

# Use of the accuracy profile for the validation of a direct determination of tagitinin C in *Tithonia diversifolia* leaves by on-line coupling of supercritical CO<sub>2</sub> extraction to FT-IR spectroscopy by means of optical fibres.

E. Ziemons<sup>a</sup>, V. Barillaro<sup>b</sup>, E. Rozet<sup>a</sup>, N. Wandji Mbakop<sup>a</sup>, R. Lejeune<sup>a</sup>, L. Angenot<sup>c</sup>,  
L. Thunus<sup>a</sup>, Ph. Hubert<sup>a</sup>

<sup>a</sup> Laboratory of Analytical Chemistry, Bioanalytical Chemistry Research Unit, Department of Pharmacy, University of Liège, Avenue de l'Hôpital 1, B 36, 4000 Liège, Belgium,

<sup>b</sup> Laboratory of Pharmaceutical Technology, Department of Pharmacy, University of Liège, Avenue de l'Hôpital 1, B36, B-4000 Liège, Belgium,

<sup>c</sup> Laboratory of Pharmacognosy, Department of Pharmacy, University of Liège, Avenue de l'Hôpital 1, B36, B-4000 Liège, Belgium.

Supercritical fluid extraction with CO<sub>2</sub> as extraction medium was on-line coupled to a FT-IR spectrometer using a tailor-made high-pressure fibre optic flow cell. This method was optimised and developed for the monitoring in real time and the quantification of dynamic extractions of tagitinin C from *Tithonia diversifolia* leaves.

In order to demonstrate the method ability to allow the direct quantification of tagitinin C in the extract medium the standard addition method was used. The area integration of curves obtained by plotting the absorbance of the highly specific C=O stretching vibration at 1668 cm<sup>-1</sup> versus time (i.e. extractograms) was used as instrumental response.

The SFE/FT-IR process was successfully validated using the accuracy profile as decision tool. On this basis, a linear regression model was chosen for the calibration curve. The method was found to be accurate as the two-sided 95 % beta-expectation tolerance interval did not exceed the acceptance limits of 85 % and 115 % on the analytical range investigated (500 to 2500 µg of added amount of tagitinin C).

The proposed method allowed the non-destructive extraction of tagitinin C and its on-line quantitative determination in less than 25 minutes thus facilitating the subsequent experiments or the pharmacological studies performed on this compound.