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Surveillance model for environmental contaminants through their monitoring in animal production

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Abstract

Background: The aim of this paper is to propose a risk based surveillance model for environmental pollutants, monitoring the presence and the quantity of toxic contaminants in cattle and sheep products.

Methods: The province of Latina was chosen as study area. The official list of polluted sites was provided by the Province. This was then integrated into a Geographic Informative System together with geographical, hydrographic, and geological data along with information regarding the farms that was obtained from the National Livestock Registry. In order to identify the monitoring priority of polluted sites, a semi-quantitative risk assessment was then set up. A score for each of the attributes considered in the risk evaluation was given to each site, based on two main criteria: the environmental hazard characterization and the quantification of the exposed animal population. The sample of farms to be monitored was defined according to statistical criteria.

Results: 12 polluted sites were chosen as "at major risk" among the 58 identified therefore, inside the respective surrounding risk areas, 24 farms were chosen to be monitored. Biological materials to be sampled were also defined. As a result, it was determined that 104 samples collected throughout a year would be sufficient to detect any contamination in the territory of the Province, with an admitted prevalence of 10% among farms at risk (CL 95%).

Conclusions: Risk based surveillance is more sensitive in detecting environmental pollution than surveillance that is randomly performed, based on the same allocation of financial resources.

Key words: risk assessment, environmental monitoring, farm animals, Geographic Information System

Introduction

The advantages of using animal sentinels, mostly invertebrates, to monitor environmental pollution have been known for a long time. In a review, Beeby [1] explains which criteria should be adopted to select sentinel species, and proposes a procedure to validate such a model.

The opportunity of using farm animals as bio-indicators for the detection of toxic substances in the environment has been suggested and partially tested by several authors [2-6]. The rationale for such use is based primarily on the accumulation mechanism of lipophilic substances, such as organochlorine pesticides and dioxins, in tissues or organs of some mammals, as well as on the natural role of "accumulator" that animals may play in the environmental cycle of several toxic molecules.

Furthermore, daily products, such as milk or eggs, together with a relative short life cycle of the animals and simultaneous availability of tested circuits for the collection and submission of biological samples, encourages the use of farm animals as sentinels for environmental pollution by providing information in a more efficient, economical and less intrusive way than detection of bio-indicators directly in the human population.

Such use, however, is not free from methodological difficulties, consisting mainly in lack of known exposure markers, lack of standardization in study design and limited availability and sharing of basic information [7].

In a review O'Brien and coll. [3] highlighted the significance of animals in epidemiological studies and have described in detail their role as "indicators" rather than "monitors" or "sentinels".

In Italy, for many years, there are two surveillance plans (PNR and PNAA), derived from

European legislation (EC Directive 96/23 [8] and Regulation EC 178/2002 [9]] that regulate the official control of food of animal origin and animal feeds, with respect to various chemical residues, including environmental contaminants.

The major criticism of the PNR plan is its progressive loss of overall sensitivity. The number and type of samples for each substance to be identified are defined annually by the Ministry of Health on the basis of directives which indicate the percentage of products to be checked for various domestic class of production (Dlgvo 158/2006 [10] and EC Decision 97/747 [11]]. Once the national sample size for each molecule and for each type of product is established, the sample is divided among individual regions, and then, using a cascade mechanism, it is further sub divided among district territories by the Local Health Authorities. This fragmentation of the samples along the distribution chain while still being able to provide information at the national level, does not allow for any significant conclusions to be drawn at lower levels.

Even if the choice of animals to be sampled has to be based on individual risk, there is no mention in the Plan of specific risks relating to geographical areas, which can be the cause of emergences being detected sometimes by serendipity [12].

Presence of chemical contaminant residues in animal products is often evidence of improper, if not clearly illegal, waste management. The issue of contaminated animal products, often ignored or underestimated by farmers, can be found wherever there is, in the same territory, inappropriate management of municipal waste (e.g. burning under uncontrolled conditions), or industrial waste (e.g. burying toxic waste) and production of animal feeds or food. The example of Beta-hexachloro-cyclohexane in the provinces of Rome and Frosinone [13] and the most recent dioxin emergency in some pig farms in Ireland [14], are a witness to this.

For these reasons and also to avoid possible effects of "dilution" due to a random sampling (or convenience ones) in geographic area, and make the best use of available resources, some Authors suggest the use of monitoring systems based on "risk" [15] to achieve better results in terms of cost-effectiveness. These systems have, for the same input data, a higher sensitivity and higher positive predictive value, under a specific prevalence of real contamination.

An effective way in managing all of the basic information to produce a risk analysis is a Geographic Information System (GIS), a tool capable of integrating geographical, geological and hydrographic features, together with the layers representing industrial and animal husbandry activities.

From available information a system of "classification" for the monitoring of polluted sites can be set through the attribution of scores for each variable taken into consideration. This can then be used for the subsequent sampling of farms within the vicinity of the selected sites.

The present works aims to:

- propose the use of farm animals in order to detect quantitative changes over time of contaminants in animal products, before they can cause significant changes firstly in the health status of the animal population exposed and secondly to the human population to which those products are destined as food;
- design a model for monitoring the occurrence and spread of toxic industrial environmental contaminants, through a sampling system of animal products based on risk. For this purpose, the presence of the contaminant in animal products will be used as a bio-indicator of environmental pollution (active surveillance), regardless of whether the law limits have been exceeded or not.

Methods

Study area

A single province, in the Region of Lazio, was chosen as the planned territory for the study, in order to work with homogeneous administrative and regulatory policies for both veterinary and environmental matters. Furthermore, the extension of the province study area was defined with an intention to include different geographical and hydrological characteristics along with the potential presence of different environmental health hazards.

In particular, the province of Latina was chosen due to the simultaneous presence of closed down industries, still operating factories, landfill sites, intensive cattle and extensive buffalo and sheep farms.

The Area is approximately 2250 km^2 wide, with a mostly flat country from the coast up to the mountain chain of Lepini-Ausoni-Aurunci, located along eastern and southern boundary, where the land becomes hilly and mountainous. The resident human population is approximately 540 thousand inhabitants, and the estimated number of cattle and sheep are, respectively, 54,261 and 28,442 [16].

Data sources

The shapefiles containing data on topography, lithology, hydrography and hydrogeology and the land uses of the area, were obtained from the SIRA (Regional Environment Information System). The following maps and linked data were selected afterwards and then included in the analysis:

- Hydrogeological map of Lazio scale 1:250:000 [17]
- Map of hydrographical features [18]
- DEM (Digital Elevation Model) [19]
- Map of administrative boundaries (20]

Geographical data (*WGS84 format of point locations*) concerning the officially censed polluted sites (ex DM 471/99) [21] were provided in shape-file format by the Ecology and Environment Section of the Province of Latina.

For each site, the attributes provided by the same bureau were: the site typology (legal landfill, illegal landfill, industrial plant) and, when available, the class of molecules detected in the surrounding environment. From the same files it was possible to derive the status of each site with respect to the step of implementation of the mandatory reclaim plan. Animal counts were established using the National Livestock Data Registry (https://anages.izs.it/home.html), the official registry of farms and animals, census data and the geographical location of cattle, buffalo, sheep and goat farms. When the number of heads were not available, it has been provided by the Veterinary Office of the Local Health Units, on request.

Molecules selection

Taking into account the type of pollutants in the examined area, the knowledge of the physiology and production of sheep and cattle, the available data on contaminants spread in soil and forages, as well as the actual availability of performing validated tests in the Region, the following molecules were proposed for monitoring: PCB, PCB-dioxin like (DL-PCBs), dioxins (PCDDs and PCDFs) and heavy metals. Moreover, these molecules are the contaminants of industrial origin that are scheduled in the Residues National Plan (PNR). It was not possible to make a specific selection of molecules for each site, due to the presence of mixed and uncertain waste in most of them, regardless they were legal or illegal wasting landfills.

Although polycyclic aromatic hydrocarbons (PAHs) constitute a frequent contaminant in the area (according to the original list), they were excluded from this study - because their effect on animal metabolism is not completely known. Therefore, about twenty sites, consisting primarily of deposits of fuel or filling stations, were excluded from the final database.

Spatial analysis

All geographical and environmental information and animal husbandry data were integrated into a GIS, using ARCGIS® 9.2.

Assuming that the concentration of an hypothetic contaminant tends to decrease gradually in the soil at progressively greater distances from the source of contamination, it was decided to use circular risk areas surrounding the sites. It was determined that the radius of the circle would be 3 km. This distance is within the range (0-5 km) used by several Authors as a limit distance for exposure in studies designed to investigate for any effects of pollution on human population health. In fact, detection of increased risk inside the range of 3 km from polluted site was demonstrated for congenital malformations [22]; while other studies aimed to measure the effect on incidence of cancer [23] or low birth weight [24] identified lower distances, respectively, of 2 km and 1 km from polluted sites, delimiting areas of increased risk.

In this model the limit of 3 km from polluted site was chosen both to maintain a conservative hypothesis, and to be able to detect the presence of contaminant traces in animal tissue, possibly earlier than any detectable effect on animal and human health would became evident.

Within the area of 3 km radius from each site (buffer), geological and hydrographic features, and number of farms and heads for each one of the following species were examined: buffalo, cattle, sheep and goats.

Exposure assessment and hazard categorization (ranking)

A semi-quantitative analysis of the risk was performed. The first step (Hazard identification) [25] consisted in the acquisition of all of the available information regarding sources of contamination identified in the territory of the province. Then a characterization of potential risk or "Hazard ranking" was carried out in the territories surrounding the individual sites, taking into account available parameters and assigning them a score.

This ranking method may be considered a simplification of proposed methods for prioritization of polluted site respect to the reclamation process (A.R.G.I.A) [26].

Two main criteria were adopted in categorization of contaminated sites: environmental risk (as assessment of possible routes of exposure) and quantification of potentially exposed animals.

Regarding the first criterion, 4 attributes were

Table 1. Environmental risk characterization.

Risk attributes	Category	Score
Type of soil	Sandstone	3
(A: "soil")	Limestone	2
	Alluvial sediments, debris and deposits	1
	Clayey deposits	0
Proximity to a water course or water basin	Within a radius of 500 m	1
(B: "Water")	Distance of any water more than 500 m	0
Depth of the ground-water	At surface level (0 m asl)	3
(C: "Aquifer")	Medium deep (0 - 21 m asl)	2
	Deep (between -21 and -100 m asl)	1
	Very deep (between - 101 and -1438 asl)	0
Status of implementation of the reclaim process	Hazard identification	4
(D: "Reclaim")	Risk characterization	3
	Reclamation project	2
	Procedure completed	1

Table 2. Quantification of exposed animals.

Animal population attributes	Category	Score
Number of cattle/buffaloes in a buffer of 3 km	> 200 heads	3
(E: "cattle")	51-200	2
	1-50	1
	0	0
Number of sheep/goats in a buffer of 3 km	> 1000 heads	4
(F:"sheep")	501-1000	3
	151-500	2
	1-150	1
	0	0

evaluated (see Table 1):
A.Type of soil;
B.Proximity to water course or water basin;
C.Depth of the ground-water;
D.Status of implementation of the reclaim process

Regarding the second criterion, two attributes were evaluated (see Table 2):
E.Number of cattle/buffaloes in a buffer of 3 km
F.Number of sheep/goats in a buffer of 3 km

Data were archived in a matrix shaped dataset, in which the polluted sites were identified as the records and each of the attributes related to risk and exposure was then represented in a field.

A score number was assigned for each attribute, and the sum of the scores was calculated for each of the two major classes of attributes (A + B + C + D and E + F). A set of 58 records was obtained using this procedure, identified with numbers from 1 to 61, each of them characterized by a risk score (ToT_Risk) and a score for the quantification of the animal population exposed (ToT_Expos).

Sampling criteria

The sampling was planned in two steps: firstly the sampling of polluted sites to be monitored, and secondly sampling of farms in the risk area around the sites. For the first aim, the median and quartiles of risk score were calculated (Median = 6, third quartile = 7). All sites in which the risk score was equal or greater than 7 were selected in first instance. Among the sites at high risk a further selection was made based on animal population exposed; sites with relative score ≥ 3 have been selected. This choice was made to give priority, for the purposes of environmental monitoring, to the sites where animal population is greater, thus giving a major probability of detecting traces of contaminants in animal products.

Once the sites were selected, the number of farms to be monitored was established in order to find at least one positive, if the prevalence exceeds the expected level. For this purpose, the total number of farms in areas at risk was used as the target population, assuming a maximum admitted prevalence of 10% and a confidence level of 95%.

The obtained number, for cattle and sheep farms, was stratified among the areas at risk. Farms with fewer than 10 animals were excluded from sampling. With regard to cattle herds, those with a predominant milk production were chosen, as they were considered to be more stable in their productive life, and related more to the local environment. In addition, the milk matrix is more suitable to monitor the presence of contaminants either hydrophilic or lipophilic, and finally sampling of mass quantities of milk is easy and not expensive In the absence of this type of farming, preference was given to herds that had a double purpose (milk and meat production). A quarterly sampling frequency was proposed in order to check, at least once a year, the total number of animals in production, considering the dry period of two months.

With regard to sheep herds, it was proposed, in addition to the quarterly sampling of mass milk during the production season (mainly from October to June in Lazio), to take samples of the sheep kidneys and fat, to be carried out at the slaughterhouse.

With regard to the animal population on which carrying out the analysis, older animals were chosen as it was supposed that they would have been exposed for a longer period of time. The organs selected were those where the accumulation of heavy metals and organochlorine compounds is known to be greater.

The sample was calculated according to the following criteria:

a. population reference = adult heads;
b. percentage of casting = 15%;
c. prevalence maximum admitted = 20%;
d. confidence level = 95%.

The obtained number of sheep to be sampled was then distributed proportionally amongst each of the farms involved.

Results

The polluted sites identified in the province of Latina (No. 58), have been plotted on the map and are shown in Figure 1.

Among the sites with a risk score (first criterion) greater or equal to 7 are 18, a further selection was made for potentially exposed animal populations that had a risk score (second criterion) ≥ 3. This led to the identification of 12 sites (Table 3)

From the complete database, initially, the total number of farms inside the buffer of 3 km, across the 12 sites chosen, was 138. Many farms appeared several times in the list, due to the overlap of some of the buffers (in fact many of the contaminated sites were concentrated in a small area), therefore a final number of 75 herds was obtained by removing those that appeared more than once.

The 12 areas at high environmental risk, encompassing 75 farms are shown in Figure 2.

According to the criteria mentioned in the previous paragraph, 24 farms were selected (32% of those at high risk) for monitoring.

Selected sites and their farms are reported in Table 4. Also in the same table the number of samples of mass milk and tissue samples from sheep and goats to be carried out at the slaughterhouse are reported. By considering the samples of mass milk collected quarterly, and making an estimation of slaughtered old animals among sheep and goats of 98, the global planned activities for bio-monitoring on an annual period is, globally, 104 samples: 91 of milk and 13 of kidney/fat.

In respect of privacy, each site was identified only by a code and each farm by an alphanumeric code indicating animal species and the sequence number in the Database (e.g. Bov1, O7, etc.).

Table 3. Scores for risk and exposed animals of high risk area (on grey ground selected sites).

Site code	Type	A Soil	B Water	C Aquifer	D Reclaim	Total score environmental risk characterization	E Cattle	F Sheep	Total score exposed animals quantification
1	Landfill	3	1	2	2	8	1	0	1
7	Illegal landfill	3	1	1	2	7	1	4	5
8	Illegal landfill	1	1	2	3	7	1	4	5
10	Polluted site	3	1	1	2	7	0	4	4
14	Illegal landfill	3	1	1	2	7	3	1	4
15	Polluted site	1	1	3	2	7	2	2	4
18	Polluted site	3	1	2	2	8	3	0	3
19	Polluted site	3	1	2	2	8	1	0	1
30	Illegal landfill	3	1	2	3	9	2	0	2
31	Illegal landfill	3	1	2	3	9	3	1	4
36	Illegal landfill	0	1	3	3	7	3	1	4
41	Illegal landfill	2	1	1	3	7	1	1	2
42	Illegal landfill	2	1	2	3	8	1	1	2
44	Illegal landfill	1	1	3	3	8	3	0	3
45	Polluted site	1	1	3	3	8	3	0	3
46	Illegal landfill	1	1	2	3	7	3	0	3
59	Illegal landfill	1	1	3	3	8	1	1	2
60	Polluted site	1	1	3	3	8	3	1	4

Figure 1. Location of polluted sites.

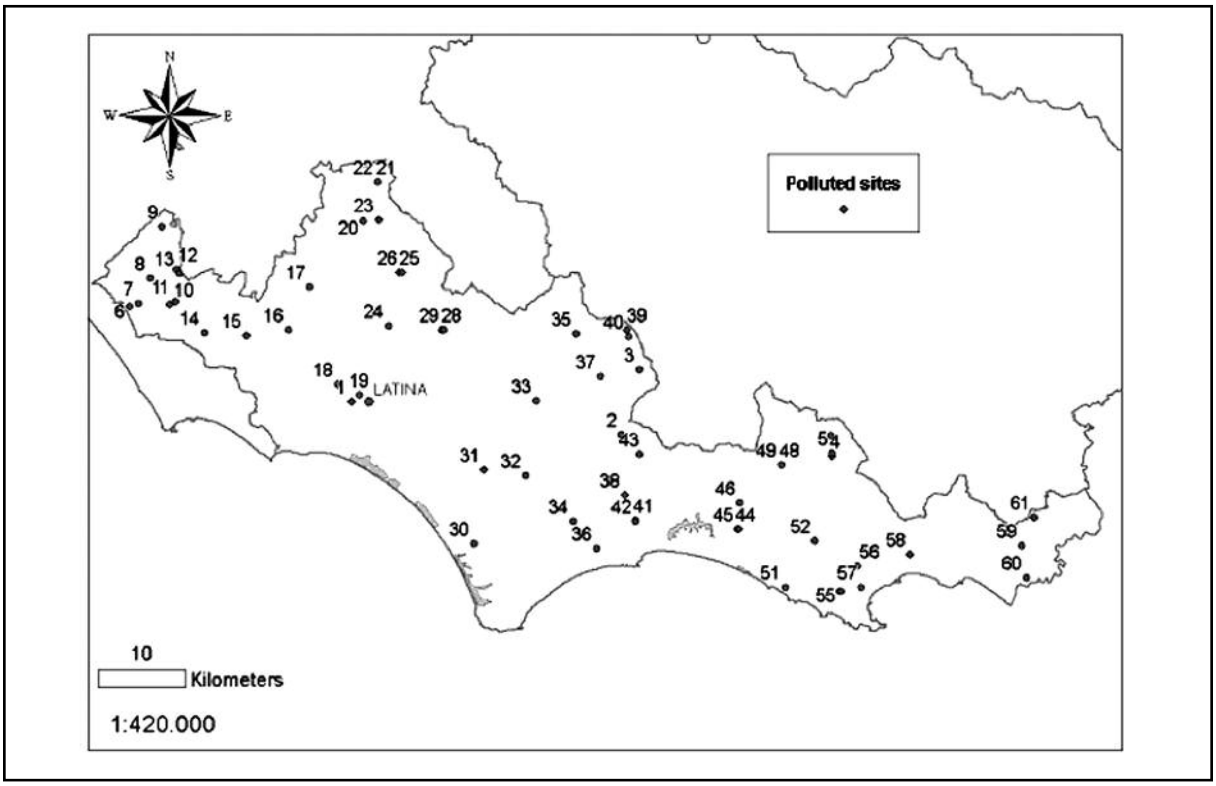

Figure 2. Location of highest risk contaminated sites, surrounded by the 3 km radius risk area with included farms.

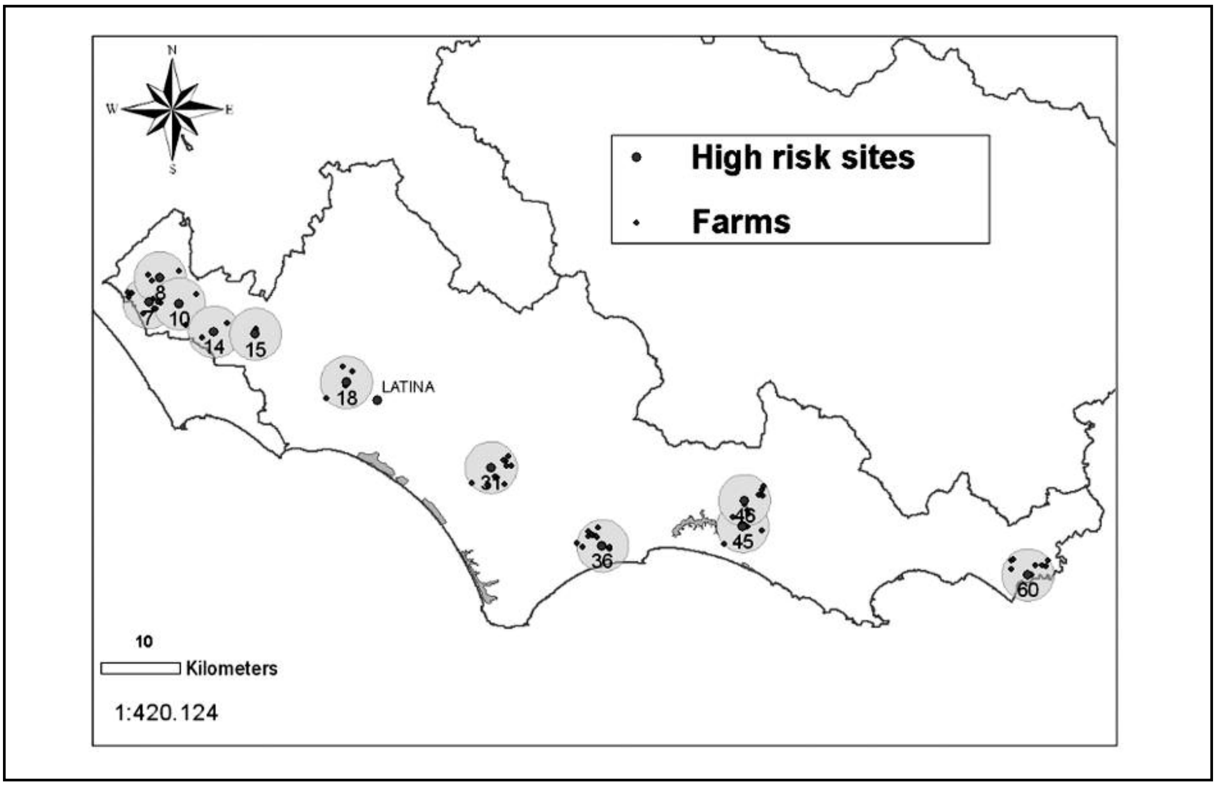

Table 4. List of farms to be monitored and summary of sampling activities on an annual period.

Site code	Farm code	Municipality	Number of animals	Number of milk samples	Casting proportion (est.)	Number of fat/kidney samples
7	O8	APRILIA	245	3	37	5
8	O12	APRILIA	219	3	33	4
10	O14	APRILIA	160	3	24	3
14	Bov4	APRILIA	36	4	NA	-
15	Bov2	APRILIA	60	4	NA	-
18	Bov43	LATINA	183	4	NA	-
18	Bov44	LATINA	78	4	NA	-
31	Bov50	PONTINIA	132	4	NA	-
31	Bov57	PONTINIA	54	4	NA	-
31	Buf75	SABAUDIA	542	4	NA	-
31	C79	SABAUDIA	11	3	2	0
36	Buf92	TERRACINA	30	4	NA	-
36	Bov96	TERRACINA	71	4	NA	-
36	Bov97	TERRACINA	76	4	NA	-
36	Bov98	TERRACINA	37	4	NA	-
36	Bov100	TERRACINA	152	4	NA	-
45	Buf23	FONDI	756	4	NA	-
45	Bov27	FONDI	35	4	NA	-
45	Bov32	FONDI	91	4	NA	-
45	Buf35	FONDI	20	4	NA	-
46	Bov25	FONDI	76	4	NA	-
46	Buf30	FONDI	57	4	NA	-
60	C48	MINTURNO	21	3	3	1
60	Bov86	SANTI COSMA E DAMIANO	10	4	NA	-
Tot.				**91**		**13**

Legend: O(n°) = code for sheep farm; Bov(n°) = code for cattle farm; Buf(n°)= code for buffalo farm; C(n°) = code form goat farm. NA: Not applicable

Note: Site 44 is not represented in table since its buffer and included farms completely overlap with those of site 45.

Discussion

The present work is an application of the linkage of data originating from environmental and health agencies, which have resulted in a unique dataset, that is more informative than the original ones.

In recent years, a more active cooperation between actors in the fields of public health and the environment has been widely promoted by international institutions, in order to unanimously reach the final goal of "one health" [27].

The proposed sampling program may be a first step for subsequent environmental-health surveillance programs, as it is able to identify situations of concern which would require specific control actions.

Among the contaminated sites several landfills were included, those that were legal landfills were probably included as they were ascertained to cause contamination to the surrounding area.

These sites may are sometimes sources of metals and PCBs, especially the older generation plants, built before promulgation of the current more protective rules for the environment [28].

The approach used in this work was a semi-quantitative risk assessment, with a scoring system applied to a grid of predetermined variables.

Emilia Romagna Region recently proposed a similar system in concept (ARGIA) [26], but much more complex for number of parameters considered and methods of calculation, aimed to evaluate the priority of clean-up operations on polluted sites.

One of the main differences between methodology used in this work and ARGIA method is the parameterisation of polluted site features (extension, volume, site vulnerability, etc.) which has been not considered in this model due to a lack of specific information. The "receptors", instead, as well as in ARGIA method, were included, being in this case animal heads.

Once again is confirmed the importance of keeping an official, complete and updated database for animal herds, including geocoding data, number of animals and typology of production.

Despite the progress achieved with the bovine Registry, in Italy the implementation of a similar Registry for other species such as sheep and goats has been delayed considerably in comparison to European standards.

The active involvement of farmers is crucial in the adoption of such surveillance programmes: they should be informed of the purposes of bio-monitoring, quite different from those of legal inspection, which always generates fear and mistrust. As a matter of general policy, farmers should be guaranteed that in case of positive detection of contaminants, their production would not be subjected to restrictive measures in marketing, except in cases of serious public health concerns.

The matrixes to be sampled suggested in this work are just indicative. For example, in case the sampling of organs at the slaughterhouse should be retained as too expensive, the analysis for metals could be carried out on samples of wool taken from live animals [29].

The entire proposed surveillance system may be improved and /or adjusted for specific situations through simple modification of the criteria evaluated and the scoring grid elaborated, depending on of the availability of information. Moreover, the occurrence of sudden environmental alarm may be the justification for a suited specific sampling programme. Other animal species or products may be used (honey, aqua-cultured fish, snails for example) [30, 31] or other contaminants could be searched in the more appropriate biological matrixes.

When the use of animals for monitoring environmental pollutants is not risk based, it is inevitably affected by a dilution factor and becomes less sensitive. For this reason, the proposed sampling protocol is totally different from that used by other Authors in random investigations [2, 32] and from the current National Surveillance Plan (PNR). In the latter, the number of samples scheduled for the same area per year, is about ten. This sampling programme, giving 104 samples (91 milk samples and 13 fat/kidney samples), is certainly more expensive for the community, but it is also more sensitive in detecting contaminants in animal products. Irrespective of this, it is possible to calibrate the method in terms of the available budget, acting on the frequency and quantity of samples, while maintaining a focus on specific risk situations.

Furthermore, the described sampling system could be adopted by Local Authorities or Farming Associations, integrating into the national monitoring plan, in order to promote and guarantee local production, particularly during the current climate of media-driven alarm, as recently witnessed in the Campania Region for buffalo "mozzarella" [33].

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