Published in final edited form as:

Psychopharmacology (Berl). 2015 September ; 232(17): 3173–3181. doi:10.1007/s00213-015-3967-1.

Developmental emergence of an obsessive-compulsive phenotype and binge behavior in rats

Nadja Freund^{1,2,3}, Britta S. Thompson¹, Kevin J. Norman¹, Patrick Einhorn^{1,4}, and Susan L. Andersen^{1,2}

¹Laboratory for Developmental Neuropharmacology, McLean Hospital, Harvard Medical School

²Department of Psychiatry, McLean Hospital, Harvard Medical School

³Department of Psychiatry and Psychotherapy, University of Tübingen, Tübingen, Germany

⁴Department of Psychology, Boston College

Abstract

Rationale—Obsessive-compulsive disorder (OCD) gradually emerges and reaches clinical significance during early adulthood. Whether a predisposition for OCD manifests as binge eating disorder earlier during adolescence is proposed.

Objectives—To further characterize how OCD-like behaviors increase across maturation and to determine whether an OCD-like predisposition increases the likelihood of binge eating during adolescence.

Methods—Male and female Sprague-Dawley rats were injected with the tricyclic antidepressant clomipramine (CMI, 15 mg/kg) or saline vehicle twice daily between postnatal day 9–15. Both groups were tested for perseverative (spontaneous alternation) and anxiety-like (elevated plus maze; marble burying) behaviors during juvenility (day 28), adolescence (day 60), and adulthood (day 90). Both motivation to eat sucrose pellets and binge eating on fat were investigated.

Results—Sex- and age-dependent increases in anxiety-like and perseverative behavior were observed in CMI subjects. Differences in consummatory behaviors emerged during late adolescence, while no significant differences in alternation or anxiety-like behaviors were detected between CMI and vehicle animals until adulthood. Adolescent CMI females consumed more sucrose pellets in 30 minutes relative to vehicle females, whereas adolescent CMI males consumed approximately half as much as vehicle males. Sucrose consumption did not differ between groups in adulthood. Adolescent CMI rats demonstrated more fat bingeing than vehicles, independent of sex.

Conclusions—OCD-like behaviors are emerging during adolescence, but sucrose consumption and fat bingeing in CMI-treated animals significantly precedes the appearance of anxiety and

Correspondence to: S.L. Andersen, McLean Hospital, 115 Mill St, Belmont MA 02478, sandersen@mclean.harvard.edu.

Financial disclosure:

The authors declare that they have no competing financial interests.

No conflict of interest exists.

perseveration. This OCD-like phenotype emerges fully during adulthood, suggesting that eating may likely serve as a coping strategy in these animals.

Keywords

anxiety; binge eating; clomipramine; maturation; obsessive-compulsive disorder; sex differences

Introduction

Multiple mental disorders like anxiety, depression, and OCD are associated with binge eating (McElroy et al. 1994; Peterson et al. 2012). Binge eating may be triggered by anxiety, but bingeing reduces anxiety and increases reward sensation during and after the episode (Kaye 2008). Similarly, anxiety is triggered in individuals with OCD during the obsessive phase and is reduced/released when the compulsive act is executed. While there is a high likelihood that OCD and eating disorders share common pathology, clinical research has been unable to establish whether OCD is a risk factor or a consequence of eating disorders (Kaye et al. 2004). One possibility is that the developmental emergence of OCD behaviors in humans predisposes an individual to binge eating despite not yet reaching clinical criteria. Binge eating typically emerges during mid-late adolescence (i.e., 16–18 years; Stice 1999), when sex differences in binge eating disorder also surface (Culbert et al. 2013). The average age of onset for late onset OCD is in the early 20's (Taylor 2011), although this predisposition for OCD may partially manifest as binge eating beforehand during late adolescence. To test this hypothesis, we determined whether neonatal clomipramine (CMI) administration in rats would increase the motivation to eat, or binge, in late adolescence prior to the later, gradual onset of OCD-like behaviors in adulthood.

Neonatal administration of the tricyclic antidepressant CMI has adverse effects on rat male behavior in adulthood (Vogel et al. 1990b) that includes an increase in OCD-like behaviors (Andersen et al. 2010; Andersen and Thompson, 2011). Exposure to CMI between postnatal day (P) 9 and P15 increases depressive-like behaviors (Mirmiran et al. 1981; Vogel et al. 1990b), anxiety-like behavior, reduces spontaneous alternation, increases hoarding, impairs working memory (Andersen et al. 2010), and increases repetitive checking behavior in adult rats (Szechtman et al. 2011; Andersen and Thompson 2011). CMI treatment also produces corticostriatal dysfunction by modulating serotonin and dopamine systems, which is also observed in individuals with eating disorders (Aveena et al. 2008) and OCD (Boker and Anderson 2014). Together these studies suggest that neonatal CMI increases OCD-like behaviors in adult males, although little is known about the developmental trajectory of these behaviors and whether sex differences exist.

The present study has two aims. First, this study investigates how OCD-like behaviors of anxiety and spontaneous alternation emerge across the course of development and sex. Second, we determine whether dysregulated food consumption is evident in CMI animals relative to controls under non-food deprived conditions. The emergence of OCD-like behaviors in concert with dysregulated eating in females and males during adolescence in CMI treated animals would support our hypotheses.

Methods

Subjects

Sprague-Dawley litters (comprised of 5 male/5 females) with their dams arrived from Charles Rivers (Wilmington, MA) at McLean Hospital at P4 and were housed on a 12h/12h light/dark cycle (lights on at 06:00h) with food and water *ad libitum*. Temperature and humidity were kept constant at $22 \pm 2^\circ$ and $55 \pm 25\%$, respectively. Between P9–16, rats were weighed daily at 09:00am and given i.p. injections of either the saline vehicle or 15 mg/kg clomipramine (Sigma-Aldrich, St. Louis, MO), followed 7 hours later by a second injection for a total of 16 injections across the eight days. Following previous protocol (Andersen et al. 2010), the second administration of CMI was given in order to maintain relatively constant drug levels throughout the day based on the half life of CMI in the rat brain (6.2 hours, Weigmann et al. 2000). Injections were given by either a 50 or 100 μ l Hamilton syringe in a 1ml/kg volume, according to our methods (Andersen et al. 2010; Andersen and Thompson 2011). We did not observe any treatment-induced or shipping-related effects of stress on body weight.

Litters were weaned at P21 and housed with same-sex littermates. Only one male and female subject from each litter was assigned to an individual condition. P25 represents childhood or juvenility, P60 is a mid to late adolescence, and P100 is adulthood (Andersen 2003). Subjects were tested ± 1 day of the stated ages. Protocols are in accordance with approved IACUC procedures at McLean Hospital and NIH guidelines.

Similar to our original paper (Andersen et al. 2010), the majority of rats were subjected to an anxiety battery (spontaneous alternation task, elevated plus maze, and marble burying) that was counterbalanced across paradigms. We have not observed any order effects in testing. A separate group of rats were used in the operant conditioning tasks. Females were not staged for two reasons: 1) vaginal swabs themselves have been known to alter cycling and even induce false pregnancy (Walker et al. 2002); and 2) the data presented are collected over three days, which encompasses and averages 75% of any of the estrogen-related differences that may have occurred (Marcondes et al. 2001). Future studies can indeed investigate the role that gonadal hormones may have in the observed effects.

Spontaneous alternation

The spontaneous alternation task was conducted in a T-maze (Andersen et al. 2010). The spontaneous alternation task is based on the assumption that rats will explore their environment to optimize the amount of area investigated (Kokkinidis and Anisman 1976). Thus, subjects are expected to alternate between right and left arm choices and inflexible behavior is indicated by the perseveration to one side. Arm choice was recorded for a total of nine trials, and the maze was cleaned with 70% ethanol solution between each trial while the subject waited in a holding cage for 1 min between trials. Different groups of animals were tested at ages P25, P60 and P100, with an $n=6-8$ for each group.

Marble burying

The test was conducted in a $41 \times 36 \times 10$ cm box cage with 5 cm of fresh hardwood chip bedding (Alpha Chips) (Andersen et al. 2010). An array of 24 standard marbles was evenly spaced over the surface. Individual subjects were placed in the test cage for 15 min. The number of marbles buried 70 % was recorded (Broekkamp et al. 1986). New bedding was used for each animal and marbles were cleaned with a 70% ethanol solution between animals. Different groups of animals were tested once at ages P25, P60 and P100, with an $n=6-8$ for each group.

Elevated plus maze (EPM)

The maze consisted of two open and two closed arms (each 10×50 cm) that were attached to a central platform and raised 50 cm above the floor (Andersen et al. 2010). Ambient light was 200–250 lux. Rats were placed on the central platform facing the closed arms and their behavior was examined over a 5-minute period. Measures of time spent in the open and closed arms as well as total arm entries were recorded. Different groups of animals were tested once at ages P25, P60 and P100, with an $n=6-8$ for each group.

Sucrose pellet consumption

Training—Subjects were not food deprived. P60 and P100 rats were used in these studies. Because juveniles did not demonstrate any behavioral change from the CMI treatment for the other metrics, we did not assess their behavior in the operant conditioning task. Subjects were initially trained on a fixed ratio (FR) 1 program in an operant conditioning chamber (Lafayette Instruments; Lafayette, IN) controlled by the Matlab (Mathworks; Natick MA) based Biopsychology Toolbox (Rose et al. 2008). A white square (7×7 cm) was displayed on the touchscreen, where a single nose poke to the stimulus resulted in the disappearance of the stimulus and presentation of 1 sucrose pellet (45 mg, Bio-Serv; Frenchtown NJ). Trials were repeated throughout the session with inter-trial-interval of 30 seconds during which the screen was black. The rats underwent one 30 min session per day, 5 days a week until they poked the target square at least 20 times within one session. These data not only provide an assessment of the ability to learn operant conditioning, but also determine general deficits in motor control, learning and motivation. An $n=6-8$ subjects were used, with the exception of the adolescent males, where $n=2$ subjects failed to learn the task after weeks. We attribute this effect to the lack of food-deprivation, commonly used to initiate responding in sucrose consumption tasks. As a result, this cell has an $n=5$. All of the females were included ($n=8$) for each condition.

Testing 1—Once the learning criterion was reached, the schedule changed to FR3 to better establish goal directed behavior. The number of sucrose pellets consumed was assessed across the final three FR3 30 min sessions, where data were collected until rats showed stable behavior (the number of completed trials was within 10% on 3 consecutive days). In this step no minimum amount of trials had to be completed.

Testing 2—Following the first test session, a progressive-ratio program (PR) was used. In this schedule of reinforcement, the number of responses required for a reward is advanced from trial to trial according to the following equation (Richardson and Roberts 1996):

$$\text{Response ratio (rounded to nearest integer)} = 5e^{[\text{trial number} \times 0.2]} - 5$$

Such that the # of responses needed followed this series: 1,2,4,6, 9, 12, 15, 20, 25, 32, 40, 50, 62,77, 95, 118, 145, 178,219, 268, 328, 402, 492, 603 Subjects remained in the operant conditioning chambers for 3 hrs/ day or until they did not respond for 30 minutes. Data were collected until the number of completed trials was within 15% across 3 consecutive days. This criterion minimized any individual differences in female behavior due to estrous cycle. The breakpoint, the highest ratio completed, is interpreted as a measure of motivation to consume sucrose (Richardson and Roberts 1996).

Crisco bingeing

Following the methods of Puhl et al. (2010), rats were allowed free access to a high fat substance (Crisco® All-Vegetable shortening, J. M. Smucker Co., Orrville, OH) for 1 hour on Monday, Wednesday, and Friday. Intermittent access produces the greatest percentage of intake change compared with constant access (Puhl et al. 2010). All subjects had continuous access to normal chow and water throughout the study. Access to fat was provided starting on P55 between 3–4 pm on access days for 5 weeks, with new shortening provided each Monday. Body weight and intake of shortening were measured after each session in order to express fat consumption in terms of grams of fat consumed per kilogram of body weight.

Statistical analyses

Data were analyzed by 3-way ANOVA with Drug, Sex and Age as variables using Stata 12 (StataCorp; College Station TX). Independent of a significant Age $\times$ Drug or Sex $\times$ Drug interaction, planned comparisons (t-tests, with Bonferroni correction) were conducted for the P100 group for spontaneous alternation, marble burying, elevated plus maze to determine whether we replicated our previous findings (Andersen et al. 2010), as well as to determine sex differences at this age. A mixed ANOVA with the between subject variables of Drug and Sex and the within-subject variable of [days] was used to analyze fat bingeing. Data were normalized to the first two weeks of intake, as previously performed (Puhl et al. 2010; Bello et al. 2014) to determine changes in % intake change, not overall consumption. Significance was considered at $p < 0.05$.

Results

Spontaneous alternation

A significant main effect of Drug ($F_{1, 83} = 8.12$, $p < 0.006$), but no interaction with Sex or Age for the number of alternations divided by the number of total turns was revealed (Figure 1). Post-hoc t-tests revealed that the adult animals drive this effect ($P < 0.005$, Bonferroni corrected).

Marble burying

A significant interaction of Sex $\times$ Age ($F_{2, 97} = 11.85$, $p < 0.0001$) and a main effect of Age ($F_{2, 96} = 74.75$, $p < 0.0001$) were found for the number of marbles buried (Figure 2a, b). While the effect of Drug only reached trend level ($p = 0.065$), planned comparisons between CMI

and vehicle animals revealed a significant difference between CMI and control in P100 males ($p<0.05$, Bonferroni corrected), but not the females.

EPM

We found a Sex $\times$ Age $\times$ Drug interaction ($F_{2, 103}=3.13$, $p<0.05$) for time spent in the open arm (Figure 3a, b). To further investigate this interaction, we analyzed males and females separately. A significant effect of Drug was observed for the males ($F_{2, 52}=4.28$, $p<0.05$) that did not interact with Age, where CMI decreased time spent in the open arm overall. Females demonstrated a 2-way interaction of Age $\times$ Drug ($F_{2, 52}=3.86$, $p<0.03$) with CMI females spending progressively less time in the open arm than vehicles. An Age $\times$ Sex $\times$ Drug interaction was observed for the number of arm entries ($F_{2, 103}=5.28$, $p<0.008$). This interaction is shown in Figure 3 c, d. CMI generally reduced arm entries in males relative to vehicles, whereas arm entries were significantly lower in CMI females during adulthood only relative to vehicles.

Sucrose consumption

A significant Sex $\times$ Drug interaction was found for the number of sucrose pellets consumed ($F_{1, 41}=7.55$, $p<0.009$) under the FR3 schedule (Figure 4a,b). CMI males decreased consumption relative to vehicles, whereas CMI females increased consumption. The breakpoint, defined as maximal number of responses performed to receive one food pellet, showed a main Sex effect ($F_{1, 41}=9.99$; $p<0.004$), where females have a higher breakpoint overall than males independent of Drug. For this reason, we analyzed the sexes in separate Age $\times$ Drug 2-way ANOVAs. A trend of a main Drug effect in males ($F_{1, 19}=3.73$, $p<0.07$), but no effects of Age were found.

Fat Bingeing

Initially, we observed a significant Sex effect of consumption when data are expressed as g/kg between Days 2–5 (day 1 is too variable; $F_{1, 26}=7.35$, $p=0.01$); females consumed more fat than males (males: 5.69 ± 1.1 vs. females 9.85 ± 1.0 g). No significant Drug effect was observed, but at this baseline assessment, vehicle subjects consumed an average of 3.02 ± 0.34 g of fat and CMI subjects consumed 2.08 ± 0.32 g of fat. To determine whether our binge escalation paradigm produced comparable sex differences to those described by others (e.g., Klump et al. 2013), a Day $\times$ Sex interaction ($F_{1, 28}=50.78$, $p<0.0001$) showed that fat consumption escalated between Day 2 and Day 15, where females consumed more than males.

To better characterize individual subject's bingeing, g/kg data were normalized as a function of this initial baseline consumption and the remaining data (Binge days 6–15) were expressed as a percent change (Figure 5). A Drug $\times$ Sex $\times$ (Binge) ANOVA was conducted, with Binge as a within-subjects variable. A significant Drug $\times$ (Binge) interaction was found for the amount of fat consumed ($F_{1, 26}=5.03$, $P<0.05$). Sex did not significantly influence fat consumption when analyzed across the days, although a Drug main effect was also observed ($F_{1, 26}=4.51$, $p<0.05$). Post-hoc comparisons with Bonferroni correction indicated that bingeing in CMI animals was significantly higher during the last three days of testing ($P<0.05$) as they entered into adulthood. Weight gain (Figure 6) did not change as a function

of Drug (nor interact with either Sex or Binge), but was significantly affected by Sex $\times$ Binge ($F_{1, 26} = 50.85$, $p < 0.0001$).

Finally, the percentage of rats reaching the upper tertile of binge behavior was investigated (Boggiano et al. 2007). As Figure 7 shows, the top 25% of all subjects of the normalized binge data (g of chow normalized to kg weight) across the last nine sessions are primarily comprised of CMI subjects, whereas the bottom tertile contains $n=2$ female CMI subjects. Based on Boggiano's classification, three of the eight CMI males are binge-prone (e.g., maintain binge eating at a high level throughout the experiment) and CMI females fail to meet this criterion.

Discussion

The results of this study show that rats treated with CMI during the neonatal period have progressively increasing anxiety-like and perseverative behaviors that fully manifest in adulthood. Binge eating is associated with the intermittent consumption of large amounts of food within a short period of time (American Psychiatric Association, 2013). Here, we show that this OCD-like preponderance produced by CMI altered eating behaviors in rats without food deprivation. CMI increased fat bingeing during late adolescence relative to vehicles in both males and females. In an operant conditioning task, consumption of sucrose was higher in female CMI than vehicle subjects with the reverse in males; with no change in motivation was observed in adolescence. These data support the hypothesis that CMI exposure predisposes towards abnormal eating.

Consistent with earlier studies (Mirmiran et al. 1981; Yannielli et al. 1999; Andersen et al. 2002; Andersen et al. 2010), neonatal CMI injections induced perseverative and anxiety-like (EPM, marble burying) behavior in adulthood. The current findings demonstrate that these behaviors gradually manifest over the course of development, but do not significantly differ between CMI pre-exposed animals and controls before adulthood. Moreover, our results parallel sex differences observed in humans, which they emerge before or during adolescence (Cyranowski et al. 2000). While males show minimal change in anxiety-like behavior across age, female rats show progressively increasing anxiety-like behavior, demonstrated by reduced time spent in the open arms of the EPM and reduced arm entries by late adolescence (P55). This observation is also consistent with (Lynn and Brown 2010) who show that female anxiety-like behavior peaks during adolescence and wanes with maturation. As a result, a predisposition exists for elevated anxiety in CMI subjects during adolescence, but the sex differences in anxiety or perseverative behavior due to CMI are most pronounced in adulthood when the trajectory likely stabilizes. This finding is in line with clinical research that shows that the age of onset for non-genetic OCD symptoms is in early adulthood (Taylor 2011).

In addition, the topography of anxiety changes between males and females. Sex differences in coping strategies have been described clinically, where boys show greater aggression than girls (Spigelman et al. 1991). The marble burying task of anxiety may be more of an adaptive, defensive coping style and is elevated in male rats in the current study. CMI males had increased marble burying relative to control males, consistent with our previous

observations (Andersen et al. 2010). Female rats, on the other hand, spent more time in the closed arm of the EPM than males, suggestive of more avoidant-type anxiety in females (Leussis and Andersen 2008). CMI females were more avoidant, but not defensive as they did not bury more marbles. This difference in the strategy of coping with anxiety in females is more consistent with harm avoidance, which has been associated with binge eating disorders (Peterson et al. 2010).

The typical developmental trajectory data may reveal a potential window of vulnerability to anxiety-disorders that interact with reward (Tomasi and Volkow 2013). The number of marbles buried increased over development in males and females independently of drug. This increase, however, progressively increased in males until adulthood, while the number of marbles buried peaked by adolescence in females. Experiences prior to what may be the pinnacle of vulnerability to develop an anxiety-related disorder may direct the trajectory towards vulnerability or resilience as females continue to mature. To this end, perseveration in the spontaneous alternation task may further telescope pathological behavior. While control males demonstrated relatively stable alternation behavior across age, CMI males progressively alternated less as they matured. A different pattern is observed in females as controls progressively alternate more across maturation, whereas CMI females do not change across maturation and, in essence, their trajectory is flat.

As anxiety progressively rises in CMI subjects, an opportunity to reduce these behaviors through sucrose or fat consumption is a characteristic of binge eating – especially within a restricted period of time in the absence of hunger (Corwin and Buda-Levin 2004; de Jong et al. 2012). Changes in reward-related behavior in CMI-treated animals bias towards disordered eating. Female CMI adolescents consumed more sucrose under an FR3 schedule than controls or CMI adult females within a defined period of time (30 minutes). This higher level of sucrose consumption was observed without food deprivation, which is commonly used in operant paradigms (Weatherly et al. 2003). This operant test assesses goal-directed behavior since the animals have to respond three times to obtain a sucrose pellet (Dezfouli and Balleine 2012). Other studies motivated binge-like eating by food deprivation (e.g., Specker et al. 1994), intermittent access to palatable food (Rossetti et al. 2013; Klump et al. 2013), or sham feeding (Kim 2012). We hypothesize that the greater consumption of sugar in female CMI adolescents is related to their anxiety-like behavior. Decreased time spent in the open arm of the EPM (while not yet reaching significance) is an internalizing, avoidant-type response that does not address the “source” of the anxiety in the same way that the CMI males defensively bury marbles. Similar to the human condition, we propose that this unresolved anxiety results in seeking comfort elsewhere. In this case, increased consumption of sugar pellets.

We also investigated fat bingeing, as operant responding for sucrose may be subject to indirect effects of CMI on learning differences. The fat bingeing paradigm measures fat consumption within a limited time (60 min) with an intermittent schedule (M, W, F). This schedule produces the greatest amount of binge behavior compared with 24-hour access one day a week or constant access (Puhl et al. 2010). An escalation of fat intake is found in binge eating (Babbs et al. 2012) and has been described in other rodent studies using a similar paradigm (Bello et al. 2014). Fat bingeing escalated across repeated days of testing

independent of sex. Similar to other studies, control females consumed more fat than males (Klump et al. 2013). We did not measure chow intake, leaving open the possibility that males compensated their caloric intake by reducing normal food consumption on testing day (Klump et al. 2013). Our CMI subjects binge on fat not for caloric benefit as they are not food-deprived, but most likely as a result of their anxiety-like predisposition. For these reasons, we believe that our model may typify an emotionally-based eating dysfunction.

Much like the developmental increase in the incidence of bingeing in humans (Schneider 2003), we hypothesize that the underlying preponderance of OCD-like behavior and the neurobiology in our model could facilitate the adolescent onset of over-eating in females. Disruptions in dopaminergic (Bello and Hajnal 2010) and serotonergic (Bailer and Kaye 2011) signaling induce binge eating. Previous studies have shown that these systems are altered by neonatal CMI (Andersen et al. 2008). Repeated exposure to intermittent sucrose (or fat) is likely to become reinforcing on its own and leads to addictive bingeing (Volkow et al. 2008). As such, dysfunction of motivational and reward systems are likely to be involved in the observed behavioral changes. For example, CMI animals are already predisposed to habitual behaviors (Andersen and Thompson 2011). Our findings of sucrose eating are in line with other studies that show only female animals demonstrated binge eating and not males. While we have shown that females overall are more motivated to work for sucrose than males, we observed no sex differences in fat bingeing. Sex differences are evident in tests following intermittent access to food with female rats 2–6 times more likely to binge than male rats (Klump et al. 2013). In the current study, females consumed roughly twice the amount of fat as males, independent of CMI exposure. Clinical studies show that female adolescents binge more than adolescent males (Croll et al. 2002). Consistent with this possibility, CMI pre-exposed adolescent males consumed less sucrose pellets compared to vehicles, which may suggest anhedonia (John et al. 2012). Other anhedonic behaviors, including decreased intracranial self-stimulation (Vogel et al. 1990a), sexual behavior (Neill et al. 1990), and reduced 24 hour sucrose consumption (Vogel et al. 1990b) have been found in adult CMI pre-exposed male rats.

This study showed that while the consequences of neonatal exposure to CMI in adulthood (i.e. OCD-like behavior) are similar in males and females, the developmental emergence of aberrant eating behavior during adolescence differs in males and females. Sucrose consumption is higher in CMI adolescent females, possibly increasing vulnerability to binge on sweet substances during adolescence in females more than males. However, CMI exposure increases binge-like eating behavior in both males and females. Clinically, the emergence of OCD during adolescence is more likely in males than females (Flament et al. 1988). These data extend that perspective: the lower rate of early onset of OCD in females than males could be explained by the fact that females more often develop binge eating during adolescence before OCD symptoms manifest fully during adulthood.

Acknowledgments

We would like to thank the Judah Family Foundation for OCD Research and the Simches Family for their unwavering support. Jodi Lukkes is acknowledged for her help with training the animals.

Abbreviations

BED	binge eating disorder
CMI	clomipramine
EPM	elevated plus maze
FR	fixed ratio
OCD	obsessive-compulsive disorder
P	postnatal day
PR	progressive ratio

References

- American Psychiatric Association. Diagnostic and statistical manual of mental disorders: DSM-V (5th ed.). Washington, DC: American Psychiatric Association; 2013.
- Andersen SL. Trajectories of brain development: point of vulnerability or window of opportunity? *Neurosci Biobehav Rev.* 2003; 27:3–18. [PubMed: 12732219]
- Andersen SL, Dumont NL, Teicher MH. Differences in behavior and monoamine laterality following neonatal clomipramine treatment. *Dev Psychobiol.* 2002; 41:50–57. [PubMed: 12115290]
- Andersen SL, Greene-Colozzi EA, Sonntag KC. A novel, multiple symptom model of obsessive-compulsive-like behaviors in animals. *Biol Psychiatry.* 2010; 68:741–747. [PubMed: 20619828]
- Andersen SL, Thompson BS. Reply to: Animal Models of Obsessive-Compulsive Disorder. *Biol Psychiatry.* 2011; 69:e31–e32. [PubMed: 21643508]
- Avena NM, Rada P, Hoebel BG. Evidence for sugar addiction: Behavioral and neurochemical effects of intermittent, excessive sugar intake. *Neurosci Biobehav Rev.* 2008; 32:20–39. [PubMed: 17617461]
- Babbs RK, Wojnicki FH, Corwin RL. Assessing binge eating. An analysis of data previously collected in bingeing rats. *Appetite.* 2012; 59:478–482. [PubMed: 22641146]
- Bailer UF, Kaye WH. Serotonin: imaging findings in eating disorders. *Curr Top Behav Neurosci.* 2011; 6:59–79. [PubMed: 21243470]
- Bello NT, Hajnal A. Dopamine and binge eating behaviors. *Pharmacol Biochem Behav.* 2010; 97:25–33. [PubMed: 20417658]
- Boggiano MM, Artiga AI, Pritchett CE, Chandler-Laney PC, Smith ML, Eldridge AJ. High intake of palatable food predicts binge-eating independent of susceptibility to obesity: an animal model of lean vs obese binge-eating and obesity with and without binge-eating. *Int J Obes (Lond).* 2007; 31:1357–1367. [PubMed: 17372614]
- Bokor G, Anderson PD. Obsessive-compulsive disorder. *Journal of pharmacy practice.* 2014; 27:116–130. [PubMed: 24576790]
- Corwin RL, Buda-Levin A. Behavioral models of binge-type eating. *Physiol Behav.* 2004; 82:123–130. [PubMed: 15234600]
- Croll J, Neumark-Sztainer D, Story M, Ireland M. Prevalence and risk and protective factors related to disordered eating behaviors among adolescents: relationship to gender and ethnicity. *J Adolesc Heal.* 2002; 31:166–175.
- Culbert KM, Breedlove SM, Sisk CL, Burt SA, Klump KL. The emergence of sex differences in risk for disordered eating attitudes during puberty: a role for prenatal testosterone exposure. *J Abnorm Psychol.* 2013; 122:420–432. doi: 10.1037/a0031791. [PubMed: 23713501]
- Cyranowski JM, Frank E, Young E, Shear MK. Adolescent onset of the gender difference in lifetime rates of major depression: a theoretical model. *Archives of General Psychiatry.* 2000; 57:21–27. [PubMed: 10632229]

- De Jong JW, Vanderschuren LJM, Adan RAH. Towards an Animal Model of Food Addiction. *Obes Facts*. 2012; 5:180–195. [PubMed: 22647301]
- Dezfouli A, Balleine BW. Habits, action sequences and reinforcement learning. *Eur J Neurosci*. 2012; 35:1036–1051. [PubMed: 22487034]
- Flament MF, Whitaker A, Rapoport JL, Davis M, Berg CZ, Kalikow K, Sceery W, Shaffer D. Obsessive compulsive disorder in adolescence: an epidemiological study. *J Am Acad Child Adolesc Psychiatry*. 1988; 27:764–771. [PubMed: 3264280]
- John CS, Smith KL, Van't Veer A, Gompf HS, Carlezon WA Jr, Cohen BM, Ongur D, Bechtholt-Gompf AJ. Blockade of astrocytic glutamate uptake in the prefrontal cortex induces anhedonia. *Neuropsychopharmacology*. 2012; 37:2467–2475. [PubMed: 22739467]
- Kaye W. Neurobiology of anorexia and bulimia nervosa. *Physiol Behav*. 2008; 94:121–135. [PubMed: 18164737]
- Kaye WH, Bulik CM, Thornton L, Barbarich N, Masters K. Comorbidity of anxiety disorders with anorexia and bulimia nervosa. *Am J Psychiatry*. 2004; 161:2215–2221. [PubMed: 15569892]
- Kim SF. Animal models of eating disorders. *Neuroscience*. 2012; 211:2–12. [PubMed: 22465439]
- Klump KL, Racine S, Hildebrandt B, Sisk CL. Sex differences in binge eating patterns in male and female adult rats. *Int J Eat Disord*. 2013; 46:729–736. [PubMed: 23625647]
- Kokkinidis L, Anisman H. Interaction between cholinergic and catecholaminergic agents in a spontaneous alternation task. *Psychopharmacology (Berl)*. 1976; 48:261–270. [PubMed: 823581]
- Leussis MP, Andersen SL. Is adolescence a sensitive period for depression? Behavioral and neuroanatomical findings from a social stress model. *Synapse New York N*. 2008; 62:22–30.
- Lynn DA, Brown GR. The ontogeny of anxiety-like behavior in rats from adolescence to adulthood. *Dev Psychobiol*. 2010; 52:731–739. [PubMed: 21117243]
- Marcondes FK, Miguel KJ, Melo LL, Spadari-Bratfisch RC. Estrous cycle influences the response of female rats in the elevated plus-maze test. *Physiol Behav*. 2001; 74:435–440.
- McElroy SL, Phillips KA, Keck PE Jr. Obsessive compulsive spectrum disorder. *J Clin Psychiatry*. 1994; 55(Suppl):33–51. discussion 52–3. [PubMed: 7961531]
- Mirmiran M, van de Poll NE, Corner MA, Uylings HB, van der Gugten J, Boer GJ. Suppression of active sleep by chronic treatment with chlorimipramine during early postnatal development: effects upon adult sleep and behavior in the rat. *Brain Res*. 1981; 204:129–146. [PubMed: 7195762]
- Neill D, Vogel G, Hagler M, Kors D, Hennessey A. Diminished sexual activity in a new animal model of endogenous depression. *Neurosci Biobehav Rev*. 1990; 14:73–76. [PubMed: 2325943]
- Peterson RE, Latendresse SJ, Bartholome LT, Warren CS, Raymond NC. Binge Eating Disorder Mediates Links between Symptoms of Depression, Anxiety, and Caloric Intake in Overweight and Obese Women. *J Obes*. 2012; 2012:407103. [PubMed: 22778917]
- Peterson CB, Thuras P, Ackard DM, Mitchell JE, Berg K, Sandager N, Wonderlich SA, Pederson MW, Crow SJ. Personality dimensions in bulimia nervosa, binge eating disorder, and obesity. *Compr Psychiatry*. 2010; 51:31–36. [PubMed: 19932823]
- Puhl MD, Cason AM, Wojnicki FH, Corwin RL, Grigson PS. A history of bingeing on fat enhances cocaine seeking and taking. *Behav Neurosci*. 2011; 125:930–942. [PubMed: 21988520]
- Richardson NR, Roberts DC. Progressive ratio schedules in drug self-administration studies in rats: a method to evaluate reinforcing efficacy. *J Neurosci Methods*. 1996; 66:1–11. [PubMed: 8794935]
- Rose J, Otto T, Dittrich L. The Biopsychology-Toolbox: a free, open-source Matlab-toolbox for the control of behavioral experiments. *J Neurosci Methods*. 2008; 175:104–107. [PubMed: 18765252]
- Rossetti C, Spena G, Halfon O, Boutrel B. Evidence for a compulsive-like behavior in rats exposed to alternate access to highly preferred palatable food. *Addict Biol*. 2013 n/a–n/a.
- Schneider M. Bulimia nervosa and binge-eating disorder in adolescents. *Adolesc Med Phila Pa*. 2003; 14:119–131.
- Specker SM, Lac ST, Carroll ME. Food deprivation history and cocaine self-administration: an animal model of binge eating. *Pharmacol Biochem Behav*. 1994; 48:1025–1029.

- Spigelman G, Spigelman A, Engleson I. Hostility, aggression, and anxiety levels of divorce and nondivorce children in response to projective tests. *J Pers Assess.* 1991; 56:438–452. [PubMed: 1865304]
- Stice E. Clinical implications of psychosocial research on bulimia nervosa and binge-eating disorder. *J Clin Psychol.* 1999; 55:675–683. [PubMed: 10445859]
- Szechtman H, Eckert MJ, Tse WS, Boersma JT, Bonura CA, McClelland JZ, Culver KE, Eilam D. Compulsive checking behavior of quinpirole-sensitized rats as an animal model of Obsessive-Compulsive Disorder (OCD): form and control. *BMC Neurosci.* 2001; 2:4. [PubMed: 11316464]
- Taylor S. Early versus late onset obsessive-compulsive disorder: evidence for distinct subtypes. *Clin Psychol Rev.* 2011; 31:1083–1100. [PubMed: 21820387]
- Tomasi D, Volkow ND. Striatocortical pathway dysfunction in addiction and obesity: differences and similarities. *Crit Rev Biochem Mol Biol.* 2013; 48:1–19. [PubMed: 23173916]
- Vogel G, Neill D, Hagler M, Kors D, Hartley P. Decreased intracranial self-stimulation in a new animal model of endogenous depression. *Neurosci Biobehav Rev.* 1990a; 14:65–68. [PubMed: 2325941]
- Vogel G, Neill D, Hagler M, Kors D. A new animal model of endogenous depression: A summary of present findings. *Neurosci Biobehav Rev.* 1990b; 14:85–91. [PubMed: 2183099]
- Volkow ND, Wang G-J, Fowler JS, Telang F. Overlapping neuronal circuits in addiction and obesity: evidence of systems pathology. *Philos Trans R Soc B Biol Sci.* 2008; 363:3191–3200.
- Walker QD, Nelson CJ, Smith D, Kuhn CM. Vaginal lavage attenuates cocaine-stimulated activity and establishes place preferences in rats. *Pharmacol Biochem Behav.* 2002; 73:743–752. [PubMed: 12213518]
- Weatherly JN, Arthur EL, Tischart LM. Alterating "motivational" variables alters induction produced by up-coming food pellet reinforcement. *Animal Cogn.* 2003; 6:17–26.
- Yannielli PC, Kargieman L, Gregoretti L, Cardinali DP. Effects of Neonatal Clomipramine Treatment on Locomotor Activity, Anxiety-Related Behavior and Serotonin Turnover in Syrian Hamsters. *Neuropsychobiology.* 1999; 39:200–206. [PubMed: 10343185]

Fig. 1.

Spontaneous alternation in males (a) and females (b) across age. A significant decrease in alternations can be seen in CMI pre-exposed rats (black bars) compared to vehicles (white bars) in both sexes. Means $\pm$ SEM are presented. * $p < 0.05$

Fig. 2.

Anxiety-like behavior tested in marble burying in males across age (a; CMI n=7, 13, and 11 and controls n=8, 11, and 13) and females (b; CMI n=6, 8, and 10 vehicle; n=6, 7, and 8). CMI pre-exposed adult males (black bars in a) bury more marbles than control animals (white bars). Means $\pm$ SEM are presented. *p < 0.05

Fig. 3.

Anxiety-like behavior in the open arm of the elevated plus maze in males (a; CMI n=7, 6, 12 and controls: n=7, 10, 9) and females (b; CMI n=7, 8, 13 and vehicle n= 7, 7, 10) and the number of arm entries in males (c) and females (d). Following an Age × Sex × Drug interaction, an overall Drug effect was observed in males, where CMI males spent less time in open compared to controls. Females were only affected by CMI pre-exposure in adulthood. Means ± SEM are presented. *p < 0.05

Fig. 4.

Sucrose consumption on an FR3 (a+b) and a PR (c+d) schedule of reinforcement. (a) On an FR3 schedule, adolescent males that were pre-exposed to CMI (black bars) consume less sucrose pellets compared to vehicle (white bars). (b) Adolescent females consume more sucrose pellets when pre-exposed with CMI compared to vehicle animals. Female animals (d) have a higher breakpoint in the PR schedule compared to males (c). CMI treated and vehicle animals do not differ in the PR schedule. Means $\pm$ SEM are presented. * $p < 0.05$

Fig. 5.

Fat bingeing (% change in g/kg consumption; normalized to weight after 2 weeks) is increased in CMI treated animals (filled boxes) relative to vehicle treated animals (open boxes). One hour access to Crisco three days a week (M, W, F) resulted in progressively more fat intake that was significantly different the last three days of assessment. Normalized means $\pm$ SEM are presented * $p < 0.05$ Bonferroni corrected.

Fig. 6.

Weight of males (squares) and females (circles) across the fat bingeing paradigm. CMI treated animals are filled symbols; vehicles are open symbols. Weight was sampled each Monday. Weight increased across days of assessment and differed between males and females. Means $\pm$ SE presented.

Fig. 7.

Histogram illustrating the number of subjects in each condition who consumed grams of chow intake across the 5 weeks normalized to their individual body weights in kg. The line represents the upper tertile (25%) of consumption of chow. The three CMI males in this upper tertile maintained binge behavior at this level for the duration of the experiment.