Determination of Propylene Glycol, Glycerol and Sorbitol in Tobacco and Tobacco Products by High Performance Liquid Chromatography with Refractive Index Detector

B. J. Rajesha, S. V. Dallakuladkar, S. K. Mehta

ITC R&D Centre, ITC Ltd., Bangalore, India

Abstract

A simple HPLC method is developed for the determination of propylene glycol (PG), Glycerol (GLY) and sorbitol in tobacco and tobacco products using amino column and mobile phase containing acetanilide and water. It involves the extraction of humectants from a sample by mobile phase and subsequent chromatographic separation on Inertsil – amino (NH2) column (4.6 x 250mm x 5 mm) using isocratic system. Mobile phase – acetanilide : water (80:20) Retention time of PG, glycerol, and sorbitol are 4.5 min, 6.0 min, and 10.8 ± 0.5 min at a flow rate of 1.0 ml/min. The method has been validated by standard validation protocols i.e. limit of detection, limit of quantification, recovery, repeatability and reproducibility. Minimum recovery of 92%, 94%, and 96% for PG, GLY and sorbitol were obtained with a linear regression coefficient of 0.9999, 0.9999 and 0.9995 respectively. Limit of detection of PG, glycerol and sorbitol were 25 ppm. Titrated method provides several advantages such as sensitivity, simple column and mobile phase.

The method has been found suitable for rapid determination of humectants for large number of tobacco and tobacco products.

Introduction

Currently standard method for the determination of humectants in tobacco is Coresta recommended method No 61. It involves use of polymer column and mobile phase containing calcium salt of EDTA, tedious multi-step sample cleaning with the use of cartridge column to remove matrix interference of tobacco. Therefore, it was felt to develop a simple, fast analytical method without any matrix interference of tobacco.

Sample preparation

Weigh about 1g ground tobacco in 250 ml Erlenmeyer flask add 100 ml mobile phase

Shake at 190 rpm for 60 min filter through 0.45 um disposable syringe filter

Filter approximately 3 ml of sample solution into a vial, seal with a crimp cap and inject to the LC system.

LC Conditions

Mobile phase – Aetionitrile : water (80:20)
Flow rate : 1.0 ml/min
Run time : 16 min
Detector cell temperature : 40°C
Detector : Refractive index

Chromatograms of Standard and Sample

Validation:

Limit of detection and limit of quantification

Determination of signal-to-noise ratio is performed by comparing measured signals from samples with known low concentration of analyte with those of blank samples and establishing the minimum concentration at which the analyte can be detected.

1. Signal to noise ratio between 3 : 1 is considered acceptable for estimating the detection limit.
2. Signal to noise ration between 10 : 1 is considered acceptable limit

a) Limit of detection of PG, GLY, and sorbitol is 25 ppm (SN ratio : 3.0 for PG, 7.4 for GLY and 4.8 for sorbitol).

b) Limit of quantification of PG, GLY, and sorbitol is 100ppm ( SN ratio : 11.9 for PG, 19.0 for GLY, and 12.9 for sorbitol).

Recoveries

Studies conducted by adding a known amount of Humectants to the tobacco sample

PG GLY Sorbitol

Repeatability

Tobacco samples of pipe tobacco were analyzed under repeatability conditions. RSD were within acceptable limits.

Reproducibility

Same tobacco samples analysed under reproducibility conditions and RSD % was well within the acceptable limits.

Conclusion

1. Easy sample preparation.
2. No matrix interference as evidenced from the recovery studies.