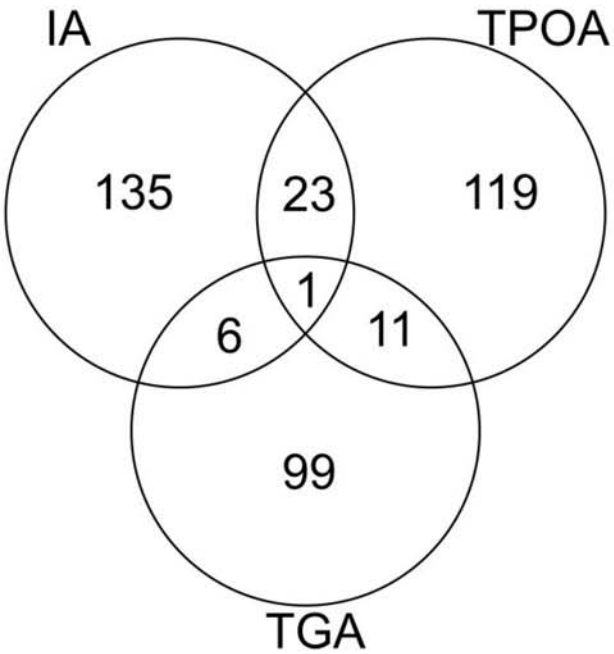
SUPPLEMENTARY DATA

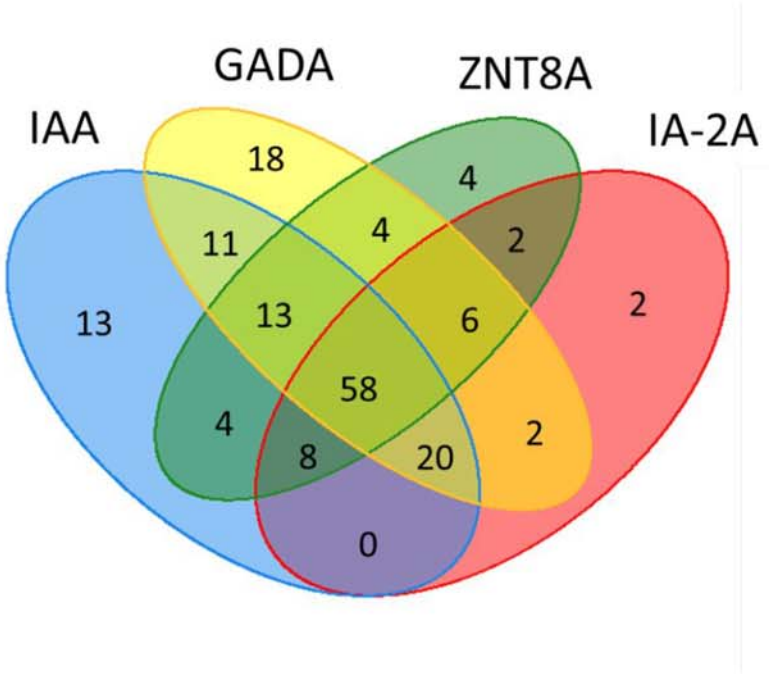
Age, HLA and sex define a marked risk of organ-specific autoimmunity in first degree relative of patients with type 1 diabetes

Christiane Winkler PhD[1,2], Manja Jolink[1], Annette Knopff[1], Nana-Adjoa Kwarteng[1], Peter Achenbach MD[1,2,3], Ezio Bonifacio PhD[4,5], Anette-G. Ziegler MD[1,2,3]

SUPPLEMENTARY DATA

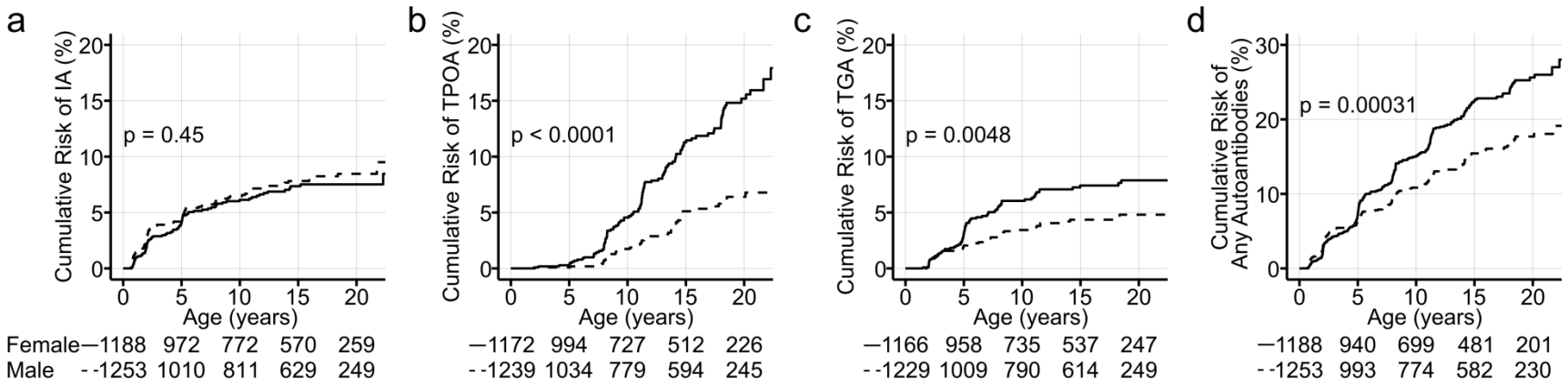
Supplementary Figure S1. Venn diagram of children with positive islet autoantibodies (IA), transglutaminase autoantibodies (TGA), and thyroid peroxidase autoantibodies (TPOA) and Venn diagram showing all islet autoantibodies positive children who developed single or multiple islet autoantibodies and the different autoantibody combination.





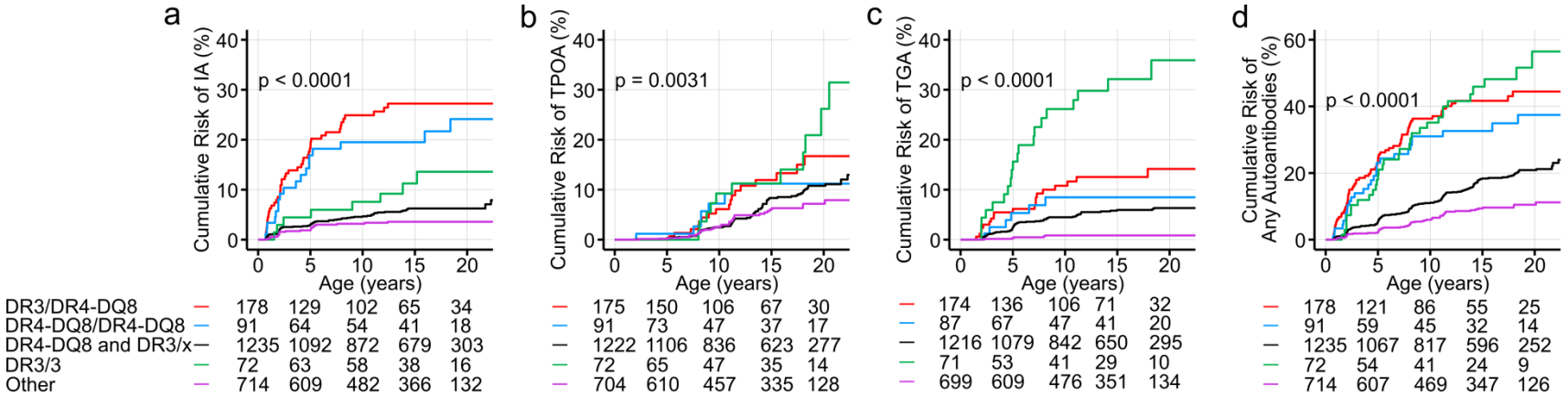
SUPPLEMENTARY DATA

Supplementary Figure S2. Cumulative risks of IA (a), TPOA (b), TGA (c), and any of the autoantibodies (d) in males (dashed curves) and females (black curves).



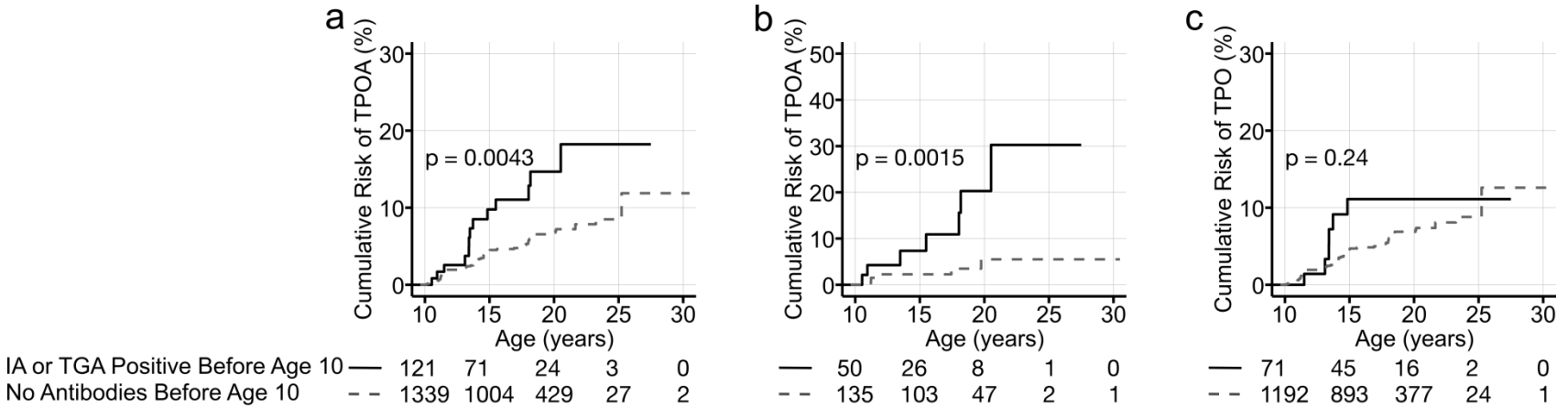
SUPPLEMENTARY DATA

Supplementary Figure S3. Cumulative risks of IA (a), TPOA (b), TGA (c), and any of the autoantibodies (d) according to HLA genotype.



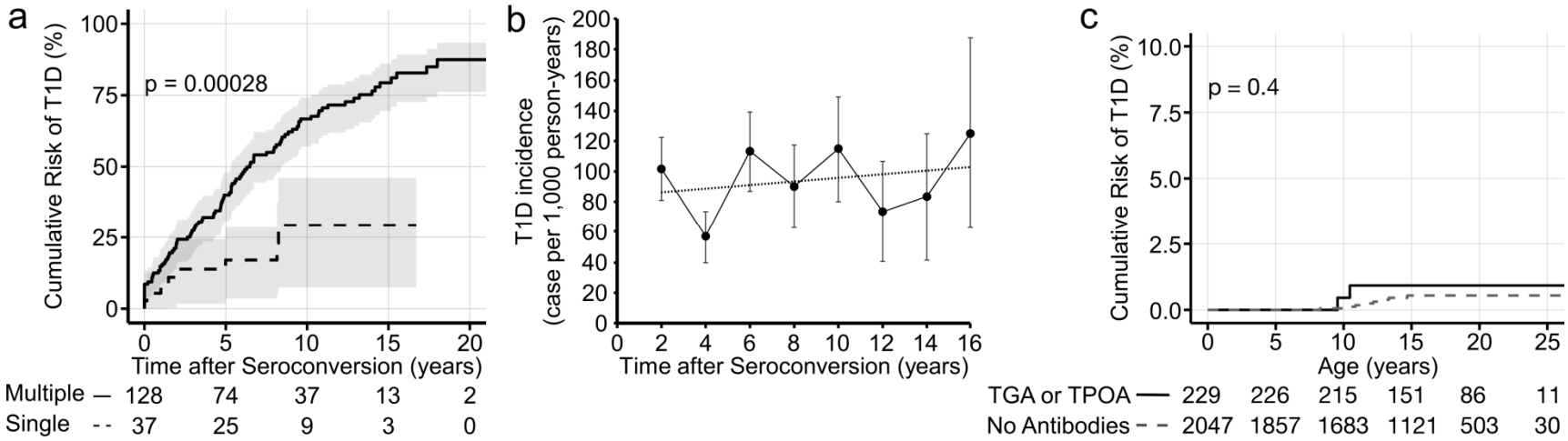
SUPPLEMENTARY DATA

Supplementary Figure S4: Cumulative risks of TPOA in participants who were positive for IA or TGA by age 10 years (black line) and in participants who were negative for IA and TGA by age 10 years (dashed line), (a) all participants; (b) in participants with the HLA DR3/3, DR3/DR4-DQ8 or DR4-DQ8/DR4-DQ8 genotype, (c) in participants with other HLA genotypes. P values were calculated with the log-rank test.



SUPPLEMENTARY DATA

Supplementary Figure S5. Cumulative risk and incidence of type 1 diabetes. (a) Kaplan-Meier curves for progression to diabetes from the time of islet autoantibody seroconversion in participants with multiple islet autoantibodies (black line) and in participants with single islet autoantibodies (gray line); (b) incidence (cases per 1,000 person-years) of progression to diabetes in children with multiple islet autoantibodies, (c) Kaplan-Meier curves for progression to diabetes from the time of transglutaminase autoantibodies or thyroid peroxidase autoantibodies seroconversion in participants without islet autoantibodies. Numbers represent participants remaining at each follow-up. Error bars represent SE. P values were calculated with the log-rank test.



SUPPLEMENTARY DATA

Supplementary Table S1. Interaction between sex and HLA in the multivariable Cox proportional hazard model for the development of autoantibodies

Step	Variable	Any autoantibody HR (95% CI)	p-value	Improvement from previous step (p-value)
1	HLA, reference x/x*			
	DR3/DR4	5.9 (4.2-8.4)	**<0.0001**	
	DR4/DR4	4.6 (2.9-7.3)	**<0.0001**	
	DR3/DR3	6.3 (4.1-9.7)	**<0.0001**	
	DR3/x, DR4/x	2.0 (1.5-2.6)	**<0.0001**	
	Sex, reference boys	1.4 (1.1-1.7)	**0.0012**	
2	HLA, reference x/x*			**0.0077**
	DR3/DR4	6.9 (4.6-10.4)	**<0.0001**	
	DR4/DR4	2.5 (1.2-5.2)	**0.015**	
	DR3/DR3	4.9 (2.6-9.3)	**<0.0001**	
	DR3/x, DR4/x	1.5 (1.0-2.1)	**0.029**	
	Interaction sex*HLA			
	Sex*DR3/DR4	0.7 (0.6-1.2)	0.23	
	Sex*DR4/DR4	2.8 (1.3-6.5)	**0.012**	
	Sex*DR3/DR3	1.6 (0.8-3.3)	0.21	
	Sex*DR3/x,DR4/x	1.7 (1.3-2.3)	**0.0002**	

*x is any haplotype other than HLA DR3 or DR4-DQ8

SUPPLEMENTARY DATA

Supplementary Table S2. 105 subjects progressed to type 1 diabetes (T1D) after seroconversion

Proband	Sex	HLA	IA status at seroconversion	IA status at T1D onset	Age (y) seroconversion,	Age (y) T1D	Seroconversion to T1D, y
1	M	DR4-DQ8/x	IAA	IA2A, GADA, ZnT8A	0.76	18.76	18.00
2	M	DR3/DR4-DQ8	IAA	IAA, IA2A, GADA, ZnT8A	0.93	18.28	17.35
3	M	DR3/x	IAA	IAA, GADA	0.80	16.31	15.51
4	M	DR3/DR4-DQ8	IAA, ZnT8A	IAA, IA2A, ZnT8A	1.11	16.28	15.17
5	M	DR4-DQ8/x	IAA	IAA, IA2A, GADA, ZnT8A	0.81	15.29	14.48
6	M	DR3/DR4-DQ8	IAA, GADA	IAA, IA2A, GADA, ZnT8A	2.25	16.43	14.18
7	F	DR4-DQ8/x	GADA, ZnT8A	IAA, GADA, ZnT8A	5.39	19.38	13.99
8	F	DR3/DR4-DQ8	IAA	GADA, ZnT8A	2.25	15.45	13.21
9	F	DR3/x	IAA	IAA, IA2A, GADA	1.00	13.88	12.88
10	M	DR4-DQ8/x	IAA, GADA	IAA, IA2A, GADA	5.23	17.58	12.34
11	M	DR3/DR4-DQ8	IAA, GADA	IAA, IA2A, GADA	5.01	16.29	11.28
12	F	DR4-DQ8/x	IAA	IAA, GADA, ZnT8A	1.03	11.95	10.93
13	M	DR4-DQ8/x	IAA, IA2A	IAA, IA2A, ZnT8A	2.15	12.88	10.73

SUPPLEMENTARY DATA

14	M	DR4-DQ8/ DR4-DQ8	IAA, IA2A, ZnT8A	IAA, IA2A, ZnT8A	4.88	15.57	10.69
15	M	DR4-DQ8/x	IAA	IAA, IA2A, GADA, ZnT8A	2.28	12.52	10.24
16	M	DR4-DQ8/x	IAA, GADA	IAA, IA2A, ZnT8A	2.01	11.60	9.60
17	M	DRx/x	IAA, IA2A, GADA	IA2A, ZnT8A	1.98	11.48	9.49
18	F	DR3/DR4-DQ8	GADA	GADA, ZnT8A	3.76	13.25	9.48
Proband	**Sex**	**HLA**	**IA status at seroconversion**	**IA status at T1D onset**	**Age (y) seroconversion,**	**Age (y) T1D**	**Seroconversion to T1D, y**
19	M	DR4-DQ8/x	GADA, ZnT8A	IAA, IA2A, GADA, ZnT8A	5.25	14.63	9.38
20	M	DR4-DQ8/ DR4-DQ8	IAA	IAA, IA2A, GADA, ZnT8A	0.83	9.93	9.10
21	F	DR3/DR4-DQ8	IAA	IAA, IA2A, ZnT8A	2.55	11.39	8.84
22	M	DR3/DR4-DQ8	IAA, GADA	IAA, GADA	2.14	10.78	8.64
23	M	DR3/DR4-DQ8	GADA	GADA	5.07	13.53	8.46
24	F	DR4-DQ8/x	IAA, IA2A, GADA	IAA, IA2A, ZnT8A	2.40	10.81	8.41
25	F	DR4-DQ8/x	IAA, GADA	GADA	3.32	11.65	8.33
26	F	DR3/3	GADA	GADA	2.43	10.67	8.24
27	M	DR3/x	GADA	GADA	9.16	17.31	8.15
28	M	DR4-DQ8/x	IA2A, ZnT8A	IA2A, ZnT8A	5.38	13.48	8.10
29	M	DR3/DR4-DQ8	IAA	IAA, GADA,	1.54	9.51	7.98

SUPPLEMENTARY DATA

				ZnT8A			
30	F	DR4-DQ8/x	IAA, IA2A, GADA, ZnT8A	IAA, IA2A, ZnT8A	4.24	12.18	7.94
31	F	DR3/DR4-DQ8	IAA, GADA	IAA, GADA	1.87	9.37	7.50
32	F	DR4/DR4-DQ8	IAA, IA2A, GADA	IAA, IA2A, ZnT8A	2.08	8.81	6.73
33	M	DR3/DR4-DQ8	IAA, GADA	IAA, IA2A, GADA, ZnT8A	2.11	8.83	6.72
34	F	DR4-DQ8/x	IAA	IAA, GADA	8.03	14.75	6.72
35	F	DR4-DQ8/ DR4-DQ8	ZnT8A	IA2A, ZnT8A	1.62	8.15	6.52
36	F	DR4-DQ8/x	ZnT8A	IAA, IA2A, GADA, ZnT8A	0.94	7.41	6.48
37	F	DR x/x	IAA	IAA, IA2A, GADA, ZnT8A	0.80	7.09	6.29
38	M	DR3/DR4-DQ8	IAA	IAA, GADA	0.72	6.87	6.16
39	F	DR4-DQ8/x	IAA, IA2A, GADA, ZnT8A	IAA, IA2A, GADA, ZnT8A	6.76	12.82	6.06
Proband	**Sex**	**HLA**	**IA status at seroconversion**	**IA status at T1D onset**	**Age (y) seroconversion,**	**Age (y) T1D**	**Seroconversion to T1D, y**
40	M	DR4-DQ8/x	IAA	IAA, IA2A, GADA	2.05	8.01	5.96
41	F	DR4-DQ8/x	IAA, IA2A, GADA	IAA, IA2A, GADA, ZnT8A	5.01	10.87	5.86
42	M	DR4-DQ8/x	IAA, GADA	IAA, IA2A, GADA	2.02	7.65	5.63
43	M	DR x/x	IAA, IA2A,	IA2A, GADA,	2.01	7.58	5.56

SUPPLEMENTARY DATA

			GADA, ZnT8A	ZnT8A			
44	F	DR4-DQ8/x	IAA, IA2A, ZnT8A	IAA, IA2A, GADA, ZnT8A	8.51	13.90	5.39
45	F	DR3/x	IAA, GADA	IAA, GADA	1.99	7.33	5.34
46	M	DR3/DR4-DQ8	IAA, IA2A, GADA, ZnT8A	IAA, IA2A, GADA, ZnT8A	5.02	10.35	5.34
47	F	DR4-DQ8/x	IAA	GADA, ZnT8A	1.88	7.21	5.33
48	M	DR3/DR4-DQ8	IAA, IA2A, ZnT8A	IAA, IA2A, GADA, ZnT8A	4.33	9.55	5.22
49	M	DR4-DQ8/ DR4-DQ8	ZnT8A	ZnT8A	4.88	9.86	4.99
50	M	DR3/DR4-DQ8	IAA, GADA	IAA, GADA, ZnT8A	4.13	9.03	4.90
51	F	DR4-DQ8/x	IAA, IA2A, GADA, ZnT8A	IAA, IA2A, GADA, ZnT8A	2.03	6.89	4.86
52	F	DR4-DQ8/ DR4-DQ8	IAA, GADA	IA2A, GADA, ZnT8A	1.93	6.74	4.80
53	F	DR3/DR4-DQ8	IAA, GADA	IAA, IA2A, GADA, ZnT8A	1.26	6.00	4.74
54	M	DR3/DR4-DQ8	IAA, IA2A, GADA, ZnT8A	IAA, IA2A, GADA, ZnT8A	2.09	6.80	4.71
55	F	DR3/3	GADA	GADA, ZnT8A	1.53	6.22	4.69
56	F	DR4-DQ8/ DR4-DQ8	IAA	IAA, IA2A, GADA, ZnT8A	0.87	5.53	4.66
57	F	DR4-DQ8/ DR4-DQ8	GADA	IAA, IA2A, GADA	4.08	8.44	4.36
58	F	DR3/3	GADA, ZnT8A	IAA, GADA,	5.05	9.33	4.27

SUPPLEMENTARY DATA

				ZnT8A			
Proband	**Sex**	**HLA**	**IA status at seroconversion**	**IA status at T1D onset**	**Age (y) seroconversion,**	**Age (y) T1D**	**Seroconversion to T1D, y**
59	M	DR3-DQ8/ DR4-DQ8	GADA	IAA, IA2A, GADA	7.90	12.13	4.24
60	M	DR3/x	GADA	IA2A, GADA	4.92	8.51	3.59
61	M	DR4-DQ8/x	IAA, IA2A, GADA, ZnT8A	IAA, IA2A, ZnT8A	2.06	5.58	3.53
62	M	DR3/DR4-DQ8	IAA, GADA, ZnT8A	IAA, IA2A, GADA, ZnT8A	2.65	5.99	3.33
63	M	DR3/DR4-DQ8	IAA	IAA, GADA, ZnT8A	0.99	4.18	3.19
64	F	DR3/x	IAA, GADA, ZnT8A	IAA, IA2A, GADA, ZnT8A	2.04	5.16	3.11
65	M	DR4-DQ8/ DR4-DQ8	IAA	IAA, IA2A, GADA, ZnT8A	0.71	3.75	3.05
66	F	DR x/x	IAA	IAA, ZnT8A	2.03	4.96	2.93
67	M	DR x/x	GADA	GADA, ZnT8A	5.35	8.22	2.87
68	F	DR4-DQ8/x	IAA, GADA	IAA, IA2A, GADA, ZnT8A	13.86	16.67	2.81
69	M	DR3/3	GADA, ZnT8A	GADA, ZnT8A	13.84	16.48	2.64
70	F	DR3/x	IAA, IA2A, GADA	IAA, IA2A, GADA, ZnT8A	5.59	7.61	2.02
71	M	DR4-DQ8/x	IAA, IA2A, GADA	IAA, IA2A, GADA, ZnT8A	2.15	4.15	1.99
72	F	DR3/DR4-DQ8	GADA	GADA	6.05	8.03	1.98

SUPPLEMENTARY DATA

73	F	DR4-DQ8/ DR4-DQ8	IAA	IAA, IA2A, GADA	1.93	3.89	1.95
74	F	DR4-DQ8/x	IAA, IA2A, GADA, ZnT8A	IA2A, GADA	0.85	2.80	1.94
75	M	DR3/DR4-DQ8	IAA, IA2A, ZnT8A	IAA, IA2A, GADA, ZnT8A	8.16	9.98	1.82
76	F	DR3/DR4-DQ8	IA2A, GADA, ZnT8A	IA2A, GADA, ZnT8A	12.05	13.77	1.72
77	M	DR x/x	IAA, IA2A, ZnT8A	IAA, IA2A, ZnT8A	2.63	4.16	1.53

Proband	**Sex**	**HLA**	**IA status at seroconversion**	**IA status at T1D onset**	**Age (y) seroconversion,**	**Age (y) T1D**	**Seroconversion to T1D, y**
78	M	DR3/DR4-DQ8	IAA, IA2A, GADA, ZnT8A	IAA, IA2A, GADA, ZnT8A	11.08	12.57	1.49
79	M	DR3/x	GADA	GADA	11.15	12.63	1.48
80	F	NA	IAA	IAA, ZnT8A	0.93	2.35	1.42
81	M	DR x/x	IAA	IAA, IA2A, ZnT8A	1.23	2.54	1.31
82	M	DR x/x	IAA	IAA, IA2A, GADA, ZnT8A	2.02	3.26	1.24
83	F	DR3/DR4-DQ8	IAA, IA2A, ZnT8A	IAA, IA2A, GADA	2.14	3.23	1.09
84	M	DR x/x	IAA	IAA	1.90	2.91	1.01
85	M	DR3/DR4-DQ8	IAA	IAA, IA2A, GADA, ZnT8A	0.88	1.84	0.96

SUPPLEMENTARY DATA

86	M	DR3/x	ZnT8A	IAA, IA2A, GADA, ZnT8A	1.40	2.36	0.96
87	F	DR x/x	IAA	IAA, IA2A, GADA	1.12	1.97	0.86
88	F	DR3/x	IAA, IA2A, GADA	IAA, IA2A, GADA	2.01	2.56	0.55
89	M	DR3/DR4-DQ8	IAA, GADA	IAA, GADA	0.84	1.34	0.51
90	M	DR3/x	IAA, IA2A, GADA	IAA, IA2A, GADA	0.64	1.11	0.46
91	F	DR4-DQ8/x	IAA, GADA	IAA, IA2A, GADA	0.82	1.29	0.46
92	M	DR4-DQ8/x	IAA, IA2A, GADA, ZnT8A	IAA, IA2A, GADA, ZnT8A	0.82	1.03	0.21
93	M	DR3/DR4-DQ8	IAA	IAA	0.86	1.01	0.15
94	F	DR3/DR4-DQ8	IAA, IA2A, GADA	IAA, IA2A, GADA	0.73	0.73	0.00
95	M	DR3/DR4-DQ8	IAA, IA2A, GADA	IAA, IA2A, GADA	1.73	1.73	0.00
96	M	DR3/3	IA2A, GADA	IA2A, GADA	1.81	1.81	0.00

Proband	**Sex**	**HLA**	**IA status at seroconversion**	**IA status at T1D onset**	**Age (y) seroconversion,**	**Age (y) T1D**	**Seroconversion to T1D, y**
97	F	DR4-DQ8/x	IAA, IA2A, GADA	IAA, IA2A, GADA	1.96	1.96	0.00
98	M	DR4-DQ8/x	IAA	IAA	2.03	2.03	0.00

SUPPLEMENTARY DATA

99	F	DR4-DQ8/ DR4-DQ8	IAA, IA2A, GADA		3.61	3.61	0.00
100	F	DR3/DR4-DQ8	GADA, ZnT8A	GADA, ZnT8A	4.28	4.28	0.00
101	F	DR3/DR4-DQ8	IAA, IA2A, GADA, ZnT8A	IAA, IA2A, GADA, ZnT8A	4.83	4.83	0.00
102	M	DR4-DQ8/x	IA2A, GADA, ZnT8A	IA2A, GADA, ZnT8A	10.44	10.44	0.00
103	F	DR3/x	IAA, IA2A, ZnT8A	IAA, IA2A, ZnT8A	11.70	11.70	0.00
104	M	NA	IA2A, GADA, ZnT8A	IA2A, GADA, ZnT8A	15.99	15.99	0.00
105	M	DR4-DQ8/x	IA2A, ZnT8A	IA2A, ZnT8A	21.74	21.74	0.00

SUPPLEMENTARY DATA

Supplementary Table S3. Five subjects with the longest periods with no progression to type 1 diabetes (T1D) after seroconversion

Proband	Sex	HLA	IA status at seroconversion	IA status last follow-up	Age (y) seroconversion	Age (y) last follow-up	Follow-up from seroconversion (y)
1	F	DR3-DQ2/DRx	IAA	GADA	0.93	24.61	23.67
2	F	DR4-DQ8/DR4-DQ8	IAA	IA2A	1.69	22.20	20.51
3	F	DR4-DQ8/DR4-DQ8	GADA	GADA, IA2A, ZnT8A	4.59	23.12	18.53
4	F	DR3-DQ8/DR4-DQ8	IAA, GADA	GADA, IA2A, ZnT8A	1.03	19.49	18.46
5	M	DR4-DQ8/x	IAA	IAA, GADA, IA2A, ZnT8A	1.76	20.20	18.44