Microchemical Journal 129 (2016) 243–248

Contents lists available at ScienceDirect

Microchemical Journal

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

Tracing the geographical origin of Argentinean lemon juices based on trace element profiles using advanced chemometric techniques

Jose E. Gaiad [a], Melisa J. Hidalgo [a], Roxana N. Villafañe [b], Eduardo J. Marchevsky [b,*], Roberto G. Pellerano [a,*]

[a] Instituto de Química Básica y Aplicada del Nordeste Argentino (IQUIBA-NEA), Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET), Facultad de Ciencias Exactas y Naturales y Agrimensura, Universidad Nacional del Nordeste (UNNE), Av. Libertad 5470, Corrientes 3400, Argentina
[b] Instituto de Química San Luis (INQUISAL), Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET), Facultad de Química, Bioquímica y Farmacia, Universidad Nacional de San Luis (UNSL), Av. Ejército de los Andes 950, San Luis 5700, Argentina

A R T I C L E I N F O

Article history:
Received 31 March 2016
Received in revised form 23 June 2016
Accepted 1 July 2016
Available online 2 July 2016

Keywords:
Citrus limon
Multi-element analysis
Geographical origin
ICP-MS
Chemometrics

A B S T R A C T

This study examines the application of chemometric techniques associated with trace element concentrations for origin evaluation of lemon juice samples. Seventy-four lemon juice samples from three different provinces of Argentina were evaluated according to their microelement contents to identify differences in patterns of elements in the three provinces. Inductively coupled plasma mass spectrometry (ICP-MS) was used for the determination of twenty-five elements (Ag, Al, As, Ba, Bi, Co, Cr, Cu, Fe, Ga, In, La, Li, Mn, Mo, Ni, Rb, Sb, Sc, Se, Sn, Sr, Tl, V, and Zn). Once the analytical data were collected, supervised pattern recognition techniques were applied to construct classification/discrimination rules to predict the origin of samples on the basis of their profiles of trace elements. Namely, linear discriminant analysis (LDA), partial least square discriminant analysis (PLS-DA), k-nearest neighbors (k-NN), random forest (RF), and support vector machine with radial basis function Kernel (SVM). The results indicated that it was feasible to attribute unknown lemon juice samples to its geographical origin. SVM had better performance compared to RF, k-NN, LDA and PLS-DA, listed in descending order. Eventually, this study verifies that trace element pattern is a powerful geographical indicator when identifying the origin of lemon juice samples by analyzing trace element data with the help of SVM technique. This level of accuracy provides an interesting foundation to propose the combination of trace element contents with SVM technique as a valuable tool to evaluate the geographical origin of lemon juice samples produced in Argentina.

1. Introduction

Natural lemon (*Citrus limon* (L.) Burm.) juices are rich sources of antioxidant nutrients such as flavonoids, carotenoids, and vitamin C; essential elements, K, Cu, Fe, Mg, and Zn; and soluble as well as insoluble dietary fiber. Together these nutrients promote several health benefits and provide protection against several illnesses [1,2]. In general, lemon fruits can be commercialized as fresh fruits, juices or oil in the international market. Today, Argentina produces around 1500×10^3 metric tons a year, mainly of the Genova and Eureka varieties. Taking the world production (around 6000×10^3 metric tons/year), Argentina's share is 20% approximately [3]. The lemon fruit annual production can vary between countries because of climatic variations.

The most important producing regions of lemon fruits in Argentina are mainly concentrated in two principal regions: *Argentine northwest (NW)* that is a region formed by three provinces, Tucuman, Salta and Jujuy, which share their borders with Bolivia and Chile; and *Argentine Northeastern* (NE) formed by three provinces, namely Entre Rios, Corrientes, and Misiones, which share their borders with Brasil, Paraguay and Uruguay. The NW region presents important competitive advantages in relation to the NE, principally because this region presents very low incidence of some important botanical diseases, that are enabling exports of lemon juices and fruits to markets such as the European Community or United States, principally [4].

For these reasons, producers, traders and consumers are especially interested in correct labelling of origin and traceability in lemon juice products. Determination of geographical origin authenticity is an important issue for the growing food industry in quality control and safety of food [5]. In Argentina, there are no established methodology to determine the traceability or geographical origins of Argentinean lemon juices [6]. In this context, the use of multielemental analytical techniques has increasingly been used to determine the geographical origin

* Corresponding authors.
E-mail addresses: eduardo.marchev@gmail.com (E.J. Marchevsky), roberto.pellerano@comunidad.unne.edu.ar (R.G. Pellerano).

http://dx.doi.org/10.1016/j.microc.2016.07.002
0026-265X/

of foods [7]. In general, the techniques to obtain elemental fingerprint of food are those with multi-element detection capability, such as ICP-based techniques [8]. Chemometric analysis of the complex element composition data obtained by these instrumental methods provides a better interpretation and the possibility to acquire relevant information about genuineness of these foods [9].

The main objectives of this work were the determination of trace element contents of lemon juice samples and the use of that chemical information to obtain adequate classification models to authenticate Argentinean lemon juice samples. Accordingly, an ICP-MS method has been proposed and the contents of 25 trace elements (Ag, Al, As, Ba, Bi, Co, Cr, Cu, Fe, Ga, In, La, Li, Mn, Mo, Ni, Rb, Sb, Sc, Se, Sn, Sr, Tl, V, and Zn) have been determined in lemon juice obtained from fruits cultivated in Argentina and derived from the three best-known lemon producing Argentinean regions: Northwest (Jujuy and Tucumán provinces) and Northeast (Corrientes province). In order to distinguish the geographical origin of the considered lemon juice samples, pattern recognition techniques such as principal component analysis (PCA), linear discriminant analysis (LDA), partial least squares discriminant analysis (PLS DA), k nearest neighbors (kNN), support vector machines (SVM), and random forest (RF) have been applied.

2. Material and methods

2.1. Reagents

All the chemicals used were of the highest purity available and all the glass materials used were washed with nitric acid and rinsed with ultrapure water. Ultrapure deionized water with a resistivity of 18.1 MΩ cm^{-1} was used exclusively. Ultrapure grade 65% (m/m) HNO_3 was acquired from Sigma (St. Louis, MO, USA). Nitric acid was further purified by sub-boiling distillation. Mono and multi-element standard solutions of trace analysis grade were purchased from Sigma-Aldrich and Agilent.

2.2. Apparatus

A high-performance microwave digestion oven, Milestone® (Chicago, USA) model Ethos One was used to digest the samples. The trace elements concentrations in digested samples has been carried out by Agilent 7700 cx (Agilent Technologies, Santa Clara, CA) ICP-MS spectrometer powered by a 27.12 MHZ radiofrequency solid-state generator at 1500 W. The ICP torch was a Fassel-type torch. The ICP torch consists of a three-cylinder assembly, with injector diameter 2.5 mm. Ni sampler and skimmer cones of 1.0 mm and 0.4 mm were used. This instrument was equipped with a MicroMist glass concentric nebulizer combined with a cooled double-pass spray chamber made of quartz. To suppress polyatomic interferences originating from sample matrix, octopole reaction system (ORS) with 5 mL/min He as collision gas and kinetic energy discrimination mode was used (collision mode). The equipment is provided with off-axis ion lens, a quadrupole mass analyzer and an electron multiplier detector. All instrument parameters were optimized daily while aspirating the tuning solution. The selected isotopes for measurement were ^{107}Ag, ^{27}Al, ^{75}As, ^{137}Ba, ^{209}Bi, ^{59}Co, ^{53}Cr, ^{63}Cu, ^{56}Fe, ^{71}Ga, ^{115}In, ^{139}La, ^{7}Li, ^{55}Mn, ^{95}Mo, ^{60}Ni, ^{85}Rb, ^{121}Sb, ^{45}Sc, ^{78}Se, ^{118}Sn, ^{88}Sr, ^{205}Tl, ^{51}V, and ^{66}Zn.

2.3. Sample collection

In this work, we analyzed 74 fresh lemon juice samples derived from Argentinean mature fruits collected from different agricultural cooperatives and producers during 2014/2015. The lemon fruits obtained correspond to three botanical varieties: Eureka ($n = 25$), Lisboa ($n = 30$) and Genova ($n = 19$). Four different locations in the north region of Argentina were selected for fruit collection (Fig. 1). Three different sites located in two provinces corresponding to NW region: Tucumán (TN-I: Tafí Viejo and TN-II: Famaillá), and Jujuy (Jy: Santa Clara) provinces and one site corresponding to NE region: Corrientes (CTE: Bella Vista) province. In order to maintain homogeneity with respect to the collection season all samples were simultaneously collected in the different sites. Finally, it is important to emphasize that all fruit considered in this study were produced under the recommendations formulated by INTA (Instituto Nacional de Tecnología Agropecuaria, Argentina) for the application of agrochemicals for this crop.

Once in the laboratory, the fruits were cleaned and washed with deionized water. Juice was extracted with a domestic plastic reamer and strained to remove seeds. The samples were not centrifuged or filtered, other than remove large particles from fresh juices. All samples were freeze-dried for a minimum of 48 h at a chamber pressure of 0.05 mbar, homogenized and stored in labelled polyethylene zipper bags.

2.4. Analytical procedures

Approximately 1.0 g of dry samples were placed into a microwave-closed vessel, added to each flasks 2.0 mL of 30% (m/m) H_2O_2 and 6.0 mL of sub-boiling HNO_3 65% (m/m) and stood for 10 min. The microwave digestion program applied included the next temperature stages:

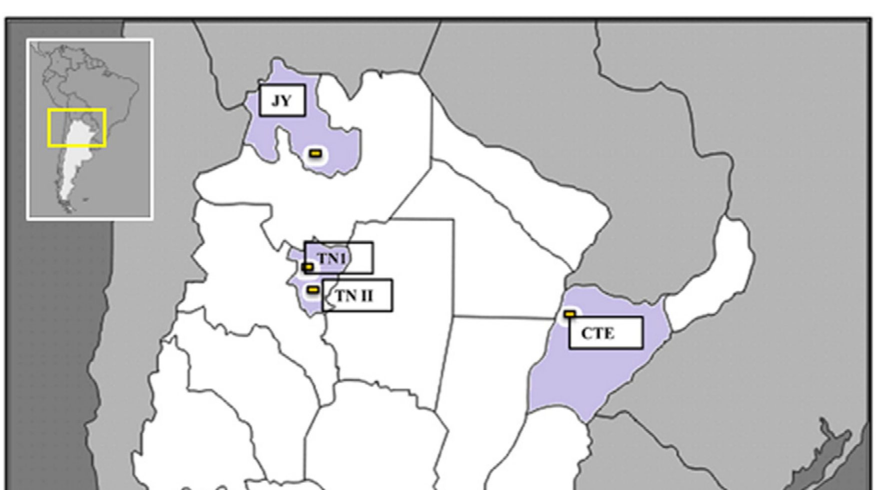

Fig. 1. Production areas of Argentinean lemon samples analyzed in this work: TN-I (Tafí Viejo, Tucumán), TN-II (Famaillá, Tucumán), JY (Santa Clara, Jujuy), and CTE (Bella Vista, Corrientes).