C-7

QUANTIFICATION OF NITRATE AND NITRITE IN SPINACH AND LETTUCE BY REVERSE-PHASE HIGH PERFORMANCE LIQUID CHROMATOGRAPHY/UV

Edgar Pinto[1], Catarina Petisca[2*], Luís Filipe Amaro[3], Olívia Pinho[4], Isabel Ferreira[5]

[1 2 3 5] REQUIMTE- Serviço de Bromatologia, Faculdade de Farmácia da Universidade do Porto, Rua Anibal Cunha 164, 4099-030 Porto; Portugal
[4] REQUIMTEFaculdade de Ciências da Nutrição e Alimentação da Universidade do Porto, Rua Dr. Roberto Frias, 4200-465 Porto, Portugal
* Corresponding author–E-mail: catarinapetisca@fcna.up.pt; Phone: +351222078929; Fax: +351222003977

Vegetables tend to concentrate nitrate ions, thus, they are a major source of human exposure to this compound. Nitrate concentrations vary significantly, ranging from 1 to 10,000 mg kg^{-1} fresh weight, while nitrite levels in fresh vegetables are low (<2 mg kg^{-1}). However, nitrite levels in vegetables may increase during post-harvest storage by the action of indigenous bacteria and/or the presence of nitrate reductase, especially when they are left at room temperature or higher. Nitrate content in vegetables is concerned with food safety. The European Commission /EC) established maximum levels of nitrate in lettuce (Lactuca sativa L) and spinach (Spinacea oleracea). The vegetables producers should gradually modify their farming methods by applying the codes of Good Agricultural Practice (GAP) recommended at national levels, so as to comply with the maximum levels to reduce nitrate levels. A rapid and cost-effective RP-HPLC method with UV detector was validated for quantification of nitrate and nitrite in spinach and lettuce. The HPLC separation conditions were optimized with respect to chemical composition of the mobile phase, flow rate, chromatographic resolution and analysis time. The chromatographic separation was achieved using a HyPurity C18, m chromatographic column with 25 cm and elution with 0.01M n-octylamine to pH 6.5. Linearity was obtained over the tested concentration range of 0.3–15 mg/L of nitrate and nitrite. The linear regression equations of nitrate and nitrite standard curves were calculated as y = 24041x + 1941 and y = 33772x -2000, respectively. The correlation coefficients were both greater than 0.999, which showed a very good linearity. The detection limit of nitrate and nitrite, defined as a signal-to-noise ratio of 3, was the same, 0.05 mg/L. The method showed good sensitivity and can detect trace levels of nitrate and nitrite. Reproducibility of the measurements was evaluated by intra-day and inter-day analysis calculated from the results of 3 replicates. Repeated trails all obtained CV values less than 2%, pointing out high degrees of reproducibility. The recoveries of nitrate and nitrite spiked into vegetable samples were higher than 96%. Extraction of nitrate and nitrite into hot water prior to filtration and measurement is the most usual process; however, several interferents appear in the chromatogram. Extraction in presence of activated charcoal of freeze-dried and frozen samples was compared. The results of analysis of samples of Lactuca sativa L, Spinacea oleracea, Tetragonia tetragonioides showed that the method was fast, reliable and sensitive.

Keywords: nitrite, nitrate, ion-pair HPLC, vegetables

H-8
SCREENING OF FURANS IN AROMATIZED COFFEE SAMPLES BY SPME-GC-MS: COMPARISON WITH CONVENTIONAL COFFEE

Catarina Petisca[1*], Olívia Pinho[2], Isabel Ferreira[3]

[1 2 3] REQUIMTE- Serviço de Bromatologia, Faculdade de Farmácia da Universidade do Porto
[*] Corresponding author–E-mail: catarinapetisca@fcna.up.pt; Phone: +351222078929; Fax: +351222003977

Furan derivatives have traditionally been used as flavouring additives to food. In addition these compounds are formed during heating of carbohydrate rich food both during the industrial processing of food and at home during cooking. For some furan compounds like 5-hydroxymethylfurfural (HMF, 5-hydroxymethyl-2-furancarboxaldehyde) the levels exceed 1 g/kg in several food items, namely coffee. Furan itself has been regarded as a prioritised substance by EU. However, there are very little knowledge a number of furan related contaminates, like for example HMF and HMF derivatives, when comes to food sources, levels in food and toxicity. A risk assessment of HMF for use as a flavouring substance was performed by EFSA in 2005, and the use of HMF as a flavouring agent was put on hold because of potential genotoxicity. Several other HMF related substances are also used as flavouring substances and the knowledge of toxicity of these substances is very limited. In addition a number of potentially genotoxic furan related contaminates are produced in food and only a few of these compounds are investigated regarding occurrence in food. Various furan contaminants in heated and flavoured foods have only been studied to a limited extent, thus, analyses of furan derivatives such as 2-furfuraldehyde, 5-methylfurfuraldeyde, 2-furan-3-carboxaldehyde and furylacrolein, among others is very important. A brand-new line of aromatized espresso coffee, mixed the aroma and the scent of vanilla, caramel, almond liquor and irish cream to offer the connoisseurs a new fragrance and a pleasant form of expression is in the market, however, no studies were published concerning its furan composition or comparison with conventional espresso coffee. A simple and sensitive method for the screening of furans in espresso coffee was optimized using headspace solid-phase microextraction (SPME) and gas chromatography with mass detection. The SPME fiber, adsorption and desorption parameters were chosen to obtain the maximum m Carboxen-PDMS sensitivity for furan compounds. Headspace SPME using a 75 fiber provided effective sample enrichment. The optimized methodology was used to compare furan composition from four aromatized espresso coffee samples (vanilla, caramel, almond liquor and irish cream coffee samples), an Arabica and a blend of Arabica and Robusta espresso coffee. Twenty eight furans were identified in coffee samples. The furans were found to be the most predominant group of compounds amongst the coffee aromatics, except for almond liquor and irish cream espresso coffee samples. Depending of the flavour added, the furan profile and quantity of compounds detected changed.

Keywords: furans, aromatized, espresso coffee