

# Need for a sustainable use of medicinal products: environmental impacts of ivermectin

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## Abstract

It is worldwide recognized that the use of pharmaceuticals for human and veterinary purposes could lead to unsustainable effects on the environment. A strategy to reduce the impact of pharmaceuticals on the environment has been recently established at European level, where guidelines to evaluate the impacts of veterinary drugs used to treat animal diseases are in place. The aim of this article is to focus on the worldwide used antiparasitic drug ivermectin (IVM) and its potential impact on the environment. A specific section is related to the IVM resistance that the massive use of this drug could generate enhancing the risk scenarios also for human health. The application of stringent measures for the veterinary use of this substance, in line with the recommendations provided by International frameworks such as One Health and EcoHealth, is recommended.

## Key words

- environment
- ecotoxicity
- resistance
- antiparasitic drug
- ivermectin

## INTRODUCTION

The treatment of many human and animal diseases relies on access to effective pharmaceuticals. At the same time, the environmental pollution caused by some pharmaceuticals is an emerging problem because of their residues discharged and released in the ecosystems during the manufacture, use and disposal. The evidence of risks for the environment is well-documented [1].

The communication about the pharmaceutical strategy on the environment supports the aim of the European Commission (EC) (COM/2019/128 final) to deliver a sustainable Europe by 2030, according to the United Nations (UN) Sustainable Development Goals (SDGs). The strategy has been established in the context of the EU Water Framework Directive that has the aim to achieve a good chemical and ecological status for all water resources. Furthermore, this strategy is a component of the Union's One Health Action Plan against Antimicrobial Resistance.

The largest source of pharmaceuticals entering the environment is represented by their use. Moreover, the general pressure of pharmaceuticals will increase due to demographic developments (e.g. increasing population density and age) and due to climate changes, an insufficient constant dilution of pharmaceutical run offs and effluents can be expected in many areas. The

route of exposure likely differs depending on whether human or veterinary use is involved. However, in some cases, up to 90% of the active ingredient is excreted (or washed off) in its original form due to the stability and the adsorption of the chemical substance. The release of veterinary drugs in the environment tends to come from untreated diffuse sources such as the spreading of manure. In this respect, the evaluation of the environmental risk of veterinary medicinal products within marketing authorisation procedures has been discussed in European Union (EU) since the mid-1990. Moreover, a first guidance document defining how to perform the environmental risk assessment (ERA) was prepared by the European Medicines Agency (EMA) in 1997. For the EU, additional guidance in support of the guidelines was provided by EMA in 2008 [2] and further revisions have been provided in 2019. In this guidance, predicted environmental concentrations (PECs) are estimated based on the dose and frequency of the product applied. If the PEC exceeds the trigger value of 100 µg/kg dry weight in the soil for intensively reared and pasture animals, studies on environmental fate and effects on selected non-target species have to be performed during the phase II. In parallel if a PEC in water compartments exceeds 100 ng/L it triggers additional risk assessment steps. In the same phase, the environmental risk is deterministically characterized by

comparing the PECs with the predicted no-effect concentrations (PNECs) in several environmental compartments.

Ivermectin (IVM) belongs to the avermectins and these are widespread used antiparasitic medical products representing a serious potential threat for the environment [3]. They have been used for several purposes for example in agriculture and horticulture for the protection of fruit, cotton, vegetables, and ornamental, thanks to their effectiveness against a wide range of nematodes, mites, and insects. Among the avermectins, IVM is mainly used for controlling livestock parasites. With over 5 billion doses worldwide sold since its market introduction in the early 1980s, this veterinary medical product has become the most widely used antiparasitic drug. It is used regularly as a pesticide for cattle, pigs, sheep, horses, and dogs. Its use is effective against internal parasites, such as gastrointestinal and pulmonary nematodes, and external parasites, such as mange mites and blood lice. IVM can be administered orally, topically or intramuscularly. Oral applications tend to result in sharp excretion peaks, with most of the dose excreted over a few days.

In connection with the excretion pathways, IVM and its metabolites were excreted mainly in faeces (90%) and only 1% in urine. Other authors who tried to determine IVM in urine did not find the parent drug nor its metabolites. Bile is the main route of excretion.

Peak elimination of injectable or topical formulations usually occurs within 2 to 7 d post-treatment, followed by a long tail that may sustain for more than 4 to 6 weeks, whereas peak elimination levels of sustained-release formulations may occur over several weeks post-treatment.

Already a decade ago, Liebig et al. [4] have demonstrated that, with regard to its environmental aspects, IVM is a substance of high concern. The environmental risk assessment of IVM was mainly performed according to the mentioned international and European guidelines, using a large number of new data on its fate and effects, and additional results from 2-species, multispecies, semifield, and field studies. This case study has clearly demonstrated unacceptable risks for living organisms (e.g., for daphnids and dung invertebrates), and the authors suggested the necessity of reassessing

IVM containing veterinary medicinal products. Moreover, when it is used in crop protection, there are evidence of ecotoxicity for non-target species, with insects generally and bees in particular being at the highest risk [5]. For human use ivermectin (IVM) has also been shown to be effective in *in vitro* tests against a broad range of viruses, including HIV, Dengue, Influenza, and Zika. Recently, a collaborative study led by the Monash Australian University and published in the journal *Antiviral Research* has showed that a single dose of IVM could stop the COVID-19 virus growing in cell cultures. The next step of this research will aim to evaluate the correct dosage [6].

## IVERMECTIN EFFECTS ON AQUATIC AND TERRESTRIAL ORGANISMS

### *Aquatic invertebrates*

Notable effects of IVM have been reported on various aquatic invertebrate species such as *Daphnia magna* and *Gammarus pulex*. A 2007 study [7] observed the extreme sensitivity of *D. magna* to this drug. 10 acute tests were carried out and a 48 h LC<sub>50</sub> of 5.7 ng/L (Table 1) was found, while chronic tests revealed high toxicity on reproduction and growth rate even at lower concentrations. The nominal LOEC obtained was 0.001 ng/L and the NOEC 0.0003 ng/L. These values are analogue to those of extremely toxic compounds that are classified as priority hazardous substances by the Water Framework Directive. Among the Amphipods, a wide order of crustaceans, high sensitivity to IVM has been reported in particular for *Gammarus pulex* and *Gammarus fossarum* [8]. A 2018 study [9] observed a 100% mortality in young American lobsters (*Homarus americanus*) exposed to ivermectin-treated sediment (61.0 and 300.0 ng/g sediment) for a 25 days period, while a prolonged exposure to lower concentrations (0-3.0, 6.4 and 14.7 ng/g sediment) induced sublethal effects IVM shows a very high acute and chronic toxicity to crustaceans in the 5.7 ng/L and 0.3 pg/L range and this should be considered in future risk management measures. At these levels no robust analytical controls can be expected. Moreover, such a very high toxicity is indicating the need of reducing any aquatic exposure of IVM to very low levels, in order to protect a wide range of aquatic invertebrates.

**Table 1**

Comparison between the effect concentrations and Initial PEC values in water and dung [4]

Compartment	Species	Effect concentration	PEC (best/worst case)
Water	<i>Daphnia magna</i>	EC <sub>50</sub> 5.7 ng/L	0.1/7.2 (IR)
		PNEC 0.0057 ng/L (acute)	0.2/2.5 (P)
		PNEC 0.00003 ng/L (chronic)	83/523 (P) ng/L
	<i>Oncorhynchus mykiss</i>	LC <sub>50</sub> 3.0 µg/L	0.1/7.2 (IR)
PNEC 3.0 ng/L		0.2/2.5 (P) 83/523 (P) ng/L	
Dung	<i>Musca autumnalis</i>	EC <sub>50(emerg.rate)</sub> 4.65 µg/kg dung fresh wt	4.8/12.7 (P) mg/kg dung fresh wt
	<i>Aphodius constans</i>	LC <sub>50</sub> 176 µg/kg dung fresh wt	4.8/12.7 (P) mg/kg dung fresh wt

PEC: predicted environmental concentrations; IR: intensively reared animals; P: pasture animals.

### Fishes

Many studies over the years focused on the consequences of the IVM use on seawater fishes: this drug is indeed commonly used to treat sea lice infestations in aquaculture, mainly in Atlantic salmon (*Salmo salar*) farms. Sea lice (*Lepeophtheirus sp.* and *Caligus sp.*) are marine ectoparasitic copepods able to induce serious health issues to fishes and consequently significant economic losses. In a 2012 [10] study, Uacán-Marín *et al.* observed several toxicological effects in salmon exposed to IVM through diet at similar concentrations to those currently used in aquaculture. The lethal concentration found was 0.174 mg/kg, only three times higher than those used in operational context, and with concentrations slightly higher than those commonly used. Effects such as lethargy, dark skin and reduced feeding behavior were reported. Negative consequences derived from the exposure to IVM were investigated also in freshwater fish. In *Oncorhynchus mykiss* a  $LC_{50}$  of 3.0 µg/L has been detected [4]. In *Danio rerio*, one of the most commonly used organisms for ecotoxicological assays Domingues *et al.* [11] observed the effects produced by a chronic exposure to concentrations between 0.25 and 25 µg/L in adult Danios. While in the acute test a 96 hours  $LC_{50}$  was calculated with higher values, the behavioural patterns of the fish changed even at the lowest concentration of 0.25 µg/L, as they tended to spend more time in the lowest part of the tank. This behavior altered their feeding ability causing a weight loss during the exposure time, more emphasized in males. At the highest concentration (25 µg/L) the effects were lethargy, reduced activity, dark coloration and mild curvature of the spine.

### Algae and plants

The stability and persistence of IVM in sediment and water, especially in wetlands, can cause constant exposure for algae and plants [12]. The toxicity effects of IVM was studied since 1989 by Halley *et al.* Phytotoxicity of IVM on algae and plants is reported in several new studies. In a study aimed to compare the possible effects of IVM on both *D. magna* and *Pseudokirchneriella subcapitata* [13] the algae showed a considerably lower sensitivity: only at the highest concentrations adopted (1250 and 4000 µg/L) a significant effect on algal growth was reported. Eichberg *et al.* [14] reported inhibition of germination in three grassland plant species (*Centaurea jacea*, *Galium verum*, *Plantago lanceolata*) from moxidectin, an antihelmintic drug similar to IVM. Another study, on *Sinapis alba*, was carried out by Vokřál *et al.* in 2019 [15] using a “seed germination and early roots growth” test with two different concentrations, 50 nM (0.044 µg mL<sup>-1</sup>) and 500 nM (0.44 µg/mL). The results of this study showed phytotoxicity for both concentrations, with a 20% growth inhibition for the lowest concentration and 24% for the highest. It is to be underlined that the exposure to a concentration of 44 µg/L can be realistically found in the environment where IVM-treated cattle excrements are present.

### Terrestrial organisms and dung insects

The large use of IVM as an anti-parasitic drug for live-

stock implies its constant release in the environment. In all the possible administration routes, the IVM is in fact largely excreted via faeces, where it can remain for a long period of time. For this reason, many insect species that live in close proximity with dung can be affected by this chemical. In particular, dung beetles tend to excavate dung in order to create brood balls in which they lay their eggs; this kind of behavior has significant positive effects on the environment, as it prevents the accumulation of dung and promotes the fertilization of the soil [16]. The toxicity of IVM on dung beetles has already been observed in several papers over the years [4]. Pecenka *et al.* [17] highlights the negative correlation of ivermectin quantity in cattle pats with dung beetle abundance and diversity. In particular, Martinez *et al.* [18] exposed the dung beetle *Euoniticellus intermedius* to IVM (3.16, 10.0, 31.6, 63.2, 100, and 316 µg/kg fresh dung) in order to determine the toxicity on adults and larvae. After 10 days of exposure, adult females showed a decrease in fecundity. Ishikawa and Iwasa [19] exposed four different species of dung beetle, *C. acutidens*, *O. bivertex*, *O. lenzii* and *P. auratus* to IVM-treated dung; the IVM was applied on cattle by pour-on formulation with the dose of 500 µg/kg of body weight and dung was collected 1, 3, 7, 14 and 21 days post-treatment. In *C. acutidens* a high mortality was observed in adults exposed to dung collected 3- or 7-days post-treatment compared to the control (exposed to non-treated dung). In the same dung, few or no brood balls were reported and in dung from 7- or 14-days post-treatment the emergence rate was significantly lower compared to the control. IVM shows a high toxicity for dung insects in the 3,16-316 µg/kg range of fresh dung, therefore, any terrestrial exposure should be minimized if possible.

### Environmental fate and behaviour

When considering the impact upon ecosystems, the environmental fate of a drug like IVM should be taken into account. The log  $K_{ow}$  of IVM of 3.2 [4] indicates bioaccumulation potential in organisms. Moreover, IVM has the potential to bioaccumulate in *L. variegatus* (biota-sediment accumulation factors ranged from 2.1 to 16.6). Several studies have shown a high persistence of this substance in mesocosms, wetland and terrestrial environment [20]. Dissipation half-lives ( $DT_{50}$ ) in soil can be rather variable depending on soil type, sorption capacity, temperature, and oxygen availability, ranging from 16 to 1520 days [4]. IVM can be also very persistent in mixtures of soil and manure or faeces (7-217 days). Log  $K_{ow}$  have ranges of 3.6-4.2 [4]. As a widely used drug in animal husbandry and farming, IVM frequently occurs in cattle faeces. Movement of IVM from dung to the underlying soil and to the nearby plants has been observed. Concentrations of IVM have been also detected in the roots of macrophytes living in wetlands subjected to different cattle uses in roots and leaves of aquatic plants in a simulated aquaculture ecosystem [21]. Moreover, IVM accumulates in dung, sediment, and water of wetlands, representing a serious threat for such ecosystems. A similar fate of IVM has been assessed in water and sediment of simulated aquaculture [21]. Overall, IVM shows

bioaccumulation and persistence indicating a need of a specific environmental risk management.

### DRUG RESISTANCE IN PARASITIC PROTOZOA

The IVM binding with ligand-gated ion channels, particularly glutamate-gated chloride channels in nematode and insects, cause an influx of negatively charged chloride ions, resulting in hyperpolarisation of synapses and paralysis of organisms. Further, recent studies in other biological systems suggest that IVM can affect additional pathways too [22]. Due to its widespread, use the protozoans developed resistance and it appeared at first time in small ruminants, then in cattle parasites and free-living *Caenorhabditis elegans*. The results of several studies suggested that both target mutation and transport alteration can lead to ivermectin resistance in particular through the changes in mRNA transcription of the ABC transport proteins and the glutathione-related genes [23, 24]. The IVM resistance is a serious problem for parasite control in livestock and there is a concern about resistance development and spread in nematode parasites of humans. Since 2000s, the World Health Organization included parasites in the list of antimicrobial resistant microorganisms together with bacteria, fungi and viruses ([www.who.int/health-topics/antimicrobial-resistance](http://www.who.int/health-topics/antimicrobial-resistance)). Studying and understanding drug resistance mechanisms of these organisms is important in terms of human and animal health.

### CONCLUSIONS

The data summarized in this brief note demonstrate that the veterinary use of IVM can lead to an unacceptable and unsustainable risk for both aquatic and terrestrial ecosystems. IVM shows a very high acute and chronic toxicity to crustaceans. In fish, IVM can cause lethargy, dark skin and reduced feeding behavior. Moreover, growth inhibition for algae and high toxicity for terrestrial organisms and dung insects were underlined. The use of this veterinary medical product, applying the ERA methodology, can cause a high risk. Specifically, for daphnids and dung organisms with a very low PNEC value, indicating a high toxicity level analogue to chemical substances classified as priority hazardous by the European Union. Based on these findings, we

suggest a reduction (or elimination) in the use of this substance as veterinary drug considering the related high risk in particular for aquatic ecosystems. If the use will be authorised more stringent measures should be applied in order to reduce and/or mitigate the impact of IVM on the environment and its organisms. For this reason, IVM should be considered in the context of the EU Water Framework Directive monitoring programmes due to the lack of monitoring data in rivers and aquatic ecosystems. The monitoring data can also improve the knowledge on the role of the environment in the development of resistance in parasitic organism described in the brief note. In some cases, due to the IVM toxicity at very low levels, the use of effect-based methods can be also recommended in areas where the presence of IVM can be predicted in order to detect the real effects on the ecosystems. In conclusion, IVM is a pharmaceutical used worldwide for its properties for the treatment of several diseases in humans and animals, an example is the recent study mentioned in the introduction about the possible use against COVID-19; but for the achievement of SDGs the potential environmental effects of IVM described in this article cannot be neglected. In particular, IVM as a very toxic, persistent and resistance building medical product with high use is maybe one of the most suitable candidates to be assessed in a sustainability strategy. These considerations are also in line with the One Health and EcoHealth approaches that are recommended by several European and international institutions (FAO, WHO, EFSA). In these international frameworks, the environment is strictly linked to human and animal health and every medicinal use authorised, should be sustainable for the environment. Moreover, the main holistic target of this approach should be the overall protection of environmental, animal, and human health.

### Conflict of interest statement

There are no potential conflicts of interest or any financial or personal relationships with other people or organizations that could inappropriately bias conduct and findings of this study.

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