Anti-Inflammatory Prodelphinidins from Black Currant (*Ribes nigrum*) Leaves

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The infusion of the leaves of *Ribes nigrum* L. (Grossulariaceae), is traditionally used in some European countries (e.g., France, Belgium) for the treatment of "rheumatic" disease. Diuretic and anti-inflammatory properties could explain this ethnopharmacologic reputation (1).

In a study to select anti-inflammatory medicinal plants, we have observed that aqueous acetone or ethanol extracts of *R. nigrum* significantly inhibited at a dose of 50 mg/kg *i.p.* the carrageenan rat paw edema (2). We have now proved that flavonoids and phenolic acids previously found in this plant (3, 4) were not responsible for this intense activity but that the most active fractions contained proanthocyanidins (oligomers and polymers). In the present study, we also report on the isolation and characterization of three bioactive molecules: two prodelphinidin dimers and a new trimeric one.

Initial fractionation of the aqueous acetone extract of the air-dried leaves was obtained by medium pressure liquid chromatography (MPLC) on RP-8 with acetone-water. This chromatographic procedure was effective for a clean-cut separation of proanthocyanidins and associated flavonoid glucosides. The fraction containing proanthocyanidins was then chromatographed on Sephadex LH-20. The main oligomeric compounds were shown to be prodelphinidins instead of procyanidins; this fact can be related to a screening on the fundamental flavan structural units of condensed tannins: delphinidin was ten times more abundant than cyanidin after acid treatment (3, 5).

The major dimers are: gallocatechin-(4α→8)-epigallocatechin (6) and gallocatechin-(4α→8)-gallocatechin (7). This is the first time that these two dimers have been isolated together. This fact explains that their separation was very difficult; combination of chromatographic systems and variation of the eluting solvents were necessary. The third compound is characterized as trimeric prodelphinidin possessing the structure gallocatechin-(4α→8)-gallocatechin-(4α→8)-gallocatechin. To the best of our knowledge, it is the first example of a natural product possessing three unsubstituted gallocatechin units. These compounds were identified by spectroscopic methods (FAB-mass, IR, ^{1}H-, ^{13}C-NMR), correlation with known proanthocyanidins, and chemical degradation studies.

Investigation of the biological activity of these three prodelphinidins showed an important dose-dependent anti-inflammatory activity after 2 h and 4 h in the carrageenan model (18 %, 40 %, and 55 % reduction of edema at respectively 5, 10, and 40 mg/kg. *i.p.*) while isoquercitrin (the major flavonoid) and phenolic acids do not show any significative result at 100 mg/kg *i.p.* In the same conditions, we observed the activity of indomethacin (44 % inhibition at 4 mg/kg), aspirin (47 % inhibition at 200 mg/kg), and crude aqueous extract (57 % inhibition at 60 mg/kg).

We can come to the conclusion that the anti-inflammatory activity of *Ribes nigrum* leaves can be at least partially explained by the presence of prodelphinidin oligomers. This biological property, as well as the positive effects of proanthocyanidins in the stabilization of collagen fibres (8), could corroborate the traditional use of this medicinal plant. Identification and quantification of these prodelphinidins could therefore be investigated for the quality control of medicinal products based on black-currant leaves.

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Xanthones from *Chironia krebsii*

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As a part of our ongoing chemical and biological studies on Gentianaceae (1), the leaves and roots of an African species, *Chironia krebsii* Griseb., have been investigated. The genus *Chironia* is distributed mainly in South Africa but several plants range nothward into tropical Africa and Madagascar. No phytochemical investigation on the genus *Chironia* has been published. Our interest in Gentianaceae arises from the fact that they contain xanthones, some of which are known to be strong and selective inhibitors of monoaminooxidases (2, 3). In addition, these compounds are useful chemotaxonomical markers (4).

Reverse phase HPLC with photodiode array detection was used to analyse the dichloromethane and methanolic extracts of aerial parts and roots of *C. krebsii* and revealed the presence of xanthones in all extracts.