**Superoxide probes: synthesis, characterization and application**

The generation of free radicals in biological systems was discovered about 60 years ago. Cellular radicals are involved in physiological processes (participation in redox signalling or immune defence). Increased production of free radicals may, however, result in the structural damage to biomolecules, leading to lipid peroxidation, posttranslational modification of proteins and DNA damage. Among the biological radicals, superoxide radical anion (O2•–) is the primary species initiating the reactive oxygen species (ROS) cascade.

Increased generation of O2•– has been implicated in numerous diseases, including neurodegeneration, cardiovascular diseases and cancer. However, in most cases, it is experimentally challenging to define the actual mechanistic role of O2•–, the inability to selectively detect O2•– is a clear limiting factor.

The chemical tools for superoxide detection are very limited, hampering progress in understanding the chemical biology of superoxide. Electron paramagnetic resonance (EPR) spin trapping technique and fluorescent detection using hydroethidine are the most rigorous techniques to detect biological radicals. Their wider applications in biology are, however, limited due to the facile cellular reduction of EPR-active nitroxide spin adducts or into non-specific oxidative products of hydroethidine. In this work, we report different approaches to improve the spin trapping and the fluorescent detections.

