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Epidemiology of childhood-onset type 1 diabetes in Azerbaijan: Incidence, clinical features, biochemistry, and HLA-DRB1 status

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Abstract

Aims: Determine the incidence and typology of diabetes in children in Azerbaijan. Methods: Clinical features, C-peptide, autoantibodies (glutamic acid decarboxylase 65 (GAD65) and islet antigen 2 (IA-2)), and HLA-DRB1 status were studied in 106 subjects <18 years of age who were recently diagnosed. 104 cases were consecutive. Incidence was determined for Baku and Absheron regions, where ascertainment is estimated to be essentially 100%.

Results: 104 of the 106 (98%) were diagnosed with type 1 diabetes, one with type 2 diabetes and one with atypical diabetes. Type 1 diabetes incidence in Baku City and Absheron was 7.05 per 100,000 population <15 years per year. Peak age of onset was 10 years. There was a slight male preponderance (male:female 1.17:1), and no temporal association with seasons.

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Almost all type 1 diabetes subjects presented with classic symptoms including a high incidence (58%) of diabetic ketoacidosis. 86% presented with low C-peptide values (<0.13 nmol/L, <0.40 ng/mL) and 74% were positive for at least one type 1 diabetes-related autoantibody.

Conclusions: Azerbaijan has a moderate type 1 diabetes incidence and clinical, biochemical and genetic features similar to that in European populations.

Keywords

Childhood diabetes; Azerbaijan; Incidence; Autoimmunity; HLA

1 Introduction

Various types of diabetes are diagnosed in childhood. The most common is type 1 diabetes, but type 2 diabetes, monogenic diabetes, and other forms also occur [1]. The observed incidence of type 1 diabetes in children varies considerably around the globe, from >60 to <1 case per 100,000 children <15 years of age in nations such as Finland and Venezuela, respectively [1]. This is thought to be due to both genetic and environmental factors, which are not fully understood [2].

However, little is known regarding the clinical characteristics of diabetes in Azerbaijan, a landlocked upper-middle income country on the eastern edge of Europe. Indeed, there are no previous published reports on incidence, prevalence, or the types of diabetes occurring in children in Azerbaijan aside from one publication in 2006 on genetic associations [3].

In order to further understand the patterns and aetiology of diabetes in children in Azerbaijan, we conducted a prospective study of consecutive new cases of diabetes diagnosed in children and adolescents <18 years at a major diabetes centre in the capital, Baku City. This study investigated the demographic, clinical, immunological and biochemical features of diabetes patients, as well as HLA-DRB1 alleles of subjects enrolled in this cohort. This was performed with the belief that improved diagnosis of the disease could lead to marked improvements in disease management.

2 Materials and methods

2.1 Study site

The study was conducted at The Endocrine Centre and 6th Children's Hospital in Baku City, Azerbaijan. All procedures were approved by relevant Ethics Committees in Azerbaijan the United States, and Australia, and performed in accordance with the Declaration of Helsinki. Written informed consent was obtained for all subjects prior to enrolment in the study. HLA genotyping was performed at Children's Hospital Oakland Research Institute with IRB approval.

2.2 Study subjects

A total of 106 subjects <18 years of age at diabetes diagnosis were enrolled. The subjects included two diagnosed in November 2013 and then a consecutive series of all subjects seen at the study institutions that were diagnosed from the 1st March 2014 to 5th March 2015.

The study institutions act as referral centres for the entire country. Date of assessment ranged from March 2014 to March 2015 with 48 (45%) study subjects assessed on the same day as diagnosis, 39 (37%) within one week, 10 (9%) within one week to one month, 8 (8%) within one to six months, and one (1%) after six months (maximum 7 months). 200 control subjects were enrolled from the general population. Control subjects were non-diabetic and unrelated to the subjects with diabetes or to each other.

2.3 Demographic data

Date of birth, sex, ethnicity, city and province of residence at time of diabetes diagnosis, date of diagnosis, as well as distance and travel time to The Endocrine Centre or 6th Children's Hospital were recorded.

2.4 Clinical parameters

Diabetes was diagnosed according to standard World Health Organisation criteria [4]. Determination of the type of diabetes was made by the local investigator according to clinical features and history. The presence of polyuria, polydipsia, weight loss, malnutrition and ketoacidosis at the time of diagnosis were recorded. Ketoacidosis was defined as presence of hyperglycemia (blood glucose > 11 mmol/l (198 mg/dl), ketone bodies in blood and/or urine, and clinical features such as Kussmaul respirations and dehydration.

The following information pertaining to diabetes care was also recorded for each subject: date of insulin commencement, commencement daily dosage of insulin and number of injections per day, type of insulin used, insulin storage method at homes, use of oral hypoglycaemic agents, and other medications or treatment. History of other medical conditions, and family history was also recorded. Body weight and height were measured by electronic scale and stadiometer respectively with subjects wearing light-weight clothing and without shoes. Body Mass Index (BMI) was then calculated. BMI standard deviation (SD) scores were calculated using the WHO standards for <5 years [5] and for >5–19 years of age [6].

2.5 Sample collection

Peripheral blood was collected by venepuncture into vacutainer tubes on the day of assessment, after an overnight fast. Serum samples were spun down immediately and stored in a –20 °C freezer. For HLA genotyping, 40–200 μl of blood from each diabetes subject were deposited into vials containing DNAgard Blood (BioMatrica Inc. San Diego, CA USA). The stabilization reagent was resuspended in the blood, then the mixture was dried before shipping. Approximately one millilitre of saliva was collected from each control subject and mixed with 0.5 ml of DNAgard Saliva (BioMatrica Inc. San Diego, CA USA) prior to shipping.

2.6 Biochemical parameters and serology

For diabetes subjects, blood glucose was measured by Bioscreen MS-200 (Erba Lachema, Brno, Czech Republic). HbA1c was measured using a Clover A1c analyser (Infopia, Anyang Gyeonggi-do, Republic of Korea). C-peptide was measured in Azerbaijan by ELISA (IBL, Hamburg, Germany) within 72 h of blood collection. Glutamic acid decarboxylase 65

(GAD65) and islet antigen 2 (IA-2) autoantibodies, were measured from frozen serum samples by commercially available ELISA kits (IBL, Hamburg, Germany) in Azerbaijan. GAD65 and IA-2 autoantibodies were considered positive if levels were   30 IU/mL, according to manufacturer's recommendation. While not directly entered from this site, similar ELISA formats have been challenged in Islet Autoantibody Standardization Programs with comparable sensitivity and specificity to radioimmunoassays [7].

2.7 HLA alleles

DNA was extracted from blood samples of 106 children with diabetes and also from saliva (BioMatrica, Inc. San Diego, CA USA) from 209 controls. The samples were genotyped for HLA alleles with high-resolution genotyping technology at Children's Hospital Oakland Research Institute in California. PCR products (amplicons) were generated from genomic DNA using DRB generic exon 2 454 fusion primers. The 454 fusion primers consist of a locus-specific primer on the 3′ end, a 10-bp multiplex ID (MID) tag, and an "A" or "B" 454-specific primer sequence on the 5′ end. The MID tag serves as a sample barcode recognized by the Conexio ASSIGN™ ATF genotyping software (version 1.1.0.35, Conexio Genomics, Freemantle, Western Australia). Amplicons were purified with AMPure beads (Becton Dickinson, Franklin Lakes, USA), quantified using the Quant-iT PicoGreen dsDNA reagent (Life Technologies, Foster City, USA), and mixed with capture beads after dilution. Individual DRB exon 2 amplicon molecules were captured by these beads, amplified in an emulsion PCR and DNA-containing beads, and subsequently analyzed by pyrosequencing to obtain sequence readings originating from a single molecule [8 9]. HLA sequence data were generated using next-generation sequencing on the Roche 454 GS Junior System (Roche, Basel, Switzerland) and analyzed using Conexio ASSIGN™ ATF to interpret HLA genotypes from the sequence files [8 9].

2.8 Statistics

Statistics were performed and graphs created using Excel software. Locus-level tests of heterogeneity and variant-level chi-squared ($\chi 2$) tests of association between 104 type 1 diabetes subjects and 200 control subjects were performed for the HLA-DRB1 locus using BIGDAWG R package [10]. Hardy-Weinberg equilibrium (HWE) proportions of HLA-DRB1 genotypes in type 1 diabetes subjects and control subjects were tested using PyPop (v0.8.0) [11]. We tested the significance of locus-level HWE deviations using Guo and Thompson's exact method [12], and identified individual genotypes deviating significantly from HWE expectations using Chen's method [13 14], using a threshold of significance of 0.05.

3 Results

3.1 Diagnosis

104 of the 106 enrolled diabetes patients were diagnosed as having type 1 diabetes. One female who presented at 8.1 years, with overweight, a positive family history, and acanthosis nigricans was diagnosed with type 2 diabetes. One female who presented with typical symptoms and ketonuria aged 11.6 years was diagnosed as having atypical diabetes as there

was no overweight, and insulin was discontinued after three months and she was managed on glimepiride.

3.2 Incidence

During the 12 month period March 2014 to February 2015, 100 subjects were diagnosed with new onset type 1 diabetes, with 91 (91%) of these <15 years of age. Forty-three of these were living in the Baku region (the capital) or the region of Absheron which surrounds Baku. Using Azerbaijan government population data [15], Baku and Absheron were calculated to have an incidence of 7.05 per 100,000. Ascertainment in these two regions is thought to be 100% or very close to that, as all new diagnoses in these two regions are referred to Children's Hospital No. 6.

3.3 Demographic characteristics

Of the 104 subjects with type 1 diabetes, 56 (54%) were males and 48 (46%) were females. There was no discernible seasonal pattern in disease incidence (data not shown). All subjects were ethnic Azeris. The mean ± SD age of type 1 diabetes diagnosis was 8.9 ± 4.4 years (range 1.0–17.3 years; Fig. 1). The median age at diagnosis was 9.3 years and the peak age of onset was 10 years. 25.0% were diagnosed at 0–4 years, 32.7% from 5 to 9 years, 33.7% from 10 to 14 years and 8.7% from 15 to 17 years of age. When examining all enrolled subjects with diabetes (n = 106), 13 (12%) subjects lived <10 km from the hospital, 29 (27%) between 50 and 100 km, 13 (12%) 100–200 km, and 51 (48%) travelled more than 200 km to access tertiary care.

3.4 Clinical parameters

3.4.1 Type 1 diabetes cases (n = 104)—The main symptoms preceding diagnosis of type 1 diabetes were polyuria (n = 104, 100%), polydipsia (n = 104, 100%) and weight loss (n = 103, 99%). Sixty (58%) subjects presented in diabetic ketoacidosis (DKA). DKA rates were 42.3% in the 0–4 year age group, 84.8% in the 5–9 year group, 52.8% in the 10–14 year group and 33.3% in the 15–19 year group. For those living in Baku City, the rates were 61.7% as compared to 54.4% outside the capital. Table 1 shows the relationships of DKA, C-peptide, and autoantibodies.

For 104 patients diagnosed with type 1 diabetes, the mean ± SD BMI was 15.5 ± 2.7 (range 9.9–30.7). BMI SD scores ranged from −6.10 to 2.61 (mean = −1.06). Three subjects had a BMI SD > 2, with a range of 2.07–2.61; all three had at least one type 1 diabetes-related autoantibody. Two subjects had a BMI SD of <5.0, ranging from −5.53 to −6.10. Both had at least one type 1 diabetes-related autoantibody and no signs of malnutrition. The mean ± SD blood glucose at type 1 diabetes diagnosis was 24.0 ± 8.0 mmol/L (range 9.4–44.4 mmol/L). Mean ± SD HbA1c was 12.2 ± 1.7% (108.7 ± 19.1 mmol/mol) with a range 5.5–14.0% (range 36.6–129.5 mmol/mol).

Three subjects (3%) had other significant medical conditions. One was diagnosed with both diabetes insipidus and type 1 diabetes at 2.3 years of age. This child was GAD65 positive, with no optic atrophy or deafness at the time of the study (Wolfram's syndrome may still be possible). Another patient had Koolen-de Vries syndrome (17q21.31) and was positive for

both type 1 diabetes–related autoantibodies. The third subject had Glucose-6-Phosphate Dehydrogenase Deficiency. Seven subjects with type 1 diabetes (7%) had a history of type 1 diabetes in the family: aunt (two subjects), sister (two), father (one), uncle (one) and cousin (one).

All type 1 subjects were treated with insulin. 60 (58%) were on long acting insulin, 61 (59%) on short acting insulin, and 44 (42%) were on analogue insulin. 51 (49%) commenced on five injections per day, 45 (43%) four, and (8%) three. All but one subject had a refrigerator at home for insulin storage.

3.4.2 Other types of diabetes (n = 2)—The subject with type 2 diabetes (BMI SD 3.36) was treated with metformin and long acting insulin (one injection per day). The subject with atypical diabetes (BMI SD −1.13) was treated with glimepiride.

3.5 C-peptide

For the 104 type 1 diabetes patients, the mean ± SD fasting C-peptide was 0.11 ± 0.10 nmol/L (0.32 ± 0.31 ng/mL). Forty subjects (38.5%) had C-peptide 0.07 nmol/L (0.20 ng/mL). Eighty-nine subjects (86%) had values < 0.13 nmol/L (<0.40 ng/mL), eight (8%) between 0.13 and 0.26 nmol/L (0.40–0.80 ng/mL) and seven (7%) between 0.26 and 0.56 nmol/L (0.80–1.70 ng/mL) (maximum C-peptide was 0.56 nmol/L (1.70 ng/mL)) (Fig. 2). The relationship of C-peptide results with autoantibodies and the presence of DKA are shown in Table 1. C-peptide was 0.07 nmol/L (0.20 ng/mL) in 18.6% of the 59 subjects aged 0–9 years, and 64.4% of the 45 subjects aged 10–19 years. The subject with type 2 diabetes had a C-peptide level of 0.53 nmol/L (1.60 ng/mL), and the subject with atypical diabetes had 0.06 nmol/L (0.18 ng/mL) circulating C-peptide.

3.6 Autoantibody status

For the 104 subjects with type 1 diabetes, 41 (39%) were IA-2 positive, 64 (62%) were GAD65 positive, 77 (74%) had either or both antibodies, and 28 (27%) were positive for both (see Table 1 for other relationships). GAD65 positivity was found in 74.6% of the 59 subjects aged 0–9 years, and 46.7% of the 45 subjects aged 10–19 years. Among autoantibody positive subjects, the mean ± SD titer for IA-2 was 112.3 ± 186.9 IU/mL and for GAD65 was 138.6 ± 168.5 IU/mL.

Autoantibodies were not detected in the subject with type 2 diabetes, but the subject with atypical diabetes was positive for IA-2 autoantibodies.

3.7 HLA results

While the DRB1 genotypes of the 200 control subjects conformed to expected HWE proportions, those of the 104 diabetes subjects deviated significantly from HWE (p-value < 1E-05). The primary contributor to this deviation was an excess of DRB1*03:01 + DRB1*04:02 heterozygotes (24 observed; 15 expected; p-value = 3.4E-03). DRB1*04:05 homozygotes were also observed to be in excess (3 observed; 0.5 expected; p-value = 7.1E-03), as were DRB1*09:01 + DRB1*07:01 heterozygotes (3 observed; 0.25 expected; p-

value = 6.0E-04). In addition, the absence of DRB1*07:01 + DRB1*03:01 heterozygotes was also significant (0 observed; 3.75 expected; p-value = 1.48E-02).

Genotyping of the DRB1 locus identified 38 alleles present in the population. Association analysis with the BIGDAWG [10] revealed that 14 of these alleles were present in sufficient frequency to assess T1D association (Table 2). The remaining 24 alleles were binned for association analyses. Significant heterogeneity was observed between diabetes subjects and control subjects at the locus level (p-value < 2.22E-16). DRB1*03:01 and DRB1*04:02 showed the strongest positive association with disease (OR 5.06 and 4.47; p = 7.77E-13 and 2.27E-10, respectively). DRB1*04:05 was also positively associated (OR 3.53; p = 1.90E-03). Six of the remaining 11 alleles showed negative disease association (protection), including alleles in the "DR2" group, DRB1*15:01 and DRB1*15:02.

4 Discussion

This study investigated the incidence as well as clinical, biochemical, and genetic characteristics of new onset diabetes in children in Azerbaijan.

No type 1 diabetes incidence data have been published previously from Azerbaijan. The 2015 International Diabetes Federation (IDF) Atlas [16] estimated a type 1 diabetes incidence of 7.0 per 100,000 <15 years of age and a prevalence of 928 cases in those <15 years of age. These numbers are based on extrapolation from data [17] from the neighbouring country of Armenia. However, the Azeris are a Turkic population [18]; hence, comparisons with other Turkic populations may be more appropriate. Data exists for two such populations, both nationwide – type 1 diabetes incidence of 10.8 per 100,000 children <18 years of age per year was reported in Turkey 2011–2103 [19], and 3.8 per 100,000 <15 years of age per year in Uzbekistan in 2014 [20]. The incidence found in the current study was 7.05 per 100,000.

It is also possible that a few cases die at type 1 diabetes onset and are misdiagnosed with another condition. Indeed, this is thought to occur in a number of less-resourced countries, including Azerbaijan [21], although it would be expected to be less likely in and near the capital. Almost all (98%) subjects in this study were diagnosed with type 1 diabetes. The pattern of type 1 diabetes of Azerbaijan is similar to that of European [1] and Turkic [19 20] populations in terms of the peak age of onset and corresponds with an almost uniform presence of classic symptoms [1]. No seasonal pattern was identified in this study, unlike a previous report from Turkey where onset was more common in winter and autumn [22]. A small female preponderance has been found in Turkey [19] and particularly in Uzbekistan [20]. However, this Azerbaijan data showed a slight male excess, in agreement with the prior published series from Azerbaijan [3].

The frequency of DKA at diagnosis varies widely around the world from 12 to 80% [23]. This study's rate of 57.7% is therefore relatively high on an international scale, although consistent with published rates of 50.8% [22] and 65.9% [24] from Turkey. Such high rates are of concern as there is a significant risk of death from DKA [25], and recent data suggest the possibility of long-term sequelae [26].

In this study, almost half of subjects with diabetes (48%) travelled >200 km to access tertiary care. Rates of DKA were similar between those diagnosed in Baku City and those outside the capital. Successful educational interventions have reduced the incidence of DKA in Italy [27] and Australia [28]. A poster campaign focused on health clinics and hospitals was distributed in Azerbaijan in 2012–13. This campaign used the six-icon poster developed by the IDF Life for a Child Program and the International Society for Pediatric and Adolescent Diabetes) [29]. Further awareness of the presenting symptoms and signs of diabetes in young people is needed in Azerbaijan to reduce these rates.

In this study, 61.5% of type 1 diabetes patients were GAD65 autoantibody positive, and 39.4% were IA-2 autoantibody positive while 26.9% had both autoantibodies. C-peptide levels were low [30] in almost all type 1 diabetes cases (86% <0.13 nmol/L [<0.40 ng/mL]). Limited comparable information is available from other Turkic populations. In Turkey, Karaguzel et al. [31] found that 63% of children with type 1 diabetes at a mean age of 11.7 years and mean duration of 3.4 years were GAD65 autoantibody positive. Marandi et al. [32] found that 27.6% of newly diagnosed type 1 diabetes subjects aged under 20 years had antibodies against GAD65, and 94.1% of subjects had a C-peptide of <0.17 nmol/L (<0.50 ng/mL) in a mainly Azeri population in Northwest Iran.

Rates of GAD65 autoantibody positivity and C-peptide values 0.07 nmol/L (0.20 ng/mL) in the current study were remarkably similar to those observed in the SEARCH Study in the United States [33]. Indeed, rates of positivity for autoantibodies against GAD65 were 61.5% and 61.4%, respectively, and rates of low to undetectable C-peptide (0.07 nmol/L [0.20 ng/mL]) were 38.5% and 38.4%, respectively.

HLA genotyping of this population reveals DRB1 alleles common to European descent populations as well as alleles common to Asian populations. The pattern of T1D association, showing disease susceptibility from "DR3" and "DR4" alleles, with protection from "DR2" alleles, is not unexpected when compared to most studies of European populations [34]. Notable, however, is the fact that the "DR4" allele that is most predisposing is DRB1*04:02, rather than the DRB1*04:01 allele common in Europeans. Azeri diabetes subjects heterozygous for the DRB1*03:01 and DRB1*04:02 alleles and homozygous for the DRB1*04:05 allele are more common than expected, given the frequencies of these predisposing alleles in the Azeri population, suggesting a role for these specific allele-combinations in diabetes. Also notable is the presence of the common European "DR2" DRB1*15:01 and the common Asian "DR2" DRB1*15:02 in the same population. This allows direct comparison of the two alleles in a single population and shows that both alleles are extremely protective for T1D.

A previous report of HLA association with type 1 diabetes, from the same diabetes center in Azerbaijan, was consistent with these results [3]. Low-resolution HLA genotyping of DQA1 and DQB1 loci showed the strongest predisposing T1D effects for DQB1*02 (commonly found in haplotype with DRB1*03:01), DQA1*03, DQB1*03:02, and DQB1*03:04 (all commonly found in haplotype with DRB1*04 alleles. DQB1*06:02, DQB1*05:03, and DQB1*06:01, all of which are found in haplotype with DRB1*15:xx or 16:xx alleles were found to be T1D protective, as was DQB1*03:01, also protective in European populations.

A small study with low-resolution genotyping on patients and controls from southeast Turkey reported a strong predisposing effect for DQB1*02 and for the presumed haplotype DRB1*03-DQB1*02 [35]. DQB1*03 was reported as significantly protective; however, the low-resolution genotyping of the study precluded distinction of the DQB1*03:01 (commonly type 1 diabetes protective) from the DQB1*03:02 (commonly type 1 diabetes predisposing) allele. Thus, the DRB1 results from this study are not inconsistent with lower-resolution genotyping results reported for similar populations.

Of note, there was one subject (0.9%) who presented with type 2 diabetes and one subject (0.9%) with atypical diabetes. This latter case may prove to be type 1 diabetes with time, or may be monogenic in cause, but genetic testing has not been carried out. No cases were diagnosed prior to 1 year of age. Importantly, in this study, only two autoantibodies could be measured due to study funding; hence, there is a need to further evaluate the Azerbaijan population for additional type 1 diabetes-related autoantibodies including those against zinc transporter 8 (ZnT8) and insulin. In a handful of cases, we were unable to measure autoantibodies immediately at diagnosis, but the maximum duration of type 1 diabetes prior to study enrolment was seven months, which is unlikely to have significantly influenced results. HLA genotyping was resource-limited to a single locus (DRB1) for this report. More extensive analyses of HLA and minor type 1 diabetes susceptibility alleles [2] will reveal additional information regarding disease risk and heritability in Azerbaijan, and we intend to conduct these studies in the future.

5   Conclusions

Diabetes in children in Azerbaijan is mainly type 1, although other types can occur. The incidence rate is mid-range in global terms. Clinical features at onset, autoimmunity, residual insulin secretion and HLA-DRB1 status are similar to European populations. It is our belief that data reported herein will likely aid future progress toward diabetes prediction, management, and education in Azerbaijan and other Turkic nations.

Data availability

The HLA-DRB1 data represent the first step of the comprehensive HLA class I and class II genotyping planned for this sample set. With the approval of the in-country investigators, and after publication of the data, the de-identified HLA will be provided to publicly available HLA databases, including allelefrequencies.net.

Acknowledgements

Literature Cited

[1]. Craig ME, Jefferies C, Dabelea D, Balde N, Seth A, and Donaghue KC: Definition, epidemiology, and classification of diabetes in children and adolescents. Pediatr Diabetes 2014; 15: pp. 4–17 [PubMed: 25182305]

[2]. Katsarou A, Gudbjörnsdottir S, Rawshani A, Dabelea D, Bonifacio E, Anderson BJ, et al.: Type 1 diabetes mellitus. Nat Rev Dis Primers 2017; 30: pp. 17016

[3]. Ahmedov G, Ahmedova L, Sedlakova P, and Cinek O: Genetic association of type 1 diabetes in an Azerbaijanian population: the HLA-DQ,-DRB1*O4, the insulin gene, and CTLA4. Pediatr Diabet 2006; 7: pp. 88–93

[4]. World Health Organization, & International Diabetes Federation. Definition and diagnosis of diabetes mellitus and intermediate hyperglycaemia; 2006 <http://www.who.int/diabetes/publications/Definition%20and%20diagnosis%20of%20diabetes_new.pdf> [retrieved February 11, 2018].

[5]. WHO Multicentre Reference Study Group: WHO child growth standards: length/height-for-age, weight-for-age, weight-for-length, weight-for-height and body mass index-for-age: methods and development. Acta Paediatr Suppl 2006; 450: pp. 76–85 [PubMed: 16817681]

[6]. de Onis M, Onyango AW, Borghi E, Siyam A, Nishida C, and Siekmann J: Development of a WHO growth reference for school-aged children and adolescents. Bull World Health Organ 2007; 85: pp. 660–667 [PubMed: 18026621]

[7]. Wasserfall C, Montgomery E, Yu L, Michels A, Gianani R, Pugliese A, et al.: Validation of a rapid type 1 diabetes autoantibody screening assay for community-based screening of organ donors to identify subjects at increased risk for the disease. Clin Exp Immunol 2016; 185: pp. 33–41 [PubMed: 27029857]

[8]. Bentley G, Higuchi R, Hoglund B, et al.: High-resolution, high-throughput HLA genotyping by next-generation sequencing. Tissue Antigens 2009; 74: pp. 393–403 [PubMed: 19845894]

[9]. Holcomb CL, Höglund B, Anderson MW, et al.: A multi-site study using high-resolution HLA genotyping by next generation sequencing. Tissue Antigens 2011; 77: pp. 206–217 [PubMed: 21299525]

[10]. Pappas DJ, Marin W, Hollenbach JA, and Mack SJ: Bridging immunogenomic data analysis workflow gaps (BIGDAWG): an integrated case-control analysis pipeline. Hum Immunol 2016; 77: pp. 283–287 [PubMed: 26708359]

[11]. Lancaster AK, Single RM, Solberg OD, Nelson MP, and Thomson G: PyPop update–a software pipeline for large-scale multilocus population genomics. Tissue Antigens 2007; 69: pp. 192–197 [PubMed: 17445199]

[12]. Guo SW, and Thompson EA: Performing the exact test of Hardy-Weinberg proportion for multiple alleles. Biometrics 1992; 48: pp. 361–372 [PubMed: 1637966]

[13]. Chen JJ, and Thomson G: The variance for the disequilibrium coefficient in the individual Hardy-Weinberg test. Biometrics 1999; 55: pp. 1269–1272 [PubMed: 11315081]

[14]. Chen JJ, Hollenbach JA, Trachtenberg EA, et al.: Hardy-Weinberg testing for HLA class II (DRB1, DQA1, DQB1, and DPB1) loci in 26 human ethnic groups. Tissue Antigens 1999; 54: pp. 533–542 [PubMed: 10674966]

[15]. The State Statistical Committee of the Republic of Azerbaijan. Population; 2017, 7 26 <https://www.stat.gov.az/source/demoqraphy/?lang=en> [retrieved February 10, 2018].

[16]. International Diabetes Federation. IDF diabetes Atlas, 7th ed Brussels: International Diabetes Federation; 2015.

[17]. Navasardyan LV: Epidemiology of type 1 diabetes in children and adolescents in the Republic of Armenia. New Armenian Med J 2014; 8: pp. 89–94

[18]. West BA: Azerbaijanis: nationality. Encyclopedia of the Peoples of Asia and Oceania. Infobase 2009; undefined: pp. 68–72

[19]. Yesilkaya E, Cinaz P, Andiran N, Bideci A, Hatun S, Sari E, et al.: First report on the nationwide incidence and prevalence of type 1 diabetes among children in Turkey. Diabet Med 2016; undefined:

[20]. Rakhimova GN, Alimova NU, Ryaboshtan A, Waldman B, Ogle GD, and Ismailov SI: Epidemiological data of type 1 diabetes mellitus in children in Uzbekistan, 1998–2014. Pediatr Diabetes 2017; undefined:

[21]. Ogle GD, Middlehurst AC, and Silink M: The IDF life for a child program index of diabetes care for children and youth. Pediatr Diabetes 2016; 17: pp. 374–384 [PubMed: 26153340]

[22]. Ardicli D, Kandemir N, Alikasifoglu A, Ozon A, and Gonc N: Clinical characteristics of type 1 diabetes over a 40 year period in Turkey: secular trend towards earlier age of onset. J Pediatr Endocrinol Metab 2014 7; 27: pp. 635–641 [PubMed: 24706426]

[23]. Usher-Smith JA, Thompson M, Ercole A, and Walter FM: Variation between countries in the frequency of diabetic ketoacidosis at first presentation of type 1 diabetes in children: a systematic review. Diabetologia 2012; 55: pp. 2878–2894 [PubMed: 22933123]

[24]. Demirbilek H, Özbek MN, and Baran RT: Incidence of type 1 diabetes mellitus in Turkish children from the south-eastern region of the country: a regional report. J Clin Res Pediatr Endocrinol 2013; 5: pp. 98–103 [PubMed: 23748062]

[25]. Rewers A: Current concepts and controversies in prevention and treatment of diabetic ketoacidosis in children. Curr Diab Rep 2012; 12: pp. 524–532 [PubMed: 22864672]

[26]. Cameron FJ, Scratch SE, Nadebaum C, Northam EA, Koves I, Jennings J, et al.: Neurological consequences of diabetic ketoacidosis at initial presentation of type 1 diabetes in a prospective cohort study of children. Diabetes Care 2014; 37: pp. 1554–1562 [PubMed: 24855156]

[27]. Vanelli M, Chiari G, Ghizzoni L, et al.: Effectiveness of a prevention program for diabetic ketoacidosis in children. An 8 year study in schools and private practices. Diabetes Care 1999; 22: pp. 7–9 [PubMed: 10333896]

[28]. King BR, Howard NJ, Verge CF, et al.: A diabetes awareness campaign prevents diabetic ketoacidosis in children at their initial presentation with type 1 diabetes. Pediatr Diabetes 2012; 13: pp. 647–651 [PubMed: 22816992]

[29]. International Diabetes Federation, & Life for a Child. DKA prevention; n.d. < http://www.lifeforachild.org/about/education-resources/dka-prevention.html> [retrieved December 10, 2017].

[30]. Jones AG, and Hattersley AT: The clinical utility of C-peptide measurement in the care of patients with diabetes. Diabet Med 2013; 30: pp. 803–817 [PubMed: 23413806]

[31]. Karagüzel G, im ek S, De er O, and Ökten A: Screening of diabetes, thyroid, and celiac diseases-related autoantibodies in a sample of Turkish children with type 1 diabetes and their siblings. Diabetes Res Clin Pract 2008; 80: pp. 238–243 [PubMed: 18241952]

[32]. Marandi LY, Rajaii M, Aliasgarzadeh A, and Sadeghi-Bazargani H: Islet cell autoantibodies in patients younger than 20 years of age with recently diagnosed diabetes in Northwest of Iran. Int J Diabetes Dev Ctries 2011; 31: pp. 70–75

[33]. Writing Group for the SEARCH for Diabetes in Youth Study Group, Dabelea D, Bell RA, D'Agostino RB, Imperatore G, Johansen JM, et al.: Incidence of diabetes in youth in the United States. JAMA 2007; 297: pp. 2716–2724 [PubMed: 17595272]

[34]. Erlich H, Valdes AM, Noble J, Carlson JA, Varney M, Concannon P, et al.: HLA DR-DQ haplotypes and genotypes and type 1 diabetes risk: analysis of the type 1 diabetes genetics consortium families. Diabetes 2008; 57: pp. 1084–1092 [PubMed: 18252895]

[35]. Keskin M, Aygün A, Pehlivan S, Keskin Ö, Kor Y, Balat A, et al.: Trends in the frequency of HLA DR-DQ haplotypes among children and adolescents with type 1 diabetes mellitus in the southeast region of Turkey. J Clin Res Pediatr Endocrinol 2012; 4: pp. 189–192 [PubMed: 23261861]
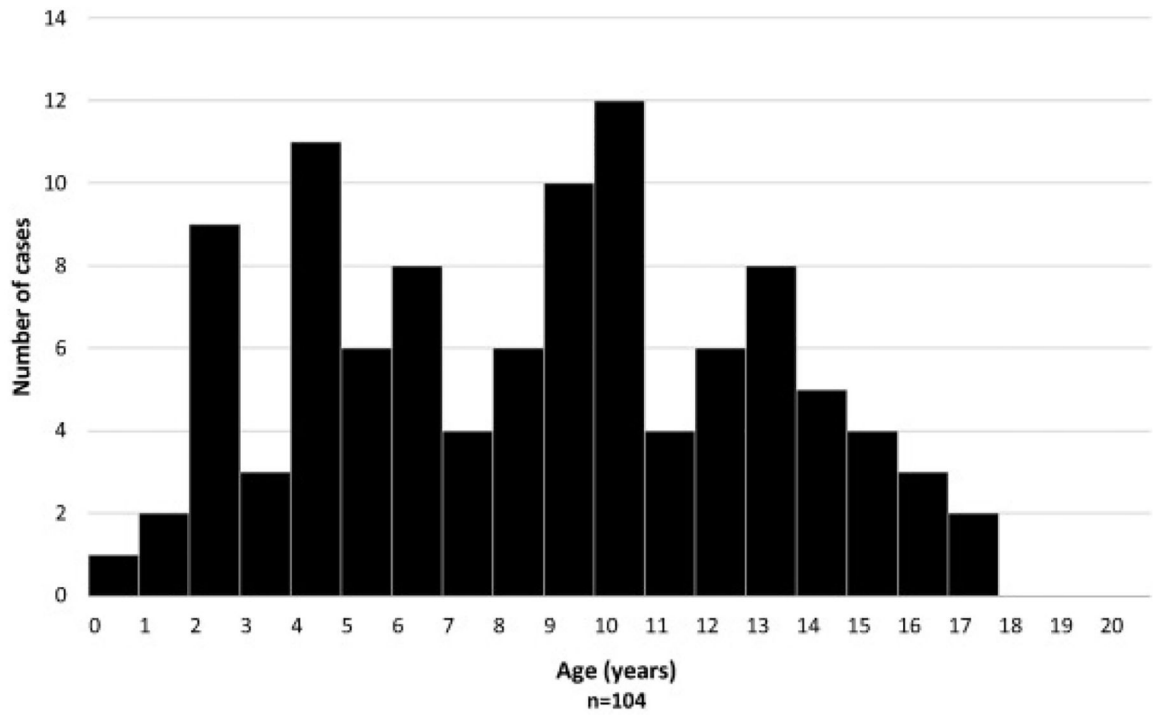
Figure 1.
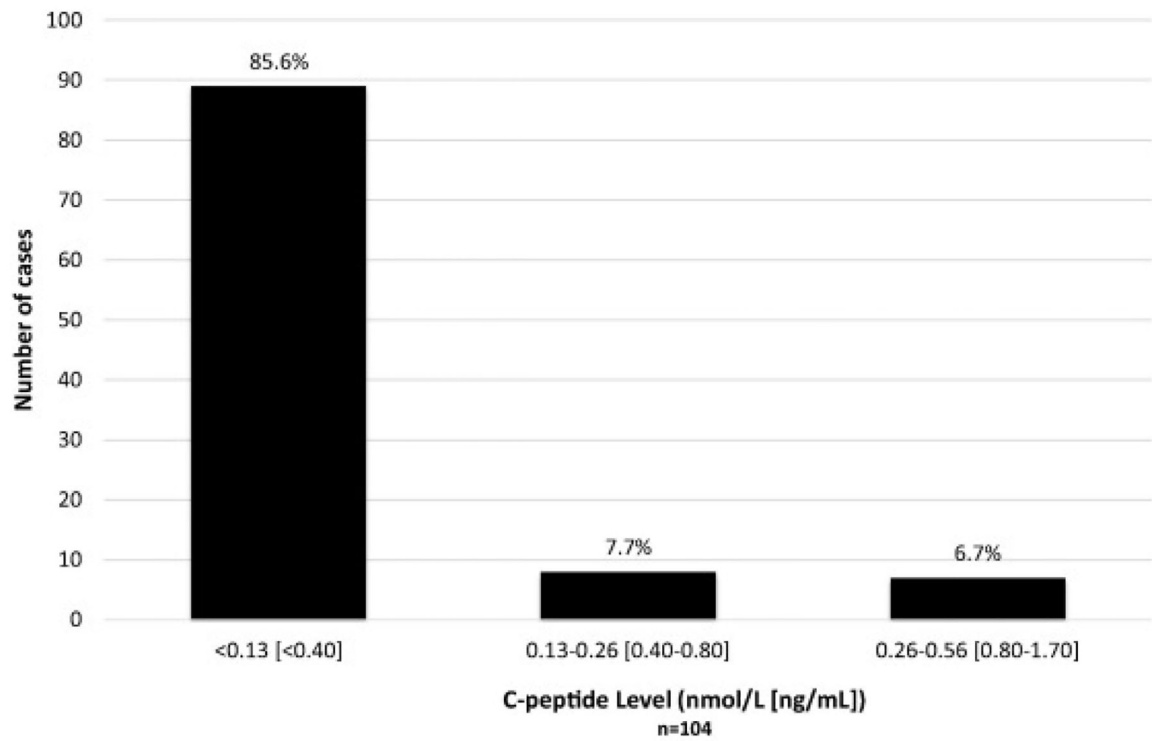The age of onset of type 1 diabetes in children <18 years in Azerbaijan.

Figure 2.
C-peptide levels at onset of type 1 diabetes.

Table 1.

The relationships of diabetic ketoacidosis, C-peptide and autoantibodies for 104 type 1 diabetes subjects.

	C-peptide	Autoantibody status						
	C-peptide < 0.13 nmol/L (<0.40 ng/mL) (n = 89)	C-peptide 0.13–0.26 nmol/L (0.40–0.80 ng/mL) (n = 8)	C-peptide 0.26–0.56 nmol/L (0.80–1.70 ng/mL) (n = 7)	GAD 30 (n = 64)	IA2 30 (n = 41)	Both autoantibodies (n = 28)	Either/both autoantibodies (n = 77)	Neither autoantibodies (n = 27)
Diabetic Ketoacidosis (n = 60)	50 (83%)	7 (12%)	3 (5%)	42 (70%)	25 (42%)	19 (32%)	48 (80%)	12 (20%)
C-peptide < 0.13 nmol/L (<0.40 ng/mL) (n = 89)				54 (61%)	33 (37%)	23 (26%)	64 (72%)	25 (28%)
C-peptide 0.13–0.26 nmol/L (0.40–0.80 ng/mL) (n = 8)				6 (75%)	4 (50%)	2 (25%)	8 (100%)	0 (0%)
C-peptide 0.26–0.56 nmol/L (0.80–1.70 ng/mL) (n = 7)				4 (57%)	4 (57%)	3 (43%)	5 (71%)	2 (29%)

Table 2.

The frequencies of HLA alleles for 104 type 1 diabetes subjects versus 209 controls.

DRB1 allele	Patient frequency (n)	Control frequency (n)	OR (95% CI)	p value	Significance
01:01	0.0385 (8)	0.0383 (16)	1.01 (0.37–2.54)	9.91E-01	NS
03:01	0.2885 (60)	0.0742 (31)	5.06 (3.08–8.4)	7.77E-13	*
04:02	0.25 (52)	0.0694 (29)	4.47 (2.67–7.57)	2.27E-10	*
04:04	0.0144 (3)	0.0311 (13)	0.46 (0.08–1.69)	2.13E-01	NS
04:05	0.0721 (15)	0.0215 (9)	3.53 (1.42–9.3)	1.90E-03	*
07:01	0.0625 (13)	0.0789 (33)	0.78 (0.37–1.56)	4.58E-01	NS
11:01	0.0144 (3	0.0789 (33)	0.17 (0.03–0.56)	1.09E-03	*
11:04	0.0144 (3)	0.0789 (33)	0.17 (0.03–0.56)	1.09E-03	*
13:01	0.0096 (2)	0.0646 (27)	0.14 (0.02–0.57)	2.05E-03	*
13:02	0.0144 (3)	0.0383 (16)	0.37 (0.07–1.31)	1.01E-01	NS
14:01	0 (0)	0.0526 (22)	0 (0–0.35)	7.56E-04	*
15:01	0.0144 (3)	0.0694 (29)	0.2 (0.04–0.65)	3.28E-03	*
15:02	0.0048 (1)	0.0646 (27)	0.07 (0–0.43)	6.53E-04	*
16:01	0.0337 (7)	0.0335 (14)	1 (0.34–2.71)	9.92E-01	NS
Binned	0.1683 (35)	0.0335 (14)	0.78 (0.49–1.23)	2.63E-01	NS