Dear Editor,

Multiple endocrine neoplasias (MENs) are genetic syndromes distinguished by specific patterns of benign and malignant tumors of endocrine glands. MEN2 is caused by autosomal dominant inheritance of a gain-of-function mutation in the RET proto-oncogene on chromosome 10 (Mulligan & Ponder 1995, Santoro et al. 1995) and is further subclassified into three syndromes based on clinical phenotype: MEN2A (medullary thyroid cancer (MTC), pheochromocytoma, primary parathyroid hyperplasia); MEN2B (MTC and pheochromocytoma), and familial medullary thyroid cancer (MTC only). MEN is first suspected when an index patient presents with one or more tumors specific to that syndrome. Of all MEN2B cases, 95% are represented by a germline point mutation affecting codon 918 (exon 16) resulting in a methionine to threonine alteration (M918T ), inducing constitutive activation of the intracellular tyrosine kinase domain of the encoded RET receptor tyrosine kinase (RTK) (Alberti et al. 2003). Tumor formation occurs in the neuroendocrine organs where constitutively activated RET is expressed. Although neuroendocrine carcinoma (NEC) of the lung has been described in MEN 1 (Farhandi et al. 1987), it has not been described in the MEN2 syndromes.

Treatment of MEN-associated tumors primarily involves surgical excision of the neoplasm. In MEN2, prophylactic thyroidectomy is recommended for family members who carry the germline mutation, ideally within the first year of life in patients with MEN2B due to the high penetrance, morbidity, and mortality of MTC associated with the M918T mutation (Skinner et al. 2005). Standard cytotoxic chemotherapy and radiotherapy are not effective in MTC (Orlandi et al. 2001); however, novel targeted therapies have been developed that inhibit the activated RET RTK, including cabozantinib (Elisei et al. 2013).

In 1994, a 40-year-old healthy, nonsmoking, Caucasian male experienced a hypertensive crisis secondary to adrenal storm during an elective hernia repair. The patient underwent bilateral adrenalectomies with pathology confirming pheochromocytoma. Germ-line genetic testing revealed the codon M918T point mutation in the RET proto-oncogene, consistent with a diagnosis of MEN2B. The patient underwent thyroidectomy in 1995; pathology demonstrated multicentric C cell hyperplasia and MTC. Additional clinicopathologic details, including peroperative calcitonin, capsular invasion, and lymph node status are unavailable due to the time interval. Although presenting at an unusually late age, the patient demonstrated other classic manifestations of MEN2B, including submucosal neuromas, Marfanoid habitus, and chronic constipation and megacolon for which they ultimately required colectomy in the context of clostridium difficile superinfection. Following thyroidectomy, annual surveillance consisted of thyroid ultrasound demonstrating no locoregional recurrence and serial calcitonin, carcinoembryonic antigen (CEA), and plasma metanephrine levels. From 2008 to 2012, the serum calcitonin ranged from 11 to 131 pg/ml (normal <10 pg/ml). CEA ranged from 0.6 to 6 ng/ml (normal 0–5 ng/ml) until March 2012, when it increased to 28.4 ng/ml. Plasma metanephrines remained normal.

In 2012, the patient developed various sites of musculoskeletal pain. A chest X-ray to evaluate rib pain incidentally revealed a density in the right middle lobe. A subsequent computed tomography (CT) scan of the chest displayed a 2.0 cm nodule in the right middle lobe associated with complete obstruction and atelectasis, suspicious for a primary lung neoplasm, as well as bulky right-sided hilar and mediastinal lymphadenopathy (Fig. 1A and B). Endobronchial biopsy was consistent with a primary neuroendocrine neoplasm of the lung, either large cell NEC or atypical carcinoid (Fig. 2). Immunohistochemical staining (IHC) was positive for
cytokeratin 7, TTF-1, synaptophysin, and Cam 5.2, consistent with primary pulmonary origin and arguing against adrenal origin. Although the patient’s tumor was not stained for CEA, staining for calcitonin and PAX-8 were both negative, the combination of which is a better marker than CEA of tissue originating from medullary thyroid cells. Furthermore, staining for CEA is not likely to be helpful in distinguishing NEC of the lung from MTC, as most neuroendocrine tumors stain for CEA (Travis et al. 1991). By 2012, the patient’s 1994–1995 surgical specimens had been destroyed, thus the lung tumor could not be compared to his prior MEN neoplasias. Prior to treatment, the patient’s serum CEA was elevated at 147.6 ng/ml, while serum calcitonin and plasma metanephrines were normal. Although the differential diagnosis included dedifferentiated MTC, primary NEC of the lung was strongly favored on the basis of dual negativity for calcitonin and PAX-8, serum tumor marker pattern, and intrathoracic radiologic pattern (Fig. 1B; solitary lung mass with bulky ipsilateral adenopathy). Staging revealed extensive osseous metastatic disease, no evidence of brain metastases, and rapid intrathoracic progression. Tumor tissue was sent for molecular testing including EGFR, ALK, KRAS, BRAF, and PIK3CA mutations per institutional standard for non-small cell lung cancer. When the first tier lung cancer panel was negative, extended mutation testing was performed by next-generation sequencing, encompassing over 700 hotspot mutations in 46 key cancer genes. This revealed only the patient’s germline RET mutation. No further testing on the tissue could be performed as the sample was exhausted.

The patient initiated first-line cytotoxic chemotheraphy with carboplatin and etoposide, a standard palliative regimen for large cell lung NEC, with best radiologic response of stable disease (Fig. 1C) and a stable

Figure 1
Serial computed tomography (CT) imaging to assess response to therapy. (A) Right middle lobe nodule with ipsilateral mediastinal lymphadenopathy, atelectasis, and bronchial obstruction prior to systemic therapy. (B) Solitary lung mass with ipsilateral hilar and mediastinal lymphadenopathy prior to systemic therapy. (C) Stable disease after four cycles of carboplatin and etoposide. (D) Significant response to therapy after 2 months of treatment with RET inhibitor cabozantinib.

Figure 2
Morphologic and immunohistochemical features of the right middle lobe mass, transbronchial biopsy. (A) Strands and solid sheets of neoplastic cells infiltrating around mucoserous glands of the bronchial wall; routine hematoxylin and eosin (H&E) staining, 100×. (B) Neoplastic cells show crush artifact, high nuclear/cytoplasmic ratio, and hyperchromatic nuclei; H&E, 600×. (C) Neuroendocrine differentiation is confirmed by positive immunohistochemical stain for synaptophysin; immunohistochemistry, 200×. (D) Positive TTF-1 immunostaining but negative PAX-8 and calcitonin immunostaining (not shown) argue in favor of primary pulmonary neuroendocrine carcinoma; immunohistochemistry, 200×.
CEA of 149.9 after four cycles. Following a 2-month treatment holiday, a CT scan revealed pulmonary disease progression, and the CEA increased to 257.2 ng/ml. Second-line therapy was initiated with cabozantinib, a small molecule inhibitor of the RTKs RET, c-MET, and VEGFR2 that improved progression-free survival (PFS) in patients with metastatic MTC during a randomized, placebo-controlled phase III trial (Elisei et al. 2013). The patient started cabozantinib at 100 mg daily and did not tolerate dose increases due to chronic ileostomy-related diarrhea, fatigue, and oral pain. Two months later, a chest CT revealed a marked response in the patient’s pulmonary disease burden (Fig. 1D), and the CEA decreased to 78.4 ng/ml. The patient’s performance status and symptoms of cough and skeletal pain improved. The pulmonary radiologic response persisted through the 18-month duration of cabozantinib treatment; however, the biochemical response persisted for only 12 months. The CEA increased to 625.3 ng/ml during the final 6 months and, ultimately, the patient deteriorated secondary to widespread skeletal progression. Overall survival from the time of diagnosis was 24 months.

Although rare, lung NEC has been described in MEN1 but not in the MEN2 syndromes (Farhandi et al. 1987). In fact, mutations in the MEN1 gene have been implicated in the pathogenesis of sporadic lung NEC. In a case series evaluating sporadic lung NECs, 4 of 11 (36%) revealed inactivation of both MEN1 alleles on chromosome 11q13 by mutation and loss of heterozygosity (Debelenko et al. 1997). This pattern of bi-allelic loss of function is consistent with a tumor suppressor, as is noted in MEN1 neoplasms, and authors proposed that MEN1 genetic alterations are the first defined pathogenic abnormality for sporadic lung NEC (Debelenko et al. 1997, Dong et al. 1997).

To our knowledge, this is the first reported case of a primary lung NEC in a patient with established MEN2B, 18 years after his index presentation with bilateral pheochromocytomas and MTC. By next-generation sequencing, the tumor harbored only the patient’s germline RET mutation. Somatic mutations in the RET proto-oncogene have been described in lung adenocarcinomas (Kohno et al. 2013) but have not previously been associated with sporadic or hereditary lung NEC. The underlying M918T RET mutation in patients with MEN2B has also been described in sporadic neuroendocrine tumors including 44% (7/16) of MTCs and 15% (3/20) of pheochromocytomas (Komminoth et al. 1996). This small series, however, did not find any RET mutations in other neuroendocrine tumors studied, including four parathyroid adenomas, eight pituitary adenomas, 17 pancreatic neuroendocrine tumors, or 11 pulmonary and ten gastrointestinal carcinoids. Given the evidence of somatic RET mutations in other sporadic neuroendocrine tumors, the question is raised whether sporadic lung NECs, in particular large cell NEC or atypical carcinoid in nonsmokers, may harbor activating RET mutations.

This case demonstrates the striking potential of personalized medicine in rare malignancies. Although our patient did not respond to conventional cytotoxic chemotherapy patterned after large cell lung NEC, they had a remarkable clinical, biochemical, and radiologic response to second-line, RET-targeted therapy. Notably, during the phase III MTC trial, cabozantinib demonstrated a differential clinical benefit in patients harboring tumoral RET mutations, in particular M918T, suggesting that RET inhibition is important to the observed clinical activity in both MTC and this case (Sherman et al. 2013). Although the response to cabozantinib was not sustained, with biochemical progression noted at 12 months and radiographic progression at 18 months, the median PFS in patients with MTC is 11.2 months (Elisei et al. 2013). Moreover, a PFS of ~1 year is consistent with mutation-targeted RTK inhibitors for EGFR or ALK-altered lung cancer, malignancies characterized by solitary oncogenic driver mutations (Zhou et al. 2011, Solomon et al. 2014). Although impossible to prove that the RET mutation was the sole oncogenic driver in this case, the absence of other driver mutations on next-generation sequencing (including MET or VEGFR2, also targeted by cabozantinib), the failure of conventional cytotoxic chemotherapy, and the dramatic response to cabozantinib do make a compelling argument.

To our knowledge, a primary lung NEC bearing a somatic or germline RET mutation has not been described. This intriguing case suggests that, although rare, primary lung NEC occurs in the setting of MEN2B, is driven by the underlying germline RET mutation, and responds to RET-targeted therapy. Evaluation of sporadic lung NECs for RET mutations appears justified. When identified, uncontrolled RET-driven malignancies should be considered for RET-targeted therapy. Indeed, this case corroborates a new paradigm in oncology: personalized, mutation-directed therapy. This strategy underlies the US National Cancer Institute MATCH trial (NCT02465060) in which patients with advanced solid tumors are selected for targeted therapy on the basis of a corresponding mutation rather than on the traditional basis of anatomic site.
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