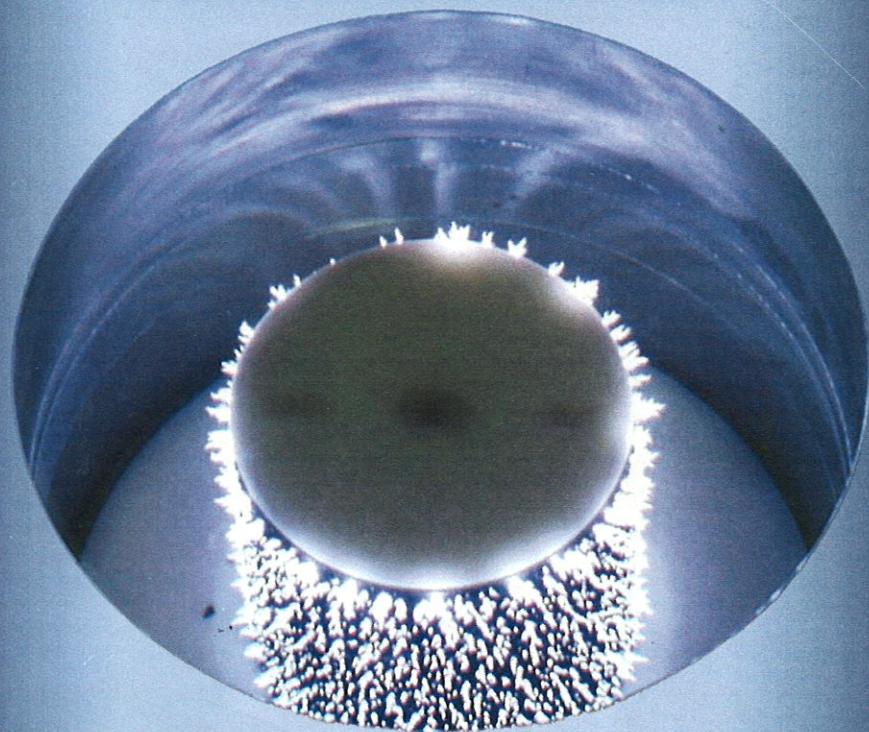


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The effect of air transient spark discharge on bovine serum albumin in liquid solutions

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Low temperature plasma (cold) is a rich source of reactive species, high energy electrons and charged particles, which form highly reactive environment at room temperature. It has a great potential in biological and medical applications as a tool for sterilization of thermosensitive materials, blood coagulation, wound healing or even cancer treatment. Many researches use cold plasma generated by gas discharges in contact with liquids and study plasma potential for decontamination of water, biocidal effects on bacteria and cytotoxic effects on cells. A lot of attention has been devoted to the effects of the plasmas on biomolecules (e.g. amino acids, proteins, DNA). Typically the oxidation and structural changes of amino acids, protein denaturation, the double strand breaks and fragmentation in DNA, can be expected. These effects are mainly caused by plasma-generated chemical species produced in the liquid environment.

We present the tests of cold plasma effects on bovine serum albumin (BSA) used as a model protein. BSA is a globular protein that is used in numerous biochemical applications due to its stability, lack of interference within biochemical reactions and low production cost. Few tests of plasma treatment on BSA have been reported, mostly with the dried protein on surfaces [1-3]. However, to understand the effect of plasma on living organisms it is also necessary to explore the plasma effect on proteins in liquid solutions.

We employed self-pulsing transient spark (TS) discharge generated in atmospheric air and treated BSA in aqueous solution. Buffered or non-buffered solutions of BSA (Sigma Aldrich A4503) of known concentration were prepared. Structural modifications of BSA induced by the TS were monitored using UV and fluorescence spectroscopy. UV absorbance at 280 nm was also measured in order to estimate the BSA concentration. The shift in maximum emission wavelength and decrease in intensity was monitored by fluorescence spectroscopy, using excitation wavelength of 280 nm with maximum fluorescence around 345 nm. To identify the secondary structure and conformation changes in the BSA we also used Raman and FTIR spectroscopy. The secondary structural changes, namely α -helix and β -sheet content, after TS treatment were compared with protein denaturation by heat and by dithiothreitol (DTT).

The effect of the applied voltage and exposure time of BSA was investigated. The fluorescence peak of BSA at 345 nm decreased with the applied voltage (e.g. by more than 50% after 6 min exposure with 18 kV) which corresponds to the protein gradual unfolding during plasma treatment. The air plasmas have shown strong biologically relevant effects - strong formation of RONS, direct contact of the discharge with the liquid solution and thus better transport of the gas-phase active species generated by the air discharge into a water solution seem to be the key factors in plasma biomedical treatments. These results successfully demonstrated a great potential of the air TS discharge as an efficient tool for biomedical applications that demonstrates an effect on biomolecules, as tested on BSA protein.

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