## Nonthermal Dielectric Barrier Discharge Plasma Enhances Skeletal Cell Differentiation and Autopod Development

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Non-thermal dielectric barrier discharge plasma (NT-Plasma) is a relatively new physicsbased technology. Although few reports exist with regard to the application of this technology to eukaryotic cells, it is thought that NT-plasma influences cell function mainly through the generation of reactive oxygen and nitrogen species (ROS and RNS). Cell functions including motility, proliferation and differentiation are directed by ROSsensitive kinases, signaling proteins and transcription factors which regulate gene expression. Furthermore, ROS generation enhances development of embryonic structures and initiates the expression of many genes linked to cell differentiation. Based on the involvement of ROS in these processes, we investigated the potential of NT-Plasma generated ROS and RNS to promote mesenchymal cell (MC) proliferation, commitment and differentiation along skeletal lineages. We asked: 1) can NT-plasma promote MC differentiation while maintaining cell viability, 2) what cell signaling pathways are activated by NT-plasma to promote cell differentiation and tissue formation, and 3) can NT-plasma ROS and redox changes enhance developmental factor expression to activate signaling cascades and promote differentiation in organ culture. The results of our study show that NT-Plasma generated ROS can be used to enhance skeletal cell differentiation by increasing intracellular ROS, which lead to the activation of kinases and transcription factors known to influence genes associated with differentiation and skeletal development. Our future goal is to investigate if NT-Plasma can positively influence MC commitment and differentiation in vivo. Development of NT-Plasma to amplify MC function will be an invaluable tool for tissue engineering and regenerative medicine.