1 Motivation

Sterilization and decontamination of medical instruments, implants, syringes, bottles, and so on in hospitals or surgeries is crucial to prevent infections of patients or employees during therapy (nosocomial infections). These infections are often caused by bacteria, fungi or proteinaceous residuals on materials in contact with the patient. Currently, conventional sterilization methods are based on treating the sample with moist heat at 134°C (autoclave), high energetic electrons (e-beam), gamma rays (γ-sterilization) or toxicants (e.g. ethylene oxide). These methods have several drawbacks and restrictions. The moist heat of an autoclave modifies or destroys the structure of high performance polymers used in implants or medical instruments and shortens the lifespan of e.g. drills or scalpels. e-beam, γ-sterilization or other radiation-based processes require high safety measures and are very expensive. Ethylene oxide, as a chemical sterilization process, is banned from use since it is highly toxic, mutagenic, explosive, and has a long degas time from porose materials (e.g. polymers in implants).

Additionally, investigations reveal deficits concerning the removal of protein residuals form surgical instruments after routine sterilization [1]. This poses a high risk to patients with respect to transmission of severe neurodegenerative diseases like Creutzfeldt-Jakob disease.

A promising alternative to conventional sterilization methods is the use of plasma for sterilization and decontamination. As plasma generates UV radiation, radical species like atomic oxygen, hydrogen or hydroxyl, and ions bombarding the surface, it is capable of deactivating and removing bacteria, fungi or proteinaceous residuals. The combined stress factors lead to a synergistic effect, enhancing the sterilization performance of each single stress. By varying the gas composition, power, or pressure, the plasma can be tuned to be most effective. Furthermore, it is possible to control the process temperature, making it applicable for heat-sensitive materials. To make plasma sterilization save for use, it is necessary to understand how biolo-
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Biological systems are influenced, deactivated, and removed by plasma. To get this far, the plasma itself as well as the biological systems have to be investigated in detail.

Fig. 2: Scanning electron microscopy images of spores on stainless steel screws [2]

2 Plasma Diagnostics

To understand the plasma itself, several diagnostic systems measuring the different parameters describing the plasma can be used. e.g. Optical Emission Spectroscopy (OES), Laser Absorption Spectrometry (LAS), Langmuir Probe (LP), Multipole Resonance Probe (MRP), or Mass Spectrometry (MS). Each system has pros and cons, why it is useful measuring the same parameters with different methods to achieve reliable results.

Optical emission spectroscopy uses spectrometers to investigate the radiation emitted by the excited species in the plasma. The amount of radiation is determined by the different plasma parameters, why it is possible to investigate the plasma without influencing it (non-invasive diagnostic method). [3] [4]

Laser absorption spectrometry uses laser radiation with a very well defined wavelength absorbed by only one specific molecule or atom. By measuring the absorption of the radiation, determination of the density of species is possible. As power is coupled into the system, it is possible to change the plasma by measuring it, why it is crucial to control the laser power very carefully.

The Langmuir probe system measures the current in a tiny wire applied with a defined voltage in the plasma. The current in the wire is caused by electrons and ions in the plasma attracted by the applied potential. By changing the potential, one can measure the electron density as well as the velocity or energy of the electrons. With this, determination of the electron energy distribution function (EEDF) is possible, showing how many electrons have which kinetic energy. [3] [4]

The multipole resonance probe emits electromagnetic waves into the plasma and measures their absorption. The absorption is caused by electrons and the frequency of maximum absorption is connected to the electron density, which can be determined investigating the absorption peak.

Mass spectrometry measures the mass of ionized particles. By this, one can determine different species which do not emit radiation in the range of the OES or can not be measured by LAS. Furthermore it can serve to check the reliability of the measured densities gained by OES and LAS.

Fig. 4: MRP in an argon plasma in the DICP (Source: www.ei.rub.de)
With this different diagnostic systems, it is possible to determine the irradiation and fluxes of radicals onto biological samples. By investigating the different damages caused by the plasma, a study and understanding of the deactivation mechanisms can be achieved.

3 Plasma & Plasma Component Sources

Plasma Sources

Two plasma systems are used studying the deactivation mechanisms. The first system is a double inductively coupled plasma (DICP) and the second a very high frequency capacitively coupled plasma (VHF-CCP).

The DICP has a discharge chamber volume of 40 liters made from stainless steel making it possible to generate very clean plasmas with a minimum of impurities. For this reason, the DICP is a reliable system to analyse the influence of plasma onto biological samples. Compared to the VHF-CCP, the DICP has a higher electron density and a lower electron temperature why both system have a different excitation of species in the plasma.

The VHF-CCP is designed to meet industrial needs. Its discharge chamber is a 4.5 liters drawer made of PEEK, a high resistant polymer, making it possible to seal the chamber after treatment and use it as sterile container. As a polymer is used as chamber material, the plasma interacts with it why many impurities are in the plasma making the diagnostic of densities and plasma parameters challenging. To achieve a reliable and fast sterilization method, the plasma process is combined with evaporation and condensation of 2 ml - 4 ml hydrogen peroxide before the plasma process. Ignition of the plasma afterwards removes hydrogen peroxide as well as biological residuals. Furthermore, the high UV and VUV radiation of a plasma inactivates biological samples that survived the condensation process.

Fig. 5: DICP with opened top

Fig. 6: Front view of the VHF-CCP

Plasma Component Source

To study the influence of single components of the plasma, another system called UV-Heat-setup exists, to simulate hydrogen plasma radiation and sample heating. The hydrogen radiation is simulated by a Hamamatsu X2D2® Deuterium lamp and the sample can be heated via a temperature controlled aluminium sample holder. Furthermore different gases can be used, like oxygen, nitrogen, hydrogen, or argon, to understand their influence onto biological systems in combination with temperature or UV and VUV radiation.
4 Biological Systems

To investigate the influence of plasma onto biological systems, spores, the most resistant form of bacteria, are treated with plasma. To understand the mechanisms leading to inactivation of spores, the different components in bacteria, like proteins, amino-acids, and DNA are also treated. By analyzing the induced damages of each component, it is possible to identify the most effective parts of a plasma for inactivation. The preparation and analysis of biological systems is done in cooperation with the Institute of Biology of Microorganisms at the Ruhr-University Bochum and with the Institute of Aerospace Medicine of the German Aerospace Center (DLR) in Cologne.

Fig. 7: Influence of hydrogen plasma and its components onto the protein GapDH [5]

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