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Visual recognition of individual conspecific males by female zebra finches, *Taeniopygia guttata*

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The aim of this study was to determine whether visual recognition of conspecifics exists in zebra finches, and if so, whether the strength of this recognition is a function of the social relationships among flock members. To achieve this aim, we trained adult female zebra finches to indicate their preference by pecking on one of two photographs presented on a digital screen. We found that female zebra finches possessed a very good ability to recognize their mates, and a slightly lower but still good ability to recognize other, socially closely related males that were members of their own small flock. Moreover, we found that the individual integrity of the small flocks continued to be maintained, as subgroups, even after these were combined to form one large flock, indicating that zebra finches demonstrate different levels of social relationships between flock members, and that these relationships are long-lasting and stable. However, when part of a larger flock, the females were unable to recognize males that were socially distant to them.

Conspecific individual recognition incorporates a subset of recognition that occurs when one organism identifies another of its own species according to its individually distinctive characteristics (Tibbetts & Dale, 2007). This type of recognition is found in various species in the animal kingdom, is displayed in various ways, and can be based on one or more senses. Emperor penguins, *Aptenodytes forsteri*, for instance, use the two–voice system to recognize one another (Aubin, Jouventin, & Hildebrand, 2000), while wasps and crayfish use visual facial characteristics to make this kind of distinction (Tibbetts, 2002; Van der Velden, Zheng, Patullo, & Macmillan, 2008). In regard to social behaviour, individual recognition is important in many aspects, such as keeping stable groups, recognition of mates and offspring, and in competitive relationships (Hurst et al., 2001).

Zebra finches (Estrildidae) are social, monogamous songbirds. They form various flock sizes, according to seasonal changes over the year: up to 10 members in a flock during winter and 10–20 during spring, while at the beginning of the summer several small flocks join together to form larger flocks of up to 300 birds. When part of a large flock, the finches can also be recognized as subgroups, whose members maintain physical proximity, especially when foraging or perching (Zann, 1996). Zebra finches display a distinct sexual dimorphism: males have distinct external characteristics, especially on the face and chest, and only the males sing (Zann, 1996).

A vocal conspecific recognition signal has been demonstrated to occur among zebra finches: males that were played 'sound supplements' (songs and calls) generated by their own colony sang more in reply than males that heard sounds generated from a different colony (Wass, Colgan, & Boag, 2005). Female zebra finches preferentially respond to the song of their own mate rather than to the song of a neighbouring conspecific male (Miller, 1979).

Regarding visual ability, it was found that female zebra finches prefer to associate with males with the reddest, brightest bills (Burley & Coopersmith, 1987), a more symmetrical chest plumage (Swaddle & Cuthill, 1994) and larger orange cheek patches (Neguib & Nemitz, 2007). It was also found that both male and female zebra finches can differentiate between zebra finches and Bengalese finches, *Lonchura striata* var. *domestica*, using both visual and acoustic traits (Campbell & Hauber, 2009a; Campbell, Shaw, & Hauber, 2009). Moreover, female zebra finches can recognize conspecifics by using a combination of visual and acoustic cues (Brazas

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& Shimizu, 2002; Campbell & Hauber, 2009b; Galoch & Bischof, 2006). The effect of visual and auditory cues in conspecific discrimination learning has also been tested in Bengalese finches (Watanabe & Jian, 1993). However, to the best of our knowledge, in zebra finches, there has been no study examining the level of conspecific recognition based only on visual information, while also correlating this recognition with their social structure and the degree of kinship between individuals.

Since in the wild the number of members in a flock of zebra finches changes significantly throughout the year, it can be assumed that different levels of recognition exist between the birds due to the different levels of relationships: a monogamous relationship between a male and a female, a relationship between flock members living together as a small group and a relationship between flock members living together as a large group. Accordingly, in this study we asked three questions. (1) Do female zebra finches recognize their mates? (2) Do female zebra finches recognize different males from their own original small flock (up to 12 individuals)? (3) Do zebra finches recognize different males from their own original flock when living in a large flock (of 30 individuals)?

METHODS

Ethical Note

The experiments were approved by the Tel-Aviv University Animal Care and Use Committee (Permit L-12-030) and carried out in accordance with its guidelines and regulations regarding the care and use of animals for experimental procedures.

General

To determine whether, based only on visual cues, individual recognition of males by female zebra finches exists, and to examine its possible different levels, we conducted three experiments. In each experiment, the birds had to learn to peck on one of the two photographs that were presented on a digital screen, in order to indicate their preference (see Visual Stimuli for each experiment). This behaviour is not spontaneous or easy for birds to learn. We therefore designed a gradual training process to enable the birds to accomplish this task successfully. Accordingly, the general protocol for all experiments consisted of three stages, each with several steps. For a schematic illustration of this protocol, see Fig. 1. Below, we give a detailed description of the specific protocol for each experiment.

Experiment 1: Mate Recognition

Subjects

Twenty-four adult zebra finches were housed in two aviaries (six males and six females in each). The aviaries (1.5 × 1.5 m and 2 m high) were located outdoors, in the Meir Segals Zoological Garden at Tel-Aviv University, exposed to ambient conditions. The birds were banded with coloured plastic rings for individual identification and provided with nesting materials. Only eight of the 12 females completed a breeding cycle with a male from their aviary and therefore participated in this experiment. The females were weighed once a week throughout the experiment and this was used as an indirect measure of their general health. Food (millet grains) and water were provided ad libitum until the beginning of the experiment. The birds were then given no access to food from 1600 hours until the beginning of testing at 0900 hours on the following day, for 5 consecutive days a week during the test period (see below).

Test cage

Tests were conducted in a cage (14 × 40 cm and 23 cm high), divided in half by an 8″ digital screen (809SL, Silver Line), which had a black card cover and faced the front part of the cage (Fig. 2). The back part of the cage contained a bowl with millet grains. The screen could be raised or lowered, in order to allow or prevent the tested female from moving from one part of the cage to the other. To the rear of the cage was a wooden diaphragm, which was used to gently push the tested female back into the front part of the cage at the conclusion of each test. A video camera (LifeCam VX3000, Microsoft) was located above the cage to record the tests. The same cage was used for all three experiments and it was placed in a room with fluorescent illumination and a temperature range of 23–25° C.

Visual stimuli

The visual stimuli comprised photographs of the mate of the tested female, plus those of a male that was a stranger to the female. The head and neck of each male was photographed in three different postures (Fig. 3), to be used in different stages of the experiments. All photographs were taken with a black background, from the same distance and with the same camera (Nikon D90, with AF-S Nikkor 18–135 mm lens, and 67 mm diameter). The photographs were edited using Photoshop software in order to show the two males being presented to the female side by side (Fig. 4), with colours and clarity remaining untouched.

Duration (days)	3	Up to 30	up to 30	Up to 30	Up to 30	Up to 30	Up to 30	1
		Training to peck on …..						
Aim	Acclimation	Grains attached to the screen	Inaccessible grains attached to the screen	An inaccessible grain attached to the target photograph	Shell of a grain attached to the target photograph	Transparent tape attached to the target photograph	Target photograph	Test
Step		2.1	2.2	2.3	2.4	2.5		
Stage	1			2			3	

Figure 1. A schematic illustration of the general protocol for all experiments. See text for more details.