Antitumor activity of non-thermal plasma: mechanisms analyses and preliminary approach using micro endoscopic plasma gun

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Background

Our group has recently showed a marked antitumor effect of DBD plasma treatment on U87 glioma bearing mice. Beside these results *in vivo*, various studies have showed an antitumor effect of plasma treatment on various cancer cells lines *in vitro* by apoptosis induction using DBD or plasma jet. Based on these interesting properties, non-thermal plasma appears to be a potential new antitumor strategy for different tumor type. The main goal of this work was to investigate the *in vitro* and *in vivo* plasma induced cell death mechanisms and to assess the possibility of lung in situ treatment using plasma gun.

Methods

Experiments were performed using a floating electrode DBD plasma reactor and the plasma gun previously described by our group. Antitumor activity was evaluated on U87 or HCT116 cancer cells previously transfected by the luciferase gene. U87 is a representative glioma model with a high chemo and radio resistance and HCT-116 is a colorectal carcinoma model which is a potential future target for *in situ* plasma application. *In vitro* effects of plasma treatment on proliferation, viability and apoptosis were assessed by bioluminescence imaging (BLI) and flow cytometer analyses with propidium iodide, annexin V or Brdu staining.

I*n vivo* DBD plasma treatments were performed on tumors heterotopically implanted in nude mice. Treatment effects on tumor cells proliferation and apoptosis were assessed by BLI, histology and by flow cytometer analyses. Plasma gun was used for lung treatment. To this end, tracheal intubation of the mouse with small diameter capillary was performed and a very low gas flow was used. Evaluation of plasma tolerance on lung tissue was done using a NF-kB-luc reported mouse. NF-kB is a transcription factor implied in the induction of inflammation process and imaging of this reported mouse by bioluminescence imaging (BLI) allows the monitoring of inflammation

Results and Discussion

Plasma treatment induced a significant antitumor effect on U87 and HCT116 cells *in vitro* and *in vivo.* Plasma also induced apoptosis *in vitro* and *in vivo*. This antitumor effect was associated to DNA damages leading to a cell cycle arrest and cell death induction by ROS generation in the vicinity of the cells.

In lung tissue, plasma gun application was successfully achieved since tracheal intubation with small capillary and use of low gas flow were well tolerated by the mouse, only a slight modification of the mouse breathing even with the presence of rare gas flow was observed. These results allow us to consider and to evaluate the interest of the plasma gun for the *in situ* treatment of lung tumor or bronchial dysplasia.

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