Clinical Heterogeneity in Monogenic Diabetes Caused by Mutations in the Glucokinase Gene (GCK-MODY)

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OBJECTIVE — To evaluate the heterogeneity in the clinical expression in a family with glucokinase mature-onset diabetes of the young (GCK-MODY).

RESEARCH DESIGN AND METHODS — Members (three generations) of the same family presented either with overt neonatal hyperglycemia, marked postprandial hyperglycemia, or glucosuria. Homeostasis model assessment of insulin resistance ($HOMA_{IR}$) and insulinogenic and disposition indexes were calculated. Oral glucose tolerance test (OGTT) results in the *GCK* mutation carriers from this family were compared with those from other subjects with *GCK* mutations in the same codon (GCK_{261}), with other missense and other types of *GCK* mutations in different codons from the European MODY Consortium database (GCK_m).

RESULTS — Mutation G261R was found in the *GCK* gene. During the OGTT, glucose ($P = 0.02$) and insulin ($P = 0.009$) response at 2 h as well as at the 2-h glucose increment (GCK_{261} versus other missense *GCK* mutations, $P = 0.003$) were significantly higher in GCK_{261} than in GCK_m carriers.

CONCLUSIONS — Differing from other GCK_m carriers, the glucose and insulin response to oral glucose was significantly higher in GCK_{261} carriers, indicating clinical heterogeneity in GCK-MODY.

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I nactivating heterozygous mutations in the glucokinase gene (*GCK*) cause a form of monogenic diabetes with autosomal dominant inheritance (GCK mature-onset diabetes of the young [MODY]) (1,2). GCK-MODY has generally been considered a phenotypically homogenous mild form of diabetes, which does not lead to marked hyperglycemia or diabetes complications and does not need treatment (2–5).

Phenotypic heterogeneity within carriers of the same *GCK* mutation has been observed only in one family (6). Here, we report a new GCK-MODY (GCK-G261R) family characterized by marked prandial hyperglycemia and unusual high levels of postprandial insulinemia.

RESEARCH DESIGN AND METHODS — The proband (online appendix Fig. A1, available at http://care.diabetesjournals.org/cgi/content/full/dc09-0681/DC1) was a firstborn child from a Finnish family with neonatal plasma glucose of 10 mmol/l. At 2 years of age, without treatment, she presented preprandial and postprandial capillary glucose of 6.5–6.8 and 8.6 mmol/l, respectively. Her younger sister had random glucose between 7 and 11.5 mmol/l as a neonate. Their mother was diagnosed with gestational diabetes and treated with insulin. After the pregnancy, she had an A1C of 5.8% without insulin treatment, but due to high postprandial plasma glucose (10–11 mmol/l), rapid-acting mealtime insulin was started. Since the second pregnancy, she is treated with diet alone. The maternal grandmother presented with hyperglycemia and glucosuria at age of 22 years and gestational diabetes during four pregnancies. She was treated with diet during the first pregnancy and with insulin during three later pregnancies, after which she had been without treatment. Her fasting capillary glucose level was normally ~6–7 mmol/l but stayed at ~10 mmol/l for nearly 2 weeks after intake of larger quantities of carbohydrates and returned to 6–7 mmol/l when carbohydrates were restricted. She takes 60 mg of nateglinide before meals. All available family members were offered an oral glucose tolerance test (OGTT) and/or genetic testing for the mutation after genetic counseling.

OGTT (except subjects <15 years) with samples drawn at −5, 0, 30, 60, 90, and 120 min was performed to determine plasma glucose and serum insulin. Insulin resistance and β-cell function was estimated using the homeostasis model assessment of insulin resistance ($HOMA_{IR}$) and the insulinogenic indexes (IG30), respectively. The disposition index (DI) was used to assess β-cell compensation. These results and those from 15 subjects with *GCK* mutations in position 261 (GCK_{261}) from the European MODY Consortium Database (EMCD) (3) were compared with that of carriers of other missense and other types of GCK

Table 1—*Clinical characteristics of patients with glucokinase inactivating mutation GCK_{261} and functional studies of recombinant human wild-type and mutants' gk*

	GCK_{261}: mutations	P_1	Other missense: GCK mutations	P_2	Other *GCK*: mutation types	P_3	Normal glucose tolerance: control subjects	P_4
n (male/female)	23 (13/10)		144 (73/71)		82 (42/40)		45 (20/25)	
BMI (kg/m²)	21.80 (7.0)	NS	20.00 (5.42)	NS	21.30 (5.73)	NS	23.7 (6.2)	NS
Age (years)	20.00 (27.0)	NS	19.00 (27.00)	NS	29.00 (28)	NS	41.6 (31)	0.001
0-min plasma glucose (mmol/l)	7.00 (0.69)	NS	6.70 (0.90)	NS	6.80 (1.01)	0.17	5.0 (0.7)	NS
120-min plasma glucose (mmol/l)	10.90 (4.13)	0.02	8.60 (2.58)	0.046	8.60 (2.98)	0.38	5.5 (1.8)	NS
2hΔPG (mmol/l)	4.12 (3.25)	0.003	2.00 (2.05)	0.046	2.50 (2.20)	0.233	0.6 (1.8)	0.004
0-min plasma insulin (mU/l)	10.00 (7.44)	NS	8.00 (6.00)	NS	9.00 (5.00)	0.41	7.7 (7.7)	NS
120-min plasma insulin (mU/l)	55.2 (28.05)	0.009	25.00 (23.50)	0.002	24.00 (13.50)	0.11	30.2 (27.4)	0.03
Incremental I/G30	8.71 (7.46)	0.106	6.09 (3.74)	0.513	4.21 (5.55)	0.672	23.7 (25.4)	0.003
HOMA$_{IR}$	3.47 (2.31)	0.568	2.68 (2.45)	0.318	2.21 (1.92)	0.259	1.7 (1.4)	0.045
DI	3.23 (2.25)	NS	2.64 (2.02)	NS	1.98 (2.18)	0.76	15.5 (13.5)	0.004

Proteins studied	Glucose $S_{0.5}$ (mmol/l)	Hill number (unit less)	ATP$_{Km}$ (mmol/l)	Turnover rate (Kcat) (sec^{-1})	Activity index (AI)	T-GSIR (mmol/l)
gk-WT	7.55 ± 0.23	1.74 ± 0.04	0.41 ± 0.03	62.3 ± 4.75	1.45 ± 0.11	5
gk-G261R	68.61 ± 16.15	1.53 ± 0.11	0.63 ± 0.10	17.03 ± 4.11	0.04 ± 0.001	7
gk-G261E	334.73 ± 26.78	1.92 ± 0.06	2.99 ± 0.37	3.72 ± 0.32	0.00	7

Upper panel: data are median (interquartile range). Clinical characteristics, glucose, and insulin values of patients with GCK-inactivating mutation GCK_{261}, other missense *GCK*-inactivating mutations, and other types of *GCK*-inactivating mutations (insertions, deletions, frame shifts, etc.). For insulin data: $n = 11$ (GCK_{261}), 36 (other missense *GCK* mutations), 45 (other types of *GCK* mutations), and 45 (normal glucose tolerant controls); P_1: GCK_{261} mutations vs. other missense *GCK* mutations; P_2: GCK_{261} mutations vs. other types of *GCK* mutations; P_3: other missense *GCK* mutations vs. other types of *GCK* mutations; P_4: GCK_{261} mutations vs. control subjects. $P < 0.05$–0.00001 are considered statistically significant. HOMA$_{IR}$ = fasting serum insulin × fasting serum glucose/22.5. Insulinogenic index (I/G30) = serum insulin at 30 min − serum insulin at 0 min/serum glucose at 30 min − serum glucose at 0 min. DI = insulinogenic index/HOMA$_{IR}$. *Lower panel*: data are means of the three independent analyses. Results of the functional studies of gk-WT and mutants gk-G261R and gk-G261E. AI, the activity index for the enzyme was calculated as previously described (8). Glucose $S_{0.5}$ of gk-G261R mutations was carried out with 25 mmol/l of MgATP, and the ATP$_{Km}$ measurement was performed at glucose concentration of 500 mmol/l. T-GSIR, threshold for glucose-stimulated insulin secretion.

mutations (insertions, deletions, etc.) (GCK_m) and normoglycemic control subjects from the Botnia Study (Table 1). The studies were approved by the institutional ethics committee. Written consent was obtained from the adults and from the parents of the children. DNA extraction, microsatellite genotyping, direct sequencing, and functional analysis of the glucokinase protein (gk), with and without gk activator, were performed as described (7–10).

RESULTS — The mutation G261R (exon 7) on *GCK* was found in the proband and in nine family members (aged 0.2–72 years) with abnormal fasting glucose. Fasting plasma glucose ranged from 6.0 to 7.6 mmol/l. The 2-h plasma glucose ranged from 9.3 to 14.5 mmol/l in the carriers, three of them presented values exceeding 13 mmol/l. All but one of the carriers had a 2-h increment in plasma glucose (2hΔPG) higher than 3 mmol/l and half higher than 6 mmol/l. Fasting insulin was 4.1–9.9 mU/l and 2-h insulin 28.8–61.9 mU/l. There was no relationship between age and glucose or insulin concentrations.

The GCK_{261} mutation carriers from our family, like those from the EMCD, had a significantly higher glucose and insulin response compared with GCK_m carriers (Table 1). Fasting plasma glucose and insulin were similar in all groups; however, the 2-h plasma glucose and insulin and 2hΔPG values were significantly higher in GCK_{261} carriers than in GCK_m carriers (Table 1). The glucose response during OGTT was higher at all time points in GCK_{261} carriers compared also with GCK_m carriers (data not shown). In 61 and 35% of GCK_{261} carriers, 2hΔPG was >3 and 4.6 mmol/l, respectively. HOMA$_{IR}$, I/G30, and DI values were higher (not significant) in the GCK_{261} carriers (Table 1), indicating possibly higher degree of β-cell compensation (Table 1).

The results from the functional studies showed that the mutations gk-G261R/E lead to a severely effected protein, with an almost negligible enzyme activity, indicating that these gk mutants cannot contribute to β-cell and hepatic glucose phosphorylation. The effect of the gk activator on the inactivating gk-G261R mutation was similar to that on the gk-WT (see online appendix Table A1).

CONCLUSIONS — The clinical phenotype of carriers from our family was heterogeneous. The proband and her sister presented with neonatal hyperglycemia, their mother with gestational diabetes, and the maternal grandmother with glucosuria. Many carriers had much higher 2hΔPG values than what is usually seen in GCK-MODY. In three carriers (one child and two adults), it exceeded 13 mmol/l and in another young carrier 12 mmol/l, indicating no relationship between high 2hΔPG values and age. A sim-

ilar pattern was seen in other carriers of the same mutation, while those with other *GCK* mutations in the MODY database had a lower glucose response during OGTT. Of note, a similar pattern of glucose response as in GCK_{261} carriers has previously been observed in GCK-L184P carriers (6). However, while the insulin response was attenuated in GCK-L184P carriers, in GCK_{261} carriers it was high and significantly different from that seen with other *GCK* mutations (11). Glucokinase is required for glycogen synthesis in liver (12). One explanation for the high 2-h glucose in GCK_{261} carriers could be reduced hepatic glycogen synthesis due to the lost of activity of GCK_{261}. Hence, the marked insulin response could be due to larger β-cell compensation in GCK_{261} carriers. Nonetheless, possible additional genetic defects could be also involved.

In summary, the clinical phenotype of patients with GCK-MODY can be heterogeneous and patients carrying severe inactivating *GCK* mutations can have high postchallenge glucose values, possibly resulting from a marked liver component of the disease.

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