A Nonclassical Model of Autoimmune Hypothyroidism

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MONONUCLEAR INFILTRATION OF THE TARGET organ is the defining feature of all cell-mediated autoimmune diseases (1) and is typically found in the thyroid of patients affected by Hashimoto thyroiditis. The infiltration is composed predominantly of T lymphocytes, B lymphocytes, and macrophages and is considered by most researchers to be the cause of follicular destruction and thus hypothyroidism.

T lymphocytes are among the first immune cells to enter the target organ, dictating the formation of ectopic lymphoid follicles within the thyroid (2). T lymphocytes can also transfer thyroiditis to normal recipients (3), leaving no doubt of their pathogenic role. B lymphocytes are mainly known as the producers of antibodies that, in Hashimoto thyroiditis, recognize thyroperoxidase and thyroglobulin. These antibodies cannot transfer disease and as such have a less clear pathogenic significance. Their binding to thyroid cells expressing thyroperoxidase and thyroglobulin can theoretically induce the death of the thyroid cell by another class of lymphocytes, called natural killers, which recognize the tail of the bound antibody and induce an antibody-dependent, cell-mediated cytotoxicity. Beside their antibody-producing activity, B cells are also important regulators of T cells, so much that agents like rituximab, a monoclonal antibody that selectively destroys CD20-positive B cells, are used for the treatment of T cell-mediated autoimmune diseases like rheumatoid arthritis and Graves ophthalmopathy (4). It is important to note that the infiltration of just lymphocytes, both T and B, in the thyroid does not lead to hypothyroidism, as shown by transgenic mice that specifically express the chemokine CCL21 in the thyroid (5). In this regard, macrophages might actually be critical for executing the damage to thyroid follicles and thus inducing hypothyroidism, although their role remains to be explored.

Nonetheless, T lymphocytes are currently considered the *prima donna* of the autoimmune infiltrate. They can destroy the target cell by two main mechanisms, all centered around apoptosis. First, CD4 and CD8 T cells activate the extrinsic pathway of apoptosis by ligation of their Fas ligand to Fas receptors expressed on thyrocytes. Second, CD8 T cells release preformed cytotoxic proteins (such as perforin, granzyme, and granulysin) that rapidly induce apoptosis.

In this classical model of autoimmune hypothyroidism, clinical hypothyroidism is the direct consequence of the loss of thyroid follicles. This loss is induced by immune-mediated

apoptosis and cytotoxic death of thyrocytes (Fig. 1, left panel). Therefore, in the classical model, failure of the thyroid gland follows directly from the *loss of thyroid follicles*. Apoptosis of thyroid cells in humans was first reported by Kotani and colleagues in 1995 (6). The authors analyzed DNA fragmentation in 16 Hashimoto thyroid specimens and found that on average between 65% and 85% of thyrocytes adjacent to the infiltrate showed fragmentation. Numerous groups have confirmed and extended these findings, and thyroid apoptosis was recently reviewed in this journal (7). Nevertheless, the extent of thyrocyte apoptosis varies greatly among thyroiditis patients. In addition, only a few studies have correlated the extent of thyroid lymphocytic infiltration to the severity of hypothyroidism, and those found only a poor correlation (8). It thus remains unclear to what extent apoptosis contributes to the hypothyroidism typical of Hashimoto thyroiditis.

We propose here a new model of autoimmune hypothyroidism. In this nonclassical model (Fig. 1, right panel), clinical hypothyroidism results from a *chronic inhibition of thyroid function*, induced for example by exposure of the thyroid to pro-inflammatory cytokines like interferon-gamma and tumor necrosis factor-alpha. These factors can be present in the thyroid also independently of infiltrating lymphocytes. The novelty of the nonclassical model is that here thyroid atrophy is the consequence of the thyroid hypofunction rather than the cause of it.

The original idea for this model came from our studies in transgenic mice that chronically express interferon-gamma in the thyrocyte (9). These mice develop many features similar to those seen in the thyroid glands of Hashimoto thyroiditis patients: goiter, disrupted thyroid architecture, macrophage infiltration, oncocytic changes of the thyrocytes, and long-lasting hypothyroidism (9,10). The thyroid phenotype is directly dependent upon interferon-gamma because it disappears when mice are crossed to mice lacking STAT1, the key signal transducer of interferon gamma (11). Strikingly, the thyroid phenotype is present even in the absence of infiltrating lymphocytes (obtained by crossing the transgenic mice to recombinaise activating gene [RAG] deficient mice) (9), suggesting that chronic exposure to a cytokine can lead *per se* to hypothyroidism, without the presence of thyroid-infiltrating lymphocytes. More recently, we have shown that the thyroid phenotype of interferon-gamma transgenic mice is dependent on LMP2 (12), a component of the immunoproteasome, and

FIG. 1. The classical and nonclassical models of autoimmune hypothyroidism. IFN, interferon; TNF, tumor necrosis factor.

that LMP2 is expressed in the thyrocytes of patients with autoimmune thyroiditis (12).

The clinical significance of a new model of autoimmune hypothyroidism is immediate. In the classical model (thyrocyte apoptosis $\rightarrow$ loss of thyroid follicles $\rightarrow$ clinical hypothyroidism) the thyroid destruction is irreversible; in the nonclassical model (chronic inhibition of thyroid function $\rightarrow$ thyroid atrophy $\rightarrow$ clinical hypothyroidism) the immune-mediated thyroid hypofunction is potentially reversible and curable.

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