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Qualitative and quantitative ana lysis of acrylamide in coffee and some of its products

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Abstract
Acrylamide is extracted efficiently due to its high polarity and water solubility (Andrzejewski et al., 2004). Nevertheless, the level of extraction is also affected by factors such as water temperature, time of contact with ground coffee and pressure applied, all of which are conditioned by the type of coffee machine used. Levels of acrylamide from 1.7 to 75.0 µg/L have been found in coffee brews prepared with different extraction methods (Andrzejewski et al., 2004; Şenyuva and Gökmen, 2005; Alves et al., 2010; Sirot et al., 2012). In agreement with Alves et al. (2010), it is more realistic to evaluate the exposure to acrylamide by analyzing the beverage rather than the ground coffee before preparation. The aim of this study was to determine levels of acrylamide in coffee acquired from coffee vending machines. These beverages make a significant contribution to the coffee intake outside the home. This research has been intentionally limited to commercially available automatic coffee vending machines placed in busy public areas, such as train and bus stations, hospitals and universities, to estimate the exposure in real life conditions where the consumer does not have any impact on the preparation of the drink. Finally, the dietary exposure to acrylamide by the Spanish population through coffee machines was estimated.
Key word: Coffee, Acrylamidede, qualitative analysis

Introduction

Coffee is generally roasted at temperatures in the range of 220–250°C, and the time and speed of roasting play an important role on the sensorial properties, such as color, aroma, and taste. These are carefully fine-tuned to a characteristic profile leading to the clear identity of the coffee product. Coffee beans are thus subjected to relatively higher temperatures than other foods, and one can expect more than one pathway leading to acrylamide besides the Maillard reaction (Stadler and Scholz, 2004). The free asparagine concentration in green coffee beans lies within a very narrow range, typically from 30 to 90 mg/100 g. Conversely, experiments have shown that acrylamide is degraded or eliminated during roasting, and the profile of acrylamide formation during the roasting of coffee reflects this effect very clearly (Taeymans *et al.*, 2004). In coffee, acrylamide is formed at the very beginning of the roasting step, reaching more than 7 mg/kg, and then declines sharply toward the end of the roasting cycle due to higher rates of elimination. These and other factors affecting acrylamide formation in coffee (Figure1) are detailed below.

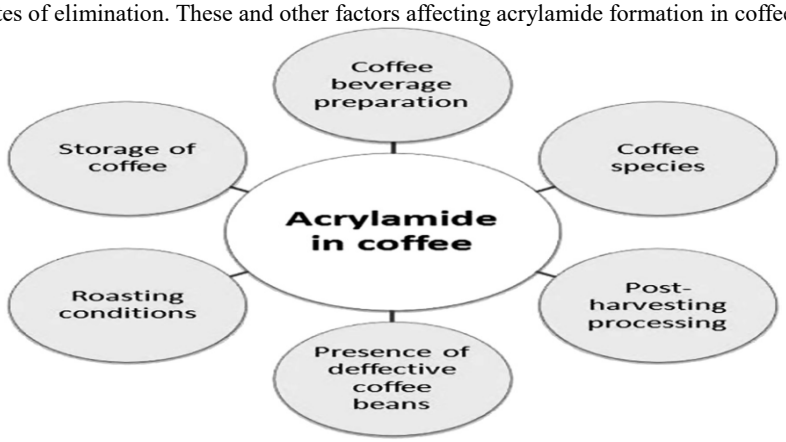

FIGURE 1 Factors affecting acrylamide formation in coffee.

MATERIALS AND METHODS

Chemicals Acrylamide standard (99%), potassium hexacyanoferrate(II) trihydrate (98%), zinc acetate dehydrate (99.5–101%) and methanol (HPLC grade) (99.9%) were purchased from Sigma–Aldrich (St. Louis, MO). [13C3]-Acrylamide (isotopic purity 99%) was from Cambridge Isotope Labs (Andover, MA). Formic acid (98%) was from Panreac (Barcelona, Spain). Milli-Q water used was produced using an Elix3 Millipore water purification system coupled to a Milli-Q module (model Advantage10; Millipore, Molsheim, France). All other chemicals, solvents and reagents were of analytical grade. Reversed-phase Oasis-HLB cartridges (30 mg, 1 mL) were from Waters (Milford, MA). Syringe filter units (0.45 µm, cellulose) were purchased from Análisis Vínicos (Tomelloso, Spain). Coffee samples Forty-one espresso coffee brew samples were acquired from different coffee vending machines in the Autonomous Community of Madrid (Spain). The vending machines were located in public areas with a high footfall of people, including hospitals, train and bus stations, universities, etc. All machines were different, involving different brewing procedures, types of coffee bean and roasting and different coffee/water ratios for extraction. The machines belonged to twelve of the most relevant companies that provide vending services in Spain. Fresh coffee samples were stored in 100 mL double-sealed containers and refrigerated at 4ºC for a maximum of three days until analysis. Volume of coffee beverage was measured. Measurement of pH pH was measured in coffee brews by using a pH meter at ambient temperature (CG-837 pH meter, Schott, Mainz, Germany).

Measurement of pH pH was measured in coffee brews by using a pH meter at ambient temperature (CG-837 pH meter, Schott, Mainz, Germany). Statistical analysis Statistical analysis was performed using Statgraphics Centurion XV (Herndon, VA). Data were expressed as mean ± standard deviation (SD). Relationships between the different parameters analyzed were evaluated by computing Pearson linear correlation coefficients at the $p < 0.05$ confidence level.

RESULTS AND DISCUSSION

Characterization of the coffee brews Coffee brews were characterized by pH, dry weight and browning. The pH values of the coffee brews ranged from 5.0 to 5.7, dry weight from 1.2% to 8.7% and browning from 1.3 to 5.8 units of absorbance (data not shown). The characteristics of coffee brews depend on variables such as green bean composition, roasting conditions, grinding, coffee/water ratio, brewing procedure, contact times and water temperature (Petracco, 2005). In the present study, coffee beverages were acquired from the most representative brands of coffee vending machines in the Spanish market. These machines differ pag. 3 M. Mesias, F.J. Morales / Journal of Food Composition and Analysis 48 (2016) 8–12 largely according to the technology applied, design, coffee origin, intensity of roasting and extraction conditions. Because of this, the coffee brews possessed different characteristics. This variability represents the broad scenario of coffee consumption from automatic vending services. Acrylamide levels in coffee brews Acrylamide levels in the coffee brews acquired from coffee vending machines ranged from 7.7 to 40.0 µg/L, with a mean value of 20.6 µg/L (Table 1,Fig. 1). All the samples contained acrylamide levels above the limit of quantification (LOQ

= 5 µg/L). Data distribution showed a high variability in the acrylamide content among samples and two outliers were identified (36.0 µg/L and 39.9 µg/L). This broad distribution in the acrylamide content is probably a consequence of the type of roasted coffee and the different conditions used to brew the coffee. The procedures to prepare a coffee brew are usually based on three different processes: decoction, infusion and pressure (Soares et al., 2015). In the decoction, ground coffee is kept in contact with water for a period of time, the extraction increasing with time of contact and water temperature. In infusion preparations, hot water flows through the ground coffee during a short period of time and, in consequence, extraction is lower than in the decoction method (Petracco, 2001). Finally, the pressure method is used to prepare espresso coffee. In this case, hot water is percolated under high pressure through ground coffee for a very short time, which may result in an incomplete extraction of acrylamide. However, due to the high coffee/water ratio used to prepare espresso coffee, the final acrylamide content may be higher than in other coffee brews. If we consider that the average coffee consumption for the Spanish population is 599 cups/person/year as reported by the Spanish Coffee Federation (SCF, 2013), corresponding to 1.64 cups of coffee/person/day, then the mean daily acrylamide intake would be 2.6 µg/person/day (range: 1.3–3.9 µg/person/day). Assuming a default value of 70 kg of body weight (bw) for an adult according to EFSA Scientific Committee (EFSA SC, 2012), the acrylamide intake from coffee would range from 0.018 to0.056 µg/kg bw/day (mean: 0.037 µg/kg bw/day). These results are similar to the daily intake of acrylamide from coffee estimated for adults from The Netherlands (0.048 µg/kg bw/day). Lower estimates have been reported for adults from Ireland (0.004 µg/kg bw/day), United Kingdom (0.005 µg/kg bw/day) and Latvia (0.029 µg/kg bw/day) whereas higher exposures have been found for Denmark (0.106 µg/kg bw/ day), France (0.168 µg/kg bw/day) and Sweden (0.171 µg/kg bw/ day) (Arisseto and Toledo, 2008). Obviously, these exposures depend on the consumption habits of the population since coffee consumption is widely variable between different countries and regions. An average consumption of coffee from 210 mL/day in the Polish adult population to as much as 1100 and 900 mL/day for Danish men and women (35–44 years age group), respectively, has been described (Granby and Fagt, 2004; Mojska et al., 2010).. In order to calculate the potential risk for human health through the consumption of acrylamide from coffee beverages, EFSA and JECFA (Joint Food and Agricultural Organization of the United Nations/World Health Organization (FAO/WHO) Expert Committee on Food Additives) have suggested using the MOE value, defining the margin of exposure (MOE) as the value calculated by dividing the dose causing cancer in animal studies by exposure of humans to acrylamide (FAO/WHO, 2011). In this respect, a MOE value of 8378 was obtained when comparing the mean exposure with the established BMDL10 (benchmark dose lower confidence limit 10%) for mammary tumors in female rats (0.31 mg/kg bw/day) and 4865 when the comparison is with the BMDL10 for Harderian gland tumors in male mice (0.18 mg/kg bw/day). The EFSA Scientific Committee considers that, for substances that are both genotoxic and carcinogenic, an MOE of 10,000 or higher is of low concern from a public health point of view (EFSA, 2015). Since the calculated MOE values are lower than 10000, it may be suggested that exposure to acrylamide through coffee consumption could imply a risk to public health. However, the maximum acrylamide exposure estimated in our study is much less than both the value of 0.2 mg/kg bw/day, defined as the lowest no observed adverse effect level (NOAEL) for a non-carcinogenic endpoint (FAO/WHO, 2011) and the value of 2.6 µg/kg bw/day estimated as the tolerable daily intake linked with the development of cancer (Tardiff et al., 2010). Therefore, it may be concluded that coffee consumption does not involve a high risk, although these conclusions should be revised for heavy coffee consumers (more than 5 cups/day). To avoid possible risk it is advisable to reduce acrylamide content in coffee and, consequently the dietary exposure to acrylamide, considering certain preventive actions and mitigation strategies.

Conclusion

The differences observed in the acrylamide content of coffee brew may result from the different roasted coffees used and the different conditions of preparation in the coffee vending machines. Consumption ofcoffee brew acquired from vending services contributes to the acrylamide exposure in humans in a way similar to other coffee brew preparations. since coffee brew is considered one of the most important contributors to acrylamide exposure in the diet of adults, it is necessary to define potential strategies to reduce the content of acrylamide in coffee. These findings are useful for risk assessment analyses in the Spanish population. Conflict of interest The authors have no conflict of interest.

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