

A NOVEL PIN-TO-HOLE SPARK DISCHARGE PLASMA PRODUCES NITRIC OXIDE FOR MEDICAL APPLICATIONS

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Nitric oxide, an important signaling molecule in endothelial cells, mediates many processes at the cellular as well as tissue levels. Several diseases such as diabetes, coronary artery diseases and hypertension are characterized by a reduction in the bioavailability of NO, which leads to endothelial dysfunction¹. The use of NO donors has several drawbacks which includes inducing systemic side effects, due to their non-localized action. We have developed a novel pin-to-hole spark discharge (PHSD) plasma device which generates nitric oxide (NO), which can be applied safely and locally to endothelial cells at low concentrations.

The spark discharge is ignited in pin-hole electrode configuration, where the central needle-like electrode is powered from the positive output of a high voltage power supply. The outer electrode is grounded. The spark discharge has the following characteristics: voltage 2 – 3 kV, pulse duration ~ 20 μ s, frequency 1 – 7 Hz, energy per pulse 0.1 J, plasma diameter 3 – 5 mm, gas temperature 300 – 330 K. In order to reduce the exposure to ultraviolet radiation produced by the spark discharge, the device was equipped with a curved, low-density polyethylene tube extension.

Using 4, 5-diaminofluorescein (DAF-2), a fluorescent dye, concentrations of ~ 1600 nM of NO was detected in liquid for a treatment of 240 pulses². Using the cell permeant version of DAF-2, a concentration of 1000 – 1200 nM of NO was detected in a monolayer of endothelial cells for a treatment of 240 pulses. The cell viability following plasma treatment was measured via cell counts and LIVE/DEAD viability assay. No significant change in cell count was observed for treatments of up to 120 pulses. Our data demonstrates that the pin-to-hole spark discharge plasma produces nitric oxide which can be applied to endothelial cells, without any loss in viability, to generate physiological responses.

1. Georg Kojda, David Harrison, "Interactions between NO and reactive oxygen species: pathophysiological importance in atherosclerosis, hypertension, diabetes and heart failure", *Cardiovascular Research*. 43: 562-571, 1999.
2. Kojima et al., "Detection and imaging of nitric oxide with novel fluorescent indicators: diaminofluoresceins", *Analytical Chemistry*. 70: 2446-2453, 1998.