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ORIGINAL RESEARCH

## Lispro administered by the QS-M Needle-Free Jet Injector generates an earlier insulin exposure

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### ABSTRACT

**Objective:** The aim of this study is to evaluate the pharmacokinetic and pharmacodynamic (PK-PD) profiles of lispro administered by the QS-M needle-free jet injector in Chinese subjects.

**Research design and methods:** A randomized, double-blind, double-dummy, cross-over study was performed. Eighteen healthy volunteers were recruited. Lispro (0.2 units/kg) was administered by the QS-M needle-free jet injector or by conventional pen. Seven-hour euglycemic clamp tests were performed.

**Results:** A larger area under the curve (AUCs) of insulin concentration and glucose infusion rate (GIR) during the first 20 minutes after lispro injection by the jet injector compared to the insulin pen was observed ( $24.91 \pm 15.25$  vs.  $12.52 \pm 7.60$  mg. kg<sup>-1</sup>,  $P < 0.001$  for  $AUC_{GIR,0-20 \text{ min}}$ ;  $0.36 \pm 0.24$  vs.  $0.10 \pm 0.04$  U min L<sup>-1</sup>,  $P < 0.001$  for  $AUC_{INS, 0-20 \text{ min}}$ ). Needle-free injection showed a shorter time to reach maximum insulin concentration ( $37.78 \pm 11.14$  vs.  $80.56 \pm 37.18$  min,  $P < 0.001$ ) and GIR ( $73.24 \pm 29.89$  vs.  $116.18 \pm 51.89$  min,  $P = 0.006$ ). There were no differences in total insulin exposure and hypoglycemic effects between the two devices.

**Conclusion:** Lispro administered by QS-M needle-free injector results in earlier and higher insulin exposure than conventional pen, and a greater early glucose-lowering effect with similar overall potency.

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## 1. Introduction

Poor absorption of insulin is an important factor in patients with diabetes [1]. Rapid-acting insulin analogs more closely approach the physiological insulin response, compared with regular human insulin [2]. Nevertheless, rapid-acting insulin analogs administered by conventional pen could not be absorbed quickly enough to replicate the physiological insulin secretion profile [3].

Needle-free jet injector delivers insulin at a high velocity into subcutaneous tissue and dispenses insulin over a larger area than a syringe [4]. Previous studies revealed that insulin administered by jet injectors is absorbed faster compared to conventional insulin pens [5–9]. A randomized, double-blind, double-dummy, crossover study conducted by Engwerda et al. [3] showed that compared to the conventional pen, enhanced insulin absorption and reduced duration of glucose-lowering action were found when aspart was administered by the jet injector. Whether the differences in the pharmacokinetic and pharmacodynamic (PK-PD) profiles can be extrapolated to other rapid-acting insulin analogs needs to be further investigated.

QS-M needle-free jet injector (Beijing QS Medical Technology Co., Ltd., China) was approved for insulin injection by the China Food and Drug Administration in 2012. Apart

from insulin aspart, lispro is a rapid-acting insulin analog often used in clinical practice [10,11]; this study aimed at comparing PK-PD profiles of lispro administered by the QS-M needle-free injector with an insulin pen in a group of Chinese subjects.

## 2. Methods

### 2.1. Study population

Eighteen volunteers (nine men and nine women) were recruited in this study. The inclusion criteria were: nonsmokers aged 18–40 years, with body mass index (BMI) of 17–24 kg/m<sup>2</sup>; subjects with normal biochemical tests, blood pressure, and electrocardiograph; subjects who signed the informed consent. The exclusion criteria were: subjects with insulin allergy or other allergic history; subjects with chronic diseases such as diabetes, cardiovascular diseases, liver or kidney disease. Subjects who used alcohol were also excluded. The study was approved by the Ethics Committee of the First Affiliated Hospital of Chongqing Medical University.

### 2.2. Study design and PK-PD sampling

This trial (NFJI-2015) is registered at ClinicalTrials.gov (ID number: NCT02443714). Double-blind, double-dummy,

randomized crossover study design was used. In phase I, nine subjects were administered with lispro (Humalog, Eli Lilly and Company, US) 0.2 IU/kg by the QS-M needle-free jet injector (Beijing QS Medical Technology Co., Ltd., China) and comparable volume of placebo solution (placebo solution consisted of normal saline, which was injected into an empty insulin vial under aseptic conditions) by an insulin pen (HumaPen Ergoll, Eli Lilly and Company, US) in the abdomen simultaneously; the remaining nine subjects were administered with lispro 0.2 IU/kg by a conventional pen and a comparable volume of placebo solution by a needle-free jet injector in the abdomen simultaneously. Seven-hour euglycemic glucose clamp tests were then performed by an experienced nurse [12–14]. Two catheters were inserted intravenously. One catheter was inserted for blood sampling, and the hand was kept in a heated box at 60°C to arterialize venous blood [15]. The other catheter was placed in an antecubital vein of the contralateral arm for administration of 20% glucose. The glucose infusion rate (GIR) was determined by plasma glucose measurements at 5-min intervals during the first 4 h and at 10-min intervals thereafter. Venous blood was sampled for detecting plasma insulin every 10 min during the first hour and every 30 min during the later 6 hours. To maintain the euglycemic level (5.0 mmol/L), GIR was regulated according to levels of plasma glucose. There was a one-week washout period between phase I and phase II of the study. In phase II, lispro and comparable volume of placebo solution were administered by jet injector or insulin pen crosswise, and other procedures were similar to phase I.

### 2.3. Assay methodology

Plasma glucose was measured by the glucose oxidase method using the Biosen 5030 Glucose Analyzer (EKF Industrie, Elektronik GmbH, Barleben, Germany). Plasma insulin was quantified using a validated lispro chemiluminescence assay [Beckman Coulter's Dxl 800 Immunoassay System, Beckman Coulter, Inc. 250 S. Kraemer Blvd. Brea, CA 92821 U.S.A.].

### 2.4. Statistical analysis

Early insulin exposure was defined as the area under the curve (AUC) of insulin and GIR from 0 to 20 min ( $AUC_{INS,0-20 \text{ min}}$  and  $AUC_{GIR,0-20 \text{ min}}$ ) [16–18]. The primary study end point was  $AUC_{GIR,0-20 \text{ min}}$ . Secondary PK–PD end points included the following:

AUC of insulin at various time intervals ( $AUC_{INS,0-20 \text{ min}}$ ,  $AUC_{INS,0-120 \text{ min}}$ , and  $AUC_{INS,0-420 \text{ min}}$ ), the time to peak plasma insulin concentrations (INS- $T_{max}$ ), the maximal insulin levels (INS-C $_{max}$ ), and 50% insulin absorption time (INS- $T_{AUC50\%}$ ); AUC of GIR at various time intervals ( $AUC_{GIR,0-120 \text{ min}}$  and  $AUC_{GIR,0-420 \text{ min}}$ ), the time to maximum GIR (GIR- $T_{max}$ ), the maximum GIR levels (GIR-C $_{max}$ ), and 50% glucose clearance time (GIR- $T_{AUC50\%}$ ).

Power Analysis and Sample Size software 11 (PASS 11; NCSS, LLC, Kaysville, UT, USA) was used to calculate the sample size. In the procedure of inequality tests for two means in a 2 × 2 crossover design using differences, a two-sided *t*-test achieved 0.84 power to infer that the mean difference was not

0 when the total sample size of a 2 × 2 crossover design was 18; the actual mean difference was 15; the standard deviation of the period differences for each subject within each sequence was 10; and the significance level was 0.05. SAS software (Version 8) was used to generate the random allocation sequence. All statistical analyses were performed using SPSS 13.0 (Chicago, IL, USA). Normally distributed variables were presented as the mean ± SD, whereas variables with a skewed distribution were analyzed after logarithmic transformation, and the results were presented as median values with the interquartile range (IQR) in parentheses. The PK–PD parameters were tested by paired *t*-tests. Pearson correlation analysis was used to test the correlations between individual variables. *P*-value < 0.05 was considered statistically significant.

## 3. Results

### 3.1. Subject disposition

The average age of the subjects was 25.76 ± 1.86 years, and the mean BMI was 21.21 ± 2.12 kg/m<sup>2</sup>. Average glycemic levels in subjects who were administered lispro using needle-free injector and insulin pen during the clamp trial were 5.13 ± 0.42 vs. 5.09 ± 0.46 mmol/L, respectively, *P* = 0.523. There was no significant difference in the average plasma glucose maintained during the glucose clamp procedure for the needle-free injector or insulin pen dosing portion of the study. All 18 subjects completed both injections and 7-hour euglycemic glucose clamp tests.

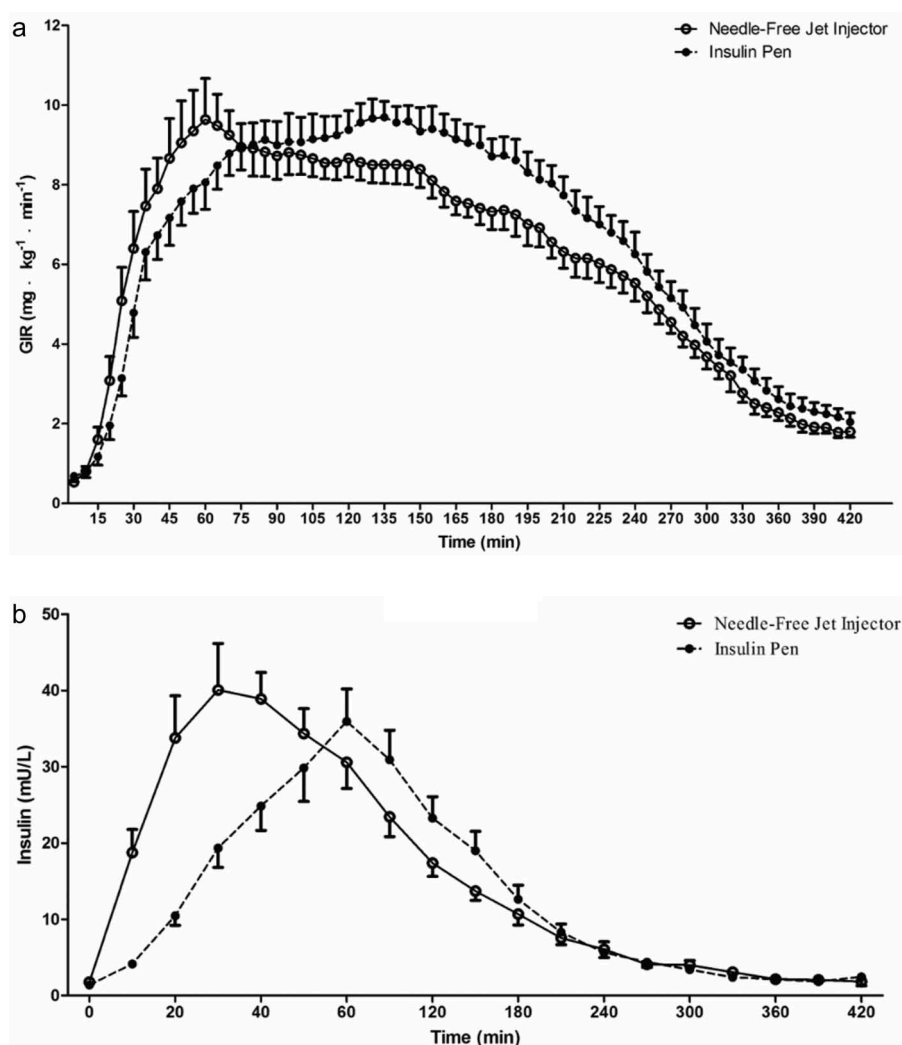
### 3.2. Pharmacodynamics

Compared to the lispro injected by conventional insulin pen, a significantly higher 20-min GIR was observed (3.71 ± 2.38 vs. 1.33 ± 0.87 mg·kg<sup>-1</sup>·min<sup>-1</sup>, *P* < 0.001), and the GIR–time curves were shifted to the left when lispro was injected by the jet injector (Figure 1(a)). A mean difference of about 40 min was observed in GIR- $T_{max}$  when lispro was administered using the jet injector compared to the conventional insulin pen (73.24 ± 29.89 vs. 116.18 ± 51.89 min, *P* = 0.006).

The faster and higher exposure of lispro in the early period translated into a greater glucose-lowering effect, illustrated as a larger AUC of GIR for the first 20 min in the subjects where lispro was injected using the jet injector ( $AUC_{GIR,0-20 \text{ min}}$  24.91 ± 15.25 vs. 12.52 ± 7.60 mg·kg<sup>-1</sup>, *P* < 0.001). GIR- $T_{AUC50\%}$  was also significantly shortened when lispro was administered by a jet injector (153.89 ± 21.60 vs. 170.00 ± 19.02 min, *P* = 0.023). The total glucose-lowering effect ( $AUC_{GIR,0-420 \text{ min}}$ ) and GIR-C $_{max}$  were similar between the two groups (Table 1).

### 3.3. Pharmacokinetics

Compared to lispro injected by the conventional insulin pen, a significantly higher 20-min insulin concentration was seen (33.79 ± 23.34 vs. 10.47 ± 5.35 mU/l, *P* < 0.001), and the concentration–time curves were shifted to the left when lispro was injected by the jet injector (Figure 1(b)); furthermore, AUC of insulin concentration for the first 20 min ( $AUC_{INS,0-20 \text{ min}}$ )



**Figure 1.** PK-PD curves of needle free jet injector and insulin pen. Glucose infusion rate–time (a) and plasma insulin concentration–time (b) profiles of lispro administered by the needle-free jet injector (○) and the insulin pen (●) during the euglycemic glucose clamp.

was significantly larger when lispro was administered by the jet injector ( $0.36 \pm 0.24$  vs.  $0.10 \pm 0.04$  U. min.  $L^{-1}$ ,  $P < 0.001$ ), and a significantly reduced  $INS-T_{max}$  and  $INS-T_{AUC50\%}$  were also observed, indicating a faster lispro exposure when injected by jet injector. Total insulin exposure ( $AUC_{INS,0-420 \text{ min}}$ ) and  $INS-C_{max}$  were similar between the two devices (Table 1).

#### 4. Discussion

This study demonstrates that lispro administered by the QS-M needle-free jet injector was more rapidly absorbed, leading to an earlier lispro exposure and therefore an earlier glucose-lowering effect, when compared with the conventional insulin pen in Chinese subjects.

Needle-free jet injectors display a cone-like dispersion pattern in the subcutaneous tissue with a relatively large surface area [4]. Rapid-acting insulin analogs administered via jet injectors are predicted to result in faster absorption and

**Table 1.** Pharmacokinetic and pharmacodynamic parameters for lispro administered by the needle-free jet injector and the insulin pen.

	Jet injector	Insulin pen	P-value
<b>Pharmacodynamic parameters</b>			
GIR- $T_{max}$ (min)	$73.24 \pm 29.89$	$116.18 \pm 51.89$	0.006
GIR- $C_{max}$ ( $mg \cdot kg^{-1} \cdot min^{-1}$ )	$11.85 \pm 3.39$	$12.13 \pm 1.97$	0.768
GIR- $T_{AUC50\%}$ (min)	$153.89 \pm 21.60$	$170.00 \pm 19.02$	0.023
$AUC_{GIR,0-20 \text{ min}}$ ( $mg \cdot kg^{-1}$ )	$24.91 \pm 15.25$	$12.52 \pm 7.60$	<0.001
$AUC_{GIR,0-120 \text{ min}}$ ( $mg \cdot kg^{-1}$ )	$886.00 \pm 216.27$	$749.14 \pm 202.07$	0.047
$AUC_{GIR,0-420 \text{ min}}$ ( $mg \cdot kg^{-1}$ )	$2393.28 \pm 518.58$	$2370.11 \pm 406.32$	0.703
<b>Pharmacokinetic Parameters</b>			
$INS-T_{max}$ (min)	$37.78 \pm 11.14$	$80.56 \pm 37.18$	<0.001
$INS-C_{max}$ ( $mU \cdot L^{-1}$ )	$48.29 \pm 23.53$	$43.06 \pm 16.54$	0.446
$INS-T_{AUC50\%}$ (min)	$81.67 \pm 20.07$	$115.00 \pm 18.66$	<0.001
$AUC_{INS,0-20 \text{ min}}$ ( $U \cdot min \cdot L^{-1}$ )	$0.36 \pm 0.24$	$0.10 \pm 0.04$	<0.001
$AUC_{INS,0-120 \text{ min}}$ ( $U \cdot min \cdot L^{-1}$ )	$3.24 \pm 1.38$	$2.87 \pm 1.25$	0.415
$AUC_{INS,0-420 \text{ min}}$ ( $U \cdot min \cdot L^{-1}$ )	$5.06 \pm 1.98$	$5.01 \pm 1.58$	0.939

GIR: glucose infusion rate; AUC: area under the curve; GIR- $T_{max}$ : time to maximum GIR; GIR- $C_{max}$ : maximum GIR levels; GIR- $T_{AUC50\%}$ : 50% glucose clearance time;  $AUC_{GIR,0-20 \text{ min}}$ : AUC of GIR from 0 to 20min;  $AUC_{GIR,0-120 \text{ min}}$ : AUC of GIR from 0 to 120min;  $AUC_{GIR,0-420 \text{ min}}$ : AUC of GIR from 0 to 420min;  $INS-T_{max}$ : time to peak plasma insulin concentrations;  $INS-C_{max}$ : maximal insulin levels;  $INS-T_{AUC50\%}$ : 50% insulin absorption time;  $AUC_{INS,0-20 \text{ min}}$ : AUC of insulin from 0 to 20min;  $AUC_{INS,0-120 \text{ min}}$ : AUC of insulin from 0 to 120min;  $AUC_{INS,0-420 \text{ min}}$ : AUC of insulin from 0 to 420min.

greater glucose-lowering effects in the early time period after administration. Previous data on the rapid-acting insulin analogs administered by the needle-free jet injector are limited. A non-randomized study has evaluated profiles of insulin lispro using jet injector technology, but the sample size was small and the dose of insulin lispro was unreasonable[5]. A study conducted by Engwerda et al. in 18 healthy volunteers, using a randomized, double-blind, double-dummy, crossover study design, found that GIR-Tmax as well as INS-Tmax were significantly shorter when aspart was injected by jet injection compared to conventional pen injection[3]. The results of our experiment were consistent with the above studies and further confirmed the advantage of the QS-M jet injector for delivering lispro in the Chinese population.

Both Engwerda's study and our study showed no significant difference in total AUC<sub>INS</sub> and AUC<sub>GIR</sub>; our study revealed that AUC<sub>GIR,0-20 min</sub> was significantly larger in the jet injector group, indicating a greater insulin exposure in the early time period after administration. In healthy individuals, there is an early-phase insulin secretion after glucose load, which is deficient in type-2 diabetes [19]. Timely insulin administration to restore early-phase insulin secretion is important to improve postprandial glucose excursions [20]. The time of physiological early-phase insulin secretion is about 30 min after oral glucose loading [21,22]. Our results showed that the average INS-Tmax of lispro insulin administered by conventional injection was 80.56 min. The average INS-Tmax was 37.78 min for jet injection, about 43 min earlier than conventional injection, suggesting that the jet injection resembles the pattern of endogenous early-phase insulin secretion more closely.

One of the major concerns with jet injector is the adverse reactions. Some studies have reported no local reactions after jet injections of insulin [3], whereas others have reported significant reaction, including immediate pain, delayed pain, bleeding, and hematomas [6,7]. In the present study, a trained nurse was arranged to perform the injection, and no hematomas, bruising, and bleeding were reported in the needle-free injection group.

There are several limitations of the current study. One is the small sample size; however, the randomized, crossover study design makes some remedies to this limitation. Besides, double-dummy ensures that both participants and investigators were blinded during the trial; therefore, potential confounders were avoided. Second, the study was performed in healthy individuals, rather than in patients with diabetes. Although the clinical relevance of the PK-PD differences between jet injector and insulin pen has been illustrated by findings from other studies in patients with diabetes [6,7,9], the advantages of QS-M jet injector in lispro administration still need to be tested in diabetic patients.

In conclusion, lispro administered by the QS-M needle-free injector results in an earlier and higher insulin exposure than the conventional pen, and a greater early glucose-lowering effect with similar overall potency. Enhanced insulin absorption by jet injection mainly occurred during the early period after administration; this closely resembles the pattern of endogenous early-phase insulin secretion. The improved profiles of lispro by jet injection might better mimic the physiological postprandial insulin secretion.

## 5. Contribution statement

Jinbo Hu and Hui Shi designed the study, oversaw the data collection, and wrote the manuscript. Changhong Zhao and Xiyue Li assisted with the data collection, and contributed to the writing and editing of the manuscript. Yue Wang and Qingfeng Cheng assisted with the data collection. Richa Goswami and Qianna Zhen contributed to the study design and data collection. Mei Mei and Ying Song provided statistical expertise, and contributed to the writing of the manuscript. Shumin Yang and Qifu Li are the guarantors of this work and, as such, had full access to all the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.

## Declaration of interest

The authors thank Laboratory of Endocrine and Laboratory of Lipid & Glucose Metabolism, the First Affiliated Hospital of Chongqing Medical University. This research was supported by the National Key Clinical Specialties Construction Program of China, the National Natural Science Foundation of China (81370954) and The Fundamental Science & Advanced Technology Research of Chongqing (Major Project, cstc2015jcyjBX0096). The authors have no other relevant affiliations or financial involvement with any organization or entity with a financial interest in or financial conflict with the subject matter or materials discussed in the manuscript apart from those disclosed.

## References

Papers of special interest have been highlighted as:

• of interest

•• of considerable interest

- Friedberg SJ, Lam YW, Blum JJ, et al. Insulin absorption: a major factor in apparent insulin resistance and the control of type 2 diabetes mellitus. *Metabolism*. 2006;55:614–619.
- Home PD. The pharmacokinetics and pharmacodynamics of rapid-acting insulin analogues and their clinical consequences. *Diabetes Obes Metab*. 2012;14:780–788.
- **PK-PD profiles of Aspart administered by jet injector in healthy participants.**
- Engwerda EE, Abbink EJ, Tack CJ, et al. Improved pharmacokinetic and pharmacodynamic profile of rapid-acting insulin using needle-free jet injection technology. *Diabetes Care*. 2011;34:1804–1808.
- Mitragotri S. Current status and future prospects of needle-free liquid jet injectors. *Nat Rev Drug Discov*. 2006;5:543–548.
- **Introduction of Needle-Free Jet Injector.**
- Sarno MJ, Bell J, Edelman SV. Pharmacokinetics and glucodynamics of rapid-, short-, and intermediate-acting insulins: comparison of jet injection to needle syringe. *Diabetes Technol Ther*. 2002;4:863–836.
- Zhou MC, Wang Y, Dong YX, et al. A comparative study of the effects of needle free (INJEX30) versus insulin pen injection on insulin absorption in diabetic patients. *Zhonghua Nei Ke Za Zhi*. 2013;52:741–744.
- Engwerda EE, Tack CJ, de Galan BE. Needle-free jet injection of rapid-acting insulin improves early postprandial glucose control in patients with diabetes. *Diabetes Care*. 2013;36:3436–3441.
- **PK-PD profiles of Aspart administered by jet injector in patients with diabetes.**
- de Galan BE, Engwerda EE, Abbink EJ, et al. Body mass index and the efficacy of needle-free jet injection for the administration of rapid-acting insulin analogs, a post hoc analysis. *Diabetes Obes Metab*. 2013;15:84–86.
- de Wit HM, Engwerda EE, Tack CJ, et al. Insulin administered by needle-free jet injection corrects marked hyperglycaemia faster in overweight or obese patients with diabetes. *Diabetes Obes Metab*. 2015;17:1093–1099.

- **Different PK-PD profiles of Aspart administered by jet injector in obese and non-obese participants.**
10. Bretzel RG, Nuber U, Landgraf W, et al. Once-daily basal insulin glargine versus thrice-daily prandial insulin lispro in people with type 2 diabetes on oral hypoglycaemic agents (APOLLO): an open randomised controlled trial. *Lancet*. 2008;29:1073–1084.
  11. Hedman CA, Lindström T, Arnqvist HJ. Direct comparison of insulin lispro and aspart shows small differences in plasma insulin profiles after subcutaneous injection in type 1 diabetes. *Diabetes Care*. 2001;24:1120–1121.
  12. Yang S, Li Q, Zhong L, et al. Serum pigment epithelium-derived factor is elevated in women with polycystic ovary syndrome and correlates with insulin resistance. *J Clin Endocrinol Metab*. 2011;96:831–836.
  - **How to perform euglycemic clamp tests.**
  13. Weiping L, Qingfeng C, Shikun M, et al. Elevated serum RBP4 is associated with insulin resistance in women with polycystic ovary syndrome. *Endocrine*. 2006;30:283–287.
  14. Zhu Q, Zhou H, Zhang A, et al. Serum LBP is associated with insulin resistance in women with PCOS. *PLoS One*. 2016 Jan 22;11:e0145337.
  15. Abdul-Ghani M, DeFronzo RA. Fasting hyperglycemia impairs glucose- but not insulin-mediated suppression of glucagon secretion. *J Clin Endocrinol Metab*. 2007;92:1778–1784.
  16. Vilsbøll T, Krarup T, Madsbad S, et al. Defective amplification of the late phase insulin response to glucose by GIP in obese Type II diabetic patients. *Diabetologia*. 2002;45:1111–1119.
  17. Cengiz E, Tamborlane WV, Martin-Fredericksen M, et al. Early pharmacokinetic and pharmacodynamic effects of mixing lispro with glargine insulin: results of glucose clamp studies in youth with type 1 diabetes. *Diabetes Care*. 2010;33:1009–1012.
  18. Heise T, Hövelmann U, Brøndsted L, et al. Faster-acting insulin aspart: earlier onset of appearance and greater early pharmacokinetic and pharmacodynamic effects than insulin aspart. *Diabetes Obes Metab*. 2015;17:682–688.
  19. Cheng K, Andrikopoulos S, Gunton JE. First phase insulin secretion and type 2 diabetes. *Curr Mol Med*. 2013;13:126–139.
  20. Weng J, Li Y, Xu W, et al. Effect of intensive insulin therapy on beta-cell function and glycaemic control in patients with newly diagnosed type 2 diabetes: a multicentre randomised parallel-group trial. *Lancet*. 2008;24(371):1753–1760.
  21. Mitrakou A, Kelley D, Mokan M, et al. Role of reduced suppression of glucose production and diminished early insulin release in impaired glucose tolerance. *N Engl J Med*. 1992;326:22–29.
  22. Del Prato S. Loss of early insulin secretion leads to postprandial hyperglycaemia. *Diabetologia*. 2003;46(Suppl 1):M2–M8.