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Metabolite profiles and their isotopomer distributions can be studied non-invasively in complex mixtures with NMR. The advent of dissolution Dynamic Nuclear Polarization (dDNP) and isotope enrichment add sensitivity and resolution to such metabolic studies. Metabolic pathways and networks can be mapped and quantified if protocols that control and exploit the ex situ signal enhancement are created. We present a sample preparation method, including cell incubation, extraction and signal enhancement, to facilitate reproducible and quantitative dDNP (qdDNP) NMR-based isotope tracer analysis. We further illustrate how qdDNP was applied to gain systematic and novel metabolic phenotypic insights into aggressive cancer cells.

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