Dose Response of Mn^{2+} on T_1 Relaxation Times in the Rat Brain after Subcutaneous Administration of $MnCl_2$

Mohammed Salman Shazeeb[1], and Christopher Sotak[2]

[1]*Radiology, University of Massachusetts Medical School, Worcester, MA, United States,* [2]*Department of Biomedical Engineering, Worcester Polytechnic Institute, Worcester, MA, United States*

Introduction: Divalent manganese ion (Mn^{2+}) is a widely used T_1 contrast agent in manganese-enhanced MRI (MEMRI) studies to visualize functional neural tracts and anatomy in the brain *in vivo*. In animal studies, the goal is to use a dose of Mn^{2+} that will maximize the contrast while minimizing its toxic effects. In rodents, systemic administration of Mn^{2+} via intravenous (IV) injection has been shown to create unique MRI contrast in the brain at a maximum dose of 175 mg/kg [1, 2]. The subcutaneous (SC) route can deliver Mn^{2+} at a maximum dose of 320 mg/kg (LD_{50} value). However, IV administration of Mn^{2+} results in faster bioelimination of excess Mn^{2+} from the plasma due to a steep concentration gradient between plasma and bile. By contrast, following SC injection, Mn^{2+} is released more slowly into the bloodstream, thus avoiding immediate hepatic elimination [3]. Therefore, SC administration of Mn^{2+} will result in prolonged accumulation of Mn^{2+} in the brain via the choroid plexus than that obtained via IV administration of Mn^{2+}. The goal of this study was to investigate the MRI dose response of Mn^{2+} in rat brain following SC administration of Mn^{2+}.

Methods: Experiments were carried out using 12 male Sprague Dawley rats weighing 200-450 g. $MnCl_2$ was administered using SC injection at three different doses: 75 (n=3), 150 (n=3), and 300 (n=7) mg/kg. All MR imaging was performed at 2.0T. Multi-slice T_1-weighted (T_1-WT) MR images (TR/TE = 700/15 ms) were acquired pre-injection and 6, 24, 72, and 168 h following the SC injection of Mn^{2+}. T_1 relaxation times were measured using an inversion recovery sequence (TR/TE = 10,000/4.8 ms, 16 inversion time (TI) points ranging from 15 ms to 3300 ms) acquired at the same time points as the T_1-WT images. Three different brain regions of interests (ROIs) were selected (cortex, sub-cortical region, and caudate nucleus) from three acquired slices. A mean ROI value from each TI point was used to calculate the respective tissue T_1 values.

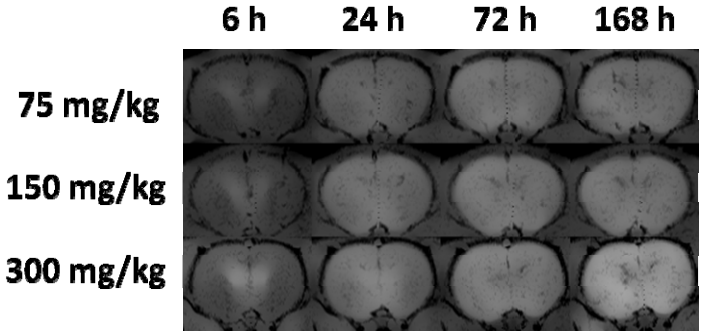

Fig. 1 – Dose dependence and time course of MEMRI contrast. T_1-weighted axial MR image sets are shown as a function of varying doses of $MnCl_2$ and as a function of time after subcutaneous injection of $MnCl_2$.

Results and Discussion: T_1-WT signal enhancement (SE) was apparent in the rat brain at 6 h which expanded from the ventricles to the sub-cortical and cortex regions. Uniform enhancement was achieved throughout the brain by the 72 h time-point at all administered Mn^{2+} doses which persisted up to 168 h. The T_1-WT SE was proportional to the dose of Mn^{2+} administered. Greater Mn^{2+} uptake occurred in the sub-cortical region than the cortex for all the SC Mn^{2+} doses administered causing greater shortening of the T_1 relaxation time in the sub-cortical region ROI than the cortex ROI (Fig. 2). ANOVA test for mixed models showed a significant effect of Mn^{2+} dose ($P < 0.01$) and time point after Mn^{2+} injection ($P < 0.0001$) on the reduction of T_1 relaxation times in the cortex (Fig. 2A) and sub-cortical (Fig. 2B) regions. Similar dose-dependent behavior of T_1 relaxation times has been observed in different regions of the mouse brain following IV injection of Mn^{2+} [2]; however, the prolonged enhancement obtained with SC Mn^{2+} injection (up to at least 168 h) is contrary to the short-term enhancement observed when Mn^{2+} was administered via IV [1] or even intrathecal injection [4].

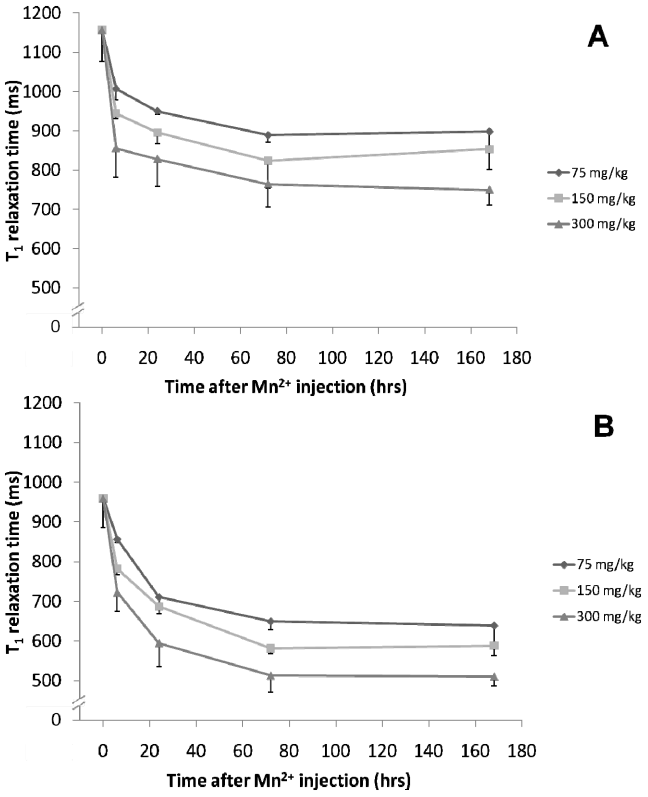

Fig. 2 – Plots of *in vivo* mean T_1 relaxation times (–1 SD) as a function of time after subcutaneous injection of $MnCl_2$ at three different doses in the **A)** cortex ROI, and **B)** sub-cortical region

Conclusion: This study is the first to demonstrate a dose-dependent response of Mn^{2+} on T_1 relaxation times in the rat brain following SC injection of Mn^{2+}. SC administration of Mn^{2+} leads to a more prolonged enhancement in the brain than IV Mn^{2+} administration which can be useful for longitudinal *in vivo* studies that require brain enhancement to persist for a long period of time to visualize neuroarchitecture like in Alzheimer's disease, Parkinson's disease, amyotrophic lateral sclerosis, and other neurodegenerative diseases.

References: [1] Aoki *et al.* (2004). NeuroImage **22**: 1046-59; [2] Lee *et al.* (2005). Magn Reson Med **53**(3): 640-648; [3] Bertinchamps *et al.* (1966). Am J Physiol **211**(1): 217-224; [4] Liu *et al.* (2004). Magn Reson Med **51**(5): 978-987.