

## Microwave assisted acid-hydrolysis of condensed tannins. Application to *Cocos nucifera* condensed tannins.

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### Abstract

Here we demonstrate that the protocol for mDP determination of condensed tannins by acid hydrolysis is improved in reaction time and yield when made under microwave radiation.

### Introdução

*Cocos nucifera* L., the coconut palm, belongs to the Arecaceae (Palmae) family and is widespread on the Brazilian coast. The coconut culture occupies a large territorial extension in Brazil (286.000 ha) producing ca. 3 million tons of fruit (4.5% of the world's production) making Brazil the 4th largest producer of coconut<sup>1</sup>.

The coconut husk fiber (mesocarp) is devoid of economic importance and is usually discarded after fruit consumption. Since 2002, our group has been investigating the pharmacological activities of the tannins from this fiber which are oligomeric and polymeric proanthocyanidins<sup>2</sup>.

In order to evaluate the extension of polymerization and monomer composition of tannins, an assay for determination of the mean degree of polymerization (mDP) is performed. This assay involves acid hydrolysis of the tannin in the presence of a nucleophile. Benzyl mercaptan and phloroglucinol are the most common nucleophilic reagents, but phloroglucinol is odorless and more selective to produce 3,4-trans adducts under lower reaction temperatures<sup>3</sup>. The methodology proposed by Kennedy & Jones (2001) is probably the most used nowadays and takes ca. 20 minutes of reaction time. Here we report the results of our attempted improvement of Kennedy & Jones reaction conditions by using microwave assistance. Factorial design was used to help optimization of time and product yield.

### Resultados e Discussão

Following the protocol created by Kennedy & Jones, 2001, we found that the tannin composition of the coconut husk fiber is catechin (as extension

unit) and catechin and epicatechin (as terminal unit), with a mDP = 5,6.

Figure 1

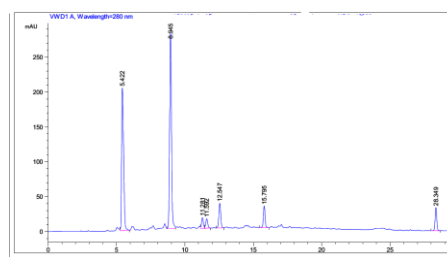


Figure 1 displays the result of a HPLC product analysis of the optimized microwave assisted acid-catalysis cleavage reaction at 100°C for 5 min with phloroglucinol as nucleophile. Legend:  $t_R=5.4$  is phloroglucinol,  $t_R=8.9$  is catechin-phloroglucinol adduct,  $t_R=12.5$  is catechin and  $t_R=15.7$  is epicatechin ( $t_R=28.3$  is an impurity from phloroglucinol). The mDP found under these conditions was the same.

Minitab<sup>®</sup> 17 software was used to design the experimental optimization, using two variables: reaction time (0,5 min as -1 and 5 min as +1), temperature (40°C as -1 and 100°C as +1)

### Conclusões

The acid hydrolysis cleavage for condensed tannin cleavage when made under microwave assistance results in comparable yields but in shorter times than under normal conditions.

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