Growth hormone and cardiovascular risk markers

Anne Klibanski *

Neuroendocrine Unit, Massachusetts General Hospital, 55 Fruit Street, BUL457B, Boston, MA 02114, USA

Abstract

The association of hypopituitarism and growth hormone (GH) deficiency with increased cardiovascular mortality has become increasingly well established [1,2]. In GH-deficient adults, whether the onset of disease is in childhood or in adult life, a higher prevalence of atherosclerotic plaques and endothelial dysfunction has been described [3–5]. Of note, high-resolution ultrasonography has shown that patients with hypopituitarism have a higher prevalence of premature atherosclerosis even in the absence of clinical symptoms [6]. Specific anatomic abnormalities also have been described in adults with GH deficiency, including an increase in intimal-medial wall thickness [3]. The contribution of specific hormonal factors and prior irradiation to the development of vascular disease remains controversial [7].

Keywords: C-reactive protein; Cardiovascular risk markers; Hypopituitarism; Growth hormone deficiency; Stroke; Androgen deficiency

1. Introduction

There are many phenotypic and biochemical abnormalities associated with growth hormone (GH) deficiency that may have an important impact on cardiovascular risk. It is well established that GH deficiency is associated with regional abnormalities in body composition that may have an impact on cardiovascular risk including truncal adiposity and increased visceral fat. These phenotypic abnormalities typically have been associated with both insulin resistance as well as dyslipoproteinemia. Although the positive effect of GH replacement on body composition suggests a causal relationship between this deficiency syndrome and abnormalities in body composition, the role of this deficiency in the development of vascular disease is not clear.

2. Inflammatory cardiovascular risk markers and GH

An important advance in our understanding of cardiovascular risk has been the understanding that inflammation plays a critical role in the pathogenesis of atherosclerosis [8]. Reports by many investigators have suggested a number of actions that GH may have on inflammatory cells. GH has been shown to enhance phagocytic activity, stimulate thymocytes, stimulate DNA synthesis by T-lymphocytes and reduce the production of cytokines in acute injury. In addition, increased monocyte production of cytokines has been reported to occur in adults with GH deficiency. The understanding that inflammation is an important cause of atherosclerosis has led to development of new considerations regarding the ramifications of GH deficiency and its replacement on cardiovascular risk. Studies of atherosclerotic lesions have shown that each such lesion represents various stages in the arterial wall inflammatory processes, and inflammatory markers have been shown to predict cardiovascular risk [9]. In these processes, atherosclerotic lesions, monocytes and lymphocytes release cytokines and growth factors.

A number of cardiovascular risk markers have been examined to assess their validity and reliability in predicting vascular events. For example, interleukin-6 is a pro-inflammatory cytokine that can be found in an atherosclerotic vessel wall. Interleukin-6 induces vascular smooth muscle proliferation and promotes lymphocyte adhesion to the endothelial wall. In addition, interleukin-6 stimulates acute-phase reactants produced by the liver, including C-reactive protein (CRP). One hypothesis that has been proposed is that cytokines such
as those occurring in inflammatory states accelerate the atherogenic process [9].

The best validated cardiovascular risk marker is CRP. Assessments of CRP have consistently found it to be an excellent predictor of cardiovascular events [10,11]. For example, in the Physicians Health Study, patients who had baseline CRP levels in the highest quartile had a threefold increased risk for future myocardial infarctions and a twofold increased risk of stroke independent of other cardiovascular risk factors [10].

To investigate the role of GH administration on inflammatory and other cardiovascular risk markers in men with GH deficiency, we assessed 40 men with adult onset GH deficiency before and after GH replacement therapy (Figs. 1–3) [12]. In this randomized, single-blind, placebo-controlled study, either GH or placebo was administered for 18 months at a dose of GH sufficient to keep serum insulin-like growth factor-I (IGF-I) levels in the appropriate normal range for men their age. The mean GH dose at study end was 4 μg/kg/day. In addition to assessments of anthropomorphic and central fat endpoints and glucose insulin and lipid levels, the main endpoints of the study were CRP, serum amyloid polypeptide A, interleukin-6 and lipoprotein(a) levels at baseline and at 6 and 18 months. We found a significant decrease in both CRP and interleukin-6 levels in patients who were treated with GH compared with control patients. Serum amyloid polypeptide A levels were not significantly different between groups (Fig. 1). Of interest, by the 18th month there were no significant differences between groups in terms of serum cholesterol, low-density lipoprotein (LDL) cholesterol and ratios of total cholesterol to high-density lipoprotein (HDL) cholesterol levels. Furthermore, we did not see a difference between groups in hemoglobin A1C levels. However, lipoprotein(a) levels were significantly increased by GH administration (Fig. 2). The observed changes in cardiovascular risk markers indicated that there was a significant difference in quartiles in comparing the initial and final study results. In our study, the mean CRP level at baseline was comparable to levels for the highest quartile of the Physicians Health Study (mean initial

Fig. 1. Central fat ratios and inflammatory markers in growth hormone (black circles) and placebo (white circles) recipients. Error bars represent 1 SD. P values for the mean difference between groups over months 6, 12 and 18 were 0.0087 for truncal fat-to-total fat ratio and 0.052 for truncal fat-to-extremity fat ratio. P values for the mean difference between groups over months 6 and 18 were 0.0027 for C-Reactive protein (CRP) levels, 0.013 for interleukin-6 levels and 0.056 for serum amyloid polypeptide A levels. (Reprinted, with permission, from Sesmilo et al., Ann. Intern. Med. 133 (2000) 111–122.)
value for the group receiving GH was 4.4 ± 4.3 mg/l ± 1 SD, versus 3.6 ± 3.3 mg/l (the placebo group). In our patients, CRP levels declined in 50% of our GH recipients by one quartile, and in 37.5% of recipients the CRP levels declined by two quartiles (12.5%) as defined by the Physicians Health Study. To put this in the context of other mechanisms and markers. Cardiovascular risk has been linked to increases in homocyst(e)ine levels in a number of studies. Homocyst(e)ine is produced during breakdown of the central amino acid methionine [15]. Homocyst(e)ine is oxidized in plasma to both homo-cyst(e)ine and cysteine-homocyst(e)ine. A number of studies have now shown that elevations in homocyst(e)ine levels may be an independent risk factor for thrombosis [15]. Studies in healthy individuals have shown that plasma homocyst(e)ine levels are useful predictors of cardiovascular mortality [16,17]. In addition, a reduction in homocyst(e)ine levels has been found to decrease carotid plaque area progression [18].

Data in the literature suggest that hormones may affect homocyst(e)ine levels. Because GH administration can increase protein synthesis, we investigated the effects of physiologic GH replacement on homocyst(e)ine levels in the previous cohort treated with GH or placebo for 18 months. We found that in the group treated with GH, Recurrent Events study [13]. The use of pravastatin in that study was associated with a 24% reduction in the incidence of death from coronary heart disease [14]. Therefore, although it is not possible to extrapolate changes from one population to another, it is important to note that, quantitatively, the change in CRP levels observed in patients treated with GH was large enough that, in other populations, such a change would be associated with a lowering of cardiovascular risk.

Endothelial dysfunction has been linked to a number of other mechanisms and markers. Cardiovascular risk has been linked to increases in homocyst(e)ine levels in a number of studies. Homocyst(e)ine is produced during breakdown of the central amino acid methionine [15]. Homocyst(e)ine is oxidized in plasma to both homo-cyst(e)ine and cysteine-homocyst(e)ine. A number of studies have now shown that elevations in homocyst(e)ine levels may be an independent risk factor for thrombosis [15]. Studies in healthy individuals have shown that plasma homocyst(e)ine levels are useful predictors of cardiovascular mortality [16,17]. In addition, a reduction in homocyst(e)ine levels has been found to decrease carotid plaque area progression [18].

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homocyst(e)ine levels decreased compared with levels for patients treated with placebo (net difference, $-1.2 \pm 0.6 \mu mol/l$). In addition, there was a negative correlation between homocyst(e)ine and plasma folate levels. Folate and vitamin B12 levels were not significantly different between the groups. However, homocyst(e)ine changes correlated negatively with changes in IGF-I: for each 1 nmol/l increase in IGF-I, there was a decrease in homocyst(e)ine levels of 0.04 ± 0.02 µmol/l. We found no correlation between changes in homocyst(e)ine and CRP or interleukin-6 levels. Our study therefore showed that GH replacement decreases fasting homocyst(e)ine levels compared with placebo (Fig. 3).

In addition to possibly playing a role in protein synthesis and influencing homocyst(e)ine levels, GH may also affect homocyst(e)ine by altering the bioavailability of other hormones such as T3. GH administration accelerates the metabolism of T4 to T3 and may increase T3 levels while causing short-term decreases in both total and free T3 levels [19]. In our study, total T3 levels increased more with GH therapy compared with placebo therapy, though all levels remained within normal range. Although there was a positive time-by-treatment interaction of total T3 levels, there was no such interaction for homocyst(e)ine. This finding is consistent with GH having an effect on homocyst(e)ine alone. However, we cannot exclude the possibility that GH may have effects on thyroid status within the normal range of homocyst(e)ine levels.

3. Cardiovascular risk markers in women with GH deficiency

Recent studies have shown that among patients with hypopituitarism, females are at particularly increased risk of death from coronary artery disease and stroke [20]. Because data regarding cardiovascular risk has been reported primarily for men, we sought to evaluate the contribution of gender-specific effects on cardiovascular risk in patients with adult-onset GH deficiency. We investigated whether inflammatory and traditional cardiovascular risk markers were different in women with hypopituitarism compared with controls. Therefore, we studied 53 women diagnosed with hypopituitarism and 11 healthy control women in a cross-sectional study. We found that interleukin-6 and CRP levels were higher in women with hypopituitarism than in healthy controls ($P < 0.0001$). Using a multivariate model, we found that CRP levels were dependent upon the diagnosis of hypopituitarism, body mass index (BMI) and estrogen use, and an important interaction was observed between the effect of BMI and hypopituitarism on CRP levels: at a high BMI, hypopituitarism was not a determinant of CRP levels. We also found that interleukin-6 levels were dependent on both hypopituitarism and BMI. Although we found that the total cholesterol levels, total cholesterol-to-HDL cholesterol (total:HDL) ratio and triglyceride levels were higher in patients with hypopituitarism than in controls, when controlling for BMI only the triglyceride level and the total:HDL ratio were dependent on hypopituitarism. Surprisingly, we found no significant difference between women with hypopituitarism and controls in lipoprotein(a) levels. However, there was a trend for lipoprotein(a) levels to be lower among patients in the hypopituitary group when estrogen use was factored in. We found a significant negative correlation between CRP and IGF-I levels.

In addition, we also found a number of interesting correlations between cardiovascular risk markers and androgens. For example, there was a negative correlation between total testosterone, free testosterone and CRP levels. There was also a negative correlation between total testosterone, androstenedione and interleukin-6 levels in women with hypopituitarism. In addition to their reported importance as a cardiovascular risk marker in men, studies have shown that CRP levels are also an important indicator of cardiovascular disease in women. The Women’s Health Study reported that the use of high-sensitivity CRP testing was an important predictor of cardiovascular risk in healthy women regardless of the LDL cholesterol level [21]. Therefore, our data showing that women with hypopituitarism have higher interleukin-6 and CRP levels compared with a control population indicate that patients with hypopituitarism and GH deficiency may indeed be at higher risk for cardiovascular disease through this inflammatory-mediated mechanism. However, at the present time there are no studies examining the effects of GH on these cardiovascular risk markers specifically in women. It is also important to emphasize that in addition to the direct effects of GH on inflammation, GH therapy has been shown in other studies to cause improvements in body composition. Changes in body composition may in turn affect cytokines such as interleukin-6. For example, approximately one-third of interleukin-6 is produced by adipose tissue [22]. Adipose tissue synthesizes both interleukin-6 and tumor necrosis factor-$\alpha$ (TNF-$\alpha$). Both of these cytokines are thought to stimulate CRP production by the liver. Therefore, in addition to the changes in inflammatory risk markers directly mediated by GH, GH’s effects on body composition, in particular visceral fat, may in turn directly affect cytokine production and function.

An interesting additional consideration is the potential impact of androgen deficiency on inflammatory cardiovascular risk markers in women. Androgens modulate interleukin-6 synthesis [23,24], and animal studies have shown that gonadectomy will increase interleukin-6 levels [24]. Androgen deficiency has now been shown to be a prevalent feature in women with
hypopituitarism [25], and therefore it is of interest that both CRP and interleukin-6 levels were found to negatively correlate with testosterone levels in women with hypopituitarism. There are a number of other hormonal mediators that may affect cardiovascular risk besides estrogen. Glucocorticoids are also known to stimulate the synthesis of interleukin-6, and although physiologic glucocorticoid administration is a target goal in treating patients with hypopituitarism, it is certainly possible that slightly supraphysiologic doses of glucocorticoids may stimulate interleukin-6 production.

The issue of inflammatory risk marker changes due to hypopituitarism versus obesity is an important and timely topic. It is well established that obesity is an important cause of increased cardiovascular risk in patients, and studies have shown that CRP levels increase as BMI increases [26]. However, despite the expected finding that women with hypopituitarism have significantly higher BMI levels than controls, we still found a significant increase in CRP levels, even when BMI is factored out. Other factors, such as lipid levels, total cholesterol and total:HDL cholesterol ratios were higher in patients and were dependent upon hypopituitarism when controlling for BMI. In contrast, insulin to glucose ratios were higher in women with hypopituitarism but not significantly higher when controlling for BMI.

4. Studies using a GH receptor antagonist

The findings that CRP levels are elevated in men with GH deficiency and that GH administration decreases CRP levels suggest an important role for GH in regulating CRP and cardiovascular risk. To further examine the relationship between GH and CRP, we investigated CRP levels and other cardiovascular risk markers in patients with acromegaly, a state of marked GH and IGF-I excess. We hypothesized that if GH therapy was an important mediator of cardiovascular risk, lowering of IGF-I levels would increase CRP levels. Thus, we expected changes opposite to those seen with GH replacement of GH-deficient adults.

The development of pegvisomant, a genetically engineered GH-receptor antagonist, enabled the study of cardiovascular risk markers before and after successful management of acromegaly. A large placebo-controlled trial of pegvisomant in 112 patients with acromegaly demonstrated a dose-dependent decrease in IGF-I levels such that they were reduced into the normal range in 89% of patients treated with 20 mg/day [27]. We studied the effects on cardiovascular risk markers of lowering IGF-I levels in 48 of the patients reported in this study. Data from 23 women and 25 men were compared with data of 47 healthy controls who were matched by age and BMI [28]. In the cross-sectional comparison, patients with acromegaly had significantly lower CRP levels than did the controls (median, 0.3 versus 2.0 mg/l). Patients with acromegaly randomized to receive 20 mg pegvisomant had increased CRP levels compared with placebo-treated patients (13.7 ± 3.6 versus 0.5 ± 3.3 mg/l, mean ± SEM) after 12 weeks. There was, however, no significant change in interleukin-6 level (Fig. 4). In contrast with the rise in lipoprotein(a) levels seen in GH-deficient patients with GH replacement therapy, patients with acromegaly demonstrated a fall in lipoprotein A levels with pegvisomant treatment (Fig. 5). Compared with published normative US population data, CRP levels in patients with acromegaly fall in the lowest quintile [29]. Given the increased prevalence of vascular disease reported among patients with acromegaly, it is likely that the favorable risk associated with low CRP levels in such patients might be counteracted by other prevalent risk factors including hypertension and diabetes. The considerable increase in CRP levels following IGF-I normalization in acromegaly is exactly opposite to the lowering in CRP levels following GH replacement in GH-deficient adults [12]. Therefore, these data strongly support a dose-dependent effect of GH action on one of the best validated cardiovascular inflammatory risk markers, CRP. In addition, they support the hypothesis that GH secretion is an important determinant of serum CRP concentrations. These data also

![Fig. 4. Mean C-reactive protein (CRP) and interleukin-6 levels in patients with acromegaly (solid bars) at baseline and after IGF-I normalization with pegvisomant. Mean levels in the group of controls from the cross-sectional study are represented with striped bars. Error bars, SEM.](image-url)
suggest that the important positive effects on cardiac disease observed with normalization of IGF-I levels are not mediated through effects on inflammatory pathways.

5. Conclusion

Traditional risk factors for cardiovascular disease have included assessment of weight, diabetes, smoking, family history and lipid levels. Increasing evidence points to the importance of CRP and other inflammatory mediators as playing a role in cardiovascular risk. In a recently published study by Ridker et al. [30] of over 29,000 women who were followed for 8 years, independent effects were observed for CRP adjusted for all comparisons of the Framingham risk score for heart disease. In fact, CRP was found to be a stronger predictor of cardiovascular events than LDL cholesterol. This risk marker is increasingly becoming a well standardized assay in clinical use. Our finding of beneficial effects of GH administration on CRP levels in GH-deficient patients may provide another useful endpoint to monitor in this population.

Therefore, in addition to conventional markers of cardiovascular risk including serum lipid levels, new insights into the mechanism of cardiovascular disease have recognized the importance of inflammation. Validated markers of inflammation including CRP have enabled the investigation of GH effects on inflammatory risk markers in GH deficiency. Elevated serum inflammatory cardiovascular risk markers have now been demonstrated in patients with adult-onset GH deficiency. Clinically favorable changes in these marker levels following GH administration suggest that GH replacement in such patients may affect the development of vascular disease. These data will need to be evaluated in long-term studies to see whether long-term GH replacement alters cardiac events in these patients.

References

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References


Fig. 5. Mean lipoprotein(a) levels in patients with acromegaly at baseline and after IGF-I normalization with pegvisomant. Mean levels in the group of controls from the cross-sectional study are represented with striped bars. Error bars, SEM. (Reprinted, with permission, from Sesmilo et al., J. Clin. Endocrinol. Metab. 87 (2002) 1692–1699.)


