

Pentagalloyl Glucose

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Introduction

Pentagalloyl glucose is easily prepared from appropriate preparations of tannic acid. The method described here is from Hagerman, A.E.; Zhao, Y.; Johanson, S. ***Methods for Determination of Condensed and Hydrolyzable Tannins***. In *Antinutrients and Phytochemicals in Foods* (Shahadi, F., ed.). American Chemical Society, Washington DC, 1997; pp. 209-222.

Reagents

- Acetate buffer (0.1 M) pH 5. Add 0.57 mL glacial acetic acid to 80 mL distilled water, titrate to pH 5 with NaOH solution, then bring volume to 100 mL with distilled water.
- Methanolysis solution: Mix 70 mL methanol with 30 mL of the acetate buffer.
- Ethyl acetate
- Diethyl ether (peroxide-free)

Method

1. In a test tube, dissolve 0.5 g tannic acid in 10 mL of the methanolysis solution. Cover, and place in 65°C water bath for 15 h.
2. Increase the pH to 6, while stirring, with 0.25 M NaOH.
3. Evaporate under reduced pressure (rotary evaporate) at temperatures below 30°C to remove methanol. As the methanol is removed, add water to maintain volume. Do not let the sample go to dryness.
4. After removing all of the methanol, extract the aqueous solution three times with diethyl ether (in a small separatory funnel). The PGG should be in the aqueous (bottom) layer. Discard the top layer after each extraction, using appropriate methods to discard ether.
5. Extract the aqueous layer three times with ethyl acetate, saving the ethyl acetate (top) layer.
6. Combine the ethyl acetate extracts and rotary evaporate to reduce the volume. Add water to maintain the volume, and continue to evaporate to remove all of the organic solvent.
7. The aqueous suspension should be centrifuged and suspended in 2% methanol /98% water, and re-centrifuged three times, saving the solid PGG each time.
8. Re-suspend the material in water, and freeze dry the suspension to yield pure white PGG.

Analysis

The starting material, and the various fractions at each step of the procedure, should be monitored by normal phase HPLC to verify that methanolysis occurred (loss of late eluting peaks, large increases in methyl gallate and PGG). The final product purity should be assessed by HPLC and by proton nmr in deuterated acetone. The assignment of peaks in the nmr will confirm that the product is PGG, possibly contaminated with a trace of methyl gallate and gallic acid.

Proton nmr assignments (from TMS)

glucose C-1, 6.3 ppm (d, 1H)

glucose C-2, C-4, 5.6 ppm (q, 2H)

glucose C-3, 6.0 ppm (t, 1H)

glucose C-5, 4.5 ppm (d, 1H)

glucose C-6, 4.4 ppm (dd, 1H)

galloyl group, between 6.9-7.2 ppm, 5 singlets (2H).