DETECTION OF TRIBUTYRIN IN BUTTER USING COLUMN CHROMATOGRAPHY (CC), THIN-LAYER CHROMATOGRAPHY (TLC) AND HIGH PERFORMANCE LIQUID CHROMATOGRAPHY (HPLC)

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ABSTRACT

DETECTION OF TRIBUTYRIN IN BUTTER USING COLUMN CHROMATOGRAPHY (CC), THIN-LAYER CHROMATOGRAPHY (TLC) AND HIGH PERFORMANCE LIQUID CHROMATOGRAPHY (HPLC)

The study of tributyrin detection in butter was done by gravimetric silica gel column chromatography, thin-layer chromatography and high performance liquid chromatography. First, column chromatography was used for selection of solvent in HPLC. Various kind of solvent were used to elute tributyrin component in butter. The eluents from column chromatography were collected and examined in TLC silica plate. The R_f value of sample and standard were calculated and compared. Selection of solvent is based on the closeness of R_f value of sample to the standard. Solvent mixture (hexane:acetonitrile/70:30) was chosen as the most suitable solvent. The R_f value of sample to the standard were 0.85 and 0.82 respectively. Hence, the selected solvent mixture was used as mobile phase in HPLC instrument. Further analysis was accomplished by HPLC techniques. The chromatographic separation in HPLC was achieved using C18 column, and the detection was accomplished using UV detector. The effect of flow rate and effect of grade of standard tributyrin were examined. Both analytical-reagent and HPLC grade were studied. HPLC grade of standard tributyrin was used rather than analytical-reagent grade. Flow rate 1.0 mLmin⁻¹ was chosen as the most suitable flow rate due to least of tailing peak. The concentration of tributyrin in butter was 0.215 mM.