Evaluation of the effect of ultrasonic frequency in the extraction of phenolic compounds of pomegranate (Punica granatum L.)

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Abstract
Nowadays, studies are in progress to elucidate the role of the components of pomegranate on human health due to the high biological potential present in its different parts, such as phenolic compounds. The study sought to obtain extracts from pomegranate peel waste using the technique of Ultrasonic Assisted Extraction (UAE), and study the effect of different extractors solvents and different frequencies of ultrasonic.

Key words: Extraction, Ultrasonic, Phenolic compounds.

Introduction
Pomegranate consumption (Punica granatum L.) is associated with possible beneficial health effects due to numerous chemical compounds of high biological potential, such as phenolic compounds present in its different parts. Phenolic compounds of pomegranate have influence on various biological factors such as the modulation of anti-inflammatory responses¹ and the endogenous antioxidant defense system enzymes². To obtain the extracts is necessary to separate the undesirable components using a solvent and using techniques of Ultrasonic Assisted Extraction, which has stood out for its high efficiency and low cost. The ultrasonic technique is based on the formation of longitudinal ultrasonic waves able to cause the cavitation phenomenon due to the formation of expansion and compression regions. The accumulation of energy due to compression and expansion cycles causes the collapse of the bubble which a resulting shock wave crosses the solvent destroying the walls of the matrix cells that release large amounts of intracellular contents to the environment, increasing compounds transfer³. The aim of the project was to study the effect of different ultrasonic frequencies in the efficiency of extraction of phenolic compounds in pomegranate fruit in order to maximize the extraction efficiency and increase the concentration of phenolic compounds and antioxidants in the extracts.

Results and Discussion
The Ultrasonic Assisted Extraction was performed from 1 gram of raw material (pomegranate peel) with 25 mL of solvent, at temperatures of 40 °C, 50 °C, 60 °C and 70 °C, and using the operation frequency of 37kHz, 80 kHz or the combination of these two frequencies. By analyzing the extracts obtained by solvent 70% ethanol, it was observed that the extraction of phenolic compounds has increased significantly when operated in the sweep condition at 80 kHz and temperature of 50°C. The analysis of antioxidant activity (DPPH) was possible to see better results in the extracts obtained in degas condition at 80 kHz and 60 °C. Further the efficiency was better when using the operation frequency of 37kHz at 60 °C. It was also observed that the extraction with water, 30% ethanol and 50% ethanol as extraction solvents, the results of total phenolic content, antioxidant activity and efficiency was lower compared to solvent extractor 70% ethanol. The results of the analysis of the best conditions are in Table 1.

Table 1. Results of analysis.

<table>
<thead>
<tr>
<th>Analysis</th>
<th>Results</th>
<th>Frequency</th>
<th>Temperature</th>
<th>Solvent</th>
</tr>
</thead>
<tbody>
<tr>
<td>Xo (%)⁴</td>
<td>59,02</td>
<td>37khz</td>
<td>60 °C</td>
<td>Ethanol 70%</td>
</tr>
<tr>
<td></td>
<td>41,94</td>
<td>37khz (sweep)</td>
<td>50 °C</td>
<td>Water</td>
</tr>
<tr>
<td>FT⁵</td>
<td>274,65</td>
<td>80khz (sweep)</td>
<td>50 °C</td>
<td>Ethanol 70%</td>
</tr>
<tr>
<td></td>
<td>175,84</td>
<td>80khz (sweep)</td>
<td>50 °C</td>
<td>Water</td>
</tr>
<tr>
<td>AA⁶</td>
<td>2796,14</td>
<td>80khz (Degas)</td>
<td>60 °C</td>
<td>Ethanol 70%</td>
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<tr>
<td></td>
<td>1823,28</td>
<td>80khz (sweep)</td>
<td>50 °C</td>
<td>Water</td>
</tr>
</tbody>
</table>

¹: Efficiency expressed in %.
²: Total phenolic content expressed in equivalent mg of gallic acid / g of dry raw material.
³: Antioxidant activity expressed in μmol equivalent of Trolox / g of dry raw material.

Conclusions
Based on these results, it can be concluded that the extraction using solvent ethanol 70%, showed better results of total phenolics, antioxidant activity and efficiency compared with the extracts obtained using as extractor solvent water, 30% ethanol and 50% ethanol.

Acknowledgement
I thank PIBIC / CNPQ for financial support, the advisor Mauricio Ariel Rostagno for the aid and opportunity and doctoral student Ana Paula Machado for their help with the preparation of experiments and analysis of results.


DOI: 10.19146/pibic-2016-51019