



# Single and Simultaneous Solid-Phase Extraction and UV–Vis Determination for Monitoring E129, E133 and E110 in Foodstuffs

Abdullah Taner Bişgin<sup>1</sup>

Received: 3 June 2020 / Accepted: 15 September 2020 / Published online: 26 September 2020  
© Shiraz University 2020

## Abstract

A column solid-phase extraction method based on adsorption of allura red (AR), brilliant blue (BB) and sunset yellow (SY) onto Dowex Optipore V493 adsorbent was developed for single and simultaneous preconcentration and spectrophotometric determination of AR, BB and SY. Effects of extraction parameters including pH, sample and eluent flow rates, amount of adsorbent, eluent type and volume, ionic strength and sample volume, etc., were investigated and optimized. At the optimum conditions, detection limits of the method were determined to be 0.55, 0.20 and 0.58  $\mu\text{g L}^{-1}$  for AR, BB and SY, respectively. Dynamic ranges were linear between 0.055–6.0  $\mu\text{g mL}^{-1}$  for AR, 0.020–4.2  $\mu\text{g mL}^{-1}$  for BB and 0.058–10.0  $\mu\text{g mL}^{-1}$  for SY. Preconcentration factor was determined to be 100. Relative standard deviations were below than 4%. In addition, adsorption isotherms with related to retentions of AR, BB and SY on the resin were investigated. Finally, recommended procedure was applied to foodstuff samples containing AR, BB and SY dyes. AR contents of real samples were determined between 20.40–27.21  $\mu\text{g/mL}$  and 6.85–21.25  $\mu\text{g/g}$  for liquid and solid samples, respectively. BB concentrations of real samples were determined between 5.06–43.30  $\mu\text{g/mL}$  and 3.15–10.05  $\mu\text{g/g}$  for liquid and solid samples, respectively. SY contents of liquid and solid samples were determined between 3.04–29.67  $\mu\text{g/mL}$  and 18.55–121.46  $\mu\text{g/g}$ , respectively. In order to prove applicability of the method, analyte addition technique was performed. Satisfactorily quantitative recovery results were obtained between 95 and 102%.

**Keywords** Allura red · Brilliant blue · Sunset yellow · Solid-phase extraction · Preconcentration

## 1 Introduction

Color is used in foodstuffs to give them charming and attractive visibility and to give them allure features (Rovina et al. 2016). Allura red (AR), brilliant blue (BB) and sunset yellow (SY) are the synthetic food dyes which are manufactured from petroleum and its derivatives and they have been known as food additives with E129, E133 and E110 food codex numbers for AR, BB and SY, respectively (Khorshidi and Niazi 2018). These are the most used and preferred dyes in foodstuffs because each of mentioned dyes have an impressive and attractive appearances. In addition, these synthetic dyes are among the third group chemicals which are listed in International Agency for

Research on Cancer (IARC) (<https://monographs.iarc.fr/list-of-classifications>). The substances which belong to list of IARC 3th Group chemicals are not classifiable as to its carcinogenicity to humans. Although these food dyes have not carcinogenic effect on human, the use of food dyes in foodstuffs could lead to some drawbacks for human health and adverse effect on human. The use and overuse of these food dyes in foodstuffs could cause to diverse health problem in adults and children who consumes excessive amount of these foodstuffs (Wang et al. 2015). The health problems including eczema, skin irritation and asthma in the adults and hyperactive behavior disorders in children could be observed (Gosetti et al. 2013). For this reason, WHO has defined the limited concentration for each food dye which can be consumed for per day. The identified limited concentration value has been known as acceptable daily intake (ADI). These ADI values are 7, 6 and 4  $\text{mg kg}^{-1}$  per day on a basis of human body weight for AR, BB and SY, respectively (ECFA 2016; ECFA 2017).

✉ Abdullah Taner Bişgin  
atbisgin@ohu.edu.tr

<sup>1</sup> Ulukışla Vocational School, Niğde Ömer Halisdemir University, 51900 Nigde, Turkey

For these reasons detection and determination of food dyes in foodstuffs are necessary and have a great importance.

Various scientific analytical instruments including UV–Vis spectrophotometer (El-Sheikh and Al-Degs 2013), high-performance liquid chromatography (Vidal et al. 2018), spectrofluorometer (Fu et al. 2015), electrochemical voltammetric analyzer (Gao et al. 2016) and mass spectrometer (Tamim et al. 2020) were used to detect and determine dye contents of foodstuffs. Nevertheless, during the measurements of dye concentrations various limitations and obstacles could be observed. One of the problems is that food samples can contain at a very low-level dye concentration which is below than dye detection limit of the instrument. Instrument cannot detect and determine dye content of food samples. Second one is the either positive or negative interference effect of matrix components (Soylak et al. 2011). Because of interfering species dye concentrations of foodstuffs can be measured higher or lower than they contain. Therefore, dye concentrations of the foodstuffs cannot be detected and determined accurately (Bişgin et al. 2015a). Due to these problems, separation and preconcentration methods are needed.

In order to solve these obstacles and limitations, analytical chemists have developed various separation, purification and preconcentration methods including solid-phase extraction (SPE) (Oymak et al. 2020), cloud point extraction (Shokrollahi and Ahmadi 2017), dispersive liquid–liquid microextraction (Unsal et al. 2015), deep eutectic solvent extraction (Uzcan and Soyvak 2020), dispersive solid-phase microextraction (Dil et al. 2016) and liquid–liquid microextraction (Rovina et al. 2017). Among these sample extraction and preconcentration methods SPE is simple, consistent, robust, precision, stable and very sensitive for organic dyes which have high adsorption affinity to solid phase (Behbahani et al. 2014a). It is possible to operate with samples which have high salinity matrices because resin can be washed with water after adsorption process of target analyte on the resin. Hence, foreign ions can be eliminated from working medium. Moreover, the SPE method provides working possibility at higher sample volume than other separation methods. Thus, higher preconcentration factors and low detection limits can be obtained. In addition, solid-phase material has high adsorption capacity and can be used many times by regenerating with appropriate solvent (Behbahani et al. 2014b).

SPE combined UV–Vis spectrophotometry is very simple, suitable, fast and practical for extraction and determination of the food dyes among these instrumental techniques and methods. In order to operate a UV–Vis spectrophotometer, high operation experience does not require and it is not needed highly experienced and skilled analyst. On the other hand, UV–Vis spectrophotometer

price is lower than other instruments and it is available in lots of laboratory. These features make the SPE combined UV–Vis spectrophotometry more attractive and preferable compared with other methods and techniques (Behbahani et al. 2015).

Dowex Optipore V493 highly cross-linked synthetic styrenic polymer which has excellent adsorption capacity for organic dyes was used as a solid-phase material. The polymeric adsorbent V493 was chosen because it has hydrophobic considerably high adsorbent surface, unique pore size and spherical beads with good physical strength. It is insoluble in strong acids, strong bases and organic solvents. V 493 is powerful adsorbent that can be recycled through a regeneration cycle prior to use to extract organic dyes from aqueous solution. The usage of highly cross-linked polymeric adsorbent in the solid-phase application for extraction, determination, preconcentration and adsorption of food dyes is of great importance. Therefore, there are many SPE applications on adsorption of food dyes onto hypercrosslinked polystyrene resin in the literature (Alkahtani et al. 2017; Tikhomirova et al. 2018).

In this study, SPE method was developed for single determination of AR, BB and SY and simultaneous determination of AR–BB and BB–SY. V493 highly cross-linked synthetic styrenic polymer was used for this purpose. Extraction parameters were separately examined for each of the dyes and optimized in detail. Foodstuff samples containing AR, BB and SY were subjected to the developed method and their dye contents were determined by UV–Vis spectrophotometry. Finally, analytical features of the method for each of the dyes were compared with each other and recent reported studies.

## 2 Materials and Methods

### 2.1 Chemicals

Dowex optipore V493 (Dow Chemical Company) commercially available polymeric adsorbent is highly cross-linked synthetic styrenic polymer resin that is insoluble in strong acid, strong base and organic solvents. It has considerably high bet surface area which is  $1100 \text{ m}^2 \text{ g}^{-1}$ . It has 20–50 mesh size and orange to brown spheres physical form (<https://www.lenntech.com/Data-sheets/Dowex-Opti-core-L493-L.pdf>). It was used throughout the extraction and preconcentration studies.

All commercial available analytical grade chemicals used in the experiments were purchased from Merck (Merck, Darmstadt, Germany) and Sigma (Sigma–Aldrich, St. Louis, MO, USA) and they were used without purification process.

In the preparation of buffer solutions,  $\text{H}_3\text{PO}_4\text{-NaH}_2\text{PO}_4$  for pH 2, 3,  $\text{HAc-NaAc}$  for pH 4, 5 and  $\text{NaH}_2\text{PO}_4\text{-Na}_2\text{HPO}_4$  for pH 6–8 were used to fix the aqueous media pH.

Each of  $100 \mu\text{g mL}^{-1}$  of AR, BB and SY stock dye solutions were prepared and stored in refrigerator at  $4^\circ\text{C}$ . More diluted dye solutions were prepared daily from these stock dye solutions.

## 2.2 Instruments

A Shimadzu Model UV-160A double-beam UV-Vis spectrophotometer (Shimadzu Corporation, Kyoto, Japan) was used to measure of absorbance and absorption spectra of AR, BB and SY. pHs of the buffer and aqueous sample solutions were measured and controlled with Hanna Model HI-2211 digital pH meter (Hanna Instruments, Woonsocket, RI, USA). The chromatographic mini glass column (Çalışkan Laboratuvar Ürünleri Ltd. Şti., Ankara, Turkey) has 1 cm in diameter, 10 cm in length and 500 mL reservoir with Teflon stopcock was used in extraction and pre-concentration procedures. In order to provide stable sample and eluent flow rate, the LongerPump YZ1515x BT100-2J model peristaltic pump (Longer Precision Pump Co., Ltd., Hebei, China) was combined with mini chromatographic glass column. Olympus SZX7 (Olympus Corporation, Tokyo, Japan) model industrial stereo microscope was used to take micrographs of the dye adsorbed form and natural form of V493 resin.

## 2.3 Solid-Phase Extraction Procedure

The chromatographic glass separation column was loaded with 350 mg of V493 resin. The resin was washed with water after each organic solvent was passed through the column. Acetone and water, methanol and water were sequentially passed through the column to remove impurities and other contaminants. Each of solvents volumes used in the experiments to remove impurities from the column was 25 mL.

Each of 25 mL of sample solutions containing  $10 \mu\text{g}$  AR,  $3.5 \mu\text{g}$  BB and  $10 \mu\text{g}$  SY dyes were prepared separately. These solutions were buffered to pH 4 which has 0.2 mol/L of buffer concentration and 0.3 mol/L ionic strength concentration by adding  $\text{HAc/NaAc}$  binary buffer chemicals and NaCl. The solutions were separately poured into reservoir of the column. Sample and eluent flow rates were controlled only using peristaltic pump and by disabling stopcock of the chromatographic column. It was permitted that solution passed through the column under vacuum by using the peristaltic pump by applying at 2 mL/min sample flow rate each of adsorbed dye molecules on the resin was eluted to 4 mL of final volume with pure methanol solution by controlling flow rate with the

peristaltic pump and applying at 2 mL/min eluent flow rate. AR, BB and SY concentrations of the eluent solutions were determined separately at 506 nm, 630 nm and 483 nm, respectively.

## 2.4 Adsorption Experiments

Batch adsorption experiments were performed separately for each of target dyes to investigate adsorption behavior between dyes and resin and to explain adsorption mechanism of the method. Experiments were conducted in a 15 mL polyethylene plastic centrifuge tube with cap, at room temperature, by using pH 4 buffer solution and contacting with 25 mg of DOV493 resin for 4 h. Adsorption equilibrium between dyes and resin was reached after shaking by a shaker for 4 h. The suspensions were centrifuged and dye concentrations of homogenous aqueous phases were determined at 506, 630 nm and 483 nm for AR, BB and SY, respectively.

## 2.5 Pretreatment of Foodstuff Samples

The certain amounts of solid foodstuff samples were dissolved in pure water. Solutions were subjected to the optimized method after filtration process and necessary dilutions.

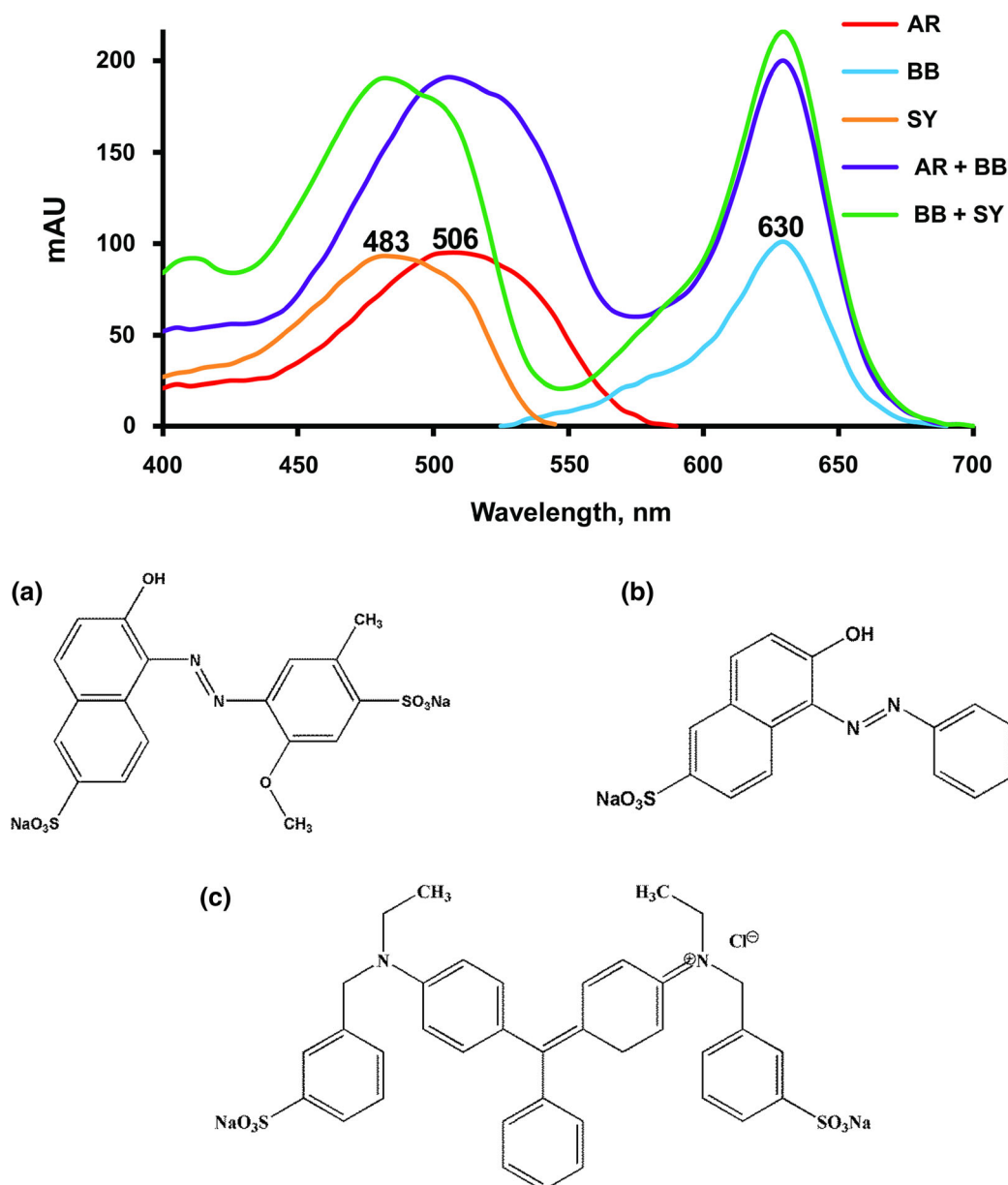
The certain volumes of liquid foodstuff samples were directly applied to the method after necessary dilutions.

## 3 Results and Discussion

In the complete study, in order to well-understand and see easily, all figures are given according to original color of the target dyes. Red, blue and yellow-orange colors symbolize AR, BB and SY, respectively. Any interference effects were observed between AR–BB and BB–SY binary dyes. The UV-Vis spectra of AR, BB, SY, mixture of AR–BB and mixture of BB–SY are given in Fig. 1 with their molecule structures. Each of experimental parameters of the method were investigated and optimized separately for each of the dyes by applying three replicates. First, whole optimization processes were performed separately to obtain quantitative extraction for each of the dyes and then simultaneous preconcentration and determination of AR–BB and BB–SY were examined and performed.

### 3.1 Effect of pH

In column SPE studies, pH is the most important parameter in view of adsorption of organic dyes onto resin. Applied pH must provide complete adsorption of dye molecules onto resin. Effect of pHs on the extraction were separately

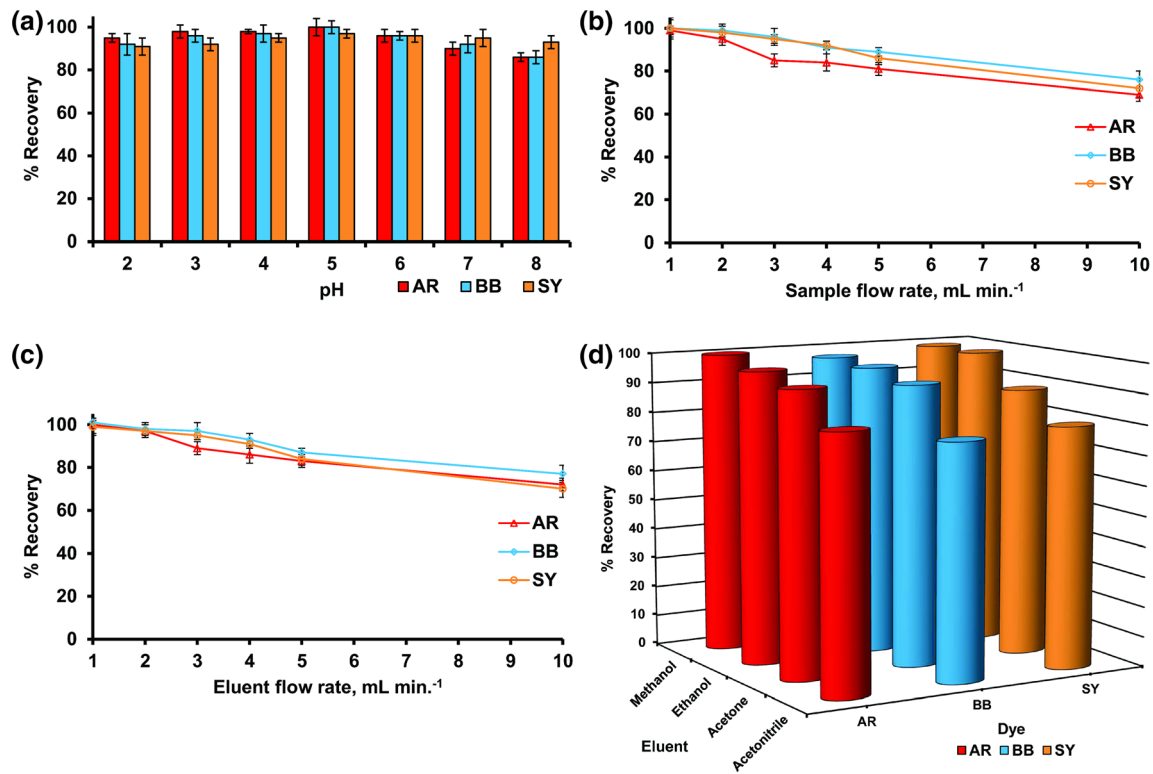


**Fig. 1** UV–Vis spectra of AR, BB, SY, mixture of AR–BB and mixture of BB–SY; molecule structures of AR (a), SY (b) and BB (c)

investigated for the each of the dyes between pH 2 and pH 8 by applying and keeping constant the optimum values of the other parameters. Results are given in Fig. 2a with bar graphic and standard deviations. Quantitative extractions were obtained between pH 2 and pH 6 for AR, pH 3–6 for BB and pH 4–7 for SY dyes. Therefore, optimum pH values were selected 5 for all the three target dyes.

Hydrophobic interactions between resin and dyes are the main leading force which is affected the adsorption. As mentioned previously in V493 characteristics, the resin has hydrophobic feature. Therefore, the resin has no  $pH_{pzc}$  thus; there are no electrostatic interactions between resin

and the dyes. The reason of difference in optimum pH ranges between AR and SY is difference of hydrophobic interactions between resin and the dyes. AR and SY dyes have the almost similar molecular structures except one methoxy and a methyl groups which belong to AR molecule. These alkoxy and alkyl groups make the AR molecule more hydrophobic than SY molecule. In addition, molecular weight of AR is slightly higher than SY molecule. Therefore, the resin has higher adsorption affinity to AR than SY. Because of all these reasons, difference in optimum pH ranges between AR and SY was observed (Bişgin 2019; Rukosueva et al. 2020).



**Fig. 2** Effect of pH (a), sample flow rate (b), eluent flow rate (c) and type of eluent (d) on the single extractions of AR, BB and SY,  $N = 3$

### 3.2 Effect of Sample Flow Rate

In column solid-phase extraction studies, sample flow rate is very important parameter to provide complete retention of analyte onto adsorbent. In order to investigate effect of sample flow rate on the extraction of AR, BB and SY, sample flow rate was applied between 1 and 10 mL min<sup>-1</sup> by keeping constant the optimum values of the other extraction parameters. Results are comparatively given in Fig. 2b for the dyes with their standard deviations. Recoveries of the examined dyes decreased with increasing sample flow rate. Quantitative recoveries were obtained between 1 and 2 mL min<sup>-1</sup> for AR, 1 and 3 mL min<sup>-1</sup> for both BB and SY dyes. For this reason, 2 mL min<sup>-1</sup> of sample flow rate was applied for three dyes in all further extraction studies to save time and to apply fast extraction.

### 3.3 Effect of Eluent Flow Rate

The eluent flow rate is important parameter in column SPE studies to obtain quantitative results because each of dyes must be eluted one by one or simultaneously as quantitative. To examine the effect of eluent flow rate on the extraction of AR, BB and SY, eluent flow rate was separately investigated for the each of the dyes between 1 and 10 mL min<sup>-1</sup> by keeping constant the optimum values of the other extraction parameters. Results are depicted in

Fig. 2c with standard deviations. Recoveries of the dyes decreased with increasing eluent flow rate. Quantitative recoveries were obtained between 1 and 2 mL min<sup>-1</sup> for AR, 1 and 3 mL min<sup>-1</sup> for both BB and SY dyes. For this reason, 2 mL min<sup>-1</sup> of sample flow rate was applied for the three dyes in all further extraction studies to save time and to apply fast extraction.

### 3.4 Effect of Eluent Type

Type of eluent solution is another important parameter to obtain high extraction efficiency and to desorb complete dye molecules from the resin. Effects of various eluents on the extraction were separately investigated for the each of the dyes by keeping constant the optimum values of the other parameters. In order to desorb AR, BB and SY molecules adsorbed on the resin, different pure organic solvents including acetonitrile, acetone, ethanol and methanol were used. Spectrophotometric absorbance measurements of AR, BB and SY were performed separately for each solvent in presence of used own elution solvent. Recovery values were given in Fig. 2d with bar graphics. Quantitative recovery results were obtained for all three target dyes when used ethanol and methanol as an elution solvent. Therefore, methanol was selected to be optimum eluent and all further preconcentration studies were performed by using methanol.

### 3.5 Effect of Dowex Optipore V493 Amount

Adsorbent amount in the column solid-phase extraction is very important to obtain both quantitative retention and quantitative elution of the dyes. Optimum adequate amount of adsorbent must be loaded to the column for retention of complete dye molecules onto resin by adsorption and to provide stabile flows through the chromatographic column for both sample and eluent solutions. Amount of V493 was examined between 50 and 400 mg by keeping constant the optimums of the other variables. Resin amount experiments were performed separately for each of the target dyes. Results are given comparatively for the three dyes in Fig. 3a. The axis of resin amount values are given in

reverse order to understand and see recovery values in detailed. Extraction efficiency increased with increasing resin amount up to 350 mg and then remained constant. Consequently, extraction column was loaded 350 mg of V493 resin.

### 3.6 Effect of Ionic Strength

Impact of ionic strength on the single extraction of AR, BB and SY was performed separately for AR, BB and SY dyes by keeping constant the other optimum variables with using NaCl at different concentration ranges between 100 and 500 mM. Results are given in Fig. 3b. The axis of ionic strength values are given in reverse order to

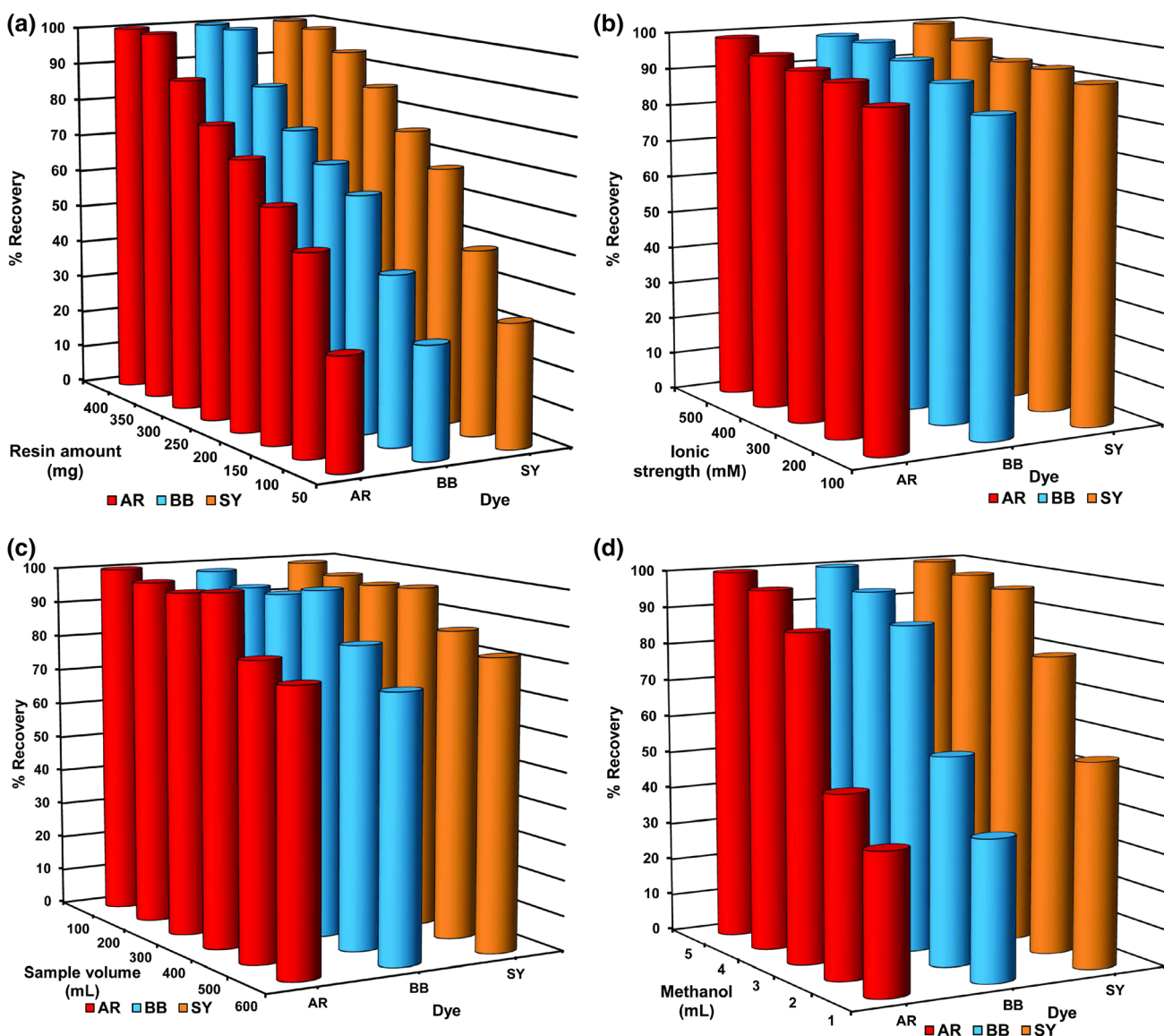


Fig. 3 Effects of resin amount (a), ionic strength (b), sample volume (c) and eluent volume (d) on the single extractions of AR, BB and SY, N = 3

understand and see recovery values in detailed. Recovery values of the all three dyes increased with increasing ionic strength concentration up to 300 mM and reached plateau and remained constant. Therefore, 300 mM ionic strength was kept optimum and the optimum value of 300 mM was applied in all further extraction studies.

### 3.7 Sample Volume

Sample volume is one of the important parameters for preconcentration studies to obtain high preconcentration factor. Detection limits of the method for the dyes decrease with increasing preconcentration factor by depending on sample volume and eluent volume. Therefore, samples containing AR, BB and SY dyes which have very low-level dye contents can be determined as more sensitive. Influence of sample volume on the simultaneous extraction of BB and SY was examined between 25 and 300 mL of sample volume by keeping constant the optimum values of the other parameters. Results are given in Fig. 3c with standard deviations. Single quantitative extraction values were observed up to 400 mL of sample volume for the three analyte dyes. At higher than 400 mL sample volume, sample solution behaved to be an eluent. Therefore, recovery results decreased with increasing sample volume at an amount higher than 400 mL. As a result of these experiments and according to 4 mL of eluent volume, 100 preconcentration factor was obtained for the three analyte dyes.

### 3.8 Eluent Volume

Eluent volume is as important as sample volume parameter in order to gain higher preconcentration factor (PF). Optimum eluent methanol was used to optimize eluent volume. For this purpose eluent volume was examined between 1 and 5 mL by keeping constant the other optimum extraction parameters. Investigation of eluent volume was performed separately for each of the dyes. Results are presented in Fig. 3d by giving methanol volume in reverse order to be shown more understandable recovery values. Quantitative extractions of AR, BB and SY dyes were obtained by applying 4 and 5 mL of eluent volume. In order to obtain higher PF, 4 mL methanol was selected as optimum elution volume.

### 3.9 Adsorption Behavior of the Method

Adsorption isotherms help to describe the interaction between sorbent and dye molecules and are useful to determine adsorption behaviors of the method. Adsorption isotherms with related to retentions of AR, BB and SY onto

V493 resin were separately investigated for each of the dyes.

Obtained experimental results and calculations have been separately applied to fit Langmuir and Freundlich adsorption isotherm models for each of the dyes. Perfect compatibilities with high correlation coefficients were observed with Freundlich model. The Freundlich model was found to be the most appropriate to describe the adsorption process of AR, BB and SY dyes on V493 with suggesting that multilayer adsorption process occurs on the heterogeneous distribution of active sites onto V493 surface. Adsorption isotherms with related to adsorption of AR, BB and SY are given comparatively in Fig. 4a–c.

In order to support adsorption of the all three dyes and mixtures of the dyes on the surface of V493, surface micrographs of the resin was also taken with industrial stereo microscope. AR, BB and SY adsorbed resins are given in Fig. 4d–f, respectively. Pure resin micrograph is given in Fig. 4g. Mixtures of AR–BB and BB–SY adsorbed resins are given in Fig. 4h, i, respectively. The dye adsorbed colorful resin micrographs showed the quantitative applicability of the method to real samples containing AR, BB, SY, mixtures of AR–BB and BB–SY.

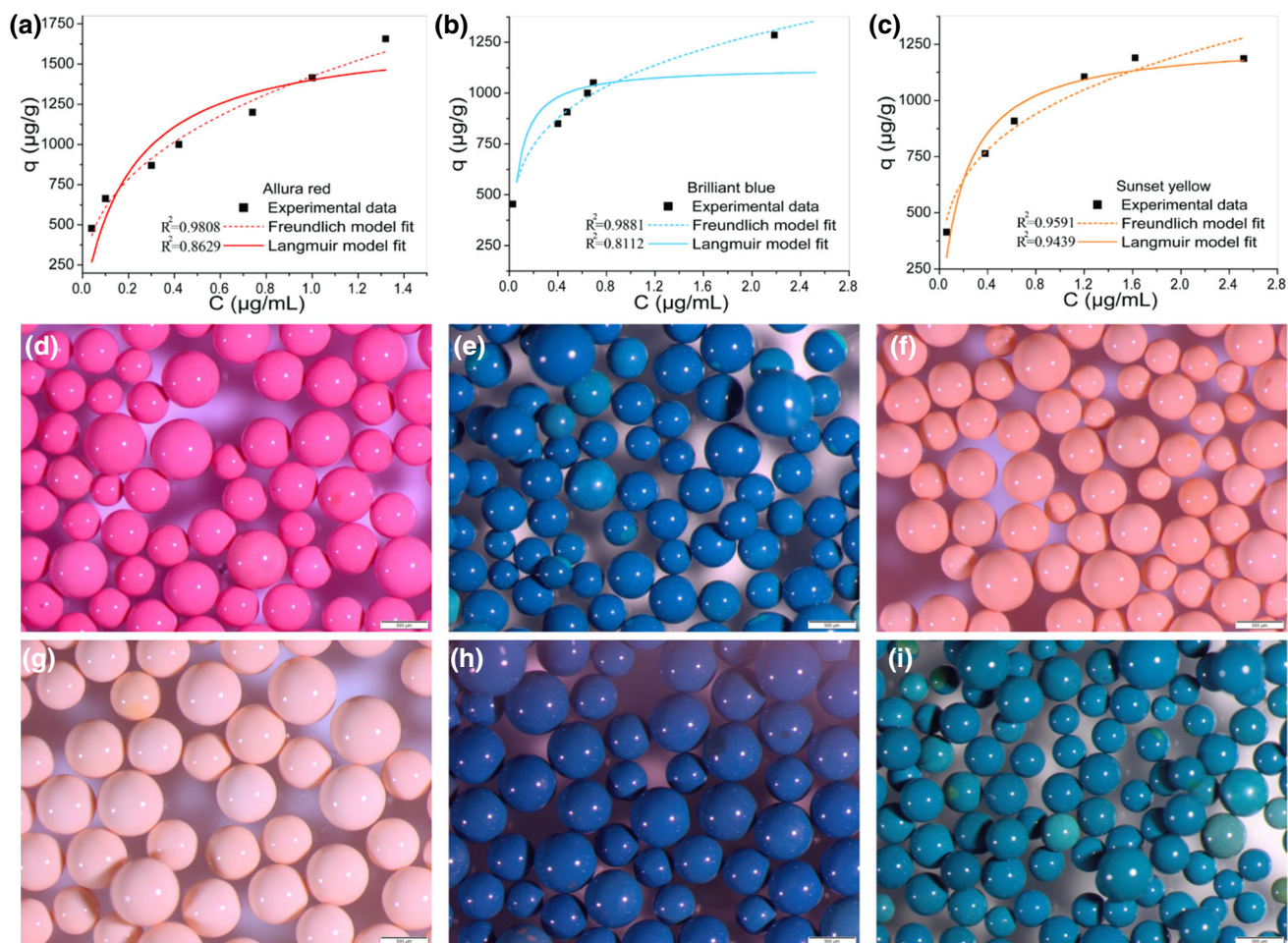
### 3.10 Effect of Matrix Components

Potentially interfering species including dyes and ions, which could be found in foodstuffs, effect the accurate determination of dye concentration. Interference effects of commonly used dyes and naturally found ions in foodstuffs were investigated by applying optimum conditions. The examined concentrations of the interfering species were also given in Table 1. Investigated interfering ions and dyes at given concentrations in Table 1 have no interference effects on the determination of target dyes. These results suggested the applicability of the developed method for the single and simultaneous extraction and determination of AR, BB and SY in food samples.

### 3.11 Application of Foodstuff Samples

In order to prove applicability of the method red energy drink, bear jelly and red candy samples containing AR dye were subjected to the method. Analyte addition/recovery tests were also applied to the samples by varying added amounts of AR between 2.00 and 4.00  $\mu\text{g}$ . Results are given in Table 2 with standard deviations. Recovery results were obtained between 95 and 101%.

In order to show applicability of the method on determination of foodstuff samples containing BB dye, optimized parameters of the method were applied to blue energy drink, citrus flavored cocktail syrup and blue candy. Analyte addition/recovery tests were also applied to the



**Fig. 4** Adsorption isotherms of the method for AR (a), BB (b) and SY (c); adsorbed AR (d), adsorbed BB (e), adsorbed SY (f), pure resin (g), adsorbed AR–BB (h) and adsorbed BB–SY (i) on the resin

**Table 1** Matrix effect of the interfering species,  $N = 3$

Ion and dye	Added species	Concentration ( $\mu\text{g/mL}$ )	% Recovery		
			AR	BB	SY
$\text{Na}^+$	$\text{NaNO}_3$	100	$98 \pm 3^a$	$97 \pm 3$	$97 \pm 2$
$\text{K}^+$	$\text{KNO}_3$	100	$99 \pm 4$	$98 \pm 3$	$99 \pm 2$
$\text{Mg}^{2+}$	$\text{Mg}(\text{NO}_3)_2 \cdot 6\text{H}_2\text{O}$	100	$96 \pm 3$	$95 \pm 4$	$100 \pm 1$
$\text{Ca}^{2+}$	$\text{CaCl}_2$	100	$102 \pm 2$	$98 \pm 3$	$96 \pm 2$
$\text{Cl}^-$	$\text{NaCl}$	100	$101 \pm 4$	$99 \pm 4$	$100 \pm 3$
$\text{NO}_3^-$	$\text{NaNO}_3$	100	$97 \pm 2$	$98 \pm 2$	$97 \pm 2$
$\text{SO}_4^{2-}$	$\text{Na}_2\text{SO}_4$	50	$98 \pm 3$	$100 \pm 4$	$99 \pm 2$
$\text{PO}_4^{3-}$	$\text{Na}_3\text{PO}_4$	50	$100 \pm 2$	$99 \pm 2$	$98 \pm 2$
Allura red	–	2	–	$101 \pm 3$	<sup>b</sup> CDA
Brilliant blue	–	2	$100 \pm 2$	–	$103 \pm 2$
Ponceau 4R	–	2	CDA	$99 \pm 3$	CDA
Sunset yellow	–	3	CDA	$98 \pm 2$	–
Tartrazine	–	4	CDA	$99 \pm 2$	CDA

<sup>a</sup>Mean  $\pm$  SD

<sup>b</sup>CDA: cannot be determined accurately



**Table 2** Analyte addition/recovery tests for determination of AR in foodstuffs,  $N = 3$ 

Sample	Red energy drink		Bear jelly		Red candy		
	Added ( $\mu\text{g}$ )	Found ( $\mu\text{g}$ )	Recovery (%)	Found ( $\mu\text{g}$ )	Recovery (%)	Found ( $\mu\text{g}$ )	Recovery (%)
–		$1.73 \pm 0.13^a$	–	$3.29 \pm 0.16$	–	$4.15 \pm 0.16$	–
2.00		$3.58 \pm 0.06$	$96 \pm 2$	$5.32 \pm 0.11$	$101 \pm 2$	$6.08 \pm 0.12$	$99 \pm 4$
4.00		$5.66 \pm 0.11$	$99 \pm 4$	$6.96 \pm 0.21$	$95 \pm 3$	$7.78 \pm 0.16$	$95 \pm 4$

<sup>a</sup>Mean  $\pm$  SD

samples by adding 1.00  $\mu\text{g}$  and 2.00  $\mu\text{g}$  of BB dye. Results are presented in Table 3 with standard deviations. Quantitative satisfactorily recovery values were obtained in the range between 95 and 97%.

The determinations of SY concentrations of orange flavored cocktail syrup, syrup medicine and peach juice powdered drink were performed by applying optimized parameters of the developed method. In order to test applicability and repeatability of the method analyte addition/recovery technique was also use. For this purpose, 2.00  $\mu\text{g}$  SY and 4.00  $\mu\text{g}$  SY were added to foodstuff samples. Results are given in Table 4 with standard deviations. Quantitative recovery values were obtained between 96 and 99%.

Finally, simultaneous extractions and determinations of AR–BB and BB–SY were also tested by adding various amounts of AR, BB and SY dyes to diverse matrices samples such as tap water and natural spring water. Results are depicted in Table 5 with standard deviations. Recovery values were obtained between 97 and 102%. These satisfactorily quantitative results showed the applicability and repeatability of the method on the simultaneous extraction and determination of AR–BB and BB–SY in different matrix.

The determined AR, BB and SY concentrations of foodstuff samples are given as  $\mu\text{g g}^{-1}$  and  $\mu\text{g mL}^{-1}$  in Table 6 with standard deviations according to their solid and liquid forms, respectively. AR contents of foodstuffs were determined between 6.85 and 21.25  $\mu\text{g g}^{-1}$  and 20.40 and 27.21  $\mu\text{g mL}^{-1}$  for solid and liquid samples, respectively. BB concentrations were between 3.15 and 10.05  $\mu\text{g g}^{-1}$  for

solid samples and 5.06 and 43.30  $\mu\text{g mL}^{-1}$  for liquid samples. SY levels of solid and liquid foodstuff samples were determined between 18.55–121.46  $\mu\text{g g}^{-1}$  and 3.04–29.67  $\mu\text{g mL}^{-1}$ , respectively.

### 3.12 Analytical Features of the Method

Optimum parameters of the method were determined separately for AR, BB and SY. Under the optimum conditions analytical characteristics including limit of detection (LOD), preconcentration factor (PF), linear dynamic ranges (LDR) were determined separately for each of the dyes. Analytical characteristics and optimum parameters of the method are given comparatively in Table 7 for AR, BB and SY dyes. Reusability study for the V493 resin was also performed. A 350 mg resin was used at least 34 cycles by applying optimum parameters of the method and using methanol as the regeneration chemical.

Analytical specifications of the method was determined and also compared in detail with reported recent studies in Table 8. LOD values of the method were determined 0.55, 0.20 and 0.58  $\mu\text{g L}^{-1}$  for AR, BB and SY, respectively. LOD values of the dyes are lower than reported studies. PF was obtained to be 100 for the all three target dyes. PF was higher than those reported studies. 0.055–6.0  $\mu\text{g mL}^{-1}$ , 0.020–4.2  $\mu\text{g mL}^{-1}$  and 0.058–10.0  $\mu\text{g mL}^{-1}$  linear dynamic ranges (LDR) were determined with correlation coefficient 0.9996, 0.9995 and 0.9993 for AR, BB and SY, respectively. LDR values of the method for the target dyes was comparable with previously reported studies. Relative standard deviations (%RSD) of the method were calculated

**Table 3** Analyte addition/recovery tests for determination of BB in foodstuffs,  $N = 3$ 

Sample	Blue energy drink		Citrus flavored cocktail syrup		Blue candy		
	Added ( $\mu\text{g}$ )	Found ( $\mu\text{g}$ )	Recovery (%)	Found ( $\mu\text{g}$ )	Recovery (%)	Found ( $\mu\text{g}$ )	Recovery (%)
–		$0.95 \pm 0.08^a$	–	$1.23 \pm 0.04$	–	$1.51 \pm 0.06$	–
1.00		$1.85 \pm 0.04$	$95 \pm 3$	$2.16 \pm 0.02$	$97 \pm 2$	$2.39 \pm 0.05$	$95 \pm 4$
2.00		$2.83 \pm 0.06$	$96 \pm 4$	$3.09 \pm 0.04$	$96 \pm 6$	$3.36 \pm 0.04$	$96 \pm 5$

<sup>a</sup>Mean  $\pm$  SD

**Table 4** Analyte addition/recovery tests for determination of SY in foodstuffs,  $N = 3$ 

Sample	Orange flavored cocktail syrup		Syrup medicine		Peach juice powdered drink		
	Added ( $\mu\text{g}$ )	Found ( $\mu\text{g}$ )	Recovery (%)	Found ( $\mu\text{g}$ )	Recovery (%)	Found ( $\mu\text{g}$ )	Recovery (%)
–		$2.17 \pm 0.06^a$	–	$2.85 \pm 0.06$	–	$3.19 \pm 0.11$	–
2.00		$4.11 \pm 0.06$	$99 \pm 4$	$4.73 \pm 0.06$	$98 \pm 2$	$4.98 \pm 0.06$	$96 \pm 6$
4.00		$6.03 \pm 0.10$	$98 \pm 6$	$6.77 \pm 0.06$	$99 \pm 4$	$7.00 \pm 0.06$	$97 \pm 3$

<sup>a</sup>Mean  $\pm$  standard deviation**Table 5** Analyte addition/recovery tests on the simultaneous extraction of AR–BB and BB–SY,  $N = 3$ 

Sample	Add ( $\mu\text{g}$ )			Found ( $\mu\text{g}$ )			% Recovery		
	AR	BB	SY	AR	BB	SY	AR	BB	SY
Tap water	–	–	–	<sup>a</sup> BDL	BDL	BDL	–	–	–
	2.00	1.00	–	<sup>b</sup> $2.01 \pm 0.02$	$0.98 \pm 0.06$	–	$100 \pm 3$	$98 \pm 4$	–
	4.00	2.00	–	$3.97 \pm 0.04$	$2.03 \pm 0.04$	–	$99 \pm 2$	$101 \pm 5$	–
	–	1.00	2.00	–	$1.01 \pm 0.05$	$2.03 \pm 0.04$	–	$101 \pm 3$	$102 \pm 3$
Natural spring water	–	2.00	4.00	–	$1.95 \pm 0.05$	$4.01 \pm 0.05$	–	$98 \pm 4$	$101 \pm 2$
	–	–	–	BDL	BDL	BDL	–	–	–
	2.00	1.00	–	$1.98 \pm 0.04$	$0.99 \pm 0.02$	–	$99 \pm 4$	$99 \pm 2$	–
	4.00	2.00	–	$3.96 \pm 0.05$	$1.94 \pm 0.04$	–	$100 \pm 3$	$97 \pm 3$	–
	–	1.00	2.00	–	$0.96 \pm 0.06$	$1.95 \pm 0.02$	–	$96 \pm 4$	$98 \pm 3$
	–	2.00	4.00	–	$2.00 \pm 0.02$	$3.87 \pm 0.02$	–	$100 \pm 2$	$97 \pm 4$

<sup>a</sup>BDL: Below detection limit<sup>b</sup>Mean  $\pm$  standard deviation**Table 6** AR, BB and SY contents of foodstuff samples

Sample	Concentration		
	AR	BB	SY
Red energy drink	<sup>a</sup> $20.40 \pm 0.61 \mu\text{g mL}^{-1}$	<sup>b</sup> ND	ND
Blue energy drink	ND	$5.06 \pm 0.12 \mu\text{g mL}^{-1}$	ND
Orange flavored cocktail syrup	ND	ND	$16.23 \pm 0.79 \mu\text{g mL}^{-1}$
Caterpillar jelly	$21.25 \pm 0.51 \mu\text{g g}^{-1}$	$3.15 \pm 0.21 \mu\text{g g}^{-1}$	ND
Citrus flavored cocktail syrup	ND	$43.30 \pm 0.81 \mu\text{g mL}^{-1}$	ND
Syrup medicine	ND	ND	$29.67 \pm 1.29 \mu\text{g mL}^{-1}$
Red candy	$8.40 \pm 0.25 \mu\text{g g}^{-1}$	ND	ND
Blue candy	ND	$9.40 \pm 0.38 \mu\text{g g}^{-1}$	ND
Peach juice powdered drink	ND	ND	$121.46 \pm 4.06 \mu\text{g g}^{-1}$
Strawberry flavored cocktail syrup	$27.21 \pm 0.57 \mu\text{g mL}^{-1}$	ND	ND
Mandarin flavored cocktail syrup	ND	ND	$3.04 \pm 0.11 \mu\text{g mL}^{-1}$
Mint flavored cocktail syrup	ND	$7.63 \pm 0.13 \mu\text{g mL}^{-1}$	ND
Worm jelly	ND	$10.05 \pm 0.95 \mu\text{g g}^{-1}$	$18.55 \pm 1.25 \mu\text{g g}^{-1}$
Bear jelly	$6.85 \pm 1.45 \mu\text{g g}^{-1}$	ND	ND

<sup>a</sup>Mean  $\pm$  SD<sup>b</sup>ND not detected

**Table 7** Analytical characteristics and optimums of the method

Parameters	AR	BB	SY
pH	2–6	3–6	4–7
Sample flow rate (mL min <sup>-1</sup> )	1–2	1–3	1–3
Eluent flow rate (mL min <sup>-1</sup> )	1–2	1–3	1–3
Eluent	Methanol	Methanol	Methanol
Eluent volume (mL)	4	4	4
Sample volume (mL)	25–400	25–400	25–400
Preconcentration factor	100	100	100
Limit of detection (µg L <sup>-1</sup> )	0.55	0.20	0.58
Linear dynamic ranges (µg mL <sup>-1</sup> )	0.055–6.0	0.020–4.2	0.058–10.0
Equation	$A = 54.103C - 2.069$	$A = 0.015C + 0.0005$	$A = 0.015C + 0.0005$
$R^2$	0.9996	0.9995	0.9993
RSD (%)	< 3	< 4	< 3

**Table 8** Comparison between analytical features of the method and recently reported methods on the extraction and determination of AR, BB and SY

Sample preparation method/detection	Extractant	Food dye	Real samples	LOD (µg/L)	LDR (µg/mL)	PF	RSD (%)	References
CPE/UV–Vis	CTAB, TX100, TX114	AR	Candy, drinks, jelly, pastel	7.80	0.00–1.40	25	3.9	Pourreza et al. (2011)
SPE/UV–Vis	Amberlite XAD16	AR	Drinks, syrup, juice powder	1.50	0.50–6.00	80	6.0	Bişgin et al. (2015b)
SPE/UV–Vis	Diaion HP20	BB	Energy drink, candy, jelly	0.19	0.05–6.00	100	3.4	Bişgin (2019)
DLLME/UV–Vis	[C10MIM][BF4]	BB	Candy, drink, jelly, ice cream	0.34	0.00–0.15	38	0.8	Guo et al. (2013)
SPE/UV–Vis	Amberlite XAD8	SY	Candy, jelly, fruit juice, syrup	0.66	0.00–10.00	80	4.5	Bişgin (2018)
SPE/UV–Vis	Alkylsilica	SY	Beverages	110	2.00–20.00	20	10.0	Rukosueva et al. (2020)
CPE/UV–Vis	TX100	SY	Soft drink, sweet, gelatin	5.00	0.02–0.45	33	1.5	El-Shahawi et al. (2013)
SPE/UV–Vis	Dowex Optipore V493	AR	Energy drinks, syrups, candies,	0.55	0.055–6.00	100	3.0	Present study
		BB	jellies, medicine	0.20	0.020–4.20	100	4.0	Present study
		SY		0.58	0.058–10.00	100	3.0	Present study

CPE cloud point extraction, UV–Vis UV–visible spectrophotometry, LOD limit of detection, SPE solid-phase extraction, RSD relative standard deviation, LDR linear dynamic range, DLLME dispersive liquid–liquid microextraction, PF preconcentration factor

below than 3%, 4% and 3% for AR, BB and SY, respectively. These RSD values were lower than previously reported studies in Table 8.

## 4 Conclusions

The proposed SPE method is easy, safe, simple, consistent, robust, precision, stable, fast and economic methodology for the either single preconcentration and determination of

AR, BB and SY and simultaneous preconcentration and determination of AR–BB and BB–SY in diverse foodstuff samples. The method offers broad application pH range between 3 and 6 for both single and simultaneous preconcentration of target dyes. Commercially available simple and a little toxic eluents such as ethanol and methanol were used. The experimental quantitative satisfactorily results and data demonstrate that single and simultaneous, reliable determination of AR, BB and SY in different food samples are applicable. Colorful surface micrographs prove

that detecting and determining AR, BB and SY dyes could be performed simultaneously and separately. The mentioned advantages and sufficient features of the method will make the method more useful for future food quality and control applications. Due to its safety, simplicity and practicality, the method can be applied to the either determination of AR, BB and SY or simultaneous determination of AR–BB and BB–SY in diverse foodstuffs.

## Compliance with Ethical Standards

**Conflict of interest** The author declares that he has no conflict of interest.

## References

- Alkahtani SA, Abu-Alrub SS, Mahmoud AM (2017) Adsorption of food coloring allura red dye (E129) from aqueous solutions using activated carbon. *Int J Food Allied Sci* 3:10–19
- Behbahani M, Tapeh NAG, Mahyari M, Pourali AR, Amin BG, Shaabani A (2014a) Monitoring of trace amounts of heavy metals in different food and water samples by flame atomic absorption spectrophotometer after preconcentration by amine-functionalized graphene nanosheet. *Environ Monit Assess* 186:7245–7257
- Behbahani M, Najafi F, Amini MM, Sadeghi O, Bagheri A, Hassanlou PG (2014b) Solid phase extraction using nanoporous MCM-41 modified with 3,4-dihydroxybenzaldehyde for simultaneous preconcentration and removal of gold(III), palladium(II), copper(II) and silver(I). *J Ind Eng Chem* 2:2248–2255
- Behbahani M, Aliakbari A, Amini MM, Behbahani AS, Omid F (2015) Synthesis and characterization of diphenylcarbazide-siliceous mesocellular foam and its application as a novel mesoporous sorbent for preconcentration and trace detection of copper and cadmium ions. *RSC Adv* 5:68500–68509
- Bişgin AT (2018) Simultaneous preconcentration and determination of brilliant blue and sunset yellow in foodstuffs by solid-phase extraction combined UV–Vis spectrophotometry. *J AOAC Int* 101:1850–1856
- Bişgin AT (2019) Simultaneous extraction and determination of allura red (E129) and brilliant blue FCF (E133) in foodstuffs by column solid-phase spectrophotometry. *J AOAC Int* 102:181–188
- Bişgin AT, Uçan M, Narin İ, Soylak M (2015a) A comparative study for separation, preconcentration and determination of tartrazine (E 102) in soft drink samples by two kinds of amberlite resins. *Food Anal Methods* 8:2141–2149
- Bişgin AT, Uçan M, Narin I (2015b) Comparison of column solid-phase extraction procedures for spectrophotometric determination of E129 (allura red) in foodstuff, pharmaceutical, and energy drink samples. *J AOAC Int* 98:946–952
- Dil EA, Ghaedi M, Asfaram A, Mehrabi F, Bazrafshan AA, Ghaedi AM (2016) Trace determination of safranin O dye using ultrasound assisted dispersive solid-phase micro extraction: artificial neural network-genetic algorithm and response surface methodology. *Ultrason Sonochem* 33:129–140
- El-Shahawi MS, Hamza A, Al-Sibaai AA, Bashammakh AS, Al-Saidi HM (2013) A new method for analysis of sunset yellow in food samples based on cloud point extraction prior to spectrophotometric determination. *J Ind Eng Chem* 19:529–535
- El-Sheikh AH, Al-Degs YS (2013) Spectrophotometric determination of food dyes in soft drinks by second order multivariate calibration of the absorbance spectra-pH data matrices. *Dyes Pigments* 97:330–339
- Evaluation of Certain Food Additives (2016) WHO technical report series 1000. World Health Organization, Geneva
- Evaluation of Certain Food Additives (2017) WHO technical report series 1007. World Health Organization, Geneva
- Fu DS, Wu PP, Zhong XD, Liu Q, Luo HD, Li YQ (2015) A simple synchronous fluorescence approach for rapid and sensitive determination of rhodamine B in chilli products. *Food Anal Methods* 8:189–194
- Gao Y, Xie Z, Zhang Y, Zou L, Ye B (2016) A simple and sensitive voltammetric method for the determination of orange II based on a functionalized graphene-modified electrode. *J AOAC Int* 99:1287–1294
- Gosetti F, Chiuminatto U, Mazzucco E, Calabrese G, Gennaro MC, Marengo E (2013) Non-target screening of allura red AC photodegradation products in a beverage through ultra high performance liquid chromatography coupled with hybrid triple quadrupole/linear ion trap mass spectrometry. *Food Chem* 136:617–623
- Guo J, Wu H, Du L, Fu Y (2013) Determination of brilliant blue FCF in food and cosmetic samples by ionic liquid independent disperse liquid–liquid micro-extraction. *Anal Methods* 5:4021–4026
- <https://monographs.iarc.fr/list-of-classifications>. Accessed 23 May 2020
- <https://www.lenntech.com/Data-sheets/Dowex-Opticore-L493-L.pdf>. Accessed 22 May 2020
- Khorshidi N, Niazi A (2018) A novel ion pair based surfactant assisted microextraction modified by orthogonal signal correction partial least squares for determination of food dyes. *J Food Meas Charact* 12:1885–1895
- Oymak T, Tokaloğlu Ş, Cam Ş, Demir S (2020) Determination of color additive tartrazine (E 102) in food samples after dispersive solid phase extraction with a zirconium-based metal–organic framework (UiO–66(Zr)–(COOH)<sub>2</sub>). *Food Addit Contam A* 37:731–741
- Poureza N, Rastegarzadeh S, Larki A (2011) Determination of allura red in food samples after cloud point extraction using mixed micelles. *Food Chem* 126:1465–1469
- Rovina K, Prabakaran PP, Siddiquee S, Shaarani SM (2016) Methods for the analysis of sunset yellow FCF (E110) in food and beverage products- a review. *Trac-Trend Anal Chem* 85:47–56
- Rovina K, Acung LA, Siddiquee S, Akanda JH, Shaarani SM (2017) Extraction and analytical methods for determination of sunset yellow (E110)—a review. *Food Anal Methods* 10:773–787
- Rukosueva EA, Aliyarova GR, Tikhomirova TI, Apyari VV, Nesterenko PN (2020) Simultaneous determination of synthetic food dyes using a single cartridge for preconcentration and separation followed by photometric detection. *Int J Anal Chem*. <https://doi.org/10.1155/2020/2409075>
- Shokrollahi A, Ahmadi S (2017) Determination of trace amounts of Brown HT as a food dye by a CPE-scanometry method. *J Taibah Univ Sci* 11:196–204
- Soylak M, Unsal YE, Yilmaz E, Tuzen M (2011) Determination of rhodamine B in soft drink, waste water and lipstick samples after solid phase extraction. *Food Chem Toxicol* 49:1796–1799
- Tamim AA, AlRabeh M, Tamimi AA, Ajlan AA, Alowaiifeer A (2020) Fast and simple method for the detection and quantification of 15 synthetic dyes in sauce, cotton candy, and pickle by liquid chromatography/tandem mass spectrometry. *Arab J Chem* 13:3882–3888



- Tikhomirova TI, Ramazanova GR, Apyari VV (2018) Effect of nature and structure of synthetic anionic food dyes on their sorption onto different sorbents: peculiarities and prospects. *Microchem J* 143:305–311
- Unsal YE, Soy lak M, Tuzen M (2015) Ultrasound-assisted ionic liquid-based dispersive liquid–liquid microextraction for pre-concentration of patent blue V and its determination in food samples by UV–visible spectrophotometry. *Environ Monit Assess* 187:1–8
- Uzcan F, Soy lak M (2020) Spectrophotometric determination of traces allura red in environmental samples after a deep eutectic solvent-based microextraction. *Int J Environ Anal Chem.* <https://doi.org/10.1080/03067319.2020.1738422>
- Vidal M, Garcia-Arrona R, Bordagaray A, Ostra M, Albizu G (2018) Simultaneous determination of color additives tartrazine and allura red in food products by digital image analysis. *Talanta* 184:58–64
- Wang M, Yang M, Sun Q, Gao Y, Zhao J (2015) Development of a facile sensor for the determination of brilliant blue FCF in beverages. *Int J Environ Anal Chem* 95:969–979