Foods 2021

Proceedings

Characterization of Dietary Fiber Extracts from Corn (*Zea mays* L.) and Cooked Common Bean (*Phaseolus vulgaris* L.) Flours and Evaluation of Their Inhibitory Potential against Enzymes Associated to Glucose and Lipids Metabolism In Vitro †

Amanda B. Serna-Perez [1], Guadalupe Loarca-Piña [1,*] and Ivan Luzardo-Ocampo [1,2,*]

[1] Research and Graduate Program in Food Science, School of Chemistry, Universidad Autonoma de Queretaro, Querétaro, Qro., Mexico; sernaamanda2@gmail.com
[2] Instituto de Neurobiología, Universidad Nacional Autónoma de Mexico (UNAM)-Campus Juriquilla, Juriquilla, Qro., Mexico
* Correspondence: loarca@uaq.mx (G.L.-P.); ivan.8907@gmail.com (I.L.-O.)
† Presented at the 2nd International Electronic Conference on Foods, 15–30 October 2021; Available online: https://foods2021.sciforum.net/.

Citation: Serna-Perez, A.B.; Loarca-Piña, G.; Luzardo-Ocampo, I. Characterization of Dietary Fiber Extracts from corn (*Zea mays* L.) and Cooked Common Bean (*Phaseolus vulgaris* L.) Flours and Evaluation of Their Inhibitory Potential against Enzymes Associated to Glucose and Lipids Metabolism In Vitro. *Proceedings* **2021**, *1*, x. https://doi.org/10.3390/xxxxx

Published: 15 October 2021

Abstract: This research aimed to characterize dietary fiber (DF)-aqueous extracts from corn (commercial and Northwestern White Population) and common beans (cv. Bayo Madero) flour blends, evaluating their inhibitory effect on enzymes involved in glucose/lipid metabolism in vitro. Insoluble fiber showed the highest content of total phenolics, being chlorogenic and ellagic acids the main identified phenolics. Soluble fiber displayed the α-glucosidase and pancreatic lipase inhibitions (20-25%). Results suggested that corn/beans DF is a functional ingredient to potentially alleviate obesity and type II diabetes.

Keywords: Corn (*Zea mays* L.); common bean (*Phaseolus vulgaris* L.); α-amylase; α-glucosidase; antioxidant capacity; dietary fiber; nixtamalization; phenolic compounds; pancreatic lipase

1. Introduction

Inadequate dietary patterns lead to the development of chronic non-communicable diseases, where obesity and type II diabetes occupy a relevant place in Mexico [1]. Since both conditions are associated with glucose and lipid metabolism, targeting these pathways and participating enzymes might be a proper way to prevent and alleviate their development [2].

It has been reported that bioactive compounds from cereals such as corn (*Zea mays* L.) and legumes such as common bean (*Phaseolus vulgaris* L.) flours could target some of the enzymes involved in glucose and lipids metabolism [3]. For instance, common bean proteins have been linked to inhibit α-amylase and improve insulin resistance *in vivo*, decreasing postprandial glucose and glycosylated hemoglobin. As antilipidemic agents, common beans have been associated to body weight regulation, lipid-lowering effects, and appetite and satiety control, mainly for their content of phenolic compounds and dietary fiber [4].

Since common beans and corn are usually consumed together in Latin America, providing bioactive compounds-rich ingredients based on these raw materials could deliver nutritionally rich, hypolipidemic, and glucose-lowering effects. The biological properties from food products combining corn and beans have demonstrated their ability to provide benefits against obesity and type II diabetes [5]. Previously, our research group has characterized the anti-lipidemic [5], anti-inflammatory [6,7], and chemoprotective effect [8] of whole nixtamalized Northwestern White Population corn and Bayo Madero beans. However, most of their bioactive compounds are retained in the non-digestible fraction, mainly composed by dietary fiber and phenolic compounds [9]. As these matrices have shown potential for the elaboration of corn/bean snacks [9] and tortillas [10], dietary fiber extracts from these materials could be used as a functional ingredient with technological potential. Hence, this research aimed to obtain and characterize the antioxidant dietary fiber extract from blends of nixtamalized corn (*Zea mays* L.) and cooked common bean (*Phaseolus vulgaris* L.) flours and evaluate their inhibitory potential on enzymes associated to glucose and lipids metabolism in vitro.

2. Materials and Methods

2.1. Biological Material and Preparation of the Flours

Commercial (A: MASECA brand flour, Mexico) and Northwestern population white (B) corn (*Zea mays* L.) flours, and common beans (C) (*Phaseolus vulgaris* L., cv. Bayo Madero) seeds were used for the preparation of flours. Corn flours were subjected to alkaline cooking or nixtamalization as previously reported using commercial lime ($Ca(OH)_2$). The resulting nixtamalized corn was milled, dehydrated, and screened through a 0.85 mm mesh [9]. For the common bean flours, beans were cooked for 2.5 h with an excess of water (1:4 common beans:water proportion), dehydrated (40 °C, 16 h), ground, and screened (0.425 mm mesh) [9]. The corn and common beans flours were blended in 70:30 and 80:20 proportions of corn and common beans flours, resulting in four experimental mixtures: 7030AC, 7030BC, 8020AC, and 8020BC.

2.2. Aqueous Extraction of Dietary Fiber (DF)

The aqueous extraction was conducted following the procedure of Benitez et al. [11] following the traditional dietary fiber extraction from the official methods of analysis of AOAC, without using enzymes. Briefly, the samples (10 g) were mixed with deionized water (40 mL) and incubated (60 °C, 60 min; 100 °C, 30 min). The water-insoluble residues (the insoluble dietary fiber or IDF) were washed with distilled water at 60 °C and ethanol (80% *v*/*v*), filtered (Whatman No. 4), and dried (60 °C, 24 h). An equal volume of ethanol (80% *v*/*v*) was added to the filtrate and was left for 12 h. Then, two ethanol (80% *v*/*v*) washes were conducted, followed by acetone and filtration (Whatman No. 42) (soluble dietary fiber of SDF).

2.3. Extraction and Quantification of Total Phenolic Compounds and Individual Phenolics from DF Fractions

Methanolic extracts from raw materials and extracted fibers were prepared as previously reported [9]. Total phenolic compounds were determined using the Folin-Ciocalteu method [12]. A high-performance liquid chromatography coupled to diode array detection (HPLC-DAD) method was used [13] to dected and quantify individual phenolic compounds. An Agilent 1100 System (Agilent Technologies, Palo Alto, CA, US) was used, and the phenolic compounds were separated in a Zorbax Eclipse XDB-C18 column (Agilent Technologies) at a flow rate of 1 mL/min, 35 ± 0.6 °C, and using standard curves from HPLC-grade standards of caffeic, chlorogenic, ellagic, ferulic, gallic; and flavonoids such as (+)-catechin and epigallocatechin gallate. Results were reported in micrograms equivalents of each phenolic compound/g dry sample.

2.4. Inhibition Potential of DF Fractions against Enzymes Linked to Glucose and Lipids Metabolism

The screening of the DF ability to inhibit enzymes linked to glucose and lipids metabolism was tested based on the residual α-amylase activity [11], α-glucosidase inhibition [14], and pancreatic lipase inhibition [11].

2.5. Statistical Analysis

The results were expressed as the means ± SD of at least two independent experiments in triplicates. An ANOVA analysis was conducted followed by a post-hoc Tukey-Kramer's test, establishing the significance at $p < 0.05$ using the JMP v. 16.0 software (SAS Institute).

3. Results and Discussion

3.1. Dietary Fiber Contents from Raw Materials and Blends

Total dietary fiber (DF) was extracted using an aqueous method from corn and common beans flours and their blends. Northwestern White population (B) corn presented the highest total dietary fiber content, followed by the commercial corn (A) and the cooked common bean flour (C) (10.65–15.76%). The 7030BC and 8020BC mixtures showed the highest fiber content, which is explained due to the higher fiber yields from their raw materials (16.30–16.67%). Despite the obtained fiber contents, values were significantly lower than reported DF values for the same corn and common beans varieties using the traditional enzymatic method [10,13]. However, IDF values (raw materials: 10.40–12.77%, blends: 10.63–15.04%) were higher ($p < 0.05$) than enzymatically-extract fibers, agreeing with previous reports using this extraction method [11].

3.2. Total Phenolic Compounds of DF Fractions

Depending on the sample was the raw materials (RM) or the fiber fractions (IDF and SDF), the content of total phenolics significantly varied ($p < 0.05$) between samples (Figure 1A). Total phenolics associated with SDF displayed the highest content in RM, while those from the fiber fractions in the blends exhibited the highest values.

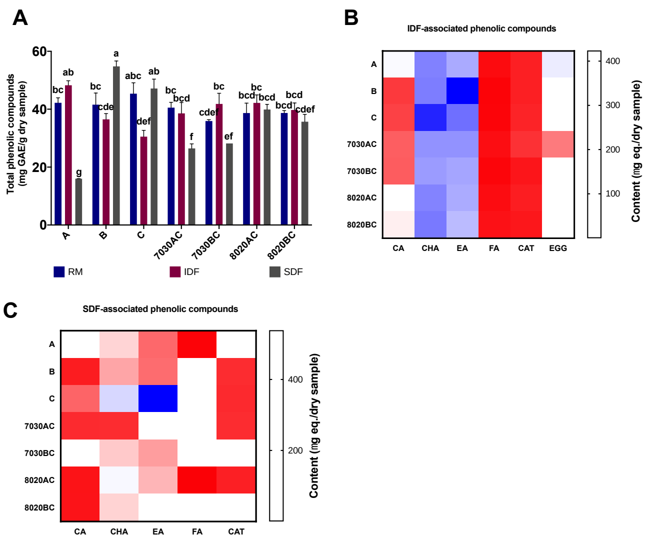

Figure 1. Content of total phenolic compounds (**A**); and individual phenolics detected from IDF (**B**) and SDF (**C**) by HPLC-DAD. The results are expressed as means ± SD of two independent experiments in triplicates. Different letters express significant differences ($p < 0.05$) by Tukey-Kramer's test. A: Nixtamalized Northwestern White Population corn flour; B: Commercial (MASECA®) nixtamalized corn; C: cooked common bean flour; CA: caffeic acid; CHA: chlorogenic acid; EA: ellagic

acid; FA: ferulic acid; CAT: (+)-catechin; EGG: epigallocatechin gallate; GAE: gallic acid equivalents; IDF: insoluble dietary fiber; RM: raw materials; SDF: soluble dietary fiber.

Results obtained for the raw samples agreed with previous reports of total phenolics for the same food matrices [15]. An overall higher amount of individual phenolics were presented in IDF (Figure 1B), which could explain the higher proportion of IDF in RM, while SDF generally accounts for 1/3 of the DF [16]. Moreover, the richness of phenolics to IDF and SDF (Figure 1C) is explained due to aromatic rings and hydrophilic groups from phenolic compounds forming strong binding to polysaccharides and proteins from cell walls of food matrices, difficulting its extraction [17]. IDF fractions mainly contained chlorogenic and ellagic acids (Figure 1B), while SDF (Figure 1C) primarily showed caffeic and ellagic acid as the most abundant phenolics. It has been reported that ferulic and gallic acid are the major free phenolic compounds from corn, while chlorogenic, p-coumaric, (+)-catechin, and rutin are the major bound phenolic compounds, but nixtamalization has a negative effect in ferulic acid, which explains its low content in the fiber fractions [18]. The concentrations of phenolics for the RM mixtures agrees with a previous report from our research group [9]. However, reports indicate that ferulic, isoferulic, vanillic, syringic, and p-coumaric acids are the primary phenolics from corn IDF. In contrast, hydroxybenzoic acid derivatives are the major phenolics from IDF and SDF fractions of cooked common beans [19], where the differences could be explained by the thermal process and the agronomic variety.

3.3. Inhibitory Activity of DF Fractions against Glucose and Lipid Metabolism Enzymes In Vitro

There were no differences in the α-amylase inhibition for both fiber fractions, but SDF fractions showed overall higher values of α-glucosidase and pancreatic lipase inhibitions (Table 1). Since it has been informed that phytochemicals exhibit a low α-amylase inhibition but strong α-glucosidase inhibition, an useful property that could be used to reduce postprandial glycemia with minimum adverse effects [20]. Although the observed results for α-amylase inhibition were lower than those reported for raw corn and common beans (20–85% and 55%, respectively) [20], our results consider cooked matrices to be used as ingredients, indicating a more realistic effect form matrices in the way they are commonly consumed.

Table 1. Inhibitory activity of IDF and SDF fractions over α-amylase, α-glucosidase, and pancreatic lipase (%).

Sample	α-Amylase Inhibition (%)	α-Glucosidase Inhibition (%)	Lipase Inhibition (%)
		IDF	
A	22.0 ± 5.1 ^{a}	48.7 ± 8.0 ^{cde}	41.6 ± 1.5 ^{e}
B	18.7 ± 1.0 ^{a}	54.0 ± 9.1 ^{bcde}	42.4 ± 3.3 ^{e}
C	20.9 ± 1.2 ^{a}	74.1 ± 3.6 ^{ab}	43.2 ± 3.0 ^{de}
7030AC	18.3 ± 1.4 ^{a}	57.1 ± 3.6 ^{bcd}	42.6 ± 4.5
7030BC	17.5 ± 3.1 ^{a}	48.0 ± 3.5 ^{cde}	55.8 ± 3.0 ^{abcd}
8020AC	18.9 ± 2.3 ^{a}	61.3 ± 7.3 ^{abcd}	57.9 ± 7.4 ^{ab}
8020BC	15.0 ± 2.3 ^{a}	41.9 ± 1.5 ^{def}	43.7 ± 0.7 ^{cde}
		SDF	
A	21.4 ± 1.9 ^{a}	80.2 ± 0.9 ^{a}	46.8 ± 2.2 ^{bcde}
B	16.1 ± 2.6 ^{a}	21.5 ± 2.4 ^{f}	41.8 ± 1.1 ^{e}
C	21.7 ± 0.7 ^{a}	50.1 ± 1.8 ^{cde}	46.6 ± 0.4 ^{bcde}
7030AC	20.7 ± 0.1 ^{a}	69.3 ± 5.7 ^{abc}	56.3 ± 0.7 ^{abc}
7030BC	19.6 ± 0.8 ^{a}	44.9 ± 3.3 ^{de}	60.8 ± 0.4 ^{a}
8020AC	14.6 ± 1.6 ^{a}	35.2 ± 4.8 ^{ef}	56.6 ± 4.1 ^{ab}
8020BC	16.4 ± 0.6 ^{a}	50.2 ± 9.4 ^{cde}	59.7 ± 3.3 ^{a}

The results are expressed as mean ± SD of two independent experiments in triplicates. Different letters express significant differences ($p < 0.05$) by Tukey-Kramer's test.

Values for α-glucosidase inhibition were higher than those informed for raw matrices (25–45%) [21], while there are no reports for specific fiber fractions from nixtamalized corn and cooked common beans. To our knowledge, there are no reports of pancreatic lipase inhibition from thermal-treated corn and common beans. Nonetheless, the importance of these food products inhibiting this enzyme suggest an important mechanism avoiding the related production of free fatty acids that could be easily absorbable at the small intestine, contributing to obesity development. Dietary fibers have been largely associated with lipase inhibition due to viscosity increase in the stomach at low pH or the ability of pectin components to protonate active serine and histidine residues from lipase, inactivating the enzyme [22].

4. Conclusions

The results suggested the feasibility of using aqueous fiber extracts from nixtamalized corn and cooked common bean as functional ingredients delivering phenolic compounds with the ability to inhibiting critical enzymes linked to glucose and lipids metabolism. Since no enzymes are used in fiber extraction, these fiber-rich ingredients could be achieved at a low-cost manufacturing process. Due to the biological potential from these extracts, further research exploring their technological properties and experiments at in vitro and in vivo levels are justified.

Author Contributions: The following statements should be used Conceptualization, I. A. L., and G.L.-P.; methodology, I.L.-O.; software, I.L.-O. and A.B.S.-P.; validation, I.L.-O., and G.L.-P.; formal analysis, I.L.-O., A.B.S.-P., and G.L.-P.; investigation, I.L.-O. and A.B.S.-P.; resources, G.L.-P.; data curation, A.B.S.-P. and I.L.-O..; writing—original draft preparation, I.L.-O. and A.B.S.-P. ; writing—review and editing, I.L.-O. and G.L.-P.; visualization, I.L.-O. and G.L.-P.; supervision, I.L.-O. and G.L.-P.; project administration, I.L.-O. and G.L.-P.; funding acquisition, I.L.-O. and G.L.-P. All authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by Fondos de Proyectos Especiales de Rectoria (FOPER) from Univrsidad Autonoma de Queretaro (grant number: FOPER-2020-FQU02143). Author Ivan Luzardo-Ocampo was supported by Consejo Nacional de Ciencia y Tecnología (CONACyT-Mexico) under the "Ayudante SNI III" Funds (grant number: 19621).

Institutional Review Board Statement: Not Applicable.

Informed Consent Statement: Not Applicable.

Data Availability Statement: Data will be available upon reasonable request.

Acknowledgments: The authors acknowledge M. Sc. David Gustavo-Gutierrez and Dr. Ruben A. Romo-Mancillas for their technical support.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Torres, F.; Rojas, A. Obesidad y salud pública en México: Transformación del patrón hegemónico de oferta-demanda de alimentos. *Probl. Desarro. Rev. Latinoam. Econ.* **2018**, *49*, 145–169, doi:10.22201/iiec.20078951e.2018.193.63185.
2. Panigrahy, S.K.; Bhatt, R.; Kumar, A. Targeting type II diabetes with plant terpenes: The new and promising antidiabetic therapeutics. *Biologia (Bratisl)* **2021**, *76*, 241–254, doi:10.2478/s11756-020-00575-y.
3. Campos-Vega, R.; Oomah, B.; Loarca-Piña, G.; Vergara-Castañeda, H. Common beans and their non-digestible fraction: Cancer inhibitory activity—An overview. *Foods* **2013**, *2*, 374–392, doi:10.3390/foods2030374.
4. Loarca-Piña, G.; Luna-Vital, D.; Mojica, L.; Ramírez-Jiménez, A.K.; Luzardo-Ocampo, I. Health benefits of beans. In *Phaseolus vulgaris: Cultivars, Production and Uses*; 2018; pp. 201–227.
5. Domínguez-Uscanga, A.; Loarca-Piña, G.; Gonzalez de Mejia, E.; Dominguez-Uscanga, A.; Loarca-Piña, G.; Gonzalez de Mejia, E. Baked corn (*Zea mays* L.) and bean (*Phaseolus vulgaris* L.) snack consumption lowered serum lipids and differentiated liver gene expression in C57BL/6 mice fed a high-fat diet by inhibiting PPARγ and SREBF2. *J. Nutr. Biochem.* **2017**, *50*, 1–15, doi:10.1016/j.jnutbio.2017.08.011.
6. Luzardo-Ocampo, I.; Campos-Vega, R.; Cuellar-Nuñez, M.L.L.; Vázquez-Landaverde, P.A.A.; Mojica, L.; Acosta-Gallegos, J.A.A.; Loarca-Piña, G. Fermented non-digestible fraction from combined nixtamalized corn (*Zea mays* L.)/cooked common bean

(*Phaseolus vulgaris* L.) chips modulate anti-inflammatory markers on RAW 264.7 macrophages. *Food Chem.* **2018**, *259*, 7–17, doi:10.1016/j.foodchem.2018.03.096.
7. Luzardo-Ocampo, I.; Campos-Vega, R.; Gonzalez de Mejia, E.; Loarca-Piña, G. Consumption of a baked corn and bean snack reduced chronic colitis inflammation in CD-1 mice via downregulation of IL-1 receptor, TLR, and TNF-α associated pathways. *Food Res. Int.* **2020**, *132*, 109097, doi:10.1016/j.foodres.2020.109097.
8. Vergara-Castañeda, H.A.; Guevara-González, R.G.; Ramos-Gómez, M.; Reynoso-Camacho, R.; Guzmán-Maldonado, H.; Feregrino-Pérez, A.A.; Oomah, B.D.; Loarca-Piña, G. Non-digestible fraction of cooked bean (*Phaseolus vulgaris* L.) cultivar Bayo Madero suppresses colonic aberrant crypt foci in azoxymethane-induced rats. *Food Funct.* **2010**, *1*, 294–300, doi:10.1039/c0fo00130a.
9. Luzardo-Ocampo, I.; Campos-Vega, R.; Gaytán-Martínez, M.; Preciado-Ortiz, R.; Mendoza, S.; Loarca-Piña, G.; Preciado-Ortíz, R.; Mendoza, S.; Loarca-Piña, G. Bioaccessibility and antioxidant activity of free phenolic compounds and oligosaccharides from corn (*Zea mays* L.) and common bean (*Phaseolus vulgaris* L.) chips during in vitro gastrointestinal digestion and simulated colonic fermentation. *Food Res. Int.* **2017**, *100*, 304–311, doi:10.1016/j.ecss.2007.10.007.
10. Treviño-Mejía, D.; Luna-Vital, D.A.; Gaytán-Martínez, M.; Mendoza, S.; Loarca-Piña, G. Fortification of commercial nixtamalized maize (*Zea mays* L.) with common bean (*Phaseolus vulgaris* L.) increased the nutritional and nutraceutical content of tortillas without modifying sensory properties. *J. Food Qual.* **2016**, *39*, 569–579, doi:10.1111/jfq.12251.
11. Benitez, V.; Rebollo-Hernanz, M.; Hernanz, S.; Chantres, S.; Aguilera, Y.; Martin-Cabrejas, M.A. Coffee parchment as a new dietary fiber ingredient: Functional and physiological characterization. *Food Res. Int.* **2019**, *122*, 105–113, doi:10.1016/j.foodres.2019.04.002.
12. Singleton, V.L.; Orthofer, R.; Lamuela-Raventós, R.M. Analysis of total phenols and other oxidation substrates and antioxidants by means of folin-ciocalteu reagent. *Methods Enzymol.* **1999**, *299*, 152–178, doi:10.1016/S0076-6879(99)99017-1.
13. Ramírez-Jiménez, A.K.; Reynoso-Camacho, R.; Mendoza-Díaz, S.; Loarca-Piña, G. Functional and technological potential of dehydrated Phaseolus vulgaris L . flours. *Food Chem.* **2014**, *161*, 254–260, doi:10.1016/j.foodchem.2014.04.008.
14. Johnson, M.H.; Lucius, A.; Meyer, T.; Gonzalez de Mejia, E. Cultivar evaluation and effect of fermentation on antioxidant capacity and in vitro inhibition of α-amylase and α-glucosidase by highbush blueberry (*Vaccinium corymbosum*). *J. Agric. Food Chem.* **2011**, *59*, 8923–8930, doi:10.1021/jf201720z.
15. Rodríguez-Salinas, P.A.; Zavala-García, F.; Urías-Orona, V.; Muy-Rangel, D.; Heredia, J.B.; Niño-Medina, G. Chromatic, nutritional and nutraceutical properties of pigmented native maize (Zea mays L.) genotypes from the northeast of Mexico. *Arab. J. Sci. Eng.* **2020**, *45*, 95–112, doi:10.1007/s13369-019-04086-0.
16. Bader Ul Ain, H.; Saeed, F.; Ahmed, A.; Asif Khan, M.; Niaz, B.; Tufail, T. Improving the physicochemical properties of partially enhanced soluble dietary fiber through innovative techniques: A coherent review. *J. Food Process. Preserv.* **2019**, *43*, 1–12, doi:10.1111/jfpp.13917.
17. Pérez-Jiménez, J.; Serrano, J.; Tabernero, M.; Arranz, S.; Díaz-Rubio, M.E.; García-Diz, L.; Goñi, I.; Saura-Calixto, F. Bioavailability of phenolic antioxidants associated with dietary fiber: Plasma antioxidant capacity after acute and long-term intake in humans. *Plant Foods Hum. Nutr.* **2009**, *64*, 102–107, doi:10.1007/s11130-009-0110-7.
18. Ramírez-Jiménez, A.K.; Rangel-Hernández, J.; Morales-Sánchez, E.; Loarca-Piña, G.; Gaytán-Martínez, M. Changes on the phytochemicals profile of instant corn flours obtained by traditional nixtamalization and ohmic heating process. *Food Chem.* **2019**, *276*, 57–62, doi:10.1016/j.foodchem.2018.09.166.
19. Dueñas, M.; Sarmento, T.; Aguilera, Y.; Benitez, V.; Mollá, E.; Esteban, R.M.; Martín-Cabrejas, M.A. Impact of cooking and germination on phenolic composition and dietary fibre fractions in dark beans (*Phaseolus vulgaris* L.) and lentils (*Lens culinaris* L.). *LWT—Food Sci. Technol.* **2016**, *66*, 72–78, doi:10.1016/j.lwt.2015.10.025.
20. Apostolidis, E.; Kwon, Y.-I.; Ghaedian, R.; Shetty, K. Fermentation of milk and soymilk by Lactobacillus bulgaricus and Lactobacillus acidophilus enhances functionality for potential dietary management of hyperglycemia and hypertension. *Food Biotechnol.* **2007**, *21*, 217–236, doi:10.1080/08905430701534032.
21. Karimi, A.; Azizi, M.H.; Ahmadi Gavlighi, H. Frationation of hydrolysate from corn germ protein by ultrafiltration: In vitro antidiabetic and antioxidant activity. *Food Sci. Nutr.* **2020**, *8*, 2395–2405, doi:10.1002/fsn3.1529.
22. Houghton, D.; Wilcox, M.D.; Chater, P.I.; Brownlee, I.A.; Seal, C.J.; Pearson, J.P. Biological activity of alginate and its effect on pancreatic lipase inhibition as a potential treatment for obesity. *Food Hydrocoll.* **2015**, *49*, 18–24, doi:10.1016/j.foodhyd.2015.02.019.