Immune checkpoints in thymic epithelial tumors: challenges and opportunities

Nicolas Girard
Head of the Thoracic Oncology Program, Institut du Thorax Curie-Montsouris, Institut Curie, Paris, France

ARTICLE INFO
Key words:
Thymoma
Thymic carcinoma
Chemotherapy
Immunotherapy
Autoimmune disorder
Myasthenia

ABSTRACT
Thymic malignancies are rare mediastinal cancers, classified according to the World Health Organization's histopathologic classification which distinguishes thymomas from thymic carcinomas. One key consideration when discussing immunotherapy for thymic epithelial tumors is that one-third of patients diagnosed with thymomas present at the time of diagnosis with autoimmune disorders, the most common being myasthenia gravis. The first step in the understanding of autoimmunity in thymic epithelial tumors is to distinguish true autoimmune disorders from paraneoplastic syndromes; besides pathophysiology, clinical correlates, impact on oncological management and survival may differ strongly. Autoimmune disorders are related to a deregulation in the physiological role of the thymus (i.e. to induce central tolerance to tissue self-antigens) through control of differentiation and subsequent positive and negative selection of immature T cells; from a clinical standpoint, in thymomas, once autoimmune disorders are present, they may not regress significantly after thymectomy. PD-L1 expression, while observed in more than 90% of epithelial cells of the normal thymus with a medullar tropism respecting Hassall's corpuscles, has also been identified in thymomas and thymic carcinomas using various immunohistochemistry protocols. Immune checkpoint inhibitors of the PD-1/PD-L1 axis have been assessed in advanced and metastatic thymic epithelial tumors, mainly thymic carcinomas. Several case reports have been published, and four trials have assessed the efficacy and safety of these inhibitors. Immunotherapy is not standard given the frequent occurrence of severe autoimmune disorders, and clinical trials are ongoing.

Thymic epithelial tumors: current treatment strategies

Thymic malignancies are rare mediastinal cancers, classified according to the World Health Organization's histopathologic classification which distinguishes thymomas from thymic carcinomas [1]; approximately 1500 patients are diagnosed every year in Europe [2]. Thymomas reproduce the architecture of the normal thymus, combining epithelial tumor cells with non-tumoral lymphocytes, and are further subdivided into subtypes (A, AB, B1, B2 and B3) based upon the degree of cell atypia, the relative proportion of the lymphocytic component and the resemblance to normal thymic architecture [1]. Thymic carcinomas are similar to their extrathymic counterparts, with the most common subtype being squamous cell carcinomas that harbor specific immunohistochemical (expression of CD5 and CD117) and molecular features. These differ from thymomas and squamous cell carcinomas originating from the lung or other organs [3,4]. However, neuroendocrine carcinomas may occur in the thymus as primary tumors. The staging of thymic tumors is currently based on the 8th edition of the American Joint Committee on Cancer/Union for International Cancer Control TNM staging classification which integrates macroscopic findings but is only assessed after surgical resection, as the classification of early-stage tumors requires information about pathological invasion of the capsule and the perithymic fat [5].

The management of thymic epithelial tumors requires cooperation between clinicians, surgeons and pathologists from establishing the diagnosis to organizing the multimodal therapeutic strategy [6]. Surgery is the mainstay of curative-intent treatment in limited-stage tumors, as complete resection represents the most favorable prognostic factor for overall survival in both thymomas and thymic carcinomas [7]. Postoperative radiotherapy is debated after complete resection of thymomas, but has been associated with better outcome in thymic carcinomas [8]. Systemic treatment may be delivered in a curative-intent approach for patients presenting with locally advanced tumors at the time of diagnosis, with invasion of intrathoracic neighboring structures and/or dissemination to the pleura and the pericardium, precluding complete resection. In such cases, chemotherapy aims to reduce the tumor burden – possibly
allowing subsequent surgery and/or radiotherapy – to achieve prolonged disease control [10,11].

For the treatment of unresectable, metastatic and recurrent tumors, which are more frequently thymic carcinomas than thymomas, systemic agents are used in a more palliative-intent setting. Historically, several consecutive lines of chemotherapy may be administered when the patient presents with tumor progression [9,10]. Recent real-life evidence provides landmark efficacy data for such strategies [10]. Targeted agents are included in the strategy, mainly consisting of the antiangiogenic agent sunitinib [11] and the MTOR inhibitor everolimus [12]; exceptional cases of thymic carcinoma harbor a druggable oncogenic alteration, mainly described in the KIT and PI3KCA genes [13–15], with opportunities for the use of specific inhibitors in a precision medicine strategy.

This review focuses on the key immunological features of thymic epithelial tumors that are relevant in the clinic for the treatment of patients with regards to the frequent occurrence of autoimmune disorders in conjunction with thymomas, and recent results with immune check-point inhibitors based on potential biomarkers.

### Autoimmune disorders in thymic epithelial tumors

One key consideration when discussing immunotherapy for thymic epithelial tumors is that one-third of patients diagnosed with thymomas present at the time of diagnosis with autoimmune disorders, the most common being myasthenia gravis [6,16–18]. Other frequent disorders include pure red cell aplasia (5% of cases) and hypogammaglobulinemia (5% of cases) (Table 1).

From a practical clinical standpoint, one recommendation of the Clinical Practice Guidelines of the European Society for Medical Oncology when thymic epithelial tumor is suspected is to conduct a systematic immunological check-up (including complete blood cell count with reticulocytes and serum protein electrophoresis, and anti-acetylcholine receptor and antinuclear antibody tests), record a complete history and conduct a full clinical examination (looking particularly at neurological signs) [6]. This is to diagnose the most common autoimmune disorders that may impact any therapeutic intervention, including surgery, radiotherapy and chemotherapy, and potential eligibility for immunotherapy.

Autoimmune disorders are usually observed in thymic carcinomas, and also in combined tumors with a thymoma component. However, pure carcinomas may be associated with true paraneoplastic syndromes related to the direct secretion of cytokines or hormones by tumor cells.

The largest report of the features of paraneoplastic and autoimmune disorders comes from the retrospective cohort of 6670 patients registered in the International Thymic Malignancy Interest Group database [18]. The results of this analysis confirm the previously reported significant association between autoimmune disorders and lymphocytie-rich, type B1 thymomas, early stage of the disease, and complete resection of the tumor; autoimmune disorders are also associated with a tendency towards a better outcome in terms of recurrence and survival.

One issue with all cohort studies is that checks for less common autoimmune disorders may not have been conducted systematically for all patients, especially in the absence of symptoms. Metachronous autoimmune disorders may also be underestimated, given the surgical orientation of the majority of databases with potentially limited follow-up of patients. Meanwhile, differential diagnosis may be challenging with postoperative treatment complications, as chemotherapy may produce cytopenias, and peripheral neurologic or cardiac side-effects. The absence of significant prognostic value of autoimmune and paraneoplastic disorders may suggest association with more aggressive tumors, as suggested by a higher rate of chromosomal alteration reported recently using data from the Cancer Genome Atlas [15]. This may be overcome by favorable clinical factors including: (i) earlier diagnosis of the tumor in the case of baseline autoimmune and paraneoplastic disorders with a higher chance of complete resection; (ii) more aggressive locoregional treatment, including thymectomy, which is known to be effective in myasthenia gravis regardless of the presence of a thymic tumor [19]; and (iii) closer clinical follow-up with potentially earlier diagnosis of recurrences.

| Neuromuscular                  | Myasthenia gravis                          |
|--------------------------------|--------------------------------------------|
|                                | Myotonic dystrophy                         |
|                                | Limbic encephalitis                        |
|                                | Peripheral neuropathy                      |
|                                | Autonomic neuropathy                       |
|                                | Acquired neuromyelitis                     |
|                                | Morvan syndrome (neuromyelitis and encephalitis) |
|                                | Stiff person syndrome                      |
|                                | Cerebellar degeneration                    |
|                                | Polymyositis (carcinomas)                  |
| Collagen and autoimmune disorders | Systemic lupus erythematous                |
|                                | Rheumatoid arthritis                       |
|                                | Sjogren syndrome                           |
|                                | Scleroderma                                |
|                                | Interstitial pneumonitis                   |
| Immune deficiency disorders    | Hypogammaglobulinemia                      |
|                                | (Good syndrome)                            |
|                                | T-cell-deficiency syndrome                 |
| Endocrine disorders            | Multiple endocrine neoplasia               |
|                                | Cushing syndrome                           |
| Dermatologic disorders         | Pemphigus                                  |
|                                | Lichen planus                              |
|                                | Chronic mucosal candidiasis                |
|                                | Alopecia areata                            |
| Miscellaneous                  | Giant cell myocarditis                     |
|                                | Nephrotic syndrome                         |
|                                | Ulcerative colitis                         |
|                                | Hypertrophic osteoarthropathy              |

### Pathogenesis of autoimmunity in thymic epithelial tumors

The first step in the understanding of autoimmunity in thymic epithelial tumors is to distinguish true autoimmune disorders from paraneoplastic syndromes; besides pathophysiology, clinical correlates, impact on oncological management and survival may differ strongly.

Autoimmune disorders are related to a deregulation in the physiological role of the thymus (i.e. to induce central tolerance to tissue self-antigens) through control of differentiation and subsequent positive and negative selection of immature T cells. This phenomenon, while primarily active during childhood, still exists in adults [20]. This process is deregulated along with thymic carcinogenesis, as:

- immature thymoma-derived lymphocytes may escape the disorganized tumor micro-environment, disrupting the journey through the thymic medulla where self-tolerance is primarily induced [21,22];

### Table 1

Autoimmune disorders associated with thymomas

| Neuromuscular                  | Myasthenia gravis |
|--------------------------------|-------------------|
|                                | Myotonic dystrophy|
|                                | Limbic encephalitis|
|                                | Peripheral neuropathy|
|                                | Autonomic neuropathy|
|                                | Acquired neuromyelitis|
|                                | Morvan syndrome (neuromyelitis and encephalitis) |
|                                | Stiff person syndrome|
|                                | Cerebellar degeneration|
|                                | Polymyositis (carcinomas) |
| Collagen and autoimmune disorders | Systemic lupus erythematous |
|                                | Rheumatoid arthritis|
|                                | Sjogren syndrome |
|                                | Scleroderma |
|                                | Interstitial pneumonitis |
| Immune deficiency disorders    | Hypogammaglobulinemia |
|                                | (Good syndrome) |
| Endocrine disorders            | Multiple endocrine neoplasia |
|                                | Cushing syndrome |
| Dermatologic disorders         | Pemphigus |
|                                | Lichen planus |
|                                | Chronic mucosal candidiasis |
|                                | Alopecia areata |
| Miscellaneous                  | Giant cell myocarditis |
|                                | Nephrotic syndrome |
|                                | Ulcerative colitis |
|                                | Hypertrophic osteoarthropathy |
medullary thymic epithelial cells have defects in antigen presentation related to loss of expression of the transcription factor AIRE (autoimmune regulator), similar to that described in autoimmune polyendocrinopathy candidiasis ectodermal dystrophy. AIRE has the unique capability to express all self-tissue-related antigens at the cell surface of thymic epithelial cells of the medulla, and the inactivation of AIRE leads to the absence of expression of some antigens and the release of self-reactive lymphocytes outside the thymus (Figure 1) [23]; and

thymic carcinogenesis may be associated with genetic changes that impair the development of T cells, and generate an increased number of self-reactive lymphocytes [24].

These mechanisms are not exclusive, as highlighted by the fact that AIRE is not inactivated in type B1 thymomas which are more frequently associated with autoimmune disorders [25], and patients can develop an autoimmune disease after thymectomy. The Cancer Genome Atlas analysis reported the overexpression of acetylcholine receptor and other related genes in myasthenia-positive thymomas, suggesting that defective negative T-cell selection is unlikely to be the sole autoimmunizing mechanism [15].

The role of PD-1/PD-L1 interaction in the non-neoplastic thymus is not fully understood; this interaction negatively regulates beta selection and modulates the positive selection [24]. PD-1 is involved in CD8+ T-cell tolerance through peripheral intrinsic mechanisms such as deletion or functional inactivation, and facilitates the peripheral differentiation of CD4+ T cells into regulatory T cells (Treg). Activation of lymphocytes through metabolic pathways is observed in expression profiling of thymomas [4].

From a clinical standpoint, in thymomas, once autoimmune disorders are present, they may not regress significantly after thymectomy.

Paraneoplastic disorders generally result from production of hormones, cytokines or peptides by tumor cells that lead to metabolic derangements; other mechanisms may include cross-reaction between tumor neoantigens and tissue-related antigens, with peripheral induction of autoantibody production stimulated by the presence of the tumor [26]. Thus, successful treatment of the underlying tumor often improves paraneoplastic syndromes, such as in neurological disorders like Lambert-Eaton myasthenia syndrome which is caused by autoantibodies to voltage-gated calcium channels in the presynaptic neuronal cell membrane, and often observed in thymic carcinomas [27]. Differential diagnosis between Lambert-Eaton syndrome and myasthenia gravis may be challenging for thoracic oncologists, especially if autoantibodies are not tested systematically. This may explain the number of patients with thymic carcinomas reported as having ‘myasthenia’.

**Immune-related biomarkers in the thymus and thymic tumors**

PD-L1 expression, while observed in more than 90% of epithelial cells of the normal thymus with a medullar tropism respecting Hassall’s corpuscles [28], has also been identified in thymomas and thymic carcinomas using various immunohistochemistry protocols (Table 2) [29–47]; overall, expression of PD-L1 is common in thymomas and thymic carcinomas, and is usually high and intense. In the micro-environment, a large

---

**Figure 1.** Inhibition of autoimmune regulator leads to release of autoreactive lymphocytes and autoimmune disorders. Adapted from Mathis D, Benoist C. Aire. Annu Rev Immunol 2009; 27:287–312.
Table 2

| Technique, antibody | Thymomas | Thymic carcinoma |
|---------------------|-----------|------------------|
|                     | PD-L1 positive (n, %) | PD-L1 positive (n, %) |
| Katoya et al. [29]  | TMA, clone E1L3 (H-score, 1% of tumor cells cut off) 101 | 22 (23%) |
|                     |                        | 38 | 26 (79%) |
| Padda et al. [30]   | TMA, clone SH1 (intensity high) 65 | 44 (68%) |
|                     |                        | 4 | 3 (75%) |
| Arbour et al. [31]  | Slides, clone E1L3 (25% of tumor cells cut off) 12 | 11 (94%) |
|                     |                        | 11 | 4 (34%) |
| Yokohama et al. [32,33] | Slides, EPR1161 (H-score, 20% of tumor cells cut off) 82 | 44 (54%) |
|                     |                        | 25 | 20 (80%) |
| Weisfieder et al. [34] | Slides, clone E1L3 (% of tumor cells cut off) 74 | 47 (64%) |
|                     |                        | 26 | 14 (54%) |
| Markeski et al. [28] | Slides, clone SP142 (1% of tumor cells cut off) 38 | 35 (92%) |
|                     |                        | 8 | 4 (50%) |
| Wei et al. [35]     | TMA, clone E1L3 (% of cells and intensity) 100 | 100 (100%: 36% low, 64 high) |
|                     |                        | 69 | 69 (100%: 64 low, 36% high) |
| Guleria et al. [36] | TMA, clone SP263 (1–25% of tumor cells cut off) 84 | 69 (82%) |
| Suster et al. [37]  | TMA, clone SP142 (1–50% of tumor cells cut off) 21 | (lymphoepithelioma like histology) 15 |
|                     |                        | (71%: 67% high, 33% low) |
| Tiseo et al. [38]   | TMA, clone E1L3 (H-score, 1% of tumor cells cut off) 87 | 16 (20%) |
|                     |                        | 25 | 13 (52%) |
| Bagir et al. [39]   | Slides, clone AM26531AF-N (intensity) 37 | 21 (57%) |
|                     |                        | 6 | 4 (67%) |
| Sakane et al. [40]  | Slides, clones SP142, SP263, 22C3, Dako 28-8 (50% of tumor cells cut off) 53 | |
|                     |                        | |
| Hakiri et al. [41]  | Slides, clone SP142 (50% of tumor cells cut off) 81 | 22 (27%) |
|                     |                        | 20 | 14 (70%) |
| Chen et al. [42]    | TMA, clone SP142 (% of cells and intensity) 50 | 24 (48%) |
|                     |                        | 20 | 2 (33%) |
| Terra et al. [43]   | Slides, clone SP263 (1–50% of tumor cells cut off) 11 | 10 (91%) |
|                     |                        | 6 | 2 (33%) |
| Owen et al. [44]    | Slides, clone 22C3 (% of tumor cells) 32 | 26 (81%) |
|                     |                        | 3 | 3 (100%) |
| Bedekovics et al. [45] | Slides, clone SP142 (1–50% of tumor cells cut off) 29 | 20 (69%: 70% high, 30% low) |
|                     |                        | 6 | 6 (86%: 17% high, 83% low) |
| Duan et al. [46]    | Slides, clone Ab88810 (% of cells and intensity) 13 | 13 (100%: 46% high, 64% low) |
|                     |                        | 20 | 20 (100%: 65 high, 35% low) |
| Funaki et al. [47]  | Slides, multiple clones (% of cells) 43 | 26 (60%) |

TMA, tissue micro-array.

series of 100 thymomas and 69 thymic carcinomas [35] reported high expression of PD-L1, IDO and FOXP3 Tregs in 36%, 13% and 16% of cases of thymoma, respectively. High expression of PD-L1, IDO and FOXP3 Tregs was associated with a higher grade of tumor histology. In patients with thymic carcinomas, high expression of PD-L1, IDO and FOXP3 Tregs was identified in 36%, 14% and 29% of cases, respectively.

The significance of this finding as a signal of active immune response against thymic epithelial tumor cells and thus a rationale for the assessment of immune checkpoint inhibitors targeting PD-1 or PD-L1 remains debatable, given: (i) the high frequency of PD-1 and PD-L1 expression in the non-neoplastic thymus; (ii) the fact that all data were actually obtained in surgically resected early-stage tumors, and not from specimens of metastatic or advanced tumors that are those potentially eligible to immune checkpoint inhibitors; (iii) in thymomas, the presence of immature and mature T cells surrounding tumor cells is part of the prototypic architecture, and not a marker of actual antitumor response; and (iv) the potential immune modulation induced by chemotherapy, radiotherapy or targeted agents, such as reported with sunitinib, that may lead to occurrence or worsening of autoimmune disorders [11], which is part of the standard treatment strategy in advanced disease, and may lead to modulation of PD-L1 expression in immune cell populations including Tregs.

In a series of 43 patients with thymic carcinomas, PD-L1 increased in patients who received induction chemotherapy, and the change was strongly correlated with epithelial mesenchyma transition status [47]. In another series, the expression of PD-L1 in tumor cells differed between primary and metastatic or recurrent tumors and between metastatic/recurrent tumors at different time points in almost 20% of cases [43].

The tumor mutation burden is emerging as a potential biomarker for immunotherapy in solid epithelial tumors. Data from the Cancer Genome Atlas and Foundation Medicine indicated a low tumor mutation burden in thymomas and thymic carcinomas; only 6% of carcinoma cases had >10 mutations/Mb and 3% had >20 mutations/Mb (Figure 2) [13,15].

Figure 2. Tumor mutation burden in thymic epithelial tumors. Reprinted from Radovich M, Pickering CR, Felau I, et al. The integrated genomic landscape of thymic epithelial tumors. Cancer Cell 2018; 33:244–258; RHAB, Rhabdoid tumors; MED, Unveal melanoma; THYM, Thymic malignancies; LAML, acute myeloid leukemia; NB, Neuroblastoma; CLL, chronic lymphoid leukemia; PRAD, Prostate adenocarcinoma; CARC, adenocortical carcinoma; BRCA, breast invasive carcinoma; MM, Malignant Mesothelioma; OV, Ovarian carcinoma; KIRC, kidney renal papillary cell carcinoma; GBM, Glioblastoma Multiform; UCEC, uterine corpus endometrial carcinoma; CRC, Colorectal cancer; DLBCL, Diffuse Large B Cell Lymphoma; HNSC, Head and Neck Squamous Cell Carcinoma; ESO, esophageal carcinoma; BLCA, bladder urothelial carcinoma; LUAD, Lung Adenocarcinoma; LUSC, Lung Squamous Cell Carcinoma; MEL, melanoma.
Immunotherapy in thymic malignancies

Approximately 20–30% of thymomas and 70–80% of thymic carcinomas present with unresectable, recurrent and/or metastatic disease [6]. Current options include cytotoxic combination regimens, combining platin with anthracyclins, etoposide or taxanes, and targeted agents such as sunitinib; response and survival rates are usually limited, ranging from 20% to 30% and from 6 to 7 months, respectively [10]. Based on results obtained in other solid, refractory tumors in adults, especially of squamous cell differentiation, and possibly considering PD-L1 expression observed in thymic epithelial tumor cells – despite its intrinsic and constitutive relationship with the thymic origin of cancer cells – immune checkpoint inhibitors of the PD-1/PD-L1 axis have been assessed in advanced and metastatic thymic epithelial tumors, mainly thymic carcinomas. Several case reports have been published, and four trials have assessed the efficacy and safety of PD-1/PD-L1 inhibitors in patients with advanced thymic epithelial tumors (Table 3) [48–58].

Clinical trials

The landmark evidence comes from a phase II trial with pembrolizumab, a fully humanized IgG4 antibody that targets the PD-1 receptor, in 40 patients with thymic carcinomas. Forty-eight percent of cases had squamous cell differentiation, and 15% of cases had a neuroendocrine phenotype [28]. The tumors were metastatic in 33 patients (median of two metastatic sites), the median number of previous systemic therapies was two, and 52% and 58% of patients had previously been operated on or had received chest radiotherapy, respectively: this suggests that the patients had been heavily treated, had a relatively limited burden of disease and had a good performance status of 0 or 1 in 95% of cases. Any history of autoimmune disease or other malignancy requiring treatment led to exclusion from this study. Pembrolizumab 200 mg was given every 3 weeks. Of the 40 patients, six (15%) developed serious autoimmune disorders: two cases of polymyositis and myocarditis (complete recovery with steroids but requirement of a pacemaker for complete auriculo-ventricular block – this correlated with the presence of T-cell-receptor clones in muscle that increased in blood with treatment with pembrolizumab); one case of pancreatitis, hepatitis and type 1 diabetes mellitus; one case of bullous pemphigoid, (recovered with steroids); one case of polymyositis and hepatitis; and one case of transaminitis. Three patients had to discontinue treatment after an adverse event. In this trial, the response rate was 23%: there was one complete response, eight partial responses and 21 (53%) patients with stable disease. The median duration of response was 23 months. Median progression-free and overall survival were 4.2 and 24.9 months, respectively. PD-L1 expression – using immunohistochemistry with DAKO 22C3 antibody – was observed in >50% of tumor cells for 10 patients, six of whom had a response to pembrolizumab; only three of the 27 patients with PD-L1 expression by tumor cells <50% had a response. Interferon gamma signature assessed using the Nanostring assay correlated with response to pembrolizumab.

Further analysis was conducted to assess the tumor mutation burden assessed by targeted sequencing as a predictor of response; this was not found to be predictive [55]. Single alterations are being investigated further.

A second trial was conducted in Korea with a similar design [52]. Of 33 patients enrolled, 26 had thymic carcinomas and seven had thymomas (four type B1, one type B2/B3 and one type B3). Three patients had a history of myasthenia gravis (considered to be controlled), 30% of patients were treated beyond third-line treatment, and 36% of patients had a history of mediastinal radiotherapy. With regards to efficacy, two (29%) patients with thymomas had a partial response, and five (72%) patients had stable disease. Of 26 patients with thymic carcinomas, five (19%) had a partial response and 14 (54%) had stable disease. The median progression-free survival was 6.1 months for both groups. Five (71%) of seven patients with thymomas and four (15%) of 26 patients...
with thymic carcinomas reported grade ≥3 immune-related adverse events, including hepatitis, myocardiitis, myasthenia gravis (some patients had pre-existing myasthenia gravis), thyroiditis, antineutrophil cytoplasmic antibody-associated rapidly progressive glomerulonephritis, colitis and subacute myoclonus; treatment of these side-effects was mainly based on steroids and immunoglobulins. Only high PD-L1 expression was predictive of a response. Subsequent analyses showed that the proliferative response of peripheral blood PD-L1+CD8+ T cells, measured as the fold change in the percentage of Ki-67+ cells 7 days after treatment, may be a useful surrogate biomarker for predicting response and prognosis [1].

A phase II trial with nivolumab, an IgG4 antibody that targets the PD-1 receptor, was conducted in Japan for patients with thymic carcinomas [53]; 15 patients were accrued in the first stage, which aimed to identify at least one patient with a response. No response was actually observed. However, 11 patients had stable disease, including five patients for ≥24 weeks. Median progression-free survival was 3.8 months. As the early termination criterion was fulfilled by patients who presented serious toxicities, patient accrual was terminated. As in other trials, patients were heavily pretreated. Only two patients presented with severe autoimmune disorders (elevated transaminases and adrenal insufficiency); it remains unclear whether these data, together with the pembrolizumab data, suggest correlation between efficacy and the occurrence of side-effects.

The fourth trial was a phase I trial with avelumab [54,56], a fully human, IgG1 anti-PD-L1 antibody. This trial reported data on eight patients: seven with thymomas (two type B3, one type B2/B3, two type B2 and one type B1) and one with thymic carcinoma. Two patients with thymomas had a confirmed partial response, two had unconfirmed responses, two (including the patient with thymic carcinoma) had stable disease, and one had progressive disease. Interestingly, three patients responded after a single dose of avelumab. Treatment-related adverse events were immune-related events, including myositis, in five patients, precluding continuation of avelumab. The trial was discontinued. Correlative studies suggested that avelumab induces infiltration of the tumor by macrophages, natural killer cells and activated T lymphocytes, suggesting real induction of antitumor responses, rather than lymphocytic depletion by treatment alone. Subsequent analysis showed that a low B-cell count in blood and pre-existing anticetacholine receptors predicted the development of myositis in these patients; interestingly, patients who developed myositis had no specific antibodies before and after avelumab treatment [57].

Single case reports

Outside of clinical trials, single case observations have been reported: a patient with type B2 thymoma treated with pembrolizumab who experienced severe mucocutaneous toxicity involving skin, mouth, esophagus, uvea and glands [48]; a patient with type B2 thymoma treated with a single dose of nivolumab who developed fatal myositis and myocarditis [58]; four patients who received nivolumab as second- or third-line treatment for PD-L1-positive thymic carcinoma with prolonged response or disease control [50]; and one patient with squamous cell thymic carcinoma who responded to pembrolizumab [49].

Key points for clinical practice

- Immune checkpoint inhibitors may represent a new option of advanced thymic epithelial tumors, but implementation in the clinic is challenging based on the biology of those malignancies.
- Efficacy is actually in line with response and progression free survival reported with other available options in advanced disease [10].
- Toxicity is a major concern, despite systematic baseline work-up for autoimmunity, with frequent occurrence – higher than in other solid tumors – of severe autoimmune adverse events, mainly myositis, myasthenia and hepatitis, possibly favored by previous treatment with anthracyclines and radiation therapy.
- Immunotherapy is contra-indicated in type B1/B2 thymoma, is not a standard-of-care in type B3 thymoma or thymic carcinoma, and should not be delivered in an off-label setting without full disclosure of risks. Clinical trials are ongoing; in Europe, the European Organization for Research and Treatment of Cancer and the European Thoracic Oncology Platform have commenced a single-arm, multicentre, phase II study (the NIVOTHYM trial) to assess the efficacy of nivolumab alone or in combination with ipilimumab in patients with advanced, refractory type B3 thymoma or thymic carcinoma. A strict autoimmune work-up is planned (NCT03134118). A phase I/II trial with pembrolizumab in thymic carcinomas and thymomas is also underway at MD Anderson Cancer Center (NCT03295227).

It remains to be determined whether immunotherapy would provide a better efficacy/safety ratio with selection of patients using biomarkers. PD-L1 expression (negative or positive) may be a predictor, while the tumor mutation burden does not seem to be relevant. Greater completion of autoimmunity checks may be useful, as suggested by post-hoc analyses of clinical trials.

In clinical practice, it is important to ensure that the primary thymic nature of a squamous cell carcinoma is identified accurately in patients who may be misdiagnosed with primary lung squamous cell carcinoma. These patients are eligible for immune checkpoint inhibitors in a routine practice setting.

To conclude, immunotherapy creates new challenges in the field of thymic epithelial tumors. There is a need for multidisciplinary clinical management and research to improve understanding of biological and immune processes regulating the interactions between thymic epithelial cells and T cells in the non-neoplastic and tumoral thymus to prevent autoimmune adverse events of immune checkpoint inhibitors, and to identify biomarkers predicting efficacy and long-term survival.

Funding

None declared.

Disclosure

Dr Girard reports grants, personal fees and non-financial support from Bristol-Myers Squibb; personal fees and non-financial support from Merck Sharp and Dohme; personal fees and non-financial support from Novartis; and grants and personal fees from Pfizer.

References

[1] WHO histological classification of tumours of the thymus. In: Travis WLB, Brambilla A, Burke AP, Marx A, Nicholson A, editors. World Health organization classification of tumours of the lung, pleura, thymus and heart. Lyon: IARC Press; 2015.
[2] Imbimbo M, Maury JM, Garassino M, Girard N. Mesothelioma and thymic tumors: Treatment challenges in (outside) a network setting. Eur J Surg Oncol 2019;45:75-80.
[3] Jha V, Sharma P, Mandal AK. Utility of Cluster of Differentiation 5 and Cluster of Differentiation 117 Immunoprofile in Distinguishing Thymic Carcinoma from Pulmonary Squamous Cell Carcinoma: A Study on 1800 Nonsmall Cell Lung Cancer Cases. Indian J Med Paediatr Oncol 2017;38:430–2.
[4] Girard N, Shen R, Guo T, Zakowski MF, Heguy A, Riely GJ, et al. Comprehensive genomic analysis reveals clinically relevant molecular distinctions between thymic carcinomas and thymomas. Clin Cancer Res 2009;15:6790–9.
[5] Thymic tumors. In: Bierley JD, Gospodarowicz MK, Wittekind C, editors. TNM classification of malignant tumours. 8th Edition. Oxford: Wiley et Sons; 2017.
[6] Girard N, Ruffini E, Marx A, Favier-Finn C, Peters S, ESMO Guidelines Committee. Thymic epithelial tumours: ESMO Clinical Practice Guidelines for diagnosis, treatment and follow-up. Ann Oncol 2015;26(Suppl 5):v40–55.
[7] Hishida T, Nomura S, Yano M, Asamura H, Yamashita M, Ohde Y, et al. Long-term outcome and prognostic factors of surgically treated thymic carcinoma: results of 306 cases from a Japanese Nationwide Database Study. Eur J Cardiothorac Surg 2016;49:835–41.
[8] Girard N, Radiation Therapy in Thymoma. In: Wenz F, editor. Radiation Oncology. Cham: Springer; 2019.

[9] Girard N, Lal R, Death W, Hirdy GJ, Looscher P.J. Chemotherapy definitions and policies for thymic malignancies. J Thorac Oncol 2011;6:5749-55.

[10] Merievskii du Vigneaux C, Dansin E, Mannaia L, Grellier L, Pichon E, Kerjoana M, et al. Systemic Therapy in Advanced Thymic Epithelial Tumors: Insights from the RYTHMIC Prospective Cohort. J Thorac Oncol 2018;13:1762-70.

[11] Thomas A, Rajan A, Berman A, Tomaia Y, Brzezinski C, Lee MJ, et al. Sunsinib in patients with chemotherapy-refractory thymoma and thymic carcinoma: an open-label phase 2 trial. Lancet Oncol 2015;16:e177-86.

[12] Zacca PD, De Paol T, Palomeni G, Favaretto A, Chella A, Tiseo M, et al. Phase II Study of Everolimus in Patients With Thymoma and Thymic Carcinoma Previously Treated With Ciclosporin-Based Chemotherapy. J Clin Oncol 2018;36:342-9.

[13] Ross JS, Vandeven Bore P, Almog N, Schrock AB, Bertolini F, Stefanini A, et al. Analysis of a panel of druggable gene mutations and of ALK and PD-L1 expression in a series of thymic epithelial tumours (TETs). Lung Cancer 2017;104:24–30.

[14] Bagir EK, Aicikalin A, Avci A, Gumurdulu D, Paydas S. PD-1 and PD-L1 expression in thymic epithelial tumors and non-neoplastic thymus. J Clin Pathol 2018;71:637–41.

[15] Watanabe T, Okuda K, Murase T, Moriya S, Haneda H, Kawano O, et al. Four immunohistostains assays to measure the PD-L1 expression in malignant pleural mesothelioma. Oncotarget 2018;9:20769-80.

[16] Hakiri S, Fukui T, Morisawa K, Nakamura S, Ozeki N, et al. Clinicopathologic Features of Thymoma With the Expression of Programmed Death Ligand 1. Ann Thorac Surg 2019;107:418–24.

[17] Chen Y, Zhang Y, Chai X, Gao J, Chen G, Zhang W, et al. Correlation between the Expression of PD-L1 and Clinicopathological Features in Patients With Thymic Epithelial Tumors. Biomed Res Int 2018. 830547.

[18] Terra SSBP, Manfield AS, Vrana JA, Roden AC. Heterogeneity of programmed death-ligand 1 expression in thymic epithelial tumours between initial specimen and synchronous or metachronous metastases or recurrences. Histopathology 2019;74:364–7.

[19] Owen D, Chu B, Lehrman AM, Annamalai L, Yearley JH, Shilo K, et al. Expression Patterns, Prognostic Value, and Intratumoral Heterogeneity of PD-L1 and PD-1 in Thymoma and Thymic Carcinoma. J Thorac Oncol 2018;13:1204–12.

[20] Bedekovics J, Beke L, Mokanatzki A, Saligay S, Mehes G. Programmed Death-ligand 1 (PD-L1) Expression in Thymic Epithelial Tumors. Appl Immunomol Mol Morphol. 2018. https://doi.org/10.1097/PAL.0000000000000699.

[21] Duan J, Liu X, Chen H, Sun Y, Liu Y, Bai H, et al. Impact of PD-L1, transforming growth factor-β expression and tumor-infiltrating CD8+ T cells on clinical outcome of patients with advanced thymic epithelial tumors. Thorac Cancer 2018;9:1341–53.

[22] Funaki S, Shintani Y, Fukui E, Yamamoto Y, Kanazaki R, Ose N, et al. The prognostic impact of programmed cell death 1 and its ligand and the correlation with epithelial-mesenchymal transition in thymic carcinoma. Cancer Med 2019;8:216–26.

[23] Zander T, Aebl S, Rast AC, Zander A, Winterhalder B, Brand C, et al. Response to Pembrolizumab in a Patient with Relapsing Thymoma. J Thorac Oncol 2016;11:e17-9.

[24] Ishikhi T, Isebo K, Tochigi N, Sunakawa M, Nakamura Y, Shibuya K, et al. Successful Use of Pembrolizumab to Treat Refractory Thymic Carcinoma with High PD-L1 Expression. Case Rep Oncol 2018;11:688–92.

[25] Uchida N, Fujita K, Okamura M, Nakatani K, Mi T, et al. The clinical benefits of immune checkpoint inhibitor for thymic carcinomas –experience of single public hospital in Japan. Respir Med Case Rep 2018;26:39–41.

[26] Giaccone G, Kim C, Thompson J, McGuire C, Kallakury B, Chahine JJ, et al. Pembrolizumab in patients with thymic carcinoma: a single-arm, single-centre, phase 2 study. Lancet Oncol 2018;19:247–55.

[27] Cho J, Kim HS, Ku BM, Choi YL, Cristescu R, Han J, et al. Pembrolizumab for Patients With Refractory or Relapsed Thymic Epithelial Tumor: An Open-Label Phase II Trial. J Clin Oncol 2019;37:2162-70. JCO2017733184.

[28] Katsuya S, Horinouchi H, Aso T, Kitahara S, Goto Y, Kanda S, et al. Expression of programmed death 1 (PD-1) and its ligand (PD-L1) in thymic epithelial tumors: impact on treatment efficacy and alteration in expression after chemotherapy. Lung Cancer 2016;99:4–10.

[29] Padda SK, Riess JW, Schwartz EJ, Tian L, Kohrt HE, Neal JW, et al. Diffuse high intensity PD-L1 staining in thymic epithelial tumors. J Thorac Oncol 2015;10:500–4.

[30] Sanders DB, Guptill JT. Myasthenia gravis and Lambert-Eaton myasthenic syndrome. Continuum 2014;20:1413–25.

[31] Marchevsky AM, Walts AE. PD-L1, PD-1, CD4, and CD8 expression in neoplastic and lymphoepithelioma-like carcinoma of the thymus. Mod Pathol 2018;31:1801–6.

[32] Suster D, Fihan G, Mackinnon AC, Suster S. Expression of PD-L1/PD-1 in lymphoepithelioma-like carcinoma of the thymus. Mod Pathol 2018;31:1801–6.

[33] Tiseo M, Damato A, Longo I, Barbieri F, Bertolini F, Stefanini A, et al. Analysis of a panel of druggable gene mutations and of ALK and PD-L1 expression in a series of thymic epithelial tumours (TETs). Lung Cancer 2017;104:24–30.

[34] Rajan A, Heery CR, Mammen AL, Pittaluga S, Lepone ML, Donahue RN, et al. B004. Prevalence of autoimmune diseases in thymic epithelial tumors insights from RYTHMIC. Mediastinum 2017;4. https://doi.org/10.21037/med.2017.AB004.

[35] Lee HS, Truong CY, Burt BM. Mutation Ensemble for Response to Programmed Cell Death 1 (PD-1) Blockade: PRIMER study. Eur J Cancer 2019;113:78e86.

[36] Rajan A, Heery CR, Mammen AL, Pittaluga S, Lepone ML, Donahue RN, et al. Systemic Therapy in Advanced Thymic Epithelial Tumors: Insights from the RYTHMIC Prospective Cohort. J Thorac Oncol 2018;13:1762-70.

[37] Suster S, Fihan G, Mackinnon AC, Suster S. Expression of PD-L1/PD-1 in lymphoepithelioma-like carcinoma of the thymus. Mod Pathol 2018;31:1801–6.

[38] Tiseo M, Damato A, Longo I, Barbieri F, Bertolini F, Stefanini A, et al. Analysis of a panel of druggable gene mutations and of ALK and PD-L1 expression in a series of thymic epithelial tumours (TETs). Lung Cancer 2017;104:24–30.