

Supporting information

Polybasic peptide-levofloxacin conjugates potentiate fluoroquinolones and other classes of antibiotics against multidrug-resistant Gram-negative bacteria

Liam Berry^a, Ronald Domalaon^a, Marc Brizuela^a, George G. Zhanel^b, Frank Schweizer^{a,b*}

^a Department of Chemistry, University of Manitoba, Winnipeg, MB, Canada

^b Department of Medical Microbiology and Infectious Diseases, University of Manitoba, Winnipeg, MB, Canada

* Address correspondence to Professor Frank Schweizer, email: schweize@cc.umanitoba.ca.

Biological Activity

Table S1: Antibacterial activity of peptide-levofloxacin conjugates against a panel of wild-type and multidrug-resistant clinical isolates of Gram-positive and Gram-negative bacteria.

Organism	Minimum inhibitory concentration (MIC) (µg/ml)												
	1	2	3	4	5	6	7	8	9	10	11	12	LVX
<i>S. aureus</i> ATCC 29213	>128	>128	>128	>128	>128	>128	>128	>128	128	128	64	>128	≤0.25
MRSA ATCC 33592	>128	>128	>128	>128	>128	>128	>128	>128	>128	>128	128	>128	≤0.25
MSSE CANWARD-2008 81388	>128	>128	>128	128	>128	>128	>128	>128	64	64	64	>128	≤0.25
MRSE CAN-ICU 61589	>128	>128	>128	>128	>128	>128	>128	>128	128	128	128	>128	>128
<i>S. pneumoniae</i> ATCC 49619	>128	>128	>128	128	>128	>128	>128	>128	>128	>128	>128	>128	1
<i>E. faecalis</i> ATCC 29212	>128	>128	>128	>128	>128	>128	>128	>128	>128	>128	>128	>128	2
<i>E. faecium</i> ATCC 27270	>128	>128	>128	>128	>128	>128	>128	>128	>128	>128	>128	>128	4
<i>E. coli</i> ATCC 25922	128	128	128	>128	>128	128	>128	128	>128	64	32	>128	≤0.25
<i>E. coli</i> CAN-ICU 61714	>128	>128	>128	>128	>128	>128	>128	>128	>128	>128	>128	>128	1
<i>E. coli</i> CAN-ICU 63074	>128	>128	>128	>128	>128	>128	>128	>128	>128	>128	>128	>128	1
<i>E. coli</i> CANWARD-2011 97615	>128	>128	>128	>128	>128	>128	>128	>128	>128	>128	>128	>128	16
<i>E. cloacae</i> 117029	>128	>128	128	128	>128	>128	64	>128	128	128	128	128	0.125
<i>P. aeruginosa</i> ATCC 27853	>128	>128	>128	>128	>128	>128	>128	>128	>128	>128	128	>128	1
<i>P. aeruginosa</i> CAN-ICU 62308	>128	>128	>128	>128	>128	>128	>128	>128	>128	>128	>128	>128	4
<i>P. aeruginosa</i> CANWARD-2011 96846	>128	>128	>128	>128	>128	>128	>128	>128	>128	>128	>128	>128	8
<i>P. aeruginosa</i> PAO1	>128	>128	>128	>128	>128	>128	>128	>128	128	128	128	64	0.5
<i>S. maltophilia</i> CAN-ICU 62584	>128	>128	>128	>128	>128	16	>128	>128	>128	>128	>128	>128	4
<i>A. baumannii</i> CAN-ICU 63169	>128	>128	>128	>128	>128	>128	>128	>128	>128	>128	>128	>128	1
<i>A. baumannii</i> ATCC 17978	>128	>128	>128	>128	>128	>128	64	>128	64	128	>128	>128	0.125
<i>K. pneumoniae</i> ATCC 13883	>128	>128	>128	>128	>128	64	>128	>128	>128	>128	>128	>128	≤0.25
<i>K. pneumoniae</i> 116381	>128	>128	>128	>128	>128	>128	>128	>128	>128	>128	>128	>128	128

Table S2: Synergy evaluation for combinations of ciprofloxacin (CIP) and conjugates **10, 11** and **12** against multidrug-resistant *P. aeruginosa*

Organism	Conjugate	MIC _{CIP} [MIC _{combo} (µg/ml)]	MIC _{conjugate} [MIC _{Combo} (µg/ml)]	FIC index	Interpretation	Absolute MIC _{CIP} ^a (µg/ml)	Potentiatio ^b
<i>P. aeruginosa</i> PA259	10	128 [8]	>128 [16]	0.063<x<0.188	Synergy	16	8-fold
	11	128 [8]	>128 [4]	0.063<x<0.094	Synergy	8	16-fold
	12	128 [4]	>128 [4]	0.031<x<0.063	Synergy	4	32-fold
<i>P. aeruginosa</i> PA260	10	32 [4]	>128 [16]	0.125<x<0.188	Synergy	4	8-fold
	11	64 [1]	>128 [16]	0.016<x<0.141	Synergy	4	16-fold
	12	32 [1]	128 [8]	0.094	Synergy	1	32-fold
<i>P. aeruginosa</i> PA262	10	32 [4]	>128 [32]	0.125<x<0.375	Synergy	32	1-fold
	11	32 [4]	>128 [8]	0.125<x<0.188	Synergy	4	8-fold
	12	32 [2]	>128 [16]	0.063<x<0.188	Synergy	4	8-fold
<i>P. aeruginosa</i> PA264	10	32 [4]	>128 [32]	0.125<x<0.375	Synergy	32	1-fold
	11	32 [4]	>128 [8]	0.125<x<0.188	Synergy	4	8-fold
	12	32 [2]	>128 [8]	0.063<x<0.125	Synergy	2	16-fold
<i>P. aeruginosa</i> PA100036	10	32 [4]	>128 [16]	0.125<x<0.250	Synergy	8	4-fold
	11	32 [8]	>128 [16]	0.125<x<0.375	Synergy	16	2-fold
	12	32 [8]	>128 [16]	0.250<x<0.375	Synergy	16	2-fold
<i>P. aeruginosa</i> PA101885	10	64 [8]	>128 [16]	0.125<x<0.250	Synergy	8	8-fold
	11	32 [8]	>128 [4]	0.250<x<0.281	Synergy	8	4-fold
	12	32 [8]	>128 [2]	0.250<x<0.266	Synergy	8	4-fold

^aMIC of FQ in presence of 8 µg/ml conjugate

^bDegree of FQ potentiation in the presence of 8 µg/ml conjugate

Table S3: Synergy evaluation for combinations of levofloxacin (LVX) and conjugates **10, 11** and **12** against multidrug-resistant *P. aeruginosa*

Organism	Conjugate	MIC _{LVX} [MIC _{combo} (µg/ml)]	MIC _{conjugate} [MIC _{Combo} (µg/ml)]	FIC index	Interpretation	Absolute MIC _{LVX} ^a (µg/ml)	Potentiatio ^b
<i>P. aeruginosa</i> PA259	10	256 [8]	>128 [32]	0.031<x<0.281	Synergy	128	2-fold
	11	256 [16]	>128 [8]	0.063<x<0.125	Synergy	16	16-fold
	12	256 [8]	>128 [16]	0.031<x<0.156	Synergy	16	16-fold
<i>P. aeruginosa</i> PA260	10	32 [1]	>128 [16]	0.031<x<0.156	Synergy	2	16-fold
	11	32 [1]	>128 [8]	0.031<x<0.094	Synergy	1	32-fold
	12	32 [0.5]	128 [16]	0.141	Synergy	1	32-fold
<i>P. aeruginosa</i> PA262	10	64 [4]	>128 [16]	0.063<x<0.189	Synergy	32	2-fold
	11	64 [4]	>128 [16]	0.063<x<0.188	Synergy	8	8-fold
	12	64 [4]	>128 [8]	0.063<x<0.125	Synergy	4	16-fold
<i>P. aeruginosa</i> PA264	10	64 [16]	>128 [32]	0.25<x<0.5	Synergy	64	1-fold
	11	64 [8]	>128 [16]	0.125<x<0.25	Synergy	16	4-fold
	12	64 [4]	>128 [4]	0.063<x<0.094	Synergy	4	16-fold
<i>P. aeruginosa</i> PA100036	10	64 [4]	>128 [16]	0.063<x<0.188	Synergy	8	8-fold
	11	64 [8]	>128 [16]	0.125<x<0.25	Synergy	16	4-fold
	12	64 [8]	>128 [32]	0.125<x<0.375	Synergy	16	4-fold
<i>P. aeruginosa</i> PA101885	10	32 [8]	>128 [16]	0.25<x<0.375	Synergy	16	2-fold
	11	32 [8]	>128 [32]	0.25<x<0.5	Synergy	16	2-fold
	12	32 [8]	>128 [32]	0.25<x<0.5	Synergy	16	2-fold

^aMIC of FQ in presence of 8 µg/ml conjugate

^bDegree of FQ potentiation in the presence of 8 µg/ml conjugate

Table S4: Synergy evaluation for combinations of moxifloxacin (MXF) and conjugates **10, 11** and **12** against multidrug-resistant *P. aeruginosa*

Organism	Conjugate	MIC _{MXF} [MIC _{combo}] (µg/ml)	MIC _{conjugate} [MIC _{Combo}] (µg/ml)	FIC index	Interpretation	Absolute MIC _{MXF} ^a (µg/ml)	Potentiatio ^b
<i>P. aeruginosa</i> PA259	10	256 [16]	>128 [16]	0.063<x<0.188	Synergy	128	2-fold
	11	512 [16]	>128 [16]	0.031<x<0.156	Synergy	32	16-fold
	12	256 [16]	>128 [4]	0.063<x<0.094	Synergy	16	16-fold
<i>P. aeruginosa</i> PA260	10	128 [2]	>128 [8]	0.016<x<0.078	Synergy	2	64-fold
	11	128 [2]	>128 [8]	0.016<x<0.078	Synergy	2	64-fold
	12	128 [1]	128 [8]	0.070	Synergy	1	128-fold
<i>P. aeruginosa</i> PA262	10	128 [16]	>128 [32]	0.125<x<0.375	Synergy	64	2-fold
	11	128 [16]	>128 [16]	0.125<x<0.250	Synergy	32	4-fold
	12	128 [8]	>128 [8]	0.063<x<0.125	Synergy	8	16-fold
<i>P. aeruginosa</i> PA264	10	128 [16]	>128 [32]	0.125<x<0.375	Synergy	64	2-fold
	11	128 [16]	>128 [16]	0.125<x<0.250	Synergy	64	2-fold
	12	128 [8]	>128 [8]	0.063<x<0.125	Synergy	8	16-fold
<i>P. aeruginosa</i> PA100036	10	128 [4]	>128 [8]	0.031<x<0.094	Synergy	4	32-fold
	11	128 [8]	>128 [16]	0.063<x<0.188	Synergy	16	8-fold
	12	128 [32]	>128 [32]	0.250<x<0.500	Synergy	128	1-fold
<i>P. aeruginosa</i> PA101885	10	128 [16]	>128 [8]	0.125<x<0.188	Synergy	16	8-fold
	11	128 [32]	>128 [2]	0.250<x<0.266	Synergy	64	2-fold
	12	128 [16]	>128 [8]	0.125<x<0.188	Synergy	16	8-fold

^aMIC of FQ in presence of 8 µg/ml conjugate

^bDegree of FQ potentiation in the presence of 8 µg/ml conjugate

Table S5: Synergy evaluation for combinations of ciprofloxacin (CIP) and conjugates **10**, **11** and **12** against wild-type and multidrug-resistant *E. coli* and *K. pneumoniae*

Organism	Conjugate	MIC _{CIP} [MIC _{combo} (μg/ml)]	MIC _{Conjugate} [MIC _{Combo} (μg/ml)]	FIC index	Interpretation	Absolute MIC _{CIP} ^a (μg/ml)	Potentiatio ^b
<i>E. coli</i> ATCC 25922	10	0.016 [0.004]	64 [32]	0.75	Additive	0.008	2-fold
	11	0.016 [0.004]	64 [16]	0.5	Synergy	0.008	2-fold
	12	0.016 [0.004]	16 [4]	0.5	Synergy	NT	NT
<i>E. coli</i> 107115	10	512 [64]	>128 [32]	0.125<x<0.375	Synergy	512	1-fold
	11	512 [512]	>128 [0.5]	1<x<1.003	Additive	512	1-fold
	12	512 [512]	>128 [0.5]	1<x<1.003	Additive	512	1-fold
<i>E. coli</i> 94393	10	1 [0.5]	64 [16]	0.75	Additive	1	1-fold
	11	1 [0.5]	128 [32]	0.75	Additive	1	1-fold
	12	1 [0.5]	128 [1]	0.508	Additive	0.5	2-fold
<i>E. coli</i> 94474	10	64 [32]	128 [8]	0.5625	Additive	32	2-fold
	11	64 [32]	>128 [8]	0.500<x<0.563	Additive	32	2-fold
	12	64 [16]	>128 [16]	0.250<x<0.125	Synergy	64	1-fold
<i>K. pneumoniae</i> 113250	10	0.125 [0.063]	>128 [0.5]	0.500<x<0.504	Additive	0.063	2-fold
	11	0.063 [0.063]	>128 [0.5]	1<x<1.004	Additive	0.063	1-fold
	12	0.125 [0.063]	>128 [0.5]	0.500<x<0.504	Additive	0.063	2-fold
<i>K. pneumoniae</i> 113254	10	0.063 [0.063]	>128 [0.5]	1<x<1.004	Additive	0.063	1-fold
	11	0.063 [0.032]	>128 [8]	0.500<x<0.563	Additive	0.032	2-fold
	12	0.063 [0.063]	>128 [0.5]	1<x<1.004	Additive	0.063	1-fold
<i>K. pneumoniae</i> 116381	10	512 [128]	>128 [8]	0.250<x<0.313	Synergy	128	4-fold
	11	512 [128]	>128 [16]	0.250<x<0.375	Synergy	256	2-fold

	12	512 [128]	>128 [8]	0.250<x<0.313	Synergy	128	4-fold
--	-----------	-----------	----------	---------------	---------	-----	--------

^aMIC of FQ in presence of 8 µg/ml conjugate

^bDegree of FQ potentiation in the presence of 8 µg/ml conjugate

Table S6: Synergy evaluation for combinations of Levofloxacin (LVX) and conjugates **10**, **11** and **12** against wild-type and multidrug-resistant *E. coli* and *K. pneumoniae*

Organism	Conjugate	MIC _{LVX} [MIC _{combo}] (µg/ml)	MIC _{Conjugate} [MIC _{Combo}] (µg/ml)	FIC index	Interpretation	Absolute MIC _{LVX} ^a (µg/ml)	Potentiation ^b
<i>E. coli</i> ATCC 25922	10	0.031 [0.008]	64 [16]	0.500	Synergy	0.008	2-fold
	11	0.031 [0.008]	64 [8]	0.375	Synergy	0.008	2-fold
	12	0.031 [0.008]	32 [8]	0.500	Synergy	0.008	2-fold
<i>E. coli</i> 107115	10	32 [8]	>128 [2]	0.250<x<0.266	Synergy	8	4-fold
	11	32 [8]	>128 [1]	0.250<x<0.258	Synergy	8	4-fold
	12	32 [8]	>128 [1]	0.250<x<0.258	Synergy	8	4-fold
<i>E. coli</i> 94393	10	1 [0.125]	64 [16]	0.375	Synergy	0.25	4-fold
	11	1 [0.25]	128 [4]	0.281	Synergy	0.25	4-fold
	12	1 [0.25]	128 [4]	0.281	Synergy	0.25	4-fold
<i>E. coli</i> 94474	10	32 [4]	128 [32]	0.375	Synergy	8	4-fold
	11	32 [4]	>128 [16]	0.125<x<0.250	Synergy	8	4-fold
	12	32 [4]	>128 [16]	0.125<x<0.250	Synergy	8	4-fold
<i>K. pneumoniae</i> 113250	10	0.125 [0.031]	>128 [4]	0.250<x<0.281	Synergy	0.031	4-fold
	11	0.125 [0.031]	>128 [2]	0.250<x<0.266	Synergy	0.031	4-fold
	12	0.125 [0.031]	>128 [1]	0.250<x<0.258	Synergy	0.063	2-fold
<i>K. pneumoniae</i> 113254	10	0.063 [0.031]	>128 [4]	0.500<x<0.531	Additive	0.031	2-fold
	11	0.063	>128 [4]	0.500<x<0.531	Additive	0.063	1-fold

		[0.031]					
	12	0.063 [0.031]	>128 [4]	0.500<x<0.531	Additive	0.031	2-fold
<i>K. pneumoniae</i> 116381	10	128 [16]	>128 [16]	0.125<x<0.250	Synergy	32	4-fold
	11	128 [32]	>128 [4]	0.250<x<0.281	Synergy	32	4-fold
	12	128 [32]	>128 [4]	0.250<x<0.281	Synergy	32	4-fold

^aMIC of FQ in presence of 8 µg/ml conjugate

^bDegree of FQ potentiation in the presence of 8 µg/ml conjugate

Table S7: Synergy evaluation for combinations of moxifloxacin (MXF) and conjugates **10**, **11** and **12** against wild-type and multidrug-resistant *E. coli* and *K. pneumoniae*

Organism	Conjugate	MIC _{MXF} [MIC _{combo}] (µg/ml)	MIC _{Conjugate} [MIC _{Combo}] (µg/ml)	FIC index	Interpretation	Absolute MIC _{MXF} ^a (µg/ml)	Potentiatio ^b
<i>E. coli</i> ATCC 25922	10	0.031 [0.004]	64 [16]	0.375	Synergy	0.008	2-fold
	11	0.031 [0.008]	64 [4]	0.313	Synergy	0.008	2-fold
	12	0.063 [0.008]	32 [4]	0.250	Synergy	0.008	4-fold
<i>E. coli</i> 107115	10	32 [2]	>128 [8]	0.063<x<0.125	Synergy	2	16-fold
	11	32 [2]	>128 [8]	0.063<x<0.125	Synergy	2	16-fold
	12	32 [4]	>128 [8]	0.125<x<0.129	Synergy	4	8-fold
<i>E. coli</i> 94393	10	1 [0.125]	64 [8]	0.250	Synergy	0.125	8-fold
	11	1 [0.125]	128 [8]	0.188	Synergy	0.125	8-fold
	12	2 [0.25]	128 [2]	0.141	Synergy	0.25	4-fold
<i>E. coli</i> 94474	10	32 [4]	128 [32]	0.313	Synergy	8	4-fold
	11	32 [4]	>128 [16]	0.125<x<0.188	Synergy	4	8-fold
	12	32 [4]	>128 [16]	0.125<x<0.188	Synergy	4	8-fold
<i>K. pneumoniae</i> 113250	10	0.125 [0.031]	>128 [8]	0.250<x<0.313	Synergy	0.031	4-fold
	11	0.125 [0.125]	>128 [0.5]	1<x<1.004	Additive	0.125	1-fold
	12	0.125 [0.125]	>128 [0.5]	1<x<1.004	Additive	0.125	1-fold
<i>K. pneumoniae</i> 113254	10	0.125 [0.063]	>128 [4]	0.500<x<0.531	Additive	0.063	2-fold
	11	0.125 [0.063]	>128 [2]	0.500<x<0.516	Additive	0.063	2-fold
	12	0.125 [0.063]	>128 [16]	0.500<x<0.625	Additive	0.125	1-fold
<i>K. pneumoniae</i> 116381	10	256 [32]	>128 [4]	0.125<x<0.156	Synergy	32	8-fold
	11	256 [32]	>128 [4]	0.125<x<0.156	Synergy	32	8-fold
	12	256 [32]	>128 [2]	0.125<x<0.141	Synergy	32	8-fold

^aMIC of FQ in presence of 8 µg/ml conjugate

^bDegree of FQ potentiation in the presence of 8 µg/ml conjugate

Table S8: Synergy evaluation for combinations of ciprofloxacin (CIP) and conjugates **10**, **11** and **12** against wild-type and multidrug-resistant *A. baumannii*

Organism	Conjugate	MIC _{CIP} [MIC _{combo} (µg/ml)]	MIC _{conjugate} [MIC _{combo} (µg/ml)]	FIC index	Interpretation	Absolute MIC _{CIP} ^a (µg/ml)	Potentiation ^b
<i>A. baumannii</i> ATCC 17978	10	0.5 [0.5]	128 [0.5]	1.004	Additive	0.5	1-fold
	11	0.5 [0.5]	128 [0.5]	1.004	Additive	0.5	1-fold
	12	05 [0.5]	>128 [0.5]	1<x<1.004	Additive	0.5	1-fold
<i>A. baumannii</i> AB027	10	32 [8]	128 [32]	0.500	Synergy	32	1-fold
	11	32 [16]	128 [2]	0.516	Additive	32	1-fold
	12	32 [16]	>128 [4]	0.500<x<0.531	Additive	32	1-fold
<i>A. baumannii</i> AB031	10	1 [0.5]	32 [0.125]	0.504	Additive	0.5	2-fold
	11	0.5 [0.125]	16 [4]	0.500	Synergy	NT	NT
	12	1 [0.016]	8 [2]	0.375	Synergy	NT	NT
<i>A. baumannii</i> LAC-4	10	32 [16]	16 [0.063]	0.504	Additive	NT	NT
	11	32 [4]	8 [2]	0.375	Synergy	NT	NT
	12	32 [4]	8 [1]	0.250	Synergy	NT	NT

^aMIC of FQ in presence of 8 µg/ml conjugate

^bDegree of FQ potentiation in the presence of 8 µg/ml conjugate

Table S9: Synergy evaluation for combinations of levofloxacin (LVX) and conjugates **10**, **11** and **12** against wild-type and multidrug-resistant *A. baumannii*

Organism	Conjugate	MIC _{LVX} [MIC _{combo}] (µg/ml)	MIC _{conjugate} [MIC _{Combo}] (µg/ml)	FIC index	Interpretation	Absolute MIC _{LVX} ^a (µg/ml)	Potentiatio ^b
<i>A. baumannii</i> ATCC 17978	10	0.125 [0.125]	128 [0.5]	1.004	Additive	0.125	1-fold
	11	0.125 [0.125]	128 [0.5]	1.004	Additive	0.125	1-fold
	12	0.125 [0.125]	>128 [0.5]	1<x<1.004	Additive	0.125	1-fold
<i>A. baumannii</i> AB027	10	8 [2]	128 [32]	0.500	Synergy	4	2-fold
	11	8 [2]	128 [32]	0.500	Synergy	4	2-fold
	12	8 [4]	>128 [0.5]	0.500<x<0.504	Additive	4	2-fold
<i>A. baumannii</i> AB031	10	0.250 [0.063]	32 [8]	0.500	Synergy	0.063	4-fold
	11	0.25 [0.031]	16 [4]	0.375	Synergy	NT	NT
	12	0.25 [0.063]	16 [0.5]	0.281	Synergy	NT	NT
<i>A. baumannii</i> LAC-4	10	2 [1]	16 [4]	0.750	Additive	NT	NT
	11	2 [0.5]	8 [2]	0.500	Synergy	NT	NT
	12	2 [0.25]	8 [2]	0.375	Synergy	NT	NT

^aMIC of FQ in presence of 8 µg/ml conjugate

^bDegree of FQ potentiation in the presence of 8 µg/ml conjugate

Table S10: Synergy evaluation for combinations of moxifloxacin (MXF) and conjugates **10**, **11** and **12** against wild-type and multidrug-resistant *A. baumannii*

Organism	Conjugate	MIC _{MXF} [MIC _{combo}] (µg/ml)	MIC _{conjugate} [MIC _{Combo}] (µg/ml)	FIC index	Interpretation	Absolute MIC _{MXF} ^a (µg/ml)	Potentiatio ^b
<i>A. baumannii</i> ATCC 17978	10	0.063 [0.063]	128 [0.5]	1.004	Additive	0.063	1-fold
	11	0.063 [0.063]	128 [0.5]	1.004	Additive	0.063	1-fold
	12	0.063 [0.063]	>128 [0.5]	1<x<1.004	Additive	0.063	1-fold
<i>A. baumannii</i> AB027	10	4 [1]	128 [16]	0.375	Synergy	2	2-fold
	11	4 [1]	128 [32]	0.500	Synergy	2	2-fold
	12	4 [2]	>128 [0.5]	0.5<x<0.504	Additive	2	2-fold
<i>A. baumannii</i> AB031	10	0.125 [0.031]	32 [8]	0.500	Synergy	0.031	4-fold
	11	0.125 [0.016]	16 [4]	0.375	Synergy	NT	NT
	12	0.125 [0.016]	16 [4]	0.375	Synergy	NT	NT
<i>A. baumannii</i> LAC-4	10	1 [0.5]	16 [4]	0.750	Additive	NT	NT
	11	1 [0.25]	8 [2]	0.500	Synergy	NT	NT
	12	1 [0.25]	8 [2]	0.500	Synergy	NT	NT

^aMIC of FQ in presence of 8 µg/ml conjugate

^bDegree of FQ potentiation in the presence of 8 µg/ml conjugate

Characterization

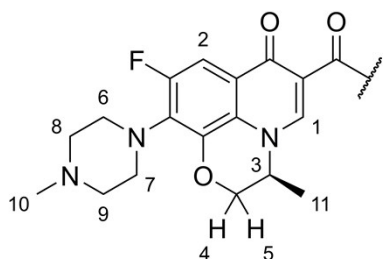


Figure S1. ^1H NMR numbering scheme used for levofloxacin portion of peptide-levofloxacin conjugates.

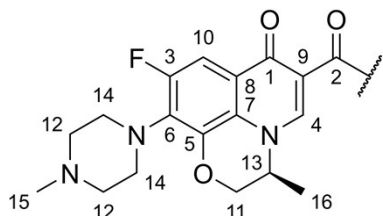


Figure S2. ^{13}C NMR numbering scheme used for levofloxacin portion of peptide-levofloxacin conjugates.

Conjugate 1: ^1H NMR (500 MHz, Methanol- d_4) δ 8.71 (d, J = 16.1 Hz, 1H, Levo-1), 7.55 – 7.38 (m, 1H, Levo-2), 4.75 – 4.68 (m, 2H, Dab- α + Levo-3), 4.56 – 4.51 (m, 1H, Levo-4), 4.43 – 4.38 (m, 1H, Levo-5), 3.64 – 3.56 (m, 6H Levo-6,7,8a,9a), 3.31 – 3.28 (m, 2H Levo8b,9b), 3.13 – 3.02 (m, 2H Dab- γ), 2.99 (s, 3H Levo-10), 2.35 – 2.26 (m, 1H Dab- β 1), 2.15 – 2.06 (m, 1H Dab- β 2), 1.55 (d, J = 6.9 Hz, 3H Levo-11). ^{13}C NMR (126 MHz, Methanol- d_4) δ 174.90(Levo-1), 173.68(Levo-2), 165.62(Levo-3), 156.53(Dab-1 carbonyl), 154.55(Levo-4), 144.96(Levo-5), 140.96(Levo-6), 129.52(Levo-7), 124.33(Levo-8), 109.56(Levo-9), 103.59(Levo-10), 68.42(Levo-11), 55.24(Levo-12), 54.09(Levo-13), 50.29(Dab- α), 47.49(Levo-14), 42.51(Levo-15), 36.31(Dab- γ), 31.03(Dab- β), 16.72(Levo-16). MALDI-TOF-MS m/z calcd for $\text{C}_{22}\text{H}_{29}\text{FN}_6\text{O}_4$ ($\text{M}+\text{H}$) $^+$ monoisotopic peak: 461.2307; found: 461.314

Conjugate 2: ^1H NMR (500 MHz, Methanol- d_4) δ 8.75 – 8.65 (m, 1H, Levo-1), 7.58 – 7.40 (m, 1H, Levo-2), 4.79 – 4.73 (m, 1H Dab $_1$ - α), 4.72 – 4.67 (m, 1H Levo-3), 4.58 – 4.49 (m, 2H Dab $_2$ - α + Levo-4), 4.43 – 4.36 (m, 1H Levo-5), 3.67 – 3.55 (m, 6H Levo-6,7,8a,9a), 3.32 – 3.28 (m, 2H Levo8b,9b), 3.15 – 3.04 (m, 4H Dab $_1$ - γ + Dab $_2$ - γ), 2.98 (s, 3H Levo-10), 2.37 – 2.19 (m, 2H, Dab $_1$ - β 1 + Dab $_2$ - β 1), 2.18 – 2.01 (m, 2H Dab $_1$ - β 2 + Dab $_2$ - β 2), 1.61 – 1.52 (m, 3H Levo-11). ^{13}C NMR (126 MHz, Methanol- d_4) δ 174.99 (Levo-1), 173.76 (Levo-2), 171.78 (Dab-1 carbonyl), 165.70 (Levo-3), 156.61 (Dab-2 carbonyl), 154.65 (Levo-4), 145.10 (Levo-5), 141.02 (Levo-6), 129.57(Levo-7), 124.35(Levo-8), 109.55(Levo-9), 103.65(Levo-10), 68.43(Levo-11), 55.31(Levo-12), 54.09(Levo-13), 50.85(Dab $_1$ - α), 50.77(Dab $_2$ - α), 47.47(Levo-14), 42.52(Levo-15), 36.42(Dab $_1$ - γ), 36.19(Dab $_2$ - γ), 30.48(Dab $_1$ - β), 29.54(Dab $_2$ - β), 16.78(Levo-16). MALDI-TOF-MS m/z calcd for $\text{C}_{26}\text{H}_{37}\text{FN}_8\text{O}_5$ ($\text{M}+\text{H}$) $^+$ monoisotopic peak: 561.2944; found: 561.313

Conjugate 3: ^1H NMR (500 MHz, Methanol- d_4) δ 8.76 – 8.73 (m, 1H, Levo-1), 7.62 – 7.58 (m, 1H, Levo-2), 4.75 – 4.70 (m, 2H, Dab $_1$ - α + Levo-3), 4.55 (dd, J = 11.6, 2.1 Hz, 1H, Dab $_2$ - α), 4.51 (dd, J = 9.2, 4.7 Hz, 1H, Dab $_3$ - α), 4.44 – 4.38 (m, 2H, Levo-4 + Levo-5), 3.63 – 3.55 (m, 6H, Levo-6,7,8a,9a), 3.33 – 3.30 (m, 1H, Levo-8b), 3.29 – 3.24 (m, 1H, Levo-9b), 3.12 – 3.05 (m, 6H, Dab $_1$ - γ + Dab $_2$ - γ + Dab $_3$ - γ), 2.98 (s, 3H, Levo-10), 2.35 – 2.22 (m, 3H, Dab $_1$ - β 1 + Dab $_2$ - β 1, Dab $_3$ - β 1), 2.15 – 2.02 (m, 3H, Dab $_1$ - β 2 + Dab $_2$ - β 2, Dab $_3$ - β 2), 1.55 (d, 3H, Levo-11). ^{13}C NMR (126 MHz, Methanol- d_4) δ 175.13(Levo-1), 173.50(Levo-2), 171.90(Dab-1 carbonyl), 171.65(Dab-2 carbonyl), 165.75(Levo-3), 156.77(Dab-3 carbonyl), 154.82(Levo-4), 145.13(Levo-5), 141.11(Levo-6), 129.64(Levo-7), 124.40(Levo-8), 109.62(Levo-9), 103.77(Levo-10), 68.39(Levo-11), 55.28(Levo-12), 54.07(Levo-13), 51.62(Dab1- α), 50.68(Dab2- α), 50.54(Dab3- α), 47.48(Levo-14), 42.48(Levo-15), 36.34(Dab1- γ), 36.33(Dab2- γ), 36.15(Dab3- γ), 30.30(Dab1- β), 29.81(Dab2- β), 28.82(Dab3- β), 16.77(Levo-16). MALDI-TOF-MS m/z calcd for $\text{C}_{30}\text{H}_{45}\text{FN}_{10}\text{O}_6$ (M+H) $^+$ monoisotopic peak: 661.3580; found: 661.352

Conjugate 4: ^1H NMR (500 MHz, Methanol- d_4) δ 8.78 (s, 1H, Levo-1), 7.69 – 7.64 (m, 1H, Levo-2), 4.75 – 4.68 (m, 2H, Dab $_1$ - α + Levo-3), 4.58 – 4.54 (m, 1H, Dab $_2$ - α), 4.50 – 4.44 (m, 2H, Dab $_3$ - α + Levo-4), 4.42 – 4.37 (m, 2H, Dab $_4$ - α + Levo-5), 3.65 – 3.53 (m, 6H, Levo-6,7,8a,9a), 3.34 – 3.30 (m, 2H, Levo-8b,9b), 3.13 – 3.03 (m, 8H, Dab $_1$ - γ + Dab $_2$ - γ + Dab $_3$ - γ + Dab $_4$ - γ), 2.98 (s, 3H, Levo-10), 2.39 – 2.31 (m, 1H, Dab $_1$ - β 1), 2.29 – 2.20 (m, 3H, Dab $_2$ - β 1 + Dab $_3$ - β 1 + Dab $_4$ - β 1), 2.16 – 2.02 (m, 4H, Dab $_1$ - β 2 + Dab $_2$ - β 2 + Dab $_3$ - β 2 + Dab $_4$ - β 2), 1.54 (d, J = 6.7 Hz, 3H, Levo-11). ^{13}C NMR (126 MHz, Methanol- d_4) δ 175.20(Levo-1), 173.55(Levo-2), 172.06(Dab-1 carbonyl), 171.75(Dab-2 carbonyl), 171.40(Dab-3 carbonyl), 165.95(Levo-3), 156.87(Dab-4 carbonyl), 154.91(Levo-4), 145.17(Levo-5), 141.11(Levo-6), 129.67(Levo-7), 124.42(Levo-8), 109.64(Levo-9), 103.87(Levo-10), 68.39(Levo-11), 55.30(Levo-12), 54.08(Levo-13), 51.66(Dab1- α), 51.14(Dab2- α), 50.83(Dab3- α), 50.70(Dab4- α), 47.68(Levo-14), 42.47(Levo-15), 36.36(Dab1- γ), 36.31(Dab2- γ), 36.28(Dab3- γ), 36.20(Dab4- γ), 30.17(Dab1- β), 29.71(Dab2- β), 29.13(Dab3- β), 28.76(Dab4- β), 16.79(Levo-16). MALDI-TOF-MS m/z calcd for $\text{C}_{34}\text{H}_{53}\text{FN}_{12}\text{O}_7$ (M+H) $^+$ monoisotopic peak: 761.4217; found: 761.411

Conjugate 5: ^1H NMR (500 MHz, Methanol- d_4) δ 8.67 (s, 1H, Levo-1), 7.39 – 7.32 (m, 1H, Levo-2), 4.69 – 4.62 (m, 2H, Levo-3 Glu- α), 4.51 (d, J = 11.5 Hz, 1H, Levo-4), 4.41 (dd, J = 11.6, 2.5 Hz, 1H, Levo-5), 3.63 – 3.56 (m, 6H, Levo-6,7,8a,9a), 3.33 – 3.28 (m, 2H, Levo-8b,9b), 3.00 (s, 3H, Levo-10), 2.46 (dd, J = 12.2, 5.2 Hz, 2H, Glu- γ), 2.27 – 2.20 (m, 1H, Glu- β 1), 2.11 – 2.04 (m, 1H, Glu- β 2), 1.51 (d, J = 6.8 Hz, 3H, Levo-11). ^{13}C NMR (126 MHz, Methanol- d_4) δ 174.84(Levo-1), 173.45(Levo-2), 165.14(Levo-3), 156.45(Glu carbonyl), 154.49(Levo-4), 144.74(Levo-5), 140.91(Levo-6), 129.30(Levo-7), 124.31(Levo-8), 109.88(Levo-9), 103.40(Levo-10), 68.40(Levo-11), 55.10(Levo-12), 54.13(Levo-13), 52.24(Glu- α), 47.51(Levo-14), 42.54(Levo-15), 29.68(Glu- γ), 28.08(Glu- β), 16.70(Levo-16). MALDI-TOF-MS m/z calcd for $\text{C}_{23}\text{H}_{28}\text{FN}_5\text{O}_6$ (M+H) $^+$ monoisotopic peak: 490.2096; found: 490.256

Conjugate 6: ^1H NMR (500 MHz, Methanol- d_4) δ 8.71 – 8.68 (m, 1H, Levo-1), 7.42 – 7.32 (m, 1H, Levo-2), 4.71 – 4.66 (m, 1H, Levo-3), 4.65 – 4.61 (m, 1H, Glu $_1$ - α), 4.54 – 4.49 (m, 1H Levo-4), 4.43 – 4.38 (m, 2H Levo-5 + Glu $_2$ - α), 3.63 – 3.56 (m, 6H, Levo-6,7,8a,9a), 3.32 – 3.27 (m, 2H Levo-8b,9b), 3.00 (s, 3H, Levo-10), 2.48 (t, J = 7.8 Hz, 2H, Glu $_1$ - γ), 2.42 (t, J = 7.7 Hz, 2H, Glu $_2$ - γ), 2.27 – 2.20 (m, 1H Glu $_1$ - β 1), 2.17 – 2.07 (m, 2H Glu $_1$ - β 2 + Glu $_2$ - β 1), 2.00 – 1.92 (m, 1H Glu $_2$ - β 2), 1.52 (d, J = 6.7 Hz, 3H Levo-11). ^{13}C NMR (126 MHz, Methanol- d_4) δ 175.04(Levo-1), 174.90(Levo-2), 174.85(Glu $_1$ -COOH), 172.33(Glu $_2$ -COOH), 172.21(Glu $_1$ -carbonyl), 165.42(Levo-3), 156.48(Glu $_2$ -carbonyl), 154.49(Levo-4), 144.86(Levo-5), 140.89(Levo-6), 129.44(Levo-7), 124.30(Levo-8), 109.83(Levo-9), 103.62(Levo-10), 68.42(Levo-11), 55.14(Levo-12), 54.15(Levo-13), 52.77(Glu $_1$ - α), 52.58(Glu $_2$ - α), 47.52(Levo-14), 42.58(Levo-15), 29.79(Glu $_1$ - γ), 29.66(Glu $_2$ - γ), 27.68(Glu $_2$ - β), 26.76(Glu $_2$ - β), 16.76(Levo-16). MALDI-TOF-MS m/z calcd for $\text{C}_{28}\text{H}_{35}\text{FN}_6\text{O}_9$ (M+H) $^+$ monoisotopic peak: 619.2522; found: 619.244

Conjugate 7: ^1H NMR (500 MHz, Methanol- d_4) δ 8.74 – 8.70 (m, 1H, Levo-1), 7.51 – 7.44 (m, 1H, Levo-2), 4.75 – 4.70 (m, 1H, Levo-3), 4.55 – 4.51 (m, 1H, Levo-4), 4.48 – 4.41 (m, 2H, Glu $_1$ - α + Levo-5), 4.37 – 4.32 (m, 2H, Glu $_2$ - α + Glu $_3$ - α), 3.63 – 3.57 (m, 6H, Levo-6,7,8a,9a), 3.32 – 3.29 (m, 2H, Levo-8b,9b), 3.00 (s, 3H, Levo-10), 2.53 – 2.48 (m, 2H, Glu $_1$ - γ), 2.46 – 2.39 (m, 4H, Glu $_2$ - γ + Glu $_3$ - γ), 2.24 – 2.10 (m, 4H, Glu $_1$ - β + Glu $_2$ - β 1 + Glu $_3$ - β 1), 2.05 – 1.95 (m, 2H Glu $_2$ - β 2 + Glu $_3$ - β 2), 1.51 (d, J = 6.8 Hz, 3H, Levo-11). ^{13}C NMR (126 MHz, Methanol- d_4) δ 175.20(Levo-1), 175.09(Levo-2), 175.01(Glu $_1$ -COOH), 174.93(Glu $_2$ -COOH), 174.72(Glu $_3$ -COOH), 173.12(Glu $_1$ -carbonyl), 172.37(Glu $_2$ -carbonyl), 165.92(Levo-3), 159.04(Glu $_3$ -carbonyl), 154.65(Levo-4), 144.95(Levo-5), 140.94(Levo-6), 129.49(Levo-7), 124.36(Levo-8), 109.83(Levo-9), 103.76(Levo-10), 68.40(Levo-11), 55.19(Levo-12), 54.15(Levo-13), 53.64(Glu $_1$ - α), 53.52(Glu $_2$ - α), 52.78(Glu $_3$ - α), 47.48(Levo-14), 42.57(Levo-15), 30.12(Glu $_1$ - γ), 29.84(Glu $_2$ - γ), 29.68(Glu $_3$ - γ), 27.13(Glu $_1$ - β), 26.79(Glu $_2$ - β), 25.90(Glu $_3$ - β), 16.90(Levo-16). MALDI-TOF-MS m/z calcd for $\text{C}_{33}\text{H}_{42}\text{FN}_7\text{O}_{12}$ (M+H) $^+$ monoisotopic peak: 748.2948; found: 748.295

Conjugate 8: ^1H NMR (500 MHz, Methanol- d_4) δ 8.77 – 8.71 (m, 1H, Levo-1), 7.55 – 7.47 (m, 1H, Levo-2), 4.76 – 4.70 (m, 1H, Levo-3), 4.57 – 4.51 (m, 1H, Levo-4), 4.48 – 4.39 (m, 2H, Glu $_1$ - α + Levo-5), 4.37 – 4.29 (m, 3H, Glu $_2$ - α + Glu $_3$ - α , Glu $_4$ - α), 3.65 – 3.57 (m, 6H, Levo-6,7,8a,9a), 3.35 – 3.30 (m, 2H, Levo-8b,9b), 3.01 (s, 3H, Levo-10), 2.54 – 2.40 (m, 8H, Glu $_1$ - γ + Glu $_2$ - γ + Glu $_3$ - γ + Glu $_4$ - γ), 2.26 – 2.08 (m, 6H, , Glu $_1$ - β + Glu $_2$ - β + Glu $_3$ - β 1 + Glu $_4$ - β 1), 2.04 – 1.95 (m, 2H, Glu $_3$ - β 2 + Glu $_4$ - β 2), 1.50 (d, J = 6.7 Hz, 3H, Levo-11). ^{13}C NMR (126 MHz, Methanol- d_4) δ 175.22(Levo-1), 175.18(Levo-2), 175.15(Glu $_1$ -COOH), 175.08(Glu $_2$ -COOH), 175.03(Glu $_3$ -COOH), 175.02(Glu $_4$ -COOH), 174.72(Glu $_1$ -carbonyl), 173.49(Glu $_2$ -carbonyl), 172.96(Glu $_3$ -carbonyl), 172.43(Glu $_4$ -carbonyl), 166.19(Levo-3), 154.66(Levo-4), 145.07(Levo-5), 140.97(Levo-6), 129.56(Levo-7), 124.37(Levo-8), 109.75(Levo-9), 103.79(Levo-10), 68.42(Levo-11), 55.22(Levo-12), 54.17(Levo-13), 54.08(Glu $_1$ - α), 54.01(Glu $_2$ - α), 53.54(Glu $_3$ - α), 52.80(Glu $_4$ - α), 47.77(Levo-14), 42.63(Levo-15), 30.20(Glu $_1$ - γ), 29.95(Glu $_2$ - γ), 29.86(Glu $_3$ - γ), 29.74(Glu $_4$ - γ), 26.96(Glu $_1$ - β), 26.67(Glu $_2$ - β), 26.47(Glu $_3$ - β), 25.69(Glu $_4$ - β), 16.96(Levo-16). MALDI-TOF-MS m/z calcd for $\text{C}_{38}\text{H}_{50}\text{FN}_8\text{O}_{15}$ (M+H) $^+$ monoisotopic peak: 877.3374; found: 877.377

Conjugate 9: ^1H NMR (500 MHz, Methanol- d_4) δ 8.73 (s, 1H, Levo-1), 7.60 (d, $J = 12.2$ Hz, 1H, Levo-2), 4.72 – 4.68 (m, 1H, Levo-3), 4.54 (dd, $J = 11.5, 2.2$ Hz, 1H, Levo-4), 4.46 (dd, $J = 8.6, 5.5$ Hz, 1H, Dab- α), 4.41 (dd, $J = 11.6, 2.6$ Hz, 1H, Levo-5), 3.63 – 3.55 (m, 6H Levo-6,7,8a,9a), 3.42 – 3.38 (m, 2H Aliphatic), 3.30 – 3.24 (m, 2H Levo8b,9b), 3.02 – 2.96 (m, 5H, Dab- γ + Levo-10), 2.26 (t, $J = 8.3, 6.9$ Hz, 2H, Aliphatic), 2.20 – 2.14 (m, 1H Dab- β 1), 2.00 – 1.94 (m, 1H, Dab- β 2), 1.63 – 1.58 (m, 4H, Aliphatic), 1.53 (d, $J = 6.8$ Hz, 3H Levo-11), 1.42 – 1.27 (m, 16H, Aliphatic). ^{13}C NMR (126 MHz, Methanol- d_4) δ 175.21(Levo-1), 174.00(Levo-2), 165.30(Levo-3), 156.70(Dab-1 carbonyl), 154.75(Levo-4), 144.70(Levo-5), 141.03(Aliphatic-carbonyl) 140.99(Levo-6), 129.45(Levo-7), 124.44(Levo-8), 110.32(Levo-9), 103.75(Levo-10), 103.56, 68.38(Levo-11), 55.14(Levo-12), 54.09(Levo-13), 50.33(Dab- α), 47.50(Levo-14), 42.50(Levo-15), 38.74(Aliphatic), 36.40(Dab- γ), 35.35(Aliphatic), 29.71(Dab- β), 29.18(Aliphatic), 29.13(Aliphatic), 29.01, 28.94, 28.93, 26.69, 25.35, 16.77(Levo-16). MALDI-TOF-MS m/z calcd for $\text{C}_{34}\text{H}_{52}\text{FN}_7\text{O}_5$ (M+H) $^+$ monoisotopic peak: 658.4087; found: 658.388

Conjugate 10: ^1H NMR (500 MHz, Methanol- d_4) δ 8.74 (s, 1H, Levo-1), 7.65 – 7.61 (m, 1H, Levo-2), 4.73 – 4.68 (m, 1H, Levo-3), 4.54 (dd, $J = 11.6, 2.2$ Hz, 1H, Levo-4), 4.47 (dd, $J = 9.1, 4.9$ Hz, 1H, Dab $_1$ - α), 4.42 – 4.37 (m, 2H, Levo-5 + Dab $_2$ - α), 3.65 – 3.55 (m, 6H Levo-6,7,8a,9a), 3.41 (t, $J = 7.0$ Hz, 2H Aliphatic), 3.31 – 3.29 (m, 2H Levo8b,9b), 3.07 – 2.99 (m, 4H Dab $_1$ - γ + Dab $_2$ - γ), 2.98 (s, 3H, Levo-10), 2.25 (t, $J = 7.4$ Hz, 2H, Aliphatic), 2.22 – 2.12 (m, 2H, Dab $_1$ - β 1 + Dab $_2$ - β 1), 2.06 – 1.98 (m, 2H Dab $_2$ - β 2 + Dab $_2$ - β 2), 1.64 – 1.57 (m, 4H, Aliphatic), 1.54 (d, $J = 6.8$ Hz, 3H, Levo-11), 1.49 – 1.10 (m, 16H, Aliphatic). ^{13}C NMR (126 MHz, Methanol- d_4) δ 175.48(Levo-1), 175.21(levo-2), 175.20(Levo-3), 173.54(Dab-1 carbonyl), 171.98(Dab-2 carbonyl), 165.33(Levo-4), 144.74(Levo-5), 141.02(Aliphatic carbonyl), 129.45(Levo-6), 129.33(Levo-7), 124.45(Levo-8), 113.47(Levo-9), 103.76(Levo-10), 68.38(Levo-11), 55.15(Levo-12), 54.09(Levo-13), 51.15(Dab $_1$ - α), 50.52(Dab $_2$ - α), 47.49(Levo-14), 42.49(Levo-15), 38.75(Aliphatic), 36.33(Dab $_1$ - γ), 36.23(Dab $_2$ - γ), 35.23(Aliphatic), 29.71(Dab $_1$ - β), 29.20(Dab $_2$ - β), 29.19(Aliphatic), 29.14(Aliphatic), 29.12, 29.00, 28.95, 28.94, 26.70, 25.34, 16.76(Levo-16). MALDI-TOF-MS m/z calcd for $\text{C}_{34}\text{H}_{52}\text{FN}_7\text{O}_5$ (M+H) $^+$ monoisotopic peak: 758.4723; found: 758.458

Conjugate 11: ^1H NMR (500 MHz, Methanol- d_4) δ 8.74 (s, 1H, Levo-1), 7.65 (d, $J = 12.2$ Hz, 1H, Levo-2), 4.73 – 4.69 (m, 1H, Levo-3), 4.54 (dd, $J = 11.5, 2.2$ Hz, 1H, Levo-4), 4.47 – 4.37 (m, 4H, Dab $_1$ - α + Levo-5 + Dab $_2$ - α + Dab $_3$ - α), 3.65 – 3.55 (m, 6H Levo-6,7,8a,9a), 3.42 (t, $J = 7.1$ Hz, 2H, Aliphatic-a), 3.30 – 3.30 (m, 2H, Levo8b,9b), 3.08 – 3.01 (m, 6H, Dab $_1$ - γ + Dab $_2$ - γ + Dab $_3$ - γ), 2.98 (s, 3H Levo-10), 2.27 – 2.15 (m, 5H, Aliphatic-b + Dab $_1$ - β 1 + Dab $_2$ - β 1 + Dab $_3$ - β 1), 2.09 – 2.01 (m, 3H, Dab $_1$ - β 2 + Dab $_2$ - β 2 + Dab $_3$ - β 2), 1.65 – 1.57 (m, 4H, Aliphatic-c+d), 1.54 (d, $J = 6.8$ Hz, 3H, Levo-11), 1.46 – 1.25 (m, 16H, Aliphatic-m). ^{13}C NMR (126 MHz, Methanol- d_4) δ 175.48(Levo-1), 175.25(levo-2), 175.23(Levo-3), 173.48(Dab-1 carbonyl), 172.16(Dab-2 carbonyl), 171.52(Dab-3 carbonyl), 165.35(Levo-4), 144.76(Levo-5), 141.06(Aliphatic carbonyl), 129.45(Levo-6), 129.32(Levo-7), 124.46(Levo-8), 110.34(Levo-9), 103.77(Levo-10), 68.38(Levo-11), 55.16(Levo-12), 54.10(Levo-13), 51.23(Dab $_1$ - α), 51.10(Dab $_2$ - α), 50.60(Dab $_3$ - α), 47.51(Levo-14), 42.48(Levo-15), 38.75(Aliphatic-a), 36.29(Dab $_1$ - γ), 36.24(Dab $_2$ - γ), 36.23(Dab $_3$ - γ), 35.24(Aliphatic-b), 29.76(Dab $_1$ - β), 29.22(Dab $_2$ - β), 29.20(Dab $_3$ - β), 29.15(Aliphatic-c), 29.13(Aliphatic-d), 29.08, 29.01, 28.96, 28.95, 26.71, 25.35, 16.74(Levo-16). MALDI-TOF-MS m/z calcd for $\text{C}_{42}\text{H}_{68}\text{FN}_{11}\text{O}_7$ (M+H) $^+$ monoisotopic peak: 858.5360; found: 858.499

Conjugate 12: ^1H NMR (500 MHz, Methanol- d_4) δ 8.74 (s, 1H, Levo-1), 7.65 (d, $J = 12.1$ Hz, 1H, Levo-2), 4.73 – 4.69 (m, 1H, Levo-3), 4.54 (dd, $J = 11.5, 2.1$ Hz, 1H, Levo-4), 4.47 (dd, $J = 9.0, 4.9$ Hz, 1H, Dab₁- α), 4.43 – 4.36 (m, 4H, Levo-5 + Dab₂- α + Dab₃- α + Dab₄- α), 3.64 – 3.54 (m, 6H Levo-6,7,8a,9a), 3.43 – 3.40 (m, 2H, Aliphatic), 3.30 – 3.30 (m, 2H, Levo8b,9b), 3.09 – 3.02 (m, 8H, Dab₁- γ + Dab₂- γ + Dab₃- γ + Dab₄- γ), 2.98 (s, 3H, Levo-10), 2.29 – 2.25 (m, 2H, Aliphatic), 2.24 – 2.13 (m, 4H, Dab₁- β 1 + Dab₂- β 1 + Dab₃- β 1 + Dab₄- β 1), 2.11 – 2.01 (m, 4H, Dab₁- β 2 + Dab₂- β 2 + Dab₃- β 2 + Dab₄- β 2), 1.65 – 1.58 (m, 4H, Aliphatic), 1.54 (d, $J = 6.8$ Hz, 3H, Levo-11), 1.45 – 1.28 (m, 16H, Aliphatic). ^{13}C NMR (126 MHz, Methanol- d_4) δ 175.62(Levo-1), 175.25(Levo-2), 175.22(Levo-3), 173.48(Dab-1 carbonyl), 172.29(Dab-2 carbonyl), 171.70(Dab-3 carbonyl), 171.48(Dab-4 carbonyl), 165.35(Levo-4), 144.76(Levo-5), 141.10(Aliphatic carbonyl), 129.45(Levo-6), 129.33(Levo-7), 124.46(Levo-8), 115.73, 110.33(Levo-9), 103.77(Levo-10), 68.38(Levo-11), 55.16(Levo-12), 54.09(Levo-13), 51.29(Dab₁- α), 51.23(Dab₂- α), 51.19(Dab₃- α), 50.60(Dab₄- α), 47.49(Levo-14), 42.48(Levo-15), 38.75(Aliphatic), 36.38(Dab₁- γ), 36.26(Dab₂- γ + Dab₃- γ + Dab₄- γ), 35.27(Aliphatic), 29.76(Dab₁- β), 29.29(Dab₂- β), 29.23(Dab₃- β), 29.21(Dab₄- β), 29.15(Aliphatic), 29.12(Aliphatic), 29.06, 29.01, 28.97, 26.72, 25.35, 16.75(Levo-16). MALDI-TOF-MS m/z calcd for C₄₂H₆₈FN₁₁O₇ (M+H)⁺ monoisotopic peak: 958.5997; found: 958.583

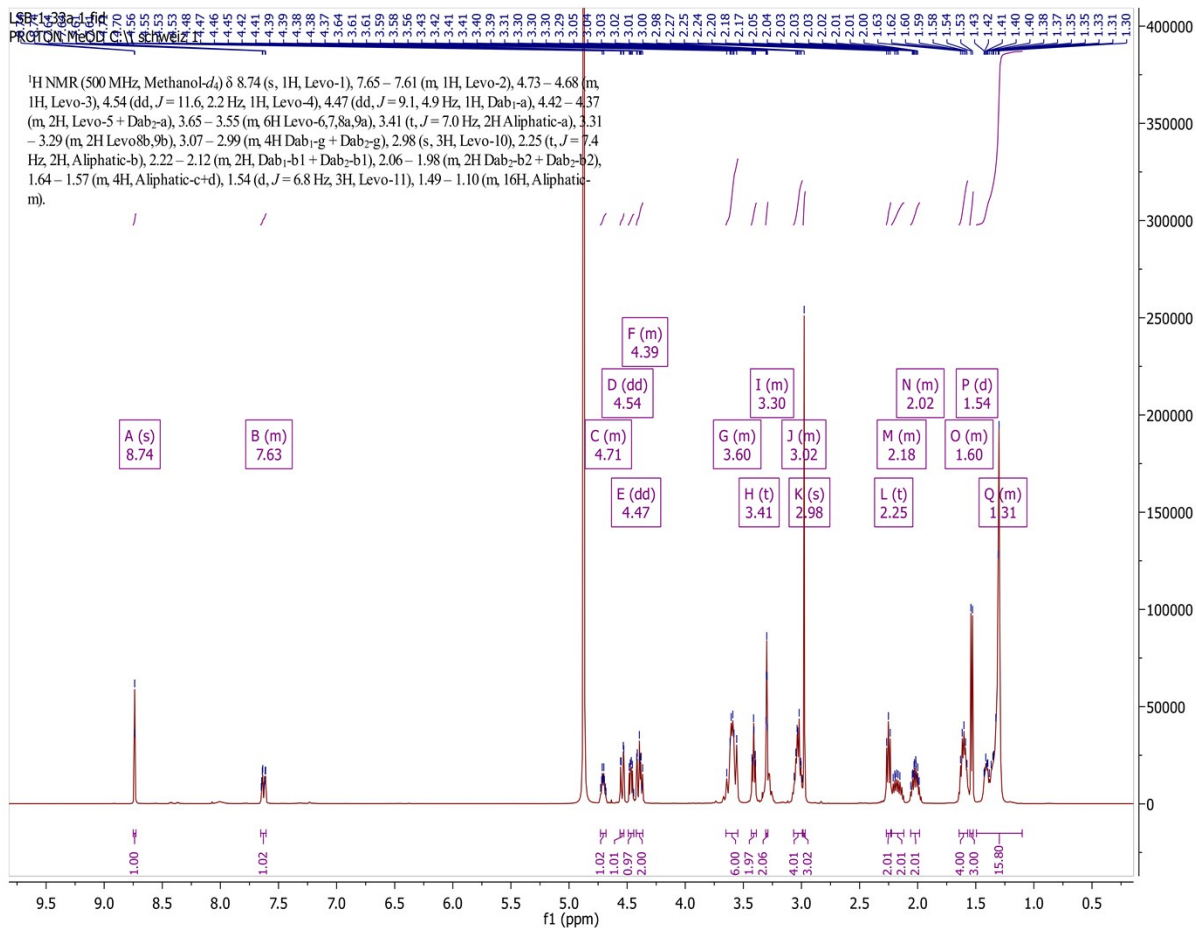


Figure S3. Conjugate 10 ¹H NMR spectra.

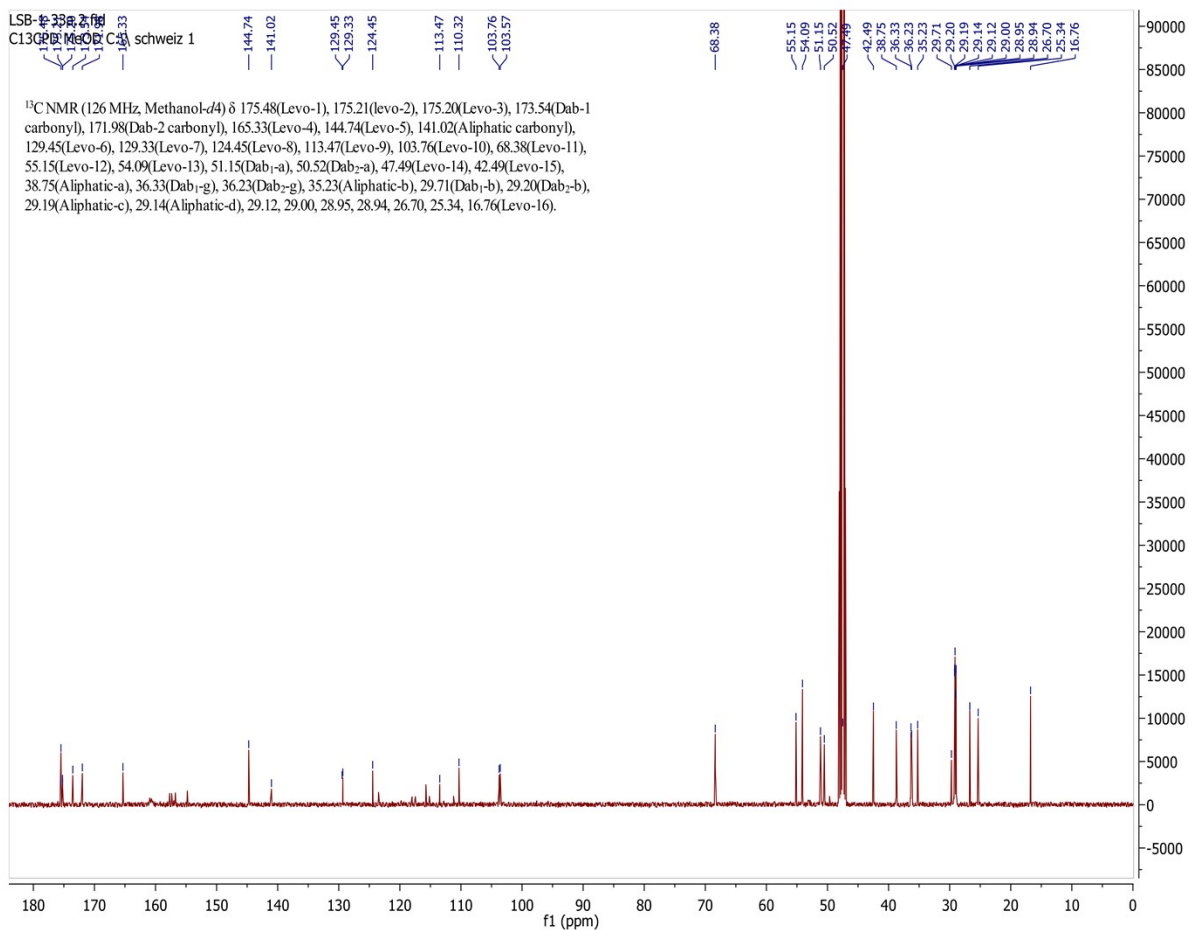


Figure S4. Conjugate 10 ^{13}C NMR spectra.

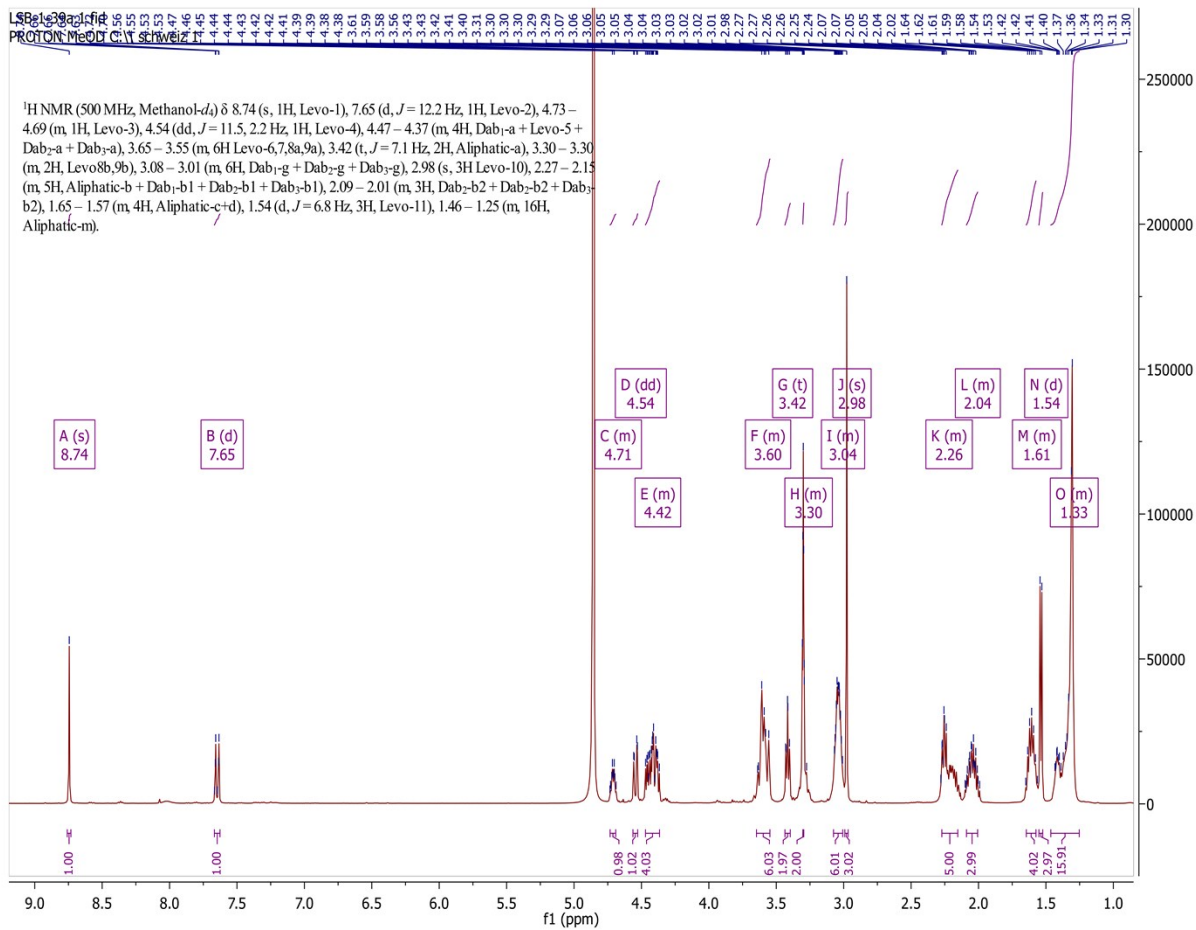


Figure S5. Conjugate **11** ¹H NMR spectra.

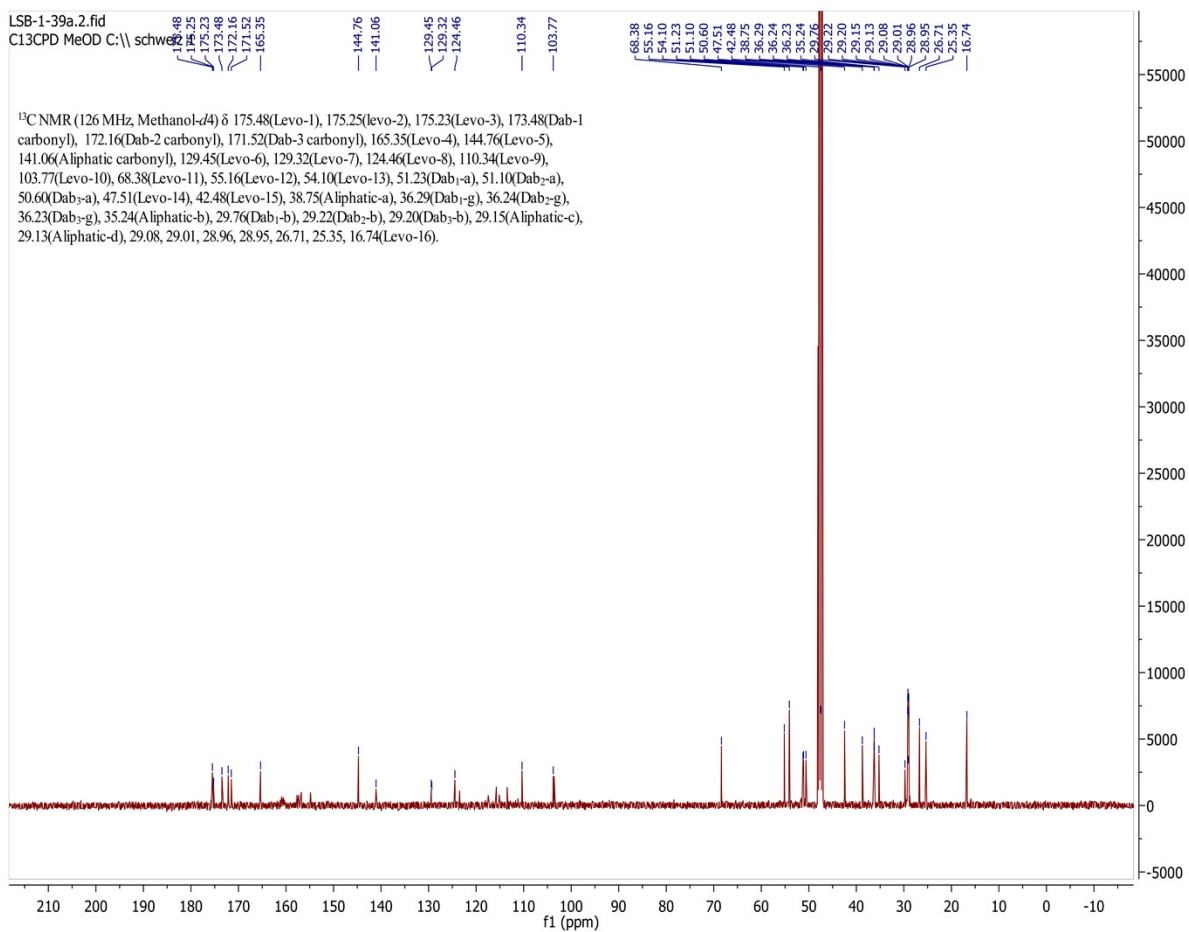


Figure S6. Conjugate **11** ^{13}C NMR spectra.

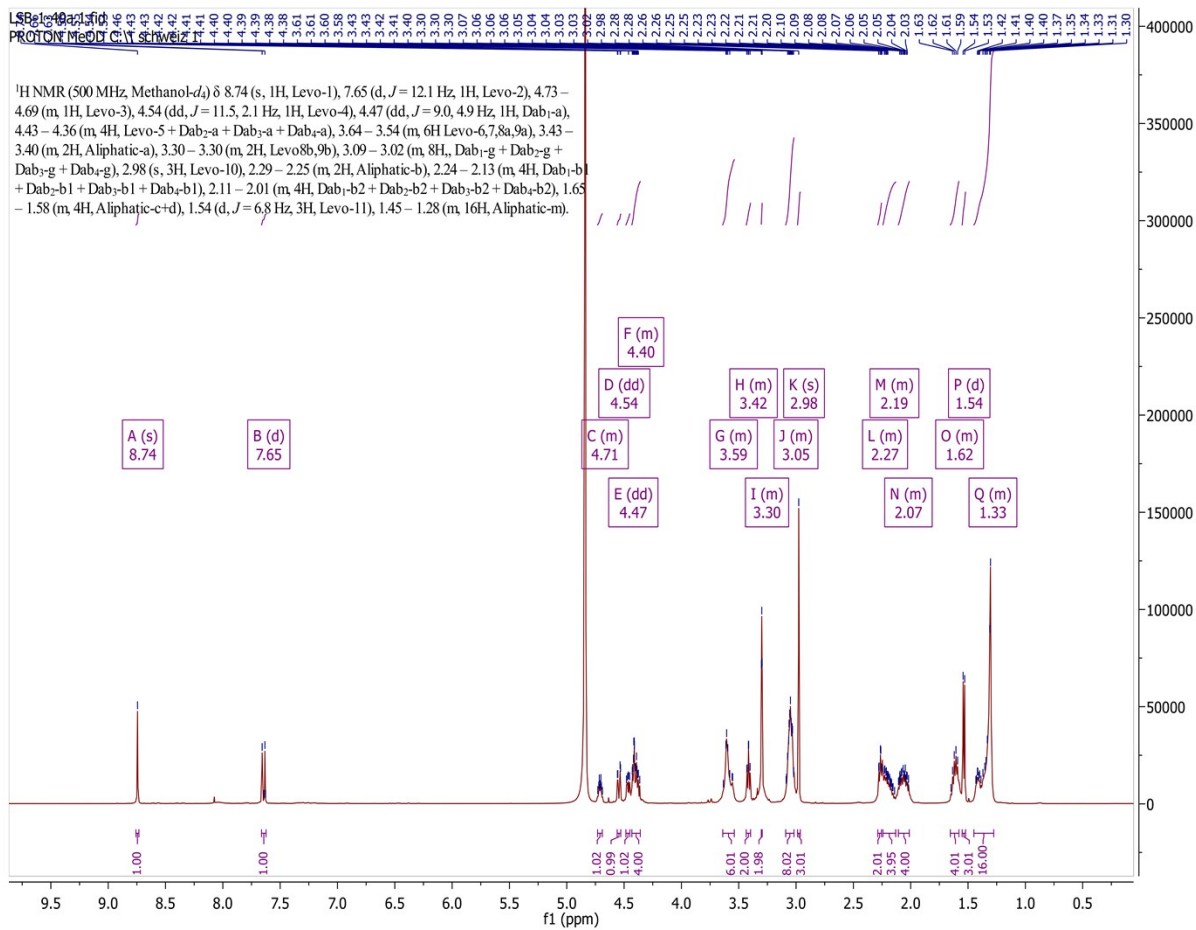


Figure S7. Conjugate 12 ¹H NMR Spectra.

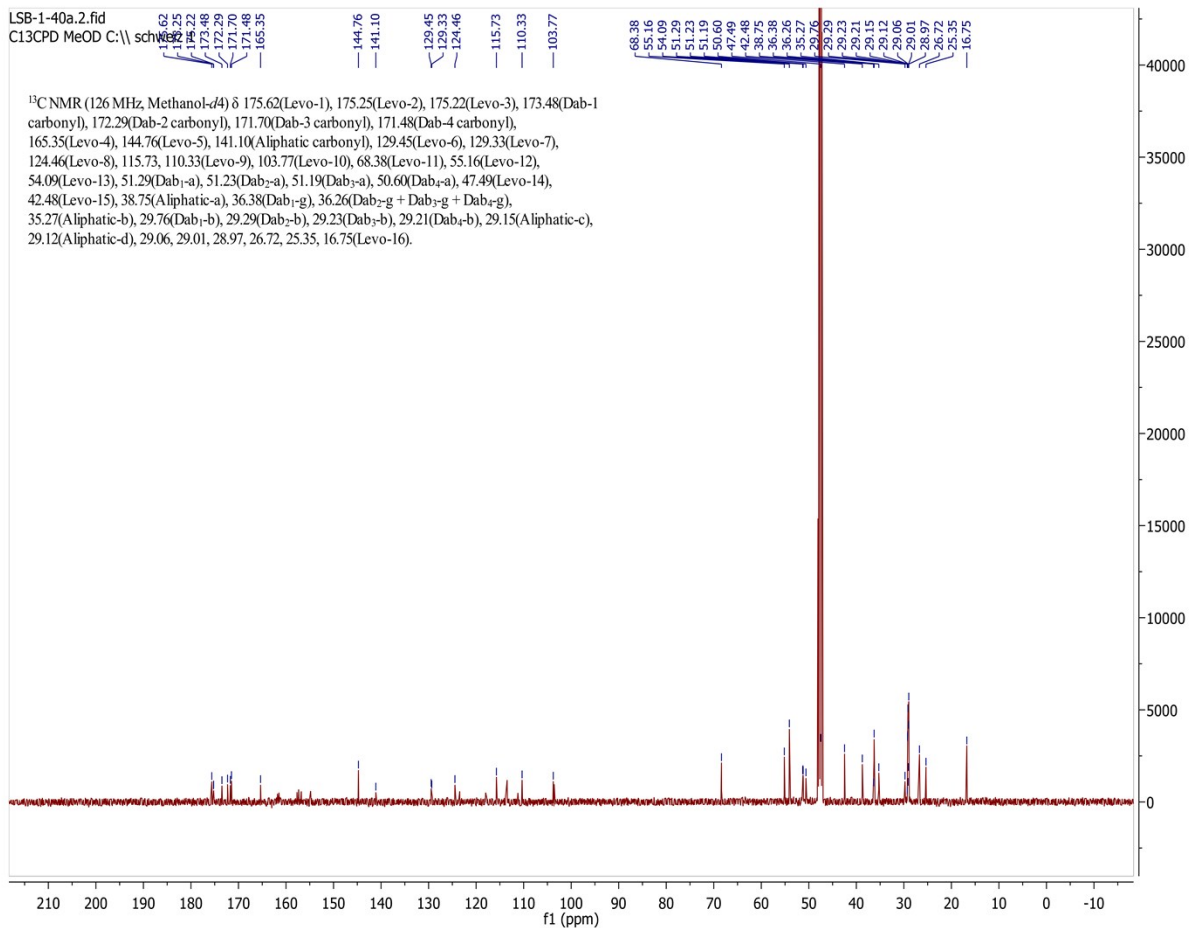


Figure S8. Conjugate **12** ¹³C NMR spectra.