

Reversed Phase HPLC of Gallotannins

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Excellent resolution of gallotannins can be achieved by gradient elution on reversed phase systems as described in Kawamoto, H.; Nakayama, M.; Murakami, K. *Phytochemistry* 1996, 41, 1427. In some cases, adequate resolution may be obtained with isocratic separations on reversed phase systems. Separation with normal phase HPLC is also possible.

Column

C-18 (ODS) such as Beckman Ultrasphere 4.6 mm x 25 cm, 5 um particles with similar C-18 precolumn.

Elution

0.1% aqueous trifluoroacetic acid

0.1% trifluoroacetic acid in HPLC grade acetonitrile

gradient, 1 mL/min, 4:1 aqueous to 3:2 aqueous over 7 min (retention time, pentagalloyl glucose, 5.9 min)

Detection

Gallotannins are conveniently detected with UV detectors. We use UV detection at 220 nm, and can easily detect ng levels of gallotannins.

Sample preparation

Gallotannins can be dissolved in methanol or acetone (gives large solvent peak). Gallotannin-bovine serum albumin precipitates can be dissolved in 1% aqueous sodium dodecyl sulfate and injected directly onto the HPLC (Hagerman, A.E.; Rice, M.E.; Ritchard, N.T. *J. Agric. Food Chem.* 1998 46, 1409-1421).