

P-028: SUBTRACTION OF BIOACTIVE CONSTITUENTS: SELECTIVE REMOVAL OF BENZOIC ACID IN ELUCIDATING THE ACTION MECHANISM OF CRANBERRY JUICE

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Cranberry (*Vaccinium macrocarpon* Ait., Ericaceae) juice (CJ) is widely used for treating urinary tract infections in women. A proposed mechanism of action is that CJ exhibits anti-adhesive effects vs. uropathogenic *E. coli*. The contained proanthocyanidins (PAs) were shown to block *E. coli* adhesion to urinary epithelial cells. Major acidic constituents, e.g. benzoic acid (BA) have also been suggested to contribute to the CJ activity due to pH effects. Guided by an *E. coli* anti-adherence bioassay, both PAs and BA were found in the active CJ fraction. To eliminate the potential interference of the bacteriostatic effect of BA on the *E. coli* anti-adherence action, we developed a countercurrent chromatography (CCC) method that allows the selective removal of BA from the active fraction in a single step. Upon BA removal, the anti-adherent activity of the fraction was fully retained. The purity of the removed BA was determined to be ~94% by LC-MS and ¹H NMR techniques. The impurity was determined to be scopoletin, which represents the first coumarin reported from cranberries. The CCC approach of selective removal of phytoconstituents has potential broad applicability in the biological evaluation of botanicals.