An Interval Tightly Linked to but Distinct From the H2 Complex Controls Both Overt Diabetes (*Idd16*) and Chronic Experimental Autoimmune Thyroiditis (*Ceat1*) in Nonobese Diabetic Mice

Olivier Boulard,[1] Diane Damotte,[2] Nathalie Deruytter,[1] Guy Fluteau,[1] Claude Carnaud,[1] and Henri-Jean Garchon[1]

The major histocompatibility complex (MHC) has long been associated with predisposition to several autoimmune diseases, including type 1 diabetes and autoimmune thyroiditis. In type 1 diabetes, a primary role has been assigned to class II genes, both in humans and in the nonobese diabetic (NOD) mouse model. However, an involvement of other tightly linked genes is strongly suspected. Here, through two independent sets of experiments, we provide solid evidence for the existence of at least one such gene. First, using a new recombinant congenic NOD strain, R114, we definitively individualized the *Idd16* locus from the MHC in a 6-cM interval proximal to *H2-K*. It affords almost complete protection against diabetes and is associated with delayed insulitis. Second, by genome scan, we mapped non-H2 genes associated with the highly penetrant form of chronic experimental autoimmune thyroiditis (EAT) that is elicited in NOD and NOD.H2^k mice by immunization with thyroglobulin. We identified one major dominant locus, *Ceat1*, on chromosome 17, overlapping with *Idd16*. Most importantly, R114 recombinant congenic mice challenged with thyroglobulin did not develop chronic EAT. This new major region defined by both *Idd16* and *Ceat1* might thus concur to the unique strength of the MHC in autoimmune susceptibility of NOD mice. *Diabetes* 51:2141–2147, 2002

Gene products from the major histocompatibility complex (MHC) play a major role in immunity and in establishment and maintenance of self-tolerance. They are associated with predisposition to most autoimmune diseases, both in humans and in experimental models (1,2). Among MHC genes, a primary role has been assigned to class I and class II genes. Their key function in antigen presentation to the T-cell receptor and their high level of polymorphism make them obvious candidates for MHC-linked predisposition to autoimmunity (3,4). In autoimmune type 1 diabetes, a disease that displays among the strongest MHC associations, specific allelic forms of class II gene products, DR and DQ, were involved in disease susceptibility (5,6). Likewise, in the mouse model of type 1 diabetes, the nonobese diabetic (NOD) strain, the unique IA molecule harbored by the NOD (H2^{g7}) haplotype was considered to encode diabetes susceptibility (7–10).

Recent data, however, suggest that class II genes do not fully explain MHC-linked predisposition to type 1 diabetes. In humans, studies based on linkage disequilibrium have pointed to an independent association of non–class II MHC genes (11–13). In NOD mice, two series of studies of H2 congenic NOD mice have also indicated a role for non–class II genes. In the first series, congenic NOD.CTS-H2 mice, which harbor an ancestrally related MHC region derived from the CTS strain, a sister strain of NOD, were protected against diabetes (14). The region lying between the *H2-K* and the *Hsp70* genes was identical between NOD and CTS (15). The protective locus, designated *Idd16*, thus mapped either proximal to *H2-K* or distal to *Hsp70*.

The second study relied on a diabetes-resistant recombinant congenic line that harbored a segment of chromosome 17 derived from the B10.A(R209) strain (16). This segment was centromeric to class II genes, the boundary mapping at a hotspot of recombination between *H2-K* and *Lmp2*. Collectively, these studies therefore pointed to a role of H2-linked genes distinct from class I and II genes in predisposition to diabetes. However, a definitive proof was still missing.

In addition to autoimmune diabetes, NOD mice spontaneously develop a mild thyroiditis with low penetrance (17). Immunization with thyroglobulin in complete Freund's adjuvant, however, results in severe and long-lasting inflammatory infiltration of the thyroid gland in almost every NOD mouse (18), contrasting with the transient inflammation observed in CBA/J, C3H, and B10.BR, which carry the predisposing H2^k haplotype and are reference models for experimental autoimmune thyroiditis (EAT) (19,20). Congenic NOD.H2^k mice are as sensitive as NOD mice to induction of chronic EAT (18). This strongly suggests that the passage of thyroid gland infiltration to

From [1]INSERM U25, Hôpital Necker-Enfants malades, Paris, France; and [2]Université René Descartes Paris V and Service d'Anatomie Pathologique, Hôpital Européen Georges Pompidou, Paris, France.

Address correspondence and reprint requests to Henri-Jean Garchon, INSERM U25, Hôpital Necker, 161 rue de Sèvres, 75743 Paris Cedex 15, France. E-mail: garchon@necker.fr.

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O.B. and D.D. contributed equally to this work.

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EAT, experimental autoimmune thyroiditis; MGD, Mouse Genome Database; MHC, major histocompatibility complex.

TABLE 1
Genetic characterization of the congenic strains

Marker	Gene	Genetic distance*	MGD distance†	NOD.H2^k	NOD.H2^b (R0)	R114	R2
D17Mit19		5.7 (19/334)	3.0	NOD	NOD	NOD	NOD
D17Mit164		7.8 (26/334)	4.1	NOD	NOD	NOD	NOD
D17Mit113		0 (0/700)	6.5	NOD	B6	B6	NOD
D17Mit114		1.3 (14/1090)	11.0	NOD	B6	B6	NOD
D17Mit260		2.3 (25/1090)	10.0	NOD	B6	B6	NOD
D17Mit100		1.75 (19/1090)	11.75	B10.BR	B6	B6	NOD
D17Mit248		0.18 (2/1090)	16.0	B10.BR	B6	B6	NOD
D17Mit81		0.09 (1/1090)	16.9	B10.BR	B6	B6	NOD
D17Mit199		0.73 (8/1090)	16.9	B10.BR	B6	B6	NOD
D17Mit16		1.3 (14/1090)	18.15	B10.BR	B6	NOD	B6
D17Mit28	*H2-K*	0.46 (5/1090)	18.44	B10.BR	B6	NOD	B6
	H2-lA	—‡	18.6	—	—	—	—
D17Nds2	*Hsp70-1*	1.55 (17/1090)	18.94	B10.BR	B6	NOD	B6
	H2-D	—	19.1	—	—	—	—
D17Mit105		1.3 (14/1090)	21.95	B10.BR	B6	NOD	B6
D17Mit36		3.3 (36/1090)	24.5	B10.BR	B6	NOD	B6
D17Mit6		1.0 (11/1090)	31.0	NOD	NOD	NOD	NOD
D17Mit7		6.4 (93/1456)	32.3	NOD	NOD	NOD	NOD
D17Mit152			37.7	NOD	NOD	NOD	NOD

*Recombination fraction (%) determined as the ratio of recombinants per meiosis in our progeny mice (given in parentheses); †Distance from the centromere (in cM) according to the MGD. ‡Not determined. Shaded data helps to visualize the segment of donor genome that has been backcrossed in NOD congenic mice.

chronicity in NOD and NOD.H2^k mice is controlled by genes located outside the H2 complex.

Two independent lines of research were undertaken, one aimed at mapping *Idd16* with new congenic recombinant strains, the other at identifying by genome scan non-MHC loci controlling EAT chronicity. The present report describes how these two lines have unexpectedly converged on the identification of a unique overlap interval closely linked to but distinct from the MHC.

RESEARCH DESIGN AND METHODS

Mice and crosses. Inbred mice (NOD, CBA/J, and C57BL/6) were housed in our animal facility under specific pathogen-free conditions and in keeping with the European Union legislation on animal care. NOD.H2^k congenic mice were previously described (18). They were intercrossed with CBA/J mice to produce F1 and F2 generations.

NOD mice congenic for the portion of chromosome 17 derived from the C57BL/6 (B6) strain surrounding the H2 complex were bred by iterative backcrossing with NOD parents and by genotypic selection of heterozygous mice at each generation. The R0 homozygous line was established by appropriate sister-brother matings at the 17th generation of backcrossing. Recombinant congenic lines were derived from the R0 line by backcrossing R0 heterozygous mice with NOD parents. The *H2-K* gene of R114 mice was of the NOD type, as indicated by typing the *D17Mit28* microsatellite, which is located in the promoter of *H2-K*. The *H2-K* gene product was also verified by immunofluorescence labeling with monoclonal antibodies specific for H2-K^d (K9.18) or for H2-K^b (5F1). Both antibodies were generous gifts from Dr. François Lemmonier (Institut Pasteur, Paris).

Assessment of phenotypes, diabetes, and thyroiditis. Development of spontaneous diabetes in females was followed weekly by testing urinary levels of glucose with Glukotest (Roche Diagnostics, Basel). Mice were classified as diabetic after producing two consecutive tests ≥3+. Incidences of diabetes were compared with the log-rank test.

Chronic EAT was induced in 6- to 8-week-old mice of both sexes as described previously (21). Briefly, porcine thyroglobulin (100 μg, purchased from Sigma, St. Louis, MO) was emulsified in 100 μl of complete Freund's adjuvant (Difco, Detroit, MI) and injected subcutaneously at four different points. Mice were boosted 2 weeks later with the same dose of antigen in incomplete Freund's adjuvant. Then, 10 weeks later, mice were killed and thyroid glands were removed, fixed in formaldehyde (4%), and embedded in paraffin. Three serial sections at four noncontiguous levels were stained with hematoxylin and eosin and examined for mononuclear cell infiltration. As described in the RESULTS section, two types of infiltrates, including clusters and foci, were observed. For each type of infiltrate, counts were summed over the four levels. Immunization of mice and scoring of thyroiditis were carried out before, and therefore blindly relative to, genotyping.

Genetic polymorphisms and maps. Microsatellite markers were drawn from the Massachusetts Institute of Technology (MIT) database (22) (found at www-genome.mit.wi.edu). Some of the markers used for the genome screen of the (NOD.H2^{k}xCBA/J)F2 cross had to be tested for polymorphism between NOD and CBA/J in addition to those previously described (23). The PCR was carried out following standard conditions as described (24). PCR products were analyzed by electrophoresis on 5–6% agarose gels. At each marker, the NOD and CBA/J alleles were designated as N and C, respectively. Orders and distances were those of the Mouse Genome Database (MGD) (25) (found at www.informatics.jax.org). Previously unordered markers in the *Idd16* region were ordered by typing recombinant haplotypes and by minimization of the number of recombination events. We found two major differences with the MGD map. First, we observed no recombination between *D17Mit113* and *D17Mit114*. These markers are 4.5 cM distant in MGD. Second, we mapped *D17Mit260* distal to *D17Mit114*, unlike MGD.

The list of microsatellites markers that were genotyped in (NOD.H2^{k}x CBA/J)F2 mice are listed in the online appendix, available at http://diabetes-.diabetesjournals.org.

Analysis of genetic data. Initial examination of the data set indicated that thyroiditis phenotypes, including foci and clusters, did not follow a normal distribution, even after logarithmic or Arcsine transformation. Data were therefore analyzed with nonparametric methods, using the "Nonparametrics and distributions" module of the Statistica package (Statsoft, Tulsa, OK). Correlation between clusters and foci was tested with the Spearman-ρ and Kendall-τ coefficients. Because the two types of infiltrate were partially correlated, they were analyzed separately and together.

Qualitative phenotypes were considered first. For clusters, mice with at least one cluster were considered as affected. Likewise, for foci, mice with at least one focus were considered as affected. The homogeneity of frequencies of affected and nonaffected mice of each genotype, NN, NC, or CC, was tested with 3×2 tables. Exact probabilities of the Pearson χ^2 distribution were calculated with the StatXact software (Cytel Software, Cambridge, MA).

A semi-quantitative analysis was also carried out on clusters, foci, and a global score using the Kruskal-Wallis test. The global score was computed by summing the numbers of foci and clusters after assigning an equivalence of five clusters to one focus. Analyses made no assumption on the mode of inheritance of loci. In Table 3, genetic models, including dominance or recessivity, were tested with the Mann-Whitney *U* test after pooling genotypic groups appropriately. Only *P* values <5% were reported. They were not corrected for the number of tests done. The threshold *P* value for calling linkage in a genome-wide scan of an F2 population was 5.2×10^{-5}, as previously recommended (26).

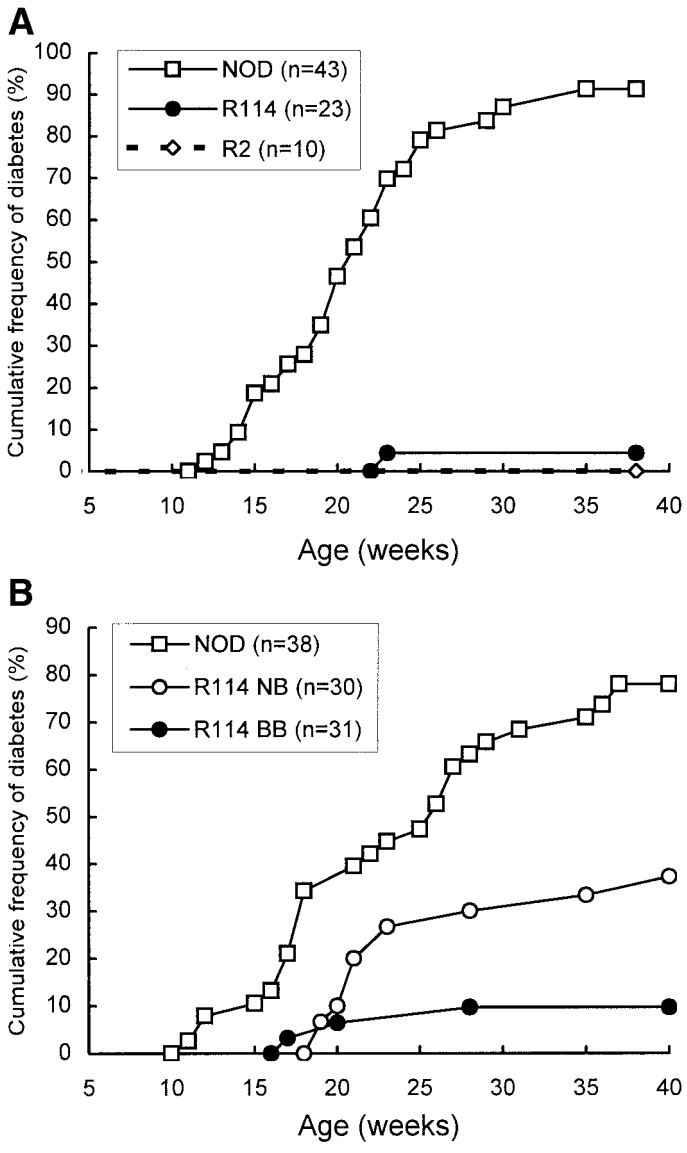

FIG. 1. Cumulative frequency of spontaneous diabetes in recombinant congenic mice. *A*: Recombinant congenic mice R114 and R2 were compared with NOD control mice ($P < 1 \times 10^{-5}$ for R114 and R2 vs. NOD with the log-rank test). *B*: Mice homozygous and heterozygous for the R114 interval were also compared with NOD control mice ($P < 1 \times 10^{-5}$ for R114 BB vs. NOD, $P = 6 \times 10^{-4}$ for R114 NB vs. NOD, and $P = 0.016$ for R114 NB vs. BB).

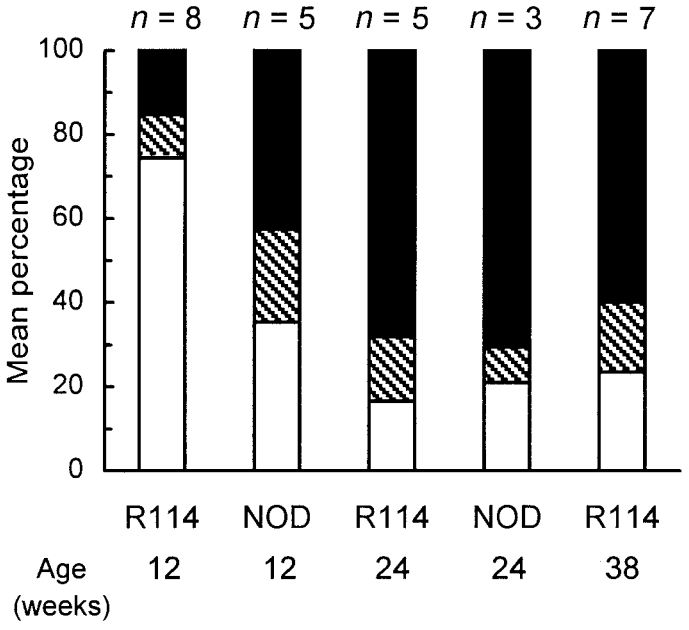

FIG. 2. Delayed insulitis in R114 mice. Infiltration of Langerhans islets was assessed at the indicated time points in nondiabetic females. Islets were classified using three levels of infiltration: none (white bar), peri-insulitis (hatched bar), and insulitis (black bar). The vertical axis shows the mean percentage of each class of islets in each group of mice.

RESULTS

Mapping of *Idd16* distinct from and proximal to H2. An NOD.H2^b congenic strain, here also designated as R0, was bred by iterative backcrossing. It carries a 27-cM segment derived from the C57BL/6 (B6) strain centered on the H2^b complex, whose approximate size is 2 cM (Table 1). At the 17th generation of backcross, two recombinant congenic lines were selected. The R2 line retains the entire H2 and the distal part of the R0 interval (~14 cM). Its proximal boundary is located between the *D17Mit199* and *D17Mit16* markers (Table 1). The latter marker maps 0.3 cM proximal to *H2-K*, the most centromeric gene of the H2 complex. The R114 line yields a mirror image of R2. The breakpoint is also between *D17Mit199* and *D17Mit16*, but the DNA is of B6 origin in the proximal part of the R0 interval (~14 cM). In this line, therefore, the H2 derives from the NOD (g7) haplotype. As shown in Fig. 1*A*, both lines are equally and strongly protected against diabetes ($P < 1 \times 10^{-5}$ for comparison of either R2 or R114 with NOD by the log-rank test). Such protection was expected for R2 mice because they carry the protective B6 MHC alleles. In addition, it was previously shown that the H2^b haplotype confers nearly dominant protection against diabetes, with almost complete penetrance in the heterozygous state (27,28). As shown in Fig. 1B, the protection afforded by the R114 interval is partially dominant.

Histopathological analysis showed that R114 mice develop an infiltration of Langerhans islets. However, it is milder than that of NOD mice at 12 weeks of age (Fig. 2). Eventually, the majority of islets of R114 mice become infiltrated at 24 and 38 weeks of age, sharply contrasting with the almost complete absence of clinical diabetes. Comparison with 24-week-old NOD mice should be made with the important reservation that only the rare NOD mice not yet diabetic can be analyzed. Therefore, R114 mice differ from previously described NOD.H2^b congenic mice (28) and also from R2 mice (data not shown) in that they develop insulitis. However, this inflammatory infiltration evolves at a slower rate than in NOD mice, unlike what is observed in NOD mice congenic at the *Idd5* or *Idd9* loci (29–31).

Phenotypic dissection of chronic EAT in (NOD.H2^{k}xCBA/J)F2 mice. In an independent line of experiments, we sought to map non-MHC genes associated with susceptibility of NOD mice to chronic EAT. Progeny mice of F1 and F2 intercrosses between NOD.H2^k and CBA/J parents were induced with thyroglobulin plus adjuvant, and histopathology of their thyroid glands was assessed 10 weeks later, at a time when inflammation is no longer visible in conventional H2^k strains.

As previously observed (18), all CBA/J mice had recovered from thyroiditis, whereas 90.3% of NOD.H2^k mice still showed severe lesions (Fig. 3*A*). These lesions consisted

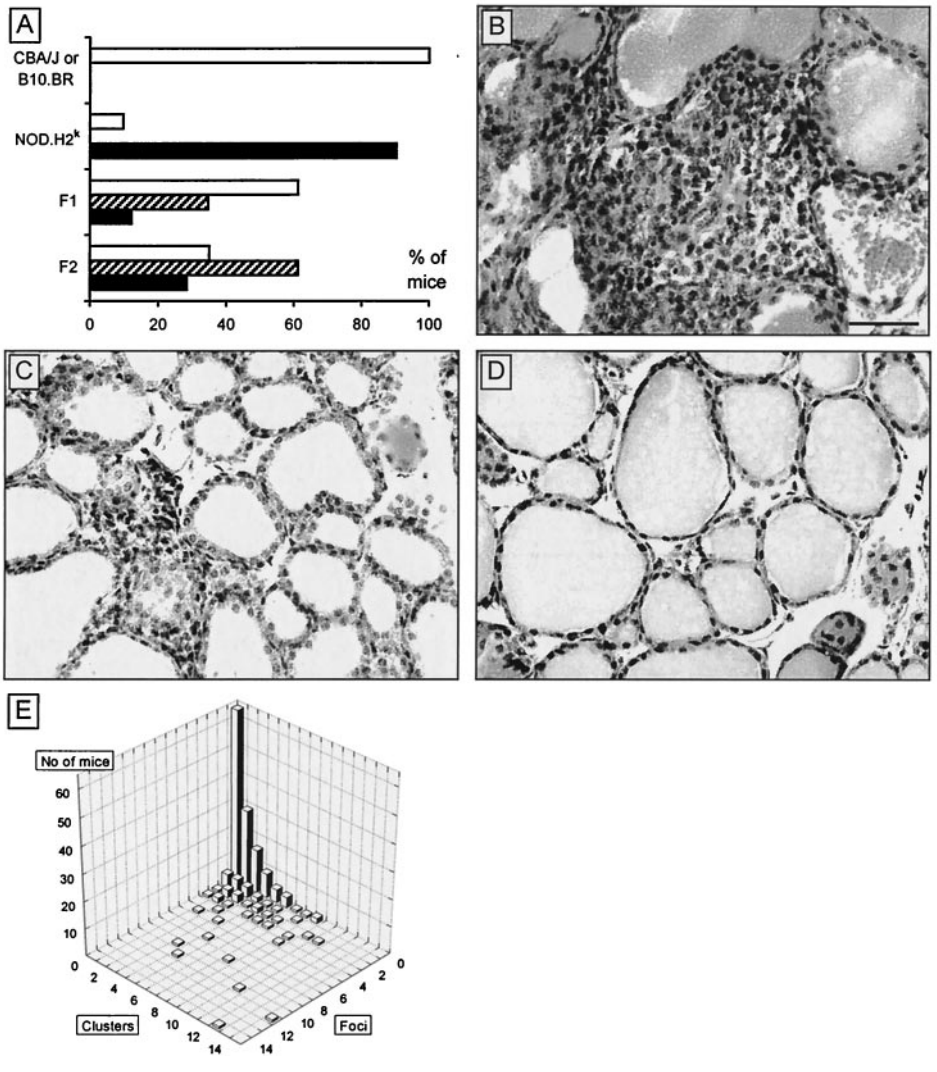

FIG. 3. Phenotypic analysis of (NOD.H2^k xCBA/J)F2 progeny mice. *A*: Percentages of animals with no thyroid infiltration (white bar), with at least one cluster (hatched bar), or with at least one focus (black bar) in NOD (n = 28), CBA/J (n = 31), and B10.BR (n = 13) parent mice and in their F1 (n = 49) and F2 (n = 183) progeny. Representative sections of thyroid glands stained with hematoxylin and eosin showing: a heavy infiltration or focus (*B*), a limited mononuclear cell infiltration or cluster (*C*), and a normal thyroid (*D*) (bar: 100 μm). *E*: Partial correlation between clusters and foci in F2 mice as shown by a 3-D histogram representation of the number of mice (vertical axis) with the numbers of clusters and foci indicated on horizontal axes.

of dense foci containing numerous (>100) mononuclear cells and disrupting the normal architecture of the thyroid gland (Fig. 3*B*). They were associated with partial or complete destruction of the thyroid vesicles. In F1 and F2 mice, the number and severity of the infiltrates were variable. Mild infiltrates were observed in addition to destructive foci. They consisted of clusters of mononuclear cells (<20) found in the interstitium between the thyroid vesicles and not associated with damage of the thyroid epithelium (Fig. 3*C*). These clusters were not seen in CBA/J or B10.BR mice 10 weeks after immunization. Among F1 mice, 34.7% (17 of 49) developed clusters, whereas a smaller proportion (12.2%, 6 of 49) had foci. This suggested a role of one or more partially dominant genes. Among F2 mice, 61.2% (112 of 183) had clusters and 28.4% (52 of 183) had foci. Although there was a significant correlation between the two types of infiltrates in these F2 mice ($r = 0.56$, $P = 5 \times 10^{-14}$), their segregation was also partially independent: 67 of the 112 mice with clusters (59.8%) displayed only clusters, and 7 of the 52 mice with foci (13.5%) had only foci (Fig. 3*E*). This suggested that the mechanisms underlying the formation of clusters and of foci were not necessarily linked. The two types of infiltrates were therefore analyzed both separately and together, using a global score and nonparametric methods.

Genetic analysis of autoimmune thyroiditis in (NOD.H2^{k}xCBA/J)F2 mice. F2 mice (n = 183) were genotyped for 81 microsatellite markers spanning the mouse genome. The frequency of mice affected for each phenotype was determined in each genotypic group. Three loci were detected (Table 2), of which only one, on chromosome 17, yielded a genome-wide significant association, defining the *Ceat1* (for chronic experimental autoimmune thyroiditis 1) locus. Four markers covering a distance of 38 cM on this chromosome influenced both clusters and foci. The strongest association was with the *D17Mit114* marker, which, as seen above, is proximal to the H2 complex. The other two loci influenced either clusters (on chr. 5) or foci (on chr. 7). Distribution of genotype frequencies suggested a dominant mode of action for *Ceat1* and for the putative locus on chromosome 5 and a recessive inheritance for that on chromosome 7. A semi-quantitative analysis was also performed (Table 3) and confirmed the models as fitted just above.

Overlap of *Idd16* and *Ceat1*. Congenic and recombinant congenic NOD mice used for mapping *Idd16* were tested for their responsiveness to induction of chronic thyroiditis. As shown in Table 4, R114 mice were fully protected from chronic EAT. This definitively demonstrated the existence of *Ceat1* and established its tight linkage to *Idd16*.

TABLE 2
Qualitative analysis of *Ceat* loci in 183 (NOD.H2^k×CBA/J)F2 mice

Chr.	Marker (distance*)	Phenotype	Number of mice			*P*‡
			NN†	NC	CC	
5	*D5Mit48* (0)	Total	45	86	50	
		clusters§	33	48	30	
		foci§	16	26	10	
		score§	35	52	31	
	D5Mit72 (9)	Total	48	85	50	
		clusters	36	43	33	0.015
		foci	16	23	13	
		score	38	47	34	0.017
	D5Mit81 (28)	Total	55	85	43	
		clusters	37	50	25	
		foci	21	21	10	
		score	41	52	26	
7	*D7Mit20* (5.5)	Total	49	83	51	
		clusters	32	53	27	
		foci	15	31	6	0.005
		score	34	57	28	
	D7Mit55 (15)	Total	52	84	47	
		clusters	36	49	27	
		foci	16	28	8	
		score	38	53	28	
17 (*Ceat1*)	*D17Mit19* (3)	Total	45	88	46	
		clusters	35	58	16	1×10^{-4}
		foci	20	25	4	5×10^{-4}
		score	39	60	17	$<1 \times 10^{-5}$
	D17Mit114 (11)	Total	45	93	45	
		clusters	35	61	16	1×10^{-4}
		foci	20	29	3	2×10^{-4}
		score	38	65	16	$<1 \times 10^{-5}$
	D17Mit7 (32.3)	Total	57	82	44	
		clusters	43	49	20	0.008
		foci	25	18	9	0.008
		score	45	53	21	0.005
	D17Mit152 (37.7)	Total	56	84	43	
		clusters	43	49	20	0.007
		foci	25	20	7	0.003
		score	45	53	21	0.004

*Distance from the centromere (in cM) according to the MGD. †Genotypic notation: NN, NOD homozygous; NC, heterozygous; CC, CBA/J homozygous. ‡For the exact χ^2 test. Only $P < 0.05$ is reported. §Mice with at least one cluster, one focus, or a thyroiditis score ≥1 are considered affected.

DISCUSSION

The MHC extends over several megabases and is one of the most gene-rich regions of the genome (32). A large proportion of MHC genes are expressed in the immune system, but only a fraction of them have a known function. Distinguishing between the contribution of these numer-

TABLE 3
Nonparametric analysis of *Ceat* loci

Chr.	MGD distance*	Marker	Phenotype	Model†		
				Model-free	Recessive	Dominant
5	9	*D5Mit72*	clusters	0.0017	0.0015	
			foci			
			score‡	0.021	0.0086	
7	5.5	*D7Mit20*	clusters			
			foci	0.0077		0.0022
			score	0.017		0.0046
17 (*Ceat1*)	11	*D17Mit114*	clusters	0.0024		7.8×10^{-4}
			foci	3.6×10^{-4}	0.0045	3.7×10^{-4}
			score	5.4×10^{-7}	7.3×10^{-4}	4.4×10^{-7}

*Distance from the centromere (in cM) according to the MGD. †*P* value of the Kruskal-Wallis test. Only $P < 0.05$ is reported. Dominance and recessivity are defined relative to NOD. ‡A global score was computed by summing the numbers of foci and clusters after assigning an equivalence of 5 clusters to 1 focus, as indicated in RESEARCH DESIGN AND METHODS.

TABLE 4
Protection of R114 congenic mice against induction of chronic EAT

Mouse strain	No. affected/total no.
NOD	8/9
R114	0/8
R0	0/32

ous candidate genes will be a considerable endeavor, especially in humans. In the mouse, breeding of congenic strains and derivation of recombinant congenics still represent the most valid approach available to a precise understanding of MHC gene contribution to complex phenotypes and diseases (33).

Based on the analysis of the R114 recombinant congenic strain, our work brings definitive evidence for the existence of at least one MHC-linked locus other than class I and class II genes in diabetes predisposition. Affording almost complete protection, this locus is one of the strongest non–H2 *Idd* loci. If B6 and CTS strains share the same alleles proximal to *H2-K*, then it is very likely that the *Idd16* MHC-linked resistance allele defined by CTS outcross (14) is also present in the B6 genome and has been positioned in our study. Of note, our present data do not rule out the existence of other loci, either proximal to *H2-K* (16) or adjacent or distal to *H2-D* (14,15). *Idd16* therefore maps proximally to the H2 complex in a region whose distal boundary is limited by the *D17Mit16* marker, and it is at least 0.3 cM proximal to the *H2-K* gene. The physical distance that separates the R114 interval from the H2 is not known precisely. Using available information from the ongoing Mouse Genome Sequencing program (found at www.ncbi.nlm.nih.gov/genome/seq/MmHome.html), it appears that a segment of 250 kb upstream of the *H2-K* gene has been sequenced and that it does not include the *D17Mit16* marker. The *Idd16* locus is therefore unambiguously distinct from the proper H2 complex.

Rather unexpectedly, whereas our genome scan was designed to identify MHC-unlinked genes for chronic EAT by crossing parent mice having a common MHC haplotype, the only genome-wide significant association was observed with the H2-linked *Ceat1* locus. There was milder evidence for two other loci, which are nevertheless worthy of consideration because they influence clusters and foci differentially, suggesting that the two types of infiltrates might arise from different molecular mechanisms.

Importantly, the existence of the *Ceat1* locus was demonstrated by the study of congenic mice. Because B10.BR mice do not develop chronic EAT (see Fig. 3*A*), *Ceat1* can be mapped to the portion of NOD.H2^k chromosome of NOD origin, that is proximal to the crossover point between NOD and B10.BR and that was mapped between *D17Mit260* and *D17Mit100*. *Ceat1* is therefore currently included in a ~8-cM interval extending from *D17Mit114* to *D17Mit100*, the latter marker being excluded.

Tight linkage of *Ceat1* and *Idd16* is consistent with the occurrence of both types of autoimmune manifestations in NOD mice and provides a genetic basis for their association. In humans also, the association of autoimmune thyroiditis and type 1 diabetes is commonly observed (34). Whether *Ceat1* and *Idd16* represent two manifestations of a single gene is now open to investigation, which notably will require the breeding of new mouse congenic lines. However, both loci influence long-term evolution and pathogenicity of inflammatory infiltrates. The effect of the NOD haplotype at the *Idd16* locus is to amplify a preexisting infiltration and to promote its development into an islet-aggressive infiltrate. In this regard, the *Ceat1* locus seems to have a similar effect on thyroidal inflammatory infiltrate, as it requires a preliminary step that is provided by immunization with thyroglobulin. Chronic EAT in NOD mice has been associated with decreased T-cell–mediated response to thyroglobulin, production of γ-interferon, and switch of antibodies to a Th1-dependent isotype (18). How *Ceat1* affects these cellular and molecular parameters will be important to determine and may provide useful clues for its molecular identification. The study of *Idd16*/*Ceat1* may also be relevant to a better understanding of the behavior of inflammatory cell infiltrates in other immunopathological situations, notably viral and parasitic infections, graft rejection, and tumor-host interaction.

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