

Suppression of KRAS-induced lung preneoplasia by inhibition of the Met receptor tyrosine kinase

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Abstract

Expression of oncogenic Kras in the alveolar epithelium is tumorigenic, but the mechanisms of Kras-induced lung tumorigenesis have not been defined. Here we investigated the role of the c-met receptor tyrosine kinase and its ligand, hepatocyte growth factor/scatter factor (HGF/SF), which have been reported to be highly expressed in cancer cells that have mutant KRAS. KrasLA1 mice, which develop multifocal lung adenocarcinomas owing to somatic mutations in KRAS, had high expression of c-Met in atypical alveolar hyperplasia (AAH) and adenomas, which precede the development of adenocarcinoma, and high concentrations of HGF/SF in bronchoalveolar lavage samples. Short-term treatment with a small molecule inhibitor of c-Met (PHA665752) reduced the phosphorylation of AKT, a pro-survival mediator of c-met, and induced apoptosis of vascular endothelial cells and alveolar epithelial cells. The pro-apoptotic effect of c-Met inhibition was observed *in vitro* in a lung adenocarcinoma cell line derived from KrasLA1 mice and in an immortalized murine vascular endothelial cell line. Thus, c-Met activates pro-survival signals in epithelial cells and vascular endothelial cells of early neoplastic lesions and thereby promotes lung tumorigenesis induced by KRAS.

Part I. C-Met and HGF expression in KrasLA1 mouse

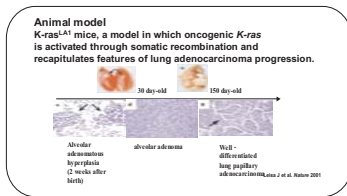
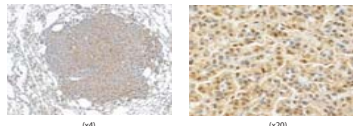
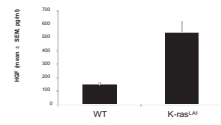


Fig. 1 (A) Immunohistochemical analysis of c-Met on lung adenoma from KrasLA1 mice



(B) HGF expression in KrasLA1 mouse lung (ELISA)



Part II. Met inhibition *in vivo*

PHA665752, a small molecule c-Met inhibitor treatment of KrasLA1 mouse.

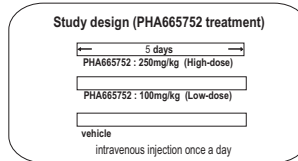
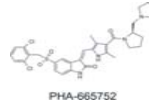
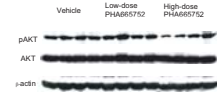
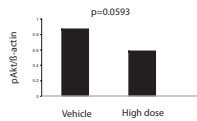


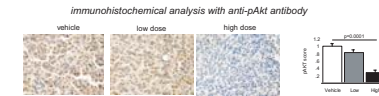
Fig. 2 (A) Akt down-regulation with Met inhibition *in vivo* (1)



Quantitation of pAkt expression using densitometry



(B) Akt down-regulation with Met inhibition *in vivo* (2)

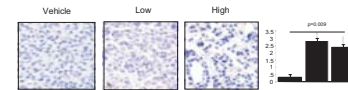


(C) Anti-angiogenesis with Met inhibition *in vivo*

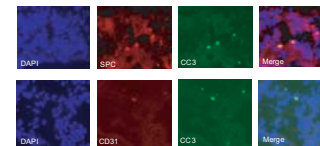


Part III. Met inhibition induced apoptosis *in vivo*

(A) Immunohistochemical analysis with anti cleaved caspase-3 antibody in mice lung tissues

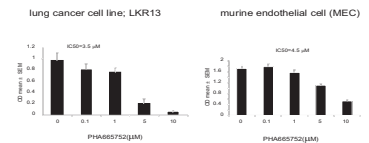


(B) Immunofluorescent analysis in mice lung tissues

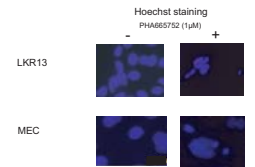


Part IV. Met inhibition induced apoptosis *in vitro*

Fig. 5 (A) Met inhibition inhibited cell growth (MTT assay)



(B) Met inhibition induced apoptosis



Summary

C-Met is expressed in alveolar preneoplasia induced by oncogenic KRAS.

A small molecule c-Met inhibitor, PHA665752, induced apoptosis of alveolar epithelial cells and endothelial cells in KrasLA1 mice.

C-Met may contribute to lung tumorigenesis induced by oncogenic KRAS.

Further studies are indicated to investigate c-Met as a target as a therapeutic or chemopreventive target in lung adenocarcinoma.