Mapping intracellular thermal response of cancer cells to magnetic hyperthermia treatment

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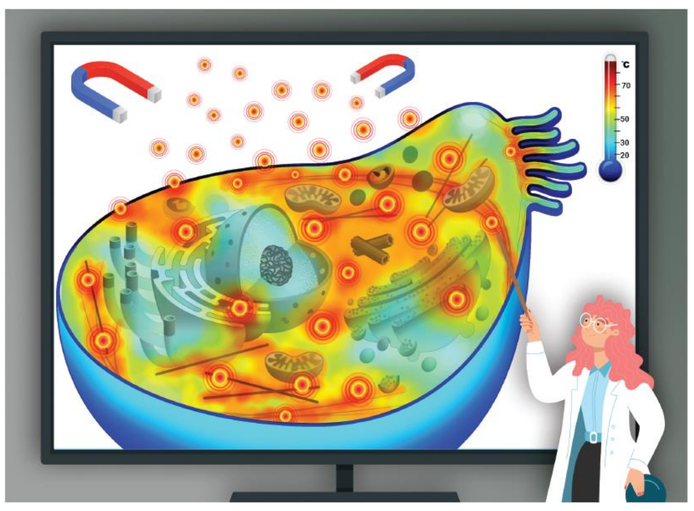
Introduction



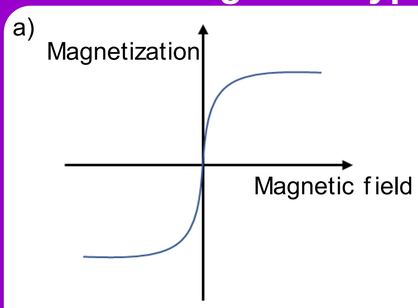
Back Cover of our ***Nanoscale* (2020)**, 12, 21647 [2].

Temperature is a key parameter for optimal cellular function and growth and can be used in cancer therapies to kill cells in tumors, a therapeutic approach called **hyperthermia**. Recently, several luminescent intracellular **nanothermometers** have been proposed [1]; however an application to sense temperature during a hyperthermia treatment is lacking.

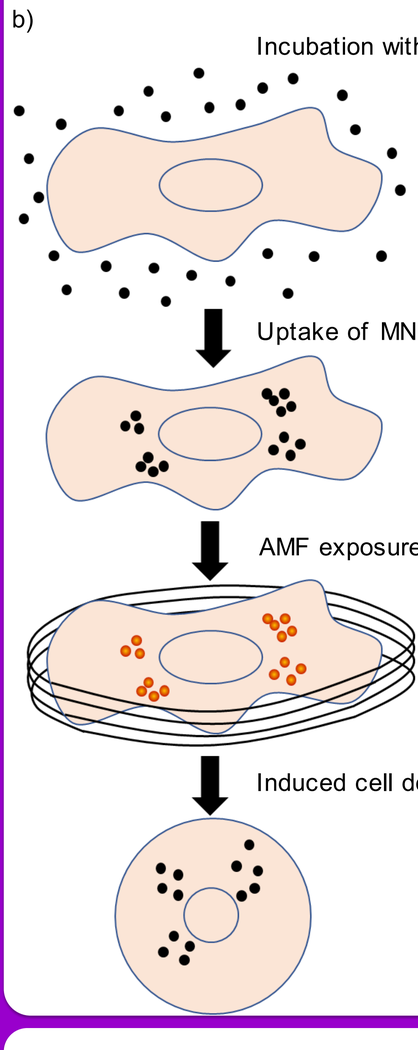
We present **green fluorescent protein (GFP)'s fluorescence lifetime parameter as a nanothermometer**. We use GFP in a bound form to actin filaments as an intracellular thermal reporter.

Furthermore, we assess intracellular temperature during **in vitro magnetothermal therapy on live HeLa cells** incubated with polyacrylic acid coated iron oxide nanoparticles. Compared to other thermosensitive materials and formulations reported so far, the **GFP nanothermosensor is easily expressed via transfection and various GFP variants are commercially available**. We foresee that the nanothermometer developed might find widespread **applications in cancer therapy research and development.**

In vitro Magnetic hyperthermia



Superpara-magnetic Fe_3O_4 MNPs are used with diameters of about 10 nm.

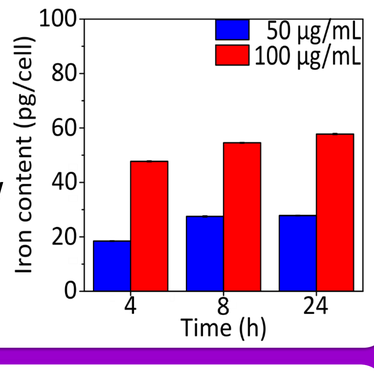


We incubate the live HeLa cells for 24 h prior experiments

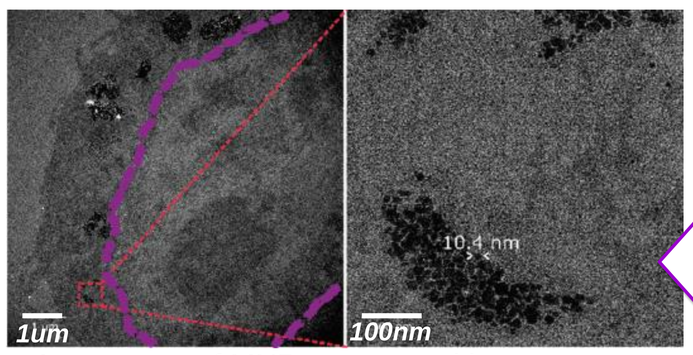
We also test the uptake is efficient with ICP and TEM***

Alternating Magnetic Field (AMF) is launched at the microscope sample plane to locally heat and induce cell death

***ICP**
Efficacy of MNP uptake is observed, at the MNP concentrations of low cytotoxicity as tested by cell viability assays [2].

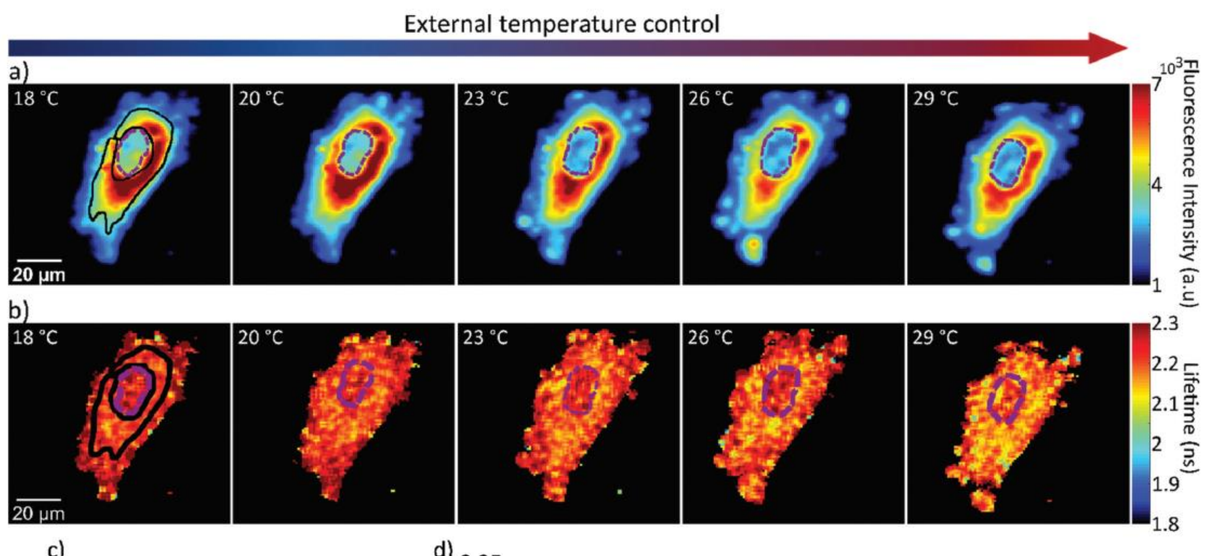


****TEM** Localization of MNPs in HeLa cells



Aggregates of MNP are seen with diameters in the micrometer range

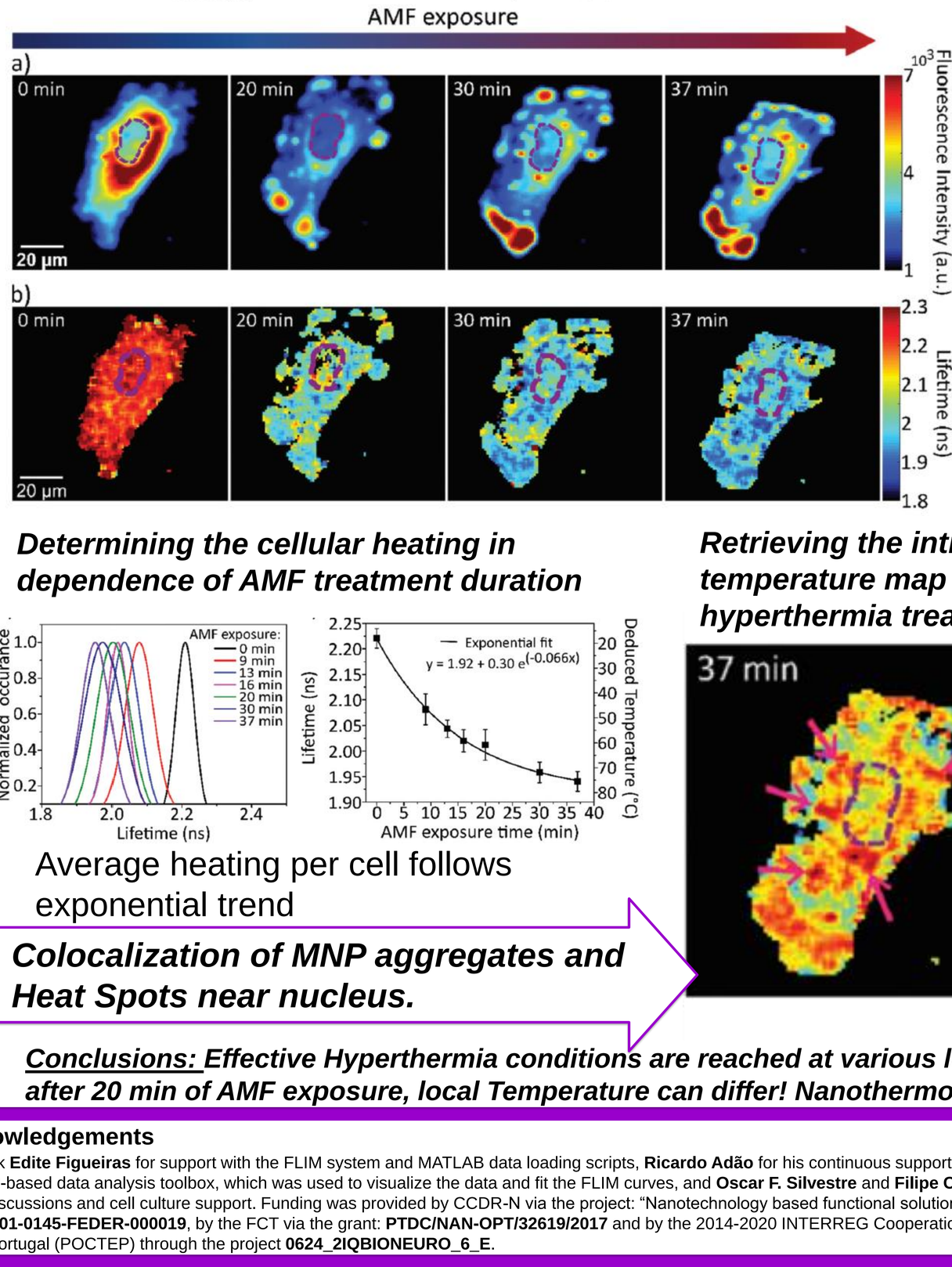
Actin-GFP as intracellular nanothermometer during in vivo magn. hyperthermia



Analyzing the temperature - dependent fluorescence properties of Actin-GFP expressed in live HeLa cells.

A linear dependence of the fluorescence lifetime $\tau_{GFP}(T)$ is observed.

Established the calibration of the GFP fluorescence lifetime τ nanothermometer.

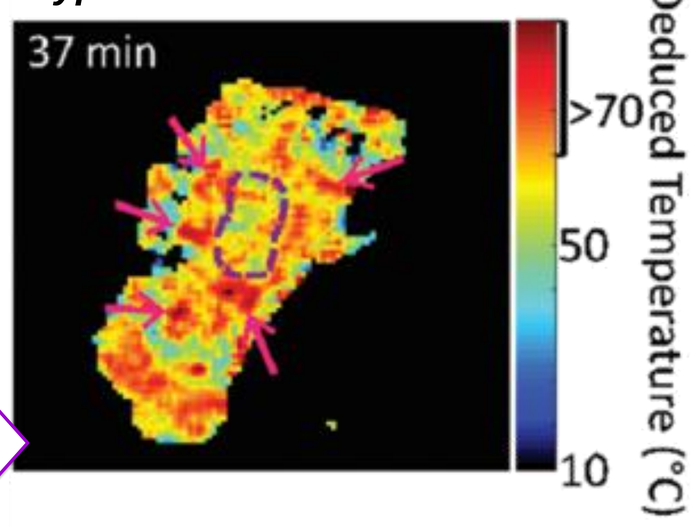


In-situ mapping of the Fluorescence intensity and lifetime changes of Action-GFP during AMF hyperthermia treatment of a HeLa live cells incubated with MNPs for 24h.

Determining the cellular heating in dependence of AMF treatment duration

Average heating per cell follows exponential trend

Retrieving the intracellular temperature map after 37 min of hyperthermia treatment



Colocalization of MNP aggregates and Heat Spots near nucleus.

***Conclusions:** Effective Hyperthermia conditions are reached at various locations of the cell after 20 min of AMF exposure, local Temperature can differ! Nanothermo-imaging needed!*

References
[1] Savchuk et al, ***Scientific Reports*** (**2019**) 9:7535, https://doi.org/10.1038/s41598-019-44023-7
[2] Silva, Savchuk, et. al., ***Nanoscale* (2020)**, 12, 21647, https://doi.org/10.1039/c9nr10370h

Acknowledgements
We thank **Edite Figueiras** for support with the FLIM system and MATLAB data loading scripts, **Ricardo Adão** for his continuous support and upgrades of a MATLAB-based data analysis toolbox, which was used to visualize the data and fit the FLIM curves, and **Oscar F. Silvestre** and **Filipe Camarneiro** for fruitful discussions and cell culture support. Funding was provided by CCDR-N via the project: "Nanotechnology based functional solutions" with grant no.: **NORTE-01-0145-FEDER-000019**, by the FCT via the grant: **PTDC/NAN-OPT/32619/2017** and by the 2014-2020 INTERREG Cooperation Programme Spain–Portugal (POCTEP) through the project **0624_2IQBIONEURO_6_E**.

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