The functional dynamics and performance of micro- and nano-structured systems are often strongly temperature-dependent. One example is the rate of cell division: determined by tissue growth and dependant on changes in the structural properties of proteins that can undergo thermal denaturation if the temperature rises above 37 ºC by just a few degrees. Aberrant and highly localised temperature irregularities are often one of the first symptoms of cellular distress and ill health ranging from inflammation to cancer. Thermal imaging of thousands of cancer cells forming a tumour with a size below 1 mm would outstrip the capabilities of conventional tomography, where the minimal detectable tumor size is on the order of several millimetres. Aside from early detection of illness, real-time accurate temperature sensing is vital in medical treatment. Cell death can be induced in cancer cells by the targeted incremental raising of their temperature to lethal levels (43-45 ºC). Proper temperature monitoring is necessary to provide the most efficient and effective treatment along with minimisation of the side-effects. Detailed knowledge of the local temperature in a biological system can be provided with high spatial resolution by the combination of cutting-edge microscopy techniques with the sensitive yet robust nanothermometers.

The aim of this project is the development of a temperature mapping method based on multi-colour fluorescence microscopy of single nanoparticles. We exploit the unique optical properties of upconverting nanoparticles (UCNPs). UCNPs can convert near-infrared light into higher energy photons in the visible range of the spectrum. The most important feature of the UCNPs luminescence is the presence of two narrow luminescence bands, whose intensity ratio is strongly temperature-dependent. A suitable optical system allows for quantification of both emissions, the calculation of their intensity ratio, and in this way, estimation of the local temperature in the nanoparticle vicinity. Our fluorescence microscope will be operating in the wide-field mode, where tens of individual nanoparticles could be recorded at once, with a time resolution of tens of milliseconds. Scanning coupled with simple on-line calculation of the nanoparticles intensity ratio converts two microscopy images into a temperature map. Thereby, a temperature of a large area could be mapped with a submicron lateral resolution. Furthermore, the UCNPs exhibit great robustness and a lack of the environmental bias, that is typical for many other nanothermometers. They are insensitive to environmental factors such as pH or changes of refraction index. The ratiometric approach is the most reliable strategy for luminescence temperature measurement as it does not depend on excitation power, photobleaching, fluorescence quenching, or scattering, especially when both bands are of similar energy as in the case of UCNPs. Additionally, with a single molecule/nanocrystal approach, the problem of nanothermometer concentration is absent, since it operates at the limit of analytical chemistry. One of the main aspects of our research is the development of homogeneous, reproducible and efficient UCNPs, that once well-characterized in a controlled environment could be transferred to biological systems for absolute temperature measurement of tissues, cells and even cell organelle.