Letters

Observations

Is there relatedness between maternal lines of Type 1 diabetic patients?

To the Editor: Type 1 diabetes is a genetically complex disease, which has been clearly shown by numerous linkage and association studies. Apart from the HLA genes, non-HLA genes are also involved in diabetes development. Until now more than 20 putative diabetes-predisposing non-HLA genes have been localised [1]. Unfortunately, the overall effect of most diabetes-predisposing non-HLA genes is weak, which could be attributed to the fact that the susceptible genes occur in different and small proportions of diabetic patients who are more or less related. It is known that diabetes susceptible genetic components aggregate in families [2], and that Type 1 diabetes at least in some of its genetic components follows a recessive model, one could imagine that relatedness between parents of diabetics could indeed be favouring diabetes development. To prove this hypothesis, we analysed mitochondrial DNA (mtDNA) of unrelated parents of Type 1 diabetic chil-

dren. Unlike nuclear DNA, mtDNA is in hundreds to thousands of copies per cell, to evolve rapidly and mutate at 5 up to 10 times faster [3]. The most variable regions of the human mtDNA are the two non-encoding hypervariable regions HV1 and HV2 of the control region, which are analysed for individualisation of forensic samples [4]. To find out whether or not there is a relation between maternal lines of unrelated diabetics we compared the sequences of those control regions to study the degree of relation between the maternal lines of mothers and maternal lines of fathers.

DNA of 80 parents of 64 Type 1 diabetic children (27 male: 37 female; 2 times 3 affected sibs, 20 times 2 affected sibs, 18 times one affected child) manifested with an age of 11 ± 6 years was isolated and the hypervariable regions HV1 (nt15998–nt16394) and HV2 (nt49–nt369) of mtDNA were amplified and sequenced. All diabetic children descended from different parts of eastern Germany and were diagnosed at the Central Institute of Diabetes “Gerhardt Katsch” (formerly GDR) and Clinic for Diabetes (>1992) and were repeatedly investigated and treated for insulin-dependent diabetes in the diabetes clinic up to 1997 by experienced diabetologists. The sequences of maternal lines were analysed with the neighbourhood-joining method as implemented in PHYLIP. This method is proposed for reconstructing trees from evolutionary distance data.

The mtDNA sequences showed that the maternal lines most likely fell into only four groups with a high probability of a common ancestral mother (Fig. 1). The first group held 7, the second group 7 and the third group 11 members. The largest

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group is the fourth with a total of 46 members. Of the sequences nine were not readily assigned to any other group. The differentiation between the groups was quite clear-cut and robust to alterations in the application of the methodology. Within each group there were maternal lines, which differed only in one or two nucleotide(s) suggesting a high degree of relation.

Our findings suggest that maternal lines of Type 1 diabetic patients are related. An observation which is supported by the analysis of 109 unrelated German Caucasoid individuals where 100 different mtDNA lineages were detected [5].

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Gender-associated differences in Type 1 diabetes risk factors?

To the Editor: Several different viruses are associated with Type 1 diabetes, but no studies have observed a gender difference in the impact of infections on diabetes. Childhood Type 1 diabetes is increasing worldwide; this has prompted several study groups to search for risk factors explaining this worldwide increase. All studies investigating risk factors for Type 1 diabetes have been matched for gender, because there is a known difference in incidence between boys and girls [1].

Several viral infections (e.g. rubella, enterovirus) in early life are associated with the risk of Type 1 diabetes [2]. Age is the most important determinant of enterovirus infections but data also supports a male predominance particularly for the more severe disease [3]. Gender differences are found in immunological response to some vaccinations given in early childhood [4].

These observations prompted us to look at gender differences in the effect of neonatal infections in a population based case-control study based on the Danish National Diabetes Register.

In 1996 a Danish national prospective register of childhood diabetes was opened. The ascertainment in the register during the first 4 years was more than 99%. All children below the age of 15 years with Type 1 diabetes diagnosed from 1996 to 1999 were invited to participate in a case-control study. Two control subjects per case were randomly selected from the National Population Register matched by date of birth and gender. Information on hospitalisation and perinatal diagnosis such as

References

1. Field LL (2002) Genetic linkage and association studies of Type 1 diabetes: challenges and rewards. *Diabetologia* 45:21–35
2. Kaprio J (2000) Science, medicine, and the future. *Genetic epidemiology*. *BMJ* 320:1257–1259
3. Parson TJ, Muniec DS, Sullivan K et al. (1997) A high observed substitution rate in the human mitochondrial DNA control region. *Nat Genet* 15:363–367
4. Pfeiffer H, Forster P, Ortmann C, Brinkmann B (2001) The results of a mtDNA study of 1,200 inhabitants of a German village in comparison to other Caucasian databases and its relevance for forensic casework. *Int J Legal Med* 114:169–172
5. Pfeiffer H, Brinkmann B, Hühne J et al. (1999) Expanding the forensic German mitochondrial DNA control region database: genetic diversity as a function of sample size and microgeography. *Int J Legal Med* 112:291–298

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preterm delivery (<37 weeks), small or large for date (>2 standard deviation from the expected mean), and neonatal jaundice and infections were obtained from the hospital discharge register. Data on maternal and paternal age and ethnicity were obtained from the Danish population register. Additionally all cases and controls received a questionnaire including questions on smoking during pregnancy, early life nutrition, growth, siblings and family history of diabetes. Immigrants and offspring of immigrants were excluded from the study. For the study 602 diabetic children were eligible and 1459 control subjects were matched by gender and date of birth. Hospital information was available for 598 cases and 1445 control subjects. Questionnaire data were available for 490 (81%) cases and 696 (48%) of control subjects. We estimated the effect of neonatal infections among boys and girls with multiple logistic regression using all cases and control with hospital information. The model included age, gender and interaction between gender and infections. Of the children 49 had an infectious diagnosis in the neonatal period and 28 of those had either meningitis or sepsis, the remaining diagnoses were upper respiratory tract infections. There was no information on treatment with antibiotics.

Interestingly we found an increased risk of diabetes in boys with any neonatal infections [OR=2.42 (CI 1.14–5.15) $p=0.02$] and a decreased risk in girls with neonatal infections: [OR=0.40 (CI 0.12–1.38) $p=0.15$]. The estimated odds ratio for boys was six times the estimated odds ratio for the girls [CI 1.41–25.5) $p=0.015$] which corresponds to the significant interaction between gender and neonatal infections. There was no association between diabetes and neonatal infections [OR=1.23 (CI 0.67–2.28) $p=0.51$] when the interaction term was omitted.

The difference between boys and girls remained after adjustment for confounders such as introduction of cow's milk (<2 month), maternal age at delivery, family history of diabetes, smoking during pregnancy and birth order with an odds ratio in boys of 4.36 (CI 1.29–14.8), and in girls of 0.49 (CI 0.10–2.36).