MANGANESE Mn(II) DOTA-MODIFIED POLYSACCHARIDE AS AN ALTERNATIVE TO GADOLINIUM-BASED CONTRAST AGENTS FOR MAGNETIC RESONANCE IMAGING (MRI)

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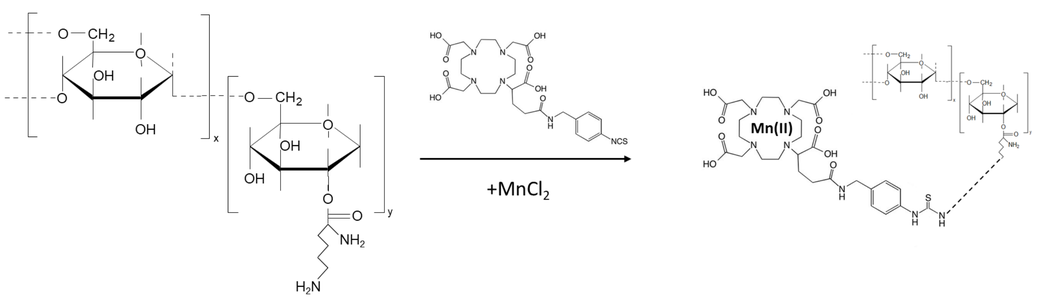
Objective

Paramagnetic manganese Mn(II) chelates emerged as suitable alternatives to the routinely applied gadolinium Gd(III)-based contrast agents for Magnetic resonance imaging (MRI). Free Mn(II) ions or in labile complexes are toxic, therefore using novel macromolecular chelators may lead to improved stability and security of novel Mn(II)-based polysaccharide complexes.

Materials and Methods

1,4,7,10-Tetraazacyclododecane-1,4,7,10-tetraacetic acid mono-N-hydroxysuccinimide ester (DOTA-NHS) and 2,2′,2"-(10-(1-carboxy-4-((4-isothiocyanatobenzyl)amino)-4-oxobutyl)-1,4,7,10-tetraazacyclododecane-1,4,7-triyl)triacetic acid (p-NCS-Bz-DOTA-GA) were conjugated to lysinated (amino) dextran with 70kDa MW under mild conditions to allow introduction of the paramagnetic Mn(II). ATR-FTIR of modified Mn(II)-DOTA-dextran conjugates was performed to confirm the successful coupling of the macrocyclic chelators to the polysaccharide molecule. Further characterization was made by PCS, ICP-MS and FPLC. *In vitro* relaxivity measurements in a high field MR scanner (9.4 Tesla, BioSpec 94/30USR, Bruker, Germany) have been carried in order to evaluate the potential of the novel Mn(II) conjugates as MR contrast agents.



Lysinated (amino) dextran | p-NCS-Bz-DOTA-GA | Mn(II)-DOTA-dextran

Results

Successful synthesis and characterization of Mn(II)-DOTA modified dextran, mediated through either DOTA-NHS ester or p-NCS-Bz-DOTA-GA was carried out. However, polydispersity due to possible cross-linking was observed by the DOTA-NHS-mediated coupling, where as a single peak fraction containing Mn(II)-p-NCS-Bz-DOTA-GA modified dextran could be collected.
ATR-FTIR spectra of the starting AD showed absorption peaks at 3322 cm-1, 2919 cm-1, 1414 cm-1 and 1348 cm-1, and 1003 cm-1 assigned to O-H, C-H, C=O, and C-O stretching respectively. After coupling to p-NCS-Bz-DOTA-GA, the C-O band disappears and it is shifted and splitted into two bands at 1047 and 1013 cm-1, corresponding to AD-p-NCS-Bz-DOTA-GA conjugation. After complexation with Mn(II) the splitting disappears and a single shifted band can be seen at 1077 cm-1, indicating coordinated Mn(II) ion (Figure 1).
In vitro MRI relaxivity studies showed superior signal enhancement of the Mn(II) polysaccharide conjugates in comparison with the low molecular $MnCl_2$ and the cyclic Gd-HPDO3A (ProHance) gadoteridol in clinical use (Figures 2 and 3).

Conclusion

Novel Mn(II)-dextran compounds were successfully prepared by using two different derivatives of the more thermodynamically stable macrocyclic DOTA chelator as alternative MR CAs. A strong signal enhancement at concentrations 50 times lower than gadoteridol, used as positive control, makes them safer potential candidates for biomedical applications.

References

[1]Devreux M, Henoumont C, Dioury F, et al. Mn2+ Complexes with Pyclen-Based Derivatives as Contrast Agents for Magnetic Resonance Imaging: Synthesis and Relaxometry Characterization. Inorg Chem. 2021;60(6):3604-3619. [2]Islam MK, Kim S, Kim HK, et al. Manganese Complex of Ethylenediaminetetraacetic Acid (EDTA)-Benzothiazole Aniline (BTA) Conjugate as a Potential Liver-Targeting MRI Contrast Agent. J Med Chem. 2017;60(7):2993-3001. [3]Troughton JS, Greenfield MT, Greenwood JM, et al. Synthesis and evaluation of a high relaxivity manganese(II)-based MRI contrast agent. Inorg Chem. 2004;43(20):6313-6323. [4]Aime S, Anelli L, Botta M, et al. Relaxometric evaluation of novel manganese(II) complexes for application as contrast agents in magnetic resonance imaging. J Biol Inorg Chem. 2002;7(1-2):58-67.

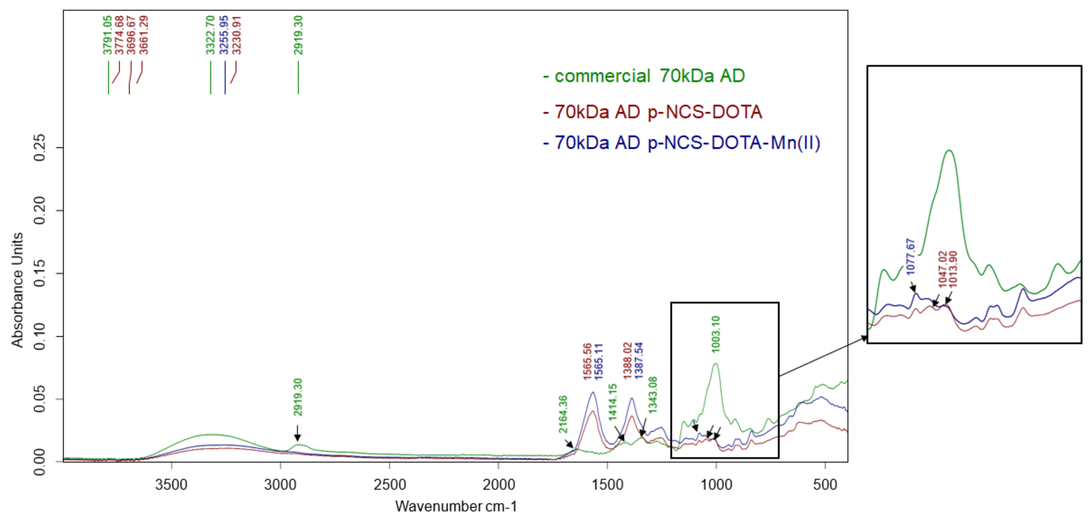


Figure 1. ATR-FTIR spectra of commercial 70kDa aminodextran (green), p-NCS-Bz-DOTA-GA-dextran conjugate (red), and the final Mn(II)-DOTA-dextran (blue).

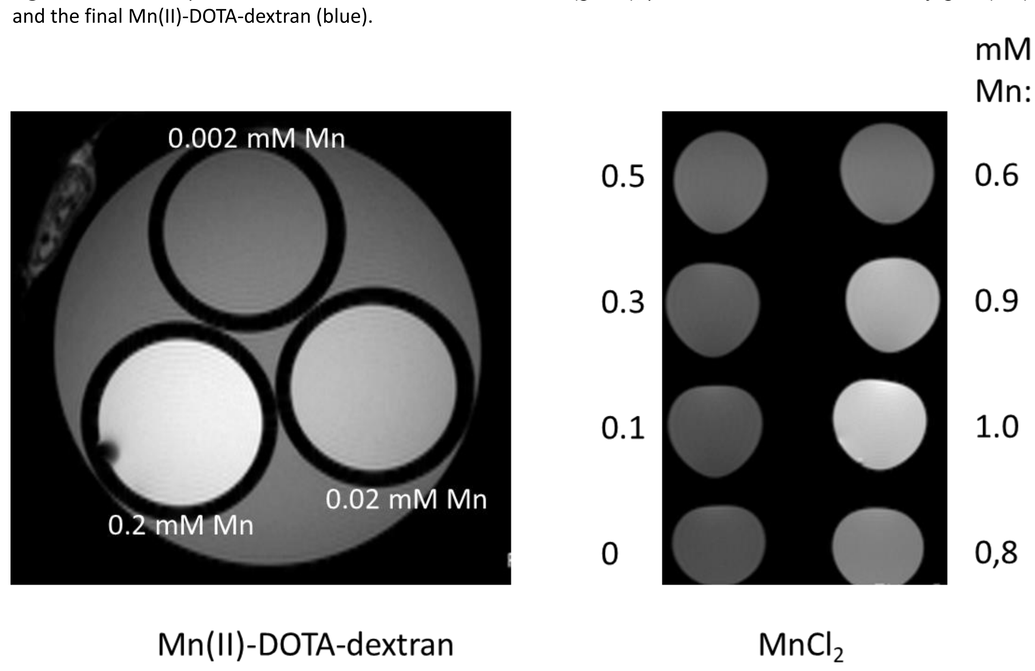


Figure 2. MRI images. Signal enhancement with Mn(II)-DOTA-dextran as contrast agent (0.2 to 0.002 mM) in comparison to $MnCl_2$ (0-1 mM)

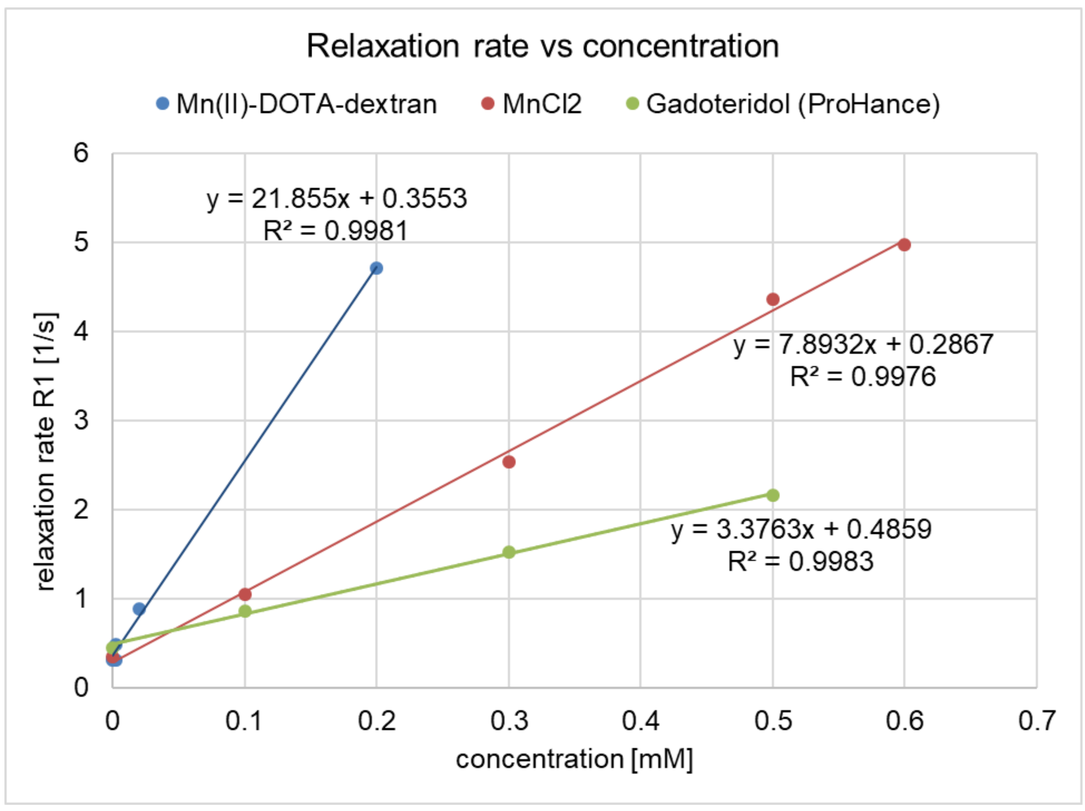


Figure 3. Plots of relaxation rate (R1, [1/s]) to concentration (mM) curves for Mn(II)-DOTA-dextran compared to low molecular weight $MnCl_2$ and clinically applied gadoteridol (ProHance, cyclic). A steep increase of the relaxation rate R1 using the Mn(II) conjugate as MR contrast agent (CA) was observed.