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Research Note

Content of Cadmium and Lead in Vegetables and Fruits Grown in the Campania Region of Italy

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ABSTRACT

Illegal practices of waste combustion and their burial in some land devoted to agricultural crops caused a severe economic crisis of the agriculture and food sector in the Campania region of Italy. To assess the levels of contamination by lead and cadmium, the only metals subject to European Union legislation, a system of monitoring of plant foods in the whole territory of the region has been promoted, with the goal of certifying productions and consumer protection. In fact, products that comply with European Union standards are assigned a Quick Response Code, which guarantees the traceability of the product (manufacturer and location). The code also ensures the safety of the product, as it allows the consumer to see the results of the analysis performed on the specific chain of production. The content of lead and cadmium was determined in 750 vegetable samples by using the atomic absorption spectrophotometry after microwave mineralization. These levels were below the maximum limits in all but three samples; two samples of tomatoes exceeded the maximum level of cadmium, and one sample of valerian contained an excess of lead.

A large area between the provinces of Naples and Caserta in the Campania region has recently caught the attention of the mass media, owing to the illegal dumping of waste—some of which is toxic—in fields or by the roadside. As this material is sometimes burnt, the area has been dubbed "Terra dei fuochi" (the land of fires). Understandably, the growing interest of the media has raised concerns that pollutants may contaminate not only the air but also foodstuffs of both plant and animal origin. The impact of this negative publicity on public opinion has led to a crisis in the agricultural and food sector in Campania, with serious consequences for the regional economy.

To provide support to local farms, the Campania region and the Istituto Zooprofilattico Sperimentale del Mezzogiorno have promoted a project for monitoring agricultural production in collaboration with the producers themselves. The project involves monitoring various environmental contaminants in foodstuffs, especially vegetables, to assess the degree of compliance with the maximum levels permitted. As part of this project, the present study ascertained the levels of lead and cadmium in various vegetables and compared these with values reported in literature, to determine the degree of risk to both human health and the environment.

The trace elements lead (Pb) and cadmium (Cd) are considered to belong to the group of nonessential, toxic metals occurring naturally in the environment and as pollutants released from industrial and agricultural sources.

An increasing number of studies have shown early adverse health effects at very low levels of cadmium exposure. Cadmium can damage the kidneys and may cause renal dysfunction *(16)*; moreover, the International Agency for Research on Cancer has classified cadmium as a human carcinogen (group I) *(13, 14)*.

Food is the main source of cadmium intake in the nonsmoking population *(15, 16)*. Indeed, the scientific report of the European Food Safety Authority on dietary exposure to cadmium in the European population *(11)* stated that the greatest impact was attributable to foods, particularly foods of vegetable origin (cereals, leafy vegetables, potatoes, and roots) that contain higher concentrations than meat, eggs, milk, dairy products, shellfish, and fish *(2, 22)*.

The presence of cadmium in foodstuffs is highly variable, depending, for instance, on the geographical location of the growing area. Indeed, cadmium occurs naturally in the Earth's crust, as part of geological materials (e.g., black shales) and as a consequence of volcanic eruptions and the exfoliation of rocks and minerals *(23)*. Other important factors are the uptake from the soil (different extent of transfer from soil to plants) and differences in patterns of accumulation by different plant varieties. For potatoes, the transfer of Cd from the root and its accumulation in the tuber could exceed food safety standards *(4)*.

Like cadmium, lead is a natural environmental contaminant. However, its former use in water pipes, paint, and petrol increased its general presence. Today, food is the main source of human exposure to lead, which accumulates in the body and most seriously affects the central nervous

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system and other organs in young children, increasing the risk of cancer *(14)*.

As already mentioned with cadmium, vegetables and vegetable products (8.4%) are among the main food categories contributing to lead exposure; in particular, potatoes and products derived from them account for 4% of dietary exposure *(10)*.

Contamination by heavy metals can reach plant products, particularly potatoes, through the use of contaminated irrigation water or sewage sludge, manure, or fertilizers made from phosphates *(5, 17, 25, 26)*. Moreover, the unlawful dumping of industrial waste is a further source. To minimize dietary exposure and safeguard public health *(8)* because foodstuffs are the main source of human intake, the European legislation has established maximum levels of cadmium and lead in various subgroups of vegetables *(6)*.

In the Campania region, the cultivation of potatoes (*Solanum tuberosum*) is one of the main sectors of agriculture, accounting for 22% of national production, while the cultivation of tomatoes (*Solanum lycopersicum*) accounts for about 8% of national production. Moreover, Campania, particularly Valle del Sele, is the chief producer of fresh-cut vegetables. Clearly then, in light of the recent media coverage of the land of fires, there is a need to ascertain the levels of contamination by cadmium and lead in vegetable products in Campania and to compare both of with the maximum permitted levels and with the reported levels in vegetables produced elsewhere in Italy and in other European countries.

In this article, we present the results of the determination of cadmium and lead in various vegetables grown in the Campania region of Italy and collected from the main areas considered to have the greatest risk of contamination.

MATERIALS AND METHODS

Research area. The area chosen for sampling covers most of the Campania region (Fig. 1). For some vegetable species, such as potatoes, the area includes fields located in those municipalities, which, according to the Ministerial Decree of 11 March 2014 fall within the zone known as the land of fires *(20)*. However, other areas in which the cultivation of these products is widespread were also sampled. For other types of products, such as fruit, salads, and tomatoes, areas located in the provinces of Naples and Salerno were examined.

Samples. All samples of vegetables analyzed in this study were grown in accordance with the typical agronomic practices on farms. From April to November 2014, during the harvest period, 750 samples of various kinds of vegetables were collected directly in the zones identified by means of a random sampling procedure. Each vegetable sample consisted of a number of subsamples taken within an area (100 by 100 m or less), with up to 1 kg being collected for each sample. The samples were handpicked by staff wearing vinyl gloves, carefully packed in clean polyethylene bags, and sent to the laboratory.

Before analysis, the samples were cleaned to remove any traces of soil; potatoes and fruits were peeled, as Regulation EC 1881/2006 stipulates that the maximum permitted level applies after washing and separation of the edible part *(6)*. Samples were then homogenized with a laboratory mill (Grindomix GM200 Retsch; Haan, Germany).

Reagents and solutions. All chemicals were of analytical reagent grade. High-purity water was produced by means of a Milli-Q Millipore deionizing system (Billerica, CA) for Graphite Furnace–Atomic Absorption Spectrophotometry (GF-AAS) determination and was used for the preparation of reference materials and sample solutions. Concentrated 67 to 69% nitric acid (HNO_3) of trace metal analysis grade and 30% hydrogen peroxide (H_2O_2) were purchased from Carlo Erba (Milan, Italy). Matrix modifiers, monobasic ammonium phosphate, and magnesium nitrate (1% Mg) were purchased from PerkinElmer (Waltham, MA). Standard solutions of lead and cadmium were prepared by dilution with 3% nitric acid of standard stock solutions at 1,000 mg/liter purchased from Merck (Darmstadt, Germany). The calibration curve was prepared by using the so-called bulk solution prepared by mixing the standard solutions and the subsequent dilution. Before use, glassware was washed with a solution of nitric acid (10%, wt/vol), rinsed with high-purity water, and dried in a desiccator sheltered from atmospheric dust.

Microwave digestion. To determine Cd and Pb concentrations, 0.5 ± 0.001 g of homogenized samples were weighed inside high-pressure Teflon vessels (Milestone, Inc., Shelton, CT); 4 ml of nitric acid (67 to 69%), 3.5 ml of oxygen peroxide (30%), and 2.5 ml of ultrapure water were then added. The vessels were placed in an Ethos E Milestone microwave oven (Milestone, Inc.), where the samples were digested under pressure for 10 min at 190°C for the analysis of trace elements. The operating conditions and the heating program used were set according to the manufacturer's recommendations. After acid digestion, the samples were cooled to room temperature and brought to 50 ml with ultrapure water in glass flasks and, finally, transferred to polypropylene vials for AAS analysis. Each sample solution was analyzed in duplicate.

Instrumentation. An atomic absorption spectrophotometer, with a transverse heated graphite furnace (GF-AAS) with Zeeman effect for electrothermal atomization and equipped with an automatic sampler, was used in this study (PinAAcle 900T, PerkinElmer). The assembly was operated from an interfaced computer running WinLab software (PerkinElmer). Hollow cathode lamps (PerkinElmer) were used as line sources for both elements. The operating conditions for GF-AAS require atomization at 1,500 and 1,600°C for Pb and Cd, respectively, after a heating phase (110 then 130°C), and pyrolysis at 850 and 500°C for Pb and Cd, respectively. The wavelengths were 283.31 nm for lead and 228.80 nm for cadmium. Argon was used as the inert gas, and internal flow was set at 250 ml/min.

Quality assurance. The analytical methods used to determine cadmium and lead in vegetables were accredited in accordance with UNI CEI EN ISO/IEC 17025:2005 ("General Requirements for the Competence of Testing and Calibration Laboratories"). Thus, appropriate quality assurance procedures and precautions were implemented to ensure the reliability of the results. The method used was validated by means of an in-house quality control procedure and through participation in an intralaboratory study.

For quantitative determination, standard solutions of each trace element were used to obtain calibration curves ranging from 0.078 to 5.0 μg/liter for cadmium and from 0.20 to 25 μg/liter for lead. All concentrations were expressed as milligram per kilogram of fresh weight. The correlation coefficients obtained for the calibration curves were all greater than 0.995. The limit of detection (LOD) and limit of quantification (LOQ) were calculated as the mean signal of five blanks plus 3 or 10 times the standard deviation, respectively. The LOQs obtained for Cd and Pb were

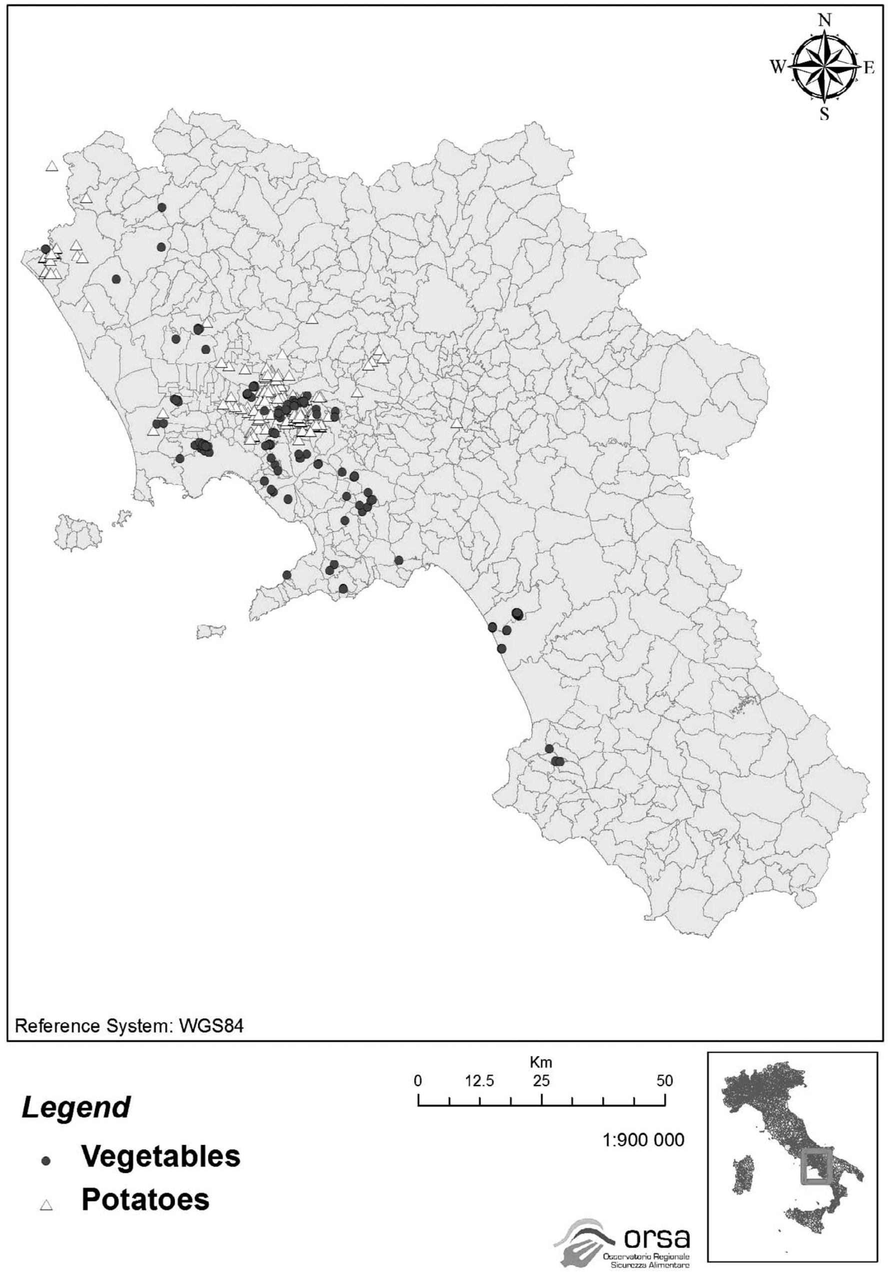

FIGURE 1. *Map of Campania showing sampling sites: white triangles indicate potato sampling; gray circles refer to all other vegetable species.*

0.010 and 0.020 mg/kg, respectively. For both Cd and Pb, the LODs and LOQs were in line with performance criteria for the methods of analysis for these elements established by the European Union *(7)*.

The accuracy of the method was validated by determining the elements recovered from the various samples spiked at different maximum residual levels set by European Union regulation. For this purpose, each spiked sample was prepared in quintuplicate: mean recoveries were 87.5% (at 0.050 mg/kg), 95.5% (at 0.100 mg/kg), 97.9% (at 0.200 mg/kg), and 99.2% (at 0.600 mg/kg) for lead and 86.0% (at 0.025 mg/kg), 81.0% (at 0.050 mg/kg), 97.8% (at 0.100 mg/kg), and 89.2% (at 0.300 mg/kg) for cadmium. Repeatability, expressed as relative standard deviation, 19% for Pb and 18.3% for Cd, was in accordance with the criteria established by the European Union regulation with corresponding HORRAT values less than 2 *(7)*.

All samples were analyzed in duplicate ($n = 2$). Chemical blank determinations were made regularly, together with each batch of samples, to check for possible contamination.

RESULTS AND DISCUSSION

In this study, 750 samples of vegetable products were analyzed for Cd and Pb. The numbers of samples of the various products are shown in Table 1, which also reports the mean, minimum, and maximum values of lead and cadmium detected.

The distribution of the samples analyzed corresponds to the distribution of the agricultural production in Campania. Thus, the highest percentage (49.8%) refers to potatoes, which mainly come from areas within the so-called land of fires (Fig. 1). This is an important consideration because some studies *(4)* have shown that an increase in cadmium and lead concentrations in soil determines an increase in their concentration in potatoes and that the contamination of potatoes, as well as constituting a risk for the health of consumers, can be regarded as an indicator of environmental pollution. In this regard, a study of potatoes grown in two

TABLE 1. *Average, minimum, and maximum concentrations of lead and cadmium in vegetables grown in the Campania region of Italy, subdivided by group*[a]

					Pb			Cd		
			No.	%	Mean	Minimum	Maximum	Mean	Minimum	Maximum
Leafy vegetables										
Cabbage	*Brassica oleracea*	*Brassicaceae*	9	1.3	0.031	<LOQ	0.062	0.006	<LOQ	0.016
Mizuna	*Brassica rapa*	*Brassicaceae*	5	0.7	0.047	<LOQ	0.076	0.012	<LOQ	0.033
Rocket	*Eruca vesicaria*	*Brassicaceae*	10	1.4	0.066	<LOQ	0.167	0.029	<LOQ	0.050
Tatsoi	*Brassica narinosa*	*Brassicaceae*	1	0.1	0.060	—[b]	—	0.011	—	—
Chard	*Beta vulgaris*	*Chenopodiaceae*	4	0.6	0.031	0.020	0.041	0.026	0.016	0.039
Spinach	*Spinacia oleracea*	*Chenopodiaceae*	8	1.1	0.045	<LOQ	0.120	0.064	<LOQ	0.184
Endive	*Cichorium endivia*	*Compositae*	11	1.6	0.059	0.040	0.142	0.012	<LOQ	0.030
Lettuce	*Lactuca sativa*	*Compositae*	28	4.0	0.069	0.020	0.234	0.043	<LOQ	0.177
Valerian	*Valerianella locusta*	*Valerianaceae*	2	0.3	0.295	0.040	0.555	0.005	<LOQ	<LOQ
Other vegetables										
Vegetable marrow	*Cucurbita pepo*	*Cucurbitaceae*	8	1.1	0.045	<LOQ	0.091	0.005	<LOQ	<LOQ
Pumpkin	*Cucurbita moschata*	*Cucurbitaceae*	2	0.3	0.018	<LOQ	0.026	0.005	<LOQ	<LOQ
Cucumbers	*Cucumis sativus*	*Cucurbitaceae*	3	0.4	0.020	<LOQ	0.039	0.005	<LOQ	<LOQ
Beans	*Phaseolus vulgaris*	*Leguminosae*	8	1.1	0.034	<LOQ	0.071	0.006	<LOQ	0.010
Potatoes	*Solanum tuberosum*	*Solanaceae*	350	49.8	0.020	<LOQ	0.092	0.008	<LOQ	0.040
Tomato	*Solanum lycopersicum*	*Solanaceae*	125	17.8	0.030	<LOQ	0.090	0.009	<LOQ	0.077
Pepper	*Capsicum spp*	*Solanaceae*	9	1.3	0.031	<LOQ	0.076	0.015	<LOQ	0.033
Eggplant	*Solanum melongena*	*Solanaceae*	8	1.1	0.035	<LOQ	0.068	0.011	<LOQ	0.030
Herbs/bulbs										
Garlic	*Allium sativum*	*Amaryllidaceae*	1	0.1	0.010	—	—	0.005	—	—
Onion	*Allium cepa*	*Amaryllidaceae*	2	0.3	0.029	0.027	0.030	0.005	—	—
Parsley	*Petroselinum crispum*	*Apiaceae*	3	0.4	0.042	<LOQ	0.092	0.007	<LOQ	0.010
Celery	*Apium graveolens*	*Apiaceae*	2	0.3	0.044	0.041	0.046	0.028	0.011	0.044
Fennel	*Foeniculum vulgare*	*Apiaceae*	15	2.1	0.037	<LOQ	0.062	0.006	<LOQ	<LOQ
Basil	*Ocimum basilicum*	*Lamiaceae*	1	0.1	0.010	—	—	0.005	—	—
Fruit										
Kiwi	*Actinidia deliciosa*	*Actinidiaceae*	1	0.1	0.028	—	—	0.005	—	—
Hazelnut	*Corylus avellana*	*Betulaceae*	1	0.1	0.010	—	—	0.005	—	—
Melon	*Cucumis melo*	*Cucurbitaceae*	8	1.1	0.024	<LOQ	0.069	0.005	<LOQ	<LOQ
Lotus	*Diospyros kaki*	*Ebenaceae*	11	1.6	0.049	<LOQ	0.069	0.005	<LOQ	<LOQ
Walnut	*Juglans regia*	*Juglandaceae*	2	0.3	0.010	<LOQ	<LOQ	0.005	<LOQ	<LOQ
Fig	*Ficus carica*	*Moraceae*	3	0.4	0.027	0.020	0.031	0.005	<LOQ	<LOQ
Olive	*Olea europea*	*Oleaceae*	4	0.6	0.014	<LOQ	0.024	0.005	<LOQ	<LOQ
Apricot	*Prunus armeniaca*	*Rosaceae*	13	1.8	0.034	<LOQ	0.060	0.006	<LOQ	0.018
Cherry	*Prunus avium*	*Rosaceae*	6	0.9	0.025	<LOQ	0.031	0.005	<LOQ	<LOQ
Strawberry	*Fragaria spp*	*Rosaceae*	11	1.6	0.036	<LOQ	0.123	0.005	<LOQ	<LOQ
Apple	*Malus domestica*	*Rosaceae*	20	2.8	0.045	<LOQ	0.092	0.008	<LOQ	0.03
Pear	*Pyrus communis*	*Rosaceae*	1	0.1	0.052	—	—	0.005	—	—
Peach	*Prunus persica*	*Rosaceae*	17	2.4	0.033	<LOQ	0.074	0.006	<LOQ	0.016
Plum	*Prunus domestica*	*Rosaceae*	13	1.8	0.042	<LOQ	0.073	0.005	<LOQ	<LOQ
Orange	*Citrus sinensis*	*Rutaceae*	2	0.3	0.032	0.026	0.037	0.005	<LOQ	<LOQ
Lemon	*Citrus limon*	*Rutaceae*	3	0.4	0.015	<LOQ	0.022	0.01	<LOQ	0.02
Grapes	*Vitis vinifera*	*Vitaceae*	19	2.7	0.052	<LOQ	0.105	0.005	<LOQ	0.013
Total			750							

[a] Values are milligram per kilogram of fresh weight.
[b] —, one measured value.

areas of Slovakia revealed cadmium and lead contamination several times higher (0.10 to 0.357 mg/kg for Cd and 0.318 to 0.483 mg/kg for Pb) than the limit value *(21)*. One of these sites was contaminated by an anthropogenic source (mining and processing of minerals), while the other suffered from natural contamination from metal-bearing rocks.

In Campania, most of the land has moderate baseline Cd values, which can be assumed to represent the natural background for siliclastic and volcanoclastic deposits throughout the region. A close relationship between phosphate fertilizers and increased baseline Cd values cannot be excluded in coastal areas, where intensive farming is practiced *(1)*.

For lead, the highest baseline values are found in stream sediments in highly urbanized areas of the Campania region. The rest of the territory, where the anthropogenic impact is less marked, displays lower baseline Pb values, which represent the natural, geogenic background for siliclastic, alluvial, and volcanoclastic deposits.

Despite the greater presence of cadmium in the soil, because of the previously mentioned factors, the results of the present study revealed a low level of contamination of potatoes. Indeed, in 274 samples of potatoes (78% of the total), cadmium levels were below the quantification limit (<0.010 mg/kg); if we assume that the cadmium level in these samples is equal to half of the LOQ *(19),* the mean value becomes 0.005 mg/kg, which is well below the maximum level permitted. Likewise, contamination by lead also proved to be very low, falling below the LOQ (<0.020 mg/kg) in 62% of cases (mean value, 0.020 mg/kg).

A similar consideration can be made for tomatoes, which accounted for 17.8% of the total number of samples analyzed. In this case, lead contamination proved to be below the LOQ (<0.020 mg/kg) in 54 of 125 samples analyzed (mean level, 0.030 mg/kg). Contamination by cadmium generally proved to be negligible, with the mean value being 0.009 mg/kg; in two samples, however, levels of 0.064 and 0.077 mg/kg were recorded, which exceed, albeit slightly, the legal limit (0.050 mg/kg).

The levels of lead and cadmium detected in samples of fruit and leafy vegetables appear to be of interest. For fruits, it seems appropriate to group together all the samples of apricots, peaches, plums, cherries, strawberries, pears, and apples, because of both the small number of samples of each fruit available and because all of these fruits belong to the same family (*Rosaceae*). Taken together, these products accounted for 60% of the fruit samples analyzed. Our analyses did not reveal any contamination by the two metals considered; their concentrations were well below the maximum permitted values, with the mean value of lead being 0.041 mg/kg (mean values ranging from 0.025 mg/kg in cherries to 0.054 mg/kg in strawberries). In this case, note that the upper limits vary from 0.10 mg/kg for fruit to 0.20 mg/kg for small fruits (strawberries and cherries). Contamination by cadmium was completely absent in these products, with values below the LOQ in almost every sample.

The same applies to the other types of fruit analyzed; all 19 samples of grapes (2.7% of all samples) proved acceptable, displaying a mean value of 0.052 mg/kg of lead, while in lotus fruits ($n = 11$) and melons ($n = 8$), the mean concentrations were 0.049 and 0.024 mg/kg, respectively. In these products, cadmium levels also proved to be below the LOQ.

For leafy vegetables, on which European Union legislation has imposed higher maximum values, only one case of irregularity was recorded; this involved a sample of valerian (*Valerianella locusta*) containing 0.55 mg/kg of lead. In all the samples, which were chiefly made up of species belonging to the family of *Compositae* (lettuce: 28 samples; endives: 11 samples) and rocket ($n = 10$)—species that constitute the fresh-cut vegetables of which Campania is among the main producers in Europe—we found no excessive levels nor high mean values (0.059 mg/kg for endives and 0.031 and 0.045 mg/kg for chard and spinach, respectively). The mean values of cadmium were equally low, with spinach showing the highest level (0.064 mg/kg). Moreover, levels of both metals below the LOQ were observed in many samples, a finding that confirms the low level of environmental contamination in the area of production situated in the Sele Plain in the province of Salerno.

Finally, the concentrations of cadmium and lead in our samples were compared with the available Italian and European literature data on similar products harvested in the field or sampled in the marketplace. Few data are available in Italy, where one study reported mean concentrations of cadmium of 0.022 mg/kg in the limited number ($n = 5$) of potato samples analyzed and cadmium values, ranging from 0.01 to 0.031 mg/kg in other vegetable products (fruit and vegetables in general) *(24)*. Moreover, an interesting comparison can be made with a study carried out on tomatoes grown in an area at high risk of pollution. While the authors found very high levels of lead (0.127 mg/kg), cadmium concentrations (0.030 mg/kg) never exceeded the maximum levels permitted *(12)*. The high levels of lead contamination in these samples were very probably due to the proximity of industrial facilities and their waste disposal plants, which polluted the soil, air, and water, thereby contaminating vegetable products grown in the area. The results of our study revealed markedly lower levels of lead and cadmium contamination than both those reported in this industrial area of Sicily *(12)* and those detected in vegetables harvested in an industrial area of Sardinia *(2)*.

For vegetables sampled in the marketplace, the mean values of cadmium and lead detected in our study were similar to those found in certified organic potatoes available on the Greek market (8.6 ± 2.7 and 11.8 ± 2.8 ng/g, respectively), while the cadmium and lead content of noncertified potatoes sold or advertised as ''organic'' on the Greek market were 60.5 ± 19.8 and 15.6 ± 2.7 ng/g, respectively *(17)*. Similar cadmium values were also detected in potatoes from the Serbian (0.009 mg/kg) and Swedish (0.013 mg/kg) markets, while lead levels quantified in both countries were very low (0.003 and 0.02 mg/kg, respectively) *(3, 27)*. Finally, in a recent study carried out on potatoes harvested in the Canary Islands, cadmium concentrations ranged from 0.006 to 0.019 mg/kg and lead concentrations from 0.007 to 0.023 mg/kg *(18)*; in this case, too, the results excluded critical contamination by cadmium.

The results of the present study show that fruit and vegetable products grown in Campania display very low levels of contamination by cadmium and lead; indeed, only two samples of tomatoes and one of valerian exceeded the limits imposed by European legislation.

These results also demonstrate that proper farming practices, which avoid the use of fertilizers that may be at risk of cadmium and lead contamination, can ensure that the concentrations of these metals in vegetable products will be very low. The results also allow us to exclude the effect of important environmental factors, such as the abundance of these elements in the soil (due to the geological features of

FIGURE 2. *Quick Response Code assigned to a producer.*

the area) or pollution of soil, air, and irrigation water resulting from the illegal disposal of industrial wastes.

The application of Quick Response Code technology enabled us to record all the results pertaining to each individual agricultural enterprise participating in the study within a specific code assigned to each organization (Fig. 2). The result of this is a system of traceability that ensures consumer safety and purchasing transparency. Indeed, only products from a chain of production that has been certified can be released onto the market, and the accompanying Quick Response Code provides complete information (producer, geographic location of cultivation, and a report of the tests carried out on the products of that specific chain). Any products not conforming to the prescribed standards are prevented from being marketed.

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