

Analysis of Tobacco Alkaloids



Introduction:

Tobacco alkaloids are extracted with a strong cation exchange sorbent using an acidic buffer, filtered and further acidified. Extraneous compounds are removed by washing the sorbent, yielding clean chromatography without loss of target analytes.

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EUBCX1HL2Z

200 mg benzenesulfonic acid
high load, 10 ml

Procedure:

1. Sample Preparation

- a) Weigh 100 mg of finely ground tobacco in a screw cap vial
- b) Add 6 mL 0.1M sodium acetate buffer (pH 4.5) and 100 μ L IS (d4- nornicotine, 1 μ g/ μ L)
- c) Mix on rotating shaker for 10 minutes
- d) Filter extract through 20 micron frit filter column
- e) Add 300 μ L glacial acetic acid and mix

2. Sample Extraction

- a) Condition column EUBCX1H2Z with 3 mL of MeOH:1.0M acetic acid (80:20)
- b) Pour sample onto column, draw through at 1-2 mL/min
- c) Wash column with 3 mL of MeOH:1.0M acetic acid (80:20)
- d) Dry column for 5-10 min using full vacuum

3. Elution

- a) Elute alkaloids with 3 mL CH_2Cl_2 /isopropanol/ NH_4OH (70:26:4) by gravity
- b) Evaporate eluant to dryness with N_2 and low heat ($< 40^\circ \text{C}$)
- c) Reconstitute with 200 μ L ethyl acetate
- d) Analyze on GC/FID/NPD or GC/MSD



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