EXTRACTION AND PURIFICATION OF NATURAL DYE
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Received on 28-07-2016

Accepted on 29-08-2016

Abstract

With the present national and international awareness of environmental ecology and pollution controls, natural dyes appear to be ideal choice since they are chosen from the non toxic sources and can be handled very easily and safely. Dyes derived from natural sources have emerged as an important alternative to synthetic dyes. In this study two different varieties of chrysanthemum (*Dendranthema grandiflora*) and peels of badam fruit (*Prunus dulcis*) were taken and dyes were extracted. The qualitative analysis of dyes was done using by Thin layer chromatography (TLC). The chromatogram was explained by Retention factor (Rf). The Rf value for each substance is the distance it has moved divided by the distance solvent front has moved.

Keywords: Natural dye, *Dendranthema grandiflora*, *Prunus dulcis*, TLC.

Introduction

Currently, ecological considerations are becoming important factors in the selection of consumer goods all over the world. Since the mid-1980s, more interest has been shown in the use of natural dyes and a limited number of commercial dyes, and small businesses have started to look at the possibility of using natural dyes for coloration of commercial dyes, and small businesses have started to look at the possibility of using natural dyes for coloration ¹. Plants are known to produce some of the most valued dyes in the world for their natural, beautiful and durable colors. Natural dyes have better biodegradability and generally have higher compatibility with the environment ².

Chrysanthemum (*Dendranthema grandiflora* T.) is one of the most important flowers crop grown commercially in India for cut and loose flowers and is also used for garden display. It belongs to family Asteraceae and is commonly called as the “Queen of the East”. The availability in range of colours and types of flowers adds to its value to be used for different
purposes. Almond (Prunus dulcis L.) is one of the species of Prunus belonging to the subfamily Prunoideae of the family Rosaceae. Nutrionally and medicinally almond is a valuable food commodity.

Chromatography is an analytic method that is widely used for separation, isolation, identification and quantification of components in a mixture. Components of the mixture are carried through the stationary phase by the flow of mobile phase. Separations are based on differences in migration rates among sample components. Thin layer chromatography (TLC) was chosen over other chromatographic methods because it is a simple, quick and inexpensive procedure that can be used for the analysis of mixture. The effectiveness of separation depends on the mixture to be separated, the choice of mobile phase and the adsorption layer. The term Retention factor ($R_f$) that is commonly used to describe the chromatographic behavior of sample solutes. The $R_f$ value for each substance is the distance it has moved divided by the distance solvent front has moved. In this study, it is attempted to extract dye from red and purple variety chrysanthemum (D. grandiflora) flower petal and peel of badam fruit (P. dulcis) and purified by thin layer chromatography (TLC) method.

Materials and methods

Plant material

The present study was conducted to extract dye from natural and herbal sources. Three plant materials were used. The flower petals from two different color of chrysanthemum (Dendranthema grandiflora) flower and peel of badam fruit (Prunus dulcis) were used (Figure 1). A dark purple and red variety chrysanthemum flowers were collected from Chennai flower market and badam fruits were collected from the badam tree in university campus.

![Figure 1: Purple (a) and red (b) variety of Chrysanthemum and badam fruit (c)](image)

**Extraction of natural dye**

2 g of each plant material was placed in a mortar-pestle and crushed with a mixture of hexane, acetone, and water in the ratio of 1:1:1. Then the extracted dye was collected by filtration and used for TLC.
**Purification of pigments by thin layer chromatography method**

The TLC plate was made using silica gel slurry. Thickness of the layer was 1 mm and pore size was moderate. Thin and uniform layer of silica was prepared. The plate was activated in an oven at 50 °C for 30 min before use. A thin line was drawn on the activated TLC plate about 1.5 cm above the bottom. A spot of the extract was placed on the line and allowed to dry. The plate was kept in the beaker containing the mobile phase (hexane: acetone: distilled water - 3:2:5) in such a way that the end near the sample application should touch the mobile phase. The chromatogram was allowed to run about 30 min. The plate was dried at RT. Then the $R_f$ value of each dye was calculated using formula:

$$R_f = \frac{\text{distance travelled by component}}{\text{distance travelled by solvent}}$$

**Results and discussion**

Thin layer chromatography is a simple and most economical method for the purification of pigments. Here three plant samples were used and dyes were extracts by using solvents. The dyes extracted from three samples were dark purple, red and pink from dark purple chrysanthemum and red chrysanthemum flower petals, peel of badam fruits, respectively. The Figure 2 shows the chromatogram of natural dyes.

![Figure 2: TLC chromatogram of natural dyes from different plant samples a) dark purple variety of chrysanthemum, b) red variety of chrysanthemum, and c) peel of badam fruit](image)

From the chromatogram, we can confirm that each dye is not a mixture of colors; they are individual, because only one spot was seen on the developed TLC plate. From the TLC plate the $R_f$ values were calculated (Table 1).

**Table-1: Determination $R_f$ values from Thin Layer Chromatogram.**

<table>
<thead>
<tr>
<th>Plant materials</th>
<th>$R_f$ Values</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dark purple petals of chrysanthemum</td>
<td>0.860</td>
</tr>
<tr>
<td>Red petals of chrysanthemum</td>
<td>0.789</td>
</tr>
<tr>
<td>Pink peels of badam fruit</td>
<td>0.815</td>
</tr>
</tbody>
</table>
The separation of natural dyes by TLC provided R<sub>f</sub> values for each. The pigment for dark purple moved and color of the spot became purple. The R<sub>f</sub> value for the moved dark purple pigment is 0.860 which may be carotene. The red dye spot turned to yellow and the R<sub>f</sub> value for this is 0.789 which may be due to presence of xanthophylls. Chrysanthemum contains a large variety of colorant secondary metabolites, such as carotenoids, flavonoids, anthocyanin. For the badam fruit, the pink dye turned to dark pink, may be because of anthocyanin and the R<sub>f</sub> value is 0.815.

**Conclusion**

Natural dyes provide an environmentally safe option for coloring of food and other materials. The present study showed that natural dye can be successfully extracted from the flowers of chrysanthemum and peel of badam fruit. The whole process of extraction is eco-friendly. TLC technique can be a good tool for the purifying of natural dyes from plant materials, and utilized for the identification of different species.

**Acknowledgements**

The authors convey their thanks to Department of Industrial Biotechnology, Bharath University, Chennai, for providing laboratory facilities.

**References**


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