Dynamic Interactions Between the Immune System and the Neuroendocrine System in Health and Disease

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The immune system and the neuroendocrine system share many common features. Both consist of diverse components consisting of receptors and networks that are widely distributed throughout the body, and both sense and react to external stimuli which, on the one hand control mechanisms of immunity, and on the other hand control and regulate growth, development, and metabolism. It is thus not surprising, therefore, that the immune system and the neuroendocrine system communicate extensively. This article will focus on bi-directional immune-endocrine interactions with particular emphasis on the hormones of the hypothalamus-pituitary-thyroid (HPT) axis. New findings will be discussed demonstrating the direct process through which the immune system-derived thyroid stimulating hormone (TSH) controls thyroid hormone synthesis and bone metamorphosis, particularly in the context of a novel splice variant of TSHβ made by peripheral blood leukocytes (PBL). Also presented are the ways whereby the TSHβ splice variant may be a contributing factor in the development and/or perpetuation of autoimmune thyroid disease (AIT), and how systemic infection may elicit immune-endocrine responses. The relationship between non-HPT hormones, in particular adipose hormones, and immunity is discussed.

Keywords: systemic regulation and adaptation, Hashimoto disease, hematopoiesis, thyroid hormones, integrated

INTRODUCTION

In its most elemental form, homeostasis can be viewed as an integrated state of equilibrium between various physical and chemical processes, not only within individual organ systems, but also throughout the body overall. Whereas most biomedical research is conducted from a highly-focused reductionist perspective given the inherent difficulties in attempting to measure and quantify multifaceted processes, there is nonetheless a need to continually reflect on the vast cross-talk of organ systems in the body.

The immune system and the neuroendocrine system both consist of widely-distributed tissues, cells, receptors, ligands, and molecules. Moreover, both systems are highly adapted to sense external signals from the environment, and to communicate information regarding those throughout the body. It is perhaps not surprising, therefore, that the immune system and the neuroendocrine...
system interact broadly at many levels. In fact, the immune system and the neuroendocrine system collectively have been referred to as a “sixth sense” based on shared neuropeptides and neurotransmitters used by the immune system (1). One of many examples of this is the dynamic set of interactions between the immune system and the neuroendocrine system in the gut. In fact, there are at least three mechanisms for detecting changes in the intestinal wall, consisting of neural sensation delivered by extrinsic and intrinsic afferent neurons, more than twenty endocrine hormones produced by the cells of the mucosal epithelium, and immune responses to local and systemic antigens (2). Collectively, these form a web of communication and defense at the level of the gut. However, many other examples of this exist, as will be discussed in the following sections.

That TSH is produced by cells of the immune system was first reported almost forty years ago (3, 4). TSH is also produced by mouse intestinal crypt enterocytes and intestinal leukocytes, particularly in “hotblocks” of experimental rotavirus and reovirus infection (5, 6). Two sets of findings opened the way for understanding a potential role for immune system TSH. First, hypophysectomized mice that are unable to make pituitary TSH had elevated levels of T4 following alloantigen priming similar to that of non-hypophysectomized animals (7). Second, bone marrow (BM) hematopoietic cells and PBL were found to produce a novel splice variant of TSHβ (8), as discussed in detail below.

**BIDIRECTIONAL IMMUNE-ENDOCRINE INTERACTIONS OF THE HPT AXIS**

The HPT axis is a critically-important hormone network for maintaining basal metabolism, growth, development, mood, and cognition. TSH is released into the circulation from the anterior pituitary followings thyrotropin releasing hormone (TRH) stimulation from the hypothalamus. TSH binds to and induces the release of the thyroid hormones (TH) thyroxine (T4) and triiodothyronine (T3) from the thyroid after binding to TSH receptors (TSHR), a seven-transmembrane domain G-protein coupled molecule on thyroid follicular cells. The majority of T4 is converted into the more biologically active T3 form following deiodination in target tissues after binding to thyroid hormone transporters (9, 10). The TSHR is also widely-distributed across many tissues outside the HPT axis (11).

Thyroid hormones have been shown to exert pleiotropic effects on PBL and on the inflammatory response. Early studies demonstrated that thymic peptides such as thymopoietin, thymulin, and thymosin produced by the thymic epithelium can have a positive effects on the secretion of hormones from the adenohypophysis (12). It was demonstrated in a series of studies that the thyroid is extensively involved in the maturation of the thymus (13–15). Conversely, THs have been shown to upregulate thymulin secretion (14). Exposure of T cells to TH has time dependent effects in that short-term exposure results in suppressed proliferation and apoptosis, whereas long-term exposure induces T cell proliferation. This appears to be regulated at least in part by activation of inducible nitric oxide synthetase (iNOS) (16–19). B cells respond differently to THs in that exposure induces development and cell-proliferation in vivo (20). T3 has direct effects on the maturation of macrophages into the M1 and M2 forms (21). T4 also has beneficial effects on the recovery from *Neisseria meningitidis* infection, mediated by iNOS production and nitric oxide mobilization (22). T4 blocks macrophage inhibitory factor proinflammatory activity in vivo and enhances survival of mice with induced sepsis (23, 24). The TSHR is expressed at high levels on a subset of murine dendritic cells (DCs), though it is minimally expressed on T cells and B cells. However, for reasons that are unclear, the TSHR is expressed on more lymph node T cells and B cells than on spleen cells (25). TSH enhances the phagocytic activity of DCs (26). TH have complex effects on the development and function of DCs, macrophages, and monocytes. Studies in which hypothyroid patients were treated with exogenous TH had increases in both plasmacytoid and myeloid DCs (26).

Adipose hormones such as adiponectin and leptin, which regulate metabolism and energy efficiency, also influence immunological function via receptors expressed on immune cells, particularly on M2-differentiated macrophages (27). Adiponectin has direct immunoregulatory activity by inhibiting the secretion of proinflammatory cytokines and increasing immunosuppressive cytokines (28, 29). Mice deficient in adiponectin fail to effectively modulate metabolic homeostasis (30). Leptin increases immune cell development, chemotaxis, and cytokine secretion (31, 32). Moreover, M1 and M2 macrophages in adipose tissues have opposing effects on insulin responses in that M1 macrophages promote insulin resistance whereas M2-macrophages enhance insulin sensitivity (33, 34). Invariant NKT (iNKT) cells and mast cells are present in adipose tissues (35, 36). Both of those are distinguished by their ability to rapidly respond to danger signals and to produce proinflammatory and regulatory cytokines. iNKT cells, in particular, are known to be a significant source of IFN-γ, IL-2, IL-4, IL-13, IL-17, and IL-21, as well as TNFα and GM-CSF, among others (37), all of which have important immunoregulatory activities and functions.

**A NOVEL TSHβ ISOFORM PRODUCED BY THE CENTRAL AND PERIPHERAL IMMUNE SYSTEM**

TSH is one of three glycoprotein hormones made in the anterior pituitary. All glycoprotein hormones share a common α-chain molecule and a unique hormone-specific β-chain component. TSHβ is highly conserved across many mammalian species. Until recently, no functional isoforms of TSHβ had been identified. We characterized a unique in-frame splice variant of TSHβ (referred to as TSHβv), which is copiously made by PBL and BM hematopoietic cells, in particular though not exclusively on myeloid cells (8, 38–40). Notably, TSHβv is stored in intracellular secretory vesicles in macrophages (39), a property that would facilitate rapid release under appropriate conditions. In that context, it will be interesting to define the signals that drive the release of intracellular TSHβv.
TSHβ is coded for by exons 2 and 3 in humans and exons 4 and 5 in mice. The splice variant is unique, however, in that in both species only the second of the two exons is used to code for TSHβv, with a small portion of the upstream intron coding for a signal peptide (Figure 1). Predictions as to the mechanisms of alternative splicing of TSHβ in leukocytes leading to the generation of TSHβv are derived from putative donor and acceptor splice sites in human intron 1 and intron 2, respectively, resulting in the elimination of exon 2 and the retention of an intron 2 associated signal peptide (Figure 2) (41).

TSHβv has been shown to be present in the human circulation (42), and to be functionally active based on cAMP signaling (8, 39) as well as to successfully couple to TSHα (42), a condition considered to be essential to achieve full biological activity (43). Moreover, TSHβv has been shown to induce TH synthesis in vivo and in vitro. T3 and T4 were elevated in the circulation of mice within one hour of injection of recombinant TSHβ, and to induce the secretion of T3 and T4 from thyroid follicular cells in vitro (44). What’s more, levels of thyroglobulin, thyroid peroxidase, and sodium-iodide supporter were elevated in thyroid follicular cells following TSHβv stimulation. Of particular interest, injection of mice with T3 and TRH caused a transient drop followed by an increase in native TSHβ though not in TSHβv in the pituitary (44).

Expression of TSHβv has been linked to the inflammatory response in AIT, in particular in Hashimoto’s thyroiditis (HT), as demonstrated by elevated transcript levels of TSHβv in PBL of

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**Figure 1**
Genetic organization of (A, B) mouse and (C, D) human native TSHβ, and (E) mouse and (F) human TSHβv.

**Figure 2**
Putative splicing mechanism used to generate human TSHβv in cells of the immune system. Donor splice sites in intron 1 and acceptor splice sites in intron 2 remove exon 2. A portion of intron 2 is used for the signal peptide as shown in Figure 1.
patients with HT compared to normal controls (42). Treatment of patients with prednisone reduced TSHβv transcript levels in persons with short duration of disease compared to persons with long duration. Additionally, TSHβv-producing plasma cells infiltrated the thyroid in HT patients (40). Recent studies demonstrate that immune system TSHβv in humans operates independently of the HPT axis and is capable of inducing TH synthesis from PBL in times of immune stress, such as during systemic infection (44). Those possibility conforms to finding in mice showing that TSHβv-producing inflammatory cells traffic to the thyroid following L. monocytogenes infection (38). Moreover, spleen cells from bacteria-infected mice, but not from non-infected mice, trafficked to the thyroid of normal non-infected mice at high density 48 hours post-transfer (Figure 3) (38). The connection between infection and AIT, while interesting, is unclear due in part to a lack of sufficient studies to draw definitive conclusions (45). Taken together, however, these findings suggest that under certain conditions TSHβv may contribute to the pathogenesis of HT and possibly other forms of AIT.

TSH has been shown to directly influence bone remodeling via TSHR expressed on osteoclasts by preventing bone resorption (46) and stimulating osteoblastic bone formation (47). In humans, there is an increased risk of bone fracture in women with low circulating TSH (48). Using Tshr−/− mice, which are incapable of TSH signaling, and WT mice that were induced to a state of hyperthyroidism by implantation of T4 pellets, Tshr−/− mice had significantly greater bone loss (49), further suggesting a role for TSH in bone restructuring. Moreover, expression of TSHβv in BM CD11b+ cells was positively rather than negatively regulated by in vivo T3 supplementation (49). This was further confirmed using human BM-derived macrophages, which had an increase in TSHβv following exposure to T3 in a dose-dependent manner (50). Those findings further indicate that the regulation of TSHβv by TH occurs independently of HPT axis control.

FIGURE 3 | Splenic leukocytes from L. monocytogenes-infected mice but not normal mice traffic to the thyroid. Immunofluorescence analysis of (A) the thyroid and (B) a thyroid perivascular lymph node from a non-infected mouse 24 hours post-cell transfer of CFSE-labeled splenic leukocytes from a L. monocytogenes-infected mouse. (C, D) Thyroid of a non-infected mouse 48 hours post-transfer of spleen cells from a L. monocytogenes-infected mouse. CFSE-labeled leukocytes are present surrounding thyroid follicles. (E, F) Thyroid of a non-infected mouse injected with CFSE-labeled spleen cells from a non-infected mouse. TF, thyroid follicle; LN, lymph node.
SUMMARY AND CONCLUSIONS

Over the past forty years, a large body of information has come forth defining an intricate nexus between the immune system and the endocrine system. Immune-endocrine pathways have effects on normal as well as pathophysiological processes, some of which is mediated by a novel alternatively-spliced form of TSHβ produced by the hematopoietic system. Indeed, a number of studies remain to be done to fully understand the biological implications of immune system TSHβ cell signaling in the thyroid and bone. For example, the extent to which native TSH and TSHβv work synergistically or antagonistically in delivering TSHR-mediated signals may provide important information into the specific role of TSHβ in AIT and osteoporosis.

AUTHOR CONTRIBUTIONS

JK is the sole author to all aspects of this article.

FUNDING

This work was funded in part by NIH grants R21AI135293 and R21 AI133313.

ACKNOWLEDGMENTS

I wish to thank Dr. Hitesh Pawar for discussions and input on the project.

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Conflict of Interest: The author declares that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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