RESEARCH PAPER

Assessment And Validation Of Analytical Methods For Determining Synthetic Food Colors In Bakery And Beverage Products: A Public Health Perspective

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ABSTRACT

Background: Synthetic food colours including Tartrazine, Erythrosine, Brilliant Blue, Allura Red, Sunset Yellow, and Ponceau 4R are extensively used in bakery products, confectioneries, beverages, and processed foods to improve their visual appeal. Although these colourants are permitted within specified regulatory limits, excessive consumption has been associated with hypersensitivity reactions, behavioural disorders in children, and other adverse health effects. Therefore this research focuses on the analysis of commonly used synthetic food colors in locally available food products, focusing on Tartrazine, Erythrosine, Brilliant Blue, Allura Red (Ponceau 4R), and Sunset Yellow. These colors are widely incorporated into bakery items, biscuits, and confectioneries to enhance their visual appeal, particularly for children, making their safety assessment critical. The study aimed to develop a precise and reliable analytical method for the detection and quantification of these dyes and to validate the method in accordance with standard parameters.

Materials and Methods: Representative samples of cakes, biscuits, candies, jellies, syrups, beverages, and confectionery products were collected from local markets. Synthetic dyes were extracted using distilled water, adsorbed onto acid-treated sheep wool, desorbed using ammonium solution, and analysed by UV–Visible spectrophotometry. The method was optimized using distilled water as the extraction solvent. Validation was performed according to ICH guidelines by evaluating linearity, precision, robustness, limit of detection (LOD), and limit of quantification (LOQ).

Results and Discussion: The optimized method showed maximum absorbance at 424.0 nm (Tartrazine), 481.5 nm (Sunset Yellow), 503.0 nm (Allura Red), 508.5 nm (Ponceau 4R), 525.8 nm (Erythrosine), and 629.6 nm (Brilliant Blue). Excellent linearity was achieved with correlation coefficients (R^2) of 0.9991 for Erythrosine and 0.9964 for Brilliant Blue. Precision studies demonstrated excellent repeatability, with intra-day and inter-day %RSD values below 2%. The method exhibited high sensitivity, with LOD and LOQ values of 0.02913 µg/mL and 0.8699 µg/mL for Erythrosine, and 0.1570 µg/mL and 0.4758 µg/mL for Brilliant Blue, respectively. Analysis of commercial food samples revealed that most products complied with Food Safety and Standards Authority of India (FSSAI) permissible limits. However, Tartrazine in Campa (13.4 mg/kg) exceeded the permissible limit of 7.5 mg/kg, while Allura Red in Sting beverage (7.6 mg/kg) exceeded the prescribed limit of 7.0 mg/kg, indicating potential regulatory non-compliance and the necessity for routine monitoring.

Conclusion: The developed UV–Visible spectrophotometric method based on sheep wool adsorption is simple, accurate, precise, sensitive, and cost-effective for the routine determination of synthetic food colours in food products. The validated method meets ICH acceptance criteria and provides a practical alternative to sophisticated chromatographic techniques for quality control laboratories. The detection of samples exceeding FSSAI limits highlights the importance of continuous surveillance, strict regulatory enforcement, and increased awareness among manufacturers and consumers to ensure the safe use of synthetic food colourants and protect public health

Keywords: Synthetic dyes, Bakery products, UV-Visible spectrophotometry, Method validation, Food safety, FSSAI.

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INTRODUCTION

Food colors are additives incorporated into food and beverages to enhance visual appeal or restore appearance lost during processing. They are broadly categorized as natural pigments or synthetic dyes. Synthetic food colors are extensively used owing to their low cost, high stability, and bright intensity. However, uncontrolled consumption of these additives has been linked to various adverse health outcomes, including behavioral changes in children, hypersensitivity reactions, gastrointestinal discomfort, and metabolic complications. [1-3]

Global food safety agencies, including the Food Safety and Standards Authority of India (FSSAI), regulate the acceptable daily intake (ADI) and maximum concentration of permitted synthetic colors. Despite these guidelines, several studies have reported their overuse in locally available food products. Analytical methods such as high-performance liquid chromatography (HPLC) and spectrophotometry are used for their detection. While HPLC is highly sensitive, it is expensive and requires trained personnel. Simpler, validated approaches that are feasible in routine laboratory practice are therefore needed. This study aimed to develop and validate a UV-Visible spectrophotometric method using sheep wool adsorption for the quantification of synthetic dyes in bakery and beverage products, and to assess their compliance with FSSAI regulations [4,5]

Colors are substances added to food to enhance its appearance, making it more visually appealing or to restore the color lost during processing. These colors can be either natural or synthetic. Figure 1& Table 1 shows two main categories of food colors: natural pigments from plants and minerals, and synthetic dyes produced by chemical methods. Examples of both groups are provided for clear comparison.[6] The unrestricted use of synthetic food colorants in commonly available bakery products may pose a range of health concerns, particularly among vulnerable groups such as children, adults, and the elderly as shown in Figure 2.

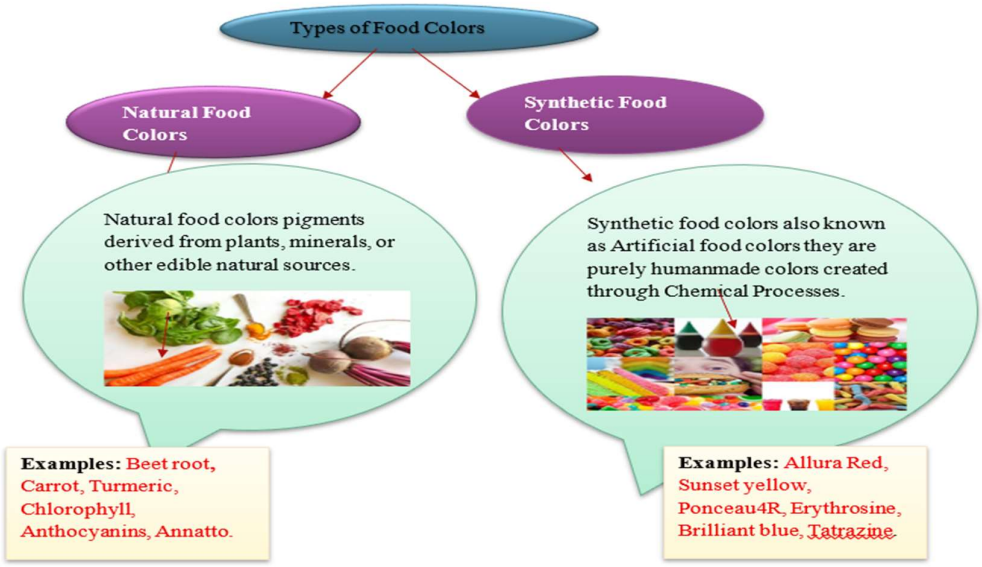

Fig. 1: Types of food colors

Table 1: List of Permitted and Non-Permitted Synthetic food colors.

Permitted Food Colors	Non - Permitted Food Colors
Brilliant blue	Rhodamine B
Erythrosine	Amaranth
Allura red	Orange 2, Orange *G*
Sun set yellow	Metanil yellow
Tartrazine	Acid magenta
Indigo carmine	Butter yellow
Ponceau4R	Malachite green

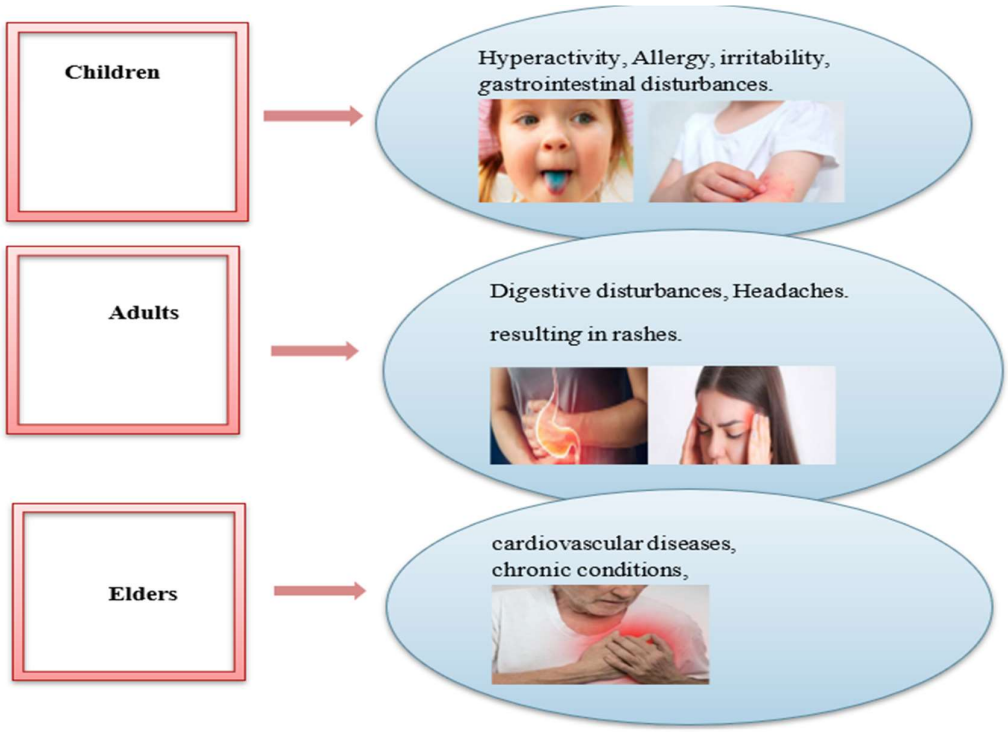

Fig. 2: Health effects of unregulated synthetic food colours on children, adults, elders.

MATERIALS AND METHODS

The sheep wool adsorption procedure was used as a sample clean-up and concentration step. Identification of the target synthetic dye was performed using product label information, visual colour assessment, and comparison of the absorption spectrum with the corresponding reference standard. The proposed method was intended for the determination of individual synthetic food colourants and is not suitable for simultaneous quantification of multiple dyes with overlapping absorption spectra without chromatographic separation.

Sample Collection and Identification

Food samples including cakes, biscuits, jams, jellies, and beverages were collected from local markets to represent commonly consumed products containing added colours. Different brands and batches were purchased in sealed original packaging and labeled with details such as name, batch number, and expiry date. Samples were transported under clean conditions, coded on arrival at the laboratory, and stored at 4 ± 2 °C until analysis. Sheep wool was also collected from local sources, washed to remove impurities, dried at room temperature, and kept in airtight containers for later use. The identification of synthetic colours involved examination of product labels, visual inspection of colour intensity, and analytical testing through spectrophotometric method with results compared against certified dye standards for confirmation. [7-9]

Sample Extraction

Approximately 5 *g* of the food sample was accurately weighed, ground into a fine powder, and transferred into a clean beaker. An appropriate extraction solvent, either ethanol or distilled water, was added, and the mixture was gently agitated for a specific period to facilitate the release of dye pigments. The extract was then filtered to eliminate insoluble residues. Sheep wool threads of about 3*cm* length were prepared by boiling in water for 3–5 minutes and subsequently conditioning with 2 M acetic acid. The treated wool was immersed in the filtrate and maintained in a water bath for about 30 minutes to allow effective adsorption of the dyes. After adsorption, the colored wool was rinsed under running water for 1–2 minutes and transferred into 2Molar ammonium solution. The threads were then reheated in hot water until complete decolorization occurred. The released dye solution was collected, centrifuged, and the clear supernatant was analyzed using UV–Visible spectrophotometry. [10-12]

Method Development

Erythrosine

In the first trial stock solution of Erythrosine was prepared by accurately weighing 10 mg of the standard dye and dissolving it in a 10 *mL* volumetric flask with methanol, resulting in a concentration of 1000 ppm. From this stock, 0.1 *mL* was pipetted and diluted to 10 *mL* with Methanol to obtain a 10ppm working solution. The prepared solution was scanned in spectrum mode to determine the absorption wave length. It was observed weak absorbance with no distinct peak formation. [13,14]

In the second trial standard stock solution of Erythrosine was prepared by accurately weighing 10 mg of the dye and dissolving it in methanol to obtain a final concentration of 1000 ppm. From this solution, 1 *mL* was further diluted to 10 *mL* with distilled water to prepare a working solution. When analyzed under the spectrophotometer, no characteristic peak absorbance was detected at the expected wavelength, indicating the absence of a distinct spectrum.

In the third trial standard stock solution of Erythrosine was prepared by accurately weighing 10 mg of the dye and dissolving it in a 10 *mL* volumetric flask with distilled water, yielding a concentration of 1000 ppm. From this stock, 0.1 *mL* was pipetted into another volumetric flask and diluted to volume with water to obtain a 10ppm working solution. The solution was then scanned in spectrum mode to determine the maximum absorbance wavelength, which was found to be 525.8 nm with an absorbance value of 1.067.

Inference: Trial 3 is optimized with water showed better peak shape and resolution for Erythrosine. Figure 3 shows Spectrum of Erythrosine showing maximum Absorbance at 525.8 nm

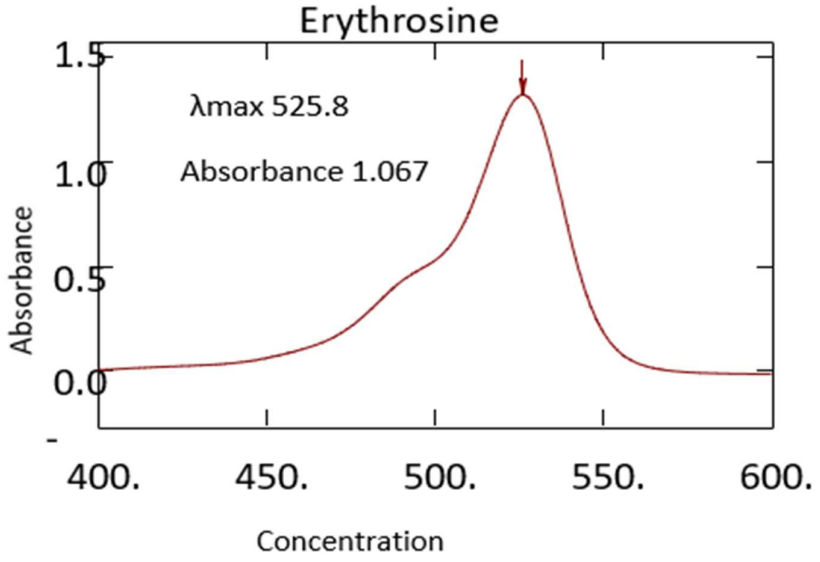

Fig. 3: Spectrum of Erythrosine

Brilliant Blue

In the first trial stock solution of brilliant blue was prepared by accurately weighing 10 mg of the standard dye and dissolving it in a 10 *mL* volumetric flask with Methanol, resulting in a concentration of 1000 ppm. From this stock, 0.1 *mL* was pipetted and diluted to 10 *mL* with Methanol to obtain a 10ppm working solution. The prepared solution was scanned in spectrum mode to determine the absorption wave length. It was observed weak absorbance with no distinct peak formation. [15,16]

In the second trial standard stock solution of brilliant blue was prepared by accurately weighing 10 mg of the dye and dissolving it in methanol to obtain a final concentration of 1000 ppm. From this solution, 1 *mL* was further diluted to 10 *mL* with distilled water to prepare a working solution. When analyzed under the spectrophotometer, no characteristic peak absorbance was detected at the expected wavelength, indicating the absence of a distinct spectrum.

A standard stock solution of brilliant blue was prepared by accurately weighing 10 mg of the dye and dissolving it in a 10 *mL* volumetric flask with distilled water, yielding a concentration of 1000 ppm. From this stock, 0.1 *mL* was pipetted into another volumetric flask and diluted to volume with water to obtain a 10ppm working solution. The solution was then scanned in spectrum mode to determine the maximum absorbance wavelength, which was found to be 629.6 nm with an absorbance value of 1.494.

Inference: Trial 3 is optimized with water showed better peak shape and resolution for brilliant blue. Figure 4 shows Spectrum of Brilliant blue showing maximum Absorbance at 629.6 nm

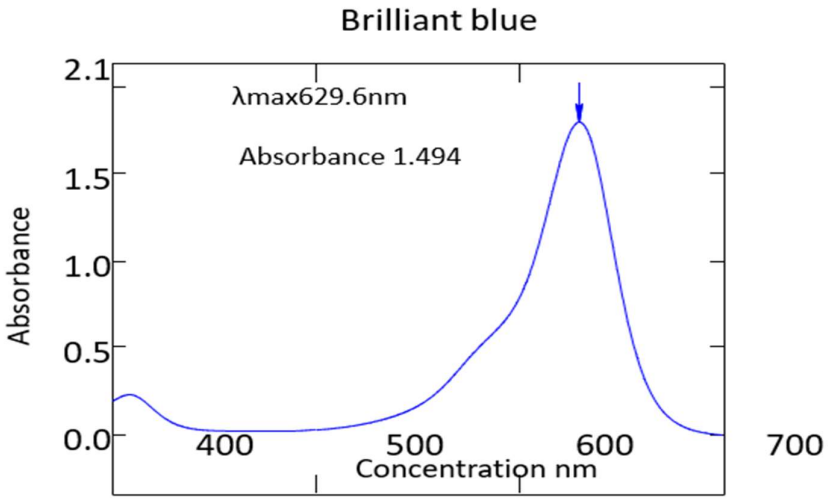

Fig. 4: Spectrum of Brilliant blue

Tartrazine

In the first trial stock solution of Tartrazine was prepared by accurately weighing 10 mg of the standard dye and dissolving it in a 10 *mL* volumetric flask with Methanol, resulting in a concentration of 1000 ppm. From this stock, 0.1 *mL* was pipetted and diluted to 10 *mL* with Methanol to obtain a 10ppm working solution. The prepared solution was scanned in spectrum mode to determine the absorption wave length. It was observed weak absorbance with no distinct peak formation. [17-19]

In the second trial standard stock solution of Tartrazine was prepared by accurately weighing 10 mg of the dye and dissolving it in Methanol to obtain a final concentration of 1000 ppm. From this solution, 1 *mL* was further diluted to 10 *mL* with distilled water to prepare a working solution. When analyzed under the spectrophotometer, no characteristic peak absorbance was detected at the expected wavelength, indicating the absence of a distinct spectrum.

In the third trial standard stock solution of Tartrazine was prepared by accurately weighing 10 mg of the dye and dissolving it in a 10 *mL* volumetric flask with distilled water, yielding a concentration of 1000 ppm. From this stock, 0.1 *mL* was pipetted into another volumetric flask and diluted to volume with water to obtain a 10ppm working solution. The solution was then scanned in spectrum mode to determine the maximum absorbance wavelength, which was found to be 424nm with an absorbance value of 0.167.

Inference: Trial 3 is optimized with water showed better peak shape and resolution for Tartrazine. Figure 5 shows Spectrum of Tartrazine showing maximum Absorbance at 424 nm

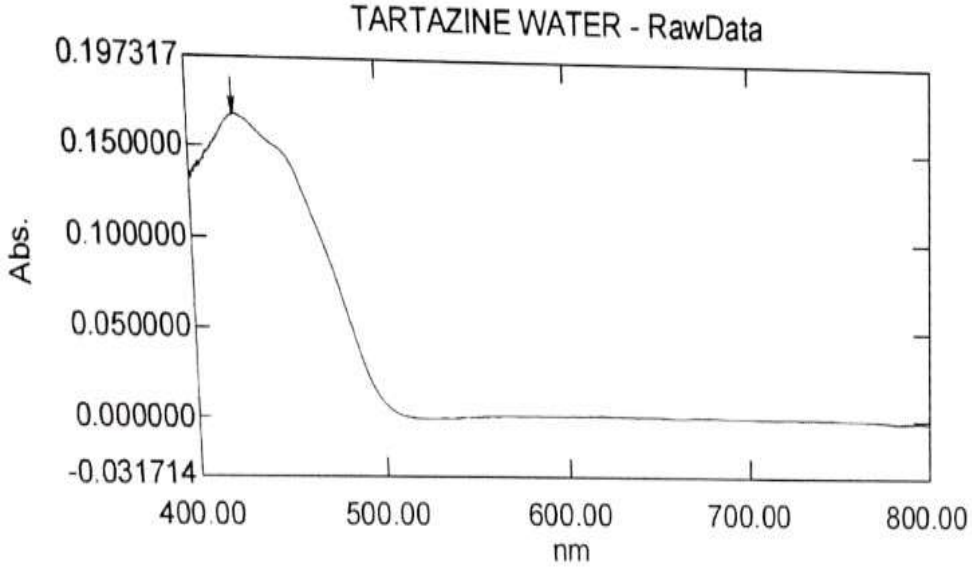

Fig. 5: Spectrum of Tartrazine

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Allura Red

For the initial experiment, a stock solution of Allura Red was obtained by precisely weighing 10 mg of the reference dye and diluting it to volume in a 10 *mL* volumetric flask with Methanol, resulting in a concentration of 1000 ppm. From this stock, 0.1 *mL* was pipetted and diluted to 10 *mL* with Methanol to obtain a 10ppm working solution. The prepared solution was scanned in spectrum mode to determine the absorption wave length. It was observed weak absorbance with no distinct peak formation.

In the second trial standard stock solution of Allura Red was prepared by accurately weighing 10 mg of the dye and dissolving it in Methanol to obtain a final concentration of 1000 ppm. From this solution, 1 *mL* was further diluted to 10 *mL* with distilled water to prepare a working solution. When analyzed under the spectrophotometer, no characteristic peak absorbance was detected at the expected wavelength, indicating the absence of a distinct spectrum.

In the third trial standard stock solution of Allura red was prepared by accurately weighing 10 mg of the dye and dissolving it in a 10 *mL* volumetric flask with distilled water, yielding a concentration of 1000 ppm. From this stock, 0.1 *mL* was pipetted into another volumetric flask and diluted to volume with water to obtain a 10ppm working solution. The solution was then scanned in spectrum mode to determine the maximum absorbance wavelength, which was found to be 503 nm with an absorbance value of 0.624.

Inference: The optimized wavelength (λmax) for Allura red was found to be 503 nm, with an absorbance of 0.624. Figure 6 shows Spectrum of Allura red showing maximum Absorbance at 503 nm

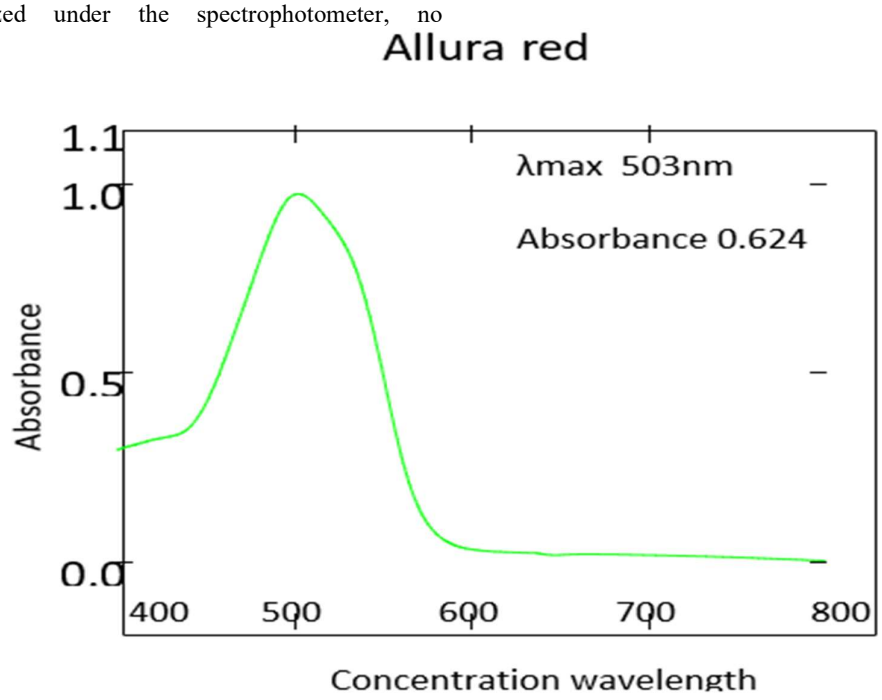

Fig. 6: Spectrum of Allura red

Sun Set Yellow

During the first trial, the stock solution of sunset yellow was prepared by carefully weighing 10 mg of the dye standard and making up the volume to 10 *mL* in a volumetric flask with Methanol, resulting in a concentration of 1000 ppm. From this stock, 0.1 *mL* was pipetted and diluted to 10 *mL* with Methanol to obtain a 10ppm working solution. The prepared solution was scanned in spectrum mode to determine the absorption wave length. It was observed weak absorbance with no distinct peak formation.

In the second trial standard stock solution of Sun set yellow was prepared by accurately weighing 10 mg of the dye and dissolving it in Methanol to obtain a final concentration of 1000 ppm. From this solution, 1 *mL* was further diluted to 10 *mL* with distilled water to prepare a working solution. When analyzed under the spectrophotometer, no characteristic peak absorbance was detected at the expected wavelength, indicating the absence of a distinct spectrum.

A standard stock solution of Sunset Yellow was prepared by accurately weighing 10 mg of the dye and dissolving it in a 10 *mL* volumetric flask with distilled water, yielding a concentration of 1000 ppm. From this stock, 0.1 *mL* was pipetted into another volumetric flask and diluted to volume with water to obtain a 10ppm working solution. The solution was then scanned in spectrum mode to determine the maximum absorbance wavelength, which was found to be 481.5 nm with an absorbance value of 0.574.

The optimized wavelength (λmax) for Sun set yellow was found to be 481.5 nm, with an absorbance of 0.574. Figure 7 shows Spectrum of Sun set yellow showing maximum Absorbance at 481.5 nm

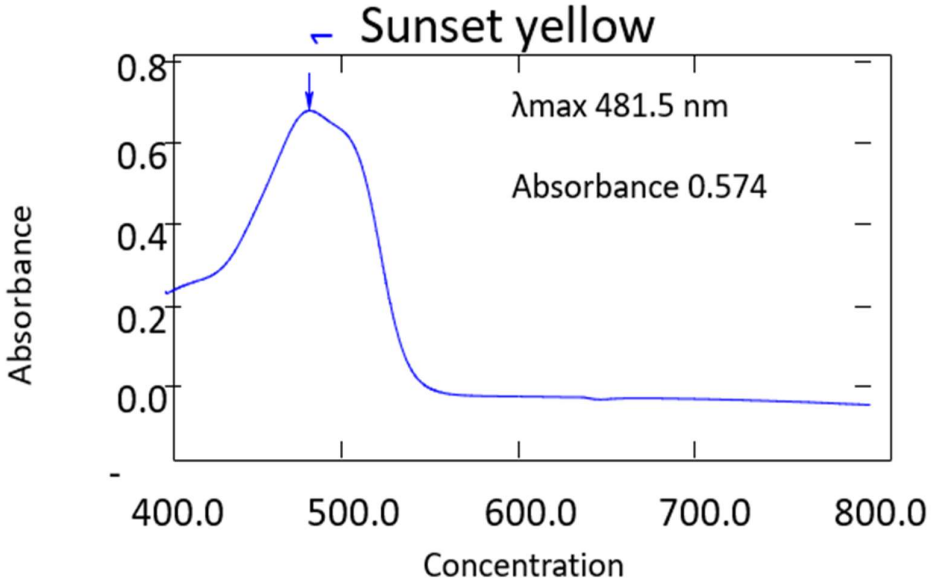

Fig. 7: Spectrum of Sun set yellow

Ponceau4r

A stock solution of Ponceau 4R was obtained in the first trial by dissolving an accurately weighed 10 mg portion of the standard dye in a 10 *mL* volumetric flask with Methanol, resulting in a concentration of 1000 ppm. From this stock, 0.1 *mL* was pipetted and diluted to 10 *mL* with Methanol to obtain a 10ppm working solution. The prepared solution was scanned in spectrum mode to determine the absorption wave length. It was observed weak absorbance with no distinct peak formation.

In the second trial standard stock solution of Ponceau4R was prepared by accurately weighing 10 mg of the dye and dissolving it in Methanol to obtain a final concentration of 1000 ppm. From this solution, 1 *mL* was further diluted to 10 *mL* with distilled water to prepare a working solution. When analyzed under the spectrophotometer, no characteristic peak absorbance was detected at the expected wavelength, indicating the absence of a distinct spectrum. [20-24]

In the third trial standard stock solution of Ponceau4R was prepared by accurately weighing 10 mg of the dye and dissolving it in a 10 *mL* volumetric flask with distilled water, yielding a concentration of 1000 ppm. From this stock, 0.1 *mL* was pipetted into another volumetric flask and diluted to volume with water to obtain a 10ppm working solution. The solution was then scanned in spectrum mode to determine the maximum absorbance wavelength, which was found to be 508.5 nm with an absorbance value of 0.658.

Inference: The optimized wavelength (λmax) for Ponceau 4R was found to be 508.5 nm, with an absorbance of 0.658. Figure 8 shows Spectrum of Ponceau 4R showing maximum Absorbance at 508.5 nm

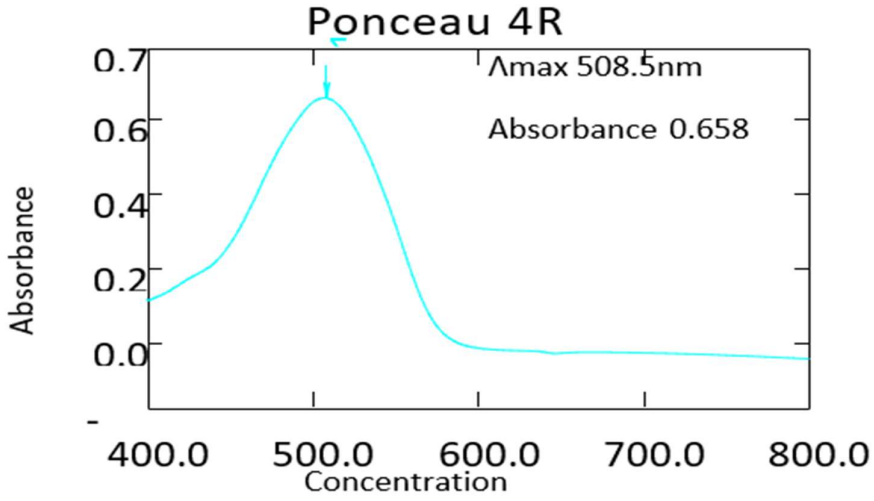

Fig. 8: Spectrum of Ponceau4R

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RESULTS AND DISCUSSION

Method Validation: Validation parameters of Erythrosine:

Linearity:

Linearity was determined by preparing different concentrations from the stock solution, and the resulting regression value met the acceptance criteria. Figure 9 shows the correlation coefficient R^2 has been determined to be 0.9991 is within the acceptable range.

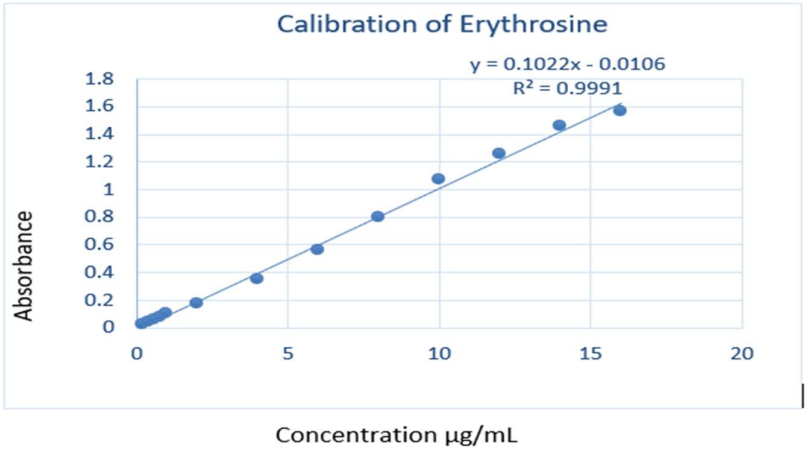

Fig. 9 : Linearity plot of Erythrosine

Precision

Precision was assessed at single concentration, with the Mean, Standard Deviation and percentage relative standard deviation (%RSD) falling within acceptable limits. Both inter-day and intra-day precision were conducted. Table 2 describes the Intra-day, inter-day, and instrument precision results showed %RSD values below 2%, confirming excellent repeatability, intermediate precision, and method reliability as per ICH guidelines

Table 2: Inter day and Intraday precision results of Erythrosine with individual values, mean, SD, and %RSD.

Concentration *µg/mL*	**Absorbance**	
	Inter day precision	**Intraday precision**
8*µg/mL*	0.737	0.748
8*µg/mL*	0.750	0.761
8*µg/mL*	0.756	0.767
8*µg/mL*	0.766	0.772
8*µg/mL*	0.713	0.726
8*µg/mL*	0.746	0.755
Mean	0.7446	0.7548
SD	0.00818	0.00737
%RSD	1.09%	0.97%

Robustness

Robustness was evaluated by analyzing the same concentration six times at varying wavelengths. And the percentage relative standard deviation (%RSD) are within limits. Table 3 describes Robustness results of Ponceau4R obtained at varied wavelengths with corresponding absorbance values. Table 4 describes LOD and LOQ. [25-26]

Table 3: Robustness results of Erythrosine obtained at varied wavelengths with corresponding absorbance values.

S.no	523.8	524	525.8	526	526.8
1	0.734	0.737	0.748	0.749	0.748
2	0.748	0.750	0.735	0.762	0.761
3	0.754	0.756	0.767	0.768	0.767
4	0.764	0.766	0.779	0.779	0.772
5	0.711	0.713	0.726	0.727	0.726
6	0.744	0.746	0.756	0.756	0.755
Mean	0.7425	0.7446	0.7518	0.7568	0.7548
SD	0.0082	0.00818	0.00883	0.0079	0.00737
%RSD	1.1%	1.09%	1.17%	1.04%	0.97%

Table 4: LOD and LOQ results of Erythrosine.

Limit of Detection	Limit of Quantification
0.02913$\mu g/mL$	0.8699$\mu g/mL$

Validation parameters of Brilliant blue:

Linearity

Linearity was determined by preparing different concentrations from the stock solution, and the resulting regression value met the acceptance criteria. Figure 10 shows the correlation coefficient R^2 has been determined to be 0.9964 is within the acceptable range.

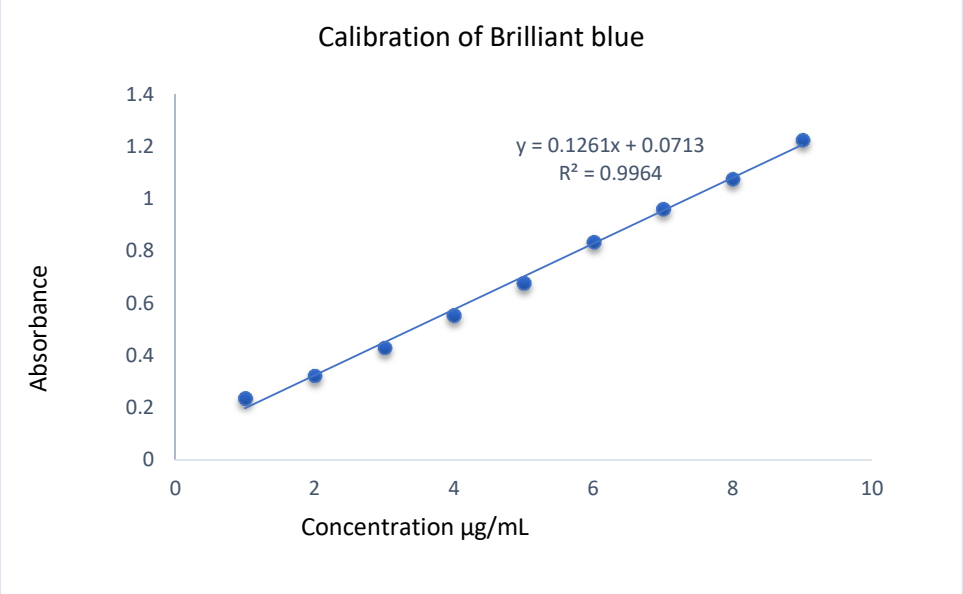

Fig. 10: Linearity plot of Brilliant blue

Precision

Precision was assessed at single concentration, with the Mean, Standard Deviation and percentage relative standard deviation (%RSD) falling within acceptable limits. Both inter-day and intra-day precision were conducted. Table 5 describes the Intra-day, inter-day, and instrument precision results showed %RSD values below 2%, confirming excellent repeatability, intermediate precision, and method reliability as per ICH guidelines. [27,28]

Table 5: Inter day and Intraday precision results of brilliant blue with individual values, mean, SD, and %RSD

Concentration *µg/mL*	**Absorbance**	
	Inter day precision	**Intraday precision**
5*µg/mL*	0.610	0.626
5*µg/mL*	0.666	0.660
5*µg/mL*	0.660	0.656
5*µg/mL*	0.659	0.652
5*µg/mL*	0.645	0.657
5*µg/mL*	0.620	0.632
Mean	0.6433	0.6471
SD	0.0103	0.0064
%RSD	1.60%	0.98%

Robustness

Robustness was evaluated by analyzing the same concentration six times at varying wavelengths. And the percentage relative standard deviation (%RSD) are within limits. Table 6 Robustness results of Ponceau4R obtained at varied wavelengths with corresponding absorbance values and Table 7 shows LOD and LOQ.

Table 6: Robustness results of brilliant blue obtained at varied wavelengths with corresponding absorbance values.

S.no	628.6	629	629.6	630	630.6
1	0.609	0.610	0.624	0.626	0.630
2	0.633	0.666	0.649	0.660	0.652
3	0.647	0.660	0.655	0.656	0.667
4	0.658	0.659	0.630	0.652	0.657
5	0.644	0.645	0.636	0.657	0.660
6	0.619	0.620	0.625	0.632	0.635
Mean	0.635	0.643	0.634	0.647	0.650
SD	0.0082	0.0103	0.0060	0.0064	0.00653
%RSD	1.29%	1.60%	0.94%	0.98%	1.0%

Table 7: LOD and LOQ results of brilliant blue

LIMIT OF DETECTION	LIMIT OF QUANTIFICATION
0.1570$\mu g/mL$	0.4758$\mu g/mL$

Experimental Procedures for Food Samples

Preparation of Sheep Wool

Sheep wool was procured and initially exposed to direct sunlight for drying. Once dried, the outer layer of hair was carefully trimmed off. The cleaned material was then thoroughly washed with water to eliminate surface impurities. After cleaning, the wool was further processed and converted into thread-like strands suitable for experimental use.

Preparation of Erythrosine, Brilliant Blue, Tatrazine Samples

Color Extraction Procedure for Beverages (Sting, Mogu Mogu, Monin curacao bleu syrup, Campa, Tata glucose)

A volume of 20 *mL* of the beverage sample was transferred into a clean container, followed by the addition of methanol and distilled water to facilitate extraction of the coloring agents. The mixture was stirred thoroughly, and if necessary, filtered to remove suspended particles. Wool threads of about 3 *cm* in length were pretreated by boiling in water for 3–5 minutes and subsequently soaking in 2 *M* acetic acid. The conditioned threads were immersed in the beverage extract and kept in a water bath for approximately 30 minutes to allow adsorption of the dyes. After adsorption, the threads were rinsed with water for 1–2 minutes and then immersed in 2 M ammonium solution. They were reheated in hot water until decolorization occurred. The released dye solution was collected, centrifuged, and the clear supernatant was subjected to spectrophotometric analysis. [29-32]

Preparation of Sun Set Yellow, Allura Red, Ponceau4r Samples

Color Extraction Procedure for Jellys, tic tac, skittles, center fruit, center fresh, custard powder

About 5 *g* of each sample was taken into a beaker, mixed with distilled water, and left for a short period with gentle shaking to extract the dyes, after which the suspension was filtered to remove residues. Wool threads of around 3 *cm* were pretreated by boiling in water for 3–5 minutes and soaking in 2 *M* acetic acid, then immersed in the sample extract and kept in a water bath for 30 minutes to enable dye adsorption. The colored threads were rinsed with water, transferred to 2Molar ammonium solution, and reheated in hot water until decolorized. The released dye solution was centrifuged, and the clear supernatant was subjected to spectrophotometric analysis as shown in Table 8. [33,34]

Table 8: Results of overall samples

S. no	Colour	Samples used	Absorbance	Weighed sample	Amount found	Limits	Inference
1	**Brilliant blue**	Lollipop	1.258	5*g*	6.9 *mg*	6*mg/kg*	Not in limits
		*G*ems	0.954	5*g*	0.006*mg*	6*mg/kg*	With in limits
		Monin curacao bleu syrup	0.935	5*mL*	0.09*mg*	6*mg/kg*	With in limits
		Center fresh	0.962	5*g*	0.001*mg*	6*mg/kg*	With in limits
		Mogu Mogu	0.242	5*mL*	0.10*mg*	6*mg/kg*	With in limits
		Tic tac	0.314	5*g*	0.003*mg*	6*mg/kg*	With in limits
2	**Erythrosine**	*G*ems	1.405	5*g*	0.02*mg*	6*mg/kg*	With in limits
		Straw berry	2.704	5*g*	0.46*mg*	6*mg/kg*	With in limits
		Juzt jelly	1.197	5*g*	0.03*mg*	6*mg/kg*	With in limits
		Tic tac	1.177	5*g*	0.01*mg*	6*mg/kg*	With in limits
3	**Tartrazine**	Campa	1.137	5*mL*	13.4*mg*	7.5*mg/kg*	Not in limits

		Sting	0.238	5*mL*	1.4*mg*	7.5*mg/kg*	With in limits
		Center fresh	2.455	5*g*	0.48*mg*	7.5*mg/kg*	With in limits
		Tic tac	1.650	5*g*	0.5*mg*	7.5*mg/kg*	With in limits
		Lollipop	0.904	5*g*	0.2*mg*	7.5*mg/kg*	With in limits
		Naturo mango	3.338	5*g*	1.18*mg*	7.5*mg/kg*	With in limits
		Custard powder	0.882	5*g*	1.86*mg*	7.5*mg/kg*	With in limits
		Skippi sticks	3.061	5*g*	0.036*mg*	7.5*mg/kg*	With in limits
		Zubi lollipop	0.953	5*g*	0.22*mg*	7.5*mg/kg*	With in limits
4	**Sun set yellow**	Pine apple	2.314	5*g*	2.8*mg*	4*mg/kg*	With in limits
		Lollipop	1.353	5*g*	0.24*mg*	4*mg/kg*	With in limits
		*G*ems	0.818	5*g*	0.077*mg*	4*mg/kg*	With in limits
		Tata glucose	2.850	5*mL*	0.35*mg*	4*μg/mL*	With in limits
		Skittles	1.960	5*g*	0.35*mg*	4*mg/kg*	With in limits
5	**Allura red**	Red velvet cake	4.000	5*g*	3.6*mg*	7*mg/kg*	With in limits
		Lollipop	0.632	5*g*	0.08*mg*	7*mg/kg*	With in limits
		Tic tac	1.313	5*g*	0.22*mg*	7*mg/kg*	With in limits
		*G*ems	1.859	5*g*	0.38*mg*	7*mg/kg*	With in limits
		Skittles	2.705	5*g*	0.31*mg*	7*mg/kg*	With in limits
		Sting	2.356	5*mL*	7.6*mg*	7*mg/kg*	Not in limits
6	**Ponceau4R**	Campa	0.856	5*mL*	0.05*mg*	6*mg/kg*	With in limits

		*G*ems	0.856	5*g*	0.001*mg*	6*mg/kg*	With in limits
		Skittles	1.305	5*g*	3.3*mg*	6*mg/kg*	With in limits
		Butterscotch cake	3.823	5*g*	7.06*mg*	6*mg/kg*	Not in limits
		Sting	2.215	5*mL*	0.31*mg*	6*mg/kg*	With in limits
		Sun feast cream biscuits	0.566	5*g*	0.01*mg*	6*mg/kg*	With in limits
		Magix cream biscuits	0.550	5*g*	0.01*mg*	6*mg/kg*	With in limits
		Sun feast biscuits	0.428	5*g*	0.01*mg*	6*mg/kg*	With in limits

DISCUSSION

The developed UV–Visible spectrophotometric method exhibited excellent linearity, with correlation coefficients (R^2) of 0.9991 for Erythrosine and 0.9964 for Brilliant Blue, which are comparable to those reported for validated RP-HPLC methods used for synthetic food colour analysis. The method showed good precision, with inter-day and intra-day %RSD values of 1.09% and 0.97% for Erythrosine and 1.60% and 0.98% for Brilliant Blue, satisfying the ICH Q2(R2) acceptance criterion of less than 2% and demonstrating reproducibility similar to RP-HPLC techniques. Robustness studies produced %RSD values between 0.97–1.17% for Erythrosine and 0.94–1.60% for Brilliant Blue, indicating that minor wavelength variations had minimal impact on analytical performance. The proposed method also showed adequate sensitivity, with LOD/LOQ values of 0.0291/0.8699 µg/mL for Erythrosine and 0.1570/0.4758 µg/mL for Brilliant Blue, although RP-HPLC generally offers lower detection limits due to its superior separation efficiency. Analysis of commercial food samples revealed that Lollipop (Brilliant Blue, 6.9 mg), Campa (Tartrazine, 13.4 mg), Sting (Allura Red, 7.6 mg), and Butterscotch Cake (Ponceau 4R, 7.06 mg) exceeded the prescribed regulatory limits, while the remaining samples complied with permissible levels. Several samples, including Red Velvet Cake (4.000 AU), Butterscotch Cake (3.823 AU), Naturo Mango (3.338 AU), and Skippi Sticks (3.061 AU), exhibited absorbance values above 2.0 AU, indicating that appropriate dilution would be necessary to obtain accurate quantitative measurements within the instrument's linear range. Overall, the developed UV–Visible method coupled with sheep wool extraction provides a rapid, economical, and reliable alternative to RP-HPLC for routine determination of synthetic food colourants, although RP-HPLC remains the preferred technique when simultaneous separation and highly sensitive quantification of multiple dyes are required

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CONCLUSION

The present study successfully developed and validated a simple, rapid, and cost-effective UV–Visible spectrophotometric method for the determination of synthetic food colourants in food and beverage products. The method demonstrated satisfactory linearity, precision, robustness, and sensitivity in accordance with ICH Q2(R2) guidelines. Analysis of commercial samples revealed that Tartrazine, Allura Red, and Ponceau 4R exceeded permissible limits in a few products, while Brilliant Blue, Erythrosine, and Sunset Yellow were generally within regulatory limits. The developed method proved to be reliable, accurate, and reproducible for routine analysis of synthetic food dyes. This validated method is reproducible, and practical for regulatory and quality control laboratories. Enhanced regulatory enforcement is essential to limit consumer exposure to excessive levels of synthetic color. The findings emphasize the importance of regular surveillance of synthetic colour additives in commercially available foods. Continuous regulatory monitoring is recommended to ensure consumer safety and compliance with food safety standards.

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ETHICAL APPROVAL

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CONSENT FOR PUBLICATION

Not applicable.

DECLARATION OF COMPETIN*G* INTEREST

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The authors have read and approved the manuscript. K. Bhavyasri designed the study. Remaining authors performed the experiment and analyzed and reviewed the data. K. Bhavyasri supervised the experiment, reviewed the data, and supported for writing the manuscript..

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