Identification of flavonoids from *Syzygium cumini* fruits by HPLC-PDA-MS/MS

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Syzygium cumini, known as jambolão, jamun, jambul, black plum, or jamblon, is a tropical tree belonging to the MYRTACEAE family, whose fruits are 2-3 cm long, with oval shape, and purple color. These fruits are commonly used for therapeutic purposes, due to their high contents of bioactive compounds, including anthocyanins and other phenolic compounds. Both anthocyanic and non-anthocyanic flavonoids from food sources have been widely studied due to their possible health benefits as dietary antioxidant and antimutagenic compounds. Anthocyanins constitute a special group of flavonoids, due to their intense color, ranging from red and violet to dark blue. Thus, high-performance liquid chromatography coupled to photodiode array detector and tandem mass spectrometry (HPLC-PDA-MS/MS), with ESI ionization source, was applied to identify the flavonoids from fruits of jambolão. The phenolic compounds were either separated on a C_{18} Shim-pack (for anthocyanins) or C_{18} Luna (other flavonoids and phenolic acids) columns, using as mobile phase linear gradient of water/methanol with 5 % formic acid and water/acetonitrile with 2 % of formic acid, respectively. For MS/MS analysis positive and negative ionization modes were used, with capillary voltage of 2.5 and 1.5 kV, respectively, and 1.2 V for MS/MS fragmentation. The identification was carried out based on the combined information provided by elution order in the reversed phase column, co-chromatography with standards, UV-visible and mass spectra characteristics compared to the literature data. Nine anthocyanins were identified, being five of them 3,5-diglucosides of the following aglycones: cyanidin, peonidin, delphinidin, petunidin and malvidin, showing *m/z* at 287, 301, 303, 317 and 331, respectively, in the positive ESI. The other four anthocyanins were 3-glucosides of the same aglycones, but peonidin. This pattern was also observed for other flavonoids, where dihexosides of dihydromyricetin (*m/z* at 303 in ESI+, and *m/z* at 301 in ESI-) and dihydroquercetin (*m/z* at 305 in ESI+, and *m/z* at 303 in ESI-) were identified, as well as two dihexosides of methylated derivatives from dihydromyricetin (one with $[M-H]^-$ at *m/z* 657, and MS/MS ions at *m/z* 495$[M-H-162]^-$, 333$[M-H-162-162]^-$ and the other one with $[M-H]^-$ at *m/z* 671, 509$[M-H-162]^-$, 347$[M-H-162-162]^-$). In addition, myricetin hexoside ($[M-H]^-$ at *m/z* 479), myricetin ($[M-H]^-$ at *m/z* 317), gallic acid ($[M-H]^-$ at *m/z* 169) and a galloyl-hexose (331$[M-H]^-$, 271, 169$[M-H-162]^-$, 125) were identified in the flavonoid extract from fruits of jambolão. For both anthocyanins and other flavonoids, the presence of glucose, or other hexose, in the molecule was assigned by the loss of 162 u. The presence of different aglycones among the flavonoids from jambolão fruits, indicates that enzymes catalyzing modification reactions, mainly hydroxylation and methylation ones, on the precursor flavonoids, play an important role in the biosynthetic pathways in this plant.

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