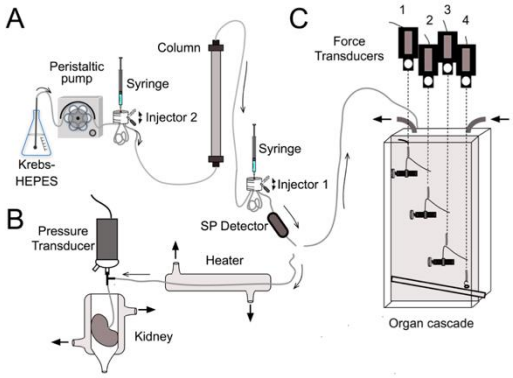
CANSCHROME/CASCADE



CANSCHROME is an original system that used liquid chromatography coupled to biological detection. When used in conjunction with CASCADE, which permits creating a pharmacological fingerprint of the eluted substances as it passes the effluent consecutively through an aorta ring, a tracheal ring chain, a vas deferens and a piece of ileum. CANSCHROME/CASCADE permits speed-up the procedure of drugs characterization either from natural products or from combinatorial chemistry. Also, it reduces the necessity of bench space saves and lab resources.

It was patented in 2012 (**Borges, R.** et al, ES2372832 A1 **27.01.2012**; **ULL**), and also was published:

CAMPUZANO-BUBLITZ, M.A. et al (2018) *Naunyn-Schmiedeberg's Ach. Pharmacol.* **391,** 9-16)

and was part of the Doctoral Thesis of José G. Hernández Jiménez. ULL in 2015.

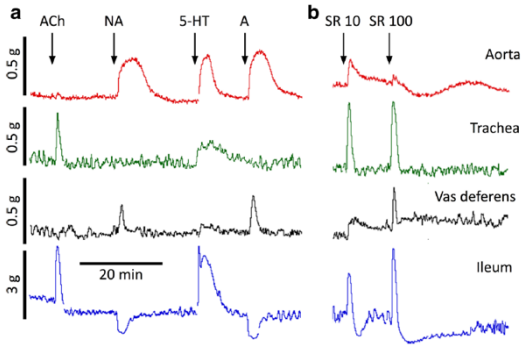


Fig. 2 Drug characterization using four-organ cascade detection. **a.** Direct injection (through injector 2, see Fig. 1) of 1 mL of acetylcholine (ACh), noradrenaline (NA), serotonin (5-HT), and adrenaline (A), all at a concentration of 10 µM. The vertical calibration bars (in grams) are for the force calibration. **b.** Contractile responses to the natural *Stevia rebaudiana* Bertoni (SR) extract at 10 and 100 mg·mL^{-1}, injecting the extract directly through injector 1

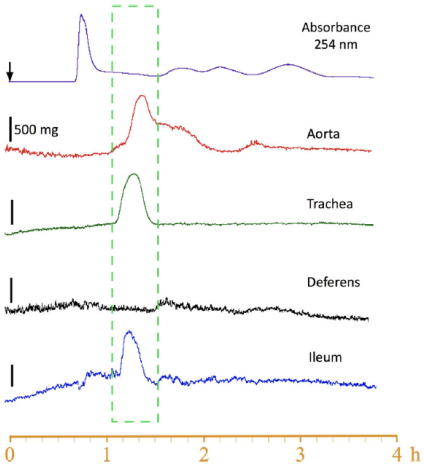


Fig. 3 On-line analysis of *Stevia rebaudiana* Bertoni plant extracts using MPLC coupled to the organ cascade. The aqueous extract (1 mL) was injected into the system and the eluate from the column was directed sequentially to an absorbance detector (set at 254 nm), rat aorta rings, rat tracheal rings change, rat vas deferens, and rat ileum. The eluate from the column that caused the peaks highlighted with a discontinuous box was collected and analyzed by mass spectrometry. The calibration bar corresponds to a 0.5 g tension/force. The figure shows one representative experiment of five. Lower axis corresponds to the time (in hour) from the injection of extract

Fig. 4 Characterization of the active fraction. The eluate from the column where the pharmacological activity was found was collected (10 mL), concentrated, and resuspended in water. **a.** An aliquot (60 µL) of this solution (SR) was added to an isolated rat ileum in a 4-mL classic organ bath chamber at the time indicated by the arrow. The traces are representative from four different experiments. **b.** Electrospray high resolution mass spectrometry spectrum of the methanol-soluble fraction. **c.** Chemical structure of rebaudioside N