Body mass index of male and female Wistar rats following administration of leptin hormone after a dietary regime

ABSTRACT

Introduction: Obesity is a problem affecting people of all ages and socioeconomic status. Leptin hormone (LH), a product of the obesity gene, is a key regulator of feeding and energy expenditure. **Aim:** The study is aimed at determining the effect of LH on body mass index (BMI) in Wistar rats after a diet regime. **Materials and Methods:** Forty rats (male [M], n = 20; female [F], n = 20), aged 9 weeks and weighing 77.2–123.0 g, were randomly divided into two Groups A (M) and B (F) and further divided into four subgroups of n = 5. They were maintained *ad libitum* on different diet and water for 10 weeks. Group 1; control (standard rat feed), Group 2 (high fat diet), Group 3 (protein diet), and Group 4 (carbohydrate diet). BMI was calculated weekly for 10 weeks (pretest). LH was injected for 2 weeks; the BMI was then calculated (posttest). Paired t-test was used to analyze the differences between the BMI pre- and post-test periods and also to analyze for sexual dimorphism for the pre- and post-test periods. Level of significance was at $P < 0.05$. **Results:** The results revealed no significant difference ($P > 0.05$) in the BMI for the pre- and post-test period and no sexual dimorphism for BMI during the pretest period. However, there was a significant difference ($P < 0.05$) and sexual dimorphism for BMI during the posttest period. **Conclusion:** This result implies that the LH is more effective in the male gender than the female when considering BMI and hence may reduce the risk associated with obesity.

Key words: Body mass index, diet regime, leptin hormone, obesity, sexual dimorphism, Wistar rats

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INTRODUCTION

Obesity is a problem affecting people of all ages, racial, ethnic background, and socioeconomic status. The obesity problem may have been distributed unevenly across populations at first, but it is largely due to urbanization, change in work structure and technology, increased access to greater amounts of food, especially more calorically dense foods, more leisure time, and sedentary activities.[1] Overweight refers to an excess amount of body weight that may come from muscles, bone, fat, and water.[2] Obesity refers to an excess amount of body fat.[2] With over one billion people now overweight or obese,[3] the obesity epidemic is not restricted to industrialized societies; this increase is often faster in developing countries than in the developed countries.[4] According to the 2010 World Health Organization (WHO) survey data on Nigeria, the prevalence of overweight was 26% and 37% in men and women, respectively, while the prevalence of obesity was 3% and 8.1% in men and women, respectively.[5,6] States that the prevalence figures for obesity within the European region confirm that in most countries, the number of obese women surpasses the number of obese men, sometimes as

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much as 2–1.[6] A study by Puepet *et al*.[7] among a sample of adult Nigerians in Jos stated that the prevalence of overweight and obesity was 21.4% in males and 23.5% in females (M:f = 1:1.3, $P < 0.05$).[7]

Leptin is a 167-amino acid peptide with a four-helix bundle moiety (motif) similar to that of a cytokine;[8,9] it is produced predominantly in the adipose tissue but is also expressed in a variety of other tissues, including placenta, ovaries, mammary epithelium, bone marrow, and lymphoid tissues.[10] Leptin acts on receptors in the hypothalamus where it inhibits appetite, counteracts the effects of neuropeptide Y (a potent feeding stimulant secreted by cells in the gut and in the hypothalamus), and also promotes the synthesis of α-melanocyte stimulating hormone; an appetite suppressant. The absence of leptin (or its receptor) leads to uncontrolled food intake and results in obesity.[11] Available evidence from human studies indicates that leptin has a mainly permissive role, with leptin administration being effective in states of leptin deficiency and less effective in states of leptin excess.[12]This adipocyte-derived hormone was once heralded to be an anti-obesity agent. While leptin is effective on certain individual bearing congenital leptin deficiencies[13] as monotherapy, in a work by Scarpace and Zhang in 2009, leptin has been shown to be disappointing in humans and rodents with common obesity, that is, obesity associated with elevated serum leptin under a normal genetic background. Leptin production increases proportionately with adiposity, and leptin levels are high in rodent and human model of diet-induced or adult-onset obesity. Yet, the increased leptin fails to curtail the progression of obesity.[14]

Obese Zucker rats had the highest plasma leptin concentrations, and Sprague-Dawley rats had the lowest [Figure 1].[15] The half-life of circulating leptin (approximately 25 min) is constant over a range of adiposity.[16] Several studies have shown that plasma leptin concentrations are appropriate for the degree of subject's adiposity in anorexia nervosa,[15] in wasting syndrome of HIV infection,[17] and in lean long-distance runners.[18]

Overweight and obesity lead to adverse and metabolic effects on blood pressure, cholesterol, triglycerides, and insulin resistance. Some of the leading causes of preventable death among adults are obesity-related conditions such as heart disease, stroke, type 2 diabetes, and some types of cancer (endometrial, breast, colon).[19] Excess weight also increases the risk of liver and gall bladder disease, sleep apnea, osteoarthritis, and gynecologic problems such as infertility.[19-22] According to the WHO as of 2005, 1.6 billion adults (over 15 years) are overweight and 400 million are obese, and as of 2015, 2.3 billion will be overweight and 700 million obese. Currently, more than 1 billion adults are overweight and at least 300 million of them are clinically obese.[23] The study is aimed at determining the effect of leptin hormone (LH) on body mass index (BMI) in Wistar rats after a diet regime.

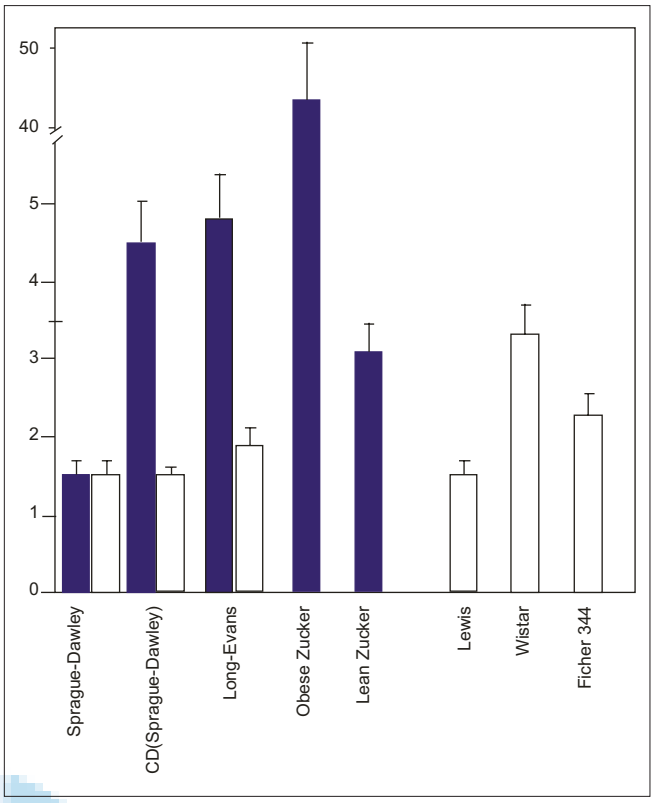

Figure 1: Plasma leptin concentrations in various strain of rats

MATERIALS AND METHODS

Animals

Forty Wistar rats (male, n = 20, female, n = 20), aged 9 weeks and weighing between 77.2 and 123.0 g, were randomly divided into two Groups A (male) and B (female); each group was further divided into four subgroups of n = 5 per group. All groups were maintained *ad libitum* on experimental diets and water for 10 weeks. They were humanely treated according to the Declaration of Helsinki (2013) for the care of animals after obtaining clearance from the Our Lady of Apostles Hospital Ethical Committee (2016).

Diets and grouping

Group 1 (A_1 and B_1), the control, received standard rat feed and water only, Group 2 (A_2 and B_2) received high-fat diet (margarine, 90%) mixed with some standard rat feed (10%), Group 3 (A_3 and B_3) received protein diet (soybeans, 100%), and Group 4 (A_4 and B_4) received carbohydrate diet (cereal, 100%).

Experimental design

The BMI $\left(\frac{\text{Weight(g)}}{\text{Length(cm}^2\text{)}}\right)$ (g/cm^2) was calculated weekly for 10 weeks (pretest) and after the LH administration (posttest)

(Normal BMI: male [0.45 ± 0.02–0.68 ± 0.05] g/cm^2,[24] female [0.4504–0.5044] g/cm^2[25]). LH was administered intraperitoneally at a dose of 3.5 μg daily at 12:00–12:30 pm for 2 weeks.

Statistical analysis

The data were analyzed using SPSS statistical package version 22.0 (IBM SPSS Inc., Chicago, IL., USA) for Windows 8.0. Paired *t*-test was used to analyze the differences between the BMI pre- and post-test periods and also to analyze for sexual dimorphism for the pre- and post-test periods. Level of significance was at $P < 0.05$.

RESULTS

The results of the experiment are presented in Tables 1-4 and Figures 1-3 below.

There was no significant difference ($P = 0.70$) in the BMI of the pre- and post-test periods and there is a weak correlation (0.03). Further, the relationship is positively correlated meaning that there was an increase of the BMI in the periods.

There was no significant difference in the BMI of pre- and post-test periods ($P = 0.56$) that means there was no significant change in the calculated BMI despite the use of the hormone when the result was paired.

There was no significant difference ($P = 0.258, 0.3$) for gender in BMI for the pretest period.

There was a significant difference for sexual dimorphism in BMI for the posttest period ($P = 0.01, 0.01$).

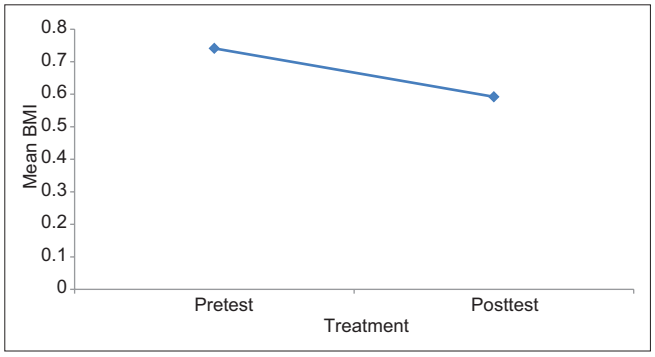

Figure 2: Comparison of the means between pre- and post-test periods

There is a slight reduction/decrease in the paired means of the two periods (pre- and post-test), but it is of no statistical significance.

A wide difference between the means of the two sexes during the pretest period is reduced when compared with the posttest period. This could suggest a more profound effect of the LH on the male gender.

DISCUSSIONS

During the research period, we noticed that the rat fed on fat diet showed hyperphagia with an initial increase in weight and calculated BMI; our observation was as reported by Wang *et al.*[26] which showed that during the first 7–10 days, the rats on fat-rich diet exhibited significant hyperphagia, an increase in both total and fat intake that produces higher body weight gain.[26]

The results from this study showed no significant difference ($P = 0.70$ and $P = 0.66$) in the BMI during the pre- and post-test period; there was no significant change in the BMI despite the injection of the LH. In earlier studies that support our findings, it was shown that initial attempts to utilize leptin as a monotherapy for obesity (including using supraphysiological doses) revealed modest, if any, weight loss and with significant variability among participants.[27-29]

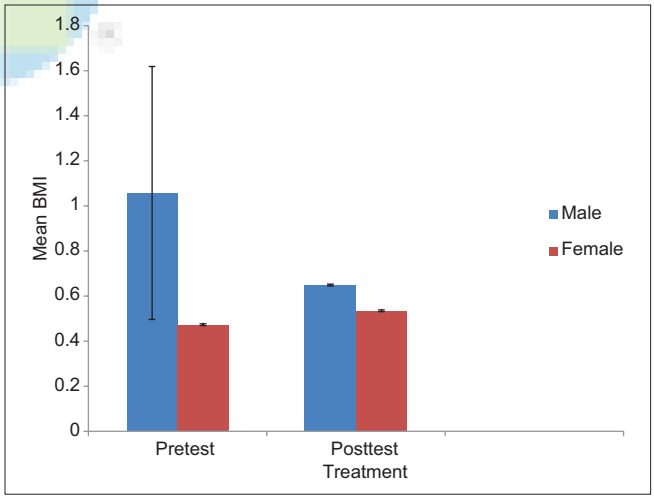

Figure 3: Gender difference in body mass index for pre- and post-test periods

Table 1: Effect of leptin hormone on pre- and post-test body mass index (correlation)

	Mean	*n*	SD	SEM	Correlation (pre- and post-test)
Pair 1					
Pretest BMI score	0.74	216	3.78	0.26	0.03
Posttest BMI score	0.59	216	0.07	0.01	

There was no significant difference ($P=0.70$) in the BMI of the pre- and post-test periods and there is a weak correlation (0.03). Further, the relationship is positively correlated meaning there was an increase of the BMI in the periods. SD=Standard deviation; SEM=Standard error of mean; BMI=Body mass index

Table 2: Effect of leptin hormone on pre- and post-test body mass index

	Mean	SD	SEM
Paired 1			
Pretest BMI score-posttest BMI score	0.15	3.78	0.26

There was no significant difference in the BMI of pre- and post-test periods (P=0.56). That means there was no significant change in the calculated BMI despite the use of the hormone when the result was paired. SD=Standard deviation; SEM=Standard error of mean; BMI=Body mass index

Table 3: Effect of leptin hormone on pretest period for gender difference in body mass index

	Sex	*n*	Mean	SD	SEM
Pretest BMI score	Male	99	1.06	5.58	0.56
	Female	117	0.5	0.05	0.00

There was no significant difference (P=0.258; 0.3) for gender in BMI for the pretest period. SD=Standard deviation; SEM=Standard error of mean; BMI=Body mass index

Table 4: Effect of leptin hormone on posttest period for gender difference in body mass index

	Sex	*n*	Mean	SD	SEM
Posttest BMI score	Male	108	0.65	0.05	0.00
	Female	117	0.54	0.05	0.00

There was a significant difference for sexual dimorphism in BMI for the posttest period (P=0.01; 0.01). SD=Standard deviation; SEM=Standard error of mean; BMI=Body mass index

In this study, when the gender differences in BMI were analyzed for the pre- and post-test periods, the results showed no significant difference ($P > 0.05$) during the pretest period and a significant difference ($P < 0.05$) during the posttest period, i.e., after the injection of the LH. The result in Figure 2 showed a reduction in the mean BMI of the male gender when compared to that of the female gender during the posttest period; this could be as a result of the excess leptin that is found in the female because it has been shown that for the same age and BMI, women have greater leptin concentrations than men.[30] Moreover, available studies indicate that the effects of leptin on weight loss and other physiological parameters appear to be more pronounced during states of relative leptin deficiency[31-33]and largely ineffective in states of leptin excess.[12] Hence, this could explain why the administered LH was more effective in the male gender.

CONCLUSION

The result implies that the LH has a sexual dimorphism in BMI, and hence, it is more effective in the male gender and may reduce the risk associated with obesity. Further studies need to be done on the role of LH in a larger population with graded LH dose and increase duration of research.

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Conflicts of interest
There are no conflicts of interest.

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