## **Plasma-Generated Reactive Species in physiological Solutions**

<u>Helena Tresp</u><sup>1,2</sup>, Malte U. Hammer<sup>1,2</sup>, Ansgar Schmidt-Bleker<sup>1,2</sup>, Jörn Winter<sup>1,2</sup>, Mareike A. Ch. Hänsch<sup>2</sup>, Kristian Wende<sup>1,2</sup>, Lucas Schaper<sup>3</sup>, Bill Graham<sup>3</sup>, Kai Masur<sup>1,2</sup>, Thomas von Woedtke<sup>2</sup>, Klaus-Dieter Weltmann<sup>2</sup>, Stephan Reuter<sup>1,2</sup>

<sup>1</sup> Centre for Innovation Competence plasmatis, Felix-Hausdorff-Str. 2, 17489 Greifswald, Germany

<sup>2</sup> Leibniz Institute for Plasma Science and Technology (INP) Greifswald, Felix-Hausdorff-Str. 2, 17489 Greifswald, Germany

> <sup>3</sup>Queen's University Belfast, University Road, Belfast BT7 1NN E-mail: <u>stephan.reuter@inp-greifswald.de</u>

For plasma medicine the determination of reactive species in plasmas and in plasma-treated liquids is essential. Especially nitric oxide and free radicals play a fundamental role in mammalian systems. Here the focus is set on plasma-generated reactive species in physiological solutions such as cell culture medium, sodium chloride solution, and phosphate buffered saline (PBS). Because nitric oxide is rapidly oxidized to nitrate and/or nitrite by oxygen (eq. 1, 2), the measurement of nitrate and nitrite concentration as the end products of NO hold as an index for the integrated nitric oxide production. Nitrite and nitrate play a key role in plasma-treated liquids [1]. For this work a colorimetric assay was used for nitrate and nitrite concentrations measurements.

$$NO + O_2^- \rightarrow ONO_2^- \xrightarrow{H^+} NO_3^- + H^+$$
(eq. 1)  
$$NO + O_2 \rightarrow N_2O_3^- \xrightarrow{2H_2O} NO_2^- + NO_3^-$$
(eq. 2)

The detection of free radicals was performed via electron paramagnetic resonance (EPR) spectroscopy. Considering the short lifetime of radicals in solution, a chemical agent – a so-called spin trap – was added to plasma-treated liquids. Here, DMPO (5,5-Dimethyl-1-pyrroline-N-oxide) is used for measurement of OH<sup>•</sup> and H<sup>•</sup> radical. This spin trap forms with radicals (more or less) stable adducts which can be determined by EPR. Additionally the pH value and concentration of  $H_2O_2$  was measured in parallel to each experiment. For these experiments two different plasma sources were used, an atmospheric pressure plasma jet (kinpen) and a pulsed discharge in liquids.

To create stable conditions for plasma treatment, the control of species, which can diffuse into the effluent of an atmospheric pressure plasma jet (humidity and air species), is necessary. A gas curtain was build and its effect on reactive species production in physiological solutions was investigated [2,3]. The gas curtain was used with varying ratios of nitrogen and oxygen as shielding gas.

## References

[1] K. Oehmigen, J. Winter, M. Hähnel, C. Wilke, R. Brandenburg, K.-D. Weltmann, Th. v. Woedtke, *Plasma Processes and Polymers* 2011, 8, p. 904-913

[2] S. Reuter, J. Winter, A. Schmidt-Bleker, H. Tresp, K.-D. Weltmann, *IEEE Transactions on Plasma Science, Special Issue on Atmospheric Pressure Plasma Jet Applications*, submitted 2012

[3] S. Reuter, H. Tresp, K. Wende, M. U. Hammer, J. Winter, K. Masur, A. Schmidt-Bleker, K.-D. Weltmann, *IEEE Transactions on Plasma Science, Special Issue on Plasmamedicine,* submitted 2012