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Growth hormone receptor modulators

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Abstract Growth hormone (GH) regulates somatic growth, substrate metabolism and body composition. Its actions are elaborated through the GH receptor (GHR). GHR signalling involves the role of at least three major pathways, STATs, MAPK, and PI3-kinase/Akt. GH receptor function can be modulated by changes to the ligand, to the receptor or by factors regulating signal transduction. Insights on the physico-chemical basis of the binding of GH to its receptor and the stoichiometry required for activation of the GH receptor-dimer has led to the development of novel GH agonists and antagonists. Owing to the fact that GH has short half-life, several approaches have been taken to create long-acting GHR agonists. This includes the pegylation, sustained release formulations, and ligand-receptor fusion proteins. Pegylation of a GH analogue (pegvisomant) which binds but not activate signal transduction forms the basis of a new successful approach to the treatment of acromegaly. GH receptors can be regulated at a number of levels, by modifying receptor expression, surface availability and signalling. Insulin, thyroid hormones and sex hormones are among hormones that modulate GHR through some of these mechanisms. Estrogens inhibit GH signalling by stimulating the expression of SOCS proteins which are negative regulators of cytokine receptor signalling. This review of GHR modulators will cover the effects of ligand modification, and of factors regulating receptor expression and signalling.

Keywords Growth hormone · GH receptor · Signalling · Insulin · Sex steroids

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1 Introduction

GH regulates somatic growth, substrate metabolism and body composition. Its actions are elaborated through the GH receptor (GHR), a member of the cytokine superfamily that includes receptors for prolactin, erythropoetin, leptin, and the interleukins. Since the cloning of the GHR over 20 years ago in 1987, major advances have been made in the understanding of its structure and function providing insights into the mechanism of GH action on cells, the signalling pathways and their regulation. Work on the physico-chemical basis of the binding of GH to its receptor and the stoichiometry required for receptor activation has led to the development of GH analogues.

These collective findings have indicated that the action of GH can be regulated at a number of levels, by modifying ligand, receptor expression and signalling. This review of GHR modulators will cover the effects of ligand modification, and of factors regulating receptor expression and signalling.

2 GHR structure and signalling

The crystallization of the extracellular domain of the GHR revealed a 1:2 stoichiometric relationship of GH to its receptor [1], indicating that dimerization of GHR is an initial and crucial event in GH signalling (Fig. 1a). GH is a four helix bundle with an unusual topology and GH binding to GHR is mediated by two asymmetric binding sites on GH [2]. In early models, GH binding to GHR monomers was thought to be sequential. The initial step of GH binding to its receptor involves high-affinity binding of site 1 to one GHR monomer followed by lower affinity binding of site 2 to a second GHR monomer [3]. Recent studies indicate that GHRs exist as pre-formed dimer, as is also the case

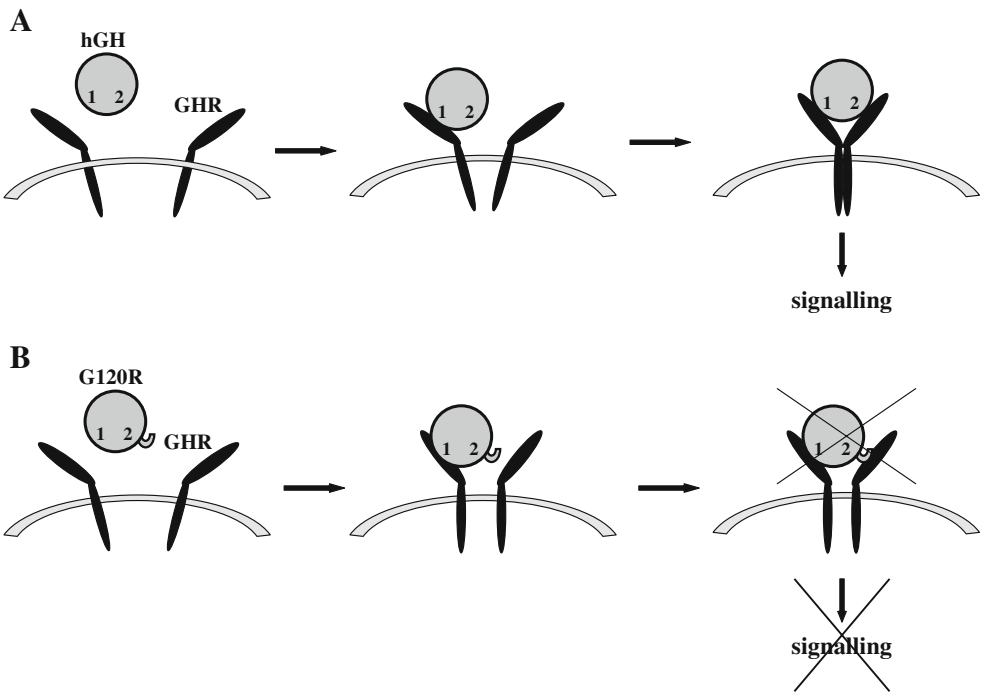

Fig. 1 The figure shows proposed principle of antagonism by B2036, a GHR antagonist. **a** Schematic representation of normal GH signalling, in which GH binds to two identical cell-surface receptors, resulting in receptor dimerization. **b** Schematic mechanism by which B2036 antagonizes GH signalling. The GH-receptor antagonist has an amino acid substitution in the region of site-2 binding that disrupts binding of GH to the second GH receptor. Dimerization is prevented and the receptor is blocked. Since the original proposal, it is now recognised that the GHR exists as preformed dimers and that ligand binding results in a conformational change that triggers signalling and this does not happen with B2036

for other class I cytokine receptors, such as the erythropoietin receptor. A conformational change in the extracellular domain of the GHR is triggered by GH binding which initiates signalling [4].

In common with cytokine receptors, the GHR is devoid of enzymatic activity with signal transduction mediated by Janus kinase (JAK) 2 [5, 6]. JAK2 activation is triggered by GH binding which induces conformational change of the GHR resulting in JAK2 transphosphorylation and catalytic activation. The phosophorylation of the receptor results in the activation of a number of signalling pathways. The JAK–STAT pathway is a major effector of GHR signalling, and necessary for the transcriptional regulation of IGF-I. The mitogen activated protein kinase (MAPK) pathway, and the phosphatidylinositol 3′-kinase (PI3K) pathway are also activated by JAK2 transphosphorylation (Fig. 2) [7]. The termination of GHR signalling is an important mechanism for controlling GH action. This is controlled by two systems, the suppressors of cytokine signalling (SOCS) proteins and the protein tyrosine phosphatases (PTPs). GH induces the expression of SOCS-1, SOCS-2, and SOCS-3, which feed back to inhibit its transcriptional action [8, 9]. SOCS2 deficient mice displayed an excessive growth phenotype [10]. Among the PTPs, SHP1 and SHP-2 inactivate the receptor by dephosphorylating JAK2 [11].

3 GHR function

Defective signalling arising from mutations of the GHR cause growth retardation. The syndrome of GH insensitivity (GHIS) was first identified in 1966 by Laron et al. in three siblings with severe growth retardation, manifesting high level of GH in circulation [12, 13]. A structural defect in GHR gene was first described in 1989 [14, 15]. Since then more than 70 GHR gene mutations have been identified [15]. The majority of the GHR abnormalities are located in the extracellular domain of the receptor [16–19]. GHR

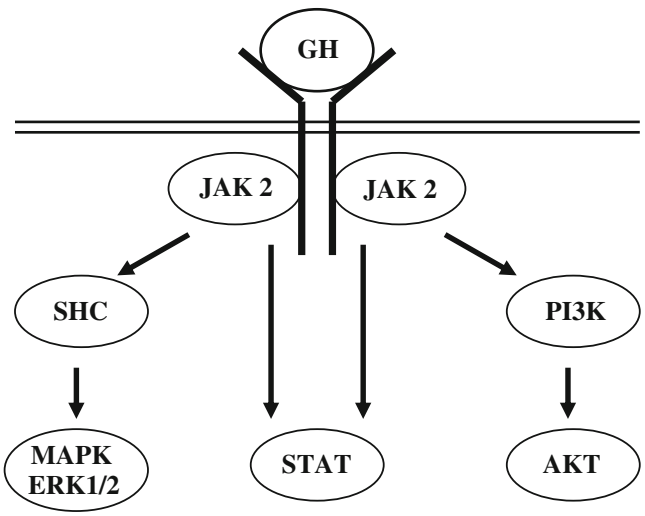

Fig. 2 This schematic diagram shows major GH receptor signalling pathways. GH binding to the GHR induces conformational change of the GHR, which activates JAK2. The phosphorylated JAK2 initiates a multitude of signalling cascades including major signalling pathways such as JAK/STAT, PI3K/AKT and MAPK pathways. Among them JAK/STAT pathway is critical for a variety of GH functions and necessary for the transcriptional regulation of IGF-I

mutations can affect the ability of the receptor to either bind GH, dimerize, anchor or migrate to the cell membrane. There are two reports of mutations of GHR gene that result in a selective loss of STAT5b signalling [20, 21].

Recent studies have shown that GHRs exist as pre-formed dimers that activate signal transduction following conformational change induced ligand binding. Interestingly a number of monoclonal antibodies directed to the GHR manifest agonistic activity. Some monoclonal antibodies against the GHR exhibit prolonged activation of the GHR [4]. The best characterised of these is mAb 263. It does not bind within the hormone binding surface but binds in a way that promotes the conformational change necessary to activate the signalling cascade [22].

Knowledge of the physical chemical properties of the binding sites has led to engineering of the molecules to produce agonist and antagonist by amino acid substitution that enhances or inhibits binding.

4 GHR ligands

4.1 GHR agonists

Growth hormone is the only natural ligand of the GHR. GH is a 191-amino acid, single chain 22 kDA polypeptide hormone, consisting of four helical structures. Two domains within the GH molecule are involved in receptor binding (see [23]). The current regimen for growth hormone replacement requires once-daily injections, which is inconvenient. Several approaches have been taken to create long-acting preparations. This includes pegylated hormones, sustained release formulations, ligand-receptor fusion proteins, and GH analogs.

4.1.1 Pegylated GH modification

Pegylation (covalent attachment of polyethylene glycol polymer chains to another molecule; PEG) increases the plasma half-life of GH by reducing renal clearance and intravascular proteolysis [24, 25]. Despite a reduction in GHR affinity, the *in vivo* efficacy of pegylated GH increases with higher level of pegylated modification and reaches an optimum at five PEG_{5000} groups per hGH [24]. In hypophysectomized rat model, injections of pegylated GH analogs increased weight gain by about 10-fold compared to that of unpegylated hGH [24]. Pegylated GH analogs are currently being evaluated in human trials. Although longer acting, pegylation reduces receptor affinity and therefore a greater dose of hormone is required. As the cost involved is high, sustained release GH formulations have also been developed as another strategy to prolong effect of GH.

4.1.2 Sustained release formulations

This approach is based on the encapsulation of GH in microspheres of biodegradable copolymers. Two sustained release preparations, Nutropin Depot® [26–29] and hGH-Biosphere® [30] have been studied the most. The latter has a superior release profile based on the IGF-I generated per mass of administered GH. Nutropin Depot® increase serum levels of GH and IGF-I in adults and children with GH deficiency [26–28, 31]. The catch-up growth observed in those children was significant, although to a lesser degree than with daily GH injections. In GH deficient adults, 8 months treatment with Nutropin Depot® decreased trunk and visceral adipose tissue and increased lean body mass as effectively as with daily GH administration [29].

However there are certain problems arising from sustained release microspheres, including initial high release and those arising from degradation of microspheres, such as acidic microenvironment and protein denaturation. Therefore other sustained release GH formulations have been developed based on hydroxyethyl methacrylated dextran [32] or sodium hyaluronate microparticles [33, 34]. However whether these are superior to Nutropin Depot® or hGH-Biosphere® is not yet known.

4.1.3 Ligand–receptor fusion proteins

The extracellular domain of the GHR is proteolytically cleaved and circulates as a GH binding protein (GHBP). When bound to GHBP, GH has delayed clearance and degradation prolonging its half life [35]. GHBP when co-administered together with GH, augments the effect of GH on weight gain and bone growth in rat models of GH deficiency [36]. Ross and colleagues [37] have fused recombinant human GH with the GHBP via a flexible linker. The clearance of the fusion protein was 300 times slower than that of GH after bolus injection in the rat, and the terminal half-life was a 100-times longer than that of GH. The authors reported that a single injection of the ligand-receptor fusion results in a weight gain of hypophysectomized rats over 10 days that was equivalent to that obtained with an equimolar dose of growth hormone injected daily. The administration of ligand-receptor fusion protein also resulted in IGF-I concentrations that were significantly greater than those seen after daily injection of GH. Thus, the ligand-receptor fusion of 75 kDa is more potent than hGH and seems promising as a potential therapeutic formulation.

4.1.4 GH analogs

One way of increasing hGH potency is by introducing mutations at the GH binding sites to enhance its binding affinity [38]. Waters and colleagues have reported that

increase in site 1 binding affinity of porcine GH improves biopotency [23, 39]. The increased biopotency of GH can be explained by a model for GH receptor activation where subunit alignment is critical for effective signalling. Substitution of four different residues in site 1 from human GH into porcine GH, increased cell proliferation when compared to porcine GH [39]. The higher potency is linked to a decreased dissociation rate between the ligand and the receptor. Thus, selected amino acid substitution in critical GH binding sites can lead to prolonged activation of the receptor and improve GH potency. Conversely, engineered amino acid substitution that result in reduced binding has been used to develop GH antagonists.

4.2 GHR antagonists

4.2.1 Development of GHR antagonists

Chen et al. pioneered the development of GH antagonist by engineering a mutation in site 2 of GH to reduce affinity to the GHR [40]. Based on early studies showing that position 120 of hGH was crucial to GH binding to the GHR (Fig. 1b) [41–43], a glycine to lysine substitution resulted in the generation of an analogue which antagonized GH-induced JAK2 activation and downstream tyrosine phosphorylation [44, 45].

Previously eight amino acids had been identified that when altered, increased the binding affinity of GH site 1 to the GHR [46]. When combined with the G120K alteration, the mutations that enhance site 1 affinity resulted in a potent antagonist, B2036. Pegylation of B2036 (B2036-PEG) increased half-life of B2036 to 72-h compared with 16 min for native GH and lowered immunogenicity. B2036-PEG (pegvisomant) binds to the GH receptor and induced receptor internalization [47]. It was introduced for human trials in the late 1990s and since then has been established as a safe and effective treatment for acromegaly [48].

Although pegvisomant effectively lowers IGF-I levels in acromegaly, a disadvantage is as administered by daily injections. Orally active GHR antagonists are under development. BVT-A ((*N*-[5-(aminosulfonyl)-2-methylphenyl]-5-bromo-2-furamide), is a small molecule which shows promise as a GH receptor antagonist *in vivo*. The small molecular weight compound down-regulates GH-stimulated IGF-I expression [49]. Administration of BVT-A suppresses serum IGF-I, hepatic mRNA levels of IGF-I, IGF-BP3, ALS, and the IGF-I and GH receptors in hypophysectomized rats [50].

4.2.2 Pegvisomant

Several studies have established pegvisomant as effective treatment of acromegaly. Daily injection of 40 mg of pegvisomant blocks the growth hormone-mediated generation of IGF-I in approximately 90% of patients, and improves soft-tissue manifestations of the disease [51–54]. Pegvisomant also improves glucose tolerance and insulin sensitivity in acromegaly [55–59]. Pegvisomant treatment is accompanied by a dose-dependent and reversible rise in GH concentration [51]. The cause has not been elucidated and could be the result of increased GH secretion or delayed clearance.

Concern has been raised as to whether the increase in circulating GH level represents tumor growth as pegvisomant dose not act directly at the tumor. Although no significant increase in tumor size was observed over 12 months of observation in a large group of patients [51, 52], longer term studies are required to ascertain whether the tumor growth is affected by pegvisomant treatment.

The therapeutic potential of pegvisomant has been explored in a number of disease states where GH or GH-dependent growth factors are thought to have a pathogenic role. Animal studies have shown that pegvisomant limits the degree of diabetic glomerulopathy [60]. Since pegvisomant has sustained suppressive effect on IGF-I and IGF-II [61], the therapeutic potential of pegvisomant in anti-cancer treatment for IGF dependent cancers, such as breast and colorectal cancer, are being investigated. Recently pegvisomant has been reported to inhibit the growth of meningioma [62], colon cancer [63] and breast cancer cells in rodents [64].

5 Factors modulating the GH receptor

Many factors are known to regulate the responsiveness of the GH receptor to GH. The most important are insulin, thyroid and sex hormones. The effects on GH receptor expression and function will be reviewed.

5.1 Insulin

The growth-promoting action of GH is mediated by IGF-I which is produced mainly in the liver, but also in extra-hepatic tissues. There is strong evidence that the anabolic action of GH requires the presence of insulin and adequate nutrition. This is exemplified in type 1 diabetes where IGF-I levels are low and longitudinal growth is impaired despite high serum levels of GH [65, 66]. These abnormalities are corrected by insulin treatment [67, 68].

5.1.1 Insulin effect on GH receptor expression

The effects of insulin on GHR expression and function are tissue specific. In cultures of rat hepatoma cells, insulin increases GHRs [69]. In animal studies, insulin deficiency

results in a decrease of GH binding and GHR expression in liver [70, 71], which can be reversed by insulin administration [70, 72]. In extra-hepatic tissues such as bone and kidney, there is evidence that insulin down-regulates GHRs [60, 70–72].

It is well established that surface membrane receptors are dynamically regulated, with cell surface abundance representing the net balance of recycling of internalised receptors and translocation of newly synthesized receptors to the cell membrane. There is recent evidence that the surface translocation of GH receptors is inhibited by insulin. Insulin dose-dependently stimulates liver GHR synthesis and GH binding (Fig. 3a), however increasing insulin concentrations reduce GHR surface translocation (Fig. 3b), which overcomes the effect on receptor synthesis [69]. These findings show that the mechanism by which insulin regulates tissue responsiveness to GH is complex and in part mediated by effects on GHR expression and surface translocation. Decrease in receptor surface availability with high dose insulin may represent rapid mechanism for insulin regulation of the GHR function.

In human studies, there is also evidence that insulin modulates the expression of GHRs. This is based on measurement of circulatory levels of GHBP. As GHBP is derived from proteolytic cleavage of the extracellular domain of the GH receptor, change in GHBP levels may reflect GH receptor status [73]. Low blood levels of GHBP occur in conditions associated with GH resistance such as malnutrition and catabolic states. This is exemplified in anorexia where GH levels are elevated, and levels of GHBP are low [74, 75]. Thus when insulin levels are low, high levels of GH does not translate into a rise in circulating IGF-I [76–82]. In type I diabetes, GHBP levels are low and associates with low IGF-I levels [83]. These investigations have also observed a significant positive correlation between levels of GHBP and total insulin dose, suggesting that GHR status in humans is dependent on adequate insulinisation [83].

In contrast, high levels of GHBP are associated with hyperinsulinaemia and obesity, with GHBP falling significantly in the obese after weight loss, with normalization of insulin levels [84, 85]. GHBP levels correlate significantly with fat mass, and because adipocytes express GHRs, it is possible that elevated GHBP levels simply reflect an increase in fat mass [84].

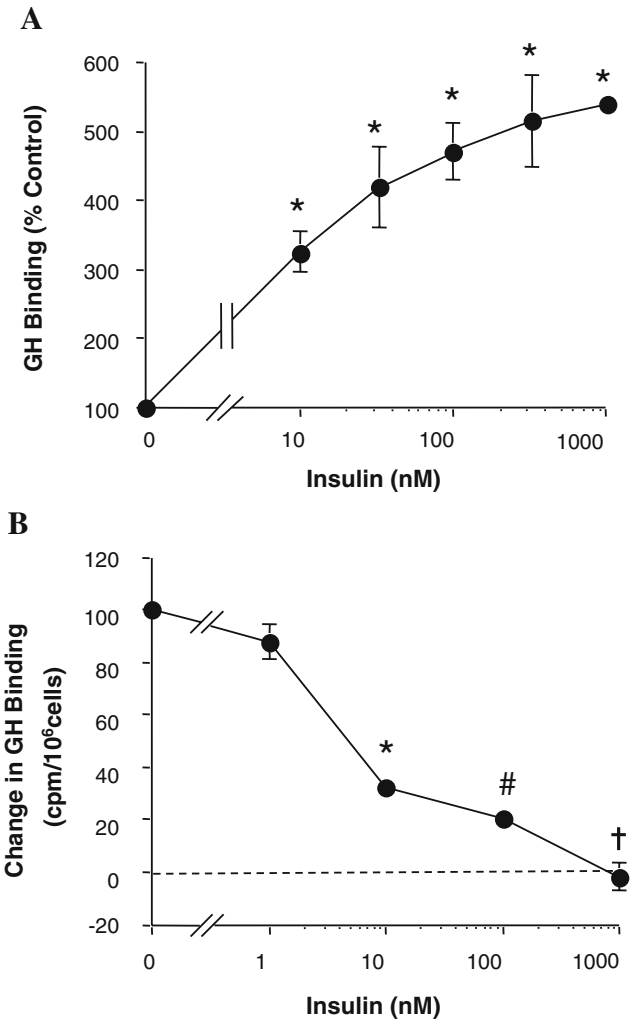

Fig. 3 Effect of insulin on intracellular GH binding and GH receptor surface translocation in human hepatoma cells (HuH7). **a** Effect of insulin on intracellular GH binding. Cells were treated with insulin at the indicated concentrations for 18 h and then GH binding was determined using ^{125}I-labeled human GH. Intercellular GH binding increased with insulin in a dose dependent manner. GH binding was expressed as percent from control. Significance vs. control: *$p<$ 0.0001. **b** Reduction in GHR surface translocation to the cell with insulin in a dose dependent manner. Cells were treated with insulin at the indicated concentrations for 15 min and then allowed to recover for 4 h. The GH binding was set up before and after the recovery. The GHR translocation was measured as the recovery of GH-binding activity of whole cells after removal of the surface GHRs by trypsin treatment. Significance vs. control: *p=0.005; #p<0.002; †p<0.0005. Adapted from [69]. Copyright 2000, The Endocrine Society

5.1.2 Insulin effect on GH receptor signalling

There is also strong evidence that insulin modulates GHR signalling in addition to the effects on receptor expression and surface translocation. In rat hepatoma cells, low dose insulin administration results in GH-induced stimulation of JAK2 phosphorylation however high dose insulin treatment results in inhibitory effect [69, 86]. The effect of insulin on GHR function appears to be mediated by the PI-3 kinase and MAPK/ERK pathways [69, 87, 88]. It has been shown that insulin increases GH signalling by enhancing GH-induced activation of MAPK/ERK pathway through post signalling cross-talk [88].

In summary, insulin regulates GHR expression, translocation and GHR function. The regulation of GH receptor

expression is complex and tissue dependent. Insulin stimulates hepatic GHR synthesis and GH binding but down-regulates GHR expression in kidney and bone tissue. In liver, high concentrations of insulin reduce GHR surface translocation, in such way regulating receptor surface availability. The effects of insulin on GHR function are mediated by stimulation of GH-induced JAK2 phosphorylation, PI-3 kinase and MAPK/ERK pathways.

5.2 Thyroid hormones

Thyroid hormones are necessary for GH dependent growth and development. Hypothyroidism in children result in impaired growth, low circulating IGF-I levels and impaired GH secretion [89]. In the hypopituitary child, GH treatment fails to normalize growth unless thyroid hormones are replaced [90].

There is strong evidence that thyroid hormones modulate the expression of GHRs. *In vitro* studies show that triiodothyronine dose-dependently upregulates GHR gene expression in human hepatoma cells [91]. In animal models, hypothyroidism is associated with decrease in liver GHR mRNA expression and GH binding, and the changes are restored by thyroxine treatment [92–94]. In human studies, the circulating level of GHBP, which may reflect GHR status, is strongly correlated to thyroid hormone status with low levels found in hypothyroidism and high in hyperthyroidism [95, 96]. Thus the evidence indicate that GHRs are positively regulated by thyroid hormones in animals and humans.

Thyroid hormones not only stimulate pituitary and liver GHR expression but also stimulate GH gene transcription [97–100] enhancing GH secretion [94, 101–103]. Thus thyroid hormones regulate the GH system through two independent mechanisms, one involving GH gene expression and the other through regulation of GH receptor expression.

To the best of our knowledge, interaction between thyroid hormones and GHRs on the signalling level has not been elucidated.

5.3 Estrogen

There is a close interaction between estrogens and GH in the regulation of growth and development. There is evidence that estrogen impairs the action of GH. Women are less responsive than men to GH treatment [104]. Estrogen administered by the oral route to hypopituitary patients suppresses GH stimulation of lipid oxidation and protein metabolism, and in postmenopausal women increases body fat and reduces lean mass [105–107]. However these metabolic and body composition effects are not seen with transdermal estrogen administration, suggesting that liver is the major site of regulatory control by estrogen.

Estrogens affects the expression and function of GHRs. In animals, the effect of estrogen receptor expression is dependent on tissue type and species. It reduces expression of GHRs in the liver of rabbits [108, 109], but exerts an opposite effect in rodents [110–112]. In rat osteosarcoma cells and human osteoblast-like cells, estrogen stimulates GH binding and GHR mRNA expression [113]. Osteoblast proliferation is enhanced by GH co-treatment with estrogen. Thus estrogen may potentiate the effect of GH on bone formation.

In human, oral estrogen administration leads to a reduction in IGF-I levels despite an increase in GH. This observation suggests that estrogen impairs the ability of GH to stimulate hepatic IGF-I production, indicating an inhibitory effect on GHR function. As discussed under GHR structure and signalling section, the JAK-STAT pathway is a major effector of GHR signalling, necessary for the transcriptional regulation of IGF-I. Estrogen inhibits GH activation of the JAK/STAT pathway. The inhibition is dose-dependent and results from suppression of GH-induced JAK2 phosphorylation, leading to reduction in transcriptional activity (Fig. 4; [114]). Estrogen does not affect phosphatase activity but stimulates expression of SOCS-2, which in turn inhibits JAK2 activation (Fig. 4). Thus, esotrogen inhibits GH receptor signalling by stimulating SOCS-2 expression (Fig. 5).

In summary, the effects of estrogen on GHRs depend on tissue type, species and route of administration. Estrogen inhibits GHR signalling by stimulating expression of SOCS-2, which in turn inhibits JAK2 phosphorylation providing a mechanism that explains inhibitory effect of estrogen on GH action.

5.4 Testosterone

Testosterone exerts growth-promoting effect in part by stimulating the GH-IGF-I system [115–119]. Testosterone enhances the secretion of GH [120], an effect mediated at the hypothalamic level by stimulation of GH releasing hormone [121]. Thus, one mechanism how testosterone regulates GH system is through stimulation of GH secretion by testosterone. However, there is some evidence that testosterone can modify GHRs directly [122].

Animal studies show that in castrated or hypophysectomized female and male rats, testosterone treatment for 2 weeks does not significantly change hepatic GHR mRNA expression or GH binding [111–123]. However in male rabbits, testosterone induced elevation in hepatic and growth plate GHR mRNA levels [108]. Testosterone significantly increased GHR mRNA in epiphyseal growth plates of hypophysectomized rats [123]. Thus peripheral action of GH on the growth plate may be modulated by testosterone by enhancing GHR expression.

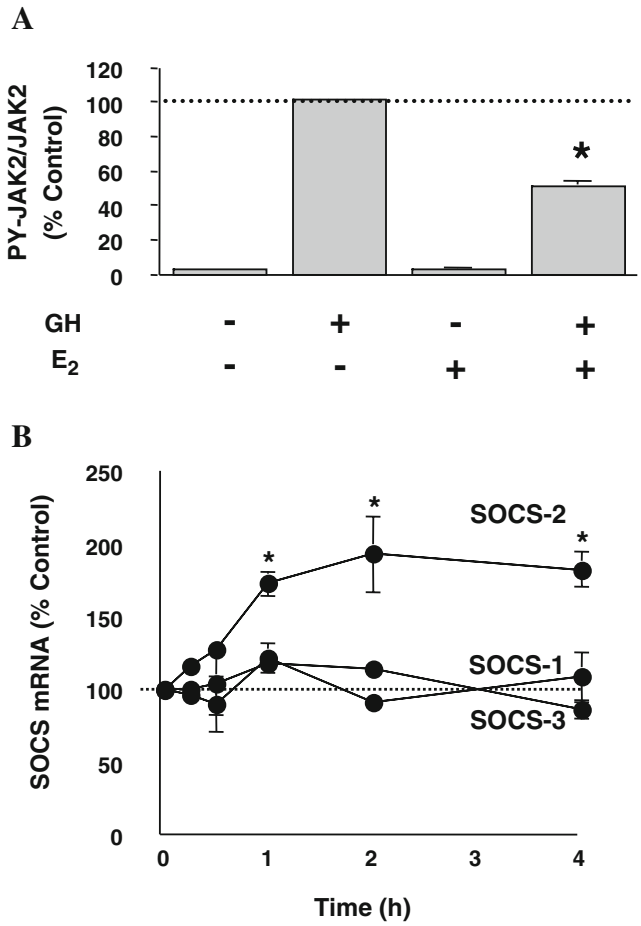

Fig. 4 The figure shows estrogen (E_2) effects on GH signalling in human kidney cells (HEK 293) stably expressing GHR. **a** E_2 effect on JAK2 phosphorylation. Protein abundance of phosphorylation and total JAK2 was determined by western analysis from cells expressing ERα pretreated with 100 nM E_2 for 2.5 h and then GH for 2 min. E_2 reduced GH induced JAK2 phosphorylation. **b** Effect of E_2 on SOCS-1, SOCS-2 and SOCS-3 mRNA expression. The mRNA abundance of SOCS-1, SOCS-2, and SOCS-3 in cells treated with 100 nM E_2 was quantified by real-time PCR. E_2 increased SOCS-2 mRNA expression over 4 h. The stimulation was acute and reached statistical significance by 1 h, and remained elevated by 4 h

Human studies show that testosterone augments the biological effects of GH. In hypopituitary children, the stimulation of growth by GH is augmented by co-treatment with testosterone [124]. In hypogonal men, testosterone treatment reduces the circulating concentration of GHBP [125] suggesting an effect on GHR expression in human. In hypopituitary men, testosterone augments the stimulation of fat oxidation, protein synthesis and fluid retention by GH [119, 126]. These observations strongly suggest that androgens regulate tissue responsiveness to GH by enhancing GHR signalling. The later is a likely mechanism given that estrogens have been shown to affect negatively GHR function. Dihydrotestosterone has been shown to enhance prolactin activation of STAT5 signalling in prostate cancer cells [127]. Since GHR is similar to prolactin receptor with regards to signalling pathways, it well may be that androgens can stimulate GHR signalling directly. Recent studies from our laboratory have observed that androgens augment the MAPK signalling of GH with the androgen receptor acting as a co-activator (Leong et al., submitted for publication).

In summary, testosterone stimulates pituitary GH secretion and GHR function. There is evidence that testosterone can stimulate GHRs directly, but the signalling mechanism remains to be elucidated.

6 Summary

The GHR is a member of the cytokine receptor superfamily. GHR function can be modulated by changes to the ligand, to the receptor and to factors regulating the signal transduction process. Targeted alterations to amino acids residing in the critical binding sites of GH have created agonists and antagonists, the pharmacokinetic properties of which can be prolonged by incorporation into sustain release formulations, by pegylation and by creation of ligand-receptor fusion proteins. The development of pegvisomant, a pegylated GHR antagonist, for acromegaly, heralds a new era of endocrine therapy.

Hormones such as insulin, thyroid hormone, gonadal steroids modulate GHR expression and function. Insulin stimulates GHR expression, however high insulin concentrations reduce GHR surface translocation, in such way regulating receptor surface availability. Thyroid hormones stimulate expression and function of GHRs. Estrogen inhibits signalling by GH via the induction of SOCS-2, a protein inhibitor for cytokine signalling. This represents a novel paradigm of steroid regulation of cytokine receptors, and is likely to have significance beyond the GH system.

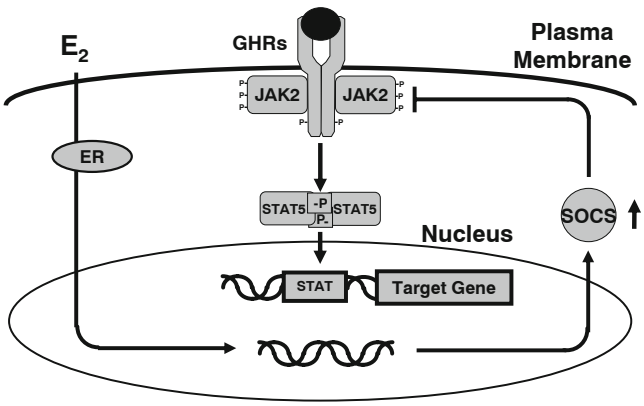

Fig. 5 This figure shows the mechanism by which estrogen inhibits GH signalling. Estrogen inhibits GH signalling via the JAK/STAT pathway by suppressing JAK2 phosphorylation, an effect exerted through stimulation of SOCS-2

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