Recombinant Human Insulin-Like Growth Factor I (IGF-I): Risks and Benefits of Normalizing Blood IGF-I Concentrations

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Abstract
Recombinant human (rh) insulin-like growth factor I (IGF-I) is being developed as a therapy for short stature caused by IGF deficiency (IGFD) and also for diabetes mellitus. To complement the human efficacy and safety data, a large amount of information is available regarding the pharmacology and toxicology of rhIGF-I in animals. This review summarizes the risks and benefits of normalizing blood IGF-I concentrations in IGFD, especially with regard to carcinogenicity, and compares and contrasts safety data for rhIGF-I, recombinant human growth hormone (rhGH), and insulin. A major difference between rhIGF-I and rhGH is that rhIGF-I (like insulin) has hypoglycaemic activity, whereas rhGH opposes insulin action and is diabetogenic. In most of their actions, GH and IGF-I are similar. IGF-I mediates most of the actions of GH, so the safety of rhGH and that of rhIGF-I also share many common features. In animals, the transgenic expression of hGH has been shown to act directly, by activating the prolactin receptor, to increase the incidence of mammary and prostate tumours. In comparison, the over-expression of IGF-I in animals or the administration of rhIGF-I does not have a carcinogenic effect. In formal toxicology and carcinogenicity studies, rhIGF-I has similar effects to insulin in that it can increase food intake, body size, and the growth rate of existing tumours. In animals and humans, IGFD has many long-term detrimental effects besides short stature: it increases the risk of diabetes, cardiovascular disease, and low bone mineral density. Therefore, a case can be made for replacement therapy with rhIGF-I to normalize blood IGF-I levels and reverse the detrimental effects of IGFD.

Introduction
The production of the protein hormone insulin-like growth factor I (IGF-I) is regulated by growth hormone (GH). IGF-I mediates many of the endocrine and paracrine effects of GH. The production and activity of IGF-I is also regulated by a variety of other factors, including insulin, nutrition, and six independently regulated IGF-binding proteins (IGFBPs) [1]. On a structural and functional basis, IGF-I, as its name implies, is closely related to insulin; the insulin receptor and the IGF-I receptor are so closely related that they can form complexes or hybrid receptors [2].

The physiology of IGF-I is also complex because it is both a hormone and a local growth factor [3]. Endocrine IGF-I is produced largely by the liver, which releases large
Fig. 1. GH is released from the pituitary gland into the blood from where it acts hormonally on many tissues, including the heart, bone, cartilage and brain, where IGF-I is also produced locally, and acts as a paracrine and autocrine growth factor. In many tissues, local and systemic IGF-I act together to affect growth and metabolism.

amounts of IGF-I into the blood from where it acts hormonally on many tissues (fig. 1). IGF-I is also produced locally in tissues (fig. 1), where it acts as a paracrine and autocrine growth factor [3]. As it is the intermediary of GH action, IGF-I acts on almost every cell and tissue in the body, having widespread effects on cellular differentiation, proliferation, growth, and apoptosis [3]. Figure 1 illustrates that, in many tissues, including the heart, bone, cartilage and brain, local and systemic IGF-I act together to affect growth and metabolism.

IGF-I can stimulate statural growth in the complete absence of GH signalling, and therefore it can be used to stimulate growth in GH-resistant patients or those suffering from primary IGF deficiency (IGFD) [4, 5]. Therefore, when considering the safety of recombinant human (rh) IGF therapy in primary IGFD, it is appropriate to study the safety record of rhGH. This is logical, because most of the systemic (or endocrine) and local actions of GH directly on tissues, in rodents and probably in humans, are mediated by the generation of IGF-I [6]. Thus, the side-effect profile and the safety of rhGH should provide a guide to the side effects and safety of rhIGF-I. It is also useful to compare the safety profiles of insulin and rhIGF-I, because they are sister hormones with many common features in their mechanisms of action and effects, especially their metabolic and hypoglycaemic actions. Accordingly, the safety of rhIGF-I can, in part, be considered in the context of the safety and toxicity of both rhGH and insulin.

The safety of long-term treatment with rhGH and rhIGF-I has generated much debate based on the correlation between blood IGF-I concentrations and risk of prostate, lung and breast cancers in some studies [7, 8]. This correlation has been debated widely because GH increases IGF-I levels and concern has been raised regarding the potential roles of rhGH or rhIGF-I treatments as cancer initiators. In IGF-I-deficient patients, however, the object of replacement therapy with rhIGF-I is to achieve normal blood IGF-I levels [9], so one might predict that rhIGF-I therapy should not alter their cancer risk compared with the normal population.

Growth Hormone Safety

The history of the formal safety testing of rhGH is complicated by some hGH preparations purified from human pituitary glands having been contaminated with Creutzfeldt-Jakob disease. The urgent withdrawal of pituitary-derived GH meant that it was seen as appropriate by regulatory agencies to approve the use of rhGH without formal long-term toxicology studies [10]. In retrospect, this decision appears to have been appropriate, as the safety record of rhGH has been good [11]. However, because of this regulatory decision, there is no formal animal toxicology data for rhGH to compare with those for rhIGF-I.

The safety of rhGH in children has been tracked in large databases [12, 13], as has the safety of pituitary-
derived hGH [14]. Recent key reports on the safety of GH use in children, especially the risk of malignancy, confirm the safety of GH use in approved indications [14]. The recent regulatory approval of the use of rhGH for the treatment of all children with short stature reflects this greater acceptance of the safety of rhGH [15].

In adults, a major safety issue arose when high-dose rhGH treatment used to induce an anabolic state in intensive care unit (ICU) patients unexpectedly and dramatically increased mortality [16]. It is possible that this increase was due to the diabetogenic effect of GH, as insulin treatment of ICU patients to produce euglycaemia dramatically lowers mortality [17]. It should be noted that rhIGF-I is insulin-like; has the opposite effect on glycaemia to rhGH; and, when administered to ICU patients suffering from head injuries, improves glucose metabolism without affecting mortality [18]. The opposing effects of rhGH and rhIGF-I on glucose metabolism represent a significant difference between the activity and the safety profile of the two hormones.

Epidemiology of GH and IGF-I Deficiencies

Adults with very low levels of GH (adult GH deficiency [GHD]) and, therefore, low IGF-I levels have an increased risk of cardiovascular (CV) disease and increased mortality from CV disease [19]. In addition, long-term high-level GH exposure, such as in acromegaly, is also associated with an increased risk of CV disease and increased mortality due to CV and respiratory disease [20]. Furthermore, there is an ongoing debate about the risk of developing cancer in acromegaly. Recent consensus reports state that a remaining area of concern in acromegaly is its association with colon cancer. The largest study so far conducted [21] found no significant increase in site-specific cancer incidence rates, and although mortality due to all cancers was not increased, mortality due to colon cancer was increased. The ongoing debate regarding GH and colon cancer has resulted in recent contrary reviews in the same journal [22, 23].

The epidemiological association between IGF-I concentrations in blood and cancer risk in middle-aged adults and the elderly has also led to GH being implicated in this association. It is clear, however, that with advancing age, the influence of GH on IGF-I levels declines. For example, the levels of IGF-I in many adult GH-deficient patients become normal for their age and gender, so that, with age, a higher and higher percentage of adult GH-deficient patients have normal IGF-I levels [24]. This probably occurs because as GH secretion declines with age, factors other than GH become more important in regulating IGF-I secretion in adults. This issue is overlooked by some authors in their discussions of the association of IGF-I levels and cancer risk [25]. The factors that probably become more important in the regulation of IGF-I levels with age are insulin and nutrition. The epidemiological relationship between IGF-I levels in blood and cancer risk [7, 8] could be indirect, with the cancers possibly caused by underlying nutritional factors. The relationship between nutrition and cancer is well documented, with several common cancers being linked to Western diets, obesity and over-nutrition, with high insulin levels being a common factor in these relationships [25]. So it is possible that the close relationship between insulin, nutrition and IGF-I leads IGF-I to be a variable associated with an increased risk of cancer. The long-term animal toxicology studies with insulin lend some support to this view (see Insulin Safety below).
The close relationship between insulin and IGF-I is reflected in IGF-I receptors (IGFR) and insulin receptors (IR) being so highly homologous in structure that insulin and IGF-I half-receptors can heterodimerize (fig. 2), leading to the formation of IGF-I/insulin hybrid receptors [2]. IRs exist in two isoforms (IR-A and IR-B), and these two isoforms can both form hybrids with the IGFR. Hybrid receptors containing IR-A (hybrid-RA) are activated by IGF-I, IGF-II and insulin, whereas hybrid receptors containing IR-B (hybrid-RB) are activated with high affinity by IGF-I, with low affinity by IGF-II, and only weakly by insulin. Cell proliferation and migration in response to both insulin and IGFs are more effectively stimulated in hybrid-RA-containing cells than in hybrid-RB-containing cells. The relative abundance of IR isoforms therefore affects activation of the IGF system through hybrid receptors, with important consequences for tissue-specific responses to both insulin and IGFs [2]. This cross-talk between the IGF-I and insulin receptors probably contributes to the similarities between the actions of IGF-I and insulin, and therefore their safety.

GH, IGF-I and IGF-II in Animals

As discussed above, there are no published formal rhGH toxicology data in animals, but it can be argued that the large amount of human safety data makes animal safety data somewhat less important. The GH data most relevant to IGF-I safety are probably those derived from studying the effect of transgenic expression of GH in animals. In mice, the over-expression of hGH, but not bovine GH (bGH), leads to mammary tumours in most animals. Similar effects are seen with prolactin (PRL) over-expression. This indicates that the effect of hGH on the mouse mammary gland is exerted via the PRL receptor rather than the GH receptor [26]. This explains why bGH did not have this effect: bGH does not bind to the mouse PRL receptor, but hGH does. IGF-I was probably not involved, because bGH and hGH increased the blood levels of IGF-I to a similar extent; yet only hGH increased the number of mammary tumours [26]. In humans, hGH also binds to both the hGH and the PRL receptors, raising the possibility that the effects of high GH levels on cancer cells in humans may not be via the GH receptor and IGF-I mediation, but via the PRL receptor. It is interesting in this regard that some of the effects of PRL are probably mediated by IGF-II [27]. The effects of IGF-II on cancer cells are widespread and recognized, especially in mice transgenic for IGF-II, which develop widespread mammary tumours and other neoplasms [28], including lung tumours [29]. The receptor that mediates such effects of IGF-II has not been defined, but recent evidence shows that a sub-type of the insulin receptor forms a hybrid with the IGFR and binds IGF-II with high affinity [2].

In mice, the over-expression of GH also can lead to renal failure and death at an early age due to glomerulosclerosis [30], possibly reflecting the adverse effects of GH on the cardiovascular system seen in humans. Mice transgenic for IGF-I do not suffer from glomerulosclerosis [31], so this adverse effect of GH does not seem to be mediated by IGF-I.

A strong interaction between diet, GHD and IGFD has been shown in the GH-deficient dwarf rat [32], which has low IGF-I levels, profound dwarfism and a relatively normal body composition when fed a normal rodent grain diet. When fed a high-fat diet, however, these rats become profoundly obese and develop insulin resistance [32]. A low level of GH and IGF-I makes the effect of a dietary change on body composition more profound. The association between IGFD and insulin resistance has also been seen in the liver-specific IGF-I-deficient (LID) mouse [33]. LID mice develop insulin resistance with age, without being on a high-fat diet, probably because of their low IGF-I and high GH status [34]. In mice, it therefore seems that a high GH level in the presence of IGFD leads to a more extreme phenotype than combined GHD and IGFD. The relevance of these data, especially to adult humans with primary IGFD, will be discussed later. However, the animal data support the human data in suggesting that IGFD is a risk factor for diabetes and implying that normalizing IGF-I levels in humans will confer a long-term health benefit by reducing the risk of diabetes.

Insulin Safety

The major safety issue with overdosing with insulin is hypoglycaemic shock. IGF-I can also cause hypoglycaemia, although the hypoglycaemic potency of IGF-I is about 10% of that of insulin. Therefore, with appropriate dosing, especially by frequent injections or the use of infusions, the acute hypoglycaemic effects of rhIGF-I can be minimized.

In human clinical trials, therapy with insulin has been implicated in the progression of retinal endothelial cell growth, neovascularization and diabetic retinopathy. The Diabetes Control and Complications Trial (DCCT) study of intensive insulin treatment found that retinopathy initially worsened, especially in patients who had a
rapid improvement in glucose control [35]. This was followed by a long-term overall improvement in measures of diabetic retinopathy. In patients with type 1 diabetes treated with rhIGF-I, there was evidence of optic disk swelling, and early progression of retinopathy was seen with high doses of rhIGF-I [36]. However, there was no evidence of retinal neovascularization. In subsequent studies in patients with type 2 diabetes using lower doses of rhIGF-I with slow dose escalation, there was little evidence of the progression of existing diabetic retinopathy (unpublished data).

Recent studies have shed more light on the effects of insulin and IGF-I on the eyes of humans with diabetes. Local transgenic over-expression of IGF-I in the eye in mice can produce neovascularization [37], whereas systemic administration of IGF-I or systemic transgenic expression has no obvious effect on the eye in mice. In another study, the insulin receptor and the IGF-I receptor were knocked out in the vascular endothelial cells of mice, and hypoxia was used to study retinal neovascularization in these knock-out mice compared with controls [38]. It was concluded that insulin and IGF-I receptors are probably both involved in retinal neovascularization, with the effect of the insulin receptor being more important in this process than the IGF-I receptor [38]. These findings suggest that in humans the safety concerns regarding insulin use and diabetic retinopathy may be similar to those for therapy with rhIGF-I.

IGF-I Epidemiology and Safety

Human Data

IGFD has been shown to be linked to the pathogenesis of atherosclerosis and ischaemic heart disease [39], as individuals with IGFD but without ischaemic heart disease (IHD) have been shown to have a significantly increased risk of developing IHD during a 15-year, follow-up period. There is also a strong epidemiological association between IGFD and the risk of CV disease [39, 40].

Blood levels of IGF-I are in part genetically determined. For example, polymorphisms in the IGF1 gene that lead to IGFD can play a role in the pathogenesis of type 2 diabetes and CV diseases. Genetically determined exposures to relatively low IGF-I levels are associated with an increased risk of type 2 diabetes and myocardial infarction [41].

The association of IGFD with increased risk of CV disease and IHD raises the issue of the effect of blood IGF-I levels on mortality. In the large, long-term Rancho Bernardo study [42], which followed 633 men and 552 post-menopausal women (mean age 74 years) for >10 years, there were 522 deaths; among these, 224 were attributed to CV disease, and 105 to IHD. IGF-I and IGFBP-1 were independently and jointly related to risk of IHD mortality. The relative risk of IHD mortality was 38% higher for every 40 ng/ml (1 SD) decrease in IGF-I, and 3.11 times greater for those in the lowest quintile of IGFBP-1 compared with those with higher IGFBP-1 levels. Low levels of IGF-I and IGFBP-1 increase the risk of fatal IHD independently of other IHD and CV disease risk factors [42]. Insulin is the primary regulator of IGFBP-1, so a low IGFBP-1 level probably reflects a high insulin tone. In the Framingham Heart Study, elderly individuals with serum IGF-I levels at or above the median value had half the risk of heart failure of those with serum IGF-I levels below the median [43]. In the same study, decreased loss of fat-free mass and higher IGF-I levels were also associated with reduced mortality during the next 2 years [44]. Thus, these studies suggest that IGFD and high insulin increase the risk of CV disease and mortality, and imply that normalizing IGF-I levels will confer a long-term health benefit by maintaining fat-free mass and reducing the risk of CV disease and CV mortality.

Blood levels of IGF-I decrease progressively with age [9], and IGFD has been associated with low bone mineral density (BMD) osteoporosis and increased fractures. In postmenopausal women, an IGF1 gene promoter polymorphism has been linked to BMD and rates of bone loss [45]. This population-based study links genetically determined levels of IGF-I to BMD in postmenopausal women and suggests that normalizing blood IGF-I levels will have beneficial effects on the ageing skeleton.

Animal Data

Transgenic and knock-out mouse models have supplied much recent data on the probable risks and benefits of rhIGF-I administration in humans. LID mice have high brain beta-amyloid levels at an early age; and in old rats, beta-amyloid in the brain can be reduced by subcutaneous injections of rhIGF-I [46]. rhIGF-I probably reduced beta-amyloid in the brain by enhancing transport of beta-amyloid carrier proteins such as albumin and transthyretin into the brain. Circulating IGF-I may be a physiological regulator of brain amyloid levels, and rhIGF-I may have therapeutic potential in Alzheimer’s disease [46]. IGFD has been shown to be a risk factor in humans for the cognitive decline of ageing [47, 48] and possibly of Alzheimer’s disease. It is possible, therefore,
that normalizing blood IGF-I concentrations by replacement therapy with rhGH or rhIGF-I may improve memory and provides a tantalizing balance to possible safety concerns regarding rhGH or rhIGF-I administration in adults.

The importance of IGF-I for CV structure and function has also been studied in young LID mice, where systolic blood pressure was increased and stroke volume and cardiac output were decreased. These effects probably occurred secondarily to the increased peripheral resistance. Liver-derived IGF-I is therefore probably involved in the regulation of blood pressure [49], which suggests a mechanism for epidemiological associations between blood IGF-I levels and CV disease and CV mortality. In an animal model of syndrome X, short-term treatment with rhIGF-I dramatically reduced food intake, improved body composition, improved glucose metabolism and reduced blood pressure [50], suggesting that rhIGF-I may have beneficial effects on several key metabolic endpoints in humans.

The global over-expression of IGF-I in mice, either by over-expressing bGH or by directly expressing IGF-I, does not lead to neoplasms. In contrast, global over-expression of IGF-II has been shown to produce neoplasms in mice. The local expression of human pre-pro IGF-I in the prostate gland or the skin does lead to neoplasms [51]. The over-expression of pre-pro IGF-I would also lead to the over-expression of the E-peptide of IGF-I.

The E-peptide of IGF-I has been shown to have mitogenic activity on a variety of cell types [52]. What effect the local expression of the E-peptide along with IGF-I might have is unclear, but it may explain the lack of effect of global over-expression of IGF-I and systemic exposure to IGF-I on the prostate compared with the effect of local expression. The transgenic mouse data appear to support the idea that increasing systemic IGF-I levels (as would also be produced by the systemic administration of rhIGF-I) would have less effect on the prostate than local IGF-I expression.

As a result of the concerns about the possible carcinogenic effects of IGF-I, a lifetime treatment study with rhIGF-I was conducted in rodents. Such animal toxicology studies, using maximally tolerated doses of drug that provide many times the human dose, are designed to provide an early assessment of possible rare events (e.g. potential carcinogenic effects) in humans. To assess the relationship between rhIGF-I exposure and its potential carcinogenicity, vehicle or rhIGF-I was administered by daily subcutaneous injection to male and female rats for up to 104 weeks [53]. The principal findings included an increase in proliferative lesions of the adrenal medulla (hyperplasia and pheochromocytoma) in males given subcutaneous doses ≥ 1.0 mg/kg/day and in females at all dose levels (0.25, 1.0, 4.0 and 10.0 mg/kg/day). Benign epithelial neoplasms of the skin (primarily keratoacanthoma and squamous cell papilloma) were increased in males given doses ≥ 4.0 mg/kg/day and were consistent with the increase in palpable masses noted in these dose groups. When adjusted for survival, there was an increase in female mammary gland neoplasms, fibroadenoma and carcinoma, at dose levels ≥ 4.0 mg/kg/day. Marginal increases in mammary gland carcinomas in males were also seen at the 10.0-mg/kg/day dose level. While the definitive mechanism(s) for these findings is uncertain, a direct mitogenic effect upon spontaneous tumours arising in these tissues cannot be discounted, or an anti-apoptotic effect, given the breadth of scientific literature that associates IGF-I and tumour proliferation/survival [25]. Still, it is conceivable that exaggerated pharmacodynamic effects such as chronic hypoglycaemia and increases in body weight and food consumption contributed to the increase in adrenal and mammary neoplasms and that mechanical trauma caused by repeated subcutaneous injections contributed to the increased incidence of skin neoplasms. This study yielded results similar to the transgenic mouse data, as it showed no evidence of treatment-related effects of rhIGF-I on the incidence of proliferative lesions of the prostate gland, colon or lung.

A comparison of the toxicology of rh insulin and rhIGF-I is instructive. Long-term toxicology (1 year) and lifetime carcinogenicity studies have been performed with insulin and insulin analogues in the mouse and rat and reported in publications and regulatory submissions [54]. The insulin analogue (NovoLog or NovoRapid, Novo Nordisk Pharmaceuticals, Hagedorn, Denmark), which has a single substitution of the amino acid proline by aspartic acid in position B28, was administered to rats subcutaneously at 10, 50 and 200 U/kg/day (~2, 8, and 32 times the human subcutaneous dose of 1.0 U/kg/day, based on U/body surface area, respectively) for 52 weeks. At a dose of 200 U/kg/day, NovoLog increased the incidence of mammary gland tumours in female rats when compared with untreated controls, but the incidence was not significantly different from that for regular human insulin, which also increased the incidence of mammary gland tumours. These data suggest that the effects of rhIGF-I and recombinant human insulins are quite similar in rat toxicological studies.
Summary

An obvious conclusion is that maintaining normal blood levels of IGF-I throughout the lifespan has many beneficial effects. Further, when blood IGF-I levels are low enough that IGFD results, then replacement therapy with rhGH or rhIGF-I appears to be a logical and justifiable treatment.

An opposing point of view has been raised by evidence that, in the fruit fly Drosophila melanogaster, the nematode Caenorhabditis elegans, and the mouse, low levels of insulin/IGF signalling prolong lifespan [55]. As discussed above, this finding does not appear to apply to humans. For example, in the Rancho Bernardo and Framingham studies, low IGF-I levels and IGFD in humans are likely to be adverse for diabetes, CV disease, Alzheimer’s disease, osteoporosis and mortality. The discrepancies between the longevity of experimental animals and data from humans have yet to be adequately explained [56], but could reflect the controlled conditions under which laboratory animals are housed or that the causes of death in humans are different from those in fruit flies, nematodes and mice.

The relationship between blood IGF-I concentrations and the risk of prostate, lung, colon and breast cancers found in case-controlled studies requires discussion. An important point, emphasized by others [57], is that epidemiological studies indicate only an association between cancer risk and blood IGF-I levels; they have not established causality. One explanation for these observations is that insulin or GH acting via IGF-I may be causative factors. Some alternative explanations for the elevated serum IGF-I levels in cancer patients have been proposed by Cohen et al. [57]. The role of GH in cancer initiation can also be questioned as, in acromegaly, the incidence of cancer, other than possibly colonic neoplasia, is not significantly increased. Furthermore, GH-transgenic mice with high IGF-I levels do not develop mammary, prostate or colonic malignancies. The long-term rodent carcinogenicity study described above reinforces these data and suggests that the cancer risk associated with replacement therapy with rhIGF-I is likely to be acceptable and similar to that of rhGH replacement therapy.

Because IGF-deficient patients treated with rhIGF-I as replacement therapy originally have subnormal IGF-I serum levels that normalize with therapy [4], one might predict that their cancer risk on rhIGF-I therapy should not increase above that of the normal population. In addition, monitoring serum IGF-I levels in recipients of rhGH or rhIGF-I can ensure that blood rhIGF-I levels are maintained within the normal range.

References

63:3991–3994.
Horm Res 2004;62(suppl 1):93–100

31 Doi T, Striker LJ, Quaife C, Conti FG, Palmi-
32 Clark RG, Mortensen DL, Carlsson LM, Carls-
33 Melmed S: Acromegaly and cancer: not a prob-
34 Haluzik M, Yakar S, Gavriloa O, Setzer J, Boss-
35 The Diabetes Control and Complications Trial
Research Group: The effect of intensive treat-
ment of diabetes on the development and pro-
gression of long-term complications in insu-
36 Thraillkill KM, Quattrin T, Baker L, Kuntze JE,
Compton PG, Martha PM Jr: Cotherapy with re-
37 Juad A, Scheike T, Davidsen M, Gyllenborg J, Jorgensen T, Ledet T, Moller N, Flyvbjerg A, Or-
skov H: Cardiovascular disease and insulin-
lke growth factor I. Circulation 2002;106:893–
895.
38 Kondo T, Vicent D, Suzzuma K, Yanagisawa M, King GL, Holzenberger M, Kahn CR: Knockout of insulin and IGF-I receptors on vascular endothelial cells protects against reti-
nal neovascularization. J Clin Invest 2003;111:
1835–1842.
39 Vaessen N, Heutink P, Janssen JA, Witteman
40 Frystyk J, Ledet T, Moller N, Flyvbjerg A, Or-
skov H: Cardiovascular disease and insulin-
lke growth factor I. Circulation 2002;106:893–
895.
41 Vaessen N, Heutink P, Janssen JA, Witteman
42 Laughlin GA, Barrett-Conner E, Criqui MH,
Kritz-Silverstein D: The prospective associa-
43 Vasan RS, Sullivan RB, D’Agostino RB, Rou-
befonoff R, Harris T, Sawyer DB, Levy D, Wil-
son PW: Serum insulin-like growth factor I and risk for heart failure in elderly individuals with-
44 Roubenoff R, Parise H, Payne HA, Abad LW,
D’Agostino R, Jacques PF, Wilson PW, Dina-
45 Rivadeneira F, Houwing-Duistermaat JJ, Vaessen N, Vergeer-Drop JM, Hofman A, Pols
HA, Van Duijn CM, Uitterlinden AG: Associa-
tion between an insulin-like growth factor I gene promoter polymorphism and bone miner-
46 Carro E, Trejo JL, Gomez-Isla T, LeRoith D,
47 Kalmijn S, Janssen JA, Pols HA, Lamberts SW,
Bretele MM: A prospective study on circulat-
ing insulin-like growth factor I (IGF-I), IGF-
binding proteins, and cognitive function in the elderly. J Clin Endocrinol Metab 2000;85:
4551–4555.
48 Dik MG, Pluijm SM, Jonker C, Deeg DJ, Lom-
49 Tivesten A, Bollano E, Anderson I, Fitzgerald S, Caudali K, Sjogren K, Skott O, Liu JL,
Mobini R, Isaksson OG, Janson JO, Ohlsson C, Bergstrom G, Isgaard J: Liver-derived insu-
lin-like growth factor-I is involved in the regu-
50 Vickers MH, Ikenasio BA, Breier BH: IGF-I treat-
51 DiGiovanni J, Kiguchi K, Frijhoff A, Wilker E,
Bo DL, Beltran L, Moats S, Ramirez A, Jorca-
no J, Conti C: Deregulated expression of insu-
52 Kuo YH, Chen TT: Novel activities of pro-
53 Dybdal N, Ewell M, Christian B, Clark R,
Fielder P, Kennedy D, Shopp D, Thakur A,
Clarke J: Lifetime (104 Week) daily subcuta-
54 Novolog product label. Available at http://
55 Murphy CT, McCarron SA, Bargmann CI,
Fraser A, Kamath RS, Ahringer J, Li H, Ke-
56 Dupont J, Holzenberger M: Biology of insulin-
like growth factors in development. Birth De-
271.
57 Cohen P, Clemmons DR, Rosenfeld RG: Does the GH-IGF axis play a role in cancer patho-
305.