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Exposure to Novelty Weakens Conditioned Fear in Long-Evans Rats

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The present study sought to determine whether post-training exposure to a novel or familiar object, encountered in either the location of the original fear conditioning (black compartment of a passive avoidance {PA} chamber) or in a neutral setting (open field where initial object training had occurred) would prove capable of reducing fear at subsequent test in a passive avoidance task. In Experiment 1, Long-Evans rats that encountered a novel object in either the black PA compartment or the open field, as well as those encountering a familiar object located in the black PA compartment all displayed weaker fear at test than did those subjects that encountered a familiar object in the open field. These effects were explained in terms of a counter-conditioning of fear resulting from the appetitive aspects of novelty exposure. Experiment 2 compared the fear-reducing capabilities of novel object exposure to a more simple extinction procedure. While both the extinction and novelty groups generally showed reduced fear compared to control animals, some evidence suggested that novel object exposure resulted in significantly less fear at test than did extinction alone.

It has long been known that conditioning processes could play some role in the acquisition of fear and phobias (Davey, 1992; but see Seligman, 1971). Watson and Rayner (1920) first demonstrated that presenting a fearevoking stimulus (e.g., a loud noise) simultaneously with a neutral stimulus (e.g., a white rat) would result in a subsequent fear of the neutral stimulus. Likewise, many treatments for conditioned fear and phobias involve various learning phenomena (for review see Miltenberger, 2004). In counter-

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conditioning, for example, a new positive (i.e., appetitive) stimulus is paired with a stimulus that had previously evoked a fear response (e.g., Wilson, 1973). For example, if one location is paired with a mild foot-shock, rats will learn to fear that location. If a positive stimulus (e.g., food) is subsequently presented in that same location, rats will learn the new positive association between that location and food, and will thus display a reduced fear response toward that space (Klein, 1969).

In the present study we attempted to examine the ability of novel objects to counter-condition a previously learned fear in a passive avoidance task (cf. Millin, 2006) in rats. Rats typically prefer exploring novel objects over familiar ones (Berlyne, 1950), and thus novel objects are generally considered appetitive in nature (e.g., Anderson, 2006ab). Indeed, novelty can be rewarding, and exposure to novel objects can instill a place preference for the location in which the novel object was encountered (Bevins, Besheer, Palmatier, Pickett, & Eurek, 2002). In the present study it was hypothesized that experiencing novel objects in the black compartment of a typical two-compartment (white/black) passive avoidance chamber that was previously paired with foot-shock would result in reduced fear of that compartment at subsequent test, as evidenced by weaker avoidance tendencies. Given that counter-conditioning can be employed in behavioral modification therapies with human patients (e.g., Croghan & Musante, 1975; de Jong, Vorage, & van den Hout, 2000), a complete understanding of its workings and potential optimization are desirable. Moreover, given the widespread use of novel object recognition procedures in learning and memory research (e.g., Ennaceur & Delacour, 1988; Anderson, Jablonski, & Klimas, 2008; Anderson, Karash, Ashton, & Riccio, 2003; for review see Ennaceur, 2010; Anderson, 2006ab), a thorough understanding of the effects of exposure to novelty on behavior is desirable.

EXPERIMENT 1

In the first experiment, rats were trained in a passive avoidance task, encountered either a novel or familiar object in either the black compartment of the passive avoidance (PA) chamber that had been previously paired with mild foot-shock or in an octagon-shaped open field where exposure to initial objects had previously occurred, and were subsequently tested for fear of the black PA compartment. The purpose of this experiment was to determine whether the type of object present (novel or familiar) and the location of the object exposure session (black PA chamber compartment or open field) would influence the degree of fear displayed by the subjects at test.

METHOD

Subjects. Subjects consisted of forty-eight adult male Long-Evans rats ranging in age (M=253.5 days, SD=72.35), which were obtained from the in-house breeding stock at Saint Joseph's University (originally derived from breeding pairs obtained from Taconic Farms Inc., Germantown, NY). Given the range of ages employed in this study, efforts were made to approximately match the four experimental groups (see procedure section below) based on age. Animals were provided *ad lib* food and water, and were housed in standard plastic shoebox cages in a room with a reversed 14/10-h light/dark schedule (all procedures occurred during the dark portion of their lighting schedule). All procedures received the approval of the Saint Joseph's University Institutional Animal Care and Use Committee (IACUC) prior to the start of the study.

Materials. Initial object exposures were conducted in a sealed, wooden, octagon-shaped open field with approximately 40cm sides and 34cm tall walls, subdivided into four zones. Small holes (approximately 1cm) were located at the center of each of the four zones in the open field, allowing for the attachment of objects (see below). The open field apparatus was located in a room containing indirect overhead fluorescent lighting.

The passive avoidance apparatus was set up on a table located in a room illuminated with standard overhead fluorescent lighting and situated immediately adjacent to the room containing the open field apparatus. Passive avoidance occurred in a typical two-compartment (white/black), Plexiglas chamber, approximately 54cm in length, 20cm in width, and 20cm One compartment of the passive avoidance apparatus in height. (approximately 26cm (L) X 20cm (W) X 20cm (H)) had a white ceiling and white walls, while the other had black walls and a black ceiling (and The two compartments were connected via a identical dimensions). guillotine door that could be opened and closed as the experiment required. The passive avoidance chamber sat atop a stainless steel grided floor with rods that were approximately 2mm in diameter and spaced approximately 1cm apart, in order to allow for the delivery of non-scrambled .5mA footshocks generated by a Harvard Instruments (Harvard, MA) shock generator (model: H.I. 3121).

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Several sets of small, plastic, easily cleaned objects served as novel and familiar objects in the present study. These included a yellow rubber duck and a clear, 120ml plastic baby bottle with a plastic cap. Each object was glued to an approximately 10cm X 10cm long, .6cm thick black Plexiglas square block that could be secured tightly to each apparatus with a screw, nut, and metal washer. Two identical versions of each object were employed so that one could be used at training, and the other during counter-conditioning.

Procedure. The procedure for Experiment 1 is depicted in Figure 1. Rats were obtained and underwent 5 days of handling and pre-exposure to the apparatus in which they were to subsequently encounter objects. Following an approximate 5-minute handling session, rats were placed in the empty octagonal-shaped open field for 5 minutes. This process was repeated across 5 consecutive days and was designed to pre-expose and habituate the animals to the octagon itself, ensuring that they would spend a sufficient amount of time exploring the objects that would be presented in that arena on subsequent stages of the experiment (cf. Besheer, & Bevins, 2000; Wilkinson, Herrman, Palmatier, & Bevins 2006). Following each pre-exposure trial, feces and urine were removed and the arena was cleaned with soapy water and dried. After the first 5 days of the procedure, 2 days of rest followed in which no procedures took place.

Following the 2-day rest period, rats underwent initial object training in which they became familiar with one particular object. Each subject was individually placed in the octagon open field for 5-min on three consecutive days at approximately the same time each day, mid-afternoon. During each of the three initial object exposure sessions, each rat repeatedly encountered a single object (duck or bottle) located in one of four possible locations within the apparatus. While the encountered object was consistent from day to day, the location of the object varied daily, with the object appearing in one of four possible locations within the arena. Objects never appeared in the same location twice over the course of training for any subject, and the overall appearance of objects in the various locations during training was counterbalanced across all subjects. The object encountered (duck or bottle) during these object familiarization training sessions was also counterbalanced across subjects. Feces and urine were removed, and the object and arena were cleaned with soapy water after each trial in order to dilute any potential scent trail that the rat may have left behind.



Figure 1. Flowchart depicting the procedure of Experiment 1.

One hour following the third object exposure, rats underwent training in a passive avoidance (PA) task. Each subject was placed in the white compartment of a two-compartment (white/black) passive avoidance apparatus. After a brief period on the white (safe) side of the box (15-sec), the guillotine door separating the two rooms was opened, allowing the rat to cross into the black compartment. Once it had crossed, the guillotine door closed, confining the animal to the black compartment. Five seconds later, it received the first of two brief (1-sec each), inescapable, 0.5-mA footshocks separated by an interval of 5-sec. Five seconds following the second foot-shock, the subject was removed from the apparatus and was returned to its home cage. Each subject's latency (in sec) to cross into the black compartment at training was recorded.

Approximately 24 hours following training, animals underwent one of four object exposure procedures (n=12; N=48). Rats either encountered the object that they had previously experienced during initial object training (Familiar Object) or an object with which they had no previous experience (Novel Object) for a period of 5-min, with the encounter occurring in either the black compartment of the passive avoidance chamber (Black PA) (i.e., counter-conditioning groups) or in the octagonal open field where the original object training had occurred (Octagon) (i.e., control groups). This

procedure resulted in four groups of subjects: Familiar Object/Black PA, Novel Object/Black PA, Familiar Object/Octagon, and Novel Object/Octagon. Those rats encountering objects in the black compartment of the passive avoidance chamber were designed to permit assessment of the effectiveness of novel and familiar object exposures to countercondition the previously learned fear. Those rats encountering objects in the octagonal open field were intended to serve as comparison groups, allowing us to control for the effects of handling and simple object exposure on subsequent fear.

Twenty-four hours following the object exposure session, all rats were returned to the passive avoidance apparatus for the assessment of their fear response. Testing involved returning each rat to the PA chamber for a period of 5-min and allowing it to choose between the fearful and nonfearful cues (i.e., one of two compartments had never been associated with shock and should be considered "safe"). Rats were placed in the white compartment of the apparatus and the guillotine door separating the white and black compartments was immediately opened. Both latency to cross into the black compartment (in sec) and total time spent in the white (safe) compartment (in sec) over the course of the 5-min test session were recorded and served as indicators of fear.

Statistical Analyses. 2x2 (Object Type {Novel/Familiar} X Encounter Location {Black PA Compartment/Octagon Open Field}) Between-Subjects ANOVAs were conducted on each of the three dependent measures: latency to cross at training, latency to cross at test, and total time spent in the white (safe) compartment at test. Statistical outliers greater than 2 *SD* away from their group's mean on the measure in question were excluded prior to running each ANOVA (i.e., outliers were only excluded from the analysis on which they differed from their fellow group members and were included in remaining analyses). All statistical analyses were performed via SPSS 16.0 for Mac, Release 16.0.2, and Levene's test was employed (following application of exclusion criterion) to examine homogeneity of variances on each measure.

RESULTS

Table 1 contains the means (and standard deviations) pertaining to the cross latency at training, cross latency at test, and total time in the white compartment at test (all in sec) of the various groups following the exclusion of statistical outliers for each measure. In regards to cross latency at training, four outliers (1 from each group) were excluded from analysis

according to the exclusion criterion outlined above. Levene's test for equality of error variances obtained evidence of heterogeneity of variances (F(3, 40)= 3.665, p=0.020), but as the groups were equal and reasonably large (n=11, following exclusion criteria), we proceeded with the ANOVA. The 2x2 ANOVA examining training cross latency failed to obtain evidence of main effects of object encounter location (Black PA Compartment/Open Field) (F(1, 40)=0.293, p=0.592) or encountered object (Novel/Familiar) (F(1, 40)=3.44, p=0.071), or for the interaction (Location X Object) (F(1,40)=0.170, p=0.682). Given the evidence of heterogeneity of variances, a series of pairwise comparisons with Mann-Whitney tests were also performed on the training cross latency data. Again, none of these tests yielded statistically significant results (p>.05).

 Table 1. Means and Standard Deviations on Each Dependent Measure
 (in sec) in Experiment 1 (following the exclusion of statistical outliers)

Group	Training Cross Latency	Testing Cross Latency	Total Time in White Compartment
Object/Location	M (SD)	M (SD)	M (SD)
Fam. Obj./Black PA	17.00 (15.74)	144.83 (128.98)	184.67 (101.22)
Novel Obj./Black PA	9.00 (7.89)	125.08 (106.07)	199.58 (93.05)
Fam. Obj./Octagon	13.63 (13.54)	207.17 (117.43)	274.64 (38.23)
Novel Obj./Octagon	8.54 (7.38)	95.17 (124.86)	174.83 (92.37)

On the measure of cross latency at test, no statistical outliers were removed according to the exclusion criterion, and Levene's test of equality of error variances failed to obtain evidence of heterogeneity of variances (F(3, 44)=.684, p=0.567). Again, no significant main effects were obtained in terms of encounter location (F(1, 44)=0.220, p=0.641) or encountered object (F(1, 44)=3.637, p=0.063), or for the interaction (Location X Object) F(1,44)=1.783, p=0.189). It is worth noting, however, that the main effect for encountered object approached significance (p=0.063), with groups encountering novel objects (M=110.125, SD=114.328) in between PA training and test sessions generally displaying less fear, as evidenced by shorter latencies at test, than those encountering familiar objects (M=176.000, SD=124.763).

One statistical outlier was removed from the Familiar Obj./Octagon Open Field group on the basis of the exclusion criterion outlined above prior to the analysis of total time in the white (safe) compartment. Levene's test on the total time in white (safe) measure suggested evidence of heterogeneity of variance (F(3, 43)=7.108, p=0.001). While the $2x^2$ ANOVA on the total time (sec) in the white compartment measure failed to obtain evidence of significant main effects for encounter location (F(1, 43)=1.696, p=0.200) or encountered object (F(1, 43)=2.872, p=0.097), a significant interaction effect was observed (Location X Object) (F(1, 43)=5.246, p=0.027). The 4 groups were reanalyzed in a One-Way ANOVA (*F*(3, 43)=3.143, *p*=0.035; Welch *F* (3, 22.169)=6.493, *p*=0.003) in order to allow for the conduction of post-hoc analyses. Least Significant Difference (LSD) post-hoc analyses revealed that the group that was exposed to the familiar object in the octagon open field displayed significantly greater fear (as measured by total time (sec) in the white compartment) than those encountering a familiar object in the black PA compartment (p=0.016), those encountering a novel object in the black PA compartment (p=0.042), or those encountering a novel object in the octagon open field (p=0.008). No other significant differences were observed according to the LSD analyses (p>.05). As evidence of heterogeneity of variance was obtained on the total time white measure, perhaps due to a ceiling effect in the group encountering the familiar object in the octagon, Games-Howell post-hoc analyses were also conducted on the total time in white (safe) measure. These analyses proved largely consistent with the LSD analyses, and again suggested that the group encountering the familiar object in the octagon was at least marginally significantly different from the group encountering the familiar object in the black PA compartment (p=0.053), the group encountering the novel object in the black PA compartment (p=0.089), and the group encountering the novel object in the octagon open field (p=0.017). No other significant differences were observed according to the Games-Howell analyses (p>.05). These effects can be seen in Figure 2.

DISCUSSION

This experiment examined the abilities of novel and familiar objects encountered in either the location in which fear conditioning had occurred or in a familiar open field twenty-four hours following training in a passive avoidance task to result in less displayed fear at subsequent test. Given the appetitive aspects of novel objects (cf., Berlyne, 1950; Bevins, Besheer, Palmatier, Pickett, & Eurek, 2002; Anderson, 2006ab), it was hypothesized that experiencing novel objects in the black compartment of the PA apparatus that had been previously associated with foot-shock would result in some degree of counter-conditioning and a reduced fear at test. The findings of this experiment generally supported this hypothesis, but they also seem to suggest that any kind of novelty exposure in between PA training and test is capable of decreasing the subsequently displayed fear response, no matter where the exposure occurred (black PA compartment or octagon open field).



Figure 2. Total time spent in the white (safe) PA compartment at test of the various groups during Experiment 1. Error bars represent SEM. Note: All non-depicted post-hoc comparisons did not achieve statistical significance (p>.05).

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When examining latency to cross over to the black compartment at test, a marginally significant (p=0.063) main effect of encountered object type was observed, with those experiencing novel objects in between training and testing displaying shorter latencies to enter the black compartment than those that had experienced familiar objects, irrespective of where the counter-conditioning object encounter had occurred (black PA compartment or octagon open field). Thus, the appetitive aspects of novelty exposure (cf., Berlyne, 1950; Anderson, 2006ab) would appear capable of generally reducing subsequent fear responses.

Moreover, when examining the total time spent in the white (safe) compartment at test, those rats encountering the novel object in the octagonal open field and those experiencing either the novel or familiar object in the black compartment of the PA chamber all displayed less fear at test than did those animals that encountered the familiar object in the octagon open field. While the counter-conditioning effects of novel objects are most obviously in line with our predictions, research has suggested that familiar objects encountered in novel locations are also preferred over familiar objects in familiar locations (e.g., Dix & Aggleton, 1999; Ennaceur, Neave, Aggleton, 1997; Beck & Luine, 2002). Thus, it seems possible that there is also a degree of appetitive novelty involved in experiencing a familiar object in the black compartment of the PA apparatus, which would constitute a novel location. Indeed, the only group that did not experience some degree of novel stimulation is the group that encountered a familiar object in the open field, which could possibly account for why their displayed fear (according to the total time in white compartment measure) approached ceiling at test. Importantly, none of the observed effects appear to be the product of general group differences in inhibitory tendencies or activity levels, which is evidenced by the lack of group differences in latency to cross into the black compartment at PA training.

Given that post-training exposure to familiar objects was, in general, shown to result in marginally greater fear (according to the test cross latency measure) than exposure to novel objects, and as rats typically prefer novel objects over familiar ones (e.g., Berlyne, 1950), indicative of their appetitive characteristics, it seems unlikely that the familiarity of the training object when encountered in the black PA compartment was somehow responsible for the reduced fear (less total time spent in white (safe) compartment) of that group. It is also worth mentioning that the lack of differences in fear reduction capabilities of novel and familiar objects encountered in the black PA compartment are not likely due to an inability to distinguish between the initial training and the counter-conditioning object. As a clear difference in the effects of novel and familiar objects encountered in the octagon open field was observed, and as Long-Evans rats appear capable of vision in low-light conditions (for review see Burn, 2008), the subjects should be capable of differential responses to the employed objects when encountered in the black compartment. Even if the objects were for some reason not recognized while in the black PA compartment (cf., Dellu, Fauchey, Moal, & Simon, 1997), however, the encountered object would essentially become novel irrespective of previous experience, and thus, along with the evidence of weaker fear in those rats encountering novel objects in the octagon open field, these data could still offer support for the notion that novelty exposure is capable of fear reduction. An alternative explanation, however, is that those subjects encountering objects in the black compartment of the PA chamber were displaying less fear at test due to extinction of fear to the black compartment that had occurred during their object exposure session. It is important to note, however, that simple extinction could not possibly account for all of the fear reducing capabilities of novelty exposure as those subjects experiencing novel objects in the octagonal open field had no additional exposure to the black PA compartment.

EXPERIMENT 2

Experiment 1 offered support for the notion that novel object exposure is more capable of counter-conditioning fear in a passive avoidance paradigm than is exposure to a familiar object. In experiment 2, we further examined whether the effects of object exposure while in the black PA compartment were resulting in greater fear reduction than would be expected by means of simple extinction of fear to the compartment.

METHOD

Subjects. Subjects consisted of thirty-six adult male Long-Evans rats ranging in age (M=345.44 days, SD= 33.23), obtained from the in-house breeding stock at Saint Joseph's University (originally derived from breeding pairs obtained from Taconic Farms Inc., Germantown, NY) and maintained in the exact fashion of Experiment 1. Again, given the range of ages employed in this study, efforts were made to approximately match the three experimental groups (see procedure section below) based on age.

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Materials. All materials were the same as those employed in the previous experiment.

Procedure. The procedure of Experiment 2 was largely consistent to that of Experiment 1, and is depicted in Figure 3. Animals were handled, pre-exposed to the octagon open field, and received both initial object training and passive avoidance training in the same fashion of the previous experiment. Approximately 24-hours following passive avoidance training, subjects underwent one of three procedures. Subjects in one group (Novel Obj./Black PA) were confined to the black PA compartment with a novel object for 5-min in the exact fashion of the previous study (n=12). A second group (Empty/Black PA) of subjects (n=12), designed to provide a measure of the effects of extinction of fear to the black PA compartment, were simply confined in the empty black compartment without any objects present for a period of 5-min. Subjects in the final group (n=12)(Empty/Octagon) were placed in the empty octagon open field without any objects present for a period of 5-min. This group was intended to provide a baseline measure of fear, while controlling for the effects of handling and transport. Approximately 24-hours later, all subjects underwent passive avoidance testing in the exact manner as Experiment 1. Latencies to cross into the black compartment at both training and test were again recorded, as was the total time spent in the white (safe) compartment during the test session, all in seconds.



Figure 3. Flowchart depicting the procedure of Experiment 2.

Statistical Analyses. One-Way ANOVAs followed by post hoc analyses on significant effects were conducted on each of the three dependent measures: latency to cross at training, latency to cross at test, and total time spent in the white (safe) compartment at test. As in the previous experiment, statistical outliers greater than 2 *SD* away from their group's mean on the measure in question were excluded prior to each analysis (i.e., outliers were only excluded from the analysis on which they differed from their fellow group members and were included in remaining analyses). All statistical analyses were performed via SPSS 16.0 for Mac, Release 16.0.2, and Levene's test was employed to examine homogeneity of variances on each measure following application of the exclusion criterion.

RESULTS

Table 2 contains the means (and standard deviations) pertaining to the cross latency at training, cross latency at test, and total time in the white compartment at test (all in sec) of the various groups following the exclusion of statistical outliers for each measure. In terms of latency to cross to the black side at training, one outlier was dropped from the empty octagon group and one was excluded from the empty black PA compartment group. Levene's test of homogeneity of variances failed to achieve significance (F(2, 31)=1.851, p=0.174). The One-Way ANOVA examining cross latency at training failed to obtain evidence of significant differences between the groups (F(2, 31)=0.057, p=0.945).

 Table 2. Means and Standard Deviations on Each Dependent Measure
 (in sec) in Experiment 2 (following the exclusion of statistical outliers)

Group	Training Cross Latency	Testing Cross Latency	Total Time in White Compartment
Object/Location	M (SD)	M (SD)	M (SD)
Empty/Octagon	7.91 (6.17)	183.42 (144.94)	259.55 (68.27)
Empty/Black PA	8.45 (3.88)	77.42 (115.04)	155.92 (91.26)
Novel Obj./Black PA	7.75 (5.34)	4.60 (5.50)	129.33 (86.02)

In regards to cross latency at test, two outliers were removed from the group that encountered the novel object in the black PA compartment. Levene's test of homogeneity of variances obtained evidence of significant differences in group variances (F(2, 31)=26.670, p=0.000). The One-Way

ANOVA examining test cross latency obtained evidence of significant group differences (F(2, 31)=7.367, p=0.002; Welch F(2, 14.732)=10.997,p=0.001). LSD post hoc analyses revealed that the group (control) that had encountered the empty octagon open field in between training and test displayed significantly slower cross latencies at test, indicating greater fear, than did the group that had encountered the empty black PA compartment (extinction) (p=0.025), or the group that had encountered the novel object in the black PA compartment (p=0.001). While the test cross latency of the group that had encountered the novel object in the black PA compartment was noticeably shorter than that of the extinction group, the difference between these two groups did not achieve statistical significance (p=0.133) according to the LSD analyses. Given the obtained evidence of homogeneity of variances, Games-Howell post hoc analyses were also employed on the test cross latency data. Interestingly, according to the Games-Howell analyses, only the group that had encountered the novel object in the black PA compartment significantly differed from the control group (p=0.003). The extinction group failed to differ from either the control group (p=0.141) or the group encountering the novel object in the black PA compartment (p=0.117). This can be seen in Figure 4.

On the measure of total time in the white (safe) compartment, one outlier was excluded from the group (control) that had encountered the empty octagon open field in between PA training and testing. Levene's test of homogeneity of variances failed to achieve significance (F(2, 32)=.416, p=0.663). The One-Way ANOVA examining total time in the white (safe) compartment at test obtained evidence of significant differences between the groups (F(2, 32)=7.821, p=0.002). LSD post hoc analyses revealed that the extinction group (p=0.005) and novel object in black PA compartment (p=0.001) groups both demonstrated significantly less fear than the control group, as indicated by fewer seconds spent in the white compartment, and the extinction and novelty groups failed to differ from one another (p=0.438). This is depicted in Figure 5.

DISCUSSION

Experiment 2 attempted to determine whether exposure to a novel object while in the black fear-evoking compartment of a PA chamber would result in greater fear reduction at test than would a more simple extinction procedure. The LSD post hoc analyses on cross latency at test and total time in the white (safe) side suggested that the group that experienced a novel object in the black PA compartment and the simple extinction group both displayed less fear than did the baseline control group. The GamesHowell post hoc analyses of test cross latency, however, suggested that only the group encountering the novel object in the black PA compartment displayed significantly less fear than controls. These results suggest that novelty exposure was more effective at reducing fear than the simple extinction procedure, and offers evidence of reduced fear as a result of novelty exposure independent of extinction. As was seen in the first experiment, these effects cannot be explained via differences in activity levels or initial inhibitory tendencies of the groups as no differences in cross latency at training were observed.



Test Cross Latency

Figure 4. Latency to cross into the black PA compartment at test of the various groups during Experiment 2. Error bars represent SEM.



Total Time in White (Safe) Compartment

Figure 5. Total time spent in the white (safe) PA compartment at test of the various groups during Experiment 2. Error bars represent SEM.

GENERAL DISCUSSION

The present study sought to determine whether post-training exposure to a novel or familiar object, encountered in either the location of the original fear conditioning (black PA compartment) or in a neutral setting (octagonal open field) would prove capable of reducing fear at subsequent test in a passive avoidance task. Indeed, evidence was found that encountering a novel object between PA training and test sessions resulted in greater fear reduction than encountering familiar ones (Exp 1). Encounter location proved to matter little to the overall amount of fear that was displayed at test as even those subjects experiencing a novel object in a neutral setting still displayed reduced fear relative to subjects experiencing familiar objects (Exp 1) in the neutral setting. Evidence was also obtained suggesting that familiar objects when encountered in the black PA compartment were more effective at reducing fear than familiar objects encountered in the octagon open field, but this seems most likely to occur due to a degree of novelty gained by the objects when encountered in a location other than that where the subjects had originally received initial object training (cf., Dix & Aggleton, 1999; Ennaceur, Neave, & Aggleton, 1997; Beck & Luine, 2002).

In Experiment 2 we specifically compared the amount of fear reduction produced by novel object exposure and a more simple extinction procedure, and found evidence that both groups resulted in reduced fear relative to a baseline control group. Moreover, according to some analyses, experiencing a novel object in the black PA compartment resulted in greater fear reduction than did the simple extinction condition. Figures 4 and 5 certainly elude to this conclusion. Additionally, given the lack of significant differences in displayed fear between those subjects encountering the novel object in the black PA compartment and those experiencing a novel object in the octagonal open field in Experiment 1, we can conclude that novelty exposure is by itself capable of reducing conditioned fear in a passive avoidance task.

These results should not come as a surprise given the appetitive qualities of interacting with novel objects (e.g., Berlyne, 1950; Bevins, Besheer, Palmatier, Pickett, & Eurek, 2002; for review see Anderson, 2006ab). Indeed, since Berlyne's (1950) original demonstration of the phenomenon, countless studies have shown that rats prefer to spend their time exploring novel objects over those with which they are familiar (e.g., Ennaceur & Delacour, 1988; Anderson, Karash, Ashton, & Riccio, 2003; Anderson, Jablonski, & Klimas, 2008). Additionally, novel objects have been known to instill a conditioned preference for the location in which they were encountered (Bevins, Besheer, Palmatier, Pickett, & Eurek, 2002), evidencing their rewarding qualities. Such place conditioning effects have not been seen with familiar objects (Bevins et al., 2002), which offers additional support for the conclusion of this study that the reduced fear demonstrated by animals experiencing a familiar object in the black PA compartment in Experiment 1 was not likely due to the familiarity of the object itself. Indeed, an explanation of this group's reduced fear based on the object regaining some degree of novelty when encountered in a different

location (cf., Beck & Luine, 2002; Dellu, Fauchey, Moal, & Simon, 1997; Dix & Aggleton, 1999; Ennaceur, Neave, & Aggleton, 1997) would appear more likely. Future research may wish to test the fear-reducing capability of other novel stimuli (e.g., additional objects, locations, etc.) interposed between initial fear conditioning and subsequent test in order to test the validity of this claim and generality of the present results.

Given the well-established appetitive aspects of novelty (e.g., Berlyne, 1950; Bevins et al., 2002), we have chosen to explain the present results in terms of a counter-conditioning of fear by the novel objects. While such an explanation appears most warranted, an alternative explanation may deserve investigation in future research. In his SOP model, Wagner (e.g., 1976, 1981) posits a model of classical conditioning that focuses on information possessing and the need for rehearsal (for a review of SOP and its implications for behavior therapy see Zinbarg, 1993). According to this model, when a surprising event occurs rehearsal mechanisms are engaged, and the activation of these mechanisms may interfere with (i.e., disrupt rehearsal of) other events (Wagner, 1971). Perhaps the lowered fear following novel object exposure in the present study is a product of novelty-induced distraction causing a disrupted rehearsal of the original fear conditioning in a manner similar to that prescribed by the SOP model, as opposed to a counter-conditioning per se. As distraction is thought to impede the long-term benefits of exposure therapy (Zinbarg, 1993), future research may wish to examine the effects demonstrated in the present report over multiple object-exposure-session-totest intervals. But again, as novel objects have themselves been shown instill a conditioned place preference for the locations in which they are encountered (Bevins et al., 2002), their ability to counter-condition fear seems to be the most likely explanation of the present results.

Irrespective of the underlying mechanism, the present results suggest that exposure to novelty between initial fear conditioning and subsequent test is capable of reducing the fear displayed at test. In addition to offering greater insight into the general behavioral processes of the animals under investigation, these results may have implications in clinical settings for humans. Indeed, the present findings suggest that clients that have conditioned fears may benefit from exposure to novel and interesting stimuli while undergoing treatment for their fear. Additional research, perhaps employing human participants, is necessary to further examine this possibility.

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