



## Determination of Tartrazine and Sunset Yellow in Foodstuffs by Derivative Spectrophotometric Method

ANTAKLI SAAD<sup>1,\*</sup>, SARKIS NAZIRA<sup>2</sup> and ALSALEH RASHA<sup>2</sup>

<sup>1</sup>Department of Chemistry, Faculty of Science, University of Aleppo, Aleppo, Syria

<sup>2</sup>Department of Analytical and Food Chemistry, Faculty of Pharmacy, University of Aleppo, Aleppo, Syria

\*Corresponding author: Tel: +963 944 814291; E-mail: antakli@scs-net.org

(Received: 8 September 2010;

Accepted: 24 December 2010)

AJC-9425

A relatively fast method was applied for determination of two synthetic food colours, tartrazine and sunset yellow, in three different kinds of foodstuffs: juice powders, flavoured juices and soft drinks. Samples preparation included degassing and/or centrifugation after dissolving the samples and showed high throughput. All studied samples showed dye levels in conformity with Syrian legislation. It was also observed that different producers use different colours and/or levels in similar products of the same flavour.

**Key Words:** Synthetic colours, Tartrazine, Sunset yellow, Derivative spectrophotometry.

### INTRODUCTION

Synthetic colours are widely used for maintenance and improvement of visual appearance in food products. Some colours can trigger adverse effects, namely tartrazine, which can cause the appearance of allergy, asthma<sup>1</sup> and childhood hyperactivity<sup>2</sup>. In addition, there are studies that prove carcinogenicity and toxicity of commonly used food colours<sup>3</sup>. Therefore, it is very important to determine these colours qualitatively and quantitatively for the assurance of food safety.

Various analytical methods have been used for determination of food colours, such as high performance liquid chromatography (HPLC)<sup>4,5</sup>, micellar electrokinetic chromatography (MEC)<sup>6,7</sup>, isotachopheresis<sup>8</sup> and differential pulse polarography<sup>9</sup>. The voltammetric methods including adsorptive voltammetry<sup>10</sup>, stripping voltammetry<sup>11</sup> and squarewave adsorptive voltammetry<sup>12</sup>, were also successfully applied to determine food colours.

To improve the efficiency and reliability of the determination, capillary electrophoresis (CE)<sup>13</sup>, high-performance ion chromatography<sup>14</sup> and reversed-phase liquid chromatography (RPLC)<sup>15,16</sup>, are successfully applied for water-soluble synthetic dyes. Some researchers have suggested MS detection<sup>17,18</sup>, which provides structural information on the dyes and their degradation products. However, most of the proposed methods for the analysis are time-consuming. Spectrophotometric<sup>19-21</sup> and derivative spectrometric methods<sup>22,23</sup>, have been used presenting high sensitivity, simplicity, rapidity and low cost.

In this study we have determined two colours: tartrazine (E102) (Fig. 1), sunset yellow (E110) (Fig. 2) and their mixtures in different foodstuffs by derivative UV/vis spectrophotometric method.

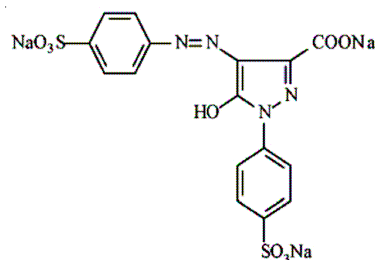


Fig. 1. Structural formula of tartrazine

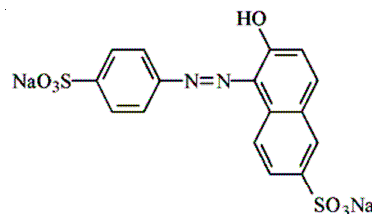


Fig. 2. Structural formula of sunset yellow

### EXPERIMENTAL

A UV/Visible spectrophotometer V-530 Jasco (Japan) was used to obtain the spectra, quartz cells 1 cm, a BP221S Sartorius analytical balance and a Power Sonic 405 ultrasonic bath were also used.

All dilutions were made using bi-distilled water, produced by Janat Instruments (Syria). Tartrazine, sunset yellow were obtained from Dynemic (India), Roha (India), respectively. All analyzed samples were purchased from Syrian local markets, included orange- and lemon-flavoured juice powders, juices and soft drinks.

**Standards solutions preparation:** Individual standard stock solutions containing each colour were prepared by dissolving 25 mg of purified colour tartrazine and sunset yellow, respectively in 250 mL volumetric flask with bi-distilled water. The solutions were kept in dark flasks. The working standard solutions of each colour were prepared by appropriate dilutions of stock solutions with bi-distilled water to give concentrations between 2 and 25  $\mu\text{g/mL}$  of tartrazine and sunset yellow, respectively, taking into consideration the purity of the colours.

**Samples solutions preparation:** The powder samples were homogenized. An amount between 0.05 and 1 g was precisely weighted and dissolved with bi-distilled water in a 25 mL volumetric flask. The sample solution was placed in ultrasonic bath for 10 min then centrifuged 15 min at 4000 rpm.

Soft drink samples were previously degassed in ultrasonic bath for 15 min. For soft drink and juice samples, an aliquot sample between 5 and 8 mL was transferred to 25 mL volumetric flask and diluted with bi-distilled water. Then centrifuged 15 min at 4000 rpm and analyzed. Some samples were analyzed without any dilution.

## RESULTS AND DISCUSSION

**Standard solutions spectra:** In the zero-order spectra there is an overlapping in peaks of the mixture 18  $\mu\text{g/mL}$  of tartrazine and 15  $\mu\text{g/mL}$  of sunset yellow as shown in (Fig. 3), so the simultaneous determination of tartrazine and sunset yellow cannot be made at this point. This was solved in the second-order derivative spectra as shown in (Fig. 4).

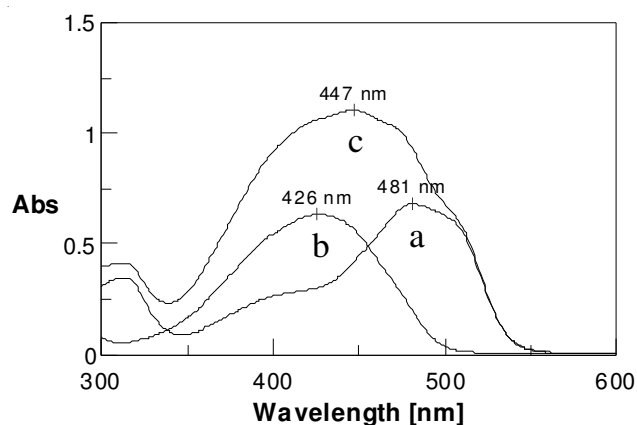


Fig. 3. Zero-order spectra of sunset yellow (a), tartrazine (b) and their mixture (c)

The absorption spectra of the solutions in the range 300-600 nm at the increasing concentrations of tartrazine and sunset yellow individually were recorded and the second derivatives were obtained. The concentration of tartrazine in the range 2-20  $\mu\text{g/mL}$  was proportional to the amplitude at 412 nm (Fig. 5). In the same way, it is found that the concen-

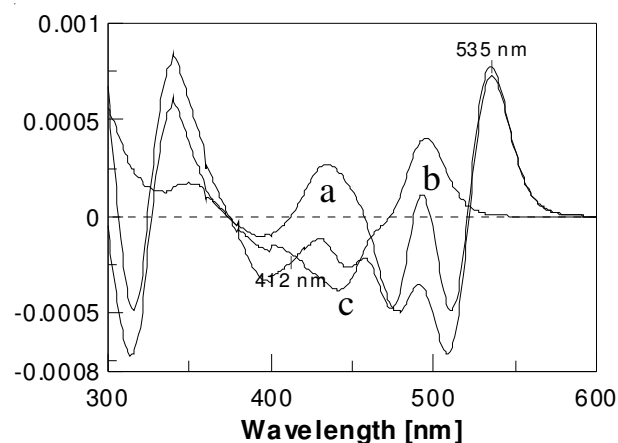


Fig. 4. Second-order spectra of sunset yellow (a), tartrazine (b) and their mixture (c)

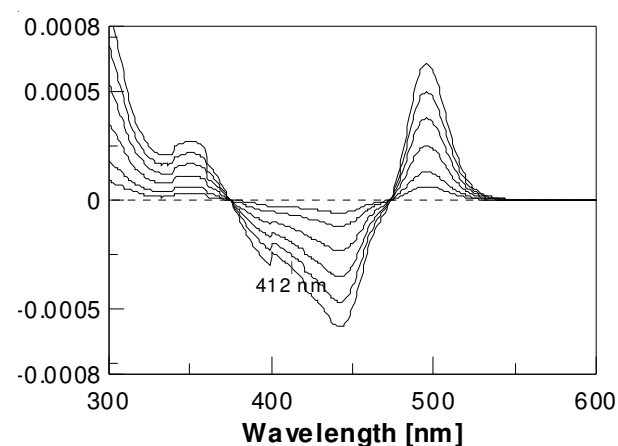


Fig. 5. Second-order derivative spectra of tartrazine

tration of sunset yellow in the range 2-24  $\mu\text{g/mL}$  was proportional to the amplitude at 535 nm (Fig. 6). In these two points, the calibration curve was obtained and used for the estimation of tartrazine and sunset yellow in studied samples.

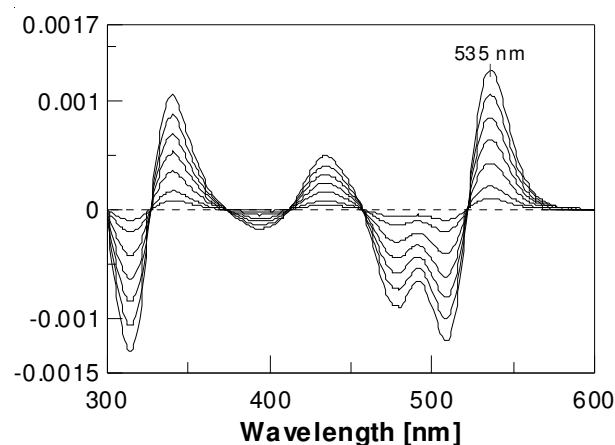


Fig. 6. Second-order derivative spectra of sunset yellow

The correlation coefficients ( $r$ ) of the calibration curves, which were constructed for five replicates measurements for each concentration in the range 2-25  $\mu\text{g/mL}$  of each colour, were above 0.998 (Figs. 7 and 8). Hence, the linearity of the calibration curve was relatively good.

TABLE-1  
CONCENTRATIONS OF COLOURS DETERMINED IN STUDIED SAMPLES

Trademark	No. of sample	Sunset yellow		Tartrazine		Trademark	No. of sample	Sunset yellow		Tartrazine	
		Concentration*	RSD (%)	Concentration*	RSD (%)			Concentration*	RSD (%)	Concentration*	RSD (%)
Crush <sup>1</sup>	1	54.96	0.26	–	–	Mountain Dew <sup>1</sup>	7	–	–	5.63	0.61
	2	54.65	0.26	–	–		8	–	–	5.72	1.14
	3	55.09	0.30	–	–		9	–	–	5.76	0.76
	4	55.31	0.39	–	–		10	–	–	5.81	1.57
	5	55.29	0.44	–	–		Range*	–	–	5.63-5.81	–
	6	51.29	0.37	–	–		1	–	–	5.86	0.56
	7	51.66	0.34	–	–		2	–	–	5.92	0.75
	8	55.57	0.52	–	–		3	–	–	5.78	0.73
	9	55.02	0.26	–	–		4	–	–	5.91	0.29
	10	55.32	0.20	–	–		5	–	–	5.99	0.92
Range*	51.29–55.57	–	–	–	6	–	–	6.01	1.48		
Local <sup>1</sup>	1	32.37	0.31	–	–	7	–	–	5.90	1.17	
	2	32.43	0.41	–	–	8	–	–	5.90	0.38	
	3	32.45	0.20	–	–	9	–	–	5.92	0.52	
	4	32.65	0.34	–	–	10	–	–	6.06	0.72	
	5	32.75	0.50	–	–	Range*	–	–	5.78-6.06	–	
	6	32.13	0.16	–	–	1	17.58	0.26	16.72	0.61	
	7	32.47	0.24	–	–	2	18.05	1.54	17.51	2.21	
	8	32.22	0.38	–	–	3	14.10	1.41	14.50	2.96	
	9	31.91	0.41	–	–	4	14.27	2.54	14.56	3.30	
	10	32.20	0.52	–	–	5	17.03	2.02	16.97	1.84	
Range*	31.91–32.75	–	–	–	6	16.99	1.10	17.13	3.10		
Master <sup>1</sup>	1	61.67	0.38	–	–	7	16.47	0.90	16.15	0.90	
	2	61.92	0.47	–	–	8	16.81	1.67	16.39	1.24	
	3	62.00	0.66	–	–	9	16.54	1.50	16.12	2.38	
	4	64.13	0.29	–	–	10	16.23	1.87	15.84	2.54	
	5	63.47	0.45	–	–	Range*	14.10–18.05	–	14.50-17.51	–	
	6	63.34	0.56	–	–	1	8.08	0.33	5.08	1.04	
	7	62.69	0.28	–	–	2	7.48	2.84	4.66	2.61	
	8	62.80	0.81	–	–	3	7.52	2.39	4.78	0.54	
	9	62.79	0.66	–	–	4	7.58	1.51	4.76	0.82	
	10	63.58	0.61	–	–	5	8.00	1.04	5.03	0.71	
Range*	61.67–64.13	–	–	–	6	8.89	0.51	5.66	1.48		
Buonasera <sup>2</sup>	1	11.83	0.25	–	–	7	7.60	0.84	4.84	1.58	
	2	11.45	0.32	–	–	8	8.15	0.23	5.10	1.48	
	3	11.77	0.24	–	–	9	8.13	1.15	5.28	2.10	
	4	11.40	0.36	–	–	10	7.51	0.58	4.80	2.25	
	5	11.99	0.35	–	–	Range*	7.48-8.89	–	4.66-5.66	–	
	6	11.95	0.25	–	–	1	25.94	0.47	4.21	1.86	
	7	11.82	0.31	–	–	2	26.43	1.85	4.12	1.12	
	8	11.82	0.23	–	–	3	25.37	0.82	4.25	3.07	
	9	11.62	0.51	–	–	4	28.73	1.48	5.04	2.71	
	10	11.93	0.22	–	–	5	24.17	3.78	3.79	2.73	
Range*	11.40-11.99	–	–	–	6	28.66	1.88	4.40	2.01		
Yomi <sup>2</sup>	1	25.51	0.39	–	–	7	30.00	1.99	4.85	2.10	
	2	25.63	0.34	–	–	8	26.02	1.25	4.73	2.96	
	3	25.47	1.23	–	–	9	28.23	0.51	4.80	2.73	
	4	25.28	0.40	–	–	10	27.45	2.07	4.74	0.64	
	5	25.29	0.38	–	–	Range*	24.17-30.00	–	3.79-5.04	–	
	6	25.34	0.27	–	–	1	15.55	1.11	10.05	1.50	
	7	24.90	1.37	–	–	2	15.87	1.88	10.39	0.73	
	8	25.21	0.35	–	–	3	13.95	2.69	8.99	3.10	
	9	25.21	0.38	–	–	4	13.03	0.40	8.72	2.10	
	10	25.38	0.58	–	–	5	12.23	1.19	8.06	3.22	
Range*	24.90-25.63	–	–	–	6	12.84	1.35	8.04	0.93		
Quatro <sup>2</sup>	1	–	–	5.68	0.37	7	15.05	0.99	8.82	0.58	
	2	–	–	5.73	1.20	8	15.80	0.74	9.10	1.93	
	3	–	–	5.63	0.43	9	15.19	2.98	9.44	0.35	
	4	–	–	5.69	0.46	10	15.07	2.80	9.44	0.62	
	5	–	–	5.76	1.61	Range*	12.23-15.87	–	8.04-10.39	–	
	6	–	–	5.70	1.08	–	–	–	–	–	
Squeeze <sup>3</sup>	1	–	–	5.68	0.37	7	15.05	0.99	8.82	0.58	
	2	–	–	5.73	1.20	8	15.80	0.74	9.10	1.93	
	3	–	–	5.63	0.43	9	15.19	2.98	9.44	0.35	
	4	–	–	5.69	0.46	10	15.07	2.80	9.44	0.62	
	5	–	–	5.76	1.61	Range*	12.23-15.87	–	8.04-10.39	–	
	6	–	–	5.70	1.08	–	–	–	–	–	
	7	–	–	5.68	0.37	7	15.05	0.99	8.82	0.58	
	8	–	–	5.73	1.20	8	15.80	0.74	9.10	1.93	
	9	–	–	5.63	0.43	9	15.19	2.98	9.44	0.35	
	10	–	–	5.69	0.46	10	15.07	2.80	9.44	0.62	
Range*	–	–	–	–	–	–	–	–	–		

\*In (µg/mL) for soft drinks and juices, (µg/g) for juice powders. <sup>1</sup>Soft drinks, <sup>2</sup>Juices, <sup>3</sup>Juice powders.

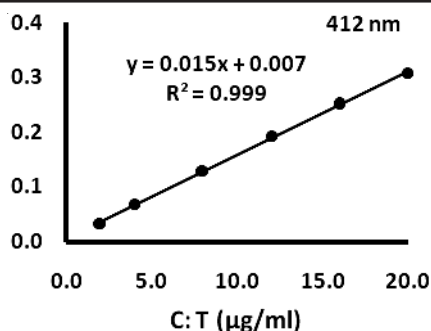


Fig. 7. Calibration curve for tartrazine

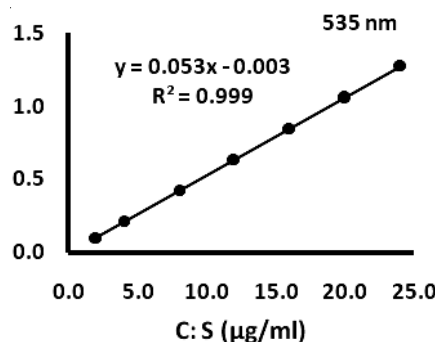


Fig. 8. Calibration curve for sunset yellow

**Sample analysis:** Table-1 summarizes the results of analysing 4 Syrian trademarks of soft drinks, 3 of juices and 4 of juice powders, each of which was analyzed in 10 independent measurements with five replicates. In the soft drinks and juices, only tartrazine or sunset yellow was detected, individually in lemon and orange flavour, respectively. While for juice powders which were all in orange flavour, both tartrazine and sunset yellow were detected. In general, the amounts used of sunset yellow were relatively higher than these of tartrazine. However, the concentrations of the detected colours were much lower than the Syrian limits; 500 µg/g for tartrazine and 200 µg/g for sunset yellow, in the ready-to-eat product<sup>24</sup>. The relative standard deviations of the quantitative results were in the range of 0.29-3.30 and 0.16-3.78 % for tartrazine and sunset yellow, respectively.

## Conclusion

We have determined levels of tartrazine, sunset yellow and their mixtures in different local foodstuffs by simple derivative spectrophotometric method. Colours levels were very lower than their maximum values which established by the Syrian legislation. Furthermore, tartrazine and sunset yellow

concentrations varied between different trademarks up to 4 times for tartrazine and 8 times for sunset yellow of the lowest levels with the same flavours. The proposed UV/visible spectrophotometric method offered high sensitivity in addition to its rapidity, accuracy, precision and low cost in either raw material or commercial products without any interference with excipients and with minimum pre-treatment procedures of samples.

## ACKNOWLEDGEMENTS

The Ministry of High Education in Syria financially and technically supported this work through Department of Analytical and Food Chemistry, Faculty of Pharmacy, University of Aleppo, Aleppo, Syria.

## REFERENCES

- J.J.B. Nevado, J.R. Flores and M.J.V. Llerena, *Fresenius J. Anal. Chem.*, **357**, 989 (1997).
- K.S. Rowe and K.J. Rowe, *J. Pediatr.*, **125**, 691 (1994).
- R.D. Combes and R.B. Haveland-Smith, *Mutat. Res.*, **98**, 101 (1982).
- M.L. Williams, *Food Chem.*, **22**, 235 (1986).
- M.C. Gennaro, E. Gioannini, S. Angelino, R. Aigotti and D. Giacosa, *J. Chromatogr. A*, **767**, 87 (1997).
- C.O. Thompson and V.C. Trenerry, *J. Chromatogr. A*, **704**, 195 (1995).
- R.A. Frazier, E.L. Inns, N. Dossi, J.M. Ames and H.E. Nursten, *J. Chromatogr. A*, **876**, 213 (2000).
- J. Karovicová, J. Polonský, A. Příbela and P. Šimko, *J. Chromatogr. A*, **545**, 413 (1991).
- S. Combeau, M. Chatelut and O. Vittori, *Talanta*, **56**, 115 (2002).
- Y.N. Ni, J.L. Bai and L. Jin, *Anal. Chim. Acta*, **329**, 65 (1996).
- M. Florian, H. Yamanaka, P.A. Carneiro and M.V.B. Zanon, *Food Addit. Contam.*, **19**, 803 (2002).
- J.J.B. Nevado, J.R. Flores and M.J.V. Llerena, *Talanta*, **44**, 467 (1997).
- L. Del Giovine and A.P. Bocca, *Food Control*, **14**, 131 (2003).
- Q.C. Chen, S.F. Mou, X.P. Hou, J.M. Riviello and Z.M. Ni, *J. Chromatogr. A*, **827**, 73 (1998).
- J.T. Hann and I.S. Gilkison, *J. Chromatogr. A*, **395**, 317 (1987).
- G.M. Greenway, N. Kometa and R. Macrae, *Food Chem.*, **43**, 137 (1992).
- T. Poiger, S.D. Richardson and G.L. Baughman, *J. Chromatogr. A*, **886**, 271 (2000).
- J. Riu, I. Schönsee and D. Barceloi, *J. Mass Spectrom.*, **33**, 653 (1998).
- J.J. Berzas, J.R. Flores, M.J.V. Llerena and N.R. Fariñas, *Anal. Chim. Acta*, **391**, 353 (1999).
- E. Dinc, E. Baydan, M. Kanbur and F. Onur, *Talanta*, **58**, 579 (2002).
- J.J.B. Nevado, C.G. Cabanillas and A.M.C. Salcedo, *Fresenius J. Anal. Chem.*, **350**, 606 (1994).
- J.J.B. Nevado, C.G. Cabanillas and A.M.C. Salcedo, *Talanta*, **42**, 2043 (1995).
- J.J.B. Nevado, J.R. Flores and M.J.V. Llerena, *Talanta*, **40**, 1391 (1993).
- Syrian Arab Organization for Standardization and Metrology, SNS 770 (1996).