MRI MONITORED UPTAKE OF MANGANESE IN THE MOUSE DURING CONTINUOUS ADMINISTRATION USING OSMOTIC INFUSION PUMPS

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Introduction

Cellular uptake of manganese (Mn^{2+}) through Ca^{2+} channels [1] combined with the paramagnetic properties of Mn^{2+}, is exploited in Mn^{2+}-enhanced magnetic resonance imaging (MEMRI) studies to provide information on brain anatomy and function of neuronal circuits. However, exposure to Mn^{2+} is known to cause cellular toxicity. In humans Mn^{2+} exposure may cause manganism, a Parkinson-like disease [2]. In rats and mice MEMRI studies currently require repeated bolus injections to optimize contrast in T_1-weighted images. Fractionated injections were shown to decrease toxicity [3]. In this study, we designed a new *in vivo* model of Mn^{2+} neurotoxicity with a constant release of the agent by implantation of mini-osmotic pumps. This would allow detailed elucidation of anatomical information and neuronal, activity-dependent, uptake while toxicity remains minimal. A study of the ion-pumps involved in uptake and clearance of Mn^{2+} in the brain and hormone-regulating glands, such as the pituitary and thyroid glands, could furthermore help in limiting the toxicity and understanding the activity-dependent neuronal uptake.

Methods

Animals and Mn^{2+} treatment:

Two month-old Swiss mice of were used for the study. Mini-osmotic pumps (Alzet) filled with 150 mg/ml $MnCl_2$ in NaCl 0.9% were implanted subcutaneously on the back of the animals, delivering a manganese dose of 30 mg/kg body weight/day (n=5; controls only saline n=3).

MRI:

T_1-weighted MR images were acquired using a Bruker Biospin 9.4 Tesla small animal scanner equipped with an actively shielded gradient set of 600 mT m-1 using a 3D gradient echo sequence (FLASH) with TR=30ms, TE=1.8ms, 60º pulse, FOV= 2.00x1.25x1.70cm with a matrix of 160x100x68 resulting in a spatial resolution of 125x125x250µm . T_1 maps were determined using a spine echo multiple TR saturation recovery approach (TE=6.5ms; TR=300, 500, 1000, 2500, 6000ms; five 300 µm thick axial slices with a resolution of 195x195 µm). For rf irradiation a 3.5cm quadrature transmit-receive coil (RAPID) was used.

Results & Discussion

We followed the tissue-specific Mn^{2+} accumulation by measurements of the T_1 relaxation times before and after 3 weeks of pump implantation (Table I). In the analyzed regions of interest, we observed a reduction of the T_1 after Mn^{2+} treatment that was related to the ion accumulation, the values being comparables with that reported upon acute injections [4]. We analyzed the time course of the Mn^{2+} accumulation in the olfactory bulb, cortex, hippocampus and cerebellum. A rapid decrease in the T_1 relaxation time was observed in the first days of exposure (Fig. 1). Some of these regions showed subareas with higher Mn^{2+} accumulation, such as the glomerular and mitral cell layers of olfactory bulb, the CA3 area in hippocampus, and the cerebellar grey matter, consistent with studies using bolus injections of Mn^{2+} [5]. Finally, an increase in signal intensity was observed in different glands, such as the anterior lobe of the pituitary gland and the thyroid gland (signal intensity in the T_1-weighted images increased 4.8 fold and 4.4 fold respectively relative to day 0), which could be related to a specific role of Mn^{2+} in secretory processes. The mechanisms of Mn^{2+} uptake and clearance and the tissue-dependent toxicity are still under investigation.

Table I. T1 values (ms) in different mouse brain areas

	Brain areas							
	OB	Cx	Str	Hp	Thal	Hyp	Cb	Mes
Control	1971 ±31	1954±43	1957±42	1973±53	1890±68	1937±39	1994±104	1763±123
Manganese	1202±15	1248±14	1111±60	1150±26	1092±30	962±53	1095±35	1100±70

Data are mean ± SE of the indicated ROIs from control and Mn^{2+}-treated mice at 21 days after implantation. OB: olfactory bulb; Cx: cortex; Str: striatum; Hp: hippocampus; Thal: thalamus; Hyp: hypothalamus; Cb: cerebellum; Mes: mesencephalon.

Fig. 1 Time-dependent effect of Mn^{2+}-treatment on the T_1 values of different regions of mouse brain. T_1 measurements were obtained from control (open symbols) and Mn^{2+}-treated (filled symbols) mice scanned at different time points after implantation of mini-osmotic pumps (day 0). Data are mean ± SE of the following ROIs: OB: olfactory bulb; Cx: cortex; Hp: hippocampus; Cb: cerebellum.

Fig. 2 T1-weighted image showing typical contrast obtained three weeks after implantation of the mini-osmotic infusion pumps. 3D FLASH image showing uptake of Mn^{2+} in olfactory bulb, and hippocampus and cerebellum.

FIG.1

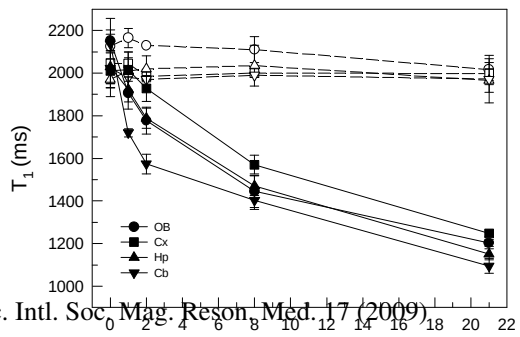
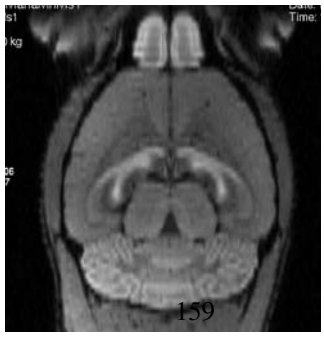


FIG.2



References

(1) Narita K *et al.* (1990) Brain Res. 510: 289-295.
(2) Olanow CW (2004) Ann N Y Acad Sci. 1012: 209-223.
(3) Bock *et al.* NMR Biomed. (2007) 21:473-478
(4) Kuo *et al.* JMRI 21:334-339 (2008) 34: 595-604.
(5) Lee *et al.* MRM (2005) 53:640-648.