Dead or Alive?

Type 1 diabetes results from an immune assault on β-cells that progresses over time until the remaining β-cells are unable to keep up with demand and the ensuing hyperglycemia leads to clinical diagnosis. Many textbooks state that this occurs when 80% of the islets are destroyed; but in truth, the amount of residual insulin secretion at the time of clinical diagnosis is variable. The standard model is that those with type 1A diabetes continue to undergo immune-mediated β-cell attack after clinical diagnosis, rapidly leading to the complete absence of β-cells (1).

In the last few decades, clinical research data have increasingly been challenging the notion that β-cells are completely destroyed soon after clinical diagnosis. Once individuals receive exogenous insulin, measurement of β-cell function requires assaying C-peptide, which is secreted in equimolar concentrations with insulin from β-cells. Stimulating the β-cell with a standard liquid "mixed meal" allows for assessment of the β-cell's ability to handle daily activities. Controlling for time of day, administration of exogenous insulin, and fasting glucose level, the mixed-meal tolerance test (MMTT) is a highly reproducible and easily performed test (2). We now know that among type 1 diabetic patients enrolled in clinical trials to preserve β-cell function, it is unusual for control or placebo-treated subjects starting with a reasonable amount of C-peptide at diagnosis to completely loose function in the first 2 years (3–13). Outside of these highly controlled clinical trial situations, residual C-peptide soon after diagnosis has been well documented (14–16). The SEARCH for Diabetes in Youth Study of antibody-positive youth with diabetes reported that more than 30% of children within the first year of diagnosis have fasting C-peptide values within the fifth percentile of normal healthy adolescents and that 11% of youth 5 or more years from diagnosis have potentially clinically significant fasting C-peptide levels (17). At the other end of the spectrum, *1*) the Joslin Medalist Study demonstrated that 64% of individuals who had lived with type 1 diabetes for more than 50 years had measureable C-peptide (18), *2*) our data of unselected subjects at least 30 years from diagnosis found detectable levels in 50% of subjects upon initial testing, and *3*) others also found persistence of C-peptide in some individuals with long-standing disease (19). Recent studies using pathologic specimens also note some patchiness to β-cell loss in those who had type 1 diabetes (1). Further, studies in pregnancy have suggested that an increase in β-cell function may occur (20,21). All these data support the concept that some β-cells may survive for a long time and that their function may wax and wane over time. Such data hold out the hope that attenuation of immune destruction could result in resurgence of endogenous islet function even in those with long-standing disease.

Many articles refer to a peak-stimulated C-peptide level of 0.2 pmol/mL as the clinically relevant value. This is due to a post hoc analysis of Diabetes Control and Complications Trial (DCCT) data in which individuals in the intensively treated group who sustained a C-peptide value of at least 0.2 pmol/mL during an MMTT had less hypoglycemia, retinopathy, and proteinuria (22). Since the DCCT excluded individuals whose C-peptide at entry was greater than 0.5 pmol/mL (23), it is not known whether greater levels of C-peptide would have even greater clinical benefit. Other data pointing to the clinical relevance of some endogenous insulin secretion come from islet transplant studies where, despite an inability to sustain glycemic control without exogenous insulin therapy, even limited function of transplanted islets attenuates major hypoglycemic episodes in this population, which is selected for transplant largely due to having hypoglycemic unawareness (24). The threshold value for such clinical relevance is unknown.

The reliability of such reports depends on robust measures of C-peptide. In recent years, the National Institute of Diabetes and Digestive and Kidney Diseases (NIDDK) has sponsored C-peptide assay standardization workshops to assure cross-laboratory reliability of data (25). The assays have been shown to reliably measure C-peptide in plasma at concentrations to a lower level of ~0.03 pmol/mL. The fact that C-peptide is reliably measured in plasma does not, of course, speak to the clinical relevance of the concentrations found.

With this backdrop, in this issue of *Diabetes Care*, Wang et al. (26) report results from individuals with type 1 diabetes using a highly sensitive C-peptide assay. This assay, performed with ELISA kits from Mercodia AB in Sweden, reportedly can reliably measure C-peptide concentrations to a lower detection limit of 1.5 pmol/L (or 0.0015 pmol/mL). This is ~20–40 times more sensitive than the standard assays. Using fasting serum samples from 182 type 1 diabetic patients recruited over a 10-year period, Wang et al. found that ~79% of subjects within 5 years of diagnosis and 10% between 31 and 40 years from diagnosis have detectable C-peptide in the ranges detectable only by the highly sensitive assay with only two subjects with detectable values who have lived with diabetes more than 40 years. As noted above, this is less than were reported in the Medalist Study, which used standard C-peptide measurements. Thus, while this study tested a large and less highly selected group, these data confirm previous studies that suggest that some β-cell secretion occurs long after diagnosis. Validating this highly sensitive assay in a workshop setting will enable other investigators to confirm these findings in defined populations. An interesting question not directly addressed in this article is the reproducibility of the assay in the same individual over time. There was clear variation in the results in the four subjects repeatedly sampled. While the authors attribute this variation to glycemic status, this is a hypothesis that could be tested by formal assessment under standardized conditions. In our own work, though 50% of subjects had detectable C-peptide in standard assays during arginine stimulation, when the same subjects were retested, this was not consistently confirmed. This variation may be a reflection of the waxing and waning of disease or issues with the assays.

As noted above, even with the conventional C-peptide assays, the clinical relevance of detecting low levels of C-peptide (less than 0.2 pmol/L) in plasma of people with type 1 diabetes is unclear. Wang et al. attempted to address the clinical relevance of the extremely low levels detected in their assay by exploring the relationship of C-peptide and glucose values in both the subjects who had multiple sampling

over time and the cohort of 182 type 1 diabetic patients described above. While these are interesting exploratory analyses, correlations of multiple variables in samples not obtained for the purpose of addressing this question should be interpreted with caution. Formal testing of the hypothesis that very low levels of C-peptide are biologically relevant will require a prospective study design controlling for multiple clinical and demographic variables, standardized testing procedures, and with prespecified outcome measures. Even then, biological relevance does not necessarily equate with clinical relevance.

This article thus serves to highlight the increasing consensus of many studies over the past decades that have found that some β-cells may function long after the clinical diagnosis of type 1 diabetes and that endogenous secretion is clinically important. Unresolved are questions about the clinical relevance of C-peptide less than 0.2 pmol/mL, and whether we can harness small amounts of β-cell function to the clinical benefit of patients.

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