Pharmacogenetic and Clinical Risk Factors for Diabetes After Transplant in a Single Pediatric Centre

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Aim:

- To study the prevalence of post kidney transplant diabetes (NODAT) in a single Asian pediatric centre.
- To identify clinical factors and single nucleotide polymorphisms (SNPs) which may predict NODAT.

Methods:

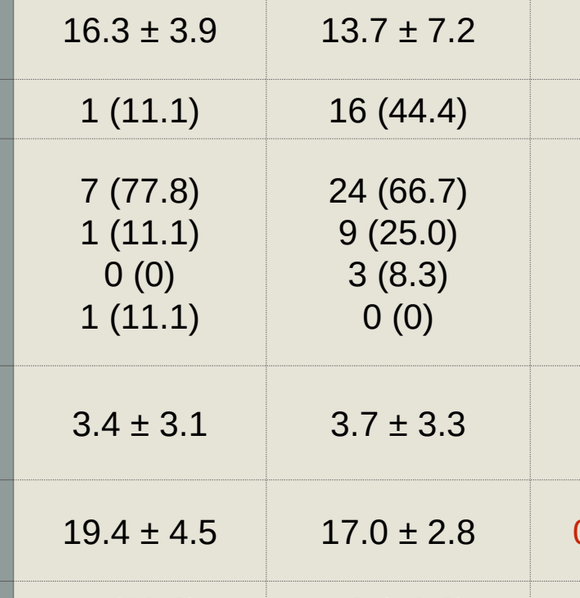
- Kidney patients from Shaw-NKF Children's Kidney Centre, National University Hospital transplanted between Feb 1989 and Oct 2017 were recruited**.**
- NODAT diagnosis was based on American Diabetes Association guidelines.
- Clinical parameters were collected retrospectively up to Jan 2018.
- Blood samples were collected after consent was obtained.
- DNA was extracted from leukocytes and amplified via polymerase chain reaction using the single base extension technique.
- After amplification, DNA was genotyped using fragment analysis.
- 20 SNPs from the following genes associated with type 2 diabetes mellitus were screened; HHEX rs7923837, HHEX rs5015480, TCF7L2 rs4506565, IGF2BP2 rs4402960, CDKAL1 rs1094398, ADIPOQ rs266729, ADIPOQ rs2241766, CDKAL1 rs2280789, CCL5 rs2280789, CCL5 rs2107538, CCL5 rs3817655, ABCB1 rs1045642, CYP3A4*1G rs2242480, PAI-1 rs1799889, KCNQ1 rs2237892, KCNQ1 2237895, KCNQ1 2237897, SLC30A8 rs13266634, HHEX rs1111875, CYP3A5 rs776746 .
- Parametric tests were used for univariate analysis between groups while Cox regression was used for multivariate analysis.

Table 1. Baseline characteristics of NODAT and non-NODAT patients

Characteristics	NODAT (n=9)	non-NODAT (n=36)	p value
Age at Tx (years) mean ± SD	16.3 ± 3.9	13.7 ± 7.2	0.31
Male, n (%)	1 (11.1)	16 (44.4)	0.06
Ethnicity Chinese, n (%) Malay, n (%) Indian, n (%) Other, n (%)	7 (77.8) 1 (11.1) 0 (0) 1 (11.1)	24 (66.7) 9 (25.0) 3 (8.3) 0 (0)	0.14
Dial years before Tx (years) mean ± SD	3.4 ± 3.1	3.7 ± 3.3	0.82
Body mass index (kg/m^2) mean ± SD	19.4 ± 4.5	17.0 ± 2.8	0.048
Dyslipidemia, n (%)	4 (50.0)	12 (46.2)	0.58
Initial immunosuppression Tacrolimus, n (%) Cyclosporine, n (%)	8 (88.9) 1 (11.1)	27 (75.0) 9 (25.0)	0.66

Tx: transplant; Dial: dialysis

Figure 1.Time from transplant to diagnosis of NODAT, comparing tacrolimus and cyclosporine as initial immunosuppression. Patients on tacrolimus were diagnosed with NODAT earlier than cyclosporine.



Results:

- 45 kidney transplant patients (14.2 ± 6.7 years at transplant), yrs) were recruited of which 9 (20%) patients had NODAT.2 patients received a second kidney transplant.
- Mean follow-up duration was 7.0 ± 7.1 years.
- NODAT was diagnosed at 2.7 ± 3.7 years post transplant.
- 1 patient had NODAT after switching maintenance cyclosporine to tacrolimus.
- On univariate analysis, NODAT patients have significantly higher body mass indices compared to non-NODAT patients (p=0.05) (Table 1). However, this significance is lost on multivariate analysis (Table 2).
- On multivariate analysis, male patients were found less likely to develop NODAT compared to females (p 0.02, 95% CI 0.0-0.6, Table 2) after adjusting for ethnicity, body mass index, CMV and dyslipidemia status.
- At 10 years post-transplant, 77.8% of patients on tacrolimus compared to 88.9% of patients on cyclosporine as initial immunosuppression were diabetes-free.
- Kaplan-Meier survival curves showed earlier onset of NODAT in patients whose initial immunosuppression was tacrolimus vs cyclosporine (Figure 1).
- IGF2BP2 rs4402960 SNP was found to be associated with NODAT. IGF2BP2 gene codes for Insulin-like growth factor 2 mRNA-binding protein 2. The T allele is reported in 38% of the local population and reported to confer risk of type 2 diabetes in East Asian, European and South Asian populations.* Frequency of rs4402960 SNPs variants in the NODAT population was significantly different from that of non-NODAT. (p=0.008, Table 3).
- IGF2BP2 rs4402960 T/T haplotype was associated with increased risk of NODAT (HR 29, CI 2.83-296.92, p=0.02).

Table 2. Multivariate analysis of pretransplant clinical risk factors for NODAT

Pretransplant factors	Hazard ratio	p value	CI (95%)
Male	0.05	0.02	0.0-0.6
Body Mass Index	1.1	0.17	0.9-1.3
Dyslipidemia	0.5	0.41	0.1-2.7
CMV D+R-#	3.4	0.18	0.6-20.8
Chinese	0.8	0.86	0.1-8.1
Malay	0.1	0.14	0.0-2.1
Indian	0.0	0.99	0.0

CMV D+R-: cytomegalovirus donor positive serology, recipient negative serology at transplant

Table 3. Distribution of IGF2BP2 variants in the transplant population

Genetic Variants	S'pore %*	NODAT n (%)	non-NODAT n (%)	p value
rs4402960 TT	3	1 (25)	0 (0)	0.008
rs4402960 GT	32	1 (25)	1 (3.8)	
rs4402960 GG	65	2 (50)	25 (96.2)	

* Percentage variants in the Singapore population, derived from Teo YY, Sim X, Ong RTH, et al. Singapore Genome Variation Project: A Haplotype map of three South-East Asian populations. Genome Research (In press).

Conclusions:

- Prevalence of NODAT in our cohort was 20%.
- The female gender is a possible risk factor in NODAT.
- s4402960 SNPs in the IGF2BP2 gene were also identified as possible risk factor.
- Patients on initial immunosuppression of tacrolimus developed NODAT earlier than cyclosporine.

This work is supported by the Venerable Yen Pei-National Kidney Foundation Research Fund (NKFRC/2016/01/07)