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## **HEAVY METAL CONTENT IN VIRGINIA AND BURLEY TOBACCO**

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## **INTRODUCTION**

The high heavy metal content in tobacco has negative effect on the biological processes in the human body. Chronic undesired effects can be expressed after years of exposure, as a result of a long-lasting intake of these elements, some of which are powerful carcinogens. The concentration of cadmium in blood is higher in smokers, while a relation between that concentration and the coronary diseases is described (Özcelik et al., 2000; Abu-Hayyeh et al., 2001). Chrome content in the lungs of smokers is more than 3 times higher than the normal. Nickel forms a toxic carbonil compound that is considered a potential carcinogen. Iron, manganese, copper and zinc are nutritive elements, which role for the plant and human organism is indisputable, but in high concentrations they are toxic as well.

The content of heavy metals in tobacco leaves is variable and depends on the growing conditions, mainly on the soil composition and properties. One of the main factors influencing the concentration of heavy metals in tobacco leaves is the soil pH (Xian and Shokonifard, 1989; King and Hajjar, 1990, Bell et al., 1992, Khan et al., 1992). Agho-Adamu (1987) and Gondola and Kadar (1993) established a negative correlation between the soil pH and the heavy metal content in tobacco. Other soil characteristics influencing the concentration of heavy metals in tobacco plant are the mechanical composition and the humus content (Adamu et al., 1989, King, 1989). Adamu et al. (1989) identified statistically significant correlation between the total Zn, Mn and Fe content in the soil and in tobacco leaves. The data of Angelova et al. (2004) demonstrated that the correlation between the total concentration of lead, cadmium and zinc in the soil and their accumulation in tobacco can be described by a linear equation for Zn and a polynomial one for Pb and Cd.

Previous studies of Bozhinova et al. (1994) identify tobacco as a crop with intensive accumulation of heavy metals. Physiological absorption in plant tissues is accompanied by accumulation on the leaf surface from the air. The content of heavy metals in tobacco leaves depends not only on the properties and composition of the soil, but on the type and variety of tobacco grown. Tso (1972) reports great differences in heavy metals content in Virginia and Burley tobacco. Tsotsolis et al. (2001) determined that the concentration of Zn and Mn varies significantly in different varieties of oriental tobacco, while no significant differences were identified for Pb and Cd.

The aim of the present study was to determine the heavy metal content in Virginia and Burley tobacco, grown under the same environmental conditions.

# **MATERIALS AND METHODS**

The present study was carried out on a gleyic chromic luvisols in Plovdiv region (Table 1). This soil is light sandy clay with an average humus content and neutral pH. Samples were

taken from 0-30 cm soil layer. Total amounts of iron, manganese, zinc, cadmium, cobalt, nickel and chrome in the soil were determined after degradation with aqua regia.

pН•	Humus	Partical size distribution, mm							
H <sub>2</sub> O	%	Големина на честичките							
		eoarse sand Kpytten necok	Fine sand Curren necos	Silt Прав	Glay Глина	Silt+ glay Прав+глина			
		2-0.2	0.2-0.02	0.02-0.002	<0.002	<0.02			
6,88	1.617	40.12	30.38	14.33	15.17	29.50			

Table 1 Some characteristics of the soil Табела 1 Некои карактеристики на почвата

Five Virginia varieties (K 326, NC 55, V 0454, PVH 19 and K 394) and 3 Burley varieties (Burley 1000, Burley 1317 and Burley 21) were tested. Leaves in full maturity from the middle stalk position were harvested for analyses. Sample preparation was done by dry burning and dilution in 3M HCL.

Atomic absorption spectrometer "Spektra AA 200" (Varian, Australia) was used for determination of the heavy metal content in the soil and plant samples at following wavelengths: Fe - 248.3 nm, Mn - 279.5 nm, Cu - 324.8 nm, Zn -213.9 nm, Pb - 217.0 nm, Cd - 228.8 nm, Co - 240.7 nm, Ni - 232.0 nm, Cr - 357.9 nm..

# **RESULTS AND DISCUSSION**

Iron (Fe). Tobacco is a crop that is particularly sensitive to a high Fe content. Toxicity symptoms become apparent at plant concentration above 1000 mg/kg and can be traced to halted root growth, leaf damage with necrotic spots on them (Kabata Pendias and Pendias, 1984)

The total Fe content in tested soil samples was 1.4% (Table 2). The concentration of this element in Virginia tobaccos varied between 94 and 155 mg/kg (Table 3), while in Burley it was between 90 and 232 mg/kg (Table 4). Observed values are lower than those in other studies. Tso (1972) recorded concentrations in Virginia tobaccos between 132 and 595 mg/kg, while in Burley concentrations were between 200 and 650 mg/ kg. Radoji?i? et al. (2003) observed between 170.72 and 995.87 mg/kg in Virginia tobacco. Probably the lower Fe concentrations observed in the present study are due to a lower soil content of this metal. Furthermore, when soil pH is above 6 the mobility of iron is reported to decrease. Availability of this element to the plants is highest at pH between 4 and 6.

Manganese (Mn). The total Mn content in the soil was 319 mg/kg (Table 1). This content is much lower than the average for the country, which is 1200 mg/kg (Brashnarova, 1981; Atlas of the soils in Bulgaria, 1998) and from around the world - about 545 mg/kg (Brashnarova, 1981, Koinov et al., 1998). Low concentrations were observed in the varieties as well. When compared, the two types of tobacco differ, with Burley accumulating more Mn - 61-92 mg/kg (Table 3 and 4). These values are close to the ones observed by Gondola and Kadar (1993) in soils with neutral or slightly alkaline reaction and significantly lower than the ones observed by other authors - 140-700 mg/kg (Tso 1978; Agro-Adamu, 1988, Bell et al., 1992). The reason for this is probably not only the lower soil content, but the neutral soil reaction as well, which accounts for the lower mobility of this element. The concentration of Mn significantly increases at acid soil reaction and could reach as high as 2400 mg/kg (Tso, 1972; Bell et al., 1992). The most typical expression of the toxic effect of this element is chlorosis. Typical dark necrotic spots appear on the leaves, with uneven chlorophyll distribution in older leaves, drying out of their edges and root withering (Kabata Pendias and Pendias, 1984).

Copper (Cu). The total copper content in the soil was 110 mg/kg and is quite higher than the average for the country (30±25 mg/kg) and for the rest of the world (20 mg/kg), but is within the acceptable levels at the respective pH (Chuljian, 1989). The concentration of copper in most of the Virginia varieties was between 16.9 and 19 mg/kg, but in V 0454 it reached 25.8 mg/ kg (Table 3). In Burley varieties it was between 20.5 and 28.7 mg/kg (Table 4). The overall copper content in all tested varieties is higher than

the one recorded by other authors (Adamu et al., 1989, Bell et al., 1992, Fischer, 1992). Our data is insufficient to positively identify the reason for these high concentrations - is it the high total copper content in the soil or it is due to specific variety responses, related to a higher accumulation of this element. However the values we observed are close to the ones reported by others. Collins et al. (1961) reported Cu plant concentration in Virginia tobaccos of 14.9-21.1 mg/kg. **Radoji~i**} et al. (2003) recorded concentrations between 16.42 and 31.45 mg/kg. According to Kabata Pendias and Pendias (1984), the normal plant concentration of copper is within 5-30 mg/ kg limits. As a lower toxicity threshold for most crops they reported 20 mg/kg. The major symptoms of the copper toxicity are dark green leaves, thick, short roots without hairs. No one of the varieties in our study showed such toxicity symptoms, which presumably means the toxicity level for these varieties was not reached.

Zinc (Zn). The total Zn content in the soil was 84 mg/kg. This is the average content for this element in Bulgarian soils (Brashnarova, 1981; Koinov et al., 1998). The zinc concentration in Virginia tobacco was between 49.2 and 82.7 mg/kg where the lowest values were measured in K 326 and the highest - in V 0454 variety respectively (Table 3). Burley tobaccos varied in their Zn content as well from 58.5 (Burley 1000) to 83.8 (Burley 21) The values observed for all varieties are within those obtained by other authors (Oso, 1972, Fischer, 1992, Radojicic et al., 2003;) and are lower than the critical for the plants 100-400 mg/kg (Kabata Pendias and Pendias, 1984). Zinc toxicity symptoms usually are manifested as chlorosis and necrosis at the leaf edges, between vein chlorosis in young leaves, growth retardation and root damage.

Lead (Pb). The toxic effects of lead resemble strongly the "frenching" symptoms (David et al., 1955) and are expressed as darkening of the leaves, sunting of older leaves, browning and shortening of the roots. Favourable phosphorus supply reduces Pb toxicity as a result of formation of insoluble phosphates in the soil and plant tissues (Kabata Pendias and Pendias, 1984).

The total lead content in the soil was 26 mg/kg (Table 2) and is within the maximum allowable content (MAC) for the neutral soil reaction (Chuldgian, 1989). The concentration of Pb in Virginia tobaccos was quite even - between 3 and 4.1 mg/kg (Table 3). In Burley varieties varying was somewhat higher - between 1.5 and 3.8 mg/kg (Table 4). Observed values are very similar to those measured by other authors (Òso, 1972; Agro-Adamu, 1988). The natural levels in

the plants from non-contaminated sites vary between 0.1 and 10 mg/kg and the concentrations considered as critical (according to Kabata Pendias and Pendias, 1984) are 30-300 mg/kg.

Cadmium (Cd). The total Cd content in the soil was 0.6 mg/kg and is within the MAC limits for this element under the respective soil reaction (Chuldgian, 1989). Observed concentration in Virginia tobaccos varies insignificantly between 0.7 and 1.0 mg/kg. The highes values observed in our investigation were found in Burley 21 variety - 2.3 mg/kg (Table 4). All these values correspond to the data of other authors (Agro-Adamu, 1988, Bell et al., 1992) and are lower than the ones considered as critical in the plant - 5-30 mg/kg (Kabata Pendias and Pendias, 1984). The apparent symptoms of increased Cd content in plants are retarded plant growth, damaged root system, leaf chlorosis, and dark redbrown coloration of leaf edges.

Cobalt (Co). The total Co content in the soil was 9.6 mg/kg (Table 2). According to Kabata Pendias and Pendias (1984) the normal concentration of Co in the soils around the world is within 1-40 mg/kg limits. For the soils in Bulgaria the observed values are 3-33 mg/kg (Enikov and Benevski, 1984, Koinov et al., 1998). In the varieties tested the concentration of this element was between 1 and 2.7 mg/kg. The lowest content was found in the K 326 (Virginia type) variety and the highest - in Burley 1317 and Burley 21 (Tables 3 and 4). According to data by Voss and Nicol (1960) the concentration of Co in Virginia tobaccos is 0.9-1.54 mg/kg. Kabata-Pendias and Pendias (1984) consider the critical concentrations of this element in the plant to be in the range of 15-50 mg/kg. Studies of Nicholas and Thomas (1953) point that the symptoms of Co toxicity in tobacco are very similar to these of iron deficiency and are expressed as chlorosis between the veins in young leaves, white leaf edges, root damage.

Nickel (Ni). The total nickel content in the soil was 48 mg/kg while the average content for Bulgarian soils is 35 mg/kg and around the world - 40 mg/kg (Brashnarova, 1981, Koinov et al., 1998, Kabata-Pendias and Pendias, 1984). Virginia tobaccos had lower Ni content (0.7 mg/ kg for K 326 and 0.9 mg/kg for PVH 19) (Table 3). Altogether Burley tobaccos accumulate more nickel - 4.8-7.8 mg/kg (Table 4). Observed values are lower than the critical ones for the plant - 10-100 mg/kg (Kabata-Pendias and Pendias, 1984) or 50 mg/kg (Alloway and Ayres, 1994). Young tobacco plants are particularly sensitive to nickel toxicity (Tso, 1972). The symptoms of poisoning are manifested as intervein chlorosis in young leaves, grayish-green leaves and dark wilting roots. These symptoms can be significantly reduced by the application of phosphate fertilizers, which form insoluble nickel phosphates (Brummer et al., 1986).

Chrome (Cr). The total Cr content in the soil was 93 mg/kg (Table 2), while the average for Bulgaria is 60-75 mg/kg (Brashnarova, 1981). Observed leaf concentrations in Virginia tobac-

cos were between 2.6 mg/kg (in PVH 19) and 4.2 mg/kg (in NC 55). The highest Cr concentration was observed in Burley 21 variety where it reached 5.1 mg/kg. Phytotoxic levels for CR in tobacco are from 18 to 24 mg/kg (Gough et al., 1979). The toxicity symptoms are wilting of the shoot and root damage. Quite typical is the chlorosis on the young leaves (Kabata-Pendias and Pendias, 1984).

Table 2. Total heavy metal content in the soilТабела 2Вкупна содржина на тешки метали во почвата

Element Елемент	Fe	Mn	Cu	Zn	Рb	Cd	Co	Ni	Cr
mg/kg	14000	319	110	84	26	0,60	9,6	48	93

Table 3. Heavy metal content in the leaves of Virginia tobaccos. Табела 3. Содржина на тешки метали во листовите на вирџиниските тутуни

Variety Copta	Fe mg/kg	Mn mg/kg	Cu mg/kg	Zn mg/kg	Pb mg/kg	Cd mg/kg	Co mg/kg	Ni mg/kg	Cr mg/kg
K326	102	29	16,9	49.2	4.0	0.7	1.0	0,7	2.9
NC55	155	39	18.4	54.1	3.2	0.7	2.1	4.7	4.2
V0454	94	57	25.8	82.7	4.1	0.9	1.4	3.5	3,8
PVH19	107	47	16,9	53.3	3.3	1.0	1.1	0,9	2.6
K394	133	47	19,0	72.6	3,0	0,8	1.4	2.9	4.2

Table 4. Heavy metal content in the leaves of Burley tobaccos. Табела 4. Содржина на тешки метали во листовите на берлејските тутуни

Variety Copta	Fe mg/kg	Mn mg/kg	Cu mg/kg	Zn mg/kg	Pb mg/kg	Cd mg/kg	Co mg/kg	Ni mg/kg	Cr mg/kg
B1317	90	92	28.7	67,3	3,8	0,6	2.7	6.1	4.5
B1000	232	71	20.5	58,5	1.5	0,6	1.7	7,8	3.7
B21	133	61	26.8	83.8	3.4	2.3	2.3	4.8	5.1

# CONCLUSION

The concentrations of heavy metals in all studied genotypes are in accordance with the data from other sources and are lower than the critical ones for the plants.

Altogether, Burley varieties accumulate higher concentrations of manganese, copper, nickel and to lower extent - zinc. No significant differences with Virginia tobaccos were observed in the accumulation of other heavy metals.

The lowest heavy metal content was observed in the Virginia tobacco variety K 326. More detailed studies are needed on the uptake and accumulation of heavy metals in tobacco as related to the environmental conditions and for selecting genotypes with decreased absorption. The application of best agricultural practices for blocking the toxic effects of heavy metals is essential for preventing accumulation of undesirable concentrations in the cigarettes and the smoke, which could be dengerous to the human health.

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# СОДРЖИНАТА НА ТЕШКИ МЕТАЛИ КАЈ ТУТУНИТЕ ВИРЏИНИЈА И БЕРЛЕЈ

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## РЕЗИМЕ

Поставен е полски експеимент на глејна хромна лувисол почва за проучување на содржината на тешки метали кај тутунот тип вирџинија (К 326, NC 55, V 0454, PVH 19 и К 394) и берлеј (В1317, В 100 и В 21). Извршена е анализа на листовите од средниот појас во техничка зрелост. Одредувањето на елементите железо, бакар, цинк, кадмиум, кобалт, никел и хром во почвени и растителни проби е изврш ено со атомски апсорпционен спектрометар. Концентрацијата на тешките метали во сите испитувани генотипови кореспондира со податоците од другите автори и е пониска од онаа што се смета како критична за растенијата. Берлејските сорти акумулираат повеќе манган, бакар, никел и цинк. Кај вирџиниските тутуни не се забележани сигнификантни разлики за другите елементи. Најниска содржина на тешки метали имаше кај вирџиниската сорта К 326.

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