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Distinct association of serum FGF21 or adiponectin levels with clinical parameters in patients with type 2 diabetes

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ABSTRACT

Fibroblast growth factor 21 (FGF21) has been identified as a novel metabolic regulator. This cross-sectional study was performed to clarify how serum FGF21 levels were associated with clinical parameters in Japanese subjects with type 2 diabetes (n = 139). Anthropometric and blood biochemical parameters, uses of drugs for diabetes, hypertension and dyslipidemia were examined regarding associations with fasting serum FGF21 concentrations. FGF21 levels were 6-times higher in those subjects taking fibrates. However, a use of thiazolidinediones did not affect serum FGF21 levels while it induced higher serum adiponectin levels. In univariate analyses, FGF21 levels showed associations with a use of fibrates, triglyceride levels, creatinine levels, waist circumference, and BMI. Multiple regression analyses adjusted for age, gender and BMI showed that a use of fibrates, triglyceride levels and creatinine levels were strong contributors to serum FGF21 levels. In contrast, a use of thiazolidinediones, HDL-cholesterol levels and fasting insulin levels were strong contributors to serum adiponectin levels. This study revealed that serum FGF21 levels were biochemical indicators correlating to a set of essential metabolic parameters, which was distinct from that correlating to serum adiponectin levels in subjects with type 2 diabetes.

1. Introduction

Fibroblast growth factor 21 (FGF21) was isolated several years ago as a new member of FGF family comprised of more than 20 FGFs [1]. FGF19, 21 and 23 are evolutionarily close to each other and involved in the metabolism of bile acids, glucose/lipids and phosphate/calcium, respectively, thus recognized as metabolic FGFs [2–5]. Human FGF21 encodes 209 amino acids and is produced mainly from liver with a minor contribution from thymus [1]. Recent reports indicated skeletal muscle and adipocytes as other important sources of FGF21 production [6–8]. Regarding the transcriptional control of FGF21 expression,

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Abbreviations: FGF21, fibroblast growth factor 21; PPARα, peroxisome proliferators-activated receptor α; FRS-2, FGF receptor substrate-2; WHR, waist–hip ratio; BP, blood pressure; FFA, free fatty acid; AST, aspartate aminotransferase; ALT, alanine aminotransferase; HbA1c, hemoglobin A1c; HOMA-R, homeostasis model assessment for insulin resistance; ARB, angiotensin receptor blocker; ACE, angiotensin converting enzyme; IGT, impaired glucose tolerance; NGT, normal glucose tolerance.

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multiple response elements for peroxisome proliferators-activated receptor α (PPARα) are designated in the promoter region of the FGF21 gene [9,10]. Starvation or antihyperlipidemic drug fibrates can stimulate transcription of FGF21 gene through the action of PPARα. Biological effects of FGF21 are exerted through a cell membrane receptor complex composed of FGF receptors and βKlotho [11,12]. Tissue-specific expressions of βKlotho and particular FGF receptor isoforms seem to define metabolic activities of FGF19 and FGF21.

Thus far, the principal target tissue of FGF21 is thought to be adipocytes and it has been reported that FGF21 induced phosphorylation of FGF receptor substrate-2 (FRS-2) and activation of Ras/MAPK, thereby increasing glucose uptake in 3T3-L1 adipocytes and primary human adipocytes [5]. Administration of recombinant FGF21 reduced blood glucose and triglyceride levels. Moreover, transgenic mice that overexpressed FGF21 in the liver showed a decrease in hepatic fat deposit, an increase in insulin sensitivity, and resistance to diet-induced obesity [5]. FGF21 is supposed to enter brain through the blood–brain barrier [13], which might be relevant to a torpor induced by FGF21 [9,14].

Although molecular mechanisms by which FGF21 production and action are regulated have been progressively elucidated in cell biology and animal model experiments, basic clinical knowledge of FGF21 in physiology and pathophysiology of metabolic diseases like diabetes, obesity or metabolic syndrome, is lacking. In this study, we measured fasting serum concentrations of FGF21 and clarified how they were associated with clinical parameters in Japanese patients with type 2 diabetes, drawing comparison with those of adiponectin.

2. Materials and methods

2.1. Recruitment and eligibility

The Jichi Medical University human research ethics committees approved the study. Participants were recruited from patients with diabetes who were admitted to the Jichi Medical University Hospital. Written informed consents were obtained from all participants. Eligibility criteria for diabetes were based on the Japan Diabetes Association guidelines on diabetes case detection and diagnosis, and included individuals with diabetes who were 20–75 years. Those individuals having gestational diabetes, active hepatitis/liver cirrhosis, or chronic renal failure on hemodialysis were precluded from the study.

2.2. Anthropometric and biochemical measurements

Height (centimeters), weight (kilograms), waist and hip circumferences (centimeters) were measured. BMI was calculated as weight in kilograms divided by the square of height in meters. Waist circumference was measured using a nonelastic measuring tape at the midpoint between the lower border of the rib cage and iliac crest. Waist–hip ratio (WHR) was calculated as waist circumference divided by hip circumference. Blood pressure (BP) was measured in the morning before breakfast.

Serum glucose, triglyceride, total cholesterol, HDL-cholesterol, free fatty acids (FFAs), acetoacetic acid, β hydroxybutyric acid, total bilirubin, aspartate aminotransferase (AST), alanine aminotransferase (ALT), uric acid, creatinine levels were measured by standard enzymatic methods. LDL-cholesterol levels were calculated from the Friedewald equation. Insulin and glucagon levels were analyzed by immunoradiometric assays. FGF21 levels were analyzed by immunoenzymatic assays (BioVendor, Modrice, Czech Republic). Adiponectin levels were analyzed by immunoenzymatic assays (Otsuka Pharmaceutical, Tokyo, Japan). Hemoglobin A1c (HbA1c) levels were analyzed by an HPLC assay. Homeostasis model assessment for insulin resistance (HOMA-R) values were calculated as fasting glucose concentration (mg/dl) × fasting insulin concentration (μU/ml)/405.

2.3. Statistical analysis

Data were shown as the mean ± SD. The statistical analysis was done with a software Statcel2 (version 2, 2004, Saitama, Japan). Results of different groups were compared with Mann–Whitney's test. Multiple linear regression analyses were performed with serum FGF21 levels or serum adiponectin levels as the dependent variables. Because serum FGF21 levels were not normally distributed, they were logarithmically converted and used for correlation analyses. In all statistical tests, P values <0.05 were considered significant.

3. Results

For an initial screening, 143 patients with type 2 diabetes, 11 patients with type 1 diabetes, and 6 patents with pancreatic diabetes ($n = 160$, M/F = 97/63) were studied. Distribution of serum FGF21 concentrations in these subjects with diabetes was broad (7–6007 pg/ml, median value 264 pg/ml), and the average value was 495 ± 751 pg/ml (mean ± SD). Serum FGF21 levels were similar between male and female subjects (437 ± 589 pg/ml vs. 585 ± 946 pg/ml, $P = 0.32$). Serum FGF21 levels were also similar among the three subtypes of diabetes (type 1, 483 ± 634 pg/ml; type 2, 497 ± 766 pg/ml; and pancreatic, 469 ± 671 pg/ml; $P = 1.00$). Serum FGF21 levels were highly correlated with serum creatinine levels ($r = 0.27$, $P = 0.00073$).

Next, we restricted study subjects to patients with type 2 diabetes whose serum creatinine levels were below 2.5 mg/dl ($n = 139$, M/F = 85/54). Main characteristics of these subjects are listed in Table 1. There was no difference in serum FGF21 levels between male and female subjects (370 ± 487 pg/ml vs. 588 ± 989 pg/ml, $P = 0.11$). In univariate correlation analyses (Table 2), serum FGF21 levels were most strongly associated with a use of fibrates ($r = 0.342$, $P = 0.000059$). Serum FGF21 levels in subjects taking fibrates were 6.0-times higher than those in subjects not taking fibrates (2188 ± 1885 pg/ml vs. 363 ± 477 pg/ml, $P = 0.000059$). The second strongest association was observed with serum triglyceride levels ($r = 0.317$, $P = 0.00019$) (Fig. 1). Exclusion of subjects taking fibrates from the analysis maintained a significant association between FGF21 and triglyceride levels ($r = 0.257$, $P = 0.0032$). Serum creatinine levels still showed a strong correlation with FGF21 levels ($r = 0.238$, $P = 0.0053$) after setting a cut-off level for creatinine below 2.5 mg/dl (Fig. 2). The other factors that showed significant correlations with FGF21 levels included