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Original Article

Simultaneous determination of some common food dyes in commercial products by digital image analysis

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ABSTRACT

A simple and relatively fast image-analysis method using digital images, obtained with a flatbed scanner, has been described. The method was used for the simultaneous determination of four common food dyes, namely, carmoisine, brilliant blue, sunset yellow, and quinoline yellow, in binary mixtures in commercial products without a need for any prior separation steps. The results obtained were validated against a standard high-performance liquid chromatography method and a good agreement was obtained. The parameters affecting the experimental results were optimized. Under the optimal conditions, the method provided acceptable linear ranges (20–250 mg/L) with correlation coefficients higher than 0.998, suitable precision (relative standard deviation $\leq$ 4.5%), and limits of detection between 4.82 and 8.05 mg/L.

1. Introduction

Food additives are commonly used in processed foodstuff to improve appearance, flavor, taste, color, texture, nutritive value, and preservation. Color is the first quality attribute of food evaluated by consumers, and is therefore an important component of food quality relevant to market acceptance. Although increasing evidence in recent years indicates that the abuse of dyes may cause adverse effects, many kinds of dyes are still widely used due to their low price, high effectiveness, and excellent stability. Therefore, determination of food dyes is important to quality control and control the amount of use permitted [1]. Numerous methods such as sensitive spectrophotometry [2], differential spectrophotometry [3], derivative spectrophotometry [4], spectrophotometry using chemometrics [5,6], colorimetry using a homemade double-beam photocolorimeter [7], reflectometry using a homemade reflectometer [8], electrochemical methods [9,10], capillary electrophoresis [11], ion-pair high-performance liquid chromatography (HPLC) [12], electrokinetic chromatography [13], supercritical fluid extraction–capillary liquid chromatography (LC) [14], thin-layer chromatography [15], HPLC–mass spectrophotometry (MS) [16], solid-phase

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extraction–HPLC [17], HPLC–diode-array detector detection [18,19], LC–MS [20], LC–hybrid linear analysis presented by Goicoechea and Olivieri [21], and gas chromatography–MS [22] have been developed for determination or resolving mixtures of colorants in foodstuff.

Image analysis is the extraction of meaningful information from images, especially from digital images by digital-image-processing techniques. Several techniques have been used for obtaining digital images for different purposes. These techniques include some types of spectroscopy and obtaining images using digital color cameras and scanners [23,24].

The aim of this work is to propose a simple and cheap image-analysis method using flatbed scanners for simultaneous determination of some food colors [quinoline yellow (QY), sunset yellow (SY), carmoisine (CA), and brilliant blue (BL)] in commercial food products. The flatbed scanners in comparison with other imaging devices are relatively inexpensive with the ability to digitize images into a stored array of pixels within a computer. Therefore, the proposed method proved to be simple, relatively fast, and economical for the analysis of the target analytes.

2. Methods

2.1. Chemicals

Dyes of purity higher than 95% were purchased from Grodab Chemie GmbH (Hamburg, Germany). The dyes used were QY, SY, CA, and BL. All other chemicals were obtained from Merck (Germany) and were of the analytical reagent grade. Standard solutions (1000 mg/L) of dyes were prepared using pure water.

2.2. Apparatus and software

The experiments were carried out using a flatbed scanner (LiDE20; Canon, China). All HPLC measurements of colors were carried out using a liquid chromatograph (Waters, Milford, MA, USA), which was equipped with the following: Waters 515 pump, Waters 717plus autosampler, and Waters 2487 dual-wavelength absorbance detector (double channel). A filter paper (ALBET ashless), a thin-chromatography Whatman paper (1 Chr Whatman paper, Thickness, 0.18 mm, UK), and a thick Whatman paper (No. 43; thickness, 0.23 mm, UK) were used as spotting surfaces. Photoshop CS5 (Adobe Systems) and MATLAB 7.0 (MathWorks, Natick, MA, USA) were used for investigating the color intensities from the saved images and calculations, respectively.

2.3. Preparation of the real sample solutions

The proposed method was applied for determination of QY, SY, CA, and BL dyes present as individual components or in binary mixtures within commercial products. The paper chromatography method was used for identification of food dyes in commercial products. In this method, the chromatogram is developed by applying aqueous samples and standard dye solutions to a normal-phase paper. The solutions were developed in the mobile phase of sodium citrate (2.4% w/v):pure ethanol (1:1). Retention factor values, that is, the ratio of distance traveled by the dye to the distance traveled by the solvent, were estimated for the samples and the standard solutions. Various solid jelly powders (Farmand; Iran) and traditional chocolate (Morvarid, Minoo, Iran) samples were purchased from local supermarkets (Tabriz, Iran). An unnamed shoddy orange soft-drink sample was supplied by the Quality Control Laboratory of the Food and Drug Department of Tabriz University of Medical Sciences. Approximately 6 g of color-coated chocolates (shells) were transferred into a flask. The colored shells were then dissolved in distilled water, centrifuged, and diluted with an equal volume of 3M CH_3COOH solution. Approximately 200 mg (per 10 mL of sample) of white commercial wool yarn was washed with detergent, rinsed with distilled water, dried, and then added to the aforementioned diluted sample, and the mixture was heated (60 minutes at 60°C or 10 minutes at 90°C). The colored yarn was then taken out, and washed with plenty of distilled water. The dyes were recovered by mixing the yarns with 10 mL of NH_3 (2M) and heating for 10 minutes at 90°C.

2.4. Analytical procedures

Different concentrations of dye samples were prepared by appropriate dilutions of stock solutions (1000 mg/L) with deionized water (Ghazi Company, Tabriz, Iran). To prepare the color spots of the dyes, the prepared solutions (20 μL) were spotted on Whatman No. 43 paper, left for a while (40 minutes), and placed on the glass plate of the flatbed scanner. The spots on the paper were separated by Microsoft Paint (XP) software. The selected images were saved in the bitmap format composed in the three channels of the RGB model using Photoshop. The magnitude of each component (red, green, and blue) was determined, and the color value was calculated by subtracting the suitable RGB channel intensities of samples from those of the white sheet. This procedure was performed using MATLAB.

2.5. High-performance liquid chromatography

The aforementioned method was validated by running a parallel HPLC test for the commercial food products. Measurements were made at 460 nm for CA, SY, and QY and at 640 nm for BL using NaH_2PO_4/Na_2HPO_4 buffer solution (pH = 6.0); acetonitrile (65:35 v/v) was used as the mobile phase with a flow rate of 0.9 mL/minute at room temperature. The retention times were 2.91, 3.06, and 3.51 minutes at the first channel (460 nm) for QY, SY, and CA, and 3.47 minutes at the second channel (640 nm) for BL. No efficient interaction between BL and CA was observed as they had similar retention times.

3. Results and discussion

3.1. The RGB color model and selection of the suitable RGB component

A digital image is intrinsically a multivariate system, which is a collection of data stored in pixels, each usually highly correlated to its neighbors [25]. The numerical information of