Insulin Receptors and Insulin Action in Heart

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Heart failure is an early and severe complication of diabetes mellitus. The endocrine system and the heart are two interrelated entities, as this is proven by the close relationship between insulin and cardiac function. Insulin significantly contributes to cardioprotection via multiple pathways and various subsequent downstream proteins. Even slight malfunction in the participating pathways can lead to myocardial dysfunction, resulting finally in overt heart failure. At this stage, the implantation of an Left ventricular assist devices (LVADs) and its contribution to the regulation of the neuromodulatory effects of insulin on the heart is pivotal and may decelerate, stabilize, or even revert the deleterious cascades that have been activated in end-stage heart failure.

insulin insulin receptors diabetes mellitus

1. Introduction

Insulin exerts a broad range of effects on the heart, implicating numerous pathways. In other words, the heart is an insulin-dependent organ, meaning that insulin promotes glucose as the primary source of cardiac energy. Insulin signaling affects a variety of myocardium cells, such as cardiomyocytes, fibroblasts, and endothelial cells. As for insulin's roles, among others, it decreases myocardial O^2 consumption, improves cardiac efficiency, assists with myocardial relaxation, and promotes increased blood flow to the myocardium [1]. Insulin signaling in the heart is achieved through insulin and IGF-1 receptors, which bind insulin, IGF-1/2, and insulin receptor substrates 1 and 2 (IRS1 and IRS2). These are the main insulin-signaling elements that regulate cellular metabolism [2]. Moreover, the phosphatidylinositide-3-dependent kinase (PI-3K) activity that controls Akt, the PKB/Akt signaling pathway, and the Janus kinase (JAK)2 pathway contribute primarily to the immediate effect of insulin on the heart muscle [2][3].

Insulin secretory dysfunction and insulin resistance can lead to diabetes mellitus (DM), a disease that accounts for increased cardiovascular morbidity, mortality, and healthcare costs. Nowadays, DM has become one of the most common diseases, and its progression can be proven fatal. The prevalence of DM has increased dramatically in all countries, regardless the income levels [4]. Both type 1 and type 2 DM are quite common, with type 2 DM being more frequent, especially in adults (>65 years), while type 1 DM is most common among younger adults (20–44 years) [5].

DM has been associated with several comorbidities, including heart failure (HF), since HF is due to poor glycemic control [6]. Ventricular assist devices (VADs) have been implanted in many patients suffering from advanced HF. VADs are a pivotal solution, either as a destination therapy or as a bridge toward transplantation, and that is

because they manage to assist cardiac circulation. Recent research indicates that VADs also help control the glycemic level, leading to lower needs for antidiabetic medication and significant improvements regarding glycated hemoglobin, insulin requirements, and glucose levels [7][8].

2. Diabetic Cardiomyopathy

The development of diabetic cardiomyopathy constitutes the leading cause of mortality in diabetic patients. Diabetic cardiomyopathy is a heart disease independent of hypertension or coronary atherosclerosis. There is a large body of evidence implicating insulin deficiency/resistance in the pathogenesis of these disorders. Other than diabetic cardiomyopathy, cardiovascular autonomic neuropathy is also a risk factor for patients with DM, as it leads to increased mortality and left ventricular diastolic and systolic dysfunction. As a matter of fact, research has proven that DM type 1 patients with diabetic autonomic neuropathy have a reduced left ventricular filling pattern, with a more intense LV working load and systolic function [9], while DM type 2 patients with diabetic autonomic neuropathy appeared to have an increased working LV workload, as well as diastolic dysfunction and an increased A/V index [10].

Diabetic myocardium exhibits characteristic fibrosis, interstitial, as well as perivascular, even when coronary disease and hypertension may be absent. Insulin has effects on the myocardium and the endothelium through multiple important pathways and mechanisms, which are analyzed below. Any occurring malfunctions of these mechanisms or abnormalities of the molecules that partake in them can have detrimental results. It is also important to note that most of the studies mentioned, which are related to the action of insulin on the heart and IRs' regulation, have been performed on animal models.

3. The Pathophysiological Pathways of Insulin Action Implicate Akt-mTOR, eNOS, and grk2

Cardiomyocyte death results from numerous cardiac injuries and has a determining role in the development of heart diseases. Therefore, cardioprotection is rather essential and is linked to many molecular and biochemical changes [11]. Specifically, when it comes to insulin and IGF-1, they have anti-apoptotic effects on the heart via mechanisms, some of which are dependent on glucose. These anti-apoptotic signals consist of multiple pathways, with one of the most important being the activation of PI3K/Akt. This pathway then triggers various downstream proteins, such as the mammalian target of rapamycin (mTOR), endothelial nitric oxide synthase (eNOS), glycogen synthase kinase (GSK)-3β, forkhead transcription factors (FOXOs), and certain Bcl-2 family members.

3.1. The Activation of PI3K/Akt

There are many classes of PI3Ks, each of which has a different structure and mode of activation [11]. More specifically, class I PI3K is divided into I_A and I_B because of the different binding subunit p110. Then, class I_A PI3K catalytic subunits include p110α, p110β, p110δ, and the regulatory subunit is mainly p85α, and class I_B PI3K consists of catalytic subunit p110γ, which is regulated with regulator protein p101. In addition, the class I_B PI3K can

be activated by the G-protein-coupled receptor (GPCR)-β, γ, subunits on p110 activation. As for Akt, mammalian genomes contain three Akt genes that encode the following isoforms: Akt1, Akt2, and Akt3.

Meanwhile, Akt/PKB is the main regulator of the signaling pathways, and it determines numerous cellular functions [11]. Akt kinase has a significant action in the cardiovascular system, such as proliferation and cell growth via mTORC1, promotes cell survival via caspase-9, YAP, Bcl-2, and Bcl-x activities, and angiogenesis, vasorelaxation, and cell metabolism via VEGF secretion and mediates eNOS phosphorylation.

Consequently, the alterations of Akt signaling play an important role in many cardiovascular pathological processes such as atherosclerosis, cardiac hypertrophy, and vascular remodeling.

The activation of the PI3K/Akt is a very important step for insulin to have a cardioprotective effect. This mechanism is a response triggered by a wide range of stimuli, such as insulin, insulin-like growth factor-1 (IGF-1), AT II, and reactive oxygen species (ROS) [11]. It is important to mention that Akt activation by insulin is mediated via tyrosine kinase activity of the insulin receptor (IR), IRS-1, and IRS-2.

At first, either insulin attaches to the insulin receptor or IGF-1/2 attaches to the IGF-1 receptor to activate the receptor. As a result of the activation, PI3K binds to the receptor as well through its p85α subunit and then activates I_A PI3K. The latter can also be activated by insulin through IRS-1 and by other tyrosine kinase receptors or cytokine receptors. As for the I_B PI3K, it is activated by GPCRs, such as adenosine and opioid receptors. The activation of PI3K subsequently activates the synthesis of PIP3 and phosphorylates phosphatidylinositol 4,5-bisphospate (PIP2), which can regenerate PIP3. After that, PIP3 can recruit phosphoinositide-dependent kinase (PDK)-1 and Akt, which leads to the phosphorylation of Akt and thus its activation [11]. It is also important to mention that the phosphorylation of Thr308 and Ser473 is needed for Akt to be fully activated. Moreover, Akt may also have the ability of autophosphorylation and to be activated independently of PI3K [11].

There are also mechanisms that negatively regulate Akt and PI3K, such as the regulation from phosphatase and tensin homolog deleted on chromosome 10 (PTEN), which converts PIP3 to PIP2 and blocks the activation of the pathway. For this reason, the reduction in PTEN can lead to activation of the pathway and therefore enhanced cardioprotection. Insulin or IGF-1/2 activation of the PI3K/Akt cascade is protective of the heart by inhibiting apoptosis and oxidative stress. While PI3-K and the atypical protein kinase C family (ζ and λ) are responsible for insulin-induced Glut4 translocation in 3T3-L1 adipocytes and L6 myocytes, Akt1 and Akt2 activity may be responsible for activating glycogen synthase [12].

Akt protein is a serine/threonine kinase and a downstream effector of PI3-K. Akt plays a central role in the metabolic actions of insulin, including glucose transport, and the synthesis of glycogen [12]. Maintaining precise physiological levels of Akt/PKB may be critical to avoiding insulin resistance. This is evidenced by studies linking impaired Akt expression and activity with type 2 diabetes [12].

The regulation of cell size by Akt is thought to be mediated by its phosphorylation and by the subsequent downstream phosphorylation of mTOR on the serine 2448 [13]. In general, the PI3K/Akt pathway manages to protect the heart via several impending signaling molecules, such as eNOS, FOXO, Bad, GSK-3β, mTOR, N-myc downstream regulated gene 2 (NDRG2) [11]. For example, the regulation of cell size by Akt is thought to be mediated by its phosphorylation and by the subsequent downstream phosphorylation of mTOR on the serine 2448 [13]. mTOR contributes to the deterrence of cardiac dysfunction in pathological hypertrophy [11]. More specifically, mTOR stimulates cell growth and metabolism and inhibits excessive hypertrophy, thus conferring cardioprotection and cardiomyocyte survival. mTOR's down activators are two important regulators: p70 S6 kinase 1 (S6K1) and the 4E binding protein 1 (4E-BP1). Out of the two, S6K1 is rather important since the PI3K/AKT/mTOR/S6K1 pathway has an essential share in cardioprotection induced by insulin [11]. As a matter of fact, the excessive activation of mTOR/S6K1 can induce cardiac insulin resistance, while the same pathway can also provide cardioprotection via increased angiotensin II (Ang II) type 2 receptor (AT2R) upregulation and adaptive hypertrophy [14].

Other than mTOR, FOXOs' phosphorylation has a cardioprotective role by restricting their transcriptional activities [11]. Moreover, Bad (one of the Bcl-2 family proteins), if maintained phosphorylated, manages to suppress apoptosis and promote cell survival [11]. GSK-3β is also essential since it is a serine/kinase that plays an important role in the regulation of glycogen synthesis and gluconeogenesis [11].

3.2. The Pathway of Foxo1

Insulin binds to an IR, while IGF-1/2 binds to an IGF-1R, either of which can phosphorylate, among others, IRS1, which induces downstream cascades, such as PI3Ks and MAPKs [15]. The PI3K/Akt pathway phosphorylates multiple pathways, with one of them being the forkhead transcription factor Foxo1. Specifically, Akt phosphorylates Foxo1 at S^{253} and constrains Foxo1's activity, which normally is to regulate physiological functions such as myocardial growth [15].

IRS1 and IRS2 are vital components of insulin signaling, and the loss of IRS1 and IRS2 mediates insulin resistance [15]. In the heart, this results in metabolic dysregulation and heart failure [16], which is interlinked with Akt inactivation and activation of Foxo1. It is important to mention that a decrease in IRS1 and IRS2 with an accompanying activation in Foxo1 is also present in the heart of animals with DM type 2 or insulin resistance [17]. In other words, the parallel inactivation of Akt, activation of Foxo1, and loss of IRS1 and IRS2 are the basis of insulin-resistant cardiomyopathy [15]. Foxo1 also triggers β-MHC gene expression in cardiomyocytes. β-MHC is a target of insulin signaling, and it manages to quell its expression via PI3K activation [15].

3.3. Insulin Growth Factor 1

Insulin-like growth factor 1 (IGF-1) is a single-chain polypeptide that is highly homologous to proinsulin. It is produced in numerous cell types and has both an autocrine and paracrine action. Its activity is arbitrated through binding to the IGF-1 receptor, which strongly binds IGF-1 and IGF-2 but has a low affinity to insulin [18]. In order for

the IGF-1 receptor to be activated, the phosphorylation of IRS is mandatory [19]. Tyrosine-phosphorylated IRS-1 and IRS-2 interact with proteins that contain SH2 domains, guiding to numerous cascade pathways. It is also important to mention the IGF-1 binding proteins (IGFBPs), which bind to IGF-1 and manage IGF-1's binding to the receptor and thus its activity [20].

The IGF-1 signaling cascades are enabled by interactions that lead to the activation of Ras, serine/threonine kinase Raf, and MAP kinase. The latter enables multiple transcription factors and therefore is involved in the IGF-1 stimulation of DNA synthesis and mitogenesis. PI3K is also vital for the metabolic growth and functional effects that insulin and IGF-1 cause, which, among others, include protein and glucose synthesis, apoptosis, and IGF-1-mediated cardiomyocyte contractility [18].

IGF-1 plays an essential role in both cardiac growth and function. When it comes to cardiac growth, IGF-1 boosts cardiac DNA and protein synthesis in isolated cardiomyocytes [21]. Specifically, IGF-1 regulates the cell cycle since, without IGF-1, the cell cannot enter the S phase [22]. Moreover, IGF-1 is associated with hypertrophy and promotes growth through signaling pathways, which include tyrosine kinase, IRS-1, PI3K, and ERK [19].

IGF-1 is both a protective and a risk factor for the heart, and even though IGF-1's role in cardiac function still remains unclear [18], it has quite the potential. For example, IGF-1 is very likely to be used as a treatment for cardiac disorders (e.g., heart failure) since it can upgrade myocardial function both in cardiac patients and healthy adults [18]. Other than that, IGF-1 may improve cardiac contractility by increasing the synthesis of contractile proteins, while at the same time, multiple IGF-1-mediated signaling pathways, such as tyrosine kinase, tyrosine kinase phosphatase, PI3K, and protein kinase C, have also been involved [23]. The activation of the pathways mentioned can lead to an increase in intracellular Ca^{2+}, which is followed by an acute positive myocardial response [18], and to the involvement of ion channels, such as T-type calcium channels [24] and cardiac K^+ channels [25].

IGF-1 can also affect the heart by preventing apoptosis [26]. This mechanism is based on signaling pathways such as tyrosine kinase, MAP kinase, and PI3K, as well as the increased expression of a member of the anti-apoptosis family of Bcl-2 proteins. IGF-1 may act as a survival factor via stimulation of the Bcl-2 family of proteins [18]. It is also important to refer to the impact of IGF-1 deficiency on the heart. Growth hormone (GH) is essential for the heart to maintain its structure and function. In humans, deficiency of IGF-1/GH with obtained GH resistance is highly related to an increased risk of cardiovascular disease [18].

In addition to everything mentioned, IGF-1 betters blood glucose control and improves insulin sensitivity. As a matter of fact, diabetes is characterized by hepatic GH resistance and high levels of IGFBPs, which result in decreased levels of total and free IGF-1 levels. In diabetes, there is also a resistance to IGF-1-mediated myocardial responses, which can be because of increased NOS activity or because of altered protein tyrosine phosphatase activity. Untypical IGF-1 activity is very likely to be associated with diabetes-related vascular disorders and may even lead to cardiac dysfunction, but it is still obscure.

IGF-1 levels and blood pressure are positively interrelated [27]. In hypertensive/hypertrophic states, there is a broad expression of IGF-1 and IGF-1 receptors. This hints that IGF-1 may be involved in pathways that lead to cardiac mechanical dysfunction. Even though insulin/IGF-1 may not play a role in hypertension, insulin/IGF-1 resistance seems to cause hypertension, especially when IGF-1 is associated with excessive NO levels in both vessels and the heart [28].

3.4. The Pathway of eNOS

eNOS, which stands for endothelial nitric oxide synthase, is usually triggered by pathophysiological stimuli, such as vascular endothelial growth factor (VEGF) and stress and is regulated by protein interactions and phosphorylation. eNOS produces nitric oxide (NO) in the endothelium, which is a very important vasoactive compound that also regulates a number of physiological and cellular mechanisms, such as angiogenesis and thrombosis [29]. The regulation of NO's expression by eNOS is rather important since NO is also highly reactive.

eNOS is a calmodulin-dependent enzyme [30]. Its activation is a result of a rise in intracellular calcium and the subsequent activation of the CaM-binding domain by calmodulin [31]. Its activity is triggered by acetylcholine, bradykinin B2 receptor, thrombin, and ATP, via intracellular elevation of calcium levels [32], and it is also dependent on protein phosphorylation and dephosphorylation. Specifically, the phosphorylation of Ser617, 635, and 1179 leads to activated eNOS, while the phosphorylation of Ser116 and Thr497 results in a decrease in functional eNOS [29]. The serine/threonine kinase Akt phosphorylates Ser1177 and Ser1179 [29].

Insulin exerts a significant cardiovascular protective effect via PI3K/Akt/eNOS-dependent signaling in addition to its metabolic modulation [11]. A dysfunction in the Akt/eNOS signaling pathway can majorly affect endothelial function in diabetes mellitus (DM) type 2. This is a result of a reduction in the Akt/eNOS phosphorylation [29]. Meanwhile, insulin binds to insulin receptors or IGFs bind to IGFRs, and signaling pathways are activated, such as the PI3K/Akt/PKB pathway, which mediates many insulin responses, such as myocardial survival and anti-apoptotic effects in endothelial cells. Therefore, the activation of the PI3K/Akt pathway by NO, derived by eNOS, can lead to enhanced endothelial function [33]. Studies have also shown that eNOS's activation is stimulated by insulin and is dependent on the phosphorylation of insulin-responsive cells [34].

NO-based therapies have been researched [35], and NO partakes in the diabetic pathology. Data have pointed out that endothelial dysfunction, abnormal eNOS expression, and NO production are characteristics of diabetes and insulin resistance [29].

3.5. The Action of G-Protein-Coupled Kinase 2

When insulin activates an IR or IGF-1/2 activates an IGF-1R, protein kinase A (PKA) and G-protein receptor kinase 2 (GRK2) phosphorylation of β2AR are induced. This reaction mediates β2AR's disassociation from the complex and its binding to the inhibitory G-protein Gi [36]. This last binding manages to alleviate adrenergic-induced cAMP activities in the heart. The phosphorylation of β2AR depends on insulin receptor substrates 1, 2 (IRS1 IRS2). As a

result, the IR-β2AR interference results in decreased β-adrenergic-induced contractile function in cardiomyocytes and perfused mouse hearts [36].

Both in diabetes and heart failure, there is an elevation of insulin levels, which constantly stimulates IRs. In DM type 2, the IRs of the heart are still sensitive to and are activated by insulin. These elevated levels of insulin can acutely impair β-adrenergic signaling pathways for contractile function in animal hearts [36].

G-protein-coupled kinase 2 (GRK2) has been proven to be a negative controller of insulin receptor signaling [37]. Even though the molecular basis of GRK2 upregulation's negative effects is still unclear, it has been proven that its inhibition in animals has led to the prevention of the development of metabolic disorders, modulating energy expenditure, brown fat function, and insulin actions in peripheral tissues [37]. In these animals, the insulin sensitivity was improved, and they displayed enhanced activation of the insulin mediated Akt pathway in muscle, adipose tissue, and liver [37].

Increased cardiac GRK2 plays a pivotal role in cardiovascular diseases such as heart failure and cardiac ischemia, and this is the reason why its inhibition in mice results in cardioprotection [37]. Other than that, increased GRK2 levels are not only a characteristic of lower cardiac functions and poorer prognosis in HF but also a characteristic of situations of systemic insulin resistance. GRK2 is also capable of inhibiting the IR pathway and regulating the adrenergic receptor signaling and thus has the ability to control body weight gain, adiposity, metabolic rate, and multiple downstream targets of the insulin cascade [38].

Decreased GRK2 levels have a cardioprotective role in many animals and maintain metabolic and pro-survival signals downstream of insulin (such as the Akt/p70S6K pathway and glucose transport) in the hearts of 9-month-old GRK2+/− animals, while the activation of ERK is not affected [37]. In adult mice, downregulation of GRK2 ignites physiological heart hypertrophy and activates a cardioprotective gene expression pathway. Moreover, this downregulation also amplifies the insulin triggered PI3K/Akt pathway [37].

The genetic deletion of GRK2 can result in a reverse of diet-induced obesity and insulin resistance [38]. More specifically, tamoxifen-induced GRK2 ablation in mice leads to a reduction in body weight gain despite the ongoing high-fat diet (HFD) feeding, higher insulin sensitivity, regulated glucose intolerance, and the prevention of adiposity and fatty liver [38].

These effects are based on tissue-specific processes such as improved insulin signaling in peripheral tissues, improved lipolysis in white adipose tissue (WAT) and brown adipose tissue (BAT), elevated expression of mRNAs that encode proteins partaking in fatty acid oxidation and thermogenesis in BAT, and lower levels of steatosis and liver inflammation [38].

References

1. Iliadis, F.; Kadoglou, N.; Didangelos, T. Insulin and the heart. Diabetes Res. Clin. Pract. 2011, 93 (Suppl. 1), S86–S91.
2. Velloso, L.A.; Carvalho, C.R.; Rojas, F.A.; Folli, F.; Saad, M.J. Insulin signalling in heart involves insulin receptor substrates-1 and -2, activation of phosphatidylinositol 3-kinase and the JAK 2-growth related pathway. Cardiovasc. Res. 1998, 40, 96–102.
3. Guo, C.A.; Guo, S. Insulin receptor substrate signaling controls cardiac energy metabolism and heart failure. J. Endocrinol. 2017, 233, R131–R143.
4. NCD Risk Factor Collaboration (NCD-RisC). Worldwide trends in diabetes since 1980: A pooled analysis of 751 population-based studies with 4.4 million participants. Lancet 2016, 387, 1513–1530.
5. Xu, G.; Liu, B.; Sun, Y.; Du, Y.; Snetselaar, L.G.; Hu, F.B.; Bao, W. Prevalence of diagnosed type 1 and type 2 diabetes among US adults in 2016 and 2017: Population based study. BMJ 2018, 362, k1497.
6. Didangelos, T.; Kantartzis, K. Diabetes and Heart Failure: Is it Hyperglycemia or Hyperinsulinemia? Curr. Vasc. Pharmacol. 2020, 18, 148–157.
7. Patel, N.; Gluck, J.A.; Radojevic, J.; Coleman, C.; Baker, W.L. Left ventricular assist device implantation improves glycaemic control: A systematic review and meta-analysis. ESC Heart. Fail. 2018, 5, 1141–1149.
8. Goetz, M.E.; Charnigo, R.; Guglin, M. Implantation of Left Ventricular Assist Device Results in Immediate Improvement of Glucose Metabolism in Patients with and Without Diabetes Mellitus. Heart Lung Circ. 2020, 29, 931–935.
9. Didangelos, T.P.; Arsos, G.A.; Karamitsos, D.T.; Athyros, V.G.; Karatzas, N.D. Left Ventricular Systolic and Diastolic Function in Normotensive Type 1 Diabetic Patients with or Without Autonomic Neuropathy. Diabetes Care 2003, 26, 1955–1960.
10. Didangelos, T.P.; Arsos, G.; Karamitsos, T.; Iliadis, F.; Papageorgiou, A.; Moralidis, E.; Athyros, V. Left Ventricular Systolic and Diastolic Function in Normotensive Type 2 Diabetic Patients with or Without Autonomic Neuropathy. Angiology 2013, 65, 877–882.
11. Yao, H.; Han, X.; Han, X. The Cardioprotection of the Insulin-Mediated PI3K/Akt/mTOR Signaling Pathway. Am. J. Cardiovasc. Drugs 2014, 14, 433–442.
12. Lee, J.H.; Ragolia, L. AKT phosphorylation is essential for insulin-induced relaxation of rat vascular smooth muscle cells. Am. J. Physiol. Physiol. 2006, 291, C1355–C1365.
13. Sharma, S.; Guthrie, P.H.; Chan, S.S.; Haq, S.; Taegtmeyer, H. Glucose phosphorylation is required for insulin-dependent mTOR signalling in the heart. Cardiovasc. Res. 2007, 76, 71–80.

14. Pulakat, L.; Demarco, V.G.; Whaley-Connell, A.; Sowers, J.R. The Impact of Overnutrition on Insulin Metabolic Signaling in the Heart and the Kidney. Cardiorenal Med. 2011, 1, 102–112.
15. Qi, Y.; Zhu, Q.; Zhang, K.; Thomas, C.; Wu, Y.; Kumar, R.; Baker, K.M.; Xu, Z.; Chen, S.; Guo, S. Activation of Foxo1 by Insulin Resistance Promotes Cardiac Dysfunction and β–Myosin Heavy Chain Gene Expression. Circ. Heart Fail. 2015, 8, 198–208.
16. Qi, Y.; Xu, Z.; Zhu, Q.; Thomas, C.; Kumar, R.; Feng, H.; Dostal, D.E.; White, M.F.; Baker, K.M.; Guo, S. Myocardial Loss of IRS1 and IRS2 Causes Heart Failure and Is Controlled by p38α MAPK During Insulin Resistance. Diabetes 2013, 62, 3887–3900.
17. Guo, S. Insulin signaling, resistance, and the metabolic syndrome: Insights from mouse models into disease mechanisms. J. Endocrinol. 2014, 220, T1–T23.
18. Ren, J.; Samson, W.K.; Sowers, J.R. Insulin-like Growth Factor I as aÈCardiac Hormone: Physiological and Pathophysiological Implications in Heart Disease. J. Mol. Cell. Cardiol. 1999, 31, 2049–2061.
19. Leroith, D. Insulin-like growth factor receptors and binding proteins. Bailliere's clinical endocrinology and metabolism. 1996, 10, 49–73.
20. Jones, J.I.; Clemmons, D.R. Insulin-Like Growth Factors and Their Binding Proteins: Biological Actions. Endocr. Rev. 1995, 16, 3–34.
21. Fuller, S.J.; Mynett, J.R.; Sugden, P.H. Stimulation of cardiac protein synthesis by insulin-like growth factors. Biochem. J. 1992, 282 Pt 1, 85–90.
22. Reiss, K.; Cheng, W.; Ferber, A.; Kajstura, J.; Li, P.; Li, B.; Olivetti, G.; Homcy, C.J.; Baserga, R.; Anversa, P. Overexpression of insulin-like growth factor-1 in the heart is coupled with myocyte proliferation in transgenic mice. Proc. Natl. Acad. Sci. USA 1996, 93, 8630–8635.
23. Florini, J.R.; Ewton, D.Z.; Coolican, S.A. Growth Hormone and the Insulin-Like Growth Factor System in Myogenesis. Endocr. Rev. 1996, 17, 481–517.
24. Piedras-Rentería, E.S.; Chen, C.-C.; Best, P.M. Antisense oligonucleotides against rat brain α1E DNA and its atrial homologue decrease T-type calcium current in atrial myocytes. Proc. Natl. Acad. Sci. USA 1997, 94, 14936–14941.
25. Guo, W.; Kada, K.; Kamiya, K.; Toyama, J. IGF-I regulates K(+)-channel expression of cultured neonatal rat ventricular myocytes. Am. J. Physiol. Content 1997, 272 Pt 2, H2599–H2606.
26. Wang, L.; Ma, W.; Markovich, R.; Chen, J.-W.; Wang, P.H. Regulation of Cardiomyocyte Apoptotic Signaling by Insulin-like Growth Factor I. Circ. Res. 1998, 83, 516–522.
27. Andronico, G.; Mangano, M.T.; Nardi, E.; Mulè, G.; Piazza, G.; Cerasola, G. Insulin-like growth factor 1 and sodium—Lithium countertransport in essential hypertension and in hypertensive left ventricular hypertrophy. J. Hypertens. 1993, 11, 1097–1101.

28. Wickman, A.; Friberg, P.; Adams, M.A.; Matejka, G.L.; Brantsing, C.; Guron, G.; Isgaard, J. Induction of Growth Hormone Receptor and Insulin-Like Growth Factor-I mRNA in Aorta and Caval Vein During Hemodynamic Challenge. Hypertension 1997, 29, 123–130.
29. Kolluru, G.K.; Siamwala, J.H.; Chatterjee, S. eNOS phosphorylation in health and disease. Biochimie 2010, 92, 1186–1198.
30. Marletta, M.A. Nitric oxide synthase: Aspects concerning structure and catalysis. Cell 1994, 78, 927–930.
31. Fleming, I.; Bauersachs, J.; Fisslthaler, B.; Busse, R. Ca2+ -Independent Activation of the Endothelial Nitric Oxide Synthase in Response to Tyrosine Phosphatase Inhibitors and Fluid Shear Stress. Circ. Res. 1998, 82, 686–695.
32. Mombouli, J.V.; Vanhoutte, P.M. Kinins and Endothelial Control of Vascular Smooth Muscle. Annu. Rev. Pharmacol. Toxicol. 1995, 35, 679–705.
33. Kawasaki, K.; Smith, R.S.; Hsieh, C.-M.; Sun, J.; Chao, J.; Liao, J.K. Activation of the Phosphatidylinositol 3-Kinase/Protein Kinase Akt Pathway Mediates Nitric Oxide-Induced Endothelial Cell Migration and Angiogenesis. Mol. Cell. Biol. 2003, 23, 5726–5737.
34. Zeng, G.; Nystrom, F.H.; Ravichandran, L.V.; Cong, L.-N.; Kirby, M.; Mostowski, H.; Quon, M.J. Roles for Insulin Receptor, PI3-Kinase, and Akt in Insulin-Signaling Pathways Related to Production of Nitric Oxide in Human Vascular Endothelial Cells. Circulation 2000, 101, 1539–1545.
35. Masters, K.S.B.; Lipke, E.A.; Rice, E.E.H.; Liel, M.S.; Myler, H.A.; Zygourakis, C.; Tulis, D.A.; West, J.L. Nitric oxide-generating hydrogels inhibit neointima formation. J. Biomater. Sci. Polym. Ed. 2005, 16, 659–672.
36. Fu, Q.; Xu, B.; Liu, Y.; Parikh, D.; Li, J.; Li, Y.; Zhang, Y.; Riehle, C.; Zhu, Y.; Rawlings, T.; et al. Insulin Inhibits Cardiac Contractility by Inducing a Gi-Biased β2-Adrenergic Signaling in Hearts. Diabetes 2014, 63, 2676–2689.
37. Lucas, E.; Jurado-Pueyo, M.; Fortuño, M.A.; Fernández-Veledo, S.; Vila-Bedmar, R.; Jiménez-Borreguero, L.J.; Lazcano, J.J.; Gao, E.; Gómez-Ambrosi, J.; Frühbeck, G.; et al. Downregulation of G protein-coupled receptor kinase 2 levels enhances cardiac insulin sensitivity and switches on cardioprotective gene expression patterns. Biochim. Biophys. Acta (BBA) Mol. Basis Dis. 2014, 1842, 2448–2456.
38. Vila-Bedmar, R.; Cruces-Sande, M.; Lucas, E.; Willemen, H.L.D.M.; Heijnen, C.J.; Kavelaars, A.; Mayor, F.; Murga, C. Reversal of diet-induced obesity and insulin resistance by inducible genetic ablation of GRK2. Sci. Signal. 2015, 8, ra73.