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Social interactions in adolescent and adult Sprague–Dawley rats: Impact of social deprivation and test context familiarity

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Abstract

Interactions with peers become particularly important during adolescence, and age differences in social interactions have been successfully modeled in rats. To determine the impact of social deprivation on social interactions under anxiogenic (unfamiliar) or non-anxiogenic (familiar) test circumstances during ontogeny, the present study used a modified social interaction test to assess the effects of 5 days of social isolation or group housing on different components of social behavior in early [postnatal day (P) 28], mid (P35), or late (P42) adolescent and adult (P70) male and female Sprague–Dawley rats. As expected, testing in an unfamiliar environment suppressed social interactions regardless of age, housing, and sex. Social deprivation drastically enhanced all forms of social behavior in P28 animals regardless of test situation, whereas depriving older animals of social interactions had more modest effects and was restricted predominantly to play fighting—an adolescent-characteristic form of social interactions. Social investigation—more adult-typical form of social behavior was relatively resistant to isolation-induced enhancement and was elevated in early adolescent isolates only. These findings confirm that different forms of social behavior are differentially sensitive to social deprivation across ontogeny.

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Keywords: Social behavior; Social interaction test; Social deprivation; Adolescence

1. Introduction

Interactions with peers during adolescence are thought to be of principal importance for social development in human adolescents, with individuals spending more time interacting with peers during adolescence than at any other developmental period [10,16]. Similarly, adolescent rats demonstrate higher levels of social behavior than younger and older animals, with these interactions being essential for developing the ability to express and understand intraspecific communication signals [11,17,30,34,35]. Interactions with peers provide a significant source of positive experiences for human adolescents [15,16] and, even in a simple model of adolescence in the rat, have been shown to be more rewarding for adolescents than for their more mature counterparts [4].

Social behavior of adolescent and adult rats is sensitive to environmental factors, with, for example, social interactions being notably lower when animals are placed together in an unfamiliar relative to a familiar testing chamber [7,27,37]. An unfamiliar testing situation is traditionally viewed as an anxiogenic condition, and the decrease of social interactions under this condition has been used as an animal model of anxiety [5,6,7,8]. Such environment-induced social inhibition has been reported to differ as a function of age and sex, with young adolescent animals and adult females less affected by an unfamiliar test situation than older adolescents and adult males. For instance, male rats tested at postnatal day (P) 35 and P60 have been reported to demonstrate a reliable decrease in time spent in social interactions under unfamiliar test circumstances, whereas P28 males showed no suppression of social interactions under these test conditions [27]. In contrast, other researchers observed environment-dependent changes in social behavior as early as P21 when play fighting was assessed separately from other types of social behavior [36]. In that study, play fighting, but not social investigation and contact behavior, was suppressed by an unfamiliar environment in P21 male rats. According to Johnston and

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File [14], adult female rats, in contrast to their male counterparts and adolescent females [9], do not differentiate between familiar and unfamiliar environments in terms of time spent in social interactions.

These previous experiments, however, assessed age- and sexrelated differences in responsiveness to familiarity of the test situation in animals that had been deprived of social contact by housing in isolation for 5-7 days prior to the social interaction test. Most studies also used only an overall measure of social behavior (i.e., overall time spent in social interactions), even though the elementary behavioral acts summed together for these assessments (e.g., sniffing, grooming, chasing, following, crawling under or over, nape attacks, boxing, and wrestling) reflect separable and differentially regulated forms of interactive social behaviors that include social investigation, contact behavior, and play fighting. Each of these forms of social activity has a distinct ethological and ontogenetic pattern [17,20,32,35,39] and is affected differentially by drug treatments [1,29,35]. For instance, the ontogeny of play fighting demonstrates an inverted U-shaped pattern, with a peak being observed around P30-35 [18,19,20,35,39], whereas social investigation is more pronounced in adult than adolescent animals [37]. Taken together, these findings suggest that different forms of social behavior may be mediated by different neural systems, and, hence, may be differentially sensitive across ontogeny to variations in familiarity of the test situation and the amount of social deprivation prior to testing.

Therefore, the main objective of the present study was to determine the impact of social deprivation on social investigation, contact behavior, and play fighting of male and female Sprague-Dawley rats under non-anxiogenic (familiar) or anxiogenic (unfamiliar) test circumstances during ontogeny. The study used a modified social interaction test [39,40] to assess the effects of 5 days of social deprivation or group housing on different components of social behavior and motivation for social contacts in adolescent and adult rats. Given previous reports that environment-dependent changes in social behavior emerge within the adolescent period [27], P28 (early), P35 (mid), and P42 (late) adolescent rats were tested in the study. This work is important, since assessment of social interactions in an anxiogenic situation has been intensively used as an animal model for evaluation of potential anxiolytic drugs in adult animals, whereas information is limited as to the appropriateness of this testing situation for such assessments among adolescent animals (see [8] for references and review).

2. Methods

2.1. Subjects

Sprague–Dawley rats bred and reared in our colony at Binghamton University were used in these experiments. A total of 384 rats derived from 48 litters were used as experimental subjects (n = 192, 96 males and 96 females) and partners (n = 192, 96 males and 96 females). All animals were housed in a temperature-controlled (22 °C) vivarium maintained on a 14-/10-h light/dark cycle (lights on at 07:00 h) with ad libitum access to food (Purina Rat Chow, Lowell, MA) and water. Pups were housed until weaning with their mothers in standard maternity cages with pine shavings as bedding material. Litters were culled to 10 (5 males and 5 females) pups on P1, the day after birth. Rats were

weaned on P21 and placed into standard plastic cages with same-sex littermates (five animals in a cage) until the onset of the differential housing manipulations for this study. In all respects, maintenance and treatment of the animals were in accord with guidelines for animal care established by the National Institutes of Health, using protocols approved by the Binghamton University Institutional Animal Care and Use Committee.

2.2. Experimental design

The design of the study was a 4 (age) \times 2 (housing) \times 2 (test situation) \times 2 (sex) factorial, with 6 animals being placed into each of the 32 experimental groups defined by this factorial design. Male and female animals were tested either on P28 (early adolescents), P35 (mid adolescents), P42 (late adolescents), or P70 (young adults). Experimental subjects were housed either in groups of three same sex littermates (group housing) or individually (isolate housing) in standard plastic cages for 5 days prior to the social interaction test. One day before testing (P27, P34, or P41 for adolescents and P69 for adults), experimental animals in the familiar test group were placed into a testing chamber for 30 min to make the testing situation familiar for them, whereas the other experimental animals were placed individually into a holding cage for the same amount of time (unfamiliar test group). To avoid the possible confounding of litter with housing and test situation effects [13], animals were assigned semi-randomly to the experimental groups, with the constraint that no more than one subject of a given sex from a given litter was assigned to a particular housing and test condition. Given that females of different age groups were included in this study, vaginal smears were not used to determine estrus cycle in the adult females.

2.3. Social interaction test

The test apparatuses consisted of Plexiglas (Binghamton Plate Glass, Binghamton, NY) chambers containing clean pine shavings that were proportionally size adjusted to provide comparable social proximity for adolescent and adult animals ($30 \text{ cm} \times 20 \text{ cm} \times 20 \text{ cm}$ for adolescents and $45 \text{ cm} \times 30 \text{ cm} \times 20 \text{ cm}$ for adolescents and $45 \text{ cm} \times 30 \text{ cm} \times 20 \text{ cm}$ for adults). Each test apparatus was divided along the long axis into two equally sized compartments by a clear Plexiglas partition that contained an aperture ($7 \text{ cm} \times 5 \text{ cm}$ for adolescents and $9 \text{ cm} \times 7 \text{ cm}$ for adults) to allow movement of the animals between compartments in a way that only one animal was able to move through the aperture at a time [39,40].

All testing procedures were conducted between 9:00 and 13:00 h under dim light (15-201x). Immediately prior to testing, each experimental subject was marked by a vertical line on the back and placed alone in a holding cage for 30 min. This pre-test social deprivation in a novel environment is a standard procedure that has been extensively used to increase baseline levels of social behavior from which inhibitory effects of different experimental manipulations, including anxiogenic compounds, may be more readily detected [6]. Each animal was then placed into the testing chamber simultaneously with a same age and sex test partner. Given that social behavior [22,23,39], social motivation [39] and rewarding properties of social interactions [4] can be dramatically modified by housing conditions of the partner, partners were always non-manipulated animals that had not been socially isolated prior to testing and who were unfamiliar with both the test apparatus and the experimental animal with which they were paired for testing. Weight differences between test subjects and their partners were minimized as much as possible, with this weight difference not exceeding 5 g for each pair of animals at P28, 10 g at P35, 15 g at P42, and 20 g at P70, and test subjects always being heavier than their partners. During the 10-min test session, the behavior of the animals was recorded by a video camera (Panasonic model AF-X8, Secaucus, NJ), with real time being directly recorded onto the videotape for later scoring (Easy Reader II Recorder; Telcom Research TCG 550, Burlington, Ontario). After each test, the apparatus was wiped with 3% peroxide hydrochloride and the shavings were replaced with fresh ones.

2.4. Behavioral measures

The frequency of a number of social activities [17,32,35,39] of each test subject was analyzed from the video recordings. Social investigation was defined as the sniffing of any part of the body of the partner. Frequency of contact behavior was scored as the sum of crawling over and under the partner and social

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