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Fakulteten för veterinärmedicin och husdjursvetenskap

IGF II: structure, function and role in disease

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IGF II: struktur, funktion och påverkan på sjukdomar

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SAMMANFATTNING

Den insulinlika tillväxtfaktorn II (IGF II) är ett proteinhormon som reglerar cellers proliferation, tillväxt, migration, differentiering och överlevnad. Den utövar sin biologiska funktion via IGF I-receptorn. Dess tillgänglighet för bindning till IGF-1R regleras av en familj bestående av sex IGF-bindarproteiner (IGFBPs). IGF II kan också inducera mitos via en alternativt splitsad variant av insulinreceptorn (insulin receptor isoform A (IR-A)). De cirkulerande nivåerna av IGF II kontrolleras genom IGF II-receptorn, som saknar signalfunktion och är homolog med mannos-6-fosfatreceptorn.

Tystad prägling av IGF II-genen är en återkommande observation vid en utvecklingsrubbning kallad Beckwith-Wiedemanns syndrom, vilket innefattar okontrollerad tillväxt och i många fall en njurtumör hos barn, Wilms' tumör. Tillväxthämning ses vid mutationer i genlocuset hos IGF II och den närliggande genen H19 vid en annan utvecklingsrubbning, Silver-Russells syndrom. IGF II utsöndras i vissa fall av mesenkymala tumörer i lungsäcken, vilket leder till låga blodsockerhalter (hypoglykemi) och Doege-Potter syndrome. IGF II har också visat sig kunna öka storleken av lesioner vid åderförkalkning.

ABSTRACT

Insulin-like growth factor II (IGF II) is a protein hormone known to regulate cell proliferation, growth, migration, differentiation and survival. It exerts its biological actions via the type 1 IGF receptor (IGF1R). Its availability for binding to IGF1R is mainly regulated by a family of six IGF binding proteins (IGFBPs). IGF II can also induce a mitogenic response via an alternatively spliced version of the insulin receptor (insulin receptor isoform A (IR-A)). The circulating and tissue levels of IGF II are further controlled by the insulin-like growth factor type II receptor (IGF2R), which is homologous to the cation-independent mannose-6-phosphate (M6P) receptor.

The gene is paternally imprinted in the sense that transcripts are almost exclusively derived from the paternal allele. Loss of imprinting of the IGF II gene is a recurrent observation in a growth disorder, Beckwith-Wiedemann syndrome, leading to overgrowth and in many cases a variety of malignant tumours. Growth retardation is observed following epimutations in the IGF II/H19 locus in another growth disorder, Silver-Russell syndrome. IGF II is sometimes secreted by non-islet cell tumours in the pleural cavity, leading to low levels of blood sugar (hypoglycemia) and Doege-Potter syndrome. IGF II has also been shown to increase the size of atherosclerotic lesions.

INTRODUCTION

Insulin-like growth factor II (IGF II) is a crucial factor in regulating cell proliferation, growth, migration, differentiation and survival. Its diverse biological activities have been characterized since the 1950s, but it was not until 1976 that the name IGF II was stipulated.

IGF II interacts with several receptors and binding proteins to exert its actions. It binds to the non-signalling insulin-like growth factor type II receptor (IGF2R) with high affinity. This receptor is homologous to the cation-independent mannose-6-phosphate (M6P) receptor. IGF II can also bind to different signalling receptors, such as the type 1 IGF receptor (IGF1R) and the insulin receptor with lower affinity, although an alternatively spliced version of the insulin receptor, named insulin receptor isoform A (IR-A) may bind IGF II with high affinity. Its availability to bind to receptors is mainly regulated by a family of six IGF binding proteins (IGFBPs) (Chao & D'Amore, 2008).

Altogether, these findings weave an intricate pattern of biological activity which this article aims to summarize. The IGF II protein, its receptors and binding proteins will be elaborated on and an attempt to establish whether or not IGF II itself or its interacting proteins may have any associations with key diseases will be made. The phylogenetic conservation of the IGF II gene between different species will also be briefly discussed.

MATERIALS AND METHODS

Articles were mainly found by using the on-line databases PubMed and Google Scholar. Due to the lack of a consistent nomenclature, several search terms were used separately as well as in combination, such as ”IGF2”, ”IGF II”, ”IGF-II”, ”IGF2R”, ”IGF AND receptor”, ”insulin-like growth factor”, sometimes along with keywords such as ”Beckwith-Wiedemann” to yield more specific results. Several reprints on the subject were also gratuitously offered by the supervisor.

LITERATURE REVIEW

Gene structure

In humans, the IGF II gene is located on chromosome 11p15.5, and stretches over approximately 30 kb DNA. There are four promoters and ten exons, giving rise to different transcripts depending on which promoter the transcript stems from (Engström et al., 1998).

Although the human IGF II gene contains ten exons, only the last three contain coding sequences. In mice, the IGF II gene resides on chromosome 7 and in rats on chromosome 1. In mouse and rat species, it contains six exons (Otte**,** 1997).

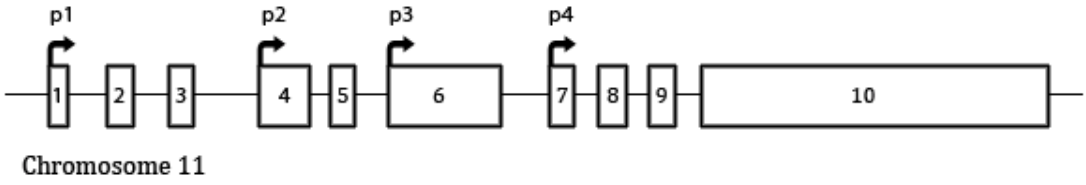

Figure 1. Illustration of the human IGF II gene structure. Exons are shown as boxes. Promoters P1-P4 are represented as arrows. Modified from Otte, 1997.

The gene structure of IGF II is strongly conserved between species, including human, rat, mouse, sheep, mink, horse, and pig.

Table 1. Homology between human and other species in regards to the mature IGF II amino acid sequence, expressed in percent (Engström et al., 1998).

Species	Amino acid sequence
Mink	100
Pig	97
Horse	95
Sheep	86
Rat	85
Mouse	84

Posttranslational processing

IGF II is first synthesized as a pro-hormone/precursor-hormone containing 180 amino acids which is subsequently processed, finally appearing as a 67 amino acid IGF II bioactive protein. This is true for all mammalian species hitherto examined, except for mink whose mature IGF II protein has a serine insertion at residue 40, therefore containing 68 amino acids (Ekström et al., 1993; Qiu et al., 2007).

At first, a signal peptide containing 24 amino acids is removed from the N-terminal, generating proIGF II (156 Aas). Subsequent cleaving of proIGF II then results in a 104 amino acid peptide product (IGF II(1-104)). Endoproteolyzation generates IGF II(1-87) and as a result, the mature IGF II peptide consists of 67 amino acids.

Both IGF II isoforms – IGF II(1-104) and IGF II(1-87) – collectively named ”big IGFs” have been found circulating in human and bovine serum (Qiu et al., 2007). They are also sometimes secreted by solitary fibrous tumours in the pleural cavity, leading to hypoglycemia and Doege-Potter syndrome (Kalebi et al., 2010).

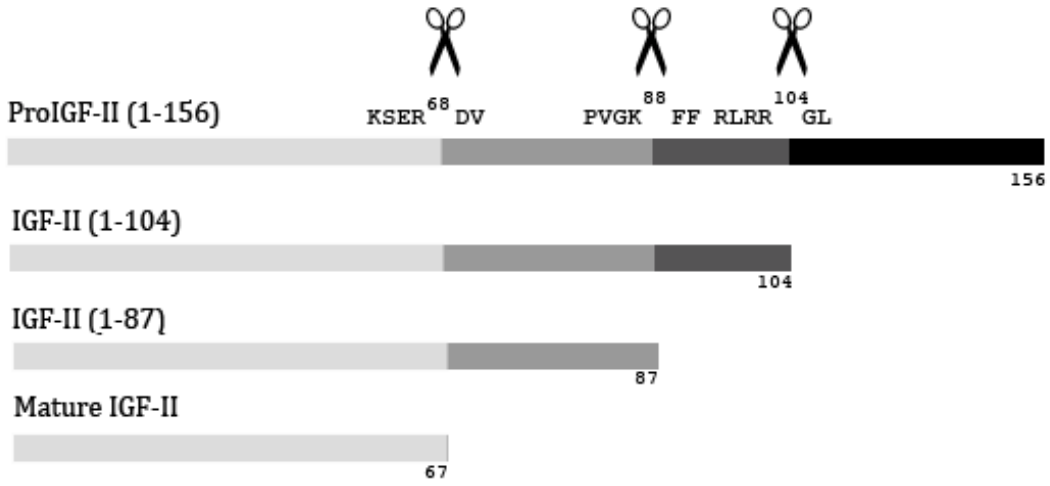

Figure 2. Illustration of the posttranslational processing of the IGF II peptide. Modified from Qiu et al., 2007.

The mature peptide

The insulin-like growth factor II (IGF II) is a polypeptide, with a composition similar to that of IGF I, relaxin and not surprisingly, insulin. In other words, these four hormones constitute a family of structurally related polypeptides (Otte, 1997). IGF II is made up of B, C, A and D domains, listed in order from the N- to the C-terminus. Three alpha-helices are located in these domains. Three disulphide bonds hold the structure together (Alvino et al., 2011).

IGF II plays a crucial role in the embryonic development of mammalian species. It is also of particular importance to placental growth (Chao & D’Amore, 2008).It works in an autocrine and paracrine fashion in different tissues, meaning that it binds to receptors on the same cell that it is produced by, or to adjacent cells (Otte, 1997). IGF II is also synthesized by the liver and released into the blood stream, acting like a classical hormone, but this doesn’t apply to all species. For instance, in embryonic mice, the expression of IGF II is high. A significant decline in circulating levels then takes place after birth, and in adult mice, its transcripts can only be found in the choroid plexus and leptomeninges (Chao & D’Amore, 2008).

By contrast, serum levels of IGF II remain high in adult humans, where it promotes the differentiation of muscle, brain and brown adipose tissue cells (Brown et al., 2009). In vitro, IGF II has shown several different biological actions, some of them seemingly contradictory. For instance, it induces apoptosis in a cell line of Wilms’

tumour named WCCS-1 but has an inhibitory effect on apoptosis in other cell systems, such as teratoma-derived cell lines. It stimulates cell proliferation via cell division (DNA-reparation and mitosis) as well as cell enlargement. It has been shown to stimulate differentiation in myoblasts (Engström et al., 1998).

Receptors

IGF2R

The insulin-like growth factor II receptor (IGF2R) is a type I transmembrane glycoprotein, composed of a large extracellular region, a small 23 residue transmembrane region, and a 167 residue cytoplasmatic tail. The extracellular region consists of a 40 residue amino signal sequence and 15 homologous extracellular repeat domains, each of them containing between 124-192 amino acids (Hassan, 2003).

Each extracellular repeat domain contains approximately 147 amino acids. Binding of IGF II occurs primarily in domain 11. The affinity of this domain for IGF II is believed to be further enhanced by a fibronectin type II-like insert in domain 13, though the exact mechanisms of this enhancement remain undiscovered. There are also questions unanswered regarding the precise binding sites within domain 11 – analyses so far have pointed towards three different proposed locations – a hydrophobic pocket, a cluster of residues and a disparate set of residues (Brown et al., 2002).

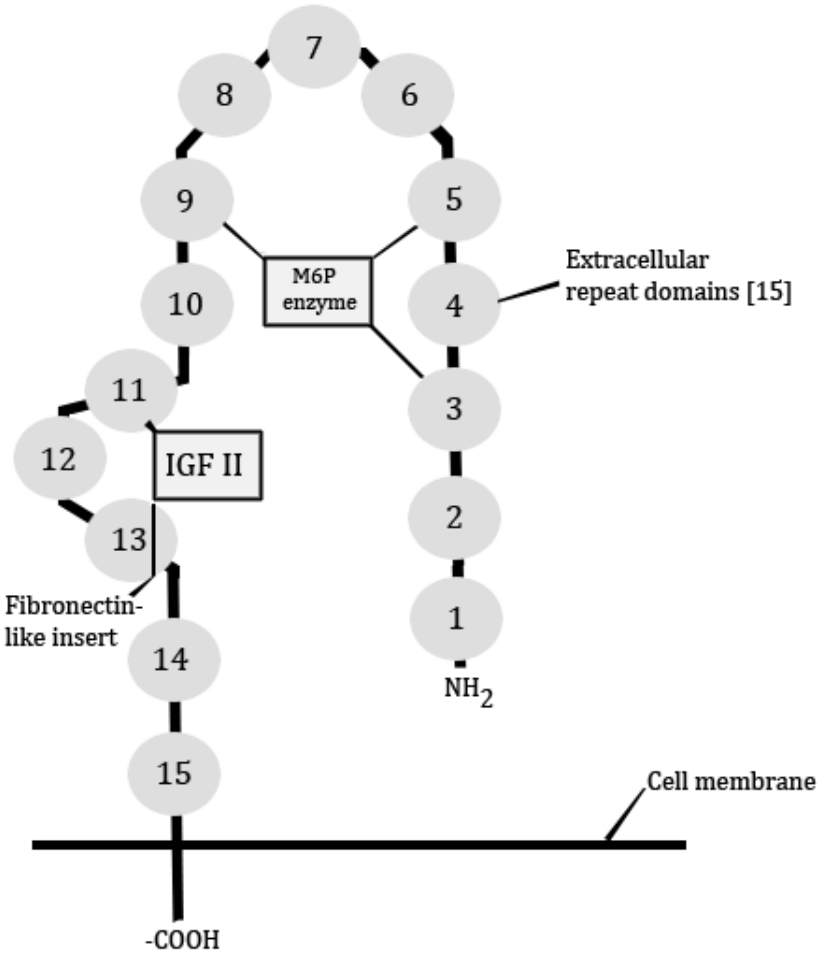

Figure 3. Illustration of the IGF II receptor structure. Based on data from Hassan, 2003; Brown et al., 2009a.

Function

IGF2R regulates the amount of circulating and tissue IGF II by transporting the ligand into the cell and degrading it (Hassan, 2003). The receptor is multifunctional and binds not only IGF II, but also mannose-6-phosphate (M6P)-marked lysosomal enzymes at domains 3, 5 and 9, enabling the transfer of newly synthesized lysosomal enzymes from the trans-Golgi network to late endosomes. Lysosomally destined enzymes are recognized by a M6P-tag, which prompts them to bind to the M6P receptor. They are then transported to late endosomes via clathrin-coated vesicles. After reaching the late endosomes, the enzymes are released and transported to their final destination – the lysosomes – whereas the M6P receptors are either headed for the cell surface or back to the Golgi network (Brown et al., 2009a).

Like IGF II, IGF2R is an imprinted gene, but while IGF II is only expressed from the allele inherited from the father, IGF2R is only expressed from the allele inherited from the mother. In mice, the imprinting of IGF2R is regulated by the intron 2 region. The paternal allele contains an anti-sense transcript, which stems from intron 2,

mediating the silencing of the paternal allele IGF2R gene. Deletion of this intron 2 region disrupts the silencing, leading to biallelic expression of paternally inherited IGF2R (Hassan, 2003).

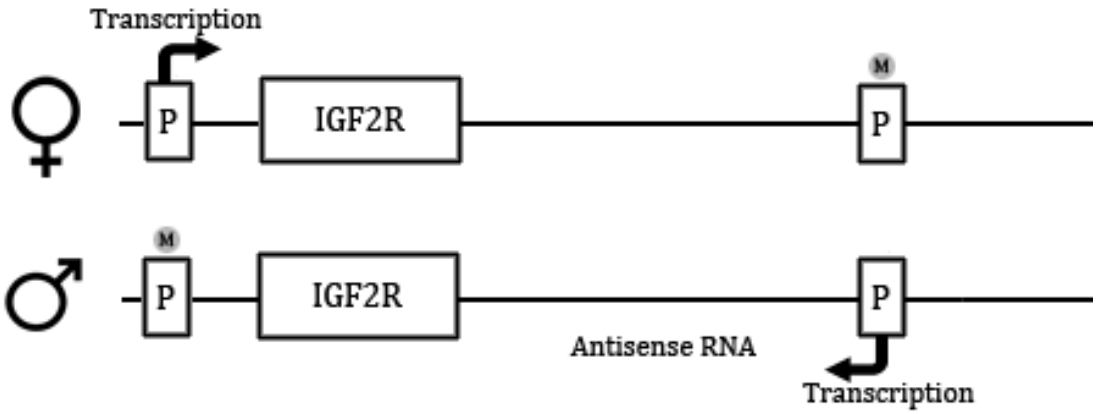

Figure 4. The imprinting regulation of the IGF II receptor. Based on data from Hassan, 2003.

Mice inheriting a disrupted IGF2R gene from their mother – thereby not expressing IGF2R in tissues – have been shown to suffer from overgrowth and perinatal lethality, presumably because of cardio-respiratory failure (due to malformed lungs and abnormalities in cardiac muscle). However, when the same gene was inherited from the father, no abnormalities in development were recorded, confirming that the IGF2R-gene is paternally imprinted. Thus, studies conducted on sheep revealed that an imprinting defect of the IGF2R gene leading to loss of IGF2R expression causes plasma levels of IGF II levels to rise. The result of this is overgrowth (Brown et al., 2009a).

Single nucleotide polymorphisms (SNP) in IGF2R lead to an increased risk of cancer and thus IGF2R has been referred to as a tumour suppressing gene. In nude mice, excessive expression of IGF2R in breast- and uterine cancer cells resulted in a lowered number of tumours and decreased tumour growth. Associations with hepatocellular, gastrointestinal, ovarian and prostate cancer have also been shown (Brown et al., 2009a).

IGF1R and insulin receptor (IR)

The insulin and IGF I receptors are both members of a receptor subfamily of transmembrane tyrosine kinases. They are structurally homologous, with the tyrosine kinase domains sharing 84 % sequence identity and the juxtamembrane and C-terminal regions possessing 61 % and 44 % sequence identity, respectively (Favelyukis et al., 2001).

Although these receptors mainly bind insulin and IGF I, they also bind IGF II with lower affinity. However, an alternatively spliced version of the insulin receptor lacking exon 11, insulin receptor isoform A (IR-A) has been shown to bind IGF II with high affinity – consequently, cell proliferation, differentiation, migration and survival may be exerted via this receptor as well. The IR-B isoform is the classical insulin receptor, which can bind IGF II, but will mainly result in a metabolic response (Chao & D'Amore, 2008).

Function

Interaction with the IGF I receptor is associated with cell proliferation, differentiation, migration and survival. When activated, the IR and IGF1R initiate a phosphorylation cascade via two different pathways. Upon ligand binding, the receptors initially undergo autophosphorylation, which enables different adapter molecules such as IRS-1, -2 and SHC to bind. The two pathways then activated are the phosphatidylinositol 3-kinase (PI3K)-AKT/protein kinase B (PKB) pathway and the mitogen-activated protein kinase (MAPK) pathway, which in this case involves Ras activity. The PI3K/PKB pathway leads to metabolic activity and the MAPK pathway regulates cell growth and differentiation, in addition to controlling the expression of certain genes (Alvino et al., 2011).

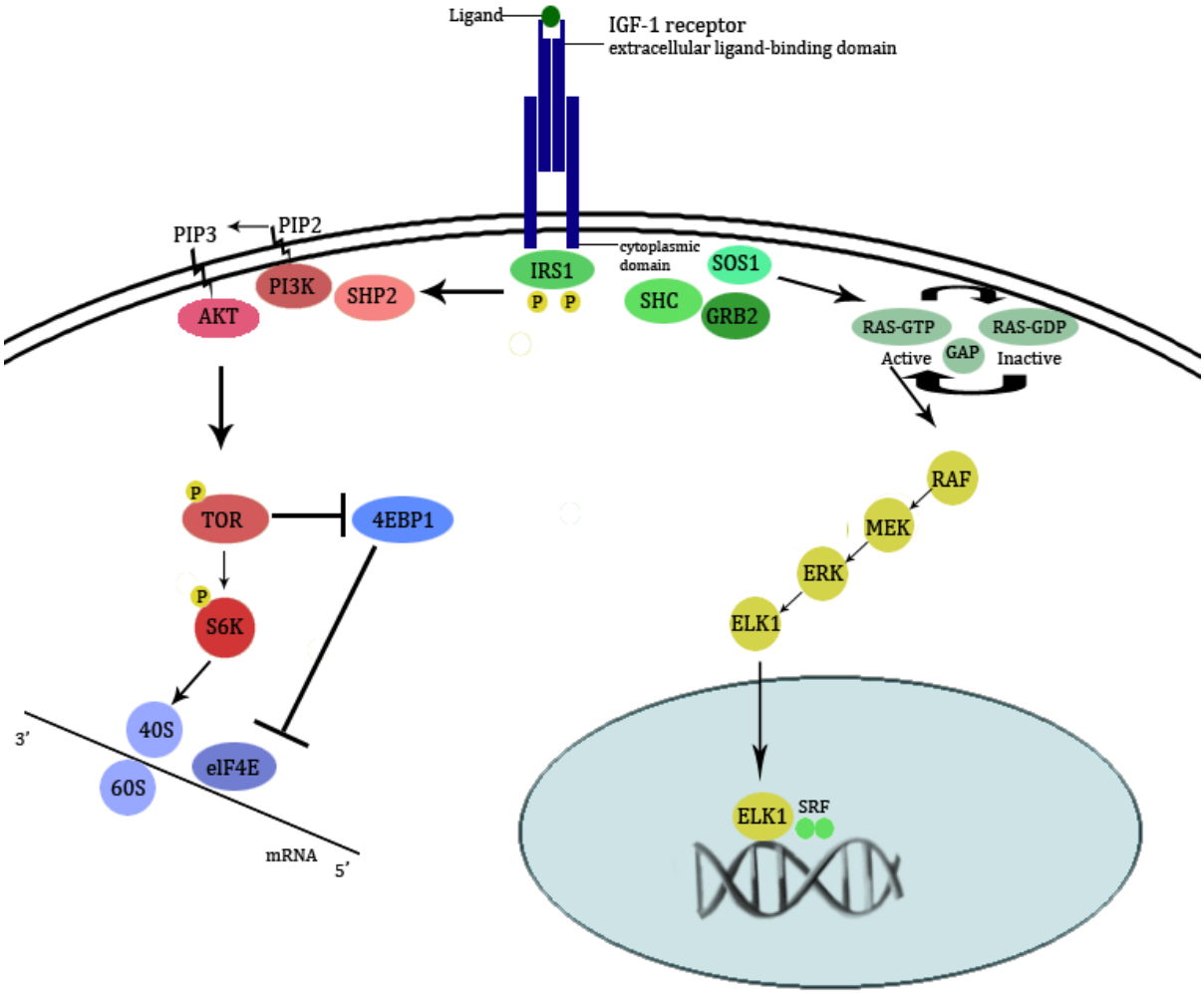

Figure 5. The IGF1R and insulin receptor signalling pathways. PKB pathway to the left and MAPK pathway to the right. Modified from Pollak, 2004.

In the PKB pathway, ligand-binding induces phosphorylation of IRS1. PI3K then catalyzes the conversion of PIP2 to PIP3, which activates AKT. Activation of AKT increases the uptake of glucose in the cell but also affects other proteins along the pathway. eIF4E is normally inhibited in a complex with its binding protein 4EBP1, but phosphorylation of 4EBP1 frees eIF4E, leading to protein synthesis. mTOR phosphorylates S6K, activating it, which also leads to protein synthesis. This pathway can also inactivate pro-apoptotic transcription factors and proteins, leading to decreased apoptosis.

In the MAPK pathway, SHC becomes phosphorylated and binds GRB2 and SOS1 in a complex which activates Ras, MAP kinases (MEK) and finally SRF and ELK1, which lead to mitogenic activity. This pathway can also regulate apoptosis (Senthil et al., 2002).

IGF binding proteins (IGFBPs)

The IGFs cannot circulate freely. Therefore they require specific IGF-binding proteins for transportation in the blood stream. There are six classical IGFBPs which bind IGFs with high affinity and have a large part of their amino acid sequence in common. More recently, a group of proteins binding IGFs with lower affinity have been discovered. Although they are structurally related to the classical IGFBPs and considered to be part of the IGFBP superfamily, due to the low binding affinity they are instead referred to as insulin-like growth factor binding protein-related proteins (IGFBP-rP) (Clemmons, 1997; López-Bermejo et al., 2000).

The availability and distribution of IGF II in different tissues is controlled through IGFBPs, (Chao & D'Amore, 2008) as well as their half-life in blood. This function makes them powerful modulators of IGF II action. Different cells and tissues synthesize different combinations of IGFBPs. For instance, IGFBP-3, -4 and -5 are typically synthesized by fibroblasts, while IGFBP-2 and -4 are synthesized by epithelial cells.

IGFBPs are in turn controlled by proteases secreted by various tissues. Proteolytic cleavage of IGFBPs affects their IGF binding affinity negatively. Phosphorylation of IGFBPs could also reduce protein binding activity, but the exact biological significance of these findings are unknown (Clemmons, 1997).

- IGFBP-1. Excess levels inhibit growth-promoting actions of IGFs.
- IGFBP-2. This binding protein inhibits IGF II-stimulated DNA synthesis in lung carcinoma cells. It also inhibits collagen synthesis.
- IGFBP-3 is synthesized in the liver. It binds 90-96 % of the IGFs in serum and potentiates the actions of IGF I.
- IGFBP-4 inhibits IGF activity consistently.
- IGFBP-5 potentiates IGF I-induced osteoblast growth and protein synthesis.
- IGFBP-6 is believed to have inhibitory effects, but studies are so far limited.
- IGFBP-rP1 is associated with different forms of cancers; may be related to apoptosis or angiogenesis. Might potentially be a tumour suppressor in prostate cancer.
- IGFBP-rP2 modulates growth of fibroblasts and epithelial cells.
- IGFBP-rP3 is believed to be involved in the regulation of prostate cell differentiation.
- IGFBP-rP4. Research indicates it might function as a tumour suppressor.

Adapted from (Clemmons, 1997; Cait et al., 2005; López-Bermejo et al., 2000).

IGFs in disease

Beckwith-Wiedemann syndrome

As previously noted, IGF II plays a pivotal role in fetal growth. Interest has therefore been focused on clinical syndromes that display aberrant growth properties. Beckwith-Wiedemann syndrome (BWS) was one of the first syndromes where IGF II-expression was linked to a growth disorder (Brown et al., 2009a).

BWS is a collective term for a number of growth disorders. Hence, the clinical manifestations of BWS may vary greatly between individuals, and there are no definite inclusion criteria for a diagnosis. However, some clinical findings commonly associated with BWS are macrosomia, macroglossia, abdominal wall defects, visceromegaly, hemihyperplasia, anterior ear creases and posterior helical pits, kidney abnormalities, cytomegaly of the adrenal fetal cortex and cleft palate (Murrell et al., 2004; Cerrato et al., 2008).

In humans, the risk of developing certain childhood cancers is significantly elevated with BWS. This is particularly true for Wilms' tumour, a type of childhood kidney cancer also known as nephroblastoma. Overexpression of IGF II is frequently observed in cases of Wilms' tumour, and for a long time, it was thought to be a tumour promoter. That was until a 2001 study surprisingly refuted these assumptions and instead identified IGF II as a source of apoptosis and necrosis in Wilms' tumour cells (Granérus et al., 2001). Embryonal tumours occur in about 5 % of BWS patients (Murrell et al., 2004), Wilms' tumour being the most prominent, but also adrenocortical carcinoma, hepatoblastoma and rhabdomyosarcoma have been reported in BWS patients (Granérus et al., 2001).

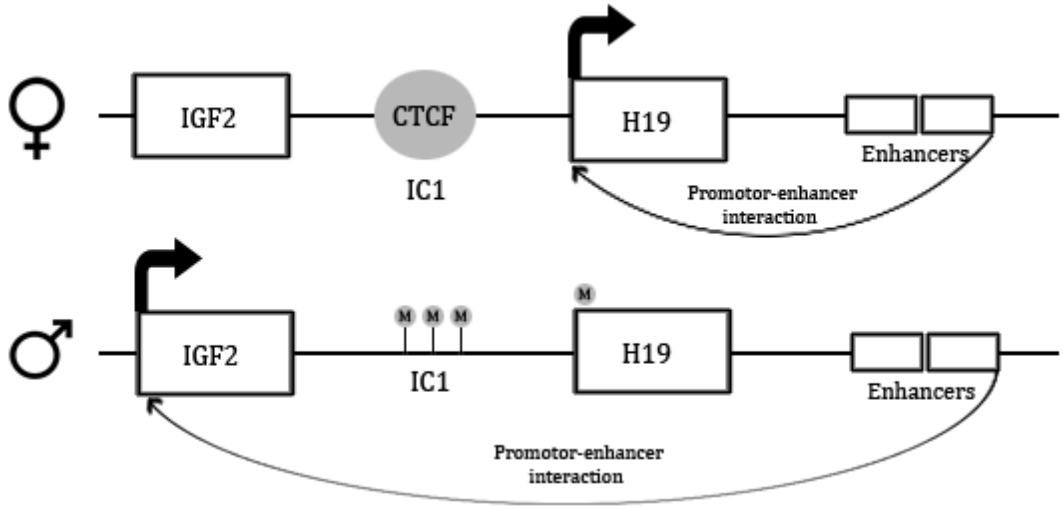

Figure 6. The imprinting regulation of the IGF II gene. Modified from Chao & D'Amore, 2008.

Human chromosome 11p15.5 contains two imprinted domains. In one of these domains, simply referred to as domain 1, IGF II and H19 genes are normally expressed. The IGF II gene is imprinted, in this case meaning that the IGF II gene is only expressed if inherited from the father. H19, on the other hand is only expressed if inherited from the mother. This imprinting is regulated by IC1. IC1 activity is, in turn, regulated by methylation. On the maternal chromosome, IC1 is unmethylated. This enables zinc finger proteins to bind, one of them named CTCF, thereby blocking IGF II promoters and enhancers from interacting. H19 is then expressed while IGF II is silent.

On the paternal chromosome, IC1 is methylated, so that CTCF can not bind; thus, IGF II promoters and enhancers can interact. This leads to IGF II being expressed, while H19 is silent. In BWS, this mechanism is dysfunctional, and there is a dysregulation of gene expression. In 5-10 % of BWS patients, there is a gain of methylation at IC1, meaning that IC1 is methylated on both the maternal and paternal chromosome – biallelic activation of IGF II and biallelic silencing of H19. This leads to a higher propensity for developing cancer and Wilms' tumours. However, there are also other genes involved in the etiology for BWS, such as KCNQ1OT1 and CDKN1C. They involve a region named IC2, which is responsible for most of the molecular defects in BWS. Paternal uniparental disomy of chromosome 11p15 is another possible cause of BWS, meaning that the affected individual inherits two copies of the chromosome from the mother and none from the father (Murrell et al., 2004, Cerrato et al., 2008).

Silver-Russell syndrome

Another growth disorder syndrome associated with mutations in chromosome 11p15 is Silver-Russell/Russell-Silver syndrome (SRS/RSS). In contrast to BWS, SRS is characterized by pre- and postnatal growth retardation. Typical clinical features include macrocephaly and an underdeveloped triangular face with ear anomalies and prominent forehead. Indeed, these abnormalities may contribute to thriving difficulties and emaciation as a consequence thereof.

Epimutations in the IGF II/H19 locus in 11p15 are believed to be one of the two major sources of SRS. The other is a maternal uniparental disomy of chromosome 7.

The epimutations in the IGF II/H 19 locus consist of hypomethylation in the IC1 region. As previously mentioned, hypomethylation would be expected to lead to biallelic silencing of IGF II and expression of H19 instead. Apart from these two main groups, there are also cases of maternal 11p15 duplications causing SRS. In this sense, SRS may well be considered to be the polar opposite of BWS, where hypermethylation and paternal duplication of 11p15 are two of the causes of the disease (Binder et al., 2011).

Doege-Potter syndrome

Doege-Potter syndrome (DPS) is characterized by one or more solitary fibrous tumors (SFT) in the pleural cavity combined with paraneoplastic hypoglycemia. SFTs are on their own rare findings; hypoglycemia has only been documented in 4 % of pleural SFT cases, making DPS an even rarer condition. These tumours secrete high molecular weight (HMW) IGF II ("big IGF") which activates the insulin receptor, thus lowering the circulating levels of glucose.

According to recent discoveries, SFT cells contain abundant levels of IGF II mRNA and diminished amounts of pro-hormone convertase 4, which could well account for reduced endoproteolyzation of pro-IGF II and consequently high levels of HMW IGF II. Normally, IGFBPs would be expected to bind secreted IGF II, but HMW IGF II has a much higher bioavailability than processed IGF II since IGFBPs are less able to bind HMW IGF II (Kalebi et al., 2010).

Atherosclerosis

IGF II promotes cell growth and differentiation by autocrine and paracrine actions, and has been observed in various forms of tumours and growth disorders. Moreover, there is some evidence for IGF II being involved in the formation of atherosclerotic lesions. However, there is a considerable lack of research in this area. Nevertheless, in 2002, a study showed (Zaina et al., 2002) that mice genetically predisposed for atherosclerosis, in combination with homozygosity for a disrupted IGF II-allele, produced aortic lesions 80 % smaller, containing 50 % less proliferating cells, in comparison with mice possessing non-disrupted IGF II-alleles.

Atherosclerotic lesions consist of smooth muscle cells (SMC), inflammatory cells, lipids and extracellular matrix. Inflammatory cells migrate from the blood stream and SMC from adjacent tissue, forming a lesion. The study showed that IGF II clearly contributed to lesion forming by promoting cell differentiation via autocrine and paracrine signalling. The circulating levels of IGF II did not affect the formation of atherosclerotic lesions, but increased local expression of IGF II in SMC resulted in focal intimal thickenings per se. The results in regards to migration of SMC and lipid circulation were inconclusive. It is assumed then that IGF II mainly acts locally by autocrine and paracrine actions in atherosclerosis, but further research is needed to determine whether any systemic effects may also be involved (Zaina et al., 2002).

DISCUSSION

The growth-promoting and beneficial functions of IGF II during embryonic development and placental growth have been well documented. In certain instances, these functions may also work to the disadvantage of the individual. One such scenario is if the mechanisms that regulate expression of IGF II are dysfunctional.

It is accepted that overexpression of the gene leads to overgrowth and in rare cases to Beckwith-Wiedemann syndrome (BWS). It is however likely that other mechanisms not related to IGF II expression are also involved in the development of BWS, since there are BWS cases unrelated to chromosome 11p15.5. Also, biallelic expression of IGF II in itself is insufficient to cause BWS and there are healthy individuals carrying imprinting defects, meaning that further research is needed to fully explain the etiology of the disease (Murrell et al., 2004).

It is also unclear to what extent and exactly how IGF II is involved in Silver-Russell syndrome (SRS). What is known is that almost 50 % of patients with SRS display DNA hypomethylation at the IGF II/H19 locus. But to this date, data reporting a deficiency of IGF II expression following IGF II/H19 disruption are sparse. This might be explained by hypomethylation not leading directly to decreased IGF II production by the liver, but rather to decreased autocrine and paracrine activity at the fetal stage (Binder et al., 2011).

With Doege-Potter syndrome having such a low incidence, and the role of IGF II only recently being implicated in the development of atherosclerotic plaques, only little research can be found on these subjects. However, there are studies suggesting a possible treatment in such cases. Since it has been shown that glucocorticoid injections can dramatically reduce the IGF II gene transcription in rats (Levinovitz & Norstedt, 1989), glucocorticoid administration could be one way of decreasing elevated IGF II levels.

It is also possible to look at the functions of IGF2R to understand the potentially pathological effects of IGF II. As IGF2R regulates the amount of circulating IGF II, disruption of IGF2R expression may cause overgrowth defects in a fashion similar to BWS. Interestingly, the IGF II and IGF2R genes are both imprinted, but on the maternal and paternal side respectively, indicating a conflict of interests between the genders. This example lies in perfect accordance with the notion that growth promoting genes are generally paternally expressed, while growth limiting genes are maternally expressed. An individual with strong IGF II expression will contribute to placental growth and increased birth weight, which is thought to be in the interest of the father, who aims to give his offspring as much resources as possible and gain success at the expense of the mother. On the other hand, the IGF2R responds to this strategy by degrading IGF II, which prevents fetal overgrowth and acts in the interest

of the mother, who aims to preserve her resources and be able to nourish current as well as possible future offspring.

Another intriguing feature of IGF II is the high degree of conservation between species, which suggests a considerable functional and/or evolutionary importance. This high level of homology begs several possible explanations. Since interaction and binding to the IGF II receptor is of utmost importance to the biological actions of IGF II, and the tertiary structure of the ligand contains the information needed for receptor binding, a high level of conservation could be favorable. Using the same logic, the important regulatory functions of IGFBPs would support this theory.

REFERENCES

Alvino, C.L., Ong, S.C., McNeil, K.A., Delaine, C., Booker, G.W., Wallace, J.C., Forbes, B.E., 2011. Understanding the mechanism of insulin and insulin-like growth factor (IGF) receptor activation by IGF-II. *PLoS ONE* 6, e27488.

Binder, G., Begemann, M., Eggermann, T., Kannenberg, K., 2011. Silver-Russell syndrome. *Best Practice & Research: Clinical Endocrinology & Metabolism* 25, 153–160.

Brown, J., Esnouf, R.M., Jones, M.A., Linnell, J., Harlos, K., Hassan, A.B., Jones, E.Y., 2002. Structure of a functional IGF2R fragment determined from the anomalous scattering of sulfur. *The EMBO Journal* 21, 1054–1062.

Brown, J., Jones, E.Y., Forbes, B.E., (2009a). Interactions of IGF-II with the IGF2R/cation independent mannose-6-phosphate receptor mechanism and biological outcomes. *Vitamins & Hormones* 80, 699–719.

Cai, Z., Chen, H.T., Boyle, B., Rupp, F., Funk, W.D., Dedera, D.A., 2005. Identification of a novel insulin-like growth factor binding protein gene homologue with tumor suppressor like properties. *Biochemical and Biophysical Research Communications* 331, 261–266.

Cerrato, F., Sparago, A., Verde, G., De Crescenzo, A., Citro, V., Cubellis, M.V., Rinaldi, M.M., Boccuto, L., Neri, G., Magnani, C., D'Angelo, P., Collini, P., Perotti, D., Sebastio, G., Maher, E.R., Riccio, A., (2008). Different mechanisms cause imprinting defects at the IGF2/H19 locus in Beckwith-Wiedemann syndrome and Wilms' tumour. *Human Molecular Genetics* 17, 1427–1435.

Chao, W., D'Amore
, P.A., (2008). IGF2: epigenetic regulation and role in development and disease. *Cytokine Growth Factor* 19, 111–120.

Clemmons, D.R., (1997). Insulin-like growth factor binding proteins and their role in controlling IGF actions. *Cytokine Growth Factor* 8, 45–62.

Ekström, T.J., Bäcklin, B.M., Lindqvist, Y., Engström, W., 1993. Insulin-like growth factor II in the mink (Mustela vison): determination of a cDNA nucleotide sequence and developmental regulation of its expression. *General and Comparative Endocrinology* 90, 243–250.

Engström, W., Shokrai, A., Otte, K., Granérus, M., Gessbo, A., Bierke, P., Madej, A., Sjölund, M., Ward, A., 1998. Transcriptional regulation and biological significance of the insulin like growth factor II gene. *Cell Proliferation* 31, 173–189.

Favelyukis, S., Till, J.H., Hubbard, S.R., Miller, W.T., 2001. Structure and autoregulation of the insulin-like growth factor 1 receptor kinase. *Nature Structural & Molecular Biology* 8, 1058–1063.

Granérus, M., Johannisson, A., Ekblom, P., Engström, W., 2001. Insulin-like growth factors I and II induce cell death in Wilms's tumour cells. *Molecular Pathology* 54, 30–35.

Hassan, A.B., (2003). Keys to the hidden treasures of the mannose 6-phosphate/insulin-like growth factor 2 receptor. *TheAmerican Journal of Pathology* 162, 3–6.

Kalebi, A.Y., Hale, M.J., Wong, M.L., Hoffman, T., Murray, J., (2009). Surgically cured hypoglycemia secondary to pleural solitary fibrous tumour: case report and update review on the Doege-Potter syndrome. *Journal of Cardiothoracic Surgery*4, 45.

Levinovitz, A., Norstedt, G., 1989. Developmental and steroid hormonal regulation of insulin-like growth factor II expression. *Molecular Endocrinology* 3, 797–804.

López-Bermejo, A., Buckway, C.K., Devi, G.R., Hwa, V., Plymate, S.R., Oh, Y., Rosenfeld, R.G., 2000. Characterization of insulin-like growth factor-binding protein-related proteins (IGFBP-rPs) 1, 2, and 3 in human prostate epithelial cells: potential roles for IGFBP-rP1 and 2 in senescence of the prostatic epithelium. *Endocrinology* 141, 4072–4080.

Murrell, A., Heeson, S., Cooper, W.N., Douglas, E., Apostolidou, S., Moore, G.E., Maher, E.R., Reik, W., (2004). An association between variants in the IGF2 gene and Beckwith-Wiedemann syndrome: interaction between genotype and epigenotype. *Human Molecular Genetics* 13, 247–255.

Otte, K., 1997. The equine IGF genes. Structural and transcriptional features. Doctor's dissertation. ISSN 1401-6257, ISBN 91-576-5418-2.

Pollak, M.N., 2004. Insulin-like growth factors and neoplasia. *Novartis Foundation Symposium*262, 84–98; discussion 98–107, 265–268.

Qiu, Q., Jiang, J.-Y., Bell, M., Tsang, B.K., Gruslin, A., 2007. Activation of endoproteolytic processing of insulin-like growth factor-II in fetal, early postnatal, and pregnant rats and persistence of circulating levels in postnatal life. *Endocrinology* 148, 4803–4811.

Senthil, D., Choudhury, G.G., Abboud, H.E., Sonenberg, N., Kasinath, B.S., 2002. Regulation of protein synthesis by IGF-I in proximal tubular epithelial cells. *American Journal of Physiology - Renal Physiology*283, F1226–1236.

Zainia, S., Pettersson, L., Ahrén, B., Brånén, L., Hassan, A.B., Lindholm, M., Mattsson, R., Thyberg, J., Nilsson, J., 2002. Insulin-like growth factor II plays a central role in atherosclerosis in a mouse model. *The Journal of Biological Chemistry* 277, 4505-4511.