

# Familial Early-Onset Type 2 Diabetes in Chinese Patients

Obesity and genetics have more significant roles than autoimmunity

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**OBJECTIVE** — We examined the prevalence of different forms of diabetes in Hong Kong Chinese patients with familial early-onset type 2 diabetes and compared their clinical features with patients with familial late-onset type 2 diabetes.

**RESEARCH DESIGN AND METHODS** — A total of 145 young patients with early-onset diabetes (age and age at diagnosis  $\leq 40$  years) and a family history of diabetes were studied. They were screened for mutations in the genes encoding glucokinase, hepatocyte nuclear factor (HNF)-4 $\alpha$ , and HNF-1 $\alpha$ . The mitochondrial DNA A $\rightarrow$ G at nucleotide 3243 (mt3243) and amylin S20G mutations were studied, and antibodies to GAD (anti-GADs) were also examined.

**RESULTS** — The prevalence of putative diabetogenic gene mutations and autoimmune markers were 4% for glucokinase, 0% for HNF-4 $\alpha$ , 5% for HNF-1 $\alpha$ , 3% for mt3243, 2% for amylin S20G, and 4% for anti-GAD. Compared with late-onset patients, the patients with early-onset diabetes had a higher prevalence of a parental history of diabetes and were generally more obese. When classified by obesity indexes (BMI and waist circumference), the obese patients, especially those with early-onset diabetes, had a clustering of cardiovascular risk factors and increased rates of retinopathy and albuminuria.

**CONCLUSIONS** — Genetic factors (up to 14%) and obesity (55%) play more significant roles than autoimmunity (4%) in familial type 2 diabetes in young Chinese patients. The significance of obesity-related genes and other gene-gene and gene-environment interactions in these young patients remains to be determined.

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The prevalence of diabetes mellitus is reaching epidemic proportions in the Hong Kong Chinese population (1). The age-adjusted population prevalence of diabetes has increased from 7.7%

in 1990 (2) to 8.9% in 1995 (3). The crude prevalence is also increasing among young people, from  $<1\%$  in those aged  $<30$  years in 1990 (2) to 1.7% in those aged 25–34 years in 1995 (4). In con-

trast to Caucasian populations, classical type 1 diabetes characterized by acute symptoms with heavy ketonuria or ketoacidosis is quite uncommon in Chinese populations. In our previous clinic-based study, type 1 diabetes was found in only 10% of the early-onset ( $<35$  years) patients and 3% of the late-onset patients. Despite having similar duration of disease, 50% of the early-onset patients and 25% of the late-onset patients required insulin therapy (5). Moreover, among young diabetic patients, 50% were obese and 50% were insulin deficient, as assessed by postglucagon plasma C-peptide levels (6). These data point to the clinical and etiological heterogeneity of diabetes in both young and late-onset patients.

Several genes that may lead to early-onset of disease under gene-gene and gene-environmental influences have now been found to be associated with diabetes. These include the five genes responsible for maturity-onset diabetes of the young (MODY), which are glucokinase (MODY2), hepatic nuclear factor (HNF)-4 $\alpha$  (MODY1), HNF-1 $\alpha$  (MODY3), HNF-1 $\beta$  (MODY5), and insulin promoter factor-1 (MODY4). Furthermore, an A3243G mutation in the mitochondrial DNA coding for tRNA<sup>eu(UUR)</sup> (mt3243) has been associated with diabetes characterized by maternal inheritance and deafness (7). Amylin is a peptide cosecreted with insulin from the pancreatic  $\beta$ -cells, and has been shown to cause  $\beta$ -cell toxicity and diabetes (8). A S20G variant of this gene has been demonstrated to enhance cytotoxicity in transfected COS-1 cells and enhance amyloidogenicity in vitro (9). The variant was also associated with development of type 2 diabetes in Japanese subjects (10). Although a similar study in Taiwanese Chinese subjects did not show cosegregation of the variant allele with glucose intolerance, normoglycemic-variant allele carriers had a mild reduction in early phase insulin secretion (11). This suggests that this variant may interact with other major genes in the pathogenesis of diabetes.

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**Abbreviations:** ACR, albumin-to-creatinine ratio; anti-GAD, antibody to GAD; BP, blood pressure; HDL-C, HDL cholesterol; LDL-C, LDL cholesterol; HOMA<sub>IR</sub>, homeostasis model assessment for insulin resistance; HNF, hepatocyte nuclear factor; MODY, maturity-onset diabetes of the young; PCR, polymerase chain reaction; PWH, Prince of Wales Hospital; RFLP, restriction fragment-length polymorphism; TC, total cholesterol; TG, triglyceride.

A table elsewhere in this issue shows conventional and Système International (SI) units and conversion factors for many substances.

Table 1—Mutations in the HNF-1 $\alpha$  and glucokinase genes in Chinese subjects with early-onset diabetes

Subject	Location	Codon/nt	Nucleotide change	Designation
HNF-1 $\alpha$ mutation				
HK90*, YDM42†	Exon 1	20	GGG (Gly)→AGG (Arg)	G20R
YDM20†	Exon 2	116	GCG (Ala)→GTG (Val)	A116V
HK10*	Intron 2/Exon 3	nt-1	AG→AA at splice acceptor site	IVS2nt-1G→A
HK54*	Exon 3	203	CGT (Arg)→CAT (His)	R203H
HK30*	Exon 6	432	TCC (Ser)→TGC (Cys)	S432C
HK92*	Exon 10	618	ATC (Ile)→ATG (Met)	I618M
Glucokinase mutation				
YDM142†	Exon 3	101	GTG (Val)→ATG (Met)	V101M
HK84*	Exon 3	110	ATC (Ile)→ACC (Thr)	I110T
HK38*	Exon 3	119	GCT (Ala)→GAT (Asp)	A119D
YDM67†, YDM144†	Exon 7	239	CAG (Gln)→CGG (Arg)	Q239R
HK15*	Exon 9	385	GGG (Gly)→GTG (Val)	G385V

\*Reported in previous studies (25, 47). †Newly found in the present study.

Antibodies to GAD (anti-GADs) are sensitive markers of type 1 diabetes in Caucasians (12), and they can be detected in some cases of late-onset type 2 diabetes with less acute presentations (13). In Asians, anti-GAD has been associated with an acute and early-onset form of diabetes with a prevalence ranging from 10 to 50%, depending on the selection criteria and assay methodologies (6,14–16).

Clinic and population-based studies reveal that ~17% of the diabetic patients in Hong Kong are diagnosed between the ages of 25 and 45 years (5,17). The increasing trend of obesity among Hong Kong children coincides with a rise in the incidence of early-onset diabetes (18). Because of their anticipated long duration of disease, it is important to classify and characterize the nature of diabetes in these young patients to facilitate early diagnosis and appropriate treatments. In this study, we have assessed the prevalence of known molecular defects, or autoimmunity as judged by detection of anti-GADs, in two separate cohorts of young Chinese patients with familial type 2 diabetes. We also compared their clinical characteristics with patients with familial late-onset diabetes.

## RESEARCH DESIGN AND METHODS

The Prince of Wales Hospital (PWH) is a regional teaching hospital in Hong Kong. Its catchment area has a population of 1.2 million, accounting for 20% of the total population in Hong Kong. Because of the lack of long-term health care programs in

Hong Kong, coverage by medical insurance is not widely available. Many patients with chronic diseases like diabetes are managed in public hospitals or clinics where they only pay a nominal fee. Hence, except for high social classes, our patients are largely representative of the diabetic population in Hong Kong. Since 1995, all patients attending the diabetes clinic of the PWH have been entered into the PWH Diabetes Registry after undergoing a structured assessment (19,20). During 1995 and 1996, a separate cohort of 150 young patients with early-onset diabetes (age  $\leq$ 40 years and age at diagnosis  $\leq$ 35 years) who underwent the structured assessment were recruited consecutively from the diabetes clinics at the PWH to form the Young Chinese Diabetes Database (6).

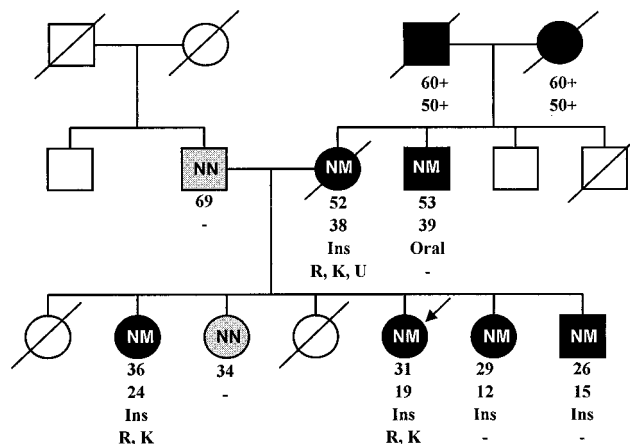
Among the 1,800 patients in the PWH Diabetes Registry recruited between 1995 and 1997 and the 150 patients in the Young Chinese Diabetes Database, 92 and 53 patients, respectively, were selected for the present study because they satisfied the enrollment criteria, which were early-onset (age and age at diagnosis  $\leq$ 40 years) type 2 diabetes (1985 WHO criteria) (21) and a positive family history (at least one first degree relative with diabetes). Patients with classic type 1 diabetes (acute ketotic presentation or continuous requirement of insulin within 1 year of diagnosis) were excluded.

We have previously reported the prevalence of anti-GAD (6), mt3243 (22,23), and amylin gene mutations (24) among patients from the Young Chinese Diabetes Database. We have also reported

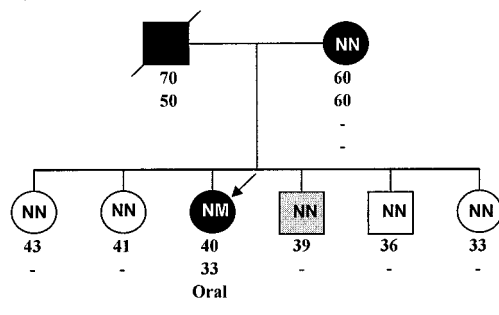
the prevalence of mt3243 (23), amylin (24), glucokinase, HNF-1 $\alpha$ , and HNF-4 $\alpha$  gene mutations (25) in a separate cohort from the PWH Diabetes Registry. In this study, screening for glucokinase and HNF-1 $\alpha$  gene mutations was extended to the 53 patients from the Young Chinese Diabetes Database. The HNF-4 $\alpha$  gene was not screened in this cohort because of the expected low frequency of mutations; none was found in the 92 patients from the PWH Diabetes Registry (25). Screening for anti-GAD was extended to the 92 patients from the PWH Diabetes Registry.

Of these 145 young patients with familial diabetes, 19 (13%) met the minimal criteria for MODY (age at diagnosis  $\leq$ 25 years and presence of diabetes in two consecutive generations). Altogether, 10 of 20 families with probands carrying putative diabetogenic gene mutations were recruited for a 75-g oral glucose tolerance test and clinical assessment. The 1999 WHO classification was used to define the glycemic status of the family members (26). For comparison of clinical characteristics of the early-onset patients, 290 sex-matched patients with late-onset diabetes (age at diagnosis  $>$ 40 years) and a family history of diabetes were randomly selected from the current 1,800 patients in the PWH Diabetes Registry. A total of 100 healthy Chinese individuals (age  $33 \pm 10$  years, 40 men and 60 women) were selected as control subjects from hospital staff and students for screening for the gene variants identified in our patients. Informed consent was obtained from each subject for a blood sample to be

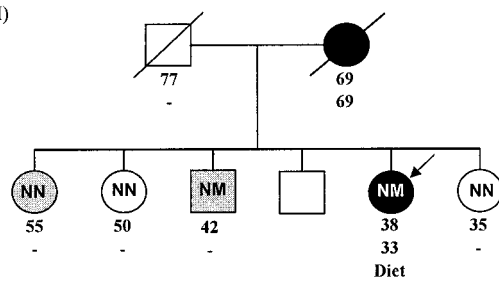
**HK10 \***  
(HNF-1 $\alpha$ , IVS2nt-1G→A)



**HK54 \***  
(HNF-1 $\alpha$ , R203H)



**YDM142**  
(GCK, V101M)



**Figure 1**—Pedigrees of 10 families carrying the HNF-1 $\alpha$ , glucokinase, mt3243, or amylin S20G gene mutations/polymorphisms. Subjects with diabetes are noted by black symbols, subjects with IFG or IGT by gray symbols, and nondiabetic and untested subjects by open symbols. The genotype of the family members is indicated by N (wild-type allele) and M (mutant/variant allele). Present age, age at diagnosis, therapy, and complications are stated in this order. The proband is indicated by an arrow. Oral, oral drugs; Ins, insulin; R, retinopathy; K, albuminuria; U, neuropathy; H, hearing impairment. \*Data were previously reported (22, 23, 25, 47).

taken for DNA extraction and measurement of biochemical indexes. This study was approved by the Clinical Research Ethics Committee of the Chinese University of Hong Kong.

**Clinical studies**

All patients underwent a structured assessment based on the Europe DiabCare Protocol (19). Their family history of diabetes, age at diagnosis, and anthropomet-

ric indexes were documented (19,20). BMI was used as an index of general obesity. Waist circumference, which is highly correlated with visceral fat accumulation measured by magnetic resonance imaging in Chinese patients (27), was used as an index of central obesity. After an overnight fast, venous blood was sampled for measurement of plasma glucose, insulin, HbA<sub>1c</sub>, total cholesterol (TC), HDL cholesterol (HDL-C), LDL cholesterol (LDL-C) (calculated), triglyceride (TG), and anti-GAD. A morning spot urine sample was collected for assessment of albuminuria. Retinopathy and sensory neuropathy was assessed as previously described (28).

General obesity was defined as a BMI  $\geq 25$  kg/m<sup>2</sup> using the recent Asian criteria (29). Albuminuria was defined as an albumin-to-creatinine ratio (ACR)  $\geq 3.5$  mg/mmol in a spot urine sample (30). The homeostasis model assessment for insulin resistance (HOMA<sub>IR</sub>) index (fasting plasma insulin  $\times$  glucose/22.5), derived from the HOMA equation, was used to assess insulin resistance (31).

**Biochemical assays**

Plasma glucose, HbA<sub>1c</sub>, lipids, urinary albumin, and creatinine were measured by routine assays in the Department of Chemical Pathology at the PWH as previously described (32). Plasma insulin was measured in non-insulin-treated patients by a radioimmunoassay (Pharmacia, Uppsala, Sweden) with intra-assay and interassay coefficients of variations of 6 and 13.8%, respectively. Anti-GAD was measured by a radioimmunoprecipitation assay (33). The upper normal limit of 18 units is applicable to Asian and European subjects (15,33).

**Genetic analysis**

The minimal promoter regions and exons of the glucokinase ( $\beta$ -cell form), HNF-1 $\alpha$ , and HNF-4 $\alpha$  (HNF-4 $\alpha$ 2 form) genes were screened for mutations by direct sequencing of polymerase chain reaction (PCR) products as described previously (34–36). The occurrence of putative mutations in other family members and control subjects was determined by PCR-restriction fragment-length polymorphism (RFLP). Mt3243A→G and amylin gene S20G mutations were determined by PCR-RFLP as described (10,22).

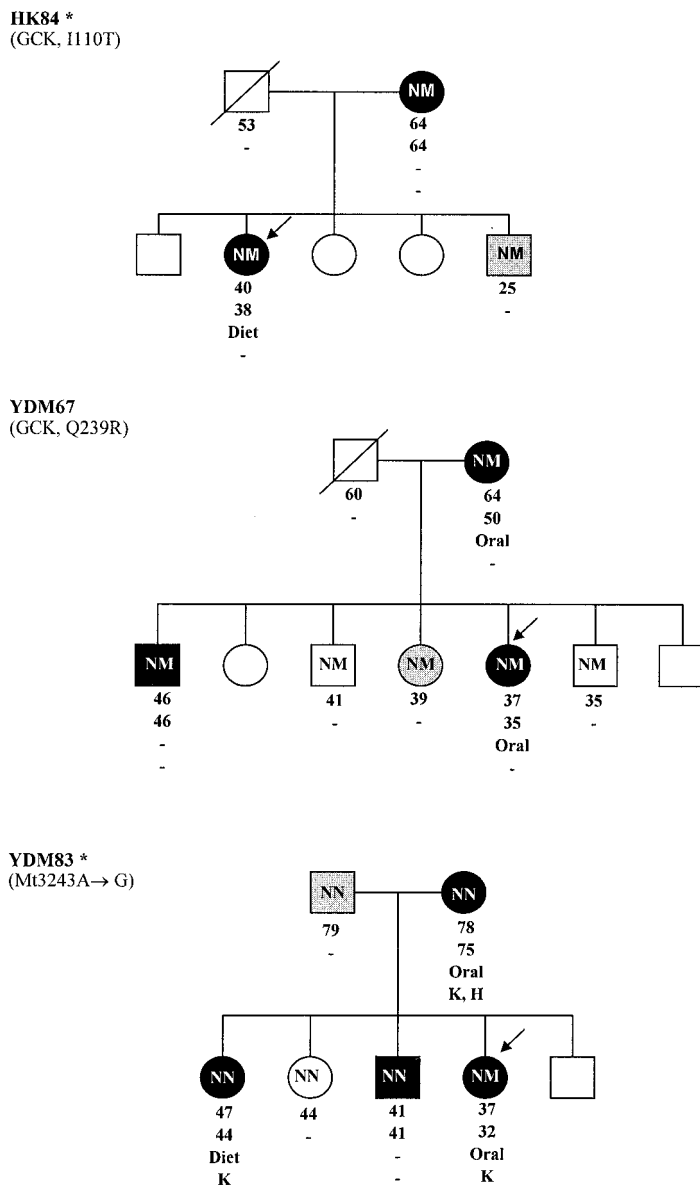


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**Statistical analysis**

Normally distributed data are expressed as means ± SD. Data with skewed distributions were normalized by logarithmic transformation. The resultant means were antilogarithmically transformed and expressed as geometric means together with 25th and 75th percentiles.  $\chi^2$  tests and Student's unpaired *t* tests were used for between-group comparisons. A *P* value <0.05 (2-tailed) was considered to be significant. All statistical analyses were performed using the Statistical Package for Social Sciences (SPSS for Windows, version 9.0).

**RESULTS**

**Prevalence of putative gene mutations and anti-GAD in patients with familial early-onset diabetes**

Among the 145 patients with familial early-onset diabetes, there were 20 (14%) with putative mutations; of these, 7 (5%) involved the HNF-1 $\alpha$  gene, 6 (4%) involved the glucokinase gene, 4 (3%) involved mt3243, and 3 (2%) involved amylin S20G. Anti-GAD was positive in six (4%). No mutation in the HNF-4 $\alpha$  gene was found in the 92 patients from the PWH Diabetes Registry. All mutations

identified in the HNF-1 $\alpha$  and glucokinase genes were novel (Table 1). The HNF-1 $\alpha$  G20R and glucokinase Q239R mutations were found in four unrelated patients. None of these mutations were found in 100 healthy control subjects.

**Family cosegregation study of gene mutations**

Among the 20 patients carrying putative gene mutations, 10 had families who were recruited for cosegregation study (Fig. 1). Cosegregation of a mutation with clinical diabetes or glucose intolerance were observed in the families of the following four patients: HK10 with HNF-1 $\alpha$  IVS2nt-1G→A, YDM142 with glucokinase V101 mol/l, HK84 with glucokinase I110T, and HK50 with mt3243. Segregation was inconclusive in the other six families. For the families of three patients (HK54 with HNF-1 $\alpha$  R203H, YDM83 with mt3243, and CX216 with amylin S20G mutations), the probands carried the gene mutations, but none of the diabetic or nondiabetic family members who presented for screening carried the mutations. For the families of three other patients (YDM67 with glucokinase Q239R, HK61 with mt3243, and YDM99 with amylin S20G mutations), all mutation carriers from the families of YDM142 and HK84 had higher fasting plasma glucose concentrations (5.8–8.9 mmol/l) than those with no mutation (4.2–5.3 mmol/l). On the other hand, the four mutation-carrying siblings of proband YDM67 had normal fasting plasma glucose concentrations (4.0–5.6 mmol/l), irrespective of their glycemic status.

**Clinical characteristics of patients with familial early-onset diabetes of unknown cause compared with familial late-onset diabetes**

Although 26 of the patients with early-onset diabetes carried putative gene mutations or the autoimmune marker, the causes of diabetes in the other 119 patients remain to be determined. These young patients with diabetes of unknown cause (age at diagnosis 30 ± 6 years) differed clinically from the 290 late-onset patients (age at diagnosis 52 ± 8 years) (Table 2). Thus, despite a positive family history of diabetes in all patients in both

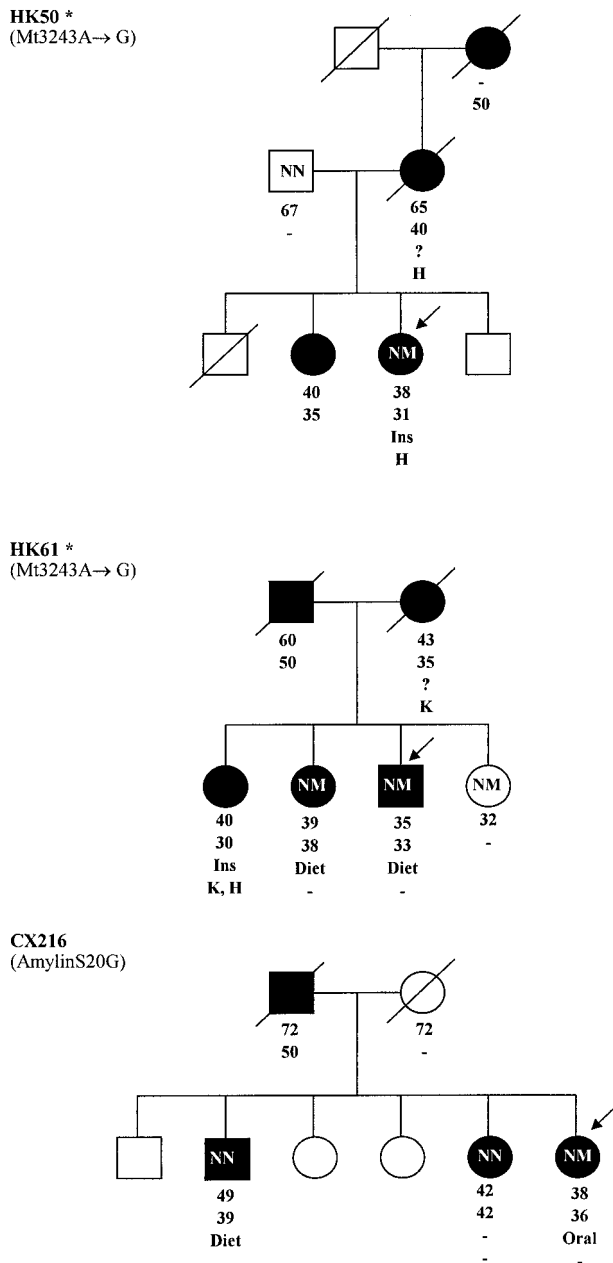


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groups, those with early-onset diabetes more frequently had a father with diabetes (39 vs. 22%) and a mother with diabetes (63 vs. 41%), but less frequently a sibling with diabetes (30 vs. 53%) ( $P < 0.001$ ). The early-onset patients had a higher BMI but lower blood pressure (BP) and lower prevalence of retinopathy and neuropathy than the late-onset patients. The early-onset patients had better glycemic control (glucose and HbA<sub>1c</sub>) as well as higher fasting insulin concentrations than the late-onset patients. Notwith-

standing a similar mean disease duration of only 4 years, both the early- and late-onset patients had a disproportionately high prevalence of albuminuria, 40 and 38%, respectively, compared with the prevalence rates of other microangiopathic complications. Insulin resistance, as assessed by the HOMA<sub>IR</sub> index, was similar between the two groups of non-insulin-treated patients. The proportion of patients treated with insulin was similar in both groups (8 vs. 7%), but fewer patients with early-onset diabetes were

treated with oral drugs (33 vs. 61%,  $P < 0.001$ ) when compared with the late-onset group.

**Clinical characteristics of the patients with familial early-onset diabetes of unknown cause and familial late-onset diabetes classified according to obesity index**

Because of the high prevalence of general obesity in both early-onset patients of unknown cause and late-onset patients (55 and 46%, respectively), we further analyzed the association of obesity with cardiovascular risk factors and complications in these patients (Table 2). Among the early-onset patients, the obese patients had worse glycemic control (HbA<sub>1c</sub>) as well as higher systolic BP levels, a more adverse lipid profile (higher TG, lower HDL-C, and higher TC/HDL-C), and higher fasting insulin than the nonobese patients. They were also more insulin-resistant (HOMA<sub>IR</sub> index) and had a higher prevalence of retinopathy and albuminuria than the nonobese patients. Among the late-onset patients, the obese patients had better glycemic control (glucose and HbA<sub>1c</sub>) than the nonobese patients. However, they had a higher systolic and diastolic BP, and a higher fasting insulin than the nonobese patients. The degree of insulin resistance and prevalence of complications were similar in the two groups.

**CONCLUSIONS**— Since 1990, our group has documented the high prevalence of diabetes and the pattern of this disease in the Hong Kong Chinese population (1,5,32,37). Using carefully phenotyped patient cohorts, we have ascertained the high prevalence of young-onset diabetes and its phenotypic heterogeneity, with obesity and family history as important features. In agreement with our previous studies (6,16,38) and other studies from Asia (14,39), only 6 (4%) of 145 young patients with familial diabetes with type 2 presentation were positive for anti-GAD, indicative of the rarity of the autoimmune form of diabetes in Chinese populations.

Our present study further demonstrates that young patients with familial early-onset diabetes are genetically heterogeneous. Up to 14% of patients might carry mutations in one of the diabetes-associated genes. Each of the novel mutations identified in the HNF-1 $\alpha$  and glucokinase genes alter conserved amino

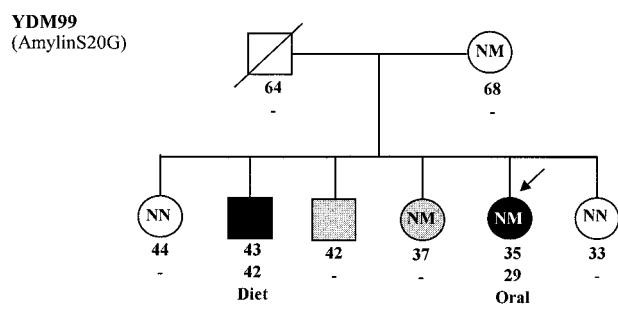


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acids in different species (35,40), suggesting that they might have functional signif-

icance. Two such mutations, HNF-1 $\alpha$  G20R and glucokinase Q239R, were

found in four unrelated patients, but haplotype analysis, which tests for a common founder effect in these mutations, was not possible because only one of the families was accessible.

Although putative genetic or autoimmune causes of diabetes were identified in nearly 20% of our familial early-onset patients, a phenotypic characterization of different types of diabetes was not feasible because of the small number of families being identified and the uncertainty of the diabetogenic role of each mutation. In this respect, despite an in vitro study demonstrating a pathogenic role of amylin S20G

Table 2—Comparison of clinical features of Chinese patients with familial type 2 diabetes according to age of diagnosis of diabetes and obesity status

Characteristics	Early-onset patients with unknown etiology	Late-onset patients	Early-onset nonobese patients	Early-onset obese patients	Late-onset nonobese patients	Late-onset obese patients
n	119	290	54	65	156	134
Sex (%)						
Men	37 (31)	98 (34)	12 (22)	25 (38)	56 (36)	42 (31)
Women	82 (69)	192 (66)	42 (78)	40 (62)	100 (64)	92 (69)
Current age (year)	34 $\pm$ 5	56 $\pm$ 9 $\ddagger$	34 $\pm$ 5	33 $\pm$ 5	56 $\pm$ 10	55 $\pm$ 9
Age at diagnosis (year)	30 $\pm$ 6	52 $\pm$ 8 $\ddagger$	31 $\pm$ 5	29 $\pm$ 6	52 $\pm$ 8	52 $\pm$ 8
Duration of disease (year)	4.0 $\pm$ 3.9	4.0 $\pm$ 4.2	3.9 $\pm$ 3.8	4.2 $\pm$ 4.0	4.5 $\pm$ 4.4	3.5 $\pm$ 3.9 $\ddagger$
Family history						
Father	46 (39)	64 (22) $\ddagger$	21 (39)	27 (42)	28 (18)	36 (27)
Mother	75 (63)	119 (41) $\ddagger$	38 (70)	39 (60)	65 (42)	54 (40)
Sibling	36 (30)	154 (53) $\ddagger$	15 (28)	20 (31)	85 (54)	69 (51)
BMI (kg/m <sup>2</sup> )	26.2 $\pm$ 4.7	25.0 $\pm$ 3.7 $\ddagger$	22.3 $\pm$ 1.8	29.5 $\pm$ 3.8 $\ddagger$	22.4 $\pm$ 1.8	28.0 $\pm$ 3.1 $\ddagger$
Waist circumference (cm)						
Men	90 $\pm$ 11	87 $\pm$ 9	78 $\pm$ 6	95 $\pm$ 9 $\ddagger$	81 $\pm$ 6	94 $\pm$ 7 $\ddagger$
Women	81 $\pm$ 11	83 $\pm$ 9	74 $\pm$ 5	89 $\pm$ 10 $\ddagger$	78 $\pm$ 6	89 $\pm$ 8 $\ddagger$
Systolic BP (mmHg)	117 $\pm$ 14	136 $\pm$ 22 $\ddagger$	114 $\pm$ 13	120 $\pm$ 14 $\ddagger$	134 $\pm$ 23	139 $\pm$ 21 $\ddagger$
Diastolic BP (mmHg)	75 $\pm$ 9	83 $\pm$ 11 $\ddagger$	74 $\pm$ 9	77 $\pm$ 10	80 $\pm$ 11	86 $\pm$ 12 $\ddagger$
TG (mmol/l)	1.4 (0.9–2.0)	1.4 (1.0–2.0)	1.0 (0.7–1.5)	1.7 (1.0–2.4) $\ddagger$	1.4 (0.9–1.9)	1.6 (1.1–2.1)
TC (mmol/l)	5.3 $\pm$ 1.2	5.6 $\pm$ 1.3	5.1 $\pm$ 1.0	5.4 $\pm$ 1.4	5.6 $\pm$ 1.3	5.5 $\pm$ 1.2
HDL-C (mmol/l)	1.2 $\pm$ 0.3	1.2 $\pm$ 0.3	1.3 $\pm$ 0.3	1.1 $\pm$ 0.3 $\ddagger$	1.3 $\pm$ 0.4	1.2 $\pm$ 0.3
TC/HDL-C	4.7 $\pm$ 1.8	4.7 $\pm$ 1.5	4.0 $\pm$ 1.0	5.3 $\pm$ 2.2 $\ddagger$	4.7 $\pm$ 1.7	4.7 $\pm$ 1.3
LDL-C (mmol/l)	3.3 $\pm$ 0.9	3.5 $\pm$ 1.0	3.2 $\pm$ 0.8	3.4 $\pm$ 1.0	3.5 $\pm$ 1.0	3.5 $\pm$ 1.0
Fasting glucose (mmol/l)	8.2 $\pm$ 3.1	9.1 $\pm$ 3.6 $\ddagger$	7.6 $\pm$ 2.8	8.7 $\pm$ 3.3	9.6 $\pm$ 3.8	8.6 $\pm$ 3.3 $\ddagger$
HbA <sub>1c</sub> (%)	7.5 $\pm$ 1.8	8.0 $\pm$ 1.9 $\ddagger$	7.1 $\pm$ 1.8	7.9 $\pm$ 1.8 $\ddagger$	8.2 $\pm$ 2.1	7.7 $\pm$ 1.5 $\ddagger$
Fasting insulin (pmol/l)*	105 (72–164)	87 (51–146) $\ddagger$	89 (60–149)	120 (78–179) $\ddagger$	76 (43–129)	99 (57–157) $\ddagger$
HOMA <sub>IR</sub> index*	34.7 (22.9–55.8)	33.1 (19.2–62.8)	27.5 (16.0–46.7)	42.7 (27.6–58.2) $\ddagger$	29.9 (14.7–61.9)	36.4 (19.7–64.2)
Urinary ACR (mg/mmol)	2.6 (0.7–6.1)	2.8 (0.8–7.1)	1.2 (0.6–1.9)	5.1 (0.9–24.1) $\ddagger$	2.6 (0.9–5.8)	3.2 (0.8–8.4)
Treatment (%)						
Diet	71 (60)	93 (32)	35 (65)	36 (55)	55 (35)	38 (28)
Oral drugs	39 (33)	176 (61) $\ddagger$	17 (31)	22 (34)	89 (57)	87 (65)
Insulin	9 (8)	21 (7)	2 (4)	7 (11)	12 (8)	9 (7)
Retinopathy (%)	10 (8)	62 (21) $\ddagger$	1 (2)	9 (14) $\ddagger$	38 (24)	24 (18)
Albuminuria (%)	48 (40)	110 (38)	8 (15)	40 (62) $\ddagger$	53 (34)	57 (43)
Neuropathy (%)	4 (3)	29 (10) $\ddagger$	2 (4)	2 (3)	13 (8)	16 (12)

Data are expressed as n, n (%), means  $\pm$  SD, or the geometric mean (25th and 75th percentiles). Data are compared between early- and late-onset patients, between early-onset nonobese and obese patients, and between late-onset nonobese and obese patients. \*Only measured in patients not treated with insulin.  $\ddagger P < 0.05$ ;  $\ddagger P < 0.001$ .

mutation (9), cosegregation findings in family studies were inconclusive (10, 11, present study). Similarly, whereas carriers of glucokinase variants from the families of YDM142 and HK84 had high fasting plasma glucose compatible with glucokinase deficiency (41), carriers of the glucokinase Q239R mutation from the YDM67 family did not have fasting hyperglycemia. In light of these findings, the prevalence of diabetogenic mutations could not be directly deduced in our population, and further functional studies are required to demonstrate their effects directly. Nevertheless, it is still plausible that these genetic mutations might represent rare variants or were affected by incomplete penetrance, phenocopies, and/or environmental factors.

Despite the familial nature of their diabetes, the remaining young patients with diabetes of unknown cause exhibited different phenotypic patterns compared with the late-onset patients. Cardiovascular risk factors (adverse lipid profiles and BP) and diabetic complications (retinopathy and neuropathy) were more frequent in late-onset patients, despite matching for sex and disease duration, perhaps because of delayed diagnosis, older age, and poorer glycemic control (42). In both early- and late-onset patients, general (BMI) and central (waist circumference) obesity clustered with cardiovascular risk factors and hyperinsulinemia. These findings were consistent with the important role of obesity in insulin-resistant and hyperinsulinemic states (43,44). The high rate of having a parental history of diabetes and obesity with its associated cardiovascular and complication risks in the early-onset patients prompts further search for other MODY- or obesity-associated genes. Our findings in early-onset patients with familial diabetes are in accordance with a recent report in Caucasian subjects, which described a high prevalence of obesity and resemblance to the metabolic syndrome in MODYx families (45). In addition, despite a mean disease duration of only 4 years, the young patients had a disproportionately higher prevalence of albuminuria (40%) compared with diabetic retinopathy and neuropathy, both of which had a <10% prevalence rate. These findings have also been reported in our previous analyses in a separate cohort of young diabetic patients (28). The cluster-

ing of obesity and albuminuria in these young patients warrants further investigation (46).

Based on these findings, we argued that if all the putative mutations found in the HNF-1 $\alpha$ , glucokinase, mt3243, and amylin S20G genes were diabetogenic, genetic factors were more significant than autoimmune markers in the pathogenesis of familial early-onset type 2 diabetes in Chinese individuals. The high prevalence of obesity, often in association with multiple cardiovascular risk factors, suggests that genetic causes for obesity require further investigation in young Chinese subjects with type 2 diabetes. Early identification of such genetic factors and an understanding of gene-gene and gene-environment interactions are important for both the diagnosis and treatment of these high-risk patients and their families (47).

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