Multifunctional Nano-Carrier as a Potential Micro-RNA Delivery Vehicle for Neuroblastoma Treatment

In this study, positively charged magnetic Nano-carriers comprising cross-linked polyethyleneimine– tripolyphosphate– Magnetic nanoparticles (PEI–TMNPs) were developed using the co-precipitation method. MNPs offer vast opportunities for drug/gene delivery. Iron oxide nanoparticles (IONPs) are available for imaging, diagnostic, and therapeutic applications.

Several types of nanoparticles (NPs) such as magnetic nanoparticles (MNPs) present promising opportunities for drug/gene delivery. Iron oxide nanoparticles (IONPs) with magnetic properties may be used for imaging, diagnostic, and therapeutic applications. These IONPs have diameters ranging from 1 to 100 nm and display a core of iron oxide (Fe_3O_4) or maghemite (Fe_2O_3) or a nonstoichiometric configuration of both. IONPs are the most potent nanomaterials in nanomedicine, owing to their exceptional physicochemical characteristics and excellent biocompatibility, non-toxicity, and stability in aqueous solutions. IONPs with bare surfaces tend to aggregate because of the strong magnetic attraction between these particles.[1–3]

The aggregation of IONPs may be resolved with its surface coating using polymer materials. Numerous studies have shown the successful delivery of PEI-based non-viral vectors into target tumors. Moreover, sodium TPP is a polyvalent anion with three negatively charged phosphate groups. TPP fragments display several attachment points that allow the fabrication of PEI–TMNP. The presence of polyanionic charges on TPP has allowed it's cross-linking with other polymers for RNA/DNA delivery.[2–5]

X-ray absorption near edge structure (XANES) and extended X-ray absorption fine structure (EXAFS) analysis were performed to understand the fine structure and Fe atomic arrangement in terms of bond distance and co-ordination number. The experiment was performed at **TLS 17C1**. Small-angle neutron scattering (SANS) measurements were obtained on **BILBY** with an OPAL research reactor at ANSTO. Scattering experiments for each sample at a continuous q range of $0.005 < q < 0.4$ Å^{-1} were performed. Structural changes were measured at temperatures of 25, 37, and 55 °C. An alternative magnetic field (AMF) was applied using in vitro magnetic field generator (MFG) 1000 (E0200, EURIS, Sweden).

SANS was used to probe changes in the core-shell structure of PEI–TMNPs before and after the addition of an external magnetic field. In this study, positively charged magnetic Nano-carriers comprising cross-linked PEI–TMNP were developed using the co-precipitation method. The PEI–TMNP samples at different PEI concentrations expressed as PEI–TMNP-x (x = 1, 2, 3 or 4).

Kuen-Song Lin (Yuan Ze University) and his group thoroughly examined the nature of the iron products using the XANES technique to obtain information related to electronic configuration, stereochemistry, and oxidation states of Fe atoms in MNP and PEI–TMNP. As shown in **Fig. 1(a)**, XANES spectra of Fe atom in MNP, and PEI–TMNP samples exhibited an absorbance feature (Fe = 7114 eV) of 1s to 3d transition. In comparison with the standard spectra, the XANES spectra of products were similar to those of the Fe_3O_4 standard. The transition intensity was very sensitive to the co-ordination symmetry. For iron oxides, 1s→3d peak intensity of Fe K-edge XANES was governed by the average coordination number and the symmetry of oxygen anions coordinated to the central iron cation.

Fe K-edge EXAFS spectroscopy may provide information about the atomic arrangement of Fe species in terms of bond distance, co-ordination number, and types of neighbors. **Fig. 1(b)** shows the fine structure parameters of MNP and PEI–TMNP at different PEI concentrations. EXAFS fitting results for the oxygen shell listed in **Table 1** suggest that MNP and PEI–TMNP have Fe atoms at the center that are primarily coordinated by Fe–O. The standard Fe–O bond distance in MNP and PEI–TMNP-x (x = 1, 2, 3, or 4) was 1.95 Å, with a coordination number of 4.30, 4.05, 4.08, 4.13, and 3.86, respectively. In all samples, the Debye-Waller factor (σ^2) was less than 0.014 (Å^2). The coordination number of the nearest O shell oscillated between samples, suggestive of the protective role of the coated polymer material.

The structural changes in PEI–TMNP-2 in response to the change in temperature and upon application of AMF at 0.5 mT external magnetic field at 37 °C were studied using SANS spectra. As shown in **Fig. 2(a)**, PEI polymer is not very sensitive to temperature change. The primary results of the AMF application highlighted the changes in temperature in response to the applied magnetic field (**Fig. 2(b)**). AMF triggered core heat generation that softened the shell. This phenomenon would enhance the gene/drug release within the tumors, leading to the inhibition of tumor growth. After the exposure to the magnetic field, more NPs were repositioned owing to the strong dipole-dipole attraction prompted by the magnetic field. However, further *in situ* experiments are necessary to comprehend the release mechanism after the AMF application.

In summary, Lin used SANS to study the possibility of integrating the AMF system and SANS, which seems promising. He further expressed that more *in-situ* experiments are necessary to comprehend the release mechanism of the Nano-carrier after its exposure to AMF. (Reported by Kuen-Song Lin, Yuan Ze University)

This report features the work of Kuen-Song Lin and his collaborators published in the J. Taiwan Inst. Chem. E. ***96****, 526 (2019).*

TLS 17C1 W200 – EXAFS
ANSTO BILBY – Small-angle Neutron Scattering

- SANS, XANES, EXAFS
- Materials Science, Chemistry, Surface, Interface and Thin-film Chemistry, Condensed-matter Physics

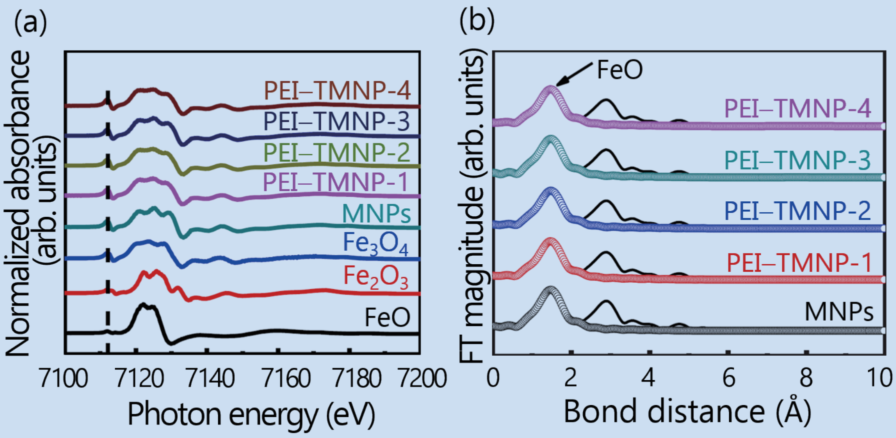

Fig. 1: (a) Fe K-edge derivative XANES spectra of FeO, Fe_2O_3, Fe_3O_4, MNPs, PEI–TMNP-x (x = 1, 2, 3 or 4). (b) Fe K -edge EXAFS Fourier transformed spectra of MNPs and PEI–TMNP-x (x = 1, 2, 3 or 4). [Reproduced from Ref. 2]

Table. 1: Fine structural parameters of MNP and PEI–TMNP-x (x = 1, 2, 3 or 4)

Samples	First Shell	CN (± 0.05)[a]	R (± 0.02 Å)[b]	$\Delta\sigma^2$ ($Å^2$)[c]
MNP	Fe–O	4.30	1.95	0.011
PEI–TMNP-1	Fe–O	4.05	1.95	0.010
PEI–TMNP-2	Fe–O	4.08	1.95	0.014
PEI–TMNP-3	Fe–O	4.13	1.95	0.013
PEI–TMNP-4	Fe–O	3.86	1.95	0.012

Notes: [a] Coordination number; [b] Bond distance; [c] Debye-Waller factor.

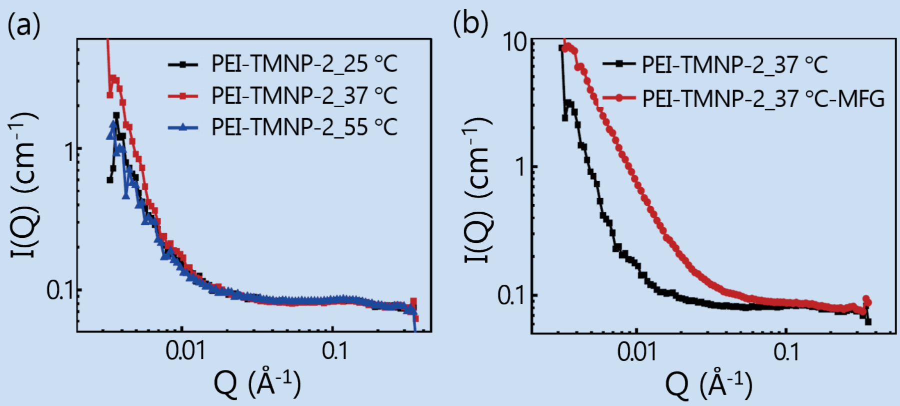

Fig. 2: The SANS spectra of (a) PEI–TMNP-2 at different temperatures and (b) PEI–TMNP-2 after applied 0.5 mT external magnetic field at 37 °C. [Reproduced from Ref. 2]

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