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Clinical Research Article

Spectrum of Clinical Presentations, Imaging Findings, and HLA Types in Immune Checkpoint Inhibitor–Induced Hypophysitis

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Abstract

Context: Hypophysitis is a known immune-related adverse event (irAE) of immune checkpoint inhibitors (CPIs), commonly associated with CTLA-4 inhibitors and less often with PD-1/PD-L1 inhibitors.

Objective: We aimed to determine clinical, imaging, and HLA characteristics of CPI-induced hypophysitis (CPI-hypophysitis).

Methods: We examined the clinical and biochemical characteristics, magnetic resonance imaging (MRI) of the pituitary, and association with HLA type in patients with CPI-hypophysitis.

Results: Forty-nine patients were identified. Mean age was 61.3 years, 61.2% were men, 81.6% were Caucasian, 38.8% had melanoma, and 44.5% received PD-1/PD-L1 inhibitor monotherapy while the remainder received CTLA-4 inhibitor monotherapy or CTLA-4/PD-1 inhibitor combination therapy. A comparison of CTLA-4 inhibitor exposure vs PD-1/PD-L1 inhibitor monotherapy revealed faster time to CPI-hypophysitis (median 84 vs 185 days, $P < .01$) and abnormal pituitary appearance on MRI (odds ratio 7.00, $P = .03$). We observed effect modification by sex in the association between CPI type and time to CPI-hypophysitis. In particular, anti-CTLA-4 exposed men had a shorter time to onset than women. MRI changes of the pituitary were most common at the time of hypophysitis diagnosis (55.6% enlarged, 37.0% normal, 7.4% empty or partially empty) but persisted in follow-up (23.8% enlarged, 57.1% normal, 19.1% empty or partially empty). HLA typing was done on 55 subjects; HLA type DQ0602 was over-represented in CPI-hypophysitis relative to the Caucasian American population (39.4% vs 21.5%, $P = 0.01$) and CPI population.

Conclusion: The association of CPI-hypophysitis with HLA DQ0602 suggests a genetic risk for its development. The clinical phenotype of hypophysitis appears heterogenous, with differences in timing of onset, changes in thyroid function tests, MRI changes, and possibly sex related to CPI type. These factors may play an important role in our mechanistic understanding of CPI-hypophysitis.

Key Words: hypophysitis, immune checkpoint inhibitors, immunotherapy, immune-related adverse events, pan-hypopituitarism, adrenal insufficiency

Abbreviations: ACTH, adrenocorticotropic hormone; CPI, immune checkpoint inhibitors; CTLA-4, cytotoxic T-lymphocyte associated protein 4; fT4, free thyroxine; HLA, human leukocyte antigen; HPA, hypothalamic-pituitary-adrenal; HPT, hypothalamus-pituitary-thyroid; irAE, immune-related adverse event; MRI, magnetic resonance imaging; PD-1, programmed cell death protein 1; PD-L1, programmed cell death ligand 1; PRL, prolactin; TSH, thyrotropin (thyroid-stimulating hormone); UCLA, University of California, Los Angeles; UCSF, University of California, San Francisco.

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The rise in use of immune checkpoint inhibitors (CPIs) has been accompanied by an increase in the incidence of hypophysitis and other endocrinopathies [1]. The occurrence of CPI-hypophysitis can lead to adrenal crisis, hospitalization, and even death [2]. Initial studies of CPI-hypophysitis suggested that the disease was restricted to treatment with ipilimumab (a cytotoxic T-lymphocyte-associated protein 4 [CTLA-4] inhibitor) and was common, occurring in 9% to 13% [1] of cancer patients. It is now also known to occur, albeit much less frequently (1%-5%) of patients, following exposure to CPIs targeting the programmed cell death protein-1/programmed death ligand 1 (PD-1/PD-L1) axis [1, 3–5]. In fact, recent studies showed that there were clinical differences among cases dependent on the type of CPI received including affected hormonal axes and imaging changes [3–6]. Additionally, it had previously been noted that imaging changes in CPI-hypophysitis were short-lived [7–9] and that the appearance of the pituitary gland returned to normal. However, in our clinical experience, imaging changes were often not observed even at the time of diagnosis, and this led us to pursue an in-depth review.

Prior to the introduction of CPIs, hypophysitis was rare, occurring in 1 person per 7 to 9 million people per year [10]. Multiple histologic subtypes exist: lymphocytic, granulomatous, xanthomatous, and plasmacytic (IgG4-related). Spontaneous lymphocytic hypophysitis, granulomatous hypophysitis, and xanthomatous hypophysitis are more common in women (approximately 3:1) and on average occur in the fourth and fifth decades of life [10]. In contrast, plasmacytic hypophysitis is more common in men (approximately 2:1) and occurs later, on average in the seventh decade of life [10]. CPI-hypophysitis is thought to be a form of lymphocytic hypophysitis driven by activation of the immune response during exposure to cancer immunotherapies [11]. Given the rarity of spontaneous lymphocytic hypophysitis, autoantibodies and genetic risk are less well established, although there are some reported associations [12, 13]; in particular, there is a demonstrated association between lymphocytic hypophysitis and HLA DR8 and DR53 [13]. Additionally, small studies have shown associations between HLA and CPI-hypophysitis in non-Caucasian populations [14, 15].

In this study, we identified cases of CPI-hypophysitis managed at our tertiary care institution. We characterized clinical differences in phenotype based on CPI type, evaluated the evolution of magnetic resonance imaging (MRI) changes during the course of hypophysitis diagnosis and treatment, and, when possible, completed HLA typing. Despite increasing awareness of immune-related adverse events (irAEs), CPI-hypophysitis often is delayed or missed which can lead to negative patient outcomes. Clarifying the variability in the clinic presentations can help increase this awareness. Risk prediction, through clinical factors and HLA typing could also help to risk stratify patients. HLA associations can identify genetic and autoimmune components that may be contributing to disease susceptibility.

Methods

UCSF Case Identification

Possible cases were identified in 3 ways (Fig. 1): (1) referral to the University of California, San Francisco (UCSF) Endocrine Clinic from 2012 to 2020; (2) database search for the word "hypophysitis" within MRI brain radiology reports performed at UCSF from 2012 through September 2019; and (3) review of all patients receiving CPIs who had abnormal cosyntropin stimulation tests (although this is not necessary for the diagnosis) at UCSF from 2012 through February 2019. After gathering these possible cases, patients were reviewed to see if they met the case definition.

Case Definition

Patients were initially identified by their oncologists as at risk for hypophysitis due to symptoms such as headache, nausea, vomiting, and fatigue. These clinical concerns led to laboratory testing, imaging and/or referral to endocrinology. Each case was individually reviewed by board-certified endocrinologists at UCSF (Z.Q., S.K., U.M.) who concurred on the diagnosis of hypophysitis with biochemical impairment, including the presence of either central adrenal insufficiency without recent glucocorticoid use or central hypothyroidism. Subjects who had MRI evidence consistent with hypophysitis but lacked biochemical evidence of hormone deficiency were not included. Subjects who had clinical symptoms consistent with hypophysitis but were either already on glucocorticoids or had incomplete laboratory assessment for anterior pituitary function (ie, adrenocorticotropic hormone [ACTH]-cortisol and thyrotropin [TSH]–free thyroxine [fT4] axis) were also excluded.

Additional cases of CPI-associated hypophysitis for HLA association studies were identified by referral from the Endocrine Division at Yale University (New Haven, CT) and a specialized onco-endocrinology clinic at the University of California, Los Angeles (UCLA, Los Angeles, CA) through 2022. Patients were confirmed to have hypophysitis by endocrinologists familiar with endocrine irAEs at their local institutions (A.L.P., M.L.). Criteria for the diagnosis of CPI-hypophysitis in these patients was congruent with the case definition above. Detailed longitudinal clinical and imaging data were not readily available to be included in the primary analysis. The group including both the UCSF cases and these subjects is referred to as the "expanded cohort."

Additional Variables of Interest

Covariates including clinical and laboratory information were extracted from the electronic health record and supplemented by directed, retrospective chart review. Outside records were reviewed as appropriate. Laboratory values that were reviewed included tests pertaining to the adrenal axis (ACTH, cortisol, and cosyntropin stimulation test results, if available); thyroid axis (TSH, fT4); and reproductive axis (follicle stimulating hormone, luteinizing hormone, and testosterone]. In an attempt to differentiate nonthyroidal illness from central hypothyroidism, we defined central hypothyroidism as someone with TSH $\leq$0.1 mIU/L (normal range, 0.45-4.12 mIU/L) and/or a fT4 $\leq$7 pmol/L (normal range, 10-18 pmol/L). Prolactin (PRL) levels before and after CPI initiation and around the time of hypophysitis diagnosis were extracted from the electronic health record and classified as low, normal, or elevated, based on respective references ranges.

Imaging Changes

Pituitary imaging performed for clinical care, including assessment for metastatic disease or of symptoms such as headache,

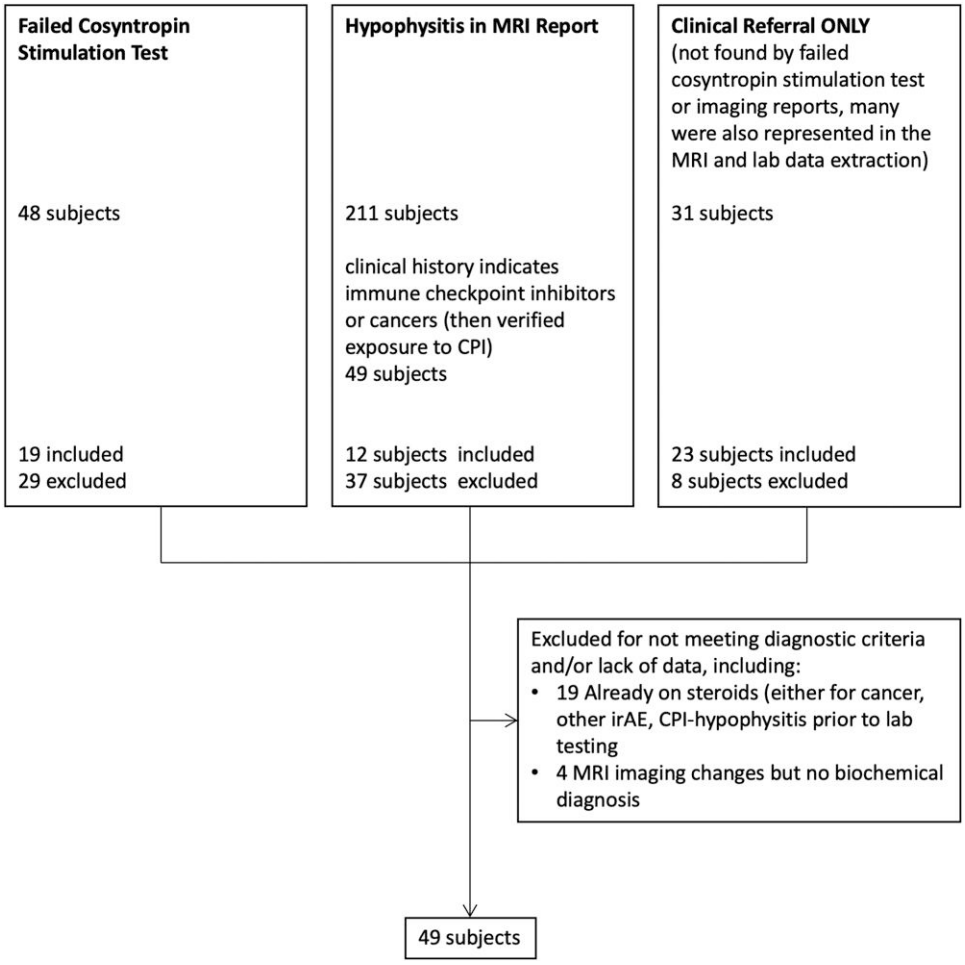

Figure 1. Study flow chart. Depiction of methods used to identify patients and exclude patients from this study.

was reviewed by a board-certified neuroradiologist (J.V.M.). Twenty-five subjects had MRI scans in at least 2 time periods defined below. Outside records were reviewed extensively to capture complete imaging data for patients. All patient MRIs were re-reviewed (J.V.M.) blinded to time of onset of hypophysitis for an unbiased approach. The pituitary gland, including the infundibulum, was categorized as being normal in appearance, enlarged, or small (labeled empty or partially empty sella). Categorization was performed on post-contrast T1-weighted images, either small-field-of-view two-dimensional (2D) sagittal and coronal imaging targeted to the pituitary gland or high-resolution isotropic 3D imaging of the brain. The timing of the MRI scans was classified relative to hypophysitis diagnosis (Fig. 2): baseline (at least 40 days prior), diagnosis (within 40 days), and follow-up (over 40 days), using time intervals suggested in the published literature based on time to resolution of imaging changes [16]. If multiple MRIs were done within the same period of time and were incongruent, the abnormal MRI was selected to represent that time period.

HLA Typing

DNA was extracted from patient blood clots using the Qiagen Gentra Puregene Blood Kit. HLA typing was carried out using single molecule, real time (SMRT) sequencing to a 4X, three-field resolution by Histogenetics. Additional control subjects were pulled from healthy controls and cancer patients exposed to CPI but without development of CPI-hypophysitis or CPI-diabetes (as there was a possibility for particular HLA associations as has previously been published [17]).

Statistical Analyses

Outcomes assessed were the time to CPI-hypophysitis diagnosis and presence and timing of imaging changes. After detailed comparisons between individual groupings of PD-1 inhibitor monotherapy, PD-L1 inhibitor monotherapy, CTLA-4 inhibitor monotherapy, and CTLA-4 combination therapy, we grouped CPI exposure into individuals with CTLA-4 exposure at CPI-hypophysitis diagnosis (CTLA-4 inhibitor monotherapy or combination therapy) and CTLA-4 unexposed at CPI-hypophysitis diagnosis (PD-1/PD-L1 inhibitor monotherapy). This grouping optimized the sample size and power for our analyses, but was supported by similarities in our data, mechanism of the agents, and common. We also evaluated the range of anterior pituitary hormone deficits and patient sex. Univariate analyses were completed using Student *t* tests and Fisher exact tests as appropriate. Median time to diagnosis was compared using Wilcoxon rank sum tests. Longitudinal analyses assessing time to diagnosis were completed using Kaplan-Meier survival curves with log rank test for equality for individual covariates and Cox proportional hazard models for multivariate analyses. Logistic regression

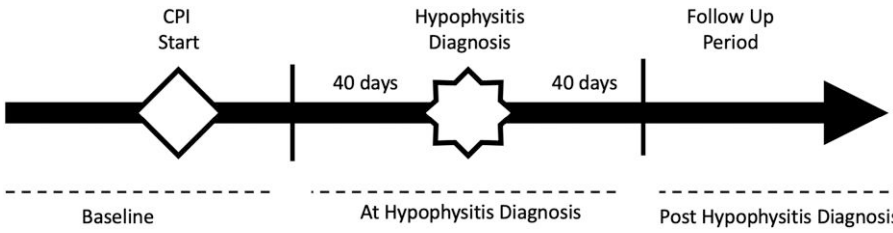

Figure 2. Schematic of time course of subjects from CPI start to hypophysitis diagnosis to follow-up. Magnetic resonance imaging (MRI) performed 40 days prior to hypophysitis diagnosis was considered *baseline* imaging. MRI performed within 40 days of diagnosis was considered *at diagnosis*. MRIs done 40 days after diagnosis were considered *post*.

was used to evaluate the odds of having an enlarged or abnormal appearing pituitary gland on imaging. Two sets of models were fit; the first included type of CPI and the second set of models included type of CPI and either age or sex or thyroid involvement. There were an insufficient number of patients with abnormal imaging be able to include all these variables in one model. Due to the predominance of women in lymphocytic hypophysitis, effect modification by sex was assessed for the time to CPI-hypophysitis diagnosis and imaging changes.

Three HLA types were tested in an a priori fashion for overrepresentation in our CPI-hypophysitis cohort: (1) HLA DQ8 (DQB1*03:02); (2) HLA DR53 (DRB4*01:01 or DRB4*01:03); and (3) HLA DQ0602 (DQB1*06:02). HLA DQ8 and DR53 were chosen based on the prior literature [13] showing an association with sporadic lymphocytic hypophysitis and HLA DQ0602 was chosen based on pilot data comparing CPI-induced diabetes mellitus, type 1 diabetes, type 2 diabetes, and CPI-hypophysitis, as HLA DQ0602 is protective again type 1 diabetes mellitus. These rates were compared to our UCSF CPI-exposed and healthy control cohort, previously published rates for sporadic lymphocytic hypophysitis, sellar masses, and 4 cases of CPI-hypophysitis at UCLA [13] using a Fisher exact test. They were compared to established rates in large cohorts of Caucasian Americans using an equality of proportions test [18, 19].

An alpha value of .05 was considered significant for all analyses aside from tests for effect modification, which used an alpha of .10. Statistics were completed using Stata 15 (College Station, TX).

Ethical conduct of research: institutional review board (IRB) approval for the retrospective case analysis was obtained from the UCSF Committee for Human Research (UCSF IRB 17-22987). Prospectively collected patient specimens for HLA studies were collected under IRB approved protocols at each center with written informed consent: UCSF IRB 10-02467, UCLA 19-000032 and 19-001708, and Yale 0608001773.

Results

Baseline Characteristics

Using the case definition criteria, 49 patients at UCSF were identified with CPI-hypophysitis (Fig. 1). The mean age of the cohort was 61.3 years (SD 10.98), 61.2% were men, 81.6% were Caucasian (Table 1). Median follow-up time after CPI-hypophysitis diagnosis was 522 days (interquartile range [IQR] 306, 775). There was a wide range of malignancies with a preponderance of melanoma (38.8%), which is the most common type of cancer treated with CPI at our institution. Three patients (6.1%) had pre-existing autoimmune thyroid disease and 13 patients (26.5%) had a known thyroid irAE prior to the diagnosis of CPI-hypophysitis and therefore central hypothyroidism could not be assessed in these patients. Of the included HLA types within this study, 39.4% had HLA DQ0602, 9.09% had DQ8, and 30.3% had DR53, the latter 2 HLA types having been associated with spontaneous hypophysitis previously [13]. Age, sex, race/ethnicity, personal and family history of autoimmune disease, prior thyroid disease, and the 3 HLA types examined did not differ by type of CPI therapy received (Table 1, Supplementary Table S1 [20]). Laboratory testing and imaging were performed as part of routine clinical care, occurring as shown in Fig. 3.

CPI Therapy

Combination CTLA-4 inhibitor therapy and PD-1 inhibitor monotherapy were the most common treatments at the time of CPI-hypophysitis diagnosis. Twenty-seven (55.5%) were exposed to CTLA-4 inhibitors, either as monotherapy (8 [16.3%]) or in combination with PD-1 inhibitors (19 [38.7%]) while 22 (44.5%) were treated with PD-1 inhibitor monotherapy (19 [38.7%]) or PD-L1 inhibitor monotherapy (3 [6.1%]). We note that PD-1 inhibitor monotherapy is the most common form of CPI used at our institution, so we have a large number of patients with CPI-hypophysitis without exposure to CTLA-4 inhibitors, despite it being less common in inhibitor exposure. Individuals with melanoma were more likely to be exposed to CTLA-4 inhibitors compared to those with other cancers ($P = .04$) (Supplementary Table S1 [20]), consistent with FDA-approved indications for CTLA-4 inhibitors.

Pituitary-Adrenal Axis

CPI-hypophysitis was identified in most patients due to symptoms of adrenal insufficiency such as fatigue, weakness, and orthostasis. Serum ACTH level was below the reference range in all 47 patients who had not already been started on glucocorticoids. Additionally, these patients showed low morning cortisol (32% only had paired basal levels) and/or failed a 250 microgram cosyntropin stimulation test (68% of the cohort). All 32 patients who underwent a cosyntropin stimulation test failed, although it is important to note that a patient early in the CPI-hypophysitis course may still have sufficient adrenal reserve for this test to be normal and a cosyntropin stimulation test is not required to diagnosis CPI-hypophysitis per National Comprehensive Cancer Network (NCCN) guidelines. No patient had recovery of the pituitary-adrenal function during follow-up (Table 2, and Fig. 3). Within the expanded cohort, adrenal insufficiency was diagnosed by either paired

Table 1. Demographic and clinical features

	Full cohort		PD-1 inhibitor monotherapy at HP Dx		PD-L1 inhibitor monotherapy at HP Dx		CTLA-4 inhibitor combination at HP DX		CTLA-4 inhibitor monotherapy at HP DX		PD-1/PD-L1 inhibitor monotherapy at HP Dx		CTLA-4 inhibitor monotherapy or combination at HP DX		*P* value: CTLA4-inhibitor–exposed vs unexposed
	n = 49		n = 19		n = 3		n = 19		n = 8		n = 22		n = 27		
	n	(%) or SD	n	(%) or SD	n	(%) or SD	n	(%) or SD	n	(%) or SD	n	(%) or SD	n	(%) or SD	
Age in years, (mean, SD)	61.25	SD 10.98	60.53	SD 12.21	62.33	SD 7.02	60.42	SD 10.68	64.50	SD 10.97	60.77	SD 11.53	61.63	SD 10.72	.79
Gender (n, %)															.24
Male	30	61.22	9	47.37	2	66.67	13	68.42	6	75.00	11	50	19	70.37	
Female	19	38.78	10	52.63	1	33.33	6	31.58	2	25.00	11	50	8	29.63	
Race (n, %)															
Caucasian	40	81.63	4	21.05	0	0.00	17	89.47	6	75.00	17	77.27	23	85.19	.53
Asian	6	12.24	14	73.68	3	100.00	1	5.26	1	12.50	4	18.18	2	7.41	
Other	3	6.12	1	5.26	0	0.00	1	5.26	1	12.50	1	4.55	2	7.41	
Ethnicity (n, %)															
Not Hispanic or Latino	47	95.92	17	89.47	3	100.00	19	100.00	8	100.00	20	90.91	27	100	.20
Hispanic or Latino	2	4.08	2	10.53	0	0.00	0	0.00	0	0.00	2	9.09	0	0	
History of Autoimmune Disease[a](n, %)															
Present	7	14.29	2	10.53	0	0.00	2	10.53	3	37.50	2	8.7	5	19.23	.42
Absent	42	86.71	17	89.47	3	100.00	17	89.47	5	62.50	20	91.3	22	80.77	
Family History of Autoimmune Disease[b](n, %)															
Present	14	28.57	4	21.05	2	66.67	5	26.32	3	37.50	6	27.27	8	29.63	.56
Absent	35	71.43	15	78.95	1	33.33	14	73.68	5	62.50	16	72.73	19	70.37	
Cancer Type[c](n, %)															
Melanoma	19	38.78	5	26.32	3	100.00	10	52.63	4	50.00	5	22.73	14	51.85	**.05**
Other Cancers	30	61.22	14	73.68		0.00	9	47.37	4	50.00	17	77.27	13	48.15	
Thyroid Disease History (n, %)															.89
No Thyroid Disease Prior to Hypophysitis	33	67.35	14	73.68	2	66.67	12	63.16	5	62.50	16	72.73	17	62.96	
Pre-existing Autoimmune Thyroid Disease	3	6.12	1	5.26	0	0.00	1	5.26	1	12.50	1	4.55	2	7.41	
Primary Thyroid irAE Prior to Hypophysitis (n, %)	13	26.53	4	21.05	1	33.33	6	31.58	2	25.00	5	22.73	8	29.63	

Features with a *P*-value less than or equal to 0.05 are noted in bold.
Abbreviations: Dx, diagnosis; HP hypophysitis.
[a]Autoimmune diseases consisted of Graves disease, Hashimoto thyroiditis, rheumatoid arthritis, and vitiligo.
[b]Family history of autoimmune diseases consisted of Graves disease, Hashimoto thyroiditis, rheumatoid arthritis, inflammatory bowel disease, vitiligo, and multiple sclerosis.
[c]Other cancers include bladder, breast, endometrial, gastric adenocarcinoma, glioblastoma, hepatocellular carcinoma, head and neck squamous cell carcinoma, mixed hepatocellular carcinoma/cholangiocarcinoma, non-small cell lung cancer, prostate cancer, colorectal cancer, mesothelioma, small bowel neuroendocrine tumor (See Supplementary Table S1 [20]).

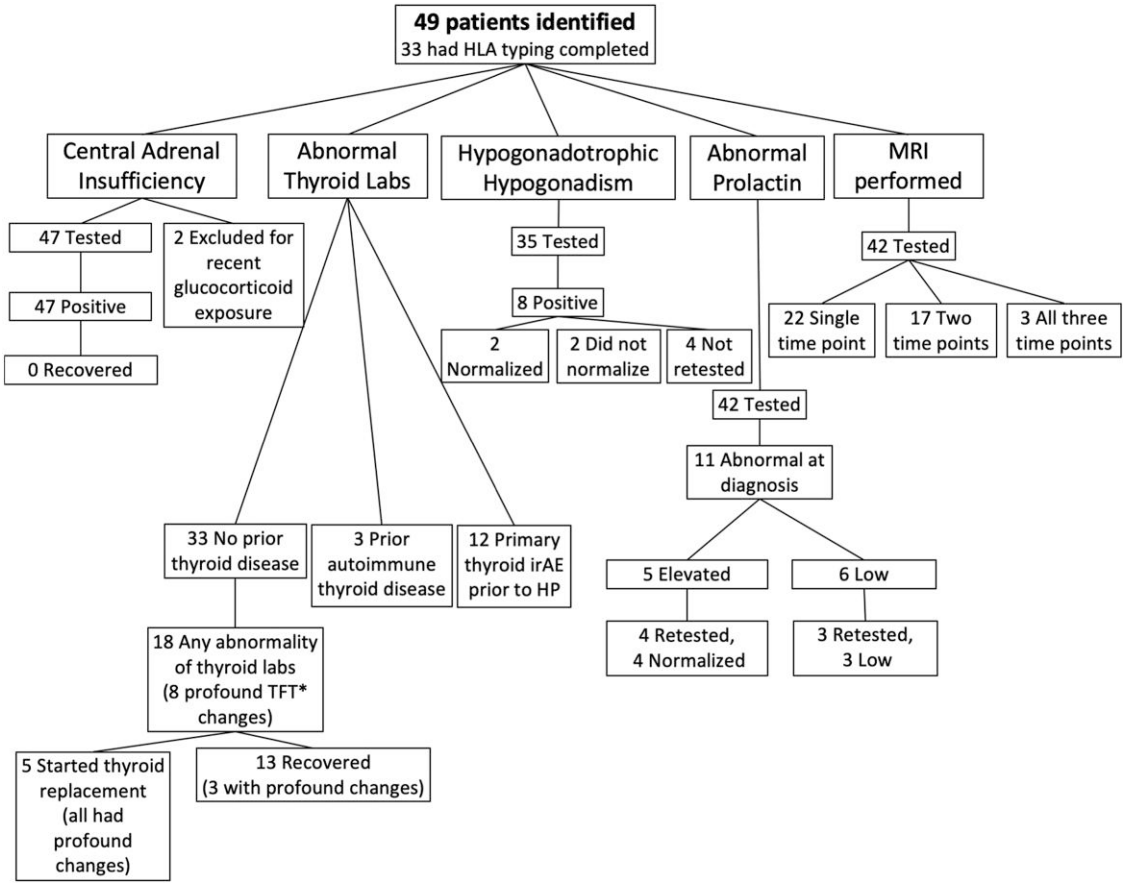

Figure 3. Distribution of patients by relevant laboratory testing and imaging. Forty-nine patients were identified but not all patients underwent laboratory testing or imaging pertinent to their hypophysitis diagnosis at the time of diagnosis or in follow-up. *Profound/clinically significant TFT changes: TSH ≤ 0.1 (0.45-4.12 mIU/L) and/or fT4 ≤ 7 (10-18 pmol/L). Abbreviations: HP, hypophysitis; irAE, immune-related adverse events; MRI, magnetic resonance imaging; TFT, thyroid function tests.

basal cortisol and ACTH levels (71.4%, 30% of whom had normal cosyntropin stimulation tests) or 250 microgram cosyntropin tests (28.6%) for all subjects from Yale and with paired cortisol and ACTH levels for all subjects from UCLA. Lab tests were initially completed or corroborated by labs at UCSF, Yale, or UCLA with cortisol cutoffs between 14 and 18 micrograms/deciliter, appropriate to the assay being used at the time.

Pituitary-Thyroid Axis

Evaluating the 33 patients with CPI-hypophysitis who did not have pre-existing thyroid disease, 18 (54%) had new thyroid hormone abnormalities of decreased TSH and/or fT4. This pattern can be seen in both central hypothyroidism and nonthyroidal illness (Table 2, and Fig. 3). Of these 18 patients, 10 had mild thyroid function abnormalities (TSH below the lower limit of normal but >0.1 mIU/L or fT4 between 7 pmol/L and the lower limit of normal) and recovered to euthyroid state without intervention (Fig. 3). This is most consistent with nonthyroidal illness. Eight patients, however, appeared to have laboratory test results consistent with central hypothyroidism (TSH ≤ 0.1 mIU/L and/or fT4 ≤ 7 pmol/L). Of these 8 patients, 3 were not treated with thyroid replacement hormone and spontaneously recovered normal thyroid status. The remaining 5 patients were treated acutely with levothyroxine, and therefore recovery of the pituitary-thyroid axis could not be determined. Abnormalities of serum TSH and fT4 were more likely to occur in patients exposed to CTLA-4 inhibitors compared with exposure to PD-1/PD-L1 inhibitors (76.5% vs 31.2% $P = .02$). All cases with laboratory changes that were suggestive of central hypothyroidism occurred in patients exposed to CTLA-4 inhibitors (Table 2). HLA type was not associated with the presence or absence of thyroid function abnormalities.

Pituitary-Gonadal Axis

The reproductive axis was evaluated in 35 of 49 patients and 8 (22.9%) had hypogonadotrophic hypogonadism (2/15 women and 6/20 men). Of the 6 who had follow-up testing, 2 showed recovery (Table 2 and Fig. 3). There was no difference in reproductive axis dysfunction between men and women or by CPI type.

PRL levels were measured in 42 of 49 subjects. Of those evaluated, 31 patients had normal PRL at the time of diagnosis, 5 (11.9%) had elevated levels, and 6 (14.3%) had decreased levels. Of those who were retested, elevated PRL levels normalized, but low levels did not (Fig. 3). Abnormal PRL levels did not differ by CPI type or by sex.

Temporal Observations

Time to onset of CPI-hypophysitis varied by type of CPI used (Supplementary Table S2 [20]). The median time to diagnosis for the cohort was 105 days. CTLA-4 inhibitor exposure at

Table 2. Pituitary axes involved and recovery

	Full cohort		PD-1 inhibitor monotherapy at HP Dx		PD-L1 inhibitor monotherapy at HP Dx		CTLA-4 inhibitor combination at HP DX		CTLA-4 inhibitor monotherapy at HP DX		PD-1/PD-L1 inhibitor monotherapy at HP Dx		CTLA-4 inhibitor monotherapy or combination at HP DX		*P* value: CTLA4-inhibitor–exposed vs unexposed
			n = 19		n = 3		n = 19		n = 8		n = 22		n = 27		
	n/n at risk	(%)	n/n at risk	(%)	n/ n at risk	(%)	n/n at risk	(%)	n/n at risk	(%)	n/n at risk	(%)	n/n at risk	(%)	
Central Adrenal Insufficiency	47/47	100.00	19/19	100.00	3/3	100.00	17/17	100.00	8/8	100.00	22/22	100.00	25/25	100.00	1.00
Recovered Adrenal	0	0.00	0	0.00	0	0.00	0	0.00	0	0.00	0	0.00	0	0.00	1.00
Abnormal Thyroid Labs	18/33	54.55	5/14	35.71	0/2		9/12	75.00	4/5	80.00	5/16	31.25	13/17	76.47	**.02**
TFT Changes Consistent with Central Hypothyroidism[a]	8/33	24.24	0/14	0.00	0/2	0.00	6/12	50.00	2/5	40.00	0/16	0.00	8/17	47.06	**.00**
Hypogonadotrophic Hypogonadism	8/35	22.86	2/15	13.33	1/2	50.00	3/13	23.08	2/5	40.00	3/17	17.65	5/18	27.78	.69
Reproductive Hormones Normalized	2/4	50.00	0/1	0.00	1/1	100.00	1/1	100.00	0/1	0.00	1/2	50.00	1/2	50.00	1.00
Abnormal Prolactin at HP diagnosis	11/42	26.19	3/16	18.75	2/3	66.67	3/14	17.65	3/6	50.00	5/19	26.32	6/17	26.09	.63
High Prolactin at HP diagnosis	5/42	11.90	2/16	12.50	2/3	66.67	0/17	0.00	1/6	16.67	4/19	21.05	1/23	4.35	
Low Prolactin at HP diagnosis	6/42	14.29	1/16	6.25	0/3	0.00	3/17	17.65	2/6	33.33	1/19	5.26	5/23	21.74	

Features with a *P*-value less than or equal to 0.05 are noted in bold.
[a]TSH ≤ 0.1 and/or Free T4 ≤ 7.

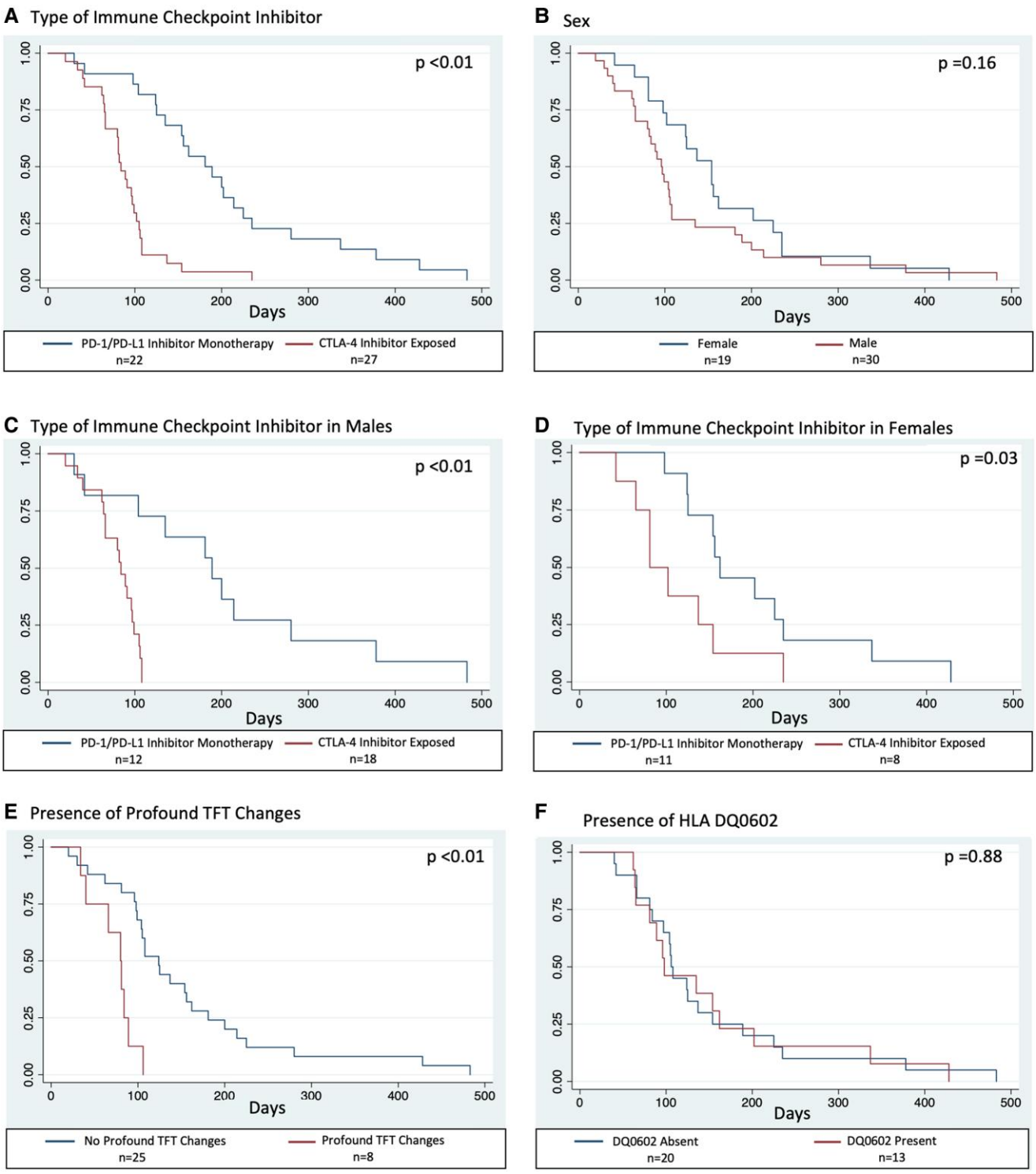

Figure 4. The type of CPI is associated with time to hypophysitis diagnosis. The interval time (days) from CPI initiation to diagnosis of hypophysitis varies based on type of CPI being used (A), type of CPI used in combination with sex (B, C, D) as well as involvement of the HPT axis (E) as represented by Kaplan-Meier Survival Estimates. Time to CPI- hypophysitis diagnosis did not vary by sex without accounting for type of CPI (B). It also did not vary by HLA type DQ0602 (F) by Kaplan Meier Survival Estimates. Abbreviations: HPT, hypothalamic-pituitary-thyroid; TFT, thyroid function tests. Clinically significant TFT changes suggestive of central hypothyroidism: TSH ≤ 0.1 (0.45-4.12 mIU/L) and/or fT4 ≤ 7 (10-18 pmol/L).

CPI-hypophysitis diagnosis was associated with earlier onset of hypophysitis compared to PD1/PD-L1 inhibitor monotherapy [84 days (IQR 65, 105) vs 185 days (IQR 125, 235; $P <$ .01)] (Fig. 4A). Similar trends were seen when looking at individual CPI therapies but with much smaller sample sizes (Supplementary Fig. S2 [20]). There was further variation in the timing of onset for CPI-hypophysitis dependent on the therapeutic target based on sex and CPI type. Men exposed to CTLA-4 inhibitors had the earliest onset of hypophysitis (84 days), followed by women exposed to CTLA-4 inhibitors (91.5 days). In contrast, women who received PD-1/PD-L1 inhibitor therapy had an earlier onset of hypophysitis (162 days)

compared to the men (189 days) (*P* for interaction between CPI type and sex = .068). For men, the hazard ratio between CTLA-4 exposed and unexposed patients was 7.87, while it was only 2.69 in women (Fig. 4B-4D).

Additionally, laboratory test results indicating central hypothyroidism at CPI-hypophysitis diagnosis were also associated with faster time to onset, but this apparent difference was attenuated by adjustment for CTLA-4 inhibitor exposure (Fig. 4E).

Presence of the HLA DQ0602 haplotype, which will be discussed in detail subsequently, was not associated with the time to CPI-hypophysitis diagnosis despite its high prevalence of this haplotype in CPI-hypophysitis patients (Fig. 4F).

Imaging

MRIs of the brain were performed either as part of routine cancer care or for workup of possible CPI-hypophysitis in 42 of the patients. Nineteen patients had an MRI performed in the 40 days prior to diagnosis of CPI-hypophysitis and 2 were abnormal (1 enlarged [moderate-severe] and 1 empty sella [empty]) (Table 3, and Fig. 5, Supplementary Fig. S1 [20]). Within the 40 days before and after diagnosis of CPI-hypophysitis, 27 patients had MRIs, of which 17 were abnormal (15 enlarged [11 moderate-severe, 4 mild] and 2 empty sella [empty]). Of the 21 MRIs performed more than 40 days (as described, based on prior literature [16]) after diagnosis of CPI-hypophysitis, 9 were abnormal (5 enlarged [3 moderate-severe, 2 mild] and 4 empty sella [2 partially empty, 2 empty]).

MRI changes in the pituitary gland were more likely at time of CPI-hypophysitis diagnosis in patients exposed to CTLA-4 inhibitors compared with those on PD-1/PD-L1 inhibitor monotherapy (Table 3). Only 22% of PD-1/PD-L1 inhibitor monotherapy patients had an enlarged gland compared with 72.22% of the CTLA-4 inhibitor–exposed subjects (Fisher exact *P* value .03). Outside of the diagnosis window (40 days before or after clinical determination of CPI-hypophysitis, there was no significant difference in pituitary appearance, even after the additional individual adjustments for age, sex, and thyroid involvement. These findings were maintained with analyses restricted to combination therapy instead of any CTLA-4 inhibitor exposure compared with PD-1 inhibitor monotherapy (data not shown). For those that did have imaging changes, there was no clear difference in the degree of change between CTLA-4 inhibitor–exposed and PD-1/PD-L1 inhibitor monotherapy patients. For patients with pituitary enlargement, there were 2 with mild and 3 with moderate-severe enlargement for PD-1/PD-L1 inhibitor monotherapy patients and 7 with mild and 7 with moderate-severe for CTLA-4 inhibitor–exposed patients. For patients with partially empty or empty sella, there were 2 with partially empty sella and 1 with an empty sella for PD-1/PD-L1 inhibitor monotherapy patients and 4 with an empty sella in CTLA-4 inhibitor–exposed patients. Interestingly, despite the difference in time to hypophysitis onset in men and women, there were no differences in imaging findings. Melanoma diagnosis, abnormal prolactin levels, thyroid hormone abnormalities, and hypogonadism were not associated with enlarged nor abnormal appearing pituitary glands at any time point. More than 40 days after hypophysitis diagnosis, a substantial minority of patients continued to have abnormal appearing pituitary glands; 23.8% had an enlarged gland and 19.0% had an empty or partially empty sella. Among the 5 patients who had persistently enlarged pituitary glands, the range of timing of the follow-up MRIs was 54 to 586 days from hypophysitis diagnosis.

HLA Types

To aid in determining if CPI-hypophysitis is induced by an autoimmune mechanism, we assessed 3 key HLA types that have been related to either spontaneous lymphocytic hypophysitis [13] or were suggested in our own pilot data and have since been seen in small studies of Asian populations [14, 15]: (1) HLA DQ8 (DQB1*03:02); (2) HLA DR53 (DRB4*01:01 or DRB4*01:03); and (3) HLA DQ0602 (DQB1*06:02). Notably, of the 3 HLA types chosen a priori, DQ0602 was the only HLA type significantly associated with an increase in hypophysitis risk relative to both UCSF controls (treated with CPI and healthy) and previously published cohorts (39.4% vs 17.9% vs 14%-21.5%, respectively) (Table 4, [13, 18, 19], and Fig. 6). HLA DR53 was significantly lower in CPI-hypophysitis subjects than in UCSF controls and the Caucasian American population (30.2% vs 55.2% vs 48.1%, respectively) (Table 4 and Fig. 6). Three of the 33 individuals self-reported as Asian, none of whom were DQ0602 positive; therefore 43.3% of the Caucasian subjects with CPI-hypophysitis have DQ0602. One of the 2 subjects of Hispanic ethnicity had HLA typing completed and was DQ0602 negative. There were 3 subjects with CPI-hypophysitis and CPI-diabetes mellitus, of whom 1 was DQ0602 positive. DQ8 had a prevalence in line with the UCSF CPI-treated and healthy, nondiabetic controls, the general population and significantly lower than in sporadic hypophysitis (9.1% vs 22.4% vs 17.5% vs 87%, respectively, Table 4, and Fig. 6). DR53 had a significantly lower prevalence than the UCSF controls, the general population, and sporadic hypophysitis (30.3% vs 55.2% vs 48.1% vs 80%, respectively, Table 4, and Fig. 6). These findings were consistent in the expanded cohort (Supplementary Table S3 [20]). There was no association between the HLA types and type of CPI at diagnosis in the UCSF cohort or the expanded cohort (Table 4, and Fig. 6). There was no association between the prevalence of HLA types within hypophysitis subjects and sex (Fig. 7). However, there was the suggestion of differences in the prevalence of DQ0602 in men and women receiving differing types of CPI. Women on PD-1/PD-L1 inhibitor monotherapy had the highest rates of DQ0602 (66.7%), followed by men exposed to CTLA-4 inhibitors (36.6%), women exposed to CTLA-4 inhibitors (33.33%), and men on PD-1/PD-L1 inhibitor monotherapy (23.08%) (Fig. 7).

Discussion

This is the first study to report on HLA associations with CPI-hypophysitis in Americans. Notably, we report a novel HLA association that is disparate from sporadic lymphocytic hypophysitis. We observed an association between HLA DQ0602 and CPI-hypophysitis, suggesting an autoimmune etiology due to HLA restricted by antigen presentation through this HLA. This HLA association presents a possible alternative mechanistic hypothesis in contrast to the proposed hypothesis that CPI-hypophysitis is due to direct binding of CTLA-4 inhibitors to pituitary cells [21]. Although not statistically significant, there was a trend of HLA DQ0602

Table 3. MRI imaging findings relative to hypophysitis diagnosis and type of immune checkpoint inhibitor

	Prior to HP						Within 40 days of HP diagnosis						Over 40 days after HP diagnosis						Ever					
	Full Cohort		PD-1/PD-L1 Inhibitor Monotherapy at HP Dx		CTLA-4 Inhibitor Monotherapy or Combination at HP DX		Full cohort		PD-1/PD-L1 Inhibitor Monotherapy at HP Dx		CTLA-4 Inhibitor Monotherapy or Combination at HP DX		Full cohort		PD-1/PD-L1 Inhibitor Monotherapy at HP Dx		CTLA-4 Inhibitor Monotherapy or Combination at HP DX		Full cohort		PD-1/PD-L1 Inhibitor Monotherapy at HP Dx		CTLA-4 Inhibitor Monotherapy or Combination at HP DX	
MRI Timing	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**
Empty or Partially Empty Sella	1	5.26	0	0.00	1	8.33	2	7.41	1	11.11	1	5.56	4	19.05	2	15.38	2	25.00	6	14.29	3	15.79	3	13.04
Normal Gland	17	89.47	6	85.71	11	91.67	10	37.04	6	66.67	4	22.22	12	57.14	8	61.54	4	50.00	16	38.10	10	52.63	6	26.09
Enlarged Gland and/or Stalk	1	5.26	1	14.29	0	0.00	15	55.56	2	22.22	13	72.22	5	23.81	3	23.08	2	25.00	21	50.00	6	31.58	15	65.22
Total	19	100	7	100	12	100	27	100	9	100	18	100	21	100	13	100	8	100	42	100	19	100	23	100
Pituitary Gland Not Imaged	23						15						21						7					
Odds of having an enlarged gland in CTLA-4 inhibitor–exposed vs PD-1/PD-L1 monotherapy subjects	Odds Ratio		*P* value		95% CI		Odds Ratio		*P* value		95% CI		Odds Ratio		*P* value		95% CI		Odds Ratio		*P* value		95% CI	
	1.00		NA		NA		**9.10**		**0.02**		1.39,	59.62	1.11		0.92		0.14,	8.68	**4.06**		**0.03**		1.11,	14.8

Features with a *P*-value less than or equal to 0.05 are noted in bold.

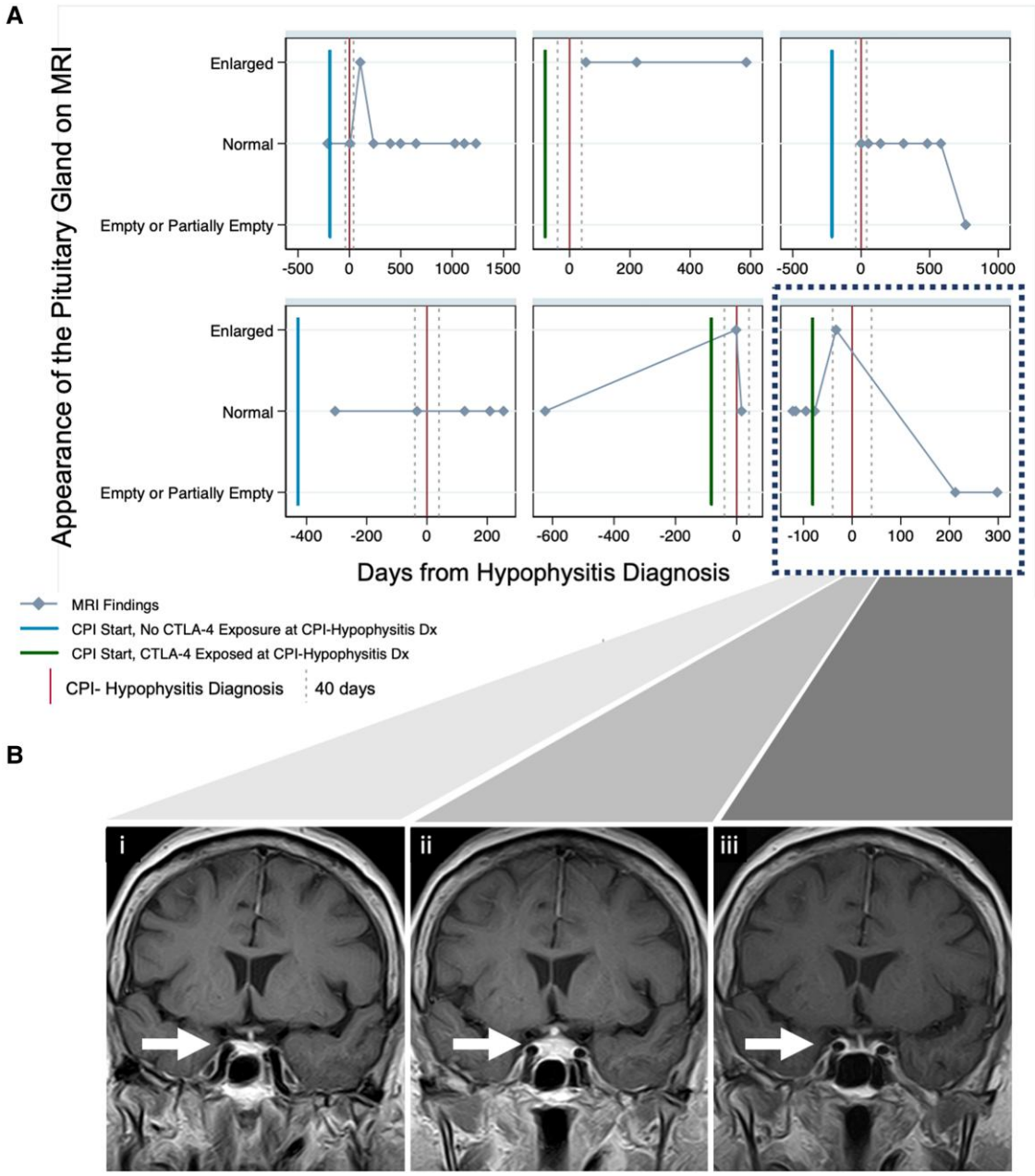

Figure 5. Representative imaging changes seen with immune checkpoint inhibitor–induced hypophysitis. (A) Representative changes in the MRI appearance in a subset of 6 subjects within this cohort relative to CPI initiation and hypophysitis diagnosis. The clinical course in the subject highlighted in (B) is outlined by the dashed line. (B) Serial coronal T1 post-contrast MRIs demonstrate a normal appearing pituitary gland and infundibulum (i), followed by an enlarged pituitary gland and thickened infundibulum (ii), and ultimately a partially empty sella (iii).

haplotype being over-represented in PD-1/PD-L1 inhibitor hypophysitis. These cases also appeared to have a slower onset and lacked imaging findings, particularly within women (Fig. 7). This HLA type is strongly protective against type 1 diabetes and is strongly associated with narcolepsy, occurring in 90% to 100% of patients with cataplectic narcolepsy [22]. It is also in linkage disequilibrium with HLA DR15, which has been reported to be associated with CPI-hypophysitis in 2 smaller Japanese cohorts [14, 15]. In both our primary and expanded cohorts, there was only 1 individual without DR15-DQ0602 linkage. The remaining HLA types reported as significant in these studies, HLAs Cw12, DQ7, DPw9, and B52 were not associated with CPI-hypophysitis in our cohorts relative to the UCSF control group and the Caucasian American population. Of note, these HLA types are relatively enriched in the Japanese population compared with the Caucasian American population. Further work is needed to better understand the impact of this association, particularly the potential differences in sex and CPI type. We hypothesize that the hypothalamic-pituitary-adrenal (HPA) involvement may be caused by an autoimmune response specifically directed at corticotrophs, analogous to the autoimmune injury directed against beta cells while sparing other islet cell populations in CPI-induced diabetes.

We also show differences in clinical presentation based on CTLA-4 and PD-1/PD-L1 inhibitor exposure. Prior studies

Table 4. HLA types in CPI-hypophysitis relative to multiple different control populations

UCSF Cohort																
	Full cohort				PD-1 inhibitor monotherapy at HP Dx		PD-L1 inhibitor monotherapy at HP Dx		CTLA-4 inhibitor combination at HP DX		CTLA-4 inhibitor monotherapy at HP DX		PD-1/PD-L1 inhibitor monotherapy at HP Dx		CTLA-4 inhibitor monotherapy or combination at HP Dx		
	n = 33			*P* value relative to UCSF CPI HP cohort	n = 14		n = 2		n = 11		n = 6		n = 16		n = 17		*P* value within HLA: CTLA4-inhibitor–exposed vs unexposed
Population	n present	n absent	(%)		n present	(%)	n present	(%)	n present	(%)	n present	(%)	n present	(%)	n present	(%)	
HLA DQ0602																	
UCSF CPI HP Cohort	13	20	39.39		5	35.71	2	100.00	4	36.36	2	33.33	7	43.75	6	35.29	.73
UCSF CPI-exposed cancer patients and healthy controls[a]	12	55	17.9	**.03**													
Caucasian Americans [19]			21.5	**.01**													
European Americans [18]			14	**<.0001**													
HLA DQ8																	
UCSF CPI HP Cohort	3	30	9.09		0	0.00	0	0.00	2	18.18	1	16.67	0	0	3	17.65	.23
UCSF CPI-exposed cancer and healthy controls[a]	15	52	22.39	.17													
Caucasian Americans [19]			17.54	.20													
Sporadic HP [13]	13	2	87	.72													
Sellar masses + CPI controls [13]	11	43	20	.23													
HLA DR53																	
UCSF CPI HP cohort	10	23	30.3		3	21.43	1	50.00	3	27.27	3	50.00	4	25.00	6	35.29	.71
UCSF CPI-exposed cancer and healthy controls[a]	37	30	55.22	**.02**													

(*continued*)

Table 4. Continued

UCSF Cohort																			
	Full cohort				PD-1 inhibitor monotherapy at HP Dx		PD-L1 inhibitor monotherapy at HP Dx		CTLA-4 inhibitor combination at HP DX		CTLA-4 inhibitor monotherapy at HP DX		PD-1/PD-L1 inhibitor monotherapy at HP Dx		CTLA-4 inhibitor monotherapy or combination at HP Dx		
	n = 33			*P* value relative to UCSF CPI HP cohort	n = 14		n = 2		n = 11		n = 6		n = 16		n = 17		*P* value within HLA: CTLA4-inhibitor–exposed vs unexposed
Population	n present	n absent	(%)		n present	(%)	n present	(%)	n present	(%)	n present	(%)	n present	(%)	n present	(%)	
Caucasian Americans[19]			48.1	.04													
Sporadic HP [13]	12	3	80	.69													
Sellar masses + CPI controls [13]	26	28	48	.08													

Expanded Cohort**																			
	Full Cohort			*P* value relative to expanded CPI HP cohort	PD-1 inhibitor monotherapy at HP Dx		PD-L1 inhibitor monotherapy at HP Dx		CTLA-4 inhibitor combination at HP DX		CTLA-4 inhibitor monotherapy at HP DX		PD-1/PD-L1 inhibitor monotherapy at HP Dx		CTLA-4 inhibitor monotherapy or combination at HP Dx		*P* value within HLA: CTLA4- inhibitor–exposed vs unexposed
	n = 55				n = 22		n = 3		n = 23		n = 7		n = 25		n = 30			
Population	n present	n absent	(%)		n present	(%)	n present	(%)	n present	(%)	n present	(%)	n present	(%)	n present	(%)		
HLA DQ0602																			
CPI-Hypophysitis Cohort	21	34	38.18		9	40.91	2	66.67	8	34.78	2	28.57	11	45.83	10	33.33	.58		
CPI-Exposed Cancer and Healthy Controls	13	55	19.12	.03															
Caucasian Americans [19]			21.5	<.01															
European Americans [18]			14	<.0001															

(*continued*)

Table 4. Continued

Expanded Cohort**																
	Full Cohort			*P* value relative to expanded CPI HP cohort	PD-1 inhibitor monotherapy at HP Dx		PD-L1 inhibitor monotherapy at HP Dx		CTLA-4 inhibitor combination at HP DX		CTLA-4 inhibitor monotherapy at HP DX		PD-1/PD-L1 inhibitor monotherapy at HP Dx		CTLA-4 inhibitor monotherapy or combination at HP Dx		*P* value within HLA: CTLA4- inhibitor–exposed vs unexposed
	n = 55				n = 22		n = 3		n = 23		n = 7		n = 25		n = 30		
Population	n present	n absent	(%)		n present	(%)	n present	(%)	n present	(%)	n present	(%)	n present	(%)	n present	(%)	
HLA DQ8																	
CPI-Hypophysitis Cohort	7	48	11.11		2	9.09	1	33.33	3	13.04	1	14.29	3	12	4	13.33	1.00
CPI-exposed cancer and healthy controls	15	53	22.06	.24													
Caucasian Americans [19]			17.54	.35													
UCLA CPI-Hypophysitis [13]	0	4	0	1.00													
Sporadic HP [13]	13	2	87	**<.0001**													
Sellar masses + CPI controls [13]	11	43	20	.31													
HLA DR53																	
CPI-Hypophysitis Cohort	14	41	25.45		4	18.18	2	66.67	5	21.74	3	42.86	6	24.00	8	26.67	.82
CPI-exposed cancer and healthy controls	37	31	54.41	**<.01**													
Caucasian Americans [19]			48.1	**<.001**													

(*continued*)

Table 4. Continued

Expanded Cohort**															
	Full Cohort			*P* value relative to expanded CPI HP cohort	PD-1 inhibitor monotherapy at HP Dx		PD-L1 inhibitor monotherapy at HP Dx		CTLA-4 inhibitor combination at HP DX		CTLA-4 inhibitor monotherapy at HP DX		PD-1/PD-L1 inhibitor monotherapy at HP Dx		CTLA-4 inhibitor monotherapy or combination at HP Dx		*P* value within HLA: CTLA4- inhibitor–exposed vs unexposed
	n = 55				n = 22		n = 3		n = 23		n = 7		n = 25		n = 30		
Population	n present	n absent	(%)		n present	(%)	n present	(%)	n present	(%)	n present	(%)	n present	(%)	n present	(%)	
UCLA CPI-Hypophysitis [13]	1	3	25	.74													
Sporadic HP [13]	12	3	80	**<.0001**													
Sellar masses + CPI controls [13]	26	28	48	**.01**													

Features with a *P*-value less than or equal to 0.05 are noted in bold.
[a]CPI-exposed cancer and healthy controls without CPI-induced diabetes and type 1 diabetes.
** the expanded cohort contains all of the UCSF patients described within the primary analysis with additional patients from UCLA, Yale University, and UCSF that we did not include in the primary analysis due to limited availability of data. Again, the CPI-induced diabetes and type 1 diabetes subjects were excluded.

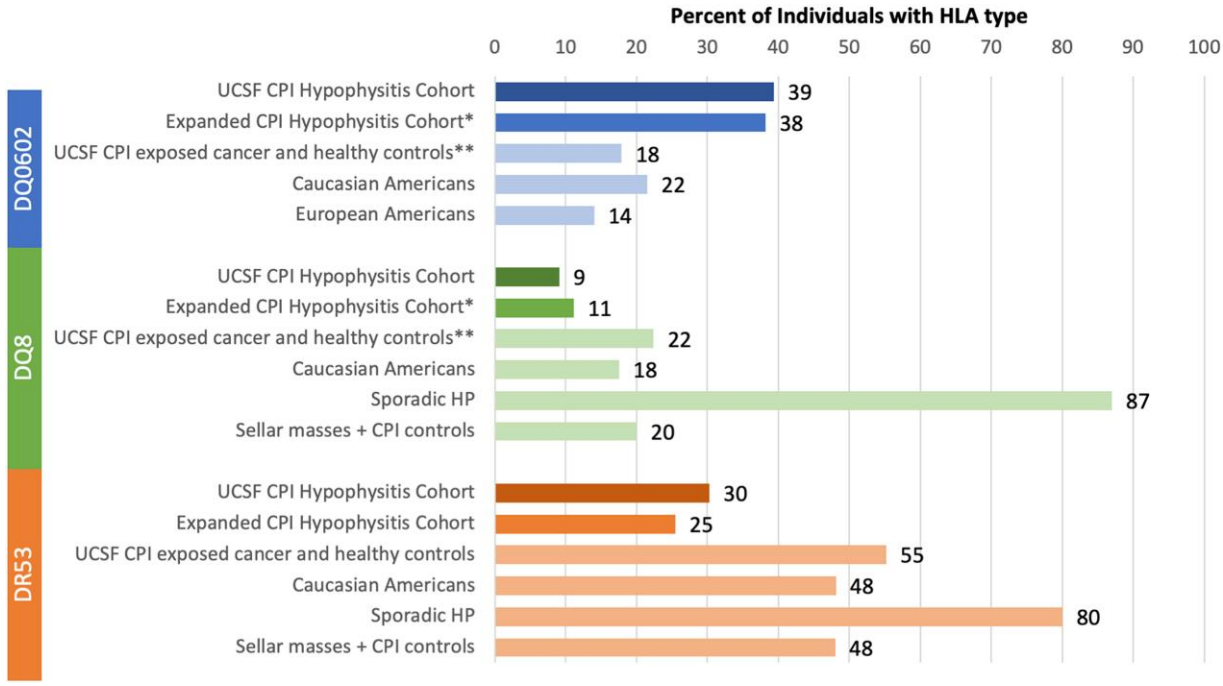

Figure 6. Prevalence of HLA types of interest across multiple cohorts. HLA DQ0602 is over-represented in CPI-hypophysitis patients. HLA DQ8 and DR53, which have previously been associated with sporadic lymphocytic hypophysitis, were not increased in CPI-hypophysitis patients. The expanded cohort contains all of the UCSF patients described within the primary analysis with additional patients from UCLA, Yale University, and UCSF that we did not include in the primary analysis due to limited availability of data. **CPI-exposed cancer and healthy controls without CPI-induced diabetes and type 1 diabetes.

report a wide spectrum of clinical presentations [3, 8, 9, 23, 24]. Many of our findings are consistent with those previously observed: CTLA-4 inhibitor associated hypophysitis is more common [3–5, 24] occurs faster [3, 5, 24], is more likely to involve the hypothalamic-pituitary-thyroid axis (HPT) [3, 5] and induce imaging [3, 4, 6] What has not been reported previously, is a difference in the time of onset between men and women on different types of CPI therapy. While both men and women exposed to CTLA-4 inhibitors developed hypophysitis faster than their counterparts on PD-1/PD-L1 monotherapy, the rates between these 4 groups varied significantly. Prior research has already established that spontaneous lymphocytic hypophysitis has a strong female predominance [10] and given its association with pregnancy [25], it could be related to changes in the immune system or hormonal alterations. Pregnancy is a time of significant change in immune tolerance to allow fetal development [26]. As an immune phenomenon, pregnancy could directly oppose changes in immune tolerance after exposure to CPI, particularly the role of regulatory T cells in pregnancy [27] and regulatory T cell expression of CTLA-4 which highlights the need to consider sex and history of pregnancy in trials. Our findings also show that the disease process may differ between sexes, including a temporal difference not previously seen, but the mechanism will need to be addressed by further studies.

We also observed a prominent evolution of imaging changes through the course of hypophysitis follow-up, particularly during diagnosis, that has not been previously reported. First, the pituitary gland frequently lacked imaging changes at CPI-hypophysitis, especially if due to PD-1/PD-L1 inhibitors. Second, on follow-up imaging over 40 days after CPI-hypophysitis diagnosis, there is substantial heterogeneity of images, with some having persistently enlarged glands and others having empty or partially empty sella. This is in contrast to the prior literature, which was predominantly CTLA-4 inhibitor–exposed patients, in which most had imaging changes at diagnosis that resolved to a normal appearing sella [8, 9, 16].

In contrast with one prior study [24], but in line with others, thyroid recovery [3, 16, 28] was common in our cohort. It has been proposed that in nonthyroidal illness there is a transient central and peripheral adaptive response to acute illness that invariably resolves on recovery from the acute illness [29, 30]. Central hypothyroidism, in contrast, occurs in patients with primary pituitary disorders and may not recover. We observed 8 cases of thyroid hormone abnormalities that were marked and consistent with central hypothyroidism. These cases occurred only in patients who had CPI-hypophysitis after CTLA-4 inhibitor exposure. In 3 cases, levothyroxine treatment was not initiated, and all recovered spontaneously on follow-up testing. This suggests that, unlike the HPA axis deficit, the HPT axis has the potential to recover. If thyroid hormone treatment is initiated acutely, then withdrawal from treatment should be considered at a future date. This is in contrast to spontaneous hypophysitis, in which isolated adrenal insufficiency is uncommon. Also, thyroid hormone abnormalities consistent with central hypothyroidism should lead to evaluation for hypophysitis and HPA axis dysfunction, especially if the patient is receiving a CTLA-4 inhibitor.

Within this study, we have provided a clinical, biochemical, and genetic characterization of hypophysitis. However, limitations do exist, and are in common in the analysis of many irAEs due to the variable nature in their presentation and timing. First, the identification of hypophysitis was made based on clinical presentation and therefore does not include cases that were mild and/or asymptomatic. In fact, we identified

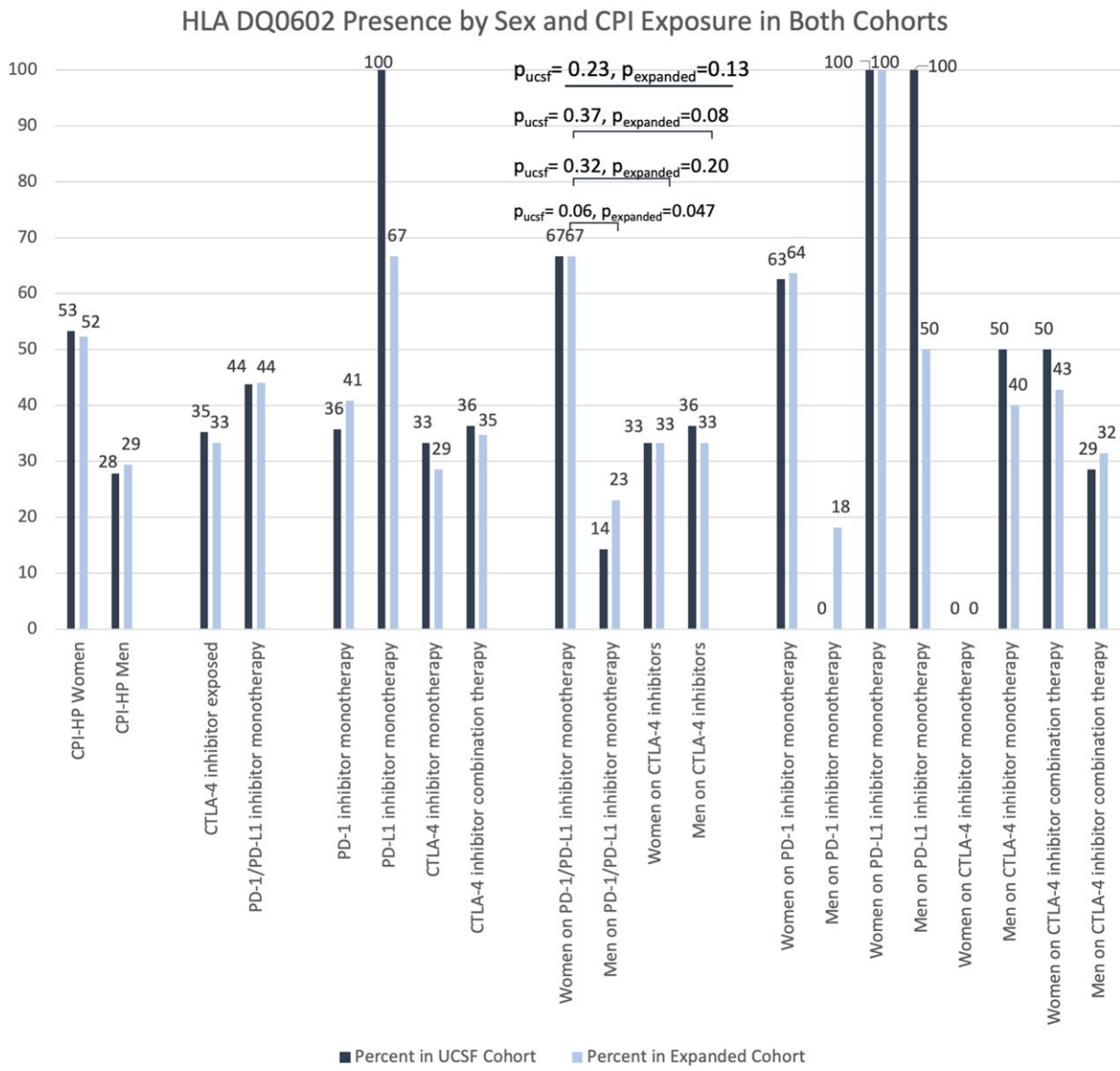

Figure 7. Prevalence of HLA DQ0602 by sex and CPI exposure. Within the cohort detailed in this paper (33 CPI-hypophysitis subjects with HLA types), there was a trend toward a higher prevalence of HLADQ0602 in women on PD-1/PD-L1 inhibitor monotherapy relative to men on PD-1/PD-L1 inhibitor monotherapy using the Fisher exact test. In the expanded cohort that includes samples from Yale and UCLA, the strength of this trend increased (55 CPI-hypophysitis subjects with HLA types).

an additional 4 patients receiving CPI treatment who had incidental findings of hypophysitis on MRI performed for other reasons and who did not have symptoms of hypophysitis nor biochemical changes consistent with hypophysitis. This raises the possibility of a subclinical form of this irAE that may either fail to progress to clinically apparent disease or may recover prior to clinical detection. In addition, MRIs and labs were done as part of clinical care, and therefore did not always contain sufficient testing to definitively confirm the CPI-hypophysitis diagnosis. In fact, 19 cases that were identified with our methods were not included because glucocorticoids were either a long-term medication for another indication or started by oncology prior to testing for adrenal insufficiency given their concern that hypophysitis was present. It is possible that oncologists would be more likely to start glucocorticoids without appropriate testing for more severe cases of CPI-hypophysitis to prevent adrenal crisis. Therefore, as this cohort excluded patients who lacked hormonal axis dysfunction and may not have included severely affected patients who were started on glucocorticoids without adequate testing, this sample may represent a moderate phenotype of CPI-hypophysitis. As described, extensive chart review of all available electronic medical records was done to find all laboratory testing and imaging that was performed. Finally, the poor health status of cancer patients often leads to high rates of hypogonadism and nonthyroidal illness, making it difficult to differentiate from CPI-hypophysitis–induced thyroid and gonadotrophic dysfunction. This highlights the need for greater surveillance and suspicion for CPI-hypophysitis and increased biochemical testing and imaging, when possible, to identify this irAE and appropriately treat it.

Within our HLA analysis, our sample size, while modest, is larger than previously published studies in this area and we

found consistent associations in both our UCSF cohort and our expanded cohort that combined the UCSF cohort with UCLA and Yale cases. We used multiple comparison groups, including a control group with cancer patients and the general population. While there may be differences in prevalence of HLA types in cancer patients compared with controls, there are no known associations between HLA DQ0602 and cancers relevant to this analysis, suggesting that the rates of HLA DQ0602 would likely be similar.

There are multiple strengths to this study. Patients were identified through a systematic approach using multiple methods in a tertiary care center that frequently uses CPI. This approach identified patients with multiple cancers and treatment types not seen in many prior studies. This highlights the consistency between our findings regarding sex, timing, and CPI type despite the variability in patients and suggests that clinical awareness should be heightened based on the CPI type and sex of the patient. Furthermore, these patients were identified as part of routine clinical care, rather than through clinical trials, making the results more generalizable to growing patient population receiving CPIs as standard of care. As described, MRIs, including those performed outside of our system, were reviewed by a neuroradiologist blinded to the time of onset of CPI-hypophysitis, decreasing the chance of bias. Finally, this is the first study to report an HLA association with CPI-hypophysitis.

Prior to the emergence of CPI therapy, hypophysitis was a rare disease. Since the advent of immune checkpoint blockade, the frequency of use has substantially increased, along with morbidity and mortality. Increased clinician awareness within oncology, emergency medicine, inpatient hospital medicine and, of course, endocrinology is essential to provide comprehensive care for these patients. Endocrinologists must closely consider the clinical situation as many of these patients may have suppressed HPA axis from exogenous glucocorticoid exposure for treatment of other irAEs or, at times, as part of their cancer treatment regimen and are at risk for nonthyroidal illness and hypogonadotropic hypogonadism. Further studies assessing the clinical heterogeneity, including on the molecular level such as the exploration of HLA types, should be pursued to improve prediction, diagnosis, and potentially, targeted treatment for CPI-hypophysitis.

The differences in hypophysitis imaging and clinical course observed based on type of CPI used suggest the possibility that there are differing disease mechanisms leading to hypophysitis. It has been reported that there is CTLA-4 expression on pituitary cells [11] and treatment with CTLA-4 inhibitors could directly target pituitary cells via destruction through the classical complement pathway and antibody-dependent cell-mediated cytotoxicity [11]. Expression of CTLA-4 on the pituitary would be unlikely to provoke this same fate with PD-1/PD-L1 inhibitor monotherapy, suggesting the potential for diverse immune-mediated mechanisms for hypophysitis development. Another proposed mechanism is that ectopic expression of ACTH on the tumor leads to the development of ACTH- primed autoantibodies which could explain the isolated adrenal insufficiency that is common to CPI-hypophysitis [31]. Further mechanistic assessment must be performed to determine whether distinct pathways are responsible for the initiation of these diseases due to differential CPI treatment, to assist in both refining clinical surveillance and treatment options.

In summary, this study shows that there is a spectrum of MRI changes as CPI-hypophysitis evolves and suggests that development of CPI-hypophysitis may be different in men and women, specifically in the time to onset and imaging appearance based on type of CPI used. We also report an association between HLA DQ0602 and CPI-hypophysitis. CPI-hypophysitis is a long-term complication from CPI treatment and can be fatal if inadequately treated. Prompt diagnosis and treatment are imperative and understanding these clinical features through parallel demographic and molecular studies will undoubtedly improve patient care.

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Disclosures

The authors have no conflicts of interest to disclose.

Data Availability

The datasets generated during and/or analyzed during the current study are not publicly available but are available from the corresponding author on reasonable request.

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