Membrane filtration for the recovery of polyphenols from distillery stillage

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Although the distillery industry makes a significant contribution to the development of the world economy, it contributes significantly to environmental pollution. For every 1 liter of alcohol produced, up to 15 liters of by-products (waste) are generated, known as distillery stillage. Due to its low pH, high content of organic compounds and nitrogen, and dark brown color, stillage poses a major challenge to the distillery industry (Fito *et al*, 2019). Environmental regulations and a greater understanding of the negative impacts of waste have made it necessary to manage the waste generated in the distillery industry in accordance with the principles of a circular economy. In line with the waste valorization approach, the recovery of value-added compounds with commercial potential from waste is a promising concept to reduce pollution and promote the economic competitiveness of the industry.

Cereal-based distillery stillage is a source of polyphenols (including phenolic acids and flavonoids), which are bioactive compounds with potential health benefits (Mikucka *et al*, 2022). Phenolic acids are derivatives of benzoic acids or cinnamic acids; they differ in the number and position of hydroxylated and methoxylated substituents on their aromatic rings, which determines the bioactive nature of phenolic acids and their antioxidant properties (Valanciene *et al*, 2020). Due to these properties, phenolic acids can be used in the pharmaceutical, cosmetic, and food industries, replacing synthetic antioxidants.

In the search for an effective method of recovering polyphenols from waste materials that provides high recovery yields and preserves the antioxidant activity of the recovered product, membrane technologies offer many advantages as alternative methods for the most common extractions. These advantages include high efficiency, simple equipment, low energy consumption, high selectivity, rapid separation, ease of combining membrane processes with conventional methods, ease of scaling, and low recovery costs (Cassano *et al* 2008). In addition, operation of the membranes at mild temperatures can preserve the biological activity of the recovered compounds. The disadvantages of membrane filtration are the low chemical and mechanical strength of the membranes and the lower efficiency caused by fouling. However, fouling of membranes can improve the separation of compounds by size exclusion, reducing the effective pore size.

Although the recovery of polyphenols from waste materials is an innovative approach to waste valorization, reports on the use of membranes in the separation of polyphenols from distillery stillage are still limited. The objective of this study was to investigate the possibility of using membrane pressure technology to recover polyphenols from distillery stillage and distillery stillage extracts. The effect of membrane pore size on total polyphenol content (TPC), total flavonoid content (TFC), phenolic acid species and contents, and antioxidant activity of the recovered compounds was investigated.

This study used distillery stillage produced during the production of concentrated raw ethyl alcohol from cereals, mainly wheat and rye. The distillery stillage had the following composition: 47,000±5,300 mg COD/L, 4345±5 mg N/L, 280±2 mg P/L, and 789±3 mg CH_3COOH/L.

Membrane filtration was performed in two variants. In variant 1, distillery stillage supernatant (from stillage centrifugation at 8,000 rpm for 10 min) was filtered directly. In variant 2, membrane filtration was used to separate polyphenols from extracts formed during microwave-assisted extraction (MAE) of stillage using ethanol as solvent. Membranes ((Amicon Ultra-15 Centrifugal Filter Devices, MERCK) characterized by *cut-offs* of 100, 30, 10, and 3 kDa were used for filtration. To obtain permeates, the filter devices were centrifuged at 5,000 rpm for 40 min.

TPC was measured based on Singletion *et al* (1999) with modifications. TFC was measured based on Quettier-Deleu *et al* (2000) with modifications. The content of total and free phenolic acids was measured by HPLC equipped with a UV-Vis detector (ProStar 325, Varian, Australia) and coupled to a Supelcosil C18 column (150 mm x 4.6 mm, 5 mm) (Sigma-Aldrich, USA). Antioxidant activity was measured by ABTS (2,2-azinobis (3-ethylbenzothiazoline-6-sulfonic acid)), DPPH (2,2-diphenyl-1-picrylhydrazyl) and FRAP (iron-reducing antioxidant activity) assays (Re *et al* (1999), Moure *et al* (2001), Benzie and Strain (1996)).

The highest TPC (4.57 mg GAE (gallic acid equivalent)/g d.m. (dry mass)) and TFC (1.08 mg QUE (quercetine equivalent)/g d.m.) were obtained in permeates from a membrane with a *cut-off* of 100 kDa (Figures 1-2). Reducing the pore size of the membrane to 3 kDa resulted in a decrease in TPC and TFC in permeates and an increase in retentates.

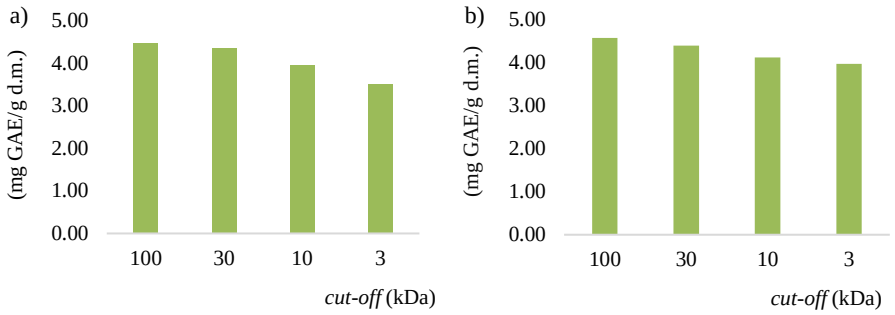

Fig. 1. TPC in permeates after membrane filtration (variant 1) (a), and in permeates after MAE and membrane filtration (variant 2) (b)

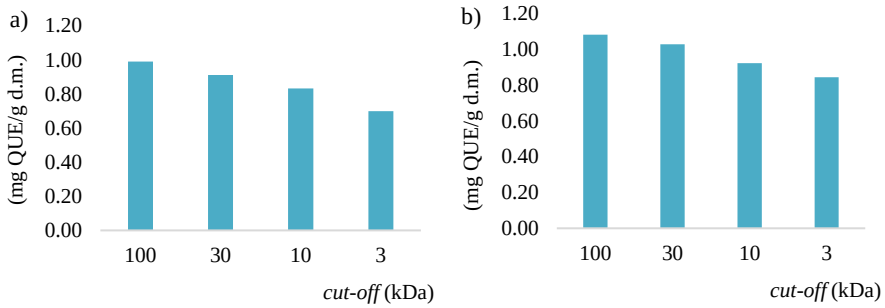

Fig. 2. TFC in permeates after membrane filtration (variant 1) (a), and in permeates after MAE and membrane filtration (variant 2) (b)

In permeates obtained with a membrane of 100 kDa, total phenolic acid content (3.86 µg/g d.m.) and antioxidant activity (DPPH, FRAP and ABTS 5.1 µmol/g d.m., 3.4 µmol/g d.m. and 25.1 µmol/g d.m., respectively) were also the highest. Antioxidant activity was positively correlated with phenolic acid content. Hydroxybenzoic acids (p-OH benzoic, vanillic, and syringic) and hydroxycinnamic acids (p-coumaric, ferulic and sinapic) were detected in permeates. Ferulic acid accounted for the largest proportion (42%) of the total phenolic acid content.

The study proves that distillery stillage is a source of valuable polyphenols. Membrane filtration can be used to concentrate polyphenols, and the method of recovery from retentates should be developed to improve the yield.

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