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EVALUATION OF CHRONIC EFFECTS OF HYPOESTROGENISM ON LEPTIN AND BODY WEIGHT. POSSIBLE PHARMACOLOGICAL MODULATION

Esposito E., 2° anno di corso del Dottorato in Scienza del Farmaco, XVI ciclo. Durata del Dottorato in anni: 3. Sede di servizio: Dipartimento di Farmacologia Sperimentale, Università degli Studi di Napoli "Federico II", Via D. Montesano,49- 80131, Napoli.

Leptin, the product of the ob gene, is thought to play a critical role in the regulation of adipose tissue mass and expenditure energy, and may directly or indirectly influence reproductive function.
Estradiol lipolytic effects and the association of leptin (LP) level with fat mass are both well-documented phenomena (Considine et al.,1996).
The present studies is undertaken to investigate the role of estrogen and raloxifene, a selective estrogen receptor modulator (SERM), in the modulation of leptin serum level and leptin receptor expression in different peripheral (subcutaneous, peritoneal, perimetral) and central (hypotalamus, hippocampus, striatum) tissues. An ovariectomy-induced obesity model is used to mimic the postmenopausal condition of hypoestrogenism in women (Chu et al., 1999).
In this investigation, a short-, middle- and long-term study (4, 7 and 22 weeks, respectively) was conducted to monitoring the change of serum leptin level in rats after the loss of estrogen (E_2), the (E_2) replacement (25 ìg/kg twice a week, s.c. in 1 ml of sesam oil) or the raloxifene treatment (3 mg/kg daily, per os in 1 ml of water).
To examine the effectiveness of ovariectomy in decreasing estrogen production and the restore by E_2 supplement, serum estradiol levels were also measured. The ovariectomy reduced significantly ($p<0.05$) the estrogen level in ovariectomized rats (Ovx) and this effect was reversed by E_2 supplement ($p<0.05$).
In all groups of animals, the increase of body weight and food intake was monitored every week, and before the sacrifice all animals were examined to determine the body fat content by analysis of bioelectrical impedance (measured by BIA 101). In the ovariectomized rats the increase of body weight was related to an increase of body fat mass ($p<0.001$).
Three weeks after the ovariectomy, the Ovx rats had significantly higher body weight than the controls (sham-operated animals) ($p<0.01$), and continued to gain more weight thereafter. Estrogen replacement and raloxifene treatment reversed significantly the weight gain and their effects were still evident in the long-term study ($p<0.001$).
Variations of LP serum levels in the different groups of animals were reported in table 1:

TAB.1: Variations of LP levels (ng/ml) in the different groups of rats in short (4 w), middle (7w) and long time study (22 w). In the two tratments (A and B) sham-operated and Ovx rats received the same vehicle of E_2 or raloxifene.

Treatment A	**Sham-operated**	**Ovx**	**Ovx+E_2**
4 weeks	1.76±0.28	1.78±0.19	1.82±0.08
7 weeks	1.08±0.27	2.30±0.44*	1.16±0.10$^{\#}$
22 weeks	2.45±0.51	5.85±0.93**	1.91±0.12$^{\#\#\#}$

Treatment B	**Sham-operated**	**Ovx**	**Ovx+Raloxifene**
7 weeks	1.21±0.14	1.84±0.23	1.20±0.18
22 weeks	2.78±0.54	4.40±0.52*	1.95±0.20$^{\#\#\#}$

*p<0.05 vs sham-operated; **p<0.01 vs sham-operated
#p<0.05 vs Ovx; ### p<0.001 vs Ovx

As evident, serum leptin levels were found to be significantly higher in Ovx rats vs controls. This effect was reversed by E_2 supplement both at 7 and 22 weeks. Raloxifene treatment significantly decreased serum leptin concentration only at 22 weeks.
By Western Blot analysis we have also evaluated the effect of ovariectomy on the expression of the functional isoform of the leptin receptor, Ob-Rb, in different adipose and cerebral tissues.
Preliminary data on subcutaneous adipose tissue show a 130-150 kDa band which probably represents the glycosylated form of the receptor Ob-Rb.
In the long-term study it is evident a decreased expression of Ob-Rb isoform in Ovx rats and this effect is reversed by E_2 supplement or raloxifene treatment in the subcutaneous adipose tissue.
We hypothesize that Ob-Rb expression is inversely related to changes in serum leptin induced by different estrogen levels.

References

1. Considine R. et al.(1996) N. Engl. J. Med. 334: 292-295

2. Chu S et al.(1999) Life Sci. 64: 2299-2306.