



TAXIFOLIN ISOLATED FROM THE SEEDS OF *Mimusops balata*: *in vitro* FORCED DEGRADATION STUDIES BY LC-UV AND LC-ESI-MS AND *in silico* STABILITY PREDICTION

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INTRODUCTION

Taxifolin (Tax) is a dihydroflavonoid with many pharmacological effects could be isolated from *Mimusops balata* seeds. This molecule undergoes degradation by intestinal microflora but has no stability data at alkaline medium. The aim of this study was to predict *in silico* degradation of Tax and to analyze its *in vitro* degradation, monitored by LC-UV and LC-ESI-MS, in alkaline medium.

MATERIAL AND METHODS

The *in silico* computational analysis was performed using Spartan® 08 version 116.2, the torsion angle was 30°, using systematic analysis by the method of functional density theory (DFT), and the radical attack were calculated the Fukui functions. For *in vitro* studies, solutions of Tax were prepared with 1M HCl, 1mM NaOH, 32% H₂O₂, and exposition to UV and visible radiation. Also to 40 °C and 40 °C/75% Relative Humidity. The exposed samples were dissolved in methanol (100 µg/mL) and analyzed by LC-UV with reverse phase and gradient system for elution. Sample prepared with 1.0 M and 0.1 M NaOH were immediately neutralized with HCl and analyzed by LC-ESI-MS.

RESULTS

In silico studies suggested that Tax is more susceptible to nucleophilic attack in C2, C4 and C7 with major values of Fukui function. The smaller BDE values were at C2, C3 and O8, being more susceptible to

autoxidation. *In vitro* studies showed that Tax is more susceptible to alkaline medium, with 16.28% of degradation, after 15 min at 1mM NaOH. Also, Tax showed degradation of 20.25% (30 min, 1M HCl), 11.73% (24h 9.76% H₂O₂), 23.09% (30 days, 40 °C), (30 days, 40 °C/ 75% RH) and 9.00% (2.4 mi lux.h). LC-ESI-MS analysis showed that Tax degraded in 0.1 M NaOH showing four additional peaks, being one with *m/z* 319.0456, formed by oxidation of Tax, other with *m/z* 337.0568, generated by water molecule addition, and two dimers formed by two molecules of Tax interaction (*m/z* 605.0964 and *m/z* 603.0787) with generation of ortho flavone dimeric of Tax.

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CONCLUSIONS

Taxifolin is a molecule susceptible to nucleophilic attack, as showed by *in vitro* and *in silico* studies and needs to be protected against alkaline medium, requiring an appropriate drug delivery system against intestinal environment.