



LEMON JUICE (*Citrus latifolia*) ON COXs ENZYMES INHIBITION

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Abstract: The excessive menstrual bleeding or abnormal shows many causes, among then the endometrial and local disorders, which causes unbalance in prostaglandins levels, being the PGF2 α e PGE2 the main found in the blood. For the treatment of excessive bleeding has been administered nonsteroidal anti-inflammatory, which significantly reduces blood loss. These are known as cyclooxygenases inhibitors (COX-1 and COX-2), in the prostaglandins pathway, these endogenous are chemical signals in inflammatory processes [1-4]. *Citrus latifolia* or Tahiti lemon (Rutaceae) belongs to *Citrus* genus, which is very known for the flavonoids biosynthesis. The most flavonoids application is in inflammatory processes, acting like inhibitors on the COXs pathways [2]. The main of this job is evaluated the supplementary feeding with Tahiti lemon juice during the menstruation, aiming the inhibition of the excessive menstrual bleeding by the action of flavonoids present in the juice. Here we present the identification some flavonoids in lemon juice by HPLC and evaluation of inhibition against COX-1 and COX-2. The juice was squeezed manually, centrifuged and the supernatant (5mL) was first treated in SPE (solid phase extraction) OASIS HLB cartridges to the sugars cleanup with H₂O (0.1% acetic acid) (3mL) and the compounds of interest was eluted with MeOH:ACN (1:1, 5mL). This fraction was analyzed by HPLC-DAD: RP HSST3 C18 column and the mobile phase was a gradient with H₂O:ACN (both with 0.1% acetic acid). Until the present moment the flavonoids narirutin, rutin, hesperidin, eriocitrin and quercetin were identified by comparison of the retention times with standards. The fresh juice was tested using COX-1 and COX-2 screening kit (Cayman) in duplicate of the following concentrations: 5%, 2.5%, 1.25%, 0.62%. The indomethacin was used as positive control. The indomethacin and the standard of major flavonoids (hesperidin, eriocitrin and rutin) in the juice were tested in duplicate of the following concentrations: 64 μ g/mL, 16 μ g/mL, 8 μ g/mL, 4 μ g/mL. The IC₅₀ was determined by the sigmoidal dose-response curve. The fresh Tahiti juice proved active against both the COXs: IC₅₀=2.18% (COX-2) and IC₅₀=2.38% (COX-1), indomethacin was active with IC₅₀=0.180 μ g/mL (COX-2) and IC₅₀=0.002 μ g/mL (COX-1). The hesperidin inhibit COX-1 (IC₅₀=30.52 μ g/mL), but the other tested flavonoids were inactive. Thus, the fresh juice proved to inhibit both COX-1 and COX-2 and can be useful to control excessive menstrual bleeding. This effect can be correlated with the flavonoid composition as hesperidin or other compounds did not tested yet.

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