

S. Choi¹, H. Cho², N. Lee³, T. Hyeon⁴, W. Moon², and C-H. Sohn¹¹Radiology, Seoul National University, Seoul, Korea, Republic of, ²Seoul National University Hospital, Korea, Republic of, ³School of Chemical and Biological Engineering, Seoul National University, Seoul, Korea, Republic of, ⁴School of Chemical and Biological Engineering, Seoul National University, Korea, Republic of**Introduction**

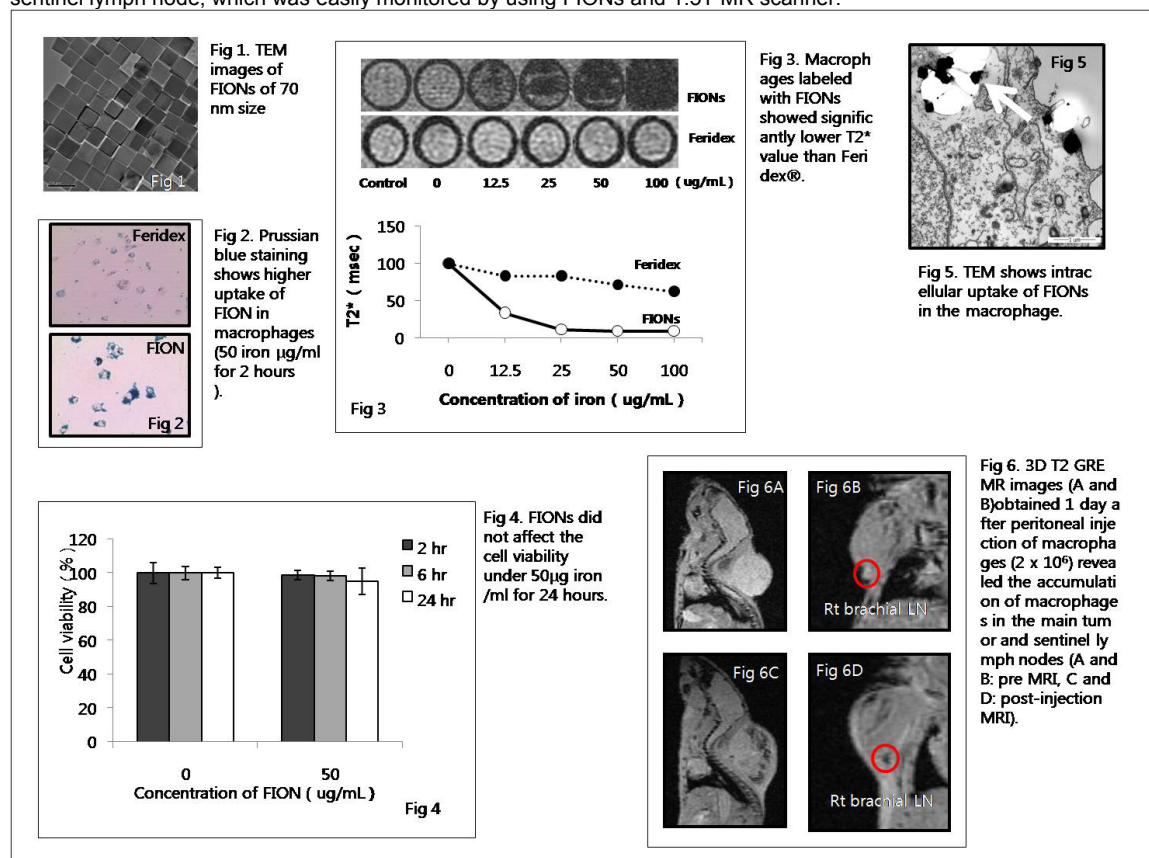
One of the body's responses to the presence of a malignant neoplasm is the recruitment of peripheral blood monocytes into the tumor, induced into the tumor mass by a chemoattractive gradient. Once the monocytes cross the endothelial basement membrane, they differentiate into macrophages. In breast cancer, macrophages may comprise up to 70% of the tumor mass. Macrophages that have infiltrated a tumor are referred to as tumor-associated macrophages or TAMs (1). There is increasing evidence that suggests that TAMs are "educated" by their microenvironment and promote tumor progression (1, 2). TAMs have been shown to foster invasion, proliferation tumor neoangiogenesis and metastasis and to suppress antitumor immune responses (3, 4). However, some reports have showed the possibility for delivery of therapeutic nanoparticles into tumors using macrophages, especially hypoxic area (5). To our knowledge, there was no report concerning the possibility of delivery of therapeutic agent to metastatic lymph nodes using macrophages. To monitor the fate of injected macrophages on MR imaging, we labeled them with ferromagnetic iron oxide nanocubes (FIONs) (Fig 1). The purpose of the present study was to confirm if metastatic lymph nodes can be targeted by macrophages labeled FIONs by using a mouse melanoma model.

Materials and Methods

Peritoneal macrophages were harvested from thioglycolate-treated Balb/c nude mice, cultured, labeled with FIONs in vitro. For labeling macrophages, they were incubated with 50 μ g iron/ml for 2 hours. To compare the cell labeling efficacy of FIONs with that of Feridex[®], macrophages were labeled with Feridex[®] under same condition, and T2* values were measured for 1 x 10⁵ macrophages labeled with FIONs and Feridex[®] by using 1.5 T MR scanner (GE health care), respectively. We used a MGRE sequence; NEX = 2, FA = 20°, TR = 800 msec, and 9 echoes ranging from 4.2 to 58.3 msec. Cell viability was measured by MTT assay. Transmission electron microscopy (TEM) was performed to confirm the phagocytosis of FIONs. Macrophages of 2x10⁶ labeled with FIONs were injected intraperitoneally (n = 5) and intravenously (n=5) into the Balb/c nude mice with melanoma tumor induced by B16F10 cell line. 3D T2 GRE MR images were obtained prior to and 1 day after the injection of macrophages: NEX = 4, FA = 10°, TR = 58 msec, and TE = 12 msec. Hematoxylin/eosin staining and Prussian blue staining were performed for the main melanoma tumor and lymph nodes.

Results and Discussion

Prussian blue staining revealed higher uptake of FIONs than Feridex[®] in macrophages (Fig 2). Macrophages labeled with FIONs showed significantly lower T2* value than Feridex[®] (Fig 3). FIONs did not affect the cell viability under 50 μ g iron/ml for 24 hours (Fig 4). TEM showed intracellular uptake of FIONs in the macrophage (Fig 5). 3D T2 GRE MR images obtained 1 day after peritoneal injection of macrophages revealed the accumulation of macrophages in the main tumor and sentinel lymph nodes (Fig 6). We believe that macrophages have the potential for the application of targeting the main tumor as well as sentinel lymph node, which was easily monitored by using FIONs and 1.5T MR scanner.

**Reference**

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