



Multimodal communication, mismatched messages and the effects of turbidity on the antipredator behavior of the Barton Springs salamander, *Eurycea sosorum*



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ABSTRACT

Prey may use multiple sensory channels to detect predators, whose cues may differ in altered sensory environments, such as turbid conditions. Depending on the environment, prey may use cues in an additive/complementary manner or in a compensatory manner. First, to determine whether the purely aquatic Barton Springs salamander, *Eurycea sosorum*, show an antipredator response to visual cues, we examined their activity when exposed to either visual cues of a predatory fish (*Lepomis cyanellus*) or a non-predatory fish (*Etheostoma lepidum*). Salamanders decreased activity in response to predator visual cues only. Then, we examined the antipredator response of these salamanders to all matched and mismatched combinations of chemical and visual cues of the same predatory and non-predatory fish in clear and low turbidity conditions. Salamanders decreased activity in response to predator chemical cues matched with predator visual cues or mismatched with non-predator visual cues. Salamanders also increased latency to first move to predator chemical cues mismatched with non-predator visual cues. Salamanders decreased activity and increased latency to first move more in clear as opposed to turbid conditions in all treatment combinations. Our results indicate that salamanders under all conditions and treatments preferentially rely on chemical cues to determine antipredator behavior, although visual cues are potentially utilized in conjunction for latency to first move. Our results also have potential conservation implications, as decreased antipredator behavior was seen in turbid conditions. These results reveal complexity of antipredator behavior in response to multiple cues under different environmental conditions, which is especially important when considering endangered species.

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1. Introduction

The use of multiple sensory channels by prey can improve localization, resolution, and amplification of predator cues (Johnstone, 1996). Aquatic prey may use a combination of cues such as visual, chemical, tactile, and electric cues in predator detection (Collin and Whitehead, 2004; Park et al., 2008; Ward and Mehner, 2010; Hettyey et al., 2012). Prey should use the cue (or multiple cues) that provides the most certainty of predator threat (Lima and Steury, 2005), which may change based on cue availability and the physical sensory environment (Endler, 1993). Further, use of multimodal cues can be integrated in an additive manner, triggering a greater response than use of unimodal cues, consistent with the sensory

complement hypothesis (Lima and Steury, 2005). Alternatively, prey may rely primarily on one sensory cue based on their perception or respond in a compensatory manner depending on how the physical environment influences perception of the cues (i.e., turbid vs clear water: Hartman and Abrahams, 2000; Weissburg et al., 2014). Some organisms rely on visual cues for predator detection due to the high information content of visual cues (Hemmi, 2005). However, in some studies with amphibians, chemical cues have been found to affect behavior more than visual cues (Stauffer and Semlitsch, 1993; Kiesecker et al., 1996; Hickman et al., 2004). Testing animals in different environments with a combination of cues can determine which hypothesis for cue use is supported for a given species.

The use of sensory modalities by prey has frequently been found to be dependent on the way the sensory environment affects each cue (Endler, 1993; Weissburg et al., 2014). Turbidity is a form of environmental noise that decreases penetrability of light within the water column and can affect the visual and chemical envi-

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ronment for prey (Utne-Palm, 2002). In habitats where turbidity reduces visual information, some fish compensate for the reduced visual information by increasing antipredator response to chemical cues (Hartman and Abrahams, 2000; Leahy et al., 2011; Ranåker et al., 2012). Further, turbidity can yield a mismatch between visual and chemical cues because turbidity can differentially attenuate certain wavelengths (Walmsley et al., 1980). This indicates that in turbid conditions there may be issues with reliance on multimodal cues for increasing resolution of predator detection because the cues are more likely to become mismatched. Ward and Mehner (2010) found that mosquitofish, *Gambusia holbrooki*, with access to mismatched cues were faster to approach predator visual cues than those with matched visual and chemical cues. These findings may be due to non-adaptive antipredator behavior in turbid conditions or due to predator inspection as an adaptive response to uncertainty. Either way, response to mismatched cues is predicted to differ from matched cues if there is an additive response, especially in clear conditions whereas if an organism uses cues compensatorily then they will only respond to the cue they perceive more strongly in a given environment and use a different cue if their primary cue is harder to perceive. Therefore, the effect of turbidity on antipredator behavior likely depends on the mode of cue use by prey.

Anthropogenic activity such as land development, severe storms, and recreation have increased turbid conditions in waterways, making turbidity an increasing issue for aquatic organisms (Henley et al., 2000; Davies-Colley and Smith, 2001). Much of the work examining behavioral and life history impacts of turbidity has been on fish (Abrahams and Kattenfeld, 1997; Gadomski and Parsley, 2005; Ferrari et al., 2010a; Swanbrow Becker and Gabor, 2012; Chivers et al., 2013), but amphibians also suffer from increased turbidity (Secondi et al., 2007; Schmutz et al., 2008). Turbidity can reduce antipredator behavior in aquatic prey (Gregory, 1993; Miner and Stein, 1993), can decrease generalized predator recognition (Ferrari and Chivers, 2009; Chivers et al., 2013), and can increase mortality (Horppila et al., 2004).

The Barton Springs salamander, *Eurycea sosorum*, is a federally endangered, IUCN red list species (IUCN, 2013) that is neotenic, fully aquatic, and endemic to Barton Springs, Austin, Travis County, Texas. In these spring habitats, fish predators abound including redbreast sunfish, *Lepomis auritus*, and largemouth bass, *Micropterus salmoides* (Labay et al., 2011). Further, *E. sosorum* is threatened by water quality degradation due to increased recreational use, increased land development, and reduced spring flow (Dries et al., 2013), which can increase turbidity (Feng et al., 2012). The turbidity levels of Barton Springs are typically <1 Nephelometric Turbidity Units (NTU), but can spike to ~40 NTU after rain events (White et al., 2003). NTU's measure how much light is reflected off of suspended solids, with higher reflectance (higher NTU's) corresponding to more suspended solids in a water column. *Eurycea sosorum* is often found in clear conditions and avoids silty turbid habitat (Dries et al., 2013). Therefore, increased turbidity might affect the ability of *E. sosorum* to detect and respond to predators if they are unable to compensate for loss of sensory information, or if the sensory information becomes mismatched, potentially decreasing their likelihood of survival in a predatory situation.

Eurycea sosorum (captive-reared) show innate antipredator response to the chemical cues of redbreast sunfish in clear water (Desantis et al., 2013). However, no study has determined whether *E. sosorum* use visual cues for antipredator response or whether turbidity has a significant effect on antipredator behavior, though Thaker et al. (2006) found that closely related *E. nana* show association preferences based on chemical cue or visual and chemical, but not visual alone. We tested whether individuals of *E. sosorum* use visual cues for predator recognition by examining activity of *E. sosorum* in response to visual cues of two sympatric species: predatory

green sunfish, *L. cyanellus* and non-predatory greenthroat darter, *Etheostoma lepidum*.

We also explored cue use of salamanders in response to predators and whether salamanders alter their behavior in response to turbidity. We exposed salamanders to visual and chemical cues of predatory and non-predatory fish, either matched or mismatched in all possible combinations, in both clear and turbid conditions. We wanted to determine a) whether salamanders use visual cues, b) the response of salamanders to mismatched vs. matched cues, and c) the effect of turbidity on response to mismatched vs. matched cues. Our experimental method will also help discern the mechanism/pattern of multimodal cue use in salamanders and whether their behavior supports the additive or compensatory hypotheses for cue use. If salamanders use cues in a complementary/additive manner, then in clear environments we predict them to show stronger antipredator response to matched visual and chemical cues of predators but less to mismatched cues. In turbid conditions we expect decreased antipredator response compared to matched cues in clear conditions. If organisms use sensory compensation, then in turbid environments where visual cues are diminished, we predict that they will compensate for loss of vision by using chemical cues.

2. Experiment 1

2.1. Animal maintenance (for both experiments)

We conducted trials at the San Marcos Aquatic Resources Center (SMARC) in San Marcos, Texas, from June–August 2014 and 24–27 April 2015 between 0800–1800 h. We housed salamanders in large flow-through fiberglass tanks (61 cm D × 81 cm W × 180 cm L) with recirculating, temperature-controlled (21–23 °C) well water. We collected three similarly sized adult green sunfish (*L. cyanellus*, mean standard length ± SE = 157 ± 5.84 mm) via hand-line on site at the SMARC, which we housed in a large flow-through tank partitioned into three equal sections. Davis et al. (2012) found that *E. nana* (a close relative to *E. sosorum*) show antipredator response to the chemical cues of green sunfish. We collected fourteen similarly sized adult greenthroat darters (*E. lepidum*: mean standard length ± SE = 49 ± 0.84 mm) via dip-net from Comal Springs, Texas, which we housed in three 9.5 l flow-through tanks. Davis & Epp (unpublished data) found that *Eurycea nana* (captive-bred) did not show antipredator response to chemical cues of the closely related fountain darters, *Etheostoma fonticola*, and that using darter chemical cue was an equivalent control to water. We maintained all animals on a 12:12 h light/dark cycle and we fed them live blackworms (*Lumbriculus variegatus*) and copepod/amphipod mixtures ad libitum.

2.2. Methods: visual cues

We tested 39 captive-bred, predator naïve, second-generation, adult *E. sosorum* in response to the visual cues of (1) four non-predatory adult greenthroat darters, and (2) one predatory adult green sunfish. Our set-up consisted of two adjacent 9.5 l drip-flow tanks filled with 4.5 l of well water with an opaque divider between the tanks. We placed the salamander in one tank and the fish in the other. Neither species of fish moved much while in the test tanks so we did not quantify their movements. To quantify antipredator behavior in each trial we first allowed each salamander to acclimate for ≥20 min. Following acclimation, we recorded prestimulus activity levels as the amount of time in ambulatory activity (i.e., swimming or walking) for 8 min (Following Epp and Gabor, 2008). We recorded activity because decreased activity is associated with decreased risk of predation in many aquatic organisms

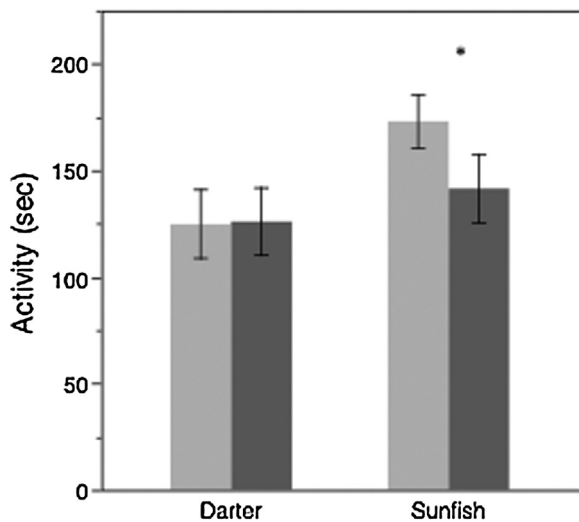


Fig. 1. Mean pre and post activity (s) \pm SE of *Eurycea sosorum* to visual cues of a predatory (sunfish) and non-predatory (darter) fish in clear water. The light color indicates the pre-stimulus activity and the dark color indicates the post-stimulus activity. Asterisks represents significant differences of $p < 0.05$.

when exposed to sit-and-wait predators (Skelly, 1994; Kats and Dill, 1998; Epp and Gabor, 2008). We then introduced visual cues by removing the opaque divider and exposing the visual stimulus of either one adult green sunfish or four adult greenthroat darters (to provide similar size stimulus to a sunfish) in a clear tank adjacent to the focal salamander. In order to be consistent with the design for experiment 2, we also injected 50 ml of clean well water (a control to simulate cue introduction) with a 60-ml syringe attached to airline tubing 10 cm below the surface of the water at a rate of 2 ml/sec (following Epp and Gabor, 2008). We then flushed the tubing with an additional 50 ml of well water at the same rate.

After removing the divider and introducing the water control, we recorded the amount of time *E. sosorum* spent active for another 8 min to determine post-stimulus antipredator behavior. Following the experiment, individuals were sexed (Gillette and Peterson, 2001) and had their snout-vent length (SVL) measured before being placed into a separate tank for tested individuals. We used 3% hydrogen peroxide and water to clean tanks of chemical cues between trials (Epp and Gabor, 2008).

2.3. Data analysis

We performed a paired *t*-test to determine whether pre-stimulus vs post-stimulus activity differed for each treatment (a significant difference in the predator treatment indicates that the pre-stimulus trial also suffices as a control).

2.4. Results

Salamanders decreased activity in the presence of the visual cues of predatory sunfish ($t = -2.68$, $df = 18$, $p = 0.02$). Salamanders did not change their activity in the presence of the visual cues of a non-predatory darter ($t = 0.12$, $df = 18$, $p = 0.91$; Fig. 1).

2.5. Discussion

We found that *E. sosorum* can use visual cues for predator recognition; in the absence of chemical cues, salamanders showed antipredator behavior (decreased activity) in response to visual cues of a predator, but did not respond to visual cues of four non-predatory darters. The response to the predatory fish is consistent with predator recognition.

3. Experiment 2

3.1. Methods: matched and mismatched visual and chemical cues

We tested 200 captive-bred, predator naïve, second-generation, adult *E. sosorum* (SVL > 19 mm; Tupa and Davis, 1976) in response to a native predator, green sunfish, and a non-predator control, greenthroat darter. We tested two visual cue treatments and two chemical cue treatments across two vision levels ($2 \times 2 \times 2$ design): (1) clear (0 NTU), and (2) turbid (mean \pm SE: 20 ± 0.4 NTU). We tested the following combination of cue treatments: (1) matched non-predator visual cues paired with non-predator chemical cues (darter control cues; $n = 25 \times 2$) (2) mismatched non-predator visual cues with predator chemical cues ($n = 25 \times 2$), (3) matched predator visual cues with predator chemical cues ($n = 25 \times 2$), (4) mismatched predator visual cues with non-predator chemical cues ($n = 25 \times 2$; Fig. 2). We did not use salamanders more than once for any treatment and we randomly assigned them to their cue treatments using a random number generator. Prior studies from our laboratory have shown that the pre- and post-stimulus activity do not differ when well water cues are introduced in this species and their close relatives, *E. nana* (Epp and Gabor, 2008; Desantis et al., 2013), therefore the pre-treatment activity functions as a control.

This experiment followed the same basic protocol as experiment 1, with the addition of latency as a response variable and turbidity as a treatment. Following introduction of the chemical and visual cues, we measured time spent active and additionally we recorded latency to first move. Latency to first move is defined as the number of seconds before ambulatory activity was observed. It is also an important measure of antipredator behavior because the longer it takes for cryptic prey to resume activity, the more likely they are to avoid predation, to an extent (Brown and Cowan, 2000; Martín et al., 2009). For turbidity treatments, we added about 2.5 g of bentonite (Sturgis Rock Solid Solutions) to well water and stirred vigorously while using an aerator to maintain suspension, prior to salamander acclimation. An aerator was placed in all treatment tanks to maintain consistency throughout the trials. Turbidity levels were measured at the end of each trial using a calibrated turbidity meter (Hach 2100N Turbidimeter).

3.2. Chemical cue acquisition

Chemical cues were collected by placing each fish in a volume of water proportional to their displacement (230 ml water per 1 ml displacement) for 24 h, after which cues were pooled across individuals of the same species, and frozen until day of use (Mathis et al., 2003; Epp and Gabor, 2008). The same individual recorded all behavior (KZ) and was blind to chemical cue treatment by coding chemical cues.

3.3. Data analysis

We conducted a repeated measures ANOVA on pre vs. post activity as response variables and cue treatment (four combinations) and turbidity as independent variables, followed by a matched pairs post hoc test. In order to determine whether there was a difference in activity due to turbidity alone, irrespective of treatment, we performed a *t*-test of all pre-treatment salamanders, using time active as the response variable and turbidity as the independent variable. We conducted a two-way ANOVA using latency to first move as a response variable and cue treatment (four combinations) and turbidity as independent variables followed by a post-hoc Tukey's HSD test and a power analysis. We tested data normality and equality of variances using a Shapiro-Wilk and Levene's tests, respectively. We used a square root transformation to improve data normality.

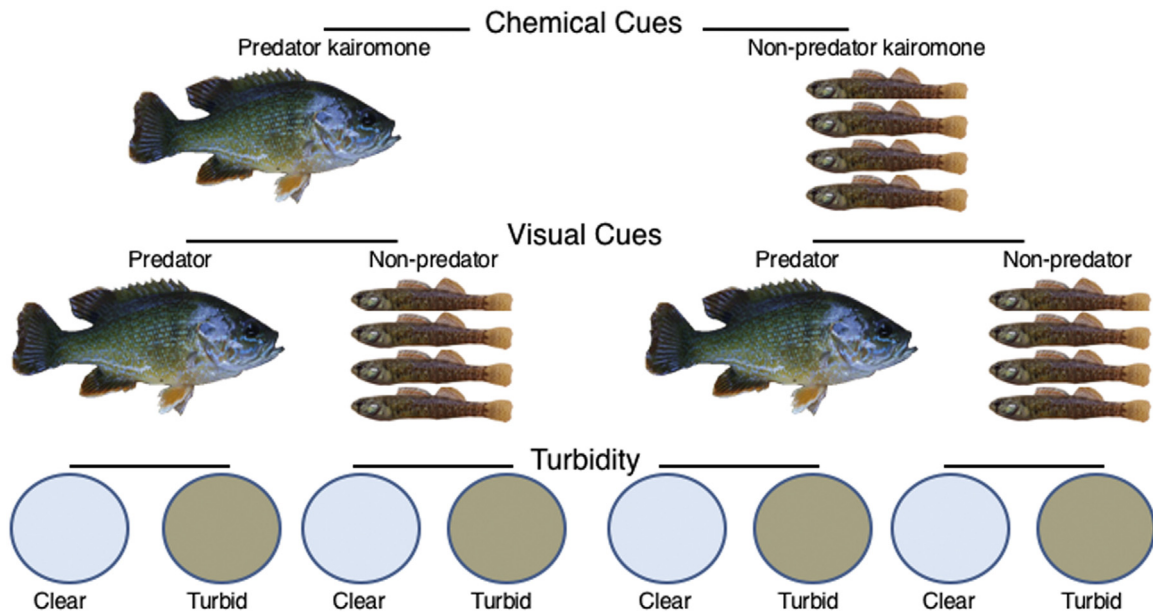


Fig. 2. Schematic of fully-crossed experimental design involving chemical cues and visual cues of predator (*Lepomis cyanellus*) vs. non-predator (*Etheostoma lepidum*), in clear (0 NTU) and turbid (~20 NTU) conditions.

We removed two values that were outliers based on Shapiro-Wilk test. All analyses were performed in JMP Pro (11.2.1).

3.4. Results

There was a significant effect of turbidity on pre and post activity, with salamanders decreasing activity more in clear vs turbid conditions ($F_{1,186} = 5.32$, $p = 0.02$; Fig. 3a). There was no significant difference in pre-treatment activity among salamanders in clear and turbid conditions ($t_{(192)} = 0.706$, $p = 0.48$, 95% CI [-10.2, 21.6]). There was also a significant interaction between activity (time) and treatment ($F_{3,186} = 4.53$, $p = 0.004$) and turbidity ($F_{1,186} = 4.93$, $p = 0.03$; Fig. 3b). We found that salamanders decreased activity in response to predator chemical cue matched with predator visual cues ($t = -4.13$, $df = 46$, $p = 0.0002$) or mismatched with non-predator visual cues ($t = -5.03$, $df = 47$, $p < 0.0001$). No other treatments were significant.

There was a significant effect of treatment ($F_{3,166} = 4.14$, $p = 0.007$) and turbidity ($F_{1,166} = 7.16$, $p = 0.008$; Fig. 3c) on latency to first move. There were no significant interactions between treatment and turbidity. Salamanders increased latency to first move more in response to predator chemical cues mismatched with non-predator visual cues than to non-predator chemical cue either matched or mismatched with visual cue ($p < 0.05$; Fig. 3c). Latency to first move did not differ for mixed and matched cues when the chemical cue was from the predator ($p > 0.05$; $\beta = 0.84$; Fig. 3c). Latency to first move did not differ for matched predator cues with mixed or matched cues when the chemical cue was from the non-predator ($p > 0.05$; Fig. 3c). Salamanders increased their latency to first move more in clear (mean \pm SE: 8.75 ± 0.57) as opposed to turbid conditions (mean \pm SE: 6.68 ± 0.53).

3.5. Discussion

Salamanders decreased their activity and increased their latency to first move more in clear conditions than in turbid conditions. Salamander pre-stimulus activity did not differ between turbid and clear conditions, indicating that the change in behavior was driven by the treatment. Salamanders also showed greater antipredator response (freezing) to chemical cues of predators when mis-

matched or matched with non-predator or predator visual cues. Salamander behavior did not change in response to any combinations containing non-predator chemical cues.

4. Discussion

We examined antipredator response of an aquatic salamander to multiple cues under different environmental conditions. We tested salamander response to mismatched and matched cues in clear and low turbidity conditions. Our results suggest that salamanders use chemical cues over visual cues preferentially, despite having the ability to use visual cues, and that they decrease activity and increase latency to respond more in clear conditions than turbid conditions.

Our findings do not support the additive or compensatory patterns for cue use in antipredator behavior. Contrary to the prediction that the interaction between sensory modalities is dependent on how the sensory environment affects each cue, we found no interaction between turbidity and cue treatment. This suggests that salamanders are not changing their response to different cues depending on the environmental conditions. Instead, salamanders only decreased total antipredator response (i.e., less decrease in activity and decreased latency to first move) in turbid water, not in clear conditions. Our results indicate that low turbidity levels are associated with a decrease in antipredator behavior of this aquatic species found in clear springs, which may contribute to population declines.

Salamanders did not decrease activity to matched non-predator chemical and visual cues, indicating that these salamanders do not perceive darters as predators, as previously found by Davis & Epp (unpublished data). Further, salamanders did not show antipredator behavior towards mismatched non-predator chemical cues with predator visual cues, but responded to mismatched and matched visual cues with chemical cues of predators. Given the results of the first experiment, this lends support that salamanders preferentially use chemical cues of predators over visual cues when both are present, instead of compensating or complimenting chemical cues with visual cues. Similarly, amphibian prey have been found to discriminate between predators and non-predators in response to chemical cues, but exhibited antipredator behavior

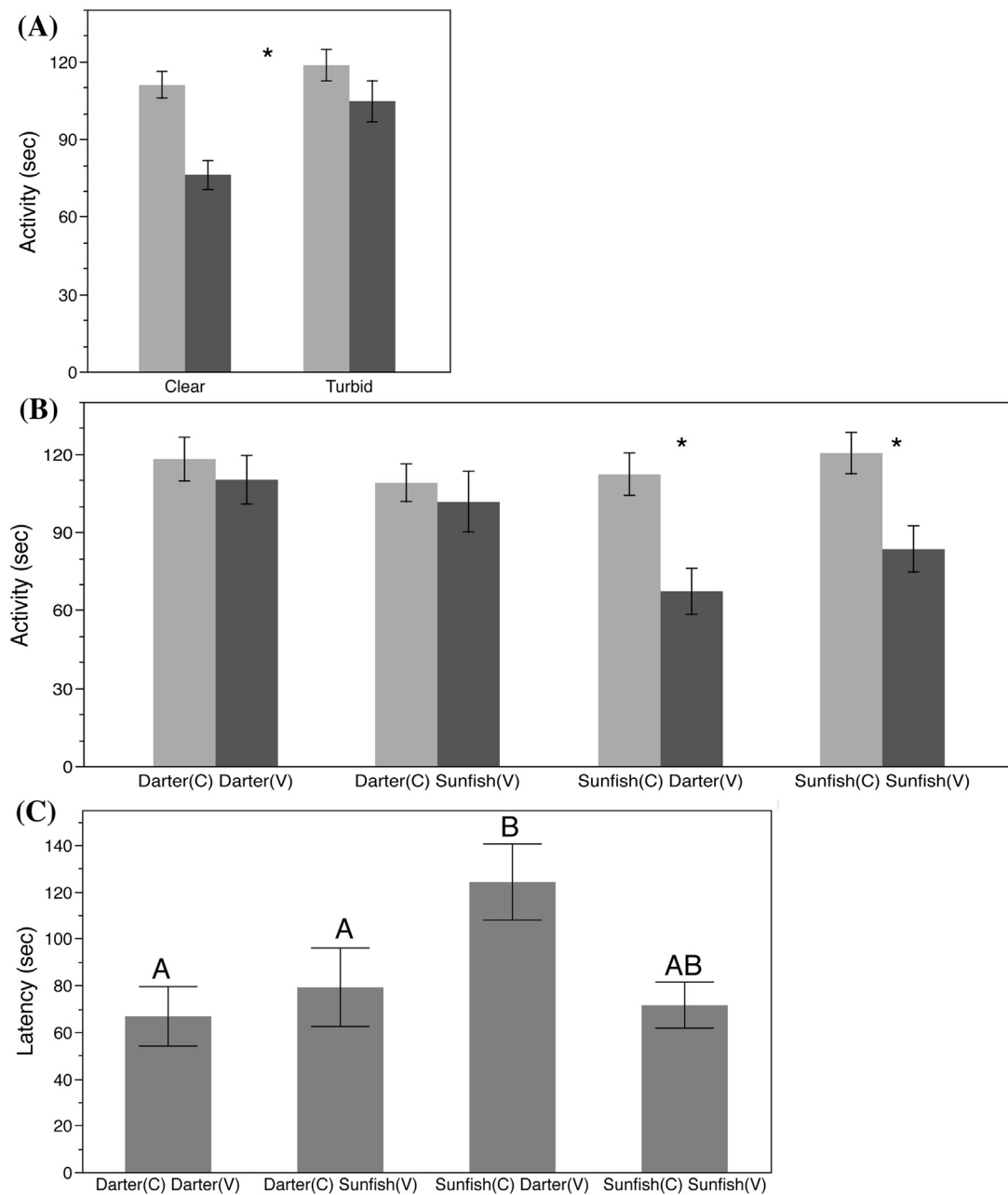


Fig. 3. (A) Mean pre and post activity (s) ± SE of *Eurycea sosorum* in clear vs turbid water. Salamanders decreased activity more in clear water than turbid water. The light color indicates the pre-stimulus activity and the dark color indicates the post-stimulus activity. Asterisks represents significant differences of $p < 0.05$. (B) Mean activity pre and post (s) ± SE of *Eurycea sosorum* in response to both predator (sunfish) and non-predator (darter) visual and chemical cues. Salamanders decreased activity more in response to the predatory chemical cues, both mismatched and matched with visual cues of predators and non-predators, compared to all other treatments. The light color indicates the pre-stimulus activity and the dark color indicates the post-stimulus activity. Asterisks represents significant differences of $p < 0.05$. (C) Mean latency (untransformed) to first show ambulatory activity (s) ± SE of *Eurycea sosorum* after introduction of chemical and visual cues of predatory sunfish and non-predatory darters in clear and turbid water. Salamanders increased latency to move in response to mismatched predator chemical cues, compared to both matched and mismatched non-predator chemical cue combinations. Salamanders decreased the latency to first move in turbid conditions. Different letters indicate significant differences of $p < 0.05$.

to both predators and non-predators in the presence of visual cues, suggesting the importance of chemical cues over visual cues for accuracy in recognition (Mathis and Vincent, 2000; Hickman et al., 2004). The reliance primarily on chemical cues may be owing to their ubiquitous nature and persistence in freshwater (Ward and Mehner, 2010) although visual cues are typically perceived quicker than chemical signals (Endler, 1993). Alternatively, it may be owing to decreased costs associated with chemical cues especially given that *E. sosorum* is usually found under rocks and has reduced eyes. If this is the case, then this species is using cues that match their perception strengths. Nonetheless, *E. sosorum* demonstrated the ability

to discriminate between the visual cues of a predator and the visual cues of four non-predatory fish, which was then abated with the addition of non-predator chemical cues.

When we explored latency to first move we found that salamander response to matched predator cues did not differ significantly from matched or mismatched non-predator chemical cues, while mismatched predator chemical cues differed significantly from non-predator chemical cues. These results suggest that although chemical cues are preferentially used for antipredator response, visual cues may still contribute to their antipredator response. The stronger response to mismatched predator chemical cues with

non-predator visual cues may be attributed to the fact that this treatment introduces a novel cue combination in a high threat environment (owing to the presence of predator chemical cues), which may induce a heightened response due to threat aversion. Recent studies have shown increased reaction to novel predator cues in high risk environments (Brown et al., 2013, 2014; Chivers et al., 2014; Brown et al., 2015), so it is possible that in such environments, signals that are not consistent with chemical cues elicit a stronger response in terms of latency to first move. Alternatively, the antipredator response is stronger because the salamanders perceived the four fish as a greater threat in the presence of predator chemical cues (and they may not have been able to identify them as predators vs non-predators).

Under turbid conditions, salamanders showed reduced antipredator behavior to both predators and non-predators, in the form of no change of activity and decreased latency to first move, as compared to in clear conditions. This suggests that either turbidity is interrupting chemical cues or that salamanders perceive turbidity as a form of cover. The ‘turbidity as cover’/visual refuge hypothesis predicts that predation risk decreases in turbid conditions due to decreased conspicuousness of prey, making reduction in antipredator behavior adaptive by reducing costs associated with unnecessary antipredator behavior (Gregory, 1993; Ajemian et al., 2015). Our results indicate that while salamanders can use both visual and chemical cues for predator recognition they may not compensate for information loss in turbid environments. Although other studies have found that in absence of complementary cues prey show a more risk-averse response (Leduc et al., 2010; Elvidge and Brown, 2014), this is not corroborated by our results showing reduced response in turbid conditions.

Although turbidity can affect foraging of predators (Johansen and Jones, 2013), we argue that it is unlikely that risk of predation on *E. sosorum* is decreased in turbid conditions. In fact, predation risk may be greater in turbid conditions in systems where only predators are adapted to turbid conditions, where turbidity does not affect the predator’s ability to forage, but decreases antipredator behavior in prey (Abrahams and Kattenfeld, 1997; Granqvist and Mattila, 2004). *Lepomis cyanellus* is tolerant of turbidity extremes: high turbidity does not affect the ability of *L. cyanellus* to forage or attack (Heimstra et al., 1969). In addition, juveniles of the related bluegill sunfish, *L. macrochirus*, had greatest foraging efficiency at a low turbidity level (~20 NTU) (Miner and Stein, 1993). Because *E. sosorum* is more adapted to clear environments, turbidity may have a greater impact on the ability of *E. sosorum* to detect predators and less of an impact on the ability of *L. cyanellus* to capture prey, thus making *E. sosorum* potentially more susceptible to predation. Effects of turbidity on other behaviors, such as foraging and mating, need to be assessed to determine the fitness impact of turbidity on this endangered species.

The diminished antipredator behavior in turbid conditions has conservation implications for *E. sosorum*. We studied a low turbidity value of 20 NTU, so our results are a conservative measure of changes in antipredator behavior because turbidity levels spike to double what we studied (White et al., 2003). Chemical cues may be affected by the same particles that increase turbidity (Ferrari et al., 2010b), which was not tested by our use of bentonite, so future studies should examine how salamanders respond to environmental changes that affect more than one sensory modality. Antipredator response to predators may be mediated by experience and turbidity could impact learning. van de Meutter et al. (2005) found that antipredator behavior is decreased if organisms are reared and subsequently tested in turbid conditions but antipredator behavior is unaffected when reared in clear conditions and tested for response in turbid conditions. Further, Munoz and Blumstein (2012) stress the importance of studying the interplay

between learning and turbidity for reintroduction purposes, indicating that determining the response of wild-caught salamanders, which are both predator-experienced and turbid-experienced, is of great importance. Therefore, experience with learning in turbid conditions may be important, which can have significant implications for hatchery-raised species, especially those that are being raised with the potential of reintroduction.

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