

Effects of Genetic Susceptibility for Type 2 Diabetes on the Evolution of Glucose Homeostasis Traits Before and After Diabetes Diagnosis

Data From the D.E.S.I.R. Study

Alain Gautier,¹ Ronan Roussel,² Céline Lange,^{3,4} Xavier Piguel,¹ Stéphane Cauchi,⁵ Sylviane Vol,⁶ Philippe Froguel,^{5,7} Beverley Balkau,^{3,4} and Fabrice Bonnet¹

OBJECTIVE—To assess the impact of genetic susceptibility on evolution toward type 2 diabetes (T2D) by analyzing time trajectories of fasting glucose, glycated hemoglobin (HbA_{1c}), insulin sensitivity (homeostasis model assessment [HOMA2%S]), and β -cell secretion (HOMA2%B) in a large nondiabetic cohort. We also examined whether baseline HbA_{1c} modified the effect of genetic predisposition on the time trajectories.

RESEARCH DESIGN AND METHODS—Time trajectories were drawn in 4,744 participants from the French Data from an Epidemiological Study on the Insulin Resistance Syndrome (D.E.S.I.R.) cohort based on samples collected every 3 years over a 9-year follow-up. Trajectories were analyzed according to the *TCF7L2* common variant, a family history of T2D, and a combination of at-risk alleles from nine T2D-associated genes.

RESULTS—There was a marked decrease in HOMA2%B in parallel to a steep increase in HbA_{1c} over the 3 years before incident diabetes, which was not influenced by genetic predisposition when considered alone. However, after the onset of T2D, the *TCF7L2* at-risk variant was associated with a greater decrease in HOMA2%B. There was a joint effect of a family history of T2D with the presence of the *TCF7L2* risk allele with a greater rise in HbA_{1c} conferred by the coexistence of a family history and the T risk allele. An HbA_{1c} $\geq 5.7\%$ at baseline was associated with a greater increase in both glycemia and HbA_{1c} levels in the presence of a combination of diabetes at-risk alleles.

CONCLUSIONS—After incident T2D, *TCF7L2* at-risk variants were associated with a faster decrease in β -cell function compared with those with the CC genotype. There was a joint effect of family history of T2D and *TCF7L2* risk variant on the rise in glycemia and the decrease in insulin secretion at the end of follow-up, suggesting the joint influence of the combination of diabetes genetic predisposition with familial factors on the evolution of glycemia over time. *Diabetes* 60:2654–2663, 2011

From the ¹Service Endocrinologie, Centre Hospitalier Universitaire Rennes, Université Rennes 1, Hôpital Sud, INSERM U991, Rennes, France; the ²Université Paris 7, Hôpital Bichat, AP-HP, INSERM U695, Paris, France; the ³Centre de Recherche en Épidémiologie et Santé des Populations, INSERM U1018, Epidemiology of Diabetes, Obesity and Chronic Kidney Disease Over the Life Course, Villejuif, France; the ⁴Université Paris-Sud 11, UMRs 1018, Villejuif, France; the ⁵Centre National de la Recherche Scientifique, UMR 8090, Institute of Biology, Lille 2 University, Pasteur Institute, Lille, France; the ⁶Institut inter Régional pour la Santé, La Riche, France; and ⁷Genomic Medicine, Hammersmith Hospital, Imperial College London, London, U.K. Corresponding author: Fabrice Bonnet, fabrice.bonnet@chu-rennes.fr. Received 11 October 2010 and accepted 11 July 2011. DOI: 10.2337/db10-1442

This article contains Supplementary Data online at <http://diabetes.diabetesjournals.org/lookup/suppl/doi:10.2337/db10-1442/-/DC1>.

*A list of the members of the D.E.S.I.R. Study Group can be found in the APPENDIX. © 2011 by the American Diabetes Association. Readers may use this article as long as the work is properly cited, the use is educational and not for profit, and the work is not altered. See <http://creativecommons.org/licenses/by-nc-nd/3.0/> for details.

Impaired fasting glucose and type 2 diabetes (T2D) arise from a failure of the β -cell to adequately compensate for insulin resistance (1–6). Previous evidence suggests that the combination of moderately elevated glycemia, increased BMI, and family history of T2D is a strong predictor of T2D in the general population (7). After a compensatory period with a slow linear increase in fasting glucose, a transient unstable period over 2 to 3 years leads to rapidly emerging hyperglycemia and overt diabetes (8). Concomitant acceleration in both β -cell dysfunction and decreasing insulin sensitivity has recently been shown to occur before the onset of T2D (9).

Previous evidence shows that genetic susceptibility for T2D could induce an early β -cell dysfunction (10–13). However, the impact of genetic predisposition to T2D on the evolution of glucose homeostasis traits over time in the years preceding T2D onset, and after T2D onset, has not been studied in a general population.

The main aim of our study was to assess the impact of genetic susceptibility on evolution toward diabetes and in the years thereafter by analyzing time trajectories of fasting glucose, glycated hemoglobin (HbA_{1c}), β -cell function (using homeostasis model assessment [HOMA]; HOMA2%B), and insulin sensitivity (HOMA2%S) in a large population without T2D at baseline. Because the *TCF7L2* polymorphism has the largest effect on T2D susceptibility among the predisposing genes discovered to date (14,15), we first analyzed the impact of the *TCF7L2* polymorphism alone on trajectories and then investigated the joint effect of the *TCF7L2* risk allele and a family history of T2D on the evolution of time trajectories for both HbA_{1c} and insulin secretion. Second, we assessed the effect of a genetic score integrating the number of at-risk alleles from nine well-defined T2D-associated variants (*TCF7L2* rs7903146, *CDKN2A/2B* rs10811661, *CDKAL1* rs7754840, *PPARG* rs1801282, *HHEX* rs1111875, *IGF2BP2* rs4402960, *KCNJ11* rs5219, *SLC30A8* rs13266634, and *WFS1* rs10010131) (15).

Finally, because recent evidence shows that glycemia influences the effects of T2D-risk genes on insulin secretion (16), we assessed whether baseline HbA_{1c} modified the impact of diabetes at-risk genes on the evolution of fasting glucose, HbA_{1c}, HOMA2%B, and HOMA2%S over the follow-up in the entire cohort.

RESEARCH DESIGN AND METHODS

We studied men and women aged 30–65 years who participated in the 9-year follow-up study Data from an Epidemiological Study on the Insulin Resistance

Syndrome (D.E.S.I.R.). Participants were recruited from volunteers offered periodic health examinations free of charge by the French Social Security in 10 health examination centers in western France. All signed an informed consent and the protocol was approved by an ethics committee. Cases of diabetes were identified by a fasting plasma glucose ≥ 7.0 mmol/L during one of the four 3-year examinations. Those who were identified as diabetic patients because of treatment for diabetes have not been included in these analyses. After exclusion of individuals with diabetes at baseline, we studied 4,577 participants with information on the *TCF7L2* rs7903146 genotype; 167 had incident diabetes.

Subjects were stratified according to 1) the *TCF7L2* rs7903146 genotype CC versus T allele carriers (4,744 participants, 167 with incident diabetes), 2) family history of T2D (4,722 participants, 167 with incident diabetes), and 3) the number of at-risk alleles (< 12 or ≥ 12 , the upper quartile) (4,055 participants, 146 with incident diabetes). Nine polymorphisms were chosen to define the number of at-risk alleles (*TCF7L2* rs7903146, *CDKN2A/2B* rs10811661, *CDKALI* rs7754840, *PPARG* rs1801282, *HHEX* rs1111875, *IGF2BP2* rs4402960, *KCNJ11* rs5219, *SLC30A8* rs1326634, and *WFS1* rs10010131) following previous analyses in this population (15). Diabetes in the family was defined as either a parent or grandparent with diabetes.

Clinical assessment. Two measures of blood pressure, using a mercury sphygmomanometer, were taken in a supine position after 5 min rest; mean values were used. Weight and height were measured in lightly clad participants, and BMI was calculated. The examining physician noted the family history of T2D in a clinical questionnaire; treatment for diabetes and hypertension were recorded. Smoking habits and degree of physical activity (at home, at work, and sport) were assessed using a self-administered questionnaire.

Biochemical measurements. All biochemical measurements were from one of four health center laboratories located in France at Blois, Chartres, La Riche, and Orléans. The interlaboratory variability for normal and pathological values was assessed monthly. Fasting plasma glucose, measured by the glucose-oxidase method, was applied to fluoro-oxalated plasma using a Technicon RA100 analyzer (Bayer Diagnostics, Puteaux, France) or a Specific or a Delta device (Konelab, Evry, France). HDL cholesterol and triglycerides were assayed with a DAX 24 (Bayer Diagnostics) or KONE analyzer (Konelab). HbA_{1c} was determined by high-performance liquid chromatography (L9100 ion-exchange analyzer; Hitachi/Merck-VWR, Fontenay-sous-Bois, France) or an

immunoassay (DCA 2000; Bayer Diagnostics). Insulin was quantified by microparticle enzyme immunoassay with an automated analyzer (IMX; Abbott, Rungis, France). Glucose, HbA_{1c}, and insulin have been standardized over laboratories and over the years of the study. Diabetes was defined to include individuals treated for diabetes and those with a fasting plasma glucose ≥ 7.0 mmol/L. Indexes of β -cell secretion and insulin sensitivity, HOMA2%B and HOMA2%S, were computed using software downloaded at <http://www.dtu.ox.ac.uk>.

Genotyping. Single nucleotide polymorphism (SNP) genotyping was performed with the SNPlex Technology (Applied Biosystems, Foster City, CA) based on the oligonucleotide ligation assay combined with multiplex PCR target amplification (<http://www.appliedbiosystems.com>).

Statistical analysis. Statistical analyses used SAS version 9.2 (SAS Institute, Cary, NC). Insulin, triglycerides, and HOMA2%B were log transformed for statistical analysis because of their skewed distributions. Baseline characteristics, means and percentages, were compared using Student *t* and χ^2 tests, respectively. A χ^2 test was used to compare those who did and did not progress to diabetes according to the *TCF7L2* genotype.

Mixed models were used to model repeated data for glucose, HbA_{1c}, HOMA2%B, and HOMA2%S, over the 3-year examinations, adjusting for age, sex, BMI, and recruitment center. Analyses in those who did and did not develop diabetes during the follow-up period were made, resetting the time-frame, considering year 0, the year of incident diabetes; for those who did not develop diabetes, year 0 corresponded to their last examination. Trajectories of mean values before and after the diagnosis of diabetes were drawn. If diabetic patients were treated by a sulfonylurea, data at the corresponding time points were not included. The slope of HbA_{1c} between different time points was tested in the mixed models using contrasts.

In the individuals who were screened with diabetes, we then studied the trajectories stratified according to the *TCF7L2* genotype, family history of T2D, and finally, the number of diabetes at-risk alleles (< 12 vs. ≥ 12). We excluded the data at the time points when diabetic patients were treated by a sulfonylurea. The sensitivity of the results was checked by including all the available data from the 167 diabetic patients, including those who were treated, and additionally, by adjusting on treatment type (lifestyle, metformin, sulfonylurea, and acarbose). We used pairwise testing between groups with *t* tests from the mixed model and also the overall tests to assess the differences between groups, over all time points. Interactions between the years and the groups

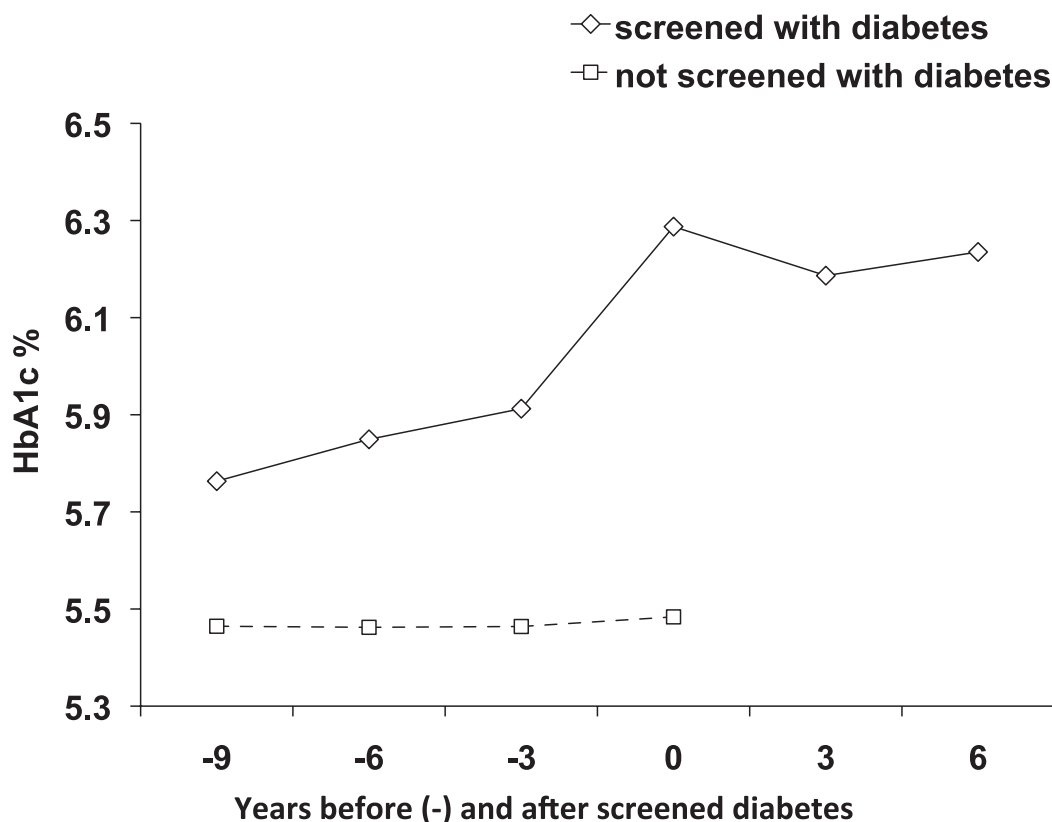


FIG. 1. Trajectories of HbA_{1c} in participants screened with incident diabetes at year 0 and in participants without diabetes, after adjustment for age, sex, BMI, and recruitment center.

were tested, over differing time periods—year -9 to $+6$, year -9 to 0 , and year 0 to $+6$ —to ascertain whether the relations over these periods differed; P values are shown in the figures.

In the entire cohort ($n = 4,080$ at baseline), logistic regression analyses were used to study 9-year incident diabetes according to family history of T2D and both *TCF7L2* and the genetic risk score, after adjusting for age, sex, BMI, and recruitment center. We also studied the evolution of HbA_{1c} over the 9 years of follow-up in all participants, stratified by the presence of a family history of diabetes, after adjustment for age, sex, BMI, and recruitment center, according to *TCF7L2* risk variant.

For the evolution of HbA_{1c} over the 9 years of the study, there was a significant interaction between baseline HbA_{1c} <5.7 and $\geq 5.7\%$ (the American Diabetes Association for the at-risk for diabetes group) (17) and the number of at-risk alleles <12 and ≥ 12 ($P = 0.0015$). We therefore stratified the cohort according to baseline HbA_{1c} to assess whether glucose metabolism at baseline modulated the impact of genetic predisposition on the evolution of fasting glucose and HbA_{1c}. Interactions were tested between at-risk allele groups, according to HbA_{1c} strata, and are presented as $P_{\text{trajectories}}$ in the figures.

RESULTS

Time trajectories of glucose homeostasis traits in individuals who did and did not progress toward diabetes. As expected, those who progressed to T2D had a worse metabolic profile at baseline compared with those who did not (Supplementary Table 1). In participants with incident T2D, a linear increase in HbA_{1c} was followed by a steep increase 3 years before diagnosis of T2D, then a small decrease 3 years after diagnosis (Fig. 1). In mixed model analysis, the HbA_{1c} time slopes between years -6 and -3 and between years -3 and 0 were significantly different ($P < 0.001$), confirming the steep increase in HbA_{1c} over the 3 years before the incidence of T2D. The same pattern was seen for fasting plasma glucose, which had been used to define incident T2D.

Time trajectories of glucose homeostasis traits according to *TCF7L2*. There was no difference between baseline characteristics according to *TCF7L2* variants for participants with incident T2D (Table 1). The frequency of the *TCF7L2* risk allele was significantly different between individuals who progressed to T2D and those who remained nondiabetic over the follow-up (60 vs. 51%, $P = 0.02$). Before T2D onset, the *TCF7L2* genotype, when considered alone, had no influence on HbA_{1c} or HOMA2%B trajectories (Fig. 2); nor did it have an influence on HOMA2%S trajectories (not shown). However, at-risk variants in the *TCF7L2* gene (CT + TT) were associated with an increase in HbA_{1c} and a faster decrease in HOMA2%B after T2D onset, with significant age-, sex-, BMI-, and recruitment center-adjusted differences 6 years after ($P = 0.03$ and $P = 0.006$, respectively) (Fig. 2). There was no significant interaction over the year 0 to $+6$ period for HbA_{1c}, but this was significant for HOMA2%B ($P = 0.03$).

Time trajectories of glucose homeostasis traits according to family history of diabetes. There was no difference between baseline characteristics according to the presence of a family history of T2D, except for a lower percentage of men ($P = 0.0006$) and a trend for a higher BMI ($P = 0.06$) (Table 1). There were no significant differences in HbA_{1c} trajectories according to family history of T2D (Fig. 3) or in fasting glucose (not shown), but there was a trend, albeit not significant, for a higher postdiagnosis HbA_{1c} in those with a family history of T2D (Fig. 3). A family history of T2D was associated with a lower HOMA2%B (at three time points, with a trend for interaction over the entire time period years -9 to $+6$: $P = 0.08$) (Fig. 3) and a lower HOMA2%S (interaction over years -9 to $+6$: $P = 0.06$)

TABLE 1

Baseline characteristics of participants with incident diabetes according to *TCF7L2* variants, family history of diabetes, and genetic score

	<i>TCF7L2</i>			Diabetes in family			Genetic score		
	CC ($n = 67$)	CT + TT ($n = 100$)	P value	No ($n = 123$)	Yes ($n = 44$)	P value	<12 ($n = 104$)	≥ 12 ($n = 42$)	P value
Age (years)	50 (9)	50 (9)	0.9	50 (9)	49 (9)	0.4	49 (9)	51 (9)	0.2
Men, n (%)	46 (69)	72 (72)	0.6	96 (78)	22 (50)	0.0006	69 (66)	35 (83)	0.04
Diabetes in family, n (%)	21 (31)	23 (23)	0.2				32 (31)	7 (17)	0.08
Current smoker, n (%)	17 (25)	37 (37)	0.1	41 (33)	13 (30)	0.6	32 (31)	15 (36)	0.6
BMI (kg/m ²)	27.7 (4.4)	27.9 (4.7)	0.8	27.4 (4.1)	28.9 (5.5)	0.06	27.8 (5)	27.1 (3.4)	0.4
Waist circumference (cm)									
Men	95 (10)	96 (10)	0.5	95 (10)	98 (10)	0.3	96 (11)	93 (8)	0.09
Women	90 (13)	89 (12)	0.8	89 (12)	90 (12)	0.6	88 (13)	93 (10)	0.3
Physical activity, n (%)									
None	15 (22)	34 (34)	0.2	39 (32)	10 (23)	0.4	26 (25)	19 (45)	0.05
Moderate	39 (58)	52 (52)		66 (54)	25 (57)		59 (57)	18 (43)	
Intensive	13 (20)	14 (14)		18 (14)	9 (20)		19 (18)	5 (12)	
sBP (mmHg)	137 (16)	140 (17)	0.3	139 (17)	138 (16)	0.5	139 (18)	138 (17)	0.7
dBp (mmHg)	83 (8)	85 (10)	0.3	84 (10)	84 (8)	0.9	84 (10)	83 (9)	0.7
Treatment for									
hypertension, n (%)	15 (22)	19 (19)	0.6	27 (22)	7 (16)	0.4	17 (16)	10 (24)	0.3
Glucose (mmol/L)	5.96 (0.50)	6.06 (0.58)	0.3	6.02 (0.55)	6.02 (0.55)	1	6.00 (0.57)	6.03 (0.49)	0.7
HbA _{1c} (%)	5.8 (0.4)	5.8 (0.5)	1	5.8 (0.5)	5.8 (0.4)	0.9	5.8 (0.5)	5.9 (0.5)	0.3
Insulin (pmol/L)	70.3 (48.3)	71.8 (47.0)	0.8	72.8 (49.4)	66.6 (41.7)	0.6	72.0 (49.2)	61.7 (39.0)	0.3
HOMA2%B (%)	88.2 (37.1)	89.4 (47.8)	0.8	89.9 (44.5)	86.2 (41.9)	0.5	89.8 (44.8)	80.5 (33.7)	0.2
HOMA2%S (%)	83.5 (40.6)	81.1 (42.5)	0.7	81.0 (42.4)	85.1 (39.9)	0.6	82.1 (41.0)	89.0 (42.8)	0.4
Triglycerides (mmol/L)	1.77 (1.79)	1.59 (0.86)	1	1.66 (1.39)	1.66 (1.07)	0.9	1.70 (1.51)	1.53 (0.99)	0.5
HDL cholesterol (mmol/L)	1.50 (0.34)	1.45 (0.36)	0.3	1.47 (0.37)	1.48 (0.31)	0.9	1.51 (0.38)	1.41 (0.28)	0.1

Data are presented as mean (SD) unless otherwise indicated. sBP, systolic blood pressure; dBp diastolic blood pressure.

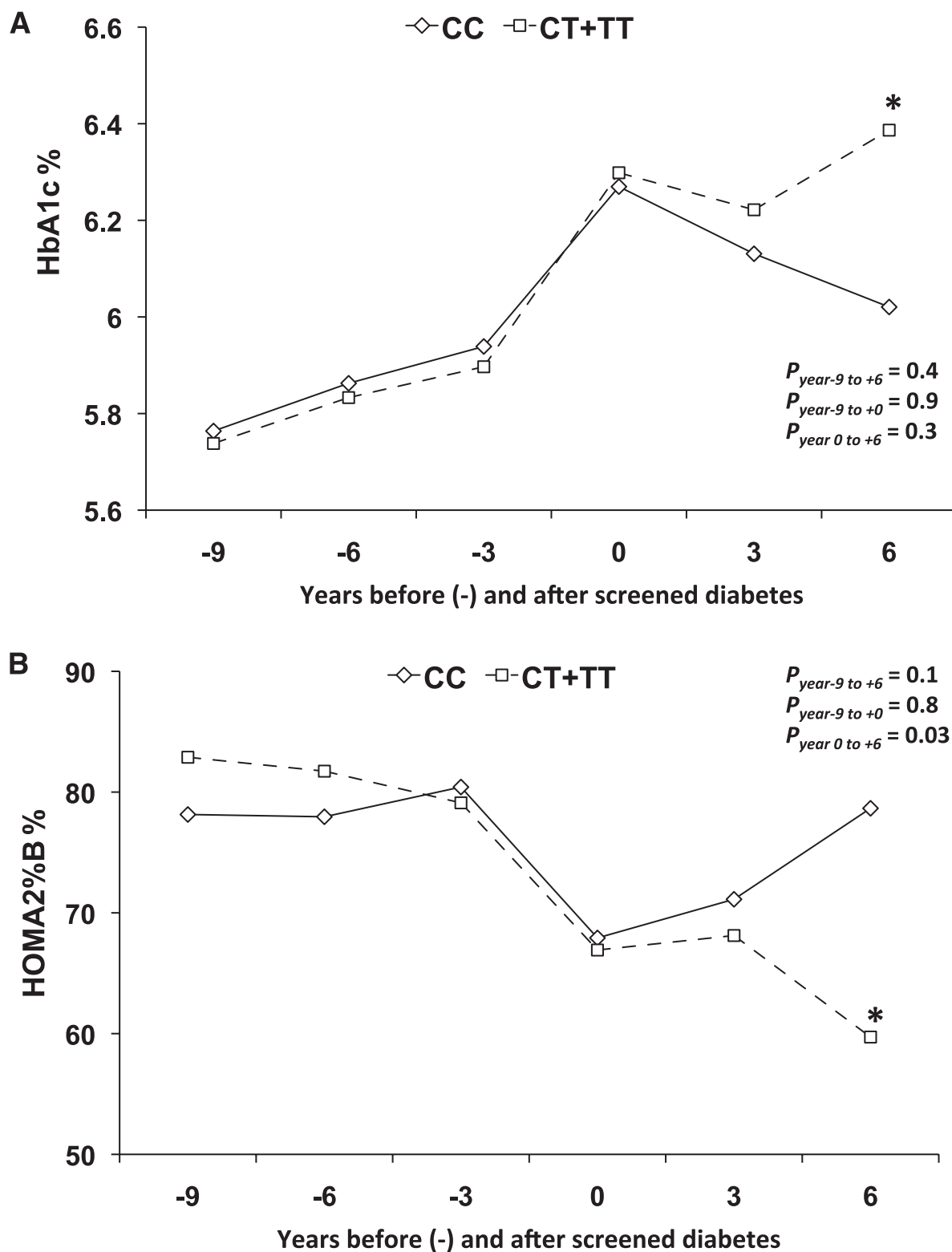


FIG. 2. Trajectories of HbA_{1c} (A) and HOMA2%B (B) in the 167 participants with incident diabetes, after adjustment for age, sex, BMI, and recruitment center, according to *TCF7L2* variants. Data after diabetes diagnosis in those treated by sulfonylureas are not included. * $P < 0.05$ for each time point. P values for the interactions between the two groups and trajectories over time are given for the entire time: from year -9 to $+6$, from year -9 to 0 before diabetes, and from year 0 to $+6$ after diabetes.

(not shown) in those with incident T2D, after adjustment for age, sex, BMI, and recruitment center.

There was a joint effect of family history of T2D and the *TCF7L2* risk variant on the HbA_{1c} trajectory with a

significantly higher HbA_{1c} and a more pronounced decline in HOMA2%B in individuals with a family history of T2D who carried the *TCF7L2* risk allele as compared with those with the CC genotype without a family history ($P = 0.003$ for

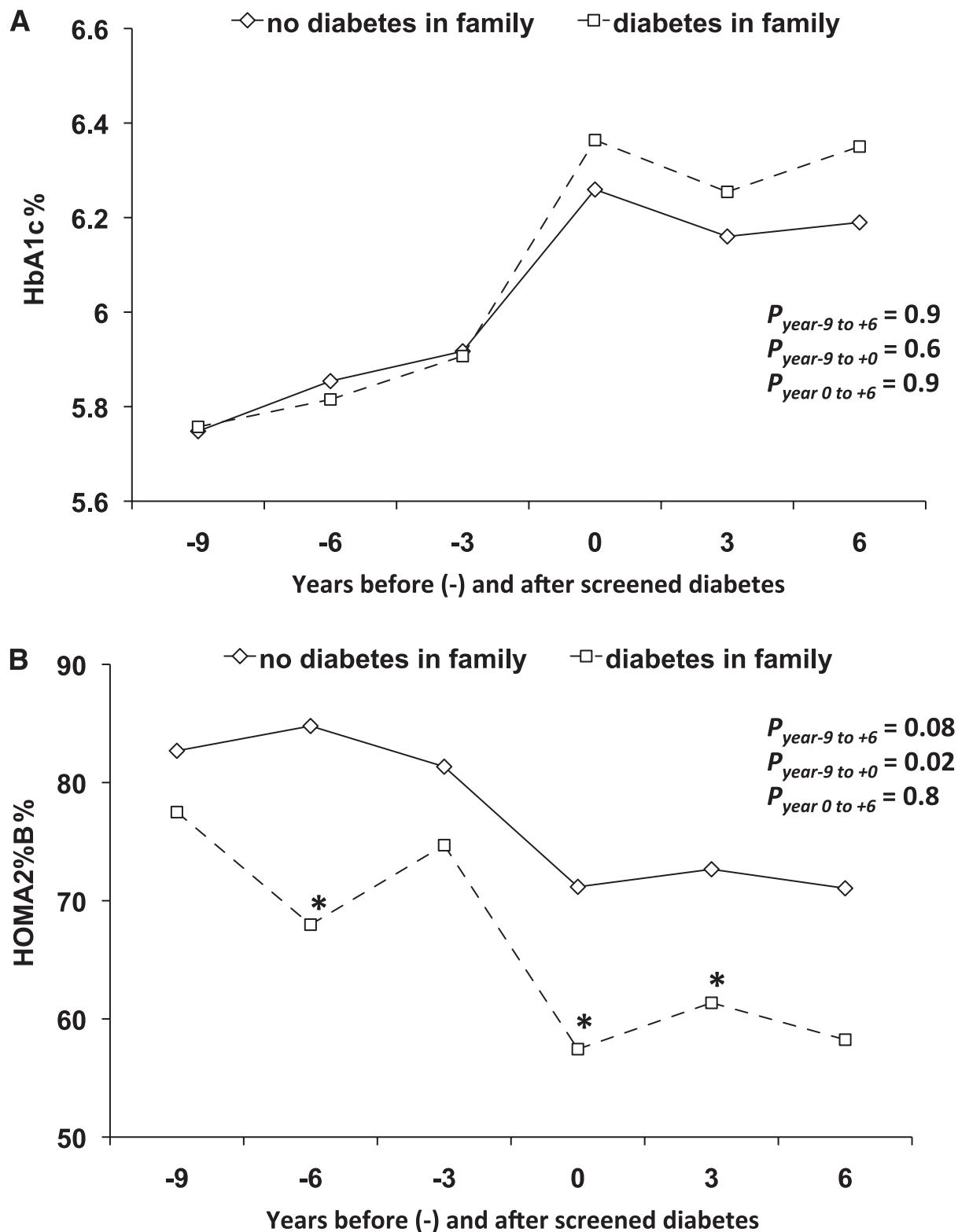


FIG. 3. Trajectories of HbA_{1c} (A) and HOMA2%B (B) in the 167 participants with incident diabetes, after adjustment for age, sex, BMI, and recruitment center, according to family history of diabetes. Data after diabetes diagnosis in those treated by sulfonylureas are not included. * $P < 0.05$ for each time point. P values for the interactions between the two groups and trajectories over time are given for the entire time: from year -9 to +6, from year -9 to 0 before diabetes, and from year 0 to +6 after diabetes.

HbA_{1c} and $P = 0.0001$ for HOMA2%B 6 years after incident T2D). Furthermore, in logistic regression on the entire cohort, the *TCF7L2* risk variant remained significantly related to the risk of 9-year incident diabetes, after

adjusting for the presence of a family history ($P = 0.01$). If we stratified the population according to the presence of a family history of T2D, those with a family history and the *TCF7L2* risk allele had higher HbA_{1c} levels at 9 years

than those with a family history and the CC genotype (Fig. 4).

Time trajectories of glucose homeostasis traits according to a genetic risk score. Among the incident cases of diabetes with ≥ 12 at-risk alleles, there was no difference between baseline characteristics according to the number of at-risk alleles, except for a higher percentage of men ($P = 0.04$) and a higher percentage of individuals with no physical activity ($P = 0.02$) (Table 1). There was no significant difference in HOMA2%B trajectories according to the genetic score (Fig. 5), and the same was the case for fasting glucose, HbA_{1c}, and HOMA2%S (data not shown). In logistic regression, a genetic score ≥ 12 was significantly associated with the risk of incident diabetes in the absence, but not in the presence, of a family history of T2D ($P = 0.0002$ and $P = 0.3$, respectively, with P -interaction = 0.04). **Impact of the glycemic status on the effects of the genetic score.** We assessed whether baseline HbA_{1c} modified the impact of diabetes risk genes on the evolution of HbA_{1c} in the entire cohort ($n = 4,080$ at baseline) during the 9 years of follow-up. Individuals with both a genetic score of ≥ 12 at-risk alleles and an HbA_{1c} $\geq 5.7\%$ at baseline had a higher plasma fasting glucose and higher HbA_{1c} at year 3 ($P = 0.05$ for glycemia, $P = 0.02$ for HbA_{1c}), year 6 ($P = 0.03$ for glycemia, $P = 0.002$ for HbA_{1c}), and year 9

($P = 0.0002$ for glycemia, $P = 0.0001$ for HbA_{1c}) than those with baseline HbA_{1c} $\geq 5.7\%$ and a genetic score < 12 (Fig. 6). The effects of a high genetic score on time trajectories for both glycemia and HbA_{1c} appear to be more pronounced for those with a slightly elevated HbA_{1c} at baseline than for those with HbA_{1c} $< 5.7\%$.

In parallel, those with both a genetic score of ≥ 12 at-risk alleles and an HbA_{1c} $\geq 5.7\%$ had a decrease in HOMA2%B at each of the 3-year examinations ($P = 0.01$ at year 3, $P = 0.03$ at year 6, and $P = 0.008$ at year 9) as compared with those with HbA_{1c} $\geq 5.7\%$ and a genetic score of < 12 at-risk alleles (data not shown). HOMA2%S did not differ at any time point according to the genetic score in either HbA_{1c} subgroup.

DISCUSSION

We describe the 9-year trajectories of fasting glycemia, HbA_{1c}, and insulin secretion and insulin sensitivity indexes before and 3 and 6 years after diabetes was screened in a large middle-aged population without T2D at baseline. For individuals who remained nondiabetic throughout the follow-up, metabolic trajectories showed a remarkable stability with little progressive increase in glycemia or HbA_{1c}—there was an increase only in BMI over time (not shown).

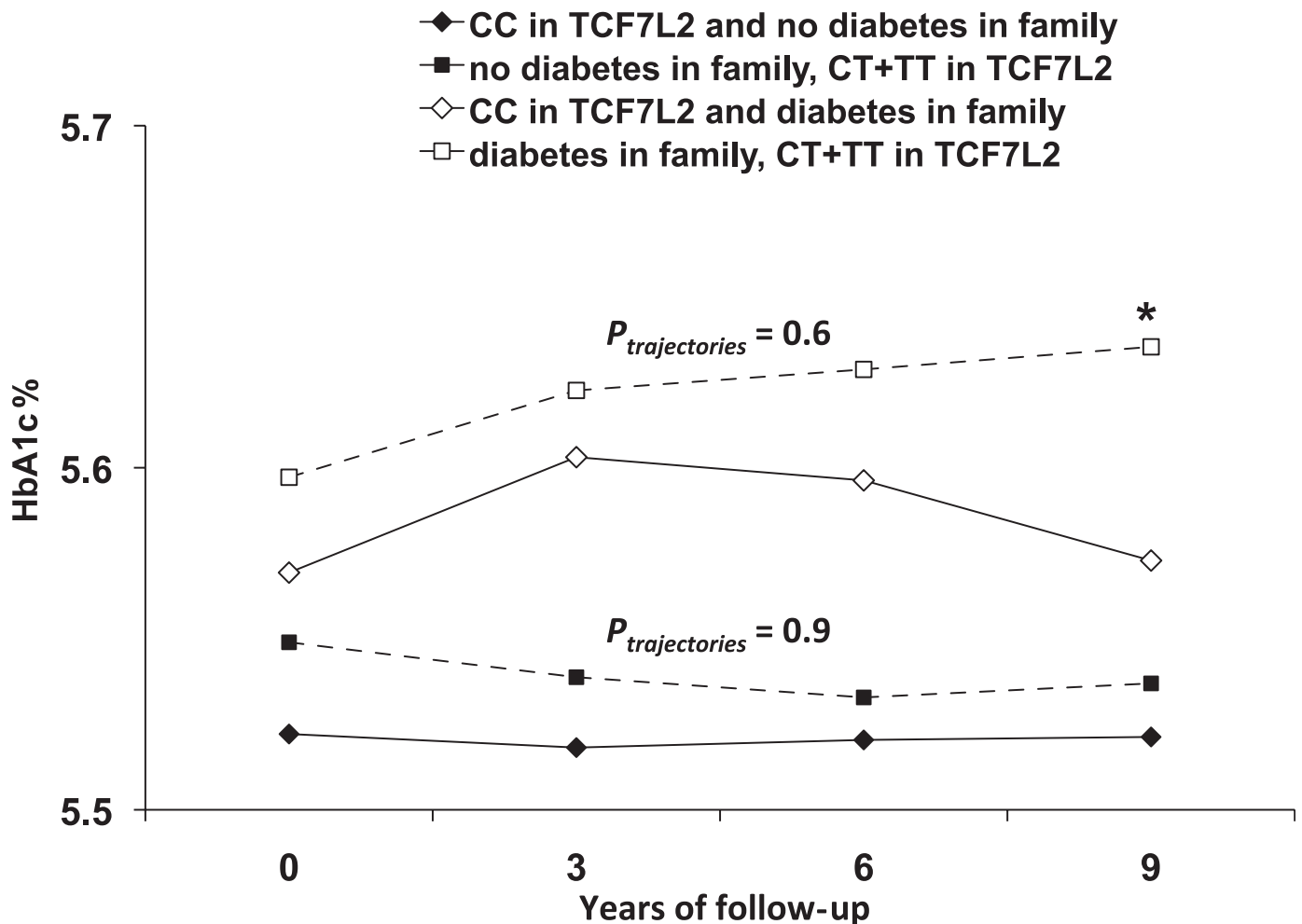


FIG. 4. Trajectories over the 9 years of follow-up of HbA_{1c} in all participants ($n = 4,080$ at baseline), stratified by the presence of a family history of diabetes, after adjustment for age, sex, BMI, and recruitment center, according to *TCF7L2* risk variant. * $P < 0.05$ for each time point. $P_{trajectories}$ is the P value for the interaction between values within each group.

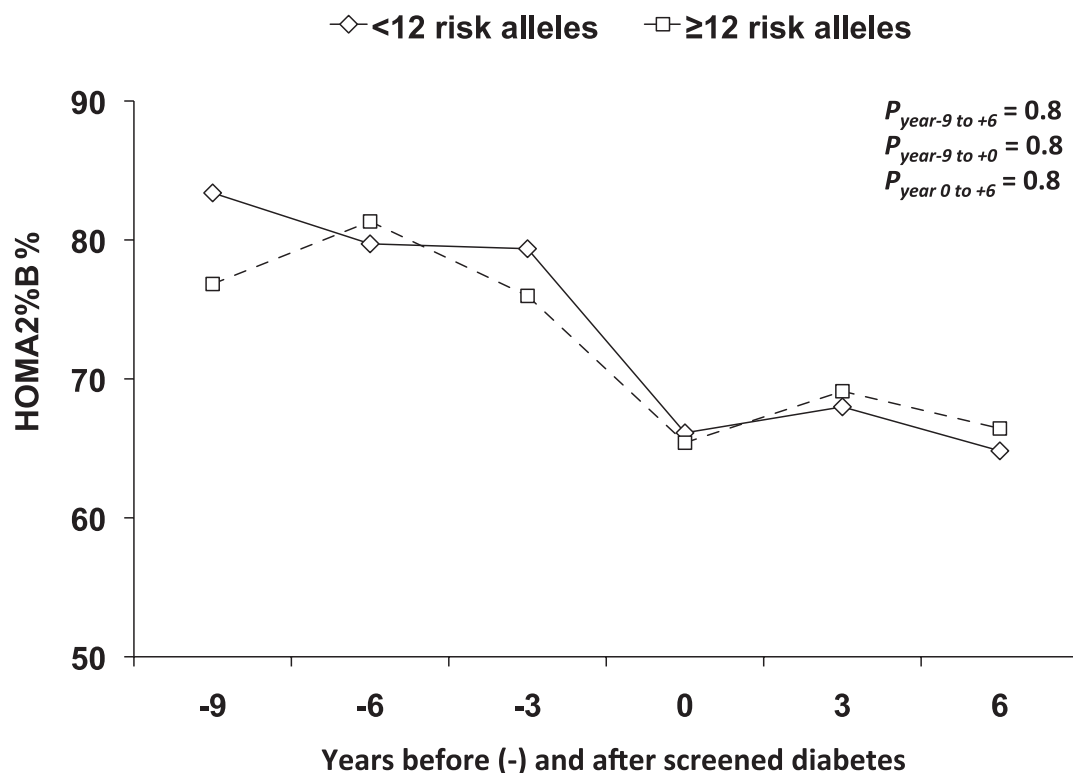


FIG. 5. Trajectories of HOMA2%B in the 146 participants with incident diabetes, after adjustment for age, sex, BMI, and recruitment center, according to a genetic score based on 12 at-risk variants. Data after diabetes diagnosis in those treated by sulfonylureas are not included. There were no differences between data at the six time points and no interaction between group and time.

Our findings are in agreement with the study by Tabák et al. (9), who described a linear increase in glycemia followed by a steep increase starting 3 years before diabetes diagnosis in parallel to a fall in insulin secretion starting 4–5 years before diabetes diagnosis. Other studies report a steep increase in fasting glycemia 1 to 3 years before diabetes diagnosis (4,18,19). We expand these findings by showing a similar pattern for HbA_{1c} with a steeper increase 3 years before T2D was screened. In contrast, HbA_{1c} values remained very stable in those who did not develop T2D over the follow-up. Our results suggest that individuals who presented with a progressive increase in HbA_{1c} values, even within the normal range, had an increased risk of developing T2D in the next 3 to 6 years and would probably benefit from a lifestyle intervention. Baseline HOMA2%B, a proxy for insulin secretion, did not differ between participants who developed T2D in comparison with those who remained nondiabetic, with a decrease in insulin secretion emerging only 3 to 6 years before diabetes was screened. This decline in insulin secretion may contribute to precipitate the onset of T2D in these patients.

We report for the first time the longitudinal effects of T2D predisposing genes on glucose homeostasis traits. Participants with at-risk variants in the *TCF7L2* gene or with a high number of at-risk alleles for nine T2D-associated genes had lower HOMA2%B after incident diabetes but similar HOMA2%S values during follow-up. This is an expected result because most of these genes are related to β -cell function (10).

In our study, the frequency of the *TCF7L2* risk allele was significantly higher in those who progressed toward T2D as compared with the nonincident cases. This is in agreement with previous findings by Lyssenko et al. (20),

who showed that the CT/TT genotypes of SNP rs7903146 strongly predicted incident T2D in two large cohorts through reduced stimulated-insulin secretion. In the Diabetes Prevention Program (DPP) study, carriers of the T allele had decreased insulin secretion accompanied by a lower BMI and an increase in insulin sensitivity, and the TT genotype was associated with progression to diabetes in the placebo group (21). Time trajectories over the years before diabetes onset were not substantially modified by genetic status, and all incident case subjects showed a similar pattern of decreased β -cell function and insulin sensitivity over time. Participants with incident T2D and carrying the risk-conferring variants of *TCF7L2* had a more pronounced decrease in HOMA2%B after T2D diagnosis. Our results persisted after adjustment for the type of treatment (lifestyle only, metformin, and acarbose), and similar findings were observed after inclusion of those with sulfonylurea treatment (data not shown). Even though the number of patients studied after diabetes diagnosis was low, these data suggest that the at-risk variant of *TCF7L2* is associated with a further decrease in β -cell function over time, despite hypoglycemic treatment.

TCF7L2, a transcription factor involved in the Wnt signaling pathway, has the largest effect on T2D susceptibility among the predisposing genes discovered to date (15,22). Although it remains to be determined exactly how a reduction of *TCF7L2* gene expression inhibits insulin secretion, a reduced sensitivity of the β -cell to incretins has been recently suggested as the main underlying mechanism (23). In vitro and animal studies of nonautoimmune diabetes show that incretin-based agents have the potential to expand β -cell mass (24–28). Thus, a reduced sensitivity of the β -cell to incretin hormones could explain the

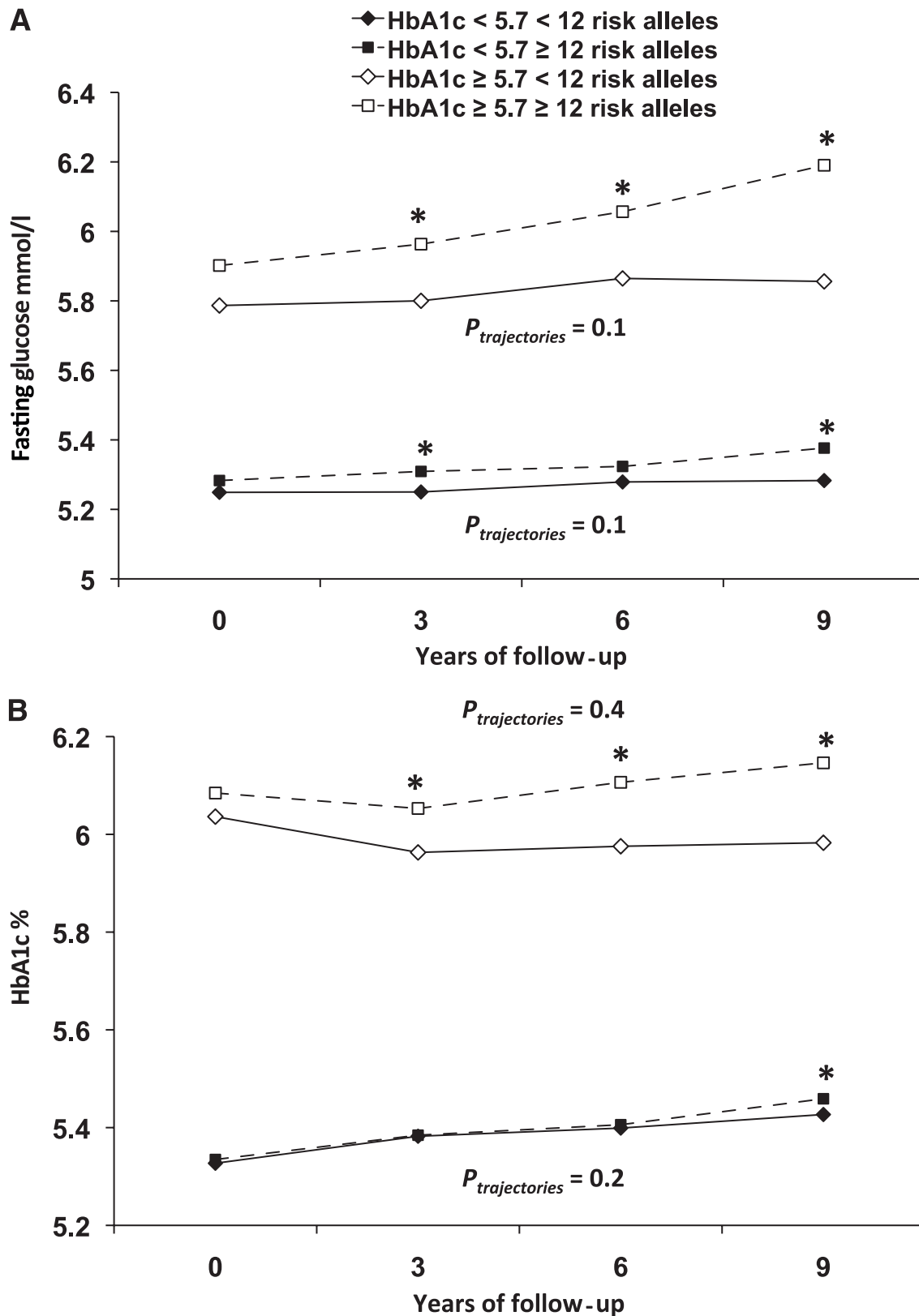


FIG. 6. Trajectories over the 9 years of follow-up of fasting glucose (A) and HbA_{1c} (B) in all participants ($n = 4,080$ at baseline), stratified by baseline HbA_{1c} <5.7 or >5.7%, after adjustment for age, sex, BMI, and recruitment center, according to a genetic score based on nine at-risk variants. Data after diabetes diagnosis in those treated by sulfonylureas are not included. * $P < 0.05$ for each time point. $P_{trajectories}$ is the P value for the interaction between values within HbA_{1c} strata.

faster deterioration in HOMA2%B after diabetes onset in *TCF7L2* at-risk genotypes. Our results support the initiation of trials testing early use of incretin drugs in these patients to prevent the decline in insulin secretion.

We observed that the time-varying effect of at-risk variants on glucose homeostasis traits seems to be magnified in those with a baseline HbA_{1c} $\geq 5.7\%$, with a progressively higher fasting glucose and HbA_{1c} in relation to decreased insulin secretion, as compared with those without a high predisposing genetic score. The effect of the genetic score was not apparent for those with a normal HbA_{1c} at baseline. This is in agreement with a recent report suggesting a greater impact of *TCF7L2* and *WFS1* in individuals who already have dysglycemia (16). We noted a similar trend, albeit not significant, when considering the *TCF7L2* at-risk variant only (data not shown). Our longitudinal findings confirm the interaction of glycemia with the modest effects of genetic variations on the evolution of glucose homeostasis traits over time in the general population. In the presence of higher baseline HbA_{1c}, both fasting glucose and HbA_{1c} levels, which were similar according to the genetic score at the inclusion, progressively increased over time in those with a high number of at-risk alleles. This is in agreement with data from the DPP study showing that among participants with both impaired glucose tolerance and elevated fasting glucose, a higher genetic score was associated with a lower probability of regression toward normoglycemia over the 3.2-year follow-up (15). This may relate to an increased vulnerability of the β -cell to glucotoxicity in individuals with a genetic predisposition to T2D.

A limitation of the study is the absence of confirmatory retesting of fasting glucose to diagnose diabetes. Variability in the measure of fasting glucose and the use of a threshold artificially accentuates the rapid rise at the time of diagnosis and the observed lower values of fasting glucose 3 years after diagnosis. Several other studies, however, report the same rapid rise in fasting glucose 2–3 years before diagnosis (4,9,18). Moreover, the parallel rise in HbA_{1c} and the decline in HOMA2%B and in HOMA2%S concomitant to the rise in fasting glucose are consistent with the steeper glycemia trajectory before T2D. Our findings on family history of T2D need to be interpreted with caution because the number of subjects in our study is low. Further epidemiological studies are needed to characterize the nature and the physiological effects of the contributing factors involved in the family predisposition of T2D.

The HOMA2%B is a practical but limited tool to quantify β -cell function. This index has previously been shown to independently predict the risk of diabetes in a large cohort, supporting its relevance to estimate insulin secretion in epidemiological studies (29). The lack of a more sensitive measure of first-phase insulin secretion would probably have masked subtle β -cell defects across declining glucose tolerance status. Although HOMA2%B is strongly correlated with the acute insulin response, defects of first-phase insulin secretion are more pronounced before diabetes onset, as compared with defects characterized by HOMA2%B (2).

In conclusion, T2D-associated variants were associated with a progressive increase in glycemia and HbA_{1c} in parallel to a lower β -cell function over time, which was more apparent in individuals who carried the predisposing *TCF7L2* rs7903146 high-risk variant and a family history of T2D. In addition, an HbA_{1c} $\geq 5.7\%$ at baseline was associated with a greater increase in both glycemia and HbA_{1c} levels in the presence of a combination of diabetes risk

alleles. These findings suggest that the combination of diabetes genetic predisposition with familial factors may help to better identify individuals at high risk to maintain hyperglycemia over time.

ACKNOWLEDGMENTS

The D.E.S.I.R. study has been supported by INSERM contracts with CNAMTS, Lilly, Novartis Pharma, and sanofi-aventis, and by INSERM (Réseaux en Santé Publique, Interactions entre les déterminants de la santé, Cohortes Santé TGIR 2008), the Association Diabète Risque Vasculaire, the Fédération Française de Cardiologie, La Fondation de France, ALFEDIAM, ONIVINS, Société Francophone du Diabète, Ardix Medical, Bayer Diagnostics, Becton Dickinson, Cardionics, Merck Santé, Novo Nordisk, Pierre Fabre, Roche, and Topcon. No other potential conflicts of interest relevant to this article were reported.

A.G. analyzed data and wrote parts of the manuscript. R.R. contributed to discussion and reviewed the manuscript. C.L. analyzed data. X.P., S.C., S.V., and P.F. reviewed the manuscript. B.B. contributed to analysis and discussion and reviewed and edited the manuscript. F.B. initiated the analysis and wrote parts of the manuscript.

APPENDIX

The D.E.S.I.R. Study Group: INSERM Centre de Recherche en Épidémiologie et Santé des Populations U1018: B. Balkau, P. Ducimetière, and E. Eschwège; INSERM U367: F. Alhenc-Gelas; Centre Hospitalier Universitaire D'Angers: Y. Gallois and A. Girault; Bichat Hospital: F. Fumeron, M. Marre, and R. Roussel; Centre Hospitalier Universitaire de Rennes: F. Bonnet; Centre National de la Recherche Scientifique UMR 8090, Lille: P. Froguel; Institute de Recherche Médecine Générale: J. Cogneau; Institute inter Regional pour la Santé: C. Born, E. Caces, M. Cailleau, J.G. Moreau, O. Lantieri, F. Rakotozafy, J. Tichet, and S. Vol.

REFERENCES

1. Festa A, Williams K, D'Agostino R Jr, Wagenknecht LE, Haffner SM. The natural course of beta-cell function in nondiabetic and diabetic individuals: the Insulin Resistance Atherosclerosis Study. *Diabetes* 2006;55:1114–1120
2. Festa A, Williams K, Hanley AJ, Haffner SM. Beta-cell dysfunction in subjects with impaired glucose tolerance and early type 2 diabetes: comparison of surrogate markers with first-phase insulin secretion from an intravenous glucose tolerance test. *Diabetes* 2008;57:1638–1644
3. Lorenzo C, Wagenknecht LE, D'Agostino RB Jr, Rewers MJ, Karter AJ, Haffner SM. Insulin resistance, beta-cell dysfunction, and conversion to type 2 diabetes in a multiethnic population: the Insulin Resistance Atherosclerosis Study. *Diabetes Care* 2010;33:67–72
4. Ferrannini E, Nannipieri M, Williams K, Gonzales C, Haffner SM, Stern MP. Mode of onset of type 2 diabetes from normal or impaired glucose tolerance. *Diabetes* 2004;53:160–165
5. Guerrero-Romero F, Rodríguez-Morán M. Assessing progression to impaired glucose tolerance and type 2 diabetes mellitus. *Eur J Clin Invest* 2006;36:796–802
6. Xiang AH, Wang C, Peters RK, Trigo E, Kjos SL, Buchanan TA. Coordinate changes in plasma glucose and pancreatic beta-cell function in Latino women at high risk for type 2 diabetes. *Diabetes* 2006;55:1074–1079
7. Lyssenko V, Almgren P, Anevski D, et al.; Botnia study group. Predictors of and longitudinal changes in insulin sensitivity and secretion preceding onset of type 2 diabetes. *Diabetes* 2005;54:166–174
8. Weir GC, Bonner-Weir S. Five stages of evolving beta-cell dysfunction during progression to diabetes. *Diabetes* 2004;53(Suppl. 3):S16–S21
9. Tabák AG, Jokela M, Akbaraly TN, Brunner EJ, Kivimäki M, Witte DR. Trajectories of glycaemia, insulin sensitivity, and insulin secretion before diagnosis of type 2 diabetes: an analysis from the Whitehall II study. *Lancet* 2009;373:2215–2221
10. Lyssenko V, Jonsson A, Almgren P, et al. Clinical risk factors, DNA variants, and the development of type 2 diabetes. *N Engl J Med* 2008;359:2220–2232

11. Munoz J, Lok KH, Gower BA, et al. Polymorphism in the transcription factor 7-like 2 (TCF7L2) gene is associated with reduced insulin secretion in nondiabetic women. *Diabetes* 2006;55:3630–3634
12. Loos RJ, Franks PW, Francis RW, et al. TCF7L2 polymorphisms modulate proinsulin levels and beta-cell function in a British Europid population. *Diabetes* 2007;56:1943–1947
13. Gloyn AL, Braun M, Rorsman P. Type 2 diabetes susceptibility gene TCF7L2 and its role in beta-cell function. *Diabetes* 2009;58:800–802
14. Helgason A, Pálsson S, Thorleifsson G, et al. Refining the impact of TCF7L2 gene variants on type 2 diabetes and adaptive evolution. *Nat Genet* 2007;39:218–225
15. Hivert MF, Jablonski KA, Perreault L, et al.; DIAGRAM Consortium; Diabetes Prevention Program Research Group. Updated genetic score based on 34 confirmed type 2 diabetes loci is associated with diabetes incidence and regression to normoglycemia in the Diabetes Prevention Program. *Diabetes* 2011;60:1340–1348
16. Heni M, Ketterer C, Thamer C, et al. Glycemia determines the effect of type 2 diabetes risk genes on insulin secretion. *Diabetes* 2010;59:3247–3252
17. American Diabetes Association. Diagnosis and classification of diabetes mellitus. *Diabetes Care* 2010;33(Suppl. 1):S62–S69
18. Laspa E, Christen A, Efstathiadou Z, Johnston DG, Godsland IF. Long-term changes and variability in diabetes risk factors prior to the development of impaired glucose homeostasis. *Diabet Med* 2007;24:1269–1278
19. Sattar N, McConnachie A, Ford I, et al. Serial metabolic measurements and conversion to type 2 diabetes in the West of Scotland Coronary Prevention Study: specific elevations in alanine aminotransferase and triglycerides suggest hepatic fat accumulation as a potential contributing factor. *Diabetes* 2007;56:984–991
20. Lyssenko V, Lupi R, Marchetti P, et al. Mechanisms by which common variants in the TCF7L2 gene increase risk of type 2 diabetes. *J Clin Invest* 2007;117:2155–2163
21. Florez JC, Jablonski KA, Bayley N, et al.; Diabetes Prevention Program Research Group. TCF7L2 polymorphisms and progression to diabetes in the Diabetes Prevention Program. *N Engl J Med* 2006;355:241–250
22. Cauchi S, Meyre D, Choquet H, et al.; DESIR Study Group. TCF7L2 variation predicts hyperglycemia incidence in a French general population: the Data from an Epidemiological Study on the Insulin Resistance Syndrome (DESIR) study. *Diabetes* 2006;55:3189–3192
23. Villareal DT, Robertson H, Bell GI, et al. TCF7L2 variant rs7903146 affects the risk of type 2 diabetes by modulating incretin action. *Diabetes* 2010;59:479–485
24. Hadjiyanni I, Baggio LL, Poussier P, Drucker DJ. Exendin-4 modulates diabetes onset in nonobese diabetic mice. *Endocrinology* 2008;149:1338–1349
25. Kim SJ, Nian C, Doudet DJ, McIntosh CH. Dipeptidyl peptidase IV inhibition with MK0431 improves islet graft survival in diabetic NOD mice partially via T-cell modulation. *Diabetes* 2009;58:641–651
26. Sherry NA, Chen W, Kushner JA, et al. Exendin-4 improves reversal of diabetes in NOD mice treated with anti-CD3 monoclonal antibody by enhancing recovery of beta-cells. *Endocrinology* 2007;148:5136–5144
27. Blandino-Rosano M, Perez-Arana G, Mellado-Gil JM, Segundo C, Aguilar-Diosdado M. Anti-proliferative effect of pro-inflammatory cytokines in cultured beta cells is associated with extracellular signal-regulated kinase 1/2 pathway inhibition: protective role of glucagon-like peptide-1. *J Mol Endocrinol* 2008;41:35–44
28. Tourrel C, Bailbe D, Lacorne M, Meile MJ, Kergoat M, Portha B. Persistent improvement of type 2 diabetes in the Goto-Kakizaki rat model by expansion of the beta-cell mass during the prediabetic period with glucagon-like peptide-1 or exendin-4. *Diabetes* 2002;51:1443–1452
29. Song Y, Manson JE, Tinker L, et al. Insulin sensitivity and insulin secretion determined by homeostasis model assessment and risk of diabetes in a multiethnic cohort of women: the Women's Health Initiative Observational Study. *Diabetes Care* 2007;30:1747–1752