Olanzapine Reduces Physical Activity in Rats Exposed to Activity-Based Anorexia: Possible Implications for Treatment of Anorexia Nervosa?

Jacquelien J.G. Hillebrand, Annemarie A. van Elburg, Martien J.H. Kas, Herman van Engeland, and Roger A.H. Adan

Background: *Anorexia nervosa (AN) patients often show extreme hypophagia and excessive physical activity. Activity-based anorexia (ABA) is considered an animal model of AN and mimics food restriction and hyperactivity in rats. This study investigated whether treatment with olanzapine (Zyprexa) reduces the development of ABA in rats. The effect of olanzapine treatment in AN patients was also evaluated in a small open-label study.*

Methods: *Rats were chronically (1 week) infused with olanzapine (7.5 mg/kg) and exposed to the ABA model or ad libitum feeding. Hyperactive AN patients were followed for up to 3 months of olanzapine treatment (5 mg/kg).*

Results: *Olanzapine treatment reduced development of ABA in rats by reducing running wheel activity, starvation-induced hypothermia and activation of the hypothalamus-pituitary-adrenal axis. Olanzapine treatment reduced activity levels of AN patients compared with untreated AN patients, without affecting body weight and plasma leptin levels.*

Conclusions: *Olanzapine treatment reduced wheel running and thereby diminished development of ABA in rats. Olanzapine treatment also reduced physical activity in hyperactive AN patients in a small open-label study. These data support the need for controlled studies investigating the putative beneficial effects of olanzapine treatment in AN patients.*

Key Words: Hyperactivity, anorexia, antipsychotic, food restriction, running wheel

Anorexia nervosa (AN) is a psychiatric disorder that is often characterized by extreme hypophagia, obsessive fears of being fat, and hyperactivity (Casper et al 1991; Davis 1997; Kron et al 1978; Walsh et al 1998). Anorexia nervosa has the highest mortality rate of psychiatric disorders (Sullivan 1995).

Some studies show that serotonin (5-hydroxytryptamine, 5-HT) signaling is altered in ill AN patients as well as in recovered AN patients (Brewerton and Jimerson 1996). For example, AN patients show decreased cerebrospinal fluid (CSF) levels of 5-HT and 5-hydroxyindole acetic acid (5-HIAA) during disease and increased levels of 5-HIAA when recovered (Kaye et al 1991). Studies on the use of serotonin reuptake inhibitors (SSRIs) in malnourished AN patients showed no benefits of treatment (Kaye et al 1998); however, SSRIs seem to be effective in preventing relapse in recovered AN patients (Kaye et al 2001). Genetic association studies showed possible implications of polymorphisms in the 5-HT_{2A} receptor, 5-HT_{2C} receptor, and serotonin transporter in AN (Collier et al 1997; Di Bella et al 2000; Hu et al 2003; Nacmias et al 1999; Westberg et al 2002), although other studies could not confirm these findings (Ando et al 2001; Gorwood et al 2002; Hinney et al 1997; Nishiguchi et al 2001).

Trials on antipsychotic treatment in AN patients have been performed previously, but only to a limited extent (Hoffman and Halmi 1993; Johnson et al 1983; Pederson et al 2003). Controlled studies on chlorpromazine (Dally and Sargant 1966) and pimozide (Vandereycken and Pierloot 1982) showed increased weight gain and a more positive attitude of AN patients toward treatment, whereas the atypical antipsychotic sulpiride did not significantly influence body weight gain and eating and body attitudes in AN patients (Vandereycken 1984).

Olanzapine (Zyprexa) is an atypical antipsychotic with a broad pharmacologic profile. This thienobenzodiazepine compound has high affinity for 5-$HT_{2A/2C}$ receptors, histamine (H1) receptors, and adrenergic (a1) receptors and moderate affinity for dopamine (D_1–D_4) receptors (Bymaster et al 1996, 1997; Moore 1999; Schotte et al 1996). Olanzapine treatment in humans causes limited extrapyramidal effects and has been associated with body weight gain (Allison and Casey 2001). A few (uncontrolled) studies already reported beneficial effects of olanzapine treatment on food intake and anxiety levels of AN patients (Barbarich et al 2004; Boachie et al 2003; Malina et al 2003; Mehler et al 2001; Powers et al 2002), but no study has reported effects of olanzapine treatment on physical activity levels.

Animal models of AN might contribute to the understanding of AN and subsequently improve AN treatment. The activity-based anorexia (ABA) model is used to study anorectic behavior in rodents and serves as an animal model of AN (Routtenberg and Kuznesof 1967). In the ABA model, scheduled feeding in combination with voluntary access to running wheels leads to a paradoxical increase in running wheel activity (RWA) and decrease in food intake (compared with food-restricted control animals; Routtenberg and Kuznesof 1967). This results in activation of the hypothalamus-pituitary-adrenal (HPA) axis and substantial body weight loss. Few studies already reported increased feeding behavior and reduced locomotor activity in rats following acute olanzapine treatment (Prinssen et al 2000; Thornton-Jones et al 2002). These effects suggest that olanzapine treatment might influence pathophysiologic processes in anorexia. Data on chronic (instead of acute) administration of olanzapine in rodents is rare (Pouzet et al 2003), however, although it is necessary for clinical comparisons (Kapur et al 2003).

In this study, the effects of chronic olanzapine treatment on development of ABA in rats as well as the putative positive

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effects of olanzapine treatment on body weight and physical activity in AN patients were investigated.

Materials and Methods

Rats

Female outbred Wistar WU rats (n = 30; Harlan, Horst, The Netherlands) weighing 160 g upon arrival were individually housed in a temperature- and humidity-controlled room (21 ± 2°C) under a 12-hour light–dark cycle (Zeitgeber [ZT]12: lights off). The University of Utrecht ethical committee on use and care of animals approved all described procedures.

Drugs

The olanzapine for the animal studies was kindly provided by Eli Lilly (Indianapolis, Indiana). Olanzapine was dissolved in a minimum quantity of acetic acid, made up to volume with sterile isotonic saline and adjusted to pH 6 with 5 M NaOH, and continuously infused (subcutaneously [sc]) during 1 week at a concentration of 7.5 mg/kg/day using osmotic minipumps (Alzet, model 2001, DURECT, Cupertino, California; Kapur et al 2003). In the human study, AN patients received olanzapine (5 mg–15 mg) tablets (Eli Lilly).

Surgical Procedures Animal Studies

Transmitters (TA10TA-F40 Data Sciences International, St. Paul, Minnesota) were placed in the abdominal cavity under fentanyl/fluanisone (Hypnorm, Janssen Pharmaceutica, Beerse, Belgium, .1 mL/100 g, intramuscular) and midazolam (Dormicum, Hoffman-LaRoche, Mijdrecht, The Netherlands, .05 mL/100 g, intraperitoneal) anesthesia. After surgery, rats were treated with buprenorphin (Temgesic, Schering-Plough, Maarssen, The Netherlands, .05 mL/100 g sc) and saline (1 mL sc) and were allowed to recover for 2 weeks.

For chronic infusions, osmotic minipumps were filled with olanzapine or vehicle and were activated by overnight incubation at 37°C. The pumps were positioned subcutaneously into the flank of the rats under Hypnorm anesthesia at the end of day –1 (ZT10). After surgery, rats were treated with Temgesic and saline as indicated earlier.

Experimental Setup Animal Studies

Olanzapine Treatment in Activity-Based Anorexia. Transmitters were implanted 1 week after arrival of the rats. After 2 weeks of recovery (day –10), the rats (n = 14, synchronized for estrous cycle) were placed in cages with running wheels for adaptation to the running wheel. During this 10-day period, food and water were available ad libitum. RWA was continuously registered using a Cage Registration Program (Department Biomedical Engineering, UMC Utrecht, The Netherlands). At day –2, transmitters were activated for baseline recordings of body temperature. At day –1 (ZT11), rats were divided into two groups, matched for body weight (vehicle: 214.7 ± 2.5 g, olanzapine: 215.0 ± 2.7 g, *ns*) and baseline RWA. Baseline RWA was determined as average RWA during the 4 days before the start of infusion (day –4 to day –1; vehicle: 5281 ± 1409 revolutions, olanzapine: 5694 ± 1496 revolutions, *ns*). Osmotic minipumps containing olanzapine or vehicle were implanted as indicated above. Immediately after surgery, food was removed (day 0, ZT12). The next days (day 1–6), rats had 1 hour access to food (ZT12–ZT13), and water was available ad libitum. Body weight and food intake were measured daily. At day 6 (ZT11), rats were decapitated, and trunk blood was collected into lithium-heparin (Sarstedt, Nümbrecht, Germany) containing tubes with 83 mmol ethylenediamine tetraacetatic acid and 1 mg aprotonin. Plasma was stored at –20°C. Retroperitoneal white adipose tissue (rWAT), interscapular brown adipose tissue (iBAT), and adrenals were collected and weighed.

Olanzapine Treatment in Ad Libitum–Fed Running Rats. This experiment was performed similarly to the previous experiment, with the only difference being that rats had ad libitum access to food throughout the experiment. Sixteen rats were divided into two groups, matched for body weight (vehicle: 236.2 ± 4.7 g, olanzapine: 231.2 ± 5.9 g, *ns*) and baseline RWA (vehicle: 6686 ± 1124 revolutions, olanzapine: 6514 ± 1590 revolutions, *ns*).

Patients

In the open-label trial AN patients were studied in a specialized treatment setting. At their entrance to the hospital, activity levels of AN patients were scored by trained nurses on a scale from 0 to 100 (score 0 = inactive, score 100 = extremely active), which has recently been validated by the use of Actiwatches (manuscript in preparation).

From a cohort of 27 AN patients, 18 patients (67%) displayed activity scores greater than 50 at their entrance to the hospital and were entitled hyperactive and included in this study. Of these 18 female AN patients, 7 received olanzapine treatment (5 mg, except for 1 patient who was treated with 15 mg), and 11 received no medication (age: olanzapine,16.0 ± 1.0 years; no medication, 17.3 ± .7 years, *ns*). Patients were assigned to the olanzapine treatment group because of anxious behavior toward eating and weight gain. All AN patients were free from other forms of pharmacotherapy. Additional treatment was generally the same for all patients and was aimed at body weight gain (.75 kg/week) followed by further normalizing of cognition and body image and treatment of possible comorbid problems. Both groups contained inpatients as well as day-treatment patients (who visited the hospital twice weekly). Every week activity levels were scored. Body weight (z scores to control for age) was measured, and blood was collected once in 2 weeks. All AN patients and parents (in case of minors) gave informed consent for participation in a clinical study, which was approved by the Ethical Committee of the University Medical Center of Utrecht.

Radioimmunoassay

In rats, plasma levels of corticosterone, adrenocorticotropic hormone (ACTH), and leptin were measured by radioimmunoassays (RIA). Plasma levels of corticosterone were measured using a commercially available rat corticosterone RIA kit (ICN Biochemicals, Costa Mesa, California). Plasma ACTH was measured using a specific rabbit antiserum directed to the midportion of ACTH, which was kindly provided by Dr. G.B. Makara (Budapest, Hungary). Synthetic human $ACTH_{(1–39)}$ (Peninsula Laboratories, Belmont, California) was labeled with ^{125}I and used as a tracer (Nijsen et al 2000). Plasma levels of leptin were measured using a commercially available rat leptin RIA kit (detection limit .5 ng/mL), according to the manufacturer's protocol (Linco Research, St. Charles, Missouri). In patients, plasma levels of leptin were analyzed by the DLR-Institute of Aerospace Medicine, Space Physiology in Cologne, Germany using a sensitive human leptin RIA assay (Mediagnost Reutlingen, Germany).

Data Analysis

Data are presented as mean ± standard error. For all measurements (in the animal and human studies), baseline levels

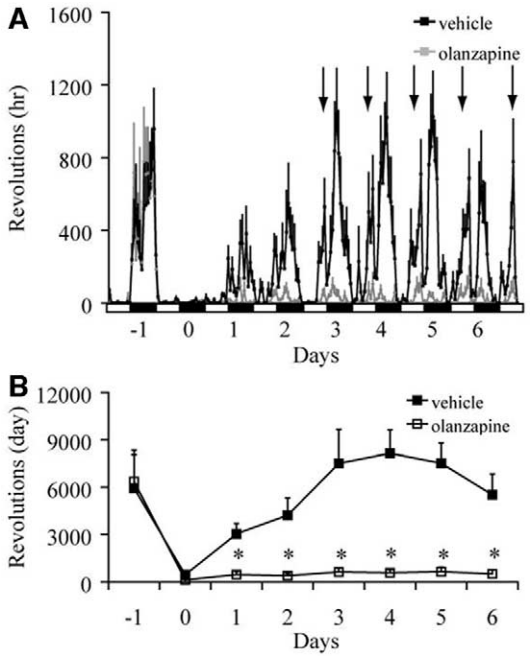

Figure 1. Running wheel activity (RWA) in activity-based anorexia rats. **(A)** RWA per hour in activity-based anorexia (ABA) rats (day –1 [baseline] through day 6) following vehicle (black) or olanzapine (grey) treatment. Arrows indicate food anticipatory activity. **(B)** Total RWA per day (day –1 [baseline] through day 6) of ABA rats following vehicle (closed squares) or olanzapine (open squares) treatment. * $p < .05$, Mann–Whitney *U* test.

were not significantly different between olanzapine- and vehicle-treated (no medication in the human study) groups.

Basal body temperature of rats was defined as mean body temperature during 30 min of inactivity in the early light phase (ZT0–ZT3) measured by telemetric devices. RWA, body weight, food intake, and basal body temperature data were first attributed to repeated measures analysis, using Huynh–Feldt correction for Mauchlys sphericity effects, followed by *t* test or Mann–Whitney *U* test when appropriate. rWAT, iBAT, plasma leptin levels, and HPA axis activation were analyzed by independent *t* tests.

Weekly activity scores of AN patients were averaged to monthly scores (to diminish within-subject variation) and were attributed to repeated measures analysis using Huynh–Feldt correction for Mauchlys sphericity effects, followed by *t* tests. Weekly body weight (*z* scores) and twice-weekly plasma leptin levels were also averaged to monthly scores and analyzed as described earlier. Differences were considered significant at $p < .05$. In two AN patients, olanzapine treatment lasted for 2 months instead of 3 months. Consequently, activity, body weight, and plasma leptin levels in month 3 were analyzed in five (instead of seven) olanzapine-treated patients.

Results

Olanzapine Treatment in Activity-Based Anorexia

Olanzapine treatment significantly decreased running wheel activity (RWA) in ABA rats [day: $F(6,66) = 8.55$; $p = .001$, day × treatment: $F(6,66) = 6.75$; $p = .001$]. Vehicle-treated ABA rats increased RWA following the start of scheduled feeding. Not only did daily revolutions increase, but the distribution of activity changed as well; a substantial part of total activity occurred before the feeding period (food anticipatory activity, FAA). FAA is one of the key phenomena occurring in ABA and (in this experimental setup) took place in the light phase. Olanzapine-treated ABA rats reduced RWA in the dark phase [day: $F(6,66) = 6.02$; $p = .002$, day × treatment: $F(6,66) = 5.23$; $p = .004$] and showed decreased FAA (day: $F(6,66) = 7.44$; $p = .002$), day × treatment: $F(6,66) = 4.68$; $p = .02$; Figure 1).

Table 1. Characteristics of Activity-Based Anorexia Rats Treated with Vehicle or Olanzapine

	Vehicle	Olanzapine
Food (g)	39.7 ± 2.4	39.5 ± 1.4
Body Weight (%)	81.7 ± 1.9	84.3 ± .4
rWAT (mg)	278.6 ± 97.2	404.0 ± 80.6
iBAT (mg)	98.9 ± 6.3	147.0 ± 15.1[a]
ACTH (pg/mL)	167.1 ± 14.4	114.7 ± 4.5[a]
Corticosterone (μg/dL)	41.1 ± 7.4	34.7 ± 5.7
Adrenal Weight (mg)	71.6 ± 3.9	65.6 ± 4.0

Cumulative (day 1– day 6) food intake, relative body weight (% of day −1), retroperitoneal white adipose tissue (rWAT) weight, interscapular brown adipose tissue weight (iBAT), plasma adrenocorticotropic hormone (ACTH) levels, plasma corticosterone levels, and adrenal weight (all analyzed at the end of day 6 [ZT11]) were measured in activity-based anorexia rats treated with vehicle or olanzapine.

[a] $p < .05$, *t* test.

One-hour food intake was not significantly affected by olanzapine treatment over time [day: $F(5,60) = 23.62$; $p = .001$, day × treatment: $F(5,60) = .84$, *ns*]. Olanzapine treatment tended to reduce body weight loss [day: $F(6,72) = 129.45$; $p = .001$, day × treatment: $F(6,72) = 3.64$; $p = .053$], whereas rWAT weight was not affected [$t(12) = –.92$, *ns*; Table 1]. Plasma leptin levels were undetectable (< .5 ng/mL) in vehicle-treated as well as olanzapine-treated ABA rats.

Basal body temperature in ABA rats was higher following olanzapine treatment as compared with vehicle treatment [day: $F(6,60) = 18.02$; $p = .001$, day × treatment: $F(6,60) = 5.43$; $p = .02$; Figure 2], and also iBAT weight was significantly increased [$t(12) = –2.94$; $p = .02$] in olanzapine-treated ABA rats.

Activation of the HPA axis was decreased following olanzapine treatment. Plasma levels of ACTH were significantly reduced [$t(12) = 3.46$; $p = .005$], and corticosterone plasma levels [$t(12) = .68$, *ns*] and adrenal weights [$t(12) = 1.08$, *ns*] were not affected (Table 1).

Thus, treating ABA rats with 7.5 mg/kg olanzapine per day reduced RWA, tended to decrease body weight loss without

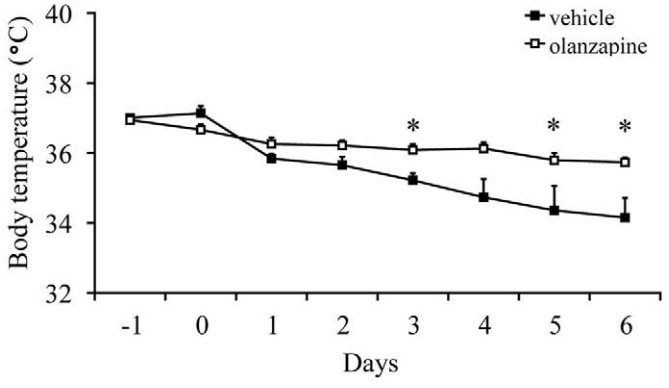

Figure 2. Basal body temperature in activity-based anorexia rats. Basal body temperature of activity-based anorexia (ABA) rats (day –1 [baseline] through day 6) following vehicle (closed squares) or olanzapine (open squares) treatment. *$p < .05$, *t* test.

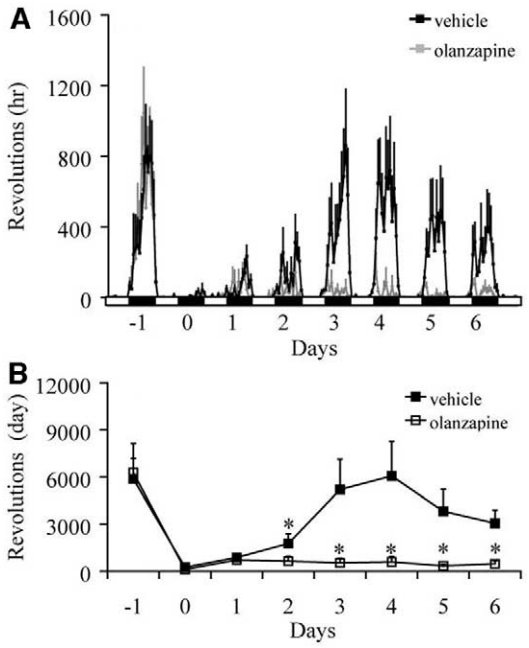

Figure 3. Running wheel activity (RWA) in ad libitum–fed rats. **(A)** RWA per hour in ad libitum–fed rats (day –1 [baseline] through day 6) following vehicle (black) or olanzapine (grey) treatment. **(B)** Total RWA (day –1 [baseline] through day 6) of ad libitum–fed rats following vehicle (closed squares) or olanzapine (open squares) treatment. *$p < .05$, Mann–Whitney *U* test.

significantly affecting food intake, decreased HPA axis activation, and reduced starvation-induced hypothermia. To investigate whether these effects of olanzapine treatment were specific for the ABA model, the experiment was repeated in ad libitum–fed running rats.

Olanzapine Treatment in Ad Libitum–Fed Running Rats

The reduction in RWA described in the previous section was not specific to ABA rats. Olanzapine treatment (7.5 mg/kg/day) also decreased total RWA [day: $F(6,84) = 5.36$; $p = .02$, day × treatment: $F(6,84) = 4.89$, $p = .02$] by reducing dark phase RWA [day: $F(6,84) = 5.38$; $p = .01$, day × treatment: $F(6,84) = 4.86$, $p = .02$] in ad libitum–fed running rats (Figure 3). Light-phase RWA of ad libitum–fed rats was negligible. Olanzapine treatment significantly increased food intake [day: $F(6,84) = 27.67$; $p = .001$, day × treatment: $F(6,84) = 3.84$, $p = .02$] and relative body weight [day: $F(6,84) = 75.08$; $p = .001$, day × treatment: $F(6, 84) = 12.51$, $p = .001$] in ad libitum–fed running rats. Plasma leptin levels [$t(14) = –3.71$ $p = .002$], but not rWAT mass [$t(14) = –.91$, *ns*], were significantly increased in olanzapine-treated ad libitum–fed running rats (Table 2). Basal body temperature as well as iBAT weight were not significantly affected by olanzapine treatment in ad libitum–fed running rats [day: $F(6,84) = 2.44$, *ns*; day × treatment: $F(6,84) = 1.02$, *ns*; Figure 4], [$t(13) = –.68$, *ns*]. Olanzapine treatment did not significantly influence plasma ACTH levels [$t(14) = .48$, *ns*] and plasma corticosterone levels [$t(14) = 1.27$, *ns*], but decreased adrenal weight ($t(14) = 2.94$, $p = .003$] in ad libitum–fed running rats (Table 2).

Olanzapine Treatment in Anorexia Nervosa Patients

Body weight of olanzapine-treated AN patients (*z* score: –3.92 ± .93) as well as plasma leptin levels (plasma leptin: 1.34 ± .40 ng/mL) were not significantly different from AN patients without medication (*z* score: –2.97 ± .36) (plasma leptin: 3.42 ± 1.18 ng/mL) at the onset of the study [$t(16) = 1.11$, *ns*; $t(15) = 1.42$, *ns*]; nevertheless, there were large within-group variations. Physical activity levels of AN patients were also not significantly different between olanzapine-treated AN patients and AN patients without medication at admittance to the hospital [olanzapine treatment: 67.57 ± 7.16; no treatment: 69.23. ± 3.92; $t(16) = .22$, *ns*].

Body weight increased in both patient groups following 3 months of treatment; however, body weight was not significantly different between olanzapine-treated AN patients (*z* score month 3: –2.40 ± .50) and AN patients without medication [*z* score month 3: –2.08 ± .23; month: $F(3,42) = 14.73$; $p = .001$, month × treatment: $F(3,42) = 2.21$, *ns*]. Likewise, plasma leptin levels significantly increased following 3 months of treatment in both groups but were not significantly different between olanzapine-treated AN patients (leptin month 3: 3.51 ± .30 ng/mL) and AN patients without medication [leptin month 3: 4.38 ± .98 ng/mL; month: $F(3,36) = 8.96$; $p = .001$, month × treatment: $F(3,36) = 2.02$, *ns*].

Olanzapine treatment significantly affected activity levels of AN patients [month: $F(3,42) = 11.45$; $p = .001$, month × treatment $F(3,42) = 6.51$; $p = .001$]. Activity levels of AN patients were significantly decreased following 2 and 3 months of olanzapine treatment compared with patients without medication (Figure 5).

Discussion

In this study, it was shown that olanzapine treatment reduces development of ABA in rats. Three important parameters of the ABA model are increased RWA, decreased food intake, and HPA axis activation. Olanzapine treatment greatly reduced RWA in restricted and ad libitum–fed rats with running wheels. Both dark-phase and light-phase activity (including FAA) of ABA rats was reduced. Previous studies have shown that once exposed to scheduled feeding in the ABA model, rats decrease food intake compared with restricted rats without running wheels (Routtenberg and Kuznesof 1967). Thus, rats in the ABA model can be called anorectic. Olanzapine treatment did not influence 1-hour food intake in ABA rats but did increase food intake and body weight gain in ad libitum–fed rats. Hence, it appears that the effects of chronic olanzapine treatment on food intake and body weight are different in ad libitum–fed rats and ABA rats (although this was not investigated in the same experiment), whereas the

Table 2. Characteristics of Ad Libitum–Fed Running Rats Treated with Vehicle or Olanzapine

	Vehicle	Olanzapine
Food (g)	141.0 ± 3.6	153.2 ± 4.6[a]
Body Weight (%)	105.9 ± 1.3	109.4 ± 0.8[a]
rWAT (mg)	2646.7 ± 300.8	3023.3 ± 288.3
iBAT (mg)	230.1 ± 20.6	261.1 ± 38.1
Leptin (ng/mL)	2.48 ± 0.45	5.14 ± .56[a]
ACTH (pg/mL)	132.6 ± 11.2	124.9 ± 11.4
Corticosterone (μg/dL)	32.2 ± 2.7	26.7 ± 3.4
Adrenal Weight (mg)	70.2 ± 2.4	58.6 ± 3.1[a]

Cumulative (day 0 – day 6) food intake, relative body weight (% of day −1), retroperitoneal white adipose tissue (rWAT) weight, interscapular brown adipose tissue weight (iBAT), plasma leptin levels, plasma adrenocorticotropic hormone (ACTH) levels, plasma corticosterone levels, and adrenal weight (all analyzed at the end of day 6 [ZT11]) were measured in ad libitum–fed rats treated with vehicle or olanzapine.

[a] $p < .05$, *t* test.

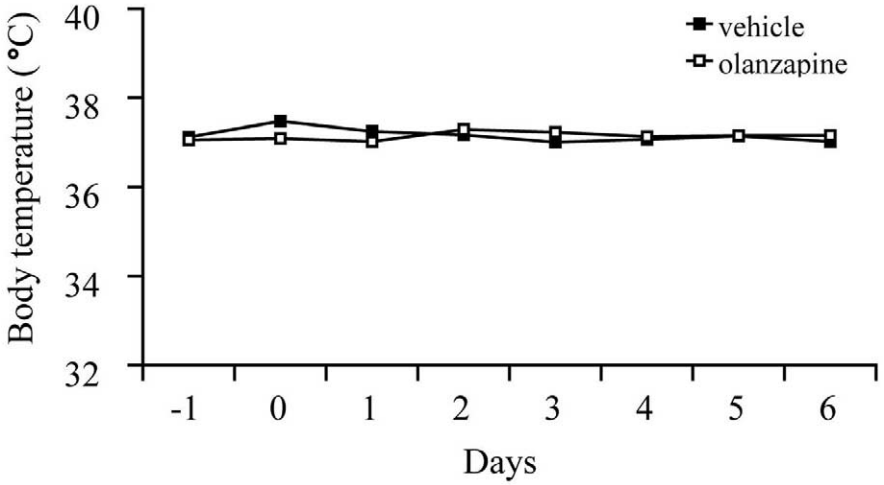

Figure 4. Basal body temperature in ad libitum–fed running rats. Basal body temperature of ad libitum–fed rats (day –1 [baseline] through day 6) following vehicle (closed squares) or olanzapine (open squares) treatment.

effect of olanzapine on RWA is independent of energy status. The absence of an effect of olanzapine treatment on food intake in food-restricted rats is unclear. It has been reported that olanzapine treatment enhances ingestive behavior in food-deprived rats (Thornton-Jones et al 2002); however, it is possible that the 1-hour feeding period in our experiment was too short to observe an orexigenic effect of olanzapine.

As a result of restricted feeding and wheel running, the HPA axis is considerably activated in ABA rats (Burden et al 1993). In our study, it was shown that olanzapine treatment decreased ACTH levels and tended to decrease corticosterone levels and adrenal weight in ABA rats, thereby reducing stress levels in ABA rats. The absence of a significant effect on corticosterone levels might be explained by the time of measurement because blood plasma was collected during the circadian peak of the corticosterone rhythm.

Hyperactivity in combination with food restriction results in a rapid exhaustion of energy stores in ABA rats. After a few days of exposure to the ABA model, energy stores are depleted, and body temperature cannot be maintained (Kas et al 2003). Here it was shown that chronic olanzapine treatment partly prevented the drop in basal body temperature in ABA rats. Basal body temperature of ad libitum–fed rats was not altered following chronic olanzapine treatment, which is in contrast with another study reporting dose-dependent hypothermia following acute olanzapine treatment in ad libitum–fed rats (Oerther and Ahlenius 2000). It has previously been shown that peripheral leptin treatment reduced hyperactivity of rats exposed to the semistarvation-induced hyperactivity model (Exner et al 2000), and it was proposed that decreases in leptin signaling might trigger hyperactivity in rats, as well as in AN patients (Hebebrand et al 1997). In our study, plasma leptin levels were undetectable in both vehicle- and olanzapine-treated ABA rats. Thus, it was shown that hyperactivity can be reduced in ABA rats without restoring plasma leptin levels. This suggests that other factors (possibly 5-HT) besides decreased plasma leptin levels may trigger development of hyperactivity. Our preliminary results from the clinical study also indicate that reduction of hyperactivity by olanzapine treatment is not significantly related to increases in plasma leptin levels, although absence of such an association might be due to the small sample size of our study.

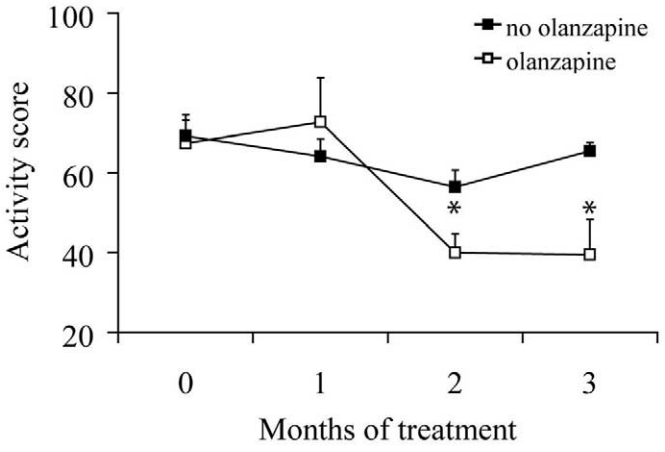

Figure 5. Effects of olanzapine treatment on physical activity in anorexia nervosa patients. Activity scores of anorexia nervosa patients with olanzapine treatment ($n = 7$, open squares) or without treatment ($n = 11$, closed squares). Activity was scored on a 0–100 scale by trained nurses. Note that at month 3, only five patients received olanzapine treatment. *$p < .05$, *t* test.

This study is one of the first to report chronic infusions of olanzapine in rats. At first glance, the dose used might appear extraordinary high and not in relation to the clinically effective dose. However, it was recently reported that chronic administration of olanzapine (by osmotic minipumps) in rats requires infusion concentrations at least 5 times higher (7.5 mg/kg, sc) than the optimal single dose (1–2 mg/kg sc) to achieve clinically comparable D_2 receptor occupancy (Kapur et al 2003). The difference in the olanzapine dose used in rats and humans is attributed to the fast biotransformation of olanzapine in rats ($T_{1/2}$ = 2.5 hours) as compared with humans ($T_{1/2}$ = 21–53 hours; Aravagiri et al 1997; Kapur et al 2003). As a result, the 7.5 mg/kg dose of rats results in clinically comparable D_2 receptor occupancy but would be supratherapeutic in (AN) patients.

The present study provides further support to the notion that olanzapine might be useful in the treatment of AN. In the small open-label study, olanzapine-treated AN patients showed a significant reduction of their physical activity levels, whereas their body weight and plasma leptin levels were not significantly different from medication-free AN patients. Reducing activity levels in AN patients who are hyperactive during admission might be crucial for further therapeutic outcome. There are several limitations to this clinical study, however. The nurses were not blind to the treatment condition when assessing activity, which might be lead to bias in the results. Because AN

patients were treated with olanzapine if they showed anxious behavior toward eating and body weight gain, it is possible that the reduction in activity levels is an indirect result of changes in anxious behavior, as has been reported earlier (Barbarich et al 2004; Boachie et al 2003; Malina et al 2003). This might suggest that AN patients attempt to alleviate anxiety through exercising. Recently, postulated that AN patients who showed dependence on exercise also showed high scores on the Beck Anxiety Inventory (Klein et al 2004). As anticipated, body weight and plasma leptin levels of AN patients were not significantly affected by olanzapine treatment. This might be explained by the relative short time of medication and the small sample size. Consequently, it also appeared that the reduction in activity was not related to changes in plasma leptin levels, similar to the results from the ABA model. Future controlled studies using a larger sample size should be performed to further explore the role of changes in plasma leptin levels in olanzapine-induced reduction of hyperactivity because a trend toward a stronger increase in plasma leptin levels in olanzapine-treated patients was present.

Only a few studies on olanzapine treatment in AN patients have been described. It has been reported that olanzapine increases body weight gain; reduces agitation, anxiety, and depression; increases ability or desire to eat; and decreases obsessive thoughts concerning body image (Barbarich et al 2004; Boachie et al 2003; Malina et al 2003; Mehler et al 2001; Powers et al 2002).

The central systems involved in the effects of olanzapine in our study are not yet clear. Olanzapine has highest affinity for 5-$HT_{2A/2C}$ receptors. It has been widely described that 5-HT pathways play an important role in energy balance. Increased levels of 5-HT or direct activation 5-$HT_{2A/2C}$ receptors reduces food intake (Clifton et al 2000; Vickers et al 2000), whereas blockade of 5-$HT_{2A/2C}$ receptors increases food intake and body weight gain (Currie et al 1996). Anorectic effects of d-fenfluramine (a 5-HT releaser and uptake inhibitor) are reduced in (obese) 5-HT_{2C} receptor deficient mice (Vickers et al 1999). Recently it was shown that 5-HT_{2C} receptors, which are located on pro-opiomelanocortin (POMC) neurons in the arcuate nucleus, mediate the appetite-suppressant effects of d-fenfluramine (Heisler et al 2002). In our study we have shown that olanzapine treatment decreases RWA; however, it was previously shown that fluoxetine (SSRI) and 5-$HT_{2A/2C}$ agonists also decrease RWA (Altemus et al 1996; Pirke et al 1993). Regarding conflicting data on serotonergic control of RWA, the possible involvement of other neurotransmitters should not be ignored. For instance, the antagonizing effects of olanzapine on the histamine 1 (H_1) receptor might also lead to reduced physical activity and increased food intake (Fukagawa et al 1989; Lozeva et al 2000). In addition, possible antagonism of the dopamine (DA) system causing decreased motor activity should not be disregarded, although it is generally stated that olanzapine has only limited extrapyramidal side effects. Modulation of motor activity following olanzapine treatment, as observed in this study, might thus be related to an altered balance between 5-HT-DA-H systems. Hence, it cannot be excluded that the observed reduction of activity in rats and patients in our study, as well as reduced anxiety levels described by others, are a result of sedation.

In conclusion, we have shown that olanzapine treatment decreased RWA in rats exposed to the ABA model as well as in ad libitum–fed rats. We also showed that olanzapine treatment in ABA rats partly prevented starvation-induced hypothermia and reduced HPA axis activation, whereas olanzapine increased food intake and body weight in ad libitum–fed rats. In addition, in a small open-label study, it was observed that hyperactive AN patients decreased physical activity following olanzapine treatment, without an influence on body weight gain and plasma leptin levels. Considering the common resistance of AN patients to therapy, the preliminary data on reduction of physical activity levels by olanzapine treatment, possibly making patients more amendable to further therapy, might be considered of importance. This finding should be considered with extreme caution, however, and underscores the need for additional, larger controlled trials on olanzapine treatment in AN patients. These trials should carefully analyze, among other factors, activity levels, body weight gain, plasma leptin levels, and therapeutic outcome. Taken together, these data on olanzapine treatment in anorectic rats and preliminary data of olanzapine treatment in hyperactive AN patients, in combination with results from other studies showing that olanzapine reduces anxiety and increases body weight gain in AN patients (Barbarich et al 2004), support the notion that olanzapine should be evaluated further as a therapeutic agent in AN patients.

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