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Environmental Risk Assessment by Fipronil and Imidacloprid Insecticides in River Shrimp (*Cryphiops caementarius*)

Evaluación del Riesgo Ambiental por los insecticidas Fipronil e Imidacloprid en el Camarón de río (*Cryphiops caementarius*)

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Abstract

Fipronil and imidacloprid currently represent approximately one third of the global insecticide market. In the present study, the environmental risk (ERA) of fipronil and imidacloprid in the postlarvae of the river shrimp (*Cryphiops caementarius*, Molina 1782) was evaluated. Short-term toxicity bioassays were performed based on LC_{50} (mean lethal concentration) (mortality) and EC_{50} (mean effective concentration) (swimming hypoactivity). PNEC (Predicted Concentration with No Known Effect) and available environmental standards for PEC (Expected Environmental Concentration) were calculated for fipronil and imidacloprid to determine risk quotient (RQ). Imidacloprid was more at risk for the aquatic environment than fipronil for the lethal response (mortality) and sublethal response (swimming hypoactivity). The observed risk difference between the two insecticides could be due to their different modes of action. *C. caementarius* should be considered as a sensitive species when defining an environmental quality standard for the conservation of the aquatic environment. Therefore, it is recommended to continue monitoring the presence of these insecticides in coastal freshwater bodies, and to reduce the use of fipronil and imidacloprid in the agricultural crops that use them.

Keywords: Environmental quality, *Cryphiops caementarius*, Aquatic ecosystem, Fipronil, swimming hypoactivity, Imidacloprid

Resumen

En la actualidad, el fipronil y el imidacloprid representan aproximadamente un tercio del mercado mundial de insecticidas. En el presente estudio se evaluó el riesgo ambiental (ERA) del fipronil e imidacloprid en las postlarvas del camarón de río (*Cryphiops caementarius*, Molina 1782). Se realizaron bioensayos de toxicidad de corta duración en base a la CL_{50} (Concentración letal media) (mortalidad) y CE_{50} (Concentración efectiva media) (hipoactividad natatoria). Se calculó la PNEC (Concentración prevista sin efecto conocido) y los estándares ambientales disponibles para la PEC (Concentración ambiental esperada) para el fipronil y el imidacloprid para determinar los cocientes de riesgo (CR). El imidacloprid resultó con mayor riesgo para el ambiente acuático que el fipronil para la respuesta letal (mortalidad) y subletal (hipoactividad natatoria). La diferencia del riesgo observada entre ambos insecticidas pudiera deberse a sus diferentes modos de acción. *C. caementarius* debería ser considerado como una especie sensible al momento de definir un estándar de calidad ambiental para la conservación del ambiente acuático. Por ende, es recomendable continuar el monitoreo para observar la presencia de estos insecticidas en los ecosistemas dulceacuícolas costeros, y reducir el uso del fipronil y del imidacloprid en los cultivos agrícolas que los emplean.

Palabras clave: Calidad ambiental, *Cryphiops caementarius*, Ecosistema acuático, Fipronil, Hipoactividad natatoria, Imidacloprid.

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1 Introduction

Imports of insecticides in Peru increased from 3481 tn to 5037 tn between 2007 and 2017 (INEI, 2018), and its use is regulated by the National Agrarian Health Service (MINAGRI, 2020) for the protection of health and the environment, because of the ecological impact insecticides have on water bodies and the impact on aquatic biodiversity (Escobar-Chávez et al., 2019; Sotelo-Vásquez and Iannacone, 2019). Currently, fipronil and imidacloprid account for approximately one third of the world's insecticide market (MINAGRI, 2020), and they work by blocking transmission into the central nervous system, but each chemical has a different mode of action (Al-Badran et al., 2018, 2019). Fipronil interferes with the passage of chloride ions by binding to a specific site within the gamma-aminobutyric acid receptor (GABA), while Imidacloprid binds to postsynaptic nicotinic acetylcholine receptors (nAChR) (Al-Badran et al., 2018).

Fipronil (phenylpirazole) and imidacloprid (neonicotinoids) are used on agricultural pests, domestic pests and ectoparasites of domestic animals (Al-Badran et al., 2018, 2019; Escobar-Chávez et al., 2019). In Peru, fipronil and imidacloprid insecticides are widely used for the pest control in agricultural crops of rice and onions (Gangwar et al., 2016; Pathak et al., 2018). Compared to other types of insecticides, fipronil and imidacloprid are considered safer due to their low toxicity in fish and mammals. Fipronil and Imidacloprid in small concentrations are very effective on arthropods (Al-Badran et al., 2018). Increased use in recent decades, moderate to high solubility and persistence in water raise serious concerns about the potential negative effects on aquatic invertebrates that are not the target of control (Al-Badran et al., 2019).

River shrimp (*Cryphiops caementarius*, Molina 1782), is a species with a biological, commercial and economic relevance in Peru, and is one of the endemic hydrobiological components maintained by the commercial research of the southern coast of Peru and northern Chile (Campos et al., 2017). Its highest populations have been observed in the rivers of the department of Arequipa (Peru), because these bodies of water have a higher flow, in addition to the predominance in this area over the cultivation of rice and onions. *Cryphiops caementarius* is used in aquaculture for food (Reyes, 2011, 2018; Romero-Camarena et al., 2013; Campos et al., 2017), subjected to a high hydrobiological exploitation in the natural environment, with impacts on habitat destruction due to natural drought and anthropic processes such as water use, agricultural and mining activities (Wasiw, 2017).

There is no research assessing the environmental risk of insecticides when using *C. caementarius* as a bioindicator species (Smit et al., 2015; Al-Badran et al., 2018, 2019). Therefore, the objective of this article was to assess the environmental risk of fipronil and Imidacloprid insecticides in *C. caementarius*.

2 Materials and methods

2.1 *Cryphiops caementarius* River Shrimp (Molina 1782)

Postlarvae of river shrimp (*C. caementarius*) Postlarvae of river shrimp (12°31′35″S 76°32′38″W) by an authorized fisherman belonging to the fishermen's guild of the area (Resolution N°83-2007-PRODUCE), Peru. Postlarvae were obtained in the closure period of this species (Baltazar and Colón, 2014; Wasiw and Yépez, 2015).

Postlarvae of *C. caementarius* were acclimatized at the Laboratory of Larviculture of the Southern Scientific University (UCSUR), Lima, Peru, two weeks before starting bioassays and following this scheme: about 1000 postlarvae were kept in a container of 750 L capacity with constant aeration at an average temperature of 21±2 °C, supplying precoated chicken liver as food every 24 hours to prevent the death of organisms. Water changes were carried out daily, and it was previously dechlorinated using sodium thiosulfate by siphon (Rice et al., 2017). Postlarvae of *C. caementarius* with an average size of 15 mm were used for bioassays (Baltazar and Colón, 2014). A calibrator (± 0.1 mm) was used to measure the total length of each postlarvae of *C. caementarius* by carefully straightening the shrimp body onto the table and measuring the total length from the tip of the head to the end of the tail (Al-Badran et al., 2019).

2.2 Insecticides

Fipronil of the Regent SC® was used at a concentration of 250 g L^{-1} and seven nominal concentrations were established (0.10 μg L^{-1}; 0.26 μg L^{-1}; 0.64 μg L^{-1}; 1.6 μg L^{-1}; 4 μg L^{-1}; 0.02 μg L^{-1} and 0.04 μg L^{-1}). Imidamin® brand was used as imidacloprid at a concentration of 350 g L^{-1} with five nominal concentrations (28.8 μg L^{-1}; 71.9 μg L^{-1}; 179.8 μg L^{-1}; 449.6 μg L^{-1} and 1124 μg L^{-1}).

2.3 Bioassays

Four replications were used for each insecticide and one control. 2 L with dilution water was placed in containers with a capacity of 3 L, which were connected to a system with constant aeration in series for each concentration and repetition. Later, 10 postlarvae specimens of *C. caementarius* were placed in each of the containers (Escobar-Chávez et al., 2019). The water used in the containers was conditioned with Nutrafin Aqua Plus® (Hagen, USA).

River shrimp postlarvae were fed every 48 hours with cooked rice to prevent their death by cannibalism. Postlarvae mortality and chronic hypoactivity (hNPL) measurements were performed at 3h, 8h, 24h, 48h, 72h and 96 h of exposure. The organisms were considered dead in the total absence of movement during 2 minutes after being gently touched with a stick. hNPL was listed as lack of displacement, lack of struggle, lack of reaction to mechanical stimuli, and lethargy. Normal postlarvae swimming activity was considered as the search for food, movement throughout the water column and rapid reaction to mechanical stimuli.

2.4 Data analysis and environmental risk assessment

Mortality and hNPL percentages of *C. caementarius*. were determined. The mean lethal concentration (LC_{50}) for mortality and mean effective concentration (EC_{50}) for hNPL was calculated with the Probit version 1.5 program with a 95% confidence level, and the regression model was verified with the Chi-square (χ^2) statistic (Rice et al., 2017). The LOEC (lowest concentration where effect is observed) and NOEC (non-observed effect concentration) parameters were calculated with Past 3.2 statistical program, using Krustal-Wallis test based on significant differences between mortality and hNPLs for the fipronil and imidacloprid concentrations used.

2.4.1 Expected concentration with no known effect (PNEC)

PNEC was found from $LC(E)_{50}$, LOEC y NOEC derived from the results of short-term toxicity tests. These parameters were given the valuation or safety factor (FV) established for toxicity tests, which was 1000 (UNEP/IPCS, 1999). With the relationship PNEC= Toxicity Parameters/Valuation Factor.

2.4.2 Expected Environmental Concentration (PEC)

The environmental quality standards (EQS) of The Netherlands legislation for fipronil (Tennekes, 2018) and imidacloprid (Smit et al., 2015) were used, in which EQS of imidacloprid presented two scenarios. Scenario 1: Short-term EQS, maximum acceptable concentration (MAC-EQS) based on acute toxicity data. Scenario 2: Long-term EQS, expressed as an average annual concentration (AA-EQS) based on chronic toxicity data with the aim of protecting the ecosystem against adverse effects resulting from long-term exposure (EC, 2011).

2.4.3 Risk characterization (PEC/PNEC)

The risk coefficient (RC) was calculated as RC= PEC/PNEC. It states that if the PEC/PNEC is <1 the fipronil and Imidacloprid evaluated are considered to have low risk; while when PEC/PNEC is >1, it is considered to have high risk (De la Torre et al., 2004).

3 Results

Mortality and hNPL increase from 3 h to 96 h of exposure in postlarvae of *C. caementarius* that presented a higher effect on fipronil at 96 h of exposure, reaching 100% of mortality and hNPL. For Imidacloprid, 87.5% and 100% were reached for mortality and hNPL, respectively. The LC_{50} and EC_{50} values for fipronil insecticides with their upper and lower limits of 95% were obtained from 3 h to 96 h of exposure, and their respective determination coefficients (R^2) (Tables 1 and 2). Similarly, LOEC and NOEC parameters for fipronil and imidacloprid are observed from 3 h to 96 h of exposure in the postlarvae of *C. caementarius* (Table 3 and 4).

Table 5 shows the values that establish the relationship between PEC and PNEC to determine the existing risk of insecticides, based on the PNEC-LC_{50}-*mortality*, PNEC-EC_{50}-*hNPL*, PNEC-LOEC-*mortality* and PNEC-LOEC-*hNPL* parameters. Values of RCs higher than one were obtained with fipronil and imidacloprid in all cases (Table 5). Imidacloprid presented a higher risk to the aquatic environment compared to fipronil for lethal (mortality) and sublethal response (postlarvae swimming hypoactivity).

Table 1. Mean lethal concentration (LC_{50}) and Mean effective concentration (EC_{50}) and upper and lower limits for lethal and sublethal parameters based on hNPL (swimming hypoactivity) in *Cryphiops caementarius* with fipronil at six different times of exposure.

Exposure time (h)	Mortality LC_{50} (μg·L^{-1})	R^2	hNPL EC_{50} (μg·L^{-1})	R^2
3 h	0.901 (0.506- 1.601)	0.98	0.252 (0.175- 0.364)	0.99
8 h	0.679 (0.354- 1.303)	0.97	0.074 (0.042- 0.131)	0.96
24 h	0.035 (0.007- 0.171)	0.87	0.003 (0.001- 0.007)	0.99
48 h	< 0.02	ND	< 0.02	ND
72 h	< 0.02	ND	< 0.02	ND
96 h	< 0.02	ND	< 0.02	ND

( ): Upper and lower limits (95 %). R^2: determination coefficient.

Table 2. Mean lethal concentration (LC_{50}) and mean effective concentration (EC_{50}) and upper and lower limits for lethal (mortality) and sublethal parameters based on hNPL (swimming hypoactivity) in *Cryphiops caementarius* with Imidacloprid at six different times of exposure.

Exposure time (h)	Mortality LC_{50} (μg·L^{-1})	R^2	hNPL EC_{50} (μg·L^{-1})	R^2
3 h	ND	ND	260.5 (82.97- 817.92)	0.83
8 h	ND	ND	246.7 (152.42- 399.42)	0.94
24 h	5353.7 (1832.17- 15643.56)	1	28.4 (6.62- 122.17)	0.97
48 h	53540.8 (1032.89- 2775350.23)	0.52	5.3 (1.11- 25.66)	0.46
72 h	13.68 (1.72- 109.11)	0.97	1.2 (0.14- 9.83)	0.93
96 h	0.23 (0.01- 6.37)	0.91	0.002 (0.00- 0.16)	0.79

( ): Upper and lower limits of 95 %. R^2: determination coefficient.

4 Discussion

No toxicity bioassays have been conducted with fipronil and imidacloprid in *C. caementarius*, but research is observed with other aquatic crustacean species (Goff et al., 2017; Al-Badran et al., 2019). *C. caementarius* had effects on mortality and swimming hypoactivity (hNPL) for both insecticides. There was a lack of reaction in hNPL to provided stimuli, reduction of movement and different swimming. The results obtained varied with concentrations and exposure times in both insecticides (Mendoza-Rodríguez, 2009).

Shan et al. (2003) when working with *Macrobrachium rosenbergii* (De Man, 1879) and *Macrobrachium niponnensis* (De Haan, 1849), found values of LC_{50} (24 h) de 6.41 μg·L^{-1} and > 25.70 μg·L^{-1}, respectively with fipronil. *C. caementarius* was more sensitive to fipronil than the two *Macrobrachium* species with a LC_{50} value of 0.035 μg·L^{-1} at 24 h of exposure. The differences obtained in LC_{50} may be due to the different protocols used in bioassays, where Shan et al. (2003) conducted a simulation of rice fields under laboratory conditions. These differences can also be explained by specific biodistribution patterns, metabolization rates, or even to the specific sensitivity of each target taxon.

Fipronil and Imidacloprid in *Palaemonetes pugio* Holthuis, 1949 at 96 h of exposure obtained a LC_{50} of 0.68 $\mu g{\cdot}L^{-1}$ for larvae and a LC_{50} of 0.32 $\mu g{\cdot}L^{-1}$ for adults with fipronil; it was significantly more toxic in the larvae with Imidaclopris (LC_{50} of 308 $\mu g{\cdot}L^{-1}$) than in adults (LC_{50} of 563.5 $\mu g{\cdot}L^{-1}$) (Key et al., 2007). In the case of *C. caementarius*, exposure to fipronil and Imidacloprid at 96 h showed mortality in shrimp postlarvae, which differs from Key et al. (2007). The LOEC and NOEC parameters for mortality were also found, which were 0.02 $\mu g{\cdot}L^{-1}$ and <0.02 $\mu g{\cdot}L^{-1}$ at 96 h of exposure with fipronil, and 28.8 $\mu g{\cdot}L^{-1}$ and <28.8 $\mu g{\cdot}L^{-1}$ (96 h) with Imidacloprid, respectively. Key et al. (2007), found lower toxicity values for *P. pugio* than those obtained in this research.

Fipronil had higher lethal toxicity than Imidacloprid based on LC_{50} *C. caementarius* at 96 h of exposure. Omar et al. (2016), in *Marsupenaeus japonicus* (Spence Bate 1888), found variable effects according to the development stage tested. The increased lethal toxicity of Fipronil compared to Imidacloprid has been observed in other species of decapod crustaceans such as *Farfantepenaeus aztecus* (Al-Badran et al., 2019). *Penaeus monodon* Fabricius, 1798 was subjected to Fipronil and Imidacloprid in the postlarvae stage at 48 h of exposure, finding LC_{50} of 0.2 $\mu g{\cdot}L^{-1}$ and 175 $\mu g{\cdot}L^{-1}$, respectively (Hook et al., 2018).

Table 3. Lower concentration where effect (LOEC) and non-observed concentration effect (NOEC) are observed for lethal (mortality) and sublethal parameters based on hNPL (swimming hypoactivity) in *Cryphiops caementarius* with fipronil at six different times of exposure.

Exposure time (h)	Mortality ($\mu g{\cdot}L^{-1}$)		hNPL ($\mu g{\cdot}L^{-1}$)	
	LOEC	NOEC	LOEC	NOEC
3 h	1.60	0.64	0.26	0.10
8 h	0.26	0.10	0.04	0.02
24 h	0.02	< 0.02	0.02	< 0.02
48 h	0.02	< 0.02	0.02	< 0.02
72 h	0.02	< 0.02	0.02	< 0.02
96 h	0.02	< 0.02	0.02	< 0.02

LOEC: Lowest concentration where effect is observed.
NOEC: Concentration of non-observed effect.

Table 4. Lower concentration where effect (LOEC) and non-observed concentration effect (NOEC) are observed for lethal (mortality) and sublethal parameters based on hNPL (swimming hypoactivity) in *Cryphiops caementarius* with imidacloprid at different exposure times.

Exposure time (h)	Mortality ($\mu g{\cdot}L^{-1}$)		hNPL ($\mu g{\cdot}L^{-1}$)	
	LOEC	NOEC	LOEC	NOEC
3 h	28.8	< 28.8	179.8	71.9
8 h	28.8	< 28.8	179.8	71.9
24 h	28.8	< 28.8	28.8	< 28.8
48 h	28.8	< 28.8	28.8	< 28.8
72 h	28.8	< 28.8	28.8	< 28.8
96 h	28.8	< 28.8	28.8	< 28.8

LOEC: Lowest concentration where effect is observed.
NOEC: Concentration of non-observed effect.

Table 5. PEC (Exposure Assessment) values, PNEC (expected concentration with no known effect) to determine the RC (Risk Coefficient) of fipronil and Imidacloprid insecticides using *Cryphiops caementarius* river shrimp.

Parameters	**Fipronil**	**Imidacloprid - Scenario 1**	**Imidacloprid - Scenario 2**
PEC	0.00007 (EQS)	0.2 (MAC-EQS)	0.0083 (AA-EQS)
PNEC (LC_{50}-$_{mortality}$)	0.00002	0.00023	0.00023
PNEC (EC_{50}-$_{hNPL}$)	0.00002	0.000002	0.000002
PNEC (LOEC-$_{mortality}$)	0.00002	0.028	0.028
PNEC (LOEC-$_{hNPL}$)	0.00002	0.028	0.028
RC (LC_{50}-$_{mortality}$)	3.5	869.56	36.08
RC (EC_{50}-$_{hNPL}$)	3.5	100.000	4.150
RC (LOEC-$_{mortality}$)	3.5	7.14	2.96
RC (LOEC-$_{hNPL}$)	3.5	7.14	2.96

EQS= Environmental Quality Standard. LC_{50} = Average lethal concentration at 96 h of exposure. EC_{50} = Average effective concentration at 96 h of exposure. LOEC= lowest concentration where effect is observed at 96 h of exposure. MAC-EQS= Maximum permissible concentration for a short-term Environmental Quality Standard. Long-term AA-EQS, expressed as an average annual concentration.

Arthropods are among the most fipronil-sensitive taxa, and related species may have very varied sensitivities to this insecticide (Stevens et al., 2011), because fipronil may be more toxic once metabolized, since fipronil sulfide and fipronil sulfone are generally two to three times more toxic than the original compound. In addition, there is a very wide range in the sensitivity of crustaceans to Imidacloprid, with LC_{50} values ranging from 1 to 52.500 $\mu g \cdot L^{-1}$ (Smit et al., 2015).

Sublethal effects of fipronil and imidacloprid on the behavior, physiology, reproduction and development of non-target aquatic invertebrates have been observed (Al-Badran et al., 2018; Sohn et al., 2018). A LC_{50} was found for the hNPL sublethal parameter, in a range from 260.5 $\mu g \cdot L^{-1}$ to 0.002 $\mu g \cdot L^{-1}$, between 3 h and 96 h with Imidacloprid, and a range of 0.252 $\mu g \cdot L^{-1}$ and <0.02 $\mu g \cdot L^{-1}$ obtained between 3 h and 96 h of exposure with fipronil. Fipronil, unlike Imidacloprid, caused erratic swimming in all directions or seizures and immediate reaction to movement stimulus, while imidacloprid caused lethargy in larvae, decreased swimming, and provoked late reaction to stimulation of movement.

For both insecticides there was a struggle for food, total absence of movement before their death, despite the attempted swimming that was observed in the mobility of the locomotive appendages. Al-Badran et al. (2019) found changes in the behavior of *F. aztecus* by the action of fipronil and imidacloprid under different exposure times depending on concentrations. Imidacloprid reduced the defense behavior of the crustacean *Orconecres rusticus* (Sohn et al., 2018). In this work imidacloprid had higher sublethal effects than fipronil. The different effects of postlarvae on both insecticides are due to action of each. fipronil is a GABA antagonist that causes hyperexcitement and seizures; while Imidacloprid is an nAChR antagonist that causes a variety of symptoms from hyperexcitation to lethargy and paralysis (Cox et al., 1998; Al-Badran et al., 2019).

In relation to the active ingredients of fipronil and imidacloprid, the results obtained with short-term RC show the existence of an environmental risk, and these are consistent with studies conducted by Van der Sluijs et al. (2015), which show the risks to biodiversity and ecosystem functioning by the widespread use of neonicotinoids such as imidacloprid and fipronil. Samples taken in groundwater and surface water have been found to exceed limits based on ecological thresholds established in different countries of North America and Europe, indicating that they exist in soils, waterways and plants in agricultural, urban and drainage areas that are contaminated with mixtures of fipronil, neonicotinoids or their metabolites (Bonmatin et al., 2015). Van der Sluijs et al. (2015) show evidence that these insecticides pose a high risk for a wide range of non-target invertebrate taxa, which would have an impact on aquatic food chains.

Pesticides can be leached in ditches and rivers by rains, and surface water can be contaminated with direct spraying by runoff and leaching of agricultural fields (Vijver and Van den Brink, 2014). The emission of fipronil and Imidacloprid to the surface waters are caused by many factors, such as the distance from the crop to the trench, mode of application, climatic conditions, etc. This is a problem if certain protocols of application are not applied or the potential effects on aquatic ecosystems are unknown (Stoorvogel et al., 2003; Pisa et al., 2015), such is the case of the river shrimp, which is often found in rivers near the rice fields, where fipronil and imidacloprid are widely use (Wasiw, 2017).

Several laboratory studies have been published on the toxicity of Imidacloprid in a variety of aquatic invertebrates and the standard test organism, *Daphnia magna* Straus, 1820, which is less toxic to neonicotinoids (Imidacloprid) compared to other invertebrates (Beketov and Liess, 2008; Escobar-Chávez et al., 2019). An acute LC_{50} of about 7 000 $\mu g{\cdot}L^{-1}$ represents more magnitude compared to the effective concentrations found for other invertebrates. This implies that *D. magna* cannot always be used as a sensitive and protective test organism for the entire aquatic species, unlike *C. caementarius* which showed greater sensitivity (Ngim and Crosby, 2001).

Although no information is available on the toxicity of these insecticides in *C. caementarius*, there are studies such as the one conducted by Van Dijk et al. (2013), where the abundance of aquatic macroinvertebrates and Imidacloprid concentrations in surface waters were evaluated. The abundance of macroinvertebrates was observed to decrease as the concentration of Imidacloprid in the aquatic environment increased (Beketov et al., 2013). According to the level of risk obtained from the literature (i.e. RQ $\geq$ 1 high risk, 0.1 $\leq$ RQ <1 medium risk, 0.01 $\leq$ RQ <0.1 low risk) (Sánchez-Bayo et al., 2002), the two insecticides presented a high risk to the aquatic ecosystem based on the lethal and sub-lethal effects of *C. caementarius.*

In general terms, the results of this research and published literature indicate that both insecticides have the potential to cause significant damage to aquatic ecosystems by provoking negative effects on individuals and populations of aquatic invertebrates at very low concentrations (Chaton et al., 2002). There would be an increased risk in *C. caementarius* from December to March, and according to Peruvian regulations this species is in a closure season because during this stage there is more presence of eggs and female carrying eggs (Baltazar and Colón, 2014).

5 Conclusions

The bioindicator *C. caementarius* allows the environmental risk of fipronil and imidacloprid to be assessed in the aquatic ecosystem by using the 96 h risk ratio of exposure based on mortality in the lethal response, as well as mortality and sub-lethality based on swimming hypoactivity. Imidacloprid presented a higher risk to the aquatic environment than fipronil for the lethal (mortality) and sublethal response (postlarvae swimming hypoactivity). The observed toxic difference between the two insecticides could be due to their different modes of action. *C. caementarius*, an invertebrate from Peru, should be considered as a sensitive species when defining an EQS for the conservation of the aquatic environment, especially from January to March, in which according to Peruvian regulations this species is in a closure period. It is advisable to continue monitoring the presence of these insecticides in coastal freshwater bodies, and to reduce the use of fipronil and imidacloprid in the agricultural crops that use them.

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