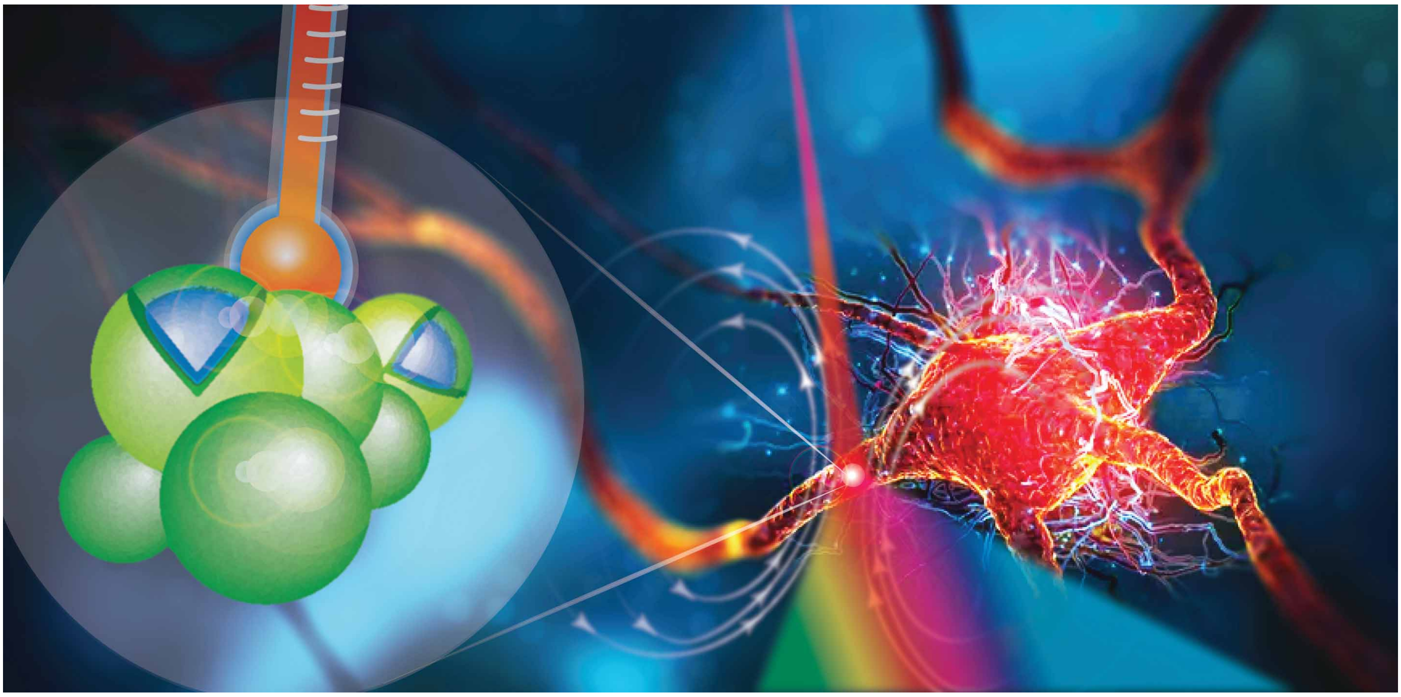
Nanoparticle-based 2D thermal bioimaging technologies (NanoTBTech)

The Horizon 2020 FET-OPEN-funded project NanoTBTech is leading the change towards new thermal bioimaging technologies



AT the dawn of medical science, way before the formal appearance of the scientific method, the Roman physician Celso/Celsus had already identified *calor* – the local increase of temperature – among the cardinal symptoms revealing inflammation process, (together with *rubor, tumor* and *dolor).* During the subsequent 2000 years of biomedical history, humankind has acquired further and deeper knowledge about the relationship between a great number of health disorders and anomalous temperature distributions in the affected organs. For instance, tumoural tissues have been seen to display different thermal relaxation dynamics than healthy ones.

At this point of the struggle, in order to solve key medical issues, researchers need to address several temperature-related fundamental biological issues. In living organisms, for instance, temperature is continuously fluctuating as it relates to many cellular functions, including gene expression, protein stabilisation, enzyme-ligand interactions, and enzyme activity. Moreover, the expression of heat-shock, cold-shock, and some virulence genes is co-ordinated in response to temperature changes. Intracellular temperature depends on the chemical reactions occurring inside cells, which are accompanied by either heat release or heat absorption, with the concomitant modification of the temperature.

Temperature changes and their spatial distribution are not just very relevant parameters that clinical physicians wish to accurately detect and quantify. Indeed, by purposely inducing a temperature change in a controlled local way, great healing effects can be achieved. In particular, hyperthermia is a harmless precision medicine method to kill malignant cells by thermal ablation and thermally-modulate the tumour microenvironment in order to have synergic effects with standard cancer treatments.

Currently, hyperthermia can be induced either by irradiation with a near infrared (NIR) laser or by an AC magnetic field. Indeed, magnetic hyperthermia already constitutes a reality that has received European regulatory approval for tumours' treatment as adjuvant or neoadjuvant therapy. However, the challenge is now to reach real-time thermal control over treated tissues, thus achieving a minimally invasive precision therapy with little collateral damage.

The challenge ahead: the right way to measure temperature for biomedical purposes

The use of temperature measurements for biomedical technology has a two-pronged promise to fulfil, namely the detection and spatial mapping of temperature gradients for a better and earlier detection of diseases, and real-time monitoring of hyperthermia treatments to avoid them causing more harm than good. To tackle those two complex issues, the key technology needs are non-contact thermometry granted with sub-micrometre resolution, providing high sensitivity thermal readout in a real-time mode (see Fig. 1).

Techniques able to go clearly below 1μm at cellular level and smaller than 1cm for *in vivo* targets are urgently needed, as the traditional contact-based sensors and mid infrared thermometers are not suitable for measurements at that tight spatial range.

Modern medicine has reached a point such that the use of traditional thermometers (e.g., liquid-filled and bimetallic thermometers, thermocouples, pyrometers, and thermistors) does not satisfy neither contactless nor submicrometric spatial resolution, that are required, for instance, to monitor the aforementioned intracellular temperature fluctuations. Although diverse biocompatible thermometers have been recently proposed, the area is still in its infancy and requires extensive and comprehensive theoretical and experimental work. Moreover, examples of *in vivo* thermal sensing are currently very scarce.

The NanoTBTech approach: a radical bet for luminescence-based nanothermometry

The main pathways (see Fig. 2) of our original approach are small size probes (luminescent nanoparticles), non-invasiveness of the methodology (NIR-to-NIR deep-tissue luminescence thermometry), and real-time readout. Thus, the planned outputs are:

- The fabrication of nontoxic, long-circulating 'stealth', functionalised, tumour-targeted and luminescent nanoparticles with high thermal sensitivity values. The temperature dependence of those nanoparticles' NIR emission spectra over time constitutes a spectroscopic fingerprint, linked to temperature changes happening around them. NanoTBTech researchers previously obtained outstanding results regarding the use of NIR-emitting nanoparticles as subcutaneous thermal probes in small animal bodies. They also applied emitting nanoparticles to unveil fundamental tissues' properties (as opposed to superficial) in *in vivo* conditions, thus evidencing the potential of NIR luminescence nanothermometry as a diagnosis tool
- Coupling luminescent 2D time-resolved thermal imaging and optical microscopy imaging under NIR irradiation (or AC magnetic field) in two different simple and compact prototypes to (a) monitor local hyperthermia in cells and (b) to study *in vivo* time-gated and 2D hyperspectral magnetic- or optically-gated thermal transient thermometry in depth tumoural models

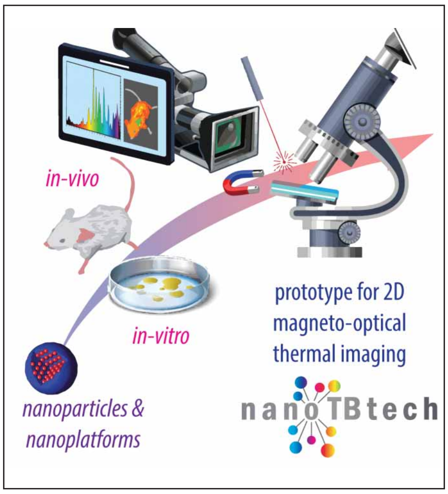
This thermal imaging modality is still less mature than many others, such as computed tomography (CT), magnetic resonance imaging (MRI) or positron emission tomography (PET), - all of them clinically applied already. However, thermal imaging based on luminescence, neither requiring long scanning times nor needing post processing analysis, provides a kind of real-time readout far from the reach of all those competing techniques.

NanoTBTech looks beyond the pure acquisition of knowledge. The effective delivery of a clean-cut breakthrough in 2-D thermal bioimaging technology will come through two impactful biomedical showcases, to demonstrate and apply the technological step forward: highly spatially-modulated intracellular magnetic/optical hyperthermia, and the *in vivo* detection and tracking of cancer.

Collective expertise to fulfil the NanoTBTech ambition

Our ambition is to develop a dedicated imaging platform with unprecedented performance leading to major advances in 2D thermal imaging technologies. Moreover, we foresee the project delivering novel insights about cell pathology and physiology, heat transfer at the nanoscale, and non-invasive detection of subcutaneous anomalies, in turn contributing to the development of novel theranostic tools. This requires a co-ordinated effort, which will not only address nanostructures' design and functionalisation but also characterisation/modelling, instrument assembling, software development, imaging interfacing, control and clinical expert inputs.

NanoTBTech brings together materials scientists, nanothermometry researchers, medical research institutes and companies devoted to nanotechnology, all of them with outstanding experience. Such a multinational consortium will supply functional technology, spreading it out through the European Research Area and to European citizens. The assembled consortium will accomplish that goal by way of moving results and discoveries captured in research papers to become prototypes that can match real life needs, first at translational centres, and tomorrow at hospitals. In the long-term, we foresee our technology as having a broad impact on non-invasive clinical imaging and theranostics.



NanoTBTech's partners

CICECO – Aveiro Institute of Materials, Universidade de Aveiro (UAVR – Portugal) is co-ordinating the project. Eight other partners participate in NanoTBTech:

- Fundacion para la Investigacion Biomedica del Hospital Universitario Ramon Y Cajal (FIBIRYCIS – Spain)
- Centre National de la Recherche Scientifique (CNRS – France)
- Agencia Estatal Consejo Superior de Investigaciones Cientificas (CSIC – Spain)
- Institut Za Nuklearne Nauke Vinca (Vinca – Serbia)
- Instytut Niskich Temperatur I Badan Strukturalnych Im. Wlodzimierza Trzebiatowskiego Polskiej Akademii Nauk (WPAS – Poland)
- Universiteit Utrecht (UU –The Netherlands)
- Nanoimmunotech SL (NIT –Spain)
- Biospace Lab Sa (Biospace Lab –France)

This project has received funding from the European Union's Horizon 2020 FET Open programme under Grant Agreement No. 801305.

Luís D Carlos
Full Professor
University of Aveiro
CICECO – Aveiro Institute of Materials (Phantom-g)

+351 234 370946

lcarlos@ua.pt
http://www.nanotbtech.eu/