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Lack of acute insulin effect on plasma endothelin-1 levels in humans

Kozo Katsumori[a], Taro Wasada[a,*], Akiko Saeki[a], Mitsuhide Naruse[b], Yasue Omori[a]

[a]*Diabetes Center, Tokyo Women's Medical College, 8-1 Kawada-cho, Shinjuku-ku, Tokyo 162 Japan*
[b]*Institute of Clinical Endocrinology, Tokyo Women's Medical College, 8-1 Kawada-cho, Shinjuku-ku, Tokyo 162 Japan*

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Abstract

Acute hyperinsulinemia does not increase circulating ET-1 levels in subjects with normal and deranged glucose metabolism.

Keywords: Plasma endothelin-1; Humans; NIDDM; Insulin

Recent in vivo studies indicate that plasma endothelin(ET)-1 levels increase during hyperinsulinemic euglycemic clamp studies in obese women [1] and lean normotensive men with NIDDM [2]. These findings are also in keeping with the in vitro data [3–5]. Since ET-1 is a potent vasoconstrictive and mitogenic endothelium-derived peptide, it may be involved in the pathogenesis of hypertension and atherosclerosis associated with insulin resistance. However, the in vivo results are still controversial [6]. To address the issue, we re-evaluated whether hyperinsulinemia modulates circulating ET-1 levels in the normotensive subjects with varying degree of glucose tolerance(NGT, IGT and NIDDM). Demographic features of these subjects are presented in Table 1. Euglycemic hyperinsulinemic clamp studies were performed after an overnight fast, as reported previously [7]. Two stepwise insulin infusion rates (1.12 and 5.0 mU/kg/min) were used with resultant hyperinsulinemia of 50–80 μU/ml and 300–500 μU/ml, respectively.

Before and during final 30 min of hyperinsulinemic euglycemia, blood samples for ET-1 were collected into chilled tube containing EDTA-2Na(2 mg/ml) and aprotinin(300 kIU/ml). In order to avoid venous stasis and venipuncture, blood samples were drawn through an indwelling heparin-lock catheter.

Plasma ET-1 levels were determined by a sensitive enzyme immunoassay as described previously [8]. This assay is based on a sandwich method that used two different anti-endothelin-1 antibodies with the first monoclonal antibody against the N-terminal portion and the second polyclonal antibody against the C-terminal heptapeptide. The

* Corresponding author.

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Table 1
Demographic features of the subjects

	NGT	IGT	NIDDM
Number (M:F)	4 (1:3)	3 (2:1)	4 (2:2)
Age (years)	39.5 (17–67)	49.7 (42–59)	57.5 (47–67)
BMI (kg/m^2)	23.1 (20.6–25.1)	27.5 (23.8–32.5)	25.1 (21.5–29.5)
FPG (mg/dl)	88 (76–98)	104 (94–107)	132 (100–164)
HbA_{1c} (%)	4.5 (4.1–5.0)	5.8 (5.0–6.4)	7.0 (5.1–9.5)
FIRI (μU/ml)	15.5 (5.8–28.4)	9.1 (8.0–10.7)	6.1 (3.2–7.6)
BP (mmHg)			
Syst	130 (118–140)	125 (100–145)	134 (128–146)
Diast	80 (70-88)	82 (70–92)	81 (70–87)
GIR (mg/kg/min)	5.1 (3.3–6.8)	4.2 (3.6–5.4)	3.9 (2.0-5.7)
Therapy			
D:OHG:Ins			2:0:2

Mean(range); D, diet alone; OHA, oral hypoglycemic agent; In, insulin; FIRI, fasting immunoreactive insulin; GIR, glucose infusion rate.

sensitivity of the assay was 0.2 pg of ET- 1/well. Although first antibody showed a cross-reactivity of 160% with ET-2 which does not exist in the blood [9], there was no significant cross-reactivity (less than 0.25%) with other related ET-family peptides.

As shown in Table 2, the mean baseline level of plasma ET-1 showed a tendency to increase along with increasing glucose intolerance. However, mean plasma ET-1 level did not show any significant change during physiological and supraphysiological degree of hyperinsulinemia. In fact, there was no individual case that showed an increase more than 50% of the baseline level. In addition, no significant relationship was found between plasma ET-1 levels and plasma insulin level or degree of insulin resistance. Therefore, the current findings cast doubt on the acute stimulation of ET-1 secretion by insulin. Lack of acute effect seems to agree with the general concept that ET synthesis and release from endothelial cells is regulated at a transcriptional step rather than during secretory process [9]. Further studies on more chronic effect are required to conclude the physiological importance of insulin in modifying plasma ET-1 levels.

Table 2
Change of plasma endothelin-1 levels during euglycemic hyperinsulinemic clamp study

	Plasma endothelin-1 level (pg/ml)		
		Insulin infusion rate (mU/kg/min)	
	Baseline	1.12	5.0
NGT(4)	1.2(0.9-1.7)	1.3(1.0-2.0)	1.1(0.5-1.6)
IGT(3)	1.9(1.4-2.8)	1.5(1.1-2.0)	1.3(0.9-1.8)
NIDDM(4)	2.1(1.0-3.5)	1.8(1.3-2.3)	1.8(1.3-2.2)

Mean (range).

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