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Accumulation of current-use pesticides, cholinesterase inhibition and reduced body condition in juvenile one-sided livebearer fish (*Jenynsia multidentata*) from the agricultural Pampa region of Argentina



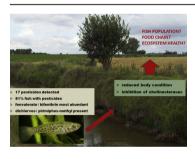
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HIGHLIGHTS

- Seventeen different pesticides were detected in one-sided livebearer fish.
- Eighty-one percent of captured animals contained at least one pesticide molecule.
- Pyrethroids fenvalerate and bifenthrin were most frequently detected.
- Restricted use organophosphate pirimiphos-methyl and dichlorvos present in fish.
- Pesticides were associated with cholinesterase inhibition and reduced body condition.

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ABSTRACT

The aim of this study was to characterize the level and nature of the pesticide contamination received by one-sided livebearer fish (*Jenynsia multidentata*) from a watercourse situated within the main agricultural region of Argentina, and to assess the effects of this contamination on fish health. Juvenile one-sided livebearer fish (*Jenynsia multidentata*) were collected in December 2011 and March 2012 from three sites along the Pergamino River. Pesticide contamination was characterized by extracting whole fish and analytically determining thirty different pesticide molecules. The biomarkers catalase, glutathione-S-transferase, and cholinesterases were assessed. Body condition was calculated as an estimate of the amount of energy reserves possessed by the fish. Seventeen different pesticides were detected in fish tissues with 81% of captured animals containing at least one pesticide molecule. The pyrethroid insecticides fenvalerate and bifenthrin were most frequently detected, being respectively found in 41.8 and 36.4% of samples tested. Highly toxic dichlorvos and pirimiphos-methyl were detected. Differential levels of contamination could not be established amongst sites but were observed within sites amongst the two

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Fungicide Aquatic sampling dates. The months when pesticide residues were most abundant from in Site A and B corresponded to the months when body condition was at its lowest in the two sites. The inhibition of Che activity in March when body condition was reduced also points to a role of insecticide contamination in the reduction of body condition. These findings provide strong new evidence that current-used agricultural pesticides can accumulate in wild fish and impact their health and energetics.

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1. Introduction

Modern agriculture is characterized by intensive crop production heavily dependent upon pesticides, fertilizers, genetically modified cultures and other technologies (Tilman et al., 2002). Despite its rural setting, intensive agriculture is a highly unnatural activity which substantially perturbs the environment (Harrison and Hester, 2012). Agricultural operations can contribute to water quality deterioration through the release of several materials into the water: sediments, pesticides, animal manures, fertilizers and other sources of inorganic and organic matter (Ongley, 1996; FAO, 2002).

The synthetic pesticides constitute the only group of chemicals created to be released into the environment to effectively kill living organisms, implying they intrinsically pose a risk to the biota (Harrison and Hester, 2012; Ongley, 1996). Pesticides enter aquatic ecosystems via diffuse entry pathways such as spray drift, accidental overspray, runoff or percolation and, hence, are called "non-point" sources of pollution. (Schäfer et al., 2011). Around the globe, freshwater systems from agricultural regions are contaminated by mixtures of diverse pesticides (Konstantinou et al., 2006; Gilliom, 2007; Giroux and Pelletier, 2012; Moreira et al., 2012; Bereswill et al., 2013; Bonansea et al., 2013; Smiley et al., 2014; De Gerónimo et al., 2014). The pollution of freshwater systems with pesticides represents a threat to biodiversity, ecosystem functions, ecosystem services (Millennium Ecosystem Assessment, 2005) and human health. Fish living in contaminated lakes and rivers are constantly exposed to pesticides either through breathing, through the food they eat or from the medium they live in (Stanley and Preetah, 2016). The European Food Safety Agency (EFSA) panel on plant protection products states that wild fish supply important ecosystem services and should be protected at the level of individuals as well as populations (EFSA, 2010).

Most of our current knowledge regarding the effects of pesticide on fish species originates from laboratory studies (Stanley and Preetah, 2016). These studies, however, strongly lack ecological realism and are poorly suitable for understanding the actual consequences of pesticide contamination on fish populations and ecosystem health (Vighi and Villa, 2013). Together with omitting to consider the structure and function of ecological communities, laboratory testing protocols also generally fail to take into account the fact that aquatic exposures to pesticides normally occur in the form of isolated or sequential pulses of mixtures of fluctuating concentrations (Eggen et al., 2004; Ashauer et al., 2006; Stehle et al., 2013; Bundschuh et al., 2014; Smiley et al., 2014). Field work examining the effects of pesticides on wild fish species are crucial to better understand the impacts of complex chronic exposures to low concentrations of various mixtures of pesticides. Nevertheless, a limited number of such studies have actually been conducted in the past (Wijeyaratne and Pathiratne, 2006; Carriquiriborde et al., 2007; Bony et al., 2008; Moreira et al., 2010). The aim of the current study was to examine whether onesided livebearer fish (*Jenynsia multidentata*) from a watercourse situated at the heart of the agricultural region of Argentina are affected by pesticide contamination.

The Pergamino River is situated at the north of Buenos Aires Province, in the geographic region known as the Rolling Pampa, which is characterized by a slightly undulated landscape where slopes do not exceed 2% (Fig. 1). The watershed of the Pergamino River covers approximately 2000 km², and is part of the Pergamino – Arrecifes system, a large watershed of 10.336 km² that belongs to the Parana River system (Reynoso and Andriulo, 2009). The natural meadows that were originally dominant within the watershed of the Pergamino River have almost completely been replaced by extensive agricultural systems, which now represents more than 91% of land cover (Cabrini and Calcaterra, 2008). Livestock and pastures occupy the rest of the surface area, being commonly predominant along watercourses and floodable sectors (Darder et al., 2012). Soybean (72% of the superficies), corn (15.3%) and wheat (13.2%) are the predominant crops (Cabrini and Calcaterra, 2008). The most used pesticides reported for the region during the spring-summer season of 2011–2012 (when fish were sampled) were respectively: glyphosate, -4 D, dicamba, chlorimuron, metsulfuron, imazethapyr, and acetochlor for herbicides, cypermethrin, chlorpyrifos, insect growth regulators, neonicotinoids and diamides for insecticides, and carbendazim and tebuconazole as fungicides (RETAA, 2013). A number of these and other pesticides have been reported in varying concentrations in water and sediments of the region's watercourses (Peruzzo et al., 2008; Aparicio et al., 2013; Bonansea et al., 2013; De Gerónimo et al., 2014; Hunt et al., 2016; Ronco et al., 2016; Caprile et al., 2016; Etchegoyen et al., 2017).

The one-sided livebearer is one of the most frequent and abundant freshwater fish in subtropical regions of South America, its distribution ranging from the southern province of Rio Negro in Argentina up to the latitude of Rio de Janeiro in Brazil (Mai et al., 2007; Goyenola et al., 2011). This small viviparous fish reproduces in spring and summer in subtropical and temperate regions. Offspring are born with a length of approximately 12 mm and the maximum size achieved by adult males and females are respectively around 48-66 and 86-91 mm, females being larger than males (Lopez-Cazorla et al., 2003; Garcia et al., 2004; Goyenola et al., 2011). In the subtropics, females reach maturity at the end of winter/early spring at a length of 35 mm-40 mm (Goyenola et al., 2011). It is an omnivorous-planktivorous fish that can feed on zooplankton, phytoplankton, periphyton, invertebrates and detritus (Iglesias et al., 2008; Cardozo et al., 2008; Quintans et al., 2009). The species is highly productive and efficient at colonizing unsaturated habitats due to its tolerance to several environmental variables (low oxygen levels, high temperatures, conductivity and turbidity) and various other life traits (small size, rapid growth, low age of maturity, high natural fertility and omnivory) that promote its resistance and resilience (Menni et al., 1996; Garcia et al., 2004; Iglesias et al., 2008; Goyenola et al., 2011).

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