ORIGINAL

Factors associated with the glucose-lowering effect of vildagliptin identified from the results of the oral glucose tolerance test in Japanese patients with type 2 diabetes

Akinobu Nakamura and Yasuo Terauchi

Department of Endocrinology and Metabolism, Graduate School of Medicine, Yokohama City University, Yokohama 236-0004, Japan

Abstract. In order to investigate the factors contributing to the glucose-lowering effect of vildagliptin, we analyzed the results of the oral glucose tolerance test together with several clinical parameters in Japanese patients with type 2 diabetes before and after 24 weeks of treatment with vildagliptin. The data of the 13 patients who satisfactorily completed the follow-up examinations were included. After 24 weeks treatment with vildagliptin, the patients were classified into a responder group (69.2%) and a non-responder group (30.8%); the responders consisting of subjects whose HbA1c decreased following 24 weeks treatment with vildagliptin, and the non-responders consisting of subjects who did not show any significant decrease of HbA1c. There were no differences in baseline characteristics between the two groups before administration of vildagliptin. After 24 weeks of treatment, HbA1c was significantly reduced from $7.3 \pm 0.5\%$ to $6.7 \pm 0.5\%$ in the responder group ($P = 0.0077$), while it tended to rather increased from $7.1 \pm 0.6\%$ to $7.5 \pm 0.7\%$ in the non-responder group ($P = 0.0679$). Also, parameters reflecting the glucose-stimulated insulin secretion, such as the insulinogenic index and oral disposition index, were significantly higher in the responder group than in the non-responder group, whereas insulin sensitivity was similar between the two groups. These results suggest that the difference in the degree of improvement of the glucose tolerance between the responder group and non-responder group in this study could be associated with the effect of vildagliptin on the glucose-stimulated insulin secretion, but not on the insulin sensitivity.

Key words: Vildagliptin, Oral glucose tolerance test, Glucose-stimulated insulin secretion, Insulin sensitivity

VILDAGLIPTIN is a novel oral anti-diabetic agent that selectively inhibits dipeptidyl peptidase-4 (DPP-4), thereby increasing the levels of active glucagon-like peptide-1 (GLP-1) and glucose-dependent insulinotropic polypeptide. Several clinical trials have shown the efficacy of vildagliptin in patients with type 2 diabetes, administered either as monotherapy or in combination with other drugs, without clinically significant weight gain and with minimal hypoglycemia [1]. Previous reports indicated that chronic administration of vildagliptin combined with metformin to patients with type 2 diabetes improved the insulin secretion and insulin sensitivity as estimated by the meal tolerance test (MTT) [2], and that chronic vildagliptin treatment

administered to patients with type 2 diabetes improved the acute insulin and C-peptide responses to glucose as estimated by the intravenous glucose tolerance test [3]. Also, Balas *et al.* showed that single administration of vildagliptin augmented insulin secretion and inhibited glucagon release, leading to enhanced suppression of endogenous glucose production during MTT, as estimated using the double tracer technique [4]. Although the oral glucose tolerance test (OGTT) is simpler, less time-consuming and less expensive to perform than the intravenous or tracer tests, and is often used in intervention studies, the efficacy of vildagliptin has not yet been adequately examined by OGTT. These conditions prompted us to investigate the factors that might contribute to the glucose-lowering effect of vildagliptin by analyzing the results of OGTT together with several clinical parameters in Japanese patients with type 2 diabetes before and after 24 weeks of treatment with vildagliptin.

Submitted Jun. 1, 2012; Accepted Aug. 22, 2012 as EJ12-0208
Released online in J-STAGE as advance publication Sep. 9, 2012

Correspondence to: Yasuo Terauchi, M.D., Ph.D., Professor, Department of Endocrinology and Metabolism, Graduate School of Medicine, Yokohama City University, 3-9 Fukuura, Kanazawa-ku, Yokohama 236-0004, Japan. E-mail: terauchi-ty@umin.ac.jp

Materials and Methods

Japanese type 2 diabetic patients over 20 years of age, who visited the outpatient clinic of the Yokohama City University Hospital or Kanagawa Cardiovascular and Respiratory Center, were recruited into the present study. The study was conducted with the approval of the institutional review board and written informed consent from the patients was obtained from all patients. Type 2 diabetes was diagnosed in accordance with the criteria proposed by the Japan Diabetes Society (JDS) [5]. The exclusion criteria were type 1 diabetes and patients receiving insulin therapy or already receiving DPP-4 inhibitor treatment. Other exclusion criteria were pregnancy, possible pregnancy and lactation. A history of severe hepatic dysfunction was also considered as an exclusion criterion.

We analyzed several clinical parameters, including HbA1c and body weight before and after 24 weeks of treatment with vildagliptin at 100 mg/day. Also, the plasma glucose and insulin were measured after an overnight 12-h fast and during a 75-g OGTT at 30, 60, 90 and 120 min before and after 24 weeks of treatment with vildagliptin. On the day of the OGTT, the medication before the OGTT was withheld. The HbA1c (%) value was estimated as a National Glycohemoglobin Standardization Program (NGSP) equivalent value (%) calculated by using the formula: $\text{HbA1c (\%)} = \text{HbA1c (JDS) (\%)} + 0.4 \%$, considering the relational expression of HbA1c (JDS) (%) measured by the previous Japanese standard substance and measurement methods and HbA1c (NGSP) [5]. Insulin sensitivity was estimated by the use of the Matsuda index [6] and homeostasis model assessment of insulin resistance (HOMA-IR) [7]. Glucose-stimulated insulin secretion was evaluated based on the insulinogenic index [8], oral disposition index [9], the insulin-to-glucose ratio determined 30 min after the glucose load (insulin/glucose 30 min) [10] and homeostasis model assessment of the beta cell function (HOMA- β) [7]. In the present study, the oral disposition index was calculated as the product of Matsuda index and insulinogenic index.

Results are expressed as means $\pm$ SD. Differences between the responder group and the non-responder group were analyzed for statistical significance by the Mann-Whitney *U*-test for unpaired comparisons. Paired analyses within groups were conducted using Wilcoxon's signed-rank test. All *P* values were

2-sided, and values less than 0.05 were considered to be statistically significant.

Results

A total of 15 patients were recruited. Of these, the examinations could not be completed satisfactorily in 2 patients. The data of the remaining 13 patients were included in the analysis. Out of the 13 patients, 8 were men and 5 were women. The mean age was 68.9 ± 6.7 years. At baseline, the mean fasting plasma glucose (FPG) was 156.9 ± 39.0 mg/dL, and the mean HbA1c was $7.2 \pm 0.5\%$. The average duration of diabetes was 6.3 ± 5.7 years, and the mean body mass index (BMI) was 22.4 ± 3.3 kg/m². Of the 13 subjects, 4 were treated with vildagliptin alone, and 4 received vildagliptin together with a sulfonylurea; the remaining were treated with vildagliptin in place of an alpha-glucosidase inhibitor ($n = 2$), glinide ($n = 1$), or a combination of an alpha-glucosidase inhibitor and a glinide ($n = 2$). After 24 weeks, the FPG decreased to 136.5 ± 34.9 mg/dL and the HbA1c decreased to $6.9 \pm 0.7\%$, with no significant increase of the body weight (Before treatment: 57.7 ± 11.8 kg, After treatment: 57.5 ± 11.3 kg).

When these results were analyzed in detail, we noted the existence of responders and non-responders: the responders consisting of subjects whose HbA1c decreased following 24 weeks treatment with vildagliptin, and the non-responders consisting of subjects who did not show any significant decrease of the HbA1c. Therefore, we analyzed the results by dividing the entire subject population into the responder and non-responder groups; after 24 weeks of treatment with vildagliptin, 4 (30.8%) patients were classified into the non-responder group. Before the start of treatment with vildagliptin, there were no differences in the baseline characteristics (Table 1) or the breakdown of other oral hypoglycemic medication (data not shown) between the two groups. After 24 weeks of treatment, HbA1c was significantly reduced from $7.3 \pm 0.5\%$ to $6.7 \pm 0.5\%$ in the responder group ($P = 0.0077$), while it tended to rather increase from $7.1 \pm 0.6\%$ to $7.5 \pm 0.7\%$ in the non-responder group ($P = 0.0679$). There were no differences in the degree of change of the body weight between the two groups (Before treatment: 56.8 ± 13.3 kg in the responder group; 59.7 ± 8.9 kg in the non-responder group, After treatment: 56.9 ± 13.1 kg in the responder group; 58.9 ± 7.5 kg in the non-responder group).

Table 1 Baseline characteristics of responders and non-responders to vildagliptin

	Responder	Non-responder	P value
Number of subjects	9 (69.2%)	4 (30.8%)	
Age (years)	68.8 ± 7.1	69.3 ± 6.8	0.8162
Sex (male/female)	5 / 4	3 / 1	
Diabetes duration (years)	5.3 ± 5.5	8.5 ± 6.4	0.2094
Body weight (kg)	56.8 ± 13.3	59.7 ± 8.9	0.4398
Body mass index (kg/m ²)	22.1 ± 2.8	23.0 ± 4.7	0.8774
Fasting plasma glucose (mg/dL)	153.1 ± 43.9	165.5 ± 28.3	0.4404
HbA1c (%)	7.3 ± 0.5	7.1 ± 0.6	0.5866

Values are means ± SD.

Table 2 Comparison of insulin secretion and insulin sensitivity estimated by OGTT between responders and non-responders pre- or post-administration of vildagliptin

		Responder	Non-responder	P value
Glucose Σ0-120 min	pre	1177 ± 133	1103 ± 167	0.6434
	post	909 ± 155**	1239 ± 282	0.0370
Insulin Σ0-120 min	pre	138 ± 54	110 ± 37	0.2801
	post	158 ± 102	104 ± 59	0.2801
Matsuda index	pre	3.66 ± 2.23	4.62 ± 3.03	0.6434
	post	4.75 ± 2.55	4.27 ± 2.36	0.6434
HOMA-IR	pre	3.49 ± 2.48	3.38 ± 3.46	0.7576
	post	2.38 ± 1.60	3.33 ± 2.87	0.3152
Insulinogenic index	pre	0.14 ± 0.07	0.10 ± 0.02	0.4404
	post	0.27 ± 0.22*	0.07 ± 0.01	0.0308
Oral disposition index	pre	0.48 ± 0.35	0.43 ± 0.24	0.7576
	post	1.12 ± 1.11	0.30 ± 0.15	0.0087
Insulin/glucose 30 min	pre	0.09 ± 0.04	0.07 ± 0.03	0.5371
	post	0.13 ± 0.06	0.06 ± 0.02	0.0896
HOMA-β	pre	38.9 ± 24.2	30.0 ± 18.8	0.4404
	post	42.1 ± 20.9	26.7 ± 16.1	0.3152

Values are means ± SD. * $P < 0.05$; ** $P < 0.01$ vs. pre

Next, we compared several parameters, including the insulin secretion and insulin sensitivity as estimated by the OGTT between the responder group and the non-responder group before and after 24 weeks of treatment with vildagliptin (Table 2). There were no differences in these parameters between the two groups before the start of administration of vildagliptin. After vildagliptin treatment, glucose Σ0-120 min was significantly lower in the responder group than in the non-responder group. Although the Matsuda index and HOMA-IR were similar in the two groups after treatment with vildagliptin, the insulinogenic index and

oral disposition index after vildagliptin treatment were significantly higher in the responder group than in the non-responder group. By contrast, the insulin/glucose ratio at 30 min and the HOMA-β were indistinguishable between the two groups after vildagliptin treatment.

Discussion

In this study, 69.2% of the patients classified as responders, were found to show improved glucose tolerance following treatment with 100 mg vildagliptin daily for 24 weeks. Moreover, the glucose-stimulated insulin secretion as estimated by the insulinogenic index and oral disposition index after treatment with vildagliptin were significantly higher in the responder group than in the non-responder group, whereas the insulin sensitivity as estimated by the Matsuda index and HOMA-IR were similar between the two groups. Since there were no differences in the baseline characteristics or any parameters estimated by the OGTT between the two groups before administration of vildagliptin, we assume that the difference in the degree of improvement of the glucose tolerance between the two groups could be associated with the effect of vildagliptin on the glucose-stimulated insulin secretion, which could not be predicted from the data before the start of vildagliptin administration. Then, what explains this phenomenon? We assume that genetic variations could have some influence on the difference between the two groups, since it has been reported that variations of the *GLP1R*, which encodes the GLP-1 receptor, may be associated with altered insulin secretion in response to GLP-1 [11].

Decreased insulin secretion might be induced not only by functional defects of the beta cells, but also by the loss of beta cell mass. Reduction of the beta cell mass has been shown in patients with type 2 diabetes [12]. Since we recently demonstrated that vildagliptin increased the beta cell mass in a diabetic mouse model [13], we compared the insulin/glucose ratio at 30 min, which reportedly is a better predictor of the beta cell mass [10], between the two groups; however, there was no difference in this ratio between the two groups. This could be explained to be a result of the duration of vildagliptin treatment being too short, because the increased beta cell secretory capacity following daily treatment with 100 mg vildagliptin even for 52 weeks was not maintained after the washout, indicating that this increased capacity may not have a disease-

modifying effect on the beta cell mass [14]. Also, it is necessary to set a sufficient period for washout to estimate the beta cell mass. Further study is needed over a longer duration of treatment and setting a sufficient period for washout.

The problem point of our results is whether improved glucose-stimulated insulin secretion could be a cause for the amelioration of hyperglycemia, or a result of this. Indeed, since it has been reported that vildagliptin treatment improved glucose-stimulated insulin secretion in patients with type 2 diabetes [3], we assume that improved glucose-stimulated insulin secretion could be a cause for the glycemic improvement. However, since it has also been reported that early-phase insulin release was significantly decreased in the diabetic range of FPG and 2-h plasma glucose estimated by OGTT [15], it could not be ruled out that improved glucose-stimulated insulin secretion was a result. Although there is no correlation between the insulinogenic index or oral disposition index and FPG or 2-h plasma glucose estimated by the OGTT in our study (data not shown), it might be due to a small sample. At this time, we could not clarify the problem point.

The limitations of our study were the relatively small sample and lack of a placebo group. Also, there was the limitation of the assessment by the OGTT. Although the oral route of glucose delivery is more physiological than intravenous glucose infusion, the OGTT results are poorly reducible on account of variations in

the glucose absorption and splanchnic glucose uptake [16]. However, OGTT is a simpler and less expensive test method that provides reasonably reliable estimates of insulin secretion and insulin sensitivity. Further large-scale randomized controlled studies are needed to obtain more detailed information.

In conclusion, our results suggest that the difference in the degree of improvement of the glucose tolerance between the responder group and non-responder group in this study could be associated with the effect of vildagliptin on the glucose-stimulated insulin secretion, but not on the insulin sensitivity.

Acknowledgments and Disclosure Statement

This work was supported in part by a Grant-in-Aid for Scientific Research (B) 21390282 from the Ministry of Education, Culture, Sports, Science and Technology (MEXT) of Japan, a Medical Award from the Japan Medical Association, and a Grant-in-Aid from the Uehara Memorial Foundation (to Y.T.) as well as a Grant-in-Aid for Young Scientists (Start-up) 21890213 and (B) 23791040 from the MEXT of Japan, a Grant-in-Aid from Yokohama General Promotion Foundation, and a Grant-in-Aid from Japan Diabetes Foundation (to A.N). The authors declare that they have no conflicts of interest.

References

1. Stamataros G, Schneider SH (2011) Vildagliptin in the treatment of type 2 diabetes mellitus. *Expert Opin Pharmacother* 12: 1967-1973.
2. Ahrén B, Pacini G, Foley JE, Schweizer A (2005) Improved meal-related beta-cell function and insulin sensitivity by the dipeptidyl peptidase-IV inhibitor vildagliptin in metformin-treated patients with type 2 diabetes over 1 year. *Diabetes Care* 28: 1936-1940.
3. D'Alessio DA, Denney AM, Hermiller LM, Prigeon RL, Martin JM, et al. (2009) Treatment with the dipeptidyl peptidase-4 inhibitor vildagliptin improves fasting islet-cell function in subjects with type 2 diabetes. *J Clin Endocrinol Metab* 94: 81-88.
4. Balas B, Baig MR, Watson C, Dunning BE, Ligueros-Saylan M, et al. (2007) The dipeptidyl peptidase IV inhibitor vildagliptin suppresses endogenous glucose production and enhances islet function after single-dose administration in type 2 diabetic patients. *J Clin Endocrinol Metab* 92:1249-1255.
5. The Committee of Japan Diabetes Society on the Diagnostic Criteria of Diabetes Mellitus (2010) Report of the Committee on the classification and diagnostic criteria of diabetes mellitus. *J Diabetes Invest* 1: 212-228.
6. Matsuda M, DeFronzo RA (1999) Insulin sensitivity indices obtained from oral glucose tolerance testing: comparison with the euglycemic insulin clamp. *Diabetes Care* 22: 1462-1470.
7. Matthews DR, Hosker JP, Rudenski AS, Naylor BA, Treacher DF, et al. (1985) Homeostasis model assessment: insulin resistance and beta-cell function from fasting plasma glucose and insulin concentrations in man. *Diabetologia* 28: 412-419.
8. Onishi Y, Hayashi T, Kogawa Sato K, Ogihara T, Kuzuya N, et al. (2010) Fasting tests of insulin secretion and sensitivity predict future prediabetes in Japanese

- with normal glucose tolerance. *J Diabetes Invest* 1: 191-195.
9. Utzschneider KM, Prigeon RL, Faulenbach MV, Tong J, Carr DB, et al. (2009) Oral disposition index predicts the development of future diabetes above and beyond fasting and 2-h glucose levels. *Diabetes Care* 32: 335-341.
 10. Meier JJ, Menge BA, Breuer TG, Müller CA, Tannapfel A, et al. (2009) Functional assessment of pancreatic beta-cell area in humans. *Diabetes* 58: 1595-1603.
 11. Sathananthan A, Man CD, Micheletto F, Zinsmeister AR, Camilleri M, et al. (2010) Common genetic variation in GLP1R and insulin secretion in response to exogenous GLP-1 in nondiabetic subjects: a pilot study. *Diabetes Care* 33: 2074-2076.
 12. Butler AE, Janson J, Bonner-Weir S, Ritzel R, Rizza RA, et al. (2003) Beta-cell deficit and increased beta-cell apoptosis in humans with type 2 diabetes. *Diabetes* 52: 102-110.
 13. Sato K, Nakamura A, Shirakawa J, Muraoka T, Togashi Y, et al. (2012) Impact of the dipeptidyl peptidase-4 inhibitor vildagliptin on glucose tolerance and beta-cell function and mass in insulin receptor substrate-2-knockout mice fed a high-fat diet. *Endocrinology* 153: 1093-1102.
 14. Foley JE, Bunck MC, Möller-Goede DL, Poelma M, Nijpels G, et al. (2011) Beta cell function following 1 year vildagliptin or placebo treatment and after 12 week washout in drug-naïve patients with type 2 diabetes and mild hyperglycaemia: a randomised controlled trial. *Diabetologia* 54: 1985-1991.
 15. Stancáková A, Javorský M, Kuulasmaa T, Haffner SM, Kuusisto J, et al. (2009) Changes in insulin sensitivity and insulin release in relation to glycemia and glucose tolerance in 6,414 Finnish men. *Diabetes* 58: 1212-1221.
 16. Muniyappa R, Lee S, Chen H, Quon MJ (2008) Current approaches for assessing insulin sensitivity and resistance in vivo: advantages, limitations, and appropriate usage. *Am J Physiol Endocrinol Metab* 294: E15-26.