

dhc Prof. Ángel A. Carbonell-Barrachina
Universidad Miguel Hernández de Elche
Departamento Tecnología Agroalimentaria

Elche, Alicante (Spain), 31.07.2023

PhD Thesis Evaluation Report

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

Author: **Joncer Naibaho**

Supervisors: **dr hab. Inż. Małgorzata Korzeniowska, prof. uczelni (promotor), and Prof. Baoru Yan, PhD (drugi promotor)**

Agricultural Sciences (field of science), Nutrition and Food Technology (scientific discipline's name) at the Wrocław University of Environmental and Life Sciences.

The selection and meaning of the topic

European countries produced an estimate of almost 360 million hectoliters of beer, producing over 7.2 million tons of brewers' spent grain (BSG), with Poland being the third producers only after Germany and UK. This material, BSG, contains significant contents of bioactive compounds, including phenolic compounds, fatty acids, amino acids, and minerals. Traditionally BSG has been used to produce enriched bread, cookies, and pasta. However, the negative effects associated with food processing and the acceptability of the final products has not been deeply studied. Thus, it is well established that the mechanisms of nutritional-related compounds release from the BSG matrix due to thermal treatments and/or extraction processes has not been addressed and deserve further research.

Formal aspects of the work

The Ph.D. thesis presented by Mgr. Inż. Joncer Naibaho entitled: "Modification of brewers' spent grain chemical composition to improve their techno-functionality and antioxidant capabilities" consists of 34 pages (plus attachments) divided to the following parts: Abstract (5 pages), Introduction (4 pages), Purpose and scope of research (1 page), Materials and Methods (2 pages), Results and Discussion (15 pages), Conclusion (2 pages), and Bibliography (8 pages), Attachment divided to the list of publications constituting the doctoral dissertation, Publications of the doctoral dissertation, Statements of the co-authors of the publications of the doctoral dissertation. Introductory part relates to the first publication, that is the complete review paper on brewers' spent grain in food systems published in a very traditional and well-known journal (IF=3.16), Journal of Food Science. The Results and Discussion section presents the main information generated during the PhD dissertation organized in 5 subsections corresponding to the 5 research manuscripts, published in highly ranked journals: (i) thermal treatments, preliminary study, publications 2 (LWT, IF 4.95) and 3 (LWT, IF 6.05), (ii) autoclave treatment, publication 4 (International Journal of Food Science and Technology, IF 3.61), (iii) water-bath treatment, publication 5 (Food Bioscience, IF 5.32), and (iv) enzymatic-protein extraction, publication 6 (Journal of Cereal Science, IF 4.07). All these parts of the R&D section are reflecting the

five manuscripts published in recognized scientific journals, papers in which Mgr. Inż. Naibaho is the first author. Conclusions together with general remark and the list of cited publications are closing the substantive part of the thesis. Additionally, the five manuscripts published in open-access are presented as attachments, with the corresponding statements signed by co-authors of selected publications.

The assessed dissertation meets the formal requirements for this type of work presented in the procedure for the doctoral degree, i.e., it is experimental and contains all the necessary chapters arranged in a typical sequence. The study is a coherent whole and is correctly written in clear and understandable language.

Content of the work

Abstract

The abstract is informative and provides a summary of all conducted experiments, their results and main conclusions.

Introduction

Prepared on the base of the first publication included in the doctoral dissertation (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) introductory section is concise but informative. It brings crucial information about the importance of beer production in Poland and the huge amount of BSG generated, and the interesting composition of this material, which includes interesting bioactive compounds, such as phenolic compounds (stage I of the PhD thesis). In addition, the main methods used to extract these bioactive compounds are introduced, including thermal treatments (stage II) and enzymatic extraction of the proteins (stage III).

Purpose and scope of research

Based on the literature data Mgr. Inż. Naibaho, the aim of the thesis was to modify the chemical composition, techno-functionality, and antioxidant activity of BSG by several techniques including physical treatments, such as autoclave heating and water-bath heating (thermal treatments) and enzymatic treatments. Later, the main hypothesis is presented "different methods could alter the chemical composition and biological properties differently due to the different stability of the specific compounds to different exposure conditions. The research was designed into three main stages (composition of the BSG, thermal treatments and enzymatic extraction of the bioactive compounds) to reach the final aim of the work and the specific objectives and hypotheses.

Materials and Methods

The materials, experimental design, and methodology are briefly introduced, giving the reader enough information to understand what is done in the different research experiments.

Results and discussion

The scientific content of the five research-articles included in the PhD dissertation are related to the effects of the thermal and/or extraction methods on the release of the bioactive compounds present in the BSG.

Publication 2 (Fiber modification of brewers' spent grain by autoclave treatment to improve its properties as a functional food ingredient, LWT, 2021, 149, 111877) aimed at modifying the dietary fiber composition of BSG by autoclave treatment (AT) to improve its functional property. Autoclave treatment reduced the dark color of BSG and decreases the water activity, thus extending the shelf-life by reducing the degradation due to microbial growth and chemical reaction. As a conclusion AT can be used to improve the safety of BSG during storage. Besides, AT also changed the dietary fiber type, by increasing the SDF (soluble dietary fiber) and decreasing the IDF (insoluble dietary fiber). In this way the AT declined the water holding capacity and oil holding capacity but increased the swelling capacity.

Questions: Do you think the use of the dietary fiber from BSG as food ingredient would change the flavor of the final product? How can you be sure about that?

Publication 3 (Antioxidant activities and polyphenolic identification by UPLC-MS/MS of autoclaved brewers' spent grain, LWT, 2022, 163, 113612), which aim was to study the effect of AT on antioxidant activity and phenolic composition of BSG. AT increased ORAC, ABTS and FRAP activities and induced the generation of key compounds, such as benzoic acid and (+)-catechin. This behavior is due to the weakening of the bounds among lignin and phenolic groups, which are released, increasing the antioxidant activity. As a conclusion AT improved the bioactivity of BSG, enhanced the amount of key phenolic compounds and released bioactive compounds from BSG matrix thus offering a high benefit for the food industry.

Questions: Do you think results would have been different if the BSG drying method was different than conventional drying at 75 °C?

Publication 4 (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), which aim was to investigate the influence of the thermal levels and water ratio on autoclave heating (AH) in altering the chemical composition, antioxidant properties, and functionality of undried fresh BSG. In this particular study, fresh/non-dried material was used to avoid the use of extra water and energy to re-dry the treated material. AH reduced ketones and furans regardless of the water ratio used, and improved the contents of flavan-3-ols, antioxidant activity, and water holding capacity of treated BSG. Besides, AH transformed saturated into unsaturated fatty acids and the higher the temperature the higher the antioxidant activity (FRAP and ABTS).

Questions: Have you done any economic study on how expensive this enzymatic treatment is? Always economy is essential for the industries and deserve further research. In some of the tables, e.g., Table

2, do you trust statistics hundred per cent? Or do you believe that some of the statistical differences have no practical meaning??

Publication 5 (Conventional water bath heating on undried brewers' spent grain: techno-functionality, fatty acids, volatiles, polyphenolic profile and antioxidant properties, Food Bioscience, 2023, 102523), which hypothesis was that heating treatments using conventional water bath heating (CWH) on BSG would modify the functionality, chemical composition and antioxidant activity. CWF decreased the content of saturated fatty acids and increased that of poly-unsaturated fatty acids, but decreased the total content of phenolic compounds, probably by migration to water. Besides, CWF increased volatile compounds leading to fruity, sweet and pleasant notes. Finally, CWF improved water-holding and oil-holding capacities. Thus, CWH a low-cost treatment improved the functionality, fatty acid nature and aromatic profile of BSG.

Questions: I guess there would be some migration of bioactive compounds to the water used in the treatment, do you think this is a practical approach or the losses due to the migration of bioactive molecules to the water could jeopardize its interest? How did you link the volatile compounds to the sensory descriptors such as fruity, sweet, etc.?

Publication 6 (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), which aim was to investigate the biological properties of the protein fraction of BSG and sediments as well as the techno-functional properties of BSG protein (BSGP). Enzymatic treatment increased the antioxidant activity of BSGP, with FRAP being related to phenolic compounds and BSGP with ORAC and ABTS capacities. BSGP had higher oil-holding capacity, foaming properties and lower emulsion capability. As a conclusion, enzymatic treatment of BSG enhanced protein functionality and bioactivity.

Questions: Have you done any economic study on how expensive this enzymatic treatment is? Which enzymatic method will you recommend? Will this recommendation depend on the final use of the protein?

Final conclusion

Summing up, I would like to state that, the assessed PhD thesis has a scientific potential with novel aspects on how to modify BSG to make it more appropriate for its use as a food ingredient to develop new products enriched with interesting bioactive compounds, such as phenolic compounds, amino acids, etc. All experiments were carefully planned and carried out with properly selected and available classic methodology and equipment. The collected results were presented clearly with appropriate tables and figures, analyzed, discussed and interpreted in a thorough manner. The results were published in renowned high ranked journals, which prove the novelty and high level of the performed research. The content of the doctoral dissertation of Mgr. Inż. Naibaho entitled: "Modification of brewers' spent grain chemical composition to improve their techno-functionality and antioxidant capabilities" and the resulting valuable practical application possibilities give me the right to conclude that it meets all the requirements specified in Art. 187 sec. 1-4 of the Act of July 20, 2018. Law on

Higher Education and Science (Journal of Laws 2023, item 742). Therefore, with full conviction, I submit an application to the Council of the discipline Nutrition and Food Technology at the Wrocław University of Environmental and Life Sciences for the admission of Mr. Joncer Naibaho to the public defense of this thesis.



dhc Prof. Ángel A. Carbonell-Barrachina