

## Report on the outcomes of a Short-Term Scientific Mission<sup>1</sup>

Action number: CA18221

Grantee name: Bárbara Santos

### **Details of the STSM**

Title: *Review on the endpoints used to assess the ecotoxicity of pesticides to reptiles needed to improve animal welfare when making risk assessment*

Start and end date: 18/07/2022 to 28/08/2022

### **Description of the work carried out during the STSM**

Description of the activities carried out during the STSM. Any deviations from the initial working plan shall also be described in this section.

*(max. 500 words)*

I spent 6 weeks at the National Institute of Biology, where I worked together with Dr Anamarija Žagar during meetings and discussions and independently when searching for literature and analysing papers. The first task aimed to do the bibliographic compilation. I revised the available literature from Web of Science and Scopus in the period between February 2016 and July 2022 using pre-selected search keywords related to two main areas: reptiles and pesticides. The period after February 2016 was selected to follow-up on the previous revision of literature done by Ortiz-Santaliestra and colleagues (1) In total, 138 publications since February 2016 until the present date were analysed and included in the database. The database was also merged with the database of Ortiz-Santaliestra et al. (1) that included 53 publications on reptiles and pesticides up to February 2016. The second task intended to provide a review on the main endpoints used. For this task, I read all papers and searched for type of endpoints selected in each study and indicating the category group they belonged (lethal or non-lethal, invasive or non-invasive, chronic or acute). From the preliminary assessment it is clear that lethal endpoints are mostly used, but comparing with other groups such as amphibians, there is still a gap in these studies in reptiles. Moreover, there has been an increase on field surveys, mainly on evaluation bioaccumulation of pesticides in tissue samples from lizards, crocodiles and turtles. The third task aimed to do a revision of the experimental approaches used in these studies. Preliminary analysis showed that since the reptiles can be exposed to environmental contaminants such as pesticides by different pathways (oral, dermal, maternal transfer), different exposure routes were used in studies and this can be further analysed to

<sup>1</sup> This report is submitted by the grantee to the Action MC for approval and for claiming payment of the awarded grant. The Grant Awarding Coordinator coordinates the evaluation of this report on behalf of the Action MC and instructs the GH for payment of the Grant.

define the study gaps. However, few studies have attempt to compare several exposure routes in the same study. For task 4, the aim was to compile a list of non-invasive methods and endpoints used. From the preliminary analysis, it is clear that non-lethal parameters are still poorly applied. Moreover, other approaches, such as, non-invasive methods (cloacal swabs, buccal swabs, pathogens load, faeces, diet assessment) are even rarer. The aim of the last task was to compile a list of research needs in order to proper include risk assessment of pesticides for reptiles and at the same time follow the implementation of the 3Rs (replacement, reduction and refinement). Due to the short period of grant work (6 weeks) it was not possible to yet analyse this data but the database is ready and this work is planned to be continued in the future.

(1) *Ortiz-Santaliestra, et al., 2017. EFSA Supporting publication EN-1251.*

### **Description of the STSM main achievements and planned follow-up activities**

Description and assessment of whether the STSM achieved its planned goals and expected outcomes, including specific contribution to Action objective and deliverables, or publications resulting from the STSM. Agreed plans for future follow-up collaborations shall also be described in this section.

*(max. 500 words)*

The grant period of 6 weeks allowed the elaboration of a complete dataset of publications on ecotoxicological studies with reptiles, with a focus on pesticides. This dataset represents the most up to date information table on relevant endpoints used in ecotoxicological studies (Action's 2<sup>nd</sup> objective). With this dataset it was already possible to make a preliminary analysis and to identify methodological gaps in risk assessment of pesticides for reptiles. For example, they are mostly focused on lizards, and a great part of the publications used the same lizard species. Some of the studies also use captive populations to assess pesticide effects which can be used as a control to compare to natural environment. Within the lizards, most publications used often only males which does not allow for a complete assessment of the toxic effects at the population level since females can respond differently. Often the studies only use one exposure pathway (oral or dermal) which also does not allow for comparison of effects and to compare to natural scenarios where individuals are often exposed to pesticides through more than one exposure route. Due to time-constraints, only very preliminary analysis was made and the plan is to follow-up with a more elaborated statistical analysis of the data compiled here in the future. Among the questions to be addressed, it is important to compare the sensitivity of different species of reptiles in order to define the most sensitive ones that could be used as representative of the taxonomic group. It is clear that there is a lack of studies in snakes and crocodiles, two groups that have lately been study using mostly bioaccumulation data but that could profit from the application of non-invasive and non-lethal methods to address pesticide toxicity due to their biological and ecological restrictions. By using these sub-lethal and non-invasive endpoints, (already used in a few studies included in this review), it will allow to minimize animal use (Action's 3<sup>rd</sup> objective). Further studies should compare in the same species, the use of lethal / non-lethal and non-invasive parameters to assess the possibility of using the latter as representative and ecological relevant in ecotoxicology assays.