Screw press extraction and ultrafiltration of flavonoids from kalanchoe leaves and stems

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Introduction

In horticulture potted flowering kalanchoe plants and cut flowers are a major product produced and handled world-wide. This production has a great business and circular economic potential, as the waste leaves and cut stems from this production are rich in alkaloids, flavonoids, steroids and terpenes, some of which have potential as anti-inflammatories, anti-microbials and anti-allergens, which could find use in medicine and cosmetics. Kahalche has previously been part of traditional natural medicine in the Americas and West Africa (Milad et al., 2014). Previous studies have shown (Rosli et al., 2012), that flavonoids and phenolic compounds can be extracted in aqueous solutions from kalanchoe tubiflora. Further, flavonoids can be separated using membrane processes such as ultrafiltration (Milani et al., 2015) and nanofiltration (Dzhonova-Atanasova et al., 2018). In the present study these methods were used to process flavonoids from kalanchoe leaves and stems. The aim was to produce a liquid fraction rich in flavonoids, which through further purification could be used as anti-microbials in the cosmetics industry.

Process Setup

The experimental setup is shown in figure 1. The membranes used are shown in table 1. The leaves and stems are first shredded and extruded into a pulp using a screw press. This is followed by extraction in water at 20 °C. After extraction the larger solids are removed by decantation. The liquid overflow from the decantation still contain fine particulates which are then removed by ultrafiltration at 20 kDa. This should allow the flavonoids to pass in the feed stream.

Table 1. Membranes used for the separation of plant polymers, flavonoids and sugars

<table>
<thead>
<tr>
<th>Membrane</th>
<th>Product and Material</th>
<th>Pore size/Molecular Weight Cut Off</th>
</tr>
</thead>
<tbody>
<tr>
<td>Microfiltration</td>
<td>NID 020 TF N</td>
<td>0.2 μm</td>
</tr>
<tr>
<td>Ultrafiltration</td>
<td>1 kDa</td>
<td>20 kDa</td>
</tr>
<tr>
<td>Ultrafiltration</td>
<td>20 kDa</td>
<td>20 kDa</td>
</tr>
<tr>
<td>Ultrafiltration</td>
<td>1 kDa</td>
<td>1 kDa</td>
</tr>
</tbody>
</table>

Results

The flux behavior during microfiltration and ultrafiltration is shown in figures 2, 3 and 4. As seen from figure 2, the flux shows the typical behavior due to solid fouling of the membrane surface during microfiltration. A fast decline in flux from 130 L·m⁻²·h⁻¹ to 30 L·m⁻²·h⁻¹ as an initial fouling cake layer builds up, followed by a slower flux decline as minor solids adsorb to the cake layer and inside the membrane pores. At a permeate flux of 68 L·m⁻²·h⁻¹ the membrane was cleared. As seen, the original flux could not be re-established, most likely due to particulates absorbed inside the membrane pores. During processing a color change of the microfiltration feed from green towards brown was observed for some of the extraction batches. This change in feed composition did not influence the microfiltration experiments, but it did influence the ultrafiltration flux, as seen in figure 3. The browning of green plant juice is either caused by polyphenol oxidase enzymes that catalyze reactions of phenolic compounds producing quinones, which then polymerize giving rise to melanins, or by Maillard polymerization reactions producing brown pigments. In both cases this leads to brown polymer pigments. The brown feed leads to a lower flux, indicating a much denser fouling layer. This is supported by the fact that the flavonoid retention for the green feed increases slightly from 40% to 44%, while for the brown feed the retention increases from 19% to 82%. The average molecular weight for the flavonoids vary between 282 and 510 Da. Flavonoids should easily permeate through the 20 kDa membrane. Since the flavonoids in both cases are retained by the 20 kDa membranes, the conclusion is that a dense fouling layer have formed. Such dense layers are usually caused by gel layer formation as polymers start to condense close to the membrane surface. The permeate from the 20 kDa ultrafiltration was processed using a 1 kDa membrane. As seen from figure 4 the flux immediately dropped from 130 to 60 L·m⁻²·h⁻¹ followed by a more gradual decline over five hours to 43 L·m⁻²·h⁻¹, after which time the permeate flux was quite stable. As seen the 1 kDa ultrafiltration turned the feed from brown towards green. As the flavonoid fraction is of interest, the ultrafiltration was continued to reach a flavonoid retention of 83%. The average molecular weight for the flavonoids vary between 282 and 510 Da. Flavonoids are a class of polyphenols and have diverse biological properties. In the present study, flavonoids were recovered from kalanchoe leaves and stems. The flavonoid fraction low in polymers, sugars and salts. A possible solution to this problem could be to combine the screw press treatment with an enzyme membrane extraction process. The success of this process depends on the flavonoid fraction low in polymers, sugars and salts. A possible solution to this problem could be to combine the screw press treatment with an enzyme membrane extraction process.

Conclusions

The combination of screw press extraction, microfiltration and ultrafiltration can produce an aqueous liquid flavonoid extract. However, the membrane separation sequence is not optimal as the fouling layer built-up does not optimally support the separation and concentration of a flavonoid fraction free in polymers, sugars and salts. A possible solution to this problem could be to combine the screw press treatment with an enzyme membrane extraction process. This combination could produce a pure flavonoid concentrate and a more stable fouling situation.