# Increased volumetric activity for hyperpolarized DNP solutions

**E. T. Peterson[1], M. G. Erickson[2], S. B. Fain[2], and I. J. Rowland[3]**

[1]Biomedical Engineering, University of Wisconsin - Madison, Madison, WI, United States, [2]Medical Physics, University of Wisconsin - Madison, Madison, WI, United States, [3]Radiology, University of Wisconsin - Madison, Madison, WI, United States

**Introduction:** Maximizing the activity of the polarization for $^{13}C$ labeled compounds is critical for in vivo or in vitro imaging or spectroscopy in hyperpolarized $^{13}C$ studies. In most systems using the dynamic nuclear polarization (DNP) method a larger volume of water containing EDTA is used to extract a concentrated smaller volume (typically less than ≤100 μl) of a frozen solution consisting of the polarized compound mixed with radical. Following extraction of the solution, the resultant volume may be significantly more dilute than desired or too large a volume for safe, rapid injection into a rodent. The result is a significant waste of the labeled compound or an inordinately large bolus depending on the desired application. To avoid this, we have investigated whether the DNP polarizer may, without modification, be used to generate smaller volumes of hyperpolarized solution more appropriate for in vivo studies. The generation of smaller volumes for injection provides a more concentrated solution, which increases SNR for both in vitro and in vivo studies. Alternatively, the approach would also enable smaller amounts of polarized compound to be used. Consequently, this study has explored the possibility of replacing a fraction of the 4ml water typically used for extraction with a chemically and biologically inert compound that is also immiscible with water and possessing a high relative density. In this study we demonstrate that by using a perfluorocarbon to replace a fraction of the water for dissolution of any polarized compound; approximately 1.5 ml of pH adjusted solution with higher polarization per unit volume (volumetric activity), may be routinely prepared.

**Methods:** To demonstrate the approach, modification of the preparation of $^{13}C$ (C-1) labeled pyruvic acid (Cambridge Isotope Laboratories, Andover, MA, USA) for injection has been investigated using the Oxford Instruments DNP polarizer (HyperSense®, Tubney Woods, Abingdon, Oxfordshire, UK). All polarization steps up to the dissolution process remain unchanged from that described in the operating manual. The efficiency of this approach was explored using a range of 0% to 100% perfluorcarbon (FC-72, 3M, St Paul, MN, USA). After loading the mixture of perfluorocarbon and water, the dissolution proceeds normally and the liquids are ejected from the polarizer into a graduated cylinder containing an approximately 300 μl solution of sodium hydroxide and sodium bicarbonate (Sigma-Aldrich, St Louis, MO, USA) for pH adjustment. Once in the graduated cylinder, the mixture rapidly settles into two layers. The dense and immiscible (10 ppm solubility in water and <5 ppm water solubility) perfluorocarbon provides a colorless lower layer. The polarized components in the colored upper layer separate completely and can be drawn off for injection. The effect of an increased radical concentration on the longitudinal relaxation time of the injectable solution was also explored. $T_1$ was measured using a pulse-acquire sequence on a Varian 4.7T imaging system, with varying TR times between 0.5 s and 2 s in order to correctly measure the separate effects of RF and $T_1$ of the polarized compound.

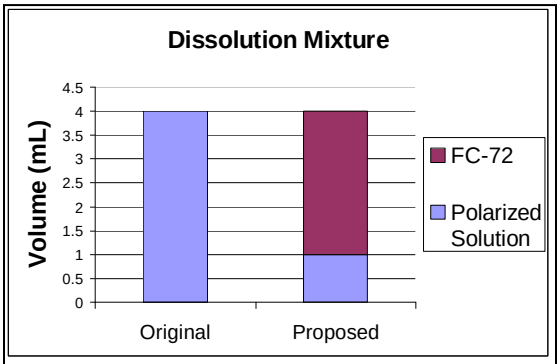


**Figure 1:** Polarizer dissolution output volume showing the relative proportions of polarized solution and FC-72 for the conventional and the mixed solution.

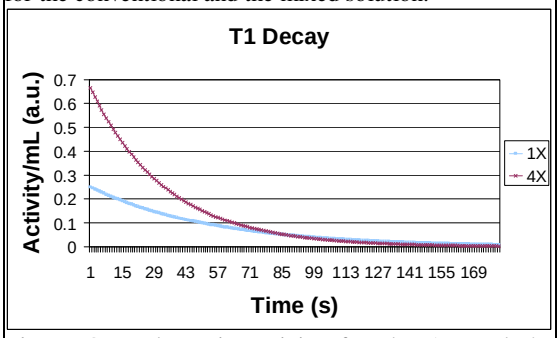


**Figure 2:** Volumetric activity for the 1X and 4X concentration polarized solutions. Note that the volumetric activity for the more concentrated solution remains higher until 86 s post-dissolution.

**Results:** The optimal empirically determined dissolution mixture of 1 ml water and 3 ml perfluorocarbon seen in **Figure 1** will flush all of the polarized compound from the polarizer and still maintain a small injection volume. This results in an injection volume of 1.5 ml after pH compensation. With increasing amounts of perfluorocarbon, the $^{13}C$ $T_1$ is reduced significantly. When 75% perfluorcarbon is used, the $T_1$ of the concentrated solution is reduced from 54 s (0% perfluorcarbon) to 34 s. However, as illustrated in **Figure 2**, the volumetric activity of the reduced volume solution (75% perfluorcarbon) remains higher than the 0% perfluorocarbon solution until 86s post dissolution. At this time, the total polarization of the solution is extremely low, clearly showing the advantages of preparing a more concentrated solution for injection. The above procedure was performed as a standard procedure whenever a polarized contrast agent is to be injected into an animal without any detrimental effects.

**Discussion and Conclusion:** Without physical modification of the DNP polarizer, the standard operating procedure may, with minimal modification, be used to routinely generate volumes of hyperpolarized labeled compound with a concentration tailored to the desired study. The reduced volume solution consisting of perfluorocarbon and water has proved to be a reliable method for reducing the volume and increasing the concentration of injected polarized material, thus decreasing the injection time and creating a more compact injection bolus. The technique is robust and applicable to most polarized materials for *in vivo* or *in vitro* applications.

**Acknowledgements:** NIH/NHLBI R01 HL080412-02, NIH/NHLBI R01 HL069116-06, GE Healthcare