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Original Article

Combined treatment with anlotinib and chemotherapy for advanced esophageal squamous cell carcinoma improved patient survival: a case report

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Abstract: Esophageal squamous cell carcinoma (ESCC) is the predominant type of esophageal cancer in Eastern Asia. Historically, advanced ESCC treatments have had low efficacy and new treatments, including immunotherapy or combination therapies, are emerging. Here, we report a special case of recurrent ESCC after surgery. The patient had a failed immunotherapy course, but benefited from anlotinib combined with chemotherapy for a fourth-line therapy. Survival after the combined therapy was greater than 19 months, and the overall patient survival was greater than 32 months.

Keywords: Esophageal squamous cell carcinoma, anlotinib, chemotherapy

Introduction

Esophageal cancer is one of the most common cancers worldwide [1]. Notably, the prevalence of the different histological subtypes is geographically distinct. In Eastern Asia and Sub-Saharan Africa, esophageal squamous cell carcinoma (ESCC) is the predominant histological subtype and comprises more than 90% of all esophageal cancer cases [1, 2]. ESCC is one of the most aggressive squamous cell carcinomas and does not have standard multidisciplinary comprehensive treatments. Radical surgery, systemic chemotherapy, and radiotherapy are often administered in clinical practice. Although radical surgical resections are recommended as the primary treatment for early esophageal cancers, disease recurrence often develops within a few years [3]. Lymph node recurrence and lung and liver oligo-metastasis are the most common post-operation recurrences [4]. Patients almost always relapse within 1-3 years, and the overall 5-year survival is less than 30% [5, 6].

In the past decade, numerous phase II/III clinical trials with targeted therapies for esophageal cancer have reported inspiring survival benefits. Both immune checkpoint inhibitors (ICIs) and small molecule multi-target tyrosine kinase inhibitors (TKIs) have been evaluated in ESCC clinical trials [7-10]. In addition, PD-1/PD-L1 inhibitors, including nivolumab, pembrolizumab, durvalumab, and SHR-1316 have also been tested in ESCC patients [7]. For example, a male Chinese patient with a post-surgical ESCC recurrence, 5% PD-L1 expression in the tumor cells, and a low tumor mutational burden (TMB) benefited from a combined treatment with nivolumab plus anlotinib [11].

Anlotinib is an oral small molecule inhibitor of multi-receptor tyrosine kinases (RTKs) that inhibits tumor angiogenesis and proliferative signaling [12]. Anlotinib targets include VEGFR1/2/3, FDFR1/2/3, c-Kit, and PDGFR α/β. The National Medical Products Administration (NMPA) China Food and Drug Administration (CFDA) approved single agent anlotinib for third-line treatment in patients with locally advanced or metastatic non-small cell lung carcinoma (NSCLC). Additionally, promising clinical trials with anlotinib in soft tissue sarcoma, advanced renal cell carcinoma, and colorectal cancer

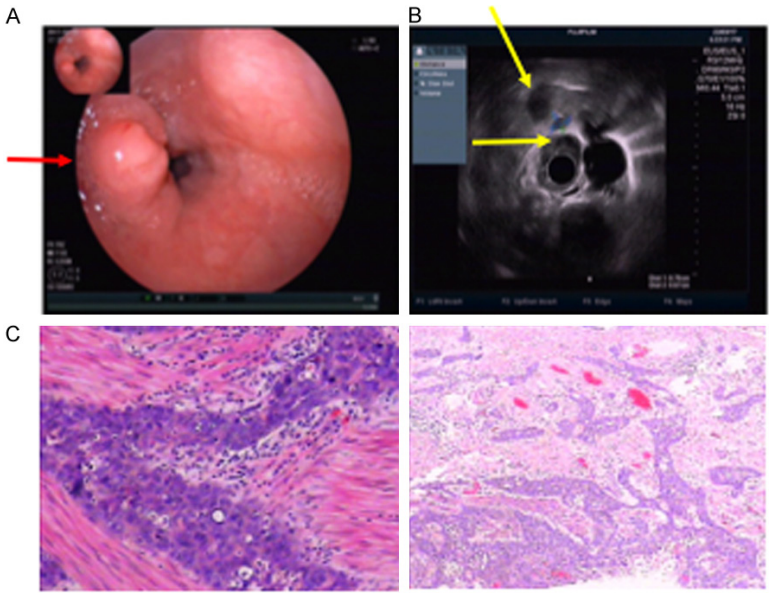

Figure 1. Clinical ESCC diagnosis. A. Endoscopic ultrasound revealed an ulcerated neoplasm in the esophagus that was 26-35 cm from the incisors. The normal esophageal wall hierarchy had disappeared and there was a low and non-uniform echo change. The tumor was approximately 2.7 × 1.5 cm, and the local adventitia was disrupted. Red arrow: Esophageal neoplasm. B. Endoscopic ultrasound depicting the swollen lymph node outside the esophageal wall. Yellow arrow: Swollen lymph node. C. Hematoxylin and eosin (H&E)-stained post-operative pathological tissue. The staining reveals poorly differentiated esophageal squamous cell carcinoma with a maximum diameter of approximately 2.5 cm. The lymph node had 1/21 metastatic cancer cells. Post-operative pathology indicated the patient was pT2N1, stage IIIA.

patients are ongoing [12, 13]. Case reports demonstrate anlotinib efficacy for treating primary osteosarcoma, recurrent glioblastoma, carcinoma ex pleomorphic adenoma, and esophageal squamous cell cancer [11, 14-16].

Here, we report a special case of ESCC in a patient where immunotherapy failed, but anlotinib plus chemotherapy for fourth-line therapy resulted in a 19-month survival.

Case presentation

A 57-year-old man had right chest pain after eating that persisted for a month. Endoscopic ultrasound revealed a neoplasm in the esophagus that was 26-35 cm from the incisors and a swollen lymph node outside the esophageal wall. Pathology in March 2017 indicated that the neoplasm was ESCC (**Figure 1**). The clinical stage of the disease at the time of diagnosis was stage III with lymphatic metastasis. The patient was in a good state of health and had an Eastern Cooperative Oncology Group Performance Status (ECOG PS) score of 1. The patient was administered docetaxel (120 mg) and lobaplatin (50 mg) as neoadjuvant therapy for 2 weeks before undergoing an esophagectomy via video-assisted thoracoscopic surgery (VATS) in May 2017. Cancerous tissue was confirmed to be poorly differentiated squamous cell carcinoma. The maximum tumor diameter was 2.5 cm and it had infiltrated underlying muscle. No additional treatment was given after the surgery.

Unfortunately, 10 months after surgery, multiple lung and lymphatic metastases were found on a PET/CT scan in March 2018 (**Figure 2A**). The patient was then treated with docetaxel (120 mg) and lobaplatin (50 mg) for first-line therapy. After six treatment cycles, the CT scan showed the lung metastases had grown and there was a new liver metastasis (**Figures 2B** and **3A**). Biopsy of the lesion indicated negative PD-1 expression, 20% PD-L1 expression, 50% ki-67 expression, VEGF expression, and EGFR amplification. Next-generation sequencing that targeted 1021 tumor-related genes in cell-free tumor DNA (ctDNA) was also completed. Although no tumor driven mutations were found, the patient had a high TMB (TMB=10), which is displayed in **Table 1** and **Figure 4A**. Based on the clinical remission rate of immune therapy in advanced esophageal cancer, for the second-line therapy the doctor prescribed the patient sequential treatment of nivolumab (200 mg) plus capecitabine (1500 mg) once every 3 weeks (Q3W). Two months later in October 2018 the patient developed multiple low-density lung nodules (**Figure 3B**). The doctor then decided to use irinotecan instead of capecitabine for the third-line treatment. The patient was given nivolumab (200 mg) and irinotecan (300 mg) as a sequential treatment once every 2 weeks (Q2W). A CT after 4 treatment cycles in December 2019 showed that the number of low-density lung nodules had increased and

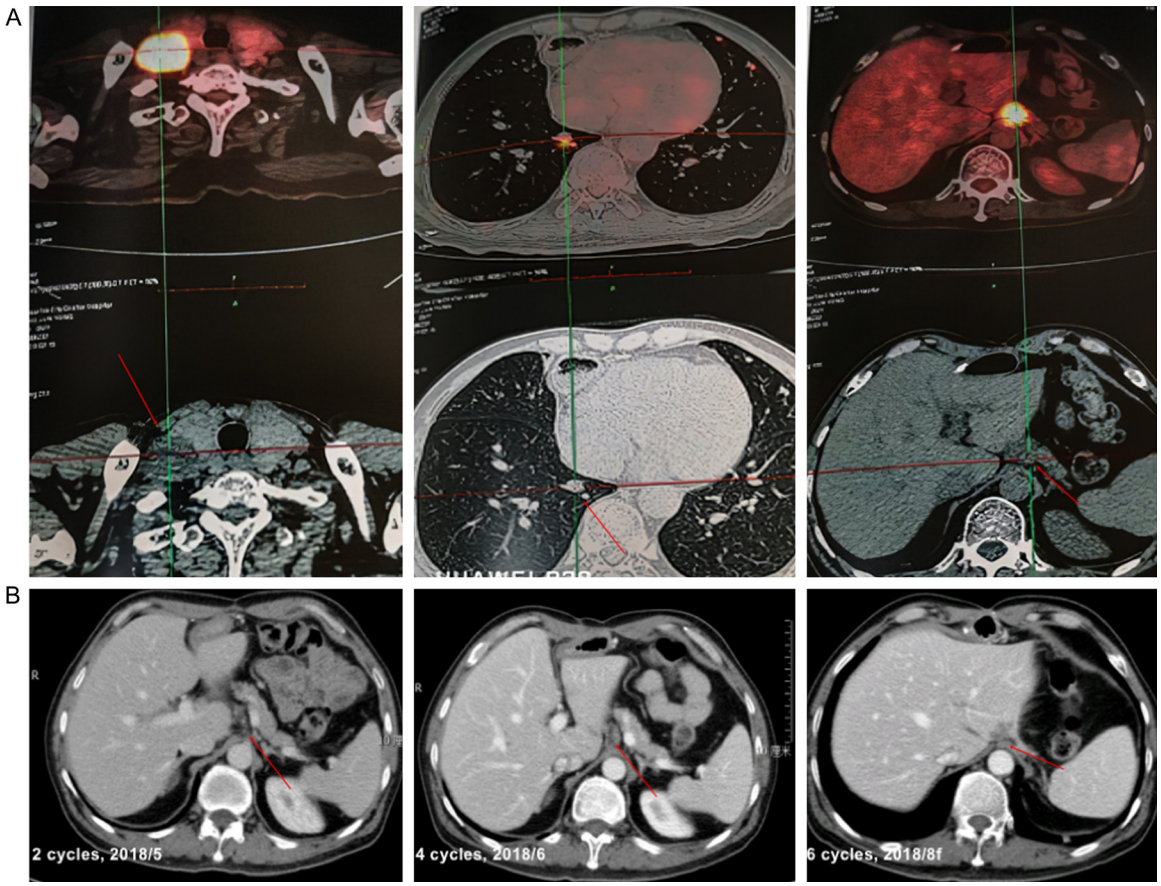

Figure 2. ESCC recurrence was confirmed by PET/CT. A. There were multiple metabolic nodes in the right interlobar, right hilar, mediastinum, peritoneal trunk, right supraclavicular fossa, and right deep neck group 10 months after surgery. Multiple lung and lymphatic metastases were considered. The patient had progressive disease and remote metastasis. Red arrow: Metabolic nodes. B. The patient received 6 cycles docetaxel and lobaplatin chemotherapy. The follow-up CT scan indicated enlarged lung metastases and a new liver metastasis. Red arrow: New liver metastasis.

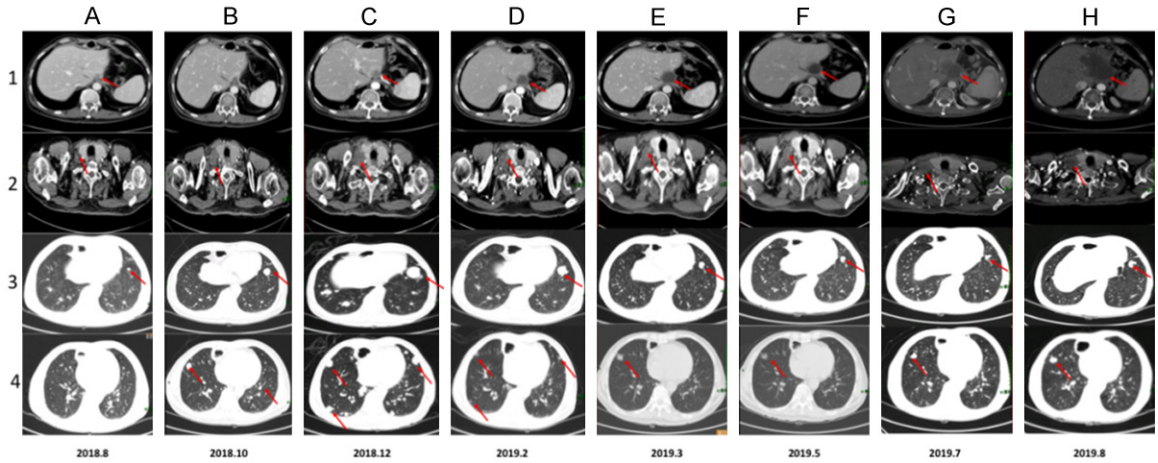

Figure 3. Second- to fifth-line therapies were monitored with CT scans. The second- to fifth-line therapy for the patient. Liver (1), lymph node (2), and lung (3 and 4) metastases are shown. A-C. After the patient developed a new liver metastasis, he received 2 lines of immunotherapy combined with chemotherapy. The disease quickly progressed, including the appearance of low-density lung nodules and a swollen neck lymph node. D-F. The patient received anlotinib combined with chemotherapy for the fourth-line therapy, and the lesions shrank. G, H. Eventually, the disease progressed to enlarged liver lesions. The patient received anlotinib and calorizumab for maintenance therapy.

Table 1. Mutations and clusters

Gene	cHGVS	pHGVS	Cluster	VAF	
				2018.8.28	2018.12.28
GNAS	c.1019T>G	p.F340C	1	9.90%	42.8%%
PDGFRA	c.1319C>T	p.T440M	2	5.40%	23.70%
DDR2	c.313C>T	p.R105C	3	4.30%	18.20%
MLL2	c.4138T>G	p.C1380G	3	3.80%	20.20%
HDAC9	c.3131T>C	p.V1044A	4	1.30%	2.90%
TP53	c.517G>T	p.V173L	5	1.30%	ND
DNMT3A	c.662T>C	p.I221T	5	1.00%	ND
MLL2	c.3496A>G	p.M1166V	5	0.80%	ND
ACIN1	c.608_610+13d elCCAGTACG TATCTCCC	-	5	0.70%	ND
ZFHX3	c.2185G>T	p.E729*	5	0.50%	ND
ETV5	c.1516G>A	p.E506K	6	ND	1.40%
CD274	c.209G>C	p.G70A	6	ND	1.30%
ARAP3	c.1537G>A	p.D513N	6	ND	1.30%
POLE	c.2246G>A	p.R749Q	6	ND	1.20%
GABRA6	c.1301T>A	p.L434H	6	ND	1.10%
FLT4	c.3092G>A	p.R1031Q	6	ND	0.80%
CDH23	c.3574G>C	p.V1192L	6	ND	0.80%
KIT	c.1942G>T	p.G648C	6	ND	0.70%
NOTCH1	c.4906G>A	p.E1636K	6	ND	0.60%
NOTCH3	c.1603G>A	p.E535K	6	ND	0.60%
CDH23	c.9502C>T	p.R3168C	6	ND	0.50%
RNASEL	c.484G>T	p.E162*	6	ND	0.50%
AR	c.967G>A	p.E323K	6	ND	0.50%
FLT1	c.2890G>A	p.E964K	6	ND	0.50%
EP300	c.5485C>T	p.R1829C	6	ND	0.50%

*protein translation is terminated.

that there were swollen neck and supraclavicular lymph nodes (**Figure 3C**). By the end of December, the amount of ctDNA had sharply increased, which indicated the disease had progressed (PD). The overall tumor clone load and the TMB (TMB=20) also increased (**Table 1** and **Figure 4B**). Unfortunately, the tumor cells were not sensitive to immunotherapy.

Based on a phase II clinical study with anlotinib (ALTER1102) where anlotinib significantly prolonged the median progression-free survival (PFS) (anlotinib: 3.0 months vs. placebo: 1.4 months) of Chinese ESCC patients, the doctor prescribed an anlotinib trial. The patient was given nedaplatin (110 mg) and raltitrexed (4 mg) plus anlotinib (12 mg, D1-D14, Q3W) for the fourth-line therapy until the disease progressed to enlarged liver lesions (**Figure 3D-G**). The patient received liver interventional treatment in July 2019 and was then administrated calorizumab (200 mg) plus anlotinib (12 mg, D1-D14, Q3W) as a maintenance treatment. A chest CT after 1 month of treatment revealed that the disease had progressed (**Figure 3H**). The ECOG PS score was 2-3. After this point, the patient was administered anlotinib and calorizumab and did not receive any other treatments. Anlotinib resulted in a better patient response for the ESCC fourth-line and follow-up maintenance therapies. At the time of publication, the patient is still alive and the overall survival time is greater than 32 months.

Discussion and conclusion

The patient experienced loco-regional, distant, or combined recurrences five times after VATS surgery. Aggressive multidisciplinary treatment including surgery and chemotherapy or immunotherapy for multiple recurrences successfully controlled the cancer, and the patient is still

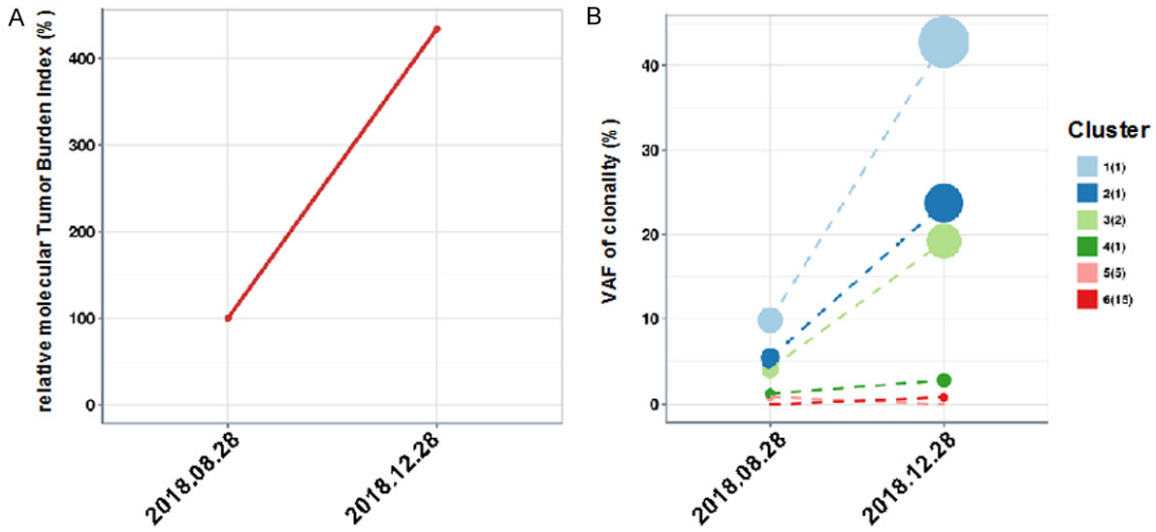

Figure 4. Gene test and clonality. Patient samples were analyzed with next-generation sequencing that targeted 1021 tumor-related genes in ctDNA. (A) The molecular tumor burden index (mTBI, mTBI=ctDNA/cfDNA) sharply increased in the second gene test. (B) All mutations came from six different clusters. The variant allele fraction (VAF) of every cluster also sharply increased in the second gene test. The results from (A and B) demonstrate that the disease progressed within this time period.

alive. Although the patient expressed PD-L1 and had a high TMB, he did not benefit from nivolumab treatment. This result reaffirms that not all the high TMB or PD-L1 expressing patients are eligible for immunotherapy. Loss of PTEN, JAK1, JAK2, and B2M function are known to influence immunotherapy efficacy in melanocytoma and lymphoma [17-19]. Two NGS sequencing efforts during immunotherapy did not detect these gene mutations in the patient. Different gene mutations may have contributed to patient response and are worthy of further study.

Anlotinib is a new, orally administered tyrosine kinase inhibitor that targets the vascular endothelial growth factor receptor (VEGFR), fibroblast growth factor receptor (FGFR), platelet-derived growth factor receptors (PDGFRs), and c-kit. Anlotinib has confirmed efficacy in advanced non-small cell lung cancer, soft tissue sarcomas, metastatic renal cell carcinoma, and advanced medullary thyroid cancer [12]. In China, anlotinib is currently in a phase II clinical study (ALTER1102) for ESCC treatment. Patients who received anlotinib exhibited a better mPFS than patients who received a placebo of the ongoing clinical trial in China. In the case presented here, although the patient was not benefited from chemotherapy or immunotherapy, the patients was benefited from the anlotinib plus chemotherapy, and the patient exhibited 6 months of partial remission. The available drugs to treat advanced ESCC are very limited. Here, the efficacy of anlotinib to treat ESCC was verified, and the results suggest a potential treatment option for patients with advanced or refractory ESCC.

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Disclosure of conflict of interest

None.

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