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Original Research

Contents of Nitrates (III) and (V), Lead and Cadmium in Select Domestic Fruits

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Abstract

This paper deals with determining nitrates (III) and (V), lead and cadmium in 10 apple, 10 plum, 12 strawberry, 12 raspberry, and 10 white grape samples. Examined products originated from allotment gardens localized in the center of Lublin as well as in its surroundings. Among the analyzed fruits, the highest nitrate (III) contents were found in plums and white grapes harvested from the allotments located outside the city. Regardless of the harvest site, the highest content of nitrates (V) was recorded in strawberries. Exceeding permissible lead levels occurred in the majority of analyzed fruit samples, while in the case of cadmium, the permissible limit was exceeded only in white grape samples.

Keywords: nitrates (III) and (V), lead, cadmium, domestic fruits, harvest site

Introduction

Fruits are a valuable source of crude fiber, vitamins, minerals, and organic acids. They also may be a source of chemical contamination in the human diet: mainly heavy metals, such as lead and cadmium, pesticides, and nitrates (III) and (V). The presence of the harmful chemicals mentioned above is one of the basic criteria of evaluating food safety. Due to the fact that fruit consumption in Poland is growing, it is necessary to limit nitrates (III) and (V) contents, namely lead and cadmium in fruits. As yet, few reports on nitrates (III) and (V) contents in fruits have been announced [1-3]. Although nitrates (V) are not an important health problem, nitrates (III) that are formed from them as a result of microbial reduction are highly toxic [4]. In addition to the fact that nitrates (III) can cause infantile methemoglobinemia, it was also shown that in vivo in humans, carcinogenic N-nitrosoamines can be formed from them. Consequently, nitrates (III) are suspected of causing gastric cancer and other malignancies [5].

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On the other hand, fruits contaminated with cadmium and lead have been widely examined in numerous experiments involving fruits harvested mainly in Poland [6-10].

Our study was aimed at evaluating the contents of nitrates (III) and (V), as well as heavy metals such as lead and cadmium in domestic fruits (apple, plum, strawberry, raspberry, and white grape) harvested from the allotments located in the center of Lublin and its surroundings.

Materials and Methods

Collecting and Preparation of Samples

Fruit samples were collected from various places in Lublin and its surroundings between July and September 2007. The following fruits were gathered: apple, plum, strawberry, raspberry and white grape. Analyzed fruit samples originated from potentially contaminated areas (allotment gardens in the center of Lublin and those adjacent to the road–100 m) and potentially not exposed to industrial and transportation pollution (allotment gardens in the Lublin surroundings far from any inter-city road

Table 1. Nitrates (III) and (V) content in the selected fruits (mg kg^{-1} of wet mass).

Harvest site	Number of samples	Fruits	Nitrates (III) ($NaNO_2$)	Median	Distant quartile	Nitrates (V) (KNO_3)	Median	Distance quartile
Allotment gardens in Lublin center	10	apple	*	*	*	2.41-5.22 **$\bar{x}$ 3.35**	2.89	1.72
	10	plum	*	*	*	1.96-6.46 **$\bar{x}$ 3.87**	3.53	2.30
	12	strawberry	0.00-0.44 **$\bar{x}$ 0.11**	0.06	0.16	2.87-243.8 **$\bar{x}$ 47.28**	37.73	58.38
	12	raspberry	0.00-0.35 **$\bar{x}$ 0.12**	0.10	0.14	5.02-27.44 **$\bar{x}$ 7.23**	5.23	4.38
	10	white grape	0.00-0.22 **$\bar{x}$ 0.19**	0.22	0.19	1.53-9.96 **$\bar{x}$ 4.23**	3.08	3.87
Allotment gardens in Lublin surroundings	10	apple	*	*	*	2.03-2.71 **$\bar{x}$ 2.33**	2.28	0.41
	10	plum	0.00-0.42 **$\bar{x}$ 0.21**	0.20	0.23	3.05-10.07 **$\bar{x}$ 4.57**	2.89	3.98
	12	strawberry	0.00-0.41 **$\bar{x}$ 0.16**	0.12	0.21	1.93-312.1 **$\bar{x}$ 55.87**	2.76	107.6
	12	raspberry	0.00-0.28 **$\bar{x}$ 0.09**	0.10	0.10	4.53-12.93 **$\bar{x}$ 6.59**	5.03	5.29
	10	white grape	0.00-0.36 **$\bar{x}$ 0.21**	0.23	0.19	1.53-10.55 **$\bar{x}$ 4.08**	2.12	4.54

$\bar{x}$ – mean value; * – lower than limit of detection
a, b – values in the same rows marked with different letters differ significantly at $p \leq 0.05$
$NaNO_2 = NO_2^-$ x 1.5
$KNO_3 = NO_3^-$ x 1.63

and 15-20 km outside Lublin – Niedrzwica Duża, Niedrzwica Kościelna, Krężnica Jara, Łuszczów, and Biskupice).

In total, 108 domestic fruits samples were examined. The numbers of samples of collected fruits were as follows: apple, 10; plum, 10; strawberry, 12; raspberry, 12; and white grape, 10 samples. The fruits were washed with de-ionized water (Millipore, France) and after removal of inedible parts, dried in the air, then dried in an electric drier at 60ºC. After complete drying (at 103ºC), the fruits were ground in a WŻ-1-type mill and stored in glass containers waiting for analyses. The dry mass content in the fruit samples was estimated by means of a drier-weight method [11].

Mineralization of Samples

For determining lead and cadmium contents, 5g fruit samples were placed into quartz crucibles and combusted in a muffle furnace at 450°C. Sample combusting is recommended for large samples of organic matter (e.g. 5-100g), [12]. After dry mineralization the resulted ash was diluted with 5 cm^3 6N spectra pure HCl (Merck). Then diluted samples were transferred quantitatively to 100 cm^3 measuring flasks and filled with water to the mark. For preparing hydrochloric acid solutions and for analysis we used water made in a Direct Q 5 deionizer.

Determination of Lead and Cadmium

Lead and cadmium contents in analyzed fruits were determined according to PN-EN 14082 in the Central Apparatus Laboratory, University of Life Science in Lublin, by means of non-flame AAS technique on Varian Spectra AA 880Z spectrometer with deuterium background correction [13]. Lead was detected at λ=217.0 nm, and cadmium at λ=228.8 nm. Detection limits for these metals were as follows: 50 μg l^{-1} and 5 μg l^{-1}, respectively. In order to make the calibration line, working solutions of examined ions prepared by dilution of standard solutions – 50 μg l^{-1} (Pb) and 2.5 μg l^{-1} (Cd), (Merck) were applied.

For calibration and validation of analytical procedures we used certified reference material (CRM) – oriental tobacco (CTTA-OTL-1, Poland).

Determination of Nitrates (III) and (V)

The determination of nitrate (III) and (V) contents was performed using the colorimetric method according to PN-92/A-75112 that consists of the measurements of intensity of color formed by nitrates (III) with Griess's agent [14].

All chemical determinations were made in two replications. The obtained numerical data were subjected to a statistical analysis by means of Statistica 6 software. The statistical significance of differences between mean values

Table 2. Lead and cadmium content in the selected fruits (mg kg^{-1} of wet mass).

Harvest site	Number of samples	Fruits	Lead	Median	Distant quartile	Numbers of samples <NDZ %	Cadmium	Median	Distance quartile	Number of samples <NDZ %
Allotment gardens in Lublin center	10	apple	0.008-0.21 **0.09**	0.07	0.16	50	0.00-0.03 $\bar{x}$ **0.009**	0.01	0.009	–
	10	plum	0.06-0.17 $\bar{x}$ **0.13**	0.14	0.08	70	0.009-0.02 $\bar{x}$ **0.01**	0.01	0.005	–
	12	strawberry	0.05-0.44 $\bar{x}$ **0.23**	0.21	0.20	75	0.03-0.05 $\bar{x}$ **0.04**[a]	0.04	0.009	–
	12	raspberry	0.12-0.39 $\bar{x}$ **0.27**[a]	0.28	0.14	75	0.008-0.02 $\bar{x}$ **0.02**	0.01	0.009	–
	10	white grape	0.06-0.09 $\bar{x}$ **0.06**	0.06	0.01	–	0.06-0.23 $\bar{x}$ **0.19**	0.21	0.13	100
Allotment gardens in Lublin surroundings	10	apple	0.01-0.20 $\bar{x}$ **0.08**	0.07	0.09	70	0.005-0.02 $\bar{x}$ **0.01**	0.01	0.007	–
	10	plum	0.04-0.27 $\bar{x}$ **0.13**	0.11	0.13	70	0.019-0.023 $\bar{x}$ **0.01**	0.01	0.006	–
	12	strawberry	0.12-0.37 $\bar{x}$ **0.31**	0.34	0.18	75	0.006-0.017 $\bar{x}$ **0.01**[b]	0.01	0.005	–
	12	raspberry	0.08-0.12 $\bar{x}$ **0.09**[b]	0.09	0.03	–	0.007-0.03 $\bar{x}$ **0.02**	0.03	0.01	–
	10	white grape	0.07-0.15 $\bar{x}$ **0.09**	0.08	0.04	–	0.11-0.29 $\bar{x}$ **0.22**	0.24	0.11	100

$\bar{x}$ – mean value
a, b – values in the same rows marked with different letters differ significantly at $p \leq 0.05$
NDZ – highest permissible content

was estimated by means of a single-factorial variance analysis ANOVA assuming significance level at 0.05.

Results and Discussion

The results related to nitrates (III) and (V) contents in selected fruits are presented in Table 1.

Nitrates (III) varied greatly in studied fruits (Table 1). Plums and grapes from gardens localized outside the city contained the highest concentration of nitrates (III) (0.21 mg $NaNO_2$ kg^{-1} FW). No nitrates (III) were found in plums harvested in the allotment gardens localized in the city center as well as in the apples, regardless of the harvest site. These results are the mean level of nitrates (III) in fruits originating from various sites, apparent differences were comparable to those achieved by Markiewicz et al. [1]. However, the studies of Sušin et al. [15] revealed much higher nitrate (III) levels in apples in relation to other fruits. When compared to those recorded only in the case of plums, however, they appeared to be statistically insignificant (Table 1).

The average nitrate (V) contents in particular fruit types from the allotment gardens in Lublin center and from its surroundings were quite similar (Table 1). Regardless of the harvest site, the highest nitrate (V) levels were found in strawberries (47.28-55.87 mg KNO_3 kg^{-1} FW). The contents of nitrate (V) in other fruits were much lower and oscillated within a relatively narrow range (from 2.33 mg KNO_3 kg^{-1} FW for apples from Lublin surroundings to 7.23 mg KNO_3 kg^{-1} FW for raspberries from Lublin center). The obtained results correspond to those reported by Nabrzyski and Gajewska [16], who recorded similar nitrate (V) contents in strawberry (58.72 mg KNO_3 kg^{-1} FW), while in other fruits the levels were within the range of 1.66-5.30 mg KNO_3 kg^{-1} FW. The studies conducted by Ognik et al. [17], related to nitrate (V) content in some berry fruits harvested in the Lublin region indicated these concentrations as 2.63-3.33 mg NO_3^- kg^{-1} FW regardless of the harvest site). According to the data achieved by Sušin et al. [15], mean nitrate (V) content in most of the fruits harvested in Slovenia in 1996-2002 was below 6.0 mg NO_3^- kg^{-1} FW in grapes, apples, and pears, whereas in strawberries it reached even up to 94.0 mg NO_3^- kg^{-1} FW. Our own study results revealed that in some strawberry samples (regardless the harvest site) even higher nitrate (V) levels were recorded (Table 1).

The contamination of fruits by nitrates (III) and (V) is negligible as compared to the values found in vegetables, namely leafy ones [18, 19]. The currently obliged EU Commission Decree (WE) No. 1881/2006 does not limit

Table 3. Lead and cadmium contents in the selected fruits (mg kg^{-1} of dry mass).

Harvest site	Number of samples	Fruits	Lead	Cadmium
Allotment gardens in Lublin center	10	apple	0.21-0.28 **$\bar{x}$ 0.25**	0.019-0.023 **$\bar{x}$ 0.02**
	10	plum	0.28-0.45 **$\bar{x}$ 0.37**	0.021-0.028 **$\bar{x}$ 0.02**
	12	strawberry	0.72-1.06 **$\bar{x}$ 0.94**	0.09-0.23 **$\bar{x}$ 0.16^{a}**
	12	raspberry	1.01-1.07 **$\bar{x}$ 1.04^{a}**	0.06-0.09 **$\bar{x}$ 0.07**
	10	white grape	0.09-0.2 **$\bar{x}$ 0.15**	0.46-0.52 **$\bar{x}$ 0.48**
Allotment gardens in Lublin surroundings	10	apple	0.21-0.24 **$\bar{x}$ 0.22**	0.019-0.027 **$\bar{x}$ 0.02**
	10	plum	0.35-0.41 **$\bar{x}$ 0.37**	0.018-0.028 **$\bar{x}$ 0.02**
	12	strawberry	1.23-1.36 **$\bar{x}$ 1.27**	0.035-0.049 **$\bar{x}$ 0.04^{b}**
	12	raspberry	0.32-0.4 **$\bar{x}$ 0.34^{b}**	0.05-0.09 **$\bar{x}$ 0.07**
	10	white grape	0.21-0.26 **$\bar{x}$ 0.23**	0.5-0.62 **$\bar{x}$ 0.56**

$\bar{x}$ – mean value
a, b – values in the same rows marked with different letters differ significantly at $p \leq 0.05$
NDZ – highest permissible content

these compounds' contents in fruits, because their combined consumption is usually much lower than the accepted daily intake (ADI), amounting to 3.65 mg kg^{-1} of body weight [20].

The results which refer to lead and cadmium contents in selected domestic fruits are presented in Table 2. For possibility of calculation the obtained results in dry and wet mass, dry mass contents in analyzed fruits samples is given in Table 3.

The analysis of mean lead content results in examined fruits indicated that it was very differentiated: from 0.06 mg kg^{-1} FW in white grapes from the gardens in the city center to 0.31 mg kg^{-1} FW in strawberries harvested from the city's suburbia (Table 2). There is no doubt that the level of contamination with lead depends on the environmental pollution and transport intensity in a given area. Such dependencies can be confirmed by results referring to raspberry fruits indicating significantly ($p \leq 0.05$) higher accumulation of the element in fruits from the city center allotment gardens in relation to those harvested from the out-of-city gardens (Table 2). The experiments carried out by Wojciechowska et al. [21], as well as Trętowska and Syrocka [22], revealed that berries are characterized by a higher vulnerability of intake and accumulation of lead and other heavy metals than the other ones. Their observations were confirmed in the present study of strawberry fruits (regardless of the harvest site), as well as the raspberries from the city center allotment gardens. Here the recorded results of fruit contamination by lead are higher or comparable to those achieved by other authors [2, 21, 23]. The observations made by Kocjan et al. [24] reveal that the apples harvested from the allotment gardens localized in Stalowa Wola city contained about 2-fold more lead as compared to those from Lublin center (Table 2). On the other hand, the results achieved by Kiczorowski and Kiczorowska [7] referring to lead concentration in apples from orchards near Lublin were lower than those in our own study. Except for raspberries from out-of-city and white grape samples regardless of the harvest site, exceeding permissible lead content was recorded, which disqualifies these fruits from consumption (Table 2). According to the requirements set in EU Commission Decree (WE) No. 1881/2006, the permissible lead content in berries and small fruits should not exceed the limit of 0.2 mg kg^{-1} FW and 0.1 mg kg^{-1} FW in the case of other fruits.

Susceptibility to cadmium accumulation in plants varies and greatly depends on the plant species [25]. It was also found that the increase of lead and cadmium intake by a plant is accompanied by the increase of soil acidity and decrease of organic matter, phosphorus, and calcium contents [26]. The results presented in Table 2 indicate the exceeding permissible levels for cadmium, regardless of the site of fruit harvest in grape (0.19-0.22 mg kg^{-1} FW), which may be an effect of the high ability to accumulate cadmium by that plant [21, 27]. According to EU Commission Decree (WE) No. 1881/2006, the permissible limit for cadmium was set as 0.05 mg kg^{-1} FW. Here the achieved results for the analyzed fruits indicate significantly ($p \leq 0.05$) a higher content of cadmium in strawberries harvested from the city center in relation to the fruits collected in the allotment gardens in Lublin surroundings (Table 2). The results related to cadmium concentration in the studied domestic fruits are higher or comparable to those recorded by Krejpcio et al. [6], Kiczorowska and Kiczorowski [8], as well as Szteke and Boguszewska [28] for seedy and berry fruits, which may prove great industrial and transportation pollution or specific soil factors existing in the studied area. Experiments performed by Rusinek et al. [23] involved in an analysis of berries harvested from the Lublin region indicated much higher cadmium contents (in raspberry and plums) and comparable ones in white grapes collected in the rural allotment gardens as compared to the results obtained here. Many times higher cadmium concentrations than those in the present study were recorded by Rusinek et al. [2] in the raspberry fruits harvested in Lublin region from the areas potentially free and potentially exposed to contamination.

In a majority of analyzed fruits, no dependence between the harvest site and nitrates (III) and (V), as well as heavy metals contents was observed. The recorded differences in lead and cadmium concentrations in the fruits harvested in allotment gardens localized in the city center and those in the city surroundings may have resulted from different levels of exposure to industrial and transportation pollution, or from different soil conditions existing in a given area.

Conclusions

1. Among the studied fruits, the highest content of nitrate (III) was found in plums and white grapes from the allotment gardens localized outside the city. Regardless of the harvest site, the largest amount of nitrates (V) was recorded in strawberries.
2. A significant dependence between the harvest site and heavy metals content was observed in the case of raspberries (lead) and strawberries (cadmium). Far higher levels of these metals were recorded in the fruits harvested in the city gardens.
3. The exceeding of permissible lead limit was recorded for a majority of the analyzed fruits, but the cadmium limit was exceeded only for white grape samples.

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