Studies and Research on Caffeine Content of Various Products

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Abstract

Caffeine is a pharmacologically active component of many foods, beverages, dietary supplements and drugs.

Coffee and tea are among the best-known beverages in the world. Most people on a global scale consume coffee daily because of their caffeine-rich content. In addition to the benefits it exerts on the human body, it also has some negative aspects: some medical conditions require a restricted caffeine diet, some drugs cannot perform their pharmacological action in the presence of caffeine, it producing addiction. To overcome these aspects, people can replace normal coffee with decaffeinated coffee or black tea.

The present study has as main objective the study of the caffeine content of different brands of coffee and black tea on the Romanian market. For the study, analyzes were made on different traditional brands and the data obtained were then compared with those obtained for decaffeinated coffee and black tea. It was found that the highest content of coffee was P1 (0.89%), followed by sample P2 (0.46%) and sample P3 (0.22%). For decaffeinated coffee, the smallest quantity is found in sample P5 (0.01%), P4 having a higher content (0.21%).

It can be said that black tea is a good substitute for coffee because the caffeine's found values are closer to those present in classic coffee, having a percentage of 0.18%.

Keywords: caffeine, UV/ Vis spectrophotometric method, coffe, tea

1. Introduction

Caffeine (1,3,7-trimethyl-2,6-purindione or, in short, 1,3,7-trimethylxanthine) is a methylxanthine-containing purine alkaloid present in the leaves, seeds or fruits of over 63 species of plants around the world, where it acts as a natural pesticide that paralyzes and kills insects feeding on the plant (Wanyika *et al.*, 2010). The most common sources of caffeine are coffee, cocoa beans, cola nuts and tea leaves.

Coffee is, after water and tea, the most consumed drink in the world and has a great cultural, social and economic importance. Coffee can be a spice when added to different products, but in most cases it is consumed as a drink to satisfy sensory pleasures and its stimulating effects (Bulancea, 2002). Coffee has a stimulating effect on the central nervous system, muscle activity and thermogenic effect, it improves visual acuity, and has rapid laxative effect and it is an important diuretic etc.; but regular consumption in large quantities also induces negative consequences such as the promotion of cardiovascular disease, increased gastric secretions, insomnia, etc. (Bulancea, 2002), (Lascu, 2008). Tea is tonic, digestive, diuretic and sudorific (Lascu, 2008).

The chemical composition of coffee (caffeine, chlorogenic acids, lipids, sucrose, fats and proteins) oscillates widely, depending on a number of factors, the most important being: geographical origin, species, processing technology, beverage preparation, etc. (Bulancea, 2002, Franca *et al.*, 2002, Wanyika *et al.*, 2010, Perrois *et al.*, 2015).

Caffeine, the bioactive substance in coffee and black tea causes a vasodilatory effect in the vessels of the muscles and increases the force of contraction of the striated muscles by increasing the levels of free fatty acids and plasma glucose, thus reducing the balance of fluids in the body, stimulating the nervous system, of gastric peristalsis and gastrointestinal secretions (Wanyika *et al.*, 2010). In the cerebral cortex, it has a psychostimulatory effect, it increases attention, it makes intellectual work easier and a more perfect association of ideas with a better appreciation of sensory stimuli in humans; it has no euphoric effect and does not produce addiction as amphetamine, and at the level of vital bulb centers there is an analeptic effect (stimulation of the respiratory center), stimulation of the vasomotor center (vasoconstriction in certain vascular areas, which can return to normal low blood pressure). Caffeine is also used in pure form for the preparation of energy drinks as well as in the pharmaceutical industry, in combination with other substances (antipyretic analgesics to enhance their effect) (Health Line, 2008).

Over three cups of coffee per day (a cup containing approximately 150 mg of caffeine), coffee consumption can cause to more sensitive people the acceleration of the pulse, tremors, insomnia and high blood pressure. It can also cause benign (non-cancerous) damage to the breasts and may aggravate premenstrual symptoms to women who use it excessively. Caffeine crosses the placenta and enters the fetal circulation, and its pharmacological use has been associated with a low birth weight. Excessive drinking during breastfeeding can cause irritability and wakefulness in a breastfed child (Wanyika *et al.*, 2010). An excessive caffeine intake in some people seems to increase the sensitivity of the heart to emotional factors and other factors and thus it increases the incidence of additional systole and other arrhythmias. To reduce such inconvenience, decaffeinated coffee may be used.

The present study has as main objective the study of the caffeine content of different brands of coffee (with caffeine and decaffeinated) and black tea on the Romanian market.

2. Materials and Methods

2.1. Materials

Different brands of coffee of the two species (*Arabica* and *Robusta*) and black tea were collected from the local market in Suceava, Romania. The coffee (5 of which two without caffeine) and the tea samples were kept at room temperature throughout the analysis. All reagents were of analytical reagent grade (Sigma-Aldrich) and all solutions were prepared using distilled water.

2.2. Standard solutions for UV/VIS Spectrophotometry method

Caffeine stock solution (400 ppm) was prepared by dissolving 40.00 mg caffeine (anhydrous) in 100 ml of distilled water. 0, 4, 8, 16, 24 and 32 ppm Caffeine working solutions were prepared by serial dilution of the stock in 25 ml volumetric flasks with addition of 1.0 ml HCl 0.01 M before topping to the mark with distilled water (Aznar, 2011). A calibration curve was constructed before analysis of the samples. Deionized water was used as the blank. Caffeine is directly analyzed by measuring the absorbance at the wavelength of 273.5 nm with a UV - VIS - NIR spectrophotometer (Shimadzu, model 3600, Japan) from the research laboratory of the Faculty of Food Engineering of Suceava (Poroch-Seritan, 2016).

2.3. Sample preparation

Approximately 0.30 g sample (coffee / tea) is refluxed with 200 ml distilled water for 15 minutes and then the solution is filtered hot, finally adding 5g Na_2CO_3 . Extraction of caffeine is accomplished by treating the filtrate twice with 15 ml chloroform by gentle agitation for a few minutes. The chloroform extracts were combined by adding small amounts of Na_2SO_4 , then filtered

and brought to dryness. Once the chloroform is removed, 50 ml distilled water is added and shaken well until complete dissolution is then carried out quantitatively in a 100 ml volumetric flask with distilled water. Finally, over 10 ml of this last solution add 1 ml of 0.01 M hydrochloric acid and make up to 25 ml with distilled water (after Aznar, 2011). The caffeine levels of the samples were calculated from the regression equation of the best line of fit of the standards with the formula: y = 0.0601, R2 = 0.9991.

3. Results and Discussion

The caffeine content varies widely depending on the type of coffee, the method of roasting green beans and the way the beverages are prepared. In general, roasted coffee has less caffeine because the roasting process reduces the caffeine content of the coffee bean (Bulancea, 2002; Franca *et al.*, 2002; Wanyika *et al.*, 2010; Perrois *et al.*, 2015).

During roasting, coffee beans lose weight due to the evaporation of water and the decomposition of some of the substances they contain, losing up to 16-18%. This explains that during the roasting the coffee components are modified and a volume increase of the coffee beans is also achieved, up to 25%. It is also noted that about 50% of the weight loss of coffee beans (during the roasting process) is due to water evaporation, and the difference is the decomposition of organic substances, especially sugar, cellulose and caffeic acid. The content of caffeine in roasted coffee decreases very little, due to the sublimation of this substance during the roasting of the green coffee. As green coffee loses an important amount of water, the caffeine content of roasted coffee beans is close to the content of this coffee bean, sometimes even higher (Măndiță, 2002).

Coffee and tea contain cellulose, which is the main structural material of all plant cells and is insoluble in water. Due to this property, cellulose does not raise problems in the process of separating the active principle - caffeine. The active compounds in plants with polyphenolic structure and with some common properties are tannins or antioxidants (coffee and tea antioxidants have high absorption in the human body), usually divided into two groups: those that can be hydrolysed (pyrogallol or acid gallic) with water and those that can not be hydrolyzed or are hydrolysed with difficulty (condensed tannins or proanthocyanidins) (Aznar, 2011).

Caffeine is perfectly soluble in hot water so it can be extracted efficiently. Caffeine passes into the aqueous solution but also accompanied by other organic compounds that are also soluble in hot water, especially tannins. When tannins are extracted with hot water, some of these compounds are partially hydrolysed to form gallic acid. The chemical treatment of vegeTable fruits (tea, coffee, etc.) with a base such as sodium carbonate converts acidic tannins into the respective sodium salts which are highly soluble in water by its ionic character. Although caffeine is water-soluble, it is more soluble in chloroform and therefore it can be extracted with this organic solvent, while the sodium salts of gallic acid and tannins remain in the aqueous phase. Thus, the extraction of the basic solution with chloroform separates almost pure caffeine. Anhydrous sodium sulphate acts by removing the salt and all water-soluble salts that are still retained in the organic solvent (organic layer) or accidentally transferred by decantation. The organic solvent can be easily removed by evaporation or by distillation to obtain caffeine. Chloroform was used because of the high solubility of most organic compounds and its low boiling point ($62^{\circ}C$) (Aznar, 2011).

Figure 1 show the absorption spectrum of the standard 16 ppm caffeine solution in the wavelength range between 300 and 220 nm, where it was found that in the UV range at the wavelength of 273.5 nm, caffeine has a maximum absorption.



Figure 1. The absorption spectrum of the standard 16 ppm caffeine solution

Concentrations of caffeine from different coffee brands (P1 - P5) and black tea (P6) respectively are represented in percent, and the variation of each compared to the others is shown in Figure 2. The diagram shows differences in caffeine concentration from the 6 samples depending on the type of analysis sample (coffee with caffeine - P1, P2, P3, decaffeinated coffee - P4, P5 or tea - P6) and coffee (*Arabica* and *Robusta*).



Figure 2. The caffeine content of the analyzed samples

It can be seen that the highest caffeine content of the studied coffee samples has P1 (0.89% caffeine), followed by sample P2 (0.46% caffeine) and sample P3 (0.22% caffeine). In terms of decaffeinated coffee, the smallest quantity is found in the P5 sample (0.01% caffeine), while sample P4 has a higher caffeine content (0.21% caffeine). The P6 black tea sample contains 0.18% caffeine.

According to some reseachers, green or roasted natural coffee has a caffeine content of between 0.8% and 2.4%, depending on the species. Hečimović *et al.* (2011), as well as other researchers, concluded that Arabica contains little caffeine, with values between 0.7% and 1.5%. In Aznar's (2011) study of 32 coffee samples, the caffeine content was between 0.88% and 1.60%. Considering these studies, we can say that the P1 sample with 0.89% caffeine content falls within the upper limits of the literature, while the P3 sample containing 0.22% of caffeine is slightly below the limits. Some of these coffee types have different countries of origin so that the caffeine levels found may vary depending on the place of origin. Also, in the absence of more information on its characteristics, some of these commercial coffe varieties may be blends of *Robusta* and *Arabica* so that the first variety could increase caffeine concentration because it has a higher content. In support of the possibility of being a mixture comes the fact that P2 has a low caffeine content of 0.46% caffeine, when it was actually a *Robusta* coffee and should have had a higher caffeine content compared to the other analyzed samples. The literature suggests a caffeine content of between 2% and 3.5% in the *Robusta* coffee category (Bondesson, 2015).

The purpose of decoffining is to produce a coffee that keeps its taste and flavor, but has a low caffeine content. European legislation permits the labeling of coffee verv (www.healthline.com/search?q1=caffeine, Directive 77/436/EEC, Directive 79/112/EEC, Directive 85/573/EEC, HOTĂRÎRE 206/11.03.2009) as decaffeinated if its maximum caffeine content does not exceed 0.12%. From Figure 2, it can be seen that only sample P5 complies with the law, with a very low caffeine content of 0.1%. In contrast, sample P4 has a caffeine content of 0.21%. Although it is not a high caffeine content, it exceeds the admissible limits. Before making any further statements, it should be noted that this non-compliance may be a consequence of the extraction method at the decaffeination stage during which a very rigorous procedure is required for the remaining caffeine content to be extremely low. In the Aznar's study (2011) on two types of decaffeinated coffee, concentrations of 0.21% and 0.39% of caffeine were obtained. The interesting fact is that the P4 sample has almost the same caffeine content as sample P3, respectively 0.21%, compared to 0.22%, as shown in Figure 2.

In terms of black tea P6, it contains 0.18% caffeine content and it can be said that there are no big differences in caffeine content of normal coffee - P3 (0.22%), decaffeinated coffee - P4 (0.21%) and black tea - P6 (0.18%). Therefore, black tea (0.18% caffeine) is an important source of caffeine, and in some studies, quantities of caffeine in black tea are even higher in the range: 1.5% to 3.3% caffeine (Yashin *et al.*, 2015).

4. Conclusion

Coffee and tea are beverages of great cultural, social and economic importance throughout the world. These drinks, depending on the concentration in their active principle - caffeine, have numerous benefits for the human body, but also a number of harmful effects. In this study, a high caffeine content in the P1 sample (0.89%) was determined, followed by sample P2 (0.46%) and sample P3 (0.22%). In terms of decaffeinated coffee, the smallest quantity is found in the assortment of sample P5 (0.01%), while in sample P4 there was a high content (0.21%) for decaffeinated coffee.

With a caffeine content of 0.18%, black tea is a good substitute for coffee, taking into account that caffeine's found values are close to those found in caffeine coffee.

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