Supplementary Information

Accessing Methyl Groups in Proteins via ^{1}H-detected MAS Solid-state NMR Spectroscopy Employing Random Protonation

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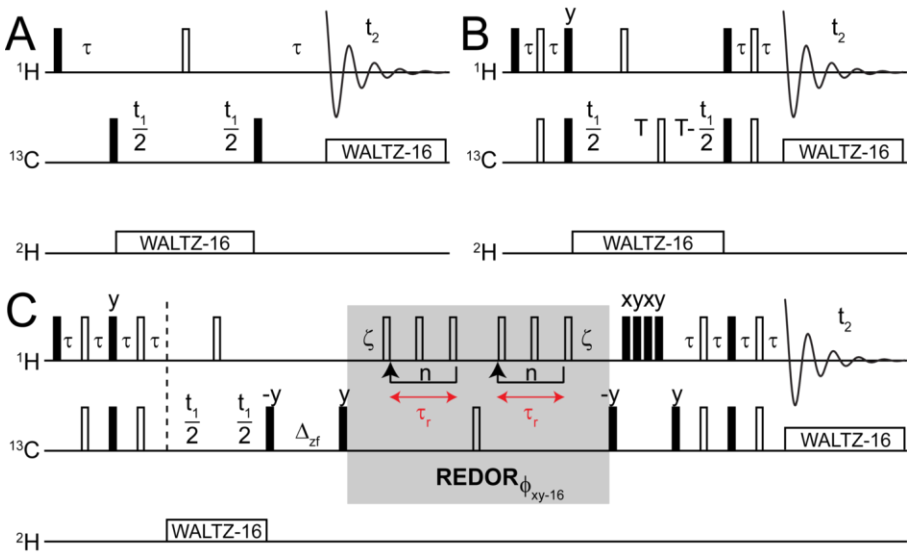

Figure S1. ^{1}H-detected pulse sequences for ^{1}H,^{13}C correlation spectroscopy and determination of ^{1}H,^{13}C dipolar coupling tensors. (A) 2D ^{1}H,^{13}C HMQC pulse sequence with $\tau = 1/2J_{HC} = 3.13$ ms[1]. (B) 2D constant-time HSQC experiment, setting the constant-time delay $T = 1/J_{C,C} = 28.6$ ms and $\tau = 1/4J_{HC} = 1.79$ ms[2]. (C) ^{1}H,^{13}C REDOR pulse sequence[3]. The ^{1}H π pulses during the REDOR period followed the xy-16 phase cycling scheme[4]. The INEPT transfer delay was set to $\tau = 1/4J_{HC} = 1.92$ ms.

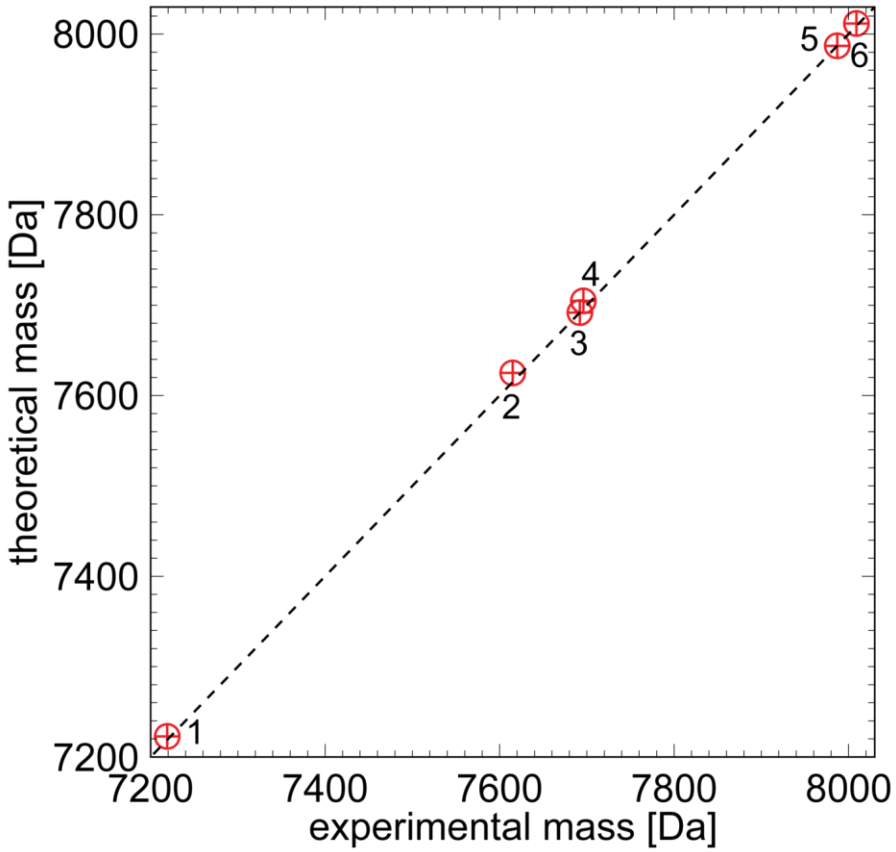

Figure S2. Linear correlation of the experimental to the theoretical mass for differently labelled samples of the SH3 domain of α-spectrin. The samples employed are from left to right: (1) unlabelled, (2) u-[^{1}H,^{13}C,^{15}N], (3) u-[^{2}H,^{15}N], (4) Leu/Val $^{13}CHD_2$ otherwise u-[^{2}H,^{12}C,^{15}N], (5) 5% GlcRAP, (6) u-[^{2}H,^{13}C,^{15}N]. The correlation coefficient R^2 was equal to 0.9998. We yielded an excellent agreement between experimental and theoretical masses, further validating the *in silico* models employed here.

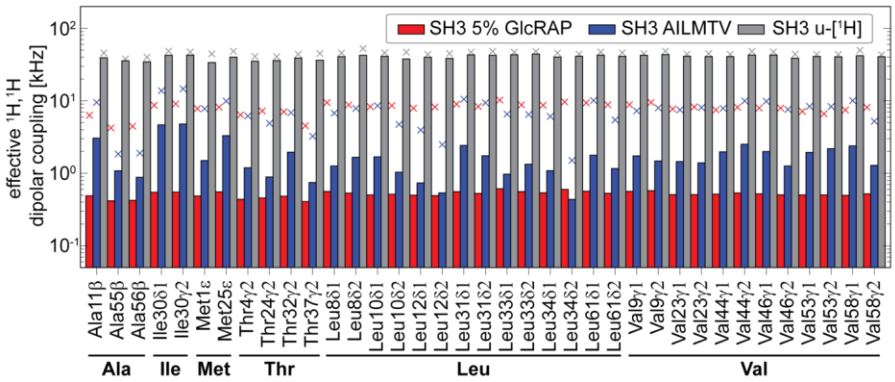

Figure S3. *In silico* calculated ^{1}H,^{1}H effective dipolar coupling for methyl groups based on the 1 μs MD relaxed crystal structure of the SH3 domain of α-spectrin (note the logarithmic y-scale). Here, calculations were carried out for the structures according to the 5% GlcRAP labelling scheme (red bars), the selective Alaβ/Ileγ2,δ1/Metε/Thrγ2/Leuδ1,δ2/Valγ1,γ2 (AILMTV) $^{13}CHD_2$ methyl labelling scheme (blue) and for the uniformly protonated structure (grey). Crosses depict the upper 2σ confidence interval. We note, that for the AILMTV labelling scheme we assumed the $^{13}CHD_2$ isotopomer for all methyl groups. The composition of methyl groups in the SH3 domain is as follows (occurrence is given in parentheses): Ala (3), Ile (1), Leu (7), Met (2), Thr (4), Val (6).

References

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