Original Article

Journal of Forensic Chemistry and Toxicology
Volume 5 Number 1, January - June 2019
DOI: http://dx.doi.org/10.21088/jfct.2454.9363.5119.4

Derivative UV-Vis Spectrophotometric Analysis of Caramel in Some Common Liquor Samples

Candida Britto[1], Praveen Kumar Yadav[2], Rakesh Mohan Sharma[3]

Author Affiliation: [1]Student [2]Research Student [3]Professor Department of Forensic Science, Punjabi University, Patiala, Punjab 147002, India.

Corresponding Author: Praveen Kumar Yadav, Research Student, Department of Forensic Science, Punjabi University, Patiala, Punjab 147002, India.

E-mail: praveenky15@yahoo.com

Received on 06.05.2019, **Accepted on** 08.06.2019

Abstract

In recent years, awareness regarding the quality of food has gradually increased as per the increase in consumer demands. One of the most important parts of the food is alcoholic beverages. Many additives such as caramel, sugar, water, etc. are added to improve the aroma volume, and color of alcoholic beverages. Caramel colors are dark brown substances often added in alcoholic beverages to give the more alluring appearance. However, many metabolites of caramel colors such as 5-hydroxymethyl furfural (5 HMF) are toxic in nature. Many studies show that in controlled concentrations, caramel colors don't pose any threat. Therefore, it is exigent to monitor the concentration of such additives. Moreover, these colors are present in trace amounts and require extensive sample preparation methods. In the present study, an attempt has been made to identify and detect caramel colors in various factory-made and commercial liquors using derivative Ultraviolet-visible (UV-Vis) spectrophotometry. It was observed that peak maxima of 281 nm of zero order spectra; in first-order spectra peak maxima of 260nm and peak minima of 290 nm can prove to be very effective in the identification of caramel colors. Derivative UV-Vis spectrophotometry thus, has proved to be an effective, non-destructive technique which requires minimal sample preparation.

Keywords: Alcoholic beverage; Caramel; Derivative UV-Vis spectrophotometry; 5-Hydroxymethyl furfuryl (5HMF).

How to cite this article:
Candida Britto, Praveen Kumar Yadav, Rakesh Mohan Sharma. Derivative UV-Vis Spectrophotometric Analysis of Caramel in Some Common Liquor Samples. J Forensic Chemistry Toxicol. 2019;5(1):21-25.

Introduction

Alcohol has been an integral part of human society since ancient times. For a chemist, it refers to a group of organic compounds containing –OH group. WHO has classified alcoholic beverages into two categories depending on whether the data pertaining to their production, sales, and consumption is available or not [1]. These categories are a) Recorded alcoholic beverages (those alcoholic beverages for which such data is available) and b) Unrecorded alcoholic beverages (those alcoholic beverages for which such data is not available). The composition of different alcoholic beverages depends on the raw material used, the methodology used during the production, and other factors such as environmental factors and additives which have been added to increase the aroma and coloration of the alcoholic beverages [2,3].

Alcoholic beverages often contain various types of additive, which are intentionally added to elaborate their aroma profile as well the appearance. One of the most commonly used additives is caramelcolor. Caramel constitutes one of mankind's oldest and

most important dietary material. Caramel colors include a family of distinct red to dark brown materials (liquid and powders) often used as additives in foods and beverages. Caramel colors are used to produce eye-pleasing colors in foods and beverages [4,5]. Caramel color is produced by the process of caramelization; the process of thermal treatment of carbohydrates. Industrial caramel colors are classified into four classes (I, II, III, IV). Class I (Plain caramel) caramel color is used in whiskey and other high proof alcohols. Class II (Sulfite caramel) caramel color is used in cognac, sherry, and vinegar. Class III (Ammonia caramel) caramel color is used in beers, whereas, class IV (Sulfite ammonia caramel) caramel color is used in non-alcoholic soft drinks [5,6].

The main use of caramel colors is to impart colors to beverages, however, they also serve other important functions. Caramel colors facilitate flavor retention and retard flavor changes in various alcoholic beverages [7]. Each class of caramel color are physically and chemically different and pose safety implications. Industrial caramel is composed of residual sucrose along with other mono-, and oligosaccharides such as glucose and fructose. And several degradation compounds mainly 5-hydroxymethyl furfural (5HMF) [8]. Such degradation compounds are present in trace amounts; as a result, it is very difficult to detect their presence in alcoholic beverages. Moreover, many of the degradation compounds show adverse effects on human health. Although caramel color containing ammonia often produce metabolites which pose health hazards, caramel colors meeting specifications can be used without any adverse effects. Therefore, it is important to detect, identify and quantify the additives in alcoholic beverages [8,9].

Many techniques including HPLC [8,10], UPLC – MS/MS [11], have been used to identify components of caramel color. Although these techniques provide reliable, sensitive, and reproducible results, they suffer from the limitation on part of their destructive, and expensive nature. Moreover, these techniques require extensive sample preparation which makes them time-consuming. Spectroscopic methods have emerged as an effective outcome for such problems in the last few decades. One such technique is Derivative UV-Vis spectrophotometry; which can be used as a technique of choice for qualitative and quantitative screening of various alcoholic beverages for caramel colors. It requires minimal sample preparation and can be used to separate unresolved bands, and for eliminating the effects of baseline shifts and baseline tilts by differentiating the normal zero order spectra [12]. In the present study, an attempt has been made to identify the caramel color in nine samples of whiskey, rum, and other commercial alcoholic beverages using Ultraviolet-visible (UV-Vis) spectrophotometry. Zero-order and first-order spectra were obtained to identify the maxima and minima peaks of caramel in various alcoholic beverages.

Materials and Methods

Collection of samples – Nine samples of alcoholic beverages were collected from the local markets of Chandigarh and Patiala. Table 1 illustrates the samples collected for the present study.

Table 1: Details of Samples Analysed

S. No.	Brand/ Description	Details				Source
		Batch No.	Mfg. Date	Proof	Distilled Blended and Bottled	
1.	Sun Caramel Ammonium Sulphate process food grade 15-4467, CML-L8156879, Colour int. 0.10-0.60 Manufacture- Sun Food Tech, Distt. Alwar, Rajasthan, India	4192 Type IV	Dec 2008			Local market, Chandigarh
2.	Officer's Choice Whiskey	72	Jan 2009	75^0	Batra Breweries	Local Market Chandigarh
3.	Officer's Choice Deluxe XXX Rum	OCR	Dec 2008	75^0	Rana Sugars Ltd. Village Louhka, Tehsil Patti, Distt. Tarn Taran, Punjab	Local Market, Chandigarh
4.	Officer's Choice Deluxe Whiskey	OCW 32	Jan 2009	75^0	Rana Sugars Ltd. Village Louhka, Tehsil Patti, Distt. Tarn Taran, Punjab	Local Market, Chandigarh

5.	Old Monk Rum	283	Jan 2009	75^0	Mohan Meakin Ltd. Mohan Gram, Bhankarpur, SAS Nagar (Mohali), Punjab	Local Market, Chandigarh
6.	Tohfa	----	----	50^0	Khasa Distillery Co. Gursimran Distilleries. KhasaDistt., Amritsar, Punjab	Local Market, Chandigarh
7.	Malwa	57	Dec 2008	50^0	Chandigarh Distilleries & Bottlers Ltd. BanurDistt. Punjab	Local Market, Patiala
8.	Lal Gulab	216	Dec 2008	50^0	Pioneer Industries Ltd. Pathankot, 145001, DisttGurdaspur, Punjab	Local Market, Patiala
9.	Santra	B122	Dec 2008	50^0	Ashoka Distilleries & Chemicals Pvt. Ltd. HathnDistt. Palwal, Haryana 121103	Local Market Patiala
10.	Jagadhri	81	Jan 2009	50^0	Haryana Distillery, Yamuna Nagar, Haryana	Local Market, Patiala

Table 2: pH of standard caramel solution and different alcoholic beverages

Code	Type	pH
1.	Standard Caramel Solution	4.48
2.	Officer's Choice Whiskey	4.85
3.	Officer's Choice Deluxe Rum	4.65
4.	Officer's Choice Deluxe Whiskey	5.02
5.	Old Monk's Rum	4.10
6.	Tohfa	4.34
7.	Malwa	4.26
8.	Lal Gulab	4.25
9.	Santra	4.28
10.	Jagadhri	8.00

Table 3: Maxima and Minima of Zero and First Order Spectra for Standard Caramel Solution and various alcoholic beverages

S. No	Spectrum for Standard Caramel Solution	Maxima (in nm)	Minima (in nm)
1.	Zero Order	281.0, 225.5	---
	First Order	260.0, 215.5	295.0, 237.5
2.	Zero Order	276.5, 217.5	---
	First Order	263.5, 214.5	295.0, 239.5
3.	Zero Order	279.5, 221.0	---
	First Order	264.5	290.5, 230.5
4.	Zero Order	280.0, 222.0	---
	First Order	342.5, 265.0	381.0, 295.0, 220.5
5.	Zero Order	279.5, 223.0	---
	First Order	264.5, 225.5	295.0, 234.5
6.	Zero Order	284.0, 235.5	---
	First Order	265.5, 225.5	299.0, 243.0
7.	Zero Order	285.0, 235.0	---
	First Order	265.5, 224.0	300.5, 243.0
8.	Zero Order	284.0, 219.0	---
	First Order	265.5, 214.5	299.0, 238.0
9.	Zero Order	284.5, 246.0, 235.0, 217.0	---
	First Order	349.0, 265.5	399.0, 299.0, 243.0
10.	Zero Order	284.0, 235.0	---
	First Order	344.5, 265.5, 223.0	375.5, 299.5, 243.0

Preparation of Standard Sample of Caramel - 500 µg of Caramel powder (manufactured by Sun Food Technology) was dissolved in a mixture of ethanol: water (1:1). Since ethanol and water have cut off wavelength at 210 and 190 nm respectively, therefore, they were used as a solvent in the present study. The pH of standard caramel solution and liquor samples were recorded using pH/Ion 510 manufactured by Eutech Instruments. The respective pH of standard caramel solution and liquor samples are provided in table 2.

Instrumental parameters and analysis – All the samples and standard caramel solution were scanned to obtain zero-order spectra in the region of 210 nm – 700 nm using Shimadzu UV-Vis 1700, Pharmaspec (Japan) spectrometer equipped with UVProbe software (Version 2.0). The samples were diluted to ten times using distilled water. Samples were analyzed in absorbance mode with a recording range of 0.00A – 1.00A and 3 scans were taken for each recording. Once the zero order spectra were recorded they were converted into first-order derivatives of their respective zero order spectra.

Results and Discussion

In the present study, an attempt has been made to identify the caramel in different types of alcoholic beverages (including whiskey, rum, and other local commercial liquors). The standard caramel solution and nine samples of different alcoholic beverages were analyzed using derivative UV-Vis spectrophotometry. Table 3 summarizes the maxima and minima results for zero order and first order analysis of standard caramel solution and nine samples of various alcoholic beverages.

a) Results of Zero Order Uv-Vis spectrophotometry.

Zero-order spectra of standard caramel solution and all samples show two maxima without any minima except for the sample of Santa which shows four maxima. Zero-order spectra of standard caramel solution demonstrate maxima at 281 nm and 225.5 nm. Officer's choice whiskey showed maxima at 276.5 nm and 217.5 nm, whereas an officer's choice deluxe whiskey showed maxima at 280 nm and 222 nm. Officer's choice deluxe rum shows maxima at 279.5 nm and 221 nm, meanwhile, old monk rum shows maxima at 279.5 nm and 223 nm. The spectra of commercial liquors show a greater variation in the wavelength at which maxima are observed. Tofa liquor displayed maxima at 284 nm and 235 nm; Malwa displayed maxima at 285 nm and 235 nm; Lal Gulab displayed maxima at 284 nm and 219 nm; Santra displayed maxima at 284.5 nm, 246 nm, 235 nm, and 217 nm; Jagadhri displayed maxima at 284 nm and 235 nm.

From the comparison of zero order spectra of standard caramel solution and zero order spectra of alcoholic beverages, it is evident that the maxima of 281 nm and 225 nm from standard caramel solution can be used for identification of caramel colors. The maxima of 281 for other alcoholic beverages fall in the range of 281 ± 5 nm, however, the maxima of 225 nm shows more variation with a range of 225 ± 10 nm. Therefore, it is safe to say that in the case of zero-order spectra, maxima 281 nm is a better peak for identification of caramel color.

b) Results of First Order Derivative UV-Vis spectrophotometry

First order spectra of standard caramel solution and nine samples of alcoholic beverages show two maxima and two minima with the exception of Officer's choice deluxe rum (one maxima and two minima), Officer's choice deluxe whiskey and Santr (two maxima and three minima each), and Jagadhri (three maxima and three minima). First order spectra of standard caramel solution showed maxima at 260 nm and 215.5 nm with minima at 295 nm and 237.5 nm. First order spectra of Officer's choice whiskey showed maxima at 263.5 nm and 214.5 nm with minima at 295 nm and 237.5 nm and Officer's choice deluxe whiskey showed maxima at 342.5 nm and 265 nm with minima at 381 nm, 295 nm, and 220.5 nm; Officer's choice deluxe rum showed maxima at 264.5 nm and minima at 290.5 nm and 230.5 nm, whereas, Old Monk rum showed maxima at 264.5 nm and 225.5 nm; Tofa showed maxima at 265.5 nm, and 225.5 nm with minima at 299 nm and 243 nm; Malwa showed maxima at 265.5 nm, and 224 nm whereas the minima was at 300.5 nm, and 243 nm; Lal Gulab showed maxima at 265.5 nm, and 214.5 nm with minima at 299 nm a, and 238 nm; Santra showed maxima at 349 nm, and 265.5 nm with minima at 399 nm, 299 nm, and 243 nm; Jagadhri showed maxima at 344.5 nm, 265.5 nm, and 223 nm with minima at 375.5 nm, 299.5 nm, and 243 nm.

Standard caramel solution shows two maxima (260 nm, and 215.5 nm) and two minima (295 nm, and 237.5 nm). The maxima at 260 nm show a range of 260 ± 1 nm for the samples, whereas, the 215.5 nm maxima show a range of 215 ± 10 nm. However, the maxima at 215.5 nm are absent in Officer's choice deluxe rum, Officer's choice deluxe whiskey, and Santra commercial liquor. In the case of minima of 295 nm, the observed range lies within 295 ± 5 nm, whereas, the range for maxima of 237.5 nm lies within 238 ± 5 nm.

Conclusion

The present study was conducted as an attempt to use derivative UV-Vis spectrophotometry to identify caramel colors in various factory-made and commercial alcoholic beverages. It can be concluded that derivative UV-Vis spectrophotometry can be used successfully to detect the caramel colors. Moreover, it is a non-destructive technique that requires minimal sample preparation. It is an easily available technique having a low per sample analysis cost.

Acknowledgments:

None to declare

Conflict of Interest:

None to declare

References

1. Chakrabarti A., Rai B. & Panda S. Producer, sellers, and drinkers - studies of noncommercial alcohol in nine countries. Int. Cent. alcohol policies Monogr. Ser. 2012;33–38.
2. Punia B.S., Yadav P.K., Bumbrah G.S. & Sharma, R.M. Analysis of Illicit liquor by headspace gas chromatography - mass spectrometry (HS-GC-MS): A Preliminary study. J. AOAC Int. 2017;100:1–17.
3. Yadav P.K. & Sharma R.M. Classification of illicit liquors based on their geographic origin using Attenuated total reflectance (ATR) – Fourier transform infrared (FT-IR) spectroscopy and chemometrics. Forensic Sci. Int. 2019;295:e1–e5.
4. Kuhnert N. Caramel: Properties and Analysis. Encyclopedia of Food and Health (2016). doi:10.1016/B978-0-12-384947-2.00112-4.
5. Vollmuth T.A. Caramel color safety – An update. Food Chem. Toxicol. 2018;111:578–96.
6. Tolgahan Kocadağlı, Vural Gökmen. Caramelization in Foods: A Food Quality and Safety Perspective. 2019.pp.18-29. DOI: 10.1016/B978-0-08-100596-5.21630-2.
7. I Chappel C. & C Howell J. Caramel colours—A historical introduction. Food and chemical toxicology : an international journal published for the British Industrial Biological Research Association 30, (1992).
8. Pons I., Garrault C., Jaubert J.N., Morel J. & Fenyo J.C. Analysis of aromatic caramel. Food Chem. 1991;39:311–20.
9. Myers D.V & Howell J.C. Characterization and specifications of caramel colours: An overview. Food and chemical toxicology: an international journal published for the British Industrial Biological Research Association 30, (1992).
10. Licht B.H. et al.Characterization of Caramel Colour IV. Food and chemical toxicology : an international journal published for the British Industrial Biological Research Association 30, (1992).
11. Fierens T. et al. Caramel colour and process contaminants in foods and beverages: Part II – Occurrence data and exposure assessment of 2-acetyl-4-(1,2,3,4-tetrahydroxybutyl)imidazole (THI) and 4-methylimidazole (4-MEI) in Belgium. Food Chem. 2018;255:372–79.
12. Bosch Ojeda, C. & Sanchez Rojas F. Recent developments in derivative ultraviolet/visible absorption spectrophotometry. Anal. Chim. Acta. 2004;518:1–24.