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IA, islet autoimmunity; T1D, type 1 diabetes.

ARTICLE HIGHLIGHTS

- Children with the HLA-DR3/4 genotype demonstrated increased risk of islet autoimmunity if solid food was introduced before 6 months of age.
- The association was not present in children who were exposed to probiotics at an early age.
- It is important to investigate the function and immune responses to the host microbiome when studying early diet, including probiotics and islet autoimmunity in genetically high-risk children.

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OBJECTIVE

To study the interaction among HLA genotype, early probiotic exposure, and timing of complementary foods in relation to risk of islet autoimmunity (IA).

RESEARCH DESIGN AND METHODS

The Environmental Determinants of Diabetes in the Young (TEDDY) study prospectively follows 8,676 children with increased genetic risk of type 1 diabetes. We used a Cox proportional hazards regression model adjusting for potential confounders to study early feeding and the risk of IA in a sample of 7,770 children.

RESULTS

Any solid food introduced early (<6 months) was associated with increased risk of IA if the child had the HLA DR3/4 genotype and no probiotic exposure during the 1st year of life. Rice introduced at 4–5.9 months compared with later in the U.S. was associated with an increased risk of IA.

CONCLUSIONS

Timing of solid food introduction, including rice, may be associated with IA in children with the HLA DR3/4 genotype not exposed to probiotics. The microbiome composition under these exposure combinations requires further study.

Class II HLA haplogenotypes account for about one-half of the genetic risk for islet autoimmunity (IA) and the later progression to type 1 diabetes (1). In addition to genes, environmental factors, including early diet, have been shown to be associated with the risk of IA (2). Probiotic use any time during the first 27 days of life was inversely associated with IA among children with the high-risk HLA DR3/4 genotype for type 1 diabetes in The Environmental Determinants of Diabetes in the Young (TEDDY) study (3). The objective of the current study was to investigate the interaction among timing of introduction of complementary foods, HLA genotype, and timing of first probiotic exposure in relation to IA in the TEDDY cohort.

RESEARCH DESIGN AND METHODS

TEDDY is a prospective cohort study involving three clinical centers in the U.S. (Colorado, Georgia/Florida, Washington State), and three in Europe (Finland, Germany,

BRIEF REPORT

and Sweden). The detailed study design and methods have been described previously (4–6). The study population is presented in Supplementary Fig. 1, and population characteristics in Supplementary Table 2. The final sample size was 7,770. The food exposures and categorization of timing are described in Table 1.

Infant gut microbiota goes through significant changes over the 1st year of life (7). Therefore, we also studied the timing of the initial probiotic exposure either from dietary supplements or from infant formula during the first 52 weeks. We also considered only early exposures before 26 weeks of age. We did not analyze findings during the first 4 weeks of life, as reported earlier (3), because these subgroup numbers were insufficient. Probiotics mainly included *Lactobacillus reuteri* and *Lactobacillus rhamnosus*. The length of probiotic use was not examined in this observational study.

IA

Persistent confirmed IA was defined by the presence of one or several autoantibodies against GAD (GADA), IA-2 antigen (IA-2A), or insulin (IAA) at each of the two TEDDY laboratories on two or more consecutive visits. The detailed study design and methods have been previously published (4,5). The timing of seroconversion was defined as the age of the first persistent confirmed autoantibody sample and the right-censored time as the age when the last blood sample available was determined as negative for IA.

Statistical Analysis

A Cox proportional hazards regression model was used to investigate the association between timing of food exposures and the risk of IA in the TEDDY cohort. Interactions between timing of food exposure and HLA genotype (DR3/4 compared with any other genotype than DR3/4) and between timing of food exposure and first probiotics were studied while controlling for country, whether any first-degree relative had type 1 diabetes, and sex of the child. Response variables included the risk of developing IA overall, IAA only as the first-appearing autoantibody (IAA-first), GADA only as the first-appearing autoantibody (GADA-first), or multiple autoantibodies appearing simultaneously. We also conducted three-way interaction models to examine whether the association between timing of selected foods and the risk of IA was modified by HLA DR3/4 and by the first exposure to probiotics. All statistical analyses were done using SAS 9.4 software (SAS/STAT 15.2).

Table 1—Food exposures

	Categorization of timing of food introduction by age (months)		
Food	Early or short duration		Late (reference)
Exclusive breastfeeding	<4		≥4
Any breastfeeding	<4		≥4
Any infant formula	<4	4 to <6	≥6
Any solid food†	<4	4 to <6	≥6
All cereals	<4	4 to <6	≥6
Gluten-containing cereals	<4	4 to <6	≥6
Nongluten-containing cereals	<4	4 to <6	≥6
Fruits and berries	<4	4 to <6	≥6
Root vegetables	<4	4 to <6	≥6
Other vegetables than roots	<4	4 to <6	≥6
Regular cow's milk	<4	4 to <6	≥6
Any meat‡	<4	4 to <6	≥6
Egg	≤9		>9
Rice*	<4	4 to <6	≥6
Oat*	<4	4 to <6	≥6

†All cereals (including gluten and nongluten), fruits and berries, all vegetables (including roots), milk products, eggs, any meat (including red meat, poultry, fish and seafood, processed meats). ‡Including all red meat, poultry, fish, and seafood. *Preliminary analyses suggested that nongluten cereals played a role in the associations between any solid food and the outcomes, and therefore, we additionally studied two of the most commonly consumed nongluten baby cereals, rice and oat, and their timing of introduction in relation to outcomes separately by country.

RESULTS

Main Effects

Early introduction of gluten-containing cereals was associated with a decreased risk of any IA, GADA-first, and multiple autoantibodies (Supplementary Tables 3–6). Wheat (consumed alone or with another cereal) accounted for 90% of the first exposures to gluten-containing cereals before 6 months of age.

Subgroups

There was an interaction between timing of introduction of fruit and berries and HLA genotype (DR3/4 vs. other) when multiple autoantibodies were studied as an outcome. Similarly, an interaction between timing of any solid food and first probiotics within the first 52 weeks in relation to multiple autoantibodies was observed. Furthermore, the interactions between timing of egg introduction and first probiotics in relation to IAA-first and GADA-first were found (Table 2).

Both HLA genotype and probiotic exposure together modified the association between timing of any solid food introduction and risk of the outcomes (Fig. 1 and Supplementary Table 7). Among children who carried HLA DR3/4 and who were not exposed to probiotics during their first 52 weeks of life, early introduction of any solid food was associated with an increased risk of any IA, IAA-first, and multiple autoantibodies. However, if probiotics were introduced before 52 weeks, none of these associations were present in the subgroup of children with HLA DR3/4 (Fig. 1). The change in direction in the association by probiotics at <52 weeks was found only among children carrying a DR3 allele. Duration of breastfeeding was not associated with the risk of IA.

Gluten-Containing Cereals, Nongluten-Containing Cereals, and Cereals Overall

Both HLA DR3/4 genotype and exposure to probiotics modified the association

Table 2—Timing of introduction of selected complementary foods and risk of developing IA by HLA genotype or by use of probiotics during the first 52 weeks of life

Timing of first food exposure (months) and outcome	HLA genotype						Use of probiotics during the first 52 weeks***					
			Other than HLA DR3/4			HLA DR3/4			No probiotic exposure before 52 weeks of age			Probiotic exposure before or at 52 weeks of age
	n	Affected, *n*	HR (95% CI), *P**	*n*	Affected, *n*	HR (95% CI), *P**	*n*	Affected, *n*	HR (95% CI), *P***	*n*	Affected, *n*	HR (95% CI), *P***
Any solid foods												
Any IA												
<4	1,840	145	0.93 (0.66, 1.30), 0.656	1,192	159	1.31 (0.90, 1.91), 0.153	2,367	234	1.20 (0.88, 1.62), 0.245	665	70	0.91 (0.57, 1.44), 0.678
4 to <6	2,393	243	1.09 (0.78, 1.50), 0.620	1,528	219	1.31 (0.91, 1.89), 0.151	3,070	364	1.31 (0.98, 1.76), 0.069	851	98	0.91 (0.59, 1.42), 0.688
≥6	495	48	1	322	36	1	621	55	1	196	29	1
Interaction *P*			0.209						0.154			
IAA-first												
<4	1,840	56	0.96 (0.54, 1.69), 0.880	1,192	60	1.78 (0.90, 3.53), 0.098	2,367	88	1.45 (0.85, 2.46), 0.174	665	28	0.90 (0.42, 1.93), 0.777
4 to <6	2,393	92	1.13 (0.65, 1.94), 0.670	1,528	76	1.70 (0.86, 3.34), 0.126	3,070	131	1.55 (0.92, 2.60), 0.101	851	38	0.90 (0.43, 1.87), 0.769
≥6	495	17	1	322	10	1	621	17	1	196	10	1
Interaction *P*			0.290						0.396			
GADA-first												
<4	1,840	55	0.84 (0.48, 1.46), 0.543	1,192	68	1.09 (0.64, 1.86), 0.754	2,367	94	0.93 (0.60, 1.44), 0.749	665	29	1.17 (0.53, 2.59), 0.692
4 to <6	2,393	118	1.28 (0.76, 2.15), 0.350	1,528	101	1.12 (0.66, 1.89), 0.667	3,070	174	1.22 (0.80, 1.86), 0.358	851	43	1.22 (0.57, 2.60), 0.609
≥6	495	18	1	322	18	1	621	27	1	196	9	1
Interaction *P*			0.307						0.732			
Multiple autoantibodies												
<4	1,840	69	0.86 (0.54, 1.38), 0.531	1,192	99	1.28 (0.81, 2.03), 0.289	2,367	122	1.29 (0.85, 1.95), 0.234	665	46	0.77 (0.45, 1.34), 0.358
4 to <6	2,393	129	1.14 (0.73, 1.76), 0.572	1,528	142	1.37 (0.87, 2.14), 0.172	3,070	212	**1.61 (1.08, 2.41), 0.020**	851	59	0.73 (0.43, 1.23), 0.234
≥6	495	27	1	322	24	1	621	29	1	196	22	1
Interaction *P*			0.236						0.028			
Cereals (24 missing)												
Any IA												
<4	1,101	81	0.86 (0.63, 1.19), 0.371	762	89	1.03 (0.74, 1.45), 0.844	1,501	135	1.06 (0.80, 1.40), 0.689	362	35	0.76 (0.48, 1.19), 0.224
4 to <6	2,807	273	1.01 (0.78, 1.32), 0.927	1,744	262	1.22 (0.91, 1.63), 0.184	3,585	428	1.25 (0.98, 1.59), 0.072	966	107	0.84 (0.59, 1.20), 0.335
≥6	805	82	1	527	62	1	952	89	1	380	55	1
Interaction *P*			0.447						0.051			
IAA-first												
<4	1,101	32	0.88 (0.52, 1.47), 0.617	762	33	1.26 (0.71, 2.25), 0.435	1,501	49	1.05 (0.67, 1.66), 0.820	362	16	1.02 (0.50, 2.07), 0.960
4 to <6	2,807	101	0.96 (0.63, 1.48), 0.858	1,744	94	1.50 (0.90, 2.52), 0.121	3,585	154	1.24 (0.84, 1.85), 0.277	966	41	0.96 (0.53, 1.73), 0.891
≥6	805	32	1	527	19	1	952	33	1	380	18	1
Interaction *P*			0.402						0.522			
GADA-first												
<4	1,101	31	0.79 (0.47, 1.32), 0.364	762	34	0.89 (0.53, 1.50), 0.252	1,501	52	0.93 (0.61, 1.44), 0.756	362	13	0.69 (0.33, 1.43), 0.315
4 to <6	2,807	130	1.16 (0.76, 1.77), 0.502	1,744	125	1.29 (0.83, 2.00), 0.697	3,585	207	1.38 (0.96, 1.99), 0.085	966	48	0.94 (0.53, 1.66), 0.832
≥6	805	30	1	527	27	1	952	37	1	380	20	1
Interaction *P*			0.957						0.368			

Continued on p. 1842

Table 2—Continued

Timing of first food exposure (months) and outcome	HLA genotype						Use of probiotics during the first 52 weeks***					
	n	Affected, *n*	Other than HLA DR3/4 HR (95% CI), *P**	*n*	Affected, *n*	HLA DR3/4 HR (95% CI), *P**	*n*	Affected, *n*	No probiotic exposure before 52 weeks of age HR (95% CI), *P***	*n*	Affected, *n*	Probiotic exposure before or at 52 weeks of age HR (95% CI), *P***
Multiple autoantibodies												
<4	1,101	40	0.82 (0.52, 1.27), 0.371	762	53	0.91 (0.60, 1.38), 0.660	1,501	70	0.95 (0.66, 1.37), 0.782	362	23	0.76 (0.44, 1.33), 0.337
4 to <6	2,807	137	0.97 (0.68, 1.39), 0.877	1,744	169	1.21 (0.85, 1.72), 0.298	3,585	238	1.24 (0.90, 1.69), 0.184	966	68	0.84 (0.54, 1.29), 0.420
≥6	805	48	1	527	43	1	952	55	1	380	36	1
Interaction *P*			0.430						0.240			
Gluten-containing cereals (134 missing)												
Any IA												
<4	294	14	**0.49 (0.28, 0.84), 0.010**	213	22	0.81 (0.52, 1.27), 0.359	410	31	**0.68 (0.46, 0.99), 0.042**	97	5	0.52 (0.21, 1.28), 0.155
4 to <6	1,624	162	0.97 (0.77, 1.21), 0.765	1,057	160	1.01 (0.80, 1.27), 0.918	2,116	254	0.95 (0.79, 1.14), 0.580	565	68	1.13 (0.82, 1.56), 0.454
≥6	2,723	259	1	1,725	230	1	3,421	365	1	1,027	124	1
Interaction *P*			0.397						0.636			
IAA-first												
<4	294	**7**	0.65 (0.295, 1.43), 0.281	213	7	0.73 (0.33, 1.62), 0.442	410	14	0.84 (0.47, 1.48), 0.539	97	0	–
4 to <6	1,624	58	0.88 (0.61, 1.28), 0.509	1,057	52	0.92 (0.63, 1.36), 0.670	2,116	83	0.84 (0.61, 1.14), 0.255	565	27	1.15 (0.70, 1.91), 0.578
≥6	2,723	100	1	1,725	86	1	3,421	138	1	1,027	48	1
Interaction *P*			0.992						0.798			
GADA-first												
<4	294	4	**0.33 (0.12, 0.90), 0.030**	213	9	0.73 (0.36, 1.47), 0.377	410	10	**0.51 (0.26, 0.98), 0.042**	97	3	0.67 (0.20, 2.21), 0.505
4 to <6	1,624	82	1.18 (0.84, 1.66), 0.330	1,057	75	1.04 (0.74, 1.46), 0.823	2,116	128	1.12 (0.86, 1.48), 0.404	565	29	1.09 (0.65, 1.82), 0.748
≥6	2,723	105	1	1,725	102	1	3,421	158	1	1,027	49	1
Interaction *P*			0.319						0.804			
Multiple autoantibodies												
<4	294	3	**0.19 (0.06, 0.59), 0.004**	213	12	0.76 (0.41, 1.38), 0.365	410	12	**0.46 (0.25, 0.83), 0.010**	97	3	0.52 (0.16, 1.67), 0.271
4 to <6	1,624	74	0.78 (0.57, 1.07), 0.127	1,057	106	1.16 (0.87, 1.53), 0.317	2,116	136	0.91 (0.71, 1.16), 0.429	565	44	1.16 (0.78, 1.72), 0.465
≥6	2,723	147	1	1,725	147	1	3,421	214	1	1,027	80	1
Interaction *P*			0.063						0.916			
Nongluten-containing cereals (29 missing)												
Any IA												
<4	1,029	79	0.89 (0.65, 1.23), 0.486	712	83	0.99 (0.71, 1.38), 0.948	1,415	128	1.01 (0.77, 1.32), 0.946	326	34	0.83 (0.53, 1.31), 0.418
4 to <6	2,830	270	0.98 (0.75, 1.27), 0.870	1,759	261	1.15 (0.87, 1.53), 0.323	3,601	424	1.17 (0.93, 1.47), 0.192	988	107	0.84 (0.59, 1.19), 0.326
≥6	850	87	1	561	69	1	1,018	100	1	393	56	1
Interaction *P*			0.523						0.092			
IAA-first												
<4	1,029	28	0.90 (0.54, 1.50), 0.675	712	35	1.08 (0.61, 1.89), 0.796	1,415	31	0.91 (0.59, 1.42), 0.696	326	32	1.19 (0.58, 2.43), 0.927
4 to <6	2,830	99	0.93 (0.61, 1.41), 0.723	1,759	107	1.29 (0.79, 2.08), 0.307	3,601	100	1.09 (0.75, 1.58), 0.648	988	106	0.97 (0.54, 1.76), 0.773
≥6	850	28	1	561	26	1	1,018	34	1	393	20	1
Interaction *P*			0.586						0.475			

Continued on p. 1843

Table 2—Continued

Timing of first food exposure (months) and outcome	HLA genotype						Use of probiotics during the first 52 weeks***					
			Other than HLA DR3/4			HLA DR3/4			No probiotic exposure before 52 weeks of age			Probiotic exposure before or at 52 weeks of age
	n	Affected, *n*	HR (95% CI), *P**	*n*	Affected, *n*	HR (95% CI), *P**	*n*	Affected, *n*	HR (95% CI), *P***	*n*	Affected, *n*	HR (95% CI), *P***
GADA-first												
<4	1,029	30	0.80 (0.48, 1.35), 0.406	712	32	0.93 (0.55, 1.56), 0.776	1,415	50	0.94 (0.62, 1.45), 0.788	326	12	0.72 (0.34, 1.52), 0.386
4 to <6	2,830	129	1.12 (0.74, 1.69), 0.595	1,759	126	1.34 (0.87, 2.07), 0.186	3,601	206	1.35 (0.95, 1.93), 0.095	988	49	0.98 (0.56, 1.73), 0.943
≥6	850	32	1	561	28	1	1,018	40	1	393	20	1
Interaction *P*			0.888						0.451			
Multiple autoantibodies												
<4	1,029	40	0.91 (0.58, 1.41), 0.660	712	49	0.88 (0.58, 1.33), 0.543	1,415	67	0.96 (0.67, 1.39), 0.844	326	22	0.81 (0.46, 1.42), 0.456
4 to <6	2,830	136	0.99 (0.69, 1.41), 0.936	1,759	169	1.17 (0.83, 1.65), 0.376	3,601	237	1.23 (0.91, 1.66), 0.188	988	68	0.82 (0.53, 1.27), 0.380
≥6	850	49	1	561	47	1	1,018	59	1	393	37	1
Interaction *P*			0.480						0.213			
Fruits and berries (37 missing)												
Any IA												
<4	1,053	69	**0.69 (0.51, 0.94), 0.017**	690	84	1.03 (0.77, 1.39), 0.835	1,341	116	0.85 (0.67, 1.09), 0.199	402	37	0.83 (0.53, 1.30), 0.413
4 to <6	2,481	248	0.96 (0.76, 1.22), 0.751	1,584	230	1.09 (0.85, 1.40), 0.514	3,124	367	1.03 (0.85, 1.25), 0.759	941	111	1.00 (0.69, 1.44), 0.987
≥6	1,169	118	1	756	98	1	1,566	167	1	359	49	1
Interaction *P*			0.120						0.617			
IAA-first												
<4	1,053	26	0.76 (0.46, 1.28), 0.303	690	32	1.12 (0.68, 1.83), 0.665	1,341	32	0.96 (0.64, 1.44), 0.843	402	14	0.81 (0.38, 1.71), 0.575
4 to <6	2,481	101	1.19 (0.80, 1.77), 0.397	1,584	89	1.12 (0.73, 1.72), 0.603	3,124	80	1.16 (0.83, 1.62), 0.375	941	45	1.11 (0.60, 2.05), 0.746
≥6	1,169	38	1	756	33	1	1,566	33	1	359	16	1
Interaction *P*			0.290						0.880			
GADA-first												
<4	1,053	29	**0.61 (0.38, 0.96), 0.034**	690	33	0.95 (0.60, 1.51), 0.834	1,341	46	0.70 (0.49, 1.02), 0.061	402	16	1.01 (0.50, 2.07), 0.970
4 to <6	2,481	106	0.83 (0.59, 1.18), 0.303	1,584	10	1.20 (0.82, 1.75), 0.348	3,124	169	0.98 (0.74, 1.30), 0.865	941	47	1.12 (0.62, 2.03), 0.710
≥6	1,169	56	1	756	43	1	1,566	81	1	359	18	1
Interaction *P*			0.283						0.772			
Multiple autoantibodies												
<4	1,053	30	**0.54 (0.34, 0.84), 0.006**	690	57	1.08 (0.75, 1.56), 0.682	1,341	64	0.84 (0.61, 1.17), 0.305	402	23	0.69 (0.40, 1.21), 0.198
4 to <6	2,481	127	0.90 (0.65, 1.25), 0.533	1,584	144	1.09 (0.80, 1.49), 0.602	3,124	202	1.04 (0.80, 1.35), 0.756	941	69	0.86 (0.55, 1.34), 0.504
≥6	1,169	67	1	756	64	1	1,566	96	1	359	35	1
Interaction *P*			0.035						0.507			
Egg (470 missing)												
Any IA												
≤9	3,098	286	0.99 (0.80, 1.23), 0.947	2,020	282	1.00 (0.80, 1.24), 0.974	4,082	445	0.94 (0.79, 1.12), 0.515	1,036	123	1.16 (0.85, 1.58), 0.350
>9	1,353	450	1	886	126	1	1,663	192	1	576	69	1
Interaction *P*			0.801						0.466			

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Table 2—Continued

Timing of first food exposure (months) and outcome	HLA genotype						Use of probiotics during the first 52 weeks***					
	n	Affected, *n*	Other than HLA DR3/4 HR (95% CI), *P**	*n*	Affected, *n*	HLA DR3/4 HR (95% CI), *P**	*n*	Affected, *n*	No probiotic exposure before 52 weeks of age HR (95% CI), *P***	*n*	Affected, *n*	Probiotic exposure before or at 52 weeks of age HR (95% CI), *P***
IAA-first												
≤9	3,098	107	0.93 (0.66, 1.31), 0.682	2,020	97	0.96 (0.67, 1.37), 0.811	4,082	166	1.09 (0.81, 1.47), 0.554	1,036	38	0.63 (0.40, 1.01), 0.053
>9	1,353	54	1	886	47	1	1,663	64	1	576	37	1
Interaction *P*			0.911						0.038			
GADA-first												
≤9	3,098	130	1.02 (0.74, 1.42), 0.898	2,020	132	1.08 (0.78, 1.50), 0.651	4,082	200	0.86 (0.67, 1.11), 0.245	1,036	62	**2.26 (1.29, 3.97), 0.004**
>9	1,353	55	1	886	53	1	1,663	91	1	576	17	1
Interaction *P*			0.904						0.004			
Multiple autoantibodies												
≤9	3,098	135	0.83 (0.63, 1.11), 0.210	2,020	176	0.95 (0.72, 1.23), 0.677	4,082	236	0.85 (0.68, 1.06), 0.153	1,036	75	1.01 (0.6, 1.48), 0.942
>9	1,353	81	1	886	86	1	1,663	119	1	576	48	1
Interaction *P*			0.321						0.475			

Boldface indicates significance at $P < 0.05$. *Adjusted for country, first-degree family member with type 1 diabetes status, sex of the child, and probiotic exposure during the 1st year of life (52 weeks). **Adjusted for country, first-degree family member with type 1 diabetes status, sex of the child, and high-risk genotype (HLA DR3/4). ***When the timing of first probiotic exposure was studied in categories <26 weeks, and ≥26 weeks, or none, slightly stronger associations were found, but they did not affect the interpretation of the results.

between early introduction of gluten-containing cereals and the outcomes (i.e., IA, GADA-first, and multiple autoantibodies) (Table 2). Children with the HLA DR3/4 genotype exposed to probiotics before the age of 52 weeks had an increased risk of IA and GADA-first if gluten-containing cereals were introduced between age 4 and 6 months compared with later (three-way interaction) (Fig. 1). However, among children with other HLA genotypes, early introduction of gluten-containing cereals was inversely associated with the risk of any IA if no probiotics were given before age of 52 weeks.

Country-Specific Analyses

There was an interaction between timing of rice introduction and country (P = 0.036) but not between timing of oat introduction and country. Only the U.S. and Sweden had a sufficient number of children in the subgroups to study the interaction. Timing of first rice cereal between age 4 and 6 months compared with later was associated with an increased risk of IA in the U.S. (hazard ratio [HR] 1.74; 95% CI 1.27, 2.38; $P < 0.0005$) but not in other countries (Table 3). U.S. children without probiotic exposure during the first 52 weeks, regardless of the HLA genotype, had an HR of 1.69 (1.22, 2.34; P = 0.0017) for the risk of any IA and 1.76 (1.10, 2.82; P = 0.019) for GADA-first when timing of rice introduction was between age 4 and 6 months compared with later.

CONCLUSIONS

As published before, early introduction of gluten-containing cereals overall was linked to a decreased risk of IA in the geographically diverse population of TEDDY (8). We also confirmed that the risk of IA related to early introduction of any solid food among children with the highest level of HLA genetic risk (DR3/4) may be modified by probiotics, although the association was not as strong as previously observed in the younger cohort of TEDDY participants (9). A novel finding was that early exposure to egg (age <9 months) is associated with an increased risk of GADA-first only in those who were exposed to probiotics.

Immune or microbiota responses to gluten-containing cereals may depend on both the HLA genotype and probiotic

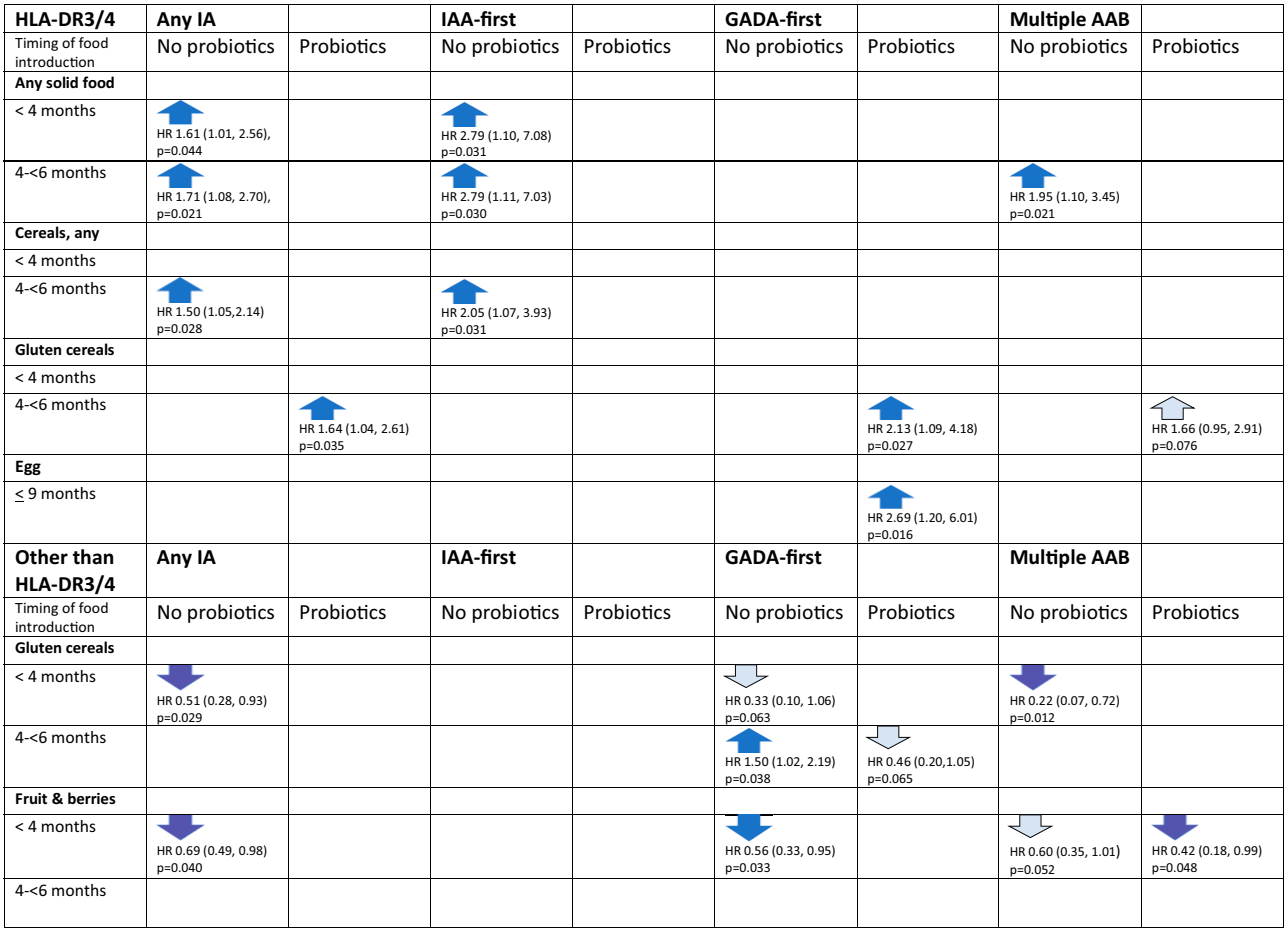

Figure 1—Timing of the introduction of foods and the risk of developing any IA, IAA-first, GADA-first, and multiple autoantibodies by HLA genotype and by probiotic exposure by 52 weeks of age, showing only the statistically significant associations. The HR from the Cox proportional hazard model (with 95% CI) uses the reference of ≥6 months, except >9 months for egg. Dark-colored arrows flag $P < 0.05$, and light-colored arrows flag $0.05 < P < 0.09$. Statistically significant three-way interactions between HLA genotype, timing of probiotic exposure, and timing of gluten cereals introduction: $P = 0.034$ for any IA and $P = 0.019$ for GADA-first, and between HLA genotype, timing of probiotic exposure, and timing of egg introduction: $P = 0.023$ for multiple autoantibodies.

exposure, and they could interact with each other. Molecular mechanisms that drive probiotic effects that may interact with genotype and food are not well understood (10). Nevertheless, gluten in cereals can act as a double-edged sword in its connection to the risk of type 1 diabetes (11,12). Gluten in wheat, barley, and rye are suggested to increase the risk of IA by promoting gut permeability and dysbiosis and to increase proinflammatory cytokines (13). Whole-grain wheat also contains several bioactive compounds promoting overall health, such as prebiotic oligosaccharides, which are linked to healthy gut microbiota (14).

The Infant Feeding Practices study (15) concluded that introduction of solid complementary foods before 4–6 months of age poses a greater risk to infant health than does infant formula. In our study, we noticed an increased risk of any IA and IAA-first with early introduction of any solid foods but only among those who were carrying the HLA DR3/4 (DR3) genotype and who did not have probiotic exposure.

The association between early timing of rice and increased risk of any IA in U.S. TEDDY children was intriguing. A somewhat toxic form of inorganic arsenic is found in relatively large quantities in rice of U.S. origin, especially if grown in southern states (16). Arsenic is a toxic trace element that can affect β-cell function and increase the risk of type 1 diabetes in youth (17) and may possibly interact with the gut microbiome (18). To decrease the potential of adverse health effects, the U.S. Food and Drug Administration has recently given guidelines for industry to reduce the arsenic content of infant rice cereals to the of level 100 parts per billion, which should be achievable under current good manufacturing practices (19). The association with the outcome was found with rice exposure between age 4 and 6 months but not earlier. During this time, children are introduced to larger quantities of solid foods. Therefore, the exposure effect of possible contaminants may be stronger than with small tastings provided earlier.

It will be important to investigate the function and immune responses of the host microbiome when studying early diet, including probiotic usage in children with a genetically increased risk of type 1 diabetes. Rice as an early food also requires further attention. The results of this study do not impose any

Table 3—Country-specific associations between timing of food introduction and IA

	U.S.			Finland			Germany			Sweden		
Timing of first food exposure (months)	Developed IA, *n* (%)	No IA, *n* (%)	HR (95% CI), *P**	Developed IA, *n* (%)	No IA, *n* (%)	HR (95% CI), *P**	Developed IA, *n* (%)	No IA, *n* (%)	HR (95% CI), *P**	Developed IA, *n* (%)	No IA, *n* (%)	HR (95% CI), *P**
Any solid food												
<4	112 (8.7)	1,169 (91.3)	**1.78 (1.17, 2.69), 0.0066**	82 (11.6)	627 (88.4)	0.67 (0.43, 1.03), 0.070	9 (6.3)	133 (93.7)	0.68 (0.31, 1.50), 0.340	101 (11.2)	799 (88.8)	0.75 (0.34, 1.62), 0.460
4 to <6	150 (10.3)	1,301 (89.7)	**1.97 (1.32, 2.96), 0.001**	100 (11.8)	751 (88.2)	**0.64 (0.41, 0.98), 0.039**	28 (12.4)	197 (87.6)	1.07 (0.61, 1.870, 0.813	184 (13.2)	1,210 (86.8)	0.82 (0.38, 1.76), 0.608
≥6	28 (5.8)	452 (94.2)	1	26 (19.0)	111 (81.0)	1	23 (13.9)	142 (86.1)	1	7 (20.0)	28 (80.0)	1
Gluten-containing cereals												
<4	8 (6.2)	122 (93.8)	0.74 (0.36, 1.49), 0.392	3 (5.3)	54 (94.7)	0.42 (0.13, 1.31), 0.132	1 (2.6)	38 (97.4)	0.30 (0.04, 2.22), 0.240	24 (8.5)	257 (91.5)	0.73 (0.46, 1.16), 0.179
4 to <6	47 (8.4)	512 (91.6)	0.91 (0.67, 1.25), 0.565	71 (13.0)	477 (87.0)	1.07 (0.80, 1.42), 0.665	6 (6.7)	83 (93.3)	0.66 (0.28, 1.54), 0.331	198 (13.3)	1,287 (86.7)	1.05 (0.80, 1.38), 0.740
≥6	234 (9.6)	2,204 (90.4)	1	133 (12.5)	935 (87.5)	1	53 (13.5)	340 (86.4)	1	69 (12.6)	480 (87.4)	1
Missing	85			24			11			14		
Nongluten-containing cereals												
<4	66 (7.7)	787 (92.3)	1.19 (0.82, 1.73), 0.363	40 (11.9)	296 (88.1)	0.86 (0.57, 1.31), 0.486	2 (4.3)	45 (95.7)	0.47 (0.11, 1.95) 0.298	54 (10.7)	451 (89.3)	0.89 (0.51, 1.56) 0.690
4 to <6	176 (10.4)	1,523 (89.6)	**1.55 (1.13, 2.14), 0.007**	118 (11.6)	903 (88.4)	0.78 (0.56, 1.09), 0.149	16 (9.4)	154 (90.6)	0.75 (0.42, 1.33), 0.323	221 13.0)	1,478 (87.0)	0.99 (0.59, 1.65), 0.962
≥6	48 (7.3)	606 (92.7)	1	50 (15.0)	284 (85.0)	1	42 (13.6)	268 (86.5)	1	16 (14.2)	97 (85.8)	1
Missing	6			6			5			12		
Rice												
<4	61 (7.8)	720 (92.2)	1.29 (0.89, 1.87), 0.185	1 (2.2)	44 (97.8)	0.20 (0.03, 1.40), 0.104	1 (2.6)	37 (97.4)	0.26 (0.04, 1.90), 0.185	23 (9.0)	233 (91.0)	0.77 (0.49, 1.21), 0.259
4 to <6	178 (10.7)	1,480 (89.3)	**1.74 (1.27, 2.38), 0.0005**	89 (12.3)	634 (87.7)	0.97 (0.73, 1.28), 0.815	15 (10.3)	131 (89.7)	0.86 (0.48, 1.55), 0.614	176 (13.1)	1,164 (86.9)	1.03 (0.80, 1.33), 0.824
≥6	51 (6.8)	705 (93.2)	1	117 (13.2)	772 (86.8)	1	44 (13.2)	289 (86.8)	1	92 (12.9)	620 (87.1)	1
Missing	17			40			15			21		
Oat												
<4	12 (6.9)	163 (93.1)	0.82 (0.46, 1.46), 0.494	4 (14.8)	23 (85.2)	1.31 (0.48, 3.59), 0.596	0	7 (100.0)	0	20 (8.4)	218 (91.6)	0.78 (0.48, 1.28), 0.327
4 to <6	84 (9.0)	849 (91.0)	0.96 (0.74, 1.24), 0.736	103 (11.4)	798 (88.6)	0.89 (0.67, 1.17), 0.402	4 (8.5)	43 (91.5)	0.87 (0.31, 2.43), 0.988	197 (13.3)	1,286 (86.7)	1.04 (0.79, 1.35), 0.796
≥6	190 (9.5)	1,816 (90.5)	1	100 (13.4)	648 (86.6)	1	55 (12.7)	378 (87.3)	1	74 (12.6)	513 (87.4)	1
Missing	98			21			45			21		
Fruits and berries**												
<4	59 (8.0)	680 (92.0)	1.16 (0.84, 1.61), 0.368	50 (11.4)	389 (88.6)	0.72 (0.48, 1.08), 0.1114	4 (5.1)	75 (94.9)	0.47 (0.17, 1.33), 0.157	40 (8.2)	446 (91.8)	**0.61 (0.39, 0.95), 0.029**
4 to <6	137 (10.5)	1,161 (89.5)	**1.42 (1.09, 1.85), 0.0087**	112 (11.4)	874 (88.6)	**0.70 (0.49, 0.98), 0.040**	15 (8.3)	165 (91.7)	0.64 (0.35, 1.17), 0.147	214 (13.4)	1,387 (86.6)	0.91 (0.64, 1.29), 0.597
≥6	93 (8.0)	1,067 (92.0)	1	45 (17.1)	219 (82.9)	1	41 (15.4)	225 (84.6)	1	37 15.7)	198 (84.3)	1
Missing	15			8			7			7		

Boldface indicates significance at $P < 0.05$. *Adjusted for first-degree family member with type 1 diabetes status, sex of the child, probiotic exposure during the 1st year of life (52 weeks), and high-risk genotype (HLA DR3/4). **Fruits and berries are often served together with baby porridge.

changes in the current recommendations on infant feeding.

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