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REVIEW ARTICLE

Insulin-like growth factor-II: its role in metabolic and endocrine disease

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Summary

Insulin-like growth factor-II (IGF-II) is a widely expressed 7·5 kDa mitogenic peptide hormone. Although it is abundant in serum, understanding of its physiological role is limited compared with that of IGF-I. IGF-II regulates foetal development and differentiation, but its role in adults is less well understood. Evidence suggests roles in a number of tissues including skeletal muscle, adipose tissue, bone and ovary. Altered IGF-II expression has been observed in metabolic conditions, notably obesity, diabetes and the polycystic ovary syndrome. This article summarizes what is known about the actions of IGF-II and its dysregulation in metabolic and endocrine diseases. The possible causes and consequences of dysregulation are discussed along with the implications for diagnostic tests and future research.

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Introduction

Research into growth hormone (GH), which started in the 1950s, culminated in 1976 in the sequencing and naming of insulin-like growth factor (IGF)-I and IGF-II. Whilst knowledge accumulated rapidly on IGF-I, the function of IGF-II remained enigmatic for many years. Uncertainty was expressed about whether it was physiologically relevant. However, recent research has revealed much about the function of IGF-II and its dysregulation in disease. A picture is emerging of a hormone important in foetal development but also in metabolism and tissue maintenance throughout life. Dysregulation of IGF-II has been reported in numerous diseases notably diabetes, obesity, polycystic ovary syndrome (PCOS), liver disease and cancer. These findings have implications for diagnostic and therapeutic approaches. This review provides an overview of the physiology of IGF-II and then updates the reader on the findings in metabolic and endocrine disease. The discussion will largely exclude cancer because this has been covered in detail elsewhere.[1]

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The IGF system

Insulin-like growth factor-I and IGF-II are key components of a complex system, which regulates growth.[2] It is necessary to briefly consider the whole system because the biological activities of the IGFs are influenced by interaction with its various other components, namely IGF-binding proteins (IGFBPs) and cell surface transmembrane receptors. The IGFBPs are a family of six proteins which sequester IGFs in serum, thereby regulating their interaction with receptors. About 75% of circulating IGFs are bound in 150-kDa ternary complexes consisting of IGF-I or IGF-II, IGFBP-3 and acid-labile subunit (ALS), an 85-kDa hepatically synthesized protein. These complexes are confined to the circulation resulting in circulating IGFs having a relatively long half-life (10–16 h). About 25% of IGFs circulate as binary complexes (40–50 kDa) with IGFBPs which cross the capillary endothelium, possibly acting as a pericellular reservoir of IGFs. Finally <1% are 'free' and bioactive, being able to interact with receptor. Free IGFs, such as insulin, have a half-life of a few minutes. The ligands of the IGF system exert their biological effects by binding to the insulin receptor (IR), the type 1 IGF receptor (IGF-1R) and hybrids thereof. There are two alternatively spliced isoforms of IR *viz* IR-A and IR-B. IR-B has 12 amino acid residues at the C-terminus of its ligand binding α subunit which are absent from IR-A. Ligand binding activates a receptor tyrosine kinase (RTK) which recruits intracellular substrate proteins leading to biological effects.[3] The components of the IGF system are illustrated in Fig. 1.

IGF-II

Insulin-like growth factor-II is a 7·5-kDa peptide consisting of 67 amino acid residues which has 67% homology with IGF-I. It

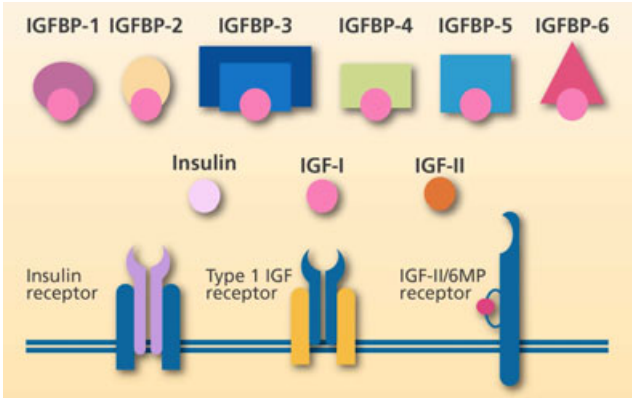

Fig. 1 Components of the insulin-like growth factor system. The ligands of the IGF system are insulin, IGF-I and IGF-II. A family of six IGF binding proteins (IGFBPs 1–6) are all able to bind both IGFs but for simplicity are shown here as binding IGF-I. A hepatically synthesized protein, acid-labile subunit (ALS), binds IGFBP-3 in serum. About 75% of serum IGFs are contained in IGF-ALS-IGFBP-3 ternary complexes. IGFs exert their biological effects by binding to the type 1 IGF receptor (IGF-1R) and insulin receptor (IR). Hybrid receptors of IGF-1R and IR also bind IGFs (not shown). The type 2 IGF receptor (IGF-2R) binds IGF-II alone. It is thought to have a role in IGF-II clearance rather than signaling. IGF-II exerts its biological effects through IGF-1R and the A isoform of IR (IR-A). Figure reproduced from Juul, 2003[2] with permission from Elsevier Press.

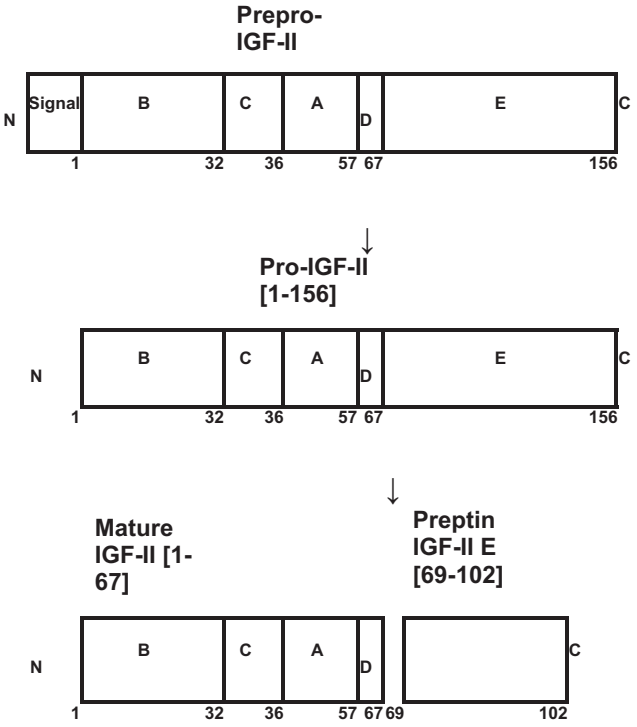

Fig. 2 Insulin-like growth factor (IGF)-II structure and processing. Prepro-IGF-II (180AAs) is the translation product of the *IGF2* gene. It has an N-terminal signal peptide (24AAs) and five domains (A–E). The signal peptide is cleaved by signal peptidase giving pro-IGF-II [1–156]. Further processing occurs following glycosylation of the E domain. Mature IGF-II [1–67] lacks the E domain. Partially processed pro-IGF-II peptides (10–18 kDa) containing part of the E domain make up 10–20% of circulating IGF-II. A fragment of the E domain [69–102] called preptin is also released into the circulation.

is expressed from the 30-kb *IGF2* gene located on 11p15·5, one of a group of genes concerned with growth. Liver is the main source of IGF-II in adults, but it is also synthesized by many other tissues, from which it is released into pericellular fluid. The foetus and placenta are also abundant sources. Its precursor molecule is prepro-IGF-II, which contains a 24-residue N-terminal signal peptide and A-E domains.[4] Cleavage of the signal peptide yields pro-IGF-II [1–156]. The E domain is then O-glycosylated permitting its further proteolysis by prohormone convertase 4 (PC4) to give mature IGF-II [1–67]. This post-translational processing is incomplete, resulting in a variety of pro-IGF-II peptides, 10–18 kDa in size, possessing all or part of the E domain. Collectively these are called 'big' IGF-II. They are secreted into blood, amounting to 10–20% of total circulating IGF-II. The physiological role of big IGF-II is uncertain. Its regulation is poorly understood but appears to be independent from that of mature IGF-II. A 34-amino acid E domain [69–102] fragment called 'preptin' was identified in β cells.[5] It is co-secreted with insulin and considered a physiological amplifier of insulin secretion. The structure of IGF-II and its post-translational processing are shown in Fig. 2.

Serum IGF-II climbs during early childhood, remains stable during adult life and then declines in old age. Its concentration in adults is about 700 ng/ml making it more abundant than IGF-I, the molar ratio being 3:1. Unlike IGF-I, its concentration remains unchanged during pregnancy. Racial differences in IGF-II concentrations have recently been reported.[6] Free IGF-II, such as insulin, circulates at picomolar concentrations and appears to be independent of age. The bioactivity of IGF-II follows its binding to IGF-1R, IR-A and hybrids of these two receptors. IGF-1R activation is thought to bring about most of the biological effects of IGF-II. IR-A binds IGF-II with higher affinity than IGF-I.[7] Its activation by IGF-II is mitogenic, whereas insulin binding causes a metabolic response.

Regulation of IGF-II

The regulation of IGF-II is complex. This complexity may enable fine tuning of its effects and avoid over-expression which could lead to disease. Transcription of the *IGF2* gene is regulated by an epigenetic mechanism called imprinting which confines expression to the paternal allele in most tissues. This is achieved by methylation of the differentially methylated region (DMR) on the maternal allele preventing its transcription. Imprinting can be considered a form of restraint on expression without which IGF-II concentrations would be excessive. There are four promoters (P1-4) from which *IGF2* is transcribed. P2-4 governs *IGF2* transcription in the embryo, whereas transcription occurs from all four promoters in the liver of adult humans. Liver expresses IGF-II biallelically which may explain why circulating IGF-II concentrations remain elevated throughout adult life.

IGFBPs antagonize the biological effects of IGF-II by binding it in serum. This prevents excessive free IGF-II which could

cause tumour development or hypoglycaemia. After IGFBP-3, IGFBP-2 accounts for most of the remaining IGF-II binding in the circulation. Although IGFBP-1 binds very little of the total circulating complement of IGF-II, it is thought to have an important role in the regulation of bioactivity. An increase in the circulating IGFBP-1 concentration suppresses free IGF-II.[2] The effects of IGF-II on a tissue depend on the number and type of receptors expressed. An increased IR-A/IR-B ratio favours IGF-II action, whereas a low ratio, observed in fully differentiated insulin target tissues, favours insulin's metabolic actions.[8] IGF-II also binds to the type 2 IGF receptor (IGF-2R) which terminates its action, thereby acting as a tumour suppressor.[9]

Insulin-like growth factor-II is regulated nutritionally. This is to be expected because both IGFs are part of a mechanism linking nutrition to growth. Their abundance signals the presence of sufficient substrate to meet the protein and energy requirements of growth. IGF-II is down-regulated during under nutrition to avoid hypoglycaemia. It is also regulated hormonally. GH increases hepatic synthesis of IGFBP-3 and ALS thereby increasing ternary complex formation and total serum IGF-II.[10] However, GH is a relatively weak positive regulator which may explain why, unlike IGF-I, the serum concentration of IGF-II fails to increase during puberty or GH administration. Insulin stimulates translocation of IGF-2R to the cell surface, promoting IGF-II clearance.

Physiological effects of IGF-II

Understanding the role of IGF-II will be important if it is to be targeted therapeutically, because disruption of physiology could cause adverse effects. Its role remains relatively poorly understood, but it appears to regulate cell growth, differentiation and metabolism, its effects overlapping with those of IGF-I.[3] It acts in an endocrine, autocrine and paracrine manner. The effect of IGF-II withdrawal on adult human physiology is unknown because no naturally occurring inherited deficiency state affecting IGF-II has been described. Presumably this is because such a state would prevent foetal survival. However, evidence suggesting physiological roles has emerged from various other sources.

Growth and development

Extensive evidence suggests that IGF-II is required for normal embryogenesis. In 1989, studies showed that it stimulated embryonic growth more potently than IGF-I.[11] Knockout studies later reported that *IGF2*$^{-/-}$ mice had severe *in utero* growth retardation, but grew normally following birth and were fertile.[12] IGF-II therefore appeared to be necessary for normal development *in utero* but unnecessary for postnatal growth. However, mice are not directly comparable to humans because, unlike humans, they normally exhibit a decrease in serum IGF-II concentrations postnatally. Subsequent, studies showed that IGF-II had a role in foetal growth in humans. It promoted formation of mesoderm, its *in utero* concentrations being ten times higher than IGF-I.[13] Its foetal actions are exerted mainly through IR-A, but also through IGF-1R. IGF-II is also more abundant than IGF-I in placenta where it is the more important regulator of growth. It also promotes nutrient transport, trophoblast invasion and proliferation and survival of cytotrophoblasts. In keeping with this role, IGF-II is most abundant in invading trophoblasts, the highest concentrations being at the maternal-foetal interface. IGF-II increases its own bioactivity by inhibiting local IGFBP-1 expression.[14]

Given the high serum concentrations present throughout life and widespread expression of its receptors, it would be anticipated that IGF-II has postnatal roles. It has an angiogenic action, central to its role in organ development and maintenance. During angiogenesis, IGF-II up regulates vascular endothelial growth factor (VEGF) and promotes differentiation of embryonic stem cells into endothelial cells.[15] Growth promoting effects of IGF-II have been observed *in vitro*. In the immune system, it promotes granulocyte macrophage colony formation and growth of B cells[16] as well as stimulating growth in erythroid and myeloid precursor cells.[17] IGF-II also promotes β-cell proliferation and survival, its expression declining during postnatal apoptosis.[18]

Insulin-like growth factor-II is required for development and maintenance of the musculoskeletal system. It promoted differentiation of chick embryo cells and C2 myoblasts. The transcription factor MyoD needed for differentiation of fibroblasts into myoblasts requires *IGF-II* expression, blockage of which inhibited differentiation.[19] IGF-II is also required for the development of bone, having a potent anabolic effect.[20] Osteoblasts synthesize more IGF-II than IGF-I. Both are osteoblast survival factors enhancing cell replication, collagen production and matrix apposition. Preptin is also osteogenic. In animals, it stimulated osteoblast proliferation and reduced apoptosis. A recent study on human osteoblast-like cells observed that preptin and IGF-II both stimulated osteoblast activity and differentiation albeit modestly compared with IGF-I.[21]

Metabolic effects

Insulin-like growth factor-II has metabolic actions on adipose tissue, skeletal muscle and liver.[22] In liver, it suppresses hepatic glucose output and increases glycogen synthesis. In peripheral target tissues, it increases glucose uptake and oxidation and increases synthesis of lipid and protein. Its ability to enhance glucose uptake is limited by stringent regulation of free IGF-II concentrations. The importance of this regulation is illustrated by the condition nonislet cell tumour hypoglycaemia (NICTH) in which unrestrained free IGF-II mimics the effect of insulin, resulting in severe hypoglycaemia.[1]

Insulin-like growth factor-II-stimulated glucose uptake has been studied extensively in cellular systems. IGF-II increased glucose uptake into 3T3-L1 adipocytes[23] and human adipocytes,[24] albeit less potently than insulin or IGF-I. The difference in potency may be attributed to IGF-II acting through the IGF-1R which binds it with relatively low affinity. The effect of IGF-II was blocked by IGFBP-1.[23] IGF-II is secreted from human adipocytes in greater quantities than IGF-I.[25] It is unclear how this IGF-II secretion is regulated, but it was reduced by TNF-α. In a separate study on human adipocytes, secretion of IGF-I increased in

response to high glucose concentrations (25 mM)[26] but it is unknown whether IGF-II secretion is similarly regulated.

Ovary

In the ovary, IGF-II appears to be more important than IGF-I. Indeed, IGF-I is not essential for follicular development because conception can occur in its deficiency.[27] Although IGF-II is absent from the ovary prepubertally, its expression climbs during puberty, exceeding that of IGF-I.[28] This increase appears to be caused by the action of gonadotrophins on granulosa cells. During normal folliculogenesis, IGF-II concentrations increase progressively in the fluid of dominant follicles.[29] This is probably as a result of intra-ovarian synthesis because the serum concentrations of IGF-II do not change during the menstrual cycle. IGF-II increases its own ovarian bioavailability by inhibiting transcription of IGFBP-1[30] and stimulating proteolysis of IGFBP-4.[31] It acts on granulosa cells to stimulate their proliferation and production of oestradiol and progesterone[32] and on thecal cells to increase androgen production.[33]

Dysregulation of IGF-II in metabolic disease

Growth disorders

Beckwith–Wiedemann syndrome (BWS) is an inherited condition characterized by excessive *IGF2* expression, usually resulting from loss of imprinting.[34] In a minority of cases, *IGF2* over-expression is caused by uniparental disomy in which two paternal gene copies are inherited. The clinical features of the syndrome include foetal overgrowth, organomegaly and an increased risk of developing tumours, but milder cases lacking these features have also been described.[35] IGF-II is believed to account for the clinical features because a similar phenotype was observed in transgenic animals over expressing *IGF2*.[36] Hypoglycaemia occurs in 50% of cases of BWS. This is caused by hyperinsulinism, rather than being a direct effect of IGF-II, but the underlying β-cell defect is unknown.

Whilst serum IGF-I is invariably increased in acromegaly, IGF-II is less consistent. One study of 60 acromegalic patients observed that IGF-II and IGFBP-2 concentrations were increased compared with healthy controls[37], but in another study, IGF-II was unchanged.[38] The conflicting data may again reflect weaker GH regulation of IGF-II. IGF-II may, via IGF-1R activation, promote development of acromegalic features, because acromegaloid skin changes have been observed in patients with severely increased IGF-II.[39] Increased IGF-I in acromegalic patients is thought to predispose to malignancy, but it is not known whether excessive IGF-II confers the same risk.

Growth hormone deficiency is associated with lower total and free IGF-II concentrations resulting from reduced hepatic synthesis of IGFBP-3 and ALS.[40] However, IGF-II is less reliable than IGF-I as a marker of GH deficiency. Only 52% of GH deficient children had low IGF-II concentrations, whereas 82% had low IGF-I.[41] Pro-IGF-II concentrations were normal in GH deficiency, consistent with its regulation being separate from that of IGF-II.[42] In GH deficiency, IGFBP-2 is the predominant IGFBP, binding more than 50% of circulating IGF-II. Individuals with Laron syndrome (GH receptor deficiency) are congenitally deficient in IGF-I and have IGF-II concentrations 25% of normal.[43] These patients are less likely to develop malignancies than their unaffected relatives although the mechanism of this protection is unknown.

Obesity

Serum IGF-II concentrations appear to increase in obesity and to correlate positively with BMI.[5] In one study comparing obese subjects with lean controls, total IGF-II was increased in the obese subjects and there was a parallel increase in free IGF-II, suggesting increased bioactivity.[44] This is probably an appropriate physiological response, which helps to promote energy storage in response to increased dietary supply. This contention is supported by the reversibility of the changes. Upon weight reduction, decreases in serum total IGF-II, IGF-2R and pro-IGF-II were observed which were independent of the type of diet used.[45] Another study also reported decreases in IGF-II and free IGF-II, but pro-IGF-II remained unchanged.[46] A prospective study in adults observed that low serum IGF-II concentrations predicted future weight gain, in both normal subjects and those with type 2 diabetes.[47] Conversely, elevated IGF-II was strongly associated with future weight reduction over a nine-year period.[48] The mechanism whereby low IGF-II predisposes to obesity is unknown, but these findings suggest that its serum concentration has prognostic value.

There has been recent interest in the association of *IGF2* polymorphisms with IGF-II concentrations and body measurements. A study of an ApaI polymorphism in middle aged males reported that serum IGF-II and BMI were increased in subjects homozygous for the G allele compared with those homozygous for the A allele.[49] A large European population study of three *IGF2* SNPs found associations with height, but not weight.[50] *IGF2* SNPs have also been associated with postprandial resting energy expenditure (REE) and fat-induced thermogenesis.[51] This is of interest because reduction in REE is a possible mechanism whereby these SNPs, along with other gene variants, could predispose to obesity.

Epidemiological studies of obesity have observed an increased risk of cancer, in which the IGF system is causally implicated. It is unknown whether the increased IGF-II concentrations observed in obesity are causally linked to cancer, but mechanisms have been described whereby a prolonged increase in its bioactivity could be carcinogenic. Firstly, when IGF-II binds to IR-A it recruits a different set of intracellular substrates from insulin, influencing gene expression in a manner favouring mitogenesis.[52] Secondly, IGF-II is less effective than insulin at terminating the signal by causing receptor internalization.[53] Thirdly, the IR-A/IR-B ratio increases with age, favouring IGF-II action over that of insulin.[54] These factors could sustain the mitogenic signal, increasing the possibility of carcinogenesis. Conversely, when insulin action is increased by treatment with metformin, there appears to be a reduced incidence of cancer and better

outcome.[55] However, it is not known whether changes in IGF-II concentrations contribute to this effect of metformin.

Parental obesity is associated with epigenetic changes in *IGF2* which may adversely affect the metabolic health of the foetus. Studies have observed that reduced methylation of the *IGF2* DMR in obese mothers was associated with increased IGF-II concentrations in cord blood.[56] Cord blood IGF-II, in turn, correlated positively with birth weight. Paternal obesity has also been associated with reduced methylation of the *IGF2* DMR in the foetus.[57] It appears therefore that parental weight at conception influences *IGF2* by epigenetic changes which could alter foetal IGF-II and weight, predisposing to metabolic disease later in the life of the infant. These observations raise the question of whether this predisposition can be prevented by optimization of parental body weight periconceptually. In humans, the answer to this question is unknown, but studies in animals have observed that periconceptual dietary restriction alters epigenetic regulation of *IGF2*.[58]

The above observations suggest potential utility of testing for IGF-II and *IGF2* polymorphisms in the management of obesity. This could help identify candidates for treatment as part of a primary prevention strategy. Aberrant DNA methylation at sequences which regulate imprinted genes could identify children at risk of developing obesity or cancer in adulthood.[59] There may also be a place for *IGF2* genetic testing in cardiovascular risk assessment. This could be carried out early in life when the potential benefit of risk-reduction therapy is greater than steps taken to prevent the development of modifiable risk factors. Before such testing can be developed, a fuller understanding is required of the link between *IGF2* genetics and metabolic disease.

Diabetes

It is well recognized that IGF-II is dysregulated in diabetes. In a study of obese subjects with and without type 2 diabetes, obese subjects had increased serum IGF-II and IGF-2R compared with normal controls.[60] The concentrations were even higher in subjects with both conditions and decreased upon weight reduction. These changes may in part reflect nutritional regulation of these proteins. Although the cause of the diabetes-related increase in IGF-II is unknown, it could be caused by increased secretion from adipose tissue in response to hyperglycaemia. Irrespective of the cause, there are concerns from epidemiological studies that persistently elevated IGF-II could be detrimental. It is implicated in the association between diabetes and increased breast cancer risk in African-American women.[61] It has also been suggested that excessive IGF-II predisposes to diabetes.[62] Excessive IGF-II is therefore potentially tumorigenic and diabetogenic.

The reported findings in type 1 diabetes are rather different from those in type 2 diabetes. Total IGF-II concentrations, in subjects with type 1 diabetes, were similar to those of controls, but free IGF-II was markedly decreased.[44] This finding may be explained by the increased IGFBP-1 observed in the same study. IGFBP-1 increases acutely during insulin deficiency because of removal of the suppressive effect of insulin on its hepatic transcription. Previous observations that IGF-II is a survival factor in β-cells led to interest in its role in type 1 diabetes. Experimental antibody-mediated neutralization of IGF-II rendered islets susceptible to cell death[63], and it was later observed that, during autoimmune destruction, β-cells had reduced *IGF2* expression, probably as a direct result of cytokine action.[64] Conversely, experimental over-expression of *IGF2* appeared to protect β-cells against IL1B-induced death.[65] However, when β-cell *IGF2* was over expressed in transgenic animals, it disrupted islet organization.[66] The animals had metabolic abnormalities indicative of insulin resistance, namely hyperinsulinaemia and mild hyperglycaemia and 30% went on to develop type 2 diabetes. These adverse effects appeared to be consequences of IGF-II-enhanced β-cell proliferation. Taken together, these observations suggest that in β-cells there is an optimal level of expression of *IGF2*.

The role in of IGF-II in neovascularization makes it a strong candidate for having a role in the pathogenesis of proliferative diabetic retinopathy (PDR). IGF-II is normally present in the vitreous of the eye at concentrations 10- to 30-fold higher than those of IGF-I. Whilst derangements of the IGF system have been observed in diabetic vitreous, relatively few studies have measured IGF-II. One study measured vitreous IGF concentrations in three groups of subjects *viz* patients with PDR, patients with retinal ischaemia and normal controls.[67] IGF-I and IGF-II were increased 2·5- and 1·4-fold, respectively, in patients with PDR compared with normal controls, although the latter increase was not statistically significant. Ischaemia potently increased the vitreous concentrations of both IGFs. A later study of PDR observed that retinal photocoagulation did not influence vitreous concentrations of either IGF.[68] Another study sought to identify the source of vitreous IGFs in PDR by comparing their concentrations with those in serum, using albumin as a marker of permeability of the blood retinal barrier.[69] Vitreous IGF concentrations paralleled those in serum, suggesting that they were determined mainly by leakage of the blood retinal barrier rather than local synthesis. These findings suggest that systemic treatments to lower IGF concentrations may be a rational approach to the treatment for PDR.

Studies of serum preptin during pregnancy observed that its concentrations were higher postpartum than in mid-pregnancy in both healthy women and those with gestational diabetes mellitus (GDM). Its concentrations were highest in of women with GDM. A recent study confirmed these findings and also showed that preptin was produced by the mammary gland.[21] Its concentrations in milk correlated with those in serum. These findings suggest clinical utility for preptin measurement in diagnosis of GDM.

Polycystic ovary syndrome

Various observations have implicated IGF-II in the pathogenesis of polycystic ovary syndrome (PCOS). Firstly, there is evidence that excessive IGF-II may disrupt folliculogenesis. In subjects with PCOS, small antral follicles had a normal cellular distribution of both IGFs.[70] However, *IGF2* expression was increased

and IGF-I undetectable in the granulosa cells of dominant follicles. In a recent study on ovarian hyperstimulation, significantly higher follicular fluid IGF-II concentrations were observed in women with PCOS than in control subjects.[71] Secondly, excessive IGF-II may pathologically increase androgen production.[33,72] Insulin resistance likely contributes to this because treatments which improve insulin sensitivity also improve hyperandrogenism.[73] It is known that hyperinsulinaemia present in insulin resistant PCOS subjects suppresses hepatic IGFBP-1 production, which results in an increase in bioavailable IGF-II.[74]

Evidence suggests that PCOS is caused by a combination of genetic predisposition and environmental influences. Variants in the *IGF2* gene cluster have been associated with the altered metabolic phenotype in PCOS.[75] These may contribute to features of PCOS such as visceral obesity, metabolic syndrome and increased cardiovascular risk. Genetic variants may also render individuals more susceptible to hyperandrogenism. Patients with PCOS are commonly homozygous for G alleles of the ApaI variant associated with increased *IGF2* expression, which may increase ovarian and adrenal androgen secretion.[76] These findings suggest that by targeting IGF-II therapeutically, it may be possible to reduce hyperandrogenism and enhance fertility in women with PCOS.

Preptin concentrations have been observed to increase in PCOS, correlating with indices of insulin resistance such as HOMA-IR.[77] The significance of this increase is unknown. However, a more recent study comparing PCOS subjects with controls from different glycaemic categories observed that subjects with impaired glucose tolerance had higher preptin concentrations than those with normal glucose tolerance, irrespective of whether they had PCOS.[78] Fasting preptin correlated positively with blood pressure, fasting triglyceride and fasting glucose. These findings suggest that increased serum preptin is a feature of insulin resistance rather than PCOS *per se*.

Metabolic bone disease

Observations that IGF-II was anabolic to bone prompted interest in its role in metabolic bone disease. Patients with osteoporosis who had suffered a femoral neck fracture had significantly lower serum concentrations of both IGFs than healthy subjects.[79] IGF concentrations were highly predictive of bone mineral density (BMD) of the femoral neck, suggesting that their deficiency may predispose to fragility fracture. A lack of IGF-II action on bone may contribute to increased fracture risk in glucocorticoid-induced osteoporosis.[80] In this condition, increased cortisol is detrimental to bone both by down-regulating IGF-II and reducing its bioactivity by causing increased IGFBP-1 expression.[20] These findings suggest clinical utility for measurement of IGF-II in the assessment of fracture risk.

Insulin-like growth factor-II has a pathological role in hepatitis C-associated osteosclerosis (HCAO), a rare metabolic disease in which there is significantly increased bone mass. Although IGF-II concentrations are normal in HCAO, pro-IGF-II is increased.[81] Its bioavailability, and hence anabolic action, appears to be enhanced by its ability to bind IGFBP-2 in a 50-kDa binary complex. The predominant form of pro-IGF-II in HCAO was later reported as pro-IGF-IIE [1–104].[82] This form does not result in increased free IGF-II in the circulation, which may explain why patients with HCAO do not suffer from hypoglycaemia.

Osteoporosis and obesity are related, in part because bone is also subject to the influence of circulating adipocyte and β-cell-derived hormones including preptin. Increased preptin in hyperinsulinaemic conditions such as obesity may, through its anabolic actions, help to preserve bone mass.[83] When administered to mice, preptin increased bone mineralization, an action that could be anticipated given that it contains the sequence of pro-IGF-II responsible for increasing bone mineralization in HCAO. In a study of males, serum preptin was lower in subjects with osteoporosis than in those with normal bone mass.[84] Preptin concentrations were positively correlated with BMD, suggesting utility for its measurement. The findings also suggest that preptin has potential as a therapeutic agent in osteoporosis. Reported findings of IGF-II dysregulation in metabolic and

Table 1. Reported insulin-like growth factor-II-related findings in metabolic endocrine disease

Disease	Finding	Reference
GH deficiency	↓IGF-II, ↓free IGF-II	40
Laron syndrome	↓IGF-II	39, 43
Acromegaly	↑↓IGF-II	37, 38
Obesity	↑IGF-II, ↑free IGF-II, *IGF2* SNPs	6, 44, 46, 49
Type 2 diabetes	↑IGF-II, ↑soluble IGF-2R	60
Type 1 diabetes	IGF-II unchanged, ↓free IGF-II	44
Gestational diabetes	↑pro-IGF-II E [69–102] (preptin)	21
Diabetic retinopathy	↑vitreous IGF-II	67
Insulin resistance	↑pro-IGF-II E [69–102] (preptin)	78
PCOS	↑follicular fluid IGF-II, *IGF2* variants	72, 75, 76
Osteoporosis	↓IGF-II	79
HCAO	↑pro-IGF-II	81

HCAO, hepatitis C-associated osteosclerosis; PCOS, polycystic ovary syndrome; ↓, decreased; ↑, increased.

Table 2. Potential clinical utility of insulin-like growth factor-II and related tests in metabolic disease

Test	Reference
Serum IGF-II	
Prediction of weight gain	47, 48, 59
Assessment of fracture risk	80
Serum Pro-IGF-II E [69–102] (preptin)	
Diagnosis of osteoporosis and monitoring of bone mass	84
Diagnosis of gestational diabetes	21
IGF2 genetic testing	
Prediction of weight gain	49
Cardiovascular risk assessment	49

endocrine disease are summarized in Table 1. Potential clinical utility for IGF-II measurement and related tests in these disorders is listed in Table 2.

Future considerations

In recent years, numerous studies of IGF-II have observed its dysregulation in metabolic and endocrine disease. However, to understand the significance of these findings, it will be necessary to have a clearer picture of the physiology of IGF-II. For example, if we are to understand the mechanism and consequences of increased serum IGF-II in obesity, we need a fuller description of the regulation of its expression in adipose tissue. IGF-II is only one of many factors linked to the pathogenesis of metabolic disease, and there is a need for metabolomic studies to study it in the context of other analytes, including other components of the IGF system which influence its action. Future genetic studies should consider *IGF2* in the wider context of other genes.

Studies have suggested clinical utility for measurement of IGF-II and related analytes in the management of patients with metabolic disease. Future research will certainly pursue new diagnostic tests. This will likely require the development of new assays. There is a need for reliable and inexpensive methods for measuring free IGF-II and for techniques which can measure its autocrine and paracrine actions. Given the high prevalence of metabolic disease, it is important that research continues to make progress in understanding its pathogenesis, the ultimate aim being prevention. In particular, the long-term consequences of excess IGF-II in obesity should be further investigated by epidemiological studies.

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