



# The content of Ca, Cu, Fe, Mg and Mn and antioxidant activity of green coffee brews



Ewelina Stelmach, Pawel Pohl\*, Anna Szymczycha-Madeja

Department of Analytical Chemistry, Faculty of Chemistry, Wrocław University of Technology, Wybrzeże Stanisława Wyspiańskiego 27, 50-370 Wrocław, Poland

## ARTICLE INFO

### Article history:

Received 17 October 2014

Received in revised form 30 December 2014

Accepted 21 February 2015

Available online 12 March 2015

### Keywords:

Green coffee

Coffee infusion

Macro- and microelements

Total phenolic compounds

Antioxidant activity

High resolution-continuum source flame atomic absorption spectrometry

## ABSTRACT

A simple and fast method of the analysis of green coffee infusions was developed to measure total concentrations of Ca, Cu, Fe, Mg and Mn by high resolution-continuum source flame atomic absorption spectrometry. The precision of the method was within 1–8%, while the accuracy was within –1% to 2%. The method was used to the analysis of infusions of twelve green coffees of different geographical origin. It was found that Ca and Mg were leached the easiest, i.e., on average 75% and 70%, respectively. As compared to the mug coffee preparation, the rate of the extraction of elements was increased when infusions were prepared using dripper or Turkish coffee preparation methods. Additionally, it was established that the antioxidant activity of green coffee infusions prepared using the mug coffee preparation was high, 75% on average, and positively correlated with the total content of phenolic compounds and the concentration of Ca in the brew.

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## 1. Introduction

Nowadays, drinking of roasted or instant coffee infusions is a habit of about two thirds' of a world population. Popularity of that beverage is a result of its stimulating effects, exceptional smell and taste. Coffee beans have a complex matrix with many substances, which can interact within the human body, e.g. improving its antioxidant activity and stimulating the neural system. During past few years, coffee drinking was also shown to be helpful in decreasing a risk of serious social disease such as diabetes type II, Parkinson and Alzheimer diseases or liver cancer (Chu, 2012). Recently, because of a pro-health and pro-healing status of antioxidants rich products, another coffee beverage has begun to enjoy a great popularity – infusion prepared using raw, unroasted green coffee beans. Green coffee infusions are believed to accelerate metabolism and in that way they can be helpful e.g., in reducing weight and preventing or overcoming obesity. Physiological studies made on mice suggest that a green coffee extract could be an effective fat absorption inhibitor and a suppressor of its metabolism in liver (Shimoda, Seki, & Aitani, 2006). In addition, it has been found to have an antihypertensive effect in rats that can last longer than 24 h after administration (Suzuki, Kagawa, Ochiai, Tokimitsu, & Saito, 2002).

The main constituents of green coffee beans are carbohydrates (up to 50% of the dry weight), both soluble (galactomannan, arabinogallactan) and insoluble in water (cellulose), phenolic species, i.e., caffeine, chlorogenic acids, in addition to polysaccharides, proteins, polyphenols, melanoids, lipids, and minerals (Bicchi, Binello, Pellegrino, & Vanni, 1995; Chu, 2012; Fischer, Reimann, Trovato, & Redgwell, 2001; Naidu, Sulochanamma, Sampathu, & Srinivas, 2008; Wei et al., 2012).

It has been recognized that benefits from consuming green coffee infusions are mainly related to the presence of phenolic compounds, especially chlorogenic acids that have a certain antioxidant activity (Bicchi et al., 1995; Naidu et al., 2008; Suzuki et al., 2002). However, it should be remembered that in case of natural products, health benefits are related to synergic effects of the whole matrix than the presence of individual substances. The consumption of the green coffee infusion is combined with the delivery to the organism of different components, also including elements. The latter constituents play a vital role in metabolism regulation and activation of enzymes; hence, they are generally required for the proper growth and development (Kaim & Schwederski, 1994). Elements present in the green coffee brew can also contribute to its recognized beneficial effects, e.g., a higher intake of Ca, K and Mg is believed to prevent hypertension (Suzuki et al., 2002). Minerals of green coffee can be present in beans at the level of 3.0–4.5 g per 100 g, which major part is attributed to K and Mg (Farah, 2012). Brews of this type of coffee, commonly prepared

\* Corresponding author. Tel./fax: +48 71 320 2494.

E-mail address: [pawel.pohl@pwr.wroc.pl](mailto:pawel.pohl@pwr.wroc.pl) (P. Pohl).

by pouring beans or ground beans with hot water and infusing for several minutes, do not contain such high amounts of elements. Hence, a moderate intake of green coffee infusions does not significantly contribute to recommended dietary intakes (RDIs) of elements as is also the case of roasted coffee (Pohl, Stelmach, Welna, & Szymczycha-Madeja, 2013). Nevertheless, small amounts of different major and minor elements in the green coffee brew may affect its sensory attributes (Farah, 2012).

Apparently from the literature, there are several studies about the mineral profile of green coffee beans (Barbosa et al., 2014; Bertrand et al., 2008; Filho, Polito, & Neto, 2007; Krivan, Barth, & Morales, 1993; Martin, Pablos, & Gonzalez, 1998; Muniz-Valencia, Jurado, Ceballos-Magana, Alcazar, & Reyes, 2013; Oleszczuk et al., 2007; Santato, Bertoldi, Perini, Camin, & Larcher, 2012) mainly undertaken in context of tracing their geographical origin. Unfortunately, the green coffee brew has never been the object of the study of its mineral content.

The objective of the present research was to retrieve the information about content of selected, physiologically and nutritionally important, macro- (Ca, Mg) and microelements (Fe, Mn, Cu) in infusions of green coffees of different origin and evaluate percentages of the leachability of these elements from beans to the infusion. For that aim, a simple and fast method of the determination of Ca, Cu, Fe, Mg and Mn in green coffee infusions by high resolution-continuum source flame atomic absorption spectrometry (HR-CS FAAS) without the need of their pre-treatment through wet oxidative digestion. Using the HR-CS FAAS instrument, it was possible to carry out the sequential multi-element analysis in a time shorter than required by ICP OES but with the suitable analytical performance in reference to the sensitivity, the accuracy and the precision (Oliveira et al., 2012; Ozdestan, 2014; Paz-Rodríguez, Domínguez-González, Aboal-Somoza, & Bermejo-Barrera, 2015). In addition, two spectrophotometric methods, i.e., Folin–Ciocalteu reagent (FCR) and free diphenylpicrylhydrazyl radical (DPPH<sup>•</sup>) assays, were applied to determinate the antioxidant activity of green coffee infusions and find out a possible correlation between the presence of phenolic and polyphenolic antioxidants and studied macro- and microelements in brews. What is more, different methods of coffee brewing, i.e., mug, Turkish, filter dripper and coffee machine coffee preparations, as well as times of brewing were used to establish conditions that can influence the final concentration of elements in green coffee infusions.

## 2. Experimental

### 2.1. Reagents and samples

Solutions of ACS grade 30% (m/m) H<sub>2</sub>O<sub>2</sub>, 37% HCl, and 65% (m/m) HNO<sub>3</sub> were supplied by J. T. Baker (Deventer, Netherlands). Solid Na<sub>2</sub>CO<sub>3</sub> and gallic acid (GA) in addition to a solution of the FCR were provided by Avantor Performance Materials (Gliwice, Poland). A solution of DPPH<sup>•</sup> was obtained from Sigma–Aldrich Chemie GmbH (Steinheim, Germany). Sigma–Aldrich TraceCERT<sup>®</sup> single-element 1000 µg ml<sup>−1</sup> standards of Ca, Cu, Fe, Mg and Mn were used to prepare multi-element standard solutions for the calibration of HR-CS FAAS. Re-distilled water was used in all preparations.

Samples of 12 green coffees from different regions of the world were provided by a café and a coffee roaster “Café Borówka” (Wrocław, Poland). It included arabica green coffee beans from Brazil [types Santos 1 (GC1) and Santos 2 (GC2)], India [type AA (GC3)], Nicaragua [type Papales (GC4)], Salvador [type SHG (GC5)], Ethiopia [type Djimmah (GC6)], Peru [type HB MCM (GC7)], Guatemala [type SHB (GC8)], Costa Rica [type SHB (GC9)],

Colombia [type Excelso (GC10)]. In addition, 2 green coffees from bioorganic farming (without artificial fertilizers) were analyzed, i.e., from Guatemala [type BIO (GC11)] and Thailand [type Doi Chaang BIO (GC12)]. The geographical origin of all collected samples of beans and their types (if applicable) were confirmed by the supplier.

### 2.2. Coffee brewing

Before brewing, coffee beans were ground in two steps: firstly in a domestic mechanical quern mill grinder with stainless steel querns (Severin, type KM3876), then, to obtain a fine-grained material of a particle size below 0.2 mm, in a manual coffee grinder with ceramic querns (Hario, Slim). Due to toughness, green coffee beans were frozen prior to grinding.

For the preparation of the mug green coffee infusion (method A), 1.0 g of ground green coffee beans was placed in a 200 ml Pyrex glass beaker and poured with 100 ml of hot (90 °C) re-distilled water and stirred. After 10 min brewing, the resulting light green infusion was separated from grounds by filtrating it through a dense filter paper. For the Turkish coffee preparation of the green coffee infusion, 1.0 g of ground (method B) or whole green coffee beans (method E) was placed in a 200 ml Pyrex glass beaker, poured with 100 ml of re-distilled water and heated to boil on a hot-plate with stirring for about 10 min. When water began to boil, resulting infusions were immediately filtered through dense filter papers. The dripped green coffee infusion (method C) was prepared in the following way: 1.0 g of ground green coffee beans was poured into a paper coffee filter (No. 2, Brigitta) placed in a plastic coffee cup dripper. Then, 100 ml of hot (90 °C) re-distilled water was dripped through the cup. At first, a small portion of water was poured and coffee was mixed into slurry with a glass rod, next, it was slowly poured and let to drip through coffee grounds. The resulting brew was collected into a glass beaker on which the dripper was mounted. Finally, to prepare the green coffee infusion using an overflow coffee machine (method D), 1.0 g of ground coffee was poured into a coffee filter (No. 2, Brigitta) placed in a filter holder of the coffee machine (Philips HD 7546/20). A water tank of the coffee machine was filled with 100 ml of re-distilled water and a brewing cycle was started. The resulting brew was collected in a jug.

Before use, utensils used in all methods of the green coffee infusion preparation were washed and finally rinsed several times with re-distilled water. For each brewing method, 3 parallel samples were prepared using fresh portions of green coffee and re-distilled water. Respective procedural blanks were also prepared and considered in final results.

### 2.3. Instrumentation

An Analytik Jena AG device (model contrAA<sup>®</sup> 700) was used to measure concentrations of Ca, Cu, Fe, Mg and Mn by HR-CS FAAS. The instrument was equipped with a xenon short-arc lamp operated in a hot-spot mode as a continuous radiation source and a high resolution Echelle type double monochromator (a pre-monochromator with a quartz prism and an Echelle grating). A charge couple device (CCD) array with high quantum efficiency and increased UV-sensitivity was used for the detection. A sample introduction system was consisted of a Ti single-slot 50 mm burner for a very sharp air-acetylene-lean flame, an adjustable nebulizer with an internal Pt/Rh capillary and a resistant ceramic impact bead and a polyphenylene sulfide (PPS) spray chamber with a mixing wing for aqueous solutions. All optimized instrumental settings used for the instrument are given in Table 1. Absorbance readings were carried out using a mean integration mode; signals were averaged over the selected integration time.

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