NEW VANADIUM-BASED MRI PROBES FOR EARLY DETECTION OF CANCER

D. Mustafi[1], S. Foxley[2], M. Zamora[2], M. W. Makinen[1], G. S. Karczmar[2]

[1]Biochemistry & Molecular Biology, The University of Chicago, Chicago, Illinois, United States, [2]Radiology, The University of Chicago, Chicago, Illinois, United States

Abstract: We have developed a novel class of MRI contrast agents, containing vanadyl (VO^{2+})-chelated organic ligands, namely *bis*(acetylacetonato)oxovanadium(IV) [$VO(acac)_2$], for magnetic resonance imaging (MRI). In separate experiments using 3T3-L1 adipocytes we have demonstrated that $VO(acac)_2$ enhances glucose uptake and tyrosine phosphorylation of the insulin receptor and insulin receptor substrate-1 synergistically with insulin. In addition, these contrast agents target specifically receptor proteins in glycolysis and are taken up by highly glycolytic cells. We have also demonstrated that $VO(acac)_2$ has no adverse effect in rats or mice after administration of a large dose. The relaxivity of $VO(acac)_2$ is comparable to that of low molecular weight gadolinium (Gd^{3+}) complexes. Furthermore, it provided excellent T_1 and T_2^* contrasts compared to that of Gd-DTPA in high-resolution MR images of rodent tumors.

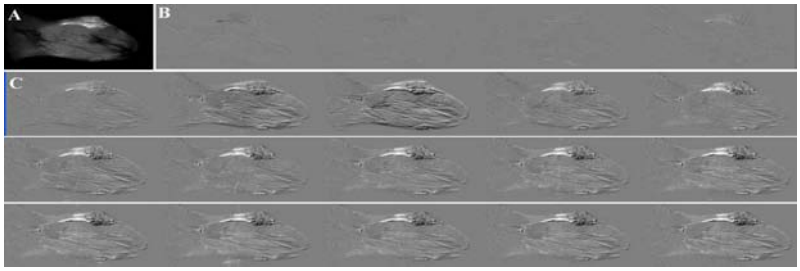
Introduction: Improved methods for early detection and accurate characterization of cancer are highly desirable for treatment of cancer. We developed a new MRI method for detecting cancer based on the use of a novel class of contrast agents that initially act as blood pool agents and then are preferentially taken up by cancer cells. Aggressive cancers are metabolically active glycolytically, resulting in increased uptake of glucose. An MRI contrast agent that enters in cells with high glycolytic activity could provide high-resolution *functional* images of tumor boundaries and internal structure, which cannot be achieved by conventional, non-specific contrast agents. These functional MR images would be analogous to PET images of [^{18}F]fluorodeoxyglucose uptake but they would have higher resolution, would provide additional information concerning blood flow and anatomy, and most importantly, would not require a radioactive tracer.

Methods: Using electron paramagnetic resonance spectroscopic and calorimetric methods, we measured the binding affinity and stoichiometry of VO^{2+}-chelates with serum albumin. By employing cell biological techniques, we measured the influence of VO^{2+}-chelates on the uptake of 2-deoxy[1-^{14}C]glucose, on glycogen synthesis, and on glycogen synthase activity in cultured 3T3-L1 adipocytes. Using a polyclonal anti-phosphotyrosine immunoblotting technique on lysates of 3T3-L1 adipocytes, we measured specificity of binding of VO^{2+}-chelates towards intracellular proteins. We monitored glucose level, heart rate, respiration, blood pressure, blood gases, and pH, in both rats and mice after administration of a large dose (1 mmol/kg) of $VO(acac)_2$ to test of short term toxicity. *In vivo* MRI experiments (n=5) were performed at 4.7 Tesla with rats bearing highly metastatic AT6.1 tumors with 0.15 mmol/kg of $VO(acac)_2$ injected I.V.

Results: Our results demonstrate that VO^{2+}-chelates bind tightly with serum albumin with 1:1 binding stoichiometry. Furthermore, the biochemical results suggest that $VO(acac)_2$ accumulated intracellularly. A high dose of $VO(acac)_2$ did not cause adverse reactions in rats and mice either acutely, or 2 days after injection. The relaxivity for $VO(acac)_2$ of 2.5 ± 0.2 $mM^{-1}s^{-1}$ is comparable to that of Gd-DTPA complex of 4.3 ± 0.2 $mM^{-1}s^{-1}$ at 4.7 Tesla. High-resolution MR images were obtained with $VO(acac)_2$ as a contrast agent. Below, the left panel (**L**) illustrates a pre-contrast image (panel A), control images ([pre-contrast] –[pre-contrast]) (panel B); and difference ([post-contrast] –[pre-contrast]) images (up to T=2 hr after the administration of $VO(acac)_2$; panel C). The uptake and clearance of $VO(acac)_2$ in muscle and tumor is compared in the plots of signal intensity vs. time shown in the right panel (**R**). The kinetic data suggest that in muscle $VO(acac)_2$ remains in the blood pool because it is bound to albumin, but there is preferential leakage from tumor blood vessels. The persistent enhancement in tumors over a period of 3-4 hr is consistent with uptake of vanadyl into tumor cells. Separate experiments showed that Gd-DTPA washout from tumors is much more rapid than that of $VO(acac)_2$.

Discussion: $VO(acac)_2$ provides excellent T_1 and T_2^* contrast; it does not have significant adverse effects in rats or mice; and it provides high-resolution MR images with selective and easily detectable enhancement of AT6.1 rodent tumors. The kinetics of uptake and washout suggest that following leakage from tumor blood vessels it is preferentially taken up by cancer cells. *This is a novel approach to cancer detection and diagnosis, since it has the potential to produce the first MRI contrast agents that are non-toxic and highly sensitive to cancer metabolism*. [Supported by NCI - CA100996].

L



R