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Heavy metal contamination of paddy soils and rice (*Oryza sativa* L.) from Kočani Field (Macedonia)

Nastja Rogan · Todor Serafimovski · Matej Dolenec · Goran Tasev · Tadej Dolenec

Abstract This research focuses on the heavy metal contamination of the paddy soils and rice from Kočani Field (eastern Macedonia) resulting from irrigation by riverine water impacted by past and present base-metal mining activities and acid mine drainage. Very high concentrations of As, Cd, Cu, Pb and Zn were found in the paddy soils (47.6, 6.4, 99, 983 and 1,245 $\mu g\ g^{-1}$) and the rice (0.53, 0.31, 5.8, 0.5 and 67 $\mu g\ g^{-1}$) in the western part of Kočani Field, close to the Zletovska River, which drains the mining facilities of the Pb–Zn mine in Zletovo. In terms of health risk, the observed highest concentrations of these elements in the rice could have an effect on human health and should be the subject of further investigations.

Keywords Heavy metals · Paddy soil · Rice (*Oryza sativa* L.) · Contamination · Mining activities · Kočani Field · Macedonia

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Introduction

Base-metal mining together with milling and grinding operations, concentrating ore and the disposal of tailings along with the acid mine and mill wastewater are some of the most important sources of heavy metals in the environment (Adriano 1986). Therefore, elevated concentrations of heavy metals can be found in and around abandoned and active mines due to the discharge and dispersion of mine waste materials, including tailings and acid mine drainage effluents into nearby agricultural soils, food crops, riverine water and stream sediments (Jung 2001; Korre et al. 2002; Lee et al. 2001; Liu et al. 2005; Lu and Zhang 2005; Simmons et al. 2005; Witte et al. 2004; Wong et al. 2002; Yang et al. 2004). It is well known that heavy metals in agricultural soils can influence both crop yield and quality, and soil containing various elements is a primary nutrient medium for plants. Food crops grown in contaminated areas may accumulate much higher than normal amounts of heavy metals, which may pose a significant public health risk to the local community.

Although numerous studies have been undertaken to investigate the heavy metal concentrations in soil and edible plants derived from the mining activities around the world, in Macedonia, such studies are very scarce (Dolenec et al. 2007). As a result, very little is known about the distribution and concentrations of heavy metals in the soil and food crops from different parts of Macedonia, which could be affected due to

historical and/or recent base-metal mining and milling operations as well as from irrigation with acid mine drainage and other mining effluents that have an impact on riverine water.

Such an example is Kočani Field in eastern Macedonia, where the base-metal mining history of the region is very long and paddy rice (*Oryza sativa* L.) is one of the main agricultural products of the region. Previous investigations have shown that the riverine water from the more polluted Zletovska River and the less polluted Bregalnica River, used for irrigation of the Kočani paddy fields, were contaminated with heavy metals as a result of mining activities and acid mine drainage from the Zletovo-Kratovo and Sasa-Toranica ore districts (Serafimovski et al. 2005). Therefore, it is very likely that the paddy soil and rice grown in this area display raised levels of heavy metals. Repeated water sampling of the Zletovska River has suggested marked chemical variations of As (1.7–25 $\mu g\ l^{-1}$), Cd (0.5–5 $\mu g\ l^{-1}$), Cu (6–10 $\mu g\ l^{-1}$), Pb (10–24 $\mu g\ l^{-1}$) and Zn (101–1,250 $\mu g\ l^{-1}$), which can be explained by the water-flow conditions and the varying degrees of mine wastes from the Pb–Zn Zletovo mine and the tailing dam leaching effluent inputs into the riverine water. The concentrations (in $\mu g\ l^{-1}$) of the aforementioned

heavy metals in the less polluted Bregalnica River, also used for the irrigation of agricultural soils, are as follows: As (0.53), Cd (0.39), Cu (3), Pb (2.4) and Zn (67) (Serafimovski et al. 2005).

Therefore, the major aim of the present study was to compare the results with permissible levels of heavy metals (As, Cd, Cu, Pb and Zn) in arable soils and crops and to reveal the influence of mining industry on the environment in Macedonia.

Study area

Kočani Field, well known for its paddy fields and thermal waters, is located in the eastern part of Macedonia, about 115 km from the capital city, Skopje. It is situated in the valley of the Bregalnica River, between the Osogovo Mountains in the north, and the Plačkovica Mountains in the south (Fig. 1). Its average length is 35 km and its width is about 5 km. The paddy soil of Kočani Field was estimated to originate from the composite material of the sediment derived from igneous, metamorphic and sedimentary rocks transported by the Bregalnica River and its tributaries and deposited in the Kočani Depression (Dolenec et al. 2007).

Fig. 1 Map of the study area showing the drainage system of the Bregalnica River and its tributaries

The broader region has a long history of mining dating to the pre-Middle Ages, with the most recent phase of mining starting after World War II. There are several Pb–Zn ore deposits and Ag, As, Cu, Sb, Ba, Au and U mineralization related to the Tertiary acidic to intermediate volcanogeno-intrusive complexes of the Besna Kobila-Osogovo Tassos metalogenic zone in the east and the Tertiary volcanogenic complexes of the Lece-Chalkidiki metalogenic zone in the north of Kočani Field (Serafimovski and Aleksandrov 1995).

The Zletovo lead–zinc deposit is situated along the active continental margin and is also intimately associated with the Tertiary volcanism and hydrothermal activity related with it. The Zletovo Mine is located in the eastern part of the Kratovo-Zletovo volcanic complex. From the geographical point of view the Zletovo Mine is located 5 km NW of the village of Zletovo and about 7 km from the city of Probistip.

The first detailed exploration started in 1928 by the English Company Selection Mines Limited and was finished in 1939 when the Zletovo deposit was prepared for exploitation with an annual capacity of 120,000 t raw ore. Nevertheless, continuous exploitation in the Zletovo Mine have started after World War II with an annual capacity of 400,000 t. Zletovo Mine is currently active with production of lead–zinc concentrate.

Mineral association comprises galena (the principal ore mineral) and sphalerite, with subordinate pyrite and lesser amounts of siderite, chalcopyrite, and occasional pyrrhotine, marcasite and magnetite. Minor occurrences of U-mineralisation have also been discovered (pitchblende).

The mining activities, the abandoned old mine sites and bare tailing, the large amounts of untreated waste material as well as the effluents from the Pb–Zn ore deposits Zletovo and Sasa have induced widespread and high heavy metal loads across the entire region. The acid mine water and the effluents from the ore processing facilities and the tailings, which were discharged untreated into the Zletovska and Bregalnica Rivers used for the irrigation of paddy soil, represented a further pollution source that could seriously affect the soil as well as the food and feed crops of Kočani Field.

The Kočanska and Orizarska Rivers, two small tributaries of the Bregalnica River, were also used downstream for irrigation drainage of the Pb–Zn mineralization of the southern part of the Osogovo Mountains, as well as the more or less untreated municipal wastes and domestic sewage of the cities of Kočani and Orizari. Therefore, the surrounding paddy fields could also be affected by the continuing usage of this riverine water for irrigation all year around.

Materials and methods

Sampling of the soil and the rice was carried out in autumn 2005 in order to investigate the distribution and concentrations of potentially toxic heavy metals, including As, Cd, Cu, Pb and Zn, related to the base-metal mining activities of the region. The sampling locations of the study area are shown in Fig. 2.

Soil sampling and analysis

Soil was collected from 38 locations using seven profiles (sections I–VII) across the Kočani paddy fields (Fig. 2). Near-surface soils were collected, because in the agricultural soil it is not possible to distinguish the A, B and C horizons. Each soil sample was made up of a composite of five subsamples taken from within a 1 × 1 m square. After air-drying at 25°C for a week, the soil samples were disaggregated, sieved to 2 mm and then ground in a mechanical agate grinder to a fine powder (<63 microns) for subsequent geochemical analysis.

All the paddy-soil samples were analysed for their As, Cd, Cu, Pb and Zn concentrations in a certified commercial Canadian laboratory (Acme Analytical Laboratories, Ltd.) after extraction for 1 h with 2-2-2-HCl–HNO$_3$–H$_2$O at 95°C by inductively coupled plasma mass spectrometry (ICP-MS). The accuracy and precision of the soil analyses were assessed by using international reference material such as Canadian certifite reference material project (CCRMRP) SO-1 (soil) and United States geological survey (USGS) G-1 (granite). The analytical precision and the accuracy were better than ±5% for the analysed elements. This was indicated by the results of duplicate measurements on ten soil samples as well as duplicate measurements on the G-1 and SO-1 standards.

Rice sampling and analysis

Rice-grain samples with a hull were collected during harvest time at the beginning of November 2005. The hulled rice grains were sampled at 14 sampling sites at the same locations as the corresponding soil samples

Fig. 2 Sampling location map of the study area: R I–VII soil samples unpolished rice samples
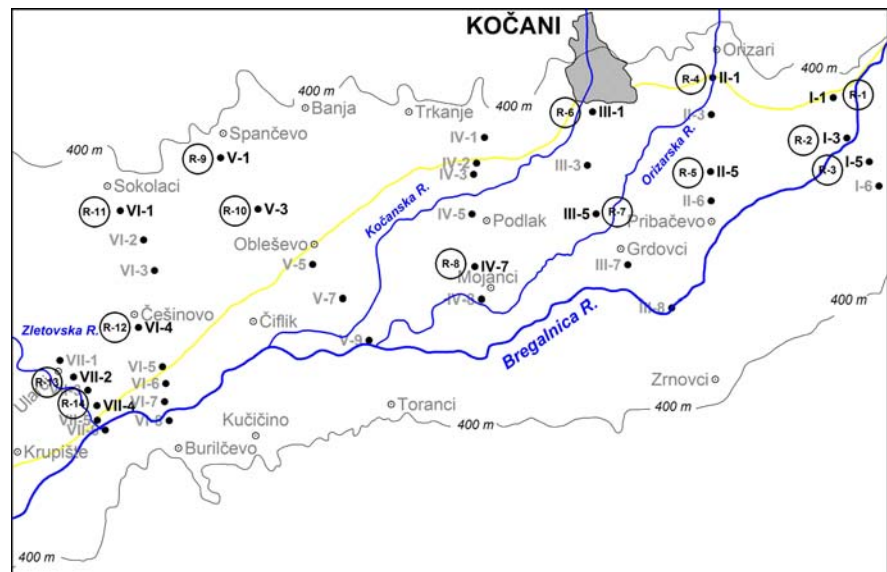
(Fig. 2). At each sampling site the rice grains were taken over an area of 10×10 m to obtain a final composite sample of about 1 kg. All the samples were collected and stored in polythene bags and brought to the laboratory for further preparation and treatment. In the laboratory the rice grains were washed three times with distilled water to eliminate soil particles and airborne pollutants, and were then oven-dried to constant weight at 75°C for 72 h. Each oven-dried sample of rice with a husk was dehulled with a ceramic pestle and mortar. The dehulled rice was ground in an agate mortar until it could pass through a 63-micron-mesh sieve. Powdered samples were then stored in clean dry glass containers before analysis.

The rice samples were measured for their As, Cd, Pb and Zn content after acid digestion in environmental-grade nitric acid and then analysed by IPC-MS in the same laboratory (Acme Analytical Laboratories, Ltd.). Quality checks included the analysis of certified reference materials (rice flour SRM 1568 from the National Bureau of Standards—NBS), blanks and duplicate samples. Reproducibility was estimated to be better than ±10%.

Results and discussion

Heavy metal concentration in the soil

Concentrations of As, Cd, Cu, Pb and Zn in the paddy soil from Kočani Field together with the assumed permissible level of heavy metals adopted by the National Environmental Protection Agency of Slovenia (Uradni list RS 1996) and their typical contents of comparable soils around the world (Bowen 1979) as well as the maximum allowable concentrations (MAC) of trace elements in agricultural soil proposed by the German Federal Ministry of Environment (1992) are presented in Table 1. Table 2 shows the main statistical parameters (mean, median, range and standard deviation [SD]) for the analysed paddy-soil samples.

The As concentration in uncontaminated, untreated soils rarely exceeded 10 µg g^{-1} (Adriano 1986). In the western Hunan mining area in China the As content in paddy soil polluted by mine tailing and mining/smelting wastewater was in the range 325–4,317 µg g^{-1} (Yang et al. 2004). In agricultural soil impacted due to past mining activities around the Songcheon Au–Ag mine in South Korea As concentrations were in the range 53–490 µg g^{-1}, with a mean value of 230 µg g^{-1} (Lee et al. 2005). In addition, Liu et al. (2005) found the mean concentration for As in agricultural soil from the Chenzhou Pb–Zn mine-affected area (Hunan, China) to be 459 µg g^{-1}. In this study, the median concentrations of As (6.7 µg g^{-1}) in the paddy soil and the range of 3.1–47.6 µg g^{-1} (Table 2) were much lower than those previously reported As concentrations in soils affected by mining activities. However, the elevated concentrations of As in the range from 20.7 to 47.6 µg g^{-1} found in the

Table 1 Total elemental concentrations in the paddy soil of Kočani Field: (1) typical contents of As, Cd, Cu, Pb and Zn in comparable soils around the world (Bowen 1979); (2) limits for elemental concentrations in soil (Environmental Protection Agency of Slovenia (Uradni list RS 1996); (3) maximum allowable concentrations (MAC) of trace elements in agricultural soils proposed by the German Federal Ministry of the Environment (1992)

Element		As (µg g⁻¹)	Cd (µg g⁻¹)	Cu (µg g⁻¹)	Pb (µg g⁻¹)	Zn (µg g⁻¹)
Location	Sample no.					
I-1	1	3.4	0.1	15	19	53
I-3	2	18.7	0.5	40	81	162
I-5	3	5.6	0.2	32	19	85
I-6	4	5.1	0.1	26	13	70
II-1	5	6.1	0.2	27	27	93
II-3	6	5.0	0.1	29	21	81
II-5	7	5.3	0.2	25	21	80
II-6	8	11.8	0.3	26	32	100
III-1	9	6.1	0.2	27	18	89
III-3	10	3.1	0.4	33	20	95
III-5	11	8.3	0.2	33	24	102
III-7	12	8.2	0.2	17	22	64
III-8	13	4.2	0.1	22	11	64
IV-1	14	3.8	0.1	29	14	74
IV-2	15	3.5	0.2	17	18	68
IV-3	16	4.3	0.3	23	19	102
IV-5	17	6.3	0.2	26	15	94
IV-7	18	8.1	0.2	26	17	83
IV-8	19	5.9	0.2	29	18	96
V-1	20	10.0	0.2	19	31	85
V-3	21	7.8	0.2	20	27	73
V-5	22	6.5	0.2	25	22	98
V-7	23	6.4	0.2	23	18	67
V-9	24	4.5	0.2	21	19	69
VI-1	25	5.0	0.2	21	24	71
VI-2	26	6.8	0.3	25	39	76
VI-3	27	10.1	0.3	34	35	86
VI-4	28	9.9	0.3	29	41	105
VI-5	29	10.5	0.3	25	40	94
VI-6	30	11.5	0.3	28	45	107
VI-7	31	7.7	0.1	19	20	66
VI-8	32	6.2	0.2	30	16	72
VII-1	33	22.2	2.7	49	412	531
VII-2	34	42.0	5.6	99	892	1,134
VII-3	35	35.1	4.5	89	727	893
VII-4	36	47.6	6.4	80	983	1,245
VII-5	37	39.6	5.0	68	745	928
VII-6	38	20.7	2.0	47	296	384
1		6.	0.35	30	35	90
2		20	1	60	85	200
3		–	1.5	60	100	200

Table 2 Basic descriptive statistics of the elemental content in the paddy soil of Kočani Field

Element	As (µg g^{-1})	Cd (µg g^{-1})	Cu (µg g^{-1})	Pb (µg g^{-1})	Zn (µg g^{-1})
Mean	11.4	0.9	33	128	206
Median	6.7	0.2	26	22	88
Minimum	3.1	0.1	15	11	53
Maximum	47.6	6.4	99	983	1,245
SD	11.3	1.7	20	260	310

paddy soils from section VII significantly exceeded the level of 6 µg g^{-1} reported by Bowen (1979), and also the limit value of 20 µg g^{-1} reported by the Environmental Protection Agency of Slovenia (Table 1).

The average content of Cd in soils was between 0.07 and 1.1 µg g^{-1} (Kabata-Pendias and Pendias 2001). In agricultural soil, the background Cd level is about 1 µg g^{-1} or less (Whitby et al. 1978). Similar background values of 0.35 µg g^{-1} were also reported for soils all around the world by Bowen (1979). According to Jung and Thornton (1996) the mean concentrations of Cd in soils in an area contaminated by past Pb–Zn mining in northwest Korea were 11.8 µg g^{-1} (0–15 cm depth), 6.0 µg g^{-1} (15–30 cm depth) and 5.1 µg g^{-1} (30–40 cm depth). Nearby uncultivated soils and household garden soils contained less than 3.3 µg g^{-1} Cd. Elevated concentrations of Cd ranging from 2.01 to 29.68 µg g^{-1} with a mean value of 13.59 µg g^{-1} were found in the paddy soil of the Lechang Pb–Zn mine area in China irrigated with mining waste water (Yang et al. 2004). The mean Cd content in the agricultural soil of 6.77 µg g^{-1} was further reported from the Chenzho Pb–Zn mine-affected area in Hunan, China (Liu et al. 2005). In the present study Cd concentrations in the paddy soil sampled were in the range 0.1–6.4 µg g^{-1}, with a median value of 0.2 µg g^{-1} (Table 2), which was less than that of 0.35 µg g^{-1}, reported for the world-average Cd content in soil by Bowen (1979). However, the highly elevated Cd concentration measured in the paddy soil in section VII, close to the Zletovska River, significantly exceeded the background Cd level of agricultural soil (1 µg g^{-1} or less) reported by Whitby et al. (1978) as well as the permitted value of 1 and 1.5 µg g^{-1}, suggested by the environmental protection agencies of Slovenia and Germany (Table 1).

The total Cu content of world soil is 30 µg g^{-1}, with values ranging from 2 to 250 µg g^{-1} (Bowen 1979). Contaminated soils around the Pb–Zn Sambo mine in Korea vary between sampling sites, ranging up to 1,400 µg g^{-1} in soils near the mine site, although most soils did not exceed 100 µg g^{-1} Cu due to the small extent of the Cu mineralization (Jung and Thornton 1996). The mean concentration of Cu in the agricultural soil around the Songcheon Au–Ag mine (South Korea) was 120 µg g^{-1} (Lee et al. 2005), while in the agricultural soil affected by the Pb–Zn mine, Chenzhou it was slightly higher, 174.03 µg g^{-1} (Liu et al. 2005). Cao and Hu (2000) also reported elevated concentrations of Cu (90.6 ± 12.9 µg g^{-1}) in paddy soil irrigated with Cu-rich wastewater. In this study, the Cu concentrations in the paddy soil samples were in the range 15–99 µg g^{-1} with a median value of 26 µg g^{-1} (Table 2), which was below the Cu content of world soil, i.e., 30 µg g^{-1} (Bowen 1979). The Cu concentration levels in the paddy soil samples in section VII were much higher, with a range of 47–99 µg g^{-1}, which exceeded the limit emission values of 60 µg g^{-1} reported by the environmental protection agencies of Slovenia and Germany (Table 1).

Agricultural soils have a wide range of Pb content, which depends on a number of factors, such as the parent material and the anthropogenic input (Adriano 1986). Jung and Thornton (1996) reported Pb concentrations in surface soils contaminated by the Pb–Zn mine in Korea ranging from 41 to 29.900 µg g^{-1}. In China, Yang et al. (2004) found Pb-contaminated paddy soil near the Lechang Pb–Zn mine, with levels of 325–4,317 µg g^{-1} due to the irrigation with mining wastewater. The mean concentrations of Pb in the agricultural soil around the Songcheon Au–Ag mine (South Korea) were reported to be 160 µg g^{-1} (Lee et al. 2005). A similar mean concentration level of 174 µg g^{-1} was also measured in the Chenzhou Pb–Zn mine (Hunan, China), which affected the agricultural soil (Liu et al. 2005). A median value of 22 µg g^{-1} for

Pb and a range of 11–983 µg g^{-1} (Table 2) were found during this study for the Kočani paddy soil. The highest concentration values were, however, measured in samples from the polluted section VII. Paddy soil from this section exhibited Pb concentrations in the range of 296−983 µg g^{-1} (Table 1), which is above the median values of Pb content in contaminated agricultural soil reported by Lee et al. (2005) and Liu et al. (2005) as well as above the limits provided by the environmental protection agencies of Slovenia and Germany (Table 1).

Average Zn concentrations of 90 µg g^{-1} have been given for world soil by Bowen (1979). More recently, Berrow and Reaves (1984) reported a mean content of 40 µg g^{-1} for world soil. However, soil highly influenced by base-metal mining in Korea had Zn concentrations close to the mine of between 329 and 25.8 µg g^{-1}, with an arithmetic mean of 8,300 µg g^{-1}. The Zn contents in other sites were, however, much lower (<230 µg g^{-1}) (Jung and Thornton 1996). The mean concentrations of Zn in paddy soils impacted by past and/or recent mining activities were, according to Lee et al. (2001), in the range from 60 to 1,064 µg g^{-1}, with a mean value of 328 µg g^{-1}, while those reported by Liu et al. (2005) ranged from 613 to 1,064 µg g^{-1}, with a mean value of 855 µg g^{-1}. Highly elevated concentrations of Zn were also found in the paddy soil from section VII, ranging from 384 to 1,245 µg g^{-1}. The Zn contents in the other sampling sections, however, were much lower (<162 µg g^{-1}). Compared with the heavy metal limits (200 µg g^{-1}) adopted by the national environmental protection agencies of Slovenia and Germany, Zn concentrations are predominantly raised only in the soil samples from section VII (Table 1).

The presented data clearly show that the paddy soil from section VII in the vicinity of the Zletovska River is highly impacted with As, Cd, Cu, Pb and Zn. Their maximum concentration values of 47.6 µg g^{-1} As, 6.4 µg g^{-1} Cd, 99 µg g^{-1} Cu, 983 µg g^{-1} Pb and 1,245 µg g^{-1} Zn are significantly higher than those from the other sampling sections and easily exceed the limit emission values of 20, 3, 100 and 300 µg g^{-1} for As, Cd, Pb and Zn, respectively, reported by the national environmental protection agencies of Slovenia and Germany (Table 1). The analysis of heavy metals measured during this study of paddy-soil samples for their possible intersection variation showed, according to a t-test, statistically significant difference at $\alpha \leq 0.05$ for As, Cd, Cu, Pb and Zn only between sampling section VII and all the other sections (I–VI) (Table 3).

In order to compare the distribution of each element in the paddy soil between the sampling sections I–VII, a box plot was employed (Fig. 3). As shown in Fig. 3, all the soil samples of the soil sections I–VI had similar median values, suggesting that their intersection differences were not substantial in most cases, while statistically significant variations existed only between sampling section VII and the other sections (I–VI).

The total concentrations of heavy metals in soils may be derived from two sources: natural and

Table 3 Results of t-test to evaluate the significant differences at $\alpha \leq 0.05$ between the sections I to VII for As, Cd, Cu, Pb and Zn

Sections	I	II	III	IV	V	VI	VII
I							As*, Cd*, Cu*, Pb*, Zn*
II				Pb*	Cu*		As*, Cd*, Cu*, Pb*, Zn*
III						Pb*	As*, Cd*, Cu*, Pb*, Zn*
IV		Pb*			Pb*	As*, Pb*	As*, Cd*, Cu*, Pb*, Zn*
V		Cu*		Pb*			As*, Cd*, Cu*, Pb*, Zn*
VI			Pb*	As*, Pb*			As*, Cd*, Cu*, Pb*, Zn*
VII	As*, Cd*, Cu*, Pb*, Zn*,	As*, Cd*, Cu*, Pb*, Zn*	As*, Cd*, Cu*, Pb*, Zn*	As*, Cd*, Cu*, Pb*, Zn*	As*, Cd*, Cu*, Pb*, Zn*	As*, Cd*, Cu*, Pb*, Zn*	As*, Cd*, Cu*, Pb*, Zn*

* Significant difference at $\alpha \leq 0.05$

Fig. 3 Whisker plots of As, Cd, Cu, Pb and Zn for soil samples from Kočani Field
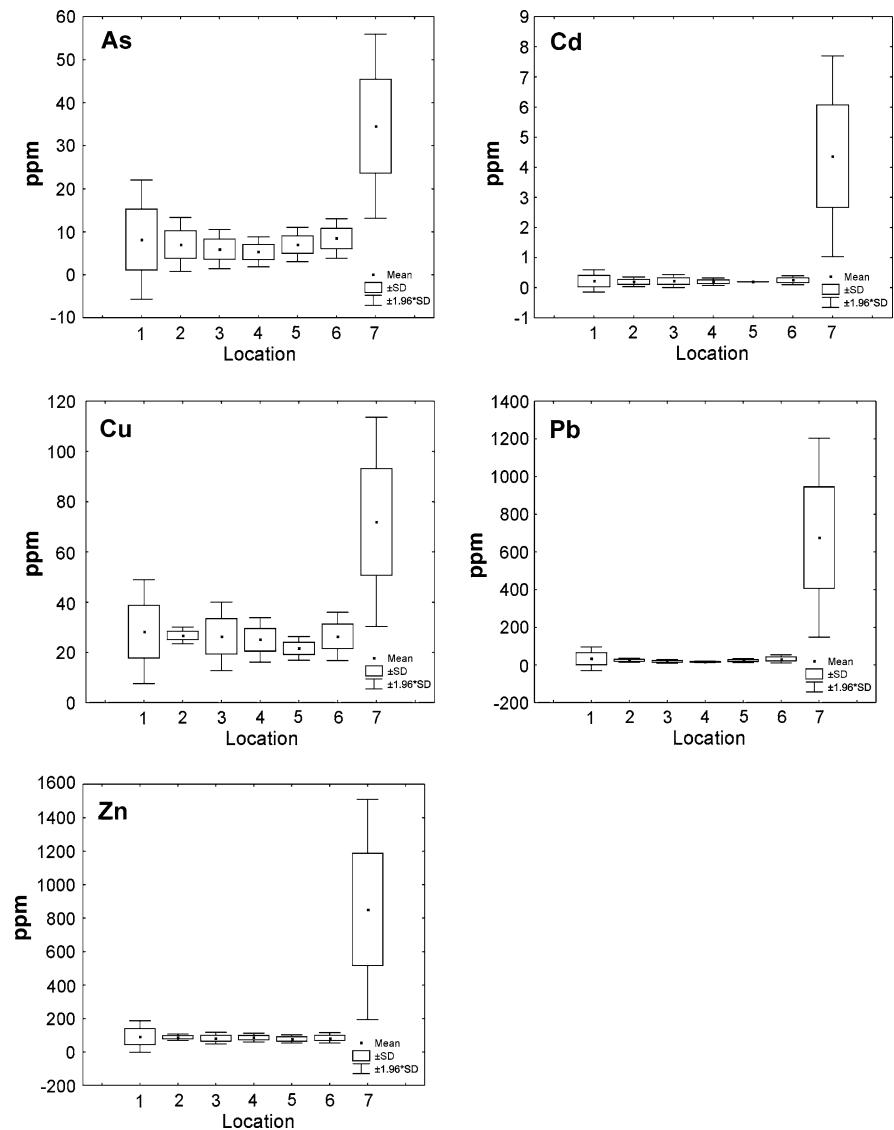
anthropogenic. Therefore, it is not easy to decide a priori which of these two contributions will represent the main source of heavy metal pollution. However, in the case of the paddy soil from section VII in the vicinity of the Zletovska River it is very clear that the anthropogenic impact due to acid mine drainage and mining activities is the main source of As, Cd, Cu, Pb and Zn contamination. All these metals are important ore-forming elements and are paragenetically related to the Pb–Zn polymetalic mineralization of the Zletovo-Kratovo ore district

predominately drained by the Zletovska River (Dolenec et al. 2007). Many studies have revealed heavy metal contamination in soil, plant, waters and sediment from metaliferous mines and mining activities throughout the world (Adriano 1986).

Heavy metal concentrations in rice

The concentrations of As, Cd, Cu Pb and Zn in unpolished rice grown on the paddy soils of Kočani Field together with the maximum permissible limit of

Table 4 Total As, Cd, Cu, Pb and Zn concentrations in unpolished rice from Kočani Field

Elements Location	$\mu g\ g^{-1}$ Sample	As	Cd	Cu	Pb	Zn
I-1	R-1	0.30	0.010	3.8	0.1	31
I-3	R-2	0.28	0.140	4.5	0.2	37
I-5	R-3	0.22	0.020	3.8	0.2	28
II-1	R-4	0.24	0.040	2.7	0.2	22
II-5	R-5	0.22	0.010	2.1	0.1	21
III-1	R-6	0.18	0.070	4.7	0.3	26
III-5	R-7	0.19	0.010	1.2	0.2	26
IV-7	R-8	0.42	0.010	2.0	0.1	15
V-1	R-9	0.28	0.150	3.2	0.1	31
V-3	R-10	0.33	0.010	1.3	0.2	19
VI-1	R-11	0.16	0.005	1.2	0.05	16
VI-4	R-12	0.15	0.005	0.8	0.1	13
VII-2	R-13	0.40	0.180	4.9	0.4	38
VII-4	R-14	0.53	0.310	5.8	0.5	67
	1		0.1		0.2	
	2			10		
	3	0.2	0.15		1.0	50

Maximum permitted levels for heavy metals in food crops: (1) Commission Regulation Directive EC (2001); (2) FAO/WHO (1992) and (3) Pilc et al. (1994)

analysed heavy metal concentrations in food crops adopted by various international and national agencies are shown in Table 4. Table 5 shows the main statistical parameters (mean, median, range and standard deviation [SD]) for the analysed unpolished rice samples.

Arsenic is a metaloid and is considered as toxic to both plants and humans. It is a naturally occurring element and the levels of As in foods generally reflect normal accumulation from the environment (Roychowdhury et al. 2003). An intake of 1 mg of inorganic As per day may give rise to skin lesions within a few years (WHO 1981). Edible plants grown on uncontaminated or unmineralized soil contain 0.01–1.5 $\mu g\ g^{-1}$ As (Kabata-Pendias and Pendias 2001). However, elevated levels of As and other heavy metals may be found in plants growing on contaminated soils. According to Lu and Zhang (2005) the As concentrations of rice from the polluted paddy soil in the Huayuan mining area (China) varied from 0.230 to 0.651 $\mu g\ g^{-1}$, with a mean value of 0.437 $\mu g\ g^{-1}$. Similar values in the range 0.216–0.551 $\mu g\ g^{-1}$ were also measured in rice from around the Chatian deposit area in China (Lu and Zhang 2005).

Our previous study of the trace metal contamination of rice from ten magazines of local farmers in Kočani Field showed that the As in unpolished rice was in the range 0.11–0.52 $\mu g\ g^{-1}$, with a median value of 0.18 $\mu g\ g^{-1}$ (Rogan et al. 2007). The As concentrations in unpolished rice (range 0.15–0.53 $\mu g\ g^{-1}$, median value 0.26 $\mu g\ g^{-1}$) found in this study (Table 5) were slightly higher than those observed in rice from the magazines. The differences and the lower concentration levels in rice seeds from the local farmers' magazines were most probably the result of the homogenisation of rice seeds from the paddy fields with different As contents. The highest concentrations (up to 0.53 $\mu g\ g^{-1}$) were measured in rice grown in the most impacted soils of section VII, close to the Zletovska River (Table 5). These values were, however, up to 2.7 times higher than the corresponding limit of 0.20 $\mu g\ g^{-1}$ for plants used for food (Pilc et al. 1994).

Cd is considered as one of the most environmentally hazardous trace metals. It is easily taken up and accumulated by plants and crops through their root systems and is present in all food (Alam et al. 2003). Research undertaken over the past 40 years has identified the irrefutable relationship between long-term consumption of Cd-contaminated rice and human Cd diseases such as itai-itai and proximal tubular renal dysfunction (Simmons et al. 2005). The background concentrations of Cd in cereal grains

Table 5 Descriptive basic statistic of the elemental content in unpolished rice from Kočani Field

Element	As ($\mu g\ g^{-1}$)	Cd ($\mu g\ g^{-1}$)	Cu ($\mu g\ g^{-1}$)	Pb ($\mu g\ g^{-1}$)	Zn ($\mu g\ g^{-1}$)
Mean	0.279	0.069	3.00	0.196	27.86
Median	0.26	0.015	2.95	0.20	26
Minimum	0.15	0.005	0.80	0.05	13
Maximum	0.53	0.310	5.80	0.50	67
SD	0.11	0.092	1.62	0.13	13.68

were, in general, in the range 0.013–0.22 µg g^{-1} (Kabata-Pendias and Pendias 2001). The Cd content in rice grown in various countries ranged from 0.01 to 0.05 µg g^{-1} (Jung 1995). According to Yang et al. (2006) the average Cd concentration level in unpolished rice contaminated with untreated mining water grown in paddy fields around the Pb–Zn mine in Lechang (southern China) was 0.24 µg g^{-1}. An extremely high concentration of Cd (6.99 µg g^{-1}) was measured in rice affected by the Chenzhou Pb–Zn mine spill in China (Liu et al. 2005).

In rice sampled during this study the Cd concentrations varied from 0.005 to 0.310 µg g^{-1}, with a median value of 0.015 µg g^{-1} (Table 5). The measured values were similar to the background levels of Cd in cereal grains reported by Kabata-Pendias and Pendias (2001). However, at some sampling sites the Cd concentrations in unpolished rice exceed the internationally recognized maximum level (ML) for Cd in rice grain of 0.2 µg g^{-1} (CAC 2002) as well as the maximum permitted level of 0.1 µg g^{-1} in cereals given by the Commission Regulation Directive EC (2001) (Table 4). Similar high concentrations of Cd (0.21 µg g^{-1}) were also determined in rice from one local magazine containing mostly the seed from paddy soils close to the Zletovska River (Rogan et al. 2007). In the study area, the highest concentrations of Cd, like other heavy metals analysed during this study, were also measured in rice grown in the vicinity of the Zletovska River.

Cu is an essential element for plant growth and development. In various plants grown in uncontaminated soils Cu levels do not generally exceed 20 µg g^{-1}. In cereal grains from different countries Cu concentrations were in the range 0.6–10.3 µg g^{-1}. In plants grown in contaminated sites impacted by the metal-processing industry Cu concentrations could be as high as 560 µg g^{-1} (Kabata-Pendias and Pendias 2001). Jung et al. (2005) reported that the Cu concentrations in polished rice from South Korea were in the range of 1.29–2.53 µg g^{-1}, with a median value of 1.85 µg g^{-1}. Elevated Cu concentration levels of up to 15.5 µg g^{-1} were measured by Cao and Hu (2000) in brown rice grown in paddy soils irrigated with Cu-rich wastewater. Lee et al. (2001) found that the Cu concentration in rice grain from heavy-metal-impacted paddy soils around the Daduk Au–Ag–Pb–Zn mine (Korea) range from 1.6 to 4.6 µg g^{-1}, with a mean value of 3.1 µg g^{-1}. According to Jung and Thornton (1996) the average Cu content in most plants grown in the contaminated soil in the vicinity of the Pb–Zn Sambo mine in Korea were, however, <20 µg g^{-1}.

The concentrations of Cu in the rice samples from the studied area varied between 0.8 and 5.8 µg g^{-1}, with a median value of 2.95 µg g^{-1} (Table 5). The highest concentrations were found in rice from the most impacted sites in section VII, close to the Zletovska River (Table 4). However, in none of the 14 sampling sites did the Cu concentration in unpolished rice exceed the maximum permissible limit of 10 µg g^{-1} Cu (WHO 1998). These results indicated that the Cu concentrations in rice from Kočani Field should not be harmful to human health through the food chain.

Pb is also thought to be one of the major chemically toxicologically dangerous trace metals. It is naturally present in small amounts in practically all environmental matrices. The Pb content in an edible portion of the plants grown in an uncontaminated area, as reported by various authors for the decade 1970–1980, range from 0.05 to 3 µg g^{-1}, while the mean Pb content calculated for cereal grains of various countries varies from 0.01 to 2.28 µg g^{-1} (Kabata-Pendias and Pendias 2001). Pb concentrations higher than 1 µg g^{-1} are, however, considered to provide consumers with an excessive amount of Pb (Witek et al. 1992). Median concentrations for Pb higher than 0.190 µg g^{-1} were found in polished rice from South Korea (Jung et al. 2005). However, elevated concentrations of Pb, which may present a health risk to humans, could be found in food crops growing in contaminated soil; for example, unpolished rice from the paddy soil around the Pb–Zn Lechang mine (South China) showed mean Pb concentrations of about 4.67 µg g^{-1} (Yang et al. 2004). Similar, high Pb contents, which were generally in the range from 0.1 to 4.3 µg g^{-1}, were also measured in cereal grains from the Chenzhou Pb–Zn mine (Hunan, China) affected area (Liu et al. 2005). According to Jung and Thornton (1996) the concentrations of Pb in rice grain grown in soils contaminated by the Pb–Zn mining activities in Korea averaged 0.22 µg g^{-1}.

The range and the median concentrations of Pb in rice sampled during this study were found to be 0.05–0.5 and 0.2 µg g^{-1}, respectively (Table 5). The highest Pb content (Table 4) was measured in rice collected from the most polluted sampling sites of

section VII with the highest concentration of Pb and other heavy metals. The measured values indicated that Pb concentrations were higher than the maximum permitted levels of 0.2 μg g^{-1} for food crops reported by the Environmental Protection Agency of Slovenia (Table 4).

Zinc is essential for both plants and animals. However, in high concentrations it can be toxic to living organisms (Dudka et al. 1995). In edible plants from uncontaminated soils the Zn concentrations usually range from 1.2 to 73 μg g^{-1} (Kabata-Pendias and Pendias 2001). Rice sold in the USA showed slightly higher average Zn concentrations of about 23 μg g^{-1} (Nriagu and Lin 1995). Similar average values of 22.5 μg g^{-1} for Zn were also measured in rice grown in soils contaminated by Pb–Zn mining activities in Korea (Jung and Thornton 1997). These values were much lower than those of about 43.19 μg g^{-1} reported for the Zn content in rice from the impacted area around the Chenzhou mine (Liu et al. 2005).

The Zn concentrations in rice measured during this study were in the range 13–67 μg g^{-1}, with a median value of 26 μg g^{-1} (Table 5), and were slightly higher relative to those (range 11.7–23.7 μg g^{-1} Zn, median 17.5 μg g^{-1} Zn) previously reported for unpolished rice sampled in the magazines of local farmers in Kočani Field (Rogan et al. 2007). Taking into account two limits proposed for Zn concentrations in crops, 50 μg g^{-1} in crops grown for food and 100 μg g^{-1} in crops grown for forage (Pilc et al. 1994), it appears that only one sample (R-4) close to the Zletovska River with Zn concentrations of 67 μg g^{-1} (Table 4) exceeded the concentration limit proposed for food crops.

The presented data suggest that unpolished rice from Kočani Field had normal concentrations of Cu, which did not exceed the critical contents of 10 μg g^{-1} (WHO 1998) while those of Zn were found to be slightly elevated above the critical values of 50 μg g^{-1} for plants used as food (Pilc et al. 1994) only at sampling site VII-4, in the vicinity of the Zletovska River. Cd, however, was elevated above its limit level for food of 0.1 μg g^{-1} (Commission Regulation Directive EC 2001) not only in the rice sampled in section VII, but also in the rice from sampling sites I-3 (sample R-2) and V-1 (sample R-9). Pb contents above the maximum permitted level of 0.2 μg g^{-1} were also measured in rice grown in

the paddy soil close to the Zletovska River (Commission Regulation Directive EC 2001). The Pb concentrations in rice from some other sampling sites were also as high as the permitted level (Table 4). In addition, As concentration that exceeded the critical content of 0.2 μg g^{-1} for food (Pilc et al. 1994) by up to 2.7 times was found in the rice samples from section VII. In some other sections (I, II, IV and V) the concentrations were, however, only slightly lower. It seems that these locations are polluted with As, not only due to irrigation with polluted riverine water, but probably also from As that could be released as a result of geogenic sources.

Heavy metal accumulation in crops is a function of complex interactions between the soil, the plant and environmental factors. It has been well documented that the content of heavy metals in crop plants is closely related to the levels in the soil (Cheng et al. 2006). It is not surprising, therefore, that As, Cd, Cu, Pb and Zn were the highest in the rice grown in the most impacted part of Kočani Field. It appears that the elevated concentrations of the analysed heavy metals were undoubtedly related to the past and present mining activities of the region, especially those in the Zletovo-Kratovo ore district.

Although the total heavy metal concentrations in the rice from Kočani Field can play an important role in their uptake into local diet, the potential for human intoxication by heavy metals is not simply related to the concentration in the food. The most important factor is the overall intake of a particular element during a specified period of time, usually a week (Dudka et al. 1995). The total dietary intake of heavy metals, however, is not only determined by their level in food, but also by the amount of consumed contaminated food in the whole diet and the intake of heavy metals from other sources such as drinking water, atmosphere, occupational exposure and everything in the environment (Dudka et al. 1995; Liu et al. 2005). Therefore, up to now it has been impossible to predict the human health hazard related to element toxicity based only on heavy metal concentrations in the rice sampled during this study. For a further discussion more detailed studies on the heavy metal concentration in agricultural soils, irrigation and drinking water, rice plant and other edible crops as well as a dietary study of the local population around Kočani Field are essential.

Conclusions

In this study we examined the distribution of heavy metals (As, Cd, Cu, Pb and Zn) in paddy soil and unpolished rice from Kočani Field. The results showed that the paddy soil of the western part of Kočani Field in the vicinity of the Zletovska River exhibited very elevated concentrations of As, Cd, Cu, Pb and Zn, with maximum values of 47.6, 6.4, 99, 983 and 1,245 $\mu g \ g^{-1}$, which significantly exceed the limits proposed by the Slovenian and German environmental agencies.

A chemical characterization of the unpolished rice also revealed the highest concentrations of As, Cd, Cu, Pb and Zn (0.53, 0.31, 5.8, 0.5 and 67 $\mu g \ g^{-1}$) for rice grown in the most polluted paddy soil in the vicinity of the Zletovska River. The concentration levels of Cu were below the maximum permitted value of 10 $\mu g \ g^{-1}$ (WHO 1998), whereas the As and Zn contents exceeded their permitted values of 0.20 and 0.50 $\mu g \ g^{-1}$, reported by Pilc et al. (1994) for food crops. Compared with the maximum permitted levels of 0.1 $\mu g \ g^{-1}$ for Cd and 0.2 $\mu g \ g^{-1}$ for Pb in cereal grains and other food crops (Commission Regulation Directive EC 2001) the Cd and Pb contents in rice grown in paddy soil close to the Zletovska River also exceeded these values. In the case of As, Cd and Pb, slightly elevated concentrations above the permitted values were also found in some other sampling sites of Kočani Field.

The elevated concentrations of As, Cd, Cu, Pb and Zn in paddy soil and rice undoubtedly indicated heavy metal contamination related to mining activities and acid mine drainage-impacted riverine water, which is used by local farmers for irrigation purposes.

To predict a dietary intake of these heavy metals by the human population and to assess the possible health risk, more detailed studies on heavy metal contamination in agricultural soils, irrigation and drinking water, rice and other edible crops as well as a dietary study of the local population are needed.

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