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Developing and testing a cognitive field test for lead-exposed wild songbirds

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Developing and testing a cognitive field test for lead-exposed wild songbirds

Abstract

Anthropogenic impacts drive global changes that can negatively impact wildlife. For example, using lead pipes in Flint, MI has resulted in contaminated drinking water that is likely to have moved into the environment through irrigation. Juvenile American robins (*Turdus migratorius*) captured in watered parks of Flint have blood-lead levels considered dangerous to brain functioning. The purpose of this study was to develop and test a feeder puzzle for assessing the cognitive effects of lead in wild urban songbirds. We predicted that songbirds could learn how to feed from a feeder puzzle, and if they visited the feeder in groups, then the solution to the puzzle could potentially be transferred by social learning within the group. Furthermore, we predicted that cognition task success could be assessed by monitoring how species interact with the feeder. The prototype was visited by American robins (*Turdus migratorius*), house sparrows (*Passer domesticus*), and European starlings (*Sturnus vulgaris*). Sparrows and starlings demonstrated the potential for social transmission by visiting the puzzle in small groups. Furthermore, starlings solved the puzzle to access food, suggesting that the feeder could be used to evaluate how lead affects songbird cognition and learning.

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DEVELOPING AND TESTING A COGNITIVE FIELD TEST FOR LEAD-EXPOSED WILD
SONGBIRDS

By

Samantha Glowacki

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Abstract

Anthropogenic impacts drive global changes that can negatively impact wildlife. For example, using lead pipes in Flint, MI has resulted in contaminated drinking water that is likely to have moved into the environment through irrigation. Juvenile American robins (*Turdus migratorius*) captured in watered parks of Flint have blood-lead levels considered dangerous to brain functioning. The purpose of this study was to develop and test a feeder puzzle for assessing the cognitive effects of lead in wild urban songbirds. We predicted that songbirds could learn how to feed from a feeder puzzle, and if they visited the feeder in groups, then the solution to the puzzle could potentially be transferred by social learning within the group. Furthermore, we predicted that cognition task success could be assessed by monitoring how species interact with the feeder. The prototype was visited by American robins (*Turdus migratorius*), house sparrows (*Passer domesticus*), and European starlings (*Sturnus vulgaris*). Sparrows and starlings demonstrated the potential for social transmission by visiting the puzzle in small groups. Furthermore, starlings solved the puzzle to access food, suggesting that the feeder could be used to evaluate how lead affects songbird cognition and learning.

Introduction

Ecosystems often pay the price for a growing human population, experiencing habitat loss, local extinction, atmospheric and water pollution, consequences of herbicides and pesticides, and overuse of natural resources, among other impacts (Western 2001). Biodiversity loss, and the loss of ecosystem services that biodiversity provides, is a consequence of human activity (Western 2001). Anthropogenic activity can also negatively impact ecosystem energy pathways and productivity (Western 2001). These influences can be further classified as activities that alter the physical environment and activities that ultimately change animal behaviors (Steidl and Powell 2006).

Changes in animal behavior that are induced by human activity can have consequences because behavior sets the foundation for ecological patterns and relationships. Wildlife conservation therefore relies on understanding behaviors and how they can be changed to positively benefit populations (Greggor et al. 2014). Importantly, animals possess the ability to adapt learning and memory processes in response to environmental triggers and stressors, such as changes to their habitats. Adaptations in cognition, like morphological adaptations, may therefore help or hinder species responses to environmental changes (Greggor et al. 2014).

Cognition and Learning

Cognition encompasses all the processes involved with the acquisition, retention, and utilization of information (Dukas 2004), and these processes define behavioral characteristics of different species (Morand-Ferron et al. 2015). The processes involved in discrimination, learning, and memory are valuable to different species depending on the context in which they are used. The costs and benefits of cognitive abilities rely on

selection to maximize the fitness of the species (Rowe and Healy 2014). Environmental changes that are induced by humans can impart selection pressures on wildlife and the natural cognitive specializations they possess (Greggor et al. 2014).

There is still a lack of understanding of the effects of individual cognitive differences within populations. One reason for the lack of research in this area is because cognition can be difficult to study in the wild (Morand-Ferron et al. 2015). However, despite the difficulty, it is important to investigate the cognitive processes underlying behaviors in order to understand the changes that occur within populations when environments change. Songbirds are sensitive to environmental changes, and they have become sentinels for the consistency of an environment due to the ease in which laboratory and field biology can be used to observe physiological and behavioral changes (Clayton et al. 2009).

Cognition in Songbirds

Learning is an important process in cognition that involves the development of neural representations for new information, which creates an association between an existing concept and new stimuli (Dukas 2004). Songbirds provide a good model for understanding cognition, learning, and behavior because they are adaptive by responding to both environmental cues and ecological niche requirements (Clayton et al. 2009). Additionally, they are relatively easy to observe in their environments and learn many different behaviors, including habitat preferences (Szymkowiak et al. 2017), foraging methods and habits (Slagsvold and Wiebe 2011), and vocal outputs (Brainard and Doupe 2013). Because of these abilities, as well as those of sensory processing and motor

output, songbird species provide a good model for the study of learning and memory disorders in humans (Brainard and Doupe 2013).

Because there is a lack of research on avian cognition in general, there are few published accounts of cognition tests designed to analyze learning, particularly in free-living birds. Some tests evaluate memory formation and the ability to learn new stimuli. Studies have focused on song production and test the neural responses to song playbacks in adult birds who have experienced early nutritional stress (Bell et al. 2018). Learning has also been tested in non-songbird species through the production of cognitive tasks, which is a useful method for observing learning and cognition in the wild. Examples of different tasks include spatial-reversal learning tasks, which involve the development of a spatial preference and then require the acceptance of its reversal. These tasks match natural scenarios where a habitat is changed and as a result, food availability and accessibility are changed too (Cauchoix et al. 2017). In this case, cognition is tested through an individual's ability to learn a "reward-stimuli association" and then how to forego a previous association and develop a novel one. Mastery of this task requires spatial associative learning, inhibitory control, and/or cognitive flexibility (Cauchoix et al. 2017).

Other examples of cognition tests evaluate motor skill learning through inhibitory control tasks. These tests analyze how long it takes for a bird to forage from an object with hidden food, which uses shaping mechanisms to train individuals to overcome the obstruction (Shaw 2017). Shaping mechanisms that involve advancing learned behaviors through the reinforcement of successive steps leading to a final desired behavior are commonly used in psychology (American Psychological Association 2018). Other

memory formation and learning tasks test color preference and discrimination, color reversal, symbol discrimination, and motivation. These are all typically accompanied by the incentive of food, where choosing the correct option in a pallet of multiple food slots rewards individuals with that food (Shaw et al. 2015). Motor tasks are also commonly used to analyze learning in birds, and performance can be influenced by previous experience and body condition; individuals who have undergone prior reinforcement perform better, as do those who have a healthier body condition (Shaw 2017).

Cognitive Effects of Heavy Metals

Certain environmental factors can disrupt cognitive processes like learning in many species. Exposure to heavy metals, such as lead, cadmium, arsenic, and methylmercury, has been correlated with negative cognitive effects and decreased neuron functionality (Karri et al. 2016). These impacts have been well described in humans, given the lack of general research on this topic in wild birds. Lead hinders cognition by disrupting various neural processes and receptors that are involved in associative learning, such as N-methyl-D-aspartate (NMDA) (Karri et al. 2016). This receptor aids memory formation by controlling long-term potentiation in the hippocampus (Newcomer et al. 2000). A functional hippocampus is critical to retaining and using information for adaptive behaviors (Rubin et al. 2014). Lead exposure has negative effects on avian physiology, but not much is known about how it impacts cognition and learning. For example, exposure can alter both the shape and function of the kidneys, bones, and the central nervous system (Haig et al. 2014). Lead exposure also interferes with synaptic signal transmission by mimicking calcium (Haig et al. 2014).

Nestling development is a time of intense neural development; thus, lead exposure may be more detrimental to nestlings than adults (Haig et al. 2014). This suggests that younger populations may be more susceptible to the cognitive consequences of lead exposure. Furthermore, neurological impacts depend on the chemical form of lead, the amount of exposure, and the length of exposure time as well as the age, sex, and overall health of the bird at hand (Haig et al. 2014). Studies involving nestlings of different bird species show that lead exposure can decrease health, hinder foraging behaviors, decrease coordination, and decrease overall survival rates (Haig et al. 2014).

Studying the Cognitive Effects of Lead Exposure

Hundreds of cities across the U.S. have been found to have lead-contaminated drinking water that is unsafe for children. While lead is unhealthy for anyone, exposure is especially detrimental to childhood development. A “safe” level of lead in the blood has not yet been identified, but a baseline of five micrograms per deciliter is now used to identify children with elevated blood-lead levels compared to their peers (Centers for Disease Control and Prevention 2019). One city that is notorious for lead-contaminated drinking water is Flint, MI. When Flint’s first water distribution system came to be in the late 1800s, the City of Flint required that all connections to the system be made with lead pipes (Masten et al. 2016). By the mid-1900s, the source of water in the City of Flint changed from the Flint River to Lake Huron water treated by the Detroit Water and Sewage Department (DWSD). This was due the river’s poor quality water from unregulated industrial waste and the need to support a growing population (Masten et al. 2016).

Although the Flint River was no longer the main source of water, it was still treated a few times a year by the Flint Water Service Center backup facility (Masten et al. 2016). In 2013, the water source switched back to the Flint River to reduce the costs of treated water by DWSD, and concerns began to arise about water quality by 2014. Water samples were taken to address health concerns, and these samples exhibited elevated levels of lead that leached from the pipes (Masten et al. 2016). While it is not known how much lead may have contaminated the surrounding environment, preliminary data suggests that American robins (*Turdus migratorius*) living near watered lawns have blood-lead levels that would be deemed dangerous in young children (J. M. Cornelius, unpublished data). Lead-contaminated water is likely to have moved into the environment through irrigation, thus exposing the local wildlife to lead as well.

The goal of this study was to develop a cognition task for wild birds that could be used to assess the cognitive abilities of various bird species. The eventual goal of this project is to assess the impacts of lead exposure on songbird development in Flint. We hypothesized that songbirds could learn how to feed from a feeder puzzle and if they visited the feeder in groups, then the solution to the puzzle could potentially be transferred by social learning within the group. Because lead hinders cellular processes in the avian nervous system (Haig et al. 2014), we speculate that lead could have negative impacts on avian cognition and learning, just as it does in humans. We further hypothesize that if exposure to lead early in development negatively impacts avian cognition, then birds with elevated blood-lead levels will perform poorly on the cognition task in comparison to healthier birds. The present study serves as an approach to address this larger hypothesis. We therefore developed a feeder puzzle cognition test to

implement in the wild for determining its efficacy in evaluating cognition in future studies.

Materials and Methods

To determine whether creating a cognition test would allow us to assess avian cognitive abilities, this study had two parts: cognition test development and implementation of the test.

Cognition Test Development

To test songbird cognition, we developed a feeding apparatus that hindered access to food. Hindering access to food is useful for studying cognition because it requires individuals to learn how to maneuver an obstruction to reach a reward. As learning abilities are indicative of cognition, observation of learning is a good measure of cognition in the wild. To study avian cognition, we developed a prototype with swinging doors and a food incentive designed to simulate a motor task used in other studies (Shaw 2017).

We created our feeder puzzle using a PVC pipe, pieces of plastic, and plywood as a foraging board. The PVC pipe (7.6cm diameter) was cut to produce a 23cm tube. A 2cm hole was carved out of either side of the tube, about 2cm from the bottom. To cover the holes, two pieces of clear plastic (4cm x 5cm) were loosely nailed into the tube so they could swing side to side in order to hide or reveal the openings. The slots were designed to reset automatically, requiring participating birds to learn how to maneuver the plastic doors in order to access food. We used clear plastic doors to allow birds to see the contents of the feeder to enhance the effects of using food as an incentive. A PVC strainer cap was placed on top of the tube to cover the opening, and to seal it off from the

environment, a piece of plastic (15cm x 15cm) was glued on top. This piece of plastic was extended past the sides of the PVC pipe to provide some rain protection to the foraging board and the contents in the tube. The whole apparatus was nailed to the plywood board using angled brackets (Figure 1A). Dried mealworms were placed inside the feeder to act as the incentive for learning the task (Figure 1B). This food was chosen due to the omnivorous nature of many birds and the accessibility in purchasing bags of dried mealworms. As these worms did have a smell to them, we expected that the scent would attract various species.



Figure 1. (A) The prototype feeder puzzle used at all locations in this study. (B) Mealworm placement inside the PVC tube of the prototype.

Testing Sites

We chose multiple testing sites for feeder implementation. Determining which locations would be best for deploying the prototype involved placing several plywood foraging boards in different areas with mealworms placed on and around them. This tactic was used to train birds to visit these areas by using mealworms as an incentive. We were also able to track the amount of bird activity by monitoring how quickly the

mealworms needed to be replenished. The three sites that were selected needed the most mealworm replenishment, which meant that activity was high. All sites were near trees on flat land and located in urban areas near where humans resided in Ypsilanti, MI and Grand Rapids, MI. The duration of time for which the cognition test was deployed at each site depended on the level of bird activity. All interactions with the prototype feeder puzzle were tracked using a motion-sensing camera trap, which was placed about one meter away to capture the apparatus and the areas surrounding it (Figure 2).



Figure 2. Experimental setup for testing cognition using the prototype feeder puzzle and motion-sensing camera trap.

Stepwise Shaping Mechanism

To test the cognitive and learning abilities of birds interacting with the feeder puzzle, a stepwise shaping mechanism was used. This mechanism followed the traditional use of shaping in psychology, which reinforces successive steps until a desired behavior is reached (American Psychological Association 2018). Step one of this process involved keeping one plastic door open while baiting the board, allowing birds full access to the contents of the feeder (Figure 3A). The method for keeping the door open changed throughout the study, but the final mechanism used involved taping the door to the tube

with masking tape. Step two involved keeping the door open but not baiting the board. Step three of the process involved keeping the plastic door halfway open while baiting the board, so birds had limited access to mealworms (Figure 3B). Step four involved keeping the door halfway open while ceasing to bait the board. Step five involved having all doors closed while baiting the board (Figure 3C), eventually leading to step six, which involved cessation of baiting once more.

A)



B)



C)



Figure 3. (A) Step one of the stepwise shaping mechanism, allowing full access to the contents of the feeder puzzle. (B) Step three of the stepwise shaping mechanism,

allowing limited access to the contents of the feeder puzzle. (C) Step five of the stepwise shaping mechanism, requiring successful maneuvering of the puzzle to access contents.

Bird activity with the feeder puzzle was measured by downloading and observing the photos taken by the camera trap. Data collection involved keeping track of which bird species approached the board and showed interest in the feeder, how many birds approached the board at once, which levels of the stepwise shaping mechanism were met, and the time required (latency) to solve the puzzle. The latency for solving the puzzle was recorded in seconds by monitoring individuals from when they landed on the board until they reached the mealworms inside the feeder. This measurement was deciphered by the timestamps on the photos taken by the camera trap. Only the birds that successfully maneuvered the feeder puzzle with closed doors were recorded for latency to solve. The potential for social transmission in groups was observed via photographs taken by the camera trap. We considered the potential for social transmission to be the presence of other individuals while a bird attempted to reach the food inside the feeder puzzle. Groups of birds were believed to be observing the task.

Results

American robins (*Turdus migratorius*), house sparrows (*Passer domesticus*), and European starlings (*Sturnus vulgaris*) interacted with the feeder puzzle. Starlings successfully solved the puzzle and approached the feeder in small groups with a maximum of three individuals on the plywood board simultaneously. Sparrows also approached the feeder in small groups with a maximum of three individuals on the plywood board simultaneously. Robins participated with the feeder puzzle individually and with less probability for social transmission.

Stepwise Shaping Mechanism

All individuals showed interest in the apparatus and ate mealworms from the foraging board base. Robins did not complete any of the shaping mechanism steps, sparrows completed steps one through four, and starlings completed steps one through six. Starlings were the only participants that retrieved food when no doors were open and the board was not baited (Table 1).

Table 1. Species completion of the feeder puzzle shaping mechanism steps.

<i>Step #:</i>	<i>Step Description:</i>	<i>Completion:</i>
Step 1	one door open, bait the board	Sparrows & Starlings
Step 2	one door open, no bait	Sparrows & Starlings
Step 3	one door halfway open, bait the board	Sparrows & Starlings
Step 4	one door halfway open, no bait	Sparrows & Starlings
Step 5	no doors open, bait the board	Starlings
Step 6	no doors open, no bait	Starlings

Completion of the Feeder Puzzle by Starlings

Starlings successfully maneuvered the feeder puzzle to access food, but sparrows and robins were unable to do so (Figure 4). Successful completion of the cognition task was characterized by the bird physically moving the door open and sticking their head inside the tube, retrieving mealworms (Figure 5). For the starlings that retrieved the food

inside the feeder, 20 visits were recorded. There was no change in the latency to solve the puzzle across trial completions (linear regression, $F_{1,19} = 0.89$, $P = 0.36$) (Figure 6).

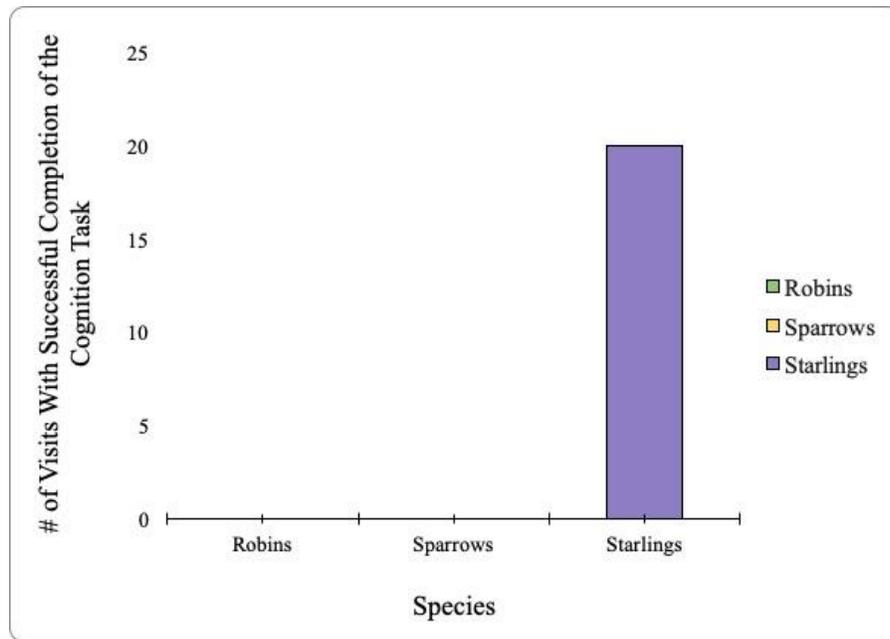


Figure 4. Number of visits with successful completion of the feeder puzzle for robins, sparrows, and starlings.



Figure 5. Example of a starling successfully accessing food from the feeder puzzle by moving the lid open and sticking its head inside.

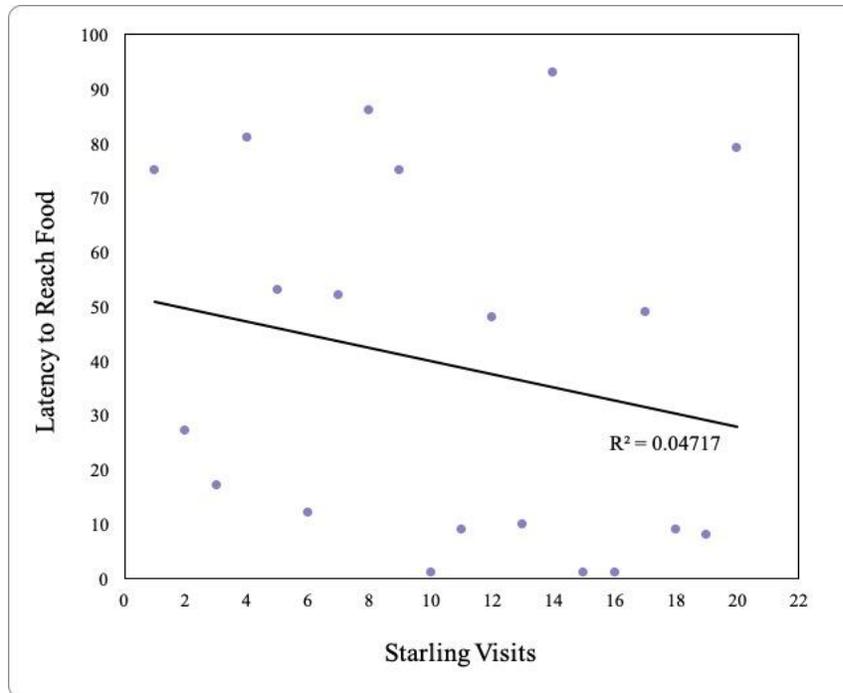


Figure 6. Latency to solve the feeder puzzle throughout different starling visits. Linear regression, $F_{1,19} = 0.89$, $P = 0.36$.

Discussion

The results of this experiment suggest that the feeder puzzle may be a promising tool for analyzing the effects of lead on avian cognition. American robins, house sparrows, and European starlings showed interest in the feeder and foraged from the base of the apparatus. While robins used the feeder individually, sparrows and starlings used the feeder in groups of up to 3 participants, suggesting the potential for social transmission of information in at least some species. This indicates that the feeder puzzle is a feasible method for recording bird behavior and cognition. Additionally, each species differed in their completion of the shaping mechanism steps and starlings were the only species to successfully maneuver the feeder puzzle to access food. Starlings may

therefore be the ideal species on which to conduct future studies concerning the impacts of lead on cognition.

The feeder puzzle may be more effective for species that have a more social foraging nature because of the social transmission of foraging skills. Sparrows and starlings demonstrated potential for social transmission of foraging strategies for interacting with the feeder. Although robins approached the board individually, sparrows and starlings approached the board in small groups and completed more of the shaping steps than the robins did. This suggests that individuals of some species may demonstrate the task for others, and by doing so, other members of these species may be able to learn to reach food on the board or inside the feeder through social transmission of information. Interestingly, European starlings were the only species that successfully maneuvered the feeder puzzle to access food from inside. This may be due to their social foraging nature. Starlings spend more time foraging under social conditions, and if conspecifics are present in a feeding area, it is likely that other starlings will return to that area to forage (Vásquez and Kacelnik 2000). This may have given starlings more opportunities to forage because they spent more time in the area when individuals continued to return to the feeder. It is possible, however, that we did not capture all social interactions near the board for other species because our camera placement may not have captured individuals who watched foraging behavior from further than a meter or two. To find other social species, we may need to observe the foraging mechanisms used by species residing in potential testing sites.

While each species that visited the feeder interacted with it by foraging on mealworms from the platform, they varied in the number of shaping mechanism steps

they successfully completed. While robins did not complete any of the steps, sparrows and starlings were able to advance through most of them. This suggests that using a shaping mechanism successfully allowed some species to learn how to maneuver the task, which may aid researchers in monitoring learning progress and cognitive abilities. Starling success of all six shaping steps indicates that this prototype can measure avian cognition in the field. Their process of accessing food could be observed and recorded via the time stamps on the camera trap. The lack of a trend in latency suggests that either different individuals were using the feeder each time or they were not learning to complete the puzzle faster. Although we recorded 20 successful starling visits, we are unsure whether it was the same few individuals or 20 different individuals. The use of banding would be beneficial in the future to track learning and compare individual cognitive abilities based on factors like body condition and blood-lead levels.

Comparing Starling, Sparrow, and Robin Performance

European Starlings

Habitat preferences and foraging methods may have played a role in European starling success with accessing food from the feeder. Starlings are found living near humans and commonly use mowed lawns, city streets, and fields for foraging (The Cornell Lab of Ornithology 2019). Therefore, starlings may be more inclined to interact with a novel stimulus in an urban setting because of their preference for this type of habitat. Starlings also may have exhibited lower neophobia – hesitation to interact with a novel stimulus (Moldoff and Westneat 2017) – which is typical of invasive, urban birds. Their exploratory foraging behavior may have aided them in completion of the task, as well as their strong jaw muscles that they use to force open their bills when they forage in

soil (The Cornell Lab of Ornithology 2019). The strength of their jaw muscles and larger size may have helped them push open the plastic doors to access the mealworms inside the feeder compared to the smaller house sparrow.

Starling success at solving the puzzle may also be explained by an operant conditioning learning model that posits that an individual is more likely to choose a foraging option when they have recently been rewarded by choosing that preference (Kacelnik and Bateson 1996). Depending on how often an individual returned to the feeder, the consistency of obtaining food when the task was completed likely increased the probability that the individual would complete the task subsequently. This shows that mealworms were a strong incentive for learning the task, since being rewarded with a high-quality food item increased the likelihood of trying to obtain food again.

Additionally, starlings are common birds to find foraging from bird feeders in backyards, especially those that are tube-shaped (The Cornell Lab of Ornithology N.d.). This may explain why they readily approached our prototype. Because previous experience is related to higher success rates for cognition tasks in other songbirds (Shaw 2017), these starlings may have gained prior experience foraging from other bird feeders and were thus able to make an association between previous and novel stimuli. Although, house sparrows and American robins may also be frequent feeders as well.

House Sparrows

House sparrows may have been less successful than starlings at solving the puzzle because they exhibit different foraging behaviors. Sparrows were able to pass through several shaping steps to access food, and they readily approached the apparatus with no signs of hesitation in retrieving mealworms from inside the tube when the doors were

open. When the doors were closed, sparrows still approached the feeder as though they would attempt to open the doors, but none of the subjects successfully did so. Urban sparrows are thought to have increased behavioral plasticity, leading to faster habituation in response to new stimuli and humans (Vincze et al. 2016). The sparrows observed in this study came from an urban setting, so their willingness to approach the feeder puzzle and forage from in and around the apparatus may be due to increased behavioral plasticity. Other studies have tested sparrow habituation, and our findings coincide with the notion that sparrows will habituate to new objects that appear suddenly near a normal food source (Moldoff and Westneat 2017).

Other behavioral factors may have influenced sparrow performance. Although urban sparrows can habituate to stimuli, they also appear to exhibit neophobia by waiting to approach a new stimulus when it is first implemented (Moldoff and Westneat 2017). Our observations are not consistent with house sparrows having high levels of neophobia, as they approached the feeder puzzle immediately and began foraging. This fast habituation may be why house sparrows advanced through so many shaping steps and why they have been such successful urban adapters. If the cognition task had been implemented for a longer period of time, sparrows may have eventually learned how to open the doors with their behavioral plasticity and ability to habituate to new situations. However, their smaller body size and bill size may have prohibited them from successfully completing the task.

Physiological factors also may have played a role in sparrow performance. The ability for sparrows to exhibit innovative behaviors exemplifying cognitive abilities and problem-solving seems to be impacted by factors like corticosterone (CORT) levels

(Bókonyi et al. 2014). House sparrows with higher levels of CORT appear to be less efficient learners and take longer to solve food acquisition tasks (Bókonyi et al. 2014). Sparrows may have performed less well on the cognition task due to higher levels of CORT impeding their problem-solving abilities for opening the doors. However, there is no reason to suspect that house sparrows at our test sites would be more or less exposed to stressors than the European starlings or American robins in this study.

American Robins

The foraging behaviors of American robins may have interfered with their completion of the cognition task. In our observation of this species, they were willing to forage from the plywood base of the apparatus when it was baited, but they did not appear to attempt the task. Motor tasks are a good measure of cognition in species using ground foraging mechanisms (Shaw et al. 2015), such as robins, and previous foraging experiences are helpful with task completion by other songbirds (Shaw 2017). Our results are not consistent with these findings because robins did not attempt to retrieve food from inside the feeder or try to open the doors to gain access. One explanation could be that the feeder puzzle differed from other bird feeders the robins may have used. Robins typically interact with platform feeders and ground feeders (The Cornell Lab of Ornithology N.d.), so the tube-shape of this cognition task may have been a novel stimulus for them. Additionally, adults tend to forage independently, possibly because of their highly territorial behavior during the spring and summer breeding seasons (Vanderhoff et al. 2020). However, when juveniles fledge, it is probable that they would forage close to their parents, thus allowing for potential social transmission of information. This

indicates that if the cognition task could be completed by robins, social transmission could be possible in a different context (i.e., post-fledge).

Time constraints and responses to other species may have also impacted the ability for robins to complete the cognition task. The test site in which robins participated was used for a limited time. If the feeder was implemented longer at this site, it is possible that robins would have eventually learned to complete the task. It is interesting, however, that robins only participated at one of the test sites and appeared to ignore the apparatus at the other sites where sparrows and starlings were present. Robins are territorial and protective of their nests (Vanderhoff et al. 2020), so it is possible that the presence of other species kept them away or the apparatus was placed outside their territories.

Evaluating Lead's Cognitive Effects

As a preliminary test for a larger hypothesis, this study provides a promising avenue for addressing the gap in our knowledge of lead's effects on avian cognition. These results suggest that using this task on juvenile starlings in Flint would be useful for testing the prediction that exposure to lead impairs cognitive development. Testing this could involve training adult starlings (or other species if they demonstrate the ability to solve the puzzle) and measuring how quickly fledged young learn the task. The development of a working prototype provides a starting point for future field studies testing cognition.

Various developmental factors are associated with adult starling cognition, and some factors can decrease cognitive abilities such as reduced telomere attrition and natal brood size (Nettle et al. 2015). Since developmental factors are related to cognitive

performance, it is worthwhile to address developmental health when studying cognition in adult starlings and other bird species. Early development was one measure we were unable to address in this study. However, the implication that developmental factors play a role in cognition suggests that there could be cognitive consequences later in life if something goes wrong as juveniles learn habits like foraging. Starlings learned how to overcome the feeder puzzle at one of our control test sites that is low in lead exposure. Their success suggests that there was no major hindrance to development at this site, but additional tests on birds of known lead exposure are necessary to determine if this task can reveal developmental deficiencies caused by lead.

This project was developed with the broader goal of assessing the effects of lead on avian cognition. We predict that birds with higher blood-lead levels will either take more time to access food from inside the feeder, or will not be able to do so at all in comparison to their low blood-lead level conspecifics. The next steps of this research involve assessing how early development plays a role in avian cognition. Lead is detrimental to human cognition because it impairs important learning processes. Children that have experienced lead exposure exhibit decreased brain volume and learning deficits as they grow older, and this is partially due to the disruption of the NMDA receptor in the hippocampus (Karri et al. 2016). Songbirds also have this NMDA receptor, and it plays an important role in normal song development and memory. Studies in zebra finches (*Poephila guttata*) that block NMDA highlight behavioral impairments with song induction, which requires sensorimotor learning and sensory acquisition (Basham et al. 1996).

Adverse physiological effects of lead have been observed in birds, but it is not known whether lead causes learning deficits the way it does in humans. Neurons function similarly across many species, including invertebrates which are models for vertebrate neural functioning. Birds and humans also share similarities in complex learning processes, which has led to the use of birds as models for human learning and memory disorders (Brainard and Doupe 2013). It is therefore likely that lead affects avian cognition in similar ways to humans. Studying birds in Flint would be helpful to understanding the impacts that the water crisis has had on bird species, and this may also highlight how lead impacts vertebrates more broadly.

Summary and Conclusions

The overall findings of this project provide insight for future studies of avian cognition, especially in populations with poor developmental health due to factors such as lead exposure. Such studies are rare because cognition is difficult to address in the wild (Morand-Ferron et al. 2016), so a strength of our study was its ability to address this gap in the literature for studying cognition. Environmental metal contamination is common in otherwise “wild” areas and is the result of various human activities, including agriculture, mining, industrial endeavors, and medical applications (Karri et al. 2016). Toxic metals like lead can be concentrated at local pollution sites, but can also spread in the atmosphere and become widely distributed. In combination with its impacts on the kidneys, liver, and brain, metal toxicity can thus have adverse effects on essential functions (Karri et al. 2016) that may occur more widely than is currently appreciated.

Since the industrial revolution, human activity has had negative impacts on ecosystems and biodiversity, due to over-hunting, habitat destruction, the use of

pesticides and herbicides, and releasing toxic compounds into the environment (Hunter 2007). For this reason, the assessment of health in various locations can be observed through sentinel species residing in these areas. The study of these sentinel species can improve the efficiency at which ecosystem health is monitored (Tabor and Aguirre 2004), which is important for maintaining human health. Birds make ideal sentinel species because of their close association with humans in urban and rural areas. Blood-lead levels of pigeons in New York City neighborhoods, for example, closely predicted the blood-lead levels of children living in those neighborhoods (Cai and Calisi 2016). Conservation requires an understanding of how changes in the environment affect wildlife behavior (Greggor et al. 2014). Behavior is the result of cognitive processes (Morand-Ferron et al. 2016) and is also a major player in the ecological patterns of a system (Greggor et al. 2014). The study of animal cognition and how environmental toxins impact cognitive development is therefore of central importance to environmental conservation that benefits both wildlife and humans.

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