Fragment Screening and Fast Nanomolar Detection on a Benchtop NMR Spectrometer Boosted by Photoinduced Hyperpolarization

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Nuclear magnetic resonance (NMR) spectroscopy has a variety of applications in drug discovery such as screening, determination of binding affinity, epitope mapping, and complex structure determination. The low sensitivity of NMR can be overcome by using photo-chemically induced dynamic nuclear polarization (photo-CIDNP), thereby reducing measurement time and consumption of protein and small molecules for screening.

We designed a dedicated photo-CIDNP fragment library of 212 compounds for screening on a high-field (600 MHz) NMR spectrometer, using low micromolar concentrations and single-scan experiments of a few seconds. [1]

The polarization yield obtained by photo-CIDNP increases inversely proportional to the magnetic field, facilitating the use of low-field benchtop magnets. Benchtop NMR spectrometers are about 20-fold cheaper than high-field spectrometers, require little maintenance, and their permanent magnets do not require cryogenic or helium cooling.

We show that photo-CIDNP-based fragment screening is possible on a cryogen-free low-field benchtop NMR spectrometer. We present a photo-CIDNP miniscreen with 30 compounds against the cancer target PIN1 measured on an 80 MHz NMR spectrometer. [2] The experiments were measured in only 3 minutes per sample using 500 µM compound and 10 µM protein concentrations and verified the fast detection of low-millimolar binders. While the concentrations used are comparable to a state-of-the-art NMR screening on high-field, the measurement time could be reduced by 5 to 10-fold. Binding could also be observed at lower concentrations down to 50 µM ligand and 1 µM protein. The detection limit for one compound was 100 nM after 6 minutes. The estimated measurement time at this concentration and field without hyperpolarization would be 450’000 hours.

The performance of screening in comparison to state-of-the-art high-field NMR reveals the advantages of our approach regarding costs and simplicity of execution. These results demonstrate the potential of photoinduced hyperpolarization to enable life science applications on cheap low-field permanent magnets and open the door to broader use of NMR in the drug discovery community.

[1] Felix Torres, Matthias Bütikofer, Gabriela R. Stadler, Alois Renn, Harindranath Kadavath, Raitis Bobrovs, Kristaps Jaudzems, Roland Riek, *J. Am. Chem. Soc.*, **2023**, 145, 22, 12066-12080.
[2] Gabriela Ruth Stadler, Takuya Fabian Segawa, Matthias Bütikofer, Venita Decker, Sandra Loss, Barbara Czarniecki, Felix Torres, Roland Pascal Riek, Fragment Screening and Fast Nanomolar Detection on a Benchtop NMR Spectrometer Boosted by Photoinduced Hyperpolarization (under review).