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# Plasma-Activated Media Induced Apoptosis in Monoculture and Co-Culture Systems of Breast Cancer Cells

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**ABSTRACT**

**Introduction:** Breast cancer remains a major challenge due to the presence of aggressive and drug-resistant subtypes. Current therapeutic strategies have limited selectivity and severe side effects. Cold atmospheric plasma (CAP), which generates reactive oxygen species, induces apoptosis in cancer cells. In this study, the impact of CAP and plasma-activated media (PAM) on MDA-MB-231 and MCF-7 breast cancer cells, as well as their co-culture with human umbilical vein endothelial cells (HUVECs), was investigated.

**Materials and Methods:** CAP was generated using a plasma jet device operating with helium and argon gases under controlled conditions. The plasma treatment was applied in vitro to MCF-7 and MDA-MB-231 cells, along with HUVEC cells and their co-cultures. To assess cell viability, angiogenesis, and gene expression, we performed MTT, tube formation, and qRT-PCR. Apoptosis in treated cells was analyzed using Annexin V staining.

**Results and Discussion:** The results demonstrated significant efficacy of PAM in both monoculture and co-culture systems compared to direct plasma exposure ($p < 0.05$). Notably, PAM induced apoptosis and altered the expression of apoptosis-related genes, resulting in treated breast cancer cells.

**Conclusion:** Our findings suggest that PAM may offer a more effective and versatile approach for cancer treatment by a reduction in cell viability and modulating key biological processes such as apoptosis and angiogenesis.

*Keywords:* Breast cancer, CASPASE3, Cold atmospheric plasma, P53, Plasma-activated media, VEGF

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