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ORIGINAL ARTICLE

Clinical Characteristics, Residual Beta-Cell Function and Pancreatic Auto-Antibodies in Thai people with Long-Standing Type 1 Diabetes Mellitus

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Abstract

Objectives. To describe the characteristics of long-standing T1DM in Thai patients and assess residual beta-cell function with status of pancreatic autoantibodies.

Methodology. This is a cross-sectional study of Thai subjects with T1DM and disease duration ≥ 25 years seen at the Theptarin Hospital. Random plasma C-peptide and pancreatic auto-antibodies (Anti-GAD, Anti-IA2, and Anti-ZnT8) were measured. Patients who developed complications were compared with those who remained free of complications.

Results. A total of 20 patients (males 65%, mean age 49.4±12.0 years, BMI 22.5±3.1 kg/m^2, A1C 7.9±1.6%) with diabetes duration of 31.9±5.1 years were studied. Half of the participants remained free from any diabetic complications while the proportions reporting retinopathy, nephropathy, and neuropathy were 40%, 30%, and 15%, respectively. HDL cholesterol was significantly higher and triglyceride concentration significantly lower in patients who were free from diabetic nephropathy but not in those who were free from other complications. The prevalence rates of anti-GAD, anti-IA2, and anti-ZnT8 were 65%, 20%, and 10%, respectively. None of the patients who tested negative for both anti-GAD and anti-IA2 was positive for anti-ZnT8. Residual beta-cell function based on detectable random plasma C-peptide (≥ 0.1 ng/mL) and MMTT was found in only 3 patients (15%). There was no relationship between residual beta-cell function and protective effects of diabetic complications.

Conclusion. Endogenous insulin secretion persists in some patients with long-standing T1DM and half of long-standing T1DM in Thai patients showed no diabetic complications. HDL cholesterol was significantly higher and triglyceride concentration significantly lower in patients who were free from diabetic nephropathy.

Key words: type 1 diabetes mellitus, long-standing, residual beta-cell function, pancreatic autoantibodies, Thai people

INTRODUCTION

Emerging evidence in Caucasian populations suggests that endogenous insulin secretion persists in long-standing type 1 diabetes mellitus (T1DM). This is protective against severe hypoglycemia and is implicated in the reduced incidence of microvascular complications.[1-3] Following onset of diabetes, patients with T1DM exhibit diverse amounts of residual c-peptide, indicating varying levels of endogenous insulin production and beta cell function. Persistence of residual c-peptide is associated with improved glycemic control and reduced risk of complications and its preservation has been used as a clinical endpoint in clinical trials. However, the clinical significance of long-duration T1DM in Asian populations remained poorly understood. A recent study of 95 Chinese people with T1DM duration of ≥30 years revealed that almost 70% of the participants remained free from diabetic complications.[4] Interestingly, residual beta-cell function assessed by plasma C-peptide ≥0.075 nmol/L was observed in 15% of study participants but pancreatic auto-antibodies had been detected in less than 20% of patients. Furthermore, favorable lipid profiles were observed in these participants and closely corresponded with the Golden Years Cohort from United Kingdom and the Joslin 50-Year Medalist cohort from United States.

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OBJECTIVES

To better understand the clinical features of long-standing T1DM in Thai people, we evaluated the clinical characteristics of long-standing T1DM (duration of diabetes ≥25 years) in Thai patients and assessed residual beta-cell function together with the status of pancreatic autoantibodies.

METHODOLOGY

A cross-sectional study of Thai participants with T1DM registered at Theptarin Hospital, a tertiary diabetes center in Bangkok was performed from January 2019 to June 2019. T1DM was defined based on the clinical presentations of abrupt onset of symptoms including polyuria, polydipsia or unexplained weight loss, diabetic ketoacidosis (DKA) and insulin requirement from the time of diagnosis for control of hyperglycemia. Plasma C-peptide was measured in all T1DM cases and potential cases of misdiagnosis of T1DM were excluded if fasting C-peptide is ≥0.2 nmol/L after several years of onset of DM.[5] If pancreatic autoantibodies were negative or unknown, then insulin must have been started at or shortly after diagnosis and used continually thereafter. Other types of diabetes including latent autoimmune diabetes in adults (LADA) and Maturity Onset Diabetes of the Young (MODY) were excluded. None of the T1DM patients in our cohort underwent islet cell transplantation or pancreatic transplantation. No HLA haplotype was done in our routine care of patients with T1DM. Long-standing T1DM was defined as disease duration ≥25 years. Demographic data, mean glycated hemoglobin (HbA1c) in the previous 12 months, lipid profiles, serum creatinine, history of acute diabetic complications including severe hypoglycemia in the previous 12 months, chronic diabetic complications, and other co-morbidities during the study period were noted. Retinopathy was detected with the regular dilated eye examinations by ophthalmologists annually. Nephropathy was defined as persistent microalbuminuria greater than 30 mg of albumin per g of creatinine from spot urine on at least 2 occasions, 3-6 months apart. Neuropathy was detected based on annual monofilament test and/or vibration perception threshold testing. Macrovascular complications including coronary artery disease, stroke, and peripheral vascular disease were noted.

Plasma C-peptide was measured by chemiluminescent immunometric assay (IMMULITE®, Siemens) with an inter-assay coefficient of variation 3.3% at plasma C-peptide 0.2 nmol/L. Mixed meal tolerance test (MMTT) was measured if random plasma C-peptide was ≥ 0.03 nmol/L. MMTT was done by ingestion of 6 mL/kg of Ensure® up to 360 mL (1 calorie/mL; 65% carbohydrates, 21% protein and 14% fat) after overnight fasting (at least 8 h) and withholding of insulin injection or oral agents (at least 12 h). Plasma C-peptide and plasma glucose were obtained at 0 and 90 min after the ingestion. Pancreatic auto-antibodies (Anti-GAD, Anti-IA2, and Anti-ZnT8) were assessed by ELISA method (RSR ®, UK). All the cut-off values for positivity of pancreatic auto-antibodies were based on the manufacturer label. Cut-off point for anti-GAD positivity is 5 U/mL with a specificity of 98% and sensitivity of 92%. Cut-off point for anti-IA2 positivity is 7.5 U/mL with a specificity of 100% and sensitivity of 68%. Cut-off value for ZnT8A positivity is 15 U/ml with a specificity of 97% and sensitivity of 76%. Participants who developed complications were compared with those that remained free of diabetic complications. All participants provided informed consent and the Ethics Committee of Theptarin Hospital approved the study (EC 09/2018).

Statistical analyses

Continuous variables were presented as mean (SD) and categorical variables were presented as proportions. Comparisons between T1DM without any complication and T1DM with complication were done using unpaired Student's t-test for continuous data and Chi-square test for categorical data. *P-value* ≤0.05 was considered statistically significant. All statistical analyses were conducted using the Statistical Package for the Social Sciences (version 17.0; SPSS, Chicago, IL, USA).

Table 1. Clinical characteristics and laboratory data of Thai people with long-standing type 1 diabetes mellitus

	All patients (n=20)	Free of any complication (n=10)	With DM complications (n=10)	*p-value*
Age (yrs)	49.4±12.0	47.3±11.9	51.4±12.3	0.459
Male/Female	13/7	8/2	5/5	0.160
Age at diagnosis (yrs)	17.5±9.4	16.4±9.2	18.5±10.0	0.632
Pre-pubertal onset (%)	35%	50%	25%	0.160
Initial presentation with DKA (%)	70%	60%	80%	0.235
Duration of DM (yrs)	31.9±5.1	30.9±5.1	32.9±5.1	0.392
Current Smoking (%)	10%	10%	10%	0.763
BMI (kg/m^2)	22.5±3.1	22.0±2.5	23.0±3.7	0.501
Daily insulin usage (unit/kg)	40.7±14.9	38.6±8.9	42.8±19.5	0.547
HbA1c (mmol/mol)	63±2	57±1	68±2	0.176
HbA1c (%)	7.9±1.6	7.4±1.1	8.4±1.9	0.176
SBP (mmHg)	120±14	118±12	121±16	0.577
DBP (mmHg)	69±9	70±10	67±8	0.459
Total Cholesterol (mmol/l)	4.6±0.8	4.9±1.0	4.2±0.4	0.088
Triglyceride (mmol/l)	0.8±0.4	0.8±0.4	0.9±0.4	0.370
HDL (mmol/l)	1.9±0.5	2.1±0.5	1.7±0.5	0.097
LDL (mmol/l)	2.7±0.8	2.9±0.9	2.5±0.5	0.237
Detectable random plasma C-peptide (%)	15%	10%	20%	0.435

Table 2. Comparisons between T1DM patients with residual beta-cell function and patents without residual beta-cell function

	T1DM with residual beta-cell function (n=3)	T1DM without residual beta-cell function (n=17)	*p-value*
Age (yrs)	41.7±8.5	50.7±12.1	0.238
Male/Female	1/2	12/5	0.234
Age at diagnosis (yrs)	11.7±5.8	18.5±9.7	0.261
Pre-pubertal onset (%)	33.3%	35.3%	0.345
Duration of DM (yrs)	30.0±3.0	32.2±5.3	0.496
BMI (kg/m^2)	23.6±4.8	22.3±2.9	0.496
Daily insulin usage (unit/kg)	36.7±15.0	41.4±15.3	0.625
HbA1c (mmol/mol)	61±1	63±1	0.862
HbA1c (%)	7.7±2.1	7.9±1.5	0.862
Free from DM complications (%)	33.3%	52.9%	0.556
Episodes of severe hypoglycemia in the past 12 months (%)	33.3%	35.3%	0.745

RESULTS

From a total of 89 T1DM cases in our hospital, 20 long-standing T1DM participants were identified and studied. Baseline clinical data (males 65%, mean age 49.4±12.0 years, BMI 22.5±3.1 kg/m^2, HbA1c 63±2 mmol/mol, 7.9±1.6%) with duration of diabetes 31.9±5.1 years were shown in Table 1. DKA was the initial presentation in 14 patients from the cohort of 20 T1DM patients (70%) with long-standing duration of diabetes. Severe hypoglycemia in the previous 12 months was found in 35% of all patients. Half of the participants remained free from any diabetic complications while the proportions reporting retinopathy, nephropathy, and neuropathy were 40%, 30%, and 15%, respectively. Even though HDL cholesterol tended to be higher in participants who were free from any diabetic complications, it did not reach statistical significance (2.1 mmol/L vs. 1.7 mmol/L, *p-value* = 0.097). However, HDL cholesterol was significantly higher (2.1 mmol/L vs. 1.5 mmol/L, *p-value* = 0.011) and triglyceride concentrations were significantly lower (1.7 mmol/L vs. 2.5 mmol/L, *p-value* = 0.036) in participants who were free from diabetic nephropathy but not in those who were free from other complications.

The prevalence rates of anti-GAD, anti-IA2, and anti-ZnT8 were 65%, 20%, and 10%, respectively. No participant who tested negative both anti-GAD and anti-IA2 was positive for anti-ZnT8. The distribution of pancreatic autoantibodies in our T1DM patients with long-standing duration is shown in Figure 1. Residual beta-cell function based on detectable random plasma C-peptide (≥0.03 nmol/L) and MMTT were found in only 3 participants (15%). The mean random plasma C-peptide was 0.07 nmol/L in these patients and the mean peak C-peptide from stimulated MMTT was 0.29 nmol/L. No relationship was observed for residual beta-cell function and the protective effects of diabetic complications as revealed in Table 2. Two of 3 participants with residual beta-cell function had proliferative diabetic retinopathy and diabetic nephropathy. Persistent secretion of C-peptide was not associated with self-reported episodes of severe hypoglycemia in the last 12-month period. The presence of any pancreatic autoantibodies was 66.7% in participants with residual beta-cell function compared with 70.6% in participants without residual beta-cell function. There was no association between pancreatic autoantibody positivity and residual beta-cell function (*p-value* = 0.270).

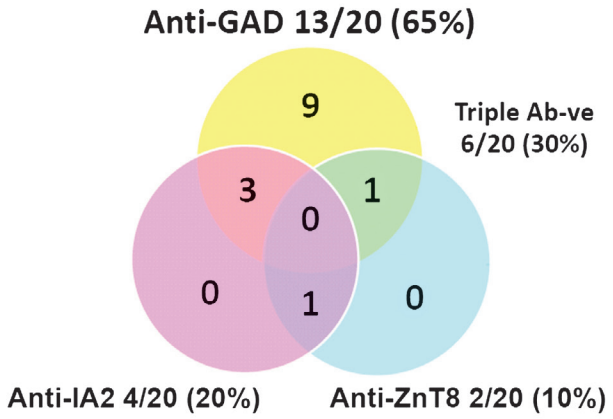

Figure 1. The distribution of pancreatic auto-antibodies in our T1DM patients with long-standing duration (N = 20 cases).

DISCUSSION

T1DM is a heterogeneous disease and the recent description of the 'endotype' has been supported using histological assessments from different ages at the onset of disease.[6] The extent of insulitis and aberrant sub-cellular distribution of proinsulin and mature insulin in the residual beta cells presented differently depending on a younger (<7 years old) or older age of onset. However, the study has been conducted exclusively in childhood-onset T1DM. The clinical profiles and immunologic studies in non-Caucasians are rarely reported. In this clinical study in Thai people with T1DM, our observations suggest that endogenous insulin secretion persists in some people with long-standing T1DM. In contrast to a previous study from China,[4] pancreatic auto-antibodies have been detected in up to 65% of the Thai participants with long-standing T1DM with anti-GAD antibodies as the most detected pancreatic auto-antibodies. Therefore, it remains unclear whether anti-ZnT8 is a useful marker for long-duration T1DM.[7]

Consistent with other studies,[1,4,7-9] almost half of our long-standing T1DM cohort showed no diabetic complication. The summarized comparison of results of our current study with other previous cohorts is shown in Table 3. Access to comprehensive diabetes services and specialists with expertise in T1DM poses several challenges in Thailand and

Table 3. Comparisons between our present study in Thai people with long-standing T1DM with other published series

Country	DM duration (yrs)	Complications	Persistent C-peptide
Joslin 50-year Medalist Study (United States, N=411)[1]	56.2±5.8	PDR 55% MAU 13% DN 61% CVD 48%	Minimal C-peptide (0.1-0.6 ng/mL) = 64.4% Sustained C-peptide (≥ 0.6 ng/mL) = 2.6%
Diabetes UK The Golden Years cohort (United Kingdom, N=400)[8]	55.8±5.4	PRP 43% DKD 36%	N/A
Chinese Study (China, N=95)[4]	37.3±6.8	DR 68% DKD 34% DN 61% CVD 14%	C-peptide ≥ 0.2 ng/mL = 14.7%
Japanese Study (Japan, N=29)[9]	55.4±3.9	PDR 59% DKD 46% CVD 25%	C-peptide > 0.4 ng/mL = 6.9%
Theptarin cohort (Thailand, N=20)	31.9±5.1	DR 40% DKD 30% DN 10% CVD 0%	C-peptide ≥ 0.1 ng/mL = 15.0%

Abbreviations: CVD – Cardiovascular Disease; DKD – Diabetic Kidney Disease; DN – Diabetic Neuropathy; DR – Diabetic Retinopathy; MAU – Microalbuminuria; PDR- Proliferative Diabetic Retinopathy; PRP – Panretinal Photocoagulation

other countries in Southeast Asia; however, T1DM patients could have long life expectancy similar to the general population if they adhere to self-diabetes management and have good support system. A recent mechanistic study among long-standing T1DM in the United States showed that elevated medium-sized HDL particles and elevated levels of HDL-associated paraoxonase 1 (PON1) which is an atheroprotective enzyme might contribute to vascular protection in this group of people.[10]

The limitations of the study should be acknowledged. First, this was a cross-sectional study from a tertiary diabetes care center in Bangkok. Our institute has an advantage as a comprehensive diabetes center in Thailand for over three decades. Therefore, to be generalizable, our findings should be confirmed in more heterogeneous healthcare services across Southeast Asia. Second, the mean HbA1c values were obtained in the past 12 months. The long-term mean glycemic control since the onset of disease might be different from the present results. Third, some residual or undocumented factors affecting diabetic complications such as the frequency and severity of DKA, distinct protective genetic factors, nutrition status or intake of various supplements could not be completely ruled out. Fourth, the modest sample size of our study would affect the statistical power. Multicenter studies are required to verify our present results and create the prospective registry for T1DM in Southeast Asia. Finally, measured conventional plasma C-peptide levels in this study might misclassify some participants who had very low level of preserved insulin secretion if ultra-sensitive plasma C-peptide measurements were used.

CONCLUSION

In conclusion, our observations highlight the emergence of long-standing type 1 diabetes mellitus in an Asian population that is considered to be under-represented. Further multi-center studies in Asian populations with ultra-sensitive plasma C-peptide measurements and detailed mechanistic study together with the assessment of genetic and epigenetic indices should be considered in these individuals with long-standing T1DM. These studies will provide a better understanding of the contributing determinants associated with long-term survival in this unique population.

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Statement of Authorship
All authors certified fulfillment of ICMJE authorship criteria.

Author Disclosure
The authors declared no conflict of interest.

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