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Non-thermal atmospheric plasma in cancer therapy: cell death mechanisms and immunity

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Motivation and Aim: Along with developing efficacious chemotherapeutic agents to treat malignant neoplasms, much attention is currently being paid to physical methods. Cold atmospheric plasma (CAP) has been shown as potential anticancer tool. CAP is an ionized gas consisting of charged particles, active uncharged particles, an electric field and UV radiation. The CAP jet is visually composed by several millions of streamers per a minute propagating from the powered electrode to the target direction with the speed of 5–20~km/c. High energy plasma electrons in the streamers head produce chemically active oxygen (ROS) and nitrogen (RNS) species in the mixture of the inert gas with air in vicinity of the bio-target surface. These active compounds are known to be the main CAP cytotoxic affecters. We have shown previously that CAP induced death of cancer cells with the hallmarks of immunogenic cell death (ICD): calreticulin and heat shock protein 70 (HSP70) externalization and high-mobility group box 1 protein (HMGB1) release [1]. The present study aimed to address how CAP treatment changes the morphology of tumor cells, affects their transcription activity, and finally stimulates cell death or survival. Moreover, we analyzed if plasma-treated cells potent to stimulate dendritic cells maturation for the forming long-term antitumor immunity.
Methods and Algorithms: In our experiments, the source of the plasma jet was a coaxial dielectric channel with an inner diameter of 8 mm with a capillary nozzle with a diameter of 2.3 mm. The models used in this study were pulmonary-originated cell lines: lung adenocarcinoma cells A549 and Wi-38 normal lung fibroblasts. Differential gene expression was assessed by mass parallel sequencing on the Illumina NextSeq platform (RNA-Seq). Functional analyses were employed to reveal the difference in normal and cancer cell response. Validation of the differential expression of certain genes was performed by RT-PCR.
Results: The CAP irradiation conditions were optimized to kill A549 cancer cells and stay alive Wi-38 cells. Under these conditions the difference in genes expression was analyzed in A549 and Wi-38 irradiated cells. We identified universal indicator RNAs whose expression is modulated differently by CAP treatment for healthy and tumor cells. The substantial differences in expression was noted for genes of transcription factors, regulating RNA polymerase II activity. Despite the fact that CAP exposure led to the appearance of ICD markers in treated cancer cells *in vitro*, such cells have stimulated only weak maturation of dendritic cells.

Conclusion: The data obtained could be a basis for the development of selective CAP treatment of solid cancers in patients. This study partly explains the mechanisms of selective action of CAP towards tumor cells. The finding the conditions of CAP treatment under whose dying cells will stimulate the dendritic cell maturation can be considered an important task for future research

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References

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