Chapter

Metabolic Impairments Caused by Pesticides in Mammals and Their Interactions with Other Pollutants

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Abstract

The biological systems are exposed to a complex environment in which the contaminants can interact in a synergistic/antagonistic fashion and for this reason, the study of "chemical cocktails" is of great interest to fully understand the final biological effect. To evaluate the final biological response of a pollutant, it is essential to have an adequate analytical methodology that allows the correct monitoring of environmental systems in order to establish their quality, and, when appropriate, the application of corrective measures. Undoubtedly, massive methods "the omics" are among the most efficient current tools. To this end, transcriptomics, proteomics, metabolomics and chemical speciation can provide very useful information, mainly when they are combined. However, the combination of proteomics with metabolomics has some drawbacks as the temporal space is different (i.e. metabolomics gives information about what happens right now, but it can be related with numerous post-translational modifications happened previously). In this sense, it seems that the combination of genomics with metabolomics is easier. Thus, when metabolomics data are interpreted in combination with genomic, transcriptomic and proteomic results, in the so-called systems biology approach, a holistic knowledge of the organism/process under investigation can be achieved.

Keywords: pesticides, mammals, metabolomics, metals, speciation, omics

1. Introduction

The evaluation of the biological response in living organisms against environmental pollution requires the use of environmental bioindicators or laboratory models of increasing complexity (mollusks, crustaceans and rodents) [1, 2]. A mammal should be always included to integrate the diverse biological filters present in humans, like the digestive tract, which regulates the passage of contaminants to be later distributed through the bloodstream to other parts of the body [3]. In

addition, cell cultures are also important in these types of studies, especially to translate the effects of pollutants to humans. The use of cell cultures also allows performing many experiments without the difficulties of the experimentation with animals [4].

Until now, there has been a great concern, mainly relying on individual environmental pollutants, to study their potential risks. However, the evaluation of health effects of chemicals by considering data obtained just from a single chemical, leads to over or underestimates the joint toxicity. In this sense, studies concerning the combined effects of pollutants better than the toxicity assessment of single chemicals, reflect the realistic impact of environmental exposures [5]. Thus, the experimentation with animal models to evaluate the biological response against environmental pollutants, and their possible translation to the effects in humans, should be carried out designing experiments that mimic as much as possible the environment. To this end, controlled exposure experiments to "chemical cocktails" that integrate several environmental pollutants of different chemical groups can be a good approximation. Although the design of experiments is significantly more complex, the obtained information is of great interest to evaluate the real effects of contaminants in the environment [6].

On the other hand, the joint toxicity of chemical mixtures can be independent one from each other (additive), stronger (synergistic) or weaker (antagonistic), depending of on the sum of effects from individual exposures or not [7].

The biological response can be evaluated using model organisms, but also with free-living species that can also serve as biomarkers of environmental pollution. Although the use of free-living organisms allows evaluating the biological response taking into account all the existing environmental factors and their interactions, and for this reason is closer to the reality, the main pitfalls are related with the difficulty to isolate the metabolic responses connected with a particular pollutant [8]. Otherwise, the study with model organisms is easier and cheaper, but requires a qualified technician to administrate single xenobiotics or their cocktails and the results can be biased by the selected administration route (e.g., subcutaneous injection), model organism used, etc., [10]. Nevertheless, the study of the biological response in free-living animals is of great interest to validate the results obtained in the exposure experiments with model organisms [11, 13].

2. Interactions of metals and chemical species through antagonistic and synergistic mechanisms

As previously commented, biological systems are exposed to a complex environment in which contaminants can interact by means of antagonistic or synergistic mechanisms making mandatory global studies (i.e.,-omics) to evaluate the biological response with an holistic view [14]. In connection to that, selenium has been claimed as the most important element regarding its antagonistic protective action against numerous contaminants (i.e., pesticides and metals). Likewise, it has been stated that cardiovascular damages caused by mercury can be antagonized by selenium [15], but also the neurotoxicity [16] and renal toxicity [17] caused by this element. Moreover, selenium present also protective character against skin cancer induced by arsenic exposure [18], chromosomal aberrations induced by cadmium [19], oxidative stress and lipid peroxidation caused by organophosphorus pesticides [20]. Moreover, the chemical form or specie of selenium is very important since the essentiality or toxicity is directly related to that. Likewise, some of them, especially selenoproteins are peroxidases or oxidoreductases, which protect against oxidative stress [21–24, 26], while inorganic selenium is toxic at high levels [14]. Our research

Mammal	Interaction	Biological effect	References
Interactions of selenium- and arsenic containing species			
Humans	A S_{etotal}/A_{stotal}	DNA hypomethylation/cancer	[39]
Humans		Se reduces the risk of As-related skin lesions and cancer	[16, 40, 41]
Rats	2−A SeO_3/iAs(III)	As prevents lethal liver damage caused by Se	[42]
Rats		As prevents Se-induced cataracts	[43]
Humans		Skin lesion/skin cancer	[44]
Mice		As prevents carcinogenic effect of Se	[45]
Rats		As prevents carcinogenic effect of Se	[46]
Rats		As protects against the toxicity of Se (growth, mortality rate, pathological condition of the liver)	[47–50]
Rats		As induces mucosal glutathione synthesis, explaining its protective effect against Se	[51]
Dogs		As antagonizes Se-induced subnormal growth and restricted food intake	[52]
Cattle		As protects against Se toxicity	[47, 53]
Hogs		As protects against Se toxicity	[48]
Steers		As protects against Se toxicity	[54]
Mice		Se prevents As-induced cytotoxicity	[55]
Mice	2 − 3−A SeO_3/ AsO_4	Se decreases the ratio of organic/inorganic As	[56]
Hamsters		Se decreases As methylation	[14]
Rats	S SeBet/iAs(III)	Coadministration enhances the tumor-suppressive effect of Se	[13]
Interactions of selenium- and mercury-containing species			
Humans	A Setotal/Hgtotal	Se prevents Hg induced cardiovascular diseases	[15]
Humans	A Setotal/$MeHg^+$	Se inhibits Hg-induced neurotoxicity	[57]
Humans		Se inhibits Hg-induced cardiovascular diseases	[58]
Rats		Se may alter the reproductive and developmental toxicity of MeHg+	[59]
Rats	2−/2+ A SeO_3 Hg	Se antagonizes Hg-induced intestinal necrosis	[60]
Rats		Se prevents Hg renotoxicity	[62]
Mice		Se prolongs the half-lives of Hg-exposed animals	[63]
Rats		Se changes the subcellular Hg distribution	[64]
Mice	2−/+ A SeO_4 MeHg	Se protects against Hg-induced neurotoxicity	[65]
Rats	A SeMet/Hg^{2+}	Se inhibits the effects of Hg on organic activities	[66]
Rats	A SeMet/$MeHg^+$	Se prevents Hg-induced porphyrinuria	[67]
Humans	A SeProt/ Hg^0	Se detoxifies Hg	[68]
Mice	A SeProt/$MeHg^+$	Hg affects the activities of selenoenzymes	[69]
Interactions of selenium and sulfur-containing species			
Sheep	A SeMet/sulfur compounds	More Se is incorporated into wool and plasma protein when dietary S is limiting	[70]

Mammal	Interaction	Biological effect	References
Interactions of selenium with species of elements			
Rats	A Se/Sb	Sb has a partially protective effect against Se toxicity	[49, 52, 71]
Rats	A Se/Bi	Bi has a partially protective effect against Se toxicity	[49, 71]
Human cells	A Se/Cd	Se protects against Cd toxicity	[72]
Rats	A Se/Cd	Se prevents Cd-induced oxidative stress	[73]
	A Se/Cd	Se protects against Cd-induced nephrotoxicity and hepatotoxicity	[74]
Mice	A Se/Cd	Se protects against Cd-induced chromosomal aberrations	[19]
Monkeys	A Se/Cd	Se protects enzyme systems	[75]
Rats	S Se/Cd	Se and Cd affect the hepatic gluconeogenic pathway	[76]
	A Se/Cd	Se partially restores Cd-induced oxidative stress and decreased sperm count and motility	[77]
	A Se/Cd	Se antagonizes the Cd-induced inhibition of hepatic drug metabolism	[78]
	A Se/Cd	Se antagonizes Cd-induced testicular damage	[79]
	A Se/Cd	Hepatoprotective effects of Se against Cd	[80]
	A Se/Ge	Ge partially protects against Se toxicity	[49, 71]
	A Se/Ni	Se may antagonize the deleterious effects of Ni during reproduction	[81, 82]
Mice	S Se/Ag	Se protects against Ag-induced lipid peroxidation in the liver	[83, 84]
	S Se/Te/Hg	Hg retention is increased by pre-administration of Te or Se	[85]
Rats	A W/Se	W partially protects against Se toxicity	[49, 71]

Table 1.
Main interaction of selenium species with other elements.

group has been working with *Mus musculus* mice exposed to the pesticide dichlorodiphenyldichloroethylene (DDE) and selenium [27] to study the join effect in the metabolome, as well as several exposure experiments to one or two metals (metalloids) in *Mus musculus* like Se-Cd [2], Hg-Se [28] and others.

Perhaps, the most studied interaction of selenium is with mercury, which was first investigated in 1967 in rats exposed to mercury chloride and selenite [29]. One of the proposed mechanism for this interaction is the formation of Hg-Se complexes, which can result in an increased whole-body retention of Hg after the co-exposure to both elements [29, 30]. However, although this interaction is well known, the mechanisms related to that remain unsolved. It has been stated that inorganic mercury can be incorporated to selenoproteins, peptides and prosthetic groups of selenoenzymes, by reaction of mercury with the selenol group of selenocysteine (SeCys) [31]. The lower pKa of SeCys makes it more reactive that Cys and for this reason, the former reacts by means of -she with Hg with stronger affinity than -SH groups. Mercury can be also incorporated into selenoproteins which play important roles in the maintenance of cellular homeostasis [26, 32]. To this end, Hg^{2-} can react with Se^{2-} (selenides), selenols or hydrogen selenide to form ternary complexes Hg—Se—S together with glutathione that finally bond to

selenoprotein P (SelP) [32–34]. The dysfunction of several proteins, like selenoproteins is in the basis of several diseases like carcinogenesis. In this sense, selenoprotein P accounts for the highest content of selenium in serum and can be also present in other selenoenzymes such as thioredoxin reductase (ThxR), glutathione peroxidase (GPx) and selenoprotein P [35, 36].

Previous studies carried out in our research group using *Mus musculus* mice as a model organism exposed to mercury and selenium demonstrate that the levels of selenoprotein P increase in the liver (extracts from hepatic cytosolic extracts) and serum after Hg exposure, and that selenite supplementation increase the effect [28]. In this context, the synthesis of SelP from selenite increases after mercury exposure since this protein serves as a vehicle for Hg detoxification [37]. This fact is also in good agreement with the decrease observed in the selenometabolites found in serum and the correlative increase in liver, where SelP synthesis takes places from selenite to be later transferred to bloodstream [38]. Moreover, our studies demonstrated that the accumulation of SelP in the liver is higher when the diet is supplemented with selenium. On the other hand, the concentration of selenoalbumin increase in liver and decrease in serum after Hg exposure to transport selenium for the synthesis of SelP [38]. Thus, these results demonstrate the interaction of mercury and selenium in the detoxification process induced by the later, the accumulation of selenoproteins in liver and bloodstream and the homeostasis of elements.

Table 1 shows the main selenium interactions with other metals in mammals.

3. Pesticides, metals and their impairments at molecular level

The joint effect induced by pesticides and metal ions increase, in general, the toxicity (synergism) like DNA damage, mortality rate and reduction in the reproduction rate, as well as changes in enzyme activities [86]. The exposure to environmental pollutants such as polychlorinated biphenyls (PCBs), organochlorine pesticides and heavy metals, has been associated to immunotoxic effects in mammals such as alterations of both the innate and adaptive arms of immune systems, which include aspects of cellular and humoral immunity [87].

Metabolic impairments caused by the join exposure of the pesticide DDE and selenium have been studied using a metabolomics approach [27]. In this study, we conclude that about 70 metabolites are altered in the most metabolically active organs, like liver and kidney, but also in brain, and that they are related with the pathways of oxidative stress, degradation of phospholipidic membrane, β-oxidation and energy metabolism, which confirm the potential of combined metabolomic platforms in environmental studies. Moreover, several metabolites present different response (increase or decrease against the control group) in the organs studied indicating a possible traffic between them. This is the case of liver and kidney, which are the most metabolically active organs and present five metabolites altered with the opposite tendency between them, namely: diacylglycerol (DAG) (18:2/18:1) and ornithine and triacylglycerols (TAG) (18:4/18:4/20:4), TAG (16:0/18:1/22:0) and TAG (18:4/18:4/22:6).

To deep insight into the protective effect of selenium against the toxic effect of DDE, we can compare mice supplemented with DDE, DDE + selenium and selenium against the control group. Selenium counteracts the effect of DDE on seven metabolites because they show a different response among the studied groups when they are compared against the control, which is illustrated in **Figure 1**. These metabolites can be used a biomarkers of the antagonistic interaction between selenium and DDE.

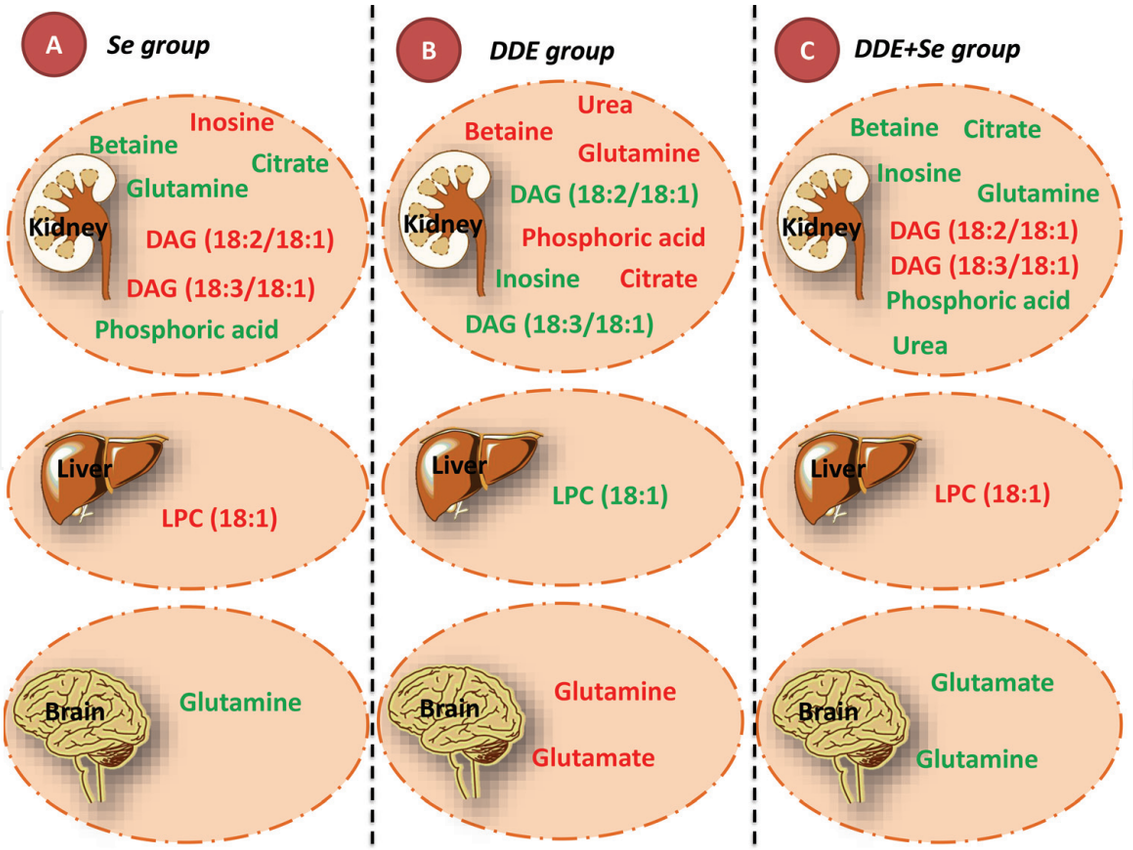

Figure 1.
Different response of metabolites in Mus musculus *mice exposed to: (A) selenium, (B) DDE, (C) selenium and DDE. Red word: Increased; Green word: Decreased.*

It is also remarkable that these antagonistic interactions between DDE and Se usually take place in kidney, since the majority of metabolites that present different response between the mice supplemented with DDE and DDE with Se occur in this organ.

4. Pharmaceutical active compounds, metals and pesticides and their impairments at molecular level

The high consumption of medicines, cosmetic products, as well as pesticides in modern agriculture or plastics in the handling and conservation of food, among others, has led to the appearance in the environment, mainly in soils and aquatic environment, of a series of compounds harmful to the living organisms that inhabit it. These are the well-known "organic micropollutants" (MCOs), a large group of substances that are continuously incorporated into the environment and that, in general, are difficult to eliminate. These substances are classified into two main groups: priority pollutants, and the so-called emerging pollutants (CE). Its detection in the environment has been possible, in many cases, thanks to the development of new and more sensitive analytical technologies that have allowed the detection of these compounds in all types of environmental samples, even in zones, apparently, not subjected to this "contaminant pressure". The analysis of "effluents of wastewater treatment plants" (WWTPs), urban and industrial, has shown, unequivocally, in general, very poor elimination of most of these substances, which is why an incorporation occurs continuously through this way to aquatic and terrestrial ecosystems [88].

Thus, besides metals and pesticides, pharmacologically active compounds (PAC), is a group of emerging contaminants, which are receiving increasing

attention because of their potential harmful effects for the environment and human health. The prevention of the emission of priority and emerging pollutants through wastewater treatment plants effluents into the aqueous environment requires the development of new treatment technologies that ensure the quality of receiving water bodies since the actual treatments are deficient and these substances are continuously incorporated into the environment [89]. Actually, analytical methodologies allow determining these substances in almost all samples and at very low levels [88, 90]. All the pollutants present in the aquatic and therefore terrestrial systems, as well as the products that are originated from them by degradation or metabolization, have an inevitable effect on the organisms that inhabit them, it can be highlighted, for example, their presence in coastal sediments or American red crab [90].

The studies related to the effect on biological responses by the presence of chemical cocktails concerning mixtures of pesticides, PACs and metals are scarce [91]. In this context, the effect that the presence of the antibiotic, ciprofloxacin has on the toxicity, distribution and accumulation of copper has been studied in earthworm (*Eisenia fetida*) [92]. However, the study of the biological response of mammals against "chemical cocktails" including metals, pesticides, and PACs has not been performed until now.

5. Omic methodologies to assess health effects of pesticides and other organic micropollutants

It is essential to have an adequate analytical methodology that allows the correct monitoring of environmental systems in order to establish their quality, and, when appropriate, the application of corrective measures. Undoubtedly, massive methods "the omics" are among the most efficient current tools. In this sense, it has been demonstrated that alterations of the homeostatic cycles are shown at the transcriptional level (transcriptomics) [93], by overexpression or inhibition of proteins (proteomics) [94] and by modifications of the metabolic cycles (metabolomics) [1, 95]. On the other hand, it has been stated that approximately 1/3 of proteins need the presence of metals as cofactors to develop their function (metalloproteins) and that metals influence on more than 50% of the proteins [96]. These metals play essential roles due to their catalytic properties or influence the structure of proteins and generally, the genome determines their presence in molecules [97]. Thus, metallomics allows understanding the distribution of elements, concentration at equilibrium of free metallic ions or free elements in a cellular compartment, cell or organism [98] and refers to the identity and/or quantity of metals/metalloids and their species [99]. Likewise, it has been proposed the integration of a global (holistic) view, much more realistic of the processes that takes place in the environment [95].

To this end, genomics decipher the information that determines cell function which is contained in the cellular core, transcriptomics reveals gene expression and proteomics make possible the examination of protein synthesis and cell signaling. On the other hand, in the establishment of transcriptional expression profiles that explain the gene function is critical in **environmental transcriptomics**, and the quantification of gene expression changes at the level of mRNA has proved to be an interesting tool in environmental approaches [100]. The transcriptomes of *M. spretus* mice captured in areas of high industrial and agricultural pollution such as the Domingo Rubio estuary or the industrial pole of Punta del Sebo (Huelva) have been determined, identifying a set of potential biomarkers of environmental contamination [93]. Likewise, the transcriptional profile of contaminants such as the DDE has been determined in controlled exposure studies in the laboratory [101].

On the other hand, proteomics is of great importance to understand cell homeostasis, to perform quantitative analysis and for the identification of potential biomarkers of *in vivo* toxicity. However, the massive number of proteins and post-translational modifications difficult the analysis. Proteomics can also assist genomic and transcriptomic studies in the efficient sequencing of complete genomes and to explain differences in susceptibility induced by polymorphisms. Nowadays is accepted that alterations in gene expression do not always induce adverse health effects due to post-translational modifications, as phosphorylation and glycosylation of proteins that determine their function, environmental factors or multigenic processes (e.g., aging and disease). For this reason, **environmental proteomics** allows understanding the mode of action of pollutants. Likewise, the use of 1st, 2nd and 3rd generation proteomic methods have been used to compare *M. spretus* mice from different areas within the Doñana National Park (PND) and surroundings, the Estero de Domingo Rubio (EDR) and the Estuary of the Guadalquivir, in order to evaluate the effects of the Aznalcóllar accident and the pesticides used around the PND [102, 103]. The information obtained by this omic can be complemented by **oxidative stress and redox proteomics**. The study of reactive oxygen species (ROS) related to oxidative damage caused by contaminants is a valuable tool in environmental studies [94]. Although most amino acids are sensitive to oxidation, the thiol group of Cys affects them especially [95] so its analysis allows determining the redox state of the thiol groups of proteins [104, 105], so its analysis allows determining the redox state of the thiol groups of proteins.

Environmental metallomics and chemical speciation requires its own methodology, generally based in the combined use of inorganic mass spectrometry (inductively coupled plasma mass spectrometry, ICP-MS) and organic MS, using a previous chromatographic separation step in order to preserve the integrity of the metal in terms of its union and location in the molecule [106]. Our research group have extensive experience in this field, especially in its projection to the environmental studies [2], and the biological response, at the metallomic and metabolomic levels of the *Mus musculus* mice exposed to As [99], Cd [2] and Hg [103]. In addition, studies with free living animals in Doñana National Park and surroundings based on these methodologies have also been performed using the *Mus spretus* mice [11] and the crab *Procambarus clarkii*.

In addition, in 1999, J. Nicholson defines metabonomics as "the quantitative measurement of the dynamic multiparametric metabolic response of living systems to pathophysiological stimuli or genetic modification" [104], and metabolomics as the measurement of all the metabolites in a specified biological sample [104]. Likewise, metabonomics allows understanding the variation in low molecular mass molecules, namely metabolites, which are the last action mechanism of the organisms, but the additional mentioned "-omics" sciences are related to cellular macromolecules. Then, the last step in the omics, directly in connection with the phenotype, **environmental metabolomics** allows obtaining a global view of the metabolic fingerprint of the biological systems exposed to contaminants, providing information at the same time that the interactions between the contaminants with the living organisms [105]. Numerous studies have been carried out on different rodents, and several exposure experiments on the *Mus musculus* laboratory mice can be highlighted. As an example, several studies carried to study of the metabolomic response of the mouse.

M. musculus exposed to inorganic As [99], Cd [95], Se-Cd [2], Hg [99], Hg-Se [28], DDE-Se [27], As, Cd, Se, Hg, deltamethrin + acrolein.

One of the most critical aspects in metabolomics is sample treatment, to extract as many metabolites as possible [10]. The analysis of biofluids for metabolomics in simpler than the extraction of metabolites from tissues and allows obtaining global

information about the state of the organism, but to obtain specific information on a specific organ the direct analysis is mandatory [106].

Finally, metabolomics allows the simultaneous measurement of hundreds of metabolites *in vitro* cell cultures, *in* vivo tissues and even in non-invasive blood and urine biofluids. However, the current drawback of this omic is the standardization of quenching, metabolite extraction procedures as well as the complexity of data analysis and interpretation. Moreover, the influence of factors in the results is important (e.g., age, gender, diet, stress, housing conditions, health status). To overcome these limitations, combination of omics seems to be the best option. However, the temporal space is different in metabolomics and proteomics (i.e., metabolomics gives information about what happens right now, but it can be related with numerous post-translational modifications happened previously). In this sense, it seems that the combination of genomics with metabolomics is easier. Thus, when metabolomics data are interpreted in combination with genomic, transcriptomic and proteomic results, in the so-called systems biology approach, a holistic view of the organism or biological process under investigation can be attained.

6. Concluding remarks

The evaluation of the real impact of a pollutant, and in particular of pesticides can be performed in the different environmental compartments or preferably in mammals to decipher the biological response. In this case, the study can be carried out in free-living animals or in laboratory mice exposed to different pollutants that should be combined to evaluate the biological response of the "chemical cocktail" since they can interact in a synergistic or antagonistic fashion. On the other hand, it is essential to have an adequate analytical methodology that allows the correct monitoring of environmental systems in order to establish their quality, and, when appropriate, the application of corrective measures. Undoubtedly, massive methods "the omics" are among the most efficient current tools.

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Conflict of interest

The authors do not have conflict of interest.

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References

[1] Gago-Tinoco A, González-Domínguez R, García-Barrera T, Blasco-Moreno J, Bebianno MJ, Gómez-Ariza J-L. Metabolic signatures associated with environmental pollution by metals in Doñana National Park using *P. clarkii* as bioindicator. Environmental Science and Pollution Research. 2014;**21**: 13315-13323. DOI: 10.1007/s11356-014-2741-y

[2] García-Sevillano MA, García-Barrera T, Gómez-Ariza JL. Application of metallomic and metabolomic approaches in exposure experiments on laboratory mice for environmental metal toxicity assessment. Metallomics. 2014;**6**:237-248. DOI: 10.1039/c3mt00302g

[3] Gómez-Ariza JL, Jahromi EZ, González-Fernández M, García-Barrera T, Gailer J. Liquid chromatography-inductively coupled plasma-based metallomic approaches to probe health-relevant interactions between xenobiotics and mammalian organisms. Metallomics. 2011;**3**:566-577. DOI: 10.1039/c1mt00037c

[4] L'Azou B, Passagne I, Mounicou S, Tréguer-Delapierre M, Puljalté I, Szpunar J, et al. Comparative cytotoxicity of cadmium forms ($CdCl_2$, CdO, CdS micro- and nanoparticles) in renal cells. Toxicology Research. 2014;**3**: 32-41. DOI: 10.1039/c3tx50063b

[5] Schnug L, Leinaas HP, Jensen J. Synergistic sub-lethal effects of a biocide mixture on the springtail Folsomia fimetaria. Environmental Pollution. 2014;**186**:158-164. DOI: 10.1016/J.ENVPOL.2013.12.004

[6] Van den Brink PJ, Tarazona JV, Solomon KR, Knacker T, Van den Brink NW, Brock TCM, et al. The use of terrestrial and aquatic microcosms and mesocosms for the ecological risk assessment of veterinary medicinal products. Environmental Toxicology and Chemistry. 2005;**24, 820**. DOI: 10.1897/04-268R.1

[7] Warne MSJ, Hawker DW. The number of components in a mixture determines whether synergistic and antagonistic or additive toxicity predominate: The funnel hypothesis. Ecotoxicology and Environmental Safety. 1995;**31**:23-28. DOI: 10.1006/EESA.1995.1039

[8] Andrews NC. Metal transporters and disease. Current Opinion in Chemical Biology. 2002;**6**:181-186. DOI: 10.1016/S1367-5931(02)00307-1

[9] García-Barrera T, Rodríguez-Moro G, Callejón-Leblic B, Arias-Borrego A, Gómez-Ariza JL. Mass spectrometry based analytical approaches and pitfalls for toxicometabolomics of arsenic in mammals: A tutorial review. Analytica Chimica Acta. 2018;**1000**:41-66. DOI: 10.1016/j.aca.2017.10.019

[10] García-Sevillano MA, González-Fernández M, Jara-Biedma R, García-Barrera T, López-Barea J, Pueyo C, et al. Biological response of free-living mouse *Mus spretus* from Doñana National Park under environmental stress based on assessment of metal-binding biomolecules by SEC-ICP-MS. Analytical and Bioanalytical Chemistry. 2012;**404**:1967-1981. DOI: 10.1007/s00216-012-6274-2

[11] González-Fernandez M, García-Sevillano MA, Jara-Biedma R, Navarro-Roldán F, García-Barrera T, López-Barea J, et al. Use of metallomics in environmental pollution assessment using mice *mus musculus/mus spretus* as bioindicators. Current Analytical Chemistry. 2013;**9**:229-243. DOI: http://www.scopus.com/inward/record.url?eid=2-s2.0-84875305764&partnerID=tZOtx3y1

[12] García-Barrera T, Gómez-Ariza JL, González-Fernández M, Moreno F, García-Sevillano MA, Gómez-Jacinto V. Biological responses related to agonistic, antagonistic and synergistic interactions of chemical species. Analytical and Bioanalytical Chemistry. 2012;**403**: 2237-2253. DOI: 10.1007/s00216-012-5776-2

[13] Park K, Mozaffarian D. Omega-3 fatty acids, mercury, and selenium in fish and the risk of cardiovascular diseases. Current Atherosclerosis Reports. 2010;**12**:414-422. DOI: 10.1007/s11883-010-0138-z

[14] Choi AL, Budtz-Jørgensen E, Jørgensen PJ, Steuerwald U, Debes F, Weihe P, et al. Selenium as a potential protective factor against mercury developmental neurotoxicity. Environmental Research. 2008;**107**: 45-52. DOI: 10.1016/J.ENVRES.2007.07.006

[15] Beyrouty P, Chan HM. Co-consumption of selenium and vitamin E altered the reproductive and developmental toxicity of methylmercury in rats. Neurotoxicology and Teratology. 2006;**28**:49-58. DOI: 10.1016/J.NTT.2005.11.002

[16] Kolachi NF, Kazi TG, Wadhwa SK, Afridi HI, Baig JA, Khan S, et al. Evaluation of selenium in biological sample of arsenic exposed female skin lesions and skin cancer patients with related to non-exposed skin cancer patients. The Science of the Total Environment. 2011;**409**:3092-3097. DOI: 10.1016/j.scitotenv.2011.05.008

[17] Mukherjee A, Sharma A, Talukder G. Effect of selenium on cadmium-induced chromosomal aberrations in bone marrow cells of mice. Toxicology Letters. 1988;**41**:23-29. DOI: 10.1016/0378-4274(88)90004-5

[18] Milošević MD, Paunović MG, Matić MM, Ognjanović BI, Saičić ZS. The ameliorating effects of selenium and vitamin C against fenitrothion-induced blood toxicity in Wistar rats. Environmental Toxicology and Pharmacology. 2017;**56**:204-209. DOI: 10.1016/j.etap.2017.09.016

[19] Lai IK, Chai Y, Simmons D, Watson WH, Tan R, Haschek WM, et al. Dietary selenium as a modulator of PCB 126-induced hepatotoxicity in male Sprague-Dawley rats. Toxicological Sciences. 2011;**124**:202-214. DOI: 10.1093/toxsci/kfr215

[20] Raines AM, Sunde RA. Selenium toxicity but not deficient or super-nutritional selenium status vastly alters the transcriptome in rodents. BMC Genomics. 2011;**12**. DOI: 10.1186/1471-2164-12-26

[21] Chen J, Berry MJ. Selenium and selenoproteins in the brain and brain diseases. Journal of Neurochemistry. 2003;**86**:1-12. DOI: 10.1046/j.1471-4159.2003.01854.x

[22] Stadtman TC. Biosynthesis and function of selenocysteine-containing enzymes. The Journal of Biological Chemistry. 1991;**266**:16257-16260

[23] Steinbrenner H, Sies H. Protection against reactive oxygen species by selenoproteins. Biochimica et Biophysica Acta, General Subjects. 2009;**1790**:1478-1485. DOI: 10.1016/j.bbagen.2009.02.014

[24] Rodríguez-Moro G, Abril N, Jara-Biedma R, Gómez-Ariza JL, García-Barrera T. Metabolic impairments caused by "chemical cocktails" in mammals using direct infusion triple quadrupole time of flight and gas chromatography mass spectrometry. Metabolomics (n.d.); 2019

[25] García-Sevillano MA, Rodríguez-Moro G, García-Barrera T, Navarro F, Gómez-Ariza JL. Biological interactions between mercury and selenium in

distribution and detoxification processes in mice under controlled exposure. Effects on selenoprotein. Chemico-Biological Interactions. 2015;**229**:82-90. DOI: 10.1016/j.cbi.2015.02.001

[26] Parziek J, Ostadalova I, Kalouskva J, Babichy A, Benes J. The detoxifying effects of selenium. Interrelation between compounds of selenium and certain metals. In: Mertz W, Cornatzer WE, editors. Newer Trace Element Nutrition. New York, USA: Marcel Dekker; 1971. pp. 85-122

[27] Carvalho CML, Lu J, Zhang X, Arnér ESJ, Holmgren A. Effects of selenite and chelating agents on mammalian thioredoxin reductase inhibited by mercury: Implications for treatment of mercury poisoning. The FASEB Journal. 2011;**25**:370-381. DOI: 10.1096/fj.10-157594

[28] Holben DH, Smith AM. The diverse role of selenium within selenoproteins. Journal of the American Dietetic Association. 1999;**99**:836-843. DOI: 10.1016/S0002-8223(99)00198-4

[29] Falnoga I, Tusek-Znidaric M. Selenium-mercury interactions in man and animals. Biological Trace Element Research. 2007;**119**:212-220. DOI: 10.1007/s12011-007-8009-3

[30] Suzuki KT, Sasakura C, Yoneda S. Binding sites for the (Hg-Se) complex on selenoprotein P. Biochimica et Biophysica Acta-Protein Structure and Molecular Enzymology. 1998;**1429**:102-112. DOI: 10.1016/S0167-4838(98)00221-0

[31] Suzuki KT, Ishiwata K, Ogra Y. Incorporation of selenium into selenoprotein P and extracellular glutathione peroxidase: HPLC-ICPMS data with enriched selenite. The Analyst. 1999;**124**:1749-1753. DOI: 10.1039/a906521k

[32] Matés JM, Segura JA, Alonso FJ, Márquez J. Roles of dioxins and heavy metals in cancer and neurological diseases using ROS-mediated mechanisms. Free Radical Biology & Medicine. 2010;**49**:1328-1341. DOI: 10.1016/j.freeradbiomed.2010.07.028

[33] Matés JM, Segura JA, Alonso FJ, Márquez J. Intracellular redox status and oxidative stress: Implications for cell proliferation, apoptosis, and carcinogenesis. Archives of Toxicology. 2008;**82**:273-299. DOI: 10.1007/s00204-008-0304-z

[34] Chen C, Yu H, Zhao J, Li B, Qu L, Liu S, et al. The roles of serum selenium and selenoproteins on mercury toxicity in environmental and occupational exposure. Environmental Health Perspectives. 2006;**114**:297-301. DOI: 10.1289/ehp.7861

[35] Shiobara Y, Suzuki KT. Binding of selenium (administered as selenite) to albumin after efflux from red blood cells. Journal of Chromatography. B, Biomedical Sciences and Applications. 1998;**710**:49-56. http://www.scopus.com/inward/record.url?eid=2-s2.0-0032511207&partnerID=tZOtx3y1 [Accessed: November 13, 2015]

[36] Richard Pilsner J, Hall MN, Liu X, Ahsan H, Ilievski V, Slavkovich V, et al. Associations of plasma selenium with arsenic and genomic methylation of leukocyte DNA in Bangladesh. Environmental Health Perspectives. 2011;**119**:113-118. DOI: 10.1289/ehp.1001973

[37] Chen Y, Hall M, Graziano JH, Slavkovich V, Van Geen A, Parvez F, et al. A prospective study of blood selenium levels and the risk of arsenic-related premalignant skin lesions. 2007; **16**:207-214. DOI: 10.1158/1055-9965.EPI-06-0581

[38] Spallholz JE, Mallory Boylan L, Rhaman MM. Environmental hypothesis: Is poor dietary selenium intake an underlying factor for

arsenicosis and cancer in Bangladesh and West Bengal, India? The Science of the Total Environment. 2004;**323**:21-32. DOI: 10.1016/J.SCITOTENV.2003.09.034

[39] Verret WJ, Chen Y, Ahmed A, Islam T, Parvez F, Kibriya MG, et al. A randomized, double-blind placebo-controlled trial evaluating the effects of vitamin E and selenium on arsenic-induced skin lesions in Bangladesh. Journal of Occupational and Environmental Medicine. 2005;**47**:1026-1035. DOI: 10.1097/01.jom.0000183095.45050.97

[40] Moxon AL. The effect of arsenic on the toxicity of seleniferous grains. Science. 1938;**88**:81

[41] Shearer TR, Anderson RS, Britton JL. Influence of selenite and fourteen trace elements on cataractogenesis in the rat. Investigative Ophthalmology and Visual Science. 1983;**24**:417-423

[42] Schrauzer GN, White DA, Mcginness JE, Schneider CJ, Bell LJ. Arsenic and cancer: Effects of joint administration of arsenite and selenite on the genesis of mammary adenocarcinoma in inbred female C_3H/St mice. Bioinorganic Chemistry. 1978;**9**:245-253. DOI: 10.1016/S0006- 3061(78)80005-2

[43] Ip C, Ganther H. Efficacy of trimethylselenonium versus selenite in cancer chemoprevention and its modulation by arsenite. Carcinogenesis. 1988;**9**:1481-1484. DOI: 10.1093/carcin/9.8.1481

[44] Dubois KP, Moxon AL, Olson OE. Further studies on the effectiveness of arsenic in preventing selenium poisoning one figure. The Journal of Nutrition. 1940;**19**:477-482. DOI: 10.1093/jn/19.5.477

[45] Moxon AL. The influence of arsenic on selenium poisoning in hogs. Proceedings of the South Dakota Academy of Science. 1941;**21**:34-36

[46] Moxon DK, Alvin L. The Journal of Nutrition. 1939:477-482

[47] Palmer IS, Thiex N, Olson OE. Dietary selenium and arsenic effects in rats. Nutrition Reports International. 1983;**27**:249-257

[48] Pisciotto PT, Graziano JH. Induction of mucosal glutathione synthesis by arsenic. Biochimica et Biophysica Acta, General Subjects. 1980;**628**:241-243. DOI: 10.1016/0304-4165(80)90371-2

[49] Rhian M, Moxon AL. Chronic selenium poisoning in dogs and its prevention by arsenic. The Journal of Pharmacology and Experimental Therapeutics. 1943;**78**:249-264 http://jpet.aspetjournals.org/content/78/3/249.abstract

[50] Olson OE. Selenium and the organic arsenicals. Feed Age. 1960;**10**:49-50

[51] Moxon AL, Rhian MA, Anderson HD, Olson OE. Growth of steers on seleniferous range. Journal of Animal Science. 1944;**3**:299-309. DOI: 10.2527/jas1944.33299x

[52] Biswas S, Talukder G, Sharma A. Prevention of cytotoxic effects of arsenic by short-term dietary supplementation with selenium in mice *in vivo*. Mutation Research - Genetic Toxicology and Environmental Mutagenesis. 1999;**441**:155-160. DOI: 10.1016/S1383-5718(99)00028-5

[53] Kenyon EM, Hughes MF, Levander OA. Influence of dietary selenium on the disposition of arsenate in the female $B_6C_3F_1$, mouse. Journal of Toxicology and Environmental Health. 1997;**51**:279-299. DOI: 10.1080/00984109708984027

[54] Holmberg RE, Ferm VH. Interrelationships of selenium,

cadmium, and arsenic in mammalian teratogenesis. Archives of Environmental Health: An International Journal. 1969;**18**:873-877. DOI: 10.1080/00039896.1969.10665508

[55] Ip C, Ganther HE. Activity of methylated forms of selenium in cancer prevention. Cancer Research. 1990;**50**: 1206-1211. http://cancerres.aacrjournals.org/content/50/4/1206.abstract

[56] Yoshizawa K, Rimm EB, Morris JS, Spate VL, Hsieh CC, Spiegelman D, Stampfer NJ, Willett WC. Mercury and the risk of coronary heart disease in men. New England Journal of Medicine.2002;**347**:1755-1760

[57] Parízek J, Ošvtádalová I. The protective effect of small amounts of selenite in sublimate intoxication. Experientia. 1967;**23**:142-143. DOI: 10.1007/BF02135970

[58] An-Sik C, Maines MD, Reynolds WA. Inhibition of the enzymes of glutathione metabolism by mercuric chloride in the rat kidney: Reversal by selenium. Biochemical Pharmacology. 1982;**31**:3093-3100. DOI: 10.1016/0006-2952(82)90085-5

[59] Parízek J, Beneš I, Oštádalová I, Babický A, Beneš J, Lener J. The effect of some inorganic and organic compounds of selenium on the metabolism of cadmium and mercury in the rat. Physiologia Bohemoslovaca. 1969;**18**:95-103

[60] Miura K, Imura N. Changes in ultrastructure and subcellular mercury distribution in rat liver after separate and combined administrations of mercuric mercury and selenite. Journal of Applied Toxicology. 1982;**2**:145-150. DOI: 10.1002/jat.2550020305

[61] Glaser V, Nazari EM, Müller YMR, Feksa L, Wannmacher CMD, Rocha JBT, et al. Effects of inorganic selenium administration in methylmercury-induced neurotoxicity in mouse cerebral cortex. International Journal of Developmental Neuroscience. 2010;**28**: 631-637. DOI: 10.1016/J.IJDEVNEU.2010.07.225

[62] Su L, Wang M, Yin S-T, Wang H-L, Chen L, Sun L-G, et al. The interaction of selenium and mercury in the accumulations and oxidative stress of rat tissues. Ecotoxicology and Environmental Safety. 2008;**70**:483-489. DOI: 10.1016/J.ECOENV.2007.05.018

[63] dos Santos APM, Mateus ML, Carvalho CML, Batoréu MCC. Biomarkers of exposure and effect as indicators of the interference of selenomethionine on methylmercury toxicity. Toxicology Letters. 2007;**169**: 121-128. DOI: 10.1016/J.TOXLET.2006.12.007

[64] Falnoga I, Tusek-Znidaric M. Selenium-mercury interactions in man and animals. Biological Trace Element Research. 2007;**119**:212-220. DOI: 10.1007/s12011- 007-8009-3

[65] Watanabe C, Yin K, Kasanuma Y, Satoh H. In utero exposure to methylmercury and Se deficiency converge on the neurobehavioral outcome in mice. Neurotoxicology and Teratology. 1999;**21**:83-88. DOI: 10.1016/S0892-0362(98)00036-1

[66] White CL. Sulfur-selenium studies in sheep. II. Effect of a dietary sulfur deficiency on selenium and sulfur metabolism in sheep fed varying levels of selenomethionine. Australian Journal of Biological Sciences. 1980;**33**:699-708. DOI: 10.1071/BI9800699

[67] Moxon AL, Jensen C. The influence of germanium gallium antimony and some organic arsenicals on the toxicity of selenium. Proceedings of the South Dakota Academy of Science. 1947:21-26

[68] Frisk P, Yaqob A, Lindh U. Indications of selenium protection

against cadmium toxicity in cultured K-562 cells. The Science of the Total Environment. 2002;**296**:189-197. DOI: 10.1016/S0048-9697(02)00080-3

[69] Messaoudi I, El Heni J, Hammouda F, Saïd K, Kerkeni A. Protective effects of selenium, zinc, or their combination on cadmium-induced oxidative stress in rat kidney. Biological Trace Element Research. 2009;**130**:152-161. DOI: 10.1007/s12011-009-8324-y

[70] Flora SJS, Behari JR, Ashquin M, Tandon SK. Time-dependent protective effect of selenium against cadmium-induced nephrotoxicity and hepatotoxicity. Chemico-Biological Interactions. 1982;**42**:345-351. DOI: 10.1016/0009-2797(82)90078-3

[71] Sidhu M, Sharma M, Bhatia M, Awasthi YC, Nath R. Effect of chronic cadmium exposure on glutathione S-transferase and glutathione peroxidase activities in Rhesus monkey: The role of selenium. Toxicology. 1993;**83**:203-213. DOI: 10.1016/0300-483X(93)90102-X

[72] Bell RR, Soliman MRI, Early JL. Acute effects of cadmium and selenium on glucose output from rat liver hepatocytes using various gluconeogenic precursors. Toxicology. 1990;**65**:161-168. DOI: 10.1016/0300-483X(90)90086-V

[73] Messaoudi I, Banni M, Saïd L, Saïd K, Kerkeni A. Involvement of selenoprotein P and GPx4 gene expression in cadmium-induced testicular pathophysiology in rat. Chemico-Biological Interactions. 2010;**188**:94-101. DOI: 10.1016/J.CBI.2010.07.012

[74] Early JL, Schnell RC. Selenium antagonism of cadmium-induced inhibition of hepatic drug metabolism in the male rat. Toxicology and Applied Pharmacology. 1981;**58**:57-66. DOI: 10.1016/0041-008X(81)90115-0

[75] Prohaska JR, Mowafy M, Ganther HE. Interactions between cadmium, selenium and glutathione peroxidase in rat testis. Chemico-Biological Interactions. 1977;**18**:253-265. DOI: 10.1016/0009-2797(77)90012-6

[76] Newairy AA, El-Sharaky AS, Badreldeen MM, Eweda SM, Sheweita SA. The hepatoprotective effects of selenium against cadmium toxicity in rats. Toxicology. 2007;**242**:23-30. DOI: 10.1016/J.TOX.2007.09.001

[77] Käkelä R, Käkelä A, Hyvärinen H. Effects of nickel chloride on reproduction of the rat and possible antagonistic role of selenium. Comparative Biochemistry and Physiology - Part C: Toxicology & Pharmacology. 1999;**123**:27-37. DOI: 10.1016/S0742-8413(99)00006-7

[78] Khandelwal S, Flora SJS, Tandon SK. Nickel-selenium interaction-time dependent biochemical alterations and metal decorporation in rats. Chemico-Biological Interactions. 1990;**75**:341-347. DOI: 10.1016/0009-2797(90)90076-Y

[79] Serafín Muñoz AH, Wrobel K, Gutierrez Corona JF, Wrobel K. The protective effect of selenium inorganic forms against cadmium and silver toxicity in mycelia of *Pleurotus ostreatus*. Mycological Research. 2007;**111**: 626-632. DOI: 10.1016/J.MYCRES.2007.03.002

[80] Rungby J. Silver-induced lipid peroxidation in mice: Interactions with selenium and nickel.Toxicology.1987; **45**:135-142

[81] Khayat A, Dencker L. Interactions between tellurium and mercury in murine lung and other organs after metallic mercury inhalation: A comparison with selenium. Chemico-Biological Interactions. 1984;**50**:123-133. DOI: 10.1016/0009-2797(84)90089-9

[82] Uwizeyimana H, Wang M, Chen W, Khan K. The eco-toxic effects of pesticide and heavy metal mixtures towards earthworms in soil. Environmental Toxicology and Pharmacology. 2017;**55**:20-29. DOI: 10.1016/j.etap.2017.08.001

[83] Desforges J-PW, Sonne C, Levin M, Siebert U, De Guise S, Dietz R. Immunotoxic effects of environmental pollutants in marine mammals. Environment International. 2016;**86**: 126-139. DOI: 10.1016/j.envint.2015.10.007

[84] Camacho-Muñoz D, Martín J, Santos JL, Aparicio I, Alonso E. Occurrence, temporal evolution and risk assessment of pharmaceutically active compounds in Doñana Park (Spain). Journal of Hazardous Materials. 2010; **183**:602-608. DOI: 10.1016/j.jhazmat.2010.07.067

[85] Deblonde T, Cossu-Leguille C, Hartemann P. Emerging pollutants in wastewater: A review of the literature. International Journal of Hygiene and Environmental Health. 2011;**214**: 442-448. DOI: 10.1016/j.ijheh.2011.08.002

[86] Kazakova J, Fernández-Torres R, Ramos-Payán M, Bello-López M. Multiresidue determination of 21 pharmaceuticals in crayfish (*Procambarus clarkii*) using enzymatic microwave-assisted liquid extraction and ultrahigh-performance liquid chromatography-triple quadrupole mass spectrometry analysis. Journal of Pharmaceutical and Biomedical Analysis. 2018;**160**:144-151. DOI: 10.1016/j.jpba.2018.07.057

[87] Cedergreen N. Quantifying synergy: A systematic review of mixture toxicit studies within environmenta toxicology. PLoSONE.2014;9:e96580.DOI:10.1371/journal.pone.0096580

[88] Huang R, Wen B, Pei Z, Shan X-Q, Zhang S, Williams PN. Accumulation, subcellular distribution and toxicity of copper in earthworm (*Eisenia fetida*) in the presence of ciprofloxacin. Environmental Science & Technology. 2009;**43**:3688-3693. DOI: 10.1021/es900061t

[89] Abril N, Ruiz-Laguna J, García-Sevillano MA, Mata AM, Gómez-Ariza JL, Pueyo C. Heterologous microarray analysis of transcriptome alterations in Mus spretus mice living in an industrial settlement. Environmental Science & Technology.2014;**48**:2183-2192. DOI: 10.1021/es4053973

[90] Montes-Nieto R, Fuentes-Almagro CA, Bonilla-Valverde D, Prieto-Álamo M-J, Jurado J, Carrascal M, et al. Proteomics in free-living *Mus spretus* to monitor terrestrial ecosystems. Proteomics. 2007;7:4376-4387. DOI: 10.1002/pmic.200700409

[91] García-Sevillano MA, Abril N, Fernández-Cisnal R, García-Barrera T, Pueyo C, López-Barea J, et al. Functional genomics and metabolomics reveal the toxicological effects of cadmium in *Mus musculus* mice. Metabolomics. 2015. DOI: 10.1007/s11306-015-0801-z

[92] Mounicou S, Szpunar J, Lobinski R. Metallomics: The concept and methodology. Chemical Society Reviews. 2009;**38**:1119-1138. DOI: 10.1039/b713633c

[93] Tainer JA, Roberts VA, Getzoff ED. Metal-binding sites in proteins. Current Opinion in Biotechnology. 1991;**2**:582-591. DOI: 10.1016/0958- 1669(91)90084-I

[94] Williams RJ. Chemical selection of elements by cells. Coordination Chemistry Reviews. 2001;**216–217**: 583-595. DOI: 10.1016/S0010-8545(00)00398-2

[95] García-Sevillano MA, García-Barrera T, Navarro F, Gómez-Ariza JL. Analysis of the biological response of mouse liver (*Mus musculus*) exposed to As_2O_3 based on integrated -omics approaches. Metallomics. 2013;**5**: 1644-1655. DOI: 10.1039/c3mt00186e

[96] Abril N, Ruiz-Laguna J, Osuna-Jiménez I, Vioque-Fernández A, Fernández-Cisnal R, Chicano-Gálvez E, et al. Omic approaches in environmental issues. Journal of Toxicology and Environmental Health. Part A. 2011;**74**: 1001-1019. DOI: 10.1080/15287394.2011.582259

[97] Morales-Prieto N, Ruiz-Laguna J, Abril N. Dietary Se supplementation partially restores the REDOX proteomic map of *M. spretus* liver exposed to p,p′-DDE. Food and Chemical Toxicology. 2018;**114**:292-301. DOI: 10.1016/j.fct.2018.02.047

[98] Bonilla-Valverde D. Contaminación de doñana: biomarcadores bioquímicos y proteomica en el ramon moruno (*Mus spretus*) y en el gorrion comun (*Passer domesticus*). Universidad de Córdoba. 2006

[99] Abril N, Chicano-Gálvez E, Michán C, Pueyo C, López-Barea J. ITRAQ analysis of hepatic proteins in free-living *Mus spretus* mice to assess the contamination status of areas surrounding Doñana National Park (SW Spain). The Science of the Total Environment. 2015;**523**:16-27. DOI: 10.1016/j.scitotenv.2015.03.116

[100] Morales-Prieto N, Abril N. REDOX proteomics reveals energy metabolism alterations in the liver of *M. spretus* mice exposed to p,p′-DDE. Chemosphere. 2017;**186**:848-863. DOI: 10.1016/j.chemosphere.2017.08.057

[101] Fernández-Cisnal R, Alhama J, Abril N, Pueyo C, López-Barea J. Redox proteomics as biomarker for assessing the biological effects of contaminants in crayfish from Doñana National Park. The Science of the Total Environment. 2014;**490**:121-133. DOI: 10.1016/j.scitotenv.2014.04.117

[102] Sperling M, Karst U. Metallomics: An emerging interdisciplinary science. Analytical and Bioanalytical Chemistry. 2013;**405**:1789-1790. DOI: 10.1007/s00216-012-6619-x

[103] García-Sevillano MA, García-Barrera T, Navarro F, Gailer J, Gómez-Ariza JL. Use of elemental and molecular-mass spectrometry to assess the toxicological effects of inorganic mercury in the mouse *Mus musculus*. Analytical and Bioanalytical Chemistry. 2014;**406**:5853-5865. DOI: 10.1007/s00216-014-8010- 6

[104] Nicholson JK, Lindon JC, Holmes E. "Metabonomics": Understanding the metabolic responses of living systems to pathophysiological stimuli *via* multivariate statistical analysis of biological NMR spectroscopic data. Xenobiotica. 1999;**29**:1181-1189. Available from: http://www.scopus.com/inward/record.url?eid=2-s2.0-0032694577&partnerID=tZOtx3y1

[105] García-Sevillano MA, García-Barrera T, Gómez-Ariza JL. Development of a new column switching method for simultaneous speciation of selenometabolites and selenoproteins in human serum. Journal of Chromatography A. 2013;**1318**: 171-179. DOI: 10.1016/j.chroma.2013.10.012

[106] Masson P, Alves AC, Ebbels TMD, Nicholson JK, Want EJ. Optimization and evaluation of metabolite extraction protocols for untargeted metabolic profiling of liver samples by UPLC-MS. Analytical Chemistry;**82**(2010): 7779-7786. DOI: 10.1021/ac101722e