Expression and role of EGFR ligands induced in airway cells by $PM_{2.5}$ and its organic components

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Urban $PM_{2.5}$ (particulate matter, aerodynamic diameter below 2.5 µm) pollution is known to be responsible for airway inflammation characterized by proinflammatory cytokine release. It is also suspected of provoking or exacerbating airway remodelling, a phenomenon characterized, especially in chronic obstructive pulmonary disease, by epithelial mucous metaplasia, inflammation, enlargement of bronchial smooth muscle mass and modifications of lung growth factor expression and/or functions. Previous study from the lab, have shown that exposure of human bronchial epithelial cells to diesel exhaust particles or $PM_{2.5}$ leads to the release of amphiregulin, a growth factor, ligand of the epidermal growth factor receptor (EGFR).

The aim of this study was to establish the expression pattern of EGFR ligands induced in human respiratory epithelial cells by $PM_{2.5}$ and its different components, and then to determine the resulting consequences, for airway epithelial cells, of increased EGFR ligand production, due to their potential involvement in pro-inflammatory responses.

The involvement of epidermal growth factor receptor (EGFR) ligands in $PM_{2.5}$-provoked airway remodeling and/or inflammation was evaluated by establishing the EGFR ligand mRNA and protein expression profile in airway epithelial cells (bronchial 16HBE 14o- cell line and nasal normal cells) exposed to native $PM_{2.5}$ or its components. Conditioned media from $PM_{2.5}$-treated cells were used to evaluate the role of EGFR ligands in airway epithelial cell proliferation and proinflammation. This study reveals for the first time mRNA and protein overexpression of several ligands (amphiregulin, TGFα and HB-EGF) by human airway epithelial cells exposed to 1 to 10 µg/cm² of $PM_{2.5}$. Moreover, we demonstrate a slight but significant contribution of aqueous compounds, attributed to their metallic content, and strong participation of PM organic components, putatively attributed to PM PAH content, since the addition of an aryl hydrocarbon receptor antagonist, α-naphthoflavone, decreased PM-induced amphiregulin and TGFα release. $PM_{2.5}$ had a moderate mitogenic effect, but this did not appear to be sustained via autocrine regulation. In contrast, $PM_{2.5}$-induced amphiregulin and TGFα secretion by bronchial epithelial cells were involved in GM-CSF secretion, suggesting an autocrine role for EGFR ligands in eliciting and sustaining the proinflammatory response. These new findings provide additional and valuable information on inflammation following chronic exposure to PM that could contribute to exacerbation of airway remodeling in respiratory-compromised individuals.

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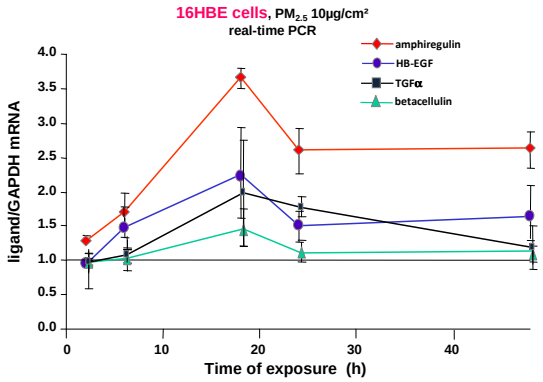
Urban PM2.5 (particulate matter, aerodynamic diameter below 2.5 µm) pollution is known to be responsible for airway inflammation characterized by proinflammatory cytokine release such as GM-CSF . It is also suspected of provoking or exacerbating airway remodelling, a phenomenon characterized, especially in chronic obstructive pulmonary disease, by epithelial mucous metaplasia, inflammation, enlargement of bronchial smooth muscle mass and modifications of lung growth factor expression and/or functions. Previous study from the lab, have shown that exposure of human bronchial epithelial cells to diesel exhaust particles or PM2.5 leads to the release of amphiregulin, a growth factor, ligand of the epidermal growth factor receptor (EGFR) (*Blanchet et al., AJRCMB, 2004, Rumelhard et al., Tox Lett., 2007*).

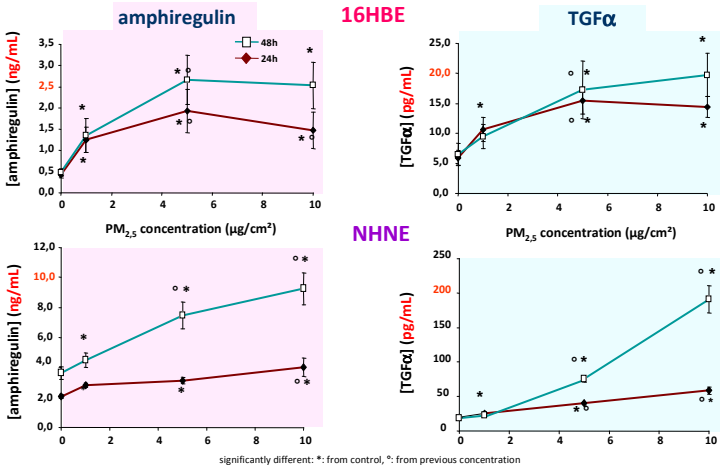
The aim of this study was to establish the expression pattern of EGFR ligands induced in human respiratory epithelial cells (bronchial epithelial cell line 16HBE, primary epithelial nasal cells NHNE) by Paris PM2.5 and its different components, and then to determine the resulting consequences, for airway epithelial cells, of increased EGFR ligand production, due to their potential involvement in pro-inflammatory responses.

PM2.5 induce mRNA expression of EGF receptor ligands



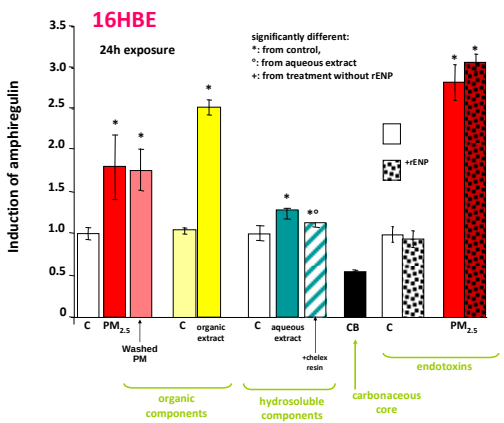
PM$_{2.5}$ induces the overexpression of EGFR ligand genes : amphiregulin, HB-EGF, TGFα
Almost no modulation of betacellulin mRNA expression.

PM2.5 induce the release of EGF receptor ligand



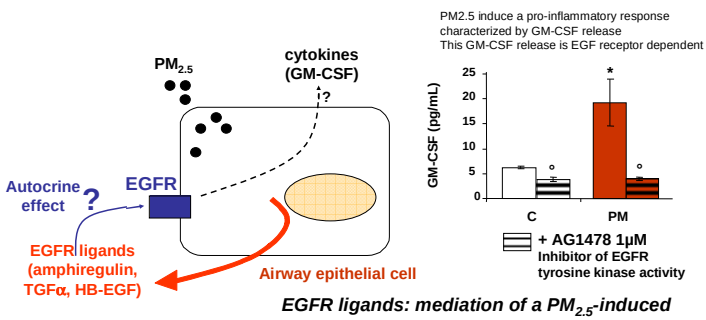
PM$_{2.5}$ induce a dose- and time-dependent release of amphiregulin and TGFα by human bronchial and nasal epithelial cells (transformed and primary cell)

Role of particle components in amphiregulin release



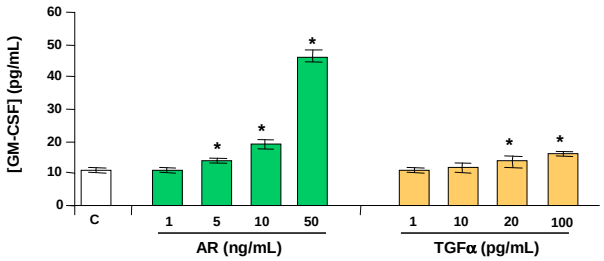
➔ Major contribution of PM2.5 organic component in amphiregulin release by 16 HBE cells
➔ Low contribution of hydrosoluble components (metals)
➔ No involvement of carbon core nor endotoxins.

Role of EGF receptor ligands?

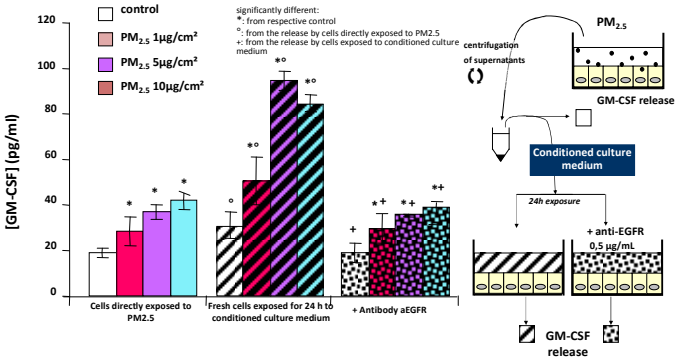


EGFR ligands: mediation of a PM$_{2.5}$-induced proinflammatory response?

EGFR ligands induce GM-CSF release



EGFR ligands mediate PM$_{2.5}$-induced GM-CSF release



EGFR ligands contribute to PM$_{2.5}$-induced GM-CSF release by autocrine action

This study reveals for the first time mRNA and protein overexpression of several ligands (amphiregulin, TGFα and HB-EGF) by human airway epithelial cells exposed to 1 to 10 µg/cm² of PM2.5.
It is mainly due to PM organic components.
PM2.5-induced amphiregulin and TGFα secretion by bronchial epithelial cells was involved in GM-CSF release, suggesting an autocrine role for EGFR ligands in eliciting and sustaining the proinflammatory response.
These new findings provide additional and valuable information on inflammation following chronic exposure to PM that could contribute to exacerbation of airway remodeling in respiratory-compromised individuals.