

Ecological implications of altered fish foraging after exposure to an antidepressant pharmaceutical



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ABSTRACT

Pharmaceutical residues are increasingly detected in environmental and biological samples, some at levels known to adversely affect non-target organisms; however, less is known of how these organism-level effects relate to the ecology of aquatic systems. Foraging processes may be used as behavioral endpoints that link effects on individuals to the population and community levels, enabling risk assessment of environmental contaminants at larger ecological scales. In this study, we performed feeding trials using juvenile Eurasian perch (*Perca fluviatilis*) exposed to the selective serotonin reuptake inhibitor (SSRI) sertraline to test the hypothesis that sertraline alters foraging ecology of the fish in terms of their functional response. We found an exposure-dependent decrease in feeding with increasing sertraline concentrations. Further experiments revealed that feeding rates decrease at both low and high prey densities, indicating effects on both attack rate and handling time, respectively. Because the functional response can shape consumer-resource dynamics, such effects may alter the stability of predator-prey systems and consequently, community structure.

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1. Introduction

Emerging organic contaminants such as pharmaceuticals are increasingly reported in environmental and biological samples, some of which have been shown to adversely affect non-target organisms (Arnold et al., 2013; Daughton and Ternes, 1999; Halling-Sørensen et al., 1998; Kidd et al., 2007; Naidoo et al., 2009). One group of these is the selective serotonin reuptake inhibitors, SSRIs, prescribed for treatment of depression and other psychological disorders. These compounds bind and block serotonin receptor sites, leading to a buildup of serotonin in nerve synapses. Given that within vertebrates, serotonin receptors are highly conserved across species (Gunnarsson et al., 2008) and because serotonin is an important biomolecule affecting a variety of physiological and behavioral functions in fish (Lillesaar, 2011), it is probable that SSRIs found in aquatic environments can result in physiological/behavioral changes in fish (Kreke and Dietrich, 2008).

In recent years, there has been a call for the incorporation of more “ecological” endpoints into research on environmental contaminants (Chapman, 2002; Relyea and Hoverman, 2006). Indeed, previous studies examining exposure of fish to SSRIs and other pharmaceuticals have noted changes in various behaviors (Bell, 2001; Brodin et al., 2013; Gaworecki and Klaine, 2008;

Nassef et al., 2010; Reyhanian et al., 2011; Valenti et al., 2012; Weinberger and Klaper, in press); however, extrapolation of sub-lethal effects on individuals to a broader ecological perspective remains limited. Our study combines behavioral and foraging ecology with ecotoxicology, providing a link to interpreting effects of environmental contaminants on population and community dynamics that can potentially be incorporated into risk assessment. We examined effects of the SSRI sertraline on foraging in European perch (*Perca fluviatilis*) and relate these to the Holling Type II functional response, known from classical ecology to be vital in shaping consumer-resource dynamics (Fryxell and Lundberg, 1998; Holling, 1959). Because serotonin has anorectic effects on appetite in fish (Kreke and Dietrich, 2008), we hypothesized that exposure to sertraline would also cause a decrease in feeding. By using two prey densities, low representing attack rate (rate of detecting/encountering prey) and high representing handling time (time to subdue/process prey), we interpret effects on individuals in a broader ecological context.

2. Materials and methods

Juvenile Eurasian perch (*P. fluviatilis*, 3.5–5.0 cm, $n = 120$) were collected in summer 2011 from Lake Krankesjön in southern Sweden. Fish were allowed to acclimate in flow-through tanks and were fed frozen chironomid larvae and live *Daphnia magna* during the acclimation/experimental periods; food provided for *D. magna* was cultured *Scenedesmus* spp. Holding and experimental

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conditions were maintained at 12°C room temperature and a 12:12 h light:dark cycle.

2.1. Functional response baseline

To establish the baseline functional response curve, individual perch were placed in feeding arenas and a randomly assigned density of live *D. magna* (10–200 in 3.5 L water; 560–900 µm) was added after 2 min (food was withheld for at least 12 h prior to trials). Fish were given 10 min maximum to begin feeding; if they did not feed within this timeframe, the trial was discarded and restarted with a new individual. Time to eat 5 prey was measured with an upper limit of 20 min after beginning feeding. Trials were replicated between 3 and 9 times at each prey density using different individuals on different dates. Feeding rates (*D. magna* min⁻¹) were calculated and a Holling Type II functional response curve (Eq. (1)) was fitted using a nonlinear least squares method in R (2012).

$$C = \frac{aN}{1 + ahN} \quad (1)$$

This is a modified form of the Holling Disk Equation (Holling, 1959) in which C represents per capita feeding rate of the predator, a represents attack rate, h is the predator's handling time, and N indicates prey density. The fitted curve was used to distinguish low and high prey densities indicating effects on a and h , respectively, used for subsequent experiments.

2.2. Exposure-response

After allowing for acclimation to individual exposure arenas (3.5 L; 24 h), fish were exposed to sertraline (CAS# 79559-97-0, Toronto Research Chemicals Inc.) in 3.5 L water at nominal exposure concentrations chosen based on prior range-finding experiments (data not shown): 89, 133, 200, 300, and 450 µg/L ($n=3$ per treatment) for a total of seven days; solvent controls (dimethyl sulfoxide) were also included. Feeding trials were conducted daily during the exposure period to account for potential learning behavior, for which fish were fed in exposure arenas at a low density of *D. magna*. Only the low prey density was used and only data from the final trial were analyzed since the aim was to establish the sertraline concentrations to be used for exposure in the following experiment (Section 2.3). Feeding trials proceeded in a similar manner as the functional response baseline experiment (Section 2.1) and feeding was scored as 0 if fish exceeded 10 min to begin feeding or if time to eat 5 prey exceeded 20 min. A log–logistic dose–response curve was fitted to the data for feeding rates at 7 d post-exposure using the *drc* package in R (2012; Ritz and Streibig, 2005).

2.3. Sertraline effects on a and h

Sertraline treatments were chosen based upon the exposure-response results: 0.12, 89, and 300 µg/L, and a solvent control. The treatment of 89 µg/L was chosen as other research also indicates the occurrence of potential effects of SSRIs in this concentration range (Mennigen et al., 2010; Valenti et al., 2009) and 300 µg/L as a positive control. The lowest treatment was included to examine potential effects at environmental concentrations (Christensen et al., 2009). One experimental trial consisted of all treatments with 3 individuals per treatment; experimental trials were replicated four times ($n=12$ per treatment). Fish were allowed to acclimate to individual exposure arenas (3.5 L) for one week, then exposed for 8 d (feeding trials were conducted throughout the acclimation and exposure periods within exposure arenas to control for potential learning behavior). Each individual received food at low and high densities (>24 h apart) at three predetermined time points (>48 h

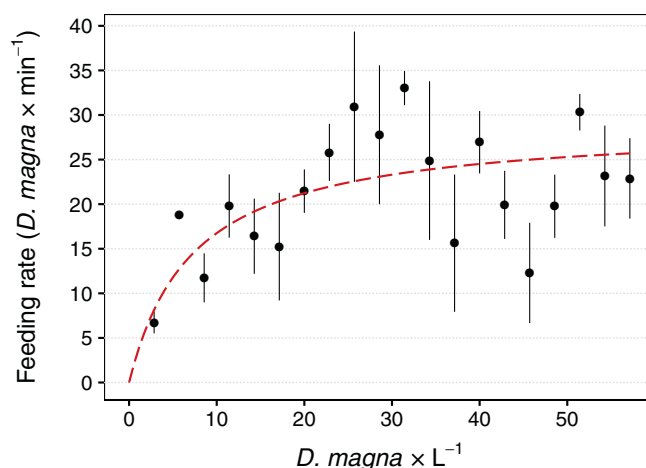


Fig. 1. Functional response baseline experiment: mean feeding rate \pm sd and fitted functional response curve (parameter estimates: $a = 3.97 \pm 1.32$, $h = 0.035 \pm 0.003$) for *P. fluviatilis* predation upon *D. magna*.

apart) within the exposure period. Feeding trials proceeded in a similar manner as the functional response baseline (Section 2.1) and the exposure-response experiments (Section 2.2.). The geometric mean of three feeding trials per individual was calculated to attain average feeding time per individual. Feeding rates were analyzed using a two-way ANOVA (factors: sertraline treatment \times prey density, block: individuals nested within sertraline treatments) in R (2012) after confirming that data met parametric assumptions of normality and homoscedasticity. Post hoc pairwise comparisons among sertraline treatments were made using linear contrasts (corrected by using the studentized range distribution).

3. Results

The Type II functional response curve (Eq. (1)) was fitted to the perch feeding data collected over a large range of prey densities; parameter estimates thereof yielded an attack coefficient $a = 3.97 \pm 1.32$ and handling time $h = 0.035 \pm 0.003$. Because aN describes the slope of the functional response curve and h determines the asymptote (Eq.(1), Fig. 1), we qualitatively chose 30 and 110 prey items per arena (i.e. ~ 9 and 31 individuals L⁻¹, respectively; Fig. 1) to represent attack rate and handling time, respectively. The exposure-response experiment indicated a negative trend in fish feeding rates with increasing sertraline concentrations at 7 d post-exposure (Fig. 2; slope = 1.86 ± 2.06 ,

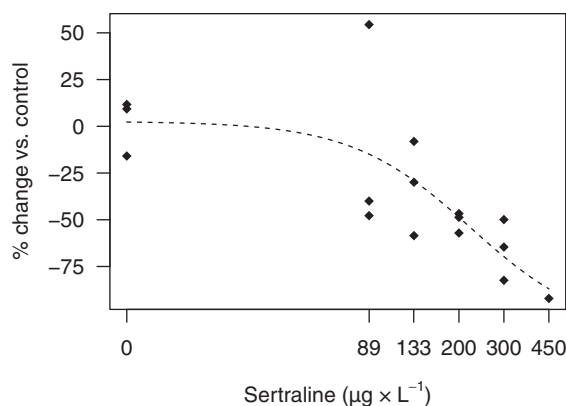


Fig. 2. Exposure-response experiment: *P. fluviatilis* feeding represented as % difference from the control mean plotted vs. sertraline concentration after 7 d exposure, with fitted dose–response curve (slope = 1.86 ± 2.06 , inflection point = 222 ± 289).

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