Usefulness of the index calculated as the product of levels of fasting plasma glucose and hemoglobin A1c for insulinoma screening

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Abstract. Hypoglycemia is the major symptom of insulinoma. Chronic and recurrent hypoglycemia leads to the disappearance of autonomic symptoms and persistence of non-specific symptoms alone, possibly contributing to the delayed diagnosis of insulinoma and accounting for several undiagnosed cases. We previously reported the usefulness of hemoglobin A1c (HbA1c) and glycated albumin as markers for early insulinoma screening; however, their diagnostic prediction performance and diagnostic performance were not satisfactory. We hypothesized that the product of fasting plasma glucose (FPG) and HbA1c levels (FPG × HbA1c index) is low in insulinoma, and this index may be a useful marker for screening. This cross-sectional multicenter study compared 82 insulinoma patients with 100 age-, sex-, and body mass index-matched controls with normal glucose tolerance based on 75-g oral glucose tolerance test. The FPG × HbA1c index was significantly lower in the insulinoma group than in the control group. Receiver operating curve analysis showed that the optimal cutoff point of the FPG × HbA1c index to diagnose insulinoma was 447.1, and the area under the curves (AUCs) of the FPG × HbA1c index and HbA1c were 0.998 and 0.966, respectively. The AUC of the index was significantly higher than that of HbA1c (p = 0.010). Conversely, no significant difference existed between the AUC of the FPG × HbA1c index and that of the FPG/fasting immunoreactive insulin index. Thus, in apparently healthy population, the product of FPG and HbA1c yields a useful index for insulinoma screening in terms of accuracy and versatility.

Key words: Insulinoma, Hemoglobin A1c, Fasting plasma glucose, Hypoglycemia

INSULINOMA is the commonest functional pancreatic neuroendocrine tumor. The predominant symptoms of the disease are autonomic and neurologic manifestations of hypoglycemia, mostly occurring in a fasting state. In cases of chronic recurrent hypoglycemia, the autonomic symptoms may disappear, and non-specific symptoms persist in patients [1, 2]. In a significant number of patients, a long period—up to 30 years in one case—elapsed between the appearance of symptoms and the diagnosis of insulinoma in Japan [3, 4], and several patients may often remain undiagnosed [5, 6].

Definitive diagnosis of insulinoma has been based on the confirmation of the presence of Whipple’s triad (hypoglycemic manifestation, plasma glucose level <50 mg/dL, and improvement of the symptom with glucose intake), 72-hour fasting test, and mixed meal test [7]. In clinical practice, studies with immunoreactive insulin (IRI) and C-peptide immunoreactivity (CPR) have been performed as effective screening tests at the early stage of the disease [8]. Additionally, we previously indicated that hemoglobin A1c (HbA1c) and glycated albumin (GA) are useful markers for screening for the early detection of insulinoma [9]; however, the sensitivity and specificity of these markers were not sufficient (HbA1c cutoff is 5.0%, sensitivity 77.4%, specificity
Materials and Methods

Study design and patients

Among all patients with insulinoma who visited one of three medical institutions included in our multicenter study (Hospital of University of Occupational and Environmental Health, Kobe University Hospital, and Keio University Hospital), 82 patients who met the inclusion criteria were included. The inclusion criteria were all ages and both sexes, patients with any FPG and HbA1c values, and a confirmed screening of benign insulinoma. A diagnosis of insulinoma is based on the patients at least one of two criteria described as following: 1. presence of hyperinsulinemic hypoglycemia during a 72-h fasting test and a pancreatic tumor identified the location by imaging examination, 2. presence of insulin secreting cells by histological analysis following the surgical resection of the tumor. Patients with a diagnosis of insulinoma and no metastasis between 2000 and 2016 were selected. Patients with malignant insulinoma, hepatic disease, renal disorder, and/or anemia were excluded. A total of 100 healthy with normal plasma glucose and insulin levels, confirmed individuals with 75-g oral glucose tolerance test in a medical check, whose characteristics [age, sex, and body mass index (BMI)] were matched to the study cases were included as the control group [10]. This study was conducted in conformity with the Declaration of Helsinki and current ethical codes and was approved by the ethics committee at the University of Occupational and Environmental Health. This study was conducted as part of standard clinical practice with intervention, and the information was retrospectively collected from the medical records. This study was not a clinical study, and the data were anonymized for retrospective analyses. Therefore, written informed consent was not obtained from the study participants.

Study outcomes

Blood collections were performed for all subjects after overnight fasting. Age, sex, BMI, levels of FPG, fasting IRI, and HbA1c, as well as the product of FPG and HbA1c (FPG × HbA1c index), were evaluated in both the study groups, and the primary outcome was the determination of optimal cutoff value of the FPG × HbA1c index for screening of insulinoma.

Statistical analyses

All the values evaluated were expressed as mean ± standard deviation (SD) or median [Q1, Q3]. The Kolmogorov-Smirnov normality test showed normal distributions for FPG and HbA1c but not for age, BMI, FPG × HbA1c index, fasting IRI and, FPG/fasting IRI index. For intergroup comparison, unpaired t tests and the Mann-Whitney U tests were performed for data with a normal distribution and for those without a normal distribution, respectively. In addition, chi-square tests were conducted to compare between both sexes. Receiver operating characteristic (ROC) curves were drawn to calculate area under the curves (AUCs) and 95% confidence intervals (95% CI) to determine the most significant marker among HbA1c, FPG × HbA1c index, and FPG/fasting IRI index for the screening of insulinoma. To compare the diagnostic discrimination among HbA1c, the FPG × HbA1c index, and the FPG/IRI index, the AUCs were compared by DeLong method. The significance level was set at p-value < 0.05. SPSS Statistical Software 25.0 (SPSS Inc., Chicago, IL) was used for the analyses.

Results

The subject characteristics are shown in Table 1. Data are mean ± SD or median [Q1, Q3]. The number of subjects in the insulinoma and control groups were 82 (24 males, 58 females) and 100 (33 males, 67 females), respectively. No significant differences in age (62.0 years vs. 59.0 years; p = 0.277) and BMI (23.6 kg/m² vs. 23.4 kg/m²; p = 0.211) were observed between the two groups. FPG was significantly lower in the insulinoma group at 50.4 ± 18.7 mg/dL than in the control group at 93.3 ± 6.5 mg/dL (p < 0.001). Conversely, fasting IRI was significantly higher in the insulinoma group at 18.7 [7.7, 24.9] μU/mL than in the control group at 4.1 [3.2, 6.0] μU/mL (p < 0.001). HbA1c was significantly lower
in the insulinoma group at 4.6 ± 0.5% than in the control group at 5.7 ± 0.3% (p < 0.001).

FPG × HbA1c index was significantly lower in the insulinoma group at 217.6 [163.6, 293.0] than in the control group at 527.5 [496.4, 560.3] (p < 0.001) (Fig. 1). The sensitivities, specificities, positive and negative predictive values, and positive and negative likelihood ratios in the ROC curve analysis obtained using different cutoff values for the FPG × HbA1c index are summarized in Table 2. The optimal cutoff value of FPG × HbA1c index for the screening of insulinoma was 447.1 (AUC = 0.998; 95% CI, 0.995–1.000) (Table 2). The FPG × HbA1c index yielded 82 true positives, 0 false negatives, 4 false positives, and 96 true negatives with the cutoff value of 447.1. There are 4 false positives in the control group, which characterized by low normal FPG levels (74–83 mg/dL), both with low IRI and FPG/fasting IRI. AUCs of HbA1c and the FPG × HbA1c index were 0.966 and 0.998, respectively (Fig. 2), showing a significant difference (p = 0.010). In contrast, AUCs of the FPG/fasting IRI index and the FPG × HbA1c index were 0.995 and 0.998, respectively (Fig. 3), showing no significant difference (p = 0.432).

Discussion

In this study, the product of FPG and HbA1c was significantly lower in the insulinoma group than in the control group, and the optimal cutoff value of the index for screening insulinoma was determined. In the ROC curve analysis, the AUC of the FPG × HbA1c index for diagnosing insulinoma was significantly higher than that of HbA1c and was comparable to that of the FPG/fasting IRI index.

This study demonstrated that the product of FPG and HbA1c is useful as a highly accurate index for diagnosing insulinoma. When chronically exposed to hypoglycemia, the autonomic symptoms associated with hypoglycemia may disappear, resulting in the persistence of non-specific symptoms alone, which are unlikely to be recognized as hypoglycemic manifestations [1, 2], leading to the unawareness of hypoglycemia in many patients with insulinoma. In such cases, because of the unawareness of hypoglycemia, subjective symptoms are often absent even when plasma glucose levels are extremely low [12]. We previously reported the usefulness of HbA1c and GA for screening of insulinoma [9]; however, there were limitations in sensitivity and specificity of these markers for screening. The lack of sufficient sensitivity and specificity was considered attributable to individual variations in

![Fig. 1](image1.png)

Comparison of the FPG × HbA1c index between the insulinoma and control groups

Data were compared using the Mann-Whitney U test for variables.

![Table 1](image2.png)

Comparison of clinical characteristics between insulinoma group and control group

|                          | Insulinoma group | Control group | p value |
|--------------------------|------------------|---------------|---------|
| n                        | 82               | 100           |         |
| Male gender, n (%)       | 24 (31.0)        | 33 (33.0)     | 0.440   |
| Age, years               | 62.0 [47.0, 75.0]| 59.0 [58.0, 59.0] | 0.277   |
| BMI score, kg/m²         | 23.6 [21.0, 27.4]| 23.4 [22.3, 24.7] | 0.211   |
| FPG, mg/dL               | 50.4 ± 18.7      | 93.3 ± 6.5    | <0.001  |
| Fasting IRI, μU/mL       | 18.7 [7.7, 24.9] | 4.1 [3.2, 6.0] | <0.001  |
| FPG/fasting IRI index    | 0.62 [0.29, 3.31]| 22.6 [16.08, 29.01] | <0.001  |
| HbA1c, %                 | 4.6 ± 0.5        | 5.7 ± 0.3     | <0.001  |

Data are mean ± SD or median [Q1, Q3]. BMI, body mass index; FPG, fasting plasma glucose; IRI, immunoreactive insulin; HbA1c, hemoglobin A1c. P values are for comparison of differences between two groups.
HbA1c and GA. However, in this study, we could obtain an index with high sensitivity and specificity using a combination of long- and short-term indices, i.e., the product of FPG and HbA1c.

In the ROC curve analysis, the AUC of the FPG × HbA1c index for predicting insulinoma was comparable to that of the FPG/fasting IRI index, indicating that the product of FPG and HbA1c is useful for the screening of insulinoma. Based on the present findings, we consider that this screening marker might be useful to screen several patients with hypoglycemia because it did not require measurement of fasting IRI. In the future, it is necessary to verify the cutoff of FPG × HbA1c index for chronic hypoglycemia. Insulinoma is suspected if Whipple’s triad is present (loss of consciousness during fasting plasma glucose of 50 mg/dL or below during attacks, and improvement of symptoms with glucose administration). Traditionally, evaluation of fasting IRI has been considered essential for screening of insulinoma because hypoglycemia usually occurs during fasting following increased autonomous secretion of insulin. To date, several indices using IRI have been suggested for screening purposes, and the FPG/fasting IRI index has been reported to be useful [8]. In this study, the AUC of the FPG/fasting IRI index for predicting insulinoma was comparable to that of the product of the FPG × HbA1c index; however, the former reflects the insulin secretion status irrespective of the hypoglycemic status, whereas the latter indicates the presence of chronic hypoglycemia. Therefore, the FPG/fasting IRI index might be

### Table 2

| Cutoff | S (%) | Sp (%) | PLR | NLR | AUC (%) |
|--------|-------|--------|-----|-----|---------|
| FPG × HbA1c | 447.1 | 100.0 | 96.0 | 25.0 | 0.00 | 0.998 |
| HbA1c | 5.1 | 84.1 | 99.0 | 84.1 | 0.16 | 0.966 |
| FPG | 75 | 90.2 | 99.0 | 90.2 | 0.10 | 0.978 |
| FPG/fasting IRI | 9.0 | 95.0 | 99.0 | 95.0 | 0.05 | 0.995 |

S, sensitivity; Sp, specificity; PLR, positive likelihood ratio; NLR, negative likelihood ratio; AUC, area under the curve.
useful for differentiating insulinoma from non-islet cell tumor hypoglycemia (NICTH), which can cause chronic hypoglycemia without excessive insulin secretion. In addition, the FPG × HbA1c index may also be useful in the screening of diseases other than insulinoma that can cause chronic hypoglycemia, such as NICTH.

This study had several limitations. First, the control group comprised individuals with normal glucose tolerance. This index is a well-known marker for screening of hypoglycemia; however, further studies that include patients with hypoglycemia (such as liver disease, renal dysfunction, hypopituitarism, adrenal insufficiency and extrapancreatic tumor) as controls is needed to validate the index to confirm hypoglycemia. Second, HbA1c levels may be affected in patients taking iron, using erythropoietin, or in patients with hemoglobinopathies. Therefore, this index cannot be applied to such patients. Third, patients with a diagnosis of insulinoma and no metastasis were selected in this study. However, not all patients had surgery, and malignant insulinoma may be mixed. Fourth, this study is a retrospective multicenter study, and HbA1c and IRI measuring instruments are different in each institution. Fifth, we need to evaluate each diagnostic ability, considering that the meaning of FPG and fasting IRI included in the gold standard of insulinoma diagnosis is different from HbA1c. Sixth, insulinoma is a rare disease, screening tests with few false positives are desirable. FPG × HbA1c has a higher false-positive rate than other indicators, and if positive, it is necessary to differentiate insulinoma using other indicators and tests. Additionally, this was a retrospective study with a small sample size. A large-scale prospective cohort study is warranted for validation of the study findings.

In conclusion, the product of FPG and HbA1c was significantly lower in insulinoma patients than in healthy controls, and the accuracy of this index was comparable to that used in the conventional screening method. We conclude that in apparently healthy population the product of FPG and HbA1c is a useful index for screening of insulinoma in terms of accuracy and versatility.

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Disclosure

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

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