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Original article

The Role of HLA Alleles and Haplotypes in Type 1 Diabetes Mellitus: A South Indian Perspective

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Abstract

Type 1 Diabetes Mellitus (T1DM) is a severe autoimmune disorder primarily affecting children, resulting from complex interactions between genetic and environmental factors. Among the genetic contributors, the Human Leukocyte Antigen (HLA) region on chromosome 6p plays a central role in disease susceptibility. This pilot case-control study aimed to investigate the association of specific HLA alleles and haplotypes with T1DM in the South Indian population and to identify the predominant HLA DR-DQ two-locus haplotypes linked to disease susceptibility. A total of 118 children were screened, from which 57 unrelated children diagnosed with T1DM and receiving care at the pediatric endocrinology clinic of Kovai Medical Centre and Hospital, Coimbatore were selected. An equal number of healthy controls without any autoimmune conditions were included for comparison. Genotyping of HLA DRB1 and DQB1* alleles was performed using PCR with sequence-specific primers (PCR-SSP), and statistical analysis was conducted using logistic regression models. Significant associations were identified between T1DM and alleles such as DRB103, DRB104, DRB107, DRB114, and DQB10301, 0302, 0304, and 0501. Among these, the haplotype DRB103-DQB106 was significantly associated with the disease. The strongest risk genotype was DRB103-DQB102, whereas others like DRB115-DQB106 and DRB115-DQB105 were associated with lower risk. This study confirms a strong link between specific HLA alleles/haplotypes and T1DM susceptibility in South Indian children, suggesting their potential as early diagnostic genetic markers.*

Key Words: *Type 1 Diabetes Mellitus; Autoimmune diseases; Haplotypes.*

Introduction

The permanent destruction of insulin-producing beta cells in the islets of Langerhans occurs because of autoimmune factors. The genetic basis determines this process that starts from one environmental stimulus or multiple such elements. The disease develops across several months to years and possibly decades when a patient maintains normal blood glucose levels along with no actual symptoms. The identification of auto antibodies occurs too late after the autoimmunity process begins but inherited auto genes exist since birth. Medical tests measuring biochemical indicators for the metabolic disorder become possible after beta cell destruction reaches a significant level but before hyperglycemia develops clinical symptoms. The extensive initial undetected phase indicates that such a high abundance of operating beta cells requires destruction to trigger hyperglycemic signs.

T1DM impacts the most number of South-East Asian children in India right now. The 6th edition of the International Diabetes Federation Diabetes Atlas reveals that India experiences three incidents of T1DM every year among 100,000 children from the 0–14 years old age group. Numerous studies indicate that Indian women have a diabetes prevalence rate of 17. A research conducted in Karnataka established 93 out of 100,000 children suffered from the condition while

the overall figure came to 3. Two separate reports found that Chennai has 2 T1DM cases per 100,000 children whereas the total number of cases in the city stands at 10. Karnal (Haryana) 2 per 100000 children 24/05/2012. This condition exists as a common disease which spreads widely although its test results tend to vary. The annual rise in T1DM cases is 3–5% but it continues its upward trend slower than the increase in T2DM cases. T1DM occurs in two distinct periods during childhood between four and six years and between ten and fourteen years old. T1DM diagnosis occurs before the tenth birthday in roughly 45% of children suffering from the disease. T1DM does not particularly favor any gender, and thus its manifestations are not inclined to be seen in a particular sex. The HLA region displays elevated LD levels that allow researchers to examine population divergence using haplotypic information together with single locus results. DNA typing has proven superior to serology by disclosing T1DM-related specific sequences and alleles which mainly occur in DRB1 and DQB1 loci.

This present research details the relationship between HLA alleles and type I diabetes mellitus in the South Indian population.

Materials and Methods

The fundamental goal of this research work is to determine connections between type 1 diabetes and HLA allele and haplotype associations. The researchers plan to execute this observational study at Kovai Medical Centre and Hospital, Coimbatore in a prospective manner. Outpatient record registrations for the study duration included 185 patients and below 18 years age patients constituted 5% of all registered cases. The researchers applied the established inclusion and exclusion criteria to select their patients. Patients with type I DM below 18 years old who obtained clinical diagnosis and laboratory findings participated in the research.

DNA extraction from whole blood

The researchers obtained peripheral blood from index finger pricks to extract DNA from 3 milliliters of blood using the salting out technique described by Syed (2008). The PBS solution along with DMEM and other stock materials went through an autoclave process at 120ºC and 15 lb pressure for 30 minutes before storing them first in ice and later at 4ºC.

An expert addition of 3000 micro litres peripheral vein blood into 12000 micro litres Red Cell Lysis Buffer occurred inside a 15 micro litre polypropylene centrifuge tube. The sample tube was positioned for five minutes to facilitate red blood cell lysis once it was turned on end. After the tube was centrifuged at 2000 rpm during a period of 10 minutes. A pipette removed the upper liquid layers after which the remaining solids were collected. When finished the pellet received 15ml RCLB for washing then proceeded to a centrifuge period of 10 minutes at 2000rpm. The WBCs settled at the bottom as clear nucleated cells so a combination of 3 ml Nuclear Lysis Buffer (NLB) and Sodium Dodecyl Sulphate (SDS) was used for cell lysis prior to the following step.

Proteins will settle as precipitate when you combine the solution with 1 ml of 6M Sodium Chloride (NaCl). The cultures underwent mixing operations following vortex mixing of the samples. Subsequently, the process involved adding 2 ml chloroform before thoroughly shaking the contents. A relatively uniform milky suspension formed as a result and underwent 10 minutes of spin at 2000 RPM. The DNA solution in aqueous form remained in the aqueous phase alongside the interphase protein boundary without causing any disruption when transferring it to a new sterile 15 ml centrifuge tube. The solution received two volume equivalents of 15% ethanol followed by slow shaking of the tube to create DNA precipitation. The DNA pellet was transferred to 1. 5 ml microcentrifuge tubes where it was spun using 2,000 rpm centrifugation for 5 minutes at 70% ethanol solution. A repeated alcohol washing step was carried out to extract maximum proteins and salts from the DNA. Before final washing with 200µl Tris-EDNA (TE) buffer, the alcohol solution was decanted away from the container.

The DNA underwent quality tests and quantity evaluations.

Optical Density Method

The purified DNA's concentration together with its protein and carbohydrate content was determined through DNA absorption measurement at 260 nm wavelength in a UV spectrophotometer. The measurement of OD at 260 nm should be 1 for a DNA solution containing 50 ng/µl (Sambrook et al. 1989) [18]. Scientists determined DNA purity by analyzing its absorption through both 260nm and 280nm wavelengths in a UV spectrophotometer. The calculation used the absorbed light measurement at 260 nm wavelength relative to 280 nm light absorbance to determine the ratio value.

DNA with the ratio of 1. 8 fractions were obtained from the division of the latter sample for comprehensive analytical purposes. The analysis of DNA concentration depended on the following formula:

The analysis of DNA concentration required the use of this calculation:

The DNA concentration formula involves multiplication of OD 260 with Dilution Factor and 50 ng/µl.

HLA-DRB1* typing by PCR-SSP

The genotyping methodology utilized PCR - SSP to analyze HLA- DRB 1* under the parameters described by Olerup&Zitterquist in (1993) [19]. The reactions contained the internal control primer derived from the human growth hormone gene to prevent any misinterpretation by excluding PCR failure while maintaining correct allele calling. PCR reactions require 5. 2 µl double distilled water along with 1 because they include internal control primers for human growth hormone gene. An additional 2 ul of 10 x PCR buffer, 0. 5 ml of 5% glycerol, 0. The dNTP solution required 2 microliters of each 10 mmole dATP, dCTP, dGTP and dTTP solution. The test solution contains 1 µl of 5 U/µl Taq DNA Polymerase enzyme together with 0.4 µl cresol red and 0.3 µl of 10mm forward and reverse primers and 4 µl of DNA in a final reaction mix. Two sets of PCR cycles using distinctive annealing temperatures were utilized for amplification of HLA-DRB1*. The first step of the reaction required a 2-minute denaturing phase at 94 degC before conducting ten denaturing cycles at 94 degC for ten seconds and ten annealing cycles at 64 degC for one minute followed by twenty more cycles that combined ten-second denaturing at 94 degC with fifty-second annealing at 61 degC and a final thirty-second extension at 72 degC.

HLA – DQB1* typing by PCR – SSP

The study employed PCR – SSP (sequence specific primer) to conduct HLA – DQB1* genotyping according to Bunce et al. (1995) [20]. The experiments required nine practitioner-designed reaction mixes at 2µM per mix for DQB1* alle typing (OccimumBiosolutions, Hyderabad, India) that used few microliters per mix from allele-specific 0. Common control at 5µM of internal control primer and 1µg of cresol red in the reaction. Internal control primer was utilized in human being growth hormone gene-based reactions to verify the accuracy of PCR in all experimental setups. Each PCR reaction required nine different additives which were prepared and placed in -20 °C freezer for future use. The components included 72 µl of double distilled water and 11. 7 µl of 10 x PCR buffer (Final conc. 1x), 5. 64 µl 100% glycerol, 2. 7 µl 10 mM dNTP mix and 22 µl of template DNA (100 ng /reaction). A mix of PCR solution was added to primer predotted PCR reaction tubes before they were incorporated into each tube. The reaction comprises 7µl of PCR solution, 5µl of primers and 2µl of the DNA template PCR in each PCR solution.

Detection of PCR products by Electrophoresis method

A 1.5% agarose solution was used for electrophoresis identification of PCR products as described in Sambrook et al. 1989 [18]. The personnel cleaned both gel platforms and gel combs with 70% alcohol. The balancing table received the platform which the combs rested in position while 70 ml of Tris-Boric Acid- EDTA (TBE) buffer received 1.05 g of agarose (1.5%) which was boiled within a microwave oven. The material reached a temperature of 60º C before 6 µl of 10 µg / ml Ethidium Bromide solution was added for mixing before being poured into the platform. Subsequently the gel solidified for 20 minutes before placement of the gel into cold 1× TBE buffer within the electrophoresis tank. The laboratory staff carefully extracted the combs from the platform after another 10-minute period. The PCR products received loading dye addition before placing them into the wells for 100 volt electrophoresis that ran for 20 minutes. The visualization process started with ultra-violet exposure of the gel followed by documentation in a gel documentation unit. The allocation of alleles followed the pattern generated by band formation during the process.

Statistical Analysis

The research data was examined through the SPSS version 16 for windows statistical software program. The descriptive analysis revealed information about frequency observations alongside mean and standard deviation distribution. Research investigators obtained allelic frequencies through direct counting while examining each allele risk by comparing its frequencies between patients and healthy participants. The chi-square test produced odds ratios OR and confidence intervals together with p values. A logistic regression method served to measure correlations between measurement data along with nominal categorization variables. The research evaluates significance at 5% statistical threshold. The Pearson chi –square results demonstrate a p-value of 0.101 that exceeds 0.05 and enables us to approve the null hypothesis (no significance).

Results

Age wise distribution of cases and controls

In the present study the total numbers of cases enrolled were 57 and controls 57. Distribution of above sample size according to age wise specified in the table 1. Large number of type 1 DM patients are present in >10yrs in both cases and controls when compared to < 10 years.

Table 1. Age distribution of cases and controls

Age	CASES		CONTROL	
	COUNT	PERCENT	COUNT	PERCENT
<10 YRS	**13**	**22.8%**	**5**	**8.8%**
>10 YRS	**44**	**77.2%**	**52**	**91.2%**
TOTAL	**57**	**100.0%**	**57**	**100.0%**

Distribution of HLA DRB1* Alleles

The comparison of essential HLA DRB1* alleles between patients and controls appears in Table 2 together with the corresponding p values in Table 3. The patients displayed higher frequency of DRB1*03 allele than controls (31.6%vs12.28%) with a (OR-3.297, p value of 0.000), and DRB1*04 allele followed with (cases 22.81%vs controls9.65%) (OR-2.767 &p value 0.007).

The patient group revealed less frequent occurrence of DRB1*07 at 2.63% compared to controls at 21.05% with a p value of 0.000 followed by DRB1*14 at 0.88% among patients against 6.14% among controls with a p value of 0.031.

The occurrence of DRB1*10 allele remains constant between patients and control groups at 11.4% and 14.04% respectively with a p value above 0.05. New data indicate that DRB1*15 occurs more often in controls than patients (cases 18.42% vs. controls 24.56%) without reaching statistical significance (0.259 > p-value 0.05).

Table 2. Frequency of HLA-DR in cases and controls

	CASES		CONTROL	
HLA-DR	**COUNT**	**PERCENT**	**COUNT**	**PERCENT**
HLA-DRB1*01	2	1.8%	2	1.8%
HLA-DRB1*03	36	31.6%	14	12.3%
HLA-DRB1*04	26	22.8%	11	9.6%
HLA-DRB1*07	3	2.6%	24	21.1%
HLA-DRB1*08	4	3.5%	4	3.5%
HLA-DRB1*09	0	0.0%	1	0.9%
HLA-DRB1*10	16	14.0%	13	11.4%
HLA-DRB1*11	0	0.0%	2	1.8%
HLA-DRB1*12	3	2.6%	5	4.4%
HLA-DRB1*13	2	1.8%	3	2.6%
HLA-DRB1*14	1	0.9%	7	6.1%
HLA-DRB1*15	21	18.4%	28	24.6%
TOTAL	**114**	**100.0%**	**114**	**100.0%**

Table 3. Frequency distribution of HLA-DRB1* alleles among cases and controls

HLA-DR	Cases (n=57)	Control (n=57)	OR	Low-CI	High-CI	χ2	p-value
DRB1*01	2 (1.75%)	2 (1.75%)	1.000	0.138	7.224	0.000	1.000
DRB1*03	36 (31.58%)	14 (12.28%)	3.297	1.662	6.538	12.399	0.000
DRB1*04	26 (22.81%)	11 (9.65%)	2.767	1.294	5.917	7.259	0.007
DRB1*07	3 (2.63%)	24 (21.05%)	0.101	0.030	0.347	18.527	0.000
DRB1*08	4 (3.51%)	4 (3.51%)	1.000	0.244	4.100	0.000	1.000
DRB1*09	0 (0%)	1 (88%)	0.000	1.763	2.289	1.004	0.316
DRB1*10	16 (14.04%)	13 (11.4%)	1.268	0.580	2.775	0.356	0.551
DRB1*11	0 (0%)	2 (1.75%)	0.000	1.769	2.302	2.018	0.155
DRB1*12	3 (2.63%)	5 (4.39%)	0.589	0.137	2.526	0.518	0.472
DRB1*13	2 (1.75%)	3 (2.63%)	0.661	0.108	4.031	0.204	0.651
DRB1*14	1 (0.88%)	7 (6.14%)	0.135	0.016	1.118	4.664	0.031
DRB1*15	21 (18.42%)	28 (24.56%)	0.694	0.367	1.312	1.274	0.259

Distribution of HLA DQB1* Alleles

The analyze of HLA DQ allele distribution presents data through figure 1 and supports data with p values in Table 4.

In the population analysis HLA DQB1*02 was detected more frequently in patients as opposed to controls (cases 41.23% vs. controls 15.79 %) (OR 3.741 & p value 0.000). While HLADQB1*06 shows greater occurrence in case patients than controls at 37.32% to 28.07% the calculated p value of 0.121 exceeds 0.05.

Among controls HLA DQB1*0301, 0304(DQ7) & DQB1*0302 (DQ8) occur 14.91% while HLA DQB1*05 distributions at 20.18% in cases but 38.60% in controls (p value=0.002).

All DRB1*and DQB1* allele distributions remain equivalent between case and control groups.Figure 1. Distribution of HLA DQ alleles in cases and control group

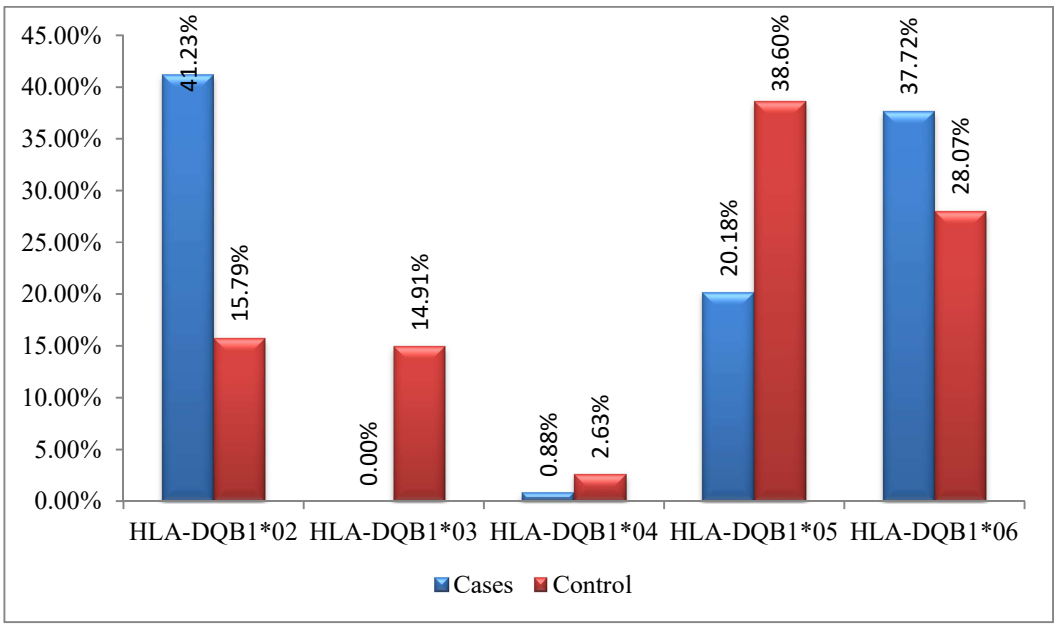

Table 4. Distribution of HLA-DQ alleles in South Indian Type 1 diabetes mellitus patients

HLA-DQ	Cases (n=57)	Control (n=57)	OR	Low-CI	High-CI	χ2	p-value
DQB1*02	47 (41.23%)	18 (15.79%)	3.741	2.000	7.000	18.098	0.000
DQB1*03	0 (0.00%)	17 (14.91%)	0.000	1.879	2.518	18.370	0.000
DQB1*04	1 (0.88%)	3 (26.3%)	0.327	0.034	3.196	1.018	0.313
DQB1*05	23 (20.18%)	44 (38.60%)	0.402	0.222	0.727	9.321	0.002
DQB1*06	43 (37.72%)	32 (28.07%)	1.552	0.889	2.709	2.404	0.121

Distribution of DRB1*-DQB1* two locus haplotype in Type 1 DM Patients:

Patients carrying the DRB1*03-DQB1*02 haplotype show a frequency increase in comparison to control subjects (cases were found in 24.56% of participants while only 4.39% of controls had this haplotype) with obtained results of OR -7.098 and p value 0.000. Data indicates that the haplotype DRB1*03-DQB1*06 exists exclusively among cases according to a significant p value of 0.024. (Table 5)

Results demonstrate DRB1*07-DQB1*02 exists more commonly in controls than in cases (cases 2.63% while controls hold 8.77%) with a statistically significant p value 0.046.($p < 0.05$). The haplotype DRB1*15-DQB1*06 appears most frequently among controls since it exists at 2.63% in cases but 26.32% in controls where p value measures 0.000. The haplotype DRB1*15-DQB1*05 displays maximal distribution within controls given the p value 0.031 ($p < 0.05$) provides.

Table 5. Haplotype frequency in South Indian type 1 diabetes patients

HLA-DR*DQ	Cases (n=57)	Control (n=57)	OR	Low-CI	High-CI	χ2	p-value
DRB1*01 - DQB1*02	2 (1.75%)	0 (0%)	0.000	1.769	2.302	2.018	0.155
DRB1*01 - DQB1*05	2 (1.75%)	0 (0%)	0.000	1.769	2.302	2.018	0.155
DRB1*03 - DQB1*02	28 (24.56%)	5 (4.39%)	7.098	2.630	19.153	18.743	0.000
DRB1*03 - DQB1*03	0 (0%)	1 (0.88%)	0.000	1.763	2.289	1.004	0.316
DRB1*03 - DQB1*04	1 (0.88%)	1 (0.88%)	1.000	0.062	16.184	0.000	1.000
DRB1*03 - DQB1*05	7 (6.14%)	2 (1.75%)	3.664	0.744	18.031	2.892	0.089
DRB1*03 - DQB1*06	5 (4.39%)	0 (0%)	0.000	1.789	2.340	5.112	0.024
DRB1*04 - DQB1*02	8 (7.02%)	2 (1.75%)	4.226	0.877	20.357	3.765	0.052
DRB1*04 - DQB1*03	2 (1.75%)	1 (0.88%)	2.018	0.180	22.571	0.338	0.561
DRB1*04 – DQB1*05	9 (7.89%)	5 (4.39%)	1.867	0.606	5.759	1.218	0.270

DRB1*04 - DQB1*06	8 (7.02%)	2 (1.75%)	4.226	0.877	20.357	3.765	0.052
DRB1*07 - DQB1*02	3 (2.63%)	10 (8.77%)	0.281	0.075	1.050	3.997	0.046
DRB1*07 - DQB1*03	3 (2.63%)	2 (1.75%)	1.514	0.248	9.233	0.204	0.651
DRB1*07 - DQB1*05	1 (0.88%)	5 (4.39%)	0.193	0.022	1.678	2.739	0.098
DRB1*07 - DQB1*06	1 (0.88%)	2 (1.75%)	0.496	0.044	5.543	0.338	0.561
DRB1*08 - DQB1*02	0 (0%)	1 (0.88%)	0.000	1.763	2.289	1.004	0.316
DRB1*08 - DQB1*04	2 (1.75%)	0 (0%)	0.000	1.769	2.302	2.018	0.155
DRB1*08 - DQB1*05	1 (0.88%)	0 (0%)	0.000	1.763	2.289	1.004	0.316
DRB1*08 - DQB1*06	3 (2.63%)	2 (1.75%)	1.514	0.248	9.233	0.204	0.651
DRB1*10 - DQB1*02	0 (0%)	1 (0.88%)	0.000	1.763	2.289	1.004	0.316
DRB1*10 - DQB1*05	7 (6.14%)	11 (9.65%)	0.613	0.229	1.641	0.965	0.326

DRB1*10 - DQB1*06	6 (5.26%)	4 (3.51%)	1.528	0.419	5.565	0.418	0.518
DRB1*11 - DQB1*03	1 (0.88%)	1 (0.88%)	1.000	0.062	16.184	0.000	1.000
DRB1*12 - DQB1*02	1 (0.88%)	2 (1.75%)	0.496	0.044	5.543	0.338	0.561
DRB1*12 - DQB1*05	0 (0%)	3 (2.63%)	0.000	1.776	2.314	3.040	0.081
DRB1*12 - DQB1*06	0 (0%)	2 (1.75%)	0.000	1.769	2.302	2.018	0.155
DRB1*13 - DQB1*05	0 (0%)	1 (0.88%)	0.000	1.763	2.289	1.004	0.316
DRB1*13 - DQB1*06	2 (1.75%)	2 (1.75%)	1.000	0.138	7.224	0.000	1.000
DRB1*14 - DQB1*05	3 (2.63%)	2 (1.75%)	1.514	0.248	9.233	0.204	0.651
DRB1*14 – DQB1*06	1 (0.88%)	2 (1.75%)	0.496	0.044	5.543	0.338	0.561
DRB1*15 - DQB1*02	0 (0%)	2 (1.75%)	0.000	1.769	2.302	2.018	0.155
DRB1*15 - DQB1*03	3 (2.63%)	3 (2.63%)	1.000	0.198	5.062	0.000	1.000

DRB1*15 - DQB1*05	1 (0.88%)	7 (6.14%)	0.135	0.016	1.118	4.664	0.031
DRB1*15 - DQB1*06	3 (2.63%)	30(26.32%)	0.076	0.022	0.256	25.829	0.000
DRB1*15 - DQB1*06	3 (2.63%)	30(26.32%)	0.076	0.022	0.256	25.829	0.000

Discussion

Type 1 Diabetes Mellitus develops during childhood or adolescence while requiring insulin treatment. This disease type functions as a non-inherited polygenic condition known as Non-Genetic Constitutional. Among multiple diseases causes stands out the HLA gene which plays a crucial role during the disease progression. The development of T1DM depends significantly on two HLA antigens including Class II human leukocyte antigen (HLA) DQ and DR antigens and Class II HLA DPA1, DPB1. There exists a common pattern in which protective DRB1* alleles pair with vulnerable DQ alleles yet protective DQ alleles link up with vulnerable DR alleles in the same population. T1DM incidence may be influenced by these protective DRB1/disease-susceptible DQB1 or protective DQA1/disease-susceptible strictly linked DQB1 combinations.

This study represents the initial observational research of HLA alleles and haplotypes relative to T1DM in children from the South Indian population. In South Indian children the genetic predisposition to T1DM mainly exists among DRB103 alleles followed by DRB104 alleles. The findings of previous research which studied T1DM patients across east and north India areas indicated DRB103 as the main allele linked to T1DM diagnosis. A north Indian and Japanese survey along with this observation found that DRB109 functioned as an effective allelic factor in triggering T1DM.

The pattern of DRB1*04 as the leading antigen has been documented similarly within different Asian ethnic groups [22]. Among Caucasians the most important genetically susceptible allele for DRB1 is DRB1*04 that stands above the secondary allele DRB1*03.

Protection from the disease rests with DRB1*07 which is followed by DRB1*14 then DRB1*15. Results from research studies with Caucasian, Asian and Indian participants revealed that DRB1*15 stood as the most protective allele followed by DRB1*07. Protection against TB among Indian children mostly depends on alleles DRB1*07 and DRB1*15. DRB1*14 serves as a protective allele in South Indian children with statistical significance (p value 0.031).

DQB1*02 leads all susceptible alleles precedence followed by DQB1*06 among the genetic locus of HLA class II in South Indian children. Studies carried out throughout India [23] found DQB1*02 to be nearly all significant in susceptibility allele testing. Vštačřina DQB1*06 appeared with a heightened frequency among cases (37.72%) yet the statistical value is above 0.05 (p value 0.121).

The protective HLA-DQB1 alleles include DQB1*05 and DQB1*0301,0304 and DQB1*0302 with p value 0.002. The population of Finnish and Swedish Caucasians showed DQB1*0302 to be a risk factor allele. Asians show evidence of DQB1*0302 acting as a susceptible allele according to literature reports [22].

The research exposed protective and susceptible haplotypes during the investigation. DRB1*03-DQB1*02 the most significant susceptible haplotype. The research findings agree with genetic haplotype data found in Caucasians and Koreans and North Indian populations [25] [21]. Research papers have identified DRB1*0405-DQB1*0401 as the main susceptible haplotype among Asians [22].

The distribution of protective haplotypes among South Indian children includes DRB1*15-DQB1*06 and DRB1*15-DQB1*05 and DRB1*07-DQB1*02 haplotypes. These genetic variations resemble testing results from both Asians and Caucasians as well as other Indian populations.

Genetic factors must be considered as the leading cause of diabetes incidence according to recommendations. The incidence of Type I DM seems to be influenced by an important gene factor which exists when susceptible DRB1 interacts with protective DQB1 or vice versa[26].

Conclusion

Research indicates how HLA alleles display multiple exceptional findings in their association with Type 1 Diabetes (TID) through three essential points: 1. The DRB1 and DQB1 risk frequency depends on particular sequences of DRB1 and DQB1 alleles instead of singular locus genotypes. 2. The occurrence of significant DR-DQ associations depends on TID risk and protection which varies based on individual DR-DQ haplotypes. 3. TID susceptibility is linked to different haplotypes whereas several haplotypes provide protection against it. The genotype pairing of DRB103-DQB102 produces a higher probability for the development of the condition. Studies have revealed that protective factors arise from three haplotypes: DRB115-DQB106, DRB115-DQB105 and DRB107-DQB102 which reduce the risk of disease development. A case-controlled multicentric genetic epidemiological study should be conducted immediately in India because this nation experiences uncommon epidemics and endemics that require further research into Type 1 Diabetes molecular pathology. Using the mentioned gene alleles medical professionals would be better able to detect Type 1 Diabetes.

References:

1. Atkinson MA, Maclaren NK. The pathogenesis of insulin-dependent diabetes mellitus. New England Journal of Medicine. 1994 Nov 24;331(21):1428-36.
2. McCulloch DK, Palmer JP. The Appropriate Use of B-cell Function Testing in the Preclinical Period of Type 1 Diabetes. Diabetic medicine. 1991 Nov 1;8(9):800-4.
3. Das AK. Type 1 diabetes in India: Overall insights. Indian journal of endocrinology and metabolism. 2015 Apr;19(Suppl 1):S31.
4. Ramachandran A, Snehalatha C, Krishnaswamy CV, Group MI. Incidence of IDDM in children in urban population in Southern India. Diabetes research and clinical practice. 1996 Oct 1;34(2):79-82.
5. Kalra S, Kalra B, Sharma A. Prevalence of type 1 diabetes mellitus in Karnal district, Haryana state, India. Diabetology & metabolic syndrome. 2010 Mar 9;2(1):14.
6. Kumar P, Krishna P, Reddy SC, Gurappa M, Aravind SR, Munichoodappa C. Incidence of type 1 diabetes mellitus and associated complications among children and young adults: results from Karnataka Diabetes Registry 1995-2008. Journal of the Indian Medical Association. 2008 Nov;106(11):708-11.
7. Felner EI, Klitz W, Ham M, Lazaro AM, Stastny P, Dupont B, White PC. Genetic interaction among three genomic regions creates distinct contributions to early-and late-onset type 1 diabetes mellitus. Pediatric diabetes. 2005 Dec 1;6(4):213-20.
8. Durruty P, Ruiz F, de los Rios MG. Age at diagnosis and seasonal variation in the onset of insulin-dependent diabetes in Chile (Southern Hemisphere). Diabetologia. 1979 Dec 1;17(6):357-60.
9. Elamin A, Omer MI, Zein K, Tuvemo T. Epidemiology of childhood type I diabetes in Sudan, 1987–1990. Diabetes care. 1992 Nov 1;15(11):1556-9.
10. Dabelea D, Bell RA, D'Agostino Jr RB, Imperatore G, Johansen JM, Linder B, Liu LL, Loots B, Marcovina S, Mayer-Davis EJ, Pettitt DJ. Incidence of diabetes in youth in the United States. Jama. 2007 Jun;297(24):2716-24.
11. Davies JL, Kawaguchi Y, Bennett ST, Copeman JB, Cordell HJ, Pritchard LE, Reed PW, Gough SC, Jenkins SC, Palmer SM. A genome-wide search for human type 1 diabetes susceptibility genes. Nature. 1994 Sep 8;371(6493):130.
12. McDevitt HO. The role of MHC class II molecules in susceptibility and resistance to autoimmunity. Current opinion in immunology. 1998 Dec 1;10(6):677-81.
13. Saiki RK, Bugawan TL, Horn GT, Mullis KB, Erlich HA. Analysis of enzymatically amplified β-globin and HLA-DQα DNA with allele-specific oligonucleotide probes. Nature. 1986 Nov 13;324(6093):163-6.
14. Erlich H, Bugawan T, Begovich AB, Scharf S, Griffith R, Saiki R, Higuchi R, Walsh PS. HLA-DR, DQ and DP typing

Using PCR amplification and immobilized probes. International Journal of Immunogenetics. 1991 Feb 1;18(1-2):33-55.

15. Bugawan TL, Erlich HA. Rapid typing of HLA-DQB1 DNA polymorphism using nonradioactive oligonucleotide probes and amplified DNA. Immunogenetics. 1991 May 1;33(3):163-70.
16. Bugawan TL, Apple R, Erlich HA. A method for typing polymorphism at the HLA-A locus using PCR amplification and immobilized oligonucleotide probes. Tissue antigens. 1994 Sep 1;44(3):137-47.
17. Syed M, A. Z., Omer, M. and Farhat, A. B. Comparison of methods for DNA extraction in a low cost laboratory setting for developing countries. Pak J pathol. 2008; 19 (1) : 6-11
18. J. Sambrook, E. F. Fritsch, T. Maniatis. Molecular cloning: a laboratory manual., 1989, No. Ed. 2, xxxviii + 1546 pp.
19. Ole Olerup, Henrik Zetterquist. HLA-DR typing by PCR amplification with sequence-specific primers (PCR-SSP) in 2 hours: An alternative to serological DR typing in clinical practice including donor-recipient matching in cadaveric transplantation. Tissue Antigens . 1992 May;39(5):225-35.
20. M Bunce , C M O'Neill, M C Barnardo, P Krausa, M J Browning, P J Morris, K I Welsh. Phototyping: comprehensive DNA typing for HLA-A, B, C, DRB1, DRB3, DRB4, DRB5 & DQB1 by PCR with 144 primer mixes utilizing sequence-specific primers (PCR-SSP). Tissue Antigens. 1995 Nov;46(5):355-67.
21. Kanga U, Vaidyanathan B, Jaini R, Menon PS, Mehra NK. HLA haplotypes associated with type 1 diabetes mellitus in North Indian children. Human immunology. 2004 Jan 31;65(1):47-53.
22. Park Y. Why is type 1 diabetes uncommon in Asia?. Annals of the New York Academy of Sciences. 2006 Oct 1;1079(1):31-40.
23. Rajalingam R, Ge P, Reed EF. A sequencing-based typing method for HLA-DQA1 alleles. Human immunology. 2004 Apr 30;65(4):373-9.
24. Rani R, Sood A, Goswami R. Molecular basis of predisposition to develop type 1 diabetes mellitus in North Indians. Tissue Antigens. 2004 Aug 1;64(2):145-55.
25. Zamani M, Pociot F, Spaepen M, Raeymaekers P, Nerup J, Cassiman JJ. Linkage and association of the HLA gene complex with IDDM in 81 Danish families: strong linkage between DR beta 1Lys71+ and IDDM. Journal of medical genetics. 1996 Nov 1;33(11):899-905.
26. Ueda H, Howson JM, Esposito L, Heward J, Chamberlain G, Rainbow DB, Hunter KM, Smith AN, Di Genova G, Herr MH, Dahlman I. Association of the T-cell regulatory gene CTLA4 with susceptibility to autoimmune disease. Nature. 2003 May 29;423(6939):506-11.