Germ-line exon 21 EGFR mutations, V843I and P848L, in nonsmall cell lung cancer patients

To the Editor:

Somatic epidermal growth factor receptor (EGFR) mutations are now routinely integrated in the molecular diagnosis of nonsmall cell lung cancers (NSCLC) [1, 2]. Thus, germ-line EGFR mutations are rarely mentioned or looked for in the context of patients with a family history of cancer, as their association with NSCLC familial cancer risk is not well established. Moreover, the predictive value of these mutations for response to EGFR tyrosine kinase inhibitors (TKIs) is not well known [3]. We report here two different heterozygous germ-line EGFR variants identified in two Caucasian NSCLC patients, who demonstrated different responses to EGFR-TKI.

A 63-year-old Caucasian, male former smoker with no family history of cancer (fig. 1a), was admitted for dyspnoea in August 2011, leading to discovery of a lower left lobe lung tumour and pleural effusion. Trans-thoracic biopsy diagnosed an invasive, acinar-predominant adenocarcinoma, classified cT2a N3 M1a (stage IV). Molecular analyses of the tumour by direct sequencing identified two concomitant heterozygous EGFR exon 21 mutations, L858R and V843I, confirmed in two independent experiments (fig. 1b). The V843I variant, but not L858R, was also detected in DNA obtained from a blood sample, with written informed consent, confirming a germ-line mutation (fig. 1c). Following treatment with cisplatin and pemetrexed, the patient relapsed in December 2012 with vertebral metastasis. An EGFR-TKI (erlotinib) was initiated, resulting in a stable disease for 9 months.

A 31-year-old Caucasian, female current smoker with a history of throat cancer in her maternal grandfather (who smoked) (fig. 1d), was admitted in February 2012 for intracranial hypertension due to a brain tumour, associated with a right upper lobe lung nodule. Surgical treatment of the two sites diagnosed an invasive, acinar-predominant adenocarcinoma, classified pT4 N0 M1b (stage IV). Molecular analyses of the primary lung tumour and the brain metastasis using direct sequencing revealed EGFR mutation P848L in exon 21 (fig. 1e), which was also detected in DNA obtained from healthy lung tissue and a blood sample, with written informed consent, confirming the germ-line mutation (fig. 1f). In July 2012, the patient relapsed, with psoas muscle metastasis. Despite treatment with cisplatin plus pemetrexed chemotherapy, the muscular lesion developed, prompting a change to erlotinib treatment in March 2013. After 4 months, the muscular metastasis progressed and erlotinib was discontinued.

Germ-line EGFR variants have rarely been described (<1 in 1000 EGFR mutations), and concern four EGFR mutations in two exons: the T790M [4–7] and R776X [3, 8] in exon 20, and the V843I [9, 10] and P848L [11, 12] in exon 21. Although somatic T790M mutations are common in patients with acquired resistance to EGFR-TKIs (50%), germ-line EGFR T790M mutations are rare, even in never-smokers (0.54%) [7]. The three other known EGFR mutations, R776X, V843I and P848L, belong to a group of very rare EGFR mutations with constitutive characteristics that are not always demonstrated. A recent study found only one EGFR germ-line mutation (R766G) among 71 lung tumour samples which was not found in 954 alleles from healthy individuals studied, leading to the conclusion that it is not a polymorphism responsible for NSCLC [3].

Germ-line EGFR mutations are rare but may contribute to oncogenesis. T790M has a moderate effect on EGFR function but, when combined with other EGFR mutations it shows a remarkable enhancement of EGFR activity. In vitro studies revealed that tyrosine autophosphorylation is enhanced in R776G-mutant EGFR when compared with wild-type EGFR and may be associated with a proliferative advantage. Germ-line EGFR mutations are often associated with another somatic EGFR mutation, such as the common L858R mutation. Among the five T790M germ-line mutated cases, three are described with L858R [7]. One of our cases harboured the V843I mutation combined with L858R; there are two other V843I mutated cases in the literature, one associated with L858R or L861Q [9] and another with L858R [10]. The mechanism through which V843I mutation confers predisposition remains unknown, but the acquisition of a second mutation must be essential for the development of lung adenocarcinoma. One possibility is that V843I causes genetic instability, thereby predisposing cells to additional mutations within the gene. This hypothesis is supported...
by the identification that the somatic L858R mutation nonrandomly occurred cis to the germ-line V843I or R776X mutations [3, 10]. R776C has also been described but as a somatic mutation in conjunction with L858R [13]. Other secondary somatic EGFR mutations have been reported with EGFR germ-line mutations, for example G719A with T790M or R776H germ-line mutations [3, 4, 7]. While cases of familial lung cancer have been occasionally reported, their genetic backgrounds remain largely unknown. Germ-line T790M mutation has been documented in five cases of four families with lung cancer susceptibility, including one family with six family members in three generations having lung cancer [4–7]. Unlike one of our cases, the germ-line V843I mutation has been reported in a family with lung cancer [10] and in a family with other cancer susceptibility [9]. Unlike somatic EGFR mutations, reported more often in Asians, females and non-smokers, few data for patients harbouring EGFR germ-line mutations are available. Five cases with T790M germ-line mutation are known, one Asian and three Caucasian (one unknown), of whom three were male and two female [7]. Three cases with R776G germ-line mutations were Caucasian, one male and two females [3, 8]. Our V848I patient was a Caucasian male, while the two previously reported cases of V848I germ-line mutations were Asian females [9, 10]. Our cases were a current and a former smoker. T790M germ-line mutations were found in three never-smokers and one smoker [7]. R776X mutations have been reported both in nonsmokers as well as in smokers, but no data are available for patients with V843I germ-line mutations [3, 8–10]. Within large cohorts of EGFR-mutated lung carcinomas, the vast majority are classified as adenocarcinomas. EGFR germ-line mutations are also more often reported in lung adenocarcinoma, as in our two cases, but one case of lung cancer harbouring R776H germ-line mutation has been reported with squamous differentiation [8]. As yet, it is hard to envisage how mutations that merely activate the kinase domain of EGFR might have differential effects on tumour-cell differentiation.

The predictive value of rare EGFR mutations is not well known. Growth inhibition assays using cell lines established from patient tissue harbouring V843I and L858R showed resistance to EGFR-TKI as well as the resistance observed in the clinical case [8]. It is widely accepted that while lung adenocarcinomas with the L858R mutation alone are susceptible to TKI therapy, gain of an additional T790M mutation overrides the effect of L858R and confers resistance to TKIs. Nevertheless, in our case, V848I was not a resistance mutation for TKI therapy, as in another case of lung adenocarcinoma with V848I EGFR mutation but without information about somatic or constitutive mutation [14]. In our second case, the unique germ-line P848L mutation is a TKI resistance mutation. R776X plus G719X mutated cell lines retain sensitivity to EGFR-TKIs [8].
We should look for germ-line EGFR mutations in cases of NSCLC with a familial history of cancers or with such very rare EGFR mutations. EGFR germ-line mutations must be confirmed in normal tissues or blood samples with written informed consent. The role of EGFR germ-line mutations in NSCLC and the familial occurrence of lung cancer or other cancers are still unclear. Further analyses are needed in order to precisely identify clinical and biological characteristics as well as the susceptibility of germ-line EGFR mutations to TKIs, particularly from the perspective of next-generation sequencing, which is able to identify such rare mutations, for giving genetic advice to the index patients and their relatives.

Correspondence: Michele Beau-Faller, Molecular Biology Laboratory, Strasbourg University Hospital, 1 avenue Molière, F-67098 Strasbourg, France. E-mail: Michele.FALLER@chru-strasbourg.fr

Nathalie Prim1,2, Michele Legrain3, Eric Guerin2, Bertrand Mennecier1, Noelle Weingartner4, Anne-Claire Voegeli5, Dominique Guenot5, Christine M. Maugard5, Anne-Elisabeth Quoix1 and Michele Beau-Faller2,3

1Chest Dept, Strasbourg University Hospital, Strasbourg, France. 2Research Unit EA 3430, Translational Medicine Federation, Strasbourg University, Strasbourg, France. 3Dept of Biochemistry and Molecular Biology, Strasbourg University Hospital, Strasbourg, France. 4Dept of Pathology, Strasbourg University Hospital, Strasbourg, France. 5UF6948, Oncogenetic Familial Evaluation and UF1422 Molecular Oncogenetic, Genetic Diagnostic Laboratory, Strasbourg University Hospital, Strasbourg, France.

Received: Dec 19 2013 | Accepted after revision: Jan 14 2014

Support statement: This work was supported by INCa (Institut National du Cancer), French National Cancer Institute.

Conflict of interest: Disclosures can be found alongside the online version of this article at err.ersjournals.com

Provenance: Submitted article, peer reviewed.

Acknowledgements: The authors wish to acknowledge the CRB (Centre de Ressources Biologiques), Biological Resource Centre, Strasbourg, France.

References

1 Girard N, Zalkman G. Topics in thoracic oncology: from surgical resection to molecular dissection. Eur Respir Rev 2013; 22: 101–102.

2 Cadranel J, Zalkman G, Sequist L. Genetic profiling and epidermal growth factor receptor-directed therapy in nonsmall cell lung cancer. Eur Respir J 2011; 37: 183–193.

3 Centeno I, Blay P, Santamaria I, et al. Germ-line mutations in epidermal growth factor receptor (EGFR) are rare but may contribute to oncogenesis: a novel germ-line mutation in EGFR detected in a patient with lung adenocarcinoma. BMC Cancer 2011; 11: 172.

4 Bell DW, Gore I, Okimoto RA, et al. Inherited susceptibility to lung cancer may be associated with the T790M drug resistance mutation in EGFR. Nat Genet 2005; 37: 1315–1316.

5 Prudkin L, Tang X, Wistuba II. Germ-line and somatic presentations of the EGFR T790M mutation in lung cancer. J Thorac Oncol 2009; 4: 139–141.

6 Tibaldi C, Giovannetti E, Vasile E, et al. Inherited germline T790M mutation and somatic epidermal growth factor receptor mutations in non-small cell lung cancer patients. J Thorac Oncol 2011; 6: 395–396.

7 Girard N, Lou E, Azzoli CG, et al. Analysis of genetic variants in never-smokers with lung cancer facilitated by an Internet-based blood collection protocol: a preliminary report. Clin Cancer Res 2010; 16: 755–763.

8 van Noesel J, van der Ven WH, van Os TA, et al. Activating germline R776H mutation in the epidermal growth factor receptor associated with lung cancer with squamous differentiation. J Clin Oncol 2013; 31: e161–e164.

9 Ikeda K, Nomori H, Mori T, et al. Novel germline mutation: EGFR V843I in patient with multiple lung adenocarcinomas and family members with lung cancer. Ann Thorac Surg 2008; 85: 1430–1432.

10 Ohtsuka K, Ohnishi H, Kurai D, et al. Familial lung adenocarcinoma caused by the EGFR V843I germ-line mutation. J Clin Oncol 2011; 29: e191–e192.

11 Sequist LV, Joshi VA, Janne PA, et al. Epidermal growth factor receptor mutation testing in the care of lung cancer patients. Clin Cancer Res 2006; 12: 4403s–4408s.

12 Wu JY, Wu SG, Yang CH, et al. Lung cancer with epidermal growth factor receptor exon 20 mutations is associated with poor gefitinib treatment response. Clin Cancer Res 2008; 14: 4877–4882.

13 Kosaka T, Yatabe Y, Endoh H, et al. Mutations of the epidermal growth factor receptor gene in lung cancer: biological and clinical implications. Cancer Res 2004; 64: 8919–8923.

14 Shih JY, Gow CH, Yu CJ, et al. Epidermal growth factor receptor mutations in needle biopsy/aspiration samples predict response to gefitinib therapy and survival of patients with advanced nonsmall cell lung cancer. Int J Cancer 2006; 118: 963–969.