Novel in vitro and in vivo data on the cellular localization of Hsp10 in smokers affected by COPD and in lung-derived cell lines exposed to cigarette smoke extract as stressor

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Cigarette smoke is a potent stressor for the respiratory system, contributing to pathogenesis, for instance in chronic obstructive pulmonary disease (COPD), but its effects on the expression, function, and cellular localization of mitochondrial chaperonins are still largely unknown. We studied in vivo (airways biopsies) the localization of Hsp10 and Hsp60 in patients (smokers and non-smokers) affected by mild-moderate COPD, and characterized the effects of non-lethal doses of cigarette smoke extract (CSE) on the expression of these molecules in two human cell lines: lung fibroblasts (HFL-1) and bronchial epithelial (16HBE). We applied various in vitro methods: immunohistochemistry (IHC), subcellular fractionation analyses (SFA), Western blotting (WB), immunocytochemistry (ICC), and transmission electron microscopy (TEM) immunogold, and used bioinformatics and databases searches to gather structural in silico data for interpreting and complementing the in vitro results. IHC showed that in smokers and non-smokers COPD patients Hsp10 was localized in both, the cytoplasm and the nucleus of epithelial and lamina propria cells, while Hsp60 was present only in the cytosol. ICC, SFA, and WB on both CSE-exposed cell lines confirmed the presence of nuclear Hsp10, with an increasing trend in parallel to CSE concentration. TEM immunogold further confirmed Hsp10 in the nucleus, in addition to its presence in the cytoplasm and mitochondria, on both cell lines. Bioinformatics and in silico structural analyses indicated that Hsp10 can localize in extramitochondrial sites, such as the nucleus, even if Hsp10 lacks known DNA-binding motifs or nuclear import signals in its primary sequence. Our data suggest a link between exposure to exogenous oxidative stress and cell response, involving Hsp10, which would play roles different from its canonical functions. It is known that Hsp10 can display an array of functions depending on its location: cytoplasm, mitochondria, or extracellular. Here, we show for the first time the presence of Hsp10 in the nucleus of epithelial and stromal human-lung cell lines, paralleling the observations in vivo in COPD patients, and indicating that intranuclear Hsp10 levels are affected by oxidative stress due to an exogenous stressor like cigarette-smoke. The questions now are by what mechanism Hsp10 becomes a resident of the nucleus and what are its functions there.