

The Insulin:Glucagon Ratio and the Choice of Glucose-Lowering Drugs

Sanjay Kalra · Yashdeep Gupta

Received: January 5, 2016 / Published online: March 10, 2016
© The Author(s) 2016. This article is published with open access at Springerlink.com

ABSTRACT

The influence of alpha and beta cells, through glucagon and insulin, on energy metabolism is well known. The insulin:glucagon ratio (IGR) is a frequently discussed entity in the medical literature. However, in recent years, focus has shifted to other pathways and markers of health and disease. This communication revisits the insulin:glucagon bipolar axis and describes the significance of the IGR. It reviews the effects of various glucose-lowering drugs on this ratio, and hypothesizes that the ratio can be used to predict the appropriate choice of drugs for managing diabetes. Drugs which increase the IGR may be beneficial in insulinopenic conditions, while those which decrease IGR may be of help in the setting of hyperinsulinemia or insulin resistance.

Enhanced content To view enhanced content for this article go to www.medengine.com/Redeem/3944F060732A6B8D.

S. Kalra (✉)
Department of Endocrinology, Bharti Hospital,
Karnal, India
e-mail: brideknl@gmail.com; brideknl-@gmail.com

Y. Gupta
Department of Endocrinology, AIIMS, New Delhi,
India

Keywords: Antidiabetic drugs; Glucagon; Insulin; Insulin:glucagon ratio; Therapy choice

THE INSULIN:GLUCAGON BIPOLAR AXIS

Glucagon and insulin are two opposing hormones which work in tandem to maintain a normal fuel balance. By modulating the relative concentrations of glucagon and insulin, the alpha and beta cells of the pancreas control endogenous glucose production, triacylglycerol deposition, and protein synthesis [1]. In simple terms, glucagon acts as a catabolic hormone, while insulin exerts anabolic effects on the body. A low insulin:glucagon ratio (IGR) stimulates mobilization of stored nutrients, increases glycogenolysis and gluconeogenesis, and promotes the breakdown of adipose tissue into free fatty acids and glycerol. A high IGR encourages biosynthesis of proteins, inhibits the production of glucose, and reduces free fatty acid release [2]. The effects of insulin and glucagon on various physiological processes are listed in Table 1.

Table 1 Biochemical effects of insulin and glucagon

Process	Insulin	Glucagon
Glycogenesis	Increased (glycogen synthase activation)	Inhibited (glycogen synthase deactivation)
Gluconeogenesis	Inhibited (inhibition of pyruvate carboxylase, phosphoenolpyruvate carboxykinase, glucose 6 phosphatase)	Increased (inhibition of pyruvate kinase, phosphofructokinase; activation of fructose 1,6-bisphosphatase)
Glycolysis	Increased (activation of glucokinase, phosphofructokinase, pyruvate kinase)	
Glycogenolysis	Inhibited (inhibition of glycogen phosphorylase)	Increased (activation of glycogen phosphorylase)
Ketogenesis	Inhibited (less substrate acetyl coenzyme A (CoA); inhibition of 3-hydroxy-3-methylglutaryl-coenzyme A (HMG CoA) synthetase)	
Lipogenesis	Increased (more substrate glycerol 3 phosphate and nicotinamide adenine dinucleotide phosphate (NADPH); activation of acetyl CoA carboxylase)	
Protein synthesis	Decreased protein degradation	Increased amino acid uptake by liver; decreased amino acids in plasma

This aspect of physiology is not, however, highlighted adequately in diabetology circles and texts. Though DeFronzo [3] included the alpha cell as part of the ominous octet, and Schwartz et al. [4] list it as one of the egregious eleven, less attention has been paid to this pathophysiological player. Some reasons for this may be the multitude of factors which affect insulin and glucagon secretion, their highly variable concentrations, the existence of a significant, and variable, porto-systemic gradient, and the difficulty involved in estimating them. In spite of extensive research, there is still controversy surrounding the absolute concentrations of insulin and glucagon in persons with diabetes. Yet another explanation may be the current interest in nonpancreatic pathophysiological pathways of diabetes. This article does not contain any new studies with human or animal subjects performed by any of the authors.

INSULIN

Ambient insulin concentrations present a spectrum of values, ranging from absolute deficiency in type 1 diabetes to hyperinsulinemia in those with type 2 diabetes. Beta-cell secretory capacity is known to fall with increasing duration of diabetes, so insulin levels may vary depending upon diabetes stage. Insulin levels are also reported to be high in obese persons, irrespective of glycemic status. This is a reflection of the insulin resistance that occurs with obesity. Insulin levels vary with meals, with specific nutrients, and with comorbid conditions. Insulinogenic stimuli, for example, include glucose as well as certain amino acids (Table 2). Alterations in growth hormone and thyroid hormone secretion also affect insulin secretion. In man, a first phase and a second

Table 2 Physiological determinants of insulin and glucagon secretion

	Insulin	Glucagon
Stimulants	Glucose; amino acids—arginine, leucine; gastrointestinal hormones—secretin, gastrin, pancreaticozym	Hypoglycemia, amino acids, low adrenaline
Inhibitors	Adrenaline	Hyperglycemia

phase of insulin can easily be identified, along with distinct basal and prandial secretion [5]. In some primates, insulin is released in oscillations which occur at intervals of roughly 9 min [6]. This phenomenon is not seen in humans.

GLUCAGON

Glucagon secretion varies from the fasting to the postprandial state according to the composition of the meal taken, and is influenced by body weight as well as comorbid illnesses [7, 8]. There are varying opinions about the concentration of glucagon in obesity and—just like diabetes—current research suggests that glucagon, or glucagon response to arginine, increases in obesity. Glucagon levels are higher in diabetes, increase on stimulation with arginine, and are not suppressed after glucose administration. Workers also suggest a resistance to glucagon in obese persons with diabetes [9].

The issue is also complicated by issues associated with the assessment of insulin and glucagon. Various researchers estimate a variety of species, such as C-peptide, insulin, pancreatic glucagon, enteroglucagon, and immunoreactive glucagon. Altered tissue response to insulin and glucagon is also reported in obesity, raising the question of whether the determination of absolute glucagon values is worth the effort. Yet another controversy relates to the systemic and portal concentrations of these antipodal hormones. As the portosystemic gradients of insulin and glucagon are dissimilar, the

relevance of testing peripheral IGR is also doubtful [10, 11].

INSULIN:GLUCAGON RATIO

The insulin:glucagon bipolar axis serves to regulate carbohydrate and lipid metabolism. The therapeutic value of glucagon in diabetes management is gradually becoming clear [12]. As these hormones work in tandem with each other, albeit in opposing directions, it may make sense to interpret the IGR instead of assessing absolute values. This concept was popularized by Unger [1], who proposed the bihormonal hypothesis. The IGR has been found to vary with the presence of, or the need for, anabolism and catabolism. The IGR varies inversely with the need for endogenous glucose production. It is lowest in total starvation and highest during loading with exogenous carbohydrate. The molecular mechanisms which control this energy homeostasis are listed in Table 1. In brief, the IGR acts as a physiological fulcrum, balancing two opposite ends of the metabolic spectrum to provide energy when needed and conserve it if possible.

GLUCOSE-LOWERING DRUGS

Recent years have witnessed the development of various classes of glucose-lowering drugs. These molecules are studied for their effect on a wide range of glucotropic hormones and metabolites, apart from their direct effect on

glucose lowering. The effect of various glucose-lowering drugs on insulin levels is well known, but their effect on glucagon levels, and specifically on the IGR, is rarely discussed. This is unfortunate, as detailed insight into this aspect of pharmacodynamics may aid the selection of the appropriate therapy for each patient based upon their insulin-glucagon homeostasis. Drugs which increase the IGR may be beneficial in insulinopenic conditions, while those which decrease the IGR may be of help in the setting of hyperinsulinemia or insulin resistance.

Recent studies have shown that metformin therapy does not lead to any change in glucagon secretion [12]. However, the insulin-sensitizing effect of metformin causes a reduction in hyperinsulinemia, and thus reduces the IGR. Similar effects are reported with pioglitazone [13].

Sulfonylureas such as glimepiride are insulinotropic agents which increase insulin secretion but have no impact whatsoever on alpha cell function or glucagon secretion. Thus, glimepiride certainly increases the IGR. In another study, glimepiride was found to increase both prandial insulin as well as prandial glucagon [area under curve (AUC), 0–2 h] [14]. The opposing effects of insulin sensitizers and insulin secretagogues are visible at the molecular level as well. While metformin activates carnitine palmitoyltransferase 1 (CPT1), glibenclamide inhibits this enzyme [15, 16]. This molecular difference may explain the effects of these drugs on weight: metformin promotes β -oxidation of free fatty acids, while glibenclamide facilitates triacylglycerol production.

The incretin-based therapies are, in general, thought to reduce plasma glucagon levels. The evidence reveals variable effects. Recently, liraglutide, a glucagon-like peptide-1 receptor agonist (GLP1RA), has been shown to increase

post-challenge glucagon levels after 12 weeks of therapy, though no such effects were noted after shorter periods of treatment. This effect persisted for 48 weeks (the duration of the trial), but was not noted in the fasting state [17]. Tachyphylaxis has been suggested as a possible mechanism to explain this phenomenon [18]. Other studies have demonstrated no effect of liraglutide on fasting glucagon, but have noted its ability to reduce 24-h glucagon levels (by reducing glucagon secretion after a protein-rich meal) [19]. The authors reported no change in the IGR with liraglutide therapy. However, in a Japanese study, liraglutide increased the IGR. While the drug reduced the incremental release of glucagon between 15 and 60 min after a meal, it did not change overall glucagon secretion. Coupled with an increase in insulin secretion, this led to an increase in the IGR with liraglutide [20].

A 28-day-long pharmacodynamics study compared the effect of lixisenatide and liraglutide on plasma insulin and glucagon concentrations, but did not statistically analyze the IGR [21]. Lixisenatide provided a significantly greater decrease in postprandial glucagon as compared to liraglutide. Postprandial insulin and C-peptide levels were also significantly reduced with lixisenatide versus liraglutide [21], while decreases in proinsulin were comparable between groups. The trend was for a higher IGR with liraglutide and a lower one with lixisenatide. In individuals without diabetes, exenatide increased the IGR at rest, but this trend reversed when exercise was performed [22]. A lowering of plasma glucagon was noted in individuals with type 2 diabetes with the same drug [23]. Dulaglutide, a once-weekly GLP1RA reduces fasting glucagon concentrations while increasing homeostatic model assessment 2 beta (HOMA2B). This implies that dulaglutide administration increases the IGR [12].

Table 3 Effects of antidiabetic drugs on the insulin:glucagon bipolar axis

Antidiabetic drugs	Insulin	Glucagon	IGR
Glimepiride	↑	No change	↑
DPP4i	↑	↓	↑
Liraglutide	↑	↓/No change	No change/ ↑
Dulaglutide	↑	↓	↑
Exenatide	↑	↓	↑
Lixisenatide	↓	↓	↓
Dapagliflozin	↑	↑	↓
Metformin	↓	No change	↓
Pioglitazone	↓	–	↓
Acarbose	↓	No change	↓

Based upon [11–14, 16, 18–22, 24–27]

↓ = reduction; ↑ = increase

DPP4i dipeptidyl peptidase-4 inhibitor, IGR insulin:glucagon ratio

Relative to the dipeptidyl peptidase inhibitor sitagliptin, exenatide has been shown to increase the IGR [24]. However, in absolute terms, sitagliptin also increases insulin secretion and reduces glucagon concentrations, thus causing a rise in the IGR. Vildagliptin, another dipeptidyl peptidase inhibitor, decreases prandial glucagon AUC (0–2 h). This effect has been reported to persist for at least 2 years, and reflects the improved alpha cell sensitivity caused by vildagliptin. Vildagliptin also increases insulin secretion, though not to the same extent as glimepiride. In total, vildagliptin increases the IGR [13]. Similar effects are noted with linagliptin and saxagliptin [25, 26].

Empagliflozin, a sodium glucose co-transporter-2 (SGLT2) inhibitor, has recently been shown to reduce the IGR. This effect is pronounced after acute administration

of the drug, but persists—to an attenuated degree—after 28-day exposure. This reduction in the IGR reflects an increased need for—and an actual increase in—endogenous glucose production, to balance the increase in glucosuria. Similar results have been reported with dapagliflozin (Table 3) [27, 28]. The possible reasons for this phenomenon have been hypothesized in detail [29].

No significant differences in serum insulin and plasma glucagon levels were documented after 1 week of colestimide administration [30]. Acarbose, an alpha-glucosidase inhibitor, reduces serum insulin levels without influencing glucagon secretion, and thus reduces the IGR [31].

IMPACT ON WEIGHT GAIN AND HYPOGLYCEMIA

Weight gain and hypoglycemia are common but unwanted corollaries of conventional glucose-lowering therapy. However, newer classes of drugs offer the advantage of weight loss along with glycemic control while also avoiding hypoglycemia. In general, drugs which increase the IGR will be associated with an anabolic effect or tendency for weight gain and an increased risk of hypoglycemia. On the other hand, drugs which reduce the IGR will facilitate weight loss and prevent hypoglycemia. Our IGR-based construct is concordant with the clinical pharmacology of directly acting insulin secretagogues such as sulfonylureas (which increase the IGR, may cause weight gain, and are associated with hypoglycemia) and novel drugs like GLP1RA and SGLT2 inhibitors (which reduce the IGR, aid weight loss, and are associated with a minimal risk of hypoglycemia).

THERAPEUTIC IMPLICATIONS

In spite of the increasing number of molecules available for diabetes management, glycemic control remains suboptimal. A large number of people with diabetes continue to be classified as “refractory” or unresponsive to treatment. Even in the best randomized controlled trials, a significant proportion of subjects remain nonresponders to allocated therapy. There is also a dearth of studies that are able to differentiate responders and nonresponders based upon their demographic characteristics, physical attributes, or biochemical phenotype. Thus, there is still an unmet need to explore and identify biochemical markers which may serve as predictors of response to chosen therapeutic interventions.

The IGR, therefore, may be used to create a matrix for choosing an antidiabetic therapy. Though data are limited and may be conflicting, one can create a model, based upon absolute and relative values of insulin and glucagon, to facilitate the selection of the appropriate therapy (Fig. 1). Persons with high insulin levels (hyperinsulinemia, secondary to insulin resistance) will benefit from drugs which lower insulin secretion (insulin sensitizers, SGLT2 inhibitors, alpha-glucosidase inhibitors). Within this group of patients, those with low

glucagon values may respond better to SGLT2 inhibitors. Incretin-based therapy has been shown to be more effective in persons with high glucagon levels [32]. As these modes of treatment cause hyperinsulinemia, incretin-based therapy may prove more suitable for persons with a normal IGR. Glimepiride may be a better choice for persons with low insulin and a low IGR. The effects of metformin and SGLT2 inhibitors on the IGR are similar to those seen with calorie restriction. Thus, clinical situations requiring calorie restriction mimicry (e.g., obese diabetes; need for geroprotection) [33] may benefit from these drug classes.

Rational combinations may also be created using knowledge of the effects of drugs on insulin and glucagon. For example, it may be beneficial to combine incretin-based therapy, which reduces glucagon concentrations, with SGLT2 inhibitors, which are known to increase glucagon.

It is not our suggestion that IGR estimation becomes an integral part of routine diabetes care; it is, and will remain, a research tool. However, clinical endocrinologists should understand this physiological concept and be able to translate it into an appropriate choice of pharmacotherapy. Preclinical and clinical development programs for modern drugs

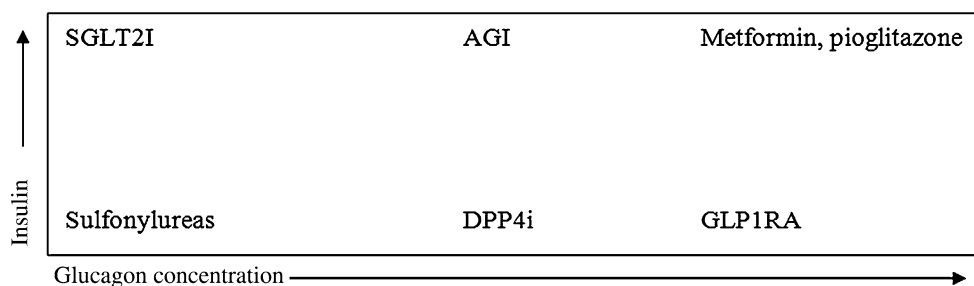


Fig. 1 Choice of glucose-lowering therapy based upon the IGR. *X*-axis: IGR; *y*-axis: insulin concentration. *AGI* alpha-glucosidase inhibitors, *DPP4i* dipeptidyl peptidase-4

inhibitor, *GLP1RA* glucagon-like peptide-1 receptor agonist, *IGR* insulin:glucagon ratio, *SGLT2i* sodium glucose transporter-2 inhibitor

should include detailed study of their effects on the insulin:glucagon axis: this will help to unravel new facets of human biochemistry and physiology.

ACKNOWLEDGMENTS

No funding was received for the publication of this article. All named authors meet the International Committee of Medical Journal Editors (ICMJE) criteria for authorship for this manuscript, take responsibility for the integrity of the work as a whole, and have given final approval for the version to be published.

Disclosures. Sanjay Kalra has received honoraria from AstraZeneca, Boehringer Ingelheim, Eli Lilly, Novo Nordisk, and Sanofi. Sanjay Kalra and Yashdeep Gupta have nothing to disclose with regards to the publication of this article.

Compliance with Ethics Guidelines. This article does not contain any new studies with human or animal subjects performed by any of the authors.

Open Access. This article is distributed under the terms of the Creative Commons Attribution-NonCommercial 4.0 International License (<http://creativecommons.org/licenses/by-nc/4.0/>), which permits any noncommercial use, distribution, and reproduction in any medium, provided you give appropriate credit to the original author(s) and the source, provide a link to the Creative Commons license, and indicate if changes were made.

REFERENCES

1. Unger RH. Glucoregulatory hormones in health and disease. A teleologic model. *Diabetes*. 1966;15:500–6.
2. Unger RH. Glucagon and the insulin: glucagon ratio in diabetes and other catabolic illnesses. *Diabetes*. 1971;20:834–8.
3. DeFronzo RA. Banting Lecture. From the triumvirate to the ominous octet: a new paradigm for the treatment of type 2 diabetes mellitus. *Diabetes*. 2009;58:773–95.
4. Schwartz SS, Epstein S, Corkey BE, Grant SF, Gavin JR 3rd, Aguilar RB. The time is right for a new classification system for diabetes: rationale and implications of the β -cell-centric classification schema. *Diabetes Care*. 2016;39(2):179–86. doi:10.2337/dc15-1585.
5. Rorsman P, Braun M. Regulation of insulin secretion in human pancreatic islets. *Annu Rev Physiol*. 2013;75:155–79.
6. Koerker DJ, Goodner CJ, Hansen BW, Brown AC, Rubenstein AH. Synchronous, sustained oscillation of C-peptide and insulin in the plasma of fasting monkeys. *Endocrinology*. 1978;102(5):1649–52.
7. Wilmore DW, Lindsey CA, Moyland JA, Faloona GR, Pruitt BA, Unger RH. Hyperglucagonaemia after burns. *Lancet*. 1974;1:73–5.
8. Kuhl C, Holst JJ. Plasma glucagon and the insulin: glucagon ratio in gestational diabetes. *Diabetes*. 1976;25:16–23.
9. Tsuchiyama N, Takamura T, Ando H, Sakurai M, Shimizu A, Kato K, et al. Possible role of alpha cell insulin resistance in exaggerated glucagon responses to arginine in type 2 diabetes. *Diabetes Care*. 2007;30:2583–7.
10. Parrilla R, Goodman MN, Toews CJ. Effect of glucagon: insulin ratios on hepatic metabolism. *Diabetes*. 1974;23:725–31.
11. Haidar A, Legault L, Messier V, Mitre TM, Leroux C, Rabasa-Lhoret R. Comparison of dual-hormone artificial pancreas, single-hormone artificial pancreas, and conventional insulin pump therapy for glycaemic control in patients with type 1

- diabetes: an open-label randomised controlled crossover trial. *Lancet Diabetes Endocrinol*. 2015;3:17–26.
12. Umpierrez G, Povedano ST, Manghi FP, Shurzinske L, Pechtner V. Efficacy and safety of dulaglutide monotherapy versus metformin in type 2 diabetes in a randomized controlled trial (AWARD-3). *Diabetes Care*. 2014;37(8):2168–76.
 13. Gastaldelli A, Casolaro A, Pettiti M, Nannipieri M, Ciociaro D, Frascerra S, et al. Effect of pioglitazone on the metabolic and hormonal response to a mixed meal in type II diabetes. *Clin Pharmacol Ther*. 2007;81(2):205–12.
 14. Ahrén B, Foley JE, Ferrannini E, Matthews DR, Zinman B, Dejager S, et al. Changes in prandial glucagon levels after a 2-year treatment with vildagliptin or glimepiride in patients with type 2 diabetes inadequately controlled with metformin monotherapy. *Diabetes Care*. 2010;33(4):730–2.
 15. Lehtihet M, Welsh N, Berggren PO, Cook GA, Sjöholm A. Glibenclamide inhibits islet carnitine palmitoyltransferase 1 activity, leading to PKC dependent insulin exocytosis. *Am J Physiol Endocrinol Metab*. 2003;285:E438–46.
 16. Zhou G, Myers R, Li Y, Chen Y, Shen X, Fenyk Melody J, et al. Role of AMP activated protein kinase in mechanism of metformin action. *J Clin Invest*. 2001;108:1167–74.
 17. Kramer CK, Zinman B, Choi H, Connelly PW, Retnakaran R. The impact of chronic liraglutide therapy on glucagon secretion in type 2 diabetes: insight from the LIBRA trial. *J Clin Endocrinol Metab*. 2015;100(10):3702–9.
 18. Kalra S, Gupta Y. Letter to the Editor: Comment on “The Impact of Chronic Liraglutide Therapy on Glucagon Secretion in Type 2 Diabetes: Insight From the LIBRA Trial” by Kramer CK, et al. *J Clin Endocrinol Metab*. 2015;100(11):116.
 19. Madsbad S, Schmitz O, Ranstam J, Jakobsen G, Matthews DR. Improved glycemic control with no weight increase in patients with type 2 diabetes after once-daily treatment with the long-acting glucagon-like peptide 1 analog liraglutide (NN2211) a 12-week, double-blind, randomized, controlled trial. *Diabetes Care*. 2004;27(6):1335–42.
 20. Matsumoto S, Yamazaki M, Kadono M, Iwase H, Kobayashi K, Okada H, et al. Effects of liraglutide on postprandial insulin and glucagon responses in Japanese patients with type 2 diabetes. *J Clin Biochem Nutr*. 2013;53(1):68–72.
 21. Kapitza C, Forst T, Coester H-V, Poitiers F, Ruus P, Hincelin-Méry A. Pharmacodynamic characteristics of lixisenatide once daily versus liraglutide once daily in patients with type 2 diabetes insufficiently controlled on metformin. *Diabetes Obes Metab*. 2013;15:642–9.
 22. Khoo EYH, Wallis J, Tsintzas K, Macdonald IA, Mansell P. Effects of exenatide on circulating glucose, insulin, glucagon, cortisol and catecholamines in healthy volunteers during exercise. *Diabetologia* 2009;53:139–43.
 23. Kolterman OG, Kim DD, Shen L, Ruggles JA, Nielsen LL, Fineman MS, et al. Pharmacokinetics, pharmacodynamics, and safety of exenatide in patients with type 2 diabetes mellitus. *Am J Health Syst Pharm*. 2005;62(2):173–81.
 24. DeFronzo RA, Okerson T, Viswanathan P, Guan X, Holcombe JH, MacConnell L. Effects of exenatide versus sitagliptin on postprandial glucose, insulin and glucagon secretion, gastric emptying, and caloric intake: a randomized, cross-over study. *Curr Med Res Opin* 2008;24(10):2943–52.
 25. Rauch T, Graefe-Mody U, Deacon CF, Ring A, Holst JJ, Woerle HJ, et al. Linagliptin increases incretin levels, lowers glucagon, and improves glycemic control in type 2 diabetes mellitus. *Diabetes Ther*. 2012;3(1):1–14.
 26. Sharma MD. Role of saxagliptin as monotherapy or adjunct therapy in the treatment of type 2 diabetes. *Ther Clin Risk Manag*. 2010;6:233.
 27. Ferrannini E, Muscelli E, Frascerra S, Baldi S, Mari A, Heise T, et al. Metabolic response to sodium glucose cotransporter 2 inhibition in type 2 diabetic patients. *J Clin Invest*. 2014;124:499–508.
 28. Merovci A, Solis Herrera C, Daniele G, Eldor R, Fiorentino TV, Tripathy D, et al. Dapagliflozin improves muscle insulin sensitivity but enhances endogenous glucose production. *J Clin Invest*. 2014;124:509–14.
 29. Kalra S, Gupta Y, Patil S. Sodium-glucose cotransporter-2 inhibition and the insulin: glucagon ratio: unexplored dimensions. *Indian J Endocr Metab*. 2015;19:426–9.
 30. Suzuki T, Oba K, Igari Y, Matsumura N, Watanabe K, Futami-Suda S, et al. Colestimide lowers plasma glucose levels and increases plasma glucagon-like PEPTIDE-1 (7–36) levels in patients with type 2 diabetes mellitus complicated by hypercholesterolemia. *J Nippon Med Sch*. 2007;74(5):338.

-
31. Dimitriadis G, Tessari P, Gerich J. Effects of the disaccharidase inhibitor acarbose on meal and intravenous glucose tolerance in normal man. *Metabolism*. 1982;31(8):841–3.
 32. Kalra S, Kalra B, Sahay R, Agrawal N. Predicting response to incretin-based therapy. *Res Rep Endocr Disord*. 2011;1:11–9.
 33. Kalra S, Unnikrishnan AG, Sahay R. Metformin and the promise of geroprotection. *Indian J Endocrinol Metab*. 2012;16:496.