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THE EFFECT OF LONG-TERM DEXFENFLURAMINE TREATMENT ON 24-HOUR ENERGY EXPENDITURE IN MAN. A DOUBLE-BLIND PLACEBO CONTROLLED STUDY

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In order to investigate the effect of long-term treatment with dexfenfluramine (dF) on 24-hour energy expenditure (EE), 10 obese females were studied in a double-blind design. Shortly before and 4 weeks after cessation of a 13 months treatment period with either dF (30 mg/day) or placebo (PL) the 24-hour EE was measured. The measurements were performed using a 24 m^3 direct heat sink calorimeter with continuous real time measurements of evaporative and sensible heat losses. The patients performed a standardized program of exercise, rest and meals. The measurements were performed at 24°C and at a humidity between 3 and 11 g/m^3. Discontinuation of dF treatment did not change energy expenditure significantly from placebo, neither when expressed in kJ/kg lean body mass nor in kJ/kg body weight. After cessation of treatment total 24-hour EE decreased likewise nonsignificantly by 2.9 percent in the dF group and by 4.0 percent in the PL group. EE measured over 24 hours was subdivided into day and night periods and into resting energy expenditure as well as a measurement of the heat losses over a period of 3 hours after a meal. This subdivision of the EE showed similar nonsignificant differences. The conclusion is therefore that dF possesses no significant thermogenic effect during long-term administration in human obese subjects.

Keywords: dexfenfluramine, obesity, thermogenesis, human, calorimeter, energy expenditure.

Introduction

The action of fenfluramine and other specific serotoninergic drugs is generally believed to be attributed to their anorectic properties. However, several studies in rodents have provided evidence of a peripheral thermogenic effect. Levitsky *et al.*[1] found that rats during prolonged treatment with fenfluramine were able to sustain a weight loss even after having returned to normal food intake. Subsequent work[2] has demonstrated that fenfluramine has no thermogenic effect in fasting animals, but potentiates significantly the thermogenic effect of carbohydrates, but not lipids. Studies failing to demonstrate a thermogenic effect of fenfluramine have used fasting subjects[3–5], whereas studies claiming an effect were performed in the postprandial phase[6,7].

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In rats, the thermogenic action of fenfluramine seems to be exerted through an increased metabolic activity in brown adipose tissue mediated by the sympathetic nervous system and inhibited by drugs like metergoline[8,9]. Likewise it has been suggested that increased lipolytic activity might explain the increased thermogenesis during physical activity after administration of dexfenfluramine in rats[10].

In humans the results are contradictory. Durnin *et al.*[3] were unable to find any effect of fenfluramine either at rest or during standardized exercise, whereas others have found an effect both on the post-prandial phase[11,12] and on the basal metabolic rate[12]. As carbohydrate *per se* activates the sympathetic nervous system[13] it is likely that fenfluramine and the more active and specific isomer, dexfenfluramine (dF), might be able to potentiate the thermogenic effect of other stimuli associated with increased sympathetic nervous system activity in humans (i.e. exercise, arousal, smoking, anxiety, cold exposure etc.). However, until now no study has examined whether dF or other serotoninergic drugs possesses a potentiating effect on the 24-hour heat loss during long-term treatment of obese humans under conditions imitating normal daily living.

Material and methods

Ten obese female patients were studied twice in a 24 m^3 whole-body calorimeter. The first examination was performed when patients were still being treated with either dF (30 mg/day) or placebo (PL) and the second examination was performed 4 weeks after having stopped the trial medication. The first examination was carried out at the end of a 13 months treatment period. In addition to the medication a dietary program had been instituted at the start of the 13 months period as the patients were prescribed a 1625 kJ formula diet (NUPO®) plus free additional food intake up to a total of 4.7 MJ a day. When a satisfactory weight loss had been achieved or when net weight loss during two consecutive months had not exceeded 3 kg, the formula diet was stopped and a 5.0 MJ conventional diet prescribed. For instruction, a isoenergetic unit system was used. Every month patients were seen at group meetings and offered patient education as well as individual counselling. After the first investigation patients were re-examined one month later in the calorimeter when the trial drug had been discontinued for 4 weeks. The study was conducted in a double-blind design. The patients were selected from a larger patient population by a third independent party, who was requested to select patients from each original randomly assigned treatment regimen in order to obtain the best possible matching with respect to sex, age and weight loss. Matching of body weight was not possible due to the limited number of patients.

Physical characteristics of the patients are shown in Table 1.

Lean body mass

At the first visit the lean body mass (LBM) was measured by bio-electric impedance as described by Lukaski[14], using the formula: LBM = 0.85 × height^2 × 1/resistance + 3.04. LBM was almost equal in the two groups as shown in Table 1.

LBM at the second examination (after discontinuation of the drug) was calculated as LBM at the first examination plus 25 percent of the weight gained between the examinations. This was done in order not to underestimate the fat free component (vessels, connective tissue etc.) of the weight gain[3,15]. The accuracy of bio-electric impedance will remain under discussion until large cross-sectional studies of obese subjects are carried out. However, as we are testing differences before and after cessation of a treatment these problems are not crucial for using the method.

Direct whole-body calorimetry

Measurements of energy expenditure (EE) as total *heat loss* were performed by direct whole body calorimetry using the 24 m^3 direct heat sink calorimeter at the Department of Physiology, Odense

Table 1. *Patient characteristics and weight changes during and after 13 months double-blind treatment with dexfenfluramine (dF) or placebo (PL) in 10 obese female patients.* All values are means ± 1 s.d.

	Dexfenfluramine group	*Placebo group*	
Age (years)	33.6 ± 9.2	32.0 ± 9.2	n.s.
Weight before start of treatment (kg)	88.6 ± 5.0	105.0 ± 17.7	*
Weight after 13 months of treatment (kg)	74.0 ± 9.5	95.7 ± 24.4	n.s.
Overweight % after 13 months of treatment	27.9 ± 18.0	38.8 ± 19.3	n.s.
Lean body mass after 13 months of treatment (kg)	54.0 ± 7.9	57.7 ± 4.9	n.s.
Lean body mass 1 month after cessation of treatment (kg)	54.7 ± 7.8	57.9 ± 4.9	n.s.
Weight loss after 9 months of treatment (kg)	15.0 ± 8.6	13.1 ± 5.8	n.s.
Weight loss after 13 months of treatment (kg)	14.6 ± 7.6	9.3 ± 6.7	n.s.
Weight gain after 1 month of cessation (kg)	2.9 ± 0.5	0.8 ± 1.3	*

* $P < 0.05$ dF *v.* PL (Student's *t* test)

University, Denmark. Details have been described elsewhere[16–18]. Briefly, the calorimeter allows continuous real time measurements of sensible (radiative, conductive and convective) and evaporative heat loss with a linear reponse between 0 and 320 W for evaporative heat with a precision varying from 4.0 to 0.6 percent over the range of 25–100 W, and between 0 and 280 W for sensible heat with a precision varying from 1.4 to 0.2 percent (range 50–200 W). The drift stability was less than 0.6 W over 24 and 72 hours. The within-subject between-weeks variability is less than 2.2 percent[17]. The response times (95 percent) was less than 15 minutes for both evaporative and sensible heat. There was no measurable 'cross talk' between evaporative and sensible heat inputs and only negligible dependency on the external air humidity, ranging from 14 to 70 percent. The experiments were performed at 24 ± 0.1°C and at an ambient humidity of 3–11 g/m^3. Familiarity with the calorimeter equipment and independence from preceding physical exercise was obtained by accommodating the patients to the calorimeter during part of the day and the night previous to the investigation, as shown in Table 2. During the 24-hour period from day 2 to day 3, patients performed a standardized program including fixed periods of exercise, rest, sleep etc. as shown in Table 2. The patients received three mixed meals and one sandwich. Totally they were equivalent to 125 kJ/kg LBM/24 hour. The patients spent 25 hours continuously in the calorimeter from 9.00 a.m. on day 2 to 10.00 a.m. on day 3. The measurements started at 10.00 a.m. after the morning meal. The subjects were provided by the depot unit with cotton underwear, jogging suit, socks and sandals and used a light blanket for bedding. Energy expenditure was measured continuously and subsequently integrated over 24 hours. This measurement was further divided into two long periods, an activity or day period and a sleep or night period. The day period lasted from 10.00 a.m. to 10.00 p.m. on day 2 plus from 6.30 a.m. to 10.00 a.m. on day 3. The night period lasted from 10.00 p.m. on day 2 to 0.6.30 a.m. on day 3. During the day period two specific measurements of EE were performed. Firstly, the resting energy expenditure (REE) was measured from 07.00 to 07.20 a.m. on day 3 while the subject remained in bed after the night's sleep and was awake 30 minutes before the measurement. Secondly, the postprandial heat loss (PPHL) was measured from 10.20 a.m. to 13.20 p.m. on day 2 after a standard meal has been given at 10.00 a.m. The meal consisted of 15.6 percent protein, 10.1 percent fat and 74.3 percent carbohydrate.

Table 2. *Schedules of activity, rest and meals before and during the 24-hour calorimeter measurements.* Energy values of the meals are given as means ± s.d.

Day 1	
1700–1720	Installation and instruction in the calorimeter
1720–1800	Sitting
1800–1820	Bicycling at low workload (25 W)
1820–1900	Sitting
1900–2200	Leave the calorimeter, dinner, watching television etc.
2200–0630	Sleeping in the calorimeter
Day 2	
0630–0720	Lying awake in the bed, listening to the radio.
0720–0900	Leaving the calorimeter, morning bath, toilet etc.
0900–1000	Installation in the calorimeter, sitting in a chair
1000–1020	Breakfast (2318 ± 159 kJ)
1020–1320	Sitting. DIT measurement.
1320–1400	Lunch, sitting, coffee or tea (2750 ± 599 kJ)
1400–1420	Bicycling
1420–1500	Sitting (reading, writing etc.)
1500–1520	Walking
1520–1600	Sitting (reading, writing etc.)
1600–1620	Bicycling
1620–1700	Sitting (reading, writing etc.)
1700–1720	Walking
1720–1800	Sitting (reading, writing etc.)
1800–1820	Bicycling
1820–1900	Sitting (reading, writing etc.)
1900–1940	Dinner (1871 ± 215 kJ)
1940–2100	Sitting (coffee or tea)
2100–2120	Walking
2120–2200	Sitting (sandwich)
2200–0630	Undressing, evening toilette, sleeping
Day 3	
0630–0720	Lying awake in bed
0720–0800	Morning toilette, sitting
0800–0820	Walking
0820–0840	Bicycling
0840–1000	Sitting (reading, writing etc.)
1000–	Leaving the calorimeter, end of investigation

Toilet activities only in connection with periods of activity.

Ethical considerations

The protocol was approved by the Municipal Ethical Committee of Copenhagen and observed the ethical rules of the Second Declaration of Helsinki.

Statistical analysis

All results are presented as means ± standard deviation. Nonparametric data analysis, Pratt's test and the Mann–Whitney rank-sum test as well as parametric analysis (Student's t test) were used for significance testing of differences within and between groups. P values below 0.05 were considered significant.

Results

Body weight

Changes in mean body weight during and after cessation of treatment are shown in Table 1.

Energy expenditure

The present paper includes results of energy expenditure obtained over five different time periods. A 24-hour period from day 2 at 10.00 a.m. to day 3 at 10.00 a.m. was divided into a daytime and a night period and EE was presented as evaporative heat loss, as sensible heat loss and as total heat loss in kJ per kg LBM (Table 3). Total EE/kg LBM decreased nonsignificantly by 2.9 ± 5.0 (s.d.) percent in the dF group and by 3.6 ± 7.2 percent in the PL group. The difference was −0.7 percent with 95 percent confidence limits of −8.7 to +7.2 percent. The data from the PPHL and REE measurements are shown in Table 4. PPHL was calculated as heat loss during the 3-hour period after the meal minus the REE value. The PPHL values increased by 7.9 percent in the dF group and decreased by 7.2 percent in the PL group. The changes were nonsignificant. Total 24-hour EE without correction for LBM changed nonsignificantly from 9783 ± 822 kJ to 9623 ± 940 kJ in the dF group and from 11 340 ± 2288 kJ to 10 886 ± 1851 kJ in the PL group. The individual values are shown in Fig. 1. When calculating the 24-EE per kg body weight (data not shown) no significant differences were demonstrated.

Table 3. *Twenty-four hour energy expenditure (EE) measured as evaporative heat loss (EHL) and sensible heat loss (SHL) during daytime (6.30 a.m. to 10.00 p.m.) and during the night period (10.00 p.m. to 6.30 a.m.).* Measurements were performed shortly before and 4 weeks after cessation of a 13 months double-blind treatment period with dexfenfluramine or placebo. All values are means ± 1 s.d. in kJ/kg lean body mass over the time periods.

		Dexfenfluramine		*Placebo*	
Period		*During dF treatment*	*After dF treatment*	*During PL treatment*	*After PL treatment*
Day	(SHL)	95.3 ± 9.7	97.0 ± 11.8	97.7 ± 19.2	96.6 ± 15.0
	(EHL)	50.7 ± 13.9	42.9 ± 12.6	58.3 ± 19.2	50.6 ± 15.9
	Total	146.0 ± 18.4	139.8 ± 23.0	156.0 ± 36.4	147.2 ± 28.9
Night	(SHL)	25.2 ± 3.7	27.7 ± 3.1	27.5 ± 3.3	28.9 ± 5.1
	(EHL)	12.2 ± 2.2	10.9 ± 1.1	14.9 ± 5.0	13.3 ± 3.1
	Total	37.4 ± 4.6	38.6 ± 4.0	42.4 ± 9.3	42.2 ± 7.7
24–hour	(SL)	120.5 ± 13.1	124.7 ± 14.5	125.3 ± 23.9	125.4 ± 20.1
	(EL)	62.9 ± 16.0	53.7 ± 13.4	73.2 ± 23.8	64.0 ± 17.9
	Total	183.4 ± 22.1	178.4 ± 26.1	198.5 ± 45.5	189.4 ± 36.1

Statistical analyses using Pratt's test and the Mann–Whitney rank-sum test showed no significant differences between and within groups.

Table 4. *Postprandial heat loss (PPHL) measured as the total postprandial heat loss after a mixed meal (TPPHL) minus the resting energy expenditure (REE) shortly before and 4 weeks after cessation of a 13 months double-blind treatment period with dexfenfluramine or placebo in obese patients.* All values are means ± 1 s.d in kJ/kg lean body mass over a 3-hour period.

	Dexfenfluramine		*Placebo*	
Period	*During dF treatment*	*After dF treatment*	*During PL treatment*	*After PL treatment*
TPPHL	20.02 ± 3.11	20.14 ± 3.62	22.26 ± 5.90	21.34 ± 4.26
REE	17.10 ± 3.30	16.99 ± 1.89	17.70 ± 3.50	17.10 ± 3.60
PPHL	2.92 ± 2.58	3.15 ± 2.17	4.57 ± 4.15	4.24 ± 3.23

Statistical analyses showed no significant differences between and within groups (Pratt, Mann–Whitney and Student's *t* tests).

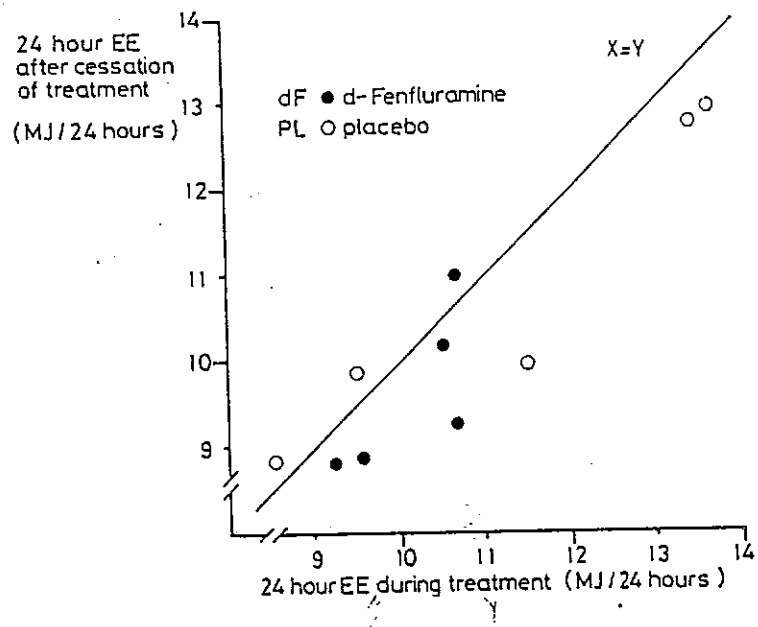

Fig. 1. *Twenty-four hour energy expenditure during and after cessation of 1 year treatment with dexfenfluramine or placebo.* Points below the line of identity indicate a decrease in energy expenditure after discontinuation of treatment.

Discussion

After introduction of specific serotoninergic drugs without hazardous side-effects, the goal of pharmacological treatment of obesity has changed from short-term support during the introduction of a diet towards long-term treatment. The aim is to maintain the patients in extended treatment programs and to prevent relapse during critical periods in which enhancement of the metabolic activity would be

especially beneficial. To our knowledge no studies have yet investigated the influence of long-term serotoninergic drug treatment on energy expenditure in humans. Fenfluramine, and more recently dF, have previously been shown to increase oxygen consumption or heat loss after carbohydrate rich meals in short-term animal experiments[2,6–8], whereas no effect has been found after intake of lipids. Furthermore, dF has not been able to increase weight loss in obese patients on a strict very low calorie diet (< 1300 kJ) used in order to eliminate the contribution of the anorectic effect of the drug[5,19]. In this investigation we used a 24 m^3 calorimeter. This provided an environmental situation in which it is possible to imitate normal daily living conditions with measurements of energy expenditure caused by major and minor exercise, i.e. fidgeting, drinking, eating, sleeping etc. The patients were adapted to the calorimeter by staying in the chamber overnight before starting the measurements. Any effect of changes in thermogenesis attributable to the menstrual cycle were avoided by repeating the measurements at approximately 4 weeks interval[20]. The increase in body weight after cessation of treatment in the dF group has been taken into consideration by calculating the results per kg LBM. This investigation was performed at the end of a 1-year treatment period, at which time tolerance to the thermogenic effect of dF may have developed. The fact that the body weight actually increased during the final 3 months of treatment in both the dF and PL groups may be taken as support for this interpretation, but changes in food intake and physical activity are also possible.

Differences between rats and humans concerning the brown adipose tissue may also be of importance since dF has been found to increase metabolic activity in BAT[8,9], which does not significantly contribute to thermogenesis in man[21]. The calorimeter measures the subject's heat losses in the chamber. From the work of Dauncey[22] such measurements, when performed over 24 hours, are equal to EE as measured by indirect calorimetry. During shorter periods, it cannot be ensured that direct and indirect calorimetry produces the equal results. However, Garby *et al.*[23], in a group of 21 females and 38 males, compared indirect measurements of EE using the conventional Douglas bag method at basal conditions with the present procedure for measuring REE. The difference between these two methods were 2.9 percent and 2.2 percent respectively, and nonsignificant. It is therefore reasonable to accept that under the present circumstances a steady state prevailed.

During shorter periods, heat losses may not be equivalent to heat production. Pittet *et al.*[24] demonstrated that heat production after a meal was larger than heat losses. At 145 minutes after the meal they found an increase in the internal core temperature of approximately 0.19°C. There was a variation related to the meals content of glucose and/or protein.

If we assume that changes of the same order of magnitude also occurred in our subjects and further that this heat storage still existed 3 hours after the meal, then the EE would be higher than the measured heat losses. Assuming a mean weight of 80 kg for the group and a similar distribution of core and shell in all of them, then a 0.19°C temperature increase will be equivalent to 3.474 kJ/°C/kg × 80 kg × 0.19°C = 52 kJ stored in the body. Over a 3 hour period the total heat loss was approximately 1600 kJ. Thus, if no change occurred during this period in the core

temperature, the error over the 3 hours is approximately 3 percent. If part of this heat storage was also lost during this period, then the error is less than 3 percent.

The limited number of patients carries an increased risk of overlooking a possible true difference (type 2 error), i.e. an effect of dF on energy expenditure. On the other hand, the validity of the results is strengthened by the very high precision of the calorimeter, as indicated in the Results section. In conclusion, the present results lend no support to the idea that dF possesses any significant thermogenic properties during long-term treatment of obese patients.

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