can be seen in the hedonic evaluation score, which decreased due to the addition of BSG (Abd EL-Moneim et al., 2015; Abd El-Moneim et al., 2018). In addition to the fact that BSG tends to be dark, the Maillard reaction could be another reason for the reduction in brightness (Odeseye et al., 2020; Waters et al., 2012). Temperature contributes to the color change due to the Maillard reaction, caramelization, hydrolysis, and pigment degradation (Ačkar et al., 2018; Żelaziński et al., 2018).

3.5 | BSGs impact on textural properties

Textural properties are a combination of mechanical, geometrical, and surface characteristics of the products (Ačkar et al., 2018). Generally, BSG increases the hardness of bread (Aprodu et al., 2017; Ktenioudaki et al., 2015; Magabane, 2017; Roth, Schuster, et al., 2016; Steinmacher et al., 2012; Waters et al., 2012), extrusion products (Ačkar et al., 2018; Kirjoranta et al., 2016; Thorvaldsson, 2020; Żelaziński et al., 2017, 2018), cookies, baked snacks, crackers and biscuits (Heredia-Sandoval et al., 2020; Ktenioudaki, Crofton, et al., 2013), and cheese blocks (Abd El-Moneim et al., 2018). Other parameters studied regarding the textural properties include elasticity for bread and pasta; fracturability for extrusion products; and crispness for cookies, baked snacks, crackers, and biscuits. In contrast to other products, it has been reported that BSG decreases the texture of the pasta, compared to the control. This phenomenon is due to the reduction in mechanical strength as an impact of interference on gluten network formation (Nocente et al., 2019).

BSG intensifies the hardness of bread products by increasing their firmness and springiness (Aprodu et al., 2017; Ktenioudaki et al., 2015; Magabane, 2017; Roth, Döring, et al., 2016; Steinmacher et al., 2012; Waters et al., 2012). Several factors responsible for this phenomenon have been identified. The amount of arabinoxylans, glucan, and xylo-oligosaccharides in BSG changes the viscoelastic properties of the dough, resulting in a challenge to dough improvement (Aprodu et al., 2017; Waters et al., 2012). This was found to be due to crosslinking gluten and gelatinized starch (Steinmacher et al., 2012). A lower pH also hardened the gluten network and, therefore, the final product (Waters et al., 2012). Furthermore, BSG decreased the crumb cell number and led to a compact texture (Torbica et al., 2019). The same result also reported denser crumbs and smaller pores due to the compact texture of the bread (Roth, Döring, et al., 2016). Compared to other byproducts, BSG had a higher impact on texture

properties than the impact of apple pomace and sugar beet pulp in bread making. BSG addition generated bread that required more chewing and was less crumbly and less cohesive (Torbica et al., 2019). Hence, improving the textural properties in BSG-enhanced bread is a challenge for further study (Gmoser et al., 2020).

Some factors influence the hardness and fracturability of extruded products, such as moisture content, temperature, screw speed, and composition of the materials (Ačkar et al., 2018). However, BSG increases the hardness in two ways: First, DF from BSG interacts with protein, leading to early disruption of gaseous cells and subsequently forming a thicker and a harder wall in the products (Ačkar et al., 2018; Żelaziński et al., 2018). Second, water bound in DF is trapped after rapid cooling, leading to incomplete water evaporation (Żelaziński et al., 2018). Hardness is inversely proportional to the expansion level and fracturability of extrudate products (Ačkar et al., 2018; Kirjoranta et al., 2016). However, a contradiction was reported in terms of fracturability, which was expressed as grindability and found to be proportional to the increase in BSG (Żelaziński et al., 2017). This contradiction can be explained by several factors that impact fracturability as mentioned previously.

A different trend was observed in cookie products, in which, after a gradual increase, hardness was found to drop slightly with the further addition of BSG (Heredia-Sandoval et al., 2020). This phenomenon appeared to be due to the brittle texture that resulted from an excessive amount of BSG, which required less force to rupture and tended to break (Heredia-Sandoval et al., 2020). BSG impacts the texture by reducing the crispness (Ktenioudaki, Crofton, et al., 2013) and breaking the strength of the cookies (Uchegbu, 2016). Moreover, the textural impact of BSG was observed for its effect of forming a closed structure and diminishing gluten development, which were shown by image analysis and peak number analysis, respectively (Ktenioudaki, Crofton, et al., 2013). As mentioned previously, the addition of BSG disrupts the formation of the network and thus generates a compact texture and, finally, hardens the texture.

4 | IMPROVING THE CHARACTERISTICS OF BSG-ENHANCED FOOD PRODUCTS BY FIBER MODIFICATION TREATMENTS

BSG influences all aspects of food, from processing to final product quality, as previously mentioned. Research indicates that only nutritional value is improved, whereas technological processing and mechanical properties, in addition to other physical properties, are negatively impacted. As shown in Table 4, the recommended amount of BSG is low, with the exception of cheese blocks. However, some modifications to processing technology and ingredient composition have been identified for their ability to improve the quality of BSG-enhanced foods. Therefore, the development of new food products with a higher level of added BSG can be expected.

Some methods have been used to improve BSG-enhanced food products, including fermentation of BSG and sourdough, in addition to enzyme treatment in bread making, temperature and screw speed modification in the extrusion process, and incorporation of additional ingredients. Generally, all of the identified techniques modify fiber characteristics, which can change other physical and processing properties, in addition to the quality of the final products. Physical properties of food to which byproducts are added are mainly influenced by the fiber composition of the byproducts (Elleuch et al., 2011) and play an important role in the food's acceptability (Torbica et al., 2019). Fiber composition is responsible for textural properties: IDF enhances rigidness, which increases hardness, whereas SDF controls softness, film formation, thickening, gelling, and foam stabilization (Föste et al., 2020). SDF is more easily mixed into the food matrix than non-SDF. As a result, SDF results in a better texture and does not affect the taste. In contrast, IDF has a low density and affects the food texture (Elleuch et al., 2011).

Sensory scores have been found to be directly related to the expansion level in extrusion products (Ačkar et al., 2018). A compact texture was also found to be negatively responsible for the release of odor compounds (Ktenioudaki, Crofton, et al., 2013) and the brightness level to be negatively related to the hardness of extrusion products (Ačkar et al., 2018; Thorvaldsson, 2020; Żelaziński et al., 2018). Hence, improving the physical properties, which can be achieved by fiber modification, can enhance the acceptability of final products from the perspective of consumers. Extrusion is able to diminish the darkening effect of BSG, which is shown by a* and b* values (Ačkar et al., 2018; Thorvaldsson, 2020; Żelaziński et al., 2018). Temperature and screw speed can modify fiber characteristics due to thermal exposure and mechanical pressure during extrusion (Ačkar et al., 2018; Thorvaldsson, 2020; Żelaziński et al., 2018).

Fermentation has been identified for its ability to increase the solubility of cell walls, which consist of DF (Magabane, 2017). Consequently, the quantity of SDF increases and leads to a change in the consistency of dough (Aprodu et al., 2017; Ktenioudaki et al., 2015; Magabane, 2017; Torbica et al., 2019). Furthermore, fermentation improves the gluten network and thus gas retention. Improving the protein network by sourdough fermentation is a result of proteolytic activity, which

modifies the physical properties of gluten. Therefore, BSG can weaken dough viscoelastic properties (Magabane, 2017). Sourdough fermentation has been observed to increase loaf volume (Aprodu et al., 2017; Magabane, 2017) and gas-holding capacity (Magabane, 2017). Additionally, fermentation has been observed to reduce the brown color of the BSG-added bread due to the reduction in free sugar by microorganisms, leading to a reduction in the Maillard reaction (Waters et al., 2012). Fermentation also reduces firmness and improves the texture quality of BSG-enhanced bread (Ktenioudaki et al., 2015; Magabane, 2017; Waters et al., 2012). The ability of sourdough fermentation to improve the properties of BSG in bread making is affected by the endoxylanase activity in the modification of the crumb hardness of the bread (Aprodu et al., 2017). However, the species of the microorganisms must be taken into account due to the differences in their ability to modify the texture (Gmoser et al., 2020). In general, lactic acid bacteria is used as an inoculum for the fermentation process (Aprodu et al., 2017; Waters et al., 2012). However, yeast and a combination of yeast and lactic acid bacteria have also been employed (Gmoser et al., 2020; Magabane, 2017).

Enzymatic treatment increases the specific volume of BSG-enhanced bread by increasing the solubility of AXs (Ktenioudaki et al., 2015; Steinmacher et al., 2012). Hence, increasing the ratio of Arb:Xyl in BSG and improving the solubility of AXs may help avoid the negative impact of BSG on the loaf volume of bread. Enzyme incorporation in BSG used in bread making has also been reported for its ability to improve textural quality (Steinmacher et al., 2012).

Incorporation of several ingredients appears to be an option in the improvement of food products containing BSG, with an expectation that other ingredients will diminish the negative impacts of BSG. For instance, the inclusion of BSG in cornbread making generated a higher loaf volume (Liu et al., 2011); in the extrusion process, pectin added into BSG extrusion thus generated a higher porosity and expansion level of extruded product (Ačkar et al., 2018). The improvement in the quality of the extruded BSG has been also reported when it is mixed with another ingredient, namely, whey protein (Kirjoranta et al., 2016). The quality of BSG-enhanced pasta has been observed to increase due to the incorporation of egg white powder (Cappa & Alamprese, 2017).

In addition to the fact that the treatments mentioned above are able to modify the properties of BSG in the food matrix, several treatments have been applied and reported to improve BSG properties as can be seen in Table 5. However, those techniques are studied in order to generate certain substances. Because they are able to improve certain properties of BSG, they can be promising methods that can be applied to BSG for food production purposes. Several treatments have been applied to other ABPs to enhance the quality in terms of their application in food production purposes.

Temperature graduation enhances the yield and properties of AXs and arabinoxylan-oligosaccharides (Coelho et al., 2014) and phenolic availability (Budaraju et al., 2018), fermentation improves the availability of phenolic and peptides (Ibarruri et al., 2019; Tišma et al., 2018; Verni et al., 2019, 2020), and hydromechanical treatment is used for protein extraction (Ibbett et al., 2019). The availability of phenolic compounds and peptides from BSG might be related to changes in the composition of DF. Phenolic compounds exist in a hydroxyl group of DF, whereas protein is entrapped in the cell wall of the materials that consist of DF. The transformation of fiber composition consequently impacts the availability of bioactive compounds. Therefore, modifying the fiber of BSG appears to be able to reduce the technical problem that occurs during processing, and thus improve the quality of final food products.

Physical treatment mainly involves high pressure and temperature, such as autoclave treatment, steam explosion, high hydrostatic pressure and high-pressure homogenization, extrusion, and mechanical pretreatment, each of which can be used to enhance fiber functionality (Kieserling et al., 2019; B. Li, Yang, et al., 2019; S. Li, Chen, et al., 2019; Xie et al., 2017; Yan et al., 2019). The chemical treatment enhances the density, water-holding capacity, and mechanical performance depending on the functional group (López Durán et al., 2018).

Fermentation is able to transform the fiber composition, including the amount of SDF, and leads to the improvement of the ratio of SDF:IDF. This is because fermentation can degrade cellulose and hemicellulose, forming a porous and loose structure. As a consequence, it enhances water- and oil-holding capacity, in addition to swelling capacity. Conversely, fermentation consequently decreases quantities of cellulose and hemicellulose, protein, and fat. Transformation of fat into fatty acid is also another benefit, which occurs because materials can bind fat and reduce losses during food processing, in addition to producing a consistent food structure (Chu et al., 2019). *Rhizopus* sp., lactic acid bacteria, and *Bacillus* sp. Are the common groups of microorganisms that have been involved in the fermentation of BSG (Chu et al., 2019; Cooray & Chen, 2018; Ibarruri et al., 2019; Tan et al., 2019; Verni et al., 2020).

High-temperature autoclave treatment and steam explosion methods degrade the molecular structure and lead to an increase in SDF, apparent viscosity, and solubility. Furthermore, they also reduce the high molecular weight. This effect on fiber is beneficial in the food matrix because the small molecular weight increases the antioxidant capacity. In addition, the viscosity and solubility enhance the food

TABLE 5 Treatments applied in enhancing the properties of BSG

Treatment	Purposes	Main finding
Thermal and enzyme treatment (Budaraju et al., 2018)	Availability of phenolic compounds	Treatment increases the free phenolic content.
Steam explosion (Kemppainen et al., 2016)	Improves the enzymatic digestibility of carbohydrate, solubility, and stability of proteins	Oligomeric non-cellulosic glucan and AXs are dissolved, and over a third of the protein is dissolved with partial degradation.
Enzymatic treatment and particle size reduction (Severini et al., 2015)	Enhances the solubility of AXs from BSG	The solubility of AXs increases due to enzyme treatment and particle size reduction.
Alkaline and enzyme treatment (Connolly et al., 2019)	Bioactivity evaluation of the extracts from treated BSG	Direct enzymatic hydrolysis of BSG without alkaline treatment is more beneficial.
Solid-state fermentation (Cooray & Chen, 2018)	Enhancing the nutritive value of BSG	Fermentation improves the nutrient content of BSG can be observed by changes in metabolites amino acids, citric acid, vitamins, and antioxidants.
Microwave superheated water and dilute alkali treatment (Coelho et al., 2014)	Extraction of AXs and arabinoxylo-oligosaccharides from BSG	The differences in extraction conditions will generate specific compounds for different applications and uses.
Wet fractionation (He et al., 2019)	Production of high protein and fiber contents of BSG	The optimal condition can be obtained in order to generate high protein and fiber content ingredients.
Solid-state fermentation (Ibarruri et al., 2019)	Enhancing the nutritive value of BSG	Significant increase in nutrition value is observed; thus, it can be applied in the alternative valorization of BSG.
Hydromechanical processing (Ibbett et al., 2019)	Improving the techno-functional protein extract	Generating rich protein extract with stabilizing properties.
Pulsed electric field treatment (Kumari et al., 2019)	Enhancing the nutritive value, polyphenolic, and bioactivities of light and dark BSG	Increase in antimicrobial, antioxidant, and biological activities.
Pulsed electric field treatment (Martín-García et al., 2020)	Improving the phenolic compounds recovery from SBG	The treatment increases the yield of total free and bound phenolic compounds.
Prehydrolysis with carbohydrases (Niemi et al., 2013)	Improving the release of protein	76% of the protein is solubilized, which is affected by time exposure to the alkaline environment.
Enzyme and ultrasonication on BSG (Yu et al., 2020)	Enhancing the recovery of protein hydrolysates	Ultrasound pretreatment increases the efficiency of protein separation, reduces enzyme loading, and decreases enzyme incubation time.
Comparison of alkaline, acid, enzyme, and hydrothermal treatment on BSG (Qin et al., 2018)	Evaluation based on protein extraction	Hydrothermal pretreatment is a more promising option for protein releasing from BSG also in terms of costs and environmental.
Ultrasound and alkaline treatment on BSG (Reis et al., 2015a)	Enhancing the yield of AXs	The differences between the methods are in the reduction time and energy used. The recovery of AX is the same.
Superfine particle of BSG (Reis et al., 2015b)	Impact of different particle size on the structure of AXs	AXs-rich extract is generated with different degrees of polymerization and degree of branching.
Solid-state fermentation (Tan et al., 2019)	Improving the nutritional profile	Enhancement of metabolites including amino acids, fatty acids, tricarboxylic acid cycle intermediates, and antioxidant activity.
Combination of enzyme and fermentation on BSG (Verni et al., 2020)	Enhancement of antioxidant activity of BSG	Bioprocessed BSG obtains enhanced biological activity; novel antioxidant peptides are observed.

(Continues)

TABLE 5 (Continued)

Treatment	Purposes	Main finding
Recyclable integrated process (E. Vieira et al., 2014)	Enhancing the availability of protein and AXs	A higher amount of protein and AXs recovery during extraction.
Several pretreatment on BSG (Ravindran et al., 2018)	Generating reducing sugar substance	Microwave-assisted alkali is found to be most efficient

structure, which is important for food acceptability and reducing food losses during processing (B. Li, Yang, et al., 2019; S. Li, Chen, et al., 2019).

A microwave-assisted alkaline treatment in fiber production from sugarcane bagasse and sugarcane tops generates fiber that produces the same texture and aroma of bread and pasta controls. This means that this method can be applied to generate high-quality food ingredients from spent grain. The mechanism involves microwave treatment to reduce the crude fiber, followed by an increase in crystallinity by alkaline treatment. As a result, the fiber digestibility increases with the increase in lignin solubility or the weakening of the bond between the lignin and phenolic group. Each step of this combination method has shown a role in generating fiber of higher quality (Gil-López et al., 2019).

These techniques appear to be promising due to their ability to improve the physical, mechanical, and processing technology properties of the fiber. As previously mentioned, the fiber physical properties are related to the quality of final BSG-added food products, such as acceptability and volume expansion, compact texture with odor compounds, and hardness with brightness. Furthermore, an improvement in biological activity, by increasing the availability of bioactive compounds, can be achieved.

5 | CONCLUSION

The incorporation of BSG in food processing is well known for its ability to enhance the biological activity of food products. Regarding the nutritional value of BSG, the potential of BSG as a functional ingredient has been identified. Seven types of food products currently use BSG to improve their nutritional properties, including bread, extrusion products, cookies, pasta, yogurt, and cheese blocks. However, sensory evaluation has shown that the addition of BSG commonly reduces the acceptability of food products, with the exception of cheese. This result is a consequence of the negative impact of BSG on physical properties, such as processing characteristics and the quality of the final products. The substitution of BSG tends to harden the texture, diminish the volume expansion, darken the color, and reduce the elasticity. However, some methods have been applied to increase the quality of BSG-enhanced food products. The potential exists to incorporate larger amounts of BSG in food processing using the proposed techniques via fiber modification, which have been applied in other ABPs to yield positive results in the improvement of fiber functionality. Although challenging, these techniques require further research to improve the quality of BSG and BSG-enhanced final food products.

ACKNOWLEDGMENTS

This work was supported by UPWR 2.0: International and interdisciplinary program of development of Wrocław University of Environmental and Life Sciences co-financed by the European Social Fund under the Operational Programme Knowledge Education Development 2014–2020: Axis III Higher education for the economy and development; Action 3.5. Comprehensive programs for schools of higher education Contract Number POWR.03.05.00-00-Z062/18.

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REFERENCES

Żelaziński, T., Ekielski, A., Siwek, A., & Dardziński, L. (2017). Characterisation of corn extrudates with the addition of brewers' spent grain as a raw material for the production of functional batters. *Acta Scientiarum Polonorum Technologia Alimentaria*, *16*(3), 247–254. https://doi.org/10.17306/J.AFS.0487

Żelaziński, T., Ekielski, A., Siwek, A., & Durczak, K. (2018). By-products from brewery industry as the attractive additives to the extruded cereals food. *Carpathian Journal of Food Science and Technology*, *10*(5), 83–97.

Ačkar, Đ., Jozinović, A., Babić, J., Miličević, B., Panak Balentić, J., & Šubarić, D. (2018). Resolving the problem of poor expansion in corn extrudates enriched with food industry by-products. *Innovative Food Science & Emerging Technologies*, *47*, 517–524. https://doi.org/10.1016/j.ifset.2018.05.004

Abd EL-Moneim, R., Shamsia, S., EL-Deeb, A., & Ziena, H. (2015). Utilization of brewers spent grain (BSG) in making functional yoghurt. *Journal of Food and Dairy Sciences*, *6*(10), 577–589. https://doi.org/10.21608/jfds.2015.50077

Abd El-Moneim, R., Shamsia, S., EL-Deeb, A., & Ziena, H. (2018). Utilization of brewers spent grain (BSG) in producing functional processed cheese 'Block.' *Journal of Food and Dairy Sciences*, *2018*(0), 103–109. https://doi.org/10.21608/jfds.2018.77764

Aggelopoulos, T., Katsieris, K., Bekatorou, A., Pandey, A., Banat, I. M., & Koutinas, A. A. (2014). Solid state fermentation of food waste mixtures for single cell protein, aroma volatiles and fat production. *Food Chemistry*, *145*, 710–716. https://doi.org/10.1016/j.foodchem.2013.07.105

Ajanaku, K. O., Dawodu, F. A., Ajanaku, C. O., & Nwinyi, O. C. (2011). Functional and nutritional properties of spent grain enhanced cookies. *American Journal of Food Technology*, *6*(9), 763–771. https://doi.org/10.3923/ajft.2011.763.771

Aliyu, S., & Bala, M.(2011). Brewer's spent grain: A review of its potentials and applications. *African Journal of Biotechnology*, *10*(3), 324–331.

Aprodu, I., Simion, A. B., & Banu, I. (2017). Valorisation of the brewers' spent grain through sourdough bread making. *International Journal of Food Engineering*, 13(10). https://doi.org/10.1515/ijfe-2017-0195

Awolu, O. O., Osemeke, R. O., & Ifesan, B. O. T. (2016). Antioxidant, functional and rheological properties of optimized composite flour, consisting wheat and amaranth seed, brewers' spent grain and apple pomace. *Journal of Food Science and Technology*, *53*(2), 1151–1163. https://doi.org/10.1007/s13197-015-2121-8

Balogun, A. O., Sotoudehniakarani, F., & McDonald, A. G. (2017). Thermo-kinetic, spectroscopic study of brewer's spent grains and characterisation of their pyrolysis products. *Journal of Analytical and Applied Pyrolysis*, *127*, 8–16. https://doi.org/10.1016/j.jaap.2017.09.009

Barbosa-Pereira, L., Bilbao, A., Vilches, P., Angulo, I., LLuis, J., Fité, B., Paseiro-Losada, P., & Cruz, J. M. (2014). Brewery waste as a potential source of phenolic compounds: Optimisation of the extraction process and evaluation of antioxidant and antimicrobial activities. *Food Chemistry*, *145*, 191–197. https://doi.org/10.1016/j.foodchem.2013.08.033

Bohnsack, C., Ternes, W., Büsing, A., & Drotleff, A. M. (2011). Tocotrienol levels in sieving fraction extracts of brewer's spent grain. *European Food Research and Technology*, *232*(4), 563–573. https://doi.org/10.1007/s00217-010-1419-z

Bonifácio-Lopes, T., Teixeira, J. A., Pintado, M. (2020). Current extraction techniques towards bioactive compounds from brewer's spent grain – A review. *Critical Reviews in Food Science and Nutrition*, *60*(16), 2730–2741. https://doi.org/10.1080/10408398.2019.1655632.

Bora, P., Ragaee, S., & Abdel-Aal, E. -S. M. (2019). Effect of incorporation of goji berry by-product on biochemical, physical and sensory properties of selected bakery products. *LWT – Food Science and Technology*, *112*, 108225. https://doi.org/10.1016/j.lwt.2019.05.123

Budaraju, S., Mallikarjunan, K., Annor, G., Schoenfuss, T., & Raun, R. (2018). Effect of pre-treatments on the antioxidant potential of phenolic extracts from barley malt rootlets. *Food Chemistry*, *266*, 31–37. https://doi.org/10.1016/j.foodchem.2018.05.110

Cappa, C., & Alamprese, C. (2017). Brewer's spent grain valorization in fiber-enriched fresh egg pasta production: Modelling and optimization study. *LWT - Food Science and Technology*, *82*, 464–470. https://doi.org/10.1016/j.lwt.2017.04.068

Carciochi, R., Sologubik, C., Fernández, M., Manrique, G., D'Alessandro, L. (2018). Extraction of Antioxidant Phenolic Compounds from Brewer's Spent Grain: Optimization and Kinetics Modeling. *Antioxidants*, *7*(4), 45https://doi.org/10.3390/antiox7040045.

Cermeño, M., Connolly, A., O'Keeffe, M. B., Flynn, C., Alashi, A. M., Aluko, R. E., FitzGerald, R. J. (2019). Identification of bioactive peptides from brewers' spent grain and contribution of Leu/Ile to bioactive potency. *Journal of Functional Foods*, *60*, 103455 https://doi.org/10.1016/j.jff.2019.103455.

Chu, J., Zhao, H., Lu, Z., Lu, F., Bie, X., & Zhang, C. (2019). Improved physicochemical and functional properties of dietary fiber from millet bran fermented by *Bacillus natto*. *Food Chemistry*, *294*, 79–86. https://doi.org/10.1016/j.foodchem.2019.05.035

Cian, R. E., Garzón, A. G., Martínez-Augustin, O., Botto, C. C., Drago, S. R. (2018). Antithrombotic Activity of Brewers' Spent Grain Peptides and their Effects on Blood Coagulation Pathways. *Plant Foods for Human Nutrition*, *73*(3), 241–246. https://doi.org/10.1007/s11130-018-0682-1.

Coelho, E., Rocha, M. A. M., Saraiva, J. A., & Coimbra, M. A. (2014). Microwave superheated water and dilute alkali extraction of brewers' spent grain arabinoxylans and arabinoxylo-oligosaccharides. *Carbohydrate Polymers*, *99*, 415–422. https://doi.org/10.1016/j.carbpol.2013.09.003

Combest, S., & Warren, C. (2019). Perceptions of college students in consuming whole grain foods made with Brewers' Spent Grain. *Food Science & Nutrition*, *7*(1), 225–237. https://doi.org/10.1002/fsn3.872

Connolly, A., Cermeño, M., Crowley, D., O'Callaghan, Y., O'Brien, N. M., FitzGerald, R. J. (2019). Characterisation of the in vitro bioactive properties of alkaline and enzyme extracted brewers' spent grain protein hydrolysates. *Food Research International*, *121*, 524–532. https://doi.org/10.1016/j.foodres.2018.12.008.

Connolly, A., O'Keeffe, M. B., Nongonierma, A. B., Piggott, C. O., FitzGerald, R. J. (2017). Isolation of peptides from a novel brewers spent grain protein isolate with potential to modulate glycaemic response. *International Journal of Food Science & Technology*, *52*(1), 146–153. https://doi.org/10.1111/ijfs.13260.

Connolly, A., O'Keeffe, M. B., Piggott, C. O., Nongonierma, A. B., FitzGerald, R. J. (2015). Generation and identification of angiotensin converting enzyme (ACE) inhibitory peptides from a brewers' spent grain protein isolate. *Food Chemistry*, *176*, 64–71. https://doi.org/10.1016/j.foodchem.2014.12.027.

Connolly, A., Piggott, C. O., FitzGerald, R. J. (2013). Characterisation of protein-rich isolates and antioxidative phenolic extracts from pale and black brewers' spent grain. *International Journal of Food Science & Technology*, *48*(8), 1670–1681. https://doi.org/10.1111/ijfs.12137.

Conway, J. (2019, August 21). Global beer production 1998-2018. *Alcohol Beverages*, https://www.statista.com/statistics/270275/worldwide-beer-production/.

Cooray, S. T., & Chen, W. N. (2018). Valorization of brewer's spent grain using fungi solid-state fermentation to enhance nutritional value. *Journal of Functional Foods*, *42*, 85–94. https://doi.org/10.1016/j.jff.2017.12.027

Crofton, E. C., & Scannell, A. G. M. (2020). Snack foods from brewing waste: Consumer-led approach to developing sustainable snack options. *British Food Journal*, *122*(12), 3899–3916. https://doi.org/10.1108/BFJ-08-2018-0527

Crowley, D., O'Callaghan, Y., McCarthy, A. L., Connolly, A., Fitzgerald, R. J., O'Brien, N. M. (2017). Aqueous and enzyme-extracted phenolic compounds from brewers' spent grain (BSG): Assessment of their antioxidant potential. *Journal of Food Biochemistry*, *41*(3), e12370. https://doi.org/10.1111/jfbc.12370.

del Río, J. C., Prinsen, P., & Gutiérrez, A. (2013). Chemical composition of lipids in brewer's spent grain: A promising source of valuable phytochemicals. *Journal of Cereal Science*, *58*(2), 248–254. https://doi.org/10.1016/j.jcs.2013.07.001

Elleuch, M., Bedigian, D., Roiseux, O., Besbes, S., Blecker, C., & Attia, H. (2011). Dietary fibre and fibre-rich by-products of food processing: Characterisation, technological functionality and commercial applications: A review. *Food Chemistry*, *124*(2), 411–421. https://doi.org/10.1016/j.foodchem.2010.06.077

Föste, M., Verheyen, C., Jekle, M., & Becker, T. (2020). Fibres of milling and fruit processing by-products in gluten-free bread making: A review of hydration properties, dough formation and quality-improving strategies. *Food Chemistry*, *306*, 125451. https://doi.org/10.1016/j.foodchem.2019.125451

Fărcaş, A. C., Socaci, S. A., Dulf, F. V., Tofană, M., Mudura, E., & Diaconeasa, Z. (2015). 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. *Journal of Cereal Science*, *64*, 34–42. https://doi.org/10.1016/j.jcs.2015.04.003

Fu, Q. Y., Yu, X. C., Li, L., Liu, G. Q., Li, B. (2011). Antioxidant Activities of Soluble Dietary Fiber Extracted from Brewers' Spent Grain. *Advanced Materials Research*, *233-235*, 2824–2827. https://doi.org/10.4028/www.scientific.net/amr.233-235.2824.

Garcia-Garcia, G., Stone, J., & Rahimifard, S. (2019). Opportunities for waste valorisation in the food industry—A case study with four UK food manufacturers. *Journal of Cleaner Production*, *211*, 1339–1356.

Gil-López, D. I. L., Lois-Correa, J. A., Sánchez-Pardo, M. E., Domínguez-Crespo, M. A., Torres-Huerta, A. M., Rodríguez-Salazar, A. E., & Orta-Guzmán, V. N. (2019). Production of dietary fibers from sugarcane bagasse and sugarcane tops using microwave-assisted alkaline treatments. *Industrial Crops and Products*, *135*, 159–169. https://doi.org/10.1016/j.indcrop.2019.04.042

Gmoser, R., Fristedt, R., Larsson, K., Undeland, I., Taherzadeh, M. J., & Lennartsson, P. R. (2020). From stale bread and brewers spent grain to a new food source using edible filamentous fungi. *Bioengineered*, *11*(1), 582–598. https://doi.org/10.1080/21655979.2020.1768694

Grasso, S. (2020). Extruded snacks from industrial by-products: A review. *Trends in Food Science & Technology*, *99*, 284–294. https://doi.org/10.1016/j.tifs.2020.03.012

Guido, L. F., Moreira, M. M. (2017). Techniques for Extraction of Brewer's Spent Grain Polyphenols: a Review. *Food and Bioprocess Technology*, *10*(7), 1192–1209. https://doi.org/10.1007/s11947-017-1913-4.

Haruna, M., Udobi, C., & Ndife, J. (2011). Effect of added brewers dry grain on the physico-chemical, microbial and sensory quality of wheat bread. *American Journal of Food and Nutrition*, *1*(1), 39–43. https://doi.org/10.5251/ajfn.2011.1.1.39.43

He, Y., Kuhn, D. D., Ogejo, J. A., O'Keefe, S. F., Fraguas, C. F., Wiersema, B. D., Jin, Q., Yu, D., Huang, H. (2019). Wet fractionation process to produce high protein and high fiber products from brewer's spent grain. *Food and Bioproducts Processing*, *117*, 266–274. https://doi.org/10.1016/j.fbp.2019.07.011.

Heredia-Sandoval, N. G., Granados-Nevárez, M. d. C., Calderón de la Barca, A. M., Vásquez-Lara, F., Malunga, L. N., Apea-Bah, F. B., Beta, T., & Islas-Rubio, A. R. (2020). Phenolic acids, antioxidant capacity, and estimated glycemic index of cookies added with brewer's spent grain. *Plant Foods for Human Nutrition*, *75*(1), 41–47. https://doi.org/10.1007/s11130-019-00783-1

Ibarruri, J., Cebrián, M., & Hernández, I. (2019). Solid state fermentation of brewer's spent grain using *Rhizopus* sp. to enhance nutritional value. *Waste and Biomass Valorization*, *10*(12), 3687–3700. https://doi.org/10.1007/s12649-019-00654-5

Ibbett, R., White, R., Tucker, G., & Foster, T. (2019). Hydromechanical processing of brewer's spent grain as a novel route for separation of protein products with differentiated techno-functional properties. *Innovative Food Science & Emerging Technologies*, *56*, 102184. https://doi.org/10.1016/j.ifset.2019.102184

Ikram, S., Huang, L., Zhang, H., Wang, J., Yin, M. (2017). Composition and Nutrient Value Proposition of Brewers Spent Grain. *Journal of Food Science*, *82*(10), 2232–2242. https://doi.org/10.1111/1750-3841.13794.

Ivanova, K., Denkova, R., Kostov, G., Petrova, T., Bakalov, I., Ruscova, M., & Penov, N. (2017). Extrusion of brewers' spent grains and application in the production of functional food. Characteristics of spent grains and optimization of extrusion. *Journal of the Institute of Brewing*, *123*(4), 544–552. https://doi.org/10.1002/jib.448

Jozinović, A., Ačkar, Đ., Panak, J., Babić, J., Miličević, B., & Šubarić, D. (2015). Fiber content in modified extruded maize flour with addition of brewers' spent grain. *Book of Abstracts* [6th International Dietary Fibre Conference, Paris, France: Poster Presentation]. https://www.bib.irb.hr/764406?%rad=764406.

Kemppainen, K., Rommi, K., Holopainen, U., Kruus, K. (2016). Steam explosion of Brewer's spent grain improves enzymatic digestibility of carbohydrates and affects solubility and stability of proteins. *Applied Biochemistry and Biotechnology*, *180*(1), 94–108. https://doi.org/10.1007/s12010-016-2085-9.

Kieserling, K., Vu, T. M., Drusch, S., & Schalow, S. (2019). Impact of pectin-rich orange fibre on gel characteristics and sensory properties in lactic acid fermented yoghurt. *Food Hydrocolloids*, *94*, 152–163. https://doi.org/10.1016/j.foodhyd.2019.02.051

Kirjoranta, S., Tenkanen, M., & Jouppila, K. (2016). Effects of process parameters on the properties of barley containing snacks enriched with brewer's spent grain. *Journal of Food Science and Technology*, *53*(1), 775–783. https://doi.org/10.1007/s13197-015-2079-6

Kitrytė, V., Šaduikis, A., Venskutonis, P. R. (2015). Assessment of antioxidant capacity of brewer's spent grain and its supercritical carbon dioxide extract as sources of valuable dietary ingredients. *Journal of Food Engineering*, *167*, 18–24. https://doi.org/10.1016/j.jfoodeng.2014.12.005.

Ktenioudaki, A., Alvarez-Jubete, L., Smyth, T. J., Kilcawley, K., Rai, D. K., & Gallagher, E. (2015). Application of bioprocessing techniques (sourdough fermentation and technological aids) for brewer's

spent grain breads. *Food Research International*, *73*, 107–116. https://doi.org/10.1016/j.foodres.2015.03.008

Ktenioudaki, A., Crofton, E., Scannell, A. G. M., Hannon, J. A., Kilcawley, K. N., & Gallagher, E. (2013). Sensory properties and aromatic composition of baked snacks containing brewer's spent grain. *Journal of Cereal Science*, *57*(3), 384–390. https://doi.org/10.1016/j.jcs.2013.01.009

Ktenioudaki, A., O'Shea, N., & Gallagher, E. (2013). Rheological properties of wheat dough supplemented with functional by-products of food processing: Brewer's spent grain and apple pomace. *Journal of Food Engineering*, *116*(2), 362–368. https://doi.org/10.1016/j.jfoodeng.2012.12.005

Kumari, B., Tiwari, B. K., Walsh, D., Griffin, T. P., Islam, N., Lyng, J. G., Brunton, N. P., Rai, D. K. (2019). Impact of pulsed electric field pre-treatment on nutritional and polyphenolic contents and bioactivities of light and dark brewer's spent grains. *Innovative Food Science & Emerging Technologies*, *54*, 200–210. https://doi.org/10.1016/j.ifset.2019.04.012.

López Durán, V., Larsson, P. A., & Wågberg, L. (2018). Chemical modification of cellulose-rich fibres to clarify the influence of the chemical structure on the physical and mechanical properties of cellulose fibres and thereof made sheets. *Carbohydrate Polymers*, *182*, 1–7. https://doi.org/10.1016/j.carbpol.2017.11.006

Lao, E. J., Dimoso, N., Raymond, J., Mbega, E. R. (2020). The prebiotic potential of brewers' spent grain on livestock's health: a review. *Tropical Animal Health and Production*, *52*(2), 461–472. https://doi.org/10.1007/s11250-019-02120-9.

Li, B., Yang, W., Nie, Y., Kang, F., Goff, H. D., & Cui, S. W. (2019). Effect of steam explosion on dietary fiber, polysaccharide, protein and physicochemical properties of okara. *Food Hydrocolloids*, *94*, 48–56. https://doi.org/10.1016/j.foodhyd.2019.02.042

Li, S., Chen, G., Qiang, S., Tang, D., Chen, Y., Zhang, Z., Lei, Z., & Chen, Y. (2019). Intensifying soluble dietary fiber production and properties of soybean curd residue via autoclaving treatment. *Bioresource Technology Reports*, *7*, 100203. https://doi.org/10.1016/j.biteb.2019.100203

Liu, S. X., Singh, M., & Inglett, G. (2011). Effect of incorporation of distillers' dried grain with solubles (DDGS) on quality of cornbread. *LWT–Food Science and Technology*, *44*(3), 713–718. https://doi.org/10.1016/j.lwt.2010.10.001

Lynch, K. M., Steffen, E. J., Arendt, E. K. (2016). Brewers' spent grain: a review with an emphasis on food and health. *Journal of the Institute of Brewing*, *122*(4), 553–568. https://doi.org/10.1002/jib.363.

Magabane, I. E. (2017). *Technologies for improving the quality of bread doughs made with barley spent grain and sorghum* (Master's thesis). University of Pretoria, South Africa. https://repository.up.ac.za/handle/2263/65907

Martín-García, B., Tylewicz, U., Verardo, V., Pasini, F., Gómez-Caravaca, A. M., Caboni, M. F., Dalla Rosa, M. (2020). Pulsed electric field (PEF) as pre-treatment to improve the phenolic compounds recovery from brewers' spent grains. *Innovative Food Science & Emerging Technologies*, *64*, 102402 https://doi.org/10.1016/j.ifset.2020.102402.

McCarthy, A. L., O'Callaghan, Y. C., Connolly, A., Piggott, C. O., FitzGerald, R. J., O'Brien, N. M. (2014). Phenolic-enriched fractions from brewers' spent grain possess cellular antioxidant and immunomodulatory effects in cell culture model systems. *Journal of the Science of Food and Agriculture*, *94*(7), 1373–1379. https://doi.org/10.1002/jsfa.6421.

McCarthy, A. L., O'Callaghan, Y. C., Connolly, A., Piggott, C. O., FitzGerald, R. J., O'Brien, N. M. (2012). Phenolic extracts of brewers' spent grain (BSG) as functional ingredients – Assessment of their DNA protective effect against oxidant-induced DNA single strand breaks in U937 cells. *Food Chemistry*, *134*(2), 641–646. https://doi.org/10.1016/j.foodchem.2012.02.133.

McCarthy, A. L., O'Callaghan, Y. C., Neugart, S., Piggott, C. O., Connolly, A., Jansen, M. A.K., Krumbein, A., Schreiner, M., FitzGerald, R. J., O'Brien, N. M. (2013). The hydroxycinnamic acid content of barley and brewers' spent grain (BSG) and the potential to incorporate phenolic extracts of BSG as antioxidants into fruit beverages. *Food Chemistry*, *141*(3), 2567–2574. https://doi.org/10.1016/j.foodchem.2013.05.048.

Meneses, N. G.T., Martins, S., Teixeira, J. A., Mussatto, S. I. (2013). Influence of extraction solvents on the recovery of antioxidant phenolic compounds from brewer's spent grains. *Separation and Purification Technology*, *108*, 152–158. https://doi.org/10.1016/j.seppur.2013.02.015.

Moreira, M. M., Morais, S., Carvalho, D. O., Barros, A. A., Delerue-Matos, C., Guido, L. F. (2013). Brewer's spent grain from different types of malt: Evaluation of the antioxidant activity and identification of the major phenolic compounds. *Food Research International*, *54*(1), 382–388. https://doi.org/10.1016/j.foodres.2013.07.023.

Morone, P., Falcone, P. M., & Tartie, V. E. (2019). Food waste valorisation: Assessing the effectiveness of collaborative research networks through the lenses of a COST action. *Journal of Cleaner Production*, *238*, 117868. https://doi.org/10.1016/j.jclepro.2019.117868

Nascimento, T. A., Calado, V., & Carvalho, C. W. P. (2017). Effect of Brewer's spent grain and temperature on physical properties of expanded extrudates from rice. *LWT–Food Science and Technology*, *79*, 145–151. https://doi.org/10.1016/j.lwt.2017.01.035

Niemi, P., Martins, D., Buchert, J., & Faulds, C. B. (2013). Prehydrolysis with carbohydrases facilitates the release of protein from brewer's spent grain. *Bioresource Technology*, *136*, 529–534. https://doi.org/10.1016/j.biortech.2013.03.076

Nigam, P. S. (2017). An overview: Recycling of solid barley waste generated as a by-product in distillery and brewery. *Waste Management*, *62*, 255–261. https://doi.org/10.1016/j.wasman.2017.02.018.

Nocente, F., Taddei, F., Galassi, E., & Gazza, L. (2019). Upcycling of brewers' spent grain by production of dry pasta with higher nutritional potential. *LWT*, *114*, 108421. https://doi.org/10.1016/j.lwt.2019.108421

Odeseye, A. A., Awonorin, S. O., Abdussalaam, R. O., Sanni, L. O., & Olayanju, T. M. A. (2020). The effect of processing variables on the biscuit-making potential of cocoyam-brewer's spent grain flour blends. *Croatian Journal of Food Science and Technology*, *12*(1), 56–66. https://doi.org/10.17508/CJFST.2020.12.1.08

Ohra-aho, T., Niemi, P., Aura, A.-M., Orlandi, M., Poutanen, K., Buchert, J., Tamminen, T. (2016). Structure of Brewer's Spent Grain Lignin and Its Interactions with Gut Microbiota in Vitro. *Journal of Agricultural and Food Chemistry*, *64*(4), 812–820. https://doi.org/10.1021/acs.jafc.5b05535.

Okpala, L. C., & Ofoedu, P. I. (2018). Quality characteristics of cookies produced from sweet potato and wheat flour blend fortified with brewer's spent grain flour. *Current Research in Nutrition and Food Science Journal*, *6*(1), 113–119. https://doi.org/10.12944/CRNFSJ.6.1.12

Parekh, I., Khanvilkar, A., Naik, A. (2017). Barley-wheat brewers' spent grain: A potential source of antioxidant rich lipids. *Journal of Food Processing and Preservation*, *41*(6), e13244 https://doi.org/10.1111/jfpp.13244.

Patel, A., Mikes, F., Bühler, S., Matsakas, L. (2018). Valorization of Brewers' Spent Grain for the Production of Lipids by Oleaginous Yeast. *Molecules*, *23*(12), 3052https://doi.org/10.3390/molecules23123052.

Patrignani, M., González-Forte, L. del S. (2021). Characterisation of melanoidins derived from Brewers' spent grain: new insights into their structure and antioxidant activity. *International Journal of Food Science & Technology*, *56*(1), 384–391. https://doi.org/10.1111/ijfs.14653.

Petrovic, J., Pajin, B., Tanackov-Kocic, S., Pejin, J., Fistes, A., Bojanic, N., & Loncarevic, I. (2017). Quality properties of cookies supplemented with fresh brewer's spent grain. *Food and Feed Research*, *44*(1), 57–63. https://doi.org/10.5937/FFR1701057P

Piggott, C. O., Connolly, A., FitzGerald, R. J. (2014). Application of ultrafiltration in the study of phenolic isolates and melanoidins from pale and black brewers' spent grain. *International Journal of Food Science & Technology*, *49*(10), 2252–2259. https://doi.org/10.1111/ijfs.12540.

Prandi, B., Faccini, A., Lambertini, F., Bencivenni, M., Jorba, M., Van Droogenbroek, B., Bruggeman, G., Schöber, J., Petrusan, J., Elst, K., & Sforza, S. (2019). Food wastes from agrifood industry as possible sources of proteins: A detailed molecular view on the composition of the nitrogen fraction, amino acid profile and racemisation degree of 39 food waste streams. *Food Chemistry*, *286*, 567–575. https://doi.org/10.1016/j.foodchem.2019.01.166

Qin, F., Johansen, A. Z., & Mussatto, S. I. (2018). Evaluation of different pretreatment strategies for protein extraction from brewer's spent grains. *Industrial Crops and Products*, *125*, 443–453. https://doi.org/10.1016/j.indcrop.2018.09.017

Rachwał, K., Waśko, A., Gustaw, K., Polak-Berecka, M. (2020). Utilization of brewery wastes in food industry. *PeerJ*, *8*, e9427https://doi.org/10.7717/peerj.9427.

Ravindran, R., Jaiswal, S., Abu-Ghannam, N., Jaiswal, A. K. (2018). A comparative analysis of pretreatment strategies on the properties and hydrolysis of brewers' spent grain. *Bioresource Technology*, *248*, 272–279. https://doi.org/10.1016/j.biortech.2017.06.039.

Reis, S. F., & Abu-Ghannam, N. (2014). Antioxidant capacity, arabinoxylans content and in vitro glycaemic index of cereal-based snacks incorporated with brewer's spent grain. *LWT–Food Science and Technology*, *55*(1), 269–277. https://doi.org/10.1016/j.lwt.2013.09.004

Reis, S. F., Coelho, E., Coimbra, M. A., Abu-Ghannam, N. (2015a). Improved efficiency of brewer's spent grain arabinoxylans by ultrasound-assisted extraction. *Ultrasonics Sonochemistry*, *24*, 155–164. https://doi.org/10.1016/j.ultsonch.2014.10.010.

Reis, S. F., Gullón, B., Gullón, P., Ferreira, S., Maia, C. J., Alonso, J. L., Domingues, F. C., Abu-Ghannam, N. (2014). Evaluation of the prebiotic potential of arabinoxylans from brewer's spent grain. *Applied Microbiology and Biotechnology*, *98*(22), 9365–9373. https://doi.org/10.1007/s00253-014-6009-8.

Reis, Sofia F., Coelho, Elisabete, Coimbra, Manuel A., Abu-Ghannam, Nissreen (2015b). Influence of grain particle sizes on the structure of arabinoxylans from brewer's spent grain. *Carbohydrate Polymers*, *130*, 222–226. https://doi.org/10.1016/j.carbpol.2015.05.031.

Ribau Teixeira, M., Guarda, E. C., Freitas, E. B., Galinha, C. F., Duque, A. F., Reis, M. A.M. (2020). Valorization of raw brewers' spent grain through the production of volatile fatty acids. *New Biotechnology*, *57*, 4–10. https://doi.org/10.1016/j.nbt.2020.01.007.

Roth, M., Döring, C., Jekle, M., & Becker, T. (2016). Mechanisms behind distiller's grains impact on wheat dough and bread quality. *Food and Bioprocess Technology*, *9*(2), 274–284. https://doi.org/10.1007/s11947-015-1620-y

Roth, M., Schuster, H., Kollmannsberger, H., Jekle, M., & Becker, T. (2016). Changes in aroma composition and sensory properties provided by distiller's grains addition to bakery products. *Journal of Cereal Science*, *72*, 75–83. https://doi.org/10.1016/j.jcs.2016.10.002

Severini, C., Azzollini, D., Jouppila, K., Jussi, L., Derossi, A., De Pilli, T. (2015). Effect of enzymatic and technological treatments on solubilisation of arabinoxylans from brewer's spent grain. *Journal of Cereal Science*, *65*, 162–166. https://doi.org/10.1016/j.jcs.2015.07.006.

Sibhatu, H. K., Anuradha Jabasingh, S., Yimam, A., Ahmed, S. (2021). Ferulic acid production from brewery spent grains, an agro-industrial waste. *LWT*, *135*, 110009 https://doi.org/10.1016/j.lwt.2020.110009.

Skendi, A., Harasym, J., & Galanakis, C. M.(2018). Recovery of high added-value compounds from brewing and distillate processing by-products. *Sustainable Recovery and Reutilization of Cereal Processing By-Products*, (189–225). Elsevier. https://doi.org/10.1016/B978-0-08-102162-0.00007-1.

Sobukola, O. P., Babajide, J. M., & Ogunsade, O. (2013). Effect of brewers spent grain addition and extrusion parameters on some properties of extruded yam starch-based pasta: Effect of BSG on yam starch-based pasta. *Journal of Food Processing and Preservation*, *37*(5), 734–743. https://doi.org/10.1111/j.1745-4549.2012.00711.x

Socaci, S. A., Fărcaş, A. C., Diaconeasa, Z. M., Vodnar, D. C., Rusu, B., Tofană, M. (2018). Influence of the extraction solvent on phenolic content, antioxidant, antimicrobial and antimutagenic activities of brewers' spent grain. *Journal of Cereal Science*, *80*, 180–187. https://doi.org/10.1016/j.jcs.2018.03.006.

Spinelli, S., Padalino, L., Costa, C., Del Nobile, M. A., & Conte, A. (2019). Food by-products to fortified pasta: A new approach for optimization. *Journal of Cleaner Production*, *215*, 985–991. https://doi.org/10.1016/j.jclepro.2019.01.117

Stefanello, F. S., dos Santos, C. O., Bochi, V. C., Fruet, A. P. B., Soquetta, M. B., Dörr, A. C., Nörnberg, J. L. (2018). Analysis of polyphenols in brewer's spent grain and its comparison with corn silage and cereal brans commonly used for animal nutrition. *Food Chemistry*, *239*, 385–401. https://doi.org/10.1016/j.foodchem.2017.06.130.

Steiner, J., Procopio, S., Becker, T. (2015). Brewer's spent grain: source of value-added polysaccharides for the food industry in reference to the health claims. *European Food Research and Technology*, *241*(3), 303–315. https://doi.org/10.1007/s00217-015-2461-7.

Steinmacher, N. C., Honna, F. A., Gasparetto, A. V., Anibal, D., & Grossmann, M. V. E. (2012). Bioconversion of brewer's spent grains by reactive extrusion and their application in bread-making. *LWT–Food Science and Technology*, *46*(2), 542–547. https://doi.org/10.1016/j.lwt.2011.11.011

Stojceska, V. (2011). Dietary Fiber from Brewer's Spent Grain as a Functional Ingredient in Bread Making Technology. *Flour and*

Breads and their Fortification in Health and Disease Prevention, 171–181. https://doi.org/10.1016/B978-0-12-380886-8.10016-9.

Stojceska, V. (2019). Brewer's Spent Grain From By-Product to Health: A Rich Source of Functional Ingredients. *Flour and Breads and their Fortification in Health and Disease Prevention*, 189–198. https://doi.org/10.1016/B978-0-12-814639-2.00015-0.

Tan, Y. X., Mok, W. K., Lee, J., Kim, J., Chen, W. N. (2019). Solid State Fermentation of Brewers' Spent Grains for Improved Nutritional Profile Using Bacillus subtilis WX-17. *Fermentation*, *5*(3), 52https://doi.org/10.3390/fermentation5030052.

Thorvaldsson, M. (2020). *Biotransformation of brewer's spent grain and application as a food ingredient in extruded breakfast cereals* (Master's thesis). Chalmers University of Technology, Sweden. https://hdl.handle.net/20.500.12380/300764

Tišma, M., Jurić, A., Bucić-Kojić, A., Panjičko, M., & Planinić, M. (2018). Biovalorization of brewers' spent grain for the production of laccase and polyphenols: Biovalorization of brewers' spent grain. *Journal of the Institute of Brewing*, *124*(2), 182–186. https://doi.org/10.1002/jib.479

Torbica, A., Škrobot, D., Janić Hajnal, E., Belović, M., & Zhang, N. (2019). Sensory and physico-chemical properties of wholegrain wheat bread prepared with selected food by-products. *LWT*, *114*, 108414. https://doi.org/10.1016/j.lwt.2019.108414

Uchegbu, N. N. (2016). Consumer acceptability of crackers produced from blend of sprouted pigeon pea, unripe plantain and brewers' spent grain and its hypoglycemic effect in diabetic rats. *World Academy of Science, Engineering and Technology, International Journal of Nutrition and Food Engineering*, *10*(6), 5.

Uchegbu, N. N., & Ishiwu, C. N. (2016). Nutritional Composition of Crackers Produced from Blend of Sprouted Pigeon Pea (Cajanus cajan), Unripe Plantain (Musa parasidiaca) and Brewers' Spent Grain Flour and Blood Glucose Level of Diabetic Rats Fed the Biscuit. *World Academy of Science, Engineering and Technology International Journal of Nutrition and Food Engineering*, *10*(1), 1–5.

Verni, M., Pontonio, E., Krona, A., Jacob, S., Pinto, D., Rinaldi, F., Verardo, V., Díaz-de-Cerio, E., Coda, R., & Rizzello, C. G. (2020). Bioprocessing of brewers' spent grain enhances its antioxidant activity: Characterization of phenolic compounds and bioactive peptides. *Frontiers in Microbiology*, *11*, 1831. https://doi.org/10.3389/fmicb.2020.01831

Verni, M., Rizzello, C. G., & Coda, R. (2019). Fermentation biotechnology applied to cereal industry by-products: Nutritional and functional insights. *Frontiers in Nutrition*, *6*, 42. https://doi.org/10.3389/fnut.2019.00042

Vieira, E. F., da Silva, D. D., Carmo, H., Ferreira, I. M.P.L.V.O. (2017). Protective ability against oxidative stress of brewers' spent grain protein hydrolysates. *Food Chemistry*, *228*, 602–609. https://doi.org/10.1016/j.foodchem.2017.02.050.

Vieira, E., Rocha, M. A.a M., Coelho, E., Pinho, O., Saraiva, J. A., Ferreira, I. M.P.L.V.O., Coimbra, M. A. (2014). Valuation of brewer's spent grain using a fully recyclable integrated process for extraction of proteins and arabinoxylans. *Industrial Crops and Products*, *52*, 136–143. https://doi.org/10.1016/j.indcrop.2013.10.012.

Vieira, E., Teixeira, J., Ferreira, I. M. P. L. V. O. (2016). Valorization of brewers' spent grain and spent yeast through protein hydrolysates with antioxidant properties. *European Food Research and Technology*, *242*(11), 1975–1984. https://doi.org/10.1007/s00217-016-2696-y.

Waters, D. M., Jacob, F., Titze, J., Arendt, E. K., & Zannini, E. (2012). Fibre, protein and mineral fortification of wheat bread through milled and fermented brewer's spent grain enrichment. *European Food Research and Technology*, *235*(5), 767–778. https://doi.org/10.1007/s00217-012-1805-9

Wen, C., Zhang, J., Duan, Y., Zhang, H., Ma, H. (2019). A Mini-Review on Brewer's Spent Grain Protein: Isolation, Physicochemical Properties, Application of Protein, and Functional Properties of Hydrolysates. *Journal of Food Science*, *84*(12), 3330–3340. https://doi.org/10.1111/1750-3841.14906.

Woomer, J. (2018). Valorization of Proso Millet and Spent Grain for Extruded Snack Development. *Thesis*, University of Kentucky. https://uknowledge.uky.edu/bae_etds/61/.

Xie, F., Li, M., Lan, X., Zhang, W., Gong, S., Wu, J., & Wang, Z. (2017). Modification of dietary fibers from purple-fleshed potatoes (Heimeiren) with high hydrostatic pressure and high pressure homogenization processing: A comparative study. *Innovative Food Science & Emerging Technologies*, *42*, 157–164. https://doi.org/10.1016/j.ifset.2017.05.012

Xiros, C., Christakopoulos, P. (2012). Biotechnological Potential of Brewers Spent Grain and its Recent Applications. *Waste and Biomass Valorization*, *3*(2), 213–232. https://doi.org/10.1007/s12649-012-9108-8.

Yan, L., Li, T., Liu, C., & Zheng, L. (2019). Effects of high hydrostatic pressure and superfine grinding treatment on physicochemical/functional properties of pear pomace and chemical composition of its soluble dietary fibre. *LWT*, *107*, 171–177. https://doi.org/10.1016/j.lwt.2019.03.019

Yu, D., Sun, Y., Wang, W., O'Keefe, S. F., Neilson, A. P., Feng, H., Wang, Z., Huang, H. (2020). Recovery of protein hydrolysates from brewer's spent grain using enzyme and ultrasonication. *International Journal of Food Science & Technology*, *55*(1), 357–368. https://doi.org/10.1111/ijfs.14314.

Zhang, H., Cao, X., Yin, M., Wang, J. (2018). Soluble dietary fiber from Qing Ke (highland barley) brewers spent grain could alter the intestinal cholesterol efflux in Caco-2 cells. *Journal of Functional Foods*, *47*, 100–106. https://doi.org/10.1016/j.jff.2018.05.046.

Zielke, C., Teixeira, C., Ding, H., Cui, S., Nyman, M., Nilsson, L. (2017). Analysis of β-glucan molar mass from barley malt and brewer's spent grain with asymmetric flow field-flow fractionation (AF4) and their association to proteins. *Carbohydrate Polymers*, *157*, 541–549. https://doi.org/10.1016/j.carbpol.2016.10.045.

Zuorro, A., Iannone, A., Lavecchia, R. (2019). Water–Organic Solvent Extraction of Phenolic Antioxidants from Brewers' Spent Grain. *Processes*, *7*(3), 126https://doi.org/10.3390/pr7030126.

How to cite this article: Naibaho, J., Korzeniowska, M. Brewers' spent grain in food systems: Processing and final products quality as a function of fiber modification treatment. *J Food Sci*. 2021; *86:* 1532–1551. https://doi.org/10.1111/1750-3841.15714

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Wroclaw, April 12 2023
(miejscowość i data)

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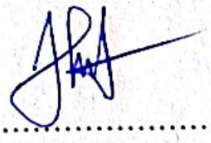
OŚWIADCZENIE

Oświadczam, że w pracy

Naibaho, J. and Korzeniowska, M. 2021. Brewers' spent grain in food systems: Processing and final products quality as a function of fiber modification treatment. Journal of Food Science 86(5), 1532–1551. https://doi.org/10.1111/1750-3841.15714 (autorzy, rok wydania, tytuł, czasopismo lub wydawca, tom, strony)

mój udział polegał na Pomysł, projekt, treść i redagowanie manuskryptu (opisać szczegółowo swój własny udział w powstaniu pracy, np. wykonaniu doświadczeń techniką ....., analizie statystycznej wyników eksperymentów zilustrowanych na ryc. ...., przygotowaniu tekstu manuskryptu zamieszczonego w rozdziale ....., kierowaniu projektem naukowym obejmującym badania opisane w tej pracy, itp.).

Wroclaw, April 12 2023



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data i podpis

Assoc. Prof. Małgorzata Korzeniowska
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OŚWIADCZENIE

Oświadczam, że w pracy

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mój udział polegał na Doskonalenie opracowanego pomysłu, treści i nadzór nad procesem pisania (opisać szczegółowo swój własny – a nie Kandydata udział w powstaniu pracy, np. wykonaniu doświadczeń techniką ....., analizie statystycznej wyników eksperymentów zilustrowanych na ryc. ...., przygotowaniu tekstu manuskryptu zamieszczonego w rozdziale ....., kierowaniu projektem naukowym obejmującym badania opisane w tej pracy, itp.).

April 12nd 2023

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data i podpis

Publication 2: Not in open access

Publication 3

LWT - Food Science and Technology 163 (2022) 113612

Contents lists available at ScienceDirect

LWT

journal homepage: www.elsevier.com/locate/lwt

Antioxidant activities and polyphenolic identification by UPLC-MS/MS of autoclaved brewers' spent grain

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ARTICLE INFO

Keywords:
Agro-industrial byproduct
Valorization
Bioactive compounds
Biological properties
Thermal exposure

ABSTRACT

Autoclave treatment (AT) modified the dietary fiber composition of brewers' spent grain (BSG), impacting its techno-processing properties. However, its impact on antioxidant properties and stability of polyphenolic compounds remain unclear. This study aimed to evaluate the influence of AT on several antioxidant activities and polyphenolic composition. The results showed that AT increased ORAC (oxygen radical absorbance capacity), ABTS (2,2′-Azinobis-(3-ethylbenzthiazoline-6-sulfonic acid), and FRAP (ferric-reducing antioxidant power) value upto 7 folds compared to the untreated BSG. The lower temperature degraded the amount of flavan-3-ols and phenolic acids. However, AT at 110 °C and 130 °C upgraded the phenolic acids upto 4 and 11 folds respectively. Higher temperature also induced the formation of benzoic acid and (+)-catechin. UPLC-MS/MS identified several phenolic acids including syringic acid, benzoic acid, coumaric acid, ferulic acid and its derivatives, and flavan-3-ols such as (+)-catechin and (−)-epicatechin. In conclusion, AT improved the bioactivity of BSG, enhanced the amount of certains phenolic compounds and released the bioactive compounds from BSG matrices thus offering a higher benefit for industry to utilise autoclaved BSG.

1. Introduction

Potential of brewers' spent grain (BSG) as food and nutraceutical ingredients has been reported due to its biological properties (Bonifácio-Lopes, Teixeira, & Pintado, 2020; Connolly, Cermeño, Alashi, Aluko, & FitzGerald, 2021; Lynch, Steffen, & Arendt, 2016; Naibaho & Korzeniowska, 2021; Patrignani, Brantsen, Awika, & Conforti, 2021). BSG possesses several biological activities such as antioxidant activities, antimicrobial properties, DNA protective and antimutagenic, anti-inflammatory as well as maintaining colon health. Those capabilities are a result of a high amount of certain bioactive compounds such as polyphenolic compounds, protein and amino acids, lipid and fatty acids and dietary fibre (Naibaho & Korzeniowska, 2021; Nigam, 2017). Therefore, the improvement of chemical-related nutritional value and certain biological properties in food products such as bread, pasta, cookies, extruded products and yoghourt, has also been investigated (Heredia-Sandoval et al., 2020; Naibaho et al., 2022; Nocente, Taddei, Galassi, & Gazza, 2019; Torbica, Škrobot, JanićHajnal, Belović, & Zhang, 2019).

The addition of BSG in food products was observed to intensify the hardness of baked food products thus potentially to regulate semi-solid food products such as yoghourt (Naibaho et al., 2022; Naibaho & Korzeniowska, 2021). This phenomenon is due to the high amount of insoluble dietary fibre (IDF) which has a high ability to absorb high amounts of water (Steiner, Procopio, & Becker, 2015) thus disrupting the network formation in baked food products. However, autoclave treatment (AT) is able to modify the amount of IDF and convert to soluble dietary fibre (SDF) (Naibaho et al., 2021b). Consequently, it

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https://doi.org/10.1016/j.lwt.2022.113612
Received 12 January 2022; Received in revised form 15 March 2022; Accepted 27 May 2022
Available online 31 May 2022

modified the physical properties of BSG such as water holding capacity and oil holding capacity (Naibaho et al., 2021b). AT degraded the hemicellulose and cellulose thus improving the amount of SDF (Li et al., 2019).

Several treatments have been reported for enhancing the yield of phenolic-rich extracts from BSG and its biological activities including pulsed electric field (Martín-García et al., 2020), solid state fermentation (Cooray & Chen, 2018; Tan, Mok, Lee, Kim, & Chen, 2019), enzyme treatments (Connolly et al., 2019, 2021), pH elevation (Connolly et al., 2021), and the combination of thermal and enzyme treatment (Budaraju, Mallikarjunan, Annor, Schoenfuss, & Raun, 2018). As mentioned earlier, AT transformed IDF in to SDF and modified the physical properties of treated BSG. Moreover, AT reduced the water activity (Aw) thus allowing a shelf life extension and safety during the storage. Surface chemistry study by Fourier transform infrared spectroscopy (FTIR) showed that AT changed the hydroxyl and acids functional groups of BSG (Naibaho et al., 2021b).

However, the influence of AT in biological activity of BSG such as antioxidant activities and polyphenolic composition remain unclear. Due to the decomposition and degradation of dietary fiber, certain polyphenolic compounds could be released from the cell wall and matrices of BSG thus improving the bioavailability. Major phenolic compounds in BSG are proto-catechuic, caffeic, *p*-coumaric and ferulic acids, catechin, and derivatives (Barbosa-Pereira et al., 2014; McCarthy et al., 2013; Moreira et al., 2013; Sibhatu, Anuradha, Yimam, & Ahmed, 2021). Polyphenolic compounds of BSG are responsible for several antioxidant activities including enzymatic antioxidants such as superoxide dismutase (SOD), catalase (CAT), glutathione (GSH) as well as non-enzymatic antioxidant activities such as total phenolic content (TPC), di(phenyl)-(2; 4;6-trinitrophenyl)iminoazanium (DPPH), ferric-reducing antioxidant power (FRAP), 2,2′-azinobis-(3-ethylbenzthiazoline-6-sulfonic acid) (ABTS), lipid peroxidation, and deoxyribose scavenging activity (Barbosa-Pereira et al., 2014; Crowley et al., 2017; Kumari et al., 2019; McCarthy et al., 2014; Moreira et al., 2013; Socaci et al., 2018; Verni et al., 2020). However, the composition and the amount of BSG polyphenolic extracts as well as its biological activities vary depending on several factors including source of origin, post handling process, extraction process and solvent used (Barbosa-Pereira et al., 2014; Bonifácio-Lopes et al., 2020; Meneses, Martins, Teixeira, & Musatto, 2013).

This study aimed to evaluate the impact of AT on BSG in terms of polyphenolic composition and its bioactivity of BSG. It is expected that several treatments might increase the antioxidant activities and improve the yield of polyphenolic compounds. Consequently, the improvement of biological properties of BSG will enhance the quality of BSG both as a food and nutraceutical ingredient. Thus, it will benefit the industry in providing a higher quality of BSG and its derivatives products.

2. Materials and methods

2.1. Materials and chemicals

BSG samples were collected from local breweries in Wroclaw, Poland. The samples were then handled as described in the previous study (Naibaho et al., 2021b). The BSG was dried using conventional drying methods at 75 °C to reach a stable moisture (approximately 2–5%). After that, the dried samples were ground by a laboratory scale mill and passed through a 385 μm laboratory scale sieve. The samples were kept in aluminium foil bags and stored at 4 °C before the treatment.

Trolox (6-hydro-2,5,7,8-tetramethylchroman-2-carboxylic acid) was purchased from Sigma-Aldrich (Steinheim, Germany). UPLC-grade water was prepared by using the HLP SMART 1000s system (Hydrolab, Gdansk, Poland). Before use, the water was filtered by a 0.22 μm membrane filter immediately. All chemicals used were analytical grade.

2.2. Experimental designs

AT was performed in an autoclave (ASL 100M, Poland) as described in the previous study (Naibaho et al., 2021b). A 75 g of distilled water was added into 25 g of BSG and mixed properly to obtain a homogenous mixture. The mixture was then packed into a polyethylene (PE) bag and vacuum sealed for the treatment. The treatment was carried out at 4 different temperatures (90 °C, 100 °C, 110 °C and 130 °C). The higher temperature (110 °C and 130 °C) was conducted with a set of 1 × 60 kPa/10 kPa pressure. All groups of treatment were conducted at different time exposure at 9, 12 and 15 min. Therefore, 12 samples were obtained in each group. After the treatment, all the samples were dried for 5 h using oven drying at 75 °C. After that, the dried samples were ground and passed through a 385 μm sieve and kept in aluminium foil bags at 4 °C before the preparation of methanol extracts. All analyses were performed in at least duplicate. BSG pale ale type from 2 different breweries was used as described in a previous study (Naibaho et al., 2021a). Therefore, the results from untreated BSG which has been reported in a previous study (Naibaho et al., 2021a) was provided as a comparison for those 2 types. BSG control was prepared by drying, milling and sieving as mentioned previously (Section 2.1). The comparison between those 2 types was provided based on the statistical analysis and the folds changes due to the treatment. The degree of changes in biological activities and phenolic compounds due to the autoclave treatment is performed in time folds.

2.3. Methanol extraction procedure

Methanol extracts from BSG were prepared for determination of antioxidant activities and identification of polyphenolic compounds. The extract was prepared following methods as described in previous studies (Turkiewicz et al., 2020). Briefly, 6 mL of methanol solution 80% (methanol/water: 80/20, *v/v*) and 7 mL of methanol 30% (methanol/water/acetic acid/ascorbic acid: 30/68/1/1, *v/v/v/m*) respectively for measurement of antioxidant activity and polyphenolic identification respectively, was added into 1 g of treated BSG and then mixed properly by using vortex for 1 min. After that, the mixture was then sonicated (Sonic 6D, Polsonic, Warsaw, Poland) for 20 min and the mixture was left 4 °C for 24 h. The mixture was then sonicated for 20 min followed by centrifugation at 19000*g* for 10 min at 4 °C. To obtain the methanol extract, the mixture was filtered using a 0.20 μm hydrophilic PTFE membrane (Millex Simplicity Filter, Merck, Germany).

2.4. Determination of antioxidants

Antioxidant activity of the extracts was assessed for ABTS, FRAP, and ORAC (Benzie & Strain, 1996; Ou, Chang, Huang, & Prior, 2013; Re et al., 1999) with slight modification with slight modification. The result was expressed as mmol Trolox equivalents/100 g dry sample.

2.4.1. ORAC

ORAC assessment was performed using fluorescence microplate method. 25 μL of samples, control, standard, and diluted buffer was added into wells containing 150 μL of fluorescein solution. The plate then was incubated for 30 min at 37 °C. After that, 25 μL of AAPH (2,2′-azobis(2-amidino-propane) dihydrochloride) solution was added to each well. As the timing is critical, the plate then immediately transferred to the plate reader and the fluorescence was measured every minute for 35 min. The calculation of standards and samples was done based on the area under the curve. The standard curve was obtained by plotting Trolox concentrations against the average of the area of two measurements for each concentration. The ORAC value was obtained using a regression equation from the standard curve.

2.4.2. ABTS

ABTS value was measured as follows: 3 mL of ABTS solution was

added into a cuvette containing 30 μL of methanol extract. Exactly after 6 min, the absorbance was measured at 734 nm wavelength spectrophotometer. The blank measurement was prepared with 30 μL of distilled water. A curve standard was prepared and measured at 734 nm wavelength spectrophotometer with absorbance range 0.700 ± 0.02.

2.4.3. FRAP

FRAP value was measured as follows. A mixture of reagent consists of acetate buffer pH 3.6, TPTZ (2,3,5-Triphenyltetrazolium chloride) dissolved in 40 mM/L HCl, and FeCl·6H20 dissolved in distilled water (10:1:1) is prepared on the same day for the measurement. A certain volume of the sample (0.1–1 mL) was mixed with distilled water to reach 1 mL mixture. 3 mL of the reagent was added into the mixture and the absorbance was measured exactly after 10 min at 593 nm wavelength with a range of 0.200–0.800 absorbance. The result then calculated as curve standard absorbance and the result was performed in Trolox equivalent.

2.5. Identification of polyphenolic compounds

Identification and quantification of flavan-3-ols and phenolic acids was conducted by Liquid Chromatography - Tandem mass Spectrometry (LC-MS-MS) following procedures as described in the previous studies (Tkacz, Wojdyło, Turkiewicz, & Nowicka, 2021; Turkiewicz et al., 2020, 2021). The profile and content were determined by ultra-performance liquid chromatography (Acquity UPLC system) with binary solvent manager and photodiode array detector PDA (Waters Corp., Milford, MA. US). The system was coupled to a Xevo™ G2 Q/TOF micro-mass spectrometer fitted with an electrospray ionisation ESI source (Waters Corp., Manchester, UK) which acts on negative modes. The analysis was carried out using full scan data dependent MS, scanning from *m/z* 100 to 1700. The characterisation of phenolic compositions was done according to the retention time and accurate molecular masses. The data were collected by a software, MassLynx™ 4.1 ChromaLynx Application Manager (Waters Corp. Milford, USA). Flavan-3-ols and phenolic acids were monitored at 280 nm and 320 wavelengths, respectively. Quantification was conducted based on the phenolic calibration standards at concentrations ranging between 0.05 and 5 mg/mL (R2 ≥ 0.9995). All the samples were analysed in triplicate and the results were performed in mg/kg dry weight sample.

2.6. Statistical analysis

The statistical analysis was carried out using Statistica software (version 13.5.0.17 by two-ways analysis of variance (ANOVA) for temperature and time exposure. Significant differences were evaluated at $p < 0.05$ by post-hoc Tukey HSD test assessment.

3. Results and discussion

3.1. Antioxidant activities

Antioxidant activities *in vitro* of BSG are shown in Table 1. In general, AT improved the antioxidant properties of treated BSG compared to that in control treatment. Furthermore, different temperatures and time exposure on AT significantly ($p < 0.05$) influenced the ORAC and ABTS value. However, different levels of temperature and time exposure had no impact on FRAP value ($p > 0.05$). ORAC value varied between 4.74 and 6.32 mmol Trolox/100 g dry weight; while ABTS and FRAP value ranged between 0.15 - 0.84 and 0.39–0.51 mmol Trolox/100 g dry weight respectively. Compared to the untreated BSG (Naibaho et al., 2021a), AT increased the antioxidant activities for ORAC, ABTS, and FRAP, and the level of improvement is shown in Fig. 1. As is shown in Fig. 1, higher temperature (110 °C and 130 °C) had the highest impact in the improvement of ORAC and ABTS value (Fig. 1a and b). Meanwhile, the highest enhancement impact on FRAP was given by the lower temperature treatments (90 °C and 100 °C). Thermal exposure on BSG by AT at 90 °C–130 °C increased the ORAC value more than 60% higher than that in control. AT was able to enhance the ABTS level remarkably up to 6–8 folds (>800%) than control, which was obtained at 130 °C. Although the combinations of temperature and time exposure statistically generated the same level of FRAP, treated BSG achieved FRAP value between 2 and 4 times higher (>250% higher) than that in untreated BSG. It was identified that untreated BSG had FRAP value at 0.1–0.2 mmol Trolox/100 g (Naibaho et al., 2021a) while current study revealed that AT-treated BSG generated FRAP level at a range of 0.39–0.51 mmol Trolox/100 g. FRAP value is reported to be related with the property of BSG in DNA protection effect (McCarthy et al., 2012).

Fig. 1 describes the level of improvement on antioxidant activities of BSG due to the AT on different temperature and time exposure. The figure shows that AT lower temperature (90 °C and 100 °C) increased FRAP higher than ABTS and ORAC value. FRAP demonstrated the ability of BSG in reducing the metal ion as a catalyst in lipid oxidation (Rahman et al., 2021). This result shows that AT at 90 °C and 100 °C improved the ability of phenolic compounds to diminish lipid oxidation. Higher temperature (110 °C and 130 °C) enhanced ABTS the most followed by FRAP and ORAC value. By this, AT improved the ability of BSG methanol extracts in electron donor for the reduction of molecular oxygen and

Table 1
Antioxidant activities *in vitro* of BSG treated with different temperature and time exposure by autoclave treatment.

Treatment	Antioxidant properties (mmol Trolox/100 g)			Polyphenolic compounds (mg/kg)		
	ORAC	ABTS	FRAP	Flavan-3-ols	Phenolic acids	Total
Group I						
Control*	2.615	0.086	0.106	824.95	100.55	925.50
90 °C; 9 min	4.738 ± 0.04^{h}	0.278 ± 0.03^{d}	0.450 ± 0.10^{a}	22.493 ± 0.01^{i}	30.021 ± 0.07^{l}	52.514
90 °C; 12 min	4.404 ± 0.09^{k}	0.303 ± 0.03^{d}	0.397 ± 0.07^{a}	22.980 ± 0.00^{h}	32.584 ± 0.00^{j}	55.563
90 °C; 15 min	4.898 ± 0.05^{g}	0.148 ± 0.03^{e}	0.390 ± 0.12^{a}	21.292 ± 0.13^{l}	30.512 ± 0.22^{k}	51.804
100 °C; 9 min	4.554 ± 0.16^{j}	0.250 ± 0.01^{de}	0.398 ± 0.07^{a}	21.774 ± 0.20^{k}	35.590 ± 0.14^{i}	57.363
100 °C; 12 min	4.572 ± 0.03^{i}	0.315 ± 0.01^{d}	0.444 ± 0.08^{a}	22.107 ± 0.01^{j}	38.478 ± 0.01^{g}	60.585
100 °C; 15 min	5.209 ± 0.05^{e}	0.283 ± 0.07^{d}	0.403 ± 0.03^{a}	24.435 ± 0.00^{g}	37.102 ± 0.01^{h}	61.537
Group II						
Control*	1.751	0.105	0.204	824.58	104.13	928.71
110 °C; 9 min	5.946 ± 0.29^{c}	0.503 ± 0.05^{c}	0.486 ± 0.04^{a}	38.230 ± 0.22^{f}	344.542 ± 0.25^{f}	382.772
110 °C; 12 min	5.956 ± 0.16^{b}	0.499 ± 0.02^{c}	0.486 ± 0.04^{a}	40.007 ± 0.09^{e}	374.444 ± 0.25^{e}	414.451
110 °C; 15 min	5.044 ± 0.15^{f}	0.511 ± 0.04^{c}	0.490 ± 0.03^{a}	41.929 ± 0.11^{d}	425.437 ± 0.19^{d}	467.366
130 °C; 9 min	5.764 ± 0.06^{d}	0.842 ± 0.03^{a}	0.509 ± 0.03^{a}	49.604 ± 0.15^{a}	1077.428 ± 0.33^{b}	1127.032
130 °C; 12 min	5.761 ± 0.12^{d}	0.683 ± 0.02^{b}	0.473 ± 0.00^{a}	46.339 ± 0.05^{c}	1057.985 ± 0.25^{c}	1104.324
130 °C; 15 min	6.321 ± 0.24^{a}	0.796 ± 0.05^{a}	0.508 ± 0.03^{a}	47.749 ± 0.01^{b}	1168.718 ± 0.39^{a}	1216.467

Note: the data is shown as mean ± standard deviation with triplicate analysis. *: the result is obtained from previous study (Naibaho et al., 2021b). except ORAC value was conducted in current study. Letters show the significant differences from other treatment in the same column ($p < 0.05$). Analysis was done in triplicate.

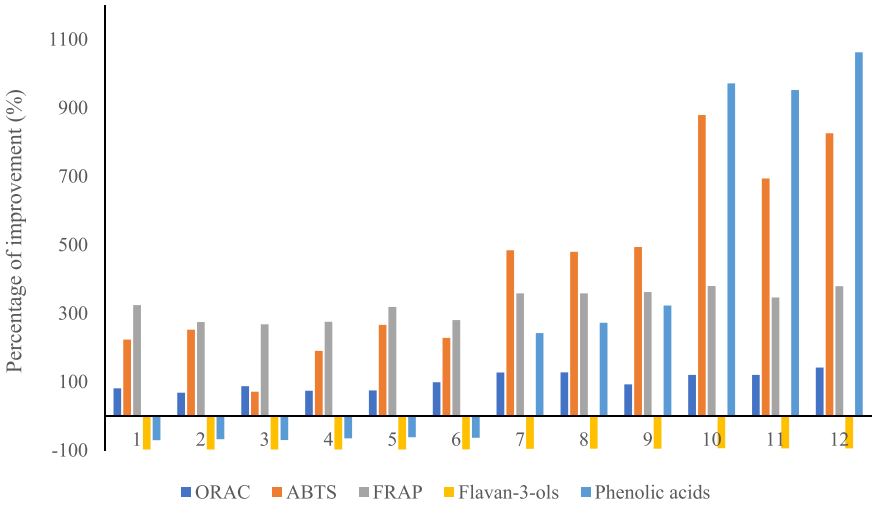

Fig. 1. The amount of improvement on biological activities and phenolic compounds of BSG due to the autoclave treatment at 90 °C (1: 9 min, 2: 12, 3: 15 min), 100 °C (4: 9 min, 5: 12, 6: 15 min), 110 °C (7: 9 min, 8: 12, 9: 15 min), and 130 °C (10: 9 min, 11: 12, 12: 15 min). The numbers were obtained based on the comparison of treated BSG (means) with untreated BSG from Table 1.

hydrogen peroxide. AT had the lowest enhancement on ORAC value regardless of the level of temperature and time exposure.

These results explain that AT strongly enhanced the biological activities of BSG. AT has been observed to enhance the amount of crude fat on BSG (Naibaho et al., 2021b) which might occur due to the release of the entrapped fat from BSG matrices. By this, the amount of extractable fat became higher. BSG consists of fatty acids such as palmitic, linoleic, oleic, and stearic acid which contribute to antioxidant properties in BSG (Fărcaş et al., 2015; Parekh, Khanvilkar, & Naik, 2017; Tan et al., 2019). The higher bioavailability of fat in BSG consequently improved its antioxidant properties. Several phenomena could occur due to the AT such as depolymerisation, debranching, and de-esterification of polysaccharides thus releasing the phenolic compounds which are responsible for biological activity. This phenomenon might be due to the transformation of insoluble dietary fibre into soluble dietary fibre (Naibaho et al., 2021b). Furthermore, FTIR analysis identified the microstructure changes in the acid functional group (Naibaho et al., 2021b) which might be the consequence of a higher presence of fatty acids or phenolic acids.

The improvement of biological properties of BSG has been observed due to the several treatments. Microwaves had 1.5 times higher %DPPH and %ABTS inhibition, and 3 times higher DPPH trolox equivalents than those in control (Budaraju et al., 2018). microwaves increased the antioxidant activity of BSG by inducing the formation of melanoidin which was formed due to the Maillard reaction (Patrignani et al., 2021). Fermentation improved the radical scavenging activity (Verni et al., 2020) and a 5.8 fold increase in total antioxidant activity has been observed (Tan et al., 2019). Aqueous pH-shift extraction increased the ORAC and TEAC (Trolox equivalent antioxidant capacity) value up to 4 and 3 times higher respectively; enzyme-aided generated 18 times higher FRAP value and had a higher *in vitro* ACE inhibitory activity (Connolly et al., 2021). Although it is reported that pH-shift improved the ORAC, TEAC, and FRAP more effectively than that in enzyme-aided treatment (Connolly et al., 2021).

However, some enzyme treatments could diminish the level of ABTS, FRAP, and ORAC (Connolly et al., 2019). Interestingly, pulsed electric fields on BSG had no impact on antioxidant activity and immunomodulatory effects (Kumari et al., 2019). Steaming (220 °C/2 min), roasting (60 °C/3 min), and autoclave (121 °C/20 min/15 psi) had no impact on the antioxidant activities and yield extracts (Budaraju et al., 2018). In current study, a fluctuation in antioxidant activities was observed. This phenomenon might depend on the stability of certain responsible compounds during the thermal exposure. Moreover, it might be also influenced by the capability of polyphenolic compounds in stabilising the specific free radical.

3.2. *Quantification and identification of flavan-3-ols and phenolic acid*

Quantitative analysis of phenolic compounds on treated BSG is presented in Table 1 and the level of modification in phenolic compounds due to the AT is performed in Fig. 1. The results showed that all the treatments significantly ($p < 0.05$) impacted the amount of flavan-3-ols and phenolic acids. Compared to the untreated BSG, AT at all temperature and time exposure ranges lowered the amount of flavan-3-ols up to −97% (Fig. 1). Interestingly, AT at 90 °C and 100 °C decreased the phenolic acid content up to −70% from the initial level, while temperature at 110 °C and 130 °C increased the amount of phenolic acids up to 1000% than that in control. The high reduction in flavan-3-ols shows that flavan-3-ols has a high instability in thermal exposure. The total amount of polyphenolic content depicts that only the highest temperature (130 °C) improved the amount of phenolic content. Although AT at 90 °C and 110 °C reduced the total amount of phenolic compounds compared to the control (untreated BSG), a significant improvement in antioxidant properties (ORAC, FRAP, and ABTS) was obtained as mentioned in the previous section. This phenomenon might be due to the depolymerisation of certain compounds such as caffeic acid into ferulic acid and led to its conversion into elementary units as reported previously (Wojdyło, Figiel, Lech, Nowicka, & Oszmiański, 2014). Moreover, a derivatization of ferulic acid into 4-vinylguaiacol due to a thermal exposure has been observed (Zago et al., 2022) and consequently declined the amounts of phenolic compounds while improving the antioxidant properties.

Higher temperatures (110 °C and 130 °C) multiplied the amount of phenolic acids due to its ability to disrupt the BSG matrices thus releasing the phenolic acids. It was observed that the high thermal exposure on BSG was able to disrupt the cell vacuoles and or cleaved the covalent bonds thus increasing the phenolic acids (Rahman et al., 2021). A reduction in the amount of phenolic acid by 4–6 times lower has been identified (Bonifácio-Lopes et al., 2020), brewing and roasting process lowered the hydroxycinnamic acid content (McCarthy et al., 2013). In addition, microwave treatment has been reported for increasing the antioxidant activity although it has no impact on the amount of bound

phenolic compounds (Budaraju et al., 2018). However, an improvement up to 2.7 folds in free phenolic acid and 1.7 folds in bound phenolic acid due to the pulsed electric field treatment has been reported (Martín-García et al., 2020) thus enhancing the amount of flavan-3-ols, phenolic acid derivatives, and flavonoids. Increase in free phenolic content has also been identified due to the thermal and enzyme treatment (Budaraju et al., 2018). The modification in phenolic compounds due to AT might be as an impact of dietary fibre transformation as reported previously (Naibaho et al., 2021b). Temperature exposure might induce the lignin solubility and polysaccharides degradation. Functional groups of lignin consist of guaiacyl and syringyl which similarly bind in all lignin. However, the amount of such compounds bound to the lignin varied depending on the extraction and isolation methods (Ohra-aho et al., 2016). By this, the variability in phenolic acids could occur due to the difference in strength binding between the matrices and phenolic compounds.

Tentative identification of phenolic compounds by LC-MS-MS is shown in Table 2. The result shows that the significant difference between flavan-3-ols and phenolic acid at 110 °C and 130 °C is due to the presence of benzoic acid and (+)-catechin which were not observed at 90 °C and 100 °C. As can be seen in Table 2, phenolic acid in BSG treated at 110 °C and 130 °C consists of syringic acid, benzoic acid, *p*-coumaric acid, ferulic-ferulic acid dimer, sinapic acid, ferulic acid, decarboxylated diferulic acid, and diferulic acid isomers; flavan-3-ols consists of (+)-catechin and (−)-epicatechin. The presence of benzoic acid and (+)-catechin was not identified at the treatment with a lower temperature (90 °C and 100 °C). According to a previous study, it was reported that hydroxybenzoic acid is the most dominant compound in BSG ohmic treated (Bonifácio-Lopes et al., 2022). By this, the low amount of phenolic acids in lower temperature treatment might be due to the absence of benzoic acid. The diminishing and inducing certain compounds have been observed in BSG due to the thermal treatment. Rahman et al. (2021) identified caffeic acid as absent in lower temperature treatment but it presented in higher temperature (>100 °C) and sinapic acid was identified at 160 °C oven heating (Rahman et al., 2021).

Furthermore, ferulic acid and its derivatives seems to be the most abundant compound which might contribute to the improvement in antioxidant properties. Ferulic acids are bound to insoluble structural cellulose or hemicellulose by ester linkages. The treatment might remove the ester-linked ferulic acid from the insoluble cellulose, insoluble hemicellulose, and lignin matrix as has been observed in previous study (Sibhatu, Anuradha Jabasingh, Yimam, & Ahmed, 2021). The temperature at 90 °C–100 °C does not efficiently rupture the cross-linking bond of ferulic acids and even caused a simplification in the structure, thus lowering the flavan-3-ols and phenolic acids. Higher temperatures allow the disruption of cross-linked bonds between insoluble ingredients and phenolic compounds thus creating the possibility to cleave the natural bonds and esterify ferulic acid. Therefore, the increase in yield extracts can be obtained (Sibhatu et al., 2021). Ferulic acid is responsible for DPPH capability due to the presence of phenolic nucleus and unsaturated side chain which form a resonance stabilised phenoxy radical (Connolly et al., 2021; Sibhatu et al., 2021). The role of hydroxycinnamic acid from BSG as an anti-inflammatory effect has been reported, due to its ability to reduce the stimulated cytokine production (McCarthy et al., 2014).

According to (Sibhatu et al., 2021), the major phenolic compounds in BSG are sinapic acid, p-coumaric acid and ferulic acid. Hydroxycinnamic acid is the most abundant phenolic acid from BSG including ferulic acid (FA), p-coumaric acid (p-CA) derivatives, FA derivatives, p-CA, caffeic acid (CA) and CA derivatives. CA regulated the antioxidant activity of phenolic extract from BSG (McCarthy et al., 2013). The presence of certain phenolic compounds in BSG depends on the solvent used (Bonifácio-Lopes et al., 2020). It is identified that extraction with 100% ethanol presented 4-hydroxybenzoic and syringic acids, while ethanol 60% identified the presence of catechin, vanillic acid, 4-hydroxybenzoic, vanillin and p-coumaric acid. Apparently, vanillin, p-coumaric acid, 4-hydroxybenzoic, catechin, ferulic acid and protocatechuic acid were observed in 80% ethanol extract. However, water extraction generated 4-hydroxybenzoic acid, p-coumaric and protocatechuic acid, vanillin, catechin, and vanillic acid. In the current study, methanol was used as a solvent during the extraction. Autoclave with different temperature and time exposure generated different levels of phenolic compounds, reduced certain compounds and induced the formation of certain compounds depending on the temperature used.

4. Conclusion

As hypothesised, AT improved the antioxidant properties in BSG including ORAC, ABTS and FRAP compared to that in untreated BSG. The study showed that the treatments at 90 °C and 100 °C enhanced the FRAP value the most while it reduced the amount of phenolic acid and flavan-3-ols. Meanwhile 110 °C and 130 °C gave the highest impact on ABTS level and multiplied the amount of phenolic acid. Tentative identification showed that AT induced depolymerisation which consequently induced certain compounds in a higher temperature such as benzoic acid and (+)-catechin. As a result, the declining of phenolic compounds at 90 °C and 100 °C was observed while at the same time it improved the antioxidant activities compared to the untreated BSG. Significant increase in antioxidant capacities and phenolic acid was observed at 110 °C and 130 °C treatments due to its ability in disrupting the BSG matrices and releasing the phenolic compounds. The study demonstrated a benefit effect in terms of the utilisation of BSG as food and or nutraceutical ingredients. Therefore, AT potentially provides the benefits for food industries with generating higher biological properties of BSG. Study on the quantitative and qualitative of observed phenolic compounds seems to be important in the near future in order to understand the impact of specific in certain biological properties.

Table 2
Identification of phenolic compounds by LC-MS-MS in treated BSG with different temperature.

Phenolic compounds	tentative identification	Retention time	MS	MS/MS	Temperature (°C)			
					90	100	110	130
phenolic acid	syringic acid	5182	197.08	153.07	✓	✓	✓	✓
	benzoic acid	5399	121.10	92.01	–	–	✓	✓
	p-coumaric acid	5576	163.06	119.04	✓	✓	✓	✓
	di-ferulic acid dimer	5731	387.10	149.07/134.01	✓	✓	✓	✓
	sinapic acid	6054	223.02	179.01	✓	✓	✓	✓
	ferulic acid	6248	193.02	134.01	✓	✓	✓	✓
	decarboxylated diferulic acid	6303	341.11	193.01/134.01	✓	✓	✓	✓
	di-ferulic acid isomers	7707	385.08	282.09/148.03	✓	✓	✓	✓
flavan-3-ols	(+)-catechin	3783	289.03	245.01	–	–	✓	✓
	(−)-epicatechin	5305	289.03	245.01	✓	✓	✓	✓

√ -present; - -absent.

CRediT authorship contribution statement

Joncer Naibaho: Conceptualization, Methodology, Validation, Formal analysis, Investigation, Writing – original draft, preparation, Writing – review & editing, Funding acquisition. **Aneta Wojdyło:** Methodology, Methodology, Validation, Formal analysis, Writing – review & editing. **Małgorzata Korzeniowska:** Conceptualization, Resources, Writing – review & editing, Supervision, Project administration, Funding acquisition. **Oskar Laaksonen:** Writing – review & editing, Project administration, Funding acquisition. **Maike Föste:** Writing – review & editing, Project administration, Funding acquisition. **Mary-Liis Kütt:** Writing – review & editing, Project administration, Funding acquisition. **Baoru Yang:** Writing – review & editing, Project administration, Funding acquisition.

Declaration of competing interest

Authors declare no conflict of interest.

Acknowledgements

This work was supported by UPWR 2.0, international and interdisciplinary programme of development of Wrocław University of Environmental and Life Sciences, co-financed by the European Social Fund under the Operational Programme Knowledge Education Development 2014–2020: Axis III Higher education for the economy and development; Action 3.5. Comprehensive programmes for schools of higher education (POWR.03.05.00–00-Z062/18).

It was also supported by ERA-NET CO-FUND Horizon 2020 - FACCE SURPLUS Sustainable and Resilient Agriculture for Food and Non-Food Systems and PROWASTE Protein-fibre biorefinery for scattered material streams (2019–2021).

References

Barbosa-Pereira, L., Bilbao, A., Vilches, P., Angulo, I., LLuis, J., Fité, B., et al. (2014). Brewery waste as a potential source of phenolic compounds: Optimisation of the extraction process and evaluation of antioxidant and antimicrobial activities. *Food Chemistry, 145*, 191–197. https://doi.org/10.1016/j.foodchem.2013.08.033

Benzie, I. F. F., & Strain, J. J. (1996). The ferric reducing ability of plasma (FRAP) as a measure of "antioxidant power": The FRAP assay. *Analytical Biochemistry, 239*(1), 70–76. https://doi.org/10.1006/abio.1996.0292

Bonifácio-Lopes, T., Teixeira, J. A., & Pintado, M. (2020). Current extraction techniques towards bioactive compounds from brewer's spent grain – a review. *Critical Reviews in Food Science and Nutrition, 60*(16), 2730–2741. https://doi.org/10.1080/10408398.2019.1655632

Bonifácio-Lopes, T., Vilas-Boas, A., Machado, M., Costa, E. M., Silva, S., Pereira, R. N., et al. (2022). Exploring the bioactive potential of brewers spent grain ohmic extracts. *Innovative Food Science & Emerging Technologies, 76*, Article 102943. https://doi.org/10.1016/j.ifset.2022.102943

Budaraju, S., Mallikarjunan, K., Annor, G., Schoenfuss, T., & Raun, R. (2018). Effect of pre-treatments on the antioxidant potential of phenolic extracts from barley malt rootlets. *Food Chemistry, 266*, 31–37. https://doi.org/10.1016/j.foodchem.2018.05.110

Connolly, A., Cermeño, M., Alashi, A. M., Aluko, R. E., & FitzGerald, R. J. (2021). Generation of phenolic-rich extracts from brewers' spent grain and characterisation of their in vitro and in vivo activities. *Innovative Food Science & Emerging Technologies, 68*, Article 102617. https://doi.org/10.1016/j.ifset.2021.102617

Connolly, A., Cermeño, M., Crowley, D., O'Callaghan, Y., O'Brien, N. M., & FitzGerald, R. J. (2019). Characterisation of the in vitro bioactive properties of alkaline and enzyme extracted brewers' spent grain protein hydrolysates. *Food Research International, 121*, 524–532. https://doi.org/10.1016/j.foodres.2018.12.008

Cooray, S. T., & Chen, W. N. (2018). Valorization of brewer's spent grain using fungi solid-state fermentation to enhance nutritional value. *Journal of Functional Foods, 42*, 85–94. https://doi.org/10.1016/j.jff.2017.12.027

Crowley, D., O'Callaghan, Y., McCarthy, A. L., Connolly, A., Fitzgerald, R. J., & O'Brien, N. M. (2017). Aqueous and enzyme-extracted phenolic compounds from brewers' spent grain (BSG): Assessment of their antioxidant potential: CROWLEY et al. *Journal of Food Biochemistry, 41*(3), Article e12370. https://doi.org/10.1111/jfbc.12370

Fărcaş, A. C., Socaci, S. A., Dulf, F. V., Tofană, M., Mudura, E., & Diaconeasa, Z. (2015). 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. *Journal of Cereal Science, 64*, 34–42. https://doi.org/10.1016/j.jcs.2015.04.003

Heredia-Sandoval, N. G., Granados-Nevárez, M. del C., Calderón de la Barca, A. M., Vásquez-Lara, F., Malunga, L. N., Apea-Bah, F. B., et al. (2020). Phenolic acids, antioxidant capacity, and estimated glycemic index of cookies added with brewer's spent grain. *Plant Foods for Human Nutrition, 75*(1), 41–47. https://doi.org/10.1007/s11130-019-00783-1

Kumari, B., Tiwari, B. K., Walsh, D., Griffin, T. P., Islam, N., Lyng, J. G., et al. (2019). Impact of pulsed electric field pre-treatment on nutritional and polyphenolic contents and bioactivities of light and dark brewer's spent grains. *Innovative Food Science & Emerging Technologies, 54*, 200–210. https://doi.org/10.1016/j.ifset.2019.04.012

Li, S., Chen, G., Qiang, S., Tang, D., Chen, Y., Zhang, Z., et al. (2019). Intensifying soluble dietary fiber production and properties of soybean curd residue via autoclaving treatment. *Bioresource Technology Reports, 7*, Article 100203. https://doi.org/10.1016/j.biteb.2019.100203

Lynch, K. M., Steffen, E. J., & Arendt, E. K. (2016). Brewers' spent grain: A review with an emphasis on food and health: Brewers' spent grain: A review with an emphasis on food and health. *Journal of the Institute of Brewing, 122*(4), 553–568. https://doi.org/10.1002/jib.363

Martín-García, B., Tylewicz, U., Verardo, V., Pasini, F., Gómez-Caravaca, A. M., Caboni, M. F., et al. (2020). Pulsed electric field (PEF) as pre-treatment to improve the phenolic compounds recovery from brewers' spent grains. *Innovative Food Science & Emerging Technologies, 64*, Article 102402. https://doi.org/10.1016/j.ifset.2020.102402

McCarthy, A. L., O'Callaghan, Y. C., Connolly, A., Piggott, C. O., FitzGerald, R. J., & O'Brien, N. M. (2012). Phenolic extracts of brewers' spent grain (BSG) as functional ingredients – assessment of their DNA protective effect against oxidant-induced DNA single strand breaks in U937 cells. *Food Chemistry, 134*(2), 641–646. https://doi.org/10.1016/j.foodchem.2012.02.133

McCarthy, A. L., O'Callaghan, Y. C., Connolly, A., Piggott, C. O., FitzGerald, R. J., & O'Brien, N. M. (2014). Phenolic-enriched fractions from brewers' spent grain possess cellular antioxidant and immunomodulatory effects in cell culture model systems: Anti-inflammatory effects of brewers' spent grain phenolic extracts. *Journal of the Science of Food and Agriculture, 94*(7), 1373–1379. https://doi.org/10.1002/jsfa.6421

McCarthy, A. L., O'Callaghan, Y. C., Neugart, S., Piggott, C. O., Connolly, A., Jansen, M. A. K., et al. (2013). The hydroxycinnamic acid content of barley and brewers' spent grain (BSG) and the potential to incorporate phenolic extracts of BSG as antioxidants into fruit beverages. *Food Chemistry, 141*(3), 2567–2574. https://doi.org/10.1016/j.foodchem.2013.05.048

Meneses, N. G. T., Martins, S., Teixeira, J. A., & Mussatto, S. I. (2013). Influence of extraction solvents on the recovery of antioxidant phenolic compounds from brewer's spent grains. *Separation and Purification Technology, 108*, 152–158. https://doi.org/10.1016/j.seppur.2013.02.015

Moreira, M. M., Morais, S., Carvalho, D. O., Barros, A. A., Delerue-Matos, C., & Guido, L. F. (2013). Brewer's spent grain from different types of malt: Evaluation of the antioxidant activity and identification of the major phenolic compounds. *Food Research International, 54*(1), 382–388. https://doi.org/10.1016/j.foodres.2013.07.023

Naibaho, J., Butula, N., Jonuzi, E., Korzeniowska, M., Laaksonen, O., Föste, M., et al. (2022). Potential of brewers' spent grain in yogurt fermentation and evaluation of its impact in rheological behaviour, consistency, microstructural properties and acidity profile during the refrigerated storage. *Food Hydrocolloids, 125*, Article 107412. https://doi.org/10.1016/j.foodhyd.2021.107412

Naibaho, J., & Korzeniowska, M. (2021). Brewers' spent grain in food systems: Processing and final products quality as a function of fiber modification treatment. *Journal of Food Science, 86*(5), 1532–1551. https://doi.org/10.1111/1750-3841.15714

Naibaho, J., Korzeniowska, M., Wojdyło, A., Figiel, A., Yang, B., Laaksonen, O., et al. (2021a). The potential of spent barley as a functional food ingredient: Study on the comparison of dietary fiber and bioactivity. *Proceedings, 70*(86), 6. https://doi.org/10.3390/foods_2020-08486

Naibaho, J., Korzeniowska, M., Wojdyło, A., Figiel, A., Yang, B., Laaksonen, O., et al. (2021b). Fiber modification of brewers' spent grain by autoclave treatment to improve its properties as a functional food ingredient. *Lebensmittel-Wissenschaft und -Technologie, 149*, Article 111877. https://doi.org/10.1016/j.lwt.2021.111877

Nigam, P. S. (2017). An overview: Recycling of solid barley waste generated as a by-product in distillery and brewery. *Waste Management, 62*, 255–261. https://doi.org/10.1016/j.wasman.2017.02.018

Nocente, F., Taddei, F., Galassi, E., & Gazza, L. (2019). Upcycling of brewers' spent grain by production of dry pasta with higher nutritional potential. *Lebensmittel-Wissenschaft und -Technologie, 114*, Article 108421. https://doi.org/10.1016/j.lwt.2019.108421

Ohra-aho, T., Niemi, P., Aura, A.-M., Orlandi, M., Poutanen, K., Buchert, J., et al. (2016). Structure of brewer's spent grain lignin and its interactions with gut microbiota in vitro. *Journal of Agricultural and Food Chemistry, 64*(4), 812–820. https://doi.org/10.1021/acs.jafc.5b05535

Ou, B., Chang, T., Huang, D., & Prior, R. L. (2013). Determination of total antioxidant capacity by oxygen radical absorbance capacity (ORAC) using fluorescein as the fluorescence probe: First action 2012.23. *Journal of AOAC International, 96*(6), 1372–1376. https://doi.org/10.5740/jaoacint.13-175

Parekh, I., Khanvilkar, A., & Naik, A. (2017). Barley-wheat brewers' spent grain: A potential source of antioxidant rich lipids. *Journal of Food Processing and Preservation, 41*(6), Article e13244. https://doi.org/10.1111/jfpp.13244

Patrignani, M., Brantsen, J. F., Awika, J. M., & Conforti, P. A. (2021). Application of a novel microwave energy treatment on brewers' spent grain (BSG): Effect on its

functionality and chemical characteristics. *Food Chemistry, 346*, Article 128935. https://doi.org/10.1016/j.foodchem.2020.128935

Rahman, M. J., Malunga, L. N., Eskin, M., Eck, P., Thandapilly, S. J., & Thiyam-Hollander, U. (2021). Valorization of heat-treated brewers' spent grain through the identification of bioactive phenolics by UPLC-PDA and evaluation of their antioxidant activities. *Frontiers in Nutrition, 8*, Article 634519. https://doi.org/10.3389/fnut.2021.634519

Re, R., Pellegrini, N., Proteggente, A., Pannala, A., Yang, M., & Rice-Evans, C. (1999). Antioxidant activity applying an improved ABTS radical cation decolorization assay. *Free Radical Biology and Medicine, 26*(9–10), 1231–1237. https://doi.org/10.1016/S0891-5849(98)00315-3

Sibhatu, H. K., Anuradha Jabasingh, S., Yimam, A., & Ahmed, S. (2021). Ferulic acid production from brewery spent grains, an agro-industrial waste. *Lebensmittel-Wissenschaft und -Technologie, 135*, Article 110009. https://doi.org/10.1016/j.lwt.2020.110009

Socaci, S. A., Fărcaş, A. C., Diaconeasa, Z. M., Vodnar, D. C., Rusu, B., & Tofană, M. (2018). Influence of the extraction solvent on phenolic content, antioxidant, antimicrobial and antimutagenic activities of brewers' spent grain. *Journal of Cereal Science, 80*, 180–187. https://doi.org/10.1016/j.jcs.2018.03.006

Steiner, J., Procopio, S., & Becker, T. (2015). Brewer's spent grain: Source of value-added polysaccharides for the food industry in reference to the health claims. *European Food Research and Technology, 241*(3), 303–315. https://doi.org/10.1007/s00217-015-2461-7

Tan, Y. X., Mok, W. K., Lee, J., Kim, J., & Chen, W. N. (2019). Solid state fermentation of brewers' spent grains for improved nutritional profile using Bacillus subtilis WX-17. *Fermentation, 5*(3), 52. https://doi.org/10.3390/fermentation5030052

Tkacz, K., Wojdyło, A., Turkiewicz, I. P., & Nowicka, P. (2021). Triterpenoids, phenolic compounds, macro- and microelements in anatomical parts of sea buckthorn (Hippophaë rhamnoides L.) berries, branches and leaves. *Journal of Food Composition and Analysis, 103*, 104107. https://doi.org/10.1016/j.jfca.2021.104107, 104107.

Torbica, A., Škrobot, D., Janić Hajnal, E., Belović, M., & Zhang, N. (2019). Sensory and physico-chemical properties of wholegrain wheat bread prepared with selected food by-products. *Lebensmittel-Wissenschaft und -Technologie, 114*, Article 108414. https://doi.org/10.1016/j.lwt.2019.108414

Turkiewicz, I. P., Tkacz, K., Nowicka, P., Michalska-Ciechanowska, A., Lech, K., & Wojdyło, A. (2021). Physicochemical characterization and biological potential of Japanese quince polyphenol extract treated by different drying techniques. *Lebensmittel-Wissenschaft und -Technologie, 152*, Article 112247. https://doi.org/10.1016/j.lwt.2021.112247

Turkiewicz, I. P., Wojdyło, A., Tkacz, K., Nowicka, P., Golis, T., & Bąbelewski, P. (2020). ABTS on-line antioxidant, α-amylase, α-glucosidase, pancreatic lipase, acetyl- and butyrylcholinesterase inhibition activity of chaenomeles fruits determined by polyphenols and other chemical compounds. *Antioxidants, 9*(1), 60. https://doi.org/10.3390/antiox9010060

Verni, M., Pontonio, E., Krona, A., Jacob, S., Pinto, D., Rinaldi, F., et al. (2020). Bioprocessing of brewers' spent grain enhances its antioxidant activity: Characterization of phenolic compounds and bioactive peptides. *Frontiers in Microbiology, 11*, 1831. https://doi.org/10.3389/fmicb.2020.01831

Wojdyło, A., Figiel, A., Lech, K., Nowicka, P., & Oszmiański, J. (2014). Effect of convective and vacuum–microwave drying on the bioactive compounds, color, and antioxidant capacity of sour cherries. *Food and Bioprocess Technology, 7*(3), 829–841. https://doi.org/10.1007/s11947-013-1130-8

Zago, E., Tiller, C., De Leener, G., Nandasiri, R., Delporte, C., Bernaerts, K. V., et al. (2022). Sustainable production of low molecular weight phenolic compounds from Belgian Brewers' spent grain. *Bioresource Technology Reports, 17*, Article 100964. https://doi.org/10.1016/j.biteb.2022.100964

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OŚWIADCZENIE

Oświadczam, że w pracy

Naibaho J., Wojdyło A., Korzeniowska M., Laaksonen O., Föste M., Kütt M.-L., & Yang B. 2022. Antioxidant activities and polyphenolic identification by UPLC-MS/MS of autoclaved brewers' spent grain. LWT – Food Science and Technology 163, 113612. https://doi.org/10.1016/j.lwt.2022.113612 (autorzy, rok wydania, tytuł, czasopismo lub wydawca, tom, strony)

mój udział polegał na Konceptualizacja, Metodologia, Walidacja, Analiza formalna, Badanie, Pisanie – oryginalny projekt, przygotowanie, Pisanie – recenzja i redakcja, Pozyskiwanie funduszy (opisać szczegółowo swój własny udział w powstaniu pracy, np. wykonaniu doświadczeń techniką ....., analizie statystycznej wyników eksperymentów zilustrowanych na ryc. ...., przygotowaniu tekstu manuskryptu zamieszczonego w rozdziale ....., kierowaniu projektem naukowym obejmującym badania opisane w tej pracy, itp.).

Wroclaw, April 12 2023

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data i podpis

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Wroclaw, April 12, 2023
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OŚWIADCZENIE

Oświadczam, że w pracy

Naibaho J., Wojdyło A., Korzeniowska M., Laaksonen O., Föste M., Kütt M.-L., & Yang B. 2022. Antioxidant activities and polyphenolic identification by UPLC-MS/MS of autoclaved brewers' spent grain. LWT – Food Science and Technology 163, 113612. https://doi.org/10.1016/j.lwt.2022.113612 (autorzy, rok wydania, tytuł, czasopismo lub wydawca, tom, strony)

mój udział polegał na Metodologia, Metodologia, Walidacja, Analiza formalna, Pisanie – recenzja i redakcja (opisać szczegółowo swój własny – a nie Kandydata udział w powstaniu pracy, np. wykonaniu doświadczeń techniką ....., analizie statystycznej wyników eksperymentów zilustrowanych na ryc. ...., przygotowaniu tekstu manuskryptu zamieszczonego w rozdziale ....., kierowaniu projektem naukowym obejmującym badania opisane w tej pracy, itp.).

Wroclaw, April 12, 2023

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mój udział polegał na Konceptualizacja, zasoby, pisanie – recenzja i redakcja, nadzór, administracja projektem i pozyskiwanie funduszy (opisać szczegółowo swój własny – a nie Kandydata udział w powstaniu pracy, np. wykonaniu doświadczeń techniką ....., analizie statystycznej wyników eksperymentów zilustrowanych na ryc. ...., przygotowaniu tekstu manuskryptu zamieszczonego w rozdziale ....., kierowaniu projektem naukowym obejmującym badania opisane w tej pracy, itp.).

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Naibaho J., Wojdyło A., Korzeniowska M., Laaksonen O., Föste M., Kütt M.-L., & Yang B. 2022. Antioxidant activities and polyphenolic identification by UPLC-MS/MS of autoclaved brewers' spent grain. LWT – Food Science and Technology 163, 113612. https://doi.org/10.1016/j.lwt.2022.113612. (autorzy, rok wydania, tytuł, czasopismo lub wydawca, tom, strony)

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mój udział polegał na: Pisanie – recenzja i redakcja, administrowanie projektem i pozyskiwanie funduszy (opisać szczegółowo swój własny – a nie Kandydata udział w powstaniu pracy, np. wykonaniu doświadczeń techniką ....., analizie statystycznej wyników eksperymentów zilustrowanych na ryc. ...., przygotowaniu tekstu manuskryptu zamieszczonego w rozdziale ....., kierowaniu projektem naukowym obejmującym badania opisane w tej pracy, itp.).

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Oświadczam, że w Naibaho J., Wojdyło A., Korzeniowska M., Laaksonen O., Föste M., Kütt M.-L., & Yang B. 2022. Antioxidant activities and polyphenolic identification by UPLC-MS/MS of autoclaved brewers' spent grain. LWT – Food Science and Technology 163, 113612. https://doi.org/10.1016/j.lwt.2022.113612 (autorzy, rok wydania, tytuł, czasopismo lub wydawca, tom, strony)

mój udział polegał na Pisanie – recenzja i redagowanie, administrowanie projektem i pozyskiwanie funduszy (opisać szczegółowo swój własny – a nie Kandydata udział w powstaniu pracy, np. wykonaniu doświadczeń techniką ....., analizie statystycznej wyników eksperymentów zilustrowanych na ryc. ...., przygotowaniu tekstu manuskryptu zamieszczonego w rozdziale ....., kierowaniu projektem naukowym obejmującym badania opisane w tej pracy, itp.).

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Naibaho J., Wojdyło A., Korzeniowska M., Laaksonen O., Föste M., Kütt M.-L., & Yang B. 2022. Antioxidant activities and polyphenolic identification by UPLC-MS/MS of autoclaved brewers' spent grain. LWT – Food Science and Technology 163, 113612. https://doi.org/10.1016/j.lwt.2022.113612 (autorzy, rok wydania, tytuł, czasopismo lub wydawca, tom, strony)

mój udział polegał na:

Pisanie – recenzja i redakcja, Zarządzanie projektem, Pozyskiwanie funduszy (opisać szczegółowo swój własny – a nie Kandydata udział w powstaniu pracy, np. wykonaniu doświadczeń techniką ....., analizie statystycznej wyników eksperymentów zilustrowanych na ryc. ...., przygotowaniu tekstu manuskryptu zamieszczonego w rozdziale ....., kierowaniu projektem naukowym obejmującym badania opisane w tej pracy, itp.).

March 27, 2023

.....

data i podpis

Publication 4

International Journal of Food Science +Technology

Institute of Food Science +Technology ifst

Original article

Chemical compositions, antioxidant activities and techno-functionality of spent grain treated by autoclave treatment: evaluation of water and temperature levels

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(Received 1 July 2022; Accepted in revised form 19 August 2022)

Summary Autoclave heating (AH) has been applied to modify the dietary fibre composition of dried brewers' spent grain (BSG) flour, which required multiple drying processes. The current study aimed to investigate the influence of the thermal levels and water ratio on AH, as an alternative, in altering the chemical compositions, antioxidant properties, and functionality of undried fresh BSG. The results showed that AH converted the saturated fatty acids into polyunsaturated fatty acids. AH reduced ketones and furans regardless of the water ratio while the amounts of aldehydes, alcohols, alkenes, and fatty acids depended on the water ratio. The elimination and formation of several volatile compounds were identified due to the AH depending on the water ratio. The total flavan-3-ols, antioxidant activities, and water-holding capacity of BSG were improved as an impact of thermal elevation and regardless of the water ratio. In conclusion, AH treatment on fresh, undried BSG showed a beneficial performance in improving the quality of BSG for further valorisation as a value-added by-product.

Keywords Agroindustrial by-products, fatty acids profile, oil-holding capacity, polyphenolic quantification, volatile compounds, water-holding capacity.

Introduction

Brewers' spent grain (BSG) has been reported for its nutritional value as well as biological properties due to the presence of polyphenolic compounds, protein, fatty acids, and dietary fibre (Lynch *et al.*, 2016; Naibaho & Korzeniowska, 2021a). The presence of polyphenolic compounds, protein, fatty acids, and dietary fibre is responsible for immunomodulatory properties as well as antimicrobial and anti-inflammatory activity. In addition to that, BSG possesses antioxidant activities such as lipid peroxidation, deoxyribose scavenging activity, superoxide dismutase, catalase, glutathione, DPPH, FRAP, and ABTS (Lynch *et al.*, 2016; Naibaho & Korzeniowska, 2021a). Although BSG possesses high potential as a food and nutraceutical ingredient, the majority of BSG still remains unused as land waste and a small fraction is used as animal/fish feed and fertiliser (Skendi *et al.*, 2018; Lao *et al.*, 2020). BSG is a complex material which is dominated by insoluble dietary fibre (Naibaho *et al.*, 2021). However, the biological properties of BSG are mostly studied due to the presence of phenolic compounds, followed by protein (Wen *et al.*, 2019; Naibaho *et al.*, 2022a, 2022b). Phenolic compounds exist in a hydroxyl group of dietary fibre while protein is entrapped in the vacuole cell wall of BSG materials, which consists of dietary fibre (Naibaho & Korzeniowska, 2021a). Besides the fact that BSG increased the nutritional value of BSG-added food products, BSG tended to inversely impact food processing aspects such as technological processing and mechanical properties which consequently diminished the physical appearance of the final products, as well as

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doi:10.1111/ijfs.16042

sensory acceptability (Naibaho & Korzeniowska, 2021a).

Aiming to improve the yield of targeted compounds as well as its biological properties, several studies have also been conducted including solid-state fermentation (Cooray & Chen, 2018; Tan *et al*., 2019), pulsed electric field (Martín-García *et al*., 2020), pH elevation (Connolly *et al*., 2021), enzyme treatments (Connolly *et al*., 2019), and the combination of thermal and enzyme treatment (Budaraju *et al*., 2018). One of the most common physical treatments on BSG is thermal exposure such as steam explosion, microwave superheating, and autoclave treatment (Coelho *et al*., 2014; Kemppainen *et al*., 2016; Naibaho *et al*., 2021, 2022b). Temperature elevation improved the yield and enhanced the functionality of arabinoxylans and arabinoxylan-oligosaccharides as well as the availability of phenolic compounds (Budaraju *et al*., 2018). Furthermore, involvement of high pressure and temperature such as steam explosion, autoclave heating, high-pressure homogenisation, extrusion, and mechanical treatment improved the fibre functionality (Xie *et al*., 2017; Kieserling *et al*., 2019; Yan *et al*., 2019; Li *et al*., 2019a, 2019b). Furthermore, the addition of BSG in food products was limited due to its insoluble dietary fibre in disrupting the food matrix formation. Therefore, dietary fibre modification of BSG was emphasised (Naibaho & Korzeniowska, 2021a).

Autoclave heating treatment (AH) on rehydrated dried BSG was reported for its ability to degrade insoluble dietary fibre and convert it into soluble dietary fibre (Naibaho *et al*., 2021), thus improving the biological properties and polyphenolic composition (Naibaho *et al*., 2022b). Moreover, AH improved the functionality of dietary fibre from soybean curd residue (Li *et al*., 2019b), increased the resistant starch content in rice grains (Zheng *et al*., 2020), and enhanced the solubility-related properties and stability of the colloidal suspension (Nawaz *et al*., 2020). Usually, BSG is dried at a high temperature and/or stored at freeze temperature before it is used for certain treatments that require energy consumption. Treatment on fresh BSG is seemingly challenging due to its more practical use for several stakeholders and low-cost production. Treatment on wet or fresh BSG has been conducted in order to improve the protein and dietary fibre composition (He *et al*., 2019) (He *et al*., 2019). AH is a simple, easy-to-operate, and low-cost instrument; it is thus promising in BSG treatment, which involves high temperature and pressure elevation. AH has never been applied on wet BSG, particularly its impact on functionality, chemical constituents, and biological properties due to the different ratio of sludging. Most of the studied treatments were evaluated on phenolic compounds and/or protein composition in addition to dietary fibre composition. The influence of the energy input such as temperature and pressure on the fatty acid profile and volatile compounds of BSG has never been investigated. Volatile profile is an important parameter due to its direct impact on food product application, in terms of the valorisation of BSG as a food ingredient.

This study aimed to evaluate the influence of AH at different thermal exposures on undried fresh BSG properties including its functionality, polyphenolic composition, fatty acid profile, aromatic compounds, and *in vitro* antioxidant activities. Based on preexperiments, the addition of water in AH on BSG is technically needed to allow a homogenous thermal exposure. However, different amounts of water in fresh BSG generated different viscosities, thus impacting the mixing process and energy. Minimising water use in industries is suggested in order to achieve more sustainable treatments and implement cleaner processing methods (Bailone *et al*., 2022). Therefore, the current study investigated different levels of water addition into BSG slurry on AH. It was hypothesised that thermal decomposition of the BSG matrix directly altered the hydroxyl groups, which are polyphenolic compounds, as well as its antioxidant properties due to the degradation of dietary fibre. Previous studies investigated the influence of thermal degradation on protein extraction. However, degradation of the vacuole cell of BSG might release fatty acids, which has never been evaluated. Therefore, the current study evaluated the fatty acid composition of BSG in addition to volatile compounds as well as water-holding capacity and oil-holding capacity as a function of dietary fibre degradation.

Materials and methods

Materials

Fresh BSG with a moisture content of approximately 70–75% was supplied by a local brewery in Wroclaw, Poland. BSG was ground to pass 0.2 mm and kept in a polyethylene bag. BSG then was stored at a freezing temperature prior to the experiment.

UPLC-grade water was prepared by using the HLP SMART 1000s system (Hydrolab, Gdansk, Poland). Immediately, before use, the water was filtered using a 0.22 μm membrane filter. Trolox (6-hydro-2,5,7,8-tetramethylchroman-2-carboxylic acid) was purchased from Sigma-Aldrich (Steinheim, Germany). All the chemicals used were analytical grade.

Experimental design

BSG was allowed to defrost at room temperature just before the treatment. BSG was mixed properly with distilled water at two different ratios 1:1 and 1:2

(BSG:distilled water, *w*/*v*). Different time exposures on BSG by autoclave treatment identified that 12 min treatment generated a medium impact on the degradation of dietary fibre (Naibaho *et al*., 2021), a higher impact on polyphenolic content at 90 °C, and a medium impact at 110 and 130 °C (Naibaho *et al*., 2022b). Therefore, the current study was conducted at 12 min time exposures at different temperatures (90, 110, and 130 °C) and different water ratios. Untreated fresh BSG was provided for comparison. Therefore, seven samples were obtained. The BSG was then dried by oven drying at 75 °C for 16 h to reach a moisture below 6% (Table 1). The sample was ground using a lab scale blender for 5 min with a 10-s pause every 1 min. Samples were packed into aluminium foil and kept at 10 °C for further analysis.

The impact of the water ratio during the AH was evaluated on volatile compositions. The analysis performed only represented the water ratio, instead of the temperature level. The sample was chosen as the medium temperature treatment, which is 110 °C at two different ratios in comparison to the untreated BSG. Therefore, three different samples were compared for their volatile profiles.

Measurement of fatty acids composition by GC-MS

Total lipid was extracted following the procedures as described previously (Fărcaş *et al*., 2015). Lipid was derivatised into fatty acid methyl esters (FAMEs) following procedures described in a previous study (Nowacki *et al*., 2017). After that, the fatty acid profile was analysed by using a gas chromatograph (GC6890) coupled with a mass spectrometer 5983 MS (Agilent Technologies Inc., Santa Clara, CA, USA) equipped with a quadrupole mass detector. Separation was performed in a capillary column HP-88 (0.25 mm × 100 m) filled with an 88:12 cyanopropyl-aryl poly-siloxane bed with a grain size of 0.2 μm. Helium (flow rate 1 mL min^{-1}) was used as the mobile phase and the sample was injected in the split mode at 4:1. The program was set with an initial temperature of 60 °C for 2 min, heating at 20 °C min^{-1} to reach 180 °C and 3 °C min^{-1} to reach 220 °C. The temperature was held for 15 min. Heating continued to reach 250 °C at a rate of 5 °C min^{-1}, and the temperature was held for 8 min. The spectra were identified using the algorithm of searching the National Institute of Standards and Technology (NIST) library (2008 version).

Analysis of volatile compounds by GC–MS

Dried sample was mixed with distilled water at a ratio of 1:2 and closed properly. The volatiles were isolated by headspace solid-phase microextraction (HS-SPME) following procedures described in previous studies (Dong *et al*., 2013; Ktenioudaki *et al*., 2013; O'Shea *et al*., 2017) by GC–MS 5975 C. The mixture was heated at 60 °C and the fibre (50/30 μm DVB/CAR/PDMS, Supelco) was exposed to the headspace for 30 min. The length of the fibre in the headspace was kept constant. The fibre was exposed to the injector of the gas chromatograph at 250 °C. The fibre was left at the port injector for 5 min to remove the contaminants. Helium was used as the carrier gas (1 mL min^{-1}). Separation of compounds was performed on a DB-5 column (30 m 0.25 mm, df = 0.25 lm, Agilent J&W, USA). The injector, ion source, and interface temperatures were set at 250, 200, and 260 °C, respectively. The mass spectrometer

Table 1 Fatty acids composition of autoclaved BSG

	BSG treatments						
Fatty acids (%)	**Control**	**90 °C/(1:1)**	**110 °C/(1:1)**	**130 °C/(1:1)**	**90 °C/(1:2)**	**110 °C/(1:2)**	**130 °C/(1:2)**
C15:0	–	–	–	29.28 ± 0.00	–	–	–
C16:0	40.22 ± 0.00	21.55 ± 0.00	21.63 ± 0.00	–	21.82 ± 0.00	21.38 ± 0.00	21.82 ± 0.00
C17:0	4.93 ± 0.00	–	–	–	–	–	–
C18:0	–	3.07 ± 0.00	2.67 ± 0.00	–	3.11 ± 0.00	3.07 ± 0.00	3.11 ± 0.00
C18:1 (n-9)	19.39 ± 0.00	17.96 ± 0.00	16.92 ± 0.00	–	17.40 ± 0.00	17.74 ± 0.00	17.40 ± 0.00
C18:2 (n-6)	32.81 ± 0.00	48.84 ± 0.00	51.67 ± 0.00	70.72 ± 0.00	48.92 ± 0.00	49.06 ± 0.00	48.92 ± 0.00
C18:3 (n-3)	2.66 ± 0.00	5.64 ± 0.00	5.40 ± 0.00	–	5.91 ± 0.00	5.90 ± 0.00	5.91 ± 0.00
C20	–	0.86 ± 0.00	–	–	0.83 ± 0.00	0.81 ± 0.00	0.83 ± 0.00
C20:1	–	2.09 ± 0.00	1.71 ± 0.00	–	2.01 ± 0.00	2.04 ± 0.00	2.01 ± 0.00
Total SFA	45.15 ± 0.00^{a}	25.47 ± 0.01^{e}	24.30 ± 0.00^{g}	29.28 ± 0.00^{b}	25.76 ± 0.00^{d}	25.26 ± 0.00^{f}	25.76 ± 0.00^{c}
Total MUFA	19.39 ± 0.00^{e}	20.05 ± 0.00^{a}	18.63 ± 0.00^{f}	0.00^{g}	19.41 ± 0.00^{d}	19.78 ± 0.00^{b}	19.41 ± 0.00^{c}
Total PUFA	35.46 ± 0.00^{g}	54.48 ± 0.00^{e}	57.07 ± 0.00^{b}	70.72 ± 0.00^{a}	54.83 ± 0.00^{d}	54.96 ± 0.00^{c}	54.83 ± 0.00^{f}

The data are shown as mean ± standard deviation with at least duplicate analysis. Letters show the significant differences from other treatment in the same row ($P < 0.05$).

was operated in the electron-impact mode with the electron energy set at 70 eV and scan range of 40–400 m/z. The oven temperature was elevated from 40 to 250 C at a rate of 4 °C min^{-1}, and the temperature was held constant for 5 min. The peak area was measured either by full scanning or by choosing specific fragments. The volatile compounds were tentatively identified using the spectra of reference compounds from NIST.

Identification of polyphenolic by UPLC–MS/MS and *in vitro* antioxidant activities

Methanol extracts of BSG were prepared following the procedures as described previously (Turkiewicz *et al.*, 2020b) with duplicates. *In vitro* antioxidant capabilities for ABTS and FRAP (Benzie & Strain, 1996; Re *et al.*, 1999) in triplicate for duplicate extracts. The identification and quantification of flavan-3-ols and phenolic acids were performed by liquid chromatography–tandem mass spectrometry (LC–MS-MS) following procedures as described in the previous studies (Turkiewicz *et al.*, 2020a, 2021; Tkacz *et al.*, 2021). The assessment was performed in duplicate.

Analysis of techno-functional properties

The water-holding capacity (WHC) and oil-holding capacity (OHC) were performed to represent the techno-functionality of BSG following the procedures as described in a previous study (Ktenioudaki *et al.*, 2013).

Statistical analysis

Statistical analysis was conducted using one-way analysis of variance (ANOVA) followed by Tukey's *post hoc* test in Statistica software version 13.5.0.17.

Results and discussion

Influence of AH on fatty acid compositions

The fatty acid composition of BSG is presented in Table 1. In general, the AH treatment reduced the amount of saturated fatty acids (SFA) and increased the level of polyunsaturated fatty acids (PUFA). The majority of AH improved the amount of monounsaturated fatty acids (MUFA) except on the ratio of 1:1 at 110 and 130 °C. The study revealed that AH decreased C17:0 at all temperatures and ratios. However, the formation of C15:0 was identified at 130 °C (1:1), C18:0 and C20:1 were observed in all treatments except at 130 °C (1:1), and C20:0 was observed at a 1:2 ratio and at 90 °C (1:1). Remarkably, the treatment at 130 °C (1:1) discharged the majority of fatty acid compared to that in untreated BSG. The results demonstrated that untreated BSG is dominated by C16:0, which is SFA; meanwhile, AH-treated BSG is dominated by C18:2 (n-6). However, in total, both treated and untreated BSG is dominated by PUFA. This result is aligned with the previous reports which identified that fatty acid of BSG is dominated by PUFA (Fărcaş *et al.*, 2015; Balogun *et al.*, 2017; Mallen & Najdanovic-Visak, 2018).

The results showed that AH allowed the rearrangement and/or depolymerisation of SFA into UFA. The modification of SFA into UFA in the current study also might be due to the release of UFA from the polysaccharides main chain due to the thermal exposure as observed previously (Rahman *et al.*, 2021). It has been reported previously that higher temperatures increased the amount of UFA and reduced the amount of SFA (Mallen & Najdanovic-Visak, 2018) due to the increasing transesterification rate, which consequently improved the mass transfer from the matrix (Mallen & Najdanovic-Visak, 2018). It is widely accepted that PUFA benefits human health while SFA is recognised to induce non-communicable diseases. Therefore, AH on BSG improves the potential application of BSG in nutraceutical and/or functional food.

Impact of AH on the profile of volatile compounds

The impact of AH on volatile compounds of BSG was investigated in one of each ratio group (1:1 and 1:2) at the medium temperature (110 °C), and the result is presented in Table 2. In general, quantitative volatile compounds on BSG are dominated by the aldehydes group. The result showed that AH reduced the amount of ketones, alcohols, and furans and increased the levels of fatty acids and aldehydes. Furthermore, besides the alteration of quantitative amounts of volatile compounds, AT on BSG with different water ratios discharged and formed several volatile compounds on BSG.

AH with a water ratio at 1:2 increased the amount of aldehydes significantly ($P < 0.05$), while a ratio of 1:1 generated the same level as in the control. AH eliminated (E)-2-hexenal regardless of the water ratio, while it presented in untreated BSG. (E)-2-hexenal has been observed as a green leaf volatile, which has antifungal properties and is responsible for an unpleasant odour which deters fungi and insects (Kunishima *et al.*, 2016). This compound might be present in BSG due to the application of pesticides during the plantation and/or storage of the grain prior to the brewing process. The present study demonstrated that AH is able to remove (E)-2-hexenal as a sign of chemical residue during the handling of grain. AH with a lower amount of water addition (1:1) removed (Z)-2-heptenal from BSG, while it was identified in untreated and 1:2

Table 2 Volatile compounds (%) of autoclave heating treated BSG (percentage of peak area)

Compounds	Treatments		
	Control	110 °C/(1:1)	110 °C/(1:2)
Aldehydes			
Butanal, 3-methyl-	4.48 ± 0.01	4.29 ± 0.23	4.23 ± 0.08
Pentanal	1.19 ± 0.01	1.33 ± 0.01	2.39 ± 0.03
Hexanal	17.30 ± 0.08	11.91 ± 0.06	16.18 ± 0.10
2-Hexenal, (E)-	0.55 ± 0.00	–	–
Heptanal	1.26 ± 0.01	1.62 ± 0.02	1.24 ± 0.06
2-Heptenal, (Z)-	1.13 ± 0.01	–	0.55 ± 0.03
2,4-Heptadienal, (E,E)-	0.53 ± 0.01	0.31 ± 0.03	0.22 ± 0.01
Octanal	–	1.34 ± 0.01	1.92 ± 0.07
2-Octenal, (E)-	3.63 ± 0.01	1.79 ± 0.11	2.42 ± 0.04
Nonanal	11.13 ± 0.15	6.17 ± 0.07	10.68 ± 0.06
2-Nonenal, (E)-	3.06 ± 0.01	0.99 ± 0.04	2.04 ± 0.02
2,4-Nonadienal, (E,E)-	0.88 ± 0.01	0.51 ± 0.02	0.49 ± 0.03
Decanal	1.92 ± 0.02	1.58 ± 0.04	1.74 ± 0.00
Dodecanal	–	0.21 ± 0.01	–
2,4-Dodecadienal, (E,E)-	–	–	0.36 ± 0.01
2,4-Decadienal, (E,E)-	–	2.75 ± 0.05	1.97 ± 0.07
Undecanal	0.36 ± 0.00	0.20 ± 0.01	–
Benzaldehyde	4.93 ± 0.03	5.57 ± 0.07	2.38 ± 0.02
Benzeneacetaldehyde	7.11 ± 0.09	4.47 ± 0.00	4.27 ± 0.07
Ketones			
2-Hexanone, 5-methyl-	–	–	0.51 ± 0.01
2-Heptanone	1.24 ± 0.05	1.30 ± 0.02	0.62 ± 0.01
5-Hepten-2-one, 6-methyl-	0.61 ± 0.05	0.38 ± 0.02	0.21 ± 0.01
3-Octen-2-one, (E)-	1.96 ± 0.14	0.70 ± 0.01	–
3,5-Octadien-2-one, (E,E)-	7.52 ± 0.47	3.41 ± 0.04	4.72 ± 0.05
5,9-Undecadien-2-one, 6,10-dimethyl-, (E)-	0.44 ± 0.02	0.48 ± 0.01	–
2(3H)-Furanone, 5-heptyldihydro-	0.62 ± 0.02	0.43 ± 0.02	0.22 ± 0.01
Alcohols			
Ethanol, 2-phenoxy-	–	0.32 ± 0.01	0.55 ± 0.01
2,4-Hexadien-1-ol	–	–	0.56 ± 0.03
2-Hexyn-1-ol	–	–	0.49 ± 0.01
1-Octen-3-ol	1.80 ± 0.14	1.57 ± 0.02	0.59 ± 0.03
2-Octen-1-ol, (Z)-	0.43 ± 0.02	–	0.22 ± 0.01
3,5-Octadien-2-ol	–	–	0.51 ± 0.03
Nona-3,5-dien-2-ol	0.56 ± 0.02	–	–
Hept-2-en-1-ol	0.39 ± 0.01	–	–
4,4,6-Trimethyl-cyclohex-2-en-1-ol	1.10 ± 0.06	0.29 ± 0.00	–
2-Butyl-2,7-octadien-1-ol	0.57 ± 0.04	–	–
1-Tetradecanol	0.38 ± 0.03	0.23 ± 0.00	–
1-Hexadecanol	1.11 ± 0.09	1.66 ± 0.02	–
2-Methoxy-4-vinylphenol	–	0.18 ± 0.00	–
Furans			
Furan, 2-pentyl-	7.60 ± 0.46	6.85 ± 0.08	6.07 ± 0.01
Furfural	–	0.31 ± 0.02	0.90 ± 0.03
Alkane			
Tridecane	4.72 ± 0.05	5.72 ± 0.07	4.18 ± 0.02
1-Tridecene	–	0.36 ± 0.02	–
Tetradecane, 2,6,10-trimethyl-	1.98 ± 0.04	0.45 ± 0.03	0.21 ± 0.00
Tetradecane	1.12 ± 0.05	1.23 ± 0.01	0.81 ± 0.03
1-Pentadecene	–	1.38 ± 0.01	0.73 ± 0.01
3-Heptadecene, (Z)-	–	0.19 ± 0.01	–
Nonadecane	0.41 ± 0.01	0.45 ± 0.02	–
Dodecane	3.39 ± 0.09	3.66 ± 0.02	3.37 ± 0.01
Octadecane, 3-ethyl-5-(2-ethylbutyl)-	–	0.40 ± 0.02	–
Undecane	–	0.25 ± 0.00	0.46 ± 0.01
Fatty acids			
Acetic acid, cyano-	–	–	0.88 ± 0.01
Hexanoic acid	0.95 ± 0.05	1.09 ± 0.01	–
Hexanoic acid, 1-cyclopentylethyl ester	–	0.18 ± 0.01	–
n-hexadecanoic acid	–	–	1.18 ± 0.06
Other			
D-Limonene	1.61 ± 0.09	2.17 ± 0.03	1.53 ± 0.01
Benzene, 1-methyl-3-(1-methylethyl)-	–	0.52 ± 0.01	0.38 ± 0.02
Benzene, 1,3-bis (1,1-dimethylethyl)-	–	–	0.24 ± 0.01
1R-α-Pinene	–	0.72 ± 0.01	0.65 ± 0.01
Total			
Aldehydes	59.47 ± 0.07^{b}	63.14 ± 0.23^{ab}	69.24 ± 0.22^{a}
Ketones	12.39 ± 0.63^{a}	6.7 ± 0.01^{b}	6.27 ± 0.07^{b}
Alcohols	6.35 ± 0.11^{a}	4.24 ± 0.04^{b}	2.91 ± 0.08^{c}
Furans	7.60 ± 0.46^{a}	7.17 ± 0.08^{b}	6.97 ± 0.01^{b}
Alkene	11.62 ± 0.02^{b}	14.06 ± 0.01^{a}	9.75 ± 0.07^{c}
Fatty acid	0.95 ± 0.05^{c}	1.28 ± 0.04^{b}	2.06 ± 0.05^{a}
Others	1.61 ± 0.16^{c}	3.41 ± 0.05^{a}	2.79 ± 0.08^{b}

The data are shown as mean ± standard deviation with at least duplicate analysis. Letters show the significant differences from other treatment in the same row ($P < 0.05$).

ratio treated BSG. (Z)-2-heptenal represents green and pungent odour perception in BSG (Dong *et al.*, 2013; Ktenioudaki *et al.*, 2013; Fărcaş *et al.*, 2015). This demonstrated that a lower amount of water (1:1) eliminated the unpleasant odour perception of BSG. AH with a higher water amount (1:2) destroyed the presence of undecanal, while it presented in untreated and 1:1 ratio treated BSG. Undecanal has never been reported on BSG; however, 2-undecenal has been reported in cells immobilised by BSG (Mallouchos *et al.*, 2007) and undecane was observed in grain (Buśko *et al.*, 2010). Undecanal is formed by the hydroformylation of decene (Kohlpaintner *et al.*, 2013). It has a pleasant odour, which is often found in perfumes (Kohlpaintner *et al.*, 2013). In other words, higher amounts of water (1:2) eliminated the pleasant odour of BSG.

The formation of octanal and (E,E)-2,4-decadienal in BSG was identified due to AH at both levels of

water addition. Dodecanal was formed at a lower water ratio (1:1) and (E,E)-2,4-dodecadienal was formed with a higher water addition (1:2). Octanal, (E, E)-2,4-decadienal, dodecanal, and (E,E)-2,4-dodecadienal has never been identified in BSG. However, octanal is present in barley and malt (Fărcaş *et al*., 2015), demonstrating fat, soap, lemon, and green odour perception (Dong *et al*., 2013). AH may have reformed the octanal as in its original form. (E, E)-2,4-decadienal was present in bread prepared with dried distilled grain, which was responsible for its rancid odour (Roth *et al*., 2016). This formation might be due to the high thermal exposure in the current study. (E,E)-2,4-decadienal has an odour activity value at 23.4% (Roth *et al*., 2016), which is much higher than the observed amount in the current study (maximum 3.34%). The influence of AH on the formation of rancid compounds ((E,E)-2,4-decadienal) can be ignored as the amount is much lower than the odour activity value. Dodecanal may be synthesised from dodecanol by dehydrogenation (Kohlpaintner *et al*., 2013), which demonstrated citrus oil odour perception; (E,E)-2,4-dodecadienal was identified in virgin olive oil (Giuffrè *et al*., 2020).

AH significantly ($P < 0.05$) reduced the amount of ketones to the same level at which both water level additions had no significant ($P > 0.05$) difference to each other. The addition of water on AH treatment eliminated 2-methyl-3-octanone. Furthermore, higher levels of water addition induced the formation of 5-methyl-2-hexanone and removed (E)-3-octen-2-one and (E)-6,10-dimethyl-5,9-undecadien-2-one. Those compounds have never been reported in BSG. 2-methyl-3-octanone was reported in processed meat products (Xia *et al*., 2020), which might be responsible for its meat-like odour perception; 5-methyl-2-hexanone was identified in black tea (Yan *et al*., 2022); (E)-3-octen-2-one is an aliphatic ketone, which was identified in pea protein isolate (Xu *et al*., 2020) and might represent rose, green and nut odour perception; (E)-6,10-dimethyl-5,9-undecadien-2-one or geranylacetone was observed as a flavour compound in mango (Pino *et al*., 2005). These results might demonstrate the ability of AH in eliminating meat-related odour perception and forming a green and fruity smell.

AH significantly reduced the amount of volatile alcohol in BSG. Regardless of the water level, AH eliminated nona-3,5-dien-2-ol, hept-2-en-1-ol, and 2-butyl-2,7-octadien-1-ol and induced the formation of 2-phenoxy-ethanol. All these eliminated alcohols were responsible for the essential oil flavour, as has been reported previously (Bannour *et al*., 2016; Vasanthakumar *et al*., 2019; Hota *et al*., 2022). However, 2-phenoxy-ethanol, as a new formed compound, has been observed in cereal grain (Buśko *et al*., 2010). Lower water addition (1:1) discharged (Z)-2-octen-1-ol and formed 2-methoxy-4-vinylphenol, which are responsible for a vinegar smell and flavouring agent compounds, respectively (Jeong *et al*., 2011; Le *et al*., 2012), while higher water addition (1:2) discharged 1-tetradecanol, 1-hexadecanol, and 4,4,6-trimethyl-cyclohex-2-en-1-ol and induced the formation of 2,4-hexadien-1-ol, 2-hexyn-1-ol, and 3,5-octadien-2-ol. The eliminated compounds are responsible for a fatty odour while the formed compounds are responsible for fruity and herbal perception (Noweck & Grafahrend, 2006; Feng *et al*., 2015; Wang *et al*., 2015; El-Tantawy *et al*., 2016; Polat *et al*., 2018; Ju *et al*., 2021). The results revealed that AH potentially removed the essential oil odour perception and dominantly formed pleasant smells including a grainy and desired flavour.

AH with a lower water addition formed several alkane compounds such as 1-tridecene, Z-3-heptadecene, and 3-ethyl-5-(2-ethylbutyl)-octadecane, in addition to 1-pentadecene and undecane, which were also formed at a higher water addition. All those formed compounds were identified as responsible for odour perception from medicinal plant extracts (Wang *et al*., 2015; Borgohain *et al*., 2022). In untreated BSG, only hexanoic acid was identified as a fatty acid, while AH (1:1) formed hexanoic acid and hexanoic acid 1-cyclopentyl-ethyl ester. AH (1:2) eliminated hexanoic acid and formed cyano-acetic acid and n-hexadecanoic acid. Furthermore, AH induced the formation of 1-methyl-3-(1-methylethyl)-benzene, 1R-α-pinene, and furfural. 1-methyl-3-(1-methylethyl)-benzene and 1R-α-pinene were identified in ginger (Ding *et al*., 2012), while furfural was reported due to the Maillard reaction in BSG-added bread (Ktenioudaki *et al*., 2013).

Seeing the significant modification in the profile of volatile compounds in BSG due to AH treatment, further investigation with the electronic nose is important. The identification of key odour compounds is suggested for further investigation to strengthen the findings in the current study.

Tentative quantification of polyphenolic compounds

The polyphenolic composition of BSG is presented in Table 3. The results revealed that the water ratio had no significant ($P > 0.05$) influence on the total flavan-3-ols and total polyphenolic composition. The higher the temperature, the higher the amount of flavan-3-ols and total polyphenol content, although 90 °C exposure led to the same level as that in control. A different pattern on the total phenolic acids was observed. The majority of the treatments increased the amount of total phenolic acids significantly to a certain level at which there was no significant difference among the treatments. These results suggested that AH at 110

Table 3 Physico-chemical and biological properties of autoclave heating treated BSG

Treatments	MC (%)	Fat (%)	WHC (g/g)	OHC (g/g)	ABTS (mmol Trolox per 100 g)	FRAP (mmol Trolox per 100 g)	Total flavan-3ols (mg kg^{-1})	Total phenolic acids (mg kg^{-1})	Total polyphenolic compounds (mg kg^{-1})
Control	5.39 ± 0.35^{a}	6.62 ± 0.31^{a}	2.90 ± 0.05^{d}	2.05 ± 0.01^{c}	0.11 ± 0.01^{c}	0.07 ± 0.01^{c}	122.79 ± 0.71^{c}	44.29 ± 2.12^{c}	167.07 ± 1.41^{d}
90 °C/(1:1)	4.79 ± 0.13^{a}	7.02 ± 0.23^{a}	3.47 ± 0.05^{bc}	2.02 ± 0.00^{e}	0.09 ± 0.02^{c}	0.05 ± 0.00^{c}	256.82 ± 15.02^{c}	69.38 ± 1.72^{c}	326.19 ± 0.85^{d}
110 °C/(1:1)	5.21 ± 0.01^{a}	8.52 ± 0.13^{a}	4.04 ± 0.01^{a}	2.07 ± 0.00^{b}	0.25 ± 0.00^{b}	0.20 ± 0.01^{b}	618.16 ± 23.42^{b}	130.41 ± 9.86^{ab}	748.56 ± 47.42^{bc}
130 °C/(1:1)	4.94 ± 0.01^{a}	9.23 ± 0.41^{a}	3.80 ± 0.03^{abc}	2.04 ± 0.00^{d}	0.33 ± 0.00^{a}	0.28 ± 0.02^{a}	996.62 ± 47.83^{a}	166.76 ± 1.35^{a}	1163.38 ± 49.18^{a}
90 °C/(1:2)	5.03 ± 0.14^{a}	6.70 ± 0.39^{a}	3.26 ± 0.07^{cd}	2.03 ± 0.00^{de}	0.09 ± 0.00^{c}	0.07 ± 0.00^{c}	216.64 ± 9.30^{c}	118.24 ± 7.15^{b}	334.89 ± 2.15^{d}
110 °C/(1:2)	5.11 ± 0.09^{a}	7.89 ± 0.11^{a}	3.88 ± 0.06^{ab}	2.05 ± 0.00^{c}	0.27 ± 0.00^{b}	0.23 ± 0.01^{b}	572.23 ± 29.73^{b}	70.18 ± 2.25^{c}	642.41 ± 31.99^{c}
130 °C/(1:2)	5.35 ± 0.05^{a}	8.51 ± 0.33^{a}	4.06 ± 0.04^{a}	2.10 ± 0.00^{a}	0.32 ± 0.02^{a}	0.29 ± 0.02^{a}	747.15 ± 5.79^{b}	128.83 ± 4.98^{ab}	875.98 ± 10.77^{b}

The data are shown as mean ± standard deviation with at least duplicate analysis. Letters show the significant differences from other treatment in the same column ($P < 0.05$).

and 130 °C is capable of increasing the release of flavan-3-ols and thus total polyphenols content up to 5–8-fold and 4–7-fold, respectively. This phenomenon might be due to the degradation of dietary fibre and/or vacuole cell disruption of BSG matrix.

The impact of thermal exposure on dried, untreated BSG by autoclave has been reported previously (Naibaho *et al*., 2021), reporting that AH transformed the insoluble dietary fibre into a soluble one. This transformation might be aligned with the increase in flavan-3-ols and total polyphenols in the current study. Thermal exposure has been identified for disrupting the cell vacuoles and/or cleaving the covalent bonds (Rahman *et al*., 2021), thus allowing the modification of lignin solubility (Ohra-aho *et al*., 2016). As a consequence, it might lead to the release of certain functional groups including flavan-3-ols and total polyphenols. The improvement of phenolic acid in BSG has been identified due to the pulsed electric field treatment and thermal exposure (Budaraju *et al*., 2018; Martín-García *et al*., 2020), which intensified up to 1.7–2.7-fold (Martín-García *et al*., 2020). The increase in the quantitative compounds in the current study due to the thermal exposure might be concomitant to the formation of certain compounds, as has been identified previously. Caffeic acid was absent at a lower temperature (<100 °C) but present at a higher temperature, while the presence of sinapic acid was observed at 160 °C oven heating (Rahman *et al*., 2021).

AH at 90 °C generated the same level of polyphenols content as in the control due to its inefficiency in rupturing the crosslinking bond between polysaccharides and phenolic compounds (Sibhatu *et al*., 2021). Meanwhile, high temperature is able to discharge ester-linked ferulic acid from polysaccharides functional groups, as reported previously (Sibhatu *et al*., 2021). The results demonstrated that the crosslink between polysaccharides and phenolic acids seems to be more stable compared to that in flavan-3-ols. Different levels of temperatures generated almost the same amount of phenolic acids, although it is remarkably higher than untreated BSG. Of note is that none of the treatments reduced the polyphenolic compounds. A decline in phenolic acids by 4–6-times lower occurred due to the extraction methods (Bonifácio-Lopes *et al*., 2020).

In vitro antioxidant capabilities

The results demonstrated that, the higher the thermal levels, the higher the increase in antioxidant activities of both FRAP and ABTS, regardless of the water ratio. However, AH at 90 °C had the same level as that in untreated BSG. This phenomenon might be aligned with the trend in the amount of flavan-3-ols, as mentioned in the previous section. Thermal

exposures at 110 and 130 °C released a higher amount of flavan-3-ols, thus enhancing the FRAP and ABTS properties of methanolic extracts from AH-treated BSG. ABTS defines the capability of the extracts in reducing the molecular oxygen and hydrogen peroxide (Benzie & Strain, 1996), and FRAP demonstrates the ability of the extracts in alleviating lipid oxidation (Rahman *et al*., 2021). By this, the current study revealed the ability of AH in improving the ability of BSG as a healthy ingredient, both as functional food and nutraceutical ingredient.

According to the previous studies, other compounds in BSG which play an important role in antioxidant capabilities of BSG include fatty acids such as palmitic, linoleic, oleic, and stearic acid (Fărcaş *et al*., 2015; Parekh *et al*., 2017; Tan *et al*., 2019). This might be slightly related to the fat content in the current study, although statistical significance was not observed. However, notably, the fat content was observed to be higher as the temperature was raised. Furthermore, as was mentioned in Influence of AH on fatty acid compositions Section, AH reduced the amount of SFA and concomitantly improved the amount of PUFA. This phenomenon might suggest an indirect link to the increase in the antioxidant activity, as discovered in this section. The improvement in antioxidant activity has also been observed previously (Budaraju *et al*., 2018). It was emphasised that the improvement in antioxidant activity had no correlation with the amount of bound phenolic compounds (Budaraju *et al*., 2018). Therefore, the improvement of antioxidant activity in the current study might be a result of free phenolic compounds. Furthermore, coumaric acid had a crucial impact on antioxidant properties of BSG (McCarthy *et al*., 2013). The specific phenolic compounds were not investigated in the current study. However, this might suggest that the antioxidant activity observed in the current study might only be due to certain compounds, which is seemingly important to investigate in the near future. Hydroxycinnamic acid is the most abundant phenolic acid from BSG including ferulic acid (FA), p-coumaric acid (p-CA) derivatives, FA derivatives, p-CA, caffeic acid (CA), and CA derivatives (McCarthy *et al*., 2013).

Impact of AH on the techno-functionality of BSG

The results showed that AH significantly ($P < 0.05$) enhanced WHC regardless of the water ratio. Statistically, the highest WHC was given by the higher temperature in both water ratios, while the lowest WHC was obtained in untreated BSG. This result demonstrated that AH improved the WHC of BSG as a sole impact of the temperature levels. The treated BSG had a range of 3.3–4.1 g/g WHC, while untreated BSG obtained WHC at 2.9 g/g. This number is aligned with the previous studies which reported that the WHC of BSG ranged from 2.9 to 4.3 g/g (Naibaho *et al*., 2021; Naibaho & Korzeniowska, 2021b). AH on dried BSG was observed to decrease the WHC of BSG (Naibaho *et al*., 2021), while the current study, which increased the WHC, was conducted on undried fresh BSG. In contrast, the majority of AH treatment decreased the OHC level of BSG. OHC in the current study appeared at the same range as previously reported, at a range of 1.9–2.2 g/g (Naibaho & Korzeniowska, 2021b). However, AH on dried BSG reduced the OHC level (Naibaho *et al*., 2021). Therefore, AH treatment on fresh BSG slurry benefits the techno-functionality of BSG. The ability of BSG in binding water is influenced by the presence of arabinoxylans (Steiner *et al*., 2015). By this, AH might have modified the polysaccharides composition of BSG, as mentioned earlier, in addition to the arabinoxylans profile. Techno-functional properties can be altered due to energy exposures. A reduction in WHC and OHC was observed due to the particle size reduction, while an increase was obtained as an impact of high-pressure treatment (Yan *et al*., 2019). The fluctuation of WHC and OHC was emphasised due to the exposure of hydrophilic groups as an impact of losing the dietary fibre structure (Yan *et al*., 2019). Improving the WHC benefits the texture and viscosity of food products (Benitez *et al*., 2019; Kieserling *et al*., 2019). Therefore, AH showed a beneficial performance in improving food structure formation.

Conclusion

The results revealed that AH is capable of reducing SFA, increasing PUFA and slightly altering the amount of MUFA. Quantitatively, AH reduced the amount of ketones, alcohols, and furans, while it intensified the aldehydes and volatile fatty acids, regardless of the water ratio. The alteration of the volatile compound profile was followed by the elimination and formation of several volatile compounds in BSG matrix depending on the water ratio. Furthermore, AH enriched the amount of total flavan-3-ols and, thus, the total polyphenolic compounds, and enhanced the antioxidant activities (ABTS and FRAP) and improved the WHC of BSG as an impact of thermal elevation and regardless of the water ratio. The study demonstrated that AH improved the quality of BSG as a functional food and nutraceutical ingredient from the perspective of bioactivity and functionality. Further investigation on polysaccharides composition, protein and amino acids profile as well as free fatty acids and storage stability related is seemingly important in order to understand the mechanisms and efficiency of AH in disrupting BSG matrix.

Acknowledgments

This work was supported by UPWR 2.0, international and interdisciplinary programme of development of Wrocław University of Environmental and Life Sciences, co-financed by the European Social Fund under the Operational Programme Knowledge Education Development 2014–2020: Axis III Higher education for the economy and development; Action 3.5. Comprehensive programmes for schools of higher education (POWR.03.05.00-00-Z062/18). It was also supported by ERA-NET CO-FUND Horizon 2020 – FACCE SURPLUS Sustainable and Resilient Agriculture for Food and Non-Food Systems and PRO-WASTE Protein-fibre biorefinery for scattered material streams (2019–2021).

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Conflict of interest

None.

Ethical approval

Ethics approval was not required for this research.

Peer review

The peer review history for this article is available at https://publons.com/publon/10.1111/ijfs.16042.

Data availability statement

Data available on request from the authors.

References

Bailone, R.L., Borra, R.C., Fukushima, H.C.S. & Aguiar, L.K. (2022). Water reuse in the food industry. *Discover Food*, **2**, 5.

Balogun, A.O., Sotoudehniakarani, F. & McDonald, A.G. (2017). Thermo-kinetic, spectroscopic study of brewer's spent grains and characterisation of their pyrolysis products. *Journal of Analytical and Applied Pyrolysis*, **127**, 8–16.

Bannour, M., Aouadhi, C., Khalfaoui, H., Aschi-Smiti, S. & Khadhri, A. (2016). Barks essential oil, secondary metabolites and biological activities of four organs of Tunisian *Calligonum azel* Maire. *Chemistry & Biodiversity*, **13**, 1527–1536.

Benitez, V., Rebollo-Hernanz, M., Hernanz, S., Chantres, S., Aguilera, Y. & Martin-Cabrejas, M.A. (2019). Coffee parchment as a new dietary fiber ingredient: functional and physiological characterization. *Food Research International*, **122**, 105–113.

Benzie, I.F.F. & Strain, J.J. (1996). The ferric reducing ability of plasma (FRAP) as a measure of "antioxidant power": the FRAP assay. *Analytical Biochemistry*, **239**, 70–76.

Bonifácio-Lopes, T., Teixeira, J.A. & Pintado, M. (2020). Current extraction techniques towards bioactive compounds from brewer's spent grain – a review. *Critical Reviews in Food Science and Nutrition*, **60**, 2730–2741.

Borgohain, P., Barua, P., Das, D. *et al.* (2022). Antifungal activity of selected medicinal plants used by indigenous people of Assam in India to treat onychomycosis. *Journal of Herbs, Spices & Medicinal Plants*, **28**, 160–178.

Budaraju, S., Mallikarjunan, K., Annor, G., Schoenfuss, T. & Raun, R. (2018). Effect of pre-treatments on the antioxidant potential of phenolic extracts from barley malt rootlets. *Food Chemistry*, **266**, 31–37.

Buśko, M., Jeleń, H., Góral, T. *et al.* (2010). Volatile metabolites in various cereal grains. *Food Additives & Contaminants: Part A*, **27**, 1574–1581.

Coelho, E., Rocha, M.A.M., Saraiva, J.A. & Coimbra, M.A. (2014). Microwave superheated water and dilute alkali extraction of brewers' spent grain arabinoxylans and arabinoxylo-oligosaccharides. *Carbohydrate Polymers*, **99**, 415–422.

Connolly, A., Cermeño, M., Crowley, D., O'Callaghan, Y., O'Brien, N.M. & FitzGerald, R.J. (2019). Characterisation of the in vitro bioactive properties of alkaline and enzyme extracted brewers'

spent grain protein hydrolysates. *Food Research International*, **121**, 524–532.
Connolly, A., Cermeño, M., Alashi, A.M., Aluko, R.E. & FitzGerald, R.J. (2021). Generation of phenolic-rich extracts from brewers' spent grain and characterisation of their in vitro and in vivo activities. *Innovative Food Science & Emerging Technologies*, **68**, 102617.
Cooray, S.T. & Chen, W.N. (2018). Valorization of brewer's spent grain using fungi solid-state fermentation to enhance nutritional value. *Journal of Functional Foods*, **42**, 85–94.
Ding, S.H., An, K.J., Zhao, C.P., Li, Y., Guo, Y.H. & Wang, Z.F. (2012). Effect of drying methods on volatiles of Chinese ginger (*Zingiber officinale* roscoe). *Food and Bioproducts Processing*, **90**, 515–524.
Dong, L., Piao, Y., Zhang, X., Zhao, C., Hou, Y. & Shi, Z. (2013). Analysis of volatile compounds from a malting process using headspace solid-phase micro-extraction and GC-MS. *Food Research International*, **51**, 783–789.
El-Tantawy, M.E., Shams, M.M. & Afifi, M.S. (2016). Chemical composition and biological evaluation of the volatile constituents from the aerial parts of *Nephrolepis exaltata* (L.) and *Nephrolepis cordifolia* (L.) C. Presl grown in Egypt. *Natural Product Research*, **30**, 1197–1201.
Fărcaş, A.C., Socaci, S.A., Dulf, F.V., Tofană, M., Mudura, E. & Diaconeasa, Z. (2015). 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. *Journal of Cereal Science*, **64**, 34–42.
Feng, X., Lian, J. & Zhao, H. (2015). Metabolic engineering of *Saccharomyces cerevisiae* to improve 1-hexadecanol production. *Metabolic Engineering*, **27**, 10–19.
Giuffrè, A.M., Capocasale, M., Macrì, R., Caracciolo, M., Zappia, C. & Poiana, M. (2020). Volatile profiles of extra virgin olive oil, olive pomace oil, soybean oil and palm oil in different heating conditions. *LWT*, **117**, 108631.
He, Y., Kuhn, D.D., Ogejo, J.A. *et al.* (2019). Wet fractionation process to produce high protein and high fiber products from brewer's spent grain. *Food and Bioproducts Processing*, **117**, 266–274.
Hota, R.N., Nanda, B.K., Behera, B.R. & Bose, A. (2022). Ameliorative effect of ethanolic extract of *Limnophila rugosa* (Scrophulariaceae) in paracetamol- and carbon tetrachloride-induced hepatotoxicity in rats. *Future Journal of Pharmaceutical Sciences*, **8**, 6.
Jeong, J.B., Hong, S.C., Jeong, H.J. & Koo, J.S. (2011). Anti-inflammatory effect of 2-methoxy-4-vinylphenol via the suppression of NF-κB and MAPK activation, and acetylation of histone H3. *Archives of Pharmacal Research*, **34**, 2109–2116.
Ju, Y., Yue, X., Cao, X., Wei, X. & Fang, Y. (2021). First study on the fatty acids and their derived volatile profiles from six Chinese wild spine grape clones (*Vitis davidii* Foex). *Scientia Horticulturae*, **275**, 109709.
Kemppainen, K., Rommi, K., Holopainen, U. & Kruus, K. (2016). Steam explosion of Brewer's spent grain improves enzymatic digestibility of carbohydrates and affects solubility and stability of proteins. *Applied Biochemistry and Biotechnology*, **180**, 94–108.
Kieserling, K., Vu, T.M., Drusch, S. & Schalow, S. (2019). Impact of pectin-rich orange fibre on gel characteristics and sensory properties in lactic acid fermented yoghurt. *Food Hydrocolloids*, **94**, 152–163.
Kohlpaintner, C., Schulte, M., Falbe, J., Lappe, P., Weber, J. & Frey, G. (2013). Aldehydes, aliphatic. In: *Ullmann's Encyclopedia of Industrial Chemistry*. Weinheim: Wiley Online Library.
Ktenioudaki, A., O'Shea, N. & Gallagher, E. (2013). Rheological properties of wheat dough supplemented with functional by-products of food processing: Brewer's spent grain and apple pomace. *Journal of Food Engineering*, **116**, 362–368.
Kunishima, M., Yamauchi, Y., Mizutani, M., Kuse, M., Takikawa, H. & Sugimoto, Y. (2016). Identification of (Z)-3:(E)-2-hexenal isomerases essential to the production of the leaf aldehyde in plants. *Journal of Biological Chemistry*, **291**, 14023–14033.
Lao, E.J., Dimoso, N., Raymond, J. & Mbega, E.R. (2020). The prebiotic potential of brewers' spent grain on livestock's health: a review. *Tropical Animal Health and Production*, **52**, 461–472.
Le, V.-D., Zheng, X.-W., Chen, J.-Y. & Han, B.-Z. (2012). Characterization of volatile compounds in *fen-Daqu* - a traditional Chinese liquor fermentation starter: characterization of volatile compounds in *fen-Daqu*. *Journal of the Institute of Brewing*, **118**, 107–113.
Li, B., Yang, W., Nie, Y., Kang, F., Goff, H.D. & Cui, S.W. (2019a). Effect of steam explosion on dietary fiber, polysaccharide, protein and physicochemical properties of okara. *Food Hydrocolloids*, **94**, 48–56.
Li, S., Chen, G., Qiang, S. *et al.* (2019b). Intensifying soluble dietary fiber production and properties of soybean curd residue via autoclaving treatment. *Bioresource Technology Reports*, **7**, 100203.
Lynch, K.M., Steffen, E.J. & Arendt, E.K. (2016). Brewers' spent grain: a review with an emphasis on food and health. *Journal of the Institute of Brewing*, **122**, 553–568.
Mallen, E. & Najdanovic-Visak, V. (2018). Brewers' spent grains: drying kinetics and biodiesel production. *Bioresource Technology Reports*, **1**, 16–23.
Mallouchos, A., Paul, L., Argyro, B., Koutinas, A. & Komaitis, M. (2007). Ambient and low temperature winemaking by immobilized cells on brewer's spent grains: effect on volatile composition. *Food Chemistry*, **104**, 918–927.
Martín-García, B., Tylewicz, U., Verardo, V. *et al.* (2020). Pulsed electric field (PEF) as pre-treatment to improve the phenolic compounds recovery from brewers' spent grains. *Innovative Food Science & Emerging Technologies*, **64**, 102402.
McCarthy, A.L., O'Callaghan, Y.C., Neugart, S. *et al.* (2013). The hydroxycinnamic acid content of barley and brewers' spent grain (BSG) and the potential to incorporate phenolic extracts of BSG as antioxidants into fruit beverages. *Food Chemistry*, **141**, 2567–2574.
Naibaho, J. & Korzeniowska, M. (2021a). Brewers' spent grain in food systems: processing and final products quality as a function of fiber modification treatment. *Journal of Food Science*, **86**, 1532–1551.
Naibaho, J. & Korzeniowska, M. (2021b). The variability of physico-chemical properties of brewery spent grain from 8 different breweries. *Heliyon*, **7**, e06583.
Naibaho, J., Korzeniowska, M., Wojdyło, A. *et al.* (2021). Fiber modification of brewers' spent grain by autoclave treatment to improve its properties as a functional food ingredient. *LWT*, **149**, 111877. The description on dietary fiber degradation due to thermal elevation on autoclave treatment on spent grain.
Naibaho, J., Korzeniowska, M., Wojdyło, A., Muchdatul Ayunda, H., Foste, M. & Yang, B. (2022a). Techno-functional properties of protein from protease-treated brewers' spent grain (BSG) and investigation of antioxidant activity of extracted proteins and BSG residues. *Journal of Cereal Science*, **107**, 103524.
Naibaho, J., Wojdyło, A., Korzeniowska, M. *et al.* (2022b). Antioxidant activities and polyphenolic identification by UPLC-MS/MS of autoclaved brewers' spent grain. *LWT*, **163**, 113612. Description in the influence of autoclave treatment in inducing the formation and discharging of polyphenolic compounds in spent grain.
Nawaz, A., Li, E., Irshad, S. *et al.* (2020). Improved effect of autoclave processing on size reduction, chemical structure, nutritional, mechanical and in vitro digestibility properties of fish bone powder. *Advanced Powder Technology*, **31**, 2513–2520.
Nowacki, D., Martynowicz, H., Skoczyńska, A. *et al.* (2017). Lecithin derived from ω-3 PUFA fortified eggs decreases blood pressure in spontaneously hypertensive rats. *Scientific Reports*, **7**, 12373.
Noweck, K. & Grafahrend, W. (2006). Fatty alcohols. In: *Ullmann's Encyclopedia of Industrial Chemistry* (edited by Wiley-VCH Verlag

GmbH & Co. KGaA). Pp. a10_277.pub2. Weinheim: Wiley-VCH Verlag GmbH & Co. KGaA.
Ohra-aho, T., Niemi, P., Aura, A.-M. *et al.* (2016). Structure of Brewer's spent grain lignin and its interactions with gut microbiota in vitro. *Journal of Agricultural and Food Chemistry*, **64**, 812–820.
O'Shea, N., Kilcawley, K.N. & Gallagher, E. (2017). Aromatic composition and physicochemical characteristics of crackers containing barley fractions. *Cereal Chemistry Journal*, **94**, 611–618.
Parekh, I., Khanvilkar, A. & Naik, A. (2017). Barley-wheat brewers' spent grain: a potential source of antioxidant rich lipids. *Journal of Food Processing and Preservation*, **41**, e13244.
Pino, J.A., Mesa, J., Muñoz, Y., Martí, M.P. & Marbot, R. (2005). Volatile components from mango (*Mangifera indica* L.) cultivars. *Journal of Agricultural and Food Chemistry*, **53**, 2213–2223.
Polat, A., Şat, İ.G. & Ilgaz, Ş. (2018). Comparison of black tea volatiles depending on the grades and different drying temperatures. *Journal of Food Processing and Preservation*, **42**, e13653.
Rahman, M.J., Malunga, L.N., Eskin, M., Eck, P., Thandapilly, S.J. & Thiyam-Hollander, U. (2021). Valorization of heat-treated Brewers' spent grain through the identification of bioactive phenolics by UPLC-PDA and evaluation of their antioxidant activities. *Frontiers in Nutrition*, **8**, 634519. Degradation mechanism on spent grain matrix in releasing the phenolic content.
Re, R., Pellegrini, N., Proteggente, A., Pannala, A., Yang, M. & Rice-Evans, C. (1999). Antioxidant activity applying an improved ABTS radical cation decolorization assay. *Free Radical Biology and Medicine*, **26**, 1231–1237.
Roth, M., Schuster, H., Kollmannsberger, H., Jekle, M. & Becker, T. (2016). Changes in aroma composition and sensory properties provided by distiller's grains addition to bakery products. *Journal of Cereal Science*, **72**, 75–83.
Sibhatu, H.K., Anuradha Jabasingh, S., Yimam, A. & Ahmed, S. (2021). Ferulic acid production from brewery spent grains, an agro-industrial waste. *LWT*, **135**, 110009.
Skendi, A., Harasym, J. & Galanakis, C.M. (2018). Recovery of high added-value compounds from brewing and distillate processing by-products. In: *Sustainable Recovery and Reutilization of Cereal Processing By-Products*. Pp. 189–225. Cambridge: Elsevier.
Steiner, J., Procopio, S. & Becker, T. (2015). Brewer's spent grain: source of value-added polysaccharides for the food industry in reference to the health claims. *European Food Research and Technology*, **241**, 303–315.
Tan, Y.X., Mok, W.K., Lee, J., Kim, J. & Chen, W.N. (2019). Solid state fermentation of Brewers' spent grains for improved nutritional profile using *Bacillus subtilis* WX-17. *Fermentation*, **5**, 52.
Tkacz, K., Wojdyło, A., Turkiewicz, I.P. & Nowicka, P. (2021). Triterpenoids, phenolic compounds, macro- and microelements in anatomical parts of sea buckthorn (*Hippophaë rhamnoides* L.) berries, branches and leaves. *Journal of Food Composition and Analysis*, **103**, 104107.
Turkiewicz, I.P., Wojdyło, A., Tkacz, K., Lech, K., Michalska-Ciechanowska, A. & Nowicka, P. (2020a). The influence of different carrier agents and drying techniques on physical and chemical characterization of Japanese quince (*Chaenomeles japonica*) microencapsulation powder. *Food Chemistry*, **323**, 126830.
Turkiewicz, I.P., Wojdyło, A., Tkacz, K., Nowicka, P., Golis, T. & Bąbelewski, P. (2020b). ABTS on-line antioxidant, α-amylase, α-glucosidase, pancreatic lipase, acetyl- and Butyrylcholinesterase inhibition activity of Chaenomeles fruits determined by polyphenols and other chemical compounds. *Antioxidants*, **9**, 60.
Turkiewicz, I.P., Wojdyło, A., Tkacz, K. & Nowicka, P. (2021). Comprehensive characterization of Chaenomeles seeds as a potential source of nutritional and biologically active compounds. *Journal of Food Composition and Analysis*, **102**, 104065.
Vasanthakumar, S., Dineshkumar, G. & Jayaseelan, K. (2019). Phytochemical screening, GC-MS analysis and antibacterial evaluation of ethanolic leaves extract of *Avicennia marina*. *Journal of Drug Delivery and Therapeutics*, **9**, 145–150.
Wang, D.C., Sun, S.H., Shi, L.N. *et al.* (2015). Chemical composition, antibacterial and antioxidant activity of the essential oils of *Metaplexis japonica* and their antibacterial components. *International Journal of Food Science & Technology*, **50**, 449–457.
Wen, C., Zhang, J., Duan, Y., Zhang, H. & Ma, H. (2019). A mini-review on Brewer's spent grain protein: isolation, physicochemical properties, application of protein, and functional properties of hydrolysates. *Journal of Food Science*, **84**, 3330–3340.
Xia, Q., Feng, T., Lou, X. *et al.* (2020). Headspace fingerprinting approach to identify the major pathway influencing volatile patterns of vinasse-cured duck processed by high pressure, as well as its impact on physicochemical and sensory attributes. *International Journal of Food Science & Technology*, **55**, 669–680.
Xie, F., Li, M., Lan, X. *et al.* (2017). Modification of dietary fibers from purple-fleshed potatoes (Heimeiren) with high hydrostatic pressure and high pressure homogenization processing: a comparative study. *Innovative Food Science & Emerging Technologies*, **42**, 157–164.
Xu, M., Jin, Z., Gu, Z., Rao, J. & Chen, B. (2020). Changes in odor characteristics of pulse protein isolates from germinated chickpea, lentil, and yellow pea: role of lipoxygenase and free radicals. *Food Chemistry*, **314**, 126184.
Yan, L., Li, T., Liu, C. & Zheng, L. (2019). Effects of high hydrostatic pressure and superfine grinding treatment on physicochemical/functional properties of pear pomace and chemical composition of its soluble dietary fibre. *LWT*, **107**, 171–177.
Yan, T., Lin, J., Zhu, J. *et al.* (2022). Aroma analysis of Fuyun 6 and Jinguanyin black tea in the Fu'an area based on E-nose and GC–MS. *European Food Research and Technology*, **248**, 947–961.
Zheng, Y., Wei, Z., Zhang, R. *et al.* (2020). Optimization of the autoclave preparation process for improving resistant starch content in rice grains. *Food Science & Nutrition*, **8**, 2383–2394. Detail understanding of thermal degradation due to autoclave treatment on grain.

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Naibaho J., Bobak L., Pudlo A., Wojdylo A., Andayani S. N., Pangestika L. M. W., Korzeniowska M., & Yang B. 2023. Chemical compositions, antioxidant activities and techno-functionality of spent grain treated by autoclave treatment: evaluation of water and temperature levels. International Journal of Food Science and Technology 58(4), 16042. https://doi.org/10.1111/ijfs.16042. (autorzy, rok wydania, tytuł, czasopismo lub wydawca, tom, strony)

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Oświadczam, że w pracy:

Naibaho J., Bobak L., Pudlo A., Wojdylo A., Andayani S. N., Pangestika L. M. W., Korzeniowska M., & Yang B. 2023. Chemical compositions, antioxidant activities and techno-functionality of spent grain treated by autoclave treatment: evaluation of water and temperature levels. International Journal of Food Science and Technology 58(4), 16042. https://doi.org/10.1111/ijfs.16042. (autorzy, rok wydania, tytuł, czasopismo lub wydawca, tom, strony)

mój udział polegał na: Konceptualizacja (wspomaganie); pozyskanie finansowania (równe); administracja projektami (lider); nadzór (równy); pisanie – projekt oryginalny (wspomagający); pisanie – recenzja i redakcja (pomoc). (opisać szczegółowo swój własny – a nie Kandydata udział w powstaniu pracy, np. wykonaniu doświadczeń techniką ....., analizie statystycznej wyników eksperymentów zilustrowanych na ryc. ...., przygotowaniu tekstu manuskryptu zamieszczonego w rozdziale ....., kierowaniu projektem naukowym obejmującym badania opisane w tej pracy, itp.).

March 27, 2023

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data i podpis

Publication 5

Food Bioscience 53 (2023) 102523

Contents lists available at ScienceDirect

Food Bioscience

journal homepage: www.elsevier.com/locate/fbio

Conventional water bath heating on undried brewer's spent grain: Functionality, fatty acids, volatiles, polyphenolic and antioxidant properties

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ARTICLE INFO

Keywords:
By-product valorization
Sustainability
Thermal exposures
Value-added production

ABSTRACT

Brewers' spent grain (BSG) contains bioactive compounds. It was hypothesized that heating treatments using conventional water bath heating (CWH) on brewers' spent grain (BSG) would modify the functionality, chemical constituents and antioxidant activities of BSG. Different temperatures and time exposures (80, 90 and 100 °C at 15, 30 and 60 min) were applied on fresh undried BSG. CWH at 80 °C increased the amount of flavan-3-ols, while 100 °C at 30 and 60 min improved the ABTS (2,2′-Azinobis-(3-ethylbenzthiazoline-6-sulfonic acid) value. CWH significantly declined saturated fatty acid and enhanced the poly-unsaturated fatty acid. Moreover, CWH discharged pungent, floral, spice and mushroom odor perceptions and formed fruity, sweet and pleasant odor perceptions, as well as essential-oil-related compounds. Additionally, CWH improved water-holding and oil-holding capacities. In conclusion, CWH as a low-cost treatment improved the functionality, fatty acid composition and aromatic profile of BSG.

1. Introduction

The valorization of brewer's spent grain (BSG) has been reviewed as a means to produce functional foods and nutraceutical ingredients (Naibaho & Korzeniowska, 2021). BSG has high nutritional value, including dietary fiber, proteins, fatty acids and phenolic acids, thus potentially possessing benefits for human health (Naibaho & Korzeniowska, 2021). Several treatments, including chemical, physical, enzymatic and combination treatments, have been applied in order to improve yield extracts, biological properties and/or the functional behavior of BSG (Naibaho & Korzeniowska, 2021). Physical treatments including steam explosions, autoclaves, particle size reduction and pulse electric fields have been applied for the treatment of BSG, improving phenolic compound contents and biological properties, intensifying the amount of soluble dietary fiber, enhancing protein functionality and modifying the structure of arabinoxylans (Connolly et al., 2019; Kumari et al., 2019; Martín-García et al., 2020; Verni et al., 2020).

Heating using a water bath is seemingly promising, as it is a low-cost processing alternative that uses simple equipment and is relatively easy to operate. Water baths have been conventionally utilized in food processing and have been compared to advanced thermal-related treatments such as microwave, ultrasound and ohmic heating techniques (Jung et al., 2020; Ye et al., 2022; Zhang et al., 2015). Obviously, newly advanced technology tends to have better performance in modifying the

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https://doi.org/10.1016/j.fbio.2023.102523
Received 11 September 2022; Received in revised form 17 February 2023; Accepted 1 March 2023
Available online 2 March 2023

qualities of food products; however, water baths are simple, low-cost and easily reproducible, in terms of their application. The use of a water bath improved the performance of microwave–ultrasound heating with respect to increasing the strength and water-holding capacity of gels (Ye et al., 2022). A water bath also influenced the performance of ultrasound-assisted treatments in improving the properties of the Maillard reaction with respect to chitosan–fructose (Zhang et al., 2015). However, to the best of our knowledge, the conventional water bath heating (CWH) treatment of BSG has not yet been reported.

Therefore, the potential of using a water bath for the thermal treatment of BSG was taken into consideration. This study aimed to evaluate the change in BSG properties due to water bath thermal heating treatments. It was hypothesized that thermal treatments using a water bath would modify functionalities such as water-holding capacity (WHC) and oil-holding capacity (OHC), antioxidant capabilities and chemical constituents, such as phenolic compounds, fatty acid composition and the volatile profile of BSG.

2. Materials and methods

2.1. Materials

2.1.1. BSG and BSG preparation

BSG wet slurry was collected from a local light-beer producer in Wroclaw, Poland. The BSG was then ground to pass 0.2 mm, kept in a polyethylene bag and stored at freezing temperatures (−20 °C) prior to the experiment.

2.1.2. Chemicals and reagents

Trolox (6-hydro-2,5,7,8-tetramethylchroman-2-carboxylic acid) and the other chemical compounds were purchased from Sigma-Aldrich (Steinheim, Germany). UPLC-grade water was prepared by using an HLP SMART 1000s system (Hydrolab, Gdansk, Poland). Immediately before use, the water was filtered using a 0.22 μm membrane filter (Millipore Sigma-Aldrich, Steinheim, Germany). All chemicals used were of analytical grade.

2.2. Experimental design

Ground wet BSG was mixed properly with distilled water at a ratio of 1:1 in a glass beaker. The mixture was then properly closed with aluminum foil. The mixture was heated in a conventional water bath (Julabo TW-12 ECO, Julabo GmbH, Germany) at different temperatures. CWH is typically applied at 85 °C for 30 min. Thus, our experiment was designed just below and above the level of technical CWH; specifically, thermal exposure was conducted at 80, 90, or 100 °C (±1 °C) for time exposures of 15, 30 or 60 min. The treated BSG was dried using an oven dryer at 75 °C overnight (±16 h) to obtain moisture contents below 6% (see Table 1). The samples were ground using a laboratory-scale blender for 5 min with a 10 s pause every 1 min. Samples were packed into aluminum foil and kept at 10 °C for further analysis.

2.3. Analysis of moisture and extracted fat content

Moisture content was measured using the oven method and fat content was measured by using the Soxhlet method (Buchi B-811, Postfach, Liechtenstein), following AOAC 2000 procedures and triplicate applications.

2.4. Analysis of WHC and OHC

The physical properties of BSG were evaluated for WHC and OHC, as described previously (Ktenioudaki et al., 2013). The analysis was conducted in triplicate.

2.5. Methanol extraction, antioxidant analysis and polyphenolic quantification

Methanol extracts were prepared following the procedure described in (Naibaho et al., 2022). *In vitro* antioxidant capabilities for ABTS (2, 2′-Azinobis-(3-ethylbenzthiazoline-6-sulfonic acid) and FRAP (ferric-reducing antioxidant power) were assessed (Naibaho et al., 2022). The extraction was performed in duplicate, and the analysis of antioxidants was performed in triplicate. The quantification of flavan-3-ols and phenolic acids was conducted by UPLC-PDA-FL (Waters Corp, Milford, MA. US) following the procedures described in (Tkacz et al., 2021) and performed in duplicate.

Table 1
Physico-chemical and biological properties of waterbath heating treated spent grain.

Treatments	MC (%)	Fat (%)	WHC (g/g)	OHC (g/g)	ABTS (mmol Trolox/Kg)	FRAP (mmol Trolox/Kg)	Total flavan-3-ols (mg/kg)	Total phenolic acids (mg/kg)	Total polyphenolic compounds (mg/kg)
Control	5.39 ± 0.35^{a}	6.62 ± 0.31^{de}	2.90 ± 0.05^{d}	2.05 ± 0.01^{f}	1.1 ± 0.00^{bc}	0.7 ± 0.01^{ab}	122.79 ± 0.71^{b}	44.29 ± 2.12^{a}	167.07 ± 1.41^{c}
80 °C/15 min	5.18 ± 0.05^{a}	5.88 ± 0.07^{f}	3.50 ± 0.05^{bc}	2.14 ± 0.00^{bc}	0.9 ± 0.00^{cd}	0.3 ± 0.00^{bc}	150.90 ± 0.71^{a}	24.34 ± 0.07^{bc}	175.24 ± 0.78^{ab}
80 °C/30 min	4.50 ± 0.09^{b}	6.40 ± 0.11^{ef}	3.74 ± 0.05^{ab}	2.13 ± 0.00^{cd}	0.9 ± 0.00^{cd}	0.2 ± 0.01^{c}	144.88 ± 0.71^{a}	25.26 ± 0.14^{bc}	170.14 ± 0.85^{bc}
80 °C/60 min	4.25 ± 0.19^{b}	6.91 ± 0.07^{cde}	3.67 ± 0.05^{abc}	2.12 ± 0.00^{d}	0.9 ± 0.00^{cd}	0.4 ± 0.00^{bc}	151.33 ± 0.71^{a}	27.23 ± 0.28^{b}	178.56 ± 0.42^{a}
90 °C/15 min	4.37 ± 0.00^{b}	6.65 ± 0.06^{de}	3.55 ± 0.02^{abc}	2.10 ± 0.00^{e}	0.7 ± 0.00^{d}	0.3 ± 0.00^{bc}	151.33 ± 0.28^{a}	27.09 ± 0.07^{b}	178.41 ± 0.35^{a}
90 °C/30 min	4.48 ± 0.11^{b}	7.35 ± 0.08^{bc}	3.69 ± 0.11^{ab}	2.16 ± 0.00^{a}	0.8 ± 0.01^{d}	0.5 ± 0.02^{abc}	112.64 ± 3.54^{c}	16.81 ± 0.71^{d}	129.45 ± 4.24^{e}
90 °C/60 min	4.34 ± 0.02^{b}	7.07 ± 0.07^{bcd}	3.77 ± 0.13^{a}	2.15 ± 0.00^{ab}	1.0 ± 0.01^{bc}	0.5 ± 0.00^{abc}	108.06 ± 1.41^{c}	22.67 ± 1.16^{c}	130.73 ± 0.26^{de}
100 °C/15 min	4.29 ± 0.22^{b}	6.95 ± 0.21^{cde}	3.59 ± 0.00^{abc}	2.13 ± 0.00^{cd}	0.9 ± 0.01^{cd}	0.3 ± 0.02^{bc}	107.99 ± 1.41^{c}	10.21 ± 0.71^{e}	118.20 ± 2.12^{f}
100 °C/30 min	4.44 ± 0.15^{b}	7.56 ± 0.08^{b}	3.60 ± 0.05^{abc}	2.09 ± 0.00^{e}	1.3 ± 0.00^{a}	0.6 ± 0.00^{ab}	124.38 ± 2.83^{b}	8.97 ± 0.22^{e}	133.35 ± 2.61^{de}
100 °C/60 min	4.25 ± 0.03^{b}	8.13 ± 0.13^{a}	3.41 ± 0.06^{c}	2.09 ± 0.00^{e}	1.3 ± 0.00^{a}	0.8 ± 0.00^{a}	125.48 ± 3.54^{b}	11.97 ± 1.41^{e}	137.45 ± 2.12^{d}

Note: the data is shown as mean ± standard deviation with at least duplicate analysis. Letters show the significant differences from other treatment in the same column ($p < 0.05$). MC: moisture content, WHC: water holding capacity, OHC: oil holding capacity, ABTS: 2,2′-Azinobis-(3-ethylbenzthiazoline-6-sulfonic acid, FRAP: ferric-reducing antioxidant power.

2.6. Analysis of fatty acids composition by GC-MS

Lipid extraction was conducted following previously described procedures (Fărcaş et al., 2015). The derivatization of lipids into fatty acid methyl esters (FAMEs) was assessed following the procedures described in previous work (Nowacki et al., 2017). The fatty acid profile was analyzed using a gas chromatograph (GC6890, Agilent Technologies Inc. CA, USA) coupled with a mass spectrometer 5983 MS equipped with a quadrupole mass detector (Agilent Technologies Inc. CA, USA). Separation was performed in a 0.25 mm × 100 m HP-88 capillary column filled with an 88:12 cyanopropyl-aryl poly-siloxane bed (grain size 0.2 μm). Helium at a flow rate of 1 mL/min was used as the mobile phase, and the sample was injected in split mode (split 4:1). The program was set as follows: initial temperature at 60 °C (2 min) and heating at 20 °C/min to reach 180 °C followed by 3 °C/min to 220 °C. The temperature was maintained for 15 min. Heating continued at a rate of 5 °C/min to reach 250 °C, and the temperature was maintained for 8 min. Spectra were identified using an algorithm for searching the National Institute of Standards and Technology's (NIST) library (version of 2008).

2.7. Analysis of volatile compounds by GC-MS

The impact of CWH on the volatile compounds of BSG was determined based on a linear function of temperature and time exposure which describes the level of exposures. The study was designed at 3 different levels of temperature and 3 different levels of time exposure. The analysis of volatile compounds was carried out to evaluate the impact of CWH at the lowest, medium, and highest exposure levels. Therefore, the profile of volatile compounds was only conducted at a combination of temperature at 80 ℃, 90 ℃, and 100 ℃ with time exposure at 15 min, 30 min, and 60 min, respectively. In addition, this hypothesis was based on the preliminary results of crude fat content as presented in Table 1. It is well known that volatile compounds are significantly related to fat composition. As presented in Table 1, the increase in temperature and time exposure enhanced the amount of fat content. Treatment at 80 ℃/15 min (lowest exposure level) represented the lowest fat content, while 90 ℃/30 min (medium exposure level) and 100 ℃/60 min (highest exposure level) represented medium and highest fat content, respectively.

The analysis of volatile compounds was carried out following previously described procedures (Dong et al., 2013; Ktenioudaki et al., 2013). Briefly, dried samples were mixed with distilled water at a ratio of 1:2 and closed properly. The volatiles were isolated by headspace solid phase microextraction (HS-SPME) using a GC-MS 5975 C (Agilent J&W, USA). The mixture was heated at 60 °C, and fiber (50/30 μm DVB/CAR/PDMS, Supelco – Sigma-Aldrich, Germany) was exposed to the headspace for 30 min. The length of the fibers in the headspace was kept constant. Helium was used as the carrier gas (1 mL/min). The separation of compounds was performed in a DB-5 column (30 m × 0.25 mm, df = 0.25 l m; Agilent J&W, USA). The injector, ion source and interface temperatures were set at 250, 200 and 260 °C, respectively. The mass spectrometer was operated in an electron impact mode, with the electron energy set at 70 eV and with a scan range of 40–400 m/z. The oven's temperature elevated from 40 to 250 °C at a rate of 4 °C/min, and the temperature was held constant for 5 min. The peak area was measured either by full scanning or by choosing specific fragments. Volatile compounds were tentatively identified using the spectra of reference compounds from NIST.

2.8. Statistical analysis

The data were collected from duplicate treatments and duplicate analyses at the minimum. One-way analysis of variance (ANOVA) and Tukey's post hoc test were performed for significant level at 95% ($p < 0.05$) using Statistica software (version 13.5.0.17, StatSoft GmbH, Germany). Furthermore, principal component analysis (PCA) was performed using the same software (Statistica software).

3. Results and discussion

3.1. Extracted fat content

As observed in Table 1, CWH at 80 °C/15 min lowered the extracted fat content statistically ($p < 0.05$), while 30 and 60 min time exposures generated the same amount of fat content ($p > 0.05$) compared to the control. Furthermore, the use of higher temperatures (90 and 100 °C) and 15 min exposure had the same level of fat content, while longer time exposures (30 and 60 min) increased the extracted fat content. The highest fat content was obtained at 100 °C/60 min, followed by a 100 °C/30 min treatment, while the lowest was at 80 °C/15 min. The fat contents in the current study ranged from 5.88% to 8.13%. The BSG in the current study had a lower value of fat content compared to previous observations, which ranged between 8% and 34% (Naibaho et al., 2021). An improvement in fat content due to the thermal exposure of BSG has been previously observed (Naibaho et al., 2021), and it was reported that this phenomenon may occur due to the ability of high temperatures to break the cell walls of the BSG matrix, thus releasing certain compounds (Ibbett et al., 2019). As a consequence, the modification of chemical constituents, such as dietary fiber, fats and fatty acids, proteins and amino acids and polyphenolic compounds, can be expected.

3.2. WHC and OHC

The results revealed that CWH significantly ($p < 0.05$) improved the WHC of treated BSG from 2.9 g/g to 3.4–3.8 g/g and increased OHC from 2.05 g/g to 2.09–2.16 g/g in the control and treated BSG, respectively. Among the treated groups, there was no significant difference ($p > 0.05$) in WHC, except between 90 and 100 °C at 60 min of heating. A higher OHC value was obtained with lower time exposures, except at 90 °C heating. The variability in WHC and OHC in the current study demonstrated that conventional water bath heating is capable of modifying the functionality of BSG, thus influencing the properties of BSG in the food matrix. This might be due to its influence on the release of certain compounds from BSG matrices, including phenolic and fatty acids and proteins. Moreover, this phenomenon led to different levels of hydroxyl group presence, hydrophobicity and lipophilicity in BSG with respect to the treatments. The WHC level change observed in the current study was in agreement with a previous report (Naibaho et al., 2021), in which the WHC value of autoclaved BSG was in the range of 2.9–3.3 g/g; meanwhile, the OHC values observed in the current study were higher than those reported previously, with OHC being in the range of 0.9–1.9 g/g in autoclaved BSG (Naibaho et al., 2021).

3.3. In vitro antioxidant capabilities

The results showed that CWH at 100 °C with time exposures of 30 or 60 min generated a significantly ($p < 0.05$) higher ABTS, heating at 90 °C with time exposures of 15 or 30 min significantly lowered the ABTS, and other treatments showed the same level as observed in the control. Furthermore, the majority of CWH treatments led to a similar FRAP value as in the control, except when heating at 80 °C for 30 min, under which a significant ($p < 0.05$) decrease was observed. These results demonstrate that CWH slightly influenced the ABTS value. The influence of thermal exposure on the ABTS capability of BSG has been previously observed: It has been reported that autoclaved BSG obtained higher ABTS and FRAP values compared to the control (Naibaho et al., 2022). It was reported that untreated BSG presented 0.8–2.1 mmol Trolox/Kg and 1.1–3.0 mmol Trolox/Kg for ABTS and FRAP, respectively (Naibaho et al., 2020). In other words, CWH-treated BSG had lower antioxidant properties in terms of ABTS and FRAP. ABTS indicates

the ability of the extract to reduce molecular oxygen and hydrogen peroxide (Benzie & Strain, 1996), while FRAP describes how the methanol extracts of BSG alleviate lipid oxidation (Rahman et al., 2021). In this regard, CWH had no influence on the lipid oxidation properties of treated BSG, and it slightly improved its ability for oxygen radical scavenging and hydrogen peroxide neutralization, particularly at 100 °C. This phenomenon may be due to the difference in material preparation, particularly in the drying process. Thermal exposure during drying induced the formation of melanoidin, which is responsible for higher antioxidant properties in BSG (Patrignani & González-Forte, 2021). In a previous report (Naibaho et al., 2020), the BSG was dried using convective drying, which required higher temperatures; in contrast, drying at lower temperatures was conducted in order to reduce the browning effect in materials.

3.4. Quantification of polyphenolic compounds

CWH at 80 °C for all time exposures significantly improved flavan-3-ol contents ($p < 0.05$), while 90 °C CWH decreased flavan-3-ols significantly, except at 60 min, which led to a higher amount than in the control. Treatments with 100 °C CWH produced the same levels of flavan-3-ols as in the control, except at 15 min, under which a lower amount was observed. Furthermore, all CWH decreased the phenolic acid contents. The impact of CWH on the total amount of polyphenolic compounds was seemingly similar as that for flavan-3-ols. The thermal exposure of BSG has been reported for its ability to disrupt cell vacuoles and/or cleave covalent bonds (Rahman et al., 2021). This phenomenon might lead to the modification of lignin solubility (Ohra-aho et al., 2016). Lignin consists of guaiacyl and syringyl functional groups, which bind similarly in all lignins (Ohra-aho et al., 2016). However, the amount and strength of the functional groups may vary (Ohra-aho et al., 2016); in this way, the variability of flavan-3-ols and the decline in phenolic acids due to CWH may have occurred as an effect of different levels of temperature and time exposures. CWH at 80 °C seemed to allow for the release of flavan-3-ols from the matrices, thus increasing their levels, while 90 and 100 °C facilitated the depolymerization and conversion of certain compounds into elementary units, thus decreasing the content of both flavan-3-ols and phenolic acid. Ferulic acids are bound to insoluble structural cellulose or hemicellulose by ester linkages (Sibhatu et al., 2021). Certain treatments might remove the ester-linked ferulic acid from insoluble cellulose, insoluble hemicellulose and lignin matrix, thus causing the content of ferulic acid to fluctuate (Sibhatu et al., 2021). The same phenomenon has been reported previously, in which caffeic acid was depolymerized into ferulic acid (Wojdyło et al., 2014) and ferulic acid was converted into 4-vinylguaiacol (Zago et al., 2022).

Hydroxycinnamic acids were the most abundant phenolic acids in BSG, including ferulic acid (FA), *p*-coumaric acid (p-CA) derivatives, FA derivatives, p-CA, caffeic acid (CA) and CA derivatives (McCarthy et al., 2013). However, their amount and/or presence depends on the extraction method and/or pre-treatment used (Rahman et al., 2021), thus leading to variable amounts of polyphenolic compounds, as was observed in the current study. For instance, it has been identified that caffeic acid was absent in lower temperature treatments, but it was present after exposure to higher temperatures (>100 °C), while sinapinic acid was identified after oven heating at 160 °C (Rahman et al., 2021). However, an investigation of specific phenolic compounds was not included in the current study. Therefore, further investigations should be conducted in order to investigate the impact of CWH on specific phenolic compounds.

3.5. Fatty acid profile

The fatty acid composition of BSG is presented in Table 2. In general, CWH significantly ($p < 0.05$) decreased the saturated fatty acid (SFA) content and increased the poly-unsaturated fatty acid (PUFA) content. The majority of treatments also enhanced the mono-unsaturated fatty acid (MUFA) content, except for the 100 °C treatment for 15 and 30 min, after which a lower amount was obtained compared to the untreated BSG. CWH discharged C17:0 at all levels of temperature and time exposures, while several fatty acids were formed depending on the specific treatment. All CWH treatments induced the formation of C18:0 and

Table 2
Fatty acids composition (% of total fatty acids) of water-bath heating treated spent grain.

Fatty acids (%)	Treatment									
	Control	80 °C/15 min	80 °C/30 min	80 °C/60 min	90 °C/15 min	90 °C/30 min	90 °C/60 min	100 °C/15 min	100 °C/30 min	100 °C/60 min
C13:0	–	0.39 ± 0.01	0.30 ± 0.01	0.29 ± 0.01	–	–	–	–	–	–
C14:0	–	0.36 ± 0.04	0.31 ± 0.01	–	–	–	–	–	–	–
C16:0	40.22 ± 0.00	19.57 ± 0.10	21.96 ± 0.01	22.29 ± 0.02	22.29 ± 0.01	21.30 ± 0.00	21.30 ± 0.01	22.22 ± 0.02	23.66 ± 0.01	21.51 ± 0.01
C16:1	–	0.49 ± 0.01	–	–	–	–	–	–	–	–
C17:0	4.93 ± 0.00	–	–	–	–	–	–	–	–	–
C18:0	–	3.18 ± 0.02	2.98 ± 0.01	3.04 ± 0.01	3.05 ± 0.01	3.13 ± 0.00	3.13 ± 0.01	3.88 ± 0.01	3.88 ± 0.01	3.43 ± 0.01
18:1 (n-9)	19.39 ± 0.00	16.17 ± 0.02	17.76 ± 0.01	17.73 ± 0.02	17.81 ± 0.01	17.90 ± 0.00	17.90 ± 0.01	17.28 ± 0.01	16.29 ± 0.01	17.46 ± 0.01
18:2 (n-6)	32.81 ± 0.00	48.83 ± 0.06	47.38 ± 0.01	47.42 ± 0.02	48.36 ± 0.01	48.11 ± 0.00	48.11 ± 0.01	48.27 ± 0.04	49.51 ± 0.01	48.94 ± 0.01
18:3 (n-3)	2.66 ± 0.00	5.20 ± 0.02	6.05 ± 0.01	5.94 ± 0.01	5.78 ± 0.01	5.95 ± 0.00	5.95 ± 0.00	5.49 ± 0.01	4.96 ± 0.01	5.77 ± 0.01
C20	–	0.79 ± 0.01	0.72 ± 0.02	0.77 ± 0.01	0.78 ± 0.01	0.82 ± 0.00	0.82 ± 0.00	0.82 ± 0.01	–	0.84 ± 0.01
C20:1	–	1.83 ± 0.01	1.81 ± 0.02	1.81 ± 0.01	1.93 ± 0.01	1.98 ± 0.00	1.98 ± 0.01	2.03 ± 0.01	1.71 ± 0.01	2.05 ± 0.01
C20:2	–	0.29 ± 0.01	–	–	–	–	–	–	–	–
C22:0	–	0.79 ± 0.01	0.71 ± 0.02	0.70 ± 0.01	–	0.80 ± 0.00	0.80 ± 0.01	–	–	–
C22:1	–	0.70 ± 0.01	–	–	–	–	–	–	–	–
C24:0	–	0.92 ± 0.01	–	–	–	–	–	–	–	–
C24:1	–	0.51 ± 0.01	–	–	–	–	–	–	–	–
Total SFA	45.15 ± 0.00^{a}	25.99 ± 0.07^{f}	26.98 ± 0.01^{d}	27.10 ± 0.01^{c}	26.12 ± 0.01^{e}	26.06 ± 0.00^{ef}	26.06 ± 0.01^{ef}	26.93 ± 0.02^{d}	27.53 ± 0.01^{b}	25.78 ± 0.01^{g}
Total MUFA	19.39 ± 0.00^{g}	19.69 ± 0.02^{c}	19.58 ± 0.01^{d}	19.54 ± 0.03^{e}	19.74 ± 0.01^{b}	19.89 ± 0.00^{a}	19.89 ± 0.01^{a}	19.31 ± 0.01^{h}	18.00 ± 0.01^{i}	19.51 ± 0.01^{f}
Total PUFA	35.46 ± 0.00^{i}	54.32 ± 0.07^{c}	53.44 ± 0.01^{g}	53.36 ± 0.02^{h}	54.14 ± 0.01^{d}	54.06 ± 0.00^{e}	54.06 ± 0.01^{e}	53.76 ± 0.05^{f}	54.47 ± 0.01^{b}	54.71 ± 0.01^{a}

Note: the data is shown as mean ± standard deviation with at least duplicate analysis. Letters show the significant differences from other treatment in the same row ($p < 0.05$). SFA: saturated fatty acid, MUFA: mono-unsaturated fatty acid, PUFA: poly-unsaturated fatty acid.

C20:1; almost all treatments formed C20:0, except for the 100 °C/30 min treatment; C22:0 was identified after 80 °C treatment for all time exposures, as well as 90 °C at 30 and 60 min; C13:0 was present after all 80 °C treatments, while 14:0 was only identified at 15 and 30 min. However, C16:1, C20:2, C22:1, C24:0 and C24:1 were only identified in BSG treated at 80 °C/15 min.

The results revealed that the main fatty acids in BSG were PUFAs C18:2(n-6) and C18:3(n-3), MUFA C18:1(n-9) and SFA C16:0. These results are in agreement with those previously reported (Mallen & Najdanovic-Visak, 2018). The increase in fatty acid yield has been observed due to an elevation in temperature (Mallen & Najdanovic-Visak, 2018) as a result of the increasing transesterification rate, which simultaneously improved the solubility and mass transfer of triglycerides (Mallen & Najdanovic-Visak, 2018). Increases in UFAs in BSG have also been identified previously as an effect of solid-state fermentation (Tan et al., 2019), which hydrolyzed lipids into fatty acids. In a similar manner, CWH might have induced the hydrolysis of lipids and/or intensified the transesterification rate, thus allowing for the formation of UFAs. Compared to the previous study, the fatty acids formed due to CWH, such as C13:0, C17:0, C20:2, C22:1 and C24:1, were not observed in BSG (Ibarruri et al., 2019); however, these fatty acids have been identified in dried and lyophilized BSG to a lesser extent (Fărcaş et al., 2015). Furthermore, the thermal exposure of BSG allowed for the release of certain functional groups from polysaccharides (Rahman et al., 2021). In this way, CWH might have induced the release of fatty acids from the functional groups of polysaccharides or from cell vacuoles. The results demonstrate that CWH improved the fatty acid properties of BSG, thus facilitating its application in industries such as pharmaceutics, cosmetics and functional foods. PUFAs are well-known for their benefits to human health, while SFAs have been recognized for their role in the induction of non-communicable diseases. Therefore, the ability of CWH to reduce SFA and enhancing PUFA contents might allow for greater benefits with respect to human health.

3.6. Volatile compounds profile

The volatile compound profile of BSG was assessed based on linear elevation treatments from the lowest to highest temperature and time exposure in order to investigate the impact of linear treatments on volatile compounds. Therefore, the evaluation of volatile compounds was carried out for the 80 °C/15 min, 90 °C/30 min and 100 °C/60 min treatments, and the results were compared with those of the untreated BSG. The results of the linear treatment's analysis regarding the volatile compound profile of BSG are presented in Table 3. The results show that the volatile compounds in BSG were quantitatively dominated by aldehydes, followed by ketones and alkanes, while several other groups were also present, including volatile fatty acids, alcohols, furans and others. In general, with higher temperatures and time exposures, the total aldehyde and furan contents will be higher. At the same time, the contents of ketones, alcohols, alkene and other groups declined.

Compared to the control, CWH significantly ($p < 0.05$) decreased the total amount of aldehydes. CWH eliminated 3-methyl-butanal, pentanal, (Z)-2-heptenal and (E)-2-hexenal in BSG. Those compounds have been identified in BSG in previous studies: 3-methyl-butanal has been reported to be responsible for buttery, oily, dark chocolate, cacao and almond odor perception; pentanal is responsible for almond, malt and pungent odor perception; (Z)-2-heptenal is responsible for green and pungent odor perception (Dong et al., 2013; Fărcaş et al., 2015; Ktenioudaki et al., 2013). However, the existence of (E)-2-hexenal has never been reported in BSG. (E)-2-hexenal has been identified as a green leaf volatile, and it has anti-fungal properties and is responsible for unpleasant odor, which deters fungi and insects (Kunishima et al., 2016). 3-Methyl-butanal, pentanal and (Z)-2-heptenal form during fermentation in the brewing process (Dong et al., 2013; Ktenioudaki et al., 2013), while (E)-2-hexenal might appear due to post-harvest handling or was naturally present in barley leaves, as many floral volatiles are present as

Table 3
Volatile compounds of water-bath heating treated spent grain (percentage of peak area).

Volatile compounds	BSG treatments			
	Control	80 °C/ 15 min	90 °C/ 30 min	100 °C/ 60 min
Aldehydes				
Butanal, 3-methyl-	4.39 ± 0.01	–	–	–
Pentanal	1.17 ± 0.01	–	–	–
Hexanal	16.94 ± 0.08	1.76 ± 0.00	15.18 ± 0.34	15.37 ± 0.09
2-Hexenal, (E)-	0.54 ± 0.00	–	–	–
Heptanal	1.23 ± 0.01	1.01 ± 0.01	1.05 ± 0.04	1.31 ± 0.07
2-Heptenal, (Z)-	1.10 ± 0.01	–	–	–
2,4-Heptadienal, (E,E)-	0.52 ± 0.00	–	–	0.38 ± 0.03
Octanal	–	–	–	1.34 ± 0.04
2-Octenal, (E)-	3.55 ± 0.01	3.15 ± 0.11	4.53 ± 0.12	2.29 ± 0.14
Nonanal	10.90 ± 0.15	10.08 ± 0.02	7.23 ± 0.09	8.98 ± 0.01
2-Nonenal, (E)-	3.00 ± 0.01	2.19 ± 0.08	3.04 ± 0.11	2.84 ± 0.07
2,4-Nonadienal, (E,E)-	0.86 ± 0.01	0.62 ± 0.04	0.53 ± 0.04	0.39 ± 0.03
Decanal	1.88 ± 0.02	1.87 ± 0.08	1.73 ± 0.08	1.50 ± 0.08
2,4-Decadienal, (E,E)-	–	–	1.42 ± 0.08	5.02 ± 0.04
Undecanal	0.35 ± 0.00	–	–	3.59 ± 0.06
Octadecanal, 2-bromo-	–	0.59 ± 0.04	–	1.78 ± 0.01
Benzaldehyde	4.83 ± 0.03	11.80 ± 0.14	4.76 ± 0.08	4.81 ± 0.00
Benzeneacetaldehyde	6.96 ± 0.09	3.34 ± 0.05	7.40 ± 0.09	7.06 ± 0.11
Ketones				
Acetophenone	–	0.31 ± 0.01	–	–
2-Hexanone, 5-methyl-	–	–	0.57 ± 0.03	–
2-Heptanone	1.21 ± 0.05	–	0.70 ± 0.04	1.09 ± 0.04
5-Hepten-2-one, 6-methyl-	0.60 ± 0.05	–	–	–
3-Octen-2-one, (E)-	1.92 ± 0.14	1.39 ± 0.08	3.38 ± 0.04	0.95 ± 0.07
3-Octanone, 2-methyl-	0.86 ± 0.02	–	–	–
3,5-Octadien-2-one, (E,E)-	7.36 ± 0.47	6.80 ± 0.21	10.04 ± 0.38	6.94 ± 0.30
5,9-Undecadien-2-one, 6,10-dimethyl-, (E)-	0.43 ± 0.02	0.31 ± 0.00	–	–
2-Undecanone	–	6.48 ± 0.15	–	2.56 ± 0.04
2(3H)-Furanone, 5-heptyldihydro-	0.61 ± 0.02	0.30 ± 0.01	0.42 ± 0.02	–
Alcohols				
Ethanol, 2-(9,12-octadecadienyloxy)-, (Z,Z)-	–	0.33 ± 0.01	–	–
1-Hexanol	–	0.29 ± 0.02	–	–
1-Hexanol, 2-ethyl-	–	4.23 ± 0.02	–	–
1-Octen-3-ol	1.76 ± 0.14	1.16 ± 0.02	2.34 ± 0.04	1.49 ± 0.04

(continued on next page)

Table 3 (*continued*)

Volatile compounds	BSG treatments			
	Control	80 °C/ 15 min	90 °C/ 30 min	100 °C/ 60 min
2-Octen-1-ol, (Z)-	0.42 ± 0.02	0.46 ± 0.01	0.43 ± 0.02	0.45 ± 0.03
5-Octen-2-yn-4-ol	–	–	0.59 ± 0.02	–
Nona-3,5-dien-2-ol	0.55 ± 0.02	–	–	–
9-Oxabicyclo[6.1.0]nonan-4-ol	–	–	0.60 ± 0.04	–
2-Nitrohept-2-en-1-ol	0.39 ± 0.01	–	–	–
4,4,6-Trimethyl-cyclohex-2-en-1-ol	1.08 ± 0.06	–	–	–
2-Butyl-2,7-octadien-1-ol	0.56 ± 0.04	–	–	–
1-Decanol, 2-hexyl-	–	–	–	0.73 ± 0.04
1-Tetradecanol	0.37 ± 0.03	–	–	–
1-Hexadecanol	1.09 ± 0.09	–	0.60 ± 0.03	–
n-Nonadecanol-1	–	–	0.42 ± 0.03	–
4,4,6-Trimethyl-cyclohex-2-en-1-ol	–	–	0.71 ± 0.01	–
9-(3,3-Dimethyloxiran-2-yl)-2,7-dimethylnona-2,6-dien-1-ol	–	–	–	0.25 ± 0.01
Furan				
Furan, 2-pentyl-	7.44 ± 0.46	7.28 ± 0.07	8.47 ± 0.16	9.22 ± 0.19
Alkane				
Tridecane	4.62 ± 0.05	5.36 ± 0.00	6.66 ± 0.05	6.91 ± 0.08
Tetradecane, 2,6,10-trimethyl-	1.94 ± 0.04	–	–	–
Tetradecane	1.09 ± 0.05	5.06 ± 0.00	0.63 ± 0.03	–
1-Pentadecene	–	–	0.44 ± 0.01	0.53 ± 0.01
Heptacosane	–	–	1.42 ± 0.08	–
Hexadecane, 1,1-bis (dodecyloxy)-	–	0.81 ± 0.08	0.37 ± 0.01	–
Nonadecane	0.40 ± 0.01	–	–	–
Dodecane	3.32 ± 0.09	7.55 ± 0.07	7.11 ± 0.11	6.69 ± 0.01
Octadecane, 3-ethyl-5-(2-ethylbutyl)-	–	2.15 ± 0.08	0.72 ± 0.01	1.14 ± 0.02
Undecane	–	–	0.58 ± 0.03	0.54 ± 0.01
Eicosane	–	–	0.51 ± 0.01	–
Heneicosane	–	–	0.97 ± 0.02	–
Fatty acids				
Hexanoic acid	0.93 ± 0.05	1.91 ± 0.04	–	1.96 ± 0.03
Dodecanoic acid, 3-hydroxy-	–	–	0.41 ± 0.02	–
Others				
D-Limonene	1.58 ± 0.09	–	1.97 ± 0.03	1.54 ± 0.05
Benzeneethanamine, 2,5-difluoro-β,3,4-trihydroxy-N-methyl-	1.26 ± 0.07	–	–	0.35 ± 0.02
Benzene, 1-methyl-3-(1-methylethyl)-	–	0.43 ± 0.01	0.62 ± 0.02	–
5-Benzylidene-3-(3,4-dimethylanilinomethyl)-2,4-thiazolidinedione	–	–	0.34 ± 0.01	–
Ethyl Acetate	–	8.36 ± 0.04	–	–
1R-α-Pinene	–	0.35 ± 0.01	–	–
Oxime-, methoxy-phenyl-	–	2.28 ± 0.04	1.09 ± 0.01	–
TOTAL				
Aldehydes	58.21 ± 0.07^{a}	36.40 ± 0.10^{d}	46.87 ± 0.08^{c}	56.67 ± 0.22^{b}
Ketones	12.99 ± 0.63^{b}	15.58 ± 0.13^{a}	15.11 ± 0.46^{a}	11.54 ± 0.29^{b}
Alcohols	6.22 ± 0.11^{a}	6.48 ± 0.03^{a}	5.70 ± 0.18^{b}	2.92 ± 0.05^{c}
Furans	7.44 ± 0.46^{bc}	7.28 ± 0.03^{c}	8.47 ± 0.16^{ab}	9.22 ± 0.19^{a}
Alkane	11.38 ± 0.02^{d}	20.93 ± 0.03^{a}	19.42 ± 0.05^{b}	15.80 ± 0.12^{c}
Fatty acid	0.93 ± 0.05^{b}	1.91 ± 0.04^{a}	0.41 ± 0.02^{c}	1.96 ± 0.03^{a}
Others	2.83 ± 0.16^{c}	11.42 ± 0.07^{a}	4.01 ± 0.07^{b}	1.89 ± 0.07^{d}

Note: the data is shown as mean ± standard deviation with at least duplicate analysis. Letters show the significant differences from other treatment in the same row ($p < 0.05$).

anti-fungal compounds in plants.

Furthermore, CWH induced the formation of octanal, (E,E)-2,4-decadienal and 2-bromo-octadecanal. Octanal has been identified in barley and malt, but it was absent in BSG (Fărcaş et al., 2015). Therefore, CWH may have reformed the octanal as in its original form. Octanal exhibits fat, soap, lemon and green odor perceptions (Dong et al., 2013). (E,E)-2,4-decadienal has not been identified in BSG; however, it was present in bread prepared with dried distilled grain, and it was responsible for rancid odor perceptions (Roth et al., 2016). However, this odor had an odor activity value at 23.4%, which is much higher than that observed in the current study. The highest amount of (E,E)-2,4-decadienal in the current study was 5.02%. In this way, rancid odor might not be detected by the human sense of smell.

CWH at 80 °C/15 min and 90 °C/30 min significantly increased ($p < 0.05$) the total ketones in BSG, while 100 °C/60 min generated the same level as in the control. CWH discharged 6-methyl-5-hepten-2-one and 2-methyl-3-octanone in all elevation levels. 6-Methyl-5-hepten-2-one is responsible for herb, oily, pungent, pear, pepper and mushroom odor perceptions (Dong et al., 2013), and it has been reported to be present in BSG-added crackers (O'Shea et al., 2017) and grain malts (Dong et al., 2013). The formation of several ketones was identified after CWH treatment, such as acetophenone (80 °C/15 min), 5-methyl-2-hexanone (90 °C/30 min) and 2-undecanone (80 °C/15 min and 100 °C/60 min). Acetophenone represents a sweet, floral and almond odor perception (Fărcaş et al., 2015). Notably, 2-undecanone and 5-methyl-2-hexanone have never been reported in BSG; however, 2-undecanone has been identified as the second most-abundant ketone in UHT milk (Dursun et al., 2017), while 5-methyl-2-hexanone was present in black tea (Yan et al., 2022). Methyl ketones predominantly originate in the lipid fraction (Dursun et al., 2017); thus, their presence may depend on lipid degradation due to thermal exposure.

CWH discharged the majority of alcohols present in untreated BSG: eight alcohols were identified in the original BSG, while only two

remained in the CWH-treated BSG, including 1-octen-3-ol and (Z)-2-octen-1-ol. Several alcohols formed due to CWH, including (Z,Z)-2-(9,12-octadecadienyloxy)-ethanol, 1-hexanol and 2-ethyl-1-hexanol at 80 °C/15 min; 5-octen-2-yn-1-ol, 9-oxabicyclo[6.1.0]nonan-4-ol, n-nonadecanol-1 and 4,4,6-trimethyl-cyclohex2-en-1-ol at 90 °C/30 min; and 2-hexyl-1-decanol and 9-(3,3-Dimethyloxiran-2-yl)-2,7-dimethylnona-2,6-dien-1-ol at 100 °C/60 min. Among those formed alcohol compounds, only 1-hexanol and 2-ethyl-1-hexanol have been identified in grain (Buśko et al., 2010; Dong et al., 2015), which are responsible for a resin, flower and green odor perception (Dong et al., 2013). Meanwhile, (Z,Z)-2-(9,12-octadecadienyloxy)-ethanol, 5-octen-2-yn-4-ol, 9-oxabicyclo[6.1.0]nonan-4-ol, N-nonadecanol-1 and 2-hexyl-1-decanol have been observed as phytochemical constituents in several plant extracts (Abirami & Gomathi, 2022; Nazir et al., 2021).

The CWH treatment generated higher levels of alkanes. 2,6,10-Trimethyl-tetradecane and nonadecane were absent due to the treatment; however, several compounds formed, including heptacosane, 1,1-bis (dodecyloxy)-hexadecane, 3-ethyl-5- (2-ethylbutyl)- octadecane, undecane, eicosane and heneicosane. These compounds have not been previously identified in BSG. However, undecane and eicosane have been reported to be present in grain (Buśko et al., 2010), while heneicosane has been observed in sorghum grain tea (Xiong et al., 2020). Other compounds, including 2,6,10-trimethyl-tetradecane, nonadecane, heptacosane, 1,1-bis(dodecyloxy)-hexadecane and 3-ethyl-5-(2-ethylbutyl)-octadecane have been identified in essential oils and as micro-organism secondary metabolites (Alqahtani et al., 2022; Wei & Fan, 2020).

Additionally, the 90 °C/30 min CWH treatment induced the formation of 3-hydroxy-dodecanoic acid and 2,4-thiazolidinedione; 80 °C/15 min CWH led to the formation of ethyl acetate and 1R-α-Pinene; 1-methyl-3-(1-methylethyl)-benzene and methoxy-phenyl-oxime were identified in both 80 °C/15 min and 90 °C/30 min treatments; 1-pentadecene (an essential oil) was identified in 90 °C/30 min and 100 °C/60 min treatments. The ability of CWH to degrade certain compounds while forming others might be beneficial for the further valorization of BSG. It was discovered that most degraded compounds are responsible for pungent, floral and spice odor perception, while the formed compounds are responsible for certain pleasant perceptions, such as fruity, sweet and essential oil odors. Therefore, such conversions are expected to broaden the utilization of BSG in food ingredients.

3.7. Principal component analysis (PCA)

PCA was conducted with respect to the linear elevation treatments, as in the volatile compound analysis in addition to the untreated BSG. As shown in Fig. 1, untreated BSG tended to be aligned with higher total polyphenolics and SFAs. CWH at lower time and thermal exposure (80 °C/15 min) significantly increased the presence of flavan-3-ols, ketones and alcohols, as well as other volatile compounds. Increasing the temperature and time exposure (to 90 °C/30 min) seemed to improve MUFA and PUFA formation, and it enhanced the functional properties of BSG. Furthermore, CWH treatments at 100 °C/60 min increased the fat content, aldehyde content and antioxidant properties of BSG.

In summary, different temperature and time exposure levels can be utilized depending on the target compounds. This study provides beneficial initial information for further investigations, including those focused on other biological properties, amino acids and peptides, aromatic compounds and specific phenolic compounds.

4. Conclusions

As was hypothesized, it was demonstrated the improvements in the functionality of BSG, including WHC and OHC, after CWH treatment. Treatment at 100 °C for 30 or 60 min generated a higher ABTS capability, while the majority of treatments had no significant influence on FRAP levels. All treatments at 80 °C led to higher total flavan-3-ol contents, while all treatments decreased the total phenolic acids. A significant increase in the amount of PUFAs was observed due to CWH treatments converting SFAs into double-bond fatty acids, thus allowing for the higher production of PUFAs from BSG and potentially promoting its use as a functional food ingredient or for nutraceutical purposes. Moreover, CWH tended to degrade the pungent, floral, spice and mushroom odor perceptions of untreated BSG while inducing the formation of compounds responsible for fruity, sweet and pleasant odors, as well as essential oil-related compounds. The further investigation of other *in vitro* antioxidant activities is seemingly important, particularly in relation to the PUFA's composition, as well as the identification of key aromatic compounds via sensory and olfactory analyses.

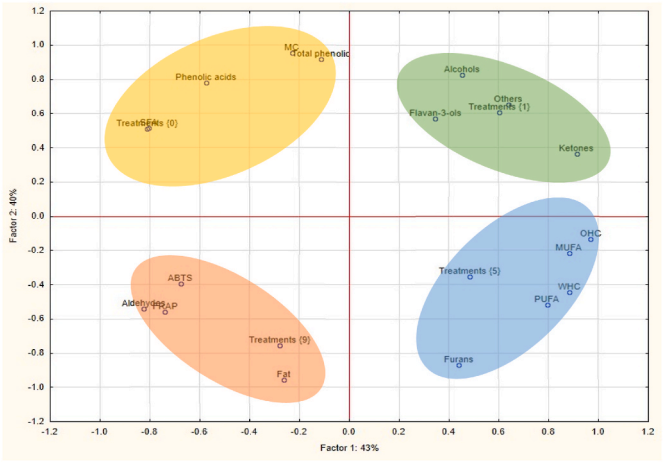

Fig. 1. Principal component analysis (PCA) of conventional water-bath heating on chemical composition, antioxidant properties and techno-functionality of treated spent grain (MC: moisture content, SFA: saturated fatty acid, ABTS: 2,2′-Azinobis-(3-ethylbenzthiazoline-6-sulfonic acid), FRAP: ferric-reducing antioxidant power, MUFA: mono-unsaturated fatty acid, PUFA: poly-unsaturated fatty acid, WHC: water holding capacity, OHC: Oil holding capacity).

Author contributions

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Declaration of competing interest

None.

Data availability

The data has been included in the manuscript

Acknowledgements

This work was supported by UPWR 2.0, international and interdisciplinary programme of development of Wrocław University of Environmental and Life Sciences, co-financed by the European Social Fund under the Operational Programme Knowledge Education Development 2014–2020: Axis III Higher education for the economy and development; Action 3.5. Comprehensive programmes for schools of higher education (POWR.03.05.00–00-Z062/18).

It was also supported by ERA-NET CO-FUND Horizon 2020 - FACCE SURPLUS Sustainable and Resilient Agriculture for Food and Non-Food Systems and PROWASTE Protein-fibre biorefinery for scattered material streams (2019–2021).

References

Abirami, D., & Gomathi, R. (2022). Target and candidate agents for diabetes treatment in the framework of the food nexus. *Energy Nexus, 5*, Article 100041. https://doi.org/10.1016/j.nexus.2022.100041

Alqahtani, S. S., Moni, S. S., Sultan, M. H., Ali Bakkari, M., Madkhali, O. A., Alshahrani, S., Makeen, H. A., Joseph Menachery, S., ur Rehman, Z., Shamsher Alam, M., Mohan, S., Eltaib Elmobark, M., Banji, D., & Z. Sayed-Ahmed, M. (2022). Potential bioactive secondary metabolites of *Actinomycetes sp.* Isolated from rocky soils of the heritage village Rijal Alma, Saudi Arabia. *Arabian Journal of Chemistry, 15*(5), Article 103793. https://doi.org/10.1016/j.arabjc.2022.103793

Benzie, I. F. F., & Strain, J. J. (1996). The ferric reducing ability of plasma (FRAP) as a measure of "antioxidant power": The FRAP assay. *Analytical Biochemistry, 239*(1), 70–76. https://doi.org/10.1006/abio.1996.0292

Buśko, M., Jeleń, H., Góral, T., Chmielewski, J., Stuper, K., Szwajkowska-Michałek, L., Tyrakowska, B., & Perkowski, J. (2010). Volatile metabolites in various cereal grains. *Food Additives & Contaminants: Part A, 27*(11), 1574–1581. https://doi.org/10.1080/19440049.2010.506600

Connolly, A., Cermeño, M., Crowley, D., O'Callaghan, Y., O'Brien, N. M., & FitzGerald, R. J. (2019). Characterisation of the in vitro bioactive properties of alkaline and enzyme extracted brewers' spent grain protein hydrolysates. *Food Research International, 121*, 524–532. https://doi.org/10.1016/j.foodres.2018.12.008

Dong, L., Hou, Y., Li, F., Piao, Y., Zhang, X., Zhang, X., Li, C., & Zhao, C. (2015). Characterization of volatile aroma compounds in different brewing barley cultivars: Characterization of volatile aroma compounds in brewing barley. *Journal of the Science of Food and Agriculture, 95*(5), 915–921. https://doi.org/10.1002/jsfa.6759

Dong, L., Piao, Y., Zhang, X., Zhao, C., Hou, Y., & Shi, Z. (2013). Analysis of volatile compounds from a malting process using headspace solid-phase micro-extraction and GC–MS. *Food Research International, 51*(2), 783–789. https://doi.org/10.1016/j.foodres.2013.01.052

Dursun, A., Güler, Z., & Şekerli, Y. E. (2017). Characterization of volatile compounds and organic acids in ultra-high-temperature milk packaged in tetra brik cartons. *International Journal of Food Properties, 20*(7), 1511–1521. https://doi.org/10.1080/10942912.2016.1213280

Fărcaş, A. C., Socaci, S. A., Dulf, F. V., Tofană, M., Mudura, E., & Diaconeasa, Z. (2015). 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. *Journal of Cereal Science, 64*, 34–42. https://doi.org/10.1016/j.jcs.2015.04.003

Ibarruri, J., Cebrián, M., & Hernández, I. (2019). Solid state fermentation of brewer's spent grain using *Rhizopus sp.* to enhance nutritional value. *Waste and Biomass Valorization, 10*(12), 3687–3700. https://doi.org/10.1007/s12649-019-00654-5

Ibbett, R., White, R., Tucker, G., & Foster, T. (2019). Hydro-mechanical processing of brewer's spent grain as a novel route for separation of protein products with differentiated techno-functional properties. *Innovative Food Science & Emerging Technologies, 56*, Article 102184. https://doi.org/10.1016/j.ifset.2019.102184

Jung, H., Moon, J. H., Park, J. W., & Yoon, W. B. (2020). Texture of surimi-canned corn mixed gels with conventional water bath cooking and ohmic heating. *Food Bioscience, 35*, Article 100580. https://doi.org/10.1016/j.fbio.2020.100580

Ktenioudaki, A., O'Shea, N., & Gallagher, E. (2013). Rheological properties of wheat dough supplemented with functional by-products of food processing: Brewer's spent grain and apple pomace. *Journal of Food Engineering, 116*(2), 362–368. https://doi.org/10.1016/j.jfoodeng.2012.12.005

Kumari, B., Tiwari, B. K., Walsh, D., Griffin, T. P., Islam, N., Lyng, J. G., Brunton, N. P., & Rai, D. K. (2019). Impact of pulsed electric field pre-treatment on nutritional and polyphenolic contents and bioactivities of light and dark brewer's spent grains. *Innovative Food Science & Emerging Technologies, 54*, 200–210. https://doi.org/10.1016/j.ifset.2019.04.012

Kunishima, M., Yamauchi, Y., Mizutani, M., Kuse, M., Takikawa, H., & Sugimoto, Y. (2016). Identification of (Z)-3:(E)-2-hexenal isomerases essential to the production of the leaf aldehyde in plants. *Journal of Biological Chemistry, 291*(27), 14023–14033. https://doi.org/10.1074/jbc.M116.726687

Mallen, E., & Najdanovic-Visak, V. (2018). Brewers' spent grains: Drying kinetics and biodiesel production. *Bioresource Technology Reports, 1*, 16–23. https://doi.org/10.1016/j.biteb.2018.01.005

Martín-García, B., Tylewicz, U., Verardo, V., Pasini, F., Gómez-Caravaca, A. M., Caboni, M. F., & Dalla Rosa, M. (2020). Pulsed electric field (PEF) as pre-treatment to improve the phenolic compounds recovery from brewers' spent grains. *Innovative Food Science & Emerging Technologies, 64*, Article 102402. https://doi.org/10.1016/j.ifset.2020.102402

McCarthy, A. L., O'Callaghan, Y. C., Neugart, S., Piggott, C. O., Connolly, A., Jansen, M. A. K., Krumbein, A., Schreiner, M., FitzGerald, R. J., & O'Brien, N. M. (2013). The hydroxycinnamic acid content of barley and brewers' spent grain (BSG) and the potential to incorporate phenolic extracts of BSG as antioxidants into fruit beverages. *Food Chemistry, 141*(3), 2567–2574. https://doi.org/10.1016/j.foodchem.2013.05.048

Naibaho, J., & Korzeniowska, M. (2021). Brewers' spent grain in food systems: Processing and final products quality as a function of fiber modification treatment. *Journal of Food Science, 86*(5), 1532–1551. https://doi.org/10.1111/1750-3841.15714

Naibaho, J., Korzeniowska, M., Wojdyło, A., Figiel, A., Yang, B., Laaksonen, O., Foste, M., Vilu, R., & Viiard, E. (2020). The potential of spent barley as a functional food ingredient: Study on the comparison of dietary fiber and bioactivity. In Proceedings of. *The 1st International Electronic Conference on Food Science and Functional Foods, 86*. https://doi.org/10.3390/foods_2020-08486

Naibaho, J., Korzeniowska, M., Wojdyło, A., Figiel, A., Yang, B., Laaksonen, O., Foste, M., Vilu, R., & Viiard, E. (2021). Fiber modification of brewers' spent grain by autoclave treatment to improve its properties as a functional food ingredient. *LWT - Food Science and Technology, 149*, Article 111877. https://doi.org/10.1016/j.lwt.2021.111877

Naibaho, J., Wojdyło, A., Korzeniowska, M., Laaksonen, O., Föste, M., Kütt, M.-L., & Yang, B. (2022). Antioxidant activities and polyphenolic identification by UPLC-MS/MS of autoclaved brewers' spent grain. *LWT - Food Science and Technology*. , Article 113612. https://doi.org/10.1016/j.lwt.2022.113612

Nazir, S., El-Sherif, A. A., Abdel-Ghani, N. T., Ibrahim, M. A. A., Hegazy, M.-E. F., & Atia, M. A. M. (2021). *Lepidium sativum* secondary metabolites (essential oils): *In vitro* and *in silico* studies on human hepatocellular carcinoma cell lines. *Plants, 10*(9), 1863. https://doi.org/10.3390/plants10091863

Nowacki, D., Martynowicz, H., Skoczyńska, A., Wojakowska, A., Turczyn, B., Bobak, Ł., Trziszka, T., & Szuba, A. (2017). Lecithin derived from ω-3 PUFA fortified eggs decreases blood pressure in spontaneously hypertensive rats. *Scientific Reports, 7*(1), Article 12373. https://doi.org/10.1038/s41598-017-12019-w

Ohra-aho, T., Niemi, P., Aura, A.-M., Orlandi, M., Poutanen, K., Buchert, J., & Tamminen, T. (2016). Structure of brewer's spent grain lignin and its interactions with gut microbiota *in vitro*. *Journal of Agricultural and Food Chemistry, 64*(4), 812–820. https://doi.org/10.1021/acs.jafc.5b05535

O'Shea, N., Kilcawley, K. N., & Gallagher, E. (2017). Aromatic composition and physicochemical characteristics of crackers containing barley fractions. *Cereal Chemistry Journal, 94*(3), 611–618. https://doi.org/10.1094/CCHEM-10-16-0256-R

Patrignani, M., González-Forte, L., & del, S. (2021). Characterisation of melanoidins derived from Brewers' spent grain: New insights into their structure and antioxidant activity. *International Journal of Food Science and Technology, 56*(1), 384–391. https://doi.org/10.1111/ijfs.14653

Rahman, M. J., Malunga, L. N., Eskin, M., Eck, P., Thandapilly, S. J., & Thiyam-Hollander, U. (2021). Valorization of heat-treated brewers' spent grain through the identification of bioactive phenolics by UPLC-PDA and evaluation of their antioxidant activities. *Frontiers in Nutrition, 8*, Article 634519. https://doi.org/10.3389/fnut.2021.634519

Roth, M., Schuster, H., Kollmannsberger, H., Jekle, M., & Becker, T. (2016). Changes in aroma composition and sensory properties provided by distiller's grains addition to bakery products. *Journal of Cereal Science, 72*, 75–83. https://doi.org/10.1016/j.jcs.2016.10.002

Sibhatu, H. K., Anuradha Jabasingh, S., Yimam, A., & Ahmed, S. (2021). Ferulic acid production from brewery spent grains, an agro-industrial waste. *LWT - Food Science and Technology, 135*, Article 110009. https://doi.org/10.1016/j.lwt.2020.110009

Tan, Y. X., Mok, W. K., Lee, J., Kim, J., & Chen, W. N. (2019). Solid state fermentation of brewers' spent grains for improved nutritional profile using *Bacillus subtilis WX-17*. *Fermentation, 5*(3), 52. https://doi.org/10.3390/fermentation5030052

Tkacz, K., Wojdyło, A., Turkiewicz, I. P., & Nowicka, P. (2021). Triterpenoids, phenolic compounds, macro- and microelements in anatomical parts of sea buckthorn (*Hippophaë rhamnoides L.*) berries, branches and leaves. *Journal of Food Composition and Analysis, 103*, Article 104107. https://doi.org/10.1016/j.jfca.2021.104107

Verni, M., Pontonio, E., Krona, A., Jacob, S., Pinto, D., Rinaldi, F., Verardo, V., Díaz-de-Cerio, E., Coda, R., & Rizzello, C. G. (2020). Bioprocessing of brewers' spent grain enhances its antioxidant activity: Characterization of phenolic compounds and bioactive peptides. *Frontiers in Microbiology, 11*, 1831. https://doi.org/10.3389/fmicb.2020.01831

Wei, Q., & Fan, W. (2020). Compositions of volatile oil from flowers, leaves and stems of *Triadica sebifera*. *Journal of Essential Oil Bearing Plants, 23*(3), 633–637. https://doi.org/10.1080/0972060X.2020.1811779

Wojdyło, A., Figiel, A., Lech, K., Nowicka, P., & Oszmiański, J. (2014). Effect of convective and vacuum–microwave drying on the bioactive compounds, color, and antioxidant capacity of sour cherries. *Food and Bioprocess Technology, 7*(3), 829–841. https://doi.org/10.1007/s11947-013-1130-8

Xiong, Y., Zhang, P., Johnson, S., Luo, J., & Fang, Z. (2020). Comparison of the phenolic contents, antioxidant activity and volatile compounds of different sorghum varieties during tea processing. *Journal of the Science of Food and Agriculture, 100*(3), 978–985. https://doi.org/10.1002/jsfa.10090

Yan, T., Lin, J., Zhu, J., Ye, N., Huang, J., Wang, P., Jin, S., Zheng, D., & Yang, J. (2022). Aroma analysis of Fuyun 6 and Jinguanyin black tea in the Fu'an area based on E-nose and GC–MS. *European Food Research and Technology, 248*, 947–961. https://doi.org/10.1007/s00217-021-03930-8

Ye, Y., Liu, X., Bai, W., Zhao, W., Zhang, Y., Dong, H., & Pan, Z. (2022). Effect of microwave-ultrasonic combination treatment on heating-induced gel properties of low-sodium tilapia surimi during gel setting stage and comparative analysis. *LWT - Food Science and Technology, 161*, Article 113386. https://doi.org/10.1016/j.lwt.2022.113386

Zago, E., Tillier, C., De Leener, G., Nandasiri, R., Delporte, C., Bernaerts, K. V., & Shavandi, A. (2022). Sustainable production of low molecular weight phenolic compounds from Belgian brewers' spent grain. *Bioresource Technology Reports, 17*, Article 100964. https://doi.org/10.1016/j.biteb.2022.100964

Zhang, H., Yang, J., & Zhao, Y. (2015). High intensity ultrasound assisted heating to improve solubility, antioxidant and antibacterial properties of chitosan-fructose Maillard reaction products. *LWT - Food Science and Technology, 60*(1), 253–262. https://doi.org/10.1016/j.lwt.2014.07.050

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March 27, 2023

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data i podpis

Publication 6

Journal of Cereal Science 107 (2022) 103524

Contents lists available at ScienceDirect

Journal of Cereal Science

journal homepage: www.elsevier.com/locate/jcs

Techno-functional properties of protein from protease-treated brewers' spent grain (BSG) and investigation of antioxidant activity of extracted proteins and BSG residues

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ARTICLE INFO

Keywords:
Protein functionality
Antioxidant activities
Polyphenolic identification
Agro-industrial byproduct

ABSTRACT

The study aimed to investigate the biological properties of the protein fraction of brewers' spent grain (BSG) and its sediments as well as the techno-functional properties of BSG protein (BSGP). BSG was incubated with 0.5% protamex and a combination of protamex and flavourzyme, in addition to the control (incubation without protease). The results showed that enzymatic treatment enhanced the antioxidant activity of BSGP. Compared to the sediment fraction, BSGPs had higher antioxidant capacities than those in sediments. The current study demonstrated that the FRAP value is aligned with the amount of the polyphenolic compounds, while BSGP is responsible for ORAC and ABTS capabilities. Enzyme treatment on BSG enhanced the antioxidant properties of BSGPs and the amount of the phenolic compounds of the sediments. BSGPs treated with proteases possessed higher oil-holding capacity, foaming properties and lower emulsion capability. In conclusion, enzymatic treatment of BSG enhanced the protein functionality and bioactivity as well as intensified the antioxidant activity of its sediments allowing further valorization.

1. Introduction

Brewers' spent grain (BSG), a byproduct of the brewery industry, consists of 15–30% protein (Naibaho and Korzeniowska, 2021a; Wen et al., 2019). BSG protein is mainly dominated by hordein, glutelin, globulin and albumin (Wen et al., 2019), and its amino acids are mainly dominated by glutamine, proline and leucine (Connolly et al., 2013). The number of amino acids in BSG has been reported to be the same as that in barley (Cermeño et al., 2019). BSG protein is reported for its benefits to human health due to its biological properties including anti-inflammatory activity, antithrombotic and blood coagulation, angiotensin-converting enzyme activity, modulation of glycemic response, dipeptidyl peptidase IV inhibitory, and protective ability against oxidant such as DPPH (di(phenyl)- (2; 4;6-trinitrophenyl)iminoazanium), FRAP (ferric-reducing antioxidant power), ABTS (2, 2′-Azinobis-(3-ethylbenzthiazoline-6-Sulfonic Acid)) and ORAC (oxygen radical absorbance capacity) (Wen et al., 2019). Therefore, protein extraction from the BSG matrix is continuously examined and optimized (Wen et al., 2019).

One of the crucial factors in protein extraction from BSG is maintaining pH at approximately 8–9 (Vieira et al., 2016), thus improving the solubility of the protein. Consequently, a higher yield will be generated (Cian et al., 2018). Furthermore, incorporation of enzymes has been observed for its ability to drop the molecular weight of the protein, thus increasing the digestibility (Vieira et al., 2016; Wen et al., 2019). Several enzymes such as flavourzyme, corolase, alcalase and Promod 144 MG

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https://doi.org/10.1016/j.jcs.2022.103524
Received 1 April 2022; Received in revised form 1 July 2022; Accepted 7 July 2022
Available online 15 July 2022

with different concentrations were observed to enhance the yield of obtained protein at a broader pH range of 2.0–12.00 (Cermeño et al., 2019; Connolly et al., 2014, 2015). The studies revealed that different enzymes and their amounts influenced the biological properties of obtained proteins. Flavourzyme (1 and 2.5%) improved the biological properties of protein from BSG (McCarthy et al., 2013). A combination of flavourzyme with alkaline and neutral protease in BSG generated a protein with a higher biological activity (Cian et al., 2018). Flavourzyme promoted protein decomposition, thus enhancing more peptides and amino acids (Yang et al., 2020). Protamex lowered the degree of hydrolysis of protein obtained from Persian lime seeds while it had a higher functionality at a broader pH range (Fathollahy et al., 2021).

A combination of flavourzyme and protamex improved the quality of BSG as an animal feed and masked the bitter taste from protamex (San Martin et al., 2020). However, the study on its impact on the functionality of extracted protein has never been reported. The study aimed to compare and evaluate the impact of protamex, flavourzyme and combined protamex–flavourzyme on the functionality of BSG extracted protein. It was expected that different enzymes would generate protein with different biological properties, polyphenolic compositions and techno-functional properties such as water solubility, foaming and emulsion formation as well as stability. The results would be beneficial for industries and scientists in terms of the further utilization of protein extracts obtained from BSG.

2. Materials and methods

2.1. Materials

The BSG samples were collected from a light-type beer-producer brewery in Poland, dried with conventional drying methods at 70 °C–75 °C for approximately 3.5 h to reach a stable moisture (2–5%), ground to a 500 mm particle size, and kept at 10 °C prior to the extract preparation. Trolox (6-hydro-2,5,7,8- tetramethylchroman-2-carboxylic acid) was purchased from Sigma-Aldrich (Steinheim, Germany). UPLC-grade water was prepared by using the HLP SMART 1000s system (Hydrolab, Gdansk, Poland). Before use, the water was filtered using a 0.22 μm membrane filter immediately. All chemicals used for analyses were analytical grade.

2.2. Experimental design

Firstly, BSG and water were mixed with a ratio of 1:10 and the mixture was separated into three group treatments: mixture without protease treatment as control (-C), treated with 0.5% protamex (-P) and treated with a combination of 0.5% protamex and 0.1% flavourzyme (-PF). The groups were treated at 50 °C for 3 h at pH 8.5, followed by heating at 90 °C in order to inactivate the enzymes. After that, the treated mixtures were cooled down to room temperature and centrifuged to separate the liquid fraction from the sediment. The sediment (BSGS) was dried using a conventional dryer at 75 °C to reach the moisture content <6%, sealed in an aluminum foil bag and stored at 10 °C for antioxidant analysis and polyphenolic identification. The liquid fraction, protein, (BSGP) was dried by a semi-pilot spray dryer (APV Anhydro A/S LAB S1 spray dryer, Denmark). The fraction was evaporated in hot air with an inlet temperature of 160–165 °C and outlet temperature of 82–85 °C. The instrument was operated with an air pressure nozzle at 2 bars and the velocity of the peristaltic pump at 2.5 L/h. The dried extract was packed into an aluminum foil bag, sealed and kept at a chilled temperature (10 °C) for further studies.

2.3. Antioxidant properties and polyphenolic identification of protein extracts and sediments

2.3.1. Methanol extraction

The extract was prepared following methods described in previous studies (Tkacz et al., 2021). The extracts were provided using methanol 80% in distilled water used for antioxidant analysis and methanol 30% (methanol/water/acetic acid/ascorbic acid: 30/68/1/1, *v/v/v/m*) for polyphenolic measurement. Antioxidant assessment and polyphenolic measurement were performed in methanol solution. Therefore, the extracts were prepared in a methanol solution. The solution was added into 1 g of sample, and it was mixed using a vortex for 1 min. The mixture was sonicated (Sonic 6D, Polsonic, Warsaw, Poland) for 20 min and left at 4 °C. After 24 h, it was again sonicated for 20 min followed by centrifugation at 19000×*g* at 4 °C for 10 min. The extract was separated using a hydrophilic PTFE membrane (0.20 μm) (Millex Simplicity Filter, Merck, Germany).

2.3.2. Assessment of antioxidants

Antioxidant activities were evaluated with 2,2′-Azinobis-(3-Ethylbenzthiazoline- 6-Sulfonic Acid) (ABTS), ferric reducing antioxidant potential (FRAP), and oxygen radical absorbance capacity (ORAC) (Benzie and Strain, 1996; Crowley et al., 2015; Ou et al., 2013; Re et al., 1999). The result was expressed as mm trolox equivalents/100 g dry sample.

2.3.3. Identification of polyphenolic compounds

Flavan-3-ols and phenolic acids were identified and quantified following procedures as described previously (Tkacz et al., 2021). The measurement was performed by ultra-performance liquid chromatography (Acquity UPLC system) with a binary solvent manager and a photodiode array detector PDA (Waters Corp., Milford, MA. US), coupled to a XevoTM G2 Q/TOF micro-mass spectrometer and fitted with an electrospray ionization ESI source (Waters Corp., Manchester, UK). A full-scan, data-dependent MS was used, scanning from *m/z* 100 to 1700. Phenolic composition was characterized according to the retention time and accurate molecular masses. Flavan-3-ols and phenolic acids were monitored at 280 nm and 320 wavelengths, respectively. The data were collected using the MassLynxTM 4.1 ChromaLynx Application Manager (Waters Corp. Milford, USA) software. Quantification was conducted based on the phenolic calibration standards at concentrations ranging from 0.05 to 5 mg/mL (R2 $\geq$ 0.9995). All the samples were analyzed in triplicate and the data were presented in g/kg of dry weight sample.

2.4. Techno-functional properties of protein extracts

2.4.1. Water solubility index (WSI)

WSI was assessed following the procedure as described previously (Jafari et al., 2017; Rashid et al., 2015) with a slight modification. A total of 2.5 g of the sample was suspended in 25 mL of distilled water at room temperature. The suspension was vortexed for 1 min and left for 30 min at room temperature (25 °C). The mixture was centrifuged at 4100×*g* for 15 min at 4 °C. The dissolved powder in the liquid fraction was dried at 105 °C to reach a constant weight. The dried supernatant was determined as the value of WSI.

2.4.2. Oil holding capacity (OHC)

OHC was determined by mixing 2.5 g of the sample with 6 mL of vegetable oil. The mixture was left at room temperature for 30 min followed by centrifugation at 3000×*g* for 15 min. The remaining oil was measured after removing the supernatant and described as OHC (Kteniouda et al., 2013).

2.4.3. Emulsifying activity (EAI) and stability index (ESI)

EAI and Esi were determined as described previously (Fathollahy et al., 2021) with a slight modification. The sample was suspended in distilled water (1 g/100 mL), mixed with 10 mL of vegetable oil and homogenized with a homogenizer at 8900×*g* for 1 min. An amount of 5 mL of 0.1% sodium dodecyl sulfate solution was added into 50 μL of the homogenized sample at 0 and 10 min after homogenization. The

absorbance was directly measured at a 500 nm wavelength. EAI and ESI were determined as Equations (1) and (2):

$$EAI = \frac{2\ x\ 2.303\ x\ A0}{0.25\ x\ sample\ (g)} \quad (1)$$

$$ESI = A0\ x\ [\frac{\triangle t}{A0 - A10}] \quad (2)$$

A0: absorbance at 0 min, A10: absorbance at 10 min, $\triangle t$: 10 min.

2.4.4. Foaming capacity (FC) and stability (FS)

An amount of 5 g/L sample in distilled water was prepared and homogenized for 10 min at 20000 rpm using a homogenizer. The change in volume before foaming, during foaming and after 30 min left at room temperature were recorded (Fathollahy et al., 2021). FC and FS were determined as Equations (3) and (4):

$$FC = \frac{B - A}{A}\ x\ 100\% \quad (3)$$

$$FS = \frac{C - A}{A}\ x\ 100\% \quad (4)$$

A: volume before whipping, B: volume just after whipping, C: volume after 30 min whipping.

2.5. Statistical analysis

Antioxidant activities and polyphenolic composition were assessed statistically by two-ways ANOVA (Analysis of Variance) while the techno-functional properties were determined by one-way ANOVA using Statistica software (version 13.5.0.17). The significant difference was determined at $p < 0.05$ followed by Tukey post hoc test.

3. Results and discussion

3.1. Antioxidant activities of protein and sediments

Antioxidant activities of BSGPs and the sediments were evaluated on ORAC, ABTS and FRAP and the results are presented in Table 1. The results revealed that BSGPs had a significantly ($p < 0.05$) higher capability in ABTS and FRAP in all observed treatments. However, only protamex treatment generated different ORAC levels, while control and protamex–flavourzyme treatment had similar levels of ORAC in protein extract and sediment. ORAC value on BSGPs ranged from 5.9 to 7.0 mmol Trolox/100 g, while its sediment had a range of 5.2–5.9 mmol Trolox/100 g. Moreover, BSGPs had ABTS capability at a range of 4.15–5.55 mmol Trolox/100 g, while its sediment was at a range of 0.32–0.63 mmol Trolox/100 g. From the perspective of FRAP value, BSGPs and sediments ranged between 0.44 and 0.53 mmol Trolox/100 g and 0.15–0.34 mmol Trolox/100 g, respectively. Within those three group treatments, BSGPs had a higher level of ORAC and ABTS. In sediments, the highest value of ORAC and ABTS was obtained in the BSGS-C and BSGS-PF, respectively. Different trends in FRAP value were identified. In protein, BSGP-C had the highest FRAP value, while its sediment (BSGS-C) had a lower FRAP capability. However, BSGP-PF had the lowest FRAP value and generated the highest FRAP value in its sediment.

The ability of protamex and protamex/flavourzyme in enhancing the ORAC and ABTS might be related to the protein content of the extracts. As mentioned above, those treatments generated a higher protein content of BSGPs. Flavourzyme and protease had been identified for their ability in reducing the molecular weight of peptides, thus generating higher amounts of low-molecular-weight amino acids (Yang et al., 2020). Therefore, they generated a higher ORAC and ABTS capability, as the current study showed. Previous study reported that untreated BSG contained ABTS and FRAP capability at a range of 0.09–0.24 mmol Trolox/100 g and 0.11–0.31 mmol Trolox/100 g, respectively (Naibaho et al., 2021). This capability is much lower than that in BSGPs and the sediments, as obtained in the current study. Additionally, it was observed that untreated dried BSG had only around 1.75–2.62 mmol Trolox/100 g ORAC value (Naibaho et al., 2022). Meanwhile, the current study revealed a higher capacity that was almost 3 times higher. The treatments on BSG, regardless of the enzyme's treatment, enhanced the antioxidant capabilities of BSGPs as well as in several BSGSs. The result revealed that protein fractions had the highest antioxidant activities compared to the sediments. This phenomenon describes the protein responsible for the antioxidant activity of BSG. The ability of protease in improving antioxidant activity of BSGPs might be aligned to its ability in generating a lower molecular weight of peptides (Abeynayake et al., 2022). Protease and flavourzyme increased the amount of amino acids in BSGPs with a lower molecular weight (Kriisa et al., 2022). FRAP presented the ability of the extracts in inhibiting lipid oxidation (Rahman et al., 2021) by reducing the metal ion. Moreover, antioxidant activity is also influenced by the ability of the extracts in donating its proton to neutralize reactive species (Abeynayake et al., 2022). ABTS demonstrated the ability of electron donors in reducing the molecular oxygen and hydrogen peroxide (Benzie and Strain, 1996). The protein generated by protease treatments in the current study revealed a higher performance in neutralizing free radicals and reducing metal ion. This performance might be aligned with the release of hydrophobic amino acids which exert proton donation capacity (Abeynayake et al., 2022). The ability of BSG protein as an antioxidant has also been reported in previous studies (Vieira et al., 2017). The antioxidant activity in the sediments might be due to the presence of dietary fiber, fatty acids and polyphenolic compounds (Naibaho and Korzeniowska, 2021b). BSG has been reported for its matrix complexity, which consequently led to an entrapment of certain bioactive compounds in the matrices (Naibaho and Korzeniowska, 2021b). The treatments in the current study might have allowed the release of bioactive compounds from the matrices, such as protein, as well as phenolic compounds. The obtained results in the current study show that both protein extracts and the sediments of the treated BSG are valuable and beneficial for food and nutraceutical

Table 1
Antioxidant properties and polyphenolic composition of the protein extracts (BSGPs) and the sediments (BSGSs) of BSG.

Treatments	ORAC (mmol Trolox/100 g)		ABTS (mmol Trolox/100 g)		FRAP (mmol Trolox/100 g)		Phenolic acids (mg/kg)		Flavan-3-ols (mg/kg)		Total polyphenolic (mg/kg)	
	BSGPs	BSGSs	BSGPs	BSGSs	BSGPs	BSGSs	BSGPs	BSGSs	BSGPs	BSGSs	BSGPs	BSGSs
Control	5.90 ± 0.00^{ab}	5.94 ± 0.00^{ab}	4.15 ± 0.03^{c}	0.59 ± 0.04^{d}	0.53 ± 0.00^{a}	0.17 ± 0.01^{e}	122.89 ± 0.00^{a}	17.05 ± 0.00^{f}	130.66 ± 0.00^{c}	73.85 ± 0.00^{f}	253.55	90.90
0.5% protamex	7.00 ± 0.48^{a}	5.52 ± 0.06^{b}	5.27 ± 0.04^{b}	0.32 ± 0.03^{e}	0.46 ± 0.02^{b}	0.34 ± 0.01^{c}	65.37 ± 0.00^{c}	26.26 ± 0.00^{d}	103.67 ± 0.00^{e}	315.22 ± 0.00^{a}	169.04	341.48
Prot + Flav	6.17 ± 0.03^{ab}	5.70 ± 0.56^{b}	5.55 ± 0.02^{a}	0.63 ± 0.01^{d}	0.44 ± 0.00^{b}	0.28 ± 0.01^{d}	70.82 ± 0.00^{b}	18.84 ± 0.00^{e}	118.42 ± 0.00^{d}	173.74 ± 0.00^{b}	189.23	192.59

Note: The data is shown as mean ± standard deviation of three replication. A different subscription letter shows a significant difference ($P < 0.05$) in the same observed parameter.

ingredients. Protein extracts can be applied into food products depending on their techno-functional properties, which will be discussed in further sections. Currently, the potential of plant-based protein as a food ingredient has been increasingly investigated due to the rise in demand in consumers' preferences (Beacom et al., 2021). The potential benefits of the biological properties of BSG protein can be incorporated into dairy products, meat analogue and other food productions as a substitution ingredient and or main ingredient. Taking into account that BSG protein might also influence the final products acceptability. According to a previous study, the incorporation of protease and flavourzyme declined the bitter taste compared to that in BSG protein prepared without enzyme incorporation (Kriisa et al., 2022). Further investigation of BSGP-added food products is seemingly important in terms of processing technology and final product acceptability. The valorization of BSG sediments into food products can be applied as a source of dietary fiber, mineral and phenolic compounds for bread, cookies, yogurts or the extrusion industry, as described in previous studies (Naibaho and Korzeniowska, 2021b).

3.2. Polyphenolic identification on protein extracts and sediments

Phenolic acids and flavan-3-ols of BSGPs and BSGSs were analyzed, and the results are presented in Table 1. The results showed that BSGPs contained a higher number of phenolic acids compared to that in sediments. The higher the phenolic acids in BSGP, the lower their number in BSGS. This phenomenon seems negatively related to the ORAC value, as described previously. BSGP-C had the highest number of phenolic acids compared to that in BSGP-P. However, its sediment obtained the lowest number of phenolic acids. BSGS-P contained the highest phenolic acid content compared to that in other sediment treatments. However, its protein extract (BSGP-P) had the lowest phenolic acids. The number of phenolic acids is seemingly related to the FRAP value both for BSGPs and BSGSs. The sample which had the highest level of phenolic acid had the highest FRAP capability. Interestingly, BSGP-P contained a lower amount of flavan-3-ols compared to that in its sediments, while BSGP-C had a higher amount of flavan-3-ols compared to its sediment. This phenomenon slightly aligned with the FRAP value as it was observed also in phenolic acid analysis. By this, polyphenolic compounds on BSGPs and BSGSs are responsible for FRAP capability. However, the ORAC and ABTS capability is aligned with its protein content, as described earlier.

BSGP-C contained a higher amount of total polyphenolic content compared to BSGS-C. In contrast, BSGP-P and BSGP-PF had a lower polyphenolic content compared to its sediments. By this, the utilization of enzymes optimized the protein extraction in BSG with a lower total polyphenolic content, particularly the 0.5% protamex treatment. Polyphenolics were entrapped in the sediment, which means that the sediment can be used as a polyphenolic source for food or nutraceutical purposes. By this, the treatment in the current study not only generated BSGPs that contained higher antioxidant capabilities, but it also produced a sediment with a higher phenolic content. Tentative identification of polyphenolic compounds was carried out in order to evaluate the presence of specific compounds both in BSGPs and BSGSs, and the result is shown in Table 2. The result showed that several phenolic acids were identified, including syringic acid, benzoic acid, coumaric acid, ferulic–ferulic acid dimer, sinapic acid, ferulic acid, diferulic acid and its isomers. Moreover, (+)-catechin and (−)-epicatechin were observed as a group of flavan-3-ols. The results explained that several compounds were absent in BSGPs as well as in BSGSs due to the treatments. Syringic acid, sinapic acid, diferulic acid and (+)-catechin were not present in BSGSs, while ferulic–ferulic acid dimer was absent in BSGPs. It has been reported that certain phenolic compounds are bound in the matrices, thus impacting their release from the BSG matrices. Sinapic acid is present in BSG in bound form, and it can be released at higher temperatures (Rahman et al., 2021). By this, the absence of mentioned compounds might be because those compounds are entrapped in BSG matrices.

Table 2
Tentative identification of polyphenolic compounds in BSGPs and BSGSs.

Tentative polyphenolic identification		BSGPs	BSGSs
phenolic acid	syringic acid	✓	–
	benzoic acid	✓	✓
	coumaric acid	✓	✓
	ferulic-ferulic acid dimer	–	✓
	sinapic acid	✓	–
	ferulic acid	✓	✓
	diferulic acid	✓	–
	diferulic acid isomers	✓	✓
flavan-3-ols	(+)-catechin	✓	–
	(−)-epicatechin	✓	✓

✓: present; -: absence.

Phenolic compounds in BSG are dominated by sinapic acid, p-coumaric acid, ferulic acid and caffeic acids as well as their derivatives (Sibhatu et al., 2021). According to Table 2, sinapic acid was not identified in the sediment while coumaric acid and ferulic acids were observed in BSGSs and BSGPs. Meanwhile, caffeic acid was not identified in either BSGPs or BSGSs. This phenomenon demonstrated the possibility of a conversion of caffeic acid into ferulic acid. It was reported that caffeic acid is responsible for the antioxidant activity of phenolic extract from BSG (McCarthy et al., 2013). By this, the antioxidant activity of the protein extracts and sediments in the current study might be ruled by ferulic acid. Previous studies have confirmed that the presence of phenolic nucleus and an unsaturated side chain in ferulic acid structure generated a resonance that stabilized the phenoxy radical, thus improving the DPPH capability of ferulic acid from BSG (Connolly et al., 2021; Sibhatu et al., 2021). Although several compounds were absent in the sediments, the total number of phenolic compounds in enzymes treated sediments was observed to be the highest compared to that in the extracts. By this, the high amount of total phenolic content in the sediments might be due to the higher content of benzoic acid, coumaric acid, ferulic acid and its isomer, as well as (−)-epicatechin.

3.3. OHC

The capability of BSGPs in binding and holding oil was measured, and the result is presented in Table 3. The OHC value in the current study ranged from 2.07 to 2.23 g/g sample. The result revealed that BSGP-P significantly ($p < 0.05$) had the highest OHC compared to other BSGPs. Meanwhile, BSGP-PF had the same OHC level as was in BSGP-C. BSGP-P contained more lipophilic fraction compared to that in other treatments. In other words, protamex increased the hydrophobicity of the BSGP. It was observed that the lipid and protein were linked by the aliphatic chain of lipids to the non-polar chain of amino acids, showing that a higher OHC described a higher hydrophobicity (Withana-Gamage et al., 2011). Therefore, the lower level of OHC control showed a higher hydrophilic fraction, which might lead to a higher water solubility. A higher OHC demonstrated a better performance in food stabilizing effect, mouthfeel and reducing loss during the food processing (Benitez et al., 2019). A higher OHC level on BSGP-P might show better performance from a food processing point of view.

The impact of protamex in enhancing protein functionality has also been identified which were extracted from persian lime. An improvement in OHC was reported due to the treatment by protamex (Fathollahy et al., 2021). The difference in OHC value might be an impact of different sources of material and extraction process. Higher OHC level might have a lower water solubility. BSGPs in the current study revealed a better performance in terms of water solubility due to it having a lower OHC, which might increase the solubilization rate during the mixing process.

Table 3
Functionality of proteins extracted from BSG.

BSGPs	Protein content (%)	Dry matter (%)	OHC (g/g)	WSI (%)	EAI (m^2/g)	ESI (min)	FC (%)	FS (%)
BSGP-C	12.6 ± 0.3	2.7 ± 0.1	2.074 ± 0.02^{b}	77.782 ± 0.28^{a}	233.065 ± 7.07^{a}	141.337 ± 7.22^{a}	27.084 ± 2.95^{b}	4.167 ± 0.00^{b}
BSGP-P	37.5 ± 0.0	3.7 ± 0.0	2.225 ± 0.03^{a}	80.967 ± 1.81^{a}	276.458 ± 0.39^{a}	74.543 ± 5.31^{b}	83.333 ± 0.00^{a}	18.750 ± 2.95^{ab}
BSGP-PF	31.4 ± 0.7	3.9 ± 0.1	2.074 ± 0.01^{b}	82.090 ± 0.96^{a}	268.675 ± 18.31^{a}	49.944 ± 3.16^{c}	81.250 ± 8.84^{a}	39.583 ± 8.84^{a}

Note: The data is shown as mean ± standard deviation of three replication. A different subscription letter shows a significant difference ($P < 0.05$) in the same observed parameter.

3.4. WSI

Although it had different levels in OHC value, BSGPs possessed the same level of WSI in which there was no substantial significant difference ($p > 0.05$). WSI was assessed to represent molecular degradation ingredient behavior in aqueous phase which describes the reconstitution of the powder (Jafari et al., 2017; Rashid et al., 2015). Difference in WSI is observed to occur due to the difference in moisture content and protein solubility (Tas et al., 2022). Since there was no difference in WSI, further investigation in protein solubility is not needed in this study.

3.5. Emulsion properties

Statistically, there was no significant difference ($p > 0.05$) in EAI in all observed groups and it ranged from 233.1 to 276.5 m^2/g. Meanwhile, a significant difference ($p < 0.05$) in ESI was identified. The difference in EAI was reported due to the elevation in hydrolysis degree (Fathollahy et al., 2021). By this, the obtained BSGPs in the current study might have the same level of hydrolysis degree. The difference in ESI is influenced by the molecular weight of amino acids (Fathollahy et al., 2021). Both BSGP-P and BSGP-PF might have had a lower molecular weight compared to the control. As reported previously that enzymatic treatment in protein extraction generated amino acids with lower molecular weight (Wen et al., 2019). Smaller molecular weight peptides had a lower efficiency in declining the interface tension. Thus, their unfolding and reorientation at the interface to stabilize the emulsion are restricted compared to the longer peptides (Fathollahy et al., 2021).

3.6. Foaming properties

The foaming properties of BSGPs are presented in Table 3. Protease-treated BSGPs had a significantly higher ($p < 0.05$) FC and FS level compared to that in control. By this, enzymatic treatments improved the foaming properties of BSGPs. The same phenomenon had been reported previously in BSG protein and lime seed protein (Connolly et al., 2014; Fathollahy et al., 2021). As foaming properties were aligned with the pH level, barley protein showed higher foaming properties at alkaline pH and a lower value at acidic pH value (Connolly et al., 2014). Foaming properties on BSGPs were not influenced by DH (Connolly et al., 2014). The foaming formation might be due to the higher solubility (WSI), as mentioned previously. This result is aligned with a previous study that reported that the foaming capacity is influenced by the protein solubility, hydrophobicity and tenderness (Li et al., 2021). The higher FS in protease-treated protein might be due to the higher amount of protein content. The current study revealed that enzymatic treatments enhanced the protein content, thus improving the foaming stability. Li et al. (2021) emphasized that the FS level is influenced by the protein concentration, hydration and molecular interaction. The increase in foaming properties is also impacted by protein flexibility, which allows the protein particle to encapsulate air particles and spread quickly on the air–water interface (Li et al., 2021). The interaction between proteins at the air–water interface is facilitated by the increase in net charge, which contributes to the higher foaming properties (Fathollahy et al., 2021). Foaming properties is an important parameter in the utilization of BSGPs as a food ingredient. Higher foaming properties show a better performance in food products. By this, the BSG-PHs obtained by protease and protease/flavourzyme potentially offer benefits for food producers.

4. Conclusion

The study revealed that enzymatic treatments on BSG improved the ORAC and ABTS value of BSG-PHs, which might be due to its impact in generating higher protein content. Protein fractions had higher antioxidant properties than that in their sediments. It was observed that polyphenolic compounds are responsible for the FRAP value of BSG and both protein fractions and sediments; meanwhile, protein content is responsible for ORAC and ABTS values. The results show that enzymatic treatments on BSG improved the antioxidant properties of its BSGPs and enhanced the phenolic compounds in its sediments. The techno-functional characterization of BSGPs demonstrated that enzymatic treatments generated a higher oil holding capacity, foaming formation capability and foaming stability, although they had a lower emulsion activity index. By this, protease-treated BSGPs might will a better performance in food processing. Further investigation on its impact on the mechanical processing of food products is seemingly important.

Author contributions

Joncer Naibaho: conceptualization, methodology, validation, formal analysis, investigation, writing-original draft preparation, writing-review and editing, funding acquisition.

Małgorzata Korzeniowska: conceptualization, methodology, validation, investigation, resources, writing - review and editing, supervision, project administration, and funding acquisition.

Aneta wojdyło: methodology, formal analysis, writing - review and editing.

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Baoru Yang: writing-review and editing, project administration, and funding acquisition.

Declaration of competing interest

None.

Acknowledgements

Authors would like to thank Prof. Adam Figiel (Institute of Agricultural Engineering, Wrocław University of Environmental and Life Sciences, Wroclaw, Poland) for the extracts drying and Prof. Oskar Laaksonen (University of Turku) for project funding.

This work was supported by UPWR 2.0, international and interdisciplinary programme of development of Wrocław University of Environmental and Life Sciences, co-financed by the European Social Fund under the Operational Programme Knowledge Education Development 2014–2020: Axis III Higher education for the economy and development; Action 3.5. Comprehensive programmes for schools of higher education (POWR.03.05.00–00-Z062/18).

It was also supported by ERA-NET CO-FUND Horizon 2020 - FACCE SURPLUS Sustainable and Resilient Agriculture for Food and Non-Food

Systems and PROWASTE Protein-fibre biorefinery for scattered material streams (2019–2021).

References

Abeynayake, R., Zhang, S., Yang, W., Chen, L., 2022. Development of antioxidant peptides from brewers' spent grain proteins. LWT - Food Sci. Technol. (Lebensmittel-Wissenschaft -Technol.) 158, 113162. https://doi.org/10.1016/j.lwt.2022.113162.

Beacom, E., Bogue, J., Repar, L., 2021. Market-oriented development of plant-based food and beverage products: a usage segmentation approach. J. Food Prod. Market. 27, 204–222. https://doi.org/10.1080/10454446.2021.1955799.

Benitez, V., Rebollo-Hernanz, M., Hernanz, S., Chantres, S., Aguilera, Y., Martin-Cabrejas, M.A., 2019. Coffee parchment as a new dietary fiber ingredient: functional and physiological characterization. Food Res. Int. 122, 105–113. https://doi.org/10.1016/j.foodres.2019.04.002.

Benzie, I.F.F., Strain, J.J., 1996. The ferric reducing ability of plasma (FRAP) as a measure of "antioxidant power": the FRAP assay. Anal. Biochem. 239, 70–76. https://doi.org/10.1006/abio.1996.0292.

Cermeño, M., Connolly, A., O'Keeffe, M.B., Flynn, C., Alashi, A.M., Aluko, R.E., FitzGerald, R.J., 2019. Identification of bioactive peptides from brewers' spent grain and contribution of Leu/Ile to bioactive potency. J. Funct.Foods 60, 103455. https://doi.org/10.1016/j.jff.2019.103455.

Cian, R.E., Garzón, A.G., Martínez-Augustin, O., Botto, C.C., Drago, S.R., 2018. Antithrombotic activity of brewers' spent grain peptides and their effects on blood coagulation pathways. Plant Foods Hum. Nutr. 73, 241–246. https://doi.org/10.1007/s11130-018-0682-1.

Connolly, A., Cermeño, M., Alashi, A.M., Aluko, R.E., FitzGerald, R.J., 2021. Generation of phenolic-rich extracts from brewers' spent grain and characterisation of their in vitro and in vivo activities. Innovat. Food Sci. Emerg. Technol. 68, 102617 https://doi.org/10.1016/j.ifset.2021.102617.

Connolly, A., O'Keeffe, M.B., Piggott, C.O., Nongonierma, A.B., FitzGerald, R.J., 2015. Generation and identification of angiotensin converting enzyme (ACE) inhibitory peptides from a brewers' spent grain protein isolate. Food Chem. 176, 64–71. https://doi.org/10.1016/j.foodchem.2014.12.027.

Connolly, A., Piggott, C.O., FitzGerald, R.J., 2014. Technofunctional properties of a brewers' spent grain protein-enriched isolate and its associated enzymatic hydrolysates. LWT - Food Sci. Technol. (Lebensmittel-Wissenschaft -Technol.) 59, 1061–1067. https://doi.org/10.1016/j.lwt.2014.06.054.

Connolly, A., Piggott, C.O., FitzGerald, R.J., 2013. Characterisation of protein-rich isolates and antioxidative phenolic extracts from pale and black brewers' spent grain. Int. J. Food Sci. Technol. 48, 1670–1681. https://doi.org/10.1111/ijfs.12137.

Crowley, D., O'Callaghan, Y., McCarthy, A., Connolly, A., Piggott, C.O., FitzGerald, R.J., O'Brien, N.M., 2015. Immunomodulatory potential of a brewers' spent grain protein hydrolysate incorporated into low-fat milk following *in vitro* gastrointestinal digestion. Int. J. Food Sci. Nutr. 66, 672–676. https://doi.org/10.3109/09637486.2015.1077788.

Fathollahy, I., Farmani, J., Kasaai, M.R., Hamishehkar, H., 2021. Characteristics and functional properties of Persian lime (Citrus latifolia) seed protein isolate and enzymatic hydrolysates. LWT - Food Sci. Technol. (Lebensmittel-Wissenschaft -Technol.) 140, 110765. https://doi.org/10.1016/j.lwt.2020.110765.

Jafari, S.M., Ghalegi Ghalenoei, M., Dehnad, D., 2017. Influence of spray drying on water solubility index, apparent density, and anthocyanin content of pomegranate juice powder. Powder Technol. 311, 59–65. https://doi.org/10.1016/j.powtec.2017.01.070.

Kriisa, M., Taivosalo, A., Föste, M., Kütt, M.-L., Viirma, M., Priidik, R., Korzeniowska, M., Tian, Y., Laaksonen, O., Yang, B., Vilu, R., 2022. Effect of enzyme-assisted hydrolysis on brewer's spent grain protein solubilization – peptide composition and sensory properties. Appl. Food Res. 2, 100108 https://doi.org/10.1016/j.afres.2022.100108.

Ktenioudaki, A., O'Shea, N., Gallagher, E., 2013. Rheological properties of wheat dough supplemented with functional by-products of food processing: brewer's spent grain and apple pomace. J. Food Eng. 116, 362–368. https://doi.org/10.1016/j.jfoodeng.2012.12.005.

Li, W., Yang, H., Coldea, T.E., Zhao, H., 2021. Modification of structural and functional characteristics of brewer's spent grain protein by ultrasound assisted extraction. LWT - Food Sci. Technol. (Lebensmittel-Wissenschaft -Technol.) 139, 110582. https://doi.org/10.1016/j.lwt.2020.110582.

McCarthy, A.L., O'Callaghan, Y.C., Neugart, S., Piggott, C.O., Connolly, A., Jansen, M.A. K., Krumbein, A., Schreiner, M., FitzGerald, R.J., O'Brien, N.M., 2013. The hydroxycinnamic acid content of barley and brewers' spent grain (BSG) and the potential to incorporate phenolic extracts of BSG as antioxidants into fruit beverages. Food Chem. 141, 2567–2574. https://doi.org/10.1016/j.foodchem.2013.05.048.

Naibaho, J., Korzeniowska, M., 2021a. The variability of physico-chemical properties of brewery spent grain from 8 different breweries. Heliyon 7, e06583. https://doi.org/10.1016/j.heliyon.2021.e06583.

Naibaho, J., Korzeniowska, M., 2021b. Brewers' spent grain in food systems: processing and final products quality as a function of fiber modification treatment. J. Food Sci. 86, 1532–1551. https://doi.org/10.1111/1750-3841.15714.

Naibaho, J., Korzeniowska, M., Wojdyło, A., Figiel, A., Yang, B., Laaksonen, O., Föste, M., Vilu, R., Viiard, E., 2021. The potential of spent barley as a functional food ingredient: study on the comparison of dietary fiber and bioactivity. Proceedings 70, 6. https://doi.org/10.3390/foods_2020-08486.

Naibaho, J., Wojdyło, A., Korzeniowska, M., Laaksonen, O., Föste, M., Kütt, M.-L., Yang, B., 2022. Antioxidant activities and polyphenolic identification by UPLC-MS/MS of autoclaved brewers' spent grain. LWT - Food Sci. Technol. (Lebensmittel-Wissenschaft -Technol.) 163, 113612. https://doi.org/10.1016/j.lwt.2022.113612.

Ou, B., Chang, T., Huang, D., Prior, R.L., 2013. Determination of total antioxidant capacity by oxygen radical absorbance capacity (ORAC) using fluorescein as the fluorescence probe: first action 2012.23. J. AOAC Int. 96, 1372–1376. https://doi.org/10.5740/jaoacint.13-175.

Rahman, Md.J., Malunga, L.N., Eskin, M., Eck, P., Thandapilly, S.J., Thiyam-Hollander, U., 2021. Valorization of heat-treated brewers' spent grain through the identification of bioactive phenolics by UPLC-PDA and evaluation of their antioxidant activities. Front. Nutr. 8, 634519 https://doi.org/10.3389/fnut.2021.634519.

Rashid, S., Rakha, A., Anjum, F.M., Ahmed, W., Sohail, M., 2015. Effects of extrusion cooking on the dietary fibre content and Water Solubility Index of wheat bran extrudates. Int. J. Food Sci. Technol. 50, 1533–1537. https://doi.org/10.1111/ijfs.12798.

Re, R., Pellegrini, N., Proteggente, A., Pannala, A., Yang, M., Rice-Evans, C., 1999. Antioxidant activity applying an improved ABTS radical cation decolorization assay. Free Radic. Biol. Med. 26, 1231–1237. https://doi.org/10.1016/S0891-5849(98)00315-3.

San Martin, D., Orive, M., Iñarra, B., Castelo, J., Estévez, A., Nazzaro, J., Iloro, I., Elortza, F., Zufía, J., 2020. Brewers' spent yeast and grain protein hydrolysates as second-generation feedstuff for aquaculture feed. Waste Biomass Valorization 11, 5307–5320. https://doi.org/10.1007/s12649-020-01145-8.

Sibhatu, H.K., Anuradha Jabasingh, S., Yimam, A., Ahmed, S., 2021. Ferulic acid production from brewery spent grains, an agro-industrial waste. LWT - Food Sci. Technol. (Lebensmittel-Wissenschaft -Technol.) 135, 110009. https://doi.org/10.1016/j.lwt.2020.110009.

Tas, O., Ertugrul, U., Grunin, L., Oztop, M.H., 2022. An investigation of functional quality characteristics and water interactions of navy bean, chickpea, pea, and lentil flours. Legume Sci. 4 https://doi.org/10.1002/leg3.136.

Tkacz, K., Wojdyło, A., Turkiewicz, I.P., Nowicka, P., 2021. Triterpenoids, phenolic compounds, macro- and microelements in anatomical parts of sea buckthorn (Hippophaë rhamnoides L.) berries, branches and leaves. J. Food Compos. Anal., 104107 https://doi.org/10.1016/j.jfca.2021.104107.

Vieira, E., da Silva, D.D., Carmo, H., Ferreira, I.M.P.L.V.O., 2017. Protective ability against oxidative stress of brewers' spent grain protein hydrolysates. Food Chem. 228, 602–609. https://doi.org/10.1016/j.foodchem.2017.02.050.

Vieira, E., Teixeira, J., Ferreira, I.M.P.L.V.O., 2016. Valorization of brewers' spent grain and spent yeast through protein hydrolysates with antioxidant properties. Eur. Food Res. Technol. 242, 1975–1984. https://doi.org/10.1007/s00217-016-2696-y.

Wen, C., Zhang, J., Duan, Y., Zhang, H., Ma, H., 2019. A mini-review on brewer's spent grain protein: isolation, physicochemical properties, application of protein, and functional properties of hydrolysates. J. Food Sci. 84, 3330–3340. https://doi.org/10.1111/1750-3841.14906.

Withana-Gamage, T.S., Wanasundara, J.P., Pietrasik, Z., Shand, P.J., 2011. Physicochemical, thermal and functional characterisation of protein isolates from Kabuli and Desi chickpea (*Cicer arietinum* L.): a comparative study with soy (*Glycine max*) and pea (*Pisum sativum* L.): characterisation of chickpea protein isolates. J. Sci. Food Agric. 91, 1022–1031. https://doi.org/10.1002/jsfa.4277.

Yang, J., Jiang, C., Bao, R., Liu, M., Lv, J., Yang, Z., Xu, W., Liang, H., Ji, C., Li, S., Zhang, S., Lin, X., 2020. Effects of flavourzyme addition on physicochemical properties, volatile compound components and microbial community succession of Suanzhayu. Int. J. Food Microbiol. 334, 108839 https://doi.org/10.1016/j.ijfoodmicro.2020.108839.

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OŚWIADCZENIE

Oświadczam, że w pracy

Naibaho, J., Korzeniowska, M., Wojdyło, A., Ayunda, HM., Foste, M., & Yang, B. 2022. Techno-functional properties of protein from protease-treated brewers' spent grain (BSG) and investigation of antioxidant activity of extracted proteins and BSG residues. Journal of Cereal Science 107, 103524. https://doi.org/10.1016/j.jcs.2022.103524 (autorzy, rok wydania, tytuł, czasopismo lub wydawca, tom, strony)

mój udział polegał na konceptualizacja, metodologia, walidacja, analiza formalna, dochodzenie, pisanie-oryginalne przygotowanie projektu, pisanie-recenzja i redakcja, pozyskiwanie funduszy (opisać szczegółowo swój własny udział w powstaniu pracy, np. wykonaniu doświadczeń techniką ....., analizie statystycznej wyników eksperymentów zilustrowanych na ryc. ...., przygotowaniu tekstu manuskryptu zamieszczonego w rozdziale ....., kierowaniu projektem naukowym obejmującym badania opisane w tej pracy, itp.).

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mój udział polegał na konceptualizacja, metodologia, walidacja, badanie, zasoby, pisanie - recenzja i redakcja, nadzór, administracja projektem i pozyskiwanie funduszy

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March 27, 2023

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(data i podpis)