NATURAL DEEP EUTECTIC SOLVENTS (NADES) AS A SUSTAINABLE ALTERNATIVE TO OBTAIN POLYPHENOL-RICH EXTRACTS FROM RED COLOURED POTATOES

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Potato (*Solanum tuberosum L.)* is one of the most important foodstuffs, mainly consumed after processing, which generates tons of waste such as the potato peel. This waste could provide a useful and inexpensive source of high-value compounds for different industry uses [1], Specifically, red-coloured potatoes are rich in anthocyanins, which can be used as additives, colorants or preservatives, in food industry. Natural Deep Eutectic Solvents (NADES) mean an opportunity to carry out green and sustainable extraction of polyphenols, in addition to improve the bioactive properties of the extracts obtained.

The extractive capacities of different natural deep eutectic solvents (NADES), some of them formed by the combination of choline chloride (ChCl) and alcohols (propanediol and butanediol) or acids (lactic and malic), were tested. The most effective one, consisting of choline chloride and lactic acid was used for anthocyanin extraction in peels from four red potato varieties (Rudolph, Memphis, Manitou and Mozart) and the amount of pelargonidin (major compound) in the different varieties were compared. The antimicrobial activity of the extracts and NADES were also evaluated according to the methos described by Pires et al [2].

The results obtained show that recovery yields are higher in the green extraction (**Figure 1**), using choline chloride and lactic acid, than in methanolic extraction.

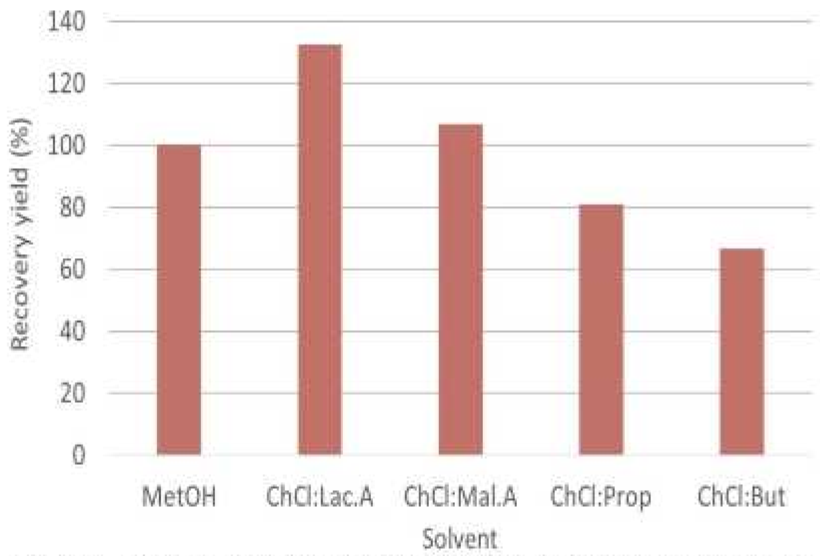

Figure 1. Comparison of Anthocyanin recovery yields between NADES and conventional extraction.

Moreover, the Rudolph variety was the richest in phenolic compounds (**Figure 2**).

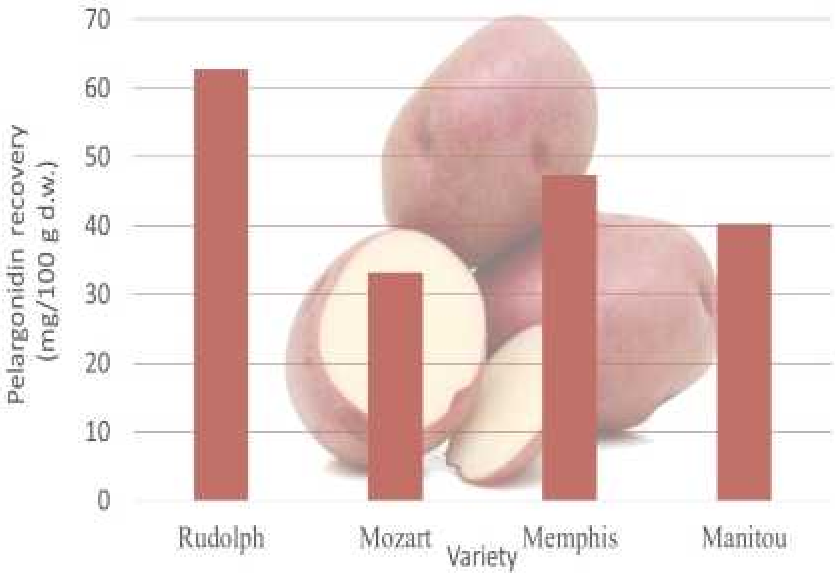

Figure 2. Comparison of Pelargonidin content between different red-colored potato varieties.

Comparing the antimicrobial activity of the extracts with that of the solvent used for extraction (**Table 1**), it can be stated that extracts rich in phenolic compounds act against clinical bacteria such as *Escherichia coli* or *Morganella morganii* and food bacteria such as *Listeria monocytogenes* or *Staphylococcus aureus*, among others. Moreover, this effect is due to the phenolic compound content and not to the action of the solvent. On the other hand, some extracts also had bactericidal activity against bacteria such as *MRSA*.

Table 1. Antimicrobial activity of red potato peel extracts from different potato varieties.

Bacteria	Solvent		Red potato varieties							
	ChCl:Lactic acid		Rudolph TPC = 0,16		Mozart TPC = 0,14		Manitou TPC = 0,11		Memphis TPC = 0,15	
	MIC	MBC	MIC	MBC	MIC	MBC	MIC	MBC	MIC	MBC
Food bacteria										
Enterobacter Cloacae	0.78	>50	**0.39**	>50	**0.39**	>50	**0.39**	>50	**0.39**	>50
Escherichia coli	0.78	>50	**0.39**	>50	**0.39**	>50	**0.39**	>50	**0.39**	>50
Pseudomonas aeruginosa	1.56	>50	1.56	>50	1.56	>50	1.56	>50	1.56	>50
Salmonella enterocolitica	0.78	>50	0.78	>50	0.78	>50	0.78	>50	0.78	>50
Yersinia enterocolitica	0.78	50	**0.39**	**1.56**	**0.39**	**1.56**	**0.39**	**1.56**	**0.39**	>50
Bacillus cereus	0.78	>50	**0.39**	>50	**0.39**	>50	**0.39**	>50	0.78	>50
Listeria monocytogenes	0.78	>50	**0.39**	>50	**0.39**	>50	**0.39**	>50	0.78	>50
Staphylococcus aureus	1.56	50	**0.78**	50	**0.39**	**25**	**0.39**	**25**	**0.39**	>50
Clinical bacteria										
Escherichia coli	0.78	>50	**0.39**	>50	**0.39**	>50	**0.39**	>50	**0.39**	>50
Klebsiella pneumoniae	0.39	>50	0.39	>50	0.39	>50	0.39	>50	0.39	>50
Morganella morganii	0.78	>50	**0.39**	>50	**0.39**	>50	**0.39**	**1.56**	0.78	>50
Proteus mirabilis	0.78	>50	**0.39**	>50	**0.39**	>50	**0.39**	>50	0.78	>50
Pseudomonas aeruginosa	0.39	>50	0.39	>50	0.39	>50	0.39	>50	0.39	>50
Enterococcus faecalis	0.78	>50	**0.39**	**0.78**	**0.39**	**0.78**	**0.39**	>50	0.78	>50
Listeria monocytogenes	0.39	>50	0.39	>50	0.39	>50	0.39	>50	0.39	>50
MRSA	0.78	>50	**0.39**	**0.78**	0.78	**0.78**	0.78	**0.78**	0.78	>50

TPC: Total phenolic content (mg Gal eq/ml); MIC: Minimum inhibitory concentration; MBC: Minimum bactericidal concentration

The results obtained show that recovery yields are higher in the green extraction, using choline chloride and lactic acid, than in methanolic extraction, being the Rudolph variety the richest. Therefore, NADES were shown to be promising alternative solvents for the isolation of phenolic compounds from by-products. Moreover, NADES extracts showed good activity against clinical and food bacteria. However, further research is needed to assess the properties and safety of the extracts, whether solvents need to be removed from the final extract, and to establish the best method for preserving them.

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Antidiabetic Potential of Cereal Byproducts

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Introduction

Type 2 diabetes mellitus is a complex metabolic disorder described by hyperglycemia and glucose intolerance. It is considered a new pandemic and its control involves numerous challenges [1]. Currently, the most common treatments consist of the use of oral hypoglycemic agents and insulin, treatments that have shown side effects (cardiovascular problems, liver and kidney disease, weight gain) [2].

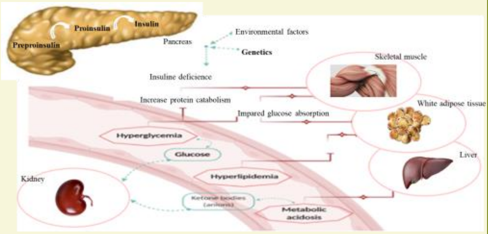

Fig. (1). Illustrative presentation of effects of insulin deficiency.

Dietary therapy displays a promising alternative for the prevention and treatment of this type of diabetes [3]. In this regard, cereals (especially whole grain with high contents of fiber or amylose content) are increasingly being incorporated into dietetic formulations to contribute to the amelioration of such disorder due to their antidiabetic potential [4].

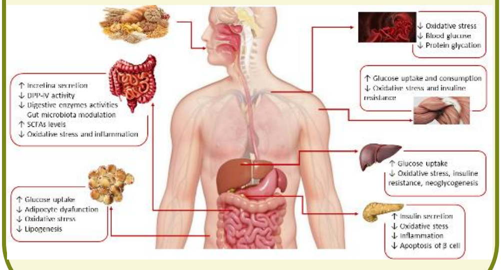

Fig. (2). Antidiabetic effects of dietary polyphenols and fibers.

Cereals as antidiabetic agents

This bioactivity is mainly due to their high content of fiber [5], which has several diabetes-related effects that include enhancement of insulin sensitivity, modulation of the secretion of certain gut hormones, and effects on various metabolic and inflammatory markers [6]. Moreover, soluble fiber inhibits the absorption of macronutrients, reduces the postprandial glucose response, and beneficially influences certain blood lipids [6,7]. Prospective cohort studies have shown that insoluble fiber intake is associated with a reduced risk of diabetes, but the mechanisms involved are unknown [6]. Antidiabetic potencial might also be ascribed to the presence of polyphenols [8], which could decrease insulin response, glycemia and insulin resistance, modulate carbohydrate and lipid metabolism, and regulate oxidative stress and inflammatory processes [9].

Use of cereal by-products

Large quantities of cereal by-products (*e.g.*, rice and rye by-products, or corn bran) are discarded worldwide and could be valorized since they are a rich source of fiber and polyphenols [8,10,11]. In this way, these by-products could be directly used in food fortification aiming to diabetic people or they could be subjected to extraction of the relevant compounds to be then used as nutraceuticals in functional foods.

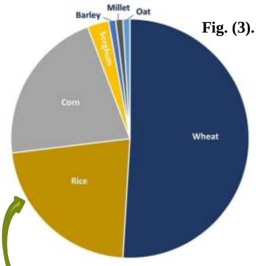

Fig. (3). Percentage of cereal waste

Agroindustrial waste	
Peel	Bark
Cake	Bran
Seed	Leaf

Conclusion

Valorization of cereal by-products offers many applications by obtaining value-added products with health-benefits for diabetic people within the circular and sustainable bioeconomy perspective. This also contribute to sustainably manage cereal wastes, resulting in the formation of value-added products within a circular and sustainable bioeconomy [12].

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Acknowledgments

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CIRCULAR USE OF CRAFT BEERS BY-PRODUCTS: CHARACTERIZATION OF THE PHENOLIC PROFILE AND ANTIOXIDANT CAPACITY OF SPENT GRAINS AND HOPS

Transcolab Summit · Trends in grains-based foods · March 2022

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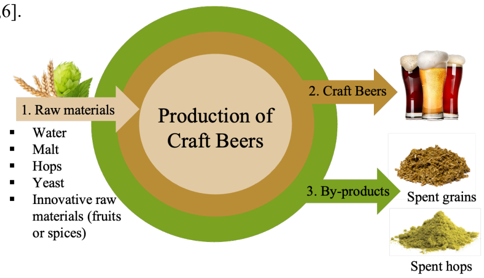
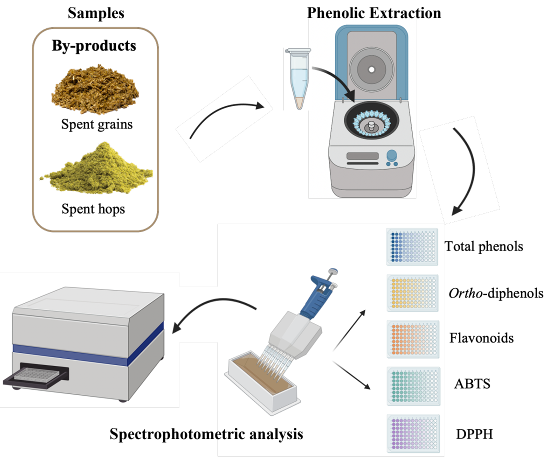
INTRODUCTION

Craft beers is a new concept introduced in the beer industry with great growth [1,2]. Although no definition legally determined, most authors define it as a product that is produced using traditional practices and using traditional raw materials, namely water, malt, hops, yeast or innovative raw materials, such as fruits or spices [3,4]. The production of craft beer includes the formation of huge amounts of various by-products such as spent grains and spent hops. The high production of theses by-products spell a major environmental problem and can be reused for microalgae production, biofuel production, extraction of polyphenolic, and antioxidants as a way of help in achieving ecological sustainability [5,6].



In this sense, the main goal of this study was to determine the phenolic composition and antioxidant capacity of two different by-products of the craft beer industry, namely spent grains and spent hops. The evaluation for each by-product was carried out by the determination of the total polyphenolic, *ortho*-diphenols, and flavonoids contents and the determination of antioxidant capacity using the ABTS and DPPH methodologies.

MATERIALS AND METHODS



ACKNOWLEDGEMENTS

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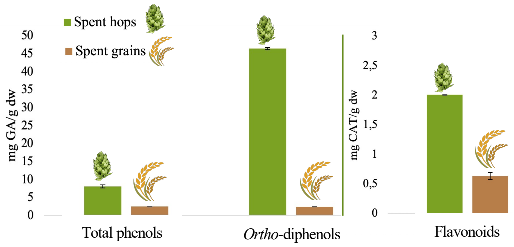
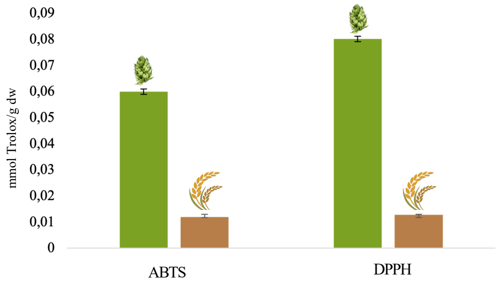
RESULTS AND DISCUSSION

Phenolic composition of by-products from craft beers.



Antioxidant capacity of by-products from craft beers.



CONCLUSIONS

Craft beer brewing generates substantial quantities of by-products, such as spent grains and spent hops, which can be reintroduced in a circular economy as a source of phenolic compounds, to be further included in healthy products, such as supplements, nutraceuticals, functional foods, and cosmetics [7].

The results obtained in this study indicate that by-products from craft beers, mainly spent hops, are a rich source of phenolic compounds. Thus, the reuse of these by-products represent sustainable and good alternative for bioactive compounds recovery and could be significantly useful in cosmetic, pharmaceutical, and food industry.

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SPONGE AND LAYER CAKE FLOURS AS A NOVEL INGREDIENT IN CAKE MAKING

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TRANSCOLAB SUMMIT
Trends in grains-based foods
MARCH 2022

Introduction

Food waste has a significantly environmental impact, which will be reduced reintroducing all waste suitable for human consumption on the food chain. Cakes are a popular bakery product that can be wasted in the manufacturing process.

The aim of this study is to evaluate how adding cake flour to sponge and layer cake formulations influence their final quality to reduce cake waste.

Materials and methods

Layer cake

Sponge cake

Drying (25ºC; 4 days)

1000 µm

Layer cake flour (LCF) → Addition 10 %, 20%, 30% → **Layer cakes**

Layer cakes	
Wheat flour	350 g
Sugar	315 g
Milk	210 g
Egg	175 g
Sunflower oil	105 g
Baking powder	10,5 g

Sponge cake flour (SCF) → Addition 10 %, 20% → **Sponge cakes**

Sponge cakes	
Egg	344 g
Wheat flour	245 g
Sugar	240,5 g
Water	55 g
Powdered milk	25 g
Emulsifier	14 g

Cake elaboration

Layer

Mixing ingredients

Sponge

Creaming- mixing

Powdered milk and flour adding

Baking

Batter characteristics

Density

Viscosity

Cake properties

Weight loss

Colour

Texture

Volume

Results

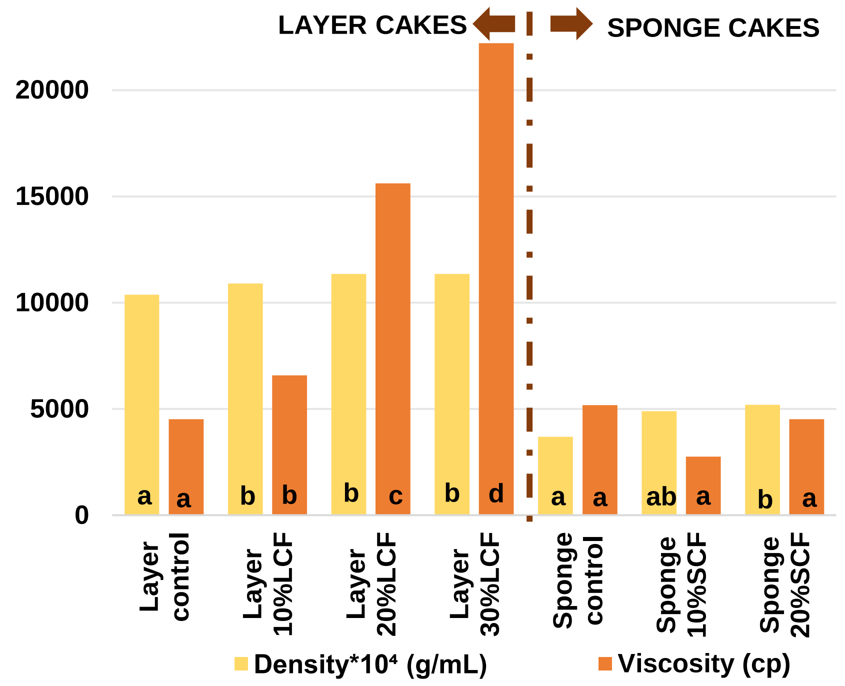

Figure 1. Density and viscosity of cake batter

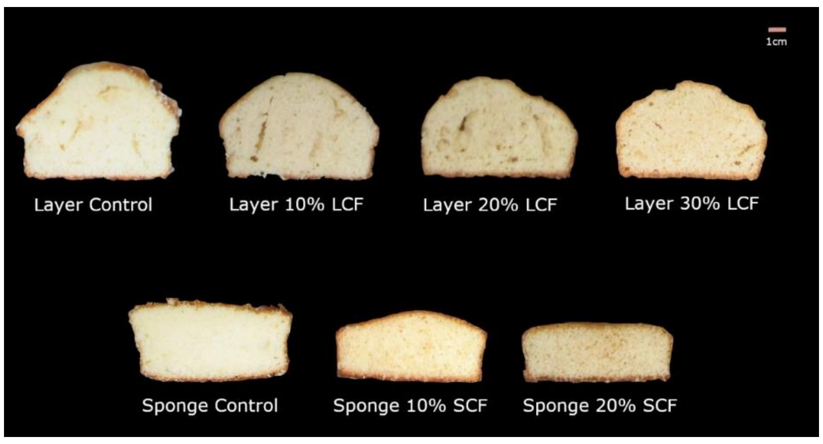

Figure 2. Images of cake slices

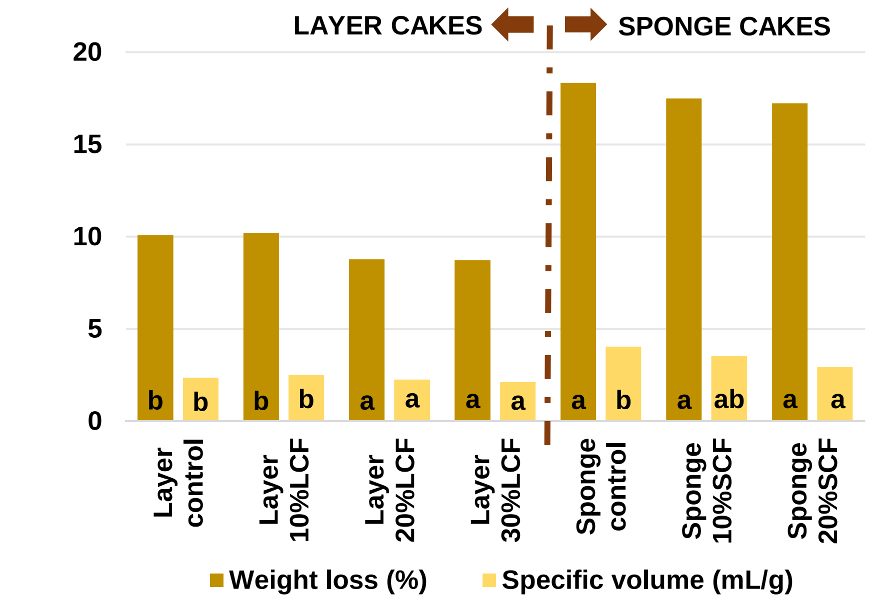

Figure 3. Weight loss and specific volume of cakes

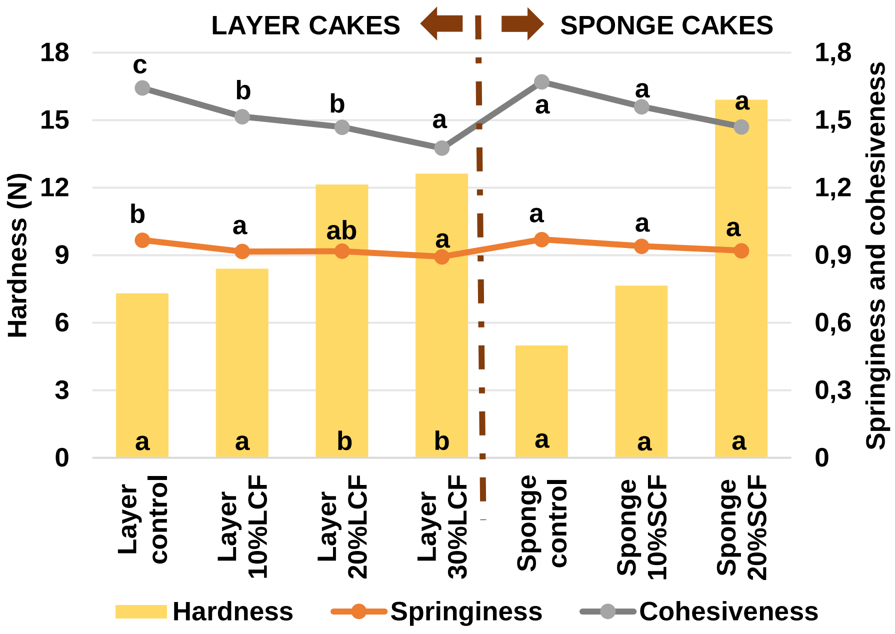

Figure 4. Hardness, springiness and cohesiveness of cakes

Conclusions: The addition of cake flour to reduce food waste in the production of cakes is possible in sponge and layer cakes. However, it is not possible adding more than 10% without significantly modifying the textural properties and the specific volume of the cakes.

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THE COMPARATIVE EFFECT OF THE USE OF THREE PULSE FLOURS ON THE QUALITY OF A REDUCED MEAT COOKED SAUSAGE

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Introduction

An excess in meat consumption can negatively affect health and environmental sustainability. One strategy to reduce meat consumption is to reduce the amount of meat in meat products by using quality plant protein sources, such as pulses [1,2]. Emulsion type sausages allows reformulation approaches and thus a reduction in the amount of meat.

Materials and methods

Reduced meat frankfurters containing contained 38% pork meat, 35% water, 20% pork backfat, 18 g common salt/kg, 3 g phosphates/kg and spices were made in duplicate following four treatments: control sausage (CON), also containing 5.3% potato starch and 1.7 % soy protein, and BEA, CHI and LEN sausages, formulated with 7 % bean, chickpea and lentil, respectively. Pulses were added as flour without more pre-treatments than milling. All the experimental treatments. The amount of starch and protein among the sausages from the different treatments were similar. The sausages were analysed for water retention, emulsion stability, instrumental texture and color, and oxidative stability after 10 days of refrigerated aerobic storage.

Results

The use of pulse flour, with respect to CON, did not excessively modify yield, texture and colour characteristics, although a lower cooking performance and cohesiveness were detected in the pulse-containing sausages (Table 1).

Table 1:

	Control	BEA	CHI	LEN	P-level
Cooking loss (%)	1.77±0.26b	4.18±0.57a	4.46±0.70a	4.56±0.16a	0.011
Cohesiveness	0.92±0.03	0.73±0.03	0.79±0.03	0.75±0.11	0.094
Oxidative stability (mg MDA/kg)[&]	0.59±0.04c	5.21±0.64a	3.63±0.06ab	2.57±0.58b	0.002

MDA: malondialdehyde
[&] Thiobarbituric reactive substances in the sausage after 10 days of refrigerated storage under aerobic conditions.
a,b Mean values with different superscript showed significant differences (Tukey test, P<0.05)

Moreover, pulses promoted the oxidation of the sau-sages during refrigerated storage with this effect being more intense for BEA sausages. This finding might be explained by the presence in the pulses of lipoxygenase activity (Shi et al., 2020), with a relatively high ther-mo-resistance, and thus resistant to the sausage pas-teurization. The effect of pulse flour on decreasing lipid oxidation stability was higher in BEA sausages than in LEN, with CHI being in an intermediate position. Further research is needed on how to ameliorate this negative effect of pulse flours as meat replacer.

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Coffee silverskin as a sustainable ingredient to prevent type 2 diabetes

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Transcolab Summit
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Introduction

Coffee silverskin (CS)

→ The major by-product of coffee roasting industries [1]

→ Valuable ingredient for functional food formulation

- Rich in protein and dietary fiber → Both reduce the glycemic response [2]
- Rich in several bioactive compounds (particularly chlorogenic acids (CGA) and caffeine)
  ↳ Modulate sugar metabolism [3]

Aim: To study the potential of CS as alternative ingredient for functional foods able to prevent or manage type 2 diabetes (DM2).

Methodologies

1

Protein [4]
(Kjeldahl method)

2

Dietary fiber [4]
(Enzymatic-gravimetric method)

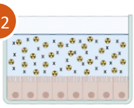
3

Extract preparation
(Ultrasound-assisted extraction)

3.1

CGA profile and caffeine content [1]
(RP-HPLC-DAD)

3.2



Effects of extract and its major compounds on intestinal sugar uptake [5]
(Quantification of ^{3}H-deoxy-D-glucose (^{3}H-DG) and ^{14}C-fructose (^{14}C-FRU) uptake by Caco-2 cells)

Results and Discussion

Table 1. Composition of CS and CS extract.

CS composition (g/100 g fresh weight)	
Protein	12.0 ± 0.3
Dietary fiber	56.4 ± 0.7
▪ Insoluble	49.1 ± 0.4
▪ Soluble	7.3 ± 0.1
CS extract composition (mg/g of freeze-dried extract)	
Caffeine	27.7 ± 0.2
Σ CGA	4.19
▪ 3-CQA	0.31 ± 0.09
▪ 4-CQA	0.15 ± 0.04
▪ 5-CQA	1.98 ± 0.05
▪ 4-FQA	0.43 ± 0.04
▪ 5-FQA	1.32 ± 0.05

Results are expressed as mean ± standard deviation. CQA, Caffeoylquinic acid; FQA, Feruloylquinic acid

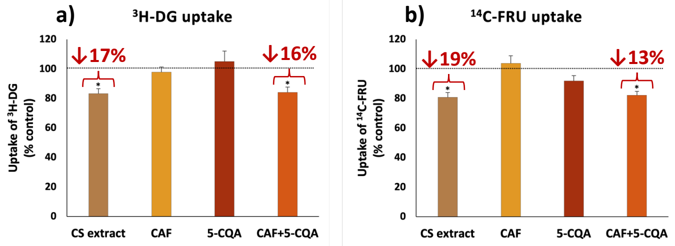


Figure 1. Effect of CS extract (1 mg/mL), caffeine (CAF), 5-caffeoylquinic acid (5-CQA), and CAF+5-CQA on (a) ^{3}H-DG and (b) ^{14}C-FRU uptakes.
Note: The effects of the standards (alone and combined) were tested at the concentrations present in 1 mg/mL of CS extract. Results are expressed as mean ± SEM. * Significantly different from control at *p*<0.05 (Student's t-test).

- Richness of CS in insoluble fiber and protein;
- In CS extract, caffeine was the major compound detected and 5-CQA was the main CGA;
- A synergism between caffeine and 5-CQA seemed to have a major role in the effects found for CS extract, although in the case of ^{14}C-FRU uptake, other compounds seem to be also involved;
- No effects were found when caffeine and 5-CQA were tested individually.

Conclusion

CS can be valued and used in food formulations with low glycemic index, to intestinal sugar uptake, and potentially and help to prevent or manage DM2.

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SUSTAINABLE FLOUR PRODUCTION OF OPUNTIA FICUS-INDICA (L.) MILLER CLADODES: A MINERAL PROFILE APPROACH

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Introduction:

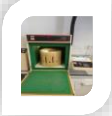
Currently, consumers are concerned about nutrition and food diversity, favoring healthier foods with proven beneficial effects on health [1]. On the other hand, consumers are increasingly demanding and concerned about sustainability and companies' sustainable practices. *Opuntia ficus-indica* (L.) Miller is a possible food alternative, being part of the dietary pattern of countries such as Mexico [2]. In Portugal, with the increase in prickly pear production, the discarded cladodes generated waste management issues, due to its invasive characteristics. However, this non-valued by-product has potential for valorization due to its nutritional/chemical composition. This work intends, therefore, to valorize cladodes for food consumption and/or applications in the food industry. One example is the production of flours for new food formulations such as bread. Hence, the mineral profile of cladodes of different varieties (2 years old) from plants cultivated in Portugal (Torres Novas) in the organic production mode were evaluated.

METHODOLOGY

SAMPLE PREPARATION

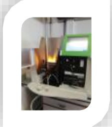
ACID DIGESTON

250 mg of cladodes
4 ml HNO_3
1 ml H_2O_2



MLS-1200 Mega
High performance microwave oven

After digestion, dilute to a final volume of 25 ml



Atomic absorption - AAS
Na, K, Ca, Mg and Fe

Thermo Scientific™ iCAP Q ICP-MS
Li, Be, B, Al, Ti, V, Cr, Mn, Co, Ni, Cu, Zn, As, Se, Rb, Sr, Zr, Mo, Cd, Sn, Sb, Te, Cs, Ba, W, Pb, Bi

31 elements analyzed

RESULTS AND DISCUSSION

Essential trace elements				
	BA	BB	L	V
Fe (µg/g)	28.37 ± 0.35	16.22 ± 0.86	14.03 ± 0.65	11.62 ± 0.90
Cu (µg/g)	0.888 ± 0.022	4.064 ± 0.049	2.994 ± 0.086	1.913 ± 0.021
Zn (µg/g)	24.83 ± 0.25	33.30 ± 0.32	7.38 ± 0.47	5.06 ± 0.16
Mn (µg/g)	80.14 ± 0.92	333.0 ± 7.4	28.64 ± 0.89	14.51 ± 0.18
Mo (µg/g)	< LoD	2.07 ± 0.32	< LoD	< LoD
Co (µg/g)	0.485 ± 0.011	0.4609 ± 0.0062	0.1730 ± 0.0057	0.09145 ± 0.00097
Cr (µg/g)	76.9 ± 1.4	50.6 ± 1.9	236.5 ± 19.8	244 ± 38
Se (µg/g)	< LoD	0.104 ± 0.041	< LoD	< LoD
Non-essential and toxic trace elements				
	BA	BB	L	V
Al (µg/g)	109.8 ± 3.3	119.9 ± 1.8	13.89 ± 0.41	5.816 ± 0.067
As (µg/g)	< LoD	0.038 ± 0.012	< LoD	< LoD
B (µg/g)	12.45 ± 0.46	16.20 ± 0.71	8.81 ± 0.43	9.64 ± 0.13
Ba (µg/g)	59.41 ± 0.70	78.27 ± 3.37	38.7 ± 1.1	19.61 ± 0.44
Be (ng/g)	1.033 ± 0.027	0.228 ± 0.0080	0.0195 ± 0.0015	0.00333 ± 0.00041
Bi (µg/g)	< LoD	< LoD	< LoD	< LoD
Cd (ng/g)	14.94 ± 0.47	34.09 ± 0.27	9.83 ± 0.56	8.69 ± 0.54
Cs (ng/g)	178.13 ± 0.34	99.5 ± 1.2	9.84 ± 0.84	8.62 ± 0.46
Li (ng/g)	12.47 ± 0.44	15.3 ± 2.7	4.53 ± 0.42	5.76 ± 0.60
Ni (µg/g)	0.367 ± 0.010	1.725 ± 0.039	0.594 ± 0.020	0.594 ± 0.019
Pb (ng/g)	585 ± 14	845.3 ± 8.9	134.4 ± 5.5	60.2 ± 1.8
Rb (µg/g)	9.13 ± 0.29	35.10 ± 0.24	6.07 ± 0.25	6.68 ± 0.06
Sb (µg/g)	< LoD	< LoD	< LoD	< LoD
Sn (ng/g)	< LoD	5.0 ± 3.5	4.88 ± 0.40	6.6 ± 1.6
Sr (µg/g)	31.6 ± 1.3	64.1 ± 2.2	32.1 ± 1.6	19.10 ± 0.19
Te (µg/g)	< LoD	0.00180 ± 0.00028	< LoD	< LoD
Ti (µg/g)	28.3 ± 1.0	20.64 ± 0.19	54.4 ± 2.6	51.5 ± 1.2
V (µg/g)	< LoD	< LoD	< LoD	< LoD
W (µg/g)	6.26 ± 0.41	< LoD	< LoD	7.15 ± 0.73
Zr (ng/g)	4.45 ± 0.54	5.27 ± 0.84	5.38 ± 0.68	4.68 ± 0.29
Macro elements				
	BA	BB	L	V
Ca (g/Kg)	66.6 ± 2.5	32.7 ± 1.3	111.4 ± 1.1	104.5 ± 5.8
K (g/Kg)	14.8 ± 1.7	53.0 ± 8.4	11.2 ± 1.3	12.9 ±2.1
Mg (g/Kg)	15.89 ± 0.46	17.35 ± 0.35	6.77 ± 0.12	4.95 ± 0.27
Na (mg/kg)	53.2 ± 5.2	56.9 ± 3.0	57.84 ± 0.37	69.5 ± 0,53

Values expressed in µg/g of dry weight (dw) (B, Al, Ti, V, Cr, Mn, Co, Ni, Cu, Zn, As, Se, Rb, Sr, Mo, Sb, Te, Ba and Bi) and in ng/g dw (Be, Li, Zr, Cd, Sn, Cs, W and Pb); Cladodes of *Opuntia ficus-indica (L.) Mille* (BA – white variety of Arrepiado; BB – white variety of Barquinha; L – Orange variety of Olaia: V – Red variety of Olaia).

Macro elements: All samples had substantial Ca contents (33-111 g/kg), high levels of K (11-53 g/kg) and Mg (5-17 g/kg), but low Na.
Non-essential trace elements: mostly Mn (15-330 µg/g), Zn (5-30 µg/g) and B (10-20 µg/g).
Essential trace elements: Fe, Cu, Zn, Mn, Co and Cr were identified in all samples in varying amounts (0.1-333 µg/kg). Mo and Se were only identified in BB.

Minerals are fundamental for maintaining organism homeostasis. When this flour is incorporated in foodstuffs these mineral contents can help reach the Daily Recommended Intakes.

CONCLUSION

According to the obtained results and following circular economy principles, the cladodes have the potential for the development of nutritionally balanced flours, enriched in minerals, which contribute to valorizing this by-product and promoting food chain sustainability.

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VALORIZATION OF PULSE BY-PRODUCTS FROM THE MEDITERRANEAN DIET

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Introduction

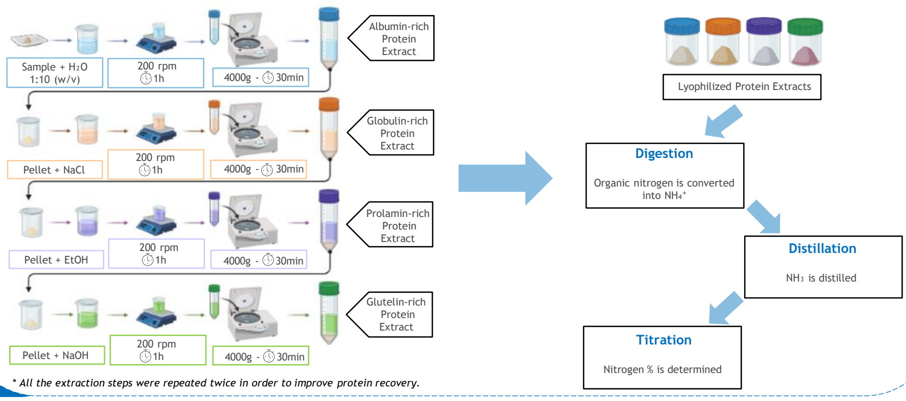
The increase in standard of living and income along with the rising population in developing countries are factors contributing to a drastic increase in the global demand for meat consumption and production. This reality poses a great concern for human health since the excessive consumption of meat is linked to cancer and several other chronic diseases. Meat production is also the principal cause for deforestation, land degradation, water pollution, desertification and, when compared to plant-based foods, production of meat results in significant higher greenhouse gas emissions. Thus, more sustainable food sources and diets are needed in order to meet future demands regarding global nutrition [1, 2]. Pulses, a low-fat source of proteins and carbohydrates, are known and for both their nutritional and environmental benefits. Not only their ability to restore nitrogen content in the soil is an appealing green solution in crop productivity, but pulse grains are also of interest in plant protein-based alternatives [3]. Protein content in pulses ranges 17-30% of dry weight, containing higher proportions of protein than other plant foods. Rich in carbohydrates, pulses also provide substantial amounts of minerals and vitamins which bioactivity might improve glycaemic control, protect against hypercholesterolemia, cancer, type 2 diabetes [4].

The present study aimed at characterizing the proximate composition of two types of dry pulses typically consumed in the Mediterranean diet, cowpea (*Vigna unguiculata*) and butter bean (*Phaseolus lunatus*), while focusing on the study of protein extracts. The future goal is to evaluate the bioactive and technological functionalities of the protein extracts obtained, which may allow for its use in food formulation.

Methods

OSBORNE SEQUENTIAL PROTEIN EXTRACTION

KJELDAHL METHOD



** All the extraction steps were repeated twice in order to improve protein recovery.*

Results

Table 1. Proximate composition (% as is) of cowpea and butter bean seed flour sample.

	Cowpea	Butter bean
Protein	19.41 ± 0.08	16.03 ± 0.08
Lipid	0.523 ± 0.198	0.264 ± 0.002
Carbohydrates	42.07 ± 0.96	43.92 ± 1.93
Ash	4.03 ± 0.18	5.15 ± 0.05
Moisture	10.93 ± 0.23	14.03 ± 0.06

Table 2. Protein contents (% as is) obtained by Kjeldahl method for cowpea and butter bean protein extracts from the Osborne extraction method.

	Protein Extract (soluble in)	Cowpea		Butter bean	
		% N	Protein	% N	Protein
Flour sample (< 0,45 mm)	H_2O	6.95	36.70	8.60	45.41
	NaCl	0.83	4.37	0.53	2.81
	EtOH	1.16	6.12	0.86	4.54
	NaOH	2.07	10.93	1.96	10.35
Flour sample (0,45-0,71 mm)	H_2O	3.72	19.64	3.88	20.49
	NaCl	0.64	3.39	0.46	2.44
	EtOH	1.43	7.55	1.65	8.71
	NaOH	1.41	7.44	2.50	13.20
Flour sample (> 0,71 mm)	H_2O	4.18	22.07	4.94	26.08
	NaCl	0.45	2.35	0.27	1.44
	EtOH	1.53	8.08	0.16	0.85
	NaOH	n.a.	n.a.	0.40	2.11

n.a., not analized due to insufficient biomass sample.
The conversion factor used was 5,28 [5].

Conclusion

- The proximal characterization of both cowpea and butter bean showed high content of carbohydrates (42.07 and 43.92 %, respectively) and proteins (19.41 and 16.03 %, respectively) and low lipid content (0.52 and 0.26 %, respectively), which is in accordance with the literature [6];
- After application of the Osborne fractionation method both matrices showed higher protein content when H_2O was used as solvent (36.70 and 45.41 %, respectively), which indicate that water soluble proteins are predominant in these kind of biomass, being NaCl and NaOH the ones showing less effectiveness regarding protein extraction;
- Further research into the functional properties and digestibility of these protein extracts will be necessary, in order to select the one with high level of nutritional composition, allowing for its application in food formulation.

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COFFEE BY-PRODUCTS: A POTENTIAL SOURCE OF ALTERNATIVE FLOURS WITH PREBIOTIC PROPERTIES

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Introduction

Coffee is one of the most popular beverages in the world, but it also produces considerable amounts of by-products such as pulp, parchment, silverskin, and defective beans. In general, all these parts are high in dietary fiber (xylans, arabinogalactans, galactomannans) and phenolic compounds (chlorogenic acids) [1]. These components have been associated with prebiotic properties, that is, they have low digestibility and stimulate the growth of beneficial bacteria in the colon [2,3]. Coffee by-products can be an interesting solution to improve the functional and nutritional properties of traditional bakery products [1]. In this work, the use of different coffee by-products as a source of prebiotic ingredients, as well as their potential application in bakery products was ascertained, having in view the economic valorisation and contribution to food security.

Methodology

Scopus

PubMed

Keywords	Results	Screening	Eligibility
"coffee" in conjunction with "prebiotic", "gut microbiota", "probiotic", "pulp", "husk", "silverskin", "spent", beans", "parchment" and "mucilage".	72	56	18

Approach: Articles that did not coincide with the theme or did not test the impact of a coffee by-product on the intestinal microbiota and/or production of SCFA were excluded.

Results and discussion

1. Dehulling

The addition of 5% coffee pulp to the diet of obesity-induced rats increases the growth of *Akkermansia muciniphila* [4].

Xylooligosaccharides from coffee husk xylan, particularly xylobiose and xylotriose, are easily metabolized by probiotic bacteria (*Lactobacillus* and *Bifidobacterium*) [5].

COFFEE CHAIN

2. Selection

3. Torrefaction

4. Brew

Mannanoligosaccharides from spent coffee grounds (SCG) promote lactic acid bacteria growth, inhibit pathogen bacteria growth, and are less digestible than inulin (a well-known commercial prebiotic) [6,7].

Silverskin increases bifidobacteria and lactobacillus levels while decreasing potentially pathogenic bacteria like clostridia and bacteroides [9].

SCG, in their natural form, increase the production of short-chain fatty acids with potential health benefits and decreases the pH [8]. SCG, in the absence of melanoidins, increase the number of *Lactobacillus* and *Bifidobacteria* [9].

According to the findings, coffee by-products are frequently used to produce oligosaccharides (structures containing 2 to 20 sugar units). These structures meet prebiotic requirements: low digestibility; promote the growth of beneficial bacteria; and inhibit the growth of pathogenic bacteria. In general, fecal bacteria have the ability to use coffee by-products as a carbon source. No research on prebiotic action of parchment, mucilage, or defective beans was found. Considering the attributes of coffee by-products, they could be used as alternative flours in bakery products. In some studies, they were included in bread and cookies, with interesting nutritional, sensory, and rheological results [1,10].

Conclusion

Using coffee by-products as an alternative flour can be a viable approach for adding value to these materials, promoting the circular economy, and obtaining a functional food at the same time.

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Tracking African beans' diversity:

a physicochemical study of *Vigna unguiculata* (L.) Walp. and *Phaseolus vulgaris* L.

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In sub-Saharan Africa, pulses, and legumes in general, are essential food sources for alleviation of malnutrition and can play an important role in sustainable agriculture, due to their ability to fix atmospheric nitrogen **[1]**. The common bean (*Phaseolus vulgaris* L.) and cowpea (*Vigna unguiculata* (L.) Walp.), are among the most important pulses in the dry areas of tropical Africa. Based on field surveys performed across three African countries (i.e., Angola – Western Africa; Mozambique – Eastern Africa and Cabo Verde Islands), we aim to investigate the patterns of physicochemical variation (i.e., seed phenotypic traits and mineral content) of the *Vigna unguiculata* and *Phaseolus vulgaris*, in order to evaluate the patterns of diversity between native and introduced species, respectively. According to our results, chemical composition of *Vigna unguiculata* has, higher content of B, Mg, S, and Zn, while *Phaseolus vulgaris* had more content of Fe, Ca, and Cu. Respecting the physical diversity, the analysis of seed phenotypic traits showed that both studied species presented a varied array of colours and seed shapes. We conclude that both species are important resources in Angola, Mozambique and Cabo Verde Islands, but native *Vigna* species are often disregarded, even though they have generally good alimentary properties **[2]**. To the best of our knowledge, this is the first comparative physicochemical study, focusing on wild and crop species, collected across different African countries. It is highlighted that interdisciplinary approaches and new data are need for the sustainable use of African plant genetic resources, contributing to achieve some of the Sustainable Development Goals, as the reduction of hunger and increasing human health.

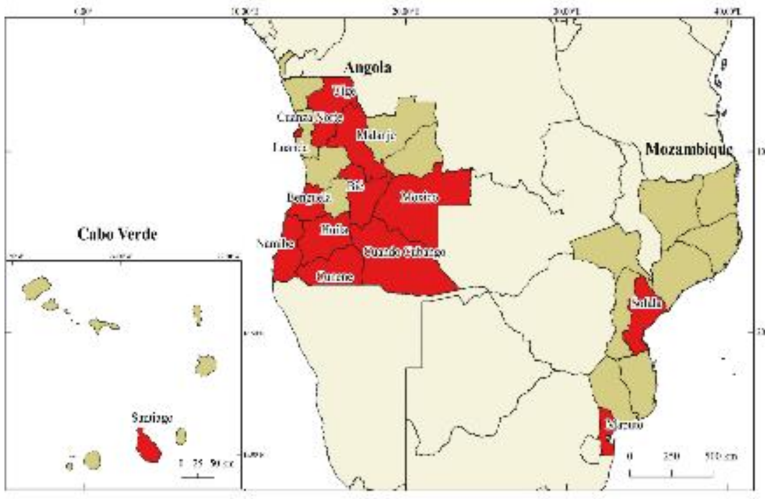
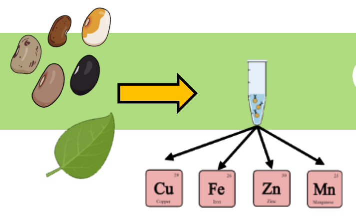


Figure 1. Countries and provinces where samples were collected (in red color): Cabo Verde (Santiago), Angola (Uíge, Cuanza Norte, Malanje, Benguela, Namibe, Bié, Moxico, Huíla, Cunene, and Cuando Cubango), and Mozambique (Sofala and Maputo).

CHEMICAL DIVERSITY



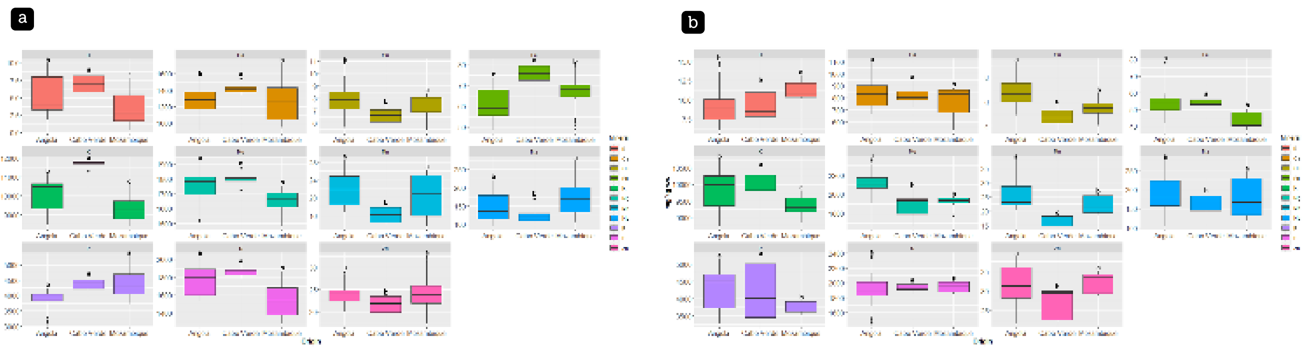


Figure 2. Variation of mineral content (mg/kg ww) and the geographical origin for the studied samples: **(a)** *Phaseolus vulgaris* (21 accessions), and **(b)** *Vigna unguiculata* (17 accessions).

Geographical origins sharing the same letter are not statistically different according to the Scott-Knott test at 5% of confidence.

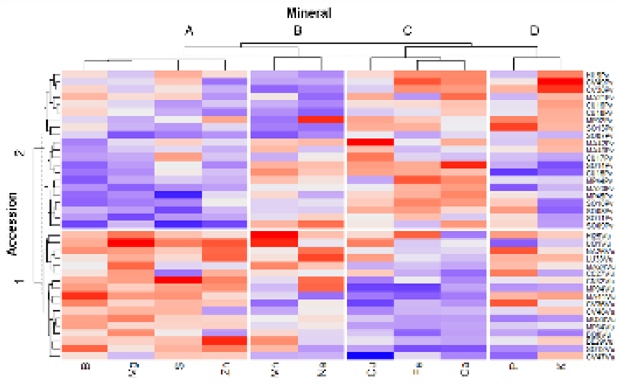


Figure 3. Heatmap of the 21 *Phaseolus vulgaris* and 17 *Vigna unguiculata* accessions obtained from the chemical characterization data for the content of 11 minerals. Red and blue boxes indicate high values and low values respectively.

PHYSICAL DIVERSITY

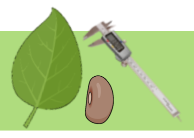


Table 1. Average values[a] (mm) ± standard deviation for morphometric measurements of *Phaseolus vulgaris* and *Vigna unguiculata* accessions from African countries.

Species	Length	Width	Height
Vigna unguiculata	13.5±2.3	7.1±0.7	5.9±3.4
Phaseolus vulgaris	8.0±1.2	6.5±1.0	4.9±0.8

[a]Average values of three separate determinations (n=10);

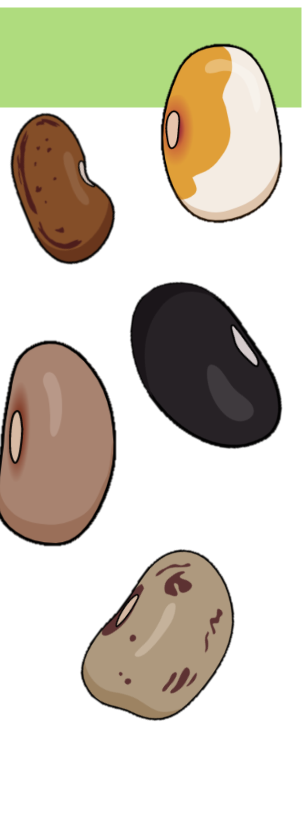


Figure 5. Photographs of the **(a)** *Phaseolus vulgaris* and **(b)** *Vigna unguiculata* accessions used in this study, collected, between 2018 and 2019, in Angola, Mozambique and Cabo Verde.

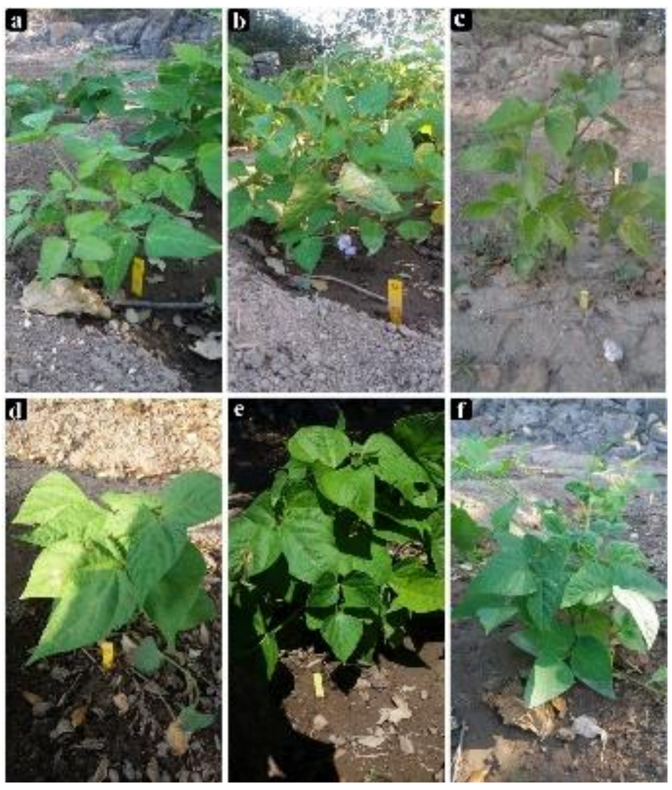


Figure 6. Cultivated samples collected in different African countries: **(a)** *Vigna unguiculata* from Malanje, Angola (MA41Vu); **(b)** *V. unguiculata* from Sofala, Mozambique (SO12Vu); **(c)** *V. unguiculata* from Santiago, Cabo Verde (CV36Vu); **(d)** *Phaseolus vulgaris* from Cunene, Angola (CU17Pv); **(e)** *P. vulgaris* from Sofala, Mozambique (SO10Pv); and **(f)** *P. vulgaris* from Santiago, Cabo Verde (CV38Pv). Plants growth under controlled conditions, between June 2019 and February 2020, at Instituto Superior de Agronomia of the University of Lisbon (ISA/UL).

NEW DATA contributes to the valorization of plant genetic resources & food security in Africa!

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IDENTIFICATION OF MOULDS ISOLATED FROM ARTISANAL AND INDUSTRIAL BREADS

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TRANSCOLAB SUMMIT
Trends in grains-based foods
MARCH 2022

INTRODUCTION

Bread is one of the main foods in the Mediterranean diet. Its environmental conditions are needed to be controled, such as high temperature or humidity which could lead to microbial growth in example moulds. This kind of microorganism are involved in formation of secundary metabolites called mycotoxines, which could cause severeal health damages. Because of this, the knowledge of the types of moulds that can be found in bread will allow to know their involvement in their impact on the hygienic quality of bread.

AIM

The main aim of this study was to identify the moulds isolated from different types of bread by Matrix-Assisted Laser Desorption/Ionization- Time of Flight Mass Spectrometry (MALDI-TOF).

MATERIAL AND METHODS

Samples: Bread samples were obtained from four artisan and four industrially produced breads. White wheat loaf, whole wheat loaf, Candeal bread, and Tin bread loaf were collected by two artisanal and two industrial bakeries twice and on different days from September until February 2021 in Valladolid city (Figure 1).
Materials: 25 g of crust or crumb were diluted in 225 ml of peptone water. Subsequently, 100 µl of appropiated diluted were grown onto dichloran glycerol (DG 18). Plates were incubated at 22°C for 5 days. After that, 2 colonies ere isolated from ecach Petri plate. Isolated were identified by MALDI-TOF using three methos of extraction: water-formic acid solution, zirconia-silica beads and extraction with an ethanol-water solution.
Analysis: mass spectrum was messured by Flex Control (version 3.1 Bruker Daltonik) and statistical analysis was processed by Excel (Office 2019) and SPSS Statistics (version 24).

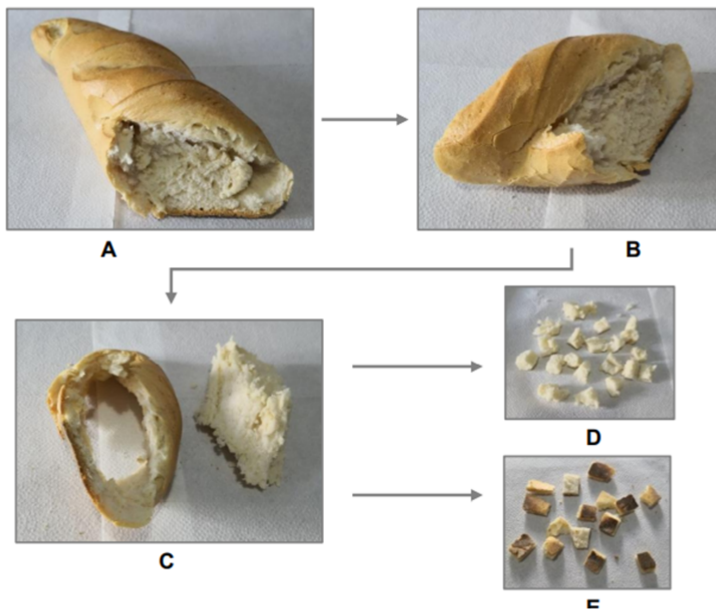
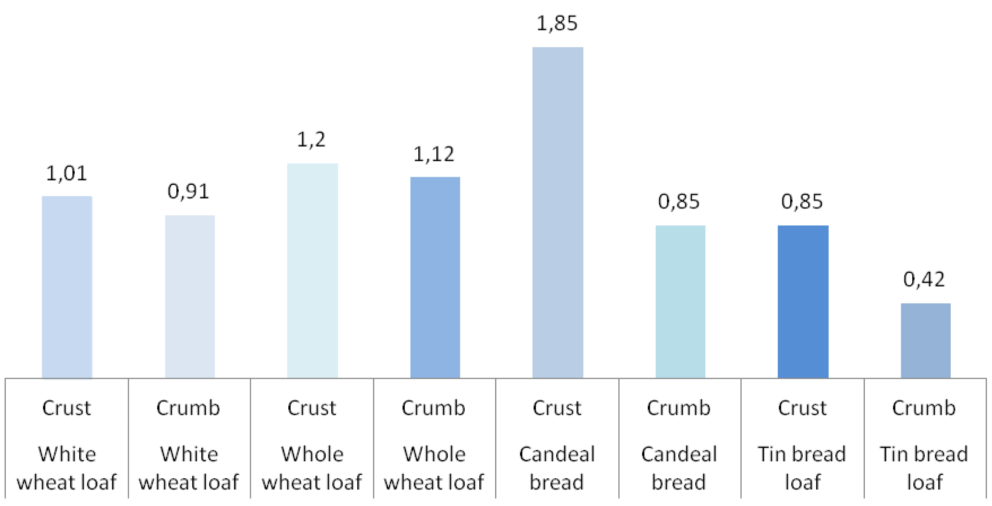


Figure 1. Process of separation of crust and crumbs in breads. A: longitudinal cutting of bread. B: size reduction. C: separation of crust and crumbs. D: reduction and homogenization of crumb size. E: reduction and homogenization of crust size.

RESULTS

A



B

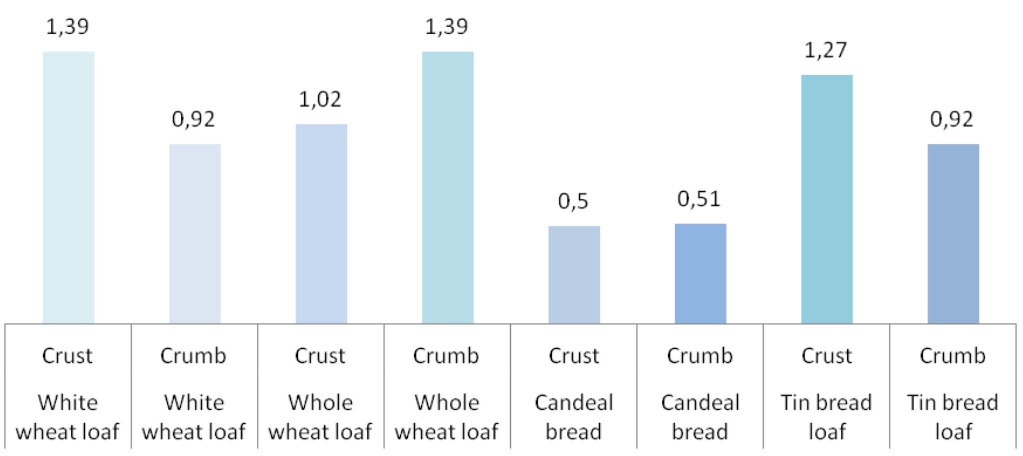


Figure 2. Counts of moulds (Log10 UFC/g) from different types of bread separating crumbs and crust. A: Artisanal bread. B: Industrial bread

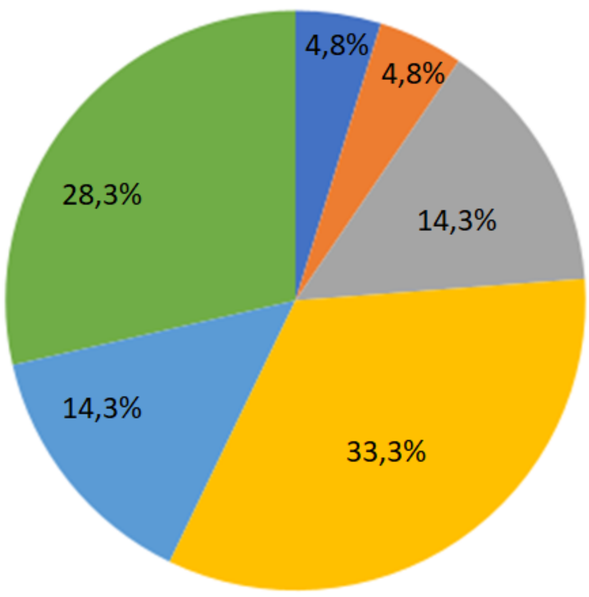


Figure 3. Main mould species identified by MALDI-TOF using two extraction methods: long extraction and zirconia-silica beads. Dark blue: *Candida solani*. Orange: *Filifactor villosus..* Grey: *Aspergillus niger*. Yellow: *Penicillium chrysogenum*. Blue: *Penicillium camemberti*. Green: no identified

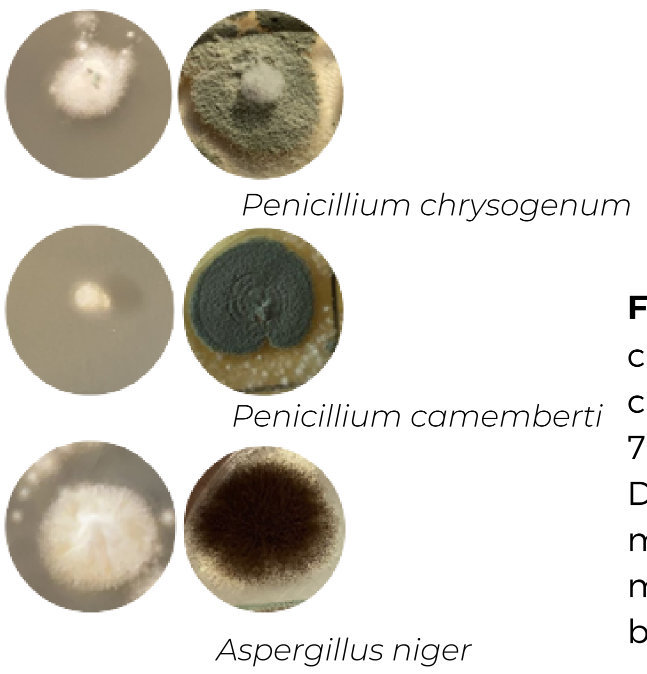


Figure 4. Morphological characteristics of colonies grown at 3 and 7 days of incubation in DG18 of the species of moulds producers of mycotoxines identified by MALDI-TOF.

CONCLUSIONS

Spectrometry techniques of masses are currently more useful than another inespecific one such as morphological exam to identify microorganisms. MALDI-TOF allowed the identification of three species of moulds which are producers of mycotoxins: *Penicillium chrysogenum*, *Penicillium camemberti* and *Aspergillus niger*.

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BIOACTIVE “FLOURS” FROM BREWING BY-PRODUCTS: A SUSTAINABLE AND HEALTHY ALTERNATIVE FOR FOOD INDUSTRY

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TRANSCOLAB SUMMIT
Trends in grains-based foods
MARCH 2022

INTRODUCTION

Major **brewing by-products**:

~85% **BSG** (brewer’s spent grain) = the insoluble part of the barley grain

~15% **BSY** (brewer’s spent yeast) = *Saccharomyces pastorianus* from Lager beer

100 L of brewed beer produce [1]:

- 20 kg of fresh **BSG**
- 2–4 kg of **BSY**

⇨ Millions of tons of by-products per year
Available at low or no cost
Represent an ecological and economical problem

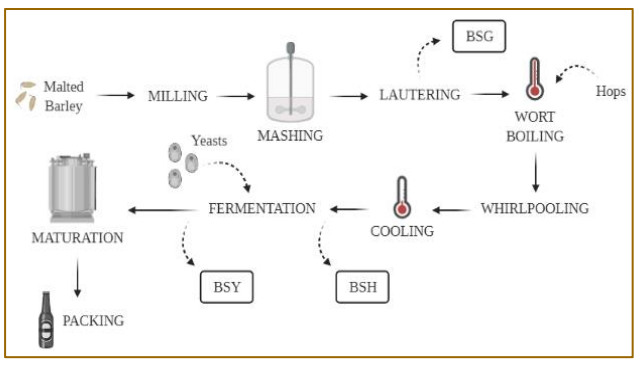


Figure 1: Brewing process [2]

BSG and **BSY** are rich in **proteins** and **fibre**

BSG: 19–30% dry weight [2]

BSY: 35–60% dry weight [2] with a high proportion of essential amino acids [3]

contain (1-3)-β-d-glucans (major structural component of cell walls), (1-3)(1-4)-β-d-Glucan (BSG) and (1-3)(1-6)-β-d-glucan (BSY) [4] ⇨ ↓ glucose
↓ choresterol abd TG
↑ imune system
etc

their use as food ingredient is **approved by EFSA**

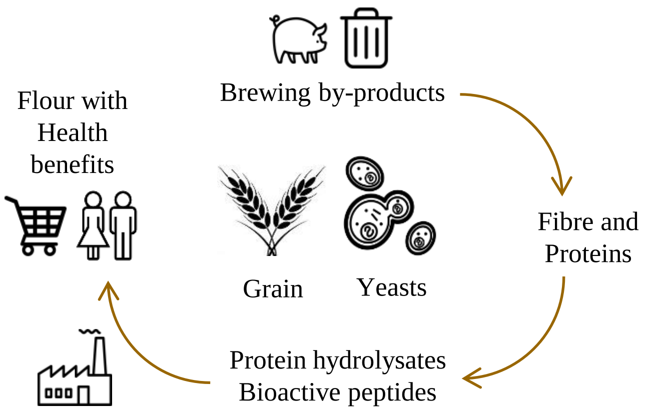
Can be hydrolysed with BSY proteolytic enzymes to produce diversified peptides presenting numerous functional activities (bioactive peptides) that enhance health, reduce the risk or ameliorate several pathological conditions [2].

⇨ Can offer additional health benefits beyond nutrition:

- Antioxidants
- ↓ Blood pressure

How to take advantage of these benefits?
Why not use to produce innovative and sustainable healthy “flours” for food industry?

The **goal** of this study is the production and characterization of the protein rich fraction from **BSY** and **BSG** targeting their use as bioactive food ingredients in flours to prevent hypertension (the main modifiable risk factor with highest prevalence and mortality worldwide)



METHODS and RESULTS

1. Extraction of proteins from brewing by-products supplied by a local beer industry: BSG (alkaline method [5]) and BSY (mechanical disruption of the cell wall [6])

2. Hydrolysis of BSG proteins (E/S 10:100 v/v at 50°C for 6h, pH 6 [5]; 15,34 mg of protein/mL*) and autolysis of BSY proteins (at 60°C for 6h, pH 6 [6]; 10 mg of protein/mL*) using BSY proteases to obtain bioactive peptides

   *BCA Protein Assay kit 23225, ThermoFisher (triplicates)

Table 1: BSG and BSY protein content, extraction and hydrolysis/autolysis

BSG		BSY		
Total initial Protein (%/dry weight)[1]	Peptides after hydrolysis (g)[2]	Total initial Protein (%/dry weight)[1]	Proteases activity (U/mL)[3]	Peptides after autolysis (g)[4]
26,90±1,51	21,14666	33,47±2,46	1,564	17,11799

[1]kjeldahl method [7] (n≥4); [2]Hydrolysis of 370mL BSG proteins; [3]Cupp-Enyard (2008) non-specific proteases assay (triplicates); [4]Autolysis of 510mL BSY proteins

DISCUSSION

✓ We were able to **recover proteins** from brewing industry waste (**BSG** and **BSY**), including viable BSY proteases that were used to produce **bioactive peptides**, thus following a **sustainable process**.

✓ The hypotensive effect of these peptides (after simulation of oral digestion) is being studied *in vitro* using vascular cells and tissues to support its effective bioactivity.

✓ After proving the anti-hypertensive potential of BSG and BSY peptides, they will be tested as new **functional ingredients** in the production of **innovative healthy “flours”** in the fight against hypertension.

✓ If succeeded, this process will be adapted to an efficient and cost-effective industrial scale, promoting **circular economy** with social and environmental benefits.

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