Determination of Trace Metal and Mineral Levels in the Tobacco and Cigarette samples using by FASS

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Summary: Cigarettes and tobacco products are consumed in large amounts by human beings in the world. Also, tobacco is one of the most important agricultural products in Turkey. The accumulation of heavy metals in tobacco leaves in accordance with a possible risk of transferring them to people by smoke is well known. This study was carried out to estimate trace metal and mineral levels in tobacco and cigarettes from Eastern and South-eastern Anatolia, Turkey. The analysis was done by flame atomic absorption spectrometry using dry ashing or wet digestion. Results obtained are in agreement with data reported in the literature.

Key Words: Tobacco, Cigarette, Flame Atomic Absorption Spectrometry, Trace Metals, Mineral.

Introduction

Tobacco is one of the most widely used commodities in the world. It has been studied extensively because of its scientific uniqueness, its economic importance in society, the health consequences of tobacco use, the economic and political importance of the industry it produced, because of its ability to generate massive revenues and due to governmental regulation [1].

A cigarette's design features influence smoke particulate mass transport through the tobacco rod and filter; thus cigarettes are characterized according to machine-smoked tar delivery categories described as full flavor, light, and ultralight [2]. Tobacco growing soils are characterized by high levels of available cadmium and lead, and thus levels of toxic metals have been found in the tobacco lamina [3-5] and in the smoke particulate [6]. The frequent intake of nicotine in the human body may cause a broad spectrum of diseases that extend towards many different organ systems. Thus, cigarette brands with similar tar deliveries could yield markedly different smoke particulate levels of heavy metals depending on filter ventilation and where the tobacco was grown. Tobacco-related disease originates from the biological consequences of repeated inhalation exposure to numerous toxic constituents in cigarette smoke, which are produced by pyrosynthesis or liberated during combustion. Tobacco smoke has toxic [7, 8], genotoxic [9], mutagenic [10] and carcinogenic properties [11] and has been linked to adverse pregnancy outcomes [2]. The accumulation of heavy metals in tobacco leaves in accordance with a possible risk of transferring them to people by smoke is well known. Its numerous deleterious health effects, combined with the substantial prevalence of cigarette smoking, make it a major worldwide cause of death [12].

Metal concentrations in tobacco have been found to be influenced by such factors as soil type, soil pH, plant genotype, stalk position, and soil and leaf residues resulting from the application of pesticides containing metals and from soil amendments, including fertilizers and municipal sludge [13, 14].

Flame atomic absorption spectrometry (FAAS) is the most widely used technique because for the analysis of trace metals in food samples [15, 16]. Furthermore, AAS is cheap and its usage is easier than other instruments.

To our knowledge, there are no literature reports on the content of trace metal and mineral levels in the tobacco and cigarette from the east and south-eastern Anatolia regions, Turkey. The aim of the current study was to investigate quantitatively trace metals (Fe, Zn, Mn, Cu, Ni, Cr, Co, Cd and Pb) and minerals (Ca, K and Mg) in the selected tobacco and cigarette samples from the east and south-eastern Anatolia regions (Turkey) using FAAS.

Results and Discussion

The human body requires both metallic and nonmetallic elements within certain permissible limits for growth and good health. Therefore, determination of element compositions in foods and related products is essential for understanding their nutritive importance [15]. In the present work, the concentrations of minerals (K, Mg, Ca) and trace metal (Fe, Zn, Cu, Mn, Ni, Cr) were determined in the tobacco and cigarette samples using dry digestion or wet digestion. The concentrations of the metals in

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the tobacco and cigarette samples are given in Table-1.

Matrix destruction based on oxidation with concentrated acids is the most widely used approach for biological samples because of its efficiency [18-21] in liberating the elements of interest from the molecular structures. Wet digestion methods favours organic matter destruction, shortens the time needed for the analysis and offers the advantage of simple, fast organic matter destruction, minimum reagent volume, reduction of possible analyte losses by volatilization or retention and elimination of the environmental contamination risks [22]. In general, dry ashing methods may present poorer accuracy and provide lower analyte recovery when compared with acid digestion methods [23, 24]. When the dry digestion procedure is compared with wet digestion procedure for testing mineral and metal concentrations, generally the standard deviation of the dry digestion procedure is higher than that of the wet digestion procedure (Table-1). In addition, the recovery of the metals in the dry digestion procedure (values in the range 85-115 %) is lower than that of the wet digestion procedure (values in the range 90-105 %). The recovery studies were carried out by adding the appropriate quantity of standard metal solutions to the previously determined metals content of the tobacco and cigarette samples. Furthermore dry digestion procedure is slow and time consuming. Therefore, the mineral and trace metal content of the tobacco and cigarette samples are given in the text, after the wet digestion procedure.

Potassium (K) is the major cation in intracellular fluid and plays a role in the regulation of osmotic pressure, blood pressure and acid–base balance [25]. Potassium ion (K^+) is necessary for the function of all living cells. K^+ diffusion is a key mechanism in nerve transmission, and K depletion in animals, including humans, results in various cardiac dysfunctions. The K contents in the tobacco and cigarette samples were found as 6689.74, 10097.22, 10086.37 and 21158.99 μg/g I, II, III and IV sites, respectively.

Magnesium (Mg) is an important element for plant, it is a component of chlorophyl and magnesium pectate, is essential for formation of carotenoids and required by a large number of enzymes connected with phosphate transfer [26]. The Mg contents in the tobacco and cigarette samples were found as 2358.86, 3583.23, 3033.28 and 3949.61 μg/g I, II, III and IV sites, respectively. Mg levels of Oriental Tobacco Leaves CTA-OTL-1 have been reported as the averaged 4519.6 μg/g [27].

Calcium (Ca) is essential for living organisms, particularly in cell physiology, where movement of the calcium ion (Ca^{2+}) into and out of the cytoplasm functions as a signal for many cellular processes. Ca is important in the development and maintenance of strong bones and teeth [28]. Ca in the plants is absorbed as Ca^{2+} ions. Ca is a component of calcium pectate, which is found in the middle lamella, acts as an activator of enzymes like ATP-ase, some kinases, phospholipases and succinate dehydrogenase and Counteracts toxicities of other metallic ions [16]. The Ca contents in the tobacco and cigarette samples were found as 1389.01, 2888.60, 3429.96 and 3279.72 μg/g I, II, III and IV sites, respectively. Ca levels of Oriental Tobacco Leaves CTA-OTL-1 have been reported as the averaged 32011.4 μg/g [27].

The appropriate content of iron in plants is essential both for the health of the plant and for the nutrient supply to man and animals. The variation among plants in their ability to absorb Fe is not always consistent and is affected by changing conditions of soil and climate and by the stages of plant growth. Generally, certain forbs, including legumes, are known to accumulate more Fe than are grasses. However, where Fe is easily soluble, plants may take up a very large amount of Fe [29]. Fe content of normal plant tissue varies according to species but, it is usually within the range 20-200 μg/g dry matter [30]. The Fe contents in the tobacco and cigarette samples were found as 212.54, 168.19, 216.42 and 127.18 μg/g I, II, III and IV sites, respectively. Fe levels of Oriental Tobacco Leaves CTA-OTL-1 have been reported as the averaged 1083.5 μg/g [27]. Fe content of tobacco leaves from Greece has been reported as 215.5 μg/g [14].

Table-1: Metal contents (μg/g) in tobacco and cigarette samples that determined after wet and dry ashing (mean ± SD), n = 3.

Sample*	Method**	K	Ca	Mg	Fe	Cu	Zn	Mn	Cr	Ni
I	a	6689.74±73.59	1389.01±4.17	2358.86±4.72	212.54±1.06	4.95±0.03	5.96±0.07	16.51±0.13	1.26±0.04	3.23±0.09
	b	6801.09±34.01	10190.91±10.19	3437.79±13.75	318.06±7.32	5.25±0.03	7.56±0.03	20.15±0.10	1.28±0.03	4.87±0.12
II	a	10097.22±100.09	2888.60±5.78	3583.23±7.17	168.19±0.51	7.61±0.08	10.38±0.19	19.42±0.12	1.23±0.02	3.44±0.01
	b	10207.82±30.62	10842.61±21.68	4128.12±8.26	354.56±1.06	8.57±0.01	13.84±0.04	22.14±0.18	1.35±0.05	4.88±0.08
III	a	10086.37±50.43	3429.96±13.72	3033.28±6.07	216.42±5.19	7.23±0.17	8.99±0.06	18.36±0.11	1.46±0.01	2.79±0.03
	b	10874.92±108.75	10674.19±42.69	4124.34±8.25	393.11±7.08	5.56±0.03	12.29±0.02	24.68±0.09	1.62±0.07	4.99±0.02
IV	a	21158.99±232.75	3279.72±13.12	3949.61±7.89	127.18±0.38	6.53±0.04	13.74±0.07	54.48±0.05	1.09±0.02	5.89±0.02
	b	16665.51±99.99	9218.78±73.75	3848.86±11.55	198.68±0.41	8.71±0.05	17.86±0.02	65.00±0.26	1.10±0.06	6.96±0.06

*I, Adıyaman; II, Bitlis; III, Diyarbakır; IV; Cigarette, **a, Wet ash; b, dry ash.

Zinc plays essential metabolic roles in the plant. The basic Zn functions in plants are related to metabolism of carbohydrates, proteins, and phosphate and also to auxins, RNA, and ribosome formations. Zn is believed to stimulate the resistance of plants to dry and hot weather and also to bacterial and fungal diseases [16]. Zn deficiency results in a wide spectrum of clinical effects depending on age, stage of development, and deficiencies of related metals. At the other extreme, excessive exposure to zinc is relatively uncommon and occurs only at very high levels [31]. The zinc content of normal plant tissues varies according to species, but, it is usually within the range 5-300 μg/g dry matter [30]. The Zn contents in the tobacco and cigarette samples were found as 5.96, 10.38, 8.99 and 13.74 μg/g I, II, III and IV sites, respectively. Zn levels of Oriental Tobacco Leaves CTA-OTL-1 have been reported as 53.5 μg/g [27]. Zn content of tobacco leaves from Greece has been reported as 23.5 μg/g [14]. Zn concentration of different brands of cigarettes in Jordan has been reported as averaged 55.62 μg/g [32]. Zn content of brands of tobacco cigarette in Nigerian has been reported as the averaged 32.56 μg/g [17].

Copper (Cu) is one of the essential micronutrients and its adequate supply for growing plants should be ensured through artificial or organic fertilnrs [33]. It may enter the food materials from soil through mineralization by crops, food processing or environmental contamination. The essential role of copper in maintaining normal health in both animals and humans has been recognized for many years [31]. Cu content of normal plant tissues varies according to species but is usually within the range 1-25 μg/g dry matter [30]. The Cu contents in the tobacco and cigarette samples were found as 4.95, 7.61, 7.23 and 6.53 μg/g I, II, III and IV sites, respectively. Cu levels of Oriental Tobacco Leaves CTA-OTL-1 have been reported as averaged 13.2 μg/g [27]. Cu content of tobacco leaves from Greece has been reported as 55 μg/g [14]. Cu concentration of different brands of cigarettes in Jordan has been reported as the averaged 12.90 μg/g [32]. Cu content of brands of tobacco cigarette in Nigerian has been reported as averaged 12.25 μg/g [17].

All plants have a specific requirement for Manganese and apparently the most important Mn function is related to the oxidation-reduction processs [16]. It is necessary for connective tissues and bones, for general metabolism and reproductive functions [34]. The Institute of Medicine recommends that intake of Mn from food; water and dietary supplements should not exceed the tolerable daily upper limit of 11 mg/day [35]. It is toxic to plants at concentration range of 1-99mg/L depending on the plant species [30]. The Mn contents in the tobacco and cigarette samples were found as 16.51, 19.42, 18.36 and 54.48 μg/g I, II, III and IV sites, respectively. Mn levels of Oriental Tobacco Leaves CTA-OTL-1 have been reported as averaged 412 μg/g [27]. Mn content of tobacco leaves from Greece has been reported as 250 μg/g [14]. Mn content of brands of tobacco cigarette in Nigerian has been reported as the averaged 128.46 μg/g [17].

Nickel (Ni) is probably an essential element for humans, with several possible roles in maintenance and production of body cells. Ni may be beneficial as an activator of some enzyme systems [36], but its toxicity at higher levels is more prominent. The most common adverse health effect of Ni in humans is an allergic reaction. Apart from the environmental contamination sources of Ni in foods, it may also be the possibility of its arising from the production or storage of foods such as drying, cooking and packaging in materials [37]. The Ni in the tobacco and cigarette samples were found as 3.23, 3.44, 2.79 and 5.89 μg/g I, II, III and IV sites, respectively. Ni levels of Oriental Tobacco Leaves CTA-OTL-1 have been reported as averaged 6.32 μg/g [27]. Ni content of tobacco leaves from Greece has been reported as 47.5 μg/g [14]. Ni content of brands of tobacco cigarette in Nigerian has been reported as the averaged 16.92 μg/g [17].

Chromium (Cr) is one of the trace metal nutrients that are essential to humans and animals, and it found widely in the environment. Good food sources of Cr include meat, whole grains, lentils, and species. Having 10 mg or less a day of Cr from food and supplements is unlikely to cause any harm [31]. The levels of chromium in plants are less than 1 or 2μg/g dry matter even when growth is reduced by toxicity [38]. The Cr contents in the tobacco and cigarette samples were found as 1.26, 1.23, 1.46 and 1.09 μg/g I, II, III and IV sites, respectively. Cr levels of Oriental Tobacco Leaves CTA-OTL-1 have been reported as the averaged 2.59 μg/g [27]. In the present study, the heavy metals Co, Cd and Pb were not detectable.

Experimental

Reagents

A standard solution of each element was prepared immediately by dilutions of a 1000 mg/L stock solution (Merck, Darmstadt, Germany) prior to use. Milli-Q deionized water (Millipore) was used

throughout this experiment. All solvents and reagents such as H_2SO_4, HNO_3, HCl, and H_2O_2 were of analytical reagent (AnalaR) grade (Merck). All the plastic and glassware containers used were washed with detergent solution followed by 10% (v/v) HNO_3 and then rinsed with tap water and finally with deionized water and dried before use.

Apparatus

Mineral and trace elements were determined using a Thermo Solar System atomic absorption spectrometer (UK), equipped with THERMO hollow cathode lamps, was used for the sample analysis. The measurements were carried out in an air/acetylene flame. The operating conditions adjusted in the spectrometer were carried out according to the standard guidelines of the manufacturers. The quantitative determinations of elements in samples were done using calibration curves obtained from diluted stock standard elements 1000 mg/L. The concentrations of analytes were obtained directly from calibration graphs after correction of the absorbance for the signal from an appropriate reagent blank. Determinations were carried out in triplicate.

Sample Preparation

The tobacco samples were collected in south-eastern (Adıyaman and Diyarbakır) and east-eastern (Bitlis) regions. Cigarette samples were purchased from the local supermarkets (Van). All samples were oven-dried at 90 ^{0}C for 24 h before grinding. They were ground with a porcelain mortar and sieved (200 mesh). Two types of digestion procedures were applied to the tobacco and cigarette samples; dry digestion (in a muffle furnace) and wet digestion (with H_2SO_4-HNO_3-H_2O_2). The precision was calculated on three replicates for all digestion procedures. Optimum digestion conditions are given below. One gram of sample was placed into a high form porcelain crucible for dry digestion. The furnace temperature was slowly increased from room temperature to 450 °C in 1 h. The sample was ashed for about 4 h until a grey ash residue was obtained. The residue was dissolved in 4 mL 3 N HCl, then the digested solutions were filtered through filter paper (Whatman no. 42) and diluted to 25 mL with deionized water. Wet digestion procedure; dried powders of tobacco and cigarette samples, 1 g, were weighed and were transferred into a glass beaker. Eight milliliters of concentrated H_2SO_4 and 16.0 mL of concentrated HNO_3 were added into the beaker. The mixture was then heated for about 10 min and after it was cooled 16.0 mL H_2O_2 was added dropwise and further heated for 10 min. The digested mixture was allowed to cool then the digested solutions were filtered through filter paper (Whatman no. 42) and diluted to 50 mL with deionized water [17]. Blank digestions were also carried out in the same way.

Conclusion

In this study, we conclude that measuring minerals and metal concentrations in tobacco and cigarette using FAAS. Our results showed that these samples contained high concentrations of K, Ca and Mg which indicated that these three elements were the most abundant elements in many plants. Furthermore, According to Wet ash method, K, Zn, Mn and Ni elements were mostly found in cigarettes; Ca, Mg, Fe, and Cr elements were found in Diyarbakir samples and Cu element was found in Bitlis samples. On the other hand according to dry ash method, K, Cu, Zn, Mn, Cr, and Ni elements were mostly found in cigarettes; Ca and Mg elements in Bitlis and Fe element was found in Diyarbakir samples. The FAAS method has the following advantages: simple, rapid and low analysis cost.

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