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Pembrolizumab-Induced Thyroiditis

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Abstract
Immune checkpoint inhibitors act to restore T cell–mediated antitumor immunity. By this nature, these cancer immunotherapy drugs are associated with various immune-related adverse events such as thyroid dysfunction. We describe a case of thyrotoxicosis secondary to a programmed cell death 1 (PD-1) immune checkpoint inhibitor, pembrolizumab. A 30-year-old female was started on pembrolizumab immunotherapy for stage III small cell carcinoma of the ovary, hypercalcemic type. Thirteen days after her second cycle of therapy, she presented with symptoms consistent with thyrotoxicosis. A thyroiditis was diagnosed by thyroid function tests and ultrasonography. She was originally treated with prednisone and metoprolol for possible Grave's disease. Pertechnetate thyroid scan was more consistent with thyroiditis secondary to pembrolizumab. She underwent a total thyroidectomy 10 days after initial presentation for refractory thyrotoxicosis despite maximal medical therapy. Her symptoms resolved and thyroid function tests significantly improved. Pathology was consistent with severe thyroiditis. Immune microenvironment may play a role in the expression of programmed cell death protein 1 ligand 1 (PD-L1). Chronic inflammation surrounding tumor upregulates PD-L1 expression on tumor cells by the release of cytokines, which acts to inhibit tumor destruction. We suggest that our patient had an undetected chronic inflammation of the thyroid, specifically Hashimoto's thyroidits, which predisposed her to thyroid destruction when taking pembrolizumab. Understanding that an inflammatory environment impacts thyroid toxicity to PD-1 inhibitor therapy is novel and should be further studied.

Keywords Pembrolizumab · Thyroiditis · Thyrotoxicosis · PD-1 inhibitor · PDL-1 inhibitor · Thyroid · Small cell carcinoma of ovary

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Case Report

Case

A 30-year-old female with stage III small cell carcinoma of the ovary, hypercalcemic type (pT3N1), on pembrolizumab presented with symptoms of thyrotoxicosis. Her oncologic treatment history was significant for total abdominal hysterectomy with bilateral salpingo-oophorectomy and two cycles of cisplatin, etoposide, and pembrolizumab. Her past medical history is significant for a 5-mm hypoechoic right thyroid nodule discovered on ultrasound 5 months earlier with a normal TSH level.

She presented 13 days after her second cycle of chemotherapy with low-grade fevers, anterior neck discomfort and fullness, tachycardia, palpitations, mild tongue tremor, and mild hyperreflexia. There was no clear source of infection and neutrophil count was normal; she was discharged on Levaquin. Thyroid function tests that resulted after her discharge were

significant for a TSH of 0.08 (0.27–4.20 μIU/mL) and free T4 of 6.63 (0.93–1.70 ng/dL), which lead to her readmission given concern for thyrotoxicosis. Her readmission diagnosis was pembrolizumab-induced thyrotoxicosis; she was treated with 20 mg of prednisone once daily and 25 mg metoprolol tartrate twice daily. However, her thyroid function tests worsened to a TSH of 0.2 (0.27–4.20 μIU/mL), total T3 of > 6.51 (0.8–2.00 ng/dL), T3 uptake of < 0.52 (0.76–1.25), total T4 of 24.86 (4.6–12.00 μg/dL), and free T4 of 7.77 (0.93–1.70 ng/dL). Her lab work was negative for anti-thyroperoxidase antibody, thyroglobulin-stimulating antibody, and TSH receptor antibody. Thyroid ultrasound showed enlargement, heterogeneity, and hyperemia of thyroid parenchyma, consistent with thyroiditis and bilateral enlarged, possibly reactive lymph nodes.

She was then treated with 30 mg methimazole once daily to treat possible Graves' disease given the rapid worsening of thyrotoxicosis, despite negative serum antibodies. Pertechnetate thyroid scan 2 days later showed no significant uptake in the thyroid gland, more consistent with thyroiditis secondary to pembrolizumab versus Graves' disease. Thyroid studies the following day showed no improvement. Methimazole was discontinued, prednisone dose was increased to 40 mg once daily, metoprolol 25 mg was increased to four times daily for continued tachycardia, and she was started on cholestyramine 4 g twice daily. Thyroid function tests remained unchanged and she began to develop orthostatic hypertension and metoprolol was decreased to 50 mg twice daily.

Ten days after initial presentation of thyrotoxicosis, she underwent a total thyroidectomy for refractory thyrotoxicosis despite maximal medical therapy. Gross pathologic examination was significant for an enlarged firm, nodular thyroid gland. Serial sectioning showed a 2-mm well-circumscribed, un-encapsulated nodule within the isthmus, but an otherwise grossly brown-tan thyroid parenchyma. Microscopically, severe thyroiditis with extensive lymphohistiocytic infiltration of thyroid parenchyma with follicular destruction and paucity of colloid was seen (Fig. 1 & 2). Three lymph nodes were negative for tumor and the 2-mm nodule found grossly in the isthmus was a fibrotic nodule. CD3 immunostain highlighted diffusely scattered T cells and CD79A highlighted foci of B cells; T:B cell ratio was 60:40. A CD68 immunostain highlighted histiocytes. A CD138 immunostain highlighted scant focal areas of plasma cells and TTF-1 and AE1-3 highlighted follicular cells underlying the inflammation. A PDL-1 immunostain highlighted predominantly histiocytes (Fig. 3).

Post-thyroidectomy, the patient's tachycardia improved. Seven days after surgery, her thyroid function tests significantly improved to euthyroid range with a free T4 of 1.07, total T4 of 6.25, T3 uptake of 1.00, and total T3 of 1.13. TSH was still low at < 0.01.

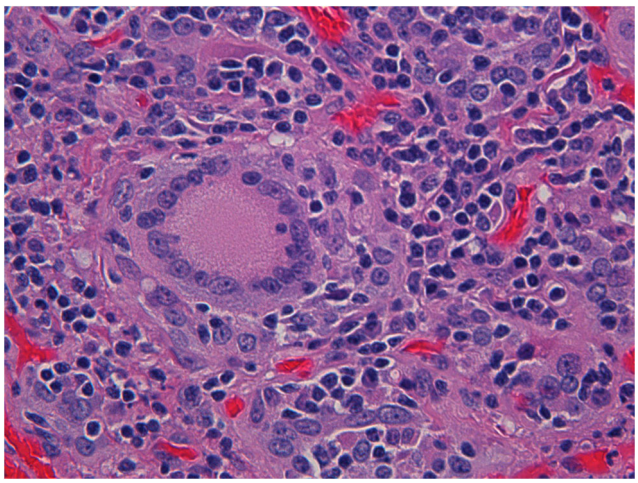

Fig. 1 This is a medium-power view of the thyroid showing a damaged follicle with residual colloid. Most of the follicular cells have enlarged nuclei with prominent nuclei and are uniform in their appearance, suggesting they are reactive to the surrounding severe inflammation. The stroma around the follicle is replaced by an intense lymphohistiocytic infiltrate with a few plasma cells (H&E, × 100)

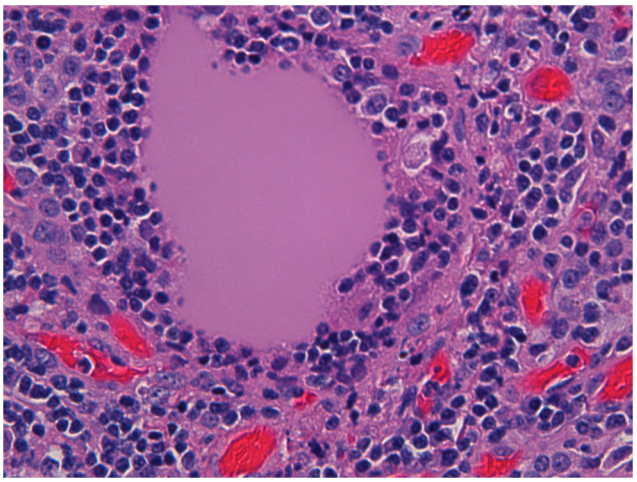

Fig. 2 At higher power, the pattern of most of the thyroid histology is seen. The illustrated follicle shows replacement of the follicular epithelial cells with lymphocytes; only a rare degenerating follicular cell remains. The intense stromal inflammatory response is noted. (H&E, × 250)

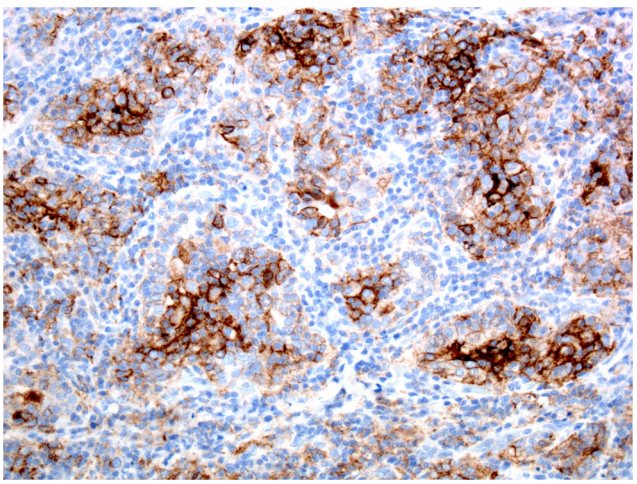

Fig. 3 Stain for PDL-1 highlights predominantly histiocytes

Discussion

Programmed cell death 1 (PD-1) transmembrane receptor is an immune checkpoint receptor on activated CD4+ and CD8+ T cells, B cells, natural killer cells, macrophages, and dendritic cells, and is important in maintaining self-tolerance. It binds two inhibitory ligands, programmed cell death protein 1 ligand 1 (PD-L1) and PD-L2. When ligated, cell function is downregulated and tolerance to self-antigens is developed [1, 2].

Tumor cells can express PD-L1 to evade immune attack. PD-L1 overexpression can be related to genetic aberrations or attributable to tumor microenvironment, both of which suppress antitumor T cell response. In fact, secretion of proinflammatory cytokines from activated T cells infiltrating tumor upregulates PD-L1 on tumor cells [3]. Various human cancers overexpress PD-L1 including head and neck cancer, breast cancer, ovarian cancer, renal cancer, pancreatic cancer, esophageal cancer, non-small cell lung cancer, melanoma, and glioblastoma. PD-L1 can also be expressed in thyroid inflammatory processes and neoplasms [4].

PD-1 inhibitors are immune checkpoint-inhibiting monoclonal antibodies that prevent T cell PD-1/tumor cell PD-L1 interaction, which restores T cell–mediated antitumor immunity [3, 5]. Pembrolizumab, a PD-1 inhibitor, is a humanized monoclonal IgG4 kappa anti-PD1 antibody. Pembrolizumab has shown efficacy in treating melanoma, non-small cell lung cancer, genitourinary cancer, Hodgkin's lymphoma, and Merkel-cell carcinoma. Immune checkpoint blockade, by the nature of restoring T cell function, is associated with immune-related adverse events including dermatologic, arthralgia/arthritis, thyroid dysfunction, gastrointestinal, hypophysitis, hepatitis, and more rarely pneumonitis, uveitis, and parotitis [3, 6].

Immune microenvironment may play a role in the expression of PD-L1. Various reports have observed increased PD-L1 expression in chronic lymphocytic thyroiditis (CLT) and Hashimoto's thyroiditis (HT), which suggests that a cytokine-rich environment provided by chronic inflammation could potentiate PD-L1 expression [4, 7–10]. Chowdury et al. found increased PD-L1 expression in CLT (60% cytoplasm positive and 40% membrane positive) and HT (80% cytoplasm positive and 20% membrane positive), while benign nodules had no detectable PD-L1 staining [4]. Chronic inflammation in the background of tumor releases certain cytokines including IFN gamma, IL-1, IL-10, and IL-6, which upregulate PD-L1 expression on tumor cells. In essence, chronic inflammation surrounding tumor acts to inhibit tumor destruction [4, 10]. Taube et al. analyzed human melanocytic lesions and found a strong association between melanocyte expression of PD-L1 and the presence of tumor-infiltrating lymphocytes. In fact, 98% of PD-L1 positive tumors were associated with tumor-infiltrating inflammation compared with only 28% of PD-L1 negative [10]. A study investigating the role of PD-L1 in differentiated thyroid carcinoma found that the presence of CD4+, CD8+, CD20+, and FoxP3+ lymphocytes; tumor-associated macrophages; and myeloid-derived suppressor cells was associated with elevated levels of PD-L1 [11]. Lubin et al. determined that the presence of CLT and HT influences the expression of PD-L1 on benign follicular metaplastic epithelium and in papillary thyroid carcinoma (PTC). In this study, thyroids lacking thyroiditis, including benign thyroids and papillary thyroid carcinomas arising in unremarkable thyroid, did not express any PD-L1. Two of the five cases with CLT demonstrated PD-L1 expression while all five cases of HT showed significant PD-L1 expression. PTC arising in a background of HT showed significant PD-L1 expression, while only one in ten cases of PTC arising in CLT expressed PD-L1 [7] Fig. 3. In comparison, we stained four in-house thyroids with autoimmune hyperthyroidism (diffuse toxic goiter) and no staining was identified in any case for PD-L1 or PD-1. Immune microenvironment indeed plays a pivotal role in PD-L1 expression.

Thyroid toxicity associated with PD-1 inhibitors is rare and poorly understood. Thyroid toxicity typical presents either as an initial thyrotoxic phase followed by hypothyroidism or isolated hypothyroidism without a previously detected thyrotoxic phase. Those who present with an initial thyrotoxic phase usually show signs of hyperthyroidism 3–6 weeks after pembrolizumab initiation and the hypothyroid phase is evident within 3 weeks of the thyroiditis [2, 6]. The literature is unclear as to which presentation is more common. According to Filette et al., the incidence of thyroid-related adverse events due to pembrolizumab therapy was 17%. Of this population, 67% presented with thyrotoxicosis and 33% presented with isolated hypothyroidism [5]. Another study found that of the 10 patients who received anti-PD-1 mAb and developed thyroid dysfunction, 60% presented with thyrotoxicosis followed by hypothyroidism, while 40% presented with isolated hypothyroidism [2]. A meta-analysis evaluating 38 randomized clinical trials determined that the incidence of presenting with hypothyroidism was actually higher than the incidence of presenting with hyperthyroidism with rates of 6.6% and 2.9%, respectively. This same study also found that the risk of hyperthyroidism was significantly greater with PD-1 inhibitors, like pembrolizumab, than with PD-L1 inhibitors. Further, when comparing two PD-1 inhibitors, it was found that pembrolizumab causes higher rates of hyperthyroidism than nivolumab (3.8% vs 2.5%) [12].

Understanding the mechanism of thyrotoxicosis as a result of PD-1 inhibitor therapy is important for treatment decisions. PD-1 inhibitor–induced thyrotoxicosis is caused by either destructive thyroiditis or autoimmunity. Destructive thyroiditis is characterized by a rise in FT3, FT4, and Tg from destroyed follicles and typically initially manifests as hyperthyroidism. As thyroid hormone stores are depleted, a period of euthyroidsm presents followed by hypothyroidism [1, 13]. Graves' disease, on the other hand, is characterized by an increase of TSAbs, which activate TSHR. Follicles are

hyperfunctioning in Graves' disease so scintigraphy shows increased uptake, whereas in thyroiditis, thyroid follicles are damaged so scintigraphy will show reduced uptake [1]. Thyroxine to triiodothyronine levels are higher in patients with thyroiditis due to the sudden release of thyroid hormones, and because thyroxine is less active than triiodothyronine, patients with thyroiditis typically present with a less severe clinical presentation compared with patients with Graves' disease [14].

Thyroiditis seen in PD-1 inhibitor therapy may be akin to the mechanism of thyroid destruction seen in various thyroid cancers and subacute thyroiditis. Thyroid cancers cause destructive thyroiditis by rapid infiltration with malignant cells causing destruction of thyroid follicles. In subacute thyroiditis, an inciting event causes proteolysis of thyroglobulin molecules. Both mechanisms result in leakage of thyroid hormones into circulation [14, 15]. Delivanis et al. determined that the mechanism of inflammatory destructive thyroiditis caused by anti-PD-1 therapy is attributable to increased circulating CD45+CD16+ natural killer cells and an elevated HLA-DR surface expression in the inflammatory intermediate CD14+CD16+ monocytes. Additionally, PD-1 blockade restores T cell function of previously suppressed T cells, which may also contribute to this drug's induced thyroiditis [16].

We believe that the hyperthyroidism/thyrotoxicosis evident in our patient on pembrolizumab was due to destructive thyroiditis and subsequent release of preformed thyroid hormones into peripheral circulation. Although our patient presented with a severe thyrotoxicosis more classic of a Graves' presentation than a thyroiditis presentation, our patient did not have any laboratory or radiologic evidence Grave's disease. Our patient did not have elevated anti-thyroid antibodies and pertechnetate thyroid scan showed no significant uptake in the thyroid gland. Immunohistochemical staining proved the presence of a mixed population of inflammatory cells and follicular destruction throughout the thyroid, characteristic of thyroiditis.

We can also suggest the possibly that our patient had an undetected sero-negative HT prior to initiation of PD-1 inhibitor therapy. Although our patient did not have elevated serum thyroid peroxidase antibody concentrations, it has been estimated that between 5 and 10% of patients with Hashimoto's thyroiditis do not have elevated antibody concentrations. From previous reports, we know that HT is associated with PD-1 expression on benign follicular metaplastic epithelium. We are suggesting that because our patient developed fulminant thyrotoxicosis shortly after the initiation of a PD-1 inhibitor, she must have had undetected HT which propagated the expression of PD-1 on her follicular epithelium. Ligation of PD-1 with PD-1 inhibitor caused follicular destruction and subsequent thyrotoxicosis [4, 7]. It is unlikely that our patient had a benign thyroid prior to PD-1 inhibitor therapy.

Conclusion

Anti-PD-1 mAb therapy can be toxic to the thyroid gland, most commonly causing thyrotoxicosis followed by hypothyroidism or just isolated hypothyroidism. Chronic inflammation in the thyroid, specifically Hashimoto's thyroiditis, has been shown to cause expression of PD-L1 on follicular epithelial cells, thus predisposing patients with this disease who are taking PD-1 inhibitors to thyroid destruction. The mechanism and histology of this thyroid destruction are poorly understood, but recognizing that an inflammatory environment impacts thyroid toxicity to PD-1 inhibitor therapy is novel and should be further studied.

Compliance with Ethical Standards

Conflict of Interest The authors declare that they have no conflict of interest.

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