

## Barley proteins: relation to malt quality in a context of climate change and need of sustainability – The new challenge

Proteins in raw materials play an important part in both malting and brewing processes, as actors of raw material biotransformation, contributors to foam, haze or even nitrogen source for the yeast.

Nonetheless, a comparable total protein content of barley can lead to various malt qualities and different proteolysis levels. In a context of climate change, of inputs reduction and need of more sustainable raw materials, there is a need to improve knowledge of proteins in barley. The collaborative "PROsIT" project (barley PROteins of InTerest) proposed to draw up the qualitative profile of the different barley proteins and tried to identify the possible relation to malt quality. The project is supported by FSOV (Found to support breeding selection in France) and all barley to beer chain (CTPS, Breeders, ARVALIS, French Maltsters & Brewers)

The analysis of barley protein fractions was developed on a sequential isolation inspired by Schalk et al. (2017) [1] followed by capillary electrophoresis analysis (Labchip, PERTEN). In more details, protein fractions are first isolated according to Osborne [2] classification: albumin, globulin, hordein, glutelin. Each of them are further separated according to molecular weight to collect, respectively 18, 10, 10 and 15 peaks.

A collection of 321 barley samples of different crops, locations, varieties and nitrogen fertilizations produced by project partners was analysed. All the samples were micro-malted at IFBM micro-malting plant and malts were characterized (extract, friability, total

& soluble proteins, wort viscosity & beta glucans content, diastatic power, etc.). A database of 1284 profiles (more than 16 000 peaks) and the corresponding malt parameters was built.

The first data treatment indicates that there is a very large diversity of barley protein composition for the same total nitrogen content with a 2 up to 3-fold factor depending the protein fraction. In other words, several barley batches could exhibit the same protein content but contain some proteins of interest and some less useful for the maltsters or brewers.

Besides, a PCA analysis on nitrogen fertilization trials on 220 samples shows an important varietal effect on protein profile. However, the crop, location or fertilizing modalities have no significant effect on protein composition in our sampling.

This major result demonstrates that the barley variety is the most significant factor triggering different protein compositions. But the different clusters gather independently some 6 row winter lines can be with some 2 row spring lines. This means that the barley specie does not drive, alone, the protein profile.

The data treatment needs to be completed but this work shows the importance of barley protein composition to assess malt quality. And the results gathered in the project is the beginning of a larger study investigating the proteins of interest for the barley to beer chain.

1. Schalk, K., Lexhaller, B., Koehler, P., & Scherf, K. A. (2017). Isolation and characterization of gluten protein types from wheat, rye, barley and oats for use as reference materials. *PLoS ONE*, 12(2), 1–20. <https://doi.org/10.1371/journal.pone.0172819>.
2. Osborne, T.B., (1907). The proteins of the wheat kernel. Publication 84. Carnegie Institute of Washington, Washington DC.

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## Brewers' spent grain for organic thin-film transistor application

Brewer's spent grain (BSG) represents the main by-product of the brewing industry, accounting for ~ 85 % of the total by-products. The annual global production of BSG is massive, that is ~ 40 million tons. In the European Union, the production is ~ 3,4 million tons/year [1]. Currently, BSG is mainly sold to farmers (~ 30 % in EU) as animal

feed, with a low market value of ~ 35 Euro/ton or landfilled. Finding alternatives, higher value uses for BSG is therefore particularly attractive from the point of view of brewery economics [1].

Besides, disposable electronics applications such as in smart food and beverage packaging require environmentally safe devices with low cost and large volume processability. Therefore, switching from nonrenewable manufacturing to sustainable processes is a major challenge for next generation electronics, and organic materials offer a unique opportunity to drive the electronic industry in an environmentally safe direction [2].

Among various organic electronic devices, thin film transistors (OTFTs) are fundamentals. They are multilayered-structured

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devices that comprises conducting electrodes, dielectrics and semiconductors. We investigated for the first time the use of two no starch polysaccharides components of BSG,  $\beta$ -glucans and arabinoxylans, as green dielectric materials for OTFTs.

Since such components possess rich polar functional groups, they can be in principle effectively employed as dielectric layers for OTFTs due to their good polarization under electric field, which would lead to high capacitance and, ultimately, to promising possibilities and applications for green devices.

The thin films were prepared by spin coating technique and, indeed, encouraging capacitance values of  $\sim 1820 \text{ nFcm}^{-2}$  at 1kHz were obtained for both components.  $\beta$ -Glucans revealed to be particularly promising when implemented in OTFTs, enabling

devices giving good response, i.e. a charge mobility of  $1.5 \text{ cm}^2/\text{Vs}$  with average current on/off ratio of  $\sim 10^4$  and a threshold voltage of  $-1 \text{ V}$ . The Ph-BTBT-C10 benchmark semiconductor was employed as active layer.

Further developments along this line are currently in progress and will include the use of the devices as platform to realize smart food and beverage package demonstrators.

1. A. Trusek et al. "Brewer's Spent Grains – Valuable Beer Industry By-Product" *Biomolecules* 2020, 10, 1669.
2. C. Kim et al. "Sustainable approaches in the design of dielectric materials for organic thin-film transistors" in A. Marrocchi (Ed) "Sustainable Strategies for Organic electronics", Woodhead publishing – Elsevier, 2022.

## Are small starch granules ruining the benefits of high gravity brewing?

The mashing process has to maintain a complex balance between starch gelatinisation and thermal inactivation of starch-hydrolysing enzymes. This balance can be affected by the intrinsic starch granule properties, extrinsic factors such as the presence of sugars and other wort components and the process parameters such as mashing thickness and the temperature-time profile. Nowadays, breweries can use high gravity brewing, which improves brewhouse efficiency. However, large proportions of small starch granules in barley malt seem to endanger this benefit. While in the past, the impact of small starch granules on the mashing process was considered neglectable because their levels in barley malt were deemed low, recent findings showed small starch granule proportions of up to 27 V/V%. Therefore, their importance during mashing has to be reconsidered. In this work, the intrinsic starch gelatinisation behaviour of barley malt starch and the impact of mash thickness on the gelatinisation of small and large starch granules was assessed. Small starch granules isolated from barley malt

had a higher peak gelatinisation temperature ( $62.5 \text{ }^\circ\text{C}$ ) than large starch granules ( $59.7 \text{ }^\circ\text{C}$ ). In addition, mainly water-extractable, non-starch components from barley malt caused an elevation of the intrinsic starch gelatinisation temperatures by  $4.6 \text{ }^\circ\text{C}$ . During the mashing process, additional water-extractable components such as sugars are produced. We, therefore, hypothesised that the impact of these components would be of great importance, especially when performing high gravity brewing. Mashing was performed with malt to water ratios varying between 1 : 6 to 1.2.5. Thicker mashes resulted in less efficient sugar production (a sugar yield of 79 % for 1 : 6 mashing compared to 66 % for 1 : 2.5 mashing), mainly due to a decrease in maltose production. These results oppose previous findings in literature. We hypothesise that this is due to delayed gelatinisation of the small starch granules during mashing, caused by wort components such as sugars. The addition of 24 °P wort to isolated starch granules resulted in a  $10 \text{ }^\circ\text{C}$  increase in the gelatinisation temperature of starch indeed. In the case of small starch granules, this resulted in a peak gelatinisation temperature of  $72.5 \text{ }^\circ\text{C}$ . This is problematic considering that malt  $\beta$ -amylase, which produces maltose, will be thermally inactivated rapidly at this temperature. Considering these results, it is clear that the small starch granule proportion in barley and malt should be introduced as a selection criterium.

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## Development of a bacterial biocontrol process applicable in the barley-malt-beer industry

Pathogenic fungi represent a generic problem for cereals as they can produce a variety of toxic secondary metabolites such as mycotoxins that represent a significant concern for the malting and brewing industries, and may affect the quality and safety of barley, malt, and beer. Besides, this situation is worsening due to the highly variable climatic conditions that favor pathogenic fungi development and the societal desire to reduce the use of phytosanitary products, including fungicides. In this context, this communication describes the development of an innovative biocontrol process applicable in malting facilities, that would contribute to guaranteeing a better hygienic and technological quality of malt, despite the increasingly complex and variable conditions for barley production. The process is based on technological bacteria, isolated from infection-resistant barley cultures, that can reduce the development of spoilage fungi and the associated mycotoxin production. The experimental approach consists of: i) determining the growth kinetics of the bacterial and fungal strains by co-culturing in order to evaluate the impact of the bacteria on the fungal pathogens; ii) carrying out a micro-malting process in order to develop

the aforementioned process, and iii) evaluating the technological and sanitary properties of the generated barley malts in order to validate the process developed. The findings highlight the ability of a barley-associated novel bacterial strain, *Erwinia gerundensis*, to inhibit the growth of fungal species and to reduce their toxigenic potential. *E. gerundensis* exhibited a significant fungistatic activity against pathogenic fungi by reducing their growth by up to 80 %, and their mycotoxin production by 70 to 100 % in liquid medium and on barley matrix. In addition, micro-malting assays carried out using naturally contaminated barley kernels have revealed that the bacterial strain was capable of reducing the fungal load and mycotoxin (eniatin) content of malt by 70 % and 50 % respectively, without any degradation of its technological quality. Based on these results, our study supports the use of *Erwinia gerundensis* as a biocontrol agent in strategies aiming at reducing the presence of pathogenic fungi and mycotoxins in cereal-based products, or as a food and feed supplement for the bio-detoxification of mycotoxins. The biocontrol process based on this bacterial strain is therefore expected to make it possible to guarantee an irreproachable hygienic and technological quality of the malt obtained from barley, thus significantly reducing the setbacks related to pathogenic fungi and mycotoxins in the brewing industry. The use of this process would also contribute to the reduction of contamination levels of malting plant effluents. Finally, future works are required to effectively evaluate the impact of *Erwinia gerundensis* during brewing and on beer quality.

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