Paracrine Regulation of Growth Hormone Secretion by Estrogen in Women

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Context: Paracrine regulation is emerging as a discrete control mechanism in the endocrine system. In hypogonadal men, stimulation of GH secretion by testosterone requires prior aromatization to estradiol, a paracrine effect unmasked by central estrogen receptor blockade with tamoxifen. In hypogonadal women, estrogen replacement via a physiological non-oral route fails to enhance GH secretion, indicating an absence of an endocrine effect. The aim was to investigate whether local estrogens produced from aromatization regulate GH secretion.

Design: We conducted an open-label, two-phase, crossover study.

Patients and Intervention: We compared the effects on GH secretion of tamoxifen with estradiol valerate in postmenopausal women. Ten women were treated with tamoxifen (10 and 20 mg/d) and estradiol valerate (2 mg/d) via oral route for 2 wk each, with a washout period of at least 6 wk.

Main Outcome Measures: We measured the GH response to arginine and circulating levels of IGF-I and SHBG, markers of hepatic estrogen effect.

Results: The GH response to arginine was reduced by 10- and 20-mg tamoxifen in a dose-dependent manner and potentiated significantly ($P < 0.05$) by estradiol valerate. Mean IGF-I concentration was reduced significantly with high-dose tamoxifen ($P < 0.01$) and estradiol valerate treatment ($P < 0.05$), whereas mean SHBG levels rose with both ($P < 0.01$).

Conclusions: Blunted GH response to stimulation occurring in the face of reduced IGF-I feedback inhibition with tamoxifen indicates that GH secretion was suppressed by estrogen receptor antagonism. Because circulating estradiol was unaffected, these data indicate a significant role of local estrogen in the central control of GH secretion. We conclude that aromatase mediates the paracrine control of GH secretion in women. **(*J Clin Endocrinol Metab* 95: 3771–3776, 2010)**

In addition to endocrine action of hormones, there is emerging evidence that paracrine regulation by hormones produced locally in the target tissue may play a major role in the regulation of the endocrine system (1). This is exemplified in men, in whom inhibition of LH secretion from the pituitary gland and stimulation of GH secretion by testosterone requires prior conversion to estradiol by the aromatase enzyme (2–4). Thus, estrogen produced locally from aromatization plays a major role in the regulation of GH secretion in men.

In women, the effect of exogenous estrogen on the regulation of GH secretion is route-dependent. Oral administration of estrogen enhances GH secretion; however, this does not happen when estrogen is replaced by a physiological non-oral route (5). Administration of estrogen by the oral route reduces IGF-I production via a first-pass hepatic effect (5–9). IGF-I, a principal marker of hepatic GH action, exerts a potent negative feedback to inhibit GH secretion (10), such that GH secretion is stimulated when circulating IGF-I levels fall. Thus, the increase in GH levels during oral estrogen ad-

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Abbreviations: CVs, Coefficients of variation; SST, somatostatin.

ministration most likely arises from the reduced negative feedback from IGF-I. This is supported by findings that transdermal estrogen replacement achieving physiological concentrations of estradiol in blood fails to reduce IGF-I levels, at the same time exerting no effect on GH secretion (5, 7). Although circulating estrogen does not appear to directly influence GH secretion in women, it does not exclude the possibility that estrogens produced locally from aromatization may do so.

Our aim was to investigate the role of local estrogen in the regulation of GH secretion. We hypothesized that estrogens produced locally from aromatization may regulate GH secretion in women. We therefore used tamoxifen to investigate a putative role of local estrogens in the regulation of GH secretion in women. Tamoxifen is a synthetic estrogen compound that exerts central estrogen receptor antagonistic (11) and hepatic estrogen agonistic effect (11, 12). This allows the investigation of central estrogen antagonism concurrently with peripheral estrogen agonist action in the liver. Because of the hepatic estrogen-like effects, both tamoxifen and oral estrogen are expected to reduce IGF-I levels, reducing the negative feedback on GH secretion. If local estrogen should play a major role in the regulation of GH secretion in women, central estrogen receptor antagonism by tamoxifen should induce a fall in GH levels despite reduced negative feedback from circulating IGF-I.

Subjects and Methods

Subjects

Ten healthy postmenopausal women (aged 56–80 yr) were recruited from the community through advertisements. Subjects were menopausal for at least 2 yr before the study, and none of them had received any form of estrogen for the last 6 months before entry into the study. All women were in good general health and had normal hematological, renal, and hepatic function documented before recruitment into the study. Exclusion criteria included body mass index of at least 30 kg/m^2, hypothalamic or pituitary disorders, diabetes mellitus, and chronic renal or hepatic illnesses. Subjects were not taking any medications known to interfere with the endocrine system. Throughout the study, subjects were instructed to follow their usual diet and to continue on their usual medications or supplements. St. Vincent's Hospital Human Research Ethics Committee approved the study, which was conducted in accordance with the principles of the Declaration of Helsinki. All study participants gave written informed consent. The study was registered with the Australian New Zealand Clinical Trials Registry (ACTRN12607000586415).

Study design

This was an open-labeled two-phase crossover study comparing the effects of tamoxifen to estradiol valerate. The tamoxifen phase consisted of a two-dose sequential evaluation of tamoxifen (Genox, Alphapharm, Carole Park, Australia) at doses of 10 and 20 mg/d for 2 wk each. The estradiol phase consisted of a 2-wk treatment with estradiol valerate (Progynova, Schering-Plough Limited, North Ryde, Australia) at a dose of 2 mg/d. The washout period between the two phases was at least 6 wk. Two of 10 subjects were unable to participate in the estradiol valerate phase for personal reasons.

For tamoxifen, we studied both 10- and 20-mg doses because they are used therapeutically, and previous studies had observed a significant reduction in IGF-I in women treated with the 20-mg dose of tamoxifen (13, 14). For estradiol valerate, we used the therapeutic dose of 2 mg/d, which previously had shown a significant dissociation of the GH-IGF-I axis (6, 15).

Subjects were studied in the Clinical Research Facility, Garvan Institute of Medical Research. All participants were instructed to fast the night before each visit. Studies were undertaken at baseline and after each of the 2-wk treatment periods. On each visit, assessment of GH status using arginine stimulation test was performed, and circulating IGF-I and SHBG levels were measured. Study bloods were collected, and serum samples were obtained by centrifugation and stored at −20 C until analysis.

Arginine stimulation test

Subjects rested on a bed for at least 30 min before the baseline blood samples were taken. Thirty grams of L-arginine hydrochloride (Phebra Pty Ltd., Lane Cove, Australia) were infused over a 30-min period. Blood samples for GH level measurements were taken at 0 and 30, 60, and 90 min after commencement of arginine infusion. Blood glucose levels were measured at baseline and 60 min after arginine infusion. No serious side effects or complications were reported during the test.

Analytical methods

All samples for any individual were measured in the same assay run for each analyte. Blood IGF-I and SHBG levels were measured at each visit. Serum IGF-I was measured by RIA after acid ethanol extraction as previously described (6, 16, 17). The coefficients of variation (CVs) for IGF-I were 8.3% at 14.7 nmol/liter and 7.4% at 28.6 nmol/liter. Serum SHBG was measured by RIA using a commercial assay (Immulite 2000; Siemens Medical Solution Diagnostics, Los Angeles, CA). The CVs for SHBG at 5.3 and 86.2 nmol/liter were 5.0 and 7.5%, respectively. GH was measured by ELISA calibrated against IS 80/505 (Bioclone Australia Pty Ltd., Marrickville, Australia) with sensitivity of less than 0.1 mIU/liter. All serum samples from five different timepoints were measured in duplicate following the manufacturer's instructions. The CVs for GH were 4.4% at 6.5 mIU/liter and 3.5% at 17.6 mIU/liter. Blood glucose was measured by glucose analyzer (YSI 2300 Stat Plus; Yellow Springs Instrument Co., Yellow Springs, OH).

Statistical analysis

The GH response to arginine was not normally distributed, and therefore data were logarithmically transformed for the analysis. Peak GH levels were calculated as maximum increment over prestimulated GH concentration. The GH response was analyzed by repeated measures ANOVA. Treatment effects of estrogen and tamoxifen on IGF-I and SHBG were assessed using paired *t* tests with Bonferroni's correction. Results were expressed as means with SE values, and a *P* value of less than 0.05 was considered to be significant. Statistical analysis was under-

taken using the statistical software package Statview 4.5 PPC (Abacus Concepts, Inc., Berkeley, CA).

Results

Mean age of subjects was 63.6 ± 2.2 yr, and mean body mass index was 24.9 ± 1.3 kg/m^2. Menopausal state was validated by measurements of circulating 17β-estradiol and FSH levels at baseline. Mean blood 17β-estradiol was 28.7 ± 2 pmol/liter, and mean FSH was 59.6 ± 9.2 IU/liter.

GH

Mean baseline GH level was 2.6 ± 0.5 mIU/liter. At baseline, 70% of subjects achieved their maximum GH stimulation at 60 min after arginine infusion. There was no significant change in blood glucose levels during the arginine stimulation test. Prestimulation GH levels did not determine GH response to arginine at baseline or during the treatment with tamoxifen or estradiol valerate. Arginine induced a significant rise in GH concentration at baseline ($P < 0.0001$), during estradiol valerate ($P < 0.0001$), and during low-dose ($P < 0.05$) but not high-dose tamoxifen treatment. When compared with baseline, GH response to arginine was attenuated by low- and high-dose tamoxifen treatment in a dose-dependent manner (mean Δ, −36% and −88%, respectively) and potentiated significantly by estradiol valerate treatment (mean Δ, 84%; $P < 0.05$; Figs. 1 and 2A). The difference in GH response was significant between tamoxifen and estradiol valerate treatments ($P < 0.05$; Fig. 1).

IGF-I

At baseline, the mean IGF-I level in blood was 14.8 ± 1.2 nmol/liter, which was reduced in a dose-dependent manner by 1.5 ± 0.7 and 3.7 ± 0.9 nmol/liter ($P < 0.01$) during low- and high-dose tamoxifen treatments, respectively (Fig. 2B). The reduction in IGF-I levels during high-dose tamoxifen was significantly greater than that during low-dose tamoxifen treatment ($P < 0.05$). Mean circulating IGF-I levels fell significantly by 3.2 ± 1.1 nmol/liter during treatment with estradiol valerate ($P < 0.05$; Fig. 2B). The effects on IGF-I levels between tamoxifen and estradiol valerate treatments were not significantly different. Peak GH response did not significantly correlate with serum IGF-I concentration at baseline or during tamoxifen or estradiol valerate treatment phases.

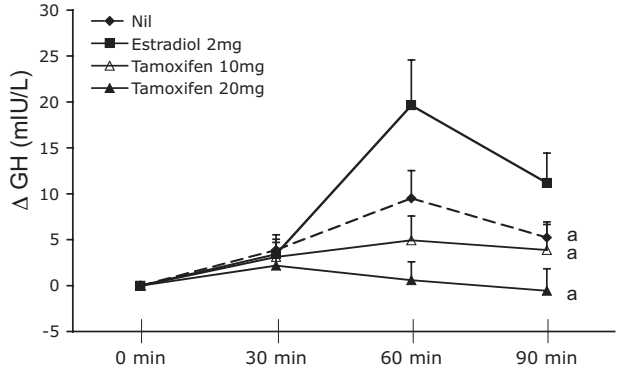

FIG. 1. Change from prestimulation serum GH levels at 30, 60, and 90 min after arginine infusion measured before (nil; *dashed line*), during oral treatment with estradiol valerate (2 mg/d), and during low-dose (10 mg/d) and high-dose tamoxifen treatment (20 mg/d). Data are expressed as means ± SEM. "a" indicates significant ($P < 0.05$) difference compared with treatment with estradiol valerate. Conversion factor: 1 mIU/liter = 0.33 μg/liter.

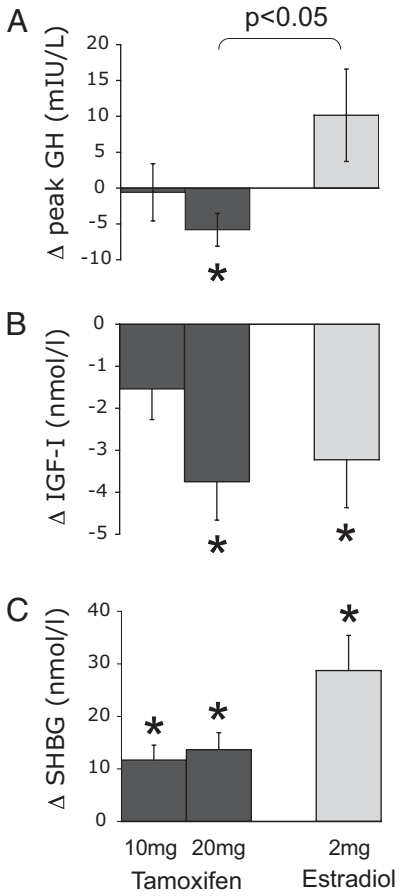

FIG. 2. A, Change from baseline in peak GH response to arginine during oral treatment with tamoxifen (10 and 20 mg/d) and estradiol valerate (2 mg/d) in postmenopausal women. Data are presented as maximum increment in serum GH over prestimulation GH levels during arginine infusion. Conversion factor: 1 mIU/liter = 0.33 μg/liter. B and C, Change from baseline in serum IGF-I levels (B) and in serum SHBG levels during low- and high-dose tamoxifen and during oral treatment with estradiol valerate. Data are expressed as means ± SEM. *, $P <$ 0.05 compared with baseline.

SHBG

At baseline, mean SHBG level was 45.5 ± 6.9 nmol/liter, which rose significantly ($P < 0.01$) by 11.7 ± 2.8 and 13.7 ± 3.2 nmol/liter during low- and high-dose tamoxifen treatment, respectively (Fig. 2C). During treatment

with estradiol valerate, mean SHBG level increased significantly by 28.7 ± 6.7 nmol/liter ($P < 0.01$; Fig. 2C). The effects on SHBG levels between tamoxifen and estradiol valerate treatments were not significantly different. There was a significant inverse correlation between the changes from baseline in SHBG and IGF-I levels; in other words, the higher the increase in SHBG, the lower the serum IGF-I levels ($r^2 = -0.21$; $P < 0.05$).

Discussion

This is the first study comparing the impact of oral estrogen and tamoxifen on the GH-IGF-I axis regulation in women. Estradiol valerate significantly enhanced, whereas tamoxifen attenuated the GH response to arginine, with the effect being significantly different between tamoxifen and estradiol valerate treatment phases. Both estradiol valerate and tamoxifen significantly reduced mean IGF-I concentration and increased mean SHBG levels, with the effect not significantly different between tamoxifen and estradiol valerate treatments.

We have investigated a putative role for local estrogen in the central regulation of GH secretion by exploiting the different tissue-specific properties of estradiol valerate and tamoxifen and comparing their respective effects on the GH-IGF-I axis. Both compounds exert a hepatic estrogen agonistic effect, whereas only tamoxifen imparts a central estrogen antagonist effect. The liver is a major source of circulating IGF-I and SHBG production. The finding of a fall in circulating IGF-I and a rise in SHBG in the present study confirms a hepatic estrogen agonist effect of tamoxifen, in line with the expected change induced by estradiol valerate (5–8, 12–14, 18).

Estradiol and tamoxifen exert opposite effects on GH secretion regulation

The most notable finding is the striking difference between estradiol valerate and tamoxifen on the GH response to arginine stimulation. Estradiol valerate enhanced, whereas tamoxifen suppressed the GH response. IGF-I exerts feedback inhibition of GH secretion (10). Thus, the increase in GH release induced by estradiol valerate is likely a consequence of the fall in circulatory IGF-I. The dissociation of the GH-IGF-I axis is a well-recognized phenomenon arising from the consequence of a first-pass effect of estrogen on hepatic IGF-I production (Fig. 3A) (5, 7). However, with tamoxifen, we observed a suppression instead of stimulation in GH response (Fig. 3B). Thus, the marked blunting of GH secretion in the face of reduced IGF-I feedback occurred with central estrogen receptor antagonism, indicating mediation by estrogen.

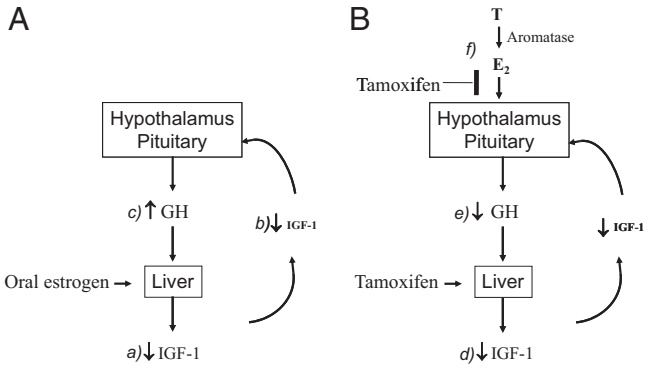

FIG. 3. A, Estrogen administered via oral route acts on the liver to reduce IGF-I production (a). The reduction in IGF-I lessens negative feedback to the hypothalamus and pituitary gland (b), and GH secretion is stimulated (c). B, Tamoxifen treatment also reduces IGF-I levels (d); however, GH secretion is attenuated (e). The finding of a blunted GH response to stimulation despite reduced IGF-I feedback inhibition indicates central suppression of GH output by tamoxifen and an important role of local estrogen in the neuroregulation of GH secretion in women (f). T, Testosterone; E2, estradiol.

What is the source of estrogen that underlies central stimulation of GH secretion, endocrine or local?

In this study, it is conceivable that estrogen derived from estradiol valerate in the systemic circulation enhanced GH secretion. However, because estradiol valerate reduced IGF-I, it is not possible to deduce whether the increase in GH secretion arose from direct estradiol stimulation or from reduced feedback inhibition. A limitation of the present study is the absence of a parenteral estradiol treatment arm. However, studies of parenteral estradiol replacement avoiding hepatic perturbation, achieving physiological concentration in peripheral blood have not observed enhancement in GH secretion (5, 7). Thus, the evidence for an endocrine drive by estradiol on GH secretion in women is poor.

Can local estrogen be driving GH secretion?

There is emerging evidence that paracrine mechanisms play a major role in the regulation of the endocrine system. Studies in men have firmly established that local estradiol derived from aromatization of testosterone regulates GH and gonadotropin secretion (3, 4, 19). There is strong evidence that estradiol derived from circulatory testosterone regulates local tissue function and growth in women, as demonstrated by clinical efficacy of aromatase inhibitor therapy for breast cancer. Because circulating estradiol was unaffected (data not shown), we propose that the blunting effect of tamoxifen arises from antagonism of estrogens produced locally from aromatization (Fig. 3B). The contrasting effects on GH status between systemic estrogen supplementation and receptor blockade suggest a paracrine mechanism and an important role of aromatase in the neuroregulation of GH secretion in women.

Clinical implications

Our findings of a possible paracrine mechanism of GH secretion in women is of clinical importance. If local estrogens play a major role in determining GH secretion, treatment with agents that block central estrogen receptors or tissue estrogen availability may result in a reduction in GH levels. It is possible that the antimitogenic effect of tamoxifen or of aromatase inhibitors is mediated in part by a down-regulation of the GH/IGF-I system. GH also exerts potent metabolic effects stimulating fat oxidation and protein anabolism, reducing fat mass, and increasing lean body mass (20, 21). Therefore, depletion of local estrogen action centrally may result in unfavorable changes in body composition and possible adverse metabolic outcomes.

Regulation of GH secretion by estrogens

Release of GH from pituitary somatotropes results from a complex interaction primarily between the stimulatory GHRH and the inhibitory somatostatin (SST). Estrogen may stimulate GH secretion by acting directly on the pituitary gland (22, 23) or the hypothalamus (24, 25). *In vitro* studies show that estrogens may regulate GH secretion by modulating the activity of hypothalamic GHRH neurons because about 70% of GHRH neurons express estrogen receptors (26). Pituitary somatotropes express high levels of SST receptor 5 mRNA (27), which are reduced when pituitary cell cultures are treated with estradiol (28, 29). Thus estrogen, by reducing SST tone, may exert a stimulatory effect on GH secretion. Strong evidence for a role of local estrogen in the regulation of GH secretion comes from the aromatase knockout mice model. In the aromatase knockout mice, GH levels are low and somatotropes are hypoplastic (30). In men with inactivating mutations of aromatase gene, GH response to stimulation is reduced (31). Thus, there is strong evidence of an important role of aromatase in the regulation of somatotroph function. We are currently undertaking studies investigating the impact of aromatase inhibitors on GH secretion.

In summary, tamoxifen reduced but estradiol valerate enhanced GH response to arginine in postmenopausal women. Both tamoxifen and estradiol valerate reduced IGF-I and increased SHBG levels, indicative of a hepatic estrogen agonist effect of tamoxifen. The finding of a blunted GH response to stimulation in the face of reduced IGF-I feedback inhibition indicates that tamoxifen inhibits its central GH output. Because circulating estradiol was unaffected, these data indicate a significant role of local estrogen in the central control of GH secretion. We conclude that aromatase mediates the paracrine control of GH secretion in women.

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References

1. **Kamegai J, Tamura H, Shimizu T, Ishii S, Tatsuguchi A, Sugihara H, Oikawa S, Kineman RD** 2004 The role of pituitary ghrelin in growth hormone (GH) secretion: GH-releasing hormone-dependent regulation of pituitary ghrelin gene expression and peptide content. Endocrinology 145:3731–3738
2. **Pitteloud N, Dwyer AA, DeCruz S, Lee H, Boepple PA, Crowley Jr WF, Hayes FJ** 2008 Inhibition of luteinizing hormone secretion by testosterone in men requires aromatization for its pituitary but not its hypothalamic effects: evidence from the tandem study of normal and gonadotropin-releasing hormone-deficient men. J Clin Endocrinol Metab 93:784–791
3. **Weissberger AJ, Ho KK** 1993 Activation of the somatotropic axis by testosterone in adult males: evidence for the role of aromatization. J Clin Endocrinol Metab 76:1407–1412
4. **Veldhuis JD, Metzger DL, Martha Jr PM, Mauras N, Kerrigan JR, Keenan B, Rogol AD, Pincus SM** 1997 Estrogen and testosterone, but not a nonaromatizable androgen, direct network integration of the hypothalamo-somatotrope (growth hormone)-insulin-like growth factor I axis in the human: evidence from pubertal pathophysiology and sex-steroid hormone replacement. J Clin Endocrinol Metab 82:3414–3420
5. **Weissberger AJ, Ho KK, Lazarus L** 1991 Contrasting effects of oral and transdermal routes of estrogen replacement therapy on 24-hour growth hormone (GH) secretion, insulin-like growth factor I, and GH-binding protein in postmenopausal women. J Clin Endocrinol Metab 72:374–381
6. **Gibney J, Johannsson G, Leung KC, Ho KK** 2005 Comparison of the metabolic effects of raloxifene and oral estrogen in postmenopausal and growth hormone-deficient women. J Clin Endocrinol Metab 90:3897–3903
7. **Bellantoni MF, Vittone J, Campfield AT, Bass KM, Harman SM, Blackman MR** 1996 Effects of oral versus transdermal estrogen on the growth hormone/insulin-like growth factor I axis in younger and older postmenopausal women: a clinical research center study. J Clin Endocrinol Metab 81:2848–2853
8. **Sonnet E, Lacut K, Roudaut N, Mottier D, Kerlan V, Oger E** 2007 Effects of the route of oestrogen administration on IGF-1 and IGFBP-3 in healthy postmenopausal women: results from a randomized placebo-controlled study. Clin Endocrinol (Oxf) 66:626–631
9. **Leung KC, Doyle N, Ballesteros M, Sjogren K, Watts CK, Low TH, Leong GM, Ross RJ, Ho KK** 2003 Estrogen inhibits GH signaling by suppressing GH-induced JAK2 phosphorylation, an effect mediated by SOCS-2. Proc Natl Acad Sci USA 100:1016–1021
10. **Chapman IM, Hartman ML, Pieper KS, Skiles EH, Pezzoli SS, Hintz**

RL, Thorner MO 1998 Recovery of growth hormone release from suppression by exogenous insulin-like growth factor I (IGF-I): evidence for a suppressive action of free rather than bound IGF-I. J Clin Endocrinol Metab 83:2836–2842
11. **Riggs BL, Hartmann LC** 2003 Selective estrogen-receptor modulators—mechanisms of action and application to clinical practice. N Engl J Med 348:618–629
12. **Löfgren L, Wallberg B, Wilking N, Fornander T, Rutqvist LE, Carlström K, von Schoultz B, von Schoultz E** 2004 Tamoxifen and megestrol acetate for postmenopausal breast cancer: diverging effects on liver proteins, androgens, and glucocorticoids. Med Oncol 21:309–318
13. **De Marinis L, Mancini A, Izzi D, Bianchi A, Giampietro A, Fusco A, Liberale I, Rossi S, Valle D** 2000 Inhibitory action on GHRH-induced GH secretion of chronic tamoxifen treatment in breast cancer. Clin Endocrinol (Oxf) 52:681–685
14. **Mandalà M, Moro C, Ferretti G, Calabro MG, Nolè F, Rocca A, Munzone E, Castro A, Curigliano G** 2001 Effect of tamoxifen on GH and IGF-1 serum level in stage I-II breast cancer patients. Anticancer Res 21:585–588
15. **Kelly JJ, Rajkovic IA, O'Sullivan AJ, Sernia C, Ho KK** 1993 Effects of different oral oestrogen formulations on insulin-like growth factor-I, growth hormone and growth hormone binding protein in postmenopausal women. Clin Endocrinol (Oxf) 39:561–567
16. **Wolthers T, Hoffman DM, Nugent AG, Duncan MW, Umpleby M, Ho KK** 2001 Oral estrogen antagonizes the metabolic actions of growth hormone in growth hormone-deficient women. Am J Physiol Endocrinol Metab 281:E1191–E1196
17. **Gibney J, Wolthers T, Johannsson G, Umpleby AM, Ho KK** 2005 Growth hormone and testosterone interact positively to enhance protein and energy metabolism in hypopituitary men. Am J Physiol Endocrinol Metab 289:E266–E271
18. **Corsello SM, Rota CA, Putignano P, Della Casa S, Barnabei A, Migneco MG, Vangeli V, Barini A, Mandalà M, Barone C, Barbarino A** 1998 Effect of acute and chronic administration of tamoxifen on GH response to GHRH and on IGF-I serum levels in women with breast cancer. Eur J Endocrinol 139:309–313
19. **Veldhuis JD, Mielke KL, Cosma M, Soares-Welch C, Paulo R, Miles JM, Bowers CY** 2009 Aromatase and 5α-reductase inhibition during an exogenous testosterone clamp unveils selective sex steroid modulation of somatostatin and growth hormone secretagogue actions in healthy older men. J Clin Endocrinol Metab 94:973–981
20. **Jorgensen JO, Pedersen SA, Thuesen L, Jorgensen J, Ingemann-Hansen T, Skakkebaek NE, Christiansen JS** 1989 Beneficial effects of growth hormone treatment in GH-deficient adults. Lancet 1:1221–1225
21. **Burt MG, Gibney J, Hoffman DM, Umpleby AM, Ho KK** 2008 Relationship between GH-induced metabolic changes and changes in body composition: a dose and time course study in GH-deficient adults. Growth Horm IGF Res 18:55–64
22. **Komolov IS, Perez-Arce JA, Fedotov VP** 1980 The effects of estradiol on prolactin and growth hormone secretion in cultured pituitary cells from intact and ovariectomized rats. Endokrinologie 75:278–284
23. **Simard J, Hubert JF, Hosseinzadeh T, Labrie F** 1986 Stimulation of growth hormone release and synthesis by estrogens in rat anterior pituitary cells in culture. Endocrinology 119:2004–2011
24. **Hassan HA, Enright WJ, Tucker HA, Merkel RA** 2001 Estrogen and androgen elicit growth hormone release via dissimilar patterns of hypothalamic neuropeptide secretion. Steroids 66:71–80
25. **Cosma M, Bailey J, Miles JM, Bowers CY, Veldhuis JD** 2008 Pituitary and/or peripheral estrogen-receptor α regulates follicle-stimulating hormone secretion, whereas central estrogenic pathways direct growth hormone and prolactin secretion in postmenopausal women. J Clin Endocrinol Metab 93:951–958
26. **Shimizu T, Kamegai J, Tamura H, Ishii S, Sugihara H, Oikawa S** 2005 The estrogen receptor (ER) α, but not ER β, gene is expressed in hypothalamic growth hormone-releasing hormone neurons of the adult female rat. Neurosci Res 52:121–125
27. **O'Carroll AM, Krempels K** 1995 Widespread distribution of somatostatin receptor messenger ribonucleic acids in rat pituitary. Endocrinology 136:5224–5227
28. **Djordjijevic D, Zhang J, Priam M, Viollet C, Gourdji D, Kordon C, Epelbaum J** 1998 Effect of 17β-estradiol on somatostatin receptor expression and inhibitory effects on growth hormone and prolactin release in rat pituitary cell cultures. Endocrinology 139:2272–2277
29. **Kimura N, Tomizawa S, Arai KN, Kimura N** 1998 Chronic treatment with estrogen up-regulates expression of sst2 messenger ribonucleic acid (mRNA) but down-regulates expression of sst5 mRNA in rat pituitaries. Endocrinology 139:1573–1580
30. **Yan M, Jones ME, Hernandez M, Liu D, Simpson ER, Chen C** 2004 Functional modification of pituitary somatotropes in the aromatase knockout mouse and the effect of estrogen replacement. Endocrinology 145:604–612
31. **Rochira V, Zirilli L, Maffei L, Premrou V, Aranda C, Baldi M, Ghigo E, Aimaretti G, Carani C, Lanfranco F** 2010 Tall stature without growth hormone: four male patients with aromatase deficiency. J Clin Endocrinol Metab 95:1626–1633