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Review Article

INSULIN: NON-ENDOCRINAL ACTIONS AND APPLICATIONS

Alka Bansal[1*], Uma Advani[2], Neha Sharma [3] , Z.Y Khan[4]
[1]Assistant Professor , Dept of Pharmacology ,SMS Medical College, Jaipur, Rajasthan.
[2]Senior Demonstrator, Dept of Pharmacology , SMS Medical College, Jaipur, Rajasthan.
[3]2nd year resident, Dept of Pharmacology, NIMS Medical College, Shobha Nagar, Jaipur-302001.
[4]Professor , Dept of Pharmacology, M.G. Medical College, Jaipur-302001, Rajasthan, India.

*Corresponding Author: Alka Bansal; Email: alkabansal04@gmail.com

Abstract: Amongst the non-endocrinal actions, insulin in cardiovascular system causes positive inotropic action & reduces peripheral vascular resistance. Due to these actions it can be useful in Congestive Heart Failure and digitalis toxicity. Glucose-Insulin-Potassium infusion enhances left ventricular function, in patients with recent myocardial infarction as well as in patients with chronic myocardial infarction. Besides these insulin has anti-inflammatory action, which can be used for rapid wound healing. Insulin also keeps blood vessels in the brain healthy and is useful in Alzheimer's Disease to improve cognitive function. It plays an important role in growth & has an anabolic action. That is why deficiency of insulin in pregnancy causes low birth weight of newborn. Insulin by suppressing ghrelin increases appetite. Insulin increases antiaging protein (K-lotho). Animal studies have shown that overexpression of K-lotho extends life span. It also potentiates the cellular responses of chemotherapeutic agents in cancer and adding intravenous insulin to standard treatment, decreases the standard doses of chemotherapeutic agents required and treatment is less toxic also. It also promotes cell growth, cell division, migration, and inhibits apoptosis. So the non endocrinal applications of insulin are as an adjuvant in congestive heart failure , arrhythmia, myocardial infarction, digitalis toxicity, wound healing, neurodegenerative diseases, stroke & along with chemotherapeutic agents in cancer.
Key words: Insulin, Nonendocrinal actions, Nonendocrinal uses

INTRODUCTION:

The discovery of insulin by Banting and Best, almost a century ago, changed the scenario in the treatment of Diabetes Mellitus (DM). Since then many scientists have worked on this molecule and confirmed the molecular mechanism of action & its role in the treatment of once considered lethal disease. But besides DM, multiple non-endocrinal uses have also been discovered and suggested. Some of these suggested non-endocrinal actions and applications of insulin are as follows-

CARDIOVASCULAR SYSTEM:

Cardiac cells have 10,000-100,000 insulin receptors per cell.[1] Insulin facilitates potassium reuptake into the myocardial cells and thus stabilizes them, which results in a lower occurrence of arrhythmias after Myocardial Infarction (MI).[2] Insulin binds to the α-subunit of the insulin receptors that activates the tyrosine kinase leading to autophosphorylation of tyrosine residues in several regions of the intracellular β-subunit and the tyrosine phosphorylation of the Insulin Receptor Substrate (IRS) proteins. Positive inotropic action of insulin was not related to catecholamine release and subsequent stimulation of beta-adrenergic receptors, but it enhances the activity of sarcoplasmic reticulum calcium ATPase and trans-sarcolemmal Ca2+ entry, mainly via reverse-mode Na+Ca2+ exchange and insulin-mediated activation of Na+H+ exchange, presumably via elevated intracellular Na+ concentration.[3,4] One of the earliest steps in the insulin signaling pathway is the activation of Phosphoinositol 3-Kinase (PI3-kinase). Insulin also increases the cardiac contractility by opening the voltage dependent Non-Selective Cation Channel (NSCC) & ligand gated channel activated by Diacylglycerol (DAG). In addition, NSCC is also activated through Phospholipase-C (PL-C) pathway,[3] as shown in figure 1.

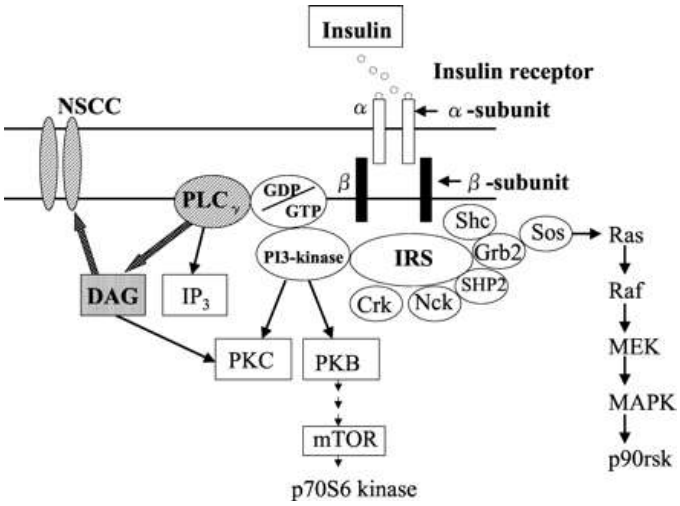

Figure 1: Insulin signaling pathway & activation of NSCC in cardiac cell.

Studies have shown that L-type Ca2+ channel blockers (e.g. diltiazem), and sarcoplasmic reticulum Ca2+ release channel blocker (e.g. ryanodine) did not affect the inotropic response to insulin. However, blockade of sarcoplasmic

reticulum Ca2+ATPase (cyclopiazonic acid), inhibition of Na+H+ exchange (HOE642) and inhibition of Na+Ca2+ exchange (SEA0400) partially prevents the inotropic response to insulin.[5]

Positive inotropy is partially dependent on increased glucose uptake. Normally free fatty acids are main substitutes metabolised by myocardial cell, but when given as Glucose-Insulin-Potassium infusion (GIK), [6, 7] glucose becomes the main substrate for energy in mycordium due to decreased availability of circulating free fatty acids & enhanced myocardial glucose uptake. This decreases the oxygen requirement.[8,9,10,11] Systemic insulin administration results in a reduction of circulating free fatty acids and an improvement in myocardial glucose uptake, which causes an efficiency improvement in the myocardial cell.

To prevent the drop in blood glucose levels, insulin administration is combined with infusion of glucose; and potassium is usually added to prevent hypokalaemia caused by simultaneous entry of potassium into the cell with glucose. During GIK administration, the uptake of free fatty acids was reduced[8] thereby decreasing the oxygen requirement which is beneficial in ischemic heart disease. In addition to above clinical applications glucose insulin infusion has also been tried in acute digitalis toxicity in rats. There it delays abnormality in cardiac conduction and serves as a cardioprotective agent. Studies have shown insulin at low concentration blunted the effect of digoxin on Na^+/K^+-ATPase activity.

Insulin also increases the expression of endogenous Nitric Oxide Synthetase enzyme (eNOS) via PI3K pathway and causes coronary vasodilatation. Intimal hyperplasia exerts a critical role in vein graft failure after arterial bypassing. Insulin induces increase in circulating IGF-1 which reduces vascular inflammatory responses, vascular oxidant stress and possibly thereby decreases intimal hyperplasia.[12] Postoperative insulin use greatly inhibits cell proliferation, reduces intimal & medial thickness, upregulates endothelial nitric oxide synthase, and [alpha]-smooth muscle actin expression. Insulin protects autologous vein grafts possibly through the phosphatidylinositol-3 kinase/Akt signaling pathway and prevents intimal hyperplasia in autologous vein grafts.[13] Insulin is a growth factor and increases proliferation of vascular smooth muscle cells in vitro. However, smooth muscle cells growth stimulation takes place at very high insulin concentrations. In contrast, low dose insulin stimulates endothelial production of nitric oxide, which leads to vasodilatation.

Rho A --- MLC--- VASODILATATION

INSULIN: ---PI3K(AKT)

HSP 90--- eNOS--- VASODILATATION

Insulin decreases C-Reactive protein around 40%. It also reduces the generation of free radicals and cytochrome C. This decreases the activation of cytochrome C dependent caspase pathway which is responsible for the apoptosis of myocytes.[7,9]

All the above cardio-vascular actions of insulin are therapeutically applied in clinical conditions like atrial fibrillation, congestive heart failure, hypertension, myocardial infarction, post MI arrhythmias & to prevent reperfusion injury in coronary bypass grafting.[8]

In a few patients with calcium channel blocker poisoning who were not responding to conventional therapy of calcium and epinephrine, high dose of insulin-dextrose infusion is found to be beneficial by inducing positive inotropic effect.[14,15]

ANTIINFLAMMATORY:

Diabetes is characterized by a proinflammatory state, and several inflammatory processes have been associated with both type 1 and type 2 diabetes and the resulting complications. The research data illustrate the anti-inflammatory effects of insulin and proinflammatory effects of glucose[10] & provide mechanistic justification for the benefits of maintaining euglycemia with insulin infusion in patients with inflammation. [16] Insulin has anti-inflammatory action predominantly by reducing the IL-6, IL-18 & TNF – alpha production via Mitogen Activated Protein Kinase (MAPK) pathway & mTOR pathway respectively. Insulin decreases the leucocyte adherence to endothelium via AKT pathway; (as shown in Figure 2) and also enhances phagocytosis thereby arresting the bacterial growth. Hence Protamine Zinc Insulin (PZI) is used to facilitate the wound healing in open wounds, surgical incision and laceration.[17,18]

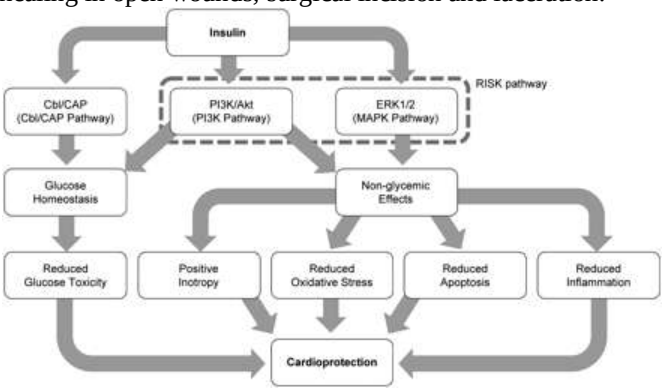

Figure 2: Insulin: modes of non endocrinal actions. {CAP indicates Cbl-associated protein; PI3K- phosphatidyl-inositol 3-kinase; ERK1/2-extracellular signal-regulated kinase; RISK- reperfusion injury salvage kinase; and MAPK- mitogen-activate protein kinase.}

Studies have proved that if insulin is added to human cell culture, it stimulates the proliferation and migration of keratinocytes and in microvascular culture it promotes migration of cells.[19,20] Studies have shown that even the topical application of insulin directly to the wound accelerates the healing process. A carbon- based nano-diamond preparation of insulin has been developed which releases insulin topically at basic pH of skin and facilitates wound healing.

CENTRAL NERVOUS SYSTEM:
Endogenous insulin keeps blood vessels to brain healthy and allows the brain to function normally. Deficiency of insulin or insulin resistance impairs the cognitive functions (especially verbal) of brain & has been implicated in the formation of plaques in Alzeimer's Disease (AD). So AD is also known as Type 3 DM.[21] Studies have shown that exogenous insulin administration improves memory both in healthy volunteers and those suffering from AD. Lack of insulin signaling in the brain may lead to changes in Akt and GSK3 activity and Tau hyperphosphorylation but must interact with other mechanisms for development of Alzheimer's disease. There is a common cellular pathway in the neurodegenerative diseases with abnormal mitochondrial function and abnormal glucose metabolism. This common pathway involves Peroxisome Proliferator Activated Receptor (PPAR)-gamma, a key regulatory enzyme involved in mitochondrial resistance and insulin resistance. So insulin supplementation can be useful in Parkinson's disease and Hungtinton's disease[22,23].

High glutamate levels are associated with increased risk of stroke. This has been reiterated by the fact that glutamate scavenger oxaloacetate decreases the risk of acute ischaemic stroke[24]. Insulin causes reduction in circulatory amino acids including glutamate & thereby proves useful in acute ischaemic stroke treatment.
Previously insulin was also used in narcoanalysis to reduce an individual's self control over one's thought and cause him to reveal the true thoughts & feelings which the person would have suppressed. Its use in Schizophrenia has been obsoleted due to availability of better drugs.[25,26]

Experimental data establish the effectiveness of insulin treatment in improving neurologic recovery in spinal cord injury , increasing the expression of anti-apoptotic bcl-2 proteins, inhibiting caspase-3 expression decreasing neuronal apoptosis, reducing the expression of proinflammatory cytokines iNOS and COX-2, and ameliorating microcirculation of injured spinal cord after moderate contusive spinal cord injury in rats. The studies report the beneficial effects of insulin in the treatment of spinal cord injury , with the suggestion that insulin should be considered as a potential therapeutic agent.[27,28]

GROWTH AND ANABOLIC ACTION-
Hormones such as insulin, insulin-like growth factors, thyroxine and the glucocorticoids act as nutritional & maturational signals and adapt fetal development to prevailing intrauterine conditions, thereby maximizing the chances of survival both in utero and at birth[29,30]. Insulin is required for the growth even in foetal stage. Its deficiency leads to low birth weight, decreased crown-rump length at birth, fetal growth retardation in uterus[29].

Insulin has been used in the provocative test for the diagnosis of Growth Hormone (GH) deficiency. When insulin is given, blood glucose levels decrease & in normal conditions GH is released because GH is counter regulatory hormone. Failure to rise of GH in response to insulin suggests GH deficiency.[31,32]

Ghrelin is an appetite suppressant and insulin (both exogenous and endogenous) causes suppression of ghrelin. So insulin increases the appetite & is abused at times for increasing body weight and muscle mass by athletes. It has also been utilized in bed ridden patients and those admitted for surgery because surgery induces a catabolic state.

ANTI-AGING EFFECT :
Klotho (KL), an antiaging protein, was named after the goddess who spins the threads of life . Mice lacking KL exhibit many changes that occur during aging include atherosclerosis, osteoporosis, infertility, and cognitive decline. They also have a shorter life span. In contrast, mice over expressing KL live 30% longer than wild-type mice and are more resistant to oxidative stress.[16] Release of insulin increases K-lotho protein.[33,34,35]

POTENTIATION OF CHEMOTHERAPEUTIC AGENTS: The effect of insulin and Insulin like Growth Factor (IGF-1) on cellular responses to chemotherapeutic drugs (5-fluorouracil, oxaliplatin and irinotecan) in colon cancer and endothelial cells was analysed *in vitro* and they were shown to modulate the cellular responses.[32,36] It might be due rapid cell division phase in which these agents act.[31] Use of low doses of chemotherapeutic agents with increased frequency along with insulin suggest another method called Insulin Potentiated Therapy (IPT) or now called Insulin Potentiated Targeted Low-Dose therapy (IPTLD) . In IPTLD 10 times lower doses of chemotherapeutic agents are given at shorter intervals. This treatment has very low toxicity and efficacy was same as that of standard chemotherapy. So it improves the quality of life. [6,16,17,28]

MISCELLANEOUS:
Insulin is also used in organ preserving solutions. Organs extracted for transplant purposes are preserved in insulin containing solutions.[18] It promotes cell growth, cell division, migration, and inhibits apoptosis. These aspects of insulin action are collectively known as the "mitogenic actions" of insulin and because they are so critical to cellular physiology.

Insulin is always present in cell culture medium for the propagation and maintenance of cells in culture.[28]

In order for all growth factors to stimulate mitogenesis, they must activate the Ras-Raf-Map kinase signalling pathway. [11] Unwanted effect of exogenous hyperinsulinemia in the setting of insulin resistance includes promotes progression of cancer and atherosclerosis via phosphorylation and activation of farnesyltransferase, a ubiquitous enzyme that farnesylates Ras protein.

Increased availability of farnesylated Ras at the plasma membrane enhances mitogenic responsiveness of cells to various growth factors, thus contributing to hypoglycemia, progression of cancer & atherosclerosis. This effect is specific to insulin, but is not related to the type of insulin used.[11]

The PI3K/AKT/mTOR pathway is an intracellular signalling pathway important in inhibiting apoptosis . It is activated by IGF-1 and has a number of downstream effects which either promote protein synthesis or inhibit protein breakdown. PI3K activation activates AKT which activates mTOR. In many cancers this pathway is overactive reducing apoptosis and allowing proliferation & is involved in progression of cancers like breast cancer[8] and non-small-cell lung cancer.[11]

CONCLUSION

The conventional role of insulin in carbohydrate metabolism is well known since decades.It has been proved that half of insulin is utilized for non-nutrient metabolic needs. These non-nutrient metabolic uses are cardioprotective in CCF, MI, anti-inflammatory, in wound healing , role in memory, anabolic action , increases antiaging protein Klotho and potentiates cellular responses of anticancer chemotherapeutic agents. So the benefit of insulin should be weighed against harmful effect of hyperinsulinemia while deciding the therapeutic dose in particular patient. Further extensive studies for nonendocrinal uses of insulin are required to weigh & establish the non-endocrinal status of insulin against its adverse effects.

REFERENCES

1. Hiraoaka Masayasu .A Novel Action of Insulin on Cardiac Membrane. *Circulation Research*., **2003**;92,7: 707-709.
2. Horst Van Der and Zijlstra F. Potential beneficial mechanisms of insulin (glucose-potassium) in acute MI. *Neth Heart J.* 2005; 13(6): 233-234, 235-238
3. Zhang Yin Hua and Hancox Jules C. A Novel, Voltage-Dependent Nonselective Cation Current Activated by Insulin in Guinea Pig Isolated Ventricular Myocytes. *Circ Res.* **2003**;92:765-768.
4. Lewinski Dirk von , Bruns Sebastian , Walther Stefanie , Kögler Harald , Pieske Burkert . Insulin Causes $[Ca^{2+}]_i$-Dependent and $[Ca^{2+}]_i$-Independent Positive Inotropic Effects in Failing Human Myocardium. *Circulation.* **2005**; 111: 2588-25955.
5. Hsu Chih-Hsueng & Wei Jeng et al. Cellular Mechanisms Responsible for the Inotropic Action of Insulin on Failing Human Myocardium. *The Journal of Heart and Lung Transplantation*. **2006**; 25, 9 , 1126-1134.
6. isser L., Zuurbier C.J., Hoek F.J.et al. GIK applied as perioperative insulinaemic normoglycemic clamp: effects on inflammatory response during coronary artery surgery. *British Journal of Anaesthesia.* **2005**; 95,4,448-57,2005.
7. Marano L, Bestetti A, Lomuscio A, Tagliabue L, Castini D, Tarricone D, et al. Effects of infusion of glucose-insulin-potassium on myocardial function after a recent myocardial infarction. *Acta Cardiol.* **2000**; 55:9-15.
8. Klein L.J. and Visser F.C. The effect of insulin on the heart Part 1: Effects on metabolism and function. *Neth Heart J.* 2010; 18(4): 197–201.
9. Cottin Y, Lhuillier I, Gilson L, Zeller M, Bonnet C, Toulouse C, et al. Glucose insulin potassium infusion improves systolic function in patients with chronic ischemic cardiomyopathy. *Eur J Heart Fail.* **2002**;4:18-4.
10. Chawla Ajay . Pleiotropic Actions of Insulin Resistance and Inflammation in Metabolic Homeostasis. Justin I. Odegaard[1], *Science*, **2013**: 339 (6116): 172-177
11. **Draznin Boris.** Mechanism of the mitogenic influence of hyperinsulinemia **.** *Diabetology & Metabolic Syndrome.* **2011**, **3**:10 (1186/175).
12. Murthy Subramanyam N. , Sukhanov Sergiy , et al. Insulin Glargine reduces carotid intimal hyperplasia after balloon catheter injury in Zucker fatty rats possibly by reduction in oxidative stress. *Mol Cell Biochem.* 2009; 330(1-2): 1–8.
13. Zhou N, Yu QJ, Si R, Gao HK, Wang T, Gao F, Wang HC, Bian JF. Postprocedure administration of insulin in canine autologous vein grafting: a potential strategy to attenuate intimalhyperplasia. *Cardiovasc Pharmacol.* **2010**;56(4):402-12.
14. Yuan TH, Kerns WP, Tomaszewski CA, Ford MD, Kline JA, Kline J. J. Insulin-glucose as adjunctive therapy for severe calcium channel antagonist poisoning. *Toxicol Clin Toxicol,* **1999**;37:463-74
15. Oubaassine R, Weckering M, Kessler L, Breidert M, Roegel JC, Eftekhari P. Insulin interacts directly with Na^+/K^+ATPase and protects from digoxin toxicity. *Toxicology.* **2012**; 4; 299(1):1-9.
16. Keng Wooi Ng, Meredith L. Allen, Ajay Desai, Duncan Macrae, Nazima Pathan, Contemporary Reviews in Cardiovascular Medicine. Cardioprotective Effects of Insulin How Intensive Insulin Therapy May Benefit Cardiac Surgery Patients. *Circulation* .**2012** ;125(5): 721-728.
17. Hoogwerf Byron J .Postoperative management of the diabetic patient. Medicinal clinics of North America. **2001**; 85,5.
18. Dagogo Samuel, George Jack K., Alberti M.M.. Management of Diabetes Mellitus in surgical patients. *Diabetes spectrum.* **2002**. 15, 44-48
19. Hayashi I, Larner J, Sato G. Hormonal growth control of cells in culture. *Cell Dev Biol*. **1978**;14:23-30
20. Sasaoka T, Rose DW, Jhun BH, Saltiel AR, Draznin B, Olefsky JM. Evidence for a functional role of Shc proteins in mitogenic signaling induced by insulin, insulin-like growth factor-1, and epidermal growth factor. *J Biol Chem* **1994**, 269:13689-13694
21. Alzeimer'-type3 DM. New York Times opininion . **2012**
22. Olmos Iciar Aviles, Limousin Patricia, Lees Andrew, Foltynie Thomas. Parkinson's Disease, Insulin Resistance and Novel Agents of Neuroprotection. *Brain.* **2013**;136 (2):374-384.
23. Wu SS, Fruch SJ. Treatment of Parkinson's disease: What is on horizon. *CNS Drugs* **2005**;19(9);723-43

24. Fanne RA, Nassar T, Heyman SN, Hijazi N, Higazi AA. Insulin and glucagon share the same mechanism of neuroprotection in diabetic rats: role of glutamate. *Am J Physiol Regul Integr Comp Physiol*. **2011;** 301(3):668-73.
25. Niazi AK, Niazi SK. A grand dame with hidden aces:The non-diabetic uses of insulin. *Indian J Endocr Metab*. **2012**:16:57-9
26. Doroshow Deborah Blythc. Schizophrenia. *J of History of Med. & allied Sciences*. **2007.** 62, 2, ,213-6
27. Wu XH, Yang SH, Duan DY, Cheng HH, Bao YT, Zhang Y. Anti-apoptotic effect of insulin in the control of cell death and neurologic deficit after acute spinal cord injury in rats. *J Neurotrauma.* **2007** ;24(9):1502-12.
28. Schubert Markus, Gautam Dinesh. Role for neuronal insulin resistance in neurodegenerative diseases. *PNAS,* **2004** ;101(9):3100-3105..
29. Fowden AL, Forhead AJ. Endocrine mechanisms of intrauterine programming. *Reproduction.* 2**004**;127(5):515-26.
30. Fowden AL. Early human development. **1992**; 29(1-3)177
31. EInzuchhi Silvio et al .Counter point: Inpatient glucose management.A premature call to arms. *Diabetes Care*. **2005**. 28, 4.
32. Fowden A, L, Forhead A, J, Horm .Endocrine Regulation of Feto-Placental Growth. Res Paediatr. 2009;72:257-265
33. Kurosu H, Yamamoto M. Suppression of aging in mice by the hormone K*lotho. Science.* **2005**; 16;309(5742):1829-33.
34. Sethi Ashish . Klotho An Anti- Aging Gene. *International Journal of Pharma and Bio Sciences.* **2011**; 2(1):498.
35. Chen CD, Podvin S, Gillespie E, Leeman SE, Abraham CR. Insulin stimulates the cleavage and release of the extracellular domain of Klotho by ADAM10 and ADAM17. *Proc Natl Acad Sci U S A* **2007**;104:19796-801
36. Damyanov C, Radoslavova M, Gavrilov V, Stoeva D. J .Low dose chemotherapy in combination with insulin for the treatment of advanced metastatic tumors: Preliminary experience. *BUON* **2009**;14:711-5