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EXTRACTION AND PURIFICATION OF CURCUMIN FROM TURMERIC: TLC AND SPECTROPHOTOMETRIC ANALYSIS

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ABSTRACT

Turmeric is obtained from the roots of Curcuma longa rhizome which belongs to family Zingiberaceae. Turmeric consist of curcuminoids which include curcumin, demethoxycurcumin and bis–demethoxycurcumin. The present work is focused on extraction of curcumin using cold extraction method by using acetone, which is a simple and economical to adopt method. The extracted curcuminoids was analysed by TLC using the mobile phase (chloroform:methanol) in ratio 98:1. Rf value showed at 0.90, 0.75, and 0.696 as curcumin, dimethoxycurcumin, bis-demethoxy curcumin. The spectrophotometric method was carried out to determine the maximum absorption wavelength which is found at 420nm. The linearity was checked with sample concentrations which obeyed Beer-lambert's law.

INTRODUCTION:

Turmeric (Indian saffron) is widely cultivated spice plant throughout India. It is used as a traditional and house hold medicine for its potential in wound healing, carminative, stomachic, blood purification and antiseptic property (1). It is also used as aesthetic in perishable food items, soften rough skin, topical creams and bath soaps. Turmeric is obtained from dried powdered rhizomes of Curcuma longa and other Curcuma species belongs to the family Zingiberacaea. Curcumin is the chief constituent (3-4%) in turmeric and is present as yellow-orange, crystalline powder with melting point at 183^0C. The chemical name of curcumin is di feruloyl methane ([1, 7-bis (4-hydroxy-3-methoxy phenyl)-1,6-heptadiene 3,5-di one]) a poly phenyl compound with molecular formula $C_{21}H_{20}O_6$ and molecular weight is 368.91. Curcumin is practically insoluble (≈11ng/ml) in water and freely soluble in organic solvents like methanol, ethanol, chloroform, benzene and DMSO. The constituents of turmeric are curcumin, de methoxy curcumin and bis dimethoxy curcumin combinely together called as curcuminoids. The other constituents in turmeric are volatile oils like turmerone, zingiberene, sesqiterpenes, sesquiphellandrine, terpinole, p-cymene, cineol, caryophyllene, nerolidol, dehydro zingirone, zerumbone, germacrene etc (2-7). Curcumin is first extracted in an impure form by Vogel et al in 1819 and its structure described initially by Lampe and Milobedeska in 1910 (8-10).

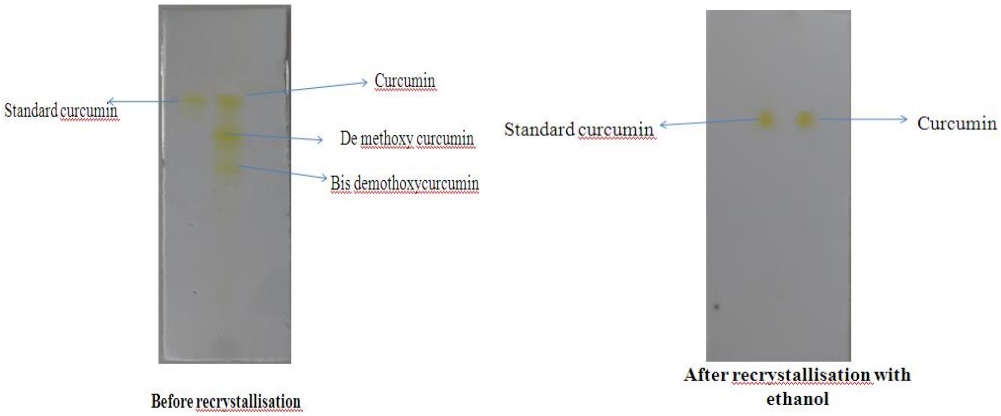

Fig 1: TLC Study before and after recrystallisation of sample

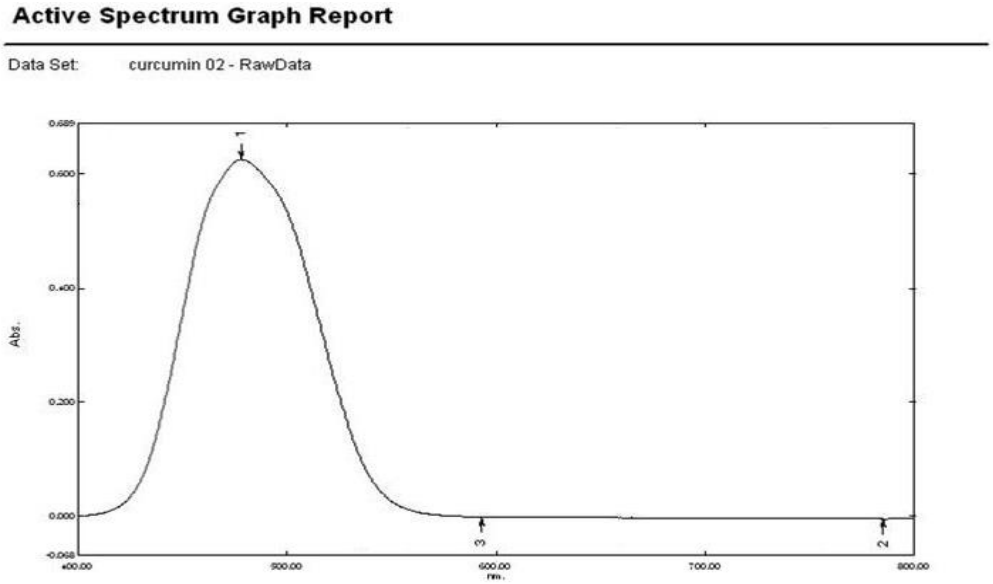

Fig 2: Maximum absorption wavelength (λmax) of curcumin at 420nm

Isolation of curcumin from turmeric powder

30g of turmeric powder soaked in 100 ml of acetone in a conical flask, wrapped to prevent the evaporation of acetone and it was kept a side for 72 hours with occasional shaking. In this time the maximum amount of curcumin dissolves in acetone and complete extraction takes place. It is filtered, concentrated; to this add n-hexane to remove resinous matter. Filter the solution and to this add 0.1N NaOH and acidify with diluted HCl, filter the solution and collect the filterate in separating funnel and separate the acetone layer. Concentrate the acetone and recrystallize with ethanol.

Separation of curcuminoids by Thin Layer Chromatography

Dissolve extracted samples (before and after recystallisation with ethanol) and standard curcumin of each milli gram separately using ethanol as a solvent in three beakers. TLC chamber saturated with chloroform and methanol (98:2) as a mobile phase. Pre coated silica gel TLC plates were activated and separated into three columns. Spots were placed on the TLC plate and develop the chromatogram.

Determine the retention time/retardation factor using the following formula. Retention Factor (R_f) = Distance travelled by sample/ Distance travelled by solvent front.

Determination of maximum wavelength

Dissolve 500mg of curcumin in ethanol, UV-Visible spectra was taken from 400-800nm wave length range and determine the maximum absorbance (λmax).

RESULTS AND DISCUSSION

In the present experiment acetone is used as a solvent as curcumin having maximum solubility in acetone. N-hexane, 0.1N NaOH and dilute HCl used to separate the resins. After treatment with alkali and acid reddish brown colour solution was obtained. It gives three different spots when separated with TLC (Figure 1**)** which indicates the presence of curcumin, demethoxy curcumin and bis demethoxy curcumin, where as single spot obtained after recrystallisation with ethanol reveals that all the remaining curcuminoids were eliminated and it matches with the standard curcumin.

The R_f values were found to be 0.90, 0.75, 0.696 for curcumin, de methoxy curcumin and bis demethoxy curcumin respectively. The curcumin UV spectrophotometric analysis conducted using ethanol as a solvent. It given maximum absorption wavelength (λmax) at 420nm (Figure 2). The linearity of the sample was done in the concentration between 10-50µg/ml and obtained 0.998 correlation co efficient.

CONCULSION:

Since ancient time curcuminoids are being used as potential therapeutic effects such as anti-tumors, anti-microbial, neuroprotective etc. A simple estimation of curcuminoids mainly curcumin was carried out by using TLC which showed Rf values at 0.90 for curcumin, 0.75 for demethoxy curcumin, 0.696 for bis-demethoxy curcumin. The maximum absorption wave length was detected by using spectro photometric method at 420nm.

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