**BRAF V600E-mutated combined large cell neuroendocrine carcinoma and adenocarcinoma responding to targeted therapy**

Tomohiro Sakamoto, Katsunori Arai, Karen Makishima, Akira Yamasaki

**SUMMARY**

We present a case of combined large cell neuroendocrine carcinoma (LCNEC), harbouring a **BRAF V600E** mutation, which significantly benefited from **BRAF**-targeted therapy. A 57-year-old woman was referred to our hospital for headache and vomiting. A head MRI showed a large tumour in her brain, and a whole-body CT revealed a tumour in the hilum of the right lung and mediastinal lymphadenopathies. Both the resected brain tumour and the mediastinal lymph node tissue contained LCNEC. Next-generation sequencing revealed a **BRAF V600E** mutation, and a combination therapy with dabrafenib and trametinib was initiated. The patient had a good response to treatment. Like non–small cell lung cancer patients, LCNEC patients should undergo multiplex somatic mutation testing.

**BACKGROUND**

Lung cancer is still the leading cause of cancer-related deaths worldwide. The treatment strategy in non–small cell lung cancer (NSCLC) is subdivided based on multiple driver oncogenes. However, the treatment for neuroendocrine lung carcinoma, particularly large cell neuroendocrine carcinoma (LCNEC), has not improved. Due to the low frequency of LCNEC, its genetic status and the significance of somatic mutation testing in daily practice are unclear.

In 2013, the nationwide Lung Cancer Genomic Screening Project for Individualized Medicine in Japan (LC-SCRUM Japan) was conducted to develop molecular-targeted therapies for NSCLC patients with rare genes. Since 2019, the OncoMine Dx Target test multi-CDx system has been used in daily practice for multiplex gene testing of advanced lung cancer in Japan. As such, lung carcinoma patients were likely to undergo somatic mutation testing.

We report a case of LCNEC, harbouring a **BRAF V600E** mutation, that responded to dabrafenib and trametinib (DT). The case demonstrated the importance of somatic mutation testing in LCNEC.

**CASE PRESENTATION**

A 57-year-old woman with a history of well-controlled bronchial asthma and smoking (25 pack-years) was referred to our hospital for headache and vomiting. One month prior to the consult, the patient noted the headache, but this was ignored. However, her symptoms did not improve spontaneously, and she was admitted to a nearby hospital after 2 weeks. A head MRI showed a large tumour in her brain, and she was referred to our neurosurgery department. On examination, she was afebrile and normotensive with normal oxygen saturation. She was noted to lose her balance occasionally while walking, but her neurological exam was unremarkable.

**INVESTIGATIONS**

The whole-body CT revealed a tumour in the right pulmonary hilum and mediastinal lymphadenopathies (figure 1A). The head MRI revealed a 45 mm cystic mass in her right cerebellum (figure 1B). The tumour was resected to control the symptoms associated with the brain tumour. The postoperative course was uneventful.

The pathological specimens of the cerebral tumour contained a mixture of adenocarcinoma-like
BRAF also suggested that both adenocarcinoma and LCNEC cells had prognosis. The mutation allele frequency (MAF) of TP53 H179 is known as a missense mutation that worsens V600E and TP53 H179Q mutation. In comprehensive Assay, Thermo Fisher Scientific), the tumour had two mutations, BRAF V600E and TP53 H179Q, in addition, the next-generation sequencing (NGS) data showed BRAF V600E mutation. In the analysis using this sample detected a BRAF V600E mutation. To decide the treatment strategy, it is necessary to clarify if the BRAF mutations are present in both LCNEC and lung adenocarcinoma. In other words, if BRAF mutation-positive lung adenocarcinoma and non-BRAF-mutated LCNEC are mixed together, BRAF-targeted therapy will have limited therapeutic effect. Therefore, it was thought that immunostaining might be useful as a means of confirming that. Immunostaining of BRAF V600E (anti-BRAF V600E rabbit monoclonal antibody clones: RM8, RevMAb Biosciences) with cerebellar tumours stain both adenocarcinoma and LCNEC cells likely harboured BRAF V600E mutations. In addition, the next-generation sequencing (NGS) data also suggested that both adenocarcinoma and LCNEC cells had BRAF mutations. Based on the NGS results (Oncomine Comprehensive Assay, Thermo Fisher Scientific), the tumour had two major somatic mutations, BRAF V600E and TP53 H179Q mutation. TP53 H179 is known as a missense mutation that worsens prognosis. The mutation allele frequency (MAF) of BRAF was 0.483 and that of TP53 was 0.916. The MAF values showed that this mutation in TP53 caused mutations in both alleles of DNA. On the other hand, BRAF V600E mutation is known to cause deletion of one allele, loss of heterozygosity, and the BRAF MAF value close to 0.5 indicated that most of the cells had the BRAF V600E mutation. Based on these, we concluded that most of the tumours were cells likely to be BRAF-dependent and that targeted therapy would be effective.

TREATMENT
DT combination therapy was started for BRAF-mutated NSCLC. Fever and rash appeared on the fourth day after treatment. Anti-histamine and topical steroids were given, but the symptoms did not improve. Furthermore, DT was temporarily suspended because of hepatic dysfunction on the sixth day. On the ninth day after DT withdrawal, adverse events improved to grade 1 or lower, and dose-reduced DT administration was resumed. On the fifth day after the resumption of administration, fever and rash reappeared, so DT was discontinued and prednisolone (PSL) (20 mg/day) was given. After that, DT was resumed, and the patient did not develop a fever or rash. PSL was gradually reduced to 7.5 mg/day.

OUTCOME AND FOLLOW-UP
The CT results 10 weeks after the start of DT revealed a decrease in the size of the tumour in the right lung hilum. This corresponded to a partial response (figure 3A). The brain tumour was surgically removed. There was no recurrence for at least a year thereafter, including in the central nervous system (figure 3B). She continues to work while receiving DT.

DISCUSSION
LCNEC is a tumour with a poor prognosis comparable with that of small cell carcinoma, with a 5-year survival rate of 40.3% for all pathological stages. According to Yamazaki et al, the response rate to cisplatin-based chemotherapy was 50%, and that of small cell carcinoma showed that the response rate to cisplatin-based chemotherapy was 50%, similar to that of small cell carcinoma. Furthermore, Fujiwara et al reported a response rate of 55.6% for cisplatin plus irinotecan. In general, lung cancer with more than one histological type has a high biological grade. However, the prognosis of combined LCNEC is reported to be similar to that of LCNEC.

The treatment strategy in NSCLC, especially in lung adenocarcinoma, was subdivided based on driver oncogenes, such as EGFR, ALK, ROS1, BRAF, NTRK, MET, and RET. Various clinical practice guidelines recommend multiple somatic mutation tests to select the initial treatment for NSCLC. However, little is known about driver genes in LCNEC. Lou et al analysed 108 cases of pulmonary neuroendocrine tumours, including LCNEC, and reported that no BRAF mutations were found. In contrast, Zakka et al analysed the cell-free tumor DNA of 320 cases of neuroendocrine carcinoma, including 70 cases of pulmonary neuroendocrine carcinoma, and found BRAF mutations in 28 cases. Furthermore, Chae et al analysed 300 cases of LCNEC and reported that 13 cases had BRAF mutations. Targeted therapy was effective in a case with a non-V600E mutation.

In clinical practice, LCNEC is often treated with cytotoxic drugs similar to those of small cell lung cancer. However, it may also be treated as NSCLC. Platinum-based chemotherapy for LCNEC was effective but insufficient. Thus, the development of additional treatments is expected. A clinical trial of DT in patients with previously treated BRAF V600E mutant metastatic NSCLC showed an overall response rate of 63.2% in a cohort of 57 patients. Furthermore, DT in patients with previously untreated BRAF V600E mutant metastatic NSCLC had an overall response rate of 64% in a cohort of 36 patients.
patient underwent routine multiplex gene analysis as NSCLC prior to initial treatment. In this case, multiplex somatic mutation testing allowed the patient to receive BRAF-targeted therapy, which resulted in improvement. This encounter showed that LCNEC should also be treated based on the results of multiplex somatic mutation testing.

Learning points

► This is a case of combined large cell neuroendocrine carcinoma in which dabrafenib and trametinib therapy was effective.
► Multiplex somatic mutation testing prior to initial treatment provided one more option before cytotoxic anticancer drugs.
► Patients with large cell neuroendocrine carcinoma should be given the same opportunity to undergo multiplex somatic mutation testing as patients with non–small cell lung carcinoma.

Case report

Figure 3  (A) A CT 10 weeks after the start of dabrafenib and trametinib therapy confirms a shrinkage of the tumour, which corresponds to a partial response. (B) An MRI showed no recurrence after surgical removal at diagnosis.

REFERENCES

1. Olivier M, Langerød A, Carrieri P, et al. The clinical value of somatic TP53 gene mutations in 1,794 patients with breast cancer. Clin Cancer Res 2006;12:1157–67.
2. Asamura H, Kameya T, Matsumo Y, et al. Neuroendocrine neoplasms of the lung: a proportionate clinical spectrum. J Clin Oncol 2006;24:70–6.
3. Yamazaki S, Sekine I, Matsumo Y, et al. Clinical responses of large cell neuroendocrine carcinoma of the lung to cisplatin-based chemotherapy. Lung Cancer 2005;49:217–23.
4. Fujiwara Y, Sekine I, Tsuta K, et al. Effect of platinum combined with irinotecan or paclitaxel against large cell neuroendocrine carcinoma of the lung. Jpn J Clin Oncol 2007;37:482–6.
5. Ruffini E, Rena O, Olario A, et al. Lung tumors with mixed histologic pattern. Clinico-pathologic characteristics and prognostic significance. Eur J Cardiothorac Surg 2002;22:701–7.
6. Battafarano RJ, Fernandez FG, Ritter J, et al. Large cell neuroendocrine carcinoma: an aggressive form of non-small cell lung cancer. J Thorac Cardiovasc Surg 2005;130:166–72.
7. Soria J-C, Ohe Y, Vansteenkiste J, et al. Osimertinib in untreated EGFR-mutated advanced non-small-cell lung cancer. N Engl J Med 2018;378:113–25.
8. Hida T, Nokihara H, Kondo M, et al. Alectinib versus crizotinib in patients with ALK-positive non-small-cell lung cancer (J-ALEX): an open-label randomised phase 3 trial. Lancet 2017;390:29–39.
9. Shaw AE, Ou SH, Bang Y-J, et al. Crizotinib in ROS1-rearranged non-small-cell lung cancer. N Engl J Med 2014;371:1963–71.
10. Planchard D, Smit EF, Groen HJM, et al. Dabrafenib plus trametinib in patients with previously untreated BRAFmutant metastatic non-small-cell lung cancer: an open-label, phase 2 trial. Lancet Oncol 2017;18:1307–16.
11. Doebele RC, Drilon A, Paz-Ares L, et al. Entrectinib in patients with advanced or metastatic NTRK fusion-positive solid tumours: integrated analysis of three phase 1-2 trials. Lancet Oncol 2020;21:271–82.
12. Paik PK, Felip E, Veillon R, et al. Tepotinib in Non-Small-Cell Lung Cancer with MET Exon 14 Skipping Mutations. N Engl J Med 2020;383:931–43.
13. Drilon A, Oxnard GR, Tan DSW, et al. Efficacy of Selpercatinib in RET Fusion-Positive Non-Small-Cell Lung Cancer. N Engl J Med 2020;383:813–24.
14. NCCN. NCCN clinical practice guidelines in oncology. Non-Small Cell Lung Cancer Ver 2020.3.
15. Akamatsu H, Ninomiya K, Kenmotsu H, et al. The Japanese lung cancer Society guideline for non-small cell lung cancer, stage IV. Int J Clin Oncol 2019;24:731–70.
16. Lou G, Yu X, Song Z. Molecular profiling and survival of completely resected primary pulmonary neuroendocrine carcinoma. Clin Lung Cancer 2017;18:e197–201.
17. Zakka K, Nagy R, Drubosky L, et al. Blood-Based next-generation sequencing analysis of neuroendocrine neoplasms. Oncotarget 2020;11:1749–57.
18. Planchard D, Besse B, Groen HIM, et al. Dabrafenib plus trametinib in patients with previously treated BRAF(V600E)-mutant metastatic non-small cell lung cancer: an open-label, multicentre phase 2 trial. Lancet Oncol 2016;17:984–93.
