

## Preface

As an alternative strategy, nanoparticles (NPs) mediated localized hyperthermia treatment modalities have shown most promising domino effect during clinical and various *in-vivo* and *in-vitro* studies. These localized treatment approach utilizes the heat generated by NPs in selective destruction of cancer cells under the application of a distant energy source. The malignant tumors are more sensitive to heat than that of the normal tissues due to its tortuous, disorganized vasculature which overflows with blind ends and abnormal bulges, these can be preferentially destroyed in 42 to 46 °C temperature range. Use of NPs offers several advantages. For example, NPs can easily pass into several tumours whose pore sizes are in 200–780 nm range increasing the effectiveness by delivering therapeutic heat directly to them. In addition, these NPs can be functionalized with cancer-specific binding agents and the possibility of NPs to form stable colloidal fluids allow for the intravenous or local hypodermal injection in the tumor tissues, resulting in obvious advantages such as less invasion, high targeting and low side effects. In the present-day nanoscale era the NPs assisted localized treatment based on magnetic- and photo-induced thermal therapy has gained much popularity in which heat is generated using either magnetic nanoparticles (MNPs) or photo-sensitive NPs, respectively. However, the use of NPs in biomedical applications is conditioned to their associated toxicity in biological systems. The magnetic-induced thermal therapy employing MNPs is known as magnetic fluid hyperthermia (MFH) while photo-induced thermal therapy with photo-sensitive NPs is known as photo-thermal therapy (PTT).

Iron oxide nanoparticles (IONPs) owing to their excellent biocompatibility are considered the most suitable candidate for MFH and render them as prospective material for

several bio-applications including drug delivery, magnetic targeting, cell separation, magnetic resonance imaging and magnetic particle imaging. Moreover, their unique magnetic property allows them to be remotely controlled and guided to a specific site (tumour region) using a magnet to provide an on-demand toxic amount of heat by applying a high frequency alternating magnetic field (AMF). When triggered with an AMF, the heat is released due to hysteresis losses which arise from phase delay in magnetisation response of the material to the applied AMF. The heat released can be improved by increasing the area under the A.C. hysteresis loop, which depends on the saturation magnetisation and coercive field of magnetic nanoparticles (MNPs).

To reduce the possible toxic effect associated with foreign particles, the usage of a minimal amount of MNPs for MFH is highly advantageous and demand of the time. It could be achieved using magnetic materials with high intrinsic loss power (ILP, a parameter which express the heating efficacy) value. With high ILP values materials, a low amount would be required to attain the therapeutic temperature even at low field intensity and frequency. Moreover, it could also aid in the likelihood of handling metastatic malignance where the accumulation of MNPs is relatively low. In the past few years various strategies, which aim at enhancing the saturation magnetisation and anisotropy of the material have been suggested to improve the heating behaviour. It includes modifying the size, shape, structure and chemical composition of the IONPs. One specific structure, magnetic nanoflowers (MNFs) of iron oxide, is quite stirring as they have shown outstanding heating performance during MFH. In addition, various other MNPs have been also evaluated and were found to display effective heating during MFH.  $\text{MnFe}_2\text{O}_4$  nanoparticles, in particular, have displayed heating and biocompatibility in line with IONPs. We believe their heating performance could further

be increased with nanoflower morphology as witnessed for iron oxide nanoflowers over iron oxide mono-core particles.

The PTT is centred on nanoparticles' ability to translate the electromagnetic radiation of near-infrared (NIR) wavelength to heat for the destruction of cancer cells. Various photo-sensitive nano-materials, such as gold, carbon nanotubes and graphene have been studied to explore their ability to generate heat during PTT. However, the clinical utilisation of these materials is limited by their non-specific distribution in normal tissues decreasing the accumulation of the photo-thermal agents in tumour sites and the potential *in-vivo* toxicity associated with non-biodegradable and bio-persistent behaviour. Recently, IONPs have demonstrated effective heating behaviour during PTT and that too at a much lower amount generally required during MFH. Moreover, a combined exposure of AMF and NIR irradiation can effectively enhance their heating output. Thus, biocompatible IONPs can substantiate as an effective photo-thermal material mitigating the limitations mentioned above. However, only a few reports are available on the heating effect of IONPs under NIR irradiation, and much of them were evaluated at higher power density than recommended for human use. A rigorous study is required to realise a high-performance MNPs which can effectively release heat under NIR irradiation of low power density, in addition to MFH.

This thesis as whole consists of six chapters:

Chapter-1 describes the literature on the structural and magnetic properties of MNPs, synthesis protocols to obtain mono-disperse MNPs, brief introduction to their application in cancer treatment through temperature rise, and factors governing the heating performance of such MNPs when exposed to AMF and NIR-laser.

Chapter- 2 provides the details of the materials and methods used for the synthesis of  $\gamma$ -Fe<sub>2</sub>O<sub>3</sub> nanoflowers, MnFe<sub>2</sub>O<sub>4</sub> nanoflowers and Fe<sub>3</sub>O<sub>4</sub> mesoporous nanoparticles. This chapter also includes the characterization techniques employed for evaluation of properties of the samples.

Chapter- 3 deals with the evaluation of heating behaviour of  $\gamma$ -Fe<sub>2</sub>O<sub>3</sub> MNFs of different size by exposing with AMF and NIR laser in separate experiments. The use of microwave energy during synthesis provided the fast reaction kinetics, reducing the synthesis time to 90 min from several hours required in the solvothermal process for getting MNFs. Varying the concentration of sodium acetate allowed us to tune the size and crystallinity of nanoflowers. Better crystallinity of the nanoflowers was found to enhance the magnetic properties (viz. higher saturation magnetisation and relatively high coercive field) leading to better heating performance during MFH. A high ILP value of  $15.21 \pm 0.34 \text{ nHm}^2\text{kg}^{-1}$  attained for these nanoflowers. Better absorbance, limited radiative emission (or higher non-radiative relaxations), agglomeration and smaller size were the factors which found to affect the heating performance of nanoflowers during PTT.

Chapter-4 deals with the study of heating performance of manganese ferrite nanoflowers, which was obtained utilizing the solvothermal process, by exposing them to AMF during MFH and NIR irradiation for PTT. The nanoflowers displayed improved saturation magnetisation,  $\sim 75 \text{ emu/g}$  which could be due to the reduced surface disorder and possible magnetic ordering within the flowers. The highest ILP value of  $5.31 \pm 0.079 \text{ nHm}^2\text{kg}^{-1}$  observed at an AMF of amplitude 170 Oe and frequency 330 kHz is comparable to the values for Fe<sub>3</sub>O<sub>4</sub> nanoflowers reported earlier. The material showed a very high heating capability under NIR irradiation of low power density. The intense NIR photo-counts is one

reason for their excellent photo-thermal performance. The aqueous suspension of the nanoflowers showed good absorbance near NIR region and a photo-thermal conversion efficiency of ~63%. Under NIR irradiation the therapeutic temperature (~42-46 °C) was achievable even at a low concentration of 0.1 mg/mL.

Chapter- 5 comprises a variety of components under one umbrella wherein a highly mono-dispersed mesoporous Fe<sub>3</sub>O<sub>4</sub> NPs through solvothermal process. The material displayed an early saturation behaviour having high saturation magnetization value of ~83 emu/g at room temperature. A high ILP value of  $\sim 5.30 \pm 0.25 \text{ nHm}^2\text{kg}^{-1}$  was obtained during MFH, at an AMF of amplitude 170 Oe and frequency 330 kHz, for the aqueous ferrofluid of concentration 0.5 mg/mL. The ILP value decreased on increasing the concentration of the ferrofluid due to the effect of enhanced dipolar effect at higher concentrations. The material showed extraordinary heating behaviour under NIR (808 nm) irradiation of  $0.33 \text{ Wcm}^{-2}$  power density. The intense NIR photo-counts observed in NIR PL experiments, due to the trapped electrons in the tetrahedral site, is the primary reason for their excellent photothermal performance. The aqueous suspension of the mesoporous nanoparticles showed good absorbance near the NIR region and photothermal conversion efficiency of ~61%. The in-vitro studies advise that the mesoporous Fe<sub>3</sub>O<sub>4</sub> nanoparticles could easily internalize in the cells and a high rate of cells death was noticed during PTT even at a concentration as low as 250 µg/mL for an exposure time of only 5 min.

Chapter-6 contains overall summary and major conclusions drawn from the present investigation. It also has some suggestions for the future work which may be explored further for better understanding as well as utilization of iron carbide based materials.

