Girls just wanna have aggression: Female Zebra Finch mate competition aggression in response to a d1 dopamine receptor antagonist

Intro

Dopamine antagonists are a classification of drugs that bind to dopamine receptors but do not activate them to block dopamine from binding and having an effect on the body [13]. There are five dopamine receptors: D1, D2, D3, D4 and D5. D1 receptors are grouped with D5—both couple to G stimulatory sites [11]. Of the five types of dopamine receptors, D1 receptors are the most abundant throughout the central nervous system and are responsible for locomotion, learning and memory, attention, impulse control, sleep, and regulation of renal function [12].

The effect of D1 Dopamine receptor antagonists has been heavily researched in conjunction with male appetitive sexual behavior in a variety of avian species. In male Japanese quail studies have indicated that male copulatory behavior is stimulated by the action of dopamine through D1 receptors. Dopamine is a prominent motivator of sexual behavior in Japanese quail [2]. In male European starlings, studies have shown that a D1 dopamine receptor antagonist disrupted male song production, supporting the theory that dopamine is involved with sexually motivated behavior in European starlings [8]. Dopamine is a significant component in the production of sexually motivated song. In male zebra finches, the idea that during mate competition, dopamine is a facilitator of male sexual behavior [4]. Little research has been done on the relationship between dopamine and mate competition aggression in female zebra finches.

In the social decision-making network, the functionality of D1 dopamine receptors in the anterior hypothalamus of male zebra finches with high competitive ability is related to the competitive ability during mate competition [4].

It is well known that the opposite sex is a potent stimulus for aggression. An opposite sex stimulus often results in same-sex aggressive behaviors [1]. For non-paired individuals in pair bonding species, acquisition of a high-quality mate is imperative, especially when potential mates are scarce [1]. The presentation of a potential mate stimulus triggers both male and female zebra finches to behave aggressively towards their same sex in an attempt to attain a mate for themselves. Female zebra finches are just as likely as males to participate in aggressive behaviors when it comes to mate competition [1].

Mate competition is a form of intrasexual mate selection. Often, this manifests in aggressive encounters between individuals vying for the same mate, thus Sexual selection is often mediated by the combat between individuals [21]. Utilization of a mate competition paradigm is one of the best ways to investigate the relationship between dopamine treatment and aggression. In this, two male zebra finch were presented a female zebra finch stimulus. This prompts the two males to court the female and compete for her attention [20].

Dopamine has also been shown to be essential in the production of male sexual behaviors, a study on male rats showed that a dopamine agonist, Amphetamine, increased the number of ejaculations by 30%, and decreased ejaculation latency and post ejaculatory interval by 40% [14]. Also in male rats, when given a mixed D1/D2 antagonist, sexual motivation was significantly decreased in a place-preference test.

The understanding of both reproductive motivation and aggression in females is less comprehensive. Numerous studies looking at the relationship between dopamine and lordosis behaviors in female rats have resulted in conflicting outcomes [14]. Many studies concerning the relationship between dopamine and female sexual motivation are oversimplified or difficult to analyze due to the complexity of the female reproductive system [15]. Despite this, in recent years, more research has presented reaffirming results in of the categories of aggression and reproductive behavior in relation to dopamine.

In female laying hens, a dopamine D1 receptor antagonist decreased aggressive behaviors of hens who were already highly aggressive [18]. This supports the hypothesis that in female avian species, dopamine is a regulator of aggression.

In terms of dopamine and sexual motivation, the introduction of a dopamine agonist in female European starlings resulted in increased interest in devices that were playing the songs of male European starlings. This supports the hypothesis that in avian species, dopamine motivates sexual behaviors in females.

It stands to reason that female mate competition aggression is modulated by the neurotransmitter dopamine. Due to the gap in knowledge surrounding dopaminergic activity in female mate competition, we sought to lessen this chasm using a pharmacological manipulation of D1 dopamine receptor antagonists on female Zebra Finches. Using female zebra finches, we examined the effects of a D1 dopamine receptor antagonist on the occurrence of aggressive behaviors during a controlled mate competition setting. We believed that subjects with their dopamine receptors inhibited would exhibit fewer aggressive behaviors towards their competition.

Methods

Housing

Birds were housed in same-sex colony cages with at least one other cage mate. Females were housed in one of four cages. Males were housed in one of three cages. All birds were housed in the same room, with males and females isolated visually but not acoustically. All male and female birds had ad lib access to both food and water throughout the course of the study. They had a consistent light/dark cycle of 14:10.

Prescreening

Both male and female Zebra Finches were prescreened for an inclination toward sexually motivated behaviors and aggressive behaviors, respectively. To perform this prescreening, four non-acquaintance females were placed in one cage, and four males (familiarity between males was not a concern) were placed in a neutral, conjoined cage with an opaque partition. The opaque partition was removed, and males and females were allowed to view each other through a wire partition for five minutes. Female aggressive behaviors and male courtship behaviors were recorded during this time. Male courtship behaviors were defined as directed song, following of females, contact with wire partition, and mount attempts. After five minutes, the wire partition was removed, allowing male and female Zebra Finches to interact. Interaction lasted 15 minutes, in which behavioral observations continued to occur.

Prescreening trial ran a total of 20 minutes (5 viewing, 15 interacting). Sixteen females were prescreened for behaviors of interest and eight were chosen as subjects. Behaviors of interest were based off the papers Adkins-Regan, E., & Robinson, T. M. (1993). Aggressive female behaviors toward the stimulus female were identified as chases and the initiation of beak contact, as well as flees, avoiding, and calls. Additional behaviors that were recorded were the number of contacts with the wire partition during the first ten minutes of data collection as well as copulation solicitation displays. Eight males were screened and five were selected as stimulus males for the present study. Male sexually motivated behaviors were identified as directed song, undirected song, mount attempts, successful mounts, and contact with wire partition during the first ten minutes of data collection.

Pairings

Using a within-groups design, each subject female underwent two trials, one with a pharmacological manipulation, and one with a control injection. In each test, one subject female was paired with one stimulus female and one stimulus male. Because selected subjects exhibited desired aggressive behaviors, subject females were also used as stimulus females. Several guidelines were established to keep trials novel while using birds that exhibited the desired behaviors. Individual females that were housed together were not paired together as subject/stimulus. To mitigate exhaustion, subjects that participated in tests were not used as stimulus (or subject) animals that same week. To preserve novelty, testing pairs were only used once, and individuals only saw each stimulus male once as either a subject or stimulus, not both. To mitigate sexual exhaustion, stimulus males were only used once per week. All pairings were randomly assigned while staying within these guidelines. .

Pharmacological Treatment

As a pharmacological manipulation, SCH-23390, a known D1-receptor dopaminergic antagonist was injected in each female subject during one trial. To prepare the D1-receptor dopaminergic antagonist solution, 5 mg of solid SCH-23390 was dissolved into 150 mL of deionized water. The dosing of this drug was based off of Schroeder, M. B., & Riters, L. V. (2006) at 1 mg SCH-23390 /kg body weight. Our subjects had an average body weight of 15 g, meaning our dosage for each trial was 0.05 mL of our dissolved SCH-23390 solution, or 0.05 mL of our control, deionized water. The injection site was the inguinal leg fold. Using a double-blind technique, pharmacological and control treatment were randomly assigned a variable that was assigned by a third party to mitigate observer bias.

Trial Procedure

Subject female was injected with 0.05 mL of either the treatment or the control solutions. The pharmacological treatment takes 15 minutes to take effect, thus subjects were placed in the trial cage for a 15-minute acclimation period, allowing the treatment to take effect. During the acclimation period, the stimulus male was placed in the conjoining cage with an opaque partition in between the two cages. Neither animal could see nor interact with the other. After 15 minutes, the stimulus female was added into the cage with the subject female and the opaque partition was removed. A wire partition remained, allowing the females to see the male and vice versa. Behavior with the wire partition was observed for 10 minutes. At the 10-minute mark, the wire partition was removed, and all three birds were able to interact. Behavior was observed and recorded throughout the 10 minutes that the birds were allowed to interact. The

total trial length (acclimation, viewing, interacting) was 35 minutes with 20 minutes of data collection.

Results

Aggression

When the number of initiated beak contacts was used as the primary aggressive behavior, we found that there was a visible difference between the number of initiated beak contacts and the treatment given. When the subject was given the D1 dopamine antagonist, they initiated fewer instances of beak contact than subjects given the control. When they were used as stimulus animals, the number of beak contacts that they initiated was not visibly different from the control subject.

Sexual motivation

When the number of partition contacts was used as a measure of sexual motivation, there was little visible difference between when subjects were given the D1 dopamine antagonist and the control treatment. This means that the male was a strong enough stimulus to trigger sexual motivation.

Movement

When the number of partition contacts was used as a measure of locomotion, there was little visible difference between when subjects were given the D1 dopamine antagonist and the control treatment. This means that the D1 dopamine antagonist was likely not inhibiting locomotion.

Stimulus Data

We can use data from each participant's stimulus behavior because of all of the guidelines set in place with our subjects and the study design as a within-subjects experiment. Subjects were 1) not from the same home cage as our subject; 2) the subject was novel to the stimulus individual; 3) the male was novel to the stimulus.

Discussion

Although males were prescreened for an affinity towards directed song and courtship behaviors, it is important to note that all of the subject participants came from the same breeding colony of 800 birds. This could mean that the males are too closely related to the females be a strong enough sexual stimulus for the female participants.

The usage of a dopamine antagonist such as SCH-23390 could have cause an impairment of locomotion. Dopamine antagonists are often given to birds as a therapeutic treatment for behaviors including auto-mutilating for their behavioral quieting effects [10]. Research shows that on a high dose of this D1 dopamine receptor antagonist, SCH-23390 acted as a sedative in male titi monkeys [9]. In this same vein, sex differences in bodyweight could have influenced the dosing of our subjects. Previous studies of dopamine antagonists using male subjects had a dosage dependent on body weight [2][8]. We implemented a similar procedure, basing our dosage on the average weight of a zebra finch. Since we observed that the male zebra finches tended to be larger and heavier than female zebra finches, this dose could have been too high, resulting in extreme effects of the drug and the observed mate competition aggression is not due to the D1 dopamine receptor antagonist, but due to general inhibition of ambulatory behaviors by the treatment.

Female aggressive behaviors may have been skewed due to body size and condition. When one female bird is larger than the other, the larger of the two is more likely to initiate aggression [5]. Additionally, when females are of similar body size and body condition, this will prolong bouts of aggressive behaviors [5]. We did not factor body size into account when pairing female subjects together. This could have caused an unknown confound, producing skewed results. In the future, to reduce the possibility of this occurring, pairing individuals of similar size would eliminate size bias among the subject and stimulus individuals.

Using a peripheral injection site, such as the inguinal fold could have had an impact on how the treatment diffused throughout the body. Drugs with a molecular weight of 400 or less are most likely to cross the blood brain barrier, our drug SCH 23390 has a molecular weight of 287.78 [16][17]. This means that the drug has a strong chance of making it past the barrier. Despite this, 98% of small molecules do not make it past the blood brain barrier [16]. We cannot ensure that the drug effectively penetrated the blood brain barrier and that receptors that were affected were in the areas of interest.

Our male stimulus individuals introduced a confounding variable. We could not ensure that they were sufficiently motivated to perform sexually motivated actions that would engage the females. Limiting data collection of female aggressive behaviors during concurrent male sexual behaviors could mitigate this.

Future research regarding mate competition aggression in female zebra finches could be strengthened in several ways. One of which is studying how female zebra finch's behavior changes in response to various levels of dosage. This would be beneficial to the body of work because we would be better able to discern whether mate competition aggression is truly reduced by the administration of a D1 dopamine receptor antagonist, or if the treatment is causing an overall sedative response.

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