Concordance rate for Type II diabetes mellitus in monozygotic twins: actuarial analysis

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Summary To determine the concordance rate for Type II (non-insulin-dependent) diabetes mellitus in monozygotic twin pairs, initially ascertained discordant for diabetes, we carried out a prospective study on 44 non-diabetic subjects, each of whom had a sibling twin with diabetes (21 men, 23 women, median age 55 years, interquartile range 47–65). The subjects were referred as discordant for Type II diabetes. The twin pairs were part of the British Diabetic Twin Study and ascertained between May 1968 and January 1998. These subjects underwent an OGTT at time of referral and periodically thereafter. The mean follow-up was 8 years (range 0–18 years) and data were collected until January 1996. The percentage of twins who developed Type II diabetes was assessed by stan-

dard actuarial life-table methods and the pairwise concordance rate, that is the proportion of concordant pairs over the sum of concordant and discordant pairs, was calculated. The observed rates of concordance for Type II diabetes at 1, 5, 10, and 15 years follow-up were 17, 33, 57, and 76 %, respectively. The concordance rate for any abnormality of glucose metabolism (either Type II diabetes or impaired glucose tolerance) at 15 years follow-up was 96 %. The concordance rate for Type II diabetes in monozygotic twins is very high even in twins initially ascertained discordant for diabetes. [Diabetologia (1999) 42: 146–150]

Keywords Type II diabetes mellitus, actuarial analysis, concordance rate, identical twins.

The pathogenesis of Type II (non-insulin-dependent) diabetes mellitus probably involves the interaction of genetic and environmental factors [1]. The nature of the genes which contribute to Type II diabetes in adults remains unclear. Genes have recently been identified which are associated with the development of Type II diabetes in children and young adults [2, 3].

Evidence that genetic factors are important in Type II diabetes derives largely from the study of twins and families; the risk in a sibling of a patient

with the disease is about 10 %, higher than the risk in the general population, which is 1 to 3 % [4, 5]. Such differences, however, could be due to shared environmental or genetic factors. Twins have been widely used to resolve the relative role of genetic and environmental factors. In the classic twin studies similarities (concordance) for a trait in identical and non-identical twin pairs are compared. A greater concordance rate in identical than non-identical twins suggests that genetic factors are important whilst similar concordance rates indicate a major role for environmental factors [5].

Early twin studies of patients with Type II diabetes indicated that the concordance rate between identical twins for Type II diabetes was very high (69 to 90 %) [5]. This observation was taken to establish a strong genetic basis for the disease and has been widely quoted as the basis for the intense current search for genes associated with Type II diabetes. These studies, however, were volunteer-based and cross-sectional

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Abbreviations: IGT, Impaired glucose tolerance; NGT, normal glucose tolerance; OGTT, oral glucose tolerance test.

with a high potential bias towards ascertaining concordant as against discordant pairs.

Type II diabetes, defined formally by a degree of oral glucose intolerance, has a peak incidence at the age of 50 years and a cumulative incidence that continues to increase into old age [4]. Thus, in any analysis of the frequency of Type II diabetes it is important to follow a cohort using oral glucose tolerance tests. There have been neither systematic prospective studies of Type II diabetic identical twins nor any using actuarial analysis. Actuarial analysis is designed to estimate event rates (in our case developing diabetes) in a cohort followed prospectively when both the duration of follow-up and number of cases under review varies during the study period. We therefore embarked on a prospective study of identical twins of Type II diabetes patients who were not diabetic at referral using actuarial analysis. The aim of the study was to determine whether the concordance rate in these identical twins was high and whether twins who remained non-diabetic had normal glucose tolerance.

Subjects and methods

As there is no twin register in Britain a systematic survey of all twins is not possible. The British Diabetic Twin Study includes data on identical twins, one or both of whom has diabetes, collected over 32 years. Twins in the study were ascertained because they were diabetic not because they were twins. To determine the lowest estimate of concordance for Type II diabetes, we selected identical twins in whom: 1. the index twin had Type II diabetes; 2. twin pairs were referred within 5 years of the diagnosis of the index twin; 3. the co-twin was not known to have diabetes at the time of referral; 4. twin pairs were all of European origin, and 5. twins were ascertained between May 1968 and January 1998. Twins were then followed prospectively until either the end of the study or development of diabetes. By ascertaining twin pairs discordant for Type II diabetes, that is only one twin had the disease, we excluded some pairs in which both twins developed diabetes at a similar time. In this way we biased ascertainment and thus we are likely to have underestimated the true level of concordance.

Of our whole series of identical twins, 44 pairs fulfilled the criteria for selection and were studied (21 males, median age at diagnosis 55 years, interquartile range 47–65). Monozygosity was established as described previously [6], using 21 genetic markers to obtain a probability of monozygosity greater than 97%. All but 3 of the 44 pairs were living apart at the start of the study. The non-diabetic twins were followed until 1 January 1998, when they had been observed for a mean of 8 years (range 0.1 to 18). Diabetes was diagnosed according to the National Diabetes Database Group Criteria [7]. All the non-diabetic twins underwent an oral glucose tolerance test (OGTT) when referred and periodically thereafter. Diabetes was diagnosed if fasting whole blood glucose was greater than 6.7 mmol/l, or 2 h whole blood glucose greater than or equal to 10.0 mmol/l and one other intervening value greater than or equal to 10 mmol/l during OGTT. Impaired glucose tolerance (IGT) was defined as a whole blood glucose concentration between 6.7 and 10 mmol/l two hours after drinking glucose. OGTT was carried out until 1979 using a 50 g glucose

load, a 75 g dose was used thereafter. These two different doses reflect the changes in the diagnostic criteria in 1979; it has been shown that doses of 50 g or 75 g given under standardised conditions, resulted in virtually identical blood glucose concentrations [7]. Before 1979 16 twin pairs were referred and 9 of them had their last follow up before that year. These 16 non-diabetic twins had an OGTT using 50 g instead of 75 g of glucose; we applied the same threshold blood glucose concentrations to twins studied in the period before and after 1979. Whole blood glucose was measured using either potassium ferricyanide until 1979 or glucose oxidase method thereafter.

Those twins who were not tested every year were instructed to do urine glucose tests periodically and to report the presence of glycosuria or the onset of symptoms suggestive of hyperglycaemia to both their General Practitioners and the investigators.

Statistical analysis. The twins were followed until the closing date or until they developed diabetes. The end-point of the study was the assessment of the pairwise concordance rate, that is the proportion of concordant pairs over the sum of concordant and discordant pairs. These data are expressed as a percentage (confidence interval). The percentage of twins who did not develop Type II diabetes was calculated by standard actuarial life-table methods [8]; the cumulative pairwise concordance rate being equal to 100 minus the probability of remaining non-diabetic twins expressed as a percentage. The standard error of a point estimate was computed using the Greenwood formula. Analysis was not done with less than five non-diabetic twins under study when any single event could have a major impact on disease-risk estimates.

Data on age at diagnosis of the index twin and age at last follow-up are expressed as means $\pm$ SD; sex is expressed as men/women ratio. Data on body mass index, lipid concentrations, blood pressure and macrovascular disease were not complete in all twins over the 16 years of study and were not analysed. Differences in age at diagnosis of the index twin, age at follow-up and sex of the twins who had normal glucose tolerance, IGT or diabetes at their last follow-up were assessed using analysis of variance (ANOVA) and Chi-squared for trend (GraphPad Instat Software V2.0.5a, 950811B, GraphPad Software Inc., San Diego, Calif., USA).

Results

Of 44 identical twins referred to the study as non-diabetic, 22 developed diabetes and 13 were found to have IGT (Tables 1 and 2). Six of the subjects died during the follow-up period. All of the twins who became diabetic developed Type II diabetes. At referral OGTT disclosed frank diabetes in seven (16%) and IGT in four (9%) subjects. The observed rates of developing Type II diabetes at 1, 5, 10, and 15 years of follow-up were 17, 33, 57, and 76%, respectively. The median period between diagnosis of Type II diabetes in the first and second twin was 6 years (range 0.1–17 years). Details of actuarial analysis for the development of diabetes are reported in Table 1 and illustrated in Figure 1.

Details of the actuarial analysis of twins developing abnormal glucose metabolism (either Type II diabetes or IGT) are presented in Table 2 and summarised in Figure 2. The rates of concordance for abnor-

Table 1. Actuarial analysis on the probability of developing Type II diabetes in monozygotic twin pairs initially discordant for the disease

Follow-up (years)	Number at risk	Type II diabetes	Lost from follow-up ¹	Proportion of subjects remaining non-diabetic	Cumulative probability of remaining non-diabetic	Confidence interval of cumulative probability
0-1	44	7	7	0.827	0.827	
1-2	30	1	3	0.964	0.798	0.671-0.880
2-3	26	1	0	0.961	0.767	0.633-0.857
3-4	25	3	2	0.875	0.671	0.548-0.768
4-5	20	0	0	1	0.671	0.501-0.794
5-6	20	1	2	0.947	0.636	0.480-0.756
6-7	17	2	2	0.875	0.556	0.413-0.678
7-8	14	2	1	0.851	0.474	0.329-0.605
8-9	11	0	1	1	0.474	0.295-0.633
9-10	10	1	0	0.9	0.426	0.270-0.574
10-11	9	0	0	1	0.426	0.247-0.594
11-12	9	3	0	0.666	0.284	0.172-0.406
12-13	6	0	0	1	0.284	0.126-0.465
13-14	6	1	0	0.833	0.237	0.108-0.393
14-15	5	0	0	1	0.237	0.093-0.417

¹ Lost from follow-up includes twins who either finished follow-up, did not wish to participate further, could no longer be located or died during the specified interval

Table 2. Actuarial analysis on the probability of developing glucose abnormalities (Type II diabetes or IGT) in monozygotic twin pairs initially discordant for Type II diabetes

Follow-up (years)	Number at risk	Type II diabetes or IGT	Lost from follow-up ¹	Proportion of subjects without Type II diabetes or IGT	Cumulative probability of not having Type II diabetes or IGT	Confidence interval of cumulative probability
0-1	44	11	3	0.741	0.741	
1-2	30	4	0	0.866	0.642	0.521-0.740
2-3	26	1	0	0.9611	0.617	0.466-0.737
3-4	25	3	2	0.875	0.540	0.406-0.656
4-5	20	0	0	1	0.540	0.382-0.674
5-6	20	1	2	0.947	0.511	0.364-0.641
6-7	17	3	1	0.818	0.418	0.295-0.537
7-8	13	3	0	0.769	0.322	0.209-0.440
8-9	10	2	0	0.8	0.257	0.146-0.384
9-10	8	1	0	0.875	0.225	0.113-0.360
10-11	7	0	0	11	0.225	0.104-0.375
11-12	7	3	1	0.538	0.121	0.058-0.208
12-13	3	0	0	1	0.121	0.039-0.251
13-14	3	0	0	1	0.121	0.039-0.251
14-15	3	3	0	0.0	0.040	0.014-0.083

¹ Lost from follow-up includes twins who either finished follow-up, did not wish to participate further, could no longer be located or died during the specified interval

mal glucose metabolism at 1, 5, 10 and 15 years follow-up were 26, 46, 78 and 96% respectively. At the end of the study, the majority (13/22) of the twins who remained non-diabetic had IGT.

There was no important difference in sex, age at diagnosis of the index twin or age at last follow-up among the twins who had normal glucose tolerance, IGT or diabetes at their last follow-up. Data is presented in Table 3 for sex, age of the index twin at diagnosis and age at last follow-up. There was no evidence that the rate of developing diabetes changed with time; thus, the rate of conversion to diabetes

was not statistically different in each 5 year period up to 15 years after the diagnosis of the index twin (that is 33, 24 and 19% respectively).

Discussion

This prospective study of a cohort of 44 identical twins of Type II diabetic patients, who were not known to have diabetes themselves on referral, showed a rate of development of Type II diabetes after 15 years follow-up of 76%. Of those twins who re-

Fig. 1. Cumulative probability of remaining non-diabetic

Fig. 2. Cumulative probability of remaining neither diabetic nor having IGT

maintained non-diabetic at the end of the study, the majority had IGT. While it is possible that these twins will remain non-diabetic, analysis of the rates of developing diabetes in the twins suggested that there was no decline in the conversion rate by the end of the study and, therefore, that the concordance rate for Type II diabetes in the twins we studied will become higher. These observations indicate that Type II diabetes is likely to develop in a subject genetically susceptible to the disease.

Our study was limited to identical twins. To estimate whether a high concordance rate in identical twins reflects a strong genetic effect it is necessary to compare the concordance rate in identical and non-identical twins. We have not done this and it will be some time before we will have studied enough non-identical pairs over a sufficiently long period to reach a firm conclusion. Our ascertainment of identical twins for this study was deliberately biased to only include twins referred discordant for Type II diabetes, thus probably underestimating the true concordance rate. Ascertainment was not population-based but by referral to the study. Only 44 twin pairs were studied. Moreover, we followed twins from time of referral for a mean of only 8 years and the majority of those who remained non-diabetic might yet develop diabetes

Table 3. Characteristics of twins at their last follow-up or when diagnosed with Type II diabetes

	NGT (<i>n</i> = 9)	IGT (<i>n</i> = 13)	Type II diabetes (<i>n</i> = 22)
Age at diagnosis of index twin (years)	54.7 ± 19.3	51.8 ± 15.1	50.1 ± 12.8
Age at last follow-up (years)	59.2 ± 19.8	57.8 ± 15.9	57.9 ± 12.9
Sex (men/women)	4/5	6/7	11/11

There was no significant difference between the three groups for any of the three parameters. Data are mean ± SD

since they already had IGT. There was no decline in the rate of twins developing diabetes by the end of the study and four of the eight non-diabetic twins, who had glucose tolerance tests after they developed IGT, subsequently developed Type II diabetes. Despite the biases in this study which could have led to an underestimation of the true concordance rate, the concordance rate for Type II diabetes was high.

Cross-sectional twin studies of the heritability of disease are flawed if the disease incidence increases with age and if ascertainment is through physician-based referral. Nevertheless, such studies of Type II diabetic twins have been widely cited to support a high concordance in identical twins [9–11]. One of these earlier studies was our own which showed a pairwise concordance rate in Type II diabetic identical twins of about 90% [9]. Two other cross-sectional studies of twins in the USA and Japan, ascertained through physician-based referral, showed a higher pairwise concordance rate in identical (69 and 83%) than non-identical (24 and 40% respectively) pairs [10, 11]. In the American study the non-diabetic twins did not have glucose tolerance tests to check for diabetes [10]. Testing twins to exclude diabetes is essential as Type II diabetes could be present, but undiagnosed. In this study 16% of twins at referral were only found to have diabetes after a glucose tolerance test.

There have been two cross-sectional population-based twin studies of Type II diabetes, one from the USA and the other from Finland [12, 13]. The non-diabetic twins were not consistently tested in either study. The American study relied on self-reporting of diabetes while the Finnish study identified twins only through prescriptions for antidiabetic drugs which excluded all patients treated by diet alone. Nevertheless, both studies reported a genetic effect determining the disease. In the Finnish study the pairwise concordance rate was higher for identical (20.4%) than non-identical (8.8%) twin pairs [12]. The American study highlighted the problem of cross-sectional studies in that initially the pairwise concordance rate for Type II diabetes was slightly higher in identical (16.7%) than in non-identical (7.7%) pairs; however, on repeat testing, approximately 10 years after the

first test, the concordance rate in identical twins was substantially higher (41.2%) than in the non-identical twins (9.5%) [13]. The rate of diabetes in the American study was probably underestimated owing to firstly, their unconventional definition of diabetes: a blood glucose greater than 13.9 mmol/l 1 h after a 50 g glucose oral load (conventional criteria would have given 65% concordance), and secondly, actuarial analysis was not done.

Genes determining susceptibility to Type II diabetes could influence insulin secretion or insulin sensitivity in predisposing to the disease, or both. The limited number of metabolic studies in non-diabetic twins of Type II diabetic patients tend to be insulin resistant with delayed insulin secretory capacity to both oral and intravenous glucose [14–16]. Studies in normal twins have shown that a substantial fraction of the variability in blood glucose concentrations, both when fasting and following an oral glucose load, can be attributed to genetic influences, however it remains unclear to what extent the metabolic changes in Type II diabetes are genetically determined [17].

Environmental factors are likely to be important in causing Type II diabetes as concordance falls short of 100% and twins differ in rates of progression to the disease. Twin studies, however, have had limited success in identifying such factors. Birth weight, physical fitness and adult weight gain have been implicated in disease risk [18–20]. Identical twins are more likely than non-identical twins to have intrauterine malnutrition. While low birth weight could be due to abnormal glucose tolerance, it did not lead to differences in glucose metabolism between identical and non-identical twins [18]. Physical fitness is also related to glucose metabolism and has been implicated as a factor in determining the risk of developing Type II diabetes [19]. Finally, adult weight gain has been found to be greater in both Type II diabetic twins and their non-diabetic twins than in control twins; thus, adult weight gain, which has been associated with an increased risk of diabetes, may be genetically determined [12, 20].

We conclude that the great majority of identical twin pairs are likely to become concordant for Type II diabetes or IGT. While environmental factors might play a role in the development of diabetes, genetic factors are important. We cannot be certain that Type II diabetes is largely genetically determined but our study shows that subjects genetically susceptible to Type II diabetes are very likely to develop the disease.

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