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Dynamic body weight and body composition changes in response to subordination stress

Kellie L. K. Tamashiro, Maria A. Hegeman, Mary M. N. Nguyen, Susan J. Melhorn, Li Yun Ma, Stephen C. Woods, and Randall R. Sakai

Department of Psychiatry and Graduate Program in Neuroscience, University of Cincinnati, Cincinnati, OH 45237, U.S.A.

Abstract

Social stress is prevalent in many facets of modern society. Epidemiological data suggest that stress is linked to the development of overweight, obesity and metabolic disease. Although there are strong associations between the incidence of obesity with stress and elevated levels of hormones such as cortisol, there are limited animal models to allow investigation of the etiology of increased adiposity resulting from exposure to stress. Perhaps more importantly, an animal model that mirrors the consequences of stress in humans will provide a vehicle to develop rational clinical therapy to treat or prevent adverse outcomes from exposure to chronic social stress. In the visible burrow system (VBS) model of chronic social stress mixed gender colonies are housed for 2 week periods during which male rats of the colony quickly develop a dominance hierarchy. We found that social stress has significant effects on body weight and body composition such that subordinate rats progressively develop characteristics of obesity that occurs, in part, through neuroendocrine alterations and changes in food intake amount. Although SUB are hyperphagic following social stress they do not increase their intake of sucrose solution as CON and DOM do suggesting that they are anhedonic. Consumption of a high fat diet does not appear to affect development of a social hierarchy and appears to enhance the effect that chronic stress has on body composition. The visible burrow system (VBS) model of social stress may be a potential laboratory model for studying stress-associated metabolic disease, including the metabolic syndrome.

Keywords

Dominance; hierarchy; social stress; obesity; food intake; high fat diet; anhedonia

Introduction

There is no question that obesity is a growing public health concern worldwide. Recent results from the NHANES studies show that the prevalence of obesity in the United States has greatly increased over the past 20 years. Using a BMI (body mass index) of 30 or greater as the criterion for obesity, the prevalence has gone from roughly 15% in 1980 to greater than 30% in 2000

Corresponding author: Randall R. Sakai, Ph.D. Department of Psychiatry – North Genome Research Institute University of Cincinnati 2170 E. Galbraith Road, E-212 Cincinnati, OH 45237 U.S.A. Phone: (513) 558-6589 Fax: (513) 558-5783 Email: randall.sakai@uc.edu. K.L.K.T. current address: Department of Psychiatry and Behavioral Sciences, Johns Hopkins University, Baltimore, MD 21205, U.S.A. M.A.H. current address: Laboratory of PsychoNeuroImmunology, University Medical Center Utrecht (UMCU), Wilhelmina's Children Hospital, Utrecht, The Netherlands.

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[1]. The increase in the prevalence of obesity has also increased the incidence of metabolic abnormalities including glucose intolerance (type 2 diabetes, impaired glucose tolerance, or impaired fasting glycemia), insulin resistance, central or abdominal obesity, dyslipidemia and hypertension [2]. These conditions are well-documented risk factors for cardiovascular disease. Estimated annual obesity-related health expenditures amounted to \$90 billion [3] and resulted in over 39 million lost work days per year [4]. It is evident that overweight and obesity affect far more than the afflicted individuals themselves and are significant public health problems. Efforts should be made to determine the reasons for the continued increases in affected individuals as well as to provide information about alleviating or even preventing the condition from occurring.

Reasons cited for the significant increases in the numbers of overweight or obese people in modern society include increased energy consumption and decreased physical activity. Additionally, daily stress arising from work and social situations interact to create an environment that is conducive to development of obesity and related metabolic disease. In humans, psychosocial stress activates the hypothalamic-pituitary-adrenal (HPA) axis causing hypersecretion of cortisol, and this in turn has been significantly correlated with hyperinsulinemia, hyperglycemia, dyslipidemia, hypertension, depression, osteoporosis and increased abdominal obesity [5-7].

Stress is well known to produce changes in body weight in animal models as well [8-16]. In non-human primates, exposure to psychosocial stress results in reduced negative feedback regulation of cortisol secretion, suppression of the hypothalamic-pituitary-gonadal (HPG) axis and reproduction, and increased depressive behavior [17,18]. In those studies, visceral obesity, insulin resistance, dyslipidemia, hypertension and coronary artery atherosclerosis were also associated with social stress [17,18]. These results are strikingly similar to those found in humans subjected to psychosocial stress and socioeconomic subordination with associated visceral obesity and metabolic disturbances.

Chronic stress (by social defeat) in subordinate tree shrews results in weight loss that has been attributed to stress-induced enhancement of metabolic activity [19,20] and to a lesser extent to reduced food intake [21]. In rats, social defeat results in decreased body weight gain following acute social defeat as well as decreased food intake that persists for several days after the defeat session [22]. In studies using restraint stress, rats lose weight and remain hypophagic for several days after the stressor has ended [23]. In contrast, rats that are food-restricted to match the lower body weight of animals subjected to restraint stress immediately become hyperphagic when allowed *ad lib* access to food suggesting that stress produces a persistent effect on food intake that extends beyond the actual stress period [23]. However, these animal models of social stress (social defeat and restraint stress) may result in different stress-induced changes compared to those elicited by social stress related to dominance hierarchy formation and maintenance [24]. In this review we will discuss stress associated changes in body weight and food intake using an animal model of social stress, the visible burrow system (VBS), and will discuss its implications and possible future directions.

The visible burrow system (VBS) model of social stress

The visible burrow system (VBS) was initially developed over 20 years ago as a neuroethological animal model of social stress to examine agonistic behaviors in rats [25,26]. More recently, we have used the VBS model to examine the effects of social dominance and subordination on the neuroendocrine systems regulating the stress response, reproduction and energy homeostasis [10,24,27-29]. The housing environment in the VBS closely represents the conditions under which rats would live in the wild and serves to facilitate expression of behavioral and physiological responses that would normally occur in that environment.

Animals compete for resources such as reproductive partners, rest areas, food and water. Food and water are provided throughout the apparatus in three areas (open surface area and two smaller chambers), but animals must still gain access to food hoppers and/or water bottles that may be guarded by one or more of the other five animals of the colony.

The VBS is a unique model since the stress imposed is derived from the natural conspecific interactions among the animals themselves and does not require excessive experimenter intervention. Certainly this does not imply that the resident-intruder or social defeat models that do involve some degree of intervention by the investigator are not applicable to the study of social stress. Rather, those models present specific situations of defeat at predetermined times arranged by the investigator, and as such the resultant behavioral and physiological responses are likely to be specific to social defeat in a contextual and temporal specific manner. In contrast, the stress imposed in the VBS is unpredictable and is based on the behavioral intensity of the social interactions of the males competing for access to the females in the colony.

The VBS apparatus has been described previously [10,28,30]. Briefly, it is constructed of opaque black Lexan and consists of a large open (i.e., uncovered) surface area (85 × 65 cm) connected to a series of covered (with transparent Lexan) tunnels and chambers. The open surface area plus each of the two other chambers contain ad lib food and water. The VBS apparatuses in our lab have been modified to incorporate a computerized food intake monitoring system which we co-developed with AccuScan Instruments, Inc. (Columbus, OH). An AccuDiet food monitor (AccuScan Instruments, Inc., Columbus, OH) is attached to each of the 3 feeding stations within a VBS. Individual animals are implanted with a subcutaneous microchip transponder that is read by scanners at each feeding station. This sophisticated system records the duration and quantity of each meal for each of the individual animals in the VBS. Food intake and identification information are then integrated using custom designed software (DietMax software v.1.7, AccuScan Instruments, Inc.) to provide a detailed food intake profile (duration and quantity of each meal) for each animal. A digital video camera with an infrared light source is mounted above each VBS and is connected to a computerized digital video recorder to allow behavioral monitoring of all animals in the VBS.

In a typical study, and in the specific studies discussed in this manuscript, 4 male and 2 female rats are group-housed in a VBS for 14 days. Control males are pair-housed with single females in conventional tub cages. A dominance hierarchy quickly forms among the males of the VBS colonies, within 3-4 days, resulting in one dominant (DOM) and three subordinate (SUB) males per colony. SUB rats show behavioral, physiological, and neuroendocrine alterations consistent with severe stress when compared with other models of stress (reviewed in [24,30]). We present a brief summary of these stress-induced changes in the next section.

During VBS housing, DOM show more offensive or aggressive behavior while SUB display primarily defensive or submissive behaviors [31,32]. Reproductive behavior and social interactions are altered in SUB after social stress [10,31]. Following 14 days of social stress in the VBS, SUB exhibit a marked elevation in basal corticosterone secretion [10,27,28,33, 34] in concert with lower corticotrophin binding globulin levels [35] compared to DOM suggesting that SUB have an overall higher level of free or biologically active glucocorticoid levels. DOM also have slightly elevated basal corticosterone levels, although not to the extent of SUB, and this is likely due to some degree of stress from his efforts to maintain DOM status. SUB have decreased plasma testosterone [10,28,29] and both DOM and SUB animals show thymus involution and adrenal hypertrophy [10,28], compared to the CON further suggesting that elevated glucocorticoids in both social groups have affected peripheral organs as well. All of these parameters indicate that both DOM and SUB indeed experience stress in the VBS. In addition, SUB animals develop enlarged spleens (possibly attributable to increased wounding)

and have decreased testes weights [10,28,29]. Body weight loss among SUB is one of the most consistent consequences of social stress in the VBS [10,27,28,34,35] and this will be discussed in further detail in this manuscript.

Social stress in the VBS produces a variety of changes in neuropeptide and neurochemical systems including altered corticotrophin releasing hormone (CRH) and vasopressin expression [36] and glucocorticoid and mineralocorticoid receptor expression in hippocampus [37]. DOM and SUB also show changes in the serotonin (5-HT) [33,34,38] and dopamine systems [38, 39]. Neuronal morphology in the hippocampus of DOM and SUB is also significantly affected by social stress [34].

Potential predictors of social status

Given the multitude of behavioral, endocrine and physiological changes resulting from social stress, an interesting question that may be asked even before a social stress experiment starts relates to the possible predictors of dominance. That is, is there any characteristic that predisposes an animal to become DOM or SUB? Any consistent predictor may then allow for better management or prevention of the negative consequences of social stress before it occurs.

Higher body weight can bias an individual to become DOM and this occurrence is evident and capitalized upon in the social defeat models of social stress where the “resident” animal is specifically selected with higher body weight and aggression to give him the upper hand in becoming DOM. This body weight bias is also apparent in colony models where it can serve as a predicting factor in determining dominance [40]. Because of this, animals used in the VBS are typically weight-matched before the colony is formed such that no individual animal has a significant weight advantage thereby allowing us to consider other factors in determining dominance and subordination.

All animals used in VBS studies are naïve and this precludes the necessity to consider the influence of previous experience on behavior and hierarchy formation. However, it is important to keep in mind that it is possible that preexisting conditions or characteristics could predispose some animals to become DOM or SUB in the VBS. Although the literature suggests that certain endocrine parameters, for example high plasma testosterone levels, may predispose an animal to express higher levels of aggression and become DOM, this is not universally the case. Likewise there is no effect of pre-grouping on the basis of the response to a dexamethasone challenge, plasma cortisol, or plasma testosterone in primates [40-42]. However, in the Common marmoset (*Callithrix jacchus*) cortisol concentrations in females prior to group formation are predictive of social status. Those females with low morning cortisol concentrations are more likely to become DOM upon group formation in a laboratory model. Once the group stabilizes, cortisol concentrations in DOM are similar to that in SUB [43]. These data suggest that basal corticosterone or testosterone may be predictive of dominance in a VBS colony. However, post hoc analysis of basal corticosterone and testosterone measured in naïve male rats prior to VBS colony formation did not differ based on eventual social status (Table 1).

We assessed plasma leptin after a 4 hr fast during the light cycle and fasted insulin and glucose levels following 16 hr food deprivation as these parameters may indicate a predisposition to metabolic disease and interactions with the stress system may serve to exacerbate or accelerate the appearance of those conditions in SUB. There were no differences among the groups according to social status (Table 1). Furthermore, we found no difference among the social groups in glucose clearance or insulin secretion in an oral glucose tolerance test administered prior to colony formation (data not shown). Other measures related to energy homeostasis such as body composition and susceptibility to weight gain on a high fat diet were also examined and are discussed in the context of the studies summarized in the remainder of this manuscript.

Stress-induced body weight and body composition changes

One of the most profound changes resulting from social subordination is decreased body weight. While DOM animals maintain their original body weight and control animals gain weight, SUB animals consistently show a rapid and sustained weight loss of up to 10-15% of their body weight that occurs over 2 weeks in the VBS [10,27,28,34,35] (Figure 1). Body composition of all animals was determined by NMR (EchoMRI, Waco, TX) prior to VBS colony formation (Day 0) and at the end of VBS housing (Day 13). These data are depicted in Figure 2. There was no significant difference in body composition prior to VBS housing suggesting that body composition prior to VBS colony formation is not a reliable predictor of social status. Plasma testosterone is inversely related to adipose tissue mass and influences fat distribution [44,45]. In addition, higher testosterone levels influence behavior and intermale aggression in rats [46,47]. These observations then suggest that perhaps leaner rats that have higher testosterone levels would be predisposed to becoming DOM of the colony. However, our data are to the contrary and suggest that neither plasma testosterone nor body composition can determine dominance in this model of social stress.

Figure 2 additionally shows that weight loss in SUB during social stress was attributable to loss of adipose tissue and maintenance of lean tissue, while DOM only lost adipose tissue. DOM did not lose weight but had decreased adiposity, suggesting that their body composition changed during social stress, shifting body weight from adipose to lean tissue.

We further analyzed adipose tissue distribution in VBS animals by separating subcutaneous fat (including the dorsosubcutaneous and inguinal fat pads) from visceral fat depots within the carcass (including retroperitoneal, perirenal, mesenteric and epididymal fat pads) using the method of Clegg, et al. [48]. Although SUB lost adipose tissue during VBS stress, they retained a higher percentage of visceral fat than CON and DOM by the end of VBS housing (Figure 3).

As shown in Table 2, DOM and SUB both had lower levels of the adiposity hormone leptin, consistent with the decreased amount of adipose tissue [49]. There was a significant correlation between these two parameters, providing a degree of cross-validity. Insulin was lower in SUB than in DOM and CON, consistent with previous reports [34]. Our data also suggest that body weight loss results from social stress associated with dominance hierarchy formation, and not merely from being housed in an “enriched environment,” since colonies composed of 4 males without females do not form hierarchies and do not display the physiological or body weight changes as with DOM and SUB in mixed-gender colonies, yet appear to be comparably active although activity levels have not been directly measured [28].

There are several possible mechanisms that may account for decreased weight in SUB. Other investigators report that chronic stress in subordinate tree shrews results in weight loss that has been attributed to stress-induced enhancement of metabolic activity [19,20] and to a lesser extent to reduced food intake [21]. In studies using restraint stress, rats lose weight and remain hypophagic until a few days after the stressor has ended [23]. Our next series of studies was aimed at determining whether the weight loss seen in SUB is the result of decreased food intake.

As described earlier, the VBSs in our laboratory are fitted with food intake monitors and microchip identification systems (AccuDiet ID system, AccuScan Instruments, Columbus, OH) that uses subcutaneous microchip identification technology to identify each animal while simultaneously recording the time and amount of food eaten. Using this system we determined that SUB lose body weight because they eat significantly less food compared to DOM beginning immediately upon entering the VBS and continuing throughout the 14 days of housing in the VBS. Thus, body weight loss in SUB can be attributed, at least in part, to decreased food intake in the VBS. Whether decreased food intake was the sole reason for body weight loss in SUB remains to be determined through implementation of an additional control

group, animals that are pair-fed to match the food intake of the SUB group. Energy expenditure may also play a significant role in weight loss in SUB and this is currently being examined in the lab.

Our behavioral studies indicate that DOM spent a majority of their time in the open surface area whereas SUB spent most time in the inner chambers and tunnels. It is important to note that SUB have adequate access to food and water even if they spend most of their time in the side chambers. Consistent with this, food intake data for individual colonies indicate that the DOM takes the majority of his meals in the open surface area while SUB eat their meals primarily in the inner chambers. Total food intake was higher for DOM compared to SUB, and the DOM did not take any meals in the inner chambers, suggesting that SUB did not decrease their intake as a result of the presence or threat of the DOM. Thus, these data suggest that although SUB have adequate access to food, they are nonetheless suppressing their food intake [50]. There may be additional differences in meal patterns among SUB rats that also impact body weight and body composition and these studies are currently ongoing in our laboratory [51,52].

In addition to determining how stress alters body weight and body composition in DOM and SUB during social housing in the VBS, it is also important to determine the long term consequences of stress on these parameters. After 14 days of social stress, SUB had elevated corticosterone and decreased testosterone compared to CON and DOM (Table 2), suggesting that SUB would be predisposed to regain body weight as adipose tissue rather than lean body mass [6]. When SUB were removed from their VBS colonies and were allowed to recover in individual cages ("recovery" period), they regained body weight quickly and by the end of 3 weeks their body weight was only slightly lower than that of CON and DOM [24](Figure 4). SUB were immediately hyperphagic during the recovery period and re-gained body weight primarily as adipose tissue although they were on a standard chow diet that is low in fat content (Harlan Teklad 7012, approximately 5.67% fat). This change in body composition was maintained or increased following a second cycle of VBS social stress and recovery [53]. Consistent with increased body fat, SUB were hyperinsulinemic and hyperleptinemic compared to DOM and CON after recovery [53]. Together, these data suggest that the endocrine and metabolic status in SUB animals following chronic social stress (i.e. high corticosterone and low testosterone) may have increased SUB susceptibility to regain body weight primarily as fat and to preferentially deposit fat viscerally rather than subcutaneously [6]. By separating subcutaneous fat (including the dorsosubcutaneous and inguinal fat pads) from visceral fat depots within the carcass (including retroperitoneal, perirenal, mesenteric and epididymal fat pads) we determined that indeed, SUB had a higher proportion of adipose tissue in the visceral fat depot compared to CON and DOM after recovery [53]. Thus, following multiple cycles of social stress and recovery, SUB accumulate more adipose tissue primarily in the visceral depot suggesting that cumulative effects of chronic social stress may result in the development of symptoms related to the metabolic syndrome.

We further determined that changes in body weight and body composition are indeed attributable to social stress in the VBS. Male rats that are food restricted to match the body weight changes of SUB ("BW MATCH") also lose a significant portion of adipose tissue and lean body mass such that their body composition is similar to that of SUB at this time point. However, BW MATCH rats did not have any significant differences in corticosterone or testosterone in response to food restriction. After 14 days, when allowed *ad libitum* access to chow, BW MATCH rats were hyperphagic similar to SUB, but they regained body weight as adipose as well as lean tissue such that their ultimate body composition did not differ significantly from that of non-restricted controls. These data suggest that changes in body composition in SUB following repeated exposures to stress and recovery cannot be attributable to weight cycling alone [54].

As mentioned earlier, we also included a second control group which consisted of VBS colonies composed of 4 males without females (“MALE ONLY”). In previous studies we determined that the males in all-male colonies do not form dominance hierarchies as mixed gender colonies do [28], but are housed in the VBS which provides a larger, more complex living area compared to conventional shoebox cages in which rats are normally housed. There was no evidence of a dominance hierarchy among the rats in MALE ONLY colonies and although all rats maintained or lost a small amount of weight compared to individually housed controls, the difference was not significant. Body composition of rats in MALE ONLY colonies resembled that of DOM in mixed gender colonies. That is, they lost adipose tissue and maintained or increased lean body mass compared to singly housed control males. These findings suggest that body composition changes seen in DOM may be attributed in part to increased activity in the VBS. In contrast, this further suggests that body weight loss followed by increased adiposity after 2 cycles of social stress and recovery in SUB is attributable to stress derived from social subordination and not to exposure to an “enriched environment” [54].

In humans, a recent study of young healthy men exposed to long-term stress revealed that long-term stress results in increased abdominal obesity and early signs of metabolic syndrome suggesting that stress plays an important role in the genesis of metabolic abnormalities [55]. In that study, subjects lost both fat and lean body mass during the first stressful episode and subsequently regained body weight as fat resulting in an overall decrease in protein mass. This is similar to SUB body weight and body composition fluctuations through cycles of social stress in the VBS and subsequent recovery further suggesting that the VBS model may provide a means to study the development of stress-induced metabolic disorders.

Sucrose Consumption Tests

Several reports suggest that stress can cause depressive-like symptoms in rodents, including anhedonia. A common measure of anhedonia in rodents is consumption of dilute sucrose solution [56]. Both decreases [56-58] and increases [58] in sucrose or saccharin solution consumption have been reported following stress, while other studies find no effect [58-60]. Although measures of anhedonia in rodents remain a topic of controversy [60,61], decreased sucrose solution consumption is generally considered to be indicative of anhedonia in rats and mice [56].

We measured consumption of 1% sucrose solution during the recovery period one and two weeks after VBS housing for 2 cycles of VBS stress and recovery. All animals were given access to a 1% sucrose solution in 25 mL sipper tubes. The test was administered in the middle of their light cycle (1300 h of a 12:12 L:D cycle with light onset at 0600 h) without food or water deprivation. Food and water were removed from the cages only during the test period and consumption of sucrose solution was measured hourly for 3 hours. SUB drank significantly less than CON and DOM (Figure 5) suggesting that they exhibit anhedonia following social stress and that this behavior persists through 2 weeks of recovery outside of the VBS. Male rats that were food restricted to match the body weight of SUB (BW MATCH) through 2 cycles of VBS and recovery did not differ in their sucrose intake when compared to *ad lib* fed control rats (data not shown). These findings suggest that decreased intake of sucrose solution by SUB is not attributable to weight cycling alone.

Further, while CON and DOM increased their intakes with repeated, weekly access to the sucrose solution SUB intakes remained at approximately the same amount. Benoit et al. have suggested that rodents normally increase their sucrose ingestion across time and that this may be a “learned response” [62]. Our data then suggest SUB may have impaired “learning” abilities. Although social stress has been found to elicit significant effects on hippocampal

dendritic morphology in DOM and SUB [34], learning and memory have not yet been assessed in VBS-housed animals.

Finally, it appears that SUB did not recognize the sucrose solution as a source of calories despite being hyperphagic on rodent chow. Other investigators find that rats subjected to chronic mild stress drink less 1% sucrose solution than non-stressed controls following water deprivation, although water intake is similar to controls [63]. Further, stress rats show a similar reduction in intake of a non-caloric saccharin solution [64] while their food intake is normal or elevated [63]. These data suggest that decreased intake of sucrose solution may be unrelated to its caloric content. This remains to be ascertained in SUB in the VBS model.

High fat diet in the VBS

The animals in all of our studies described above were maintained on a standard chow diet. Despite the relatively low percentage of dietary fat, our data suggest that chronic stress leads to an altered metabolic state; one that is consistent with those of humans that develop symptoms of the metabolic syndrome. Because consumption of a HF diet in the absence of psychosocial stress also predisposes rats to obesity, glucose intolerance and other symptoms of the metabolic syndrome, we assessed the effect of consuming a HF diet in the VBS paradigm. Animals were maintained on a HF diet (Research Diets, 40% kcal dietary fat) for 3 weeks prior to entering the VBS colony, and this continued while in the VBS and recovery.

There are conflicting reports about the stress responsiveness of rats maintained on HF diet [65-68] and the differences may be accounted for by the variety of animal models and stress paradigms used. HF animals in our pilot study had an enhanced corticosterone response to 1-hour restraint stress prior to colony formation, but they subsequently had typical behavioral responses to VBS. Specifically, the dominance hierarchy was established within a few days of colony formation and aggressive and defensive behaviors exhibited by DOM and SUB animals were comparable in number and intensity (as assessed by number of bite wounds) as those in chow-fed VBS colonies. Furthermore, consumption of HF diet prior to and during social stress did not prevent significant weight loss by SUB. Thus, in the VBS paradigm, maintenance on a HF diet did not significantly affect social behavior and hierarchy formation and makes this a useful model in examining the physiological and metabolic consequences of social stress using diets of varying fat composition.

Interestingly, post hoc analysis revealed that susceptibility to gain weight on the HF diet was a predictor of subsequent dominance formation. That is, the animals that gained the least amount of adipose tissue while on HF diet prior to being placed in the VBS became DOM while animals that gained more adipose weight on the HF diet became SUB. Taken together, these preliminary data also suggest that adipose tissue gain on a HF-diet prior to colony formation may be a predictor of social status. As discussed earlier, higher testosterone levels may influence behavior and aggression in male rats and plasma testosterone has an inverse relationship with adipose tissue mass and influences fat distribution [44,45]. Based on this, leaner rats that have higher testosterone levels may be predisposed to becoming DOM. However, in this study there was no significant correlation between the amount of adipose mass and plasma testosterone. DOM did not have higher testosterone than SUB prior to colony formation similar to findings in animals fed standard chow diet. Thus, it is unlikely that lean rats had higher testosterone prior to VBS exposure which predisposed them to becoming DOM and an alternative mechanism is likely to be responsible [69,70].

Stress can affect the consumption of “palatable” foods (i.e. foods that are calorically dense and have high amounts of carbohydrates or fat or both) such that it decreases total food intake on standard chow, but increases consumption of more “palatable” foods thus yielding visceral obesity. It has been suggested that individuals consume more palatable food during stress in

an attempt to reduce the negative effects of a chronic stressor [71]. Whether this is true in SUB has not been determined. However, our data show that SUB had decreased food intake even when maintained on HF diet in the VBS. Although our sucrose consumption data suggest that SUB do not increase their consumption of a carbohydrate solution, this behavior may be specific to the solution's macronutrient composition. It may be possible that SUB behavior will change if they are presented with diets composed of different macronutrients to consume, such as a high fat (HF) diet, or if they are provided with a choice of macronutrients. It has been suggested that having a choice of which macronutrient to consume can play a significant role in macronutrient influences on the negative effects of stress [68]. These are interesting considerations that warrants future study.

Concluding remarks

The VBS model of social stress was initially developed as a neuroethological animal model to study agonistic behavior [26]. The experiments discussed in this review replicate and extend the initial findings in the VBS model. The data summarized here are the first to document the effects of chronic social stress in the VBS model on behavioral, physiological and endocrine parameters of energy homeostasis. Our examination of potential predictors of social status did not reveal any significant differences according to social status and thus suggest that social stress derived from social subordination is likely to be the primary cause of physiological and endocrine differences between DOM and SUB following colony housing. We have also begun to examine the long-term consequences of stress on body weight and body composition in SUB. Some alterations persist even after animals have been allowed to recover for a significant amount of time.

VBS SUB have increased visceral adiposity that appears to be enhanced with repeated chronic stress challenge. Visceral adiposity is a significant risk factor in the development of other symptoms of the metabolic syndrome. The metabolic syndrome, a clustering of metabolic abnormalities such as insulin resistance, hyperlipidemia, and hypertension, has been associated with stress and chronic activation of the HPA axis. Whether these symptoms will eventually be manifest in VBS SUB remains to be determined. Our data suggest that these disease states may ensue either with additional social stress challenges or if SUB are permitted to continue in the metabolic state that they end up in following 2 cycles of VBS stress and recovery. Conversely, if SUB animals do not progress to developing symptoms of the metabolic syndrome, it will be important to determine why this is so given that changes in their body composition and endocrine status strongly suggest that they would be susceptible. Finally, we are assessing brain expression of neuropeptides and neurochemicals that are known to be involved in the regulation of energy homeostasis to determine how these are altered by social stress to influence body weight and body composition.

Although the consequences of the metabolic syndrome are well known, the mechanisms contributing to its development are not [2]. Genetic animal models of obesity are available and these display symptoms of the metabolic syndrome. However, an animal model, such as the VBS, that more closely represents the way in which humans develop the syndrome would be valuable.

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Figure 1. Body weight during VBS. SUB in colonies lost a significant amount of body weight over 14-days in the VBS compared to DOM. * $P < 0.05$ vs. CON; ** $P < 0.05$ vs. CON and DOM.

Figure 2.

Body composition prior to (Day 0) and on Day 13 of VBS housing measured by NMR. Following 14 days in the VBS, both DOM and SUB had significantly decreased % body fat. SUB did not gain lean body mass as CON and DOM did. DOM did not lose body weight and appears to have shifted fat to lean body mass. * $P < 0.05$ vs. DAY 0; # $P < 0.05$ vs. CON on DAY 13.

Figure 3.
Adipose tissue distribution of VBS rats sacrificed following 14 days in VBS (n = 4 colonies).
* $P < 0.05$ vs. CON.

Figure 4.

Percentage of original body weight over two cycles of chronic social stress in the VBS (14 days each) and recovery (21 days each) in individual cages. SUB rats lost a significant amount of body weight during both VBS 1 and VBS 2, while DOM lost little or maintained their weight and CON continued to gain weight. During the recovery periods, SUB regained lost weight but did not attain the same body weight level as CON. [Adapted from Sakai and Tamashiro, 2005]

Figure 5.

Intake of 1% sucrose solution. CON and DOM significantly increase their intake of sucrose solution with each trial. In contrast SUB consistently have lower sucrose intake than CON and DOM and do not increase their intakes across trials. * $P < 0.05$ vs. CON; ** $P < 0.05$ vs. CON and DOM.

Table 1

Endocrine parameters prior to VBS colony formation.

	Pre-VBS (according to social status determined at the end of 14 days in the VBS)		
	CON	DOM	SUB
Basal corticosterone (ug/dl)	1.95 ± 1.17	2.96 ± 1.82	1.40 ± 0.48
Testosterone (ng/ml)	4.2 ± 1.0	5.2 ± 1.0	4.9 ± 0.9
Leptin (ng/ml)	6.3 ± 0.6	6.9 ± 1.4	5.8 ± 0.6
Fasted insulin (pmol)	94 ± 9	79 ± 11	66 ± 9
Fasted glucose (mg/dl)	113 ± 3	115 ± 4	113 ± 2

Data are presented as mean ± S.E.M; n = 8 colonies (8 CON, 8 DOM, 24 SUB).

There were no significant differences among the groups in any measure.

Table 2

Endocrine parameters following 14 days of social stress in the VBS. Adapted from [28].

	After 14 day VBS		
	CON	DOM	SUB
Testosterone (ng/ml)	4.9 ± 0.6	5.0 ± 1.0	3.0 ± 0.4 [†]
Leptin (ng/ml)	3.7 ± 0.7	1.9 ± 0.5 [*]	0.8 ± 0.2 [†]
Insulin (pmol)	271 ± 36	264 ± 36	170 ± 17 [†]

Data are presented as mean ± S.E.M; n = 12 colonies (12 CON, 12 DOM, 35 SUB).

* $P < 0.05$ vs. CON[†] $P < 0.05$ vs. CON, DOM