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Risk assessment methodologies in the field of contaminants, food contact materials, technological ingredients and nutritional risks

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Abstract

The programme aimed at training the fellow in the risk assessment guidelines proposed by the EFSA in the field of contaminants, food contact materials, technological ingredients and nutritional risks. It had a modular 'learning by doing' approach and a balanced learning/case studies and theory. Module 1 offered an insight into chemical risk assessment and conferred transferable skills for a proper application of the framework. The hands-on activities consisted of three case studies that went from a simple exercise on an official opinion, to working in a team with experts to produce a new opinion, to an individual work to obtain a publishable review manuscript. Module 2 was a training in experimental toxicology designed to create a toxicological basis and to enable the fellow to perform toxicological studies for risk assessment purposes. She joined the team working on cyanotoxins, gained experience with both EFSA and Organization of Economic Cooperation and Development (OECD) guidelines on genotoxicity and an insight into the developing of analytical methods suitable for risk assessment purposes. During module 3, the fellow was trained in nutritional risk assessment and involved in experimental work in chemical characterisation, biomarkers and mechanisms of action of bioactive compounds. This developed the critical perspective when assessing nutritional and health claims related the design of experiments, methods used, interpretation of results and human relevance. Module 4 provided a 'hand-on experience' in scientific risk communication as the fellow was encouraged and supported in the participation at local, national and international workshops and congresses presenting the outcomes of the three modules. Thus, the fellow was successfully integrated in the day-by-day workflow of the department, gaining first-hand practical experience in risk assessment in a multicultural and interdisciplinary context. This enabled a productive exchange of good practices and contributed to building a European risk assessment community.

Keywords: cyanotoxins, anthocyanins, EU-FORA, nanomaterials, nutrition, risk assessment, toxicology

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1. Introduction

The European Food Risk Assessment Fellowship (EU-FORA) Programme was created by EFSA to build a European pool of expert risk assessors and a knowledge community. It is intended for scientists with less than 15 years of experience in various fields: biology, chemistry, veterinary or human medicine, food technology, toxicology, agricultural or environmental science and offers dedicated training and hands-on experience in chemical and microbiological risk assessment (Bronzwaer et al., 2016).

The training programme 'Risk assessment methodologies in the field of contaminants, food contact materials, technological ingredients and nutritional risks' was developed and implemented at the Department of Nutrition and Bromatology, Toxicology and Legal Medicine (DNBTLM) Faculty of Pharmacy, Universidad de Sevilla (US), as hosting site, under the supervision of Profs. Angeles Jos and Ana Mª Troncoso. The team at the DNBTLM has extensive expertise in the field of experimental toxicology; nanomaterials; nutrition; risk assessment of food contaminants; and assessment of nutritional and health claims. Thus, they were able to offer a complementary insight and transferable practical skills to the EU-FORA fellow who had a very different background – food technology – and was coming from a life science university as sending organisation (University of Agricultural Sciences and Veterinary Medicine Cluj-Napoca, Romania).

The programme was designed starting from methodologies and guidelines proposed by EFSA and adapted to the expertise of the team at DNBTLM. The objectives of the training were considered in relation to the two groups at DNBTLM: Area of Toxicology and Area of Nutrition and Food Science, while the activities were grouped in modules based on common intended outcomes. A 'learning by doing' approach was consistent for all the modules.

2. Description of work programme

2.1. Aims

The aim of the working programme proposed by the DNBTLM was to train the EU-FORA fellow in the risk assessment methodologies and guidelines proposed by EFSA in the field of contaminants, food contact materials, technological ingredients and nutritional risks. To achieve this purpose, six learning objectives were formulated: (1) to efficiently use the main information sources and databases useful for risk assessment purposes; (2) to be able to identify the hazards of main concern for chemical and nutritional risk assessment; (3) to acquire substantial knowledge on the different phases of the food safety risk assessment of contaminants, food contact materials, technological ingredients and nutritional risks; (4) to acquire practical skills in experimental toxicology (*in vitro* assays, genotoxicity assays, analytical determinations etc.) for hazard characterisation; (5) to be able to interpret and discuss experimental results in a risk assessment perspective; (6) to gain oral and written communication skills to present scientific and risk assessment results. The working programme had a modular 'learning by doing' approach to meet the six objectives. For each of the four modules, a balance between interactive learning/case studies and theory was guaranteed.

2.2. Module 1 – Insight into chemical food safety risk assessment

Module 1 was focused on chemical risk assessment and included both activities designed to deepen the theoretical knowledge of the fellow and 'learning by doing' case studies. These activities were designed to provide the fellow a proper insight into the four parts of chemical risk assessment and confer transferable skills that will enable the proper application of the framework to any other chemicals.

The fellow benefited from an individual introductory session with the supervisor prof. Jos at the beginning of the fellowship which complemented the initial training at EFSA. The goal was to discuss and clarify aspects of the chemical risk assessment; EFSA's framework; and reliable scientific sources and databases (EFSA guidelines, OCDE protocols, European Commission, IARC, Codex Alimentarius) for risk assessment purposes. During the whole year, the fellow was engaged in meetings and discussions with the team of experts from the DNBTLM for a deeper insight on particular topics of the risk assessment of contaminants. In addition, she attended two undergraduate courses from the Degree in Pharmacy curricula: Food Safety; Toxicology and one course in Technological ingredients for food processing from the line of study on Food and Health included in Professional Master's in Pharmacy. The courses provided a complementary overview to the fellow's background in food technology.

The hands-on experience on chemical food safety risk assessment consisted of three case studies that went from a simple exercise on an already published official opinion, to working in a team with experts with the aim of producing a new opinion to an individual work under the guidance of the supervisor prof. Jos with the aim of obtaining a publishable review manuscript.

The first activity was an in-depth study of EFSA's opinion on acrylamide in food (EFSA, 2015) because it presented all the steps in the risk assessment framework of contaminants, with detailed methodological explanation that could be easily followed and replicated. In addition, the management approach is detailed in the Commission Regulation (EU) 2017/2158 establishing mitigation measures and benchmark levels for the reduction of acrylamide in food (Commission Regulation, 2017).

The case study was valorised by two lectures given by the fellow to the participants of two workshops organised by the Area of Toxicology from the DNBTLM, US (Table 1). The attendance to the workshops provided complementary information for the chemical risk assessment and a proper background for debating and networking activities. Following her participation at the IIIrd Workshop on Toxicology, the fellow co-authored a poster on creative education in Pharmacy (Table 1).

The second case study involved the fellow in the workings of the Scientific Committee of the Spanish Agency for Food Safety and Nutrition (AESAN) on emerging risks. The chemical hazards of interest in food safety in Spain were already identified by the Committee, and the fellow was assigned to work on the emerging *Fusarium* mycotoxins enniatins (ENNs) and nivalenol (NIV); and pyrrolizidine alkaloids (PAs). ENNs and NIV were of interest because either they are not yet regulated, and/or they can co-occur with other mycotoxins (CONTAM Panel, 2014, 2017a), while PAs are a large group of plant secondary metabolites highly toxic to humans and animals (CONTAM Panel, 2017b). A literature search was conducted to identify relevant scientific data on ENNs, NIV and PAs relevant for food safety in Spain. The results and the conclusion obtained under the supervision of prof. Jos, the coordinator of the working group, were approved by the Committee and published in a report on emerging chemical hazards of concern in Spain (Jos et al., 2018) (Table 1).

The third case study required the fellow to apply EFSA's guidance on risk assessment of the application of nanoscience and nanotechnologies in the food and feed chain covering mainly the human exposure via the oral route (EFSA Scientific Committee, 2018) on a nanomaterial used in food contact materials. The aim of the activity was to develop a scientific opinion in the form of a publishable manuscript in a peer-reviewed scientific journal. The topic was chosen because of the safety concerns associated with the characteristics of nanomaterials related to their nanoscale, both physicochemical (size, chemical composition, aspect ratio, surface properties, crystallinity, solubility, clustering properties) and biological (particle–cell interactions, mobility, persistence in the body, bioavailability). The guidelines are recent and there is no concern of producing a research paper over imposing with other current publication. In addition, the supervisor prof. Jos has ample expertise in the toxicology and safety of food contact materials and can proficiently guide the fellow. A systematic search of the scientific literature was performed using Web of Science, Science Direct, PubMed and PubChem electronic databases. The scientific data were extracted and structured according to EFSA's three-step framework proposed for hazard identification and characterisation of nanoparticles with a preliminary step (Step 0) that firstly determines the existence of nanospecific properties (EFSA Scientific Committee, 2018). In Step 0, data were collected and evaluated to determine the *in vitro* rate of degradation of the nanomaterial to the non-nanomaterial under representative gastrointestinal tract conditions. In Step 1, a literature review was performed on the carcinogenicity, mutagenicity, reprotoxicology of the nanomaterial. The aim was to either provide sufficient information to perform the risk assessment or to identify key toxicological aspects needed to be explored in the *in vivo* oral study (Step 2). Step 2 comprises of the review of data on *in vivo* studies: pilot 14-day studies aimed at dose finding and assessment of absorption, tissue distribution and accumulation, elimination phase (Step 2a) and modified 90-day toxicity tests in rodents with emphasis on liver, brain, testis and spleen (Step 2b). The goal was to identify a reference point: lower boundary of the benchmark dose confidence interval (BMDL) or a no observed adverse effect level (NOAEL). Step 3 consisted of the reviewing of in-depth targeted studies (long-term exposure toxicokinetic studies, neurotoxicity, immunotoxicity or endocrine-mediated effects) designed to decreasing the uncertainty of the risk assessment. In addition, data were collected on the effects on the gut microbiome as the nanomaterial showed antimicrobial activity. Partial results of the study will be presented in a poster at the XXIIIrd Spanish Congress of Toxicology at the end of June 2019 (Table 1).

A valuable insight into the field of nanotechnology was offered by the participation at the Workshop on Nanotechnology in Food Industry organised by the National Network of Food Nanotechnology and the Platform Food4Life Spain at the Ministry of Science in Madrid in the 14 March 2019.

2.3. Module 2 – Experimental toxicology in risk assessment

Module 2 was focused on a 'hands-on' training in experimental toxicology offered by the Area of Toxicology from the DNBTLM, to complement the fellow's background. The aim of the module was to create a sound toxicological basis and to enable her to appropriately and critically evaluate toxicological studies for risk assessment.

Similar to module 1, the fellow was systematically engaged into meetings and discussions with the supervisor prof. Jos and the team of experts from the DNBTLM for a deeper insight into experimental toxicology: protocols; methods and assays; doses and administration routes; analysing of samples and data interpretation in the framework of risk assessment. The attendance to the undergraduate course on Toxicology completed her theoretical training.

The fellow joined the team working on the toxicity of cyanotoxins, mainly cylindrospermopsin (CYN) and mixtures of microcystins (MCs) and CYN. This topic was chosen because these toxins pose rising health-related concerns because of the increasing spread and frequency of cyanobacteria blooms caused by to eutrophication and climate change (Buratti et al., 2017). Humans may be exposed to cyanotoxins via many routes, but the oral exposure by contaminated water and foods (mainly fish, sea food and vegetables) is prevalent (Testai et al., 2016).

CYN is considered by an External scientific EFSA Report (Testai et al., 2016) as a potential emerging risk because of its cytotoxicity, neurotoxicity, pro-genotoxicity and potential carcinogenicity (Puerto et al., 2018). However, the mode of action is not yet fully understood, mainly in respect to its genotoxicity, which is a key point in the risk assessment. Some *in vitro* studies suggested genotoxicity due to DNA fragmentation, mediated by previous metabolism (Puerto et al., 2018). In addition, EFSA recommended the performing of new *in vivo* genotoxicity studies of CYN because previous results were either inconclusive and contradictory (Testai et al., 2016). Thus, the fellow was involved in the study of CYN genotoxicity *in vivo* following the EFSA guidelines on genotoxicity testing (EFSA Scientific Committee, 2011). Rats were used as experimental model and the induced potential genotoxic effects were evaluated after oral administration of pure CYN standard, by application of a combined micronucleus (MN)-comet assay following OECD 474 (OECD, 2016a) and OECD 489 (OECD, 2016b) guidelines. The fellow gained experience in working with both EFSA and OECD guidelines on genotoxicity testing and was trained in the using of OLYMPUS BX61 microscope for MN assay; the microscope together with the CometAssay IV software for comet assay; and Graph-Pad InStat software for the statistical analysis of the results. In addition, there was an active exchange of best practices in scientific reference and data management between the fellow and the team of experts at the DNBTLM. Some of the results of this experiment were already presented in two posters (Table 1) and included in a manuscript co-authored by the fellow: '*In vivo* genotoxicity evaluation of Cylindrospermopsin in rats using a combined micronucleus and comet assay' currently under review for a peer-reviewed journal. In addition, results will be presented as an additional poster at the EUROTOX2019 Congress in September (Table 1).

CYN and MCs frequently co-occur (Testai et al., 2016) and mixtures are a more probable exposure scenario. In addition, the toxic effects may be different than those observed for single cyanotoxins. An External scientific EFSA Report has concluded that more data are needed on the toxicity of cyanotoxin mixtures (Testai et al., 2016). The fellow is involved in a second ongoing *in vivo* genotoxicity testing on mixtures of CYN and MCs, following the previously protocol described. Partial results will be presented as a poster at the XXIIIrd Spanish Congress of Toxicology at the end of June 2019.

The same External scientific EFSA Report (Testai et al., 2016) showed the need for new analytical methods for sample preparation, routine detection and quantification of CYN and mixtures of MCs – CYN in complex matrices for both control procedures and risk assessment. Thus, the fellow participated in the activities related to the development and optimisation of a method to determine the content of CYN and MCs – CYN in mussel matrix based on a solid-phase extraction method and quantification by ultra-performance liquid chromatography–tandem mass spectrometry previously optimised and validated by the team at DNBTLM (Diez-Quijada et al., 2018; Prieto et al., 2018). The fellow gained a close insight into the developing and validating of analytical methods with suitable limit of quantification (LOQ) and limit of detection (LOD) for risk assessment purposes. The fellow was proactively involved in the design of the experiment for the optimisation study by carrying out the response surface methodology.

The fellow had the opportunity to be involved in a biotransformation and bioaccessibility study under simulated digestion. The experiment was an *in vitro* digestion model, including salivary; gastric

and duodenal phases; and colonic fermentation under lactic acid bacteria (Maisanaba et al., 2018). Thus, the fellow had an insight into the protocol, design of experiments and data interpretation that conferred skills to critically evaluate simulated digestion studies for risk assessment purposes.

In addition, both the fellow and the team from Area of Toxicology from the DNBTLM have already undergone steps in the continuing of their collaboration: the fellow applied for funding for a future mobility to the US after the ending of the fellowship, while the members of Area of Toxicology included the fellow as an external collaborator in a funding proposal.

2.4. Module 3 – Insight into nutritional risk assessment

In module 3, the Area of Nutrition and Food Science of the DNBTLM offered training in exposure assessment, nutritional risk assessment and assessment of nutritional and health claims. Similar to the previous two modules engaged the fellow in both theoretical and practical aspects of: food composition and food consumption databases; novel foods; dietary reference values; nutrients bioavailability; biomarkers; mechanisms of action of bioactive compounds; nutritional and health claims.

The fellow participated in discussions on specific characteristics of the nutritional risk assessment with the supervisor prof. Troncoso and the team of experts from the DNBTLM. In addition, she attended the undergraduate course from the Degree in Pharmacy curricula on Nutrition, Dietetics and Dietotherapy and two Master courses of the line of study of Food and Health included in Professional Master's in Pharmacy: Bioactive compounds and functional foods; Nutritional risk assessment. During the courses the fellow received a complementary theoretic background, and practical skills during the seminars and practical courses on: evaluating and designing diets; working with food consumption database; assessing nutritional labels and health claims of foodstuffs; identifying and quantifying biomarkers in human samples. The fellow had the opportunity to engage with both Spanish and international Erasmus students and scholars during the group activities, contributing to her working experience in multicultural environments and sharing of best practices.

In addition, the fellow collaborated with the experts from the Area of Nutrition and Food Science of the DNBTLM in their ongoing experimental work in the field of chemical characterisation, biomarkers and mechanisms of bioactive compounds. In particular, she was involved in two experiments.

The first study dealt with the assessment of health protective properties of dietary polyphenols consumed as part of human diets in cardiovascular diseases. The topic was chosen because the scientific evidence is the most robust compared to the multitude of other biological activities demonstrated *in vitro*. In most of the *in vitro* studies, the bioactivity was observed in supra-physiological concentrations and/or for non-physiological compounds and the bioactivity remained theoretical with no demonstrated *in vivo* mechanisms (Cerezo et al., 2015). The study aimed to identify the molecular mechanism of polyphenols (melatonin and hydroxytyrosol) related to the inhibition of angiogenesis, a process involved in the development and destabilisation of atherosclerotic plaques. The vascular endothelial growth factor (VEGF) – the most important pro-angiogenic growth factor in humans – exerts its angiogenic effects by stimulating VEGF receptor 2 (VEGFR-2) (Cerezo et al., 2015). The inhibition of VEGF-induced VEGFR-2 activity was tested on human umbilical vein endothelial cells (HUVECs). The fellow participated in the preparing and treatment of HUVECs; protein content determination; phosphorylated VEGFR-2 in lysates measurement by ELISA; and Western blot analysis for VEGR-2 endothelial nitric oxide synthase (eNOS) following the procedures described by (Cerezo et al., 2015, 2017; Moyle et al., 2015).

The second study focused on the nutritional value of anthocyanins extracted from blueberries because of the growing scientific and economic interest in their potential beneficial effects on human health. Anthocyanins are water-soluble plant pigments found in red, blue or purple fruits and vegetables. They are present predominantly in the skin of the fruit, but in berries, they are present in both the skin and flesh (Cassidy, 2018). They are glycosides of anthocyanidins and only six of them seem to be relevant to the human diet (cyanidin, delphinidin, malvidin, pelargonidin, peonidin, petunidin) (Cassidy, 2018). However, the anthocyanin content and profile vary widely with growing and storage condition; thus, it is of interest to evaluate local varieties. Thus, the aim of the study was to determine the content of anthocyanins from four Spanish blueberry varieties; to identify the anthocyanin profile; and to investigate the *in vitro* bioactivity in relation to EFSA's Guidance for the scientific requirements for health claims related to antioxidants, oxidative damage and cardiovascular health (EFSA Panel on Dietetic Products Nutrition, 2018). The anthocyanin fraction was prepared and the antioxidant activity was performed (Cerezo et al., 2010). The experiment is ongoing, and the total anthocyanin content and determination and quantification of the individual anthocyanin compounds will follow shortly. Other

assays related to their antibacterial activity will be performed in the fellow's home institution, as this experiment builds on her previous experience in the study of anthocyanins and their health benefits. The results of the case study are to be included in a scientific manuscript co-authored by researchers from both sending and hosting institution, contributing to the continuous co-operation.

In addition, the fellow was invited to attend the Workshop 'Towards the search for new bioactive properties in fermented foods' organised by the Area of Nutrition and Food Science of the DNBTLM on 24 May 2019 with the aim of completing her theoretical and practical experience and contributing to the extension of her scientific network.

The fellow gained practical experience with the testing of bioactive compounds for their bioactivity, bioavailability and mechanisms of action. This contributed to the development of a critical perspective on studies needed to assess nutritional and health claims in relation to the design of experiment, methods used, interpretation of results and extrapolation for human relevance. In addition, a productive exchange of good practices took place between the fellow and the experts of the DNBTLM in relation to laboratory practices and data processing. Moreover, the fellow shared her personal practices in references management in a seminar on EndNote reference manager software (Table 1).

2.5. Module 4 – Scientific risk communication

Module 4 was designed to provide a 'hand-on experience' in scientific risk communication. The fellow was encouraged and supported in the participation at local, national and international workshops and congresses (Table 1). Thus, all the case studies used for the training of the fellow were materialised into different forms of scientific communication: lectures, posters oral, presentations, scientific opinions and research manuscripts.

In addition, the fellow was able to gain experience by being involved in the organisation of a scientific congress as the DNBTLM was the host of the XXIII Spanish and VII Iberoamerican Congress of Toxicology, organised by the Spanish Association of Toxicology (AETOX) and the Area of Toxicology of the Faculty of Pharmacy of the US in 26-28.06.2019.

Table 1: Scientific communications delivered or co-authored by the EU-FORA fellow

Date of communication	Type of communication	Scientific context	Title of communication
19.11.2018	lecture	VIIIth Workshop on Food Safety: Risk assessment, [VIII Jornadas de Seguridad Alimentaria: Análisis de Riesgos], DNBTLM, US, Spain	Acrylamide formation in foods during thermal processing
28.11.2018	scientific report	Revista del Comité Científico de la AESAN	Report of the Scientific Committee of the Spanish Agency for Food Safety and Nutrition (AESAN) on the prospection of chemical hazards of interest in food safety in Spain
2-5.04.2019	poster	European Biophysical Societies' Association EBSA22 Conference Bucharest, Romania	*In vivo* genotoxicity of cylindrospermopsin by the comet assay
26.04.2019	lecture	IIIrd Workshop on Toxicology and Society: Abuse drugs and molecular toxicology (Forum and toxicology colloquium), [III Jornadas de Toxicología y Sociedad: Drogas de Abuso y Toxicología Molecular (Foro y Olimpiadas de Toxicología)], DNBTLM, US, Spain	Acrylamide formation in foods during thermal processing
5-10.05.2019	poster	The 11th International Conference on Toxic cyanobacteria, in Krakow, Poland	DNA damage induced by cylindrospermopsin in rats

Date of communication	Type of communication	Scientific context	Title of communication
15-17.05.2019	poster	The 25th European Association of Faculties of Pharmacy (EAFP) Annual Conference 2019: Creative education: Towards competences in patient-oriented pharmacy education, Krakow, Poland	Celebration of the III Meeting on Toxicology and Society: Drugs of abuse and molecular toxicology (Toxicology Forum and olympiad)
11.06.2019	seminar	DNBTLM, US, Spain	Overview of using reference management software: EndNote Case Study
26-28.06.2019	oral presentation	XXIIIrd Spanish and VIIth Iberoamerican Congress of Toxicology; section: toxicology education, [XXIII Congreso Español de Toxicología y VII Iberoamericano; sección de Educación en Toxicología], US, Spain	Experience of an European food risk assessment (EU-FORA) fellow at the University of Seville
26-28.06.2019	poster	XXIIIrd Spanish and VIIth Iberoamerican Congress of Toxicology; section: food safety, [XXIII Congreso Español de Toxicología y VII Iberoamericano; sección de Educación en Toxicología], US, Spain, sección de Seguridad Alimentaria, US, Spain	EFSA Scientific Committee's (2018) stepwise framework for nano-related hazard identification and characterisation in food and feed
	poster		Analysis of DNA damage in rats by simultaneous exposure to cylindrospermopsin and microcystin-LR [Análisis del daño en el ADN en ratas tras la exposición simultánea a cilindrospermopsina y microcistina-LR]
8-11.09.2019	poster	55th Congress of the European Societies of Toxicology, EUROTOX2019: Toxicology – Science Providing Solutions, Helsinki, Finland	Cylindrospermopsin induces genotoxic damage in rats by the comet and micronucleus tests

3. Conclusions

The working programme 'Risk assessment methodologies in the field of contaminants, food contact materials, technological ingredients and nutritional risks' managed by the DNBTLM, US, employed a modular 'learning by doing' approach. The four modules (chemical risk assessment; experimental toxicology; nutritional risk assessment and risk communication) included activities that successfully trained the EU-FORA fellow in: the efficient use of scientific databases for risk assessment; identifying hazards; using the food safety risk assessment framework for contaminants, food contact materials, technological ingredients and nutritional risks; acquiring practical skills in experimental toxicology; experimental data interpretation for risk assessment; and communication of risk assessment results.

This is supported by the outcomes of the working programme: two lectures on risk assessment in thematic workshops; co-authoring as an external collaborator of a published Report of the Scientific Committee of the Spanish Agency for Food Safety and Nutrition (AESAN) on the prospection of chemical hazards of interest in food safety in Spain; six posters at international conferences on risk assessment topics; one oral presentation at an international conference on risk assessment training; a submitted manuscript to a peer-reviewed journal and other papers on preparation; and participation at four thematic workshops.

The activities were designed to provide the fellow a proper insight into risk assessment and confer transferable skills. In addition, she was successfully integrated in the day-by-day workflow of the department, gaining first-hand experience. The fellow attended both undergraduate and Master courses that were complementary to the fellow's background in food science and technology. This created a good environment for professional and social interaction between the team at the DNBTLM, the fellow and the students. It helped with the integration of the fellow in the daily routine of the DNBTLM, but also provided a multicultural and interdisciplinary context. This enabled a fruitful

exchange of good practices in teaching, laboratory and communicating in a higher education environment in general, and risk assessment in particular.

Thus, the professional and pleasant working environment of the DNBTLM has guaranteed the success of the EU-FORA training programme. It has set the stage for future collaboration between the fellow and her home institution, on one side, and the team at DNBTLM and the hosting institution, on the other side, such as common project proposals that currently under evaluation; opinion and research manuscripts under preparation and future European funding opportunities accessed in consortium. This contributes significantly to building a European risk assessment community by engaging in common goals and using harmonised practices.

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Abbreviations

AESAN	Spanish Agency for Food Safety and Nutrition
BMDL	Benchmark dose confidence interval
CYN	Cylindrospermopsin
DNBTLM	Department of Nutrition and Bromatology, Toxicology and Legal Medicine
ENNs	Enniatins
eNOS	Endothelial nitric oxide synthase
HUVEC	Human umbilical vein endothelial cells
LOD	Limit of detection
LOQ	Limit of quantification
MCs	Microcystin
MN	Micronucleus
NIV	Nivalenol
NOAEL	No observed adverse effect level
OECD	Organization of Economic Cooperation and Development
PAs	Pyrrolizidine alkaloids
US	Universidad de Sevilla
VEGF	Vascular endothelial growth factor
VEGFR-2	Vascular endothelial growth factor receptor 2