Analytica Chimica Acta 940 (2016) 104–112

Contents lists available at ScienceDirect

Analytica Chimica Acta

journal homepage: www.elsevier.com/locate/aca

Paper spray mass spectrometry and PLS-DA improved by variable selection for the forensic discrimination of beers

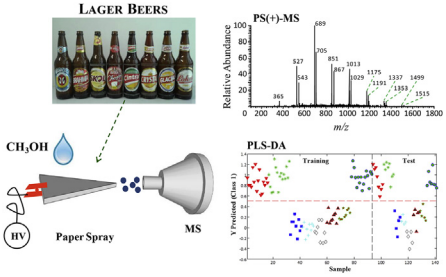
Hebert Vinicius Pereira [a], Victória Silva Amador [a], Marcelo Martins Sena [a, b], Rodinei Augusti [a], Evandro Piccin [a, *]

[a] Departamento de Química, Instituto de Ciências Exatas, Universidade Federal de Minas Gerais, Belo Horizonte, MG, Brazil
[b] Instituto Nacional de Ciência e Tecnologia em Bioanalítica, Campinas, SP, Brazil

HIGHLIGHTS

- For the first time a supervised classification multivariate method was used for PS-MS.
- Direct analysis by PS-MS combined with PLS-DA was used for fast beer differentiation.
- Differentiation was studied in a forensic context for beers of a single type (lagers).
- Variable selection was utilised for improving the PLS-DA model.
- OPS (ordered predictors selection) provided a method with 100% of reliability rate.

GRAPHICAL ABSTRACT



ARTICLE INFO

Article history:
Received 14 June 2016
Received in revised form
2 August 2016
Accepted 3 August 2016
Available online 6 August 2016

Keywords:
Ambient ionization
Paper spray
Mass spectrometry
Beer
Food and beverage analysis
Partial least squares discriminant analysis
Forensic analysis

ABSTRACT

Paper spray mass spectrometry (PS-MS) combined with partial least squares discriminant analysis (PLS-DA) was applied for the first time in a forensic context to a fast and effective differentiation of beers. Eight different brands of American standard lager beers produced by four different breweries (141 samples from 55 batches) were studied with the aim at performing a differentiation according to their market prices. The three leader brands in the Brazilian beer market, which have been subject to fraud, were modeled as the higher-price class, while the five brands most used for counterfeiting were modeled as the lower-price class. Parameters affecting the paper spray ionization were examined and optimized. The best MS signal stability and intensity was obtained while using the positive ion mode, with PS(+) mass spectra characterized by intense pairs of signals corresponding to sodium and potassium adducts of malto-oligosaccharides. Discrimination was not apparent neither by using visual inspection nor principal component analysis (PCA). However, supervised classification models provided high rates of sensitivity and specificity. A PLS-DA model using full scan mass spectra were improved by variable selection with ordered predictors selection (OPS), providing 100% of reliability rate and reducing the number of variables from 1701 to 60. This model was interpreted by detecting fifteen variables as the most significant VIP (variable importance in projection) scores, which were therefore considered diagnostic ions for this type of beer counterfeit.

* Corresponding author. Universidade Federal de Minas Gerais, Av. Pres. Antônio Carlos, 6627, Pampulha, 31270-901, Belo Horizonte, Minas Gerais, Brazil.
E-mail address: evandropiccin@gmail.com (E. Piccin).

http://dx.doi.org/10.1016/j.aca.2016.08.002
0003-2670/

1. Introduction

Brewing industry is a huge global business and one of the most important sectors in the food and beverage industry. In fact, after water and tea, beer is considered the most popular drink, being consumed all over the world. The beer market is expected to generate about US$ 690 billion in sales by 2020, with China, United States, and Brazil as the leading producers. Lager beers represent more than 90% of the total production [1–3].

Four basic raw ingredients are needed for beer production: water, malt, hops, and yeast. The main stages of the brewing process include malting, milling, mashing, wort separation, boiling, fermentation, and clarification. The properties, quality, and amounts of the ingredients together with the way to perform the stages of the brewing process strongly affect the beer characteristics. The fermentation stage, for example, defines the type of the beer. While lager beers are brewed at low temperatures using bottom-fermenting yeast, ale beers are fermented at high temperatures and the yeast remains on the surface of the liquid during the process. Previous to fermentation, other important stages of brewing are the malting and mashing, where polysaccharides from the cereals are hydrolyzed to low molecular weight sugars as well as to malto-oligosaccharides. The final bottled product, i.e. the beer, is a complex mixture containing several inorganic salts and more than 800 organic compounds [1,4].

Similarly to many other countries, the Brazilian beer market is dominated by large companies that basically commercialize American standard lagers. Only four companies (Anheuser-Busch InBev, Grupo Petrópolis, Brasil Kirin, and Heineken) produce around 96% of the beer consumed in Brazil [5]. Anheuser-Busch InBev has alone 68% of this market, and produces the three most consumed brands. Owing to their effective marketing, customer's preference, higher quality and prices in their category, these brands have been the subject of frauds. In fact, several cases of beer counterfeiting have been recently reported, wherein the counterfeiters have switched the labels and bottle caps of less expensive brands by the labels and caps of the more expensive market leader brands [6–8]. This type of fraud is attractive when involving a large amount of bottles, since the most consumed beers in Brazil are commercialized in similar standard bottles of 600 mL. Therefore, the development of fast and reliable analytical methods for distinguishing beers of similar characteristics, such as between American standard lagers from different brands and/or brewers, is very important for ensuring the security of both the consumers and the producers. Furthermore, authenticity studies using advanced analytical techniques are of major importance as they usually establish the differences on the composition of food products.

Many analytical techniques have been utilised to differentiate and classify beers, including gas chromatography coupled to flame ionization detection (GC-FID) [9] and mass spectrometry (GC-MS) [10,11], liquid chromatography coupled to ultraviolet detection (LC-UV) [12] and to mass spectrometry (LC-MS) [13,14], spectrofluorimetry [15], infrared spectroscopy [16–19], inductively coupled plasma - mass spectrometry (ICP-MS) [20], nuclear magnetic resonance (NMR) spectrometry [21,22], potentiometric [23] and voltammetric [24] electronic tongues, and mass spectrometry [25–29]. More recently, data fusion of different analytical techniques, such as spectrofluorimetry, mid-infrared, near-infrared, and UV-visible spectroscopies, has been used aiming at improving the discrimination of beers, in comparison to individual techniques [30–33]. However, this strategy has the disadvantages of being slower and more laborious.

The chemical discrimination of beers is multivariate in nature. Thus, almost all of the analytical approaches developed using the techniques mentioned above employed chemometric tools, such as principal component analysis (PCA), linear discriminant analysis (LDA), or partial least squares discriminant analysis (PLS-DA). It is important to highlight that the majority of the methods focused on the classification and differentiation between special and regular beers, as well as on the discrimination among beers of different styles [10–13,16,17,19,23–26,29]. However, less attention has been given to studies involving the differentiation of beers of the same style [9,14,15,20,27,28], a more difficult task from the analytical point of view.

Since the introduction of paper spray mass spectrometry (PS-MS) around six years ago [34], there is a steady increase in the development of tools and analytical methodologies involving this technology. As an ambient MS technique [35,36], PS-MS provides qualitative and quantitative chemical analyses of complex mixtures in the open atmosphere with no or minimal sample preparation. PS-MS is also characterized as rapid, cost-effective, and reliable [34,37]. Recent advances in PS-MS instrumentation include the introduction of the zero volt form of PS [38], low voltage PS with carbon nanotubes impregnated on the paper surface [39], the fabrication of cartridges for integrated solid phase extraction [40] and with continuous solvent supply capabilities [41,42], and the use of paraffin barriers on the paper substrate for improving PS performance [43]. PS-MS has proven to be suitable for a diverse array of analytical applications [44–48]. In food analysis, PS-MS has been utilised for the determination of additives and contaminants [49,50], and for the characterization of coffees and teas [51,52].

In spite of its great potential, a very small number of papers have applied PS-MS in combination with chemometric tools. And these applications have been limited to PCA, an unsupervised chemometric method [51–53]. Although unsupervised multivariate methods are more reliable to observe true differences among different samples, they are not the most appropriate tool for developing models that allow previous designations of specific sets of samples and determine precise thresholds for classification. For this aim, supervised classification multivariate methods are required, such as PLS-DA. Thus, this works aims at developing a simple and rapid analytical method for discriminating different brands of American standard lager beers commercialized in Brazil by combining PS-MS and a PLS-DA model in an application focussed on a specific forensic problem. The three most consumed brands of Brazilian beers, which are also the main targets of counterfeiting, were jointly modeled as the adulterated class. On the other hand, five of the cheaper brands most frequently reported as adulterants [6–8] were modeled as the adulterant class. The PLS-DA classification model were improved by variable selection using the recently proposed ordered predictors selection (OPS) [54], and the model was interpreted by detecting the most discriminant spectral variables in a search for diagnostic ions.

2. Materials and methods

2.1. Samples and materials

A total of 141 bottled American lager beers (600 mL) of eight different brands produced by four different breweries were purchased at local stores. Table 1 presents the detailed information of the samples. For the brands produced by Anheuser-Busch InBev (Brahma, Antarctica, and Skol), 10 different batches were analyzed, with one of the batches containing 6 bottles and the other 9 batches containing 2 bottles each (total of 24 bottles for each brand). Beers produced by Brasil Kirin (Cintra and Glacial), Grupo Petrópolis (Crystal and Lokal), and Cervejaria Krill (A Outra) were represented by 5 batches per brand, with one batch containing 6 bottles and the other 4 batches containing 2 bottles each (except for the brand Crystal, which one of the batches was represented by only one