IMAGE-GUIDED PRO-ANGIOGENIC THERAPY IN DIABETIC STROKE MOUSE MODEL WITH A MULTI-MODAL NANOPROBE

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Target audience: Neurologist, Endocrinologist

Purpose: The objective of this study is to develop a non-invasive imaging strategy to assess the efficacy of pro-angiogenic therapy in diabetic stroke mouse model.

Methods: Ischemic stroke was induced by photothrombosis in adult male C57BL/6 and *db/db* mice (8-week old, n = 24). Endothelial progenitor cells (EPCs) isolated from bone marrow of C57BL/6 mice (male, 5-week old, n = 48) were intra-arterial injected into mice 24 h after surgery. The cyclic peptide (cRGDyK), Gd^{3+}-DTPA, IR783 and rhodamine were functionalized into the fifth generation (G5) PAMAM dendrimer to offer the $\alpha_v\beta_3$ integrin-targeted nanoprobe. Control nanoprobe that has similar chemical structure but without the cyclic peptide targeting domain was also prepared. The home-made paramagnetic/optical nanoprobe [1] was intravenously injected into mice at day 10 after stroke. Magnetic resonance (MR) and near-infrared (NIR) fluorescence imaging were performed at 24 h post-injection.

Results: The signal intensity on MR and NIR imaging in ischemic-angiogenic area of *db/db* mice was significantly lower than in wild type mice after $\alpha_v\beta_3$ integrin-targeted nanoprobe injection. After EPCs transplantation, the signal intensity was both increased in two kinds of mice compared to mice treated with saline. The enhancement of signal intensity in *db/db* mice was not as large as in wide type mice. Furthermore, histological analysis revealed that the microvessel density was consistent with the signal intensity from MR and NIR images.

Discussion: Pro-angiogenic therapy plays a critical role in the treatment of stroke patients with diabetes mellitus because the deficient angiogenesis after ischemia may result in worse outcomes [2]. However, the efficacy of pro-angiogenic therapies is hardly to be evaluated with current diagnostic modalities. While current clinical techniques for monitoring therapeutic efficacy, such as blood flow measurements, are capable of detecting the large caliber vessels that form at the late stages of revascularization, molecular imaging with targeted contrast agents can map the early signatures of angiogenesis. The $\alpha_v\beta_3$ integrin, differentially up-regulated in proliferating versus quiescent endothelial cells, is ideally suited for detection of angiogenesis [3]. This study demonstrated for the first time that noninvasive imaging with targeted multi-modal nanoprobe could be utilized to detect angiogenesis in ischemic stroke. This non-invasive imaging method indicates that bone marrow derived EPCs could successfully augment the angiogenic response in wild type mice and *db/db* mice, although the therapeutic response was less satisfactory in *db/db* mice than that of wild type mice. It might be explained that partial transplanted cells were functionally impaired in diabetes [4].

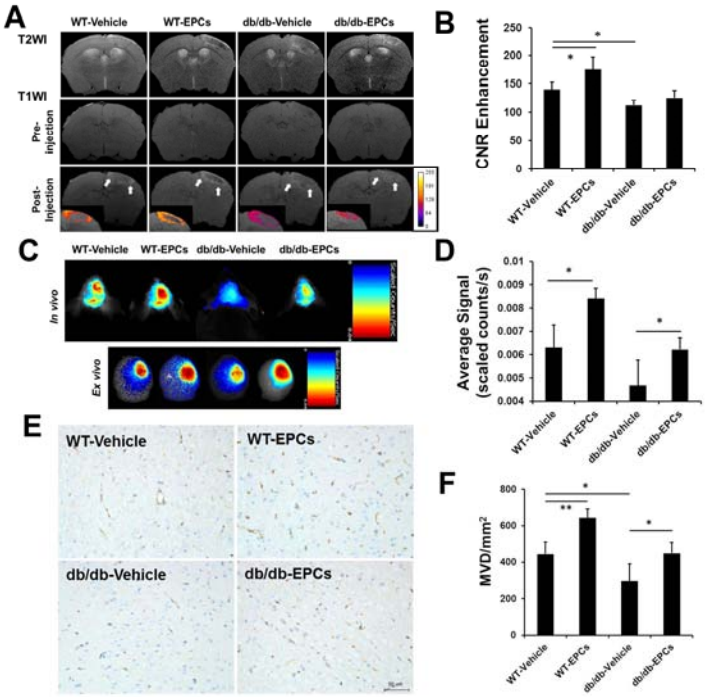

Figure: MR and NIR imaging of pro-angiogenic therapy in ischemic stroke mice. (A) Representative MR images of wild type and *db/db* mice treated with EPCs or saline. Arrows show high signal on T1-weighted imaging. The corresponding infract areas are shown on T2-weighted imaging and colored scale represents the Δ signal between pre- and post-injection. (B) The CNR enhancement in wild type mice treated with EPCs was significantly higher than mice treated with saline. In *db/db* mice, EPCs treatment also caused a moderate increase of CNR on T1-weighted imaging (n=3). (C, D) Staining for microvasculature also showed a larger number of capillaries in EPCs treated mice compared to saline treatment (n=3). Scaled bar = 50 μm, * = p < .05, ** = p < .01.

Conclusion: This non-invasive imaging method shows potential for early and accurate detection of therapeutic response in diabetic stroke patients, enabling individualized optimization for a variety of treatment strategies.

References:

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