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Ecological Thresholds for a Fenthion-Based Veterinary Pharmaceutical in Tropical Soil Using Species Sensitivity Distribution (SSD) Modeling

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Abstract

Fenthion is a veterinary pharmaceutical (VP) used in the control of ecto- and endoparasites in livestock. This active substance may reach the soil through the animals' urine and feces, but the ecotoxicological data of this compound on non-target soil fauna is limited. This study aimed to evaluate the chronic ecotoxicity of a fenthion-based veterinary formulation on six species of soil invertebrates. Ecotoxicological tests were performed in a natural tropical soil (Entisol) with earthworms *Eisenia andrei*, enchytraeids *Enchytraeus crypticus* and *Enchytraeus bigeminus* and collembolans *Folsomia candida*, *Sinella curviseta* and *Proisotoma minuta*, following ISO protocols. The species sensitivity distribution (SSD) approach was employed to establish protective concentrations (PCs) for fenthion in soil. Collembolans were the most sensitive organisms; the EC50 for *F. candida*, *S. curviseta* and *P. minuta* were 0.72, 0.93 and 0.48 mg kg^{-1}, respectively. Earthworms *E. andrei* presented intermediate sensitivity (EC50 = 28.40 mg kg^{-1}). Enchytraeids were the least sensitive animals (EC50 for *E. crypticus* and *E. bigeminus* of 233.66 and 221.47 mg kg^{-1}, respectively). SSD-derived PCs for 95%, 90%, 80% and 50% of soil species were estimated in 0.017, 0.054, 0.225 and 3.432 mg kg^{-1}, respectively. The predicted environmental concentration (PEC) of fenthion in soil via veterinary use (0.032 mg kg^{-1}) is almost twice the PC95 value (0.017 mg kg^{-1}), indicating potential ecological risk of fenthion to soil species. This study brings new information regarding the ecotoxicological profile and ecological risk of a relevant VP to the edaphic community.

Keywords: springtails; enchytraeids; earthworms; soil fauna; ecotoxicity; ecological risk

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Academic Editor: Ewald Schnug

Received: 21 June 2026
Revised: 29 July 2026
Accepted: 31 July 2026
Published: 3 August 2026

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1. Introduction

Livestock farming represents a major segment of Brazil's agribusiness sector. According to the Brazilian Institute of Geography and Statistics (IBGE) [1], the value of the main livestock products reached R$ 122.4 billion in 2023, with a growth of 5.4% compared to the previous year. In terms of cattle herd size in the country, estimations indicate a total of 227 million animals in 2022 [2,3]. Despite the overall high productivity, the livestock

activity is significantly impaired by parasitism. For cattle, for example, the parasitism by endo- and ectoparasites is estimated to cause annual losses as high as USD 13.9 billion [4].

Parasite control in cattle is carried out through integrated management techniques that utilize chemical products, such as anthelmintics, insecticides and acaricides, playing a central role in the treatment and control of ruminant parasites [5]. These products account for 29% of all the veterinary pharmaceuticals (VPs) sold in the country [6], and they can be administered orally, topically by spraying or pour-on and via intraruminal bolus or injection. After treatment, parasiticides are excreted through urine or feces, with excretion rates depending on the compound, the dosage applied and the body size of the animal receiving the drug. However, for some VPs, it is known that between 40% and 90% of the administered compound is excreted unmetabolized [7,8].

Among the VPs used to control parasites in cattle, organophosphate (OP) insecticides (such as chlorpyrifos and fenthion) have gained space in the livestock field due to their outstanding efficacy against ectoparasites, such as ticks [9]. Fenthion is a long-acting OP [10] used to control ectoparasites such as warble flies, biting flies, lice and larvae in dairy cattle. It is rapidly absorbed through the skin and used topically, with application doses ranging from 10 to 15 mg of active ingredient (a.i.) per kilogram of body weight [11].

Due to the indiscriminate use of VPs in large herds, and considering the high rate of excretion of these compounds in unmetabolized form by treated cattle [7,8], it is likely that these substances reach the soil. In the case of fenthion, the compound has high lipophilicity (log Kow = 4.84 [12]) and moderate persistence in the soil (half-life of 47 days), potentially remaining for over 180 days [13]. Given its mechanism of action on the nervous system of target organisms, it may also pose toxicity risks to non-target soil invertebrates. However, studies assessing the toxic effects of this active ingredient on soil fauna bioindicators remain scarce and limited to earthworms [14,15].

Due to their representativeness and ecological functions within the soil ecosystem, soil fauna organisms such as collembolans, earthworms and enchytraeids have been widely used as model organisms in ecotoxicological tests (ISO 11267 [16], ISO 11268-2 [17], ISO 16387 [18]). However, data generated from these studies are typically based on a limited number of species, which may not accurately reflect environmental exposure under field conditions, where a large number of taxa can be affected, and the magnitude of impacts varies according to species sensitivity. One approach to improve the extrapolation of laboratory results to more ecologically realistic scenarios is the use of species sensitivity distributions (SSDs), which integrate ecotoxicological test results across multiple species, accounting for interspecific variability and taxonomic diversity in toxicity responses [19]. Due to these advantages, SSDs are widely used by environmental regulatory agencies to establish protection thresholds and support decision-making [20].

Although Brazil currently has no regulation establishing how SSDs should be elaborated, the Australian National Environment Protection Council (NEPC) recommends having ecotoxicity values for at least five species belonging to at least three distinct taxonomic groups [21]. In this line, earthworms, enchytraeids and collembolans are amongst the most commonly used and recommended soil faunal groups [16–18] for the determination of toxicity thresholds of contaminants in soil.

The objective of this study is to conduct an ecotoxicological assessment of the VP fenthion toward non-target soil invertebrates. Ecotoxicological tests were conducted with collembolans, enchytraeids and earthworms following standardized international test (ISO) protocols, and the ecotoxicological data were used to construct a species sensitivity distribution (SSD) curve to derive protective concentrations (PCs) for fenthion in soil.

2. Materials and Methods

2.1. Test Soil

A natural sandy soil (Entisol) sampled in Araranguá, Santa Catarina, Brazil (29°00′19.98″ S, 49°31′03.84″ W), was used as a test substrate for ecotoxicity bioassays. The soil, which was obtained from the top layer (0–20 cm), was sieved (2 mm mesh aperture), air-dried and defaunated through three freezing–thawing cycles (ISO 11268-2 [17]). Physicochemical characteristics of Entisol (60.7% sand, 35.2% silt, 4.1% clay, 0.9% soil organic matter, 2.12 cmolc dm^{-3} cation exchange capacity, pH 4.02) were determined in the EPAGRI soil laboratory, following Tedesco et al. [22]. This soil was selected for the ecotoxicological bioassays, as this class of soils is representative of Brazilian sandy soils, occupying approximately 15% of the national territory [23], and likely represents a worst-case scenario for evaluating the toxicity of organic chemicals in Brazilian scenarios [24]. The soil samples used on the bioassays had their moisture adjusted to approximately 60% of their maximum water-holding capacity (WHC).

2.2. Test Substance

A fenthion-based veterinary commercial formulation was used to artificially spike the soil samples for ecotoxicity bioassays. Tiguvon® Spot-on (150 g fenthion L^{-1}) is recommended for the control of larvae, mosquitoes, lice and ticks in both dairy and beef cattle herds [11]. While the product label describes the carrier generically as vehicle q.s.p. [11], equivalent Tiguvon® Spot-on formulations employ dipropylene glycol methyl ether as the main organic solvent vehicle [25]. Soil samples were spiked with 0.1, 1, 10, 100, 500 and 1000 mg active ingredient (a.i.) kg^{-1} dry soil (mg kg^{-1}) for the bioassays with earthworms *Eisenia andrei* and enchytraeids *Enchytraeus crypticus* and *Enchytraeus bigeminus*; 0.5, 1, 2, 4 and 8 mg kg^{-1} for collembolans *Folsomia candida*; and 0.25, 1 and 8 mg kg^{-1} for the alternative species of collembolans *Sinella curviseta* and *Proisotoma minuta*. The range of concentrations was defined based on range-finding bioassays and previous sensitivity assessments with alternative species [26] and was selected in order to allow a good estimation of the effective concentrations (EC10, EC20 and EC50 values) for each species.

2.3. Test Organisms

Earthworms *E. andrei*, enchytraeids *E. crypticus* and *E. bigeminus* and collembolans *F. candida*, *S. curvisetta* and *P. minuta* were maintained in a climate-controlled (20 ± 2 °C, 12 h photoperiod) laboratory room, according to ISO protocols (ISO 11268-2 [17], ISO 16387 [18] and ISO 11267 [16], respectively). Specific details about culture media composition, feeding frequency and moisture replenishment can be found in Bandeira et al. [27].

2.4. Chronic Toxicity Test with Earthworms E. andrei

Adult earthworms (with a clitellum) weighing 380 ± 107 mg were selected for the assay. The pool of earthworms was previously acclimated in the Entisol (control wet soil) for 24 h before the test began. Six fenthion concentrations, plus a negative control with distilled water, were tested. Ten earthworms were placed in each experimental unit, which consisted of plastic recipients containing 800 g of wet soil. The oligochaetes were fed with defaunated horse manure (approximately 5 g) weekly, and a few drops of distilled water were used to replenish moisture loss once a week. Adult survival was measured after 28 days through manual counting, whereas juvenile production was assessed after 56 days through warm extraction, following the procedure described in Alves et al. [28].

2.5. Chronic Toxicity Test with Enchytraeids E. crypticus and E. bigeminus

Bioassays with enchytraeids were performed with the standard species *E. crypticus* and the alternative species *E. bigeminus*. Organisms with an approximate length of 1 cm and a visible clitellum were used in the bioassays. In summary, ten organisms were manually pinched from the culture medium (agar-based) and added to each replica containing 30 g of wet soil (contaminated or control). Six fenthion concentrations were tested, as well as a negative control. Once a week, soil moisture was adjusted with distilled water, and around 5 mg of finely ground oat flakes was offered as food. After 21 days, enchytraeids were fixed in 70% ethanol, and Bengal rose solution (1%) was used to stain the organisms, which were then counted on a stereoscopic microscope [26]. Detailed information about the procedures can be found in Alves et al. [29].

2.6. Chronic Toxicity Test with Collembolans F. candida, S. curviseta and P. minuta

Bioassays with collembolans were performed with the standard soil species (*F. candida*) as well as two alternative species (*S. curviseta* and *P. minuta*). Age-synchronized collembolans (10–12 days old) were used in the chronic ecotoxicity bioassays, as established by ISO 11267 [16]. Briefly, ten collembolans were added to snap cap recipients containing 30 g of wet soil (contaminated or control soil). Five concentrations were selected for the bioassay with the standard species *F. candida*, and the outcomes with this species were used to choose priority concentrations for the species *S. curviseta* and *P. minuta* to avoid unnecessary exposure of organisms. A control treatment receiving only distilled water was set up for the assays with all species. The exposure period lasted 28 days, with weekly checks and adjustments of soil moisture, internal air renewal of replicas and replenishment of food (*Saccharomyces cerevisiae*, approximately 5 mg). At the end of the exposure period, the surviving adults and generated juveniles were counted as described in Bandeira et al. [27] through image assessment in ImageJ version 1.54g from the high-resolution pictures taken from floating collembolans in each replica.

2.7. Data Analysis

2.7.1. Estimation of Effect Concentrations

The Kolmogorov–Smirnov test and the Bartlett's test were used to verify the normality and homoscedasticity of the data of each assay individually. Data was log-transformed (to fulfill ANOVA's assumptions) and then submitted to Dunnett's post hoc test ($p < 0.05$) to identify the no observed effect concentration (NOEC) as well as the lowest observed effect concentration (LOEC). Exponential, Hormesis, Logistic and Gompertz regression models were fitted to the data, and the best-fit model was selected for the estimation of effective concentrations of 10% (EC10), 20% (EC20) and 50% (EC50) of reduction in reproduction. All statistical analyses were performed using Statistica v7.0.61.0 software.

2.7.2. Species Sensitivity Distribution (SSD) Modeling

A species sensitivity distribution (SSD) curve was elaborated using the EC20 values obtained from the bioassays with each one of the six organisms tested. The USEPA Species Sensitivity Distribution generator [30] was used to generate the curve through a log-normal model fitting. Protective concentrations for 95% (PC95), 90% (PC90), 80% (PC80) and 50% (PC50) were derived from the curve.

3. Results

3.1. Chronic Ecotoxicity Tests

The chronic ecotoxicity tests with the standard species met the validation criteria established by ISO protocols. In the control treatments, more than 100 juveniles of *F. candida* and

a coefficient of variation (CV) below 30% were recorded, as well as more than 30 juveniles of *E. andrei*, *E. crypticus* and *E. bigeminus*, with CVs below 20% and 50%, respectively, in the test controls (Table S1). Adult mortality remained below 20% in all control treatments for collembolans (with no mortality in the earthworm and enchytraeids test). For *S. curviseta*, the number of juveniles in the control was lower than 100 (60 ± 8 juveniles), which is somewhat expected, as this species has lower reproduction rates compared to the standard species [26].

Fenthion caused a significant reduction in the production of juveniles of all bioassays performed, but the magnitude of the toxic effect varied with species (Figure 1). *E. bigeminus* was only significantly affected by 500 mg kg^{-1}, whereas *E. crypticus* and *E. andrei* had their reproduction significantly reduced by 100 mg kg^{-1}.

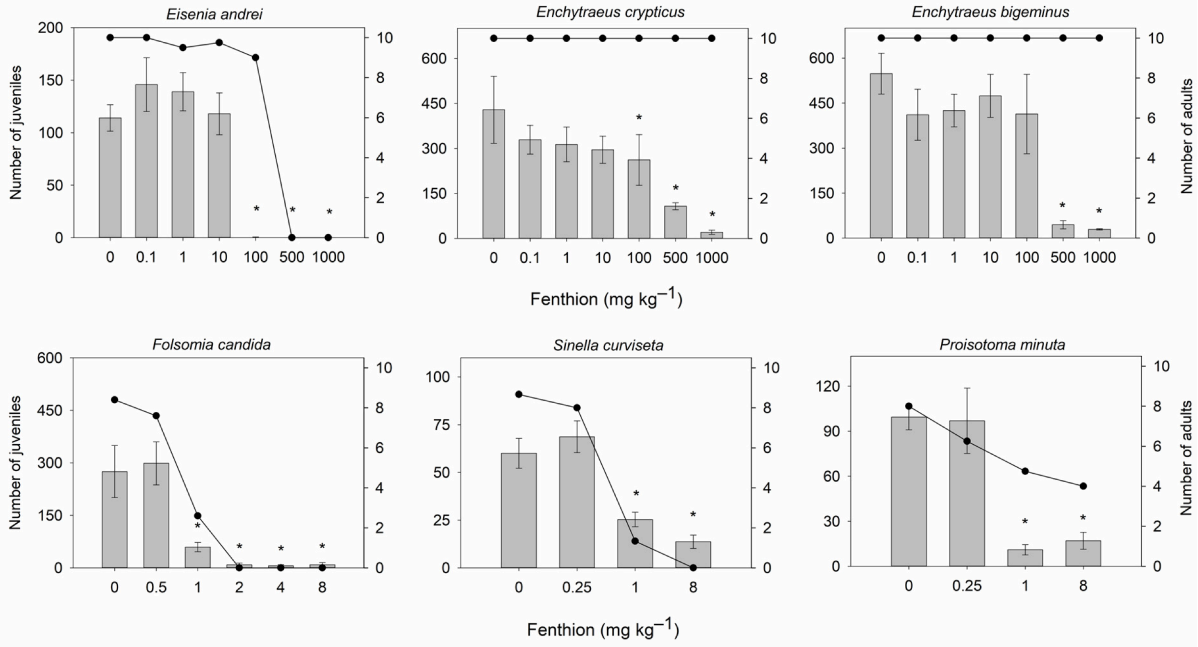

Figure 1. Mean number of juveniles (bars ± SD, left *y*-axis) and adults (dots with line, right *y*-axis) of earthworms *Eisenia andrei* (*n* = 4), *Enchytraeus crypticus* and *Enchytraeus bigeminus* (*n* = 4) and collembolans *Folsomia candida* (*n* = 5), *Sinella curviseta* (*n* = 3) and *Proisotoma minuta* (*n* = 3) found in Entisol contaminated with increasing concentrations of a fenthion-based veterinary formulation. Asterisks (*) indicate a significant reduction in the number of generated juveniles compared to the control treatment (Dunnett's post hoc test, $p < 0.05$).

Collembolans were the most sensitive group, with significant reductions in reproduction starting at 1 mg kg^{-1} for all the species studied (Figure 1, Table 1). For *F. candida*, reproduction was strongly inhibited (78%) by 1 mg kg^{-1} onwards, with almost no offspring produced from 2 mg kg^{-1}.

Table 1. Reproduction-based ecotoxicological parameters (NOEC, LOEC, EC10, EC20 and EC50, expressed as mg kg^{-1}, with 95% confidence limits in parentheses) from ecotoxicity assays with earthworms *Eisenia andrei*, enchytraeids *Enchytraeus crypticus* and *Enchytraeus bigeminus* and collembolans *Folsomia candida*, *Sinella curviseta* and *Proisotoma minuta* exposed to Entisol contaminated with increasing concentrations of a fenthion-based veterinary formulation.

Parameter	Species					
	E. andrei	*E. crypticus*	*E. bigeminus*	*F. candida*	*S. curviseta*	*P. minuta*
NOEC	10	10	100	0.50	0.25	0.25
LOEC	100	100	500	1	1	1

Table 1. *Cont.*

Parameter	Species					
	E. andrei	*E. crypticus*	*E. bigeminus*	*F. candida*	*S. curviseta*	*P. minuta*
EC10	3.50 (0.33–6.68)	43.5 (a)	94.7 (21.8–167)	0.08 (a)	0.35 (a)	0.12 (a)
EC20	8.49 (4.19–12.8)	80.90 (a)	130 (46.6–212)	0.21 (0.10–0.33)	0.46 (0.03–0.90)	0.19 (0.06–0.34)
EC50	28.4 (14.0–42.8)	234 (88.4–379)	221 (107–336)	0.72 (0.41–1.03)	0.93 (0.19–1.67)	0.48 (0.18–0.78)

(a) Data did not allow the estimation of the 95% confidence limits.

3.2. Species Sensitivity Distribution (SSD)

The SSD revealed a clear distinction of sensibility amongst the groups studied. Enchytraeids were clearly the least sensitive group to fenthion (Figure 2); a 20% reduction in the reproduction of species is expected only from concentrations of 80 mg kg^{-1} and above. Earthworms *E. andrei*, on the other hand, presented intermediate sensibility, with an EC20 9 to 15 times lower than those estimated for enchytraeids. Collembolans were relatively more sensitive to fenthion (Figure 2), with EC20 values in the range of 0.19–0.46 mg kg^{-1} (Table 1).

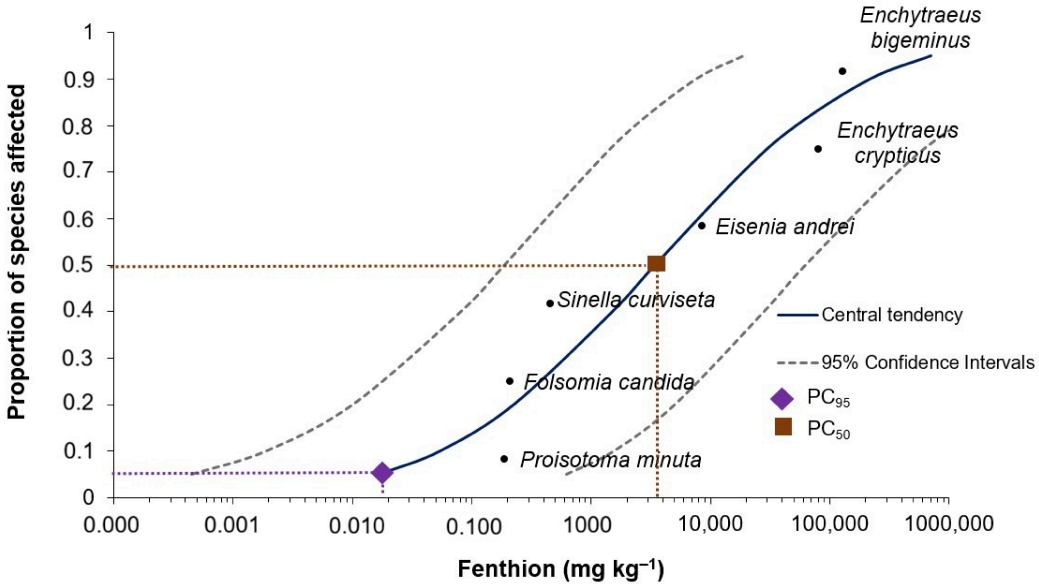

Figure 2. Species sensitivity distribution (SSD) curve elaborated with the EC20 values derived from chronic ecotoxicity assays with fenthion for six different species of soil invertebrates. PCx: protective concentrations for x% of soil invertebrate species.

The protective concentration estimation revealed that a concentration of 0.017 mg kg^{-1} is the safe fenthion threshold to guarantee the protection of 95% of soil species. PC90, PC80 and PC50 values are also presented in Table 2.

Table 2. Fenthion protective concentrations for x % of soil species (PC95, PC90, PC80 and PC50) estimated from species sensitivity distribution curve elaborated with chronic toxicity assays with six species of soil invertebrates.

Parameter	Value (mg kg^{-1}) [a]
PC95	0.017 (0.001–0.621)
PC90	0.054 (0.002–1.550)
PC80	0.225 (0.010–5.028)
PC50	3.432 (0.188–62.688)

[a] The 95% confidence intervals were presented in parentheses.

4. Discussion

Regarding the EC_{50} values (Table 1), the values for both enchytraeid species are similar, with their 95% confidence intervals overlapping, indicating no significant difference in ecotoxicity between the two species. Despite that, Freitas et al. [31] observed that *E. bigeminus* was the most sensitive species to copper compared to *E. crypticus* and *E. dudichi*. In addition, this species can be found in different land-use scenarios of agricultural areas, which makes this species a realistic model for soil ecotoxicity bioassays [32].

Collembolans were significantly affected from 1 mg kg^{-1}. From an ecosystem perspective, concentrations of this magnitude may represent a significant ecological risk for the species, since continuous exposure to these levels could lead to a decline in *F. candida* populations over time. The risk is also relevant for *S. curviseta*, as although this species still showed reproduction when exposed to 1 and 8 mg/kg, it reproduces sexually, generally producing fewer juveniles compared to *F. candida*, which reproduces through parthenogenesis [33,34].

In addition, although the EC50 values of the three collembolan species overlapped, we observed a 1.9- to 2.9-fold difference in the mean ECx values for *P. minuta* compared to *S. curviseta*, revealing that even within the same taxonomic group, different species can exhibit distinct responses to fenthion exposure in soil. These differences are likely related to the larger body size and consequently lower surface-area-to-volume ratio of *S. curviseta*, which limits cuticular toxicant uptake [35], combined with species-specific differences in habitat use and detoxification capacity documented for Collembola [36,37].

Studies evaluating the ecotoxicity of fenthion on soil invertebrates are scarce, but Márquez-Lázaro et al. [14] verified that fenthion did not cause growth inhibition of *Eisenia fetida* up to 50 μg kg^{-1} and that 75 μg kg^{-1} induced a statistically significant increase in earthworm's growth compared to control. On the other hand, muscle protein carbonylation was significantly increased from 25 μg kg^{-1}, indicating that biomarkers of oxidative stress might be considered when assessing the toxicity of fenthion toward soil invertebrates. In the environment, fenthion undergoes oxidative and hydrolytic degradation into metabolites such as fenthion phenol sulfoxide and fenthion phenol sulfone, which QSAR model estimations suggest may exert even higher ecotoxicity than the parent compound [38], although experimental validation of their effects on soil invertebrates remains absent in the literature.

Reis et al. [39] conducted a study in which the toxicity of amitraz—an antiparasitic compound that, like fenthion, acts on the arthropod nervous system—was tested for *F. candida* and *E. crypticus* in a tropical soil and found that the toxic effects of amitraz were more pronounced in collembolans than in enchytraeids, a pattern similar to the results obtained in the present experiment. The greater toxicity of amitraz and fenthion to collembolans compared to oligochaetes may be attributed to the fact that arthropods are one of the main targets of these active ingredients [40,41]. In general, synthetic compounds that act on acetylcholinesterase (AChE) tend to be more toxic to arthropods than to oligochaetes because these substances are designed to affect the insect nervous system, which shares significant similarities with that of other arthropods [26].

The recommended commercial dose of TIGUVON is approximately 7.5 g of active ingredient per animal (50 mL of commercial formulation recommended for one animal; TIGUVON contains 15 g a.i. per 100 mL). In a worst-case scenario, a maximum cattle density of 3.6 cattle units per hectare is expected in Brazilian pasturelands under intensification scenarios [42], and an excretion rate of 90% of the active ingredient [7,8] can be assumed. In this scenario, the estimated amount of fenthion reaching the soil in one hectare is about 24.3 g a.i. Assuming an incorporation depth of 5 cm and a soil density of 1500 kg m^{-3} [43], the estimated environmental concentration of fenthion after application of the commercial

dose—assuming uniform distribution of the feces over one hectare—is 0.032 mg kg^{-1} (see Equation (S1) in the Supplementary Material for detailed calculation). Therefore, although this concentration is not expected to cause a significant reduction in species reproduction on chronic toxicity assays (Figure 1), it is almost twice higher than the PC95 value (0.017 mg kg^{-1}). According to EC [44], the ecological risk of a certain compound is considered significant when the PEC/PNEC (predicted no-effect concentration) ratio is equal to or greater than 1; in our case, assuming the PC95 as the PNEC value, this ratio is 1.88, indicating that the ecological risk of fenthion to the species is significant/unacceptable. Therefore, our PC95 is likely a good initial protective threshold for soil invertebrates, but the veterinary use of this compound can result in contamination scenarios that equal or even exceed these limits.

It is also important to consider that fenthion can also be used as a pesticide, and its concentration in the soil may be higher when applied in this form. For example, Mahmud et al. [45] reported fenthion concentrations ranging from 1.65 to 1.81 μg g^{-1} (equivalent to mg kg^{-1}) after pesticide application in agricultural soils used for watermelon cultivation. These concentrations would likely have a significant impact on collembolan reproduction and are higher than the PC80 (0.225 mg kg^{-1}), indicating that more than 20% of species would be at risk.

Using commercial formulations rather than pure active ingredients in ecotoxicological evaluations provides a more realistic scenario for users and yields ecotoxicity outcomes that are more ecologically relevant from a regulatory point of view. Despite that, it must be recognized that fenthion-based formulations may contain organic solvents used to dissolve and stabilize the active ingredient (e.g., dipropylene glycol methyl ether; [25]). Therefore, a potential influence of the formulation vehicle on the final ecotoxicity cannot be completely ruled out. For future studies with fenthion, it is recommended to include a vehicle control to properly rule out any vehicle-related toxic effects.

Our experiments were conducted under standard conditions of soil moisture (e.g., 60% WHC) and temperature (20 °C). However, it must be noted that environmental conditions, particularly soil moisture, can directly influence the ecotoxicity of soil contaminants. Previous works already demonstrated a higher ecotoxicity of organic contaminants in scenarios of reduced soil moisture (e.g., 30–45% WHC) compared to standard soil moisture (50–60% WHC; [46,47]). Considering that drought scenarios are expected to be more frequent in a climate change world, drought stress could still amplify the toxic effects of fenthion observed in this study.

Our study brings new information about the toxicity thresholds for fenthion in soil, contributing to filling an existing regulatory gap in Brazil. Data from our study are also relevant for other countries, given that there is evidence that fenthion is also used in other regions around the world, such as North America [48] and Europe [49,50]. Although preliminary, the PCx values derived from our SSD are likely a reliable starting point for establishing environmentally protective concentrations of fenthion, once the modeling accounts for species covering at least three different taxonomic groups and with effective concentrations estimated for a natural sandy soil that potentially represents a worst-case scenario of exposure. The addition of ecotoxicity data obtained from other soil invertebrate species in our SSD, particularly other earthworm ecological guilds (e.g., endogeic or anecic species) rather than only the standard epigeic *E. andrei*, as well as other taxonomic groups such as Acari and Isopoda, is recommended as a continued step of research, in order to improve the accuracy and representativeness of the derived protective concentrations. Finally, future research should explore the cumulative effects of fenthion over time, as well as its combined impacts with other pollutants under field conditions.

5. Conclusions

Fenthion caused ecotoxicity in all the species tested, but the effects were higher for collembolans than for earthworms (intermediate sensitivity) and enchytraeids (least sensitive group). The protective concentration for 95% of soil species (0.017 mg kg^{-1}) was lower than the predicted concentration following veterinary use, indicating that the risk for soil invertebrates is not negligible. Our results provide a more integrated understanding of the ecotoxicity of fenthion to soil faunal invertebrates, which could be helpful for environmental agencies in the establishment of threshold limits of this active ingredient in soil compartments.

Supplementary Materials: The following supporting information can be downloaded at https://www.mdpi.com/article/10.3390/soilsystems10080088/s1, Table S1: Adult survival (%), number of juveniles (±standard deviation) and coefficient of variation (CV) of the control treatments of chronic ecotoxicity bioassays conducted with earthworms *Eisenia andrei*, enchytraeids *Enchytraeus crypticus* and *Enchytraeus bigeminus* and collembolans *Folsomia candida*, *Sinella curviseta* and *Proisotoma minuta* exposed to fenthion in Entisol. Equation S1: Estimated environmental concentration of Fenthion in soil following veterinary application.

Author Contributions: Conceptualization, F.O.B. and D.B.; data curation, I.V.; formal analysis, F.O.B. and D.B.; investigation, I.V., Í.L.G. and R.P.; resources, D.B.; writing—original draft preparation, I.V. and F.O.B.; writing—review and editing, C.R.D.M.B., P.R.L.A. and D.B.; supervision, F.O.B. and D.B.; project administration, F.O.B. and D.B.; funding acquisition, D.B. All authors have read and agreed to the published version of the manuscript.

Funding: The authors thank the Fundacao de Amparo a Pesquisa e Inovacao do Estado de Santa Catarina (FAPESC) for the financial support (project number 2024TR002026, Edital 33/2024; Edital 35/2025). R.P. and F.O.B. thank the FAPESC for the SET-E and DCR-C grants (process numbers 1712/2024 and 910/2025, respectively).

Data Availability Statement: The data presented in this study will be made available as Supplementary Materials associated with the published article.

Acknowledgments: D.B. and C.R.D.M.B. thank the National Council for Scientific and Technological Development (CNPq) for their scientific productivity grants (process numbers 3081895939/2022-1 and 30249483/2022-0, respectively). During the preparation of this work, the authors used generative AI tools to assist in language revision, grammar checking and text improvement. The authors take full responsibility for the content of the publication and have carefully reviewed and edited all outputs.

Conflicts of Interest: The authors declare no conflicts of interest.

Abbreviations

The following abbreviations are used in this manuscript:

WHC Water-Holding Capacity

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