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Medical treatment involving investigational drugs and genetic profile of thymic carcinoma

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ABSTRACT

Background: Thymic carcinoma is a rare neoplasm of the thymus, and information regarding its genetic profile and optimal medical treatment is limited. We sought to characterize the genetic profile of thymic carcinoma and to evaluate the efficacy of various medical treatments, including treatment with tyrosine kinase inhibitors (TKIs), cytotoxic agents, and immune checkpoint inhibitors.
Methods: We retrospectively reviewed medical records of 64 consecutive patients with thymic carcinoma at the National Cancer Center Hospital between April 1973 and March 2014. We analyzed treatment course of patients who underwent medical treatment involving investigational drugs. For patients with available tissue samples, targeted sequencing of 50 cancer-related genes using next-generation sequencing was performed.
Results: Thirty-six patients had received chemotherapy. Median progression-free survival in patients receiving first-line chemotherapy was 7.07 months (95% confidence interval, 5.67–8.93). Median survival time was 32.6 months (95% confidence interval, 23.2–43.4). As second- or later-line chemotherapy, a total of 13 patients were treated with 24 investigational drugs, including 8 multi-targeted TKIs, 5 cytotoxic agents, and 2 immune checkpoint inhibitors. Six (24%) of the patients treated with investigational drugs maintained disease control for at least 6 months. Tissue samples of 52 patients (81.3%) were available for targeted sequencing, consisting of 52 formalin-fixed, paraffin-embedded (FFPE) and 16 fresh frozen tissue samples. The genetic alterations of *TP53*, *KRAS*, *FBXW7*, and *NRAS* were detected in 7 patients (13.5%), and no *KIT* mutations were noted.
Conclusions: Multi-targeted TKIs exhibited potential clinical efficacy for previously-treated thymic carcinoma. The frequency of genetic alterations in this study was low, with no apparent relationship with the efficacy of chemotherapy.

1. Introduction

Thymic carcinoma is a rare neoplasm with a poor prognosis. Thymic epithelial tumors, including thymoma and thymic carcinoma, account for about 20% of mediastinal tumors, with an incidence of approximately 0.15 per 100,000 person-years in the United States [1]. Thymic carcinoma accounts for approximately 10–15% of all thymic epithelial tumors and is distinguished from thymoma by its malignant nature and poor prognosis [2–4]. Five-year survival rates are reported to be 80% for stage I/II, 63% for stage III, 42% for stage IVa, and 30% for stage IVb [4].

Data regarding the clinical course and prognosis of thymic carcinoma are limited, because of its relatively low incidence. Chemotherapy regimens including platinum or anthracyclines are

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widely utilized based on several phase II trials for treatment-naïve advanced or recurrent thymic carcinoma, although no randomized controlled trials have been conducted [5–9]. However, clinical evidence supporting second- or later line chemotherapy is much more limited. There are some promising reports of multi-targeted tyrosine kinase inhibitors (TKIs) including vascular endothelial growth factor receptor (VEGFR) and stem cell factor receptor (KIT) [10,11].

While recent progress in cancer genome research has revealed relationships between tumor genetic alterations and clinical outcome, information regarding the genetic profile of thymic carcinoma is still limited [12–14]. Here, we retrospectively evaluated the genetic profiles of thymic carcinoma patients treated at our institution and assessed the therapeutic responses to chemotherapy, including experimental regimens.

2. Patients and methods

2.1. Patients

Sixty-four consecutive patients with thymic carcinoma treated at the National Cancer Hospital between April 1973 and March 2014 were included in this study. Clinical information was collected and analyzed. For patients whose tissue samples were available, targeted next-generation sequencing of tumor tissue was performed. This study was approved by the institutional review board at the National Cancer Center Hospital.

2.2. Clinical data

Age at diagnosis, sex, medical history, concomitant diseases, smoking status, histology, diagnostic methodology, treatment modality, and response to treatment were obtained from medical records. Diagnosis was made from tissue samples obtained via computed tomography (CT)-guided needle biopsy, endobronchial ultrasound-guided transbronchial needle aspiration biopsy, transbronchial lung biopsy for lung metastases, or surgically resected specimens. Clinical staging was reevaluated per the Masaoka staging system using CT, magnetic resonance imaging, positron-emission tomography, and histology [16]. For patients who had received chemotherapy, we recorded the treatment regimen, the best overall response as determined by the investigator in the case of experimental therapy, and dates of the treatment initiation and conclusion.

2.3. DNA preparation

Freshly frozen and formalin-fixed, paraffin embedded (FFPE) tissue samples were used in this study. Genomic DNA was extracted from freshly frozen tissues using a kit (QIAamp DNA mini Kit®; QIAGEN, Hilden Germany) and from FFPE tissues using a dFFPE kit (QIAamp DNA FFPE Tissue Kit®; QIAGEN). DNA was quantified via fluorometry (Qubit® Fluorometer; Thermo Fisher Scientific, Waltham MA, USA).

2.4. Targeted deep sequencing of mutation hot spots in 50 cancer-related genes

We used 10-ng DNA samples for library preparation (Ion Ampliseq Cancer Hotspot Panel v2®; Thermo Fisher Scientific), which enables detection of mutations at 2790 hot spots in 50 cancer-related genes (Table 1) [17]. The library DNA samples were prepared by amplifying targeted regions using multiple polymerase chain reactions (PCRs) followed by adopter DNA ligation. Concentrations of library DNA were evaluated by quantitative real-time PCR (Ion Library Quantitation Kit®; Thermo Fisher Scientific) and sequencing (Ion Proton platform®; Thermo Fisher Scientific). Variations in allele frequency <4% were excluded in this analysis.

Table 1
List of cancer-related genes examined in this study.

ABL1	*EGFR*	*GNA11*	*KRAS*	*PTPN11*
AKT	*ERBB2*	*GNAQ*	*MET*	*RB1*
ALK	*ERBB4*	*HNF1A*	*MLH1*	*RET*
APC	*EZH2*	*HRAS*	*MPL*	*SMAD4*
ATM	*FBXW7*	*IDH1*	*NOTCH1*	*SMARCB1*
BRAF	*FGFR1*	*IDH2*	*NPM1*	*SMO*
CDH1	*FGFR2*	*JAK2*	*NRAS*	*SRC*
CDKN2A	*FGFR3*	*JAK3*	*PDGFRA*	*STK11*
CSF1R	*FLT3*	*KDR*	*PIK3CA*	*TP53*
CTNNB1	*GNAS*	*KIT*	*PTEN*	*VHL*

2.5. Statistical analysis

Time to treatment failure (TTF) was defined as the time from initiation of each treatment to discontinuation for any reason, including disease progression, treatment toxicity, patient preference, or death. Progression-free survival (PFS) was defined as the time from the date of initiation of each treatment to that of disease progression or death from any cause with censoring of patients who were lost to follow-up. Overall survival (OS) was defined as the duration from the date of diagnosis to that of death from any cause, with censoring of patients who were lost to follow-up. Survival curves were calculated using the Kaplan–Meier method. All statistical analyses were performed using commercial software (STATA version 13.1®; StataCorp, College Station TX, USA).

3. Results

3.1. Patient characteristics

Sixty-four consecutive patients with thymic carcinoma were treated at National Cancer Hospital between April 1973 and March 2014. Patient characteristics are summarized in Table 2. The median age was 57 years (range, 22–77), and 38 patients (59.3%) were male. The most common histology was squamous cell carcinoma (47 patients, 73.4%), followed by poorly differentiated carcinoma (7 patients, 10.9%), and thymic carcinoma, not otherwise specified (NOS) (7 patients, 10.9%). The most common clinical stages were stage III (18 patients, 28.1%), IVa (18 patients, 28.1%), and IVb (20 patients; 31.3%). Three patients were stage I, and five were stage II. Current and former smokers accounted for about half of all patients. Almost all patients (96.8%) were assigned a performance score (PS) of 0–1. The clinical characteristics of the 36 patients who received chemotherapy are also shown in Table 2, with characteristics similar to those of the entire group.

3.2. First- and second-line chemotherapy

Clinical data were accessible for 35 out of 36 patients receiving chemotherapy from 1978 to 2014. Carboplatin and paclitaxel were administrated as first-line treatment in 24 patients (66.7%), followed by other platinum regimens (11 patients, 30.6%), and non-platinum regimens (1 patient, 2.8%). Only two patients had received regimens containing anthracyclines. The median PFS and median survival time (MST) in patients undergoing first-line chemotherapy were 7.07 months (95% confidence interval [CI], 5.67–8.93 months) and 32.6 months (95% CI, 23.2–43.6 months), respectively (Fig. 1). The response rate (RR) and disease control rate (DCR) of carboplatin and paclitaxel were 27.8% (95% CI, 12.3–48.6%) and 86.1% (95% CI, 73.1–100%), respectively. Median PFS and MST with first-line car-