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SPECTROPHOTOMETRIC ANALYSIS OF COLOURANT EXTRACTS FROM THREE Different COLOURED Flowers of *Tagetus erecta* (L) AT DIFFERENT pH VALUES

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ABSTRACT:

Spectrophotometric analysis of the natural colourants from three different coloured flowers of *Tagetus erecta* (L) were carried out at different pH values. The results show that *Tagetus erecta* (L) extract (colourant) was stable at the experimental pH range of 2.60 – 9.36 (Light yellow coloured flowers) and 4.0- 12.0 (Orange coloured flowers) and 2.48 – 8.40 (Brown coloured flowers). This was indicated by the similar peaks of absorption maxima at the different pH values. Beer's law is valid over the concentration range of 50 - 400 ppm. The colourant extract has absorption maximum at 470 nm, 410 nm and 480 nm respectively. *Tagetus erecta* (L) extract, with further studies could be used as a colourant in pharmaceutical formulations.

KEY WORD: Spectrophotometric analysis, *Tagetus erecta*, Colourant, Ph Effect.

INTRODUCTION:

Marigold is a plant of the genus *Tagetus* of the family Asteraceae, mostly cultivated as garden flower which is one of the natural sources for achieving yellow color. The use of colourants in cosmetics, foods and drugs dates back to antiquity. As colourants abound, so are the reasons for using them. Colours could be used as a means of identification, improvement of aesthetics, and protection

from various factors of degradation, warning and concealment, to mention but a few [1]. Natural dyes of plant origin are present in plants as glycosides or glycones which are free or bound to sugars [2]. Natural dyes seem to be less harmful on humans and environment, and nowadays they could be considered as a better alternative to synthetic dyes [3-8]. Flowers vary in colour from yellow and gold to orange, red and mahogany. The taller and larger-flowered *Tagetes erecta* was often called African Marigold and the smaller *Tagetes patula* was known as French Marigold. But Marigolds have been cultivated all over the world and lots of similar hybrid varieties have been developed from the two species [10,11,12]. Marigold flowers contain compounds called carotenoids. Lutein ($C_{40}H_{56}O_2$) is a natural pigment of the carotenoid family. It is widely found in nature. Lutein and its isomer zeaxanthin are also known as oxycarotenoids. They are the main xanthophylls in Marigold flowers and also in vegetables such as spinach and kale. It is also present in foods such as corn and egg yolks and can be found in the eye, skin, cervix and the breast. It is a powerful antioxidant and helps in maintaining healthy eyes [9,13]. The effect of pH changes on the colourant is the main focus of the present study.

MATERIALS AND METHOD:

All materials/chemicals used as procured without further purification, were of AR grade (Merck). Distilled water was obtained from an all glass still.

Preparation of *Tagetes* Extract:

20 gms of dry flowers of *Tagetes* were dispersed in 1 dm³ ethanol and heated to above 50 to 60 °C in Soxhlet extractor for five hours. All the colourants were extracted from flowers by the end of five hours, after extraction the extract was vacuum distilled and recovered to dryness. 100 ml of distilled water was added to this extract and subsequent aliquots were prepared using this stock solution and stored in amber-coloured bottles to prevent darkening which often occurs when the material is exposed to light.

Absorption spectra of *Tagetes* extract at different pH values:

A 50-100 ppm concentration of the colourant was made in aqueous solution and pH values were adjusted to 2.60, 6.95, 8.20, 9.36 (Sample-1), 4.00, 6.15, 8.00, 12.00 (Sample-2) and 2.48, 3.15, 4.06, and 8.40 (Sample-3) respectively. These solutions were scanned in turn at intervals of 1 nm using UV – VISIBLE spectrophotometer (Equiptronics). Their peak absorbances were determined.

Beer's plot of Tagetus colourant solution:

A stock solution of Tagetus colourant (1000 ppm) was made in distilled water. A serial dilution of the stock was made to obtain concentrations of 50-400 ppm. The maximum absorbances of these solutions were determined using the UV –VISIBLE spectrophotometer at 470 nm 480 nm and 410 nm respectively and plotted against the concentration to obtain a standard plot

RESULT AND DISCUSSION:

A straight line passing through the origin was obtained for the Beer's plot (Fig.1 , Fig. 2 & Fig. 3)). This show that light absorbance of standard solution of Tagetus colourant extract can be used in quantitative analysis of the crude extract. Tables 1 to 3 show the absorption peaks of the Tagetus extract solution prepared at different pH values. Tagetus colourant solution, within the limits of pH range chosen, showed consistency in its wavelength of the peak of absorption of light spectrum. This shows that a single chromophore was contained in the Tagetus extract probably with a similar or different substituents in different position of the chromophoric nucleus. Had it been composed of several chromophores, it would have shown less variation of peak absorptions. Thus, it seems that they all may contain the same basic chromophoric or structural unit.

Results also indicate the stability of the colourant within the stipulated pH range. Different colours have their different and characteristic wavelength of peak absorption. It was found that Tagetus colourant extract was stable at the experimental pH-range of 2.6 – 9.36 for sample – 1 and 4.0 to 12.0 for sample – 2 and 2.48 to 8.40 for sample -3 respectively. The spectrophotometer analysis does not show significant instability as a result of the pH modifications. Due to the adverse effects of some synthetic colourants, which include carcinogenicity, tetratogenicity and various other toxic effects natural colourant such as *Tagetus erecta* can be used without all these undesired effects. Natural colourants have wider margin of safety than synthetic colourants.

CONCLUSION:

It has been seen that the natural pigments showed the sensitivity to different pH values and the structural changes in chromophore system of dyes caused by changes in pH but in the present study, we observed no such changes in the properties of the extracts of *Tagetus erecta* with the change in pH.

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Table 1: Effect of pH on the peak absorption values (Sample1-Light yellow coloured)

S.No.	pH values	Peak absorption (nm)
1	2.60	470
2	6.95	470
3	8.20	470
4	9.36	470

Table2 : Effect of pH on the peak absorption values(Sample2-Orange coloured)

S.No.	pH values	Peak absorption (nm)
1	4.00	480
2	6.15	480
3	8.00	480
4	12.00	480

Table3 : Effect of pH on the peak absorption values(Sample3- Brown coloured)

S.No.	pH values	Peak absorption (nm)
1	2.48	410
2	3.15	410
3	4.06	410
4	8.40	410



