Deuterium and tritium labelling of carbohydrates and glycoconjugates have wide applicability in the bio-chemical and biophysical fields. Carbohydrates function as receptors for a variety of hormones, and play critical roles in cell recognition and differentiation. Labelled carbohydrates may serve as convenient probes into the molecular organization and structural dynamics of micelles and membranes, or as metabolic and biosynthetic probes of cellular activity. Further, deuterium labeled compounds are frequently utilized in the conformational analysis of oligo- and polysaccharides, as well as for glycoproteins, glycolipids, and cardioglycosides, to help alleviate resonance overcrowding in $^1$H NMR spectroscopy.

We are continuing to explore the empirical parameters attendant to the microwave-promoted, stereospecific $^1$H $\rightarrow$ $^2$H catalytic exchange technique using Raney nickel® catalysts that we have developed. This facile technique is mild, tolerant of thermally-sensitive moieties, and since substrate epimerization, racemization, and degradation are absent, product isolation is generally quantitative.

Microwave irradiation reactions are becoming increasingly popular, and several types of microwave apparati have been marketed that facilitate cleaner reactions, higher yields and more uniform results in a shorter reaction time. In a preliminary study, we found that the use of a simple, multi-mode domestic microwave oven for the irradiation of a pre-activated Raney nickel catalyst and a deuterium isotope source readily promoted stereospecific C-H $\rightarrow$ C-D exchange in non-reducing carbohydrates. One model carbohydrate used to evaluate the extent of stereospecific isotopic exchange promoted by microwave irradiation is 1-O-methyl-$\beta$-D-galactopyranoside:

![Galactopyranoside Structure](image)

The percentage incorporation of $^2$H into each of the exchangeable vicinal C-H positions in this simple model, as well as $^2$H incorporation into di-, tri, tetra-, penta- and hexa-1-O-methyl oligosaccharides, were assessed using 720 MHz $^1$H NMR spectroscopy at the National High Magnetic Field Laboratory (NHMFL, Tallahassee, FL). High-field $^1$H NMR is required for all but the simplest of spin-coupled systems, due to the general clustering of resonances within a narrow chemical shift range ($\sim$ 3-5ppm). In general, the relative rates of stereospecific exchange are equatorial-H $>$ axial-H $>$ syn-axial-H. Furthermore, the overall reaction sequence is reversible ($^2$H $\rightarrow$ $^1$H; if H$_2$O is used as the $^1$H isotope source starting with pre-labeled $^2$H substrates). Experiments incorporating $^2$H and $^3$H into complex oligosaccharides (i.e., gangliosides) are currently underway.

Acknowledgements

This work was supported by Pfizer, Incorporated and the University of South Alabama Research Council.

References