Genetic Similarities Between Latent Autoimmune Diabetes and Type 1 and Type 2 Diabetes

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In this issue of *Diabetes*, Cervin et al. (1) investigated whether patients with latent autoimmune diabetes in adults (LADA) (defined as age at diabetes onset >35 years, GADA positive), analyzing known risk alleles for type 1 and type 2 diabetes, share genetic polymorphisms with type 1 diabetes (age at onset <35 years) and/or type 2 diabetes (age at onset >35 years, GADA negative). In their LADA patients, they confirmed an increased frequency of type 1 diabetes–associated genetic risk factors including the heterozygous HLA-DQB1*0201/*0302 genotype, insulin AA genotype (rs689), and increased CT and TT genotypes of the *PTPN22* gene (rs2476601) (2). Somewhat surprisingly, in LADA patients they also found a higher frequency of the type 2 diabetes–associated CT and TT genotypes of the TCF7L2 gene (rs7903146) (3), therefore concluding that LADA shares genetic features with both type 1 and type 2 diabetes.

The article by Cervin et al. (1) uses the term LADA and immediately abandons the standard meaning of the term, instead equating LADA with GAD-positive patients who developed diabetes after age 35 years. LADA has been defined as the presence of GAD antibodies in patients with age of onset of diabetes after 35 years and insulin independence for at least 6 months after diagnosis (4–6). We agree with the authors in abandoning the standard definition of LADA and would agree with a number of other reviews that suggest one should abandon the term LADA altogether (7,8). A simpler alternative would be islet autoantibody–positive diabetes, which would likely come close to equating with type 1A diabetes, depending upon the specificity of the assays used. Unfortunately, in the current article (1), specificities of the GAD assay varied over time in DASP (Diabetes Autoimmunity Standardization Program) workshops, being as low as 87% (13% false positives), and given Bayes' theorem and simple calculations, the major conclusions of the study may be in doubt with such a false-positive rate (Fig. 1). We would suggest the hypothesis that in the current article the group termed LADA represents an unknown mixture of type 1A and type 2 diabetes. This might be consistent with a marked increase in high-risk HLA genotype DQB1*0201/DQB1*0302, decreased DQB1*0602, increased PTPN22 and insulin risk polymorphisms (provided by patients with type 1A diabetes), and the type 2 diabetes–associated TCF7L2 polymorphism, with a modest increase ($P = 0.03$) in patients termed GAD positive, but many of whom we believe might be negative with a more consistently specific GAD assay. This alternative hypothesis is difficult to test without improved and more definitive assays for both type 1A and type 2 diabetes.

The above comment raises a number of important issues. *1*) Can we accurately diagnose type 1A diabetes, and how good are the diagnostic criteria short of pancreatic histology? *2*) Can we distinguish LADA from type 1A diabetes occurring in adults or type 2 diabetes? *3*) Are the distinctions important and what do they tell us about the pathogenesis of type 1A diabetes?

There are now assays for autoantibodies reacting with four major islet autoantigens, confirmed in DASP workshops. Multiple laboratories have assays with high sensitivities and specificities often exceeding 98% for autoantibodies reacting with GAD65, IA-2, insulin, and the newest autoantigen Znt8 (9). In that type 1A diabetes occurs in less than 1% of most populations and multiple autoantibodies are often measured, assays with high specificity are essential for both individual diagnosis and epidemiologic studies. Even a specificity of 99% for a single autoantibody assay is insufficient for the prediction of high diabetes risk. Having persistent autoantibodies reacting to at least two of the four major autoantibodies, however, defines groups with high risk (10,11). GAD65 autoantibodies remain the most sensitive marker for adult-onset "autoimmune" diabetes. Though having diabetes by Bayes' theorem increases the probability of a true GAD-positive result, we would suggest it is insufficient with a false-positive rate approaching 10%. Figure 1 illustrates hypothetical overall HLA DR3/4 and TCF7L2 genotype frequencies in GAD-positive versus GAD-negative diabetic subjects assuming a 10% false-positive rate for GAD and a mixture of patients with type 1A and type 2 diabetes. With this hypothetical mixture of GAD-positive type 1A and type 2 diabetes, HLA DR3/4 frequency is still increased to 19.8%, similar to the 27% frequency reported for LADA in Cervin's article (1) (driven by type 1A diabetes). With a false-positive rate approaching 10%, GAD positivity will not exclude type 2 diabetes.

The lack of prediction of diabetes with presence of a single autoantibody is consistent with analysis of pancreatic histology of cadaveric donors. It is likely that expression of a single islet autoantibody will rarely be associated with insulitis, while expression of multiple biochemical autoantibodies will be (12,13). A much larger number of pancreases need evaluation, and the Juvenile Diabetes Research Foundation has initiated a program (nPOD: Network for Pancreatic Organ Donors with Diabetes) headed by Mark Atkinson. A Web site is available for "real-time" viewing of nPOD pancreatic histology (www.jdrfnpod.org). Progressive loss of C-peptide is the outcome of such β-cell loss and was observed in studies of

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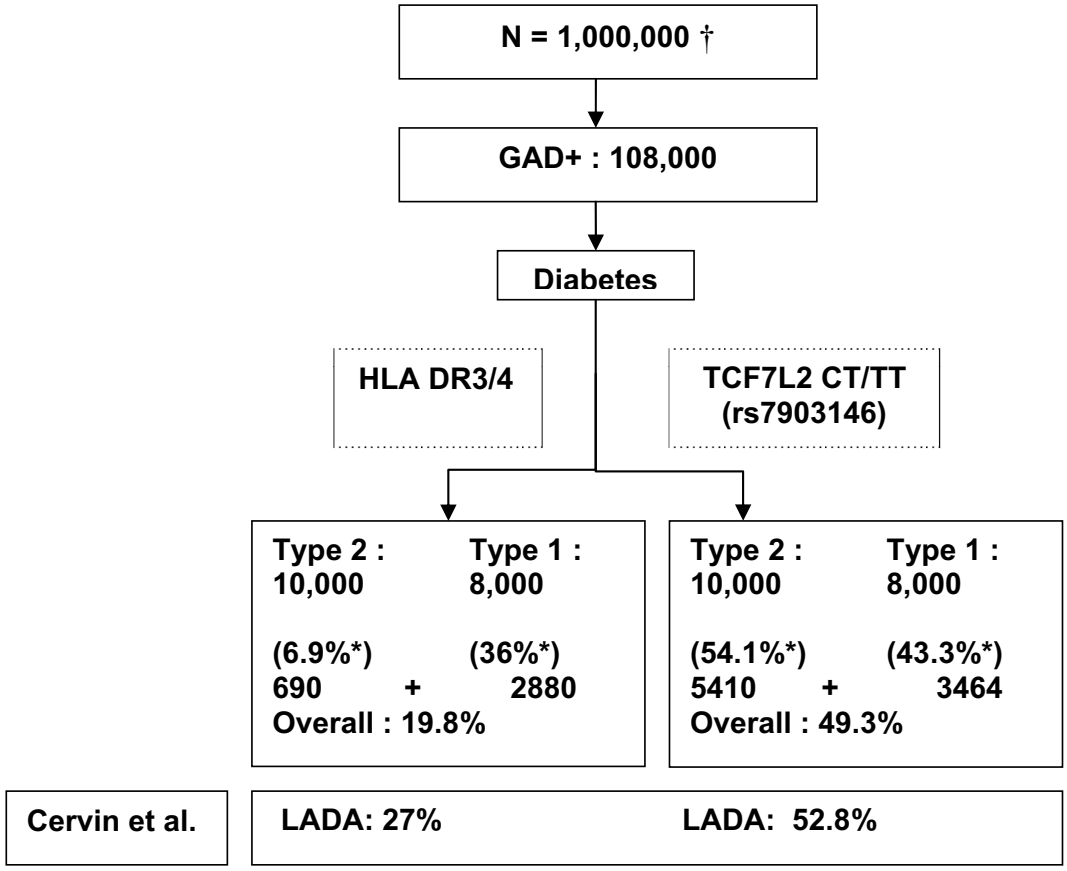

FIG. 1. Bayes' theorem prediction of genetic results with 10% false-positive rate for GAD autoantibody determination and mixture of type 1A and type 2 diabetes. *HLA DR3/4 and rs7903146 frequencies from Cervin et al. (1) for type 1A and type 2 diabetes. †Assumptions for Fig. 1 are the following: 10% false-positive rate for GAD, 80% GAD-positive rate in type 1A diabetes, and frequency of 1 and 10% for type 1A and type 2 diabetes, respectively.

islet autoantibody–positive patients with type 2 diabetes (LADA) (14,15).

Given the above information, can we distinguish type 1A diabetes developing in adults from LADA? An immediate difficulty arises from the observation that type 1A diabetes is not a monomorphic disorder, even within pairs of monozygotic twins. As much as 33 and 37 years can elapse between the appearance of anti-islet autoantibodies and the diagnosis of type 1A diabetes, respectively, in one twin and their twin mate (M. Redondo, unpublished data). There are characteristic immune and genetic correlations with age of onset of type 1A diabetes, including lack of expression of insulin autoantibodies in adolescents and adults compared with young children and decreased percentages of the highest risk HLA genotypes. Publications on LADA analyzing the above parameters are usually consistent with older age of onset of type 1A diabetes and as indicated would not be a firm basis for distinction (16,17).

There clearly are multiple different and crucially important varieties of both type 1 and type 2 diabetes. Neonatal permanent diabetes perhaps provides the clearest examples of clinical relevance in terms of accurate diagnosis. This includes mutations of the FOXP3 gene, which controls development of regulatory T-cells and leads to diabetes and overwhelming autoimmunity (IPEX syndrome) (18). Patients with mutations of the sulfonylurea receptor respond to oral medication for both diabetes and neurologic disorders (19). Thus, distinguishing different forms of diabetes will be important, and within late-onset autoantibody-positive diabetes, knowledge of whether genes predisposing to type 2 diabetes contribute to pathogenesis is an important question. To date, the spectrum of multiple polymorphisms associated with type 1 diabetes appear to all relate to immune function (20). Abandoning the classic definition of LADA is probably wise and certainly expedient, but then immunogenetic and autoimmune measures with high specificity will be needed to exclude the more common type 2 diabetes in epidemiologic studies. Although type 2 diabetes is much more common than type 1A diabetes in adults, more than 40% of type 1A diabetes develops after age 30 years (21). In that monozygotic twins can differ by decades in onset, defining the determinants of age of onset of type 1A diabetes will be a difficult task. It is likely that some of the adolescents followed from birth who are now anti-islet autoantibody positive will not develop diabetes until they are adults and even adults over age 35 years.

REFERENCES

1. Cervin C, Lyssenko V, Bakhtadze E, Lindholm E, Nilsson P, Tuomi T, Cilio CM, Groop L: Genetic similarities between latent autoimmune diabetes in adults, type 1 diabetes, and type 2 diabetes. *Diabetes* 57:1433–1437, 2008
2. Petrone A, Suraci C, Capizzi M, Giaccari A, Bosi E, Tiberti C, Cossu E, Pozzilli P, Falorni A, Buzzetti R. The protein tyrosine phosphatase nonreceptor 22 (*PTPN22*) is associated with high GAD antibody titre in latent autoimmune diabetes in adults (NIRAD study 3). *Diabetes Care* 31:534–538, 2007
3. Field SF, Howson JM, Smyth DJ, Walker NM, Dunger DB, Todd JA: Analysis of the type 2 diabetes gene, TCF7L2, in 13,795 type 1 diabetes cases and control subjects. *Diabetologia* 50:212–213, 2007

4. Fourlanos S, Dotta F, Greenbaum CJ, Palmer JP, Rolandsson O, Colman PG, Harrison LC: Latent autoimmune diabetes in adults (LADA) should be less latent. *Diabetologia* 48:2206–2212, 2005
5. Tuomi T, Carlsson A, Li H, Isomaa B, Miettinen A, Nilsson A, Nissen M, Ehrnstrom BO, Forsen B, Snickars B, Lahti K, Forsblom C, Saloranta C, Taskinen MR, Groop LC: Clinical and genetic characteristics of type 2 diabetes with and without GAD antibodies. *Diabetes* 48:150–157, 1999
6. Tuomi T, Groop LC, Zimmet PZ, Rowley MJ, Knowles W, Mackay IR: Antibodies to glutamic acid decarboxylase reveal latent autoimmune diabetes mellitus in adults with a non–insulin-dependent onset of disease. *Diabetes* 42:359–362, 1993
7. Gale EA: Latent autoimmune diabetes in adults: a guide for the perplexed. *Diabetologia* 48:2195–2199, 2005
8. Stenstrom G, Gottsater A, Bakhtadze E, Berger B, Sundkvist G: Latent autoimmune diabetes in adults: definition, prevalence, β-cell function, and treatment. *Diabetes* 54 (Suppl. 2):S68–S72, 2005
9. Wenzlau JM, Juhl K, Yu L, Moua O, Sarkar SA, Gottlieb P, Rewers M, Eisenbarth GS, Jensen J, Davidson HW, Hutton JC: The cation efflux transporter ZnT8 (Slc30A8) is a major autoantigen in human type 1 diabetes. *Proc Natl Acad Sci USA* 104:17040–17045, 2007
10. Verge CF, Gianani R, Kawasaki E, Yu L, Pietropaolo M, Jackson RA, Chase HP, Eisenbarth GS: Prediction of type 1 diabetes in first-degree relatives using a combination of insulin, GAD, and ICA512bdc/IA-2 autoantibodies. *Diabetes* 45:926–933, 1996
11. Barker JM, Barriga KJ, Yu L, Miao D, Erlich HA, Norris JM, Eisenbarth GS, Rewers M: Prediction of autoantibody positivity and progression to type 1 diabetes: Diabetes Autoimmunity Study in the Young (DAISY). *J Clin Endocrinol Metab* 89:3896–3902, 2004
12. Gianani R, Putnam A, Still T, Yu L, Miao D, Gill RG, Beilke J, Supon P, Valentine A, Iveson A, Dunn S, Eisenbarth GS, Hutton J, Gottlieb P, Wiseman A: Initial results of screening of nondiabetic organ donors for expression of islet autoantibodies. *J Clin Endocrinol Metab* 91:1855–1861, 2006
13. Oling V, Marttila J, Knip M, Simell O, Ilonen J: Circulating CD4+CD25 high regulatory T cells and natural killer T cells in children with newly diagnosed type 1 diabetes or with diabetes-associated autoantibodies. *Ann N Y Acad Sci* 1107:363–372, 2007
14. Turner R, Stratton I, Horton V, Manley S, Zimmet P, Mackay IR, Shattock M, Bottazzo GF, Holman R: UKPDS 25: autoantibodies to islet-cell cytoplasm and glutamic acid decarboxylase for prediction of insulin requirement in type 2 diabetes: UK Prospective Diabetes Study Group. *Lancet* 350:1288–1293, 1997
15. Borg H, Gottsater A, Fernlund P, Sundkvist G: A 12-year prospective study of the relationship between islet antibodies and β-cell function at and after the diagnosis in patients with adult-onset diabetes. *Diabetes* 51:1754–1762, 2002
16. Hosszufalusi N, Vatay A, Rajczy K, Prohaszka Z, Pozsonyi E, Horvath L, Grosz A, Gero L, Madacsy L, Romics L, Karadi I, Fust G, Panczel P: Similar genetic features and different islet cell autoantibody pattern of latent autoimmune diabetes in adults (LADA) compared with adult-onset type 1 diabetes with rapid progression. *Diabetes Care* 26:452–457, 2003
17. Desai M, Zeggini E, Horton VA, Owen KR, Hattersley AT, Levy JC, Walker M, Gillespie KM, Bingley PJ, Hitman GA, Holman RR, McCarthy MI, Clark A: An association analysis of the HLA gene region in latent autoimmune diabetes in adults. *Diabetologia* 50:68–73, 2007
18. Wildin RS, Smyk-Pearson S, Filipovich AH: Clinical and molecular features of the immunodysregulation, polyendocrinopathy, enteropathy, X linked (IPEX) syndrome. *J Med Genet* 39:537–545, 2002
19. Sagen JV, Raeder H, Hathout E, Shehadeh N, Gudmundsson K, Baevre H, Abuelo D, Phornphutkul C, Molnes J, Bell GI, Gloyn AL, Hattersley AT, Molven A, Sovik O, Njolstad PR: Permanent neonatal diabetes due to mutations in KCNJ11 encoding Kir6.2: patient characteristics and initial response to sulfonylurea therapy. *Diabetes* 53:2713–2718, 2004
20. Todd JA, Walker NM, Cooper JD, Smyth DJ, Downes K, Plagnol V, Bailey R, Nejentsev S, Field SF, Payne F, Lowe CE, Szeszko JS, Hafler JP, Zeitels L, Yang JH, Vella A, Nutland S, Stevens HE, Schuilenburg H, Coleman G, Maisuria M, Meadows W, Smink LJ, Healy B, Burren OS, Lam AA, Ovington NR, Allen J, Adlem E, Leung HT, Wallace C, Howson JM, Guja C, Ionescu-Tirgoviste C, Simmonds MJ, Heward JM, Gough SC, Dunger DB, Wicker LS, Clayton DG: Robust associations of four new chromosome regions from genome-wide analyses of type 1 diabetes. *Nat Genet* 39:857–864, 2007
21. Molbak AG, Christau B, Marner B, Borch-Johnsen K, Nerup J: Incidence of insulin-dependent diabetes mellitus in age groups over 30 years in Denmark. *Diabet Med* 11:650–655, 1994