Reviewer's report

Title: 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

Version: 1 Date: 26 January 2011

Reviewer: Elisa Giovannetti

Reviewer's report:

The present work describes the analysis of EGFR gene mutations in a population of 71 patients diagnosed with lung adenocarcinoma from Northern Spain, as well as in 912 individuals with lung cancer recruited from the CAPUA study, 477 unrelated healthy donor individuals and 32 individuals with other types of cancer. Furthermore the study was aimed at 1) determining the frequency of a new germ-line mutation found (p.R776G) during the study, as well as the frequency of three other EGFR germ-line mutations detected by other authors; and 2) evaluating whether the novel mutation detected may have a functional effect on the EGFR protein.

Overall, the manuscript deals with an issue of topical interest, which has not yet been the focus of many studies. Screening of EGFR activating mutation in NSCLC specimens is now commonly used to tailor the treatment with EGFR-tyrosine kinase inhibitors. However, analysis of germ-line mutations can increase the knowledge on cancer susceptibility alleles, while the evaluation of their role in response to treatment, if further developed, might lead to clinically relevant applications.

The experiments are well planned and conducted and the results summarized appropriately.

There are only a few minor revisions that might clarify some points to the reader:

1) In order to assist the reader, the Figures 1, and 2 should be enlarged and modified by using more readable labels for easy identification of bp numbers, restriction enzymes and wt vs. mt status.

2) The analysis of the role of p.R776G EGFR mutations in vitro (eg, using EGFR mutant vector constructions based on a pcDNA3.1 plasmid containing human wild-type EGFR cDNA where the EGFR tyrosine kinase domain was replaced with the corresponding p.R776G mutant variant and the resultant construction was used to transflect 293EBNA and COS7 cells) is well performed but time consuming. In the discussion the Authors should provide examples of alternative analysis, such as transfection of an YFP-tagged fragment of the EGFR intracellular domain (YFP-EGFR-ICD), followed by immunofluorescence microscopy analysis (see: de Gunst et al., Mol Cancer 2007)
5) References should take into account also the most recently published literature in the field (eg, Tibaldi et al., J Thor Oncol 2011)

**Level of interest:** An article of importance in its field

**Quality of written English:** Acceptable

**Statistical review:** No, the manuscript does not need to be seen by a statistician.

**Declaration of competing interests:**

'I declare that I have no competing interests'