Supplementary Information

Statistic Evaluation of Simulated NMR Data of Flexible Molecules

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This supplementary contains the MDOC parameters and NMR data of the belizentrin precursor. The data for sagittamide A are collected in the supplementary of the paper of Sternberg *et al.*[1] and the data of the 1,4-diketone and of mandelalide A in the supplementary of the paper of Farès *et al.*[2]

Table S1: General parameters for the MDOC simulations

Parameter	Value
Target temperature	280 K
MD time step	0.5 fs
BPT atomic charge calculation	At start
Coupling time η to the heat bath	0.02 ps
Memory decay time τ for the property average	200 ps
Time constant ρ for the exponential rise of pseudo-forces	200 ps
Order parameter of the alignment medium S_{am}	0.01
Enhancement factor for the torsion of π-bonds*	2
Use error as width	true

** The torsion barrier for π-bonds with valence 2.0 is stepped up from 94.403 kJ/mol to 188.806 kJ/mol. This enhances the stiffness of π-systems and prevents too large distortions by orientational pseudo-forces.*

Table S2: Width and weight parameters for the pseudo forces[1)].

Belizentrin Derivate MDOC Simulations	
Parameter	Value
Pseudo-force width *ΔD* for the one bond CH couplings	Exp. errors as width / Hz
Weight parameter k_D (one bond - in kJ mol^{-1} Hz^{-1})	0.0006
Pseudo-force width *ΔD* for the one long range couplings	Exp. errors as width / Hz
Weight parameter k_D (long range - in kJ mol^{-1} Hz^{-1})	0.0006
Pseudo-force width *ΔR* for the NOE distance constraints	Exp. errors as width /Å
Weight parameter k_R (NOE distances - in kJ mol^{-1}Å^{-1})	0.25
Pseudo-force width *ΔJ* for the $^3J_{HH}$ coupling constraints	Exp. errors as width / Hz
Weight parameter k_J ($^3J_{HH}$ couplings - in kJ mol^{-1}Hz^{-1})	6.0
Total MD duration	20 ns

1) U. Sternberg, P. Tzvetkova, and C. Muhle-Goll, The Simulation of NMR Data of Flexible Molecules - Sagittamide A as Example for MD Simulations with Orientational Constraints; Phys. Chem. Chem. Phys., 2020, DOI: 10.1039/D0CP01905D.

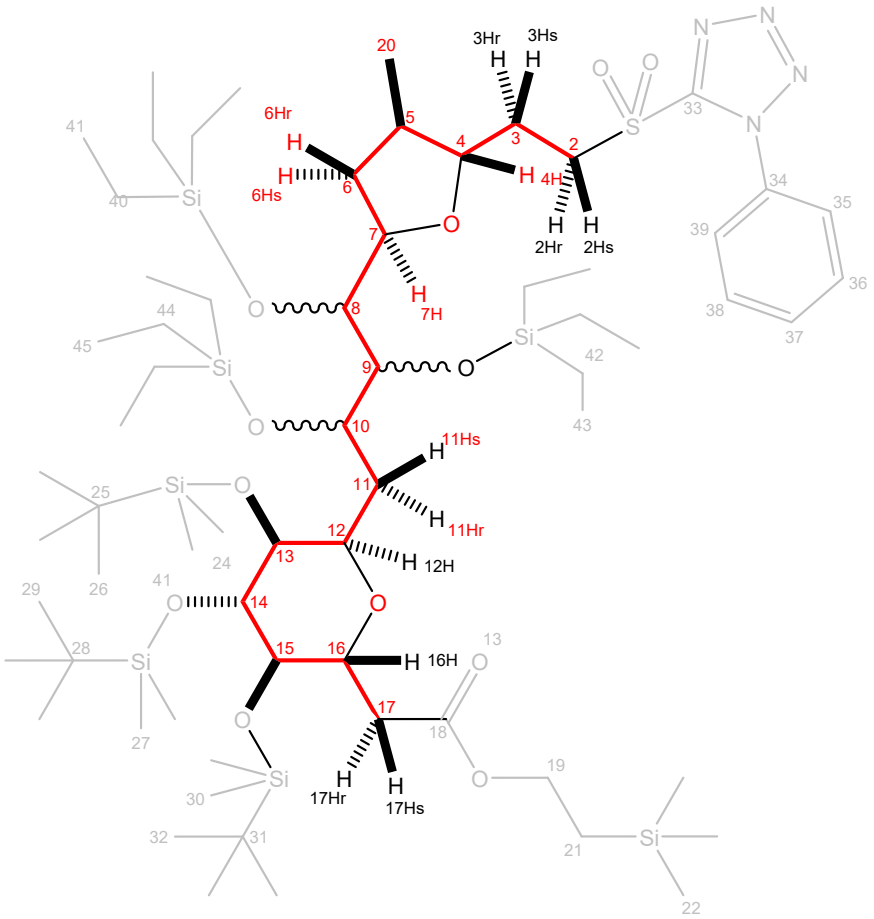

Figure S1: Derivate of belizentrin precursor with (trimethylsilyl)ethyl, triethylsilyl, and *tert*-butyldimethylsilyl protecting groups. The CH_2 carbon atoms of the triethylsiliyl groups are denoted using big letters A, B and C and the protons using small letters a, b and c.

Table S3: Experimental RDC values and MDOC simulated values of the derivate of belizentrin precursor in C8(R), C9(R) and C10(S) configuration. Only values connected to the linker carbons C8-C11 and protons are included. Outlier $(1/\chi)^2 < 1$ in red.

Atom A	Atom B	RDC / Hz Exp.	RDC / Hz MDOC Sim.	Error / Hz	Diff. / Hz Exp.-Sim.	$(1/\chi_i)^2$
H10	C10	1.67	1.6806	0.47	-0.011	1954.907
H11proR	C11	6.56	6.2216	0.31	0.338	0.839
H11proR	H11proS	6.88	6.7909	1.04	0.089	136.315
H11proR	H12	0.4	0.2116	0.4	0.188	4.508
H11proS	C11	5.71	5.2422	0.41	0.468	0.768
H9	C9	-0.08	-0.2441	0.54	0.164	10.834
H8	C8	8.66	8.2555	0.37	0.404	0.837
H43Bc	C43B	-0.49	-0.4813	0.41	-0.009	2208.483
H43Ba	C43B	-0.49	-0.4759	0.41	-0.014	845.968
H43Bb	C43B	-0.49	-0.4833	0.41	-0.007	3725.98
H43Aa	C43A	-0.49	-0.4818	0.41	-0.008	2522.946
H43Ab	C43A	-0.49	-0.4766	0.41	-0.013	936.743
H43Ac	C43A	-0.49	-0.4877	0.41	-0.002	30891.805
H43Cc	C43C	-0.49	-0.4786	0.41	-0.011	1290.007
H43Ca	C43C	-0.49	-0.4823	0.41	-0.008	2805.45
H43Cb	C43C	-0.49	-0.4903	0.41	0	1.52367E6
H45Cc	C45C	-0.38	-0.3763	0.45	-0.004	14521.457
H45Cb	C45C	-0.38	-0.3727	0.45	-0.007	3771.893
H45Ca	C45C	-0.38	-0.3809	0.45	1E-3	269508.188
H45Ac	C45A	-0.38	-0.3719	0.45	-0.008	3080.861
H45Aa	C45A	-0.38	-0.372	0.45	-0.008	3134.721
H45Ab	C45A	-0.38	-0.3827	0.45	0.003	28823.498

H45Bc	C45B	-0.38	-0.3762	0.45	-0.004	14354.966
H45Ba	C45B	-0.38	-0.3753	0.45	-0.005	9031.995
H45Bb	C45B	-0.38	-0.3767	0.45	-0.003	19062.629
H41Aa	C41A	-0.11	-0.1082	0.17	-0.002	8457.638
H41Ac	C41A	-0.11	-0.1049	0.17	-0.005	1129.193
H41Ab	C41A	-0.11	-0.1096	0.17	0	221504.562
H41Ca	C41C	-0.11	-0.102	0.17	-0.008	453.156
H41Cb	C41C	-0.11	-0.1072	0.17	-0.003	3796.396
H41Cc	C41C	-0.11	-0.1099	0.17	0	1.9716E6
H41Bc	C41B	-0.11	-0.1124	0.17	0.002	4907.831
H41Ba	C41B	-0.11	-0.1101	0.17	0	7.03012E6
H41Bb	C41B	-0.11	-0.1026	0.17	-0.007	527.27

Nr. Values/χ^2: 34/4.022 = 8.454 Number of outliers: 3

Minimum 1/ χ_i^2: 0.768206

Table S4: Experimental NOE distances and MDOC simulated values of the derivate of belizentrin precursor in C8(R), C9(R) and C10(S) configuration. Only values of linker carbons C8-C11 and protons are included. Outlier $(1/\chi)^2 < 1$ in red.

Atom A	Atom B	NOE Dist. / Å Exp.	NOE Dist. / Å MDOC Sim.	Error / Å	Diff. / Å Exp.-Sim.	$(1/\chi_i)^2$
H7	H8	3.3	2.704	0.8	0.596	1.803
H4	H8	3.6	3.518	0.7	0.082	72.396
H10	H11proR	2.8	2.6	0.7	0.2	12.306
H10	H11proS	2.9	2.632	0.7	0.268	6.812
H10	H12	3.2	3.028	0.6	0.172	12.135
H10	H17proR	4	4.104	0.7	-0.104	45.643
H10	H8	3.2	3.027	0.6	0.173	12.01
H10	H26Ba	6.8	6.522	1.6	0.278	33.237
H10	H26Bb	6.8	7.323	1.6	-0.523	9.364
H10	H26Bc	6.8	6.306	1.6	0.494	10.505
H10	H26Ca	6.8	6.595	1.6	0.205	60.835
H10	H26Cb	6.8	6.976	1.6	-0.176	82.639
H10	H26Cc	6.8	6.397	1.6	0.403	15.793
H10	H26Aa	6.8	6.89	1.6	-0.09	314.111
H10	H26Ab	6.8	6.701	1.6	0.099	261.252
H10	H26Ac	6.8	6.465	1.6	0.335	22.855
H10	H42Aa	4.4	4.379	1.1	0.021	2700.684
H10	H42Ab	4.4	4.366	1.1	0.034	1075.311
H10	H42Ca	4.4	4.425	1.1	-0.025	1989.449
H10	H42Cb	4.4	4.422	1.1	-0.022	2431.587
H10	H42Bb	4.4	4.498	1.1	-0.098	125.589
H10	H42Ba	4.4	4.478	1.1	-0.078	198.384
H10	H44Ba	3.6	3.537	0.9	0.063	202.497
H10	H44Bb	3.6	3.588	0.9	0.012	5967.334
H10	H44Ab	3.6	3.631	0.9	-0.031	823.843
H10	H44Aa	3.6	3.555	0.9	0.045	405.702
H10	H44Ca	3.6	3.575	0.9	0.025	1259.006
H10	H41Aa	5.1	5.292	1.2	-0.192	39.043
H10	H41Ac	5.1	5.3	1.2	-0.2	35.931
H10	H41Ab	5.1	5.325	1.2	-0.225	28.555
H10	H41Ca	5.1	5.295	1.2	-0.195	37.946
H10	H41Ca	5.1	5.303	1.2	-0.203	34.869
H10	H41Cc	5.1	5.322	1.2	-0.222	29.272
H10	H41Bc	5.1	5.3	1.2	-0.2	35.9
H10	H41Ba	5.1	5.302	1.2	-0.202	35.347
H10	H41Bb	5.1	5.318	1.2	-0.218	30.275
H11proR	H8	2.8	2.926	0.5	-0.126	15.756
H11proR	H44Ba	3.8	3.764	0.9	0.036	619.785
H11proR	H44Bb	3.8	3.77	0.9	0.03	912.898
H11proR	H44Ab	3.8	3.785	0.9	0.015	3822.26
H11proR	H44Aa	3.8	3.754	0.9	0.046	375.162
H11proR	H44Ca	3.8	3.724	0.9	0.076	140.933

H11proR	H41Aa	6	6.055	1.4	-0.055	636.927
H11proR	H41Ac	6	6.059	1.4	-0.059	567.098
H11proR	H41Ab	6	6.071	1.4	-0.071	390.935
H11proR	H41Ca	6	6.038	1.4	-0.038	1338.421
H11proR	H41Cb	6	6.047	1.4	-0.047	880.089
H11proR	H41Cc	6	6.06	1.4	-0.06	544.366
H11proR	H41Bc	6	6.06	1.4	-0.06	547.375
H11proR	H41Ba	6	6.076	1.4	-0.076	336.356
H11proR	H41Bb	6	6.079	1.4	-0.079	314.792
H11proS	H12	2.8	2.551	0.7	0.249	7.878
H11proS	H13	2.8	2.708	0.5	0.092	29.633
H11proS	H15	3.5	5.225	0.6	-1.725	0.121
H11proS	H29Bc	7.2	6.852	1.7	0.348	23.832
H11proS	H29Bb	7.2	7.81	1.7	-0.61	7.768
H11proS	H29Ba	7.2	6.826	1.7	0.374	20.646
H11proS	H29Aa	7.2	7.39	1.7	-0.19	80.1
H11proS	H29Ac	7.2	7.17	1.7	0.03	3158.117
H11proS	H29Ab	7.2	6.893	1.7	0.307	30.638
H11proS	H29Ca	7.2	6.809	1.7	0.391	18.889
H11proS	H29Cb	7.2	7.107	1.7	0.093	337.567
H11proS	H29Cc	7.2	7.393	1.7	-0.193	77.428
H11proS	H42Aa	4.1	4.115	1	-0.015	4406.154
H11proS	H42Ab	4.1	4.091	1	0.009	11297.828
H11proS	H42Ca	4.1	4.079	1	0.021	2372.369
H11proS	H42Cb	4.1	4.062	1	0.038	680.359
H11proS	H42Bb	4.1	4.026	1	0.074	184.583
H11proS	H42Ba	4.1	4.063	1	0.037	728.432
H11proS	H44Ba	3.9	3.828	0.9	0.072	157.857
H11proS	H44Bb	3.9	3.762	0.9	0.138	42.46
H11proS	H44Ab	3.9	3.724	0.9	0.176	26.158
H11proS	H44Aa	3.9	3.813	0.9	0.087	106.887
H11proS	H44Ca	3.9	3.769	0.9	0.131	46.879
H11proS	H41Aa	6.7	6.468	1.6	0.232	47.418
H11proS	H41Ac	6.7	6.468	1.6	0.232	47.52
H11proS	H41Ab	6.7	6.482	1.6	0.218	53.928
H11proS	H41Ca	6.7	6.465	1.6	0.235	46.495
H11proS	H41Cb	6.7	6.468	1.6	0.232	47.449
H11proS	H41Cc	6.7	6.48	1.6	0.22	53.019
H11proS	H41Bc	6.7	6.421	1.6	0.279	32.775
H11proS	H41Ba	6.7	6.446	1.6	0.254	39.658
H11proS	H41Bb	6.7	6.452	1.6	0.248	41.777
H8	H6proR	2.8	2.759	0.5	0.041	150.492
H7	H8	3.3	2.704	0.8	0.596	1.803

Nr. Values/χ^2: 84/10.819 = 7.764 Number of outliers: 1(red)

Minimum 1/ χ^2: 0.120995

Table S5: Experimental $^3J_{HH}$ and MDOC simulated values of the derivate of belizentrin precursor in C8(R), C9(R) and C10(S) configuration. Only values of linker carbons C8-C11 and protons are included. Outlier $(1/\chi)^2 < 1$ in red.

Atom A	Atom B	$^3J_{HH}$ coup./ Hz Exp.	$^3J_{HH}$ coup./ Hz MDOC sim.	Error / Hz	Diff. / Hz Sim.- Exp.	$(1/\chi_i)^2$
H7	H8	7.3	7.34	1	0.04	629.883
H10	H9	4.3	4.372	1.1	0.072	236.675
H10	H11proS	8.2	8.132	1.1	-0.068	259.334
H10	H11proR	1.3	2.817	1.1	1.517	0.526
H11proS	H12	1.3	3.257	1.3	1.957	0.441
H9	H8	3.4	4.06	1.2	0.66	3.307

Nr. Values/χ^2: 6/4.480 = 1.339 Number of outliers: 2 (red)

Minimum 1/ χ^2: 0.441295

Table S6: χ-probabilities and n/χ^2 values from MDOC simulation of 8 linker configurations of C8, C9 and C10 of the derivate of belizentrin and 8 linker configurations with interchanged assignment of NMR data to the prochiral protons H11proS and H11proR. Only values of linker carbons C8-C11 and protons are included. Probabilities larger one in color (color scheme as used for the columns in Fig. 7 of the paper)

Linker Conf.	NMR Data Ass.[1)]	Probability % RDC	Probability % RDC+ NOE-R	Probability % RDC+NOE-R+ $^3J_{HH}$	n/χ^2 RDC	n/χ^2 RDC+ NOE-R	n/χ^2 RDC+ NOE-R+ $^3J_{HH}$
RRR	Original	0.000111954	0.	$2.4451*10^{-9}$	6.82507	6.00733	4.80282
RRS	Original	0.0207417	100.	99.9999	8.45419	10.3051	7.78358
RSR	Original	0.103971	0.	$3.24923*10^{-9}$	9.07945	5.83469	4.97735
RSS	Original	14.3816	0.	$2.32981*10^{-7}$	12.3442	6.23716	5.19537
SRR	Original	78.1138	0.	$2.63553*10^{-7}$	14.103	6.53878	5.26571
SRS	Original	0.000781544	0.	$3.71121*10^{-13}$	7.30243	5.83564	4.57441
SSR	Original	0.291541	0.	0.0000117827	9.80178	6.46294	5.39065
SSS	Original	6.83822	0.	$2.84426*10^{-11}$	12.0915	6.16841	4.72633
RRR	Ex11	0.000946909	0.	$1.48275*10^{-6}$	7.19894	6.27571	5.27176
RRS	Ex11	0.00195017	0.	$1.0623*10^{-10}$	7.67583	6.28445	4.66083
RSR	Ex11	0.0225336	0.	$2.83727*10^{-6}$	8.46939	6.42406	5.49821
RSS	Ex11	0.110606	0.	$1.01223*10^{-8}$	9.0976	6.40655	5.14541
SRR	Ex11	0.0108537	0.	$8.45117*10^{-13}$	8.22427	6.19789	4.43679
SRS	Ex11	$1.9048*10^{-9}$	0.	$2.35494*10^{-16}$	4.68119	5.41305	4.00337
SSR	Ex11	0.0131899	0.	$8.20619*10^{-11}$	8.10673	6.2535	4.69297
SSS	Ex11	0.0891395	0.	$3.48751*10^{-10}$	9.09274	6.41119	4.72253
σ[2)]		0.33	0.40	0.45			

[1)] Assignment of NMR to the prochiral protons H11proS and H11proR : Original - data as given in Tables S3, S4 and S5. Ex11 – prochiral assignment of the two protons H11proS and H11proR exchanged.

[2)] Standard deviation σ of the χ values as used for the χ-probability calculation

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[1] Sternberg, U.; Tzvetkova, P.; Muhle-Goll, C., The simulation of NMR data of flexible molecules: sagittamide A as an example for MD simulations with orientational constraints. *Phys Chem Chem Phys* **2020,** *22* (30), 17375-17384.

[2] Farès, C.; Lingnau, J. B.; Wirtz, C.; Sternberg, U., Conformational Investigations in Flexible Molecules Using Orientational NMR Constraints in Combination with (3)J-Couplings and NOE Distances. *Molecules* **2019,** *24* (23).