PHYTOCHEMICAL ANALYSIS OF THE HYDROETHANOLIC EXTRACT OF PEELS FROM *Passiflora edulis* fo. *flavicarpa* Degener AND POTENTIAL VASORELAXANT EFFECT *EX VIVO*.

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Abstract

P. edulis is popularly known as yellow passion fruit. The objective of this study was to perform a phytochemical analysis of the hydroethanolic extract of flour of the peels (AFM) and evaluate its cardiovascular properties. The AFM was analyzed by Thin Layer Chromatography (TLC) and Ultra-High Performance Liquid Chromatography (UHPLC-UV-DAD). The total phenolics and flavonoids were quantified by colorimetric methods. The mesenteric artery was utilization for observation of the vascular reactivity. Phytochemical analysis allowed the identification from flavonoids (Orientin and isoorientin) in AFM. The pharmacological study showed that AFM has action in the vascular system causing relaxation not dependent on the endothelium.

Keywords: yellow passion fruit; peels; flavonoids; relaxing.

Introduction

P. edulis (Passifloraceae) is popularly known as yellow passion fruit and stands between Passiflora species (SATO et al., 1992). The peels of the fruits represent about 65-70 % of their weight. It is currently considered a residue in the food industry, however it could be reused as raw material for the preparation of new products (REIS, 2000). Different flours of passion fruit peel have been used to treat diabetes, high blood pressure and cholesterol (Jamir et al., 1999; Janebro et al., 2008).

So, the objective of this study was to perform a phytochemical analysis of the hydroethanolic extract of the flour of the peels from *P. edulis* by TLC and UHPLC-UV-DAD and to evaluate its cardiovascular properties in *ex vivo* model of mesenteric artery isolated.

Methodology

P. edulis peels was dried in a circulating air heater circulating air heater (55ºC) and triturated to obtain the flour. The flour was extracted in ethanol 50% (1:20, p/v), by maceration for seven days, filtered, concentrated by rotaevaporator and freeze-dried, obtained the hydroethanolic extract (AFM).

TLC was carried out on silica gel F254 the mobile phase: EtOH: CH_2O_2: H_2O: MeOH (10: 0.5: 0.6: 0.2; v/v/v/v). For this analysis, the extracts were fractionated by liquid-liquid partition with solvents of increasing polarity in order to obtain the acetate (EtAcO), and n-butanol (BuOH) fractions. After development, the plates were dried and sprayed with developer NP reagent (1% diphenylboryloxyethylamine), visualized under UV-365 nm.

The AFM was analyzed by liquid chromatography using a UHPLC Shimadzu, with UV-DAD detector. Chromatographic analyzes were performed using a reversed phase column (Phenomenex®) C-18 (4.6 mm x 250 mm, 5 µm) in gradient elution system (A: formic acid 0.3%; B: acetonitrile 0.3%). The chromatograms were recorded at 340 nm, whereas the UV

spectra were monitored at wavelength of 400-800 nm. The peaks were characterized by comparison of their retention times and UV spectra with the reference standards and by a co-injection (extract + reference standard). The total phenolics and flavonoids of the AFM extract were quantified by UV spectroscopy using colorimetric methods (Singleton and Rossi 1965).

The evaluation of the vasorelaxant effect of the extract was realized using mesenteric artery for observation of the vascular reactivity. Statistic analysis was conducted by Student's t-test. Statistical significance was considered of 95% ($p < 0.05$).

Results and discussions

Through TLC analyse was possible observe the presence of phenolic compounds and flavonoids, according the color of the spots. Some reference standards as orientin and isoorientin were used in TLC analyse and spots with Rf similar these compounds were observed in AFM extract (Figure 1 A).

The qualitative analysis by UHPLC confirmed the presence of the flavonoids orientin and isoorientin when compared the T_R with the reference standards and when was performed a co-injection analyse (extract + standard), by observation of the increase of area peak (Figura 1 B). A phenolic content of 7.71 ± 0.12 mg/g and a flavonoid content of 3.5 ± 0.14 mg/g in AFM extract were verified.

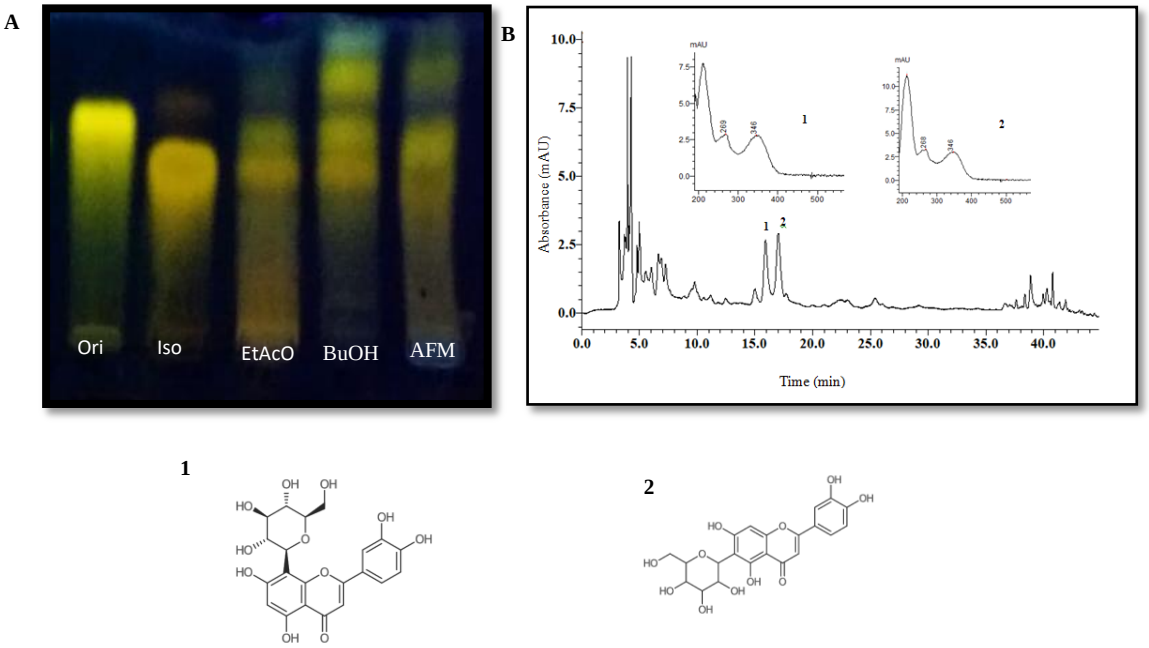

Figure 1: (A) Chromatogram obtained by TLC of AFM and pulverizado com reagente NP, visualized under UV-365 nm. Isoorientin (Iso) and Orientin (Ori) standards were used. (B) Chromatogram at 340 nm obtained by UHPLC-DAD of AFM (B). Orientin (1) and Isoorientin (2).

The pharmacological assays showed that AFM extract has a vascular relaxation effect depending on the concentration in the artery containing functional endothelium (E+) reaching a (Emax = 34.26 ± 10.4%) and in the absence of a functional endothelium (E) reached (Emax = 61.09 ± 7.5%). There was no difference in relaxation with vascular endothelial removal ($p > 0.05$), indicating that the vasodilator effect does not depend directly on the endothelium derivatives.

In our study, the chemistry profile reveals the presence of phenolic compounds especially flavonoids in extract of the flour of the peels from *P. edulis*. According to the iterature, the flavonoids and phenolics in general are involved in vascular relaxation activities

as well as present potent anti-inflammatory and antioxidant activities (Xu et al., 2007; Lubrano et al., 2007; Paredes et al., 2018).

So the presence of these compounds in AFM extract could justify, at least partially, the vasodilatory activities presented .

Conclusion

From this work it was possible to conclude that AFM extract show action on the vascular system causing relaxation not dependent on the endothelium. The phytochemical analysis allowed identify the compounds orientin and isoorientin. Therefore, our results demonstrated the potential efect vasorelaxant of AFM suggesting its potential as a source of bioactive molecules. For this reason the peels of *P. edulis* shows be promise to develop a herbal drug.

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