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Chronic paroxetine infusion influences macronutrient selection in male Sprague–Dawley rats

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Abstract

Decreased intake and weight loss are among the side effects frequently reported with chronic selective serotonin reuptake inhibitor (SSRI) use in both humans and animals. In an earlier study, we documented that paroxetine administered for several weeks induced a weight loss of greater than 10% in some male Sprague–Dawley rats (Pharmacol. Biochem. Behav. 63 (1999) 435). As a follow-up to that work, we investigated in this study whether such treatment influenced dietary macronutrient selection. Animals were first habituated to foods containing principally either proteins, fats, or carbohydrates in a self-selection paradigm, after which they were implanted intraperitoneally with osmotic minipumps that delivered either paroxetine (7.5 mg/kg/day) or vehicle (50:50 ethanol:water) for 28 days; food intake and weight changes were documented during this period. No acute effects of the drug were apparent. By the fifth day of treatment, significant differences in weight gain between groups were observed and thereafter generally maintained for the remainder of the study, with animals receiving paroxetine showing about an 8% decrease in weight gain overall. Carbohydrate and fat intakes were significantly reduced, whereas preference was unchanged in fats and proteins and initially decreased in carbohydrates; in the latter, this pattern reversed and exceeded vehicle animals for the second half of the study. Several hypotheses are discussed with respect to specific and nonspecific effects of paroxetine on feeding and macronutrient selection.

Keywords: Selective serotonin reuptake inhibitor; Paroxetine; Carbohydrates; Fats; Proteins; Anorexia; Macronutrients

1. Introduction

Selective serotonin reuptake inhibitors (SSRIs), a novel class of antidepressants, are purported to be the treatment of choice for most patients clinically diagnosed with depression (Harvey and Bouwer, 2000; Nelson, 1999). In this class, paroxetine (Paxil), fluoxetine (Prozac), sertraline (Zoloft), fluvoxamine (Luvox), and, the newest, citalopram, have been found to be relatively effective in alleviating or managing depressive symptoms with generally fewer side effects than those associated with the traditional tricyclic compounds (Hiemke and Härtter, 2000; Nelson, 1999). However, there have been gastric problems reported with SSRI use, both in the human and animal literature, usually in the form of weight loss and gastrointestinal bleeding (Aranth and Lindberg, 1992; Edwards and Anderson, 1999; Fava et al., 2000; Harvey and Bouwer, 2000; Jose de Abajo et al., 1999; Konkle and Bielajew, 1999; Ottervanger et al., 1994; Spigset and Martensson, 1999). Because such effects can influence the interpretation of both clinically and experimentally based results, the mechanism of SSRI action and its role in producing these unwanted byproducts have been vigorously explored in recent years.

A variety of animal models have been designed to evaluate the effects of SSRIs and other serotonergic agents on food intake and weight change (Warwick and Schiffman, 1992). Modifying serotonin levels, either directly by adding 5-HT precursors such as 5-hydroxytryptophan, or indirectly by blocking the reuptake of the neurotransmitter itself, alters food intake both quantitatively and qualitatively (Li and Anderson, 1984; Simansky, 1996; Stallone and Nicolaidis, 1989). For instance, whereas increasing 5-HT availability in the neuronal synapses results in weight loss (Luo and Li, 1991), the opposite pattern—that of weight gain—occurs when 5-HT levels are reduced (Li and Anderson, 1984). In animals, central (hypothalamic) and systemic injections of SSRIs induce appetite changes for specific foods (Blundell, 1984; Leibowitz and Alexander, 1998) via a proposed

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control of carbohydrate selection (Harvey and Bouwer, 2000). Likewise, serotonergic drugs appear, in the short term, to suppress food intake in humans, due to a preferential reduction in carbohydrate consumption resulting in weight loss (Blundell, 1992). However, more recent clinical data suggest that some SSRIs, including paroxetine, induce weight gain in certain populations, a phenomenon that tends to occur with longer treatments (Fava et al., 2000; Harvey and Bouwer, 2000; Weber et al., 2000).

It is well known that SSRIs increase serotonergic activity by inhibiting the presynaptic reuptake mechanism. One idea is that among the many consequences of increased serotonin availability are a suppression in food intake, an overall increase in energy expenditure, and, thus, a reduction in body weight (Blundell, 1984; Harvey and Bouwer, 2000; Simansky, 1996). This anorexic effect is attributed to changes in food preference—more specifically, a selective decrease in the consumption of high-carbohydrate diets while "sparing" protein intake (Luo and Li, 1991; Stallone and Nicolaidis, 1989; Wurtman and Wurtman, 1979). The latter finding is commonly reported and has led to the suggestion that serotonin plays a role in regulating the relative proportions of macronutrients that an animal ingests (Stallone and Nicolaidis, 1989). Others (Harvey and Bouwer, 2000) have extended this idea to propose that enhancing 5-HT reduces carbohydrate intake, contributing to weight loss, and that inhibiting this mechanism promotes carbohydrate "craving," resulting in weight gain; whether this scenario occurs via SSRI-induced 5-HT needs to be explored. In one study, for example, Blundell and Hill (1989) reported only a decrease in fat intake with no reduction in carbohydrates and proteins, following the administration of D-fenfluramine; similar results have been reported by other laboratories (Kanarek and Dushkin, 1988). However, two recent studies by Heisler et al. (1997, 1999), using fluoxetine, noted weight loss in both male and female rats associated with fat and protein reductions; carbohydrate preference remained constant throughout the study. They interpreted their data to suggest that SSRIs altered macronutrient preference, rather than a specific effect on carbohydrate intake.

The present study is a follow-up to one conducted earlier in our laboratory (Konkle and Bielajew, 1999) in which several doses of the potent SSRI, paroxetine, were systemically administered under a chronic regime and their influence on the rewarding effects of brain stimulation were assessed. Among the observations was a substantial weight loss in a subset of rats receiving the highest dose (7.5 mg/kg/day). Note that this occurred only in the nonstimulation or control group of animals and led to their removal before the completion of the study due to weight loss greater than 10% during the drug treatment phase. Subsequent necropsy reports of these rats indicated a greater prevalence of liver dysfunction (i.e., discolouration and abnormal contouring of lobes) and kidney damage (Konkle and Bielajew, 1999). Gastrointestinal bleeding, a commonly reported hematological side effect of both fluoxetine (Aranth and Lindberg, 1992) and paroxetine (Ottervanger et al., 1994), is thought to reflect the impairment of the haemostatic function of blood platelet transporters to uptake serotonin (Jose de Abajo et al., 1999). After several weeks of treatment, this depletion of serotonin in the platelet's cell membranes leads to an increased risk of internal bleeding due to poor platelet aggregation (Jose de Abajo et al., 1999; Ottervanger et al., 1994). Aranth and Lindberg (1992) described a clinical case involving spontaneous bleeding (bruises and menorrhagia) in a woman treated with fluoxetine, who experienced heavy menstrual flow and had a large platelet count (as observed by a palpable spleen). Similarly, Ottervanger et al. (1994) reported that paroxetine decreases both whole blood serotonin and its uptake into the platelets; when drug use is discontinued, the symptoms disappear.

The question of interest in the present study was whether reduction in food intake and weight loss are related to the influence of paroxetine treatment on dietary choices. In order to explore this issue further, we used a dietary self-selection paradigm (Shor-Posner et al., 1991) in which rats were permitted to choose among three pure (90%) macronutrients in addition to their regular diet. Both the acute and chronic effects of paroxetine, administered via Alzet osmotic minipumps placed directly into the intraperitoneal cavity, were evaluated. We chose to examine only the dose (7.5 mg/kg) previously associated with a significant weight loss (Konkle and Bielajew, 1999). Animals were provided with a choice of three diets rich in one of three macronutrients (carbohydrates, fats, or proteins) as an alternative to their regular diet of Purina rat chow. Our objective was to evaluate whether paroxetine, infused chronically, acts to decrease overall energy consumption, or leads to a selective reduction in the intake/preference for a particular macronutrient.

2. Methods

2.1. Subjects and surgery

Fifteen male Sprague–Dawley rats (Charles River Laboratories), weighing between 150 and 175 g at the time of their arrival, were individually housed in plastic cages in a temperature-controlled facility. The animals were maintained on a reverse 12-h light/12-h dark cycle with lights off at 0800 h. Tap water and standard Purina rat chow were available ad libitum for 3 weeks, after which three macronutrient-enriched test diets (Standard and Custom Diets for Laboratory Animals, New Brunswick, NJ) consisting of 90% carbohydrates (D11512), 90% proteins (D11057), and 90% fats (D12267) were introduced. Regular chow continued to be available in the cage. The exact composition of the three diets appears in Table 1. (Standard Purina rat chow in the form of large pellets comprises roughly 70% carbohydrates, 25% proteins, and 5% fats.) Approximately 1

Table 1
Macronutrient selection diet composition for rodents

Ingredients	Principal macronutrient (90% of kcal)		
	Proteins (3.72 kcal/g) [g]	Carbohydrates (3.77 kcal/g) [g]	Fats (6.76 kcal/g) [g]
Casein, 30 mesh	**875**	50	50
D,L-methionine	**13.1**	0.75	0.75
Corn starch	20	**432.5**	20
Sucrose	20	**432.5**	20
Cellulose, BW200	50	50	50
Corn oil	22.2	22.2	22.2
Crisco	–	–	**366.7**
Mineral Mix S10001	35	35	35
Vitamin Mix V1001	10	10	10
Choline bitartrate	2	2	2
FD&C Red Dye 40	–	–	**0.1**
FD&C Blue Dye 1	–	**0.1**	–
FD&C Yellow Dye 5	**0.1**	–	–

The dashed lines indicate that the ingredient was absent in this diet. Differences in the components of the three test diets are highlighted in **bold**. The protein (yellow powder) and the carbohydrate (blue powder) diets were mixed with tap water; the fat diet (pink) was in a prepared ready-to-eat form. Values indicated are reproduced from the information supplied by E.A. Ulman, PhD, Research Diets.

month later, under halothane anesthesia, an Alzet osmotic minipump (Model 2ML4; Alza, Palo Alto, CA) was implanted in each rat, delivery portal first, into the peritoneal cavity. Analgesia was provided by rectal administration of Tylenol gel (50 mg/ml) immediately and 8 h postsurgery. The rats were distributed into three groups consisting of paroxetine ($n=6$), vehicle ($n=6$), and saline-treated ($n=3$) animals; the saline group was included to assess potential vehicle (50% ethanol) effects. The range of weights in each group was similar. Throughout the study, animals were closely monitored by the veterinary staff for signs of abnormal thermoregulation, activity, grooming, drinking, and elimination.

2.2. Drug

Paroxetine hydrochloride was generously donated by SmithKline Beecham Pharmaceuticals (Harlow, Essex, UK). The powdered compound was dissolved in 50% ETOH, which required brief stirring and mild heating, to achieve a final dose of 7.5 mg/kg. The pump was rated to provide a constant infusion rate of 2.5 μl/h with a stable drug concentration for 28 days.

2.3. Measurements and procedure

Body weight, macronutrient intake, latency to feed, and first food ingested were the four measures of interest. Body weights were determined near the end of the light cycle between 0600 and 0730 h, at which time macronutrient intake for that day was also recorded. Fresh diets were provided at the beginning of each dark phase. Latency to feed and first food sampled were determined upon the introduction of the food dishes (using a random sequence) to the cage.

Baseline measures were taken every second day for 1 month. On the first day of drug administration (the acute phase), body weight and macronutrient intake were observed at 4, 6, 8, 10, and 24 h postsurgery; note that the Alzet pump has a reported initial delay of 4–6 h to drug release (Alza) if not preactivated in warm physiological saline, which it was not in this case. The chronic phase lasted 28 days during which time all four measures were recorded daily for a period of nine consecutive days and then triweekly for the remainder of the experiment.

2.4. Pathology

At the end of the treatment phase, a semiquantitative biochemical test for whole blood urea nitrogen (BUN), using Azostix reagent strips from Ames, was performed on each rat from tail vein blood; the results provide information on renal function. The animals were then sacrificed by intraperitoneal injection of a lethal dose of sodium pentobarbital (Somnotol). A necropsy report was generated for every animal in order to evaluate the general condition of the internal organs, particularly the liver, kidneys, and the gastrointestinal tract; paroxetine is reportedly metabolized in both liver and kidneys (Adler and Angrist, 1995; Hiemke and Härtter, 2000).

3. Results

3.1. Baseline data

For the 3 days preceding surgery, mean baseline values of each group's body weight, latency to feed, first food ingested, and amount consumed of each diet were determined. The data were analyzed with and without the inclusion of the saline group and no significant effects were found either way. The values across groups were nearly identical. During this phase, equal proportions based on caloric content of the protein and carbohydrate diets were selected (almost 45% each) and the rest of the diet (10%) were fats. The average total amount ingested was 40–42 g across groups. Feeding was initiated, on average, within 1 min of food presentation, and carbohydrates were consistently selected first in all but two rats. Throughout the study (baseline, acute, and chronic phases), the 30-min criterion to commence eating was not met six times, or 3% of the data collected, and did not occur systematically in any one animal. The regular chow available in the cage went untouched. Data associated with the saline group were not

considered in the remaining analyses, nor were they shown in the figures.

3.2. Acute data

Acute data were analyzed using a two-factor split-plot ANOVA with Group as the independent factor (paroxetine and vehicle) and Time as the repeated factor (6, 8, 10, and 24 h postsurgery); Greenhouse–Geisser (G–G) corrections were applied to the degrees of freedom of the repeated factor in order to correct for any violations to the assumption of sphericity (Howell, 1997). The two groups did not differ significantly in carbohydrate and fat consumption, but protein intake was significantly decreased in the case of the paroxetine group throughout the acute phase [$F(1,10) = 6.86$, $P = .025$]. Not surprisingly, both groups decreased intake

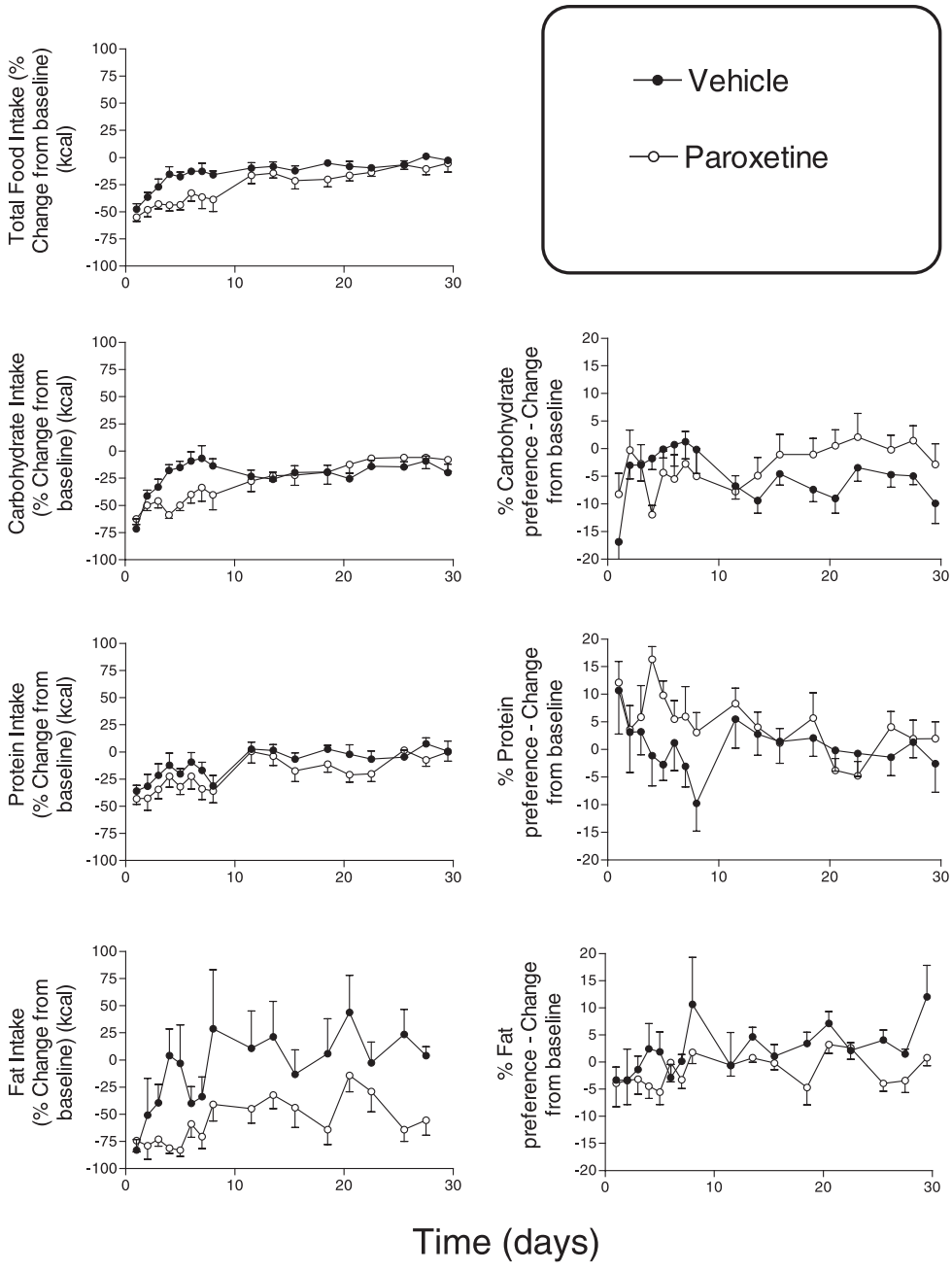

Fig. 1. The left panel shows average food intake (percent change from baseline), expressed in kilocalories, in the vehicle (filled circles) and paroxetine (unfilled circles) groups over time. The 0% point on the ordinate represents the baseline average. Changes in total food consumed appear in the top panel, followed by changes in specific macronutrients—carbohydrates, proteins, and fats on the bottom. The right panel illustrates macronutrient preference (percent mean macronutrient intake divided by total intake) in the paroxetine and vehicle groups over the same time period. Carbohydrate preference appears on the top right panel, protein preference in the middle panel, and fat preference on the bottom.

overall and to a similar degree, which we attribute to the surgery experience. The cumulative intake during the acute phase is the value associated with the first day in each graph in Fig. 1.

3.3. Chronic data

The data collected during the month-long chronic phase of the study are shown in Figs. 1 and 2. A split-plot ANOVA (with G–G correction) was used to analyze changes in intake (overall and specific macronutrients), macronutrient preference, and weight gain with Time as the repeated factor (17 levels) and Group as the independent factor (two levels). The left panel of Fig. 1 illustrates the change in mean intake of each macronutrient plotted separately across groups. Significant Group differences were observed in total food [$F(1,10)=7.66$, $P=.02$; vehicle>paroxetine] and fat intake [$F(1,9)=6.3$, $P=.03$; vehicle>paroxetine]. The pattern of fat intake was stable over time, whereas it was significantly altered in the case of proteins [$F(5,50)=10.65$, $P=1\times10^{-5}$], carbohydrates [$F(6,55)=9.17$, $P=1\times10^{-5}$], and total food intake [$F(5,47)=16.39$, $P=1\times10^{-5}$], with the vehicle group consuming more than the paroxetine group in every instance. An interaction between Group and Time was observed only in the case of carbohydrate intake [$F(6,55)=3.25$, $P=.01$]. However, when the analyses were redone, separating the data into two periods (Days 1–12 and 13–28), only group total food intake was significantly altered during the first phase [$F(1,10)=12.95$, $P=5\times10^{-3}$] and group protein intake during the second phase of the study [$F(1,10)=9.92$, $P=.01$]. Animals in the paroxetine group consumed a daily average of 19 g of proteins vs. 24 g in the vehicle group.

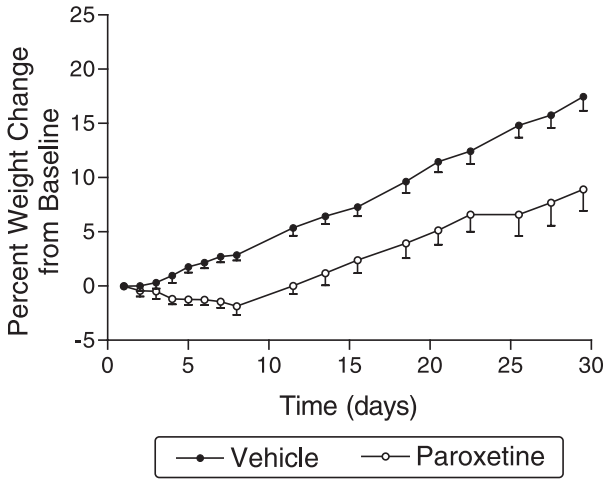

Fig. 2. Average weight change expressed as a percent difference from baseline for each group—vehicle (filled circle) and paroxetine (unfilled circle)—during the chronic phase of the study. Baseline levels for each group are set at 0%.

The right panel of Fig. 1 depicts the change in macronutrient preference of each group. The preference of each macronutrient was significantly altered during the course of the study (time factor); however, only the values associated with carbohydrate preference gave rise to a significant interaction [$F(11,114)=1.90$, $P=.04$] when the less conservative Hundt–Felt correction factor was applied (Howell, 1997). Overall, the protein- and carbohydrate-enriched diets remained the most preferred—and fats the least preferred—throughout the study. While the data suggest that protein preference was initially elevated in the paroxetine group relative to the vehicle condition, this effect just failed to meet significance (uncorrected $P=.058$). Preference for the carbohydrate-enriched diet was reduced for about 10 days postsurgery in the paroxetine group and thereafter was significantly greater than the vehicle group; the latter showed a 5–10% decline in preference over time.

Fig. 2 shows the changes in body weight of the two groups during the chronic phase. All factors achieved significance: Group [$F(1,10)=18.06$, $P=.002$]; Time [$F(1,14)=88.38$, $P=1\times10^{-6}$]; Group × Time [$F(1,14)=6.71$, $P=.01$]. Note that in further analyses, the interaction was present only during the first phase or Days 1–12 of the study [$F(2,20)=8.95$, $P=.002$]. Initial weight loss is apparent (up to the eighth session) in the paroxetine-treated animals as compared to the steady increase in weights of the other group. Post-hoc tests revealed statistical differences between the two groups starting at Session 5 and thereafter for the remaining sessions. By the end of the chronic phase, the difference in weight gain between the two groups was about 9%.

First food ingested was monitored from the beginning of baseline to the end of drug infusion. The pattern observed during baseline tests—that carbohydrates were typically selected first—remained stable during the drug phase of the study, as confirmed by chi-square analyses. For frequency of carbohydrates vs. other diets, the result was $\chi^2(1)=294$, $P=1\times10^{-5}$, and this pattern was consistent over time (baseline vs. treatment phase).

3.4. Biochemistry and necropsy data

The semiquantitative test for whole BUN revealed no apparent differences between the groups; the estimated values were similar: 23 mg/dl (paroxetine) and 20 mg/dl (vehicle). Only one animal in the paroxetine group had a slightly elevated level (more than 26 mg/dl). The results of the necropsy report showed that all groups had some minor degree of liver discolouration and, for the most part, the bladders and kidneys appeared normal. However, half of those animals receiving the antidepressant drug revealed abnormal blood coagulation, adhesions of the pumps to the mesentery, and development of large cysts surrounding the opening end of the pumps. There was no consistent correspondence between animals with these problems and weight loss or reduced carbohydrate intake. The paroxetine group was divided into animals with and without tissue abnormal-

ities, and their weights were examined at the point of least weight gain (Session 8) and at the end of the study (see Fig. 2). The "normal" paroxetine group showed a weight gain of −1% and +10% and the other group −3% and +8% at these two time points.

4. Discussion

Chronic administration of paroxetine was shown to reduce total caloric intake in paroxetine-treated rats, particularly during the first 10 days of drug treatment. Specific consumption of fats and carbohydrates was most affected, and fat intake never resumed baseline values in this group, remaining consistently at 25–50% below that recorded during the pretreatment phase. Protein intake, in contrast, was not significantly altered between groups, but was overall reduced during the first 10 days. Coincident with the period of reduced food intake was a small weight *loss* in the paroxetine group and a subsequent slower rate of weight gain as compared to the control animals. No particular difference in the latency to feed was evident in any of the rats over the 2 months of data collection—equal baseline and treatment period. Although animals typically ate the carbohydrate meal first, overall preference, expressed as the ratio of specific macronutrient to total food consumed, was similar for carbohydrate- and protein-rich diets, about 45%, and the remaining 10% in fats. Preference during the first half of the chronic phase shifted, however, in the paroxetine group; the reduction in fats and carbohydrates was balanced by an increase in protein preference. In the long-term, the preference patterns are quite similar between the two groups; however, whereas carbohydrate preference is reduced in vehicle-treated animals during the latter half of the chronic period, such changes are not observed with paroxetine treatment.

One interpretation of these data, given the results of the necropsy report, is that nonspecific malaise accounted for the weight loss and changes in intake and dietary preferences in the paroxetine animals. Note that three of these animals displayed some degree of gastrointestinal pathology. In order to address this possibility, we compared the intake of each macronutrient separately between the two paroxetine-treated subgroups (i.e., animals with adhesions and those without). We found no difference between these two subgroups in carbohydrate intake (average intake of 11.87 and 12.77 g for groups with and without adhesions, respectively), evaluated over the first 12 days of drug treatment, or fat intake, evaluated over the full treatment period, suggesting that factors related to gastrointestinal pathology were not obviously influencing intake. Note that in the case of carbohydrate intake, the magnitude of the subgroup difference (animals with and without adhesions) was 0.332 (based on Cohen's *d*), and 0.223 in the analyses done on the intact groups (all subjects included)—relatively comparable values. As reported above, weight gain in the two subgroups was likewise similar during the session associated with the least weight gain and the end of the study. Ideally, necropsy assessment should be conducted during the period of least weight gain in a subset of animals rather than at the end of the study as was done here.

There is also concern that the infusion of paroxetine, 4–6 h following surgery (pump was not prewarmed), might have interfered with wound healing. Although drug release is typically begun immediately following surgery in such studies, a better alternative would be to preload the pump with several days worth of vehicle solution before commencing drug infusion, if the intraperitoneal route is desired, as was the case here. This is relevant given that paroxetine has been reported to cause platelet dysfunction (Ottervanger et al., 1994), although normal platelet aggregation and coagulation have also been observed (Alderman et al., 1996), hence its favorable safety profile. We also compared the pattern of weight gain in our groups to that reported by others using alternative routes of constant paroxetine infusion. For example, Yamane et al. (2001) observed, at the end of 14 days via subcutaneous infusion, a 21-g difference between the vehicle and drug groups, comparable to our 16-g difference for the same time frame. Note that our dose of paroxetine was a little lower (7.5 mg/kg instead of their 10 mg/kg dose) and our rats were slightly heavier. Despite these arguments, we cannot rule out that, at least in some animals, weight loss includes factors unrelated to paroxetine's primary effect on feeding. Closer examination of meal microstructure elements, for example, would be warranted in this case. In addition, it may be that paroxetine induces a general state of malaise, unrelated to problems discussed above, such as wound healing, formation of adhesions, route of administration, etc., which contribute to its anorectic effects.

A second possibility is that paroxetine had a specific effect on macronutrient consumption, supporting the contention that serotonin agonists decrease carbohydrate intake (Luo and Li, 1990, 1991; Mok et al., 2000; Weiss et al., 1991; Wurtman and Wurtman, 1977), at least in the short run. It has been suggested that a carbohydrate–serotonin negative feedback loop mechanism is responsible for this alteration (Wurtman and Wurtman, 1979)—the idea being that consumption of carbohydrates will ultimately increase cerebral tryptophan (precursor to serotonin) levels, consequently facilitating serotonin synthesis. This increase in serotonin provides feedback to the organism, allowing for the proper adjustments in carbohydrate intake. Our data suggest that artificially increasing serotonin synthesis via drug administration may regulate carbohydrate intake in a similar fashion. Furthermore, as paroxetine typically requires 10–14 days to reach steady-state plasma levels (Tulloch and Johnson, 1992), this time frame coincides with the return of carbohydrate consumption to basal levels observed here. The synchrony of these two events may signify the involvement of a negative carbohydrate–serotonin feedback loop mechanism in mediating the anorectic action of paroxetine and the specificity of this action to carbohydrates.

At least one group has reported instead decreases in fat and/or protein intake following the administration of serotoninergic agonists (Heisler et al., 1997, 1999; Orthen-Gambill and Kanarek, 1982). Similarly, we found fat intake to be significantly suppressed throughout the month-long drug infusion period, but not fat preference, which recovered to baseline during the second half of this period. It has been suggested that an anorectic action of the drug would be most discernable in the preferred macronutrient (Heisler et al., 1997, 1999), and their work demonstrates this phenomenon, in that carbohydrates, the least preferred macronutrient, was least affected by fluoxetine treatment in both male and female rats. In our study, proteins and carbohydrates were equally preferred—yet only intake of the latter macronutrient and fats, the least preferred, was affected. We examined individual differences in preference and found great consistency across animals with almost all consuming about equal proportions of proteins and carbohydrates. While our rats were habituated to a diet containing roughly 70% carbohydrates, 25% proteins, and 5% fats (percent macronutrients of standard Purina rat chow) before baseline collection commenced, their selection during and after baseline did not perfectly parallel that breakdown, and showed a preference for proteins and fats relative to their standard chow. The latter, although available, was never eaten.

Others have suggested that the source of macronutrient may play an important role in its preference. In one study, for example, rats preferentially chose a high-carbohydrate diet vs. a high-fat diet when the source of carbohydrates was either sucrose or Polycose, but not when it was corn starch (Glass et al., 1997). More recently, this group (Glass et al., 1999) has evaluated a variety of lipid sources including lard, vegetable shortening, and corn oil, and found the first two to be preferred to a high-carbohydrate diet, and the reverse to occur when the fat source is corn oil. In our study, the carbohydrate source incorporated both sucrose and corn starch in equal amounts. Similar arguments regarding the source of macronutrient as a determinant of preference have been made for proteins and fat consumption (reviewed in Kanarek, 1985; Sclafani, 1987). Such factors may be extremely important in weeding out the discrepancies between studies.

One group (Blundell and Hill, 1989) has suggested that the long-term administration of drugs that activate serotonergic receptors, such as D-fenfluramine, is most potent in rats made hyperphagic by a cafeteria-style diet—carbohydrates (icing sugar) and fats (beef drippings)—prior to drug experience. In their study, after 90 days of this regime, little tolerance to the drug-induced anorexia was exhibited and was not dependent on reduction of either carbohydrates or fats. While our animals were introduced to the self-selection paradigm 1 month before the drug phase of the study, obesity was not established and their weight gain during baseline tests was similar to animals fed a standard chow diet (Konkle and Bielajew, 1999). The degree of the anorectic effect would be expected to be greater in hyperphagic rats and may also be influenced by the nature and familiarity of the diet, as discussed above.

As in humans, the results of the present study suggest that giving animals the choice of macronutrients provides a healthier nutrient opportunity as well as being more ecologically relevant. In our previous study (Konkle and Bielajew, 1999), we examined the effects of chronic paroxetine administration on several measures, including percent efficiency of food intake (takes into account the amount ingested as a function of weight gain); only access to standard rat chow was available, a food that is primarily composed of carbohydrates ($\sim$70%). Compared to that study, the animals in this one fared much better. Weight loss was substantially less and, in fact, the animals allowed a self-selective diet recovered from the anorectic effect of the drug in about 10 days of intake, evaluated over the first 12 days of drug treatment, or fat intake, evaluated over the full treatment period.

The effect of serotonergic agents on feeding mechanisms is complex, and a number of variables need to be acknowledged in designing relevant experiments, including diet composition, taste, texture, etc. Employment of the self-selection paradigm is one way to address these issues and provides a richer context in which to study these effects. In addition to food characteristics, factors such as species (Feigin et al., 1987), strain, age (Leibowitz et al., 1991), and sex (Archer, 1975; reviewed in Kanarek, 1985) likely influence feeding patterns and preferences, making it challenging to design animal paradigms to model human behavior. Particularly in chronic investigations of drug effects, the use of self-selection procedures should provide more information about the role of specific macronutrients in regulating food intake and energy expenditure.

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