RAPID COMMUNICATION

Careful readings for a flash glucose monitoring system in nondiabetic Japanese subjects: individual differences and discrepancy in glucose concentration after glucose loading

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Abstract. The FreeStyle Libre Flash Glucose Monitoring System (FGM), which can continuously measure glucose concentration in the interstitial fluid glucose (FGM-ISFG), has been in clinical use worldwide. However, it is not clear how accurately FGM-ISFG reflects plasma glucose concentration (PG). In the present study, we examined the clinical utility of FGM by oral glucose tolerance test (OGTT). In eight healthy volunteers (3 males; mean age, 41.8 y) wearing FGM sensors for 14 days, OGTT was performed during days 1–7 and days 8–14, and then both FGM-ISFG and PG were compared. Parkes error grid analysis indicated that all of 65 FGM-ISFG values were within Zone A (no effect on clinical action) and Zone B (little or no effect on clinical outcome). However, in OGTT, the mean FGM-ISFG was higher than the mean actual PG at 30, 60, and 90 minutes after loading (155.5 vs. 139.2 mg/dL, 166.2 vs. 139.2 mg/dL, 149.5 vs. 138.2 mg/dL, respectively; $p < 0.05$). Moreover, the area under the curve of FGM-ISFG was also significantly larger than that of PG (17,626.2 vs. 15,195.0 min·mg/dL; $p < 0.05$). In four of eight subjects, FGM-ISFG tended to be higher than PG in both OGTTs, and the greatest difference between the two values was 58 mg/dL. FGM is useful for glycemic control, whereas it is not appropriate to change therapeutic regimens based on the judgment of nocturnal hypoglycemia and postprandial hyperglycemia by FGM-ISFG. Careful attention is required for proper application of FGM.

Key words: Flash glucose monitoring, Interstitial fluid glucose

GLUCOSE MEASUREMENT using capillary samples is useful for blood glucose control in patients with diabetes mellitus. Recently, continuous glucose monitoring system (CGM), which can provide information on blood glucose levels by continuously measuring interstitial fluid glucose concentration (ISFG), is one of the most commonly used tools to detect nocturnal hypoglycemia and postprandial hyperglycemia. Although there is a time lag between fluctuation in ISFG and that in blood glucose level, ISFG is generally thought to be an acceptable indicator for blood glucose control [1].

In conventional CGM, it is necessary to measure self-monitoring capillary glucose concentrations 3–4 times a day for calibration of ISFG. However, the FreeStyle Libre Flash Glucose Monitoring System

(FGM), which can monitor ISFG for 14 days with only attachment of a small sensor patch to the upper arm without calibration, was recently approved for use in Japan. The system is so easy to handle that it can be widely used in clinical settings requiring glycemic control in prediabetic and diabetic subjects [2].

However, little information is available regarding the how accurately ISFG measured by FGM (FGM-ISFG) reflects plasma glucose concentration (PG). Therefore, we examined the clinical utility of FGM in non-diabetic subjects by comparing FGM-ISFG with PG, followed by oral glucose tolerance test (OGTT).

Subjects and Methods

Eight healthy volunteers without diabetes participated in this study. This study was approved by the Shinshu University Medical Ethics Committee, and written informed consent was obtained from all subjects. FGM was precisely attached according to the package insert and ISFG was monitored for 14 days.

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ISFG is measured automatically every 10 minutes and can be displayed on a remote device when necessary. For estimation of FGM performance, 75-g OGTT was conducted during the first period from days 1–7 after installing FGM and a second from days 7–14 day. Venous blood was sampled at 0, 30, 60, 90, and 120 minutes for PG measurements, and FGM-ISFG was recorded simultaneously. PG was measured by the glucose oxidation method. Unfortunately, some problems with FGM occurred in three subjects: in case 2, a scan failure on day 10 led to cancellation of the second OGTT; in case 4, a new sensor was reattached and then remeasured because FGM-ISFG was continuously low, and greatly deviated away from the capillary blood glucose level; in case 8, FGM-ISFG was always about 80–100 mg/dL higher than the blood glucose from SMBG, exceeding 200 mg/dL with no relation to meals, which resulted in cancellation of OGTT. Therefore, a total of 13 OGTTs were performed in seven subjects. Using the Japan Diabetes Society criteria [3], participants were categorized as having the following glucose tolerance based on fasting plasma glucose levels (FPG) and 2-h post-load plasma glucose levels (2-h PG) in 75-g OGTT: normal glucose tolerance, NGT (FPG <110 mg/dL and 2-h PG <140 mg/dL); impaired glucose tolerance, IGT (FPG <126 mg/dL and 2-h PG 140–199 mg/dL; diabetes mellitus, DM (FPG ≥126 mg/dL and/or 2-h PG ≥200 mg/dL). Parkes error grid analysis was performed to evaluate the consistency of FGM-ISFG with actual PG. In this analysis, actual PG and FGM-ISFG were plotted on the abscissa and ordinate, respectively. The zones in the graph indicate clinical usefulness as follows: Zone A: no effect on clinical action; Zone B: altered clinical action (little or no effect on clinical outcome); Zone C: altered clinical action (likely to affect clinical outcome); Zone D: altered clinical action. Next, we analyzed the individual data. The differences between FGM-ISFG and PG were examined at 0, 30, 60, 90, and 120 minutes. Pearson's product-moment correlation coefficient was calculated to determine the correlation between FGM-ISFG and PG. The Mann–Whitney U test was used to detect statistical significance of differences, and $p < 0.05$ was taken to indicate significance.

Results

The seven participants (3 men and 4 women) had a mean age of 41.4 years (range: 25–56 years). Four

were judged as having normal glucose tolerance, and the others had impaired glucose tolerance (Table 1). There was a significant and strong correlation between FGM-ISFG and PG obtained from OGTT in all participants ($r = 0.850$, $p < 0.001$). The following correlation formula was determined:

$$\text{FGM-ISFG (mg/dL)} = 24.942 + 0.94707 \times \text{Actual PG (mg/dL)}$$

As shown in Fig. 1, plotting actual PG and FGM-ISFG by the Parkes error grid method indicated that 64 (98.5%) of 65 data sets fell within Zone A, and the remaining one data set (1.5%) fell within with Zone B. There were no FGM-ISFG values in Zones C and D.

The overall result of OGTT is shown in Fig. 2. FGM-ISFG was significantly higher than PG at 30, 60, and 90 minutes after glucose loading (155.5 *vs.* 139.2 mg/dL, 166.2 *vs.* 139.2 mg/dL, 149.5 *vs.* 138.2 mg/dL, respectively; $p < 0.05$). The area under the curve of FGM-ISFG was also significantly larger than that of PG (17,626.2 *vs.* 15,195.0 min·mg/dL; $p < 0.05$). Overall mean absolute difference (MAD) and mean absolute relative difference (MARD) between FGM-ISFG and actual PG were 19.6 ± 10.0 mg/dL and $17.1 \pm 10.0\%$, respectively. MAD and MARD in each OGTT were indicated in Table 1. MAD ranged from 5.4 to 37.0 mg/dL and MARD also varied from 3.5 to 33.5%. The differences between FGM-ISFG and actual PG in each subject are shown in Fig. 3. The mean values of

Table 1 Mean absolute difference and mean absolute relative difference between interstitial fluid glucose and actual plasma glucose

Case	Age/Sex	Glucose tolerance status	MAD (mg/dL)	MARD (%)
1	51/M	IGT	16.4	9.9
			5.4	3.5
2	25/F	NGT	5.4	5.2
			N.A.	N.A.
3	27/F	NGT	22.2	20.9
			27.0	31.7
4	47/F	NGT	27.2	27.1
			37.0	33.5
5	56/M	IGT	24.0	19.3
			29.6	22.0
6	51/F	IGT	25.8	20.9
			16.0	13.0
7	33/M	NGT	8.6	6.0
			10.8	9.0

MAD, mean absolute difference; MARD, mean absolute relative difference; IGT, impaired glucose tolerance; NGT, normal glucose tolerance; N.A., not available.

Fig. 1 Parkes error grid analysis

The validity of FGM was determined by examining the Parkes error grid on which actual PG and FGM-ISFG data in OGTTs were plotted. All data fell within Zones A and B. FGM, flash glucose monitoring system; PG, plasma glucose concentration; FGM-ISFG, interstitial fluid glucose concentration by FGM.

Fig. 2 Overall results of OGTTs

FGM-ISFG was significantly higher than PG at 30, 60, and 90 minutes after glucose loading. All data are expressed as means $\pm$ standard deviation (SD). * $p < 0.05$. OGTT, oral glucose tolerance test; FGM-ISFG, interstitial fluid glucose concentration by flash glucose monitoring; PG, plasma glucose concentration.

Fig. 3 Differences between FGM-ISFG and actual PG in OGTTs

Over half of the subjects showed higher FGM-ISFG than actual PG. OGTT, oral glucose tolerance test; FGM-ISFG, interstitial fluid glucose concentration obtained by flash glucose monitoring; PG, plasma glucose concentration; N/A, not available.

FGM-ISFG and PG obtained from two OGTTs were used in this analysis. In Cases 2–5, FGM-ISFG tended to be higher than PG in both OGTTs. The greatest difference between FGM-ISFG and actual PG was 58 mg/dL at 60 minutes after glucose loading in Case 4. Furthermore, a line graph of all measured FGM-ISFG and actual PG revealed that FGM-ISFG maintained higher levels than actual PG regardless of a delayed change of ISFG (Fig. 4). In Case 7, FGM-ISFG was higher than PG in the initial OGTT, but the two values were similar in the second OGTT. In Case 1, both values in the OGTTs were almost identical.

Discussion

FGM provides accurate measurements of ISFG corresponding to capillary PG reference values, remaining stable over 14 days of wear and unaffected by patient characteristics [4]. Furthermore, in patients with type 1 diabetes, this system had a mean absolute relative difference of 13.2% and mean absolute difference of 19.8 mg/dL for the entire study period, which was similar to CGMs reported in previous studies under at-home conditions [5]. In two randomized controlled studies performed in insulin-treated adult patients with type 1 or 2 diabetes, FGM intervention failed to decrease HbA1c levels, but achieved a 38–43% reduction of time in hypoglycemia (<70 mg/dL) with less undesirable sensor-wear reactions [6, 7]. Thus, it is clinically helpful for meticulous glucose control and prevention of hypoglycemia for patients with diabetes.

We found that FGM-ISFG could be used to evaluate PG in terms of clinical decision making because all data were plotted in Zones A and B on Parkes error grid analysis. The 2013 version of ISO15197 requires $\geq 95\%$ of measured glucose values to be within Zone A or B on the Parkes error grid [8]. The FGM system was found to meet the criteria and to be fully available, indicating that the system is beneficial for non-diabetic Japanese to get information about variation patterns of blood glucose. However, it is a serious clinical problem that FGM-ISFG was higher than actual PG overall. Physiologically, it is unlikely that the glucose concentration in the interstitial fluid is higher than that in the blood. Glucose monitoring using subcutaneous interstitial fluid is less invasive, but this method can delay the detection of glycemic variability and decrease the sensitivity to local glycemic fluctuations related to the insertion sites of sensor, leading to unexplained

Fig. 4 Variation of FGM-ISFG and PG during OGTT in Case 4. FGM-ISFG levels were always higher than PG levels. OGTT, oral glucose tolerance test; FGM-ISFG, interstitial fluid glucose concentration by flash glucose monitoring; PG, plasma glucose concentration.

fluctuations of measured values [9, 10]. As these features are considered equally true in non-diabetic as well as diabetic individuals, FGM-ISFG in non-diabetic persons may tend to be displayed at a higher level due to measurement problems. Interestingly, some subjects showed little difference between FGM-ISFG and PG, whereas others showed marked differences with no tendencies related to gender, age, or glucose tolerance. Bailey *et al.* [4] reported that the accuracy of the sensor was unaffected by factors such as BMI, age, type of diabetes, insertion site, presence of insulin injection, or HbA1c. This strongly suggests that absolute values of FGM-ISFG should not be used for clinical decision making, at least in some subjects, without calibration of the values. The reasons for individual differences are unknown at present, but each showed a similar tendency in the first and second tests. Thus, appropriate calibration of absolute values may overcome these discrepancies. FGM-ISFG was higher than PG in all OGTTs. This tendency was particularly prominent with time points after glucose loading. Theoretically, the rise in FGM-ISFG should be delayed compared to that in PG at 30 minutes after glucose loading, suggesting that accuracy is particularly problematic for values after loading.

Another study showed similar findings that called into question the reliability of FGM. In a study performed in 20 subjects (8 with type 1 diabetes mellitus, 12 with type 2 diabetes mellitus), the overall correlation of FGM-ISFG showed lower values from

FGM than expected based on point of care (POC) glucose measurement in the lower glucose range, and higher than expected values in the higher ranges. Furthermore, 75-g OGTT resulted in a slower rise in glucose level as determined by FGM than that obtained by the perchloric acid hexokinase method, POC testing, test strip, and conventional CGM during the first 45–60 minutes after glucose loading [11]. Moreover, a study using CGM including FGM for type 1 diabetes patients indicated that all sensors performed less accurately during hypoglycemia and best during hyperglycemia [12]. These observations strongly suggest that values obtained from FGM should be examined in a clinical context. FGM allows blood glucose fluctuation to be detected easily, and this technique may become the central method of evaluating glycemic control of diabetes in future. However, at present, it is not appropriate to change therapeutic regimens based on the judgment of nocturnal hypoglycemia and postprandial hyperglycemia by FGM-ISFG *per se*. Combined use of more reliable devices, including instruments for self-monitoring of blood glucose and subsequent appropriate calibration, is required if FGM is adopted. In some cases, it may be necessary to measure actual PG proactively if there are doubts regarding the results obtained with FGM.

According to the current diagnostic criteria in Japan, diagnosis of diabetes is made partly based on fasting plasma glucose level, 75 g OGTT 2-hour value, and casual plasma glucose level [3]. In a potential

individual with diabetes, a missed diagnosis may occur if blood glucose levels at the points described above do not happen to meet the criteria. Meanwhile, continuous blood glucose measurement with the FGM systems may be surely allowed to catch glucose variation pattern including hyperglycemia required for diagnosis. Moreover, adoption of dynamic index such as AUC obtained from FGM-based glucose values may lead to more early and correct diagnosis of diabetes. The FGM system will have potential to become an alternative diagnostic tool for diabetes by improvement of overestimation of IFSG-based glucose levels ISFGs after glucose loading.

This study had a small number of subjects and did not include diabetic patients. To confirm our findings and obtain more detailed information on which subjects exhibit large discrepancies in FGM-ISFG and PG, it is desirable to perform clinical experiments in large numbers of subjects, including those with diabetes mellitus.

In conclusion, the individual differences in FGM-ISFG in terms of consistency with actual PG were large, and FGM-ISFG tended to be higher. This tendency became more pronounced after glucose loading. Careful attention is required for proper application of FGM.

Disclosure

None of the authors have any potential conflicts of interest associated with this research.

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