Supplemental Experimental Procedures

Stereochemical Control of Splice Modulation in FD-895 Analogues

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Figure S1. Synthesis of FD-895 (1) from aldehyde **15** and core **5**. The entire synthetic pathway is shown using the methods and materials prepared in Chan *et al.* 2020.²⁵ In this publication, a central route was developed to enable preparation of **14** and **36**, these steps are shown in grey. Steps used specifically to prepare **1** from **14** and **36** are shown in black. Experimental procedures are provided for these (black) steps.

Position ^a	$\delta_{\rm H}$, mult (<i>J</i> in Hz)	$\delta_{\rm C}$	¹ H, ¹ H-COSY	¹ H, ¹ H-NOESY	¹ H, ¹³ C-HMBC
1		172.1			
2'	2.30, dd (14.8, 3.9)	20 (2"w, 3	2", 3	1, 3, 4
2"	2.20, dd (14.8, 3.0)	38.0	2', 3	2', 3, 4', 5'	1, 3w
3	3.51, m	69.4	3OHw	2', 2'', 4', 5', 5'', 60H, 170H	
3OH	3.66, d (11.0)		3	3, 60Н, 170Н	
4'	1.60, m	20.4	3w, 4", 5'	2', 3, 8, 24	3, 5
4"	1.25, m	30.4	3, 5'	3w, 5', 7	3, 5
5'	1.54, m	25.0	5"	2", 3, 5', 8	4, 6, 24w
5"	1.23, m	35.9	4'	3, 4', 7	4, 6, 7, 24w
6		73.3			
6OH	1.85, s			3, 30Н, 170Н	
7	5.26, s	79.2	8	4'', 5'', 8, 24	8, 9, 24, 26, 29
8	5.83, dd (15.2, 9.8)	126.5	7,9	4'w, 5', 7, 9, 10	6w, 9, 10
9	5.63, dd (15.2, 10.0)	140.7	8, 10	4'', 5'', 7, 8, 10, 11, 25	7, 8, 10, 11w, 25w
10	2.38, m	41.1	11, 25	7, 8, 9, 11w, 25, 26	8, 9, 11, 25
11	5.24, d (1.8)	82.6	10	9, 10w, 13, 25, 26w	1, 9, 10, 12, 14, 25, 26
12		131.6			
13	6.10, dd (10.9, 1.5)	131.7	14, 26	11, 14, 15, 25, 26w, 28w	11, 14, 15, 26
14	6.23, dd (15.0, 10.8)	126.2	13, 15	13, 15, 16, 26, 27	12, 13, 16
15	5.53, dd (15.0, 8.7)	138.3	14, 16	13, 14, 16, 17, 18, 27	13, 14, 16, 17, 27
16	2.39, m	42.6	17, 27	14, 15, 17, 18, 27	13w, 14, 15, 17, 18, 27
17	3.10, m	75.3	16, 18	15, 16, 18, 19, 27	16w, 27w
17OH	2.14, bs		17	3w, 3OH, 6OH, 17w	
18	2.66, dd (5.8, 2.2)	60.6	17, 19	15w, 16, 17, 20, 27, 28	17, 20
19	2.87, dd (8.3, 2.3)	59.7	18w, 20	17, 20, 21, 28, 31	17, 20, 21, 28
20	1.26, m	39.4	19, 28	18, 19, 21, 23, 28	18, 19, 21, 23, 27, 28
21	3.15, td (6.4, 4.1)	83.6	20, 22', 22''	19, 20, 22', 22'', 23, 28	19, 20, 22w, 23, 28, 31
22'	1.63, m	22.0	22", 23	21, 22", 23	20, 21, 23
22"	1.37, ddd (14.0, 7.6, 6.7)	25.9	22', 23	21, 22', 23	20, 21, 23
23	0.84, t (7.5)	10.0	22', 22''	21, 22', 22''	21, 22
24	1.02, s	24.8		4', 5', 6OH, 7	5, 6, 7
25	0.70, d (6.8)	16.4	10	9, 10, 11, 13w, 26	9, 10, 11
26	1.57, d (1.3)	11.9		10, 11, 14, 25	11, 12, 13, 14, 15
27	1.14, d (6.8)	17.0	16	14, 15, 16, 17, 18w	15, 16, 17
28	0.83, d (7.1)	10.6	20	17, 18, 19, 21, 31w	19, 20, 21
29		169.0			
30	1.62, s	20.7			29
31	3.25, s	57.8		19, 23	21

Table S1. NMR data for FD-895 (1) in C_6D_6

^a Position numbering is provided in Supporting Figure S1.



Figure S2. Synthesis of 3S-FD-895 (1a) from side chain 4 and core 5a. The entire synthetic pathway is shown using the methods and materials (4 and 5a) prepared in Chan *et al.* 2020.²⁵ In this publication, a central route was developed to enable preparation of analogues of FD-895 (1), these steps are shown in grey. Steps used specifically to prepare 1a are shown in black. Experimental procedures are provided for these (black) steps.

Position	$\delta_{\rm H}$, mult (<i>J</i> in Hz)	δ_{C}	¹ H, ¹ H-COSY	¹ H, ¹ H-NOESY	¹ H, ¹³ C-HMBC
1		169.6			
2'	2.47, m	40.0	2", 3	3, 5'	1, 3, 4w
2"	2.38, m	40.0	2', 3	3	1, 3, 4
3	4.25, bs	67.3	2', 2''w	2'w, 2'', 4', 4''	
3OH					
4'	1.35, m	27.5	3w, 4'', 5''	3, 5', 5'', 7	2w, 3w, 5w
4"	1.33, m	27.5	3w, 4', 5'	3, 5', 5''	3w, 5w
5'	1.88, dt (13.5, 6.5)	20.7	4", 5"	4''	4, 6, 24
5"	1.82, m	30.7	4', 5'	4', 24	4, 6, 7, 24
6		73.4			
6OH					
7	5.26, d (9.8)	79.3	8	4', 8, 9, 24	8, 9, 24w, 29
8	5.93, dd (15.2, 9.8)	126.6	7, 9, 10w	5'w, 7, 9, 10	6w, 10
9	5.62, dd (15.3, 10.0)	140.4	8, 10	7, 8, 10, 11, 25	7, 10, 11w, 25w
10	2.47, m	41.2	9w, 11, 25	8w, 25	8, 9, 11, 25w
11	5.20, d (10.7)	82.2	10	10w, 13, 25	1, 9, 10, 12, 13, 25w, 26
12		132.1			
13	6.17, d (11.0)	131.4	14, 26w	8, 15, 27w	11, 15, 26
14	6.26, dd (15.0, 10.9)	126.3	13, 15	15, 27	12, 13, 16
15	5.54, dd (15.0, 8.7)	137.9	14, 16	7, 13, 14, 16w, 27	12, 13, 16, 17, 27
16	2.38, m	42.6	15, 17w, 27	14,15,27	14, 15, 17, 18, 27
17	3.10, t (6.6)	75.2	16, 18	16w, 18w 27	15, 16, 18, 27
17OH					
18	2.68, dd (5.8, 1.9)	60.6	17, 19w	17, 19, 20, 28	17, 19w, 20w
19	2.86, dd (8.2, 2.3)	59.6	18w, 20	20w, 28	17w, 18w, 20, 21w
20	1.27, ddd (8.2, 6.9, 4.1)	39.3	19, 28	18, 21, 22', 22'', 28	19, 28w
21	3.14, td (6.3, 4.1)	83.5	20, 22', 22''	20, 22', 22'', 23, 28	19, 20w, 22w, 23, 28, 31
22'	1.63, m	22.0	21, 22'', 23	22", 23w	20, 21, 23
22"	1.39, m	23.8	21, 22', 23	22', 23	20, 21, 23
23	0.84, t (7.5)	10.0	22', 22''	21, 22', 22''	21, 22
24	1.11, s	24.8		2"	5, 6, 7
25	0.73, d (6.8)	16.5	10	10	9, 10, 11
26	1.60, s	11.9	13	10, 14	11, 12, 13
27	1.15, d (6.7)	17.1	16	15, 16, 17	15, 16, 17
28	0.85, d (7.8)	10.6	20	20, 21	19, 20
29		169.1			
30	1.66, s	20.8			29
31	3.25, s	57.7		21, 22', 23, 24	21

Table S2. NMR data for 3S-FD-895 (1a) in C₆D₆

^a Position numbering is provided in Supporting Figure S2.



Figure S3. Synthesis of 7R-FD-895 (1b) from core 5b, prepared in 4 steps from intermediate 35, and side chain 4. The entire synthetic pathway is shown using the methods and materials prepared in Chan *et al.* 2020.²⁵ In this publication, a central route was developed to enable preparation of analogues of FD-895 (1), these steps are shown in grey. Steps used specifically to prepare 1b are shown in black. Experimental procedures are provided for these (black) steps.

Position	$\delta_{\rm H}$, mult (<i>J</i> in Hz)	$\delta_{\rm C}$	¹ H, ¹ H-COSY	¹ H, ¹ H-NOESY	¹ H, ¹³ C-HMBC
1		172.3			
2'	2.35, m	39.0	2', 3	3, 4''w, 5'	1, 3, 4
2"	2.29, dd (14.6, 3.1)	57.0	2", 3	3, 4'', 5'	1, 3, 4
3	3.57, bs	69.6	3OH, 4'	2', 2'', 4', 4''w, 5'w, 5'', 24	N/A
3OH	3.67, d (10.6)		3	4', 5''	conformers
4'	1.66, m	20.6	3, 4", 5', 5"	3, 30H, 5', 5'', 7w	conformers
4"	1.82, m	50.0	3, 4', 5', 5''	3w, 5', 5'', 7w	2
5'	1.58, m	26.5	4', 4'', 5''	4'	conformers
5"	0.95, m	30.3	4', 4'', 5'	4"	conformers
6		73.8			
6OH					
7	5.40, m	77.9	8	4', 8, 10, 15, 24, 27w	5, 6, 8, 9, 29
8	5.92, dd (15.4, 2.3)	128.0	7,9	7, 9, 10, 11, 13w, 14w, 15w, 22	7, 10
9	5.38, m	130.7	8, 10	4', 8, 10, 11, 15, 18, 25	7, 10
10	2.47, tq (10.1, 6.7)	41.0	11, 25	8, 9, 25, 26	8, 9, 11, 25
11	5.36, d (10.7)	82.9	10	4', 8, 10, 13, 25	1, 9, 10, 12w, 13, 25, 26
12		131.9			
13	6.15, d (10.9)	131.5	14, 26w	8, 9, 11, 15, 28	11, 15, 26
14	6.26, dd (15.0, 10.8)	126.2	13, 15	15, 16, 22', 26, 27	12, 13, 16
15	5.50, dd (15.0, 8.7)	138.2	14, 16	8w, 13, 14, 16, 17, 18w, 27	13, 16, 17, 27
16	2.37, m	42.7	15, 17, 26	14, 15, 27	14, 15, 17, 18, 27
17	3.06, t (6.6)	75.3	16, 18	16w, 19, 27	15, 16, 18, 19, 27
17OH					
18	2.65, dd (5.7, 2.2)	60.7	17	10, 15, 16, 19, 20, 27w, 28w	17, 19, 20w
19	2.85, dd (8.2, 2.3)	59.7	20	10w, 16w, 17w, 18, 20w, 21w, 22'w, 28	18, 20, 28
20	1.30, m	39.4	19, 28	18, 23, 28	18, 19, 27, 28
21	3.14, td (6.3, 4.0)	83.6	20, 22', 22''	19, 20, 22', 22''w, 23, 28	19w, 20, 23, 28, 31
22'	1.57, m	22.0	23	21, 22β, 23	20, 21, 23
22"	1.36, m	23.8	23	21, 22α, 23	20, 21, 23
23	0.84, t (7.5)	10.0	22', 22''	21, 22', 22''	21, 22
24	1.00, s	24.7		3OH, 5', 5'', 7	5, 6, 7
25	0.76, d (6.8)	17.1	10	9, 10, 11, 20, 26	9, 10, 11
26	1.62, bs	11.5		10, 11, 14	11, 12, 13, 14, 15
27	1.12, d (6.8)	16.9	16	14, 15, 16, 17w	15, 16, 17
28	0.84, d (7.0)	10.6	20	19, 21	19, 20, 21
29		169.2			
30	1.67, s	20.4			29
31	3.24. s	57.8		19, 21, 22', 23, 28	21

Table S3. NMR data for 7*R*-FD-895 (1b) in C₆D₆

^a Position numbering is provided in Supporting Figure S3.



Figure S4. Synthesis of 10R, 11R-FD-895 (1c) from core 5c, prepared in 5 steps from 25 and 32, and side chain 4. The entire synthetic pathway is shown using the methods and materials prepared in Chan *et al.* 2020.²⁵ In this publication, a central route was developed to enable preparation of analogues of FD-895 (1), these steps are shown in grey. Steps used specifically to prepare 1c are shown in black. Experimental procedures are provided for these (black) steps

Position	$\delta_{\rm H}$, mult (<i>J</i> in Hz)	$\delta_{\rm C}$	¹ H, ¹ H-COSY	¹ H, ¹ H-NOESY	¹ H, ¹³ C-HMBC
1		169.6			
2'	2.38, m	40.1	2', 3	3w, 5'	1, 3, 4
2"	2.25, dd (13.3, 5.3)	40.1	2', 3	3	1, 3, 4
3	4.14, m	67.8	2', 2''w	2'w, 2'', 4', 4'', 5'w, 24	
3OH					
4'	1.29, m	27.5	3, 4", 5', 5"	3, 4'', 5', 5''	5, 3
4"	1.20, m	27.5	3w, 4', 5', 5''	3, 4', 5', 5'', 24	5
5'	1.83, m	20.7	4', 4'', 5''	2', 4', 4'', 8, 25	3w, 4, 6, 7w, 24
5"	1.75, dt (13.4, 6.7)	30.7	4', 4'', 5'	4', 4'', 24	3w, 4, 6, 7, 24, 30
6		73.5			
6OH	1.75, s				
7	5.23, d (9.8)	79.3	8	4', 8w, 9, 24	8, 9, 24w, 29
8	5.90, dd (15.2, 9.8)	126.6	7, 9, 10w	5', 7, 9, 10	6, 10
9	5.58, dd (15.3, 10.0)	140.3	8, 10	4', 7, 8, 10, 11, 25	7, 10
10	2.43, m	41.2	11, 25	8, 9, 11, 25, 26	8, 9, 11, 25
11	5.18, d (10.8)	82.1	10	9, 10w, 13, 25, 26w	1, 9, 10, 12, 13, 25, 26
12		132.2			
13	6.17, d (10.8)	131.3	14, 26w	9, 11, 15, 25, 26	11, 15, 26
14	6.27, dd (15.1, 10.8)	126.5	13, 15	15, 16, 26, 27	12, 13, 16
15	5.53, dd (15.1, 8.8)	137.8	14, 16	13, 14, 16, 17, 18w, 27	13, 16, 17
16	2.36, m	42.8	15, 17, 27	14, 15, 17, 18, 27	14, 15, 17, 18, 27
17	3.12, m	74.8	16, 18	15, 16, 18, 19, 27	15w, 16, 19, 25
17OH	1.55, bs				
18	2.69, dd (5.1, 2.2)	60.3	17, 19w	14w, 15, 16, 17, 19, 20, 27, 28	17, 19, 20
19	2.90, dd (8.1, 2.3)	59.4	18w, 20	17, 18w, 20, 22'w, 28	17w, 18, 20, 21w
20	1.31, m	39.2	19, 28	19, 21, 28	19, 23
21	3.14, m	83.6	20, 22', 22''	20, 22', 22'', 23	19, 20, 22w, 23, 28 31
22'	1.60, m	22.0	21, 22", 23	21, 22", 23	20, 21, 23
22"	1.39, m	23.9	21, 22', 23	19w, 21, 22', 23, 31	20, 21, 23
23	0.84, t (7.5)	10.1	22', 22''	20, 21, 22', 22'', 31w	21, 22
24	1.06, s	24.8		4'w, 4'', 5'w, 5'', 7	5, 6, 7
25	0.72, d (6.7)	16.5	10	5', 9, 10, 11,13, 26	9, 10, 11
26	1.58, s	12.0	13	10, 14, 25w	11, 12, 13, 14
27	1.11, d (6.8)	16.9	16	14, 15, 16, 17, 18w	15, 16, 17
28	0.87, d (7.0)	10.5	20	18, 19, 20, 21, 22'	19, 20, 21
29		169.2			
30	1.63, s	20.8			29
31	3.24, s	57.7		19w, 21, 22'', 23	21

Table S4. NMR data for 10*R*,11*R* -FD-895 (1c) in C₆D₆

^a Position numbering is provided in Supporting Figure S4.



Figure S5. Synthesis of 17*S*-FD-895 (1d) from core 5 and side chain 4d. The entire synthetic pathway is shown using the methods and materials prepared in Chan *et al.* $2020.^{25}$ In this publication, a central route was developed to enable preparation of analogues of FD-895 (1), these steps are shown in grey. Steps used specifically to prepare 1d are shown in black. Experimental procedures are provided for these (black) steps

Position	$\delta_{\rm H}$, mult (<i>J</i> in Hz)	$\delta_{\rm C}$	¹ H, ¹ H-COSY	¹ H, ¹ H-NOESY	¹ H, ¹³ C-HMBC
1		171.8			
2'	2.29, dd (14.8, 3.9)	20.2	2', 3	3, 30Hw, 4'/5'w	3
2"	2.19, dd (14.8, 3.0)	36.2	2", 3	3, 4'/5'	1
3	3.49, td (11.1, 3.5)	69.0	3OH, 4'w, 4''	2', 2'', 3OH, 4'/5', 5''	
3OH	3.63, d (11.2)		3	2', 3, 4'', 6OH	3
4'	1.57, m	20.0	3w, 4", 5', 5"	2", 3, 5", 24	
4"	1.25, m	30.0	3, 4', 5', 5''	30Hw, 5', 7, 9, 24w	3w
5'	1.55, m	25.5	3, 4"	2'', 3OH, 4'', 6OH, 8	
5"	1.20, m	33.3	5'	3, 4', 7w, 24	4w, 6w, 7w
6		72.5			
6OH	1.75, s			3OH, 5''w, 7w, 8, 24	5, 6, 17
7	5.26, d (1.5)	78.8	8	4'', 8, 9w, 24	8, 29
8	5.83, dd (15.2, 9.8)	140.3	7,9	4'/5', 7, 9, 10w, 25	7, 10w
9	5.62, dd (15.2, 10.0)	126.0	8, 10	4'/5', 7, 8, 10, 25	10, 25w
10	2.39, m	40.8	9, 11, 25	7w, 8, 9w, 11w, 25, 26	11
11	5.24, d (2.4)	82.2	10	9, 10, 13, 25, 26	1, 12, 13, 26
12		131.0			
13	6.11, d (10.7)	131.4	14, 26w	11, 14, 15, 25, 26w	11, 14w, 15, 26
14	6.26, dd (15.2, 10.8)	126.1	13, 15	13, 15, 16, 26, 27w	12w, 13w
15	5.80, dd (15.2, 8.3)	137.6	14, 16	13, 14, 27	12, 13, 14, 16, 27
16	2.36, m	41.2	15, 17, 27	14, 17, 27	15
17	3.42, q (3.7)	73.0	16, 17OH, 18	15, 16, 17OH, 18, 19, 27	
17OH	1.55, bs			17, 19	16, 17
18	2.56, dd (3.8, 2.2)	57.3	17, 19w	15w, 16, 17, 19w, 20, 27, 28	
19	3.01, dd (8.3, 2.3)	59.3	18, 20	15w, 17, 17OH, 18w, 20, 21, 28	20
20	1.33, m	38.9	19, 21, 28	18, 19w, 21, 23, 28	
21	3.15, m	83.4	22', 22''	19, 20, 22', 22'', 23	19, 28
22'	1.63, m	22.5	21, 23	21w, 22'', 23	20w, 21, 23
22"	1.40, dt (14.0, 6.9)	23.3	21, 23	21w, 22', 23	20w, 21, 23
23	0.85, t (7.5)	9.7	22', 22''	20, 21, 22', 22''	21, 22
24	1.00, s	24.4		4'/5', 4''w, 5'', 6OH, 7	5, 6,7
25	0.70, d (6.7)	16.1	10	7, 9, 10, 13, 26	8, 10, 11
26	1.59, d (1.3)	11.5	13	10, 11w, 14, 25	11, 12, 13, 14w, 15w
27	1.12, d (7.0)	16.9	16	9, 14, 15, 16, 17, 18	15, 16, 17
28	0.88, d (6.9)	10.5	20	18, 19, 20, 21	19, 20, 21
29		168.7			
30	1.61, s	20.4			29
31	3.23, s	57.4		21, 22', 22'', 23	21

Table S5. NMR data for 17S-FD-895 (1d) in C₆D₆

^a Position numbering is provided in Supporting Figure S5.



Figure S6. Synthesis of 17-O-methyl-FD-895 (1e) from core **5** and side chain **4e**, prepared in 1 step from **4**. The entire synthetic pathway is shown using the methods and materials prepared in Chan *et al.* 2020.²⁵ In this publication, a central route was developed to enable preparation of analogues of FD-895 (1), these steps are shown in grey. Steps used specifically to prepare **1e** are shown in black. Experimental procedures are provided for these (black) steps.

Position	$\delta_{\rm H}$ mult (<i>I</i> in Hz)	δο	¹ H ¹ H-COSY	¹ H ¹ H-NOESY	$^{1}\text{H}^{13}\text{C-HMBC}$
1		172.1	11, 11 0001		
2,	2.29 dd(14.8, 3.8)	172.1	2", 3	3.4'w	1. 3. 4w
2"	2.20, dd (14.8, 3.0)	38.6	2'. 3	3. 5'	1
3	3.48. m	69.3	2'w, 2''w, 30H, 4''	2', 2'', 4', 5''	
30H	3.62. d(11.1)	0515	3	3, 60H	
4'	1.57. m		3. 4", 5', 5"	3, 5", 7, 8	5, 6, 3, 24
4''	1.26. m	30.4	4', 5', 5''	2", 3, 5", 9	3
5'	1.54, m		4', 4'', 5''	2", 4", 8	4, 6, 24
5''	1.18, m	35.9	4', 4'', 5'	4'	6, 7
6		73.6			
6OH	1.77. s				
7	5.24, d (5.5)	79.2	8	4"/5", 8, 9, 24w, 26	8w, 9, 24w, 29
8	5.82, dd (15.2, 9.8)	126.4	7,9	4'/5', 7w, 9, 10, 13w	6w, 9w, 10
9	5.62, dd (15.1, 10.9)	140.7	8, 10	4"/5", 7, 8, 10, 11, 25	7, 8w, 10, 25
10	2.39, tq (10.4, 6.8)	41.1	9, 11, 25	8, 9w, 25, 26	8, 9w, 11, 12, 25
11	5.26, d (6.3)	82.6	10	10, 13, 25, 26	1, 9, 10, 13, 25w, 26
12		131.7			
13	6.12, dd (10.9, 1.5)	131.4	14, 26	8w, 11, 14, 15, 25, 26w	11, 15, 26
14	6.23, ddd (15.1, 10.9, 1.04)	125.7	13, 15	8w, 13, 15, 16, 26	13w, 16
15	5.64, dd (15.2, 10.2)	138.7	14, 16	13, 14, 16, 17	12, 13, 14w, 16, 27
16	2.48, dq (14.4, 7.0)	42.6	15, 17, 27	14, 15, 17w, 27w, 32w	14, 15, 17, 18, 27
17	2.62, t (7.1)	86.2	16, 18	15w, 16, 20, 32	15, 16, 18, 27, 32
17OH					
18	2.68, m	60.6	17	15, 16, 20, 32	17, 19w, 20
19	2.68, m	59.1	20	17, 20, 28w,	20, 21w
20	1.22, m	39.6	19, 28	17w, 21w, 22', 22''	19
21	3.20, m	83.5	20, 22', 22''	20, 22', 22'', 23	19w, 20w, 23w
22'	1.64, m	22.7	21w, 22'', 23	21, 22''	20, 21, 23
22"	1.37, dp (17.0, 6.7, 6.1)	23.7	22', 23	21, 22'	20, 21, 23
23	0.82, t (7.4)	10.0	22', 22''	20, 21, 22'w, 22''w	21, 22
24	1.01, s	24.8		4'/5', 7	5, 6, 7
25	0.71, d (6.8)	16.4	10	9w, 10, 11w, 13, 26	9, 10, 11
26	1.59, d (1.3)	11.8	13	8, 10, 11, 14, 25	11, 12, 13, 14w, 15w
27	1.14, d (6.8)	17.0	16	14, 15, 16, 17	15, 16, 17
28	0.87, d (7.1)	10.8	20	22', 22''	19, 20, 21
29		169.0			
30	1.61, s	20.7			29
31	3.25, s	57.7		20w, 22', 22''w, 23w, 26w, 32	21
32	3.50, s	58.5		60H, 17, 31w	17

Table S6. NMR data for 17-O-Me-FD-895 (1e) in C₆D₆

^a Position numbering is provided in Supporting Figure S6.



Figure S7. Synthesis of 17-O-methyl-17*S*-FD-895 (1f) from core **5** and side chain 4f, prepared from **4d.** The entire synthetic pathway is shown using the methods and materials prepared in Chan *et al.* 2020.²⁵ In this publication, a central route was developed to enable preparation of analogues of FD-895 (1), these steps are shown in grey. Steps used specifically to prepare 1f are shown in black. Experimental procedures are provided for these (black) steps

Position	$\delta_{\rm H}$, mult (<i>J</i> in Hz)	$\delta_{\rm C}$	¹ H, ¹ H-COSY	¹ H, ¹ H-NOESY	¹ H, ¹³ C-HMBC
1		172.1			
2'	2.29, dd (14.8, 3.8)	20 6	2", 3	3, 4'w	1, 3, 4w
2"	2.20, dd (14.8, 3.0)	38.0	2', 3	3, 5'	1
3	3.48, m	69.3	2'w, 2''w, 30H, 4''	2', 2'', 4', 5''	
3OH	3.62, d (11.1)		3	3, 6OH	
4'	1.57, m	20.4	3, 4", 5', 5"	3, 5'', 7, 8	5, 6, 3, 24
4''	1.26, m	30.4	4', 5', 5''	2'', 3, 5'', 9	3
5'	1.54, m	25.0	4', 4'', 5''	2'', 4'', 8	4, 6, 24
5"	1.18, m	55.9	4', 4'', 5'	4'	6, 7
6		73.6			
6OH	1.77, s				
7	5.24, d (5.5)	79.2	8	4''/5'', 8, 9, 24w, 26	8w, 9, 24w, 29
8	5.82, dd (15.2, 9.8)	126.4	7,9	4'/5', 7w, 9, 10, 13w	6w, 9w, 10
9	5.62, dd (15.1, 10.9)	140.7	8, 10	4"/5", 7, 8, 10, 11, 25	7, 8w, 10, 25
10	2.39, tq (10.4, 6.8)	41.1	9, 11, 25	8, 9w, 25, 26	8, 9w, 11, 12, 25
11	5.26, d (6.3)	82.6	10	10, 13, 25, 26	1, 9, 10, 13, 25w, 26
12		131.7			
13	6.12, dd (10.9, 1.5)	131.4	14, 26	8w, 11, 14, 15, 25, 26w	11, 15, 26
14	6.23, ddd (15.1, 10.9, 1.04)	125.7	13, 15	8w, 13, 15, 16, 26	13w, 16
15	5.64, dd (15.2, 10.2)	138.7	14, 16	13, 14, 16, 17	12, 13, 14w, 16, 27
16	2.48, dq (14.4, 7.0)	42.6	15, 17, 27	14, 15, 17w, 27w, 32w	14, 15, 17, 18, 27
17	2.62, t (7.1)	86.2	16, 18	15w, 16, 20, 32	15, 16, 18, 27, 32
17OH					
18	2.68, m	60.6	17	15, 16, 20, 32	17, 19w, 20
19	2.68, m	59.1	20	17, 20, 28w,	20, 21w
20	1.22, m	39.6	19, 28	17w, 21w, 22', 22''	19
21	3.20, m	83.5	20, 22', 22''	20, 22', 22'', 23	19w, 20w, 23w
22'	1.64, m	22.7	21w, 22", 23	21, 22"	20, 21, 23
22''	1.37, dp (17.0, 6.7, 6.1)	23.7	22', 23	21, 22'	20, 21, 23
23	0.82, t (7.4)	10.0	22', 22''	20, 21, 22'w, 22''w	21, 22
24	1.01, s	24.8		4'/5', 7	5, 6, 7
25	0.71, d (6.8)	16.4	10	9w, 10, 11w, 13, 26	9, 10, 11
26	1.59, d (1.3)	11.8	13	8, 10, 11, 14, 25	11, 12, 13, 14w, 15w
27	1.14, d (6.8)	17.0	16	14, 15, 16, 17	15, 16, 17
28	0.87, d (7.1)	10.8	20	22', 22''	19, 20, 21
29		169.0			
30	1.61, s	20.7			29
31	3.25, s	57.7		20w, 22', 22''w, 23w, 26w, 32	21
32	3.50, s	58.5		60H, 17, 31w	17

Table S7. NMR data for 17S-O-Me-17S-FD-895 (1f) in C₆D₆

^a Position numbering is provided in Supporting Figure S7.



Figure S8. Synthesis of 3S,17S-FD-895 (**1g**) from core **5a** (see Section D) and side chain **4d** (see Section G). The entire synthetic pathway is shown using the methods and materials prepared in Chan *et al.* 2020.²⁵ In this publication, a central route was developed to enable preparation of analogues of FD-895 (**1**), these steps are shown in grey. Steps used specifically to prepare **1g** are shown in black. Experimental procedures are provided for these (black) steps. **Stille coupling of stannane 4d and core 5a to afford** 3S,17S-FD-895 (**1**).

Position	$\delta_{\rm H}$, mult (<i>J</i> in Hz)	δ _C , Type	¹ H, ¹ H-COSY	¹ H, ¹ H-NOESY	¹ H, ¹³ C-HMBC
1		169.9			
2'	2.43, dd (13.3, 11.3)	40.2	2", 3	3w, 5'	1, 3, 4w
2"	2.30, dd (13.1, 5.4)	40.2	2', 3	3	1, 3, 4
3	4.20, m	67.8	2', 2''w	2'w, 2'', 4', 4''	
3OH					
4'	1.34, m	27.6	4'', 5', 5''	3, 5", 7	
4"	1.29, m	27.0	3w, 4', 5'w, 5''w	3, 24	2, 3w, 5w
5'	1.85, td (13.4, 4.0)	20.0	4', 4'', 5''	2', 4'', 8	4w, 6, 24w
5"	1.78, td (13.6, 4.8)	30.9	4', 4'', 5'	4', 4'', 24	4, 6, 7, 24w
6		73.6			
6OH	1.92, bs			5", 24	
7	5.25, d (9.8)	79.3	8	4', 8, 9, 24	8, 9, 29
8	5.91, dd (15.2, 9.8)	126.5	7, 9, 10w	5', 7, 9, 10	6w, 10
9	5.61, dd (15.2, 10.0)	140.5	8, 10	4', 7, 8, 10w, 25	7, 10, 25w
10	2.47, m	41.3	9, 11, 25	8w, 25, 26	8, 9, 11, 25w
11	5.20, d (10.6)	82.3	10	9, 10w, 13, 25, 26w	1, 9w, 10, 12, 13, 26
12		131.6			
13	6.19, d (10.9, 1.5)	131.8	14, 26w	11, 15, 25w	11, 15, 26
14	6.27, ddd (15.1, 10.8, 1.0)	126.6	13, 15	15, 16, 26, 27	12, 13, 16
15	5.81, dd (15.0, 8.4)	137.6	14, 16	14, 16, 27	12, 13, 16, 17w, 27
16	2.36, m	41.5	15, 17w, 27	14, 15, 17, 27	14, 15, 17w, 18w, 27
17	3.44, t (3.4)	73.0	16, 18	15, 16, 18, 19, 27	15, 16w, 18, 19, 27w
17OH					
18	2.57, dd (3.8, 2.2)	59.7	17, 19	17, 20, 27, 28	17w
19	3.02, dd (8.2, 2.3)	57.7	18, 20	17, 20w, 28	20
20	1.33, m	39.2	19, 21, 28	18, 21, 28	18w, 19, 21w, 28w
21	3.14, td (6.2, 4.4)	83.8	20, 22', 22''	20, 22'w, 22'', 23	19, 20w, 23
22'	1.63, m	22.0	21, 22", 23	21w, 22", 23	20, 21
22"	1.41, m	23.9	21, 22', 23	22', 23	20, 21, 23
23	0.86, t (7.4)	10.2	22', 22''	20, 21, 22', 22''	21, 22
24	1.08, s	24.8		5", 7	5, 6, 7
25	0.73, d (6.8)	16.5	10	9, 10, 11	9, 10, 11
26	1.61, d (1.3)	12.0	13	10, 14	11, 12, 13
27	1.12, d (7.0)	17.3	16	14, 15, 16, 17, 18w	15, 16, 17
28	0.89, d (7.0)	10.9	20	18w, 19w, 20, 22'	19, 20, 21
29		169.4			
30	1.64, s	20.9			29
31	3.23, s	57.7		19w, 21, 22", 23	21

Table S8. NMR data for 3*S*,17*S*-FD-895 (1g) in C₆D₆

^a Position numbering is provided in Supporting Figure S8.



Figure S9. Synthesis 7*R*,17*S*-FD-895 (**1h**) from core **5b** (Section E) and side chain **4d** (Section G). The entire synthetic pathway is shown using the methods and materials prepared in Chan *et al.* 2020.²⁵ In this publication, a central route was developed to enable preparation of analogues of FD-895 (**1**), these steps are shown in grey. Steps used specifically to prepare **1h** are shown in black. Experimental procedures are provided for these (black) steps

Position	$\delta_{\rm H}$, mult (<i>J</i> in Hz)	$\delta_{\rm C}$	¹ H, ¹ H-COSY	¹ H, ¹ H-NOESY	¹ H, ¹³ C-HMBC
1	4.65, d (9.3)	172.3			
2'	2.35, m	20.2	2', 3	3, 4''w	1, 3, 4
2"	2.30, dd (14.6, 3.2)	39.3	2", 3	3, 4''	1, 3, 4w
3	3.58, ddt (10.8, 7.4,	69.6	2'',w3OH, 4', 4''w	30H, 4'', 170H, 24w	
3OH	3.70, d (10.7)		3	2'., 2'', 3, 170H, 24	2w, 3
4'	1.80, m	20.0	3, 4", 5', 5"	30Hw, 5', 7w, 24	conformers
4"	1.62, m	30.6	3w, 4', 5', 5''	2'w, 5''	conformers
5'	1.58, m	265	4'w , 4'', 5''	4', 5''	conformers
5"	0.97, m	36.5	4', 4'', 5'	3w, 4''	conformers
6		73.8			
6OH	1.68, bs			3OH, 24	
7	5.41, m	82.9	8	8, 10w, 24, 26	5, 6, 8, 9, 14, 29
8	5.93, dd (15.3, 2.5)	127.9	7,9	4''/5', 7, 9, 10	7, 10
9	5.39, m	130.7	8, 10	4', 8, 10, 25	7, 8, 10
10	2.48, tq (10.3, 6.7)	41.1	11, 25	8, 9w, 25, 26	8, 9, 11, 25w
11	5.36, d (10.6)	78.0	10	10w, 13, 25, 26w	1, 10, 12, 13, 26
12		131.6			
13	6.17, d (10.7)	131.6	14, 26w	11, 14, 15, 16, 25, 26w	11, 15, 26
14	6.29, dd (15.1, 10.8)	126.4	13, 15	15, 16, 26, 27	12, 13, 16
15	5.80, dd (15.1, 8.4)	137.8	14, 16	13, 14, 16, 17w, 27	12, 13, 14, 10w, 17w, 27w
16	2.38, m	41.5	15, 17, 27	14, 15w, 17, 18, 27	14, 15, 17, 18
17	3.43, m	72.8	16, 17OH, 18	15, 16, 17OH, 18, 19, 27	15, 18, 19
17OH	1.67, m		17	3, 3OH, 24	
18	2.57, dd (3.8, 2.3)	59.6	17	15, 17, 20, 27, 28	17
19	3.02, dd (8.2, 2.2)	57.6	20	17, 17OH, 20, 28	17w, 18w, 20, 21w, 23w
20	1.33, m	39.0	19, 28	18, 21, 23	19, 28
21	3.15, m	83.7	20, 22', 22''	22', 22'', 23	19, 20, 22w, 23
22'	1.63, m	22.0	21, 22", 23	20w, 22''	20, 21, 23
22"	1.40, dp (14.2, 7.3)	23.8	21, 22', 23	22', 23	20, 21, 23
23	0.85, t (7.4)	10.0	22', 22''	20, 21, 22'w, 22''	21, 22
24	1.02, s	24.7		3, 30Н, 60Н, 7, 170Н	5, 6, 7
25	0.77, d (6.8)	16.9	10	9, 10, 11, 26w	9, 10, 11
26	1.64, d (1.2)	11.9		10, 14	11, 12, 13
27	1.12, d (7.0)	17.3	16	14w, 15w, 16, 17	15, 16, 17
28	0.89, d (7.0)	10.8	20	18, 19, 20, 21	19, 20, 21
29		169.3			
30	1.68, s	20.4			29
31	3.24, s	57.7		22', 22'', 23	21

Table S9. NMR data for 7*R*, 17*S* -FD-895 (1h) in C₆D₆

^a Position numbering is provided in Supporting Figure S9.



Figure S10. Synthesis of 17*S*,18*S*,19*S*-FD-895 (1i) from core 5 and side chain 4i, prepared in 4 steps from 13. The entire synthetic pathway is shown using the methods and materials prepared in Chan *et al.* 2020.²⁵ In this publication, a central route was developed to enable preparation of analogues of FD-895 (1), these steps are shown in grey. Steps used specifically to prepare 1i are shown in black. Experimental procedures are provided for these (black) steps.

Position	$\delta_{\rm H}$, mult (<i>J</i> in Hz)	δ_{C}	¹ H, ¹ H-COSY	¹ H, ¹ H-NOESY	¹ H, ¹³ C-HMBC
1		172.1			
2'	2.31, dd (14.8, 3.8)	20.5	2', 3	3w	1, 3, 4
2"	2.22, dd (14.8, 2.8)	38.5	2", 3	3w, 4'/5'	1
3	3.52, m	69.3	2', 2'', 4'w, 4''	2'', 4'/5'w, 5''	
ЗОН			3	3, 24, 30	
4'	1.58, m	20.2	4", 5', 5"	2", 5", 24	3w, 4w, 6, 24
4''	1.33, m	30.2	3, 4'	4', 5'	3, 5
5'	1.55, m	25.0	4'w, 4'', 5''	2", 4', 8w	4, 6, 24w
5''	1.27, m	35.8	4'	7	4, 6, 7, 24w
6		73.3			
6OH				3OH, 24	
7	5.25, d (10.6)	79.2	8	4'/5', 5'', 8w, 9, 24	8, 9, 24, 29
8	5.83, dd (15.2, 9.8)	126.5	7,9	4'/5', 7, 9, 10	6w, 10
9	5.64, (15.2, 10.0)	140.6	8, 10	5", 8, 10, 11, 25	7, 10, 11w, 25w
10	2.41, m	41.0	9, 11, 25	7w, 8, 25, 26	8, 9, 11, 25
11	5.27, d (11.1)	82.6	10	10, 13, 25	1, 9, 10, 12/13, 25, 26
12		131.7			
13	6.17, d (10.9)	131.3	14, 26w	11, 14, 15, 25	11, 14, 15, 26
14	6.32, dd (15.1, 10.8)	126.1	13, 15	13, 15, 16, 26, 27	12, 13, 16
15	5.75, dd (15.1, 8.2)	138.4	14, 16	13, 14, 16, 17w, 18w, 27	12, z13, 16, 17, 27
16	2.44, m	41.8	15, 17, 27	14, 15, 17, 27	14, 15, 17, 18, 27
17	3.48, dd (6.6, 3.6)	72.6	16, 18	15, 16, 18, 19w, 27	15w, 16w, 19, 27w
17OH					
18	2.78, dd (3.6, 2.3)	59.2	17	15w, 16, 17, 20, 27, 28	17
19	3.01, dd (7.4, 2.2)	57.3	20	17, 20, 28	18w, 20, 28
20	1.52, m	38.0	19, 28	18, 19w, 21, 28	19, 28
21	2.83, dt (7.5, 4.6)	84.7	20w, 22'	20, 22', 22''w, 23, 28, 31	23, 31
22'	1.49, m	22.5	21, 22", 23	21w, 23w	20, 21, 23
22"	1.38, m	23.3	23	21w, 23w	20w, 21w, 23
23	0.83, t (7.4)	10.6	22', 22''	21, 22'w, 22''	21, 22
24	1.02, s	24.7		4'/ 5', 7	5, 6, 7
25	0.72, d (6.8)	16.4	10	9, 10, 11, 26w	9, 10, 11
26	1.61, d (1.3)	11.8	13	10, 11w, 14	11, 12, 13, 15
27	1.20, d (6.9)	16.3	16	14, 15, 16, 17, 18w	15, 16, 17
28	1.02, d (6.9)	12.4	20	20, 21	19, 20, 21
29		169.1			
30	1.63, s	20.8			29
31	3.11, s	57.1		20, 21	21

Table S10. NMR data for 17*S*, 18*S*, 19*S*-FD-895 (1i) in C₆D₆

^a Position numbering is provided in Supporting Figure S10.



Figure S11. Synthesis of **17***S***,20***S***,21***S***-FD-895 (1j)** from core **3** and side chain **2j**, prepared in 10 steps from **7**. The entire synthetic pathway is shown using the methods and materials prepared in Chan *et al.* 2020.²⁵ In this publication, a central route was developed to enable preparation of analogues of FD-895 (1), these steps are shown in grey. Steps used specifically to prepare **1j** are shown in black. Experimental procedures are provided for these (black) steps.

Position	$\delta_{\rm H}$ mult (<i>L</i> in Hz)	δα	¹ H ¹ H-COSY	¹ H ¹ H-NOFSY	¹ H ¹³ C-HMBC
1	on, mun (o m mz)	172.1			
2,	2 29 dd (14 8 3 8)	172.1	3	3	134
2,,	2.29, dd (14.8, 3.8)	38.2	3	3 4'/5'	1, 5, 4
3	3 50 m	69.0	30H 4''	$2, \frac{1}{2}, \frac{1}{2}$	1
3 30Н	3.62 bs	07.0	3011, 4 3w/ 4''	3	
JOII 1'	1.56 m		3 1, 5,	2'' 8w 24	3w - 6 - 2Aw
+ 1''	1.30, m	30.0	3,4,5	2, 0w, 24 5' 7 0 24w	5 5 5
4 5,	1.23, III 1.54 m		<i>3</i> , <i>4</i> , <i>3 4</i> , <i>3 7 7 7 7 7 7 7 7 7 7</i>	2'' 4'' 8w	3
5,,	1.34, III	35.6	4,4,5	2,4,0W	3w, 4, 0, 24w
5	1.20, m	72.4	4,4,5	5,4,7,24W	4, 0W, /W
0		/3.4			
60H		70.0	0		0.0.04.00
7	5.24, d (6.6)	78.9	8	4', 4'', 8w, 9, 24	8, 9, 24w, 29
8	5.82, dd (15.2, 12.3)	126.2	7,9	4'/5', 7w, 9, 10	6w, 10
9	5.62, dd (15.2, 10.0)	140.5	8, 10	4", 8, 10, 11, 25	7, 10w, 25
10	2.38, m	41.0	9, 11, 25	8, 25, 26	11w, 25
11	5.26, d (7.5)	82.4	10	9, 10, 13, 25	1, 12, 26
12		131.4			
13	6.17, d (10.8)	131.4	14, 26	10w, 11, 14, 15, 25, 26w	11, 14, 15, 26
14	6.32 dd (15.2, 10.8)	126.2	13, 15	13, 15, 16, 26, 27	13, 16
15	5.83, dd (15.2, 10.4)	137.9	14, 16	13, 14, 16, 17w, 18w, 27	13, 16, 27w
16	2.44, tq (6.6, 6.3)	41.4	15, 17, 27	17w, 27	14, 15, 17, 18w, 27
17	3.36, t (4.9)	73.4	16, 18	15w, 16, 18, 19, 27	15w, 18
17OH					
18	2.74, dd (4.5, 2.3)	58.6	17, 19	16, 17, 20, 27, 28	17
19	2.95, dd (7.0, 2.3)	57.4	18, 20	17, 20, 28	20
20	1.55, m	37.8	19, 21, 28	18, 21, 28	19
21	2.84, dt (7.4, 4.7)	84.4	20, 22', 22''	20, 22', 22'', 23, 28, 31	19
22'	1.52, m	22.4	20, 21, 23	23w	23
22"	1.38, m	23.4	20, 21, 23	20, 21w, 28	23
23	0.85, t (7.4)	10.2	22', 22''	21w, 22', 22''	21, 22
24	1.00, s	24.5	,	4'/5', 7	5, 6, 7
25	0.70, d (6.7)	16.2	10	9, 10, 11, 13w	9, 10, 11
26	1.58, d (1.2)	11.6	13	10, 11, 14, 15, 25	11, 12, 13
27	1.14. d (6.9)	16.3	16	14, 15, 16, 17, 18w	15, 16, 17
28	1.02, d (7.5)	12.0	20	18w. 20. 21w	19, 20, 21
29		169.0			
30	161 s	20.4			29
31	3.11. s	56.9		20w. 21	21
51	····, ·	50.7			

Table S11. NMR data for 17*S*,20*S*,21*S*-FD-895 (1j) in C₆D₆

^a Position numbering is provided in Supporting Figure S11.

Compound	GI ₅₀ value (nM)	95% confidence interval
FD-895 (1)	1.92	1.00 to 2.83
3S-FD-895 (1a)	2.75	1.45 to 4.04
7 <i>R</i> -FD-895 (1b)	588.2	176.9 to 999.4
10 <i>R</i> ,11 <i>R</i> -FD-895 (1c)	41300	26500 to 56100
17S-FD-895 (1d)	2.17	1.47 to 2.86
17-O-Me-FD-895 (1e)	2.64	1.10 to 4.18
17S-O-Me-17S-FD-895 (1f)	40.5	25.8 to 55.2
3 <i>S</i> ,17 <i>S</i> -FD-895 (1g)	142.9	86.7 to 199.0
7 <i>R</i> ,17 <i>S</i> -FD-895 (1h)	858.5	553.9 to 1163.0
17 <i>S</i> , 18 <i>S</i> , 19 <i>S</i> -FD-895 (1i)	470.2	319.8 to 620.6
17 <i>S</i> ,20 <i>S</i> ,21 <i>S</i> -FD-895 (1j)	285.2	178.6 to 391.8

Table S12. GI₅₀ values for 1 and analogues 1a-1j.^a

^a HCT-116 cells were treated with **1a** or analogues **1a-1e** ranging from 0.1 to 5000 nM for 72 h, then cell viability was measured by Cell Titer MTS Assay (see Methods).

qPCR primers		
Primer	5' Sequence 3'	
Aurora A FP	CCACCTTCGGCATCCTAATA	
Aurora A RP	TCCAAGTGGTGCATATTCCA	
PLK-1 FP	CTCAACACGCCTCATCCTC	
PLK-1 RP	GTGCTCGCTCATGTAATTGC	
Aurora B FP	GGAGAGCTTAAAATTGCAGATTTTG	
Aurora B RP	TGCAGCTCTTCTGCAGCTCCT	
PHF5a FP	P GTTGCCATCGGAAGACTGT	
PHF5a RP	GCCCCTGGTAAGATCCATAGT	
SF3A1 FP	GGTGTTCCCAGAGAGCAGTAG	
SF3A1 RP	GCTGGGGCCATGTCTGTTTT	
Aurora A exon 3- exon 4 FP	AATTCTTCCCAGCGCATTC	
Aurora A exon 3- exon 4 RP	TCCTCAGGATTATTTTCAGGTG	
Aurora A exon 3- exon 7 FP	CTGCCATCGGCACCTGTATAT	
Aurora A exon 3- exon 7 RP	GCATGTACTGACCACCCAAA	
SF3B1 FP	GGGCTACTGATTTGGGGAGA	
SF3B1 RP	CATGAACCATAGCCTGTCAGC	
MCL-1 long FP	GTGCCTTTGTGGCTAAACACT	
MCL-1 long RP	AGTCCCGTTTTGTCCTTACGA	
MCL-1 short FP	GGCCTTCCAAGGATGGGTTT	
MCL-1 short RP	ACTCCAGCAACACCTGCAAAA	
DNAJB1 FP	GGCCTGATGGGTCTTATCTATGG	
DNAJB1 RP	TTAGATGGAAGCTGGCTCAAGAG	
ASF/SF2 FP	GAGTTCGAGGACCCGCGAG	
ASF/SF2 RP	CGGCCGCTTCGAGGAAACTC	
Aurora A FP	CCACCTTCGGCATCCTAATA	
Aurora A RP	TCCAAGTGGTGCATATTCCA	
PLK-1 FP CTCAACACGCCTCATCCTC		
PLK-1 RP GTGCTCGCTCATGTAATTGC		
FP: forward primer; RP reverse primer		

Table S13. Sequences of the qPCR primers used in this study.



Figure S12. Cell viability studies conducted on treatments HCT-116 cells for **a**) 4 h at ~20 times the GI_{50} value (upper left, Fig. 4) or **b**) 24 h at 500 nM. Cells were stained and data was collected using the Cell Titer MTS Assay (see Methods).

```
^{1}\text{H-NMR} (500 MHz) and ^{13}\text{C-NMR} (125 MHz) spectra of FD-895 (1) in C_{6}D_{6}
```



1 H, 1 H-gCOSY (500 MHz) spectrum of FD-895 (1) in C₆D₆



1 H, 1 H-NOESY (500 MHz) spectrum of FD-895 (1) in C₆D₆





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 1 H-NMR (600 MHz) and 13 C-NMR (125 MHz) spectra of 3*S*-FD-895 (**1a**) in C₆D₆












```
^{1}H-NMR (600 MHz) and ^{13}C-NMR (125 MHz) spectra of 7R-FD-895 (1b) in C<sub>6</sub>D<sub>6</sub>
```



¹H,¹H–gCOSY (500 MHz) spectrum of 7R–FD–895 (**1b**) in C₆D₆









¹H-NMR (600 MHz) and ¹³C-NMR (125 MHz) spectra of 10R, 11R-FD-895 (1c) in C₆D₆



¹H,¹H–gCOSY (500 MHz) spectrum of 10R,11R–FD–895 (**1c**) in C₆D₆



¹H,¹H–NOESY (500 MHz) spectrum of 10R,11R–FD–895 (**1c**) in C₆D₆





¹H,¹³C-HMBC (500 MHz) spectrum of 10R,11R-FD-895 (**1c**) in C₆D₆



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<sup>1</sup>H-NMR (500 MHz) and <sup>13</sup>C-NMR (125 MHz) spectra of 17S-FD-895 (1d) in C_6D_6
```



1 H, 1 H–gCOSY (500 MHz) spectrum of 17*S*–FD–895 (**1d**) in C₆D₆











 1 H-NMR (600 MHz) and 13 C-NMR (125 MHz) spectra of 17-O-methyl-FD-895 (1e) in C₆D₆



1 H, 1 H–COSY (500 MHz) spectrum of 17–O–methyl–FD–895 (1e) in C₆D₆





 1 H, 13 C-HSQC (600 MHz) spectrum of 17-O-methyl-FD-895 (1e) in C₆D₆



1 H, 13 C-HMBC (600 MHz) spectrum of 17-O-methyl-FD-895 (1e) in C₆D₆



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¹H-NMR (600 MHz) and ¹³C-NMR (125 MHz) spectra of 17-O-methyl-17*S*-FD-895 (**1f**) in C₆D₆ $_{175_{FD895_{K4_{17M}}}}$











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¹H-NMR (600 MHz) and ¹³C-NMR (125 MHz) spectra of 7*S*,17*S*-FD-895 (1h) in C_6D_6






















¹H-NMR (600 MHz) and ¹³C-NMR (125 MHz) spectra of 17*S*,20*S*,21*S*-FD-895 (1j) in C₆D₆













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Data File D:\DATA\2023\BURKART\BURKART-HPLC-01042023-A 2023-01-04 13-04-53\J5.D Sample Name: J5

Peak	RetTime	Туре	Width	Area	Height	Area	
#	[min]		[min]	[mAU*s]	[mAU]	00	
3	9.799	VB	0.0643	6278.65576	1401.18237	95.5469	
4	10.153	BB	0.0741	57.52671	11.16870	0.8754	
5	10.497	BB	0.0594	47.07749	11.58256	0.7164	
6	11.065	BB	0.0541	27.89825	7.73891	0.4245	
7	12.544	BB	0.0545	5.74987	1.65786	0.0875	
Tota	ls :			6571.28040	1466.52441		
=====	=========	=====	=========	=======================================	=================		

Data File D:\DATA\2023\BURKART\BURKART-HPLC-01042023-A 2023-01-04 13-04-53\A5-B.D Sample Name: A5: 4.0 ug loaded ______ Acq. Operator : Y. Su Seq. Line : 13 Acq. Instrument : Instrument 1 Location : Vial 2 Injection Date : 1/4/2023 5:32:29 PM Inj : 1 Inj Volume : 20.0 µl Different Inj Volume from Sequence ! Actual Inj Volume : 40.0 µl : D:\DATA\2023\BURKART\BURKART-HPLC-01042023-A 2023-01-04 13-04-53\HPLC-UV-MS-Acq. Method 2021-A.M Last changed : 1/4/2023 12:59:41 PM by Y. Su Analysis Method : C:\CHEM32\1\METHODS\HPLC-DATA-ANALYSIS-2022-A.M Last changed : 1/4/2023 1:51:37 PM by Y. Su (modified after loading) *MWD1 B, Sig=254,4 Ref=off (D:\DATA\2023\BURKART\BURKART-HPLC-01042023-A 2023-01-04 13-04-53\A5-B.D - D:\DATA\2023\BURK mAU 7R-FD-895 (1b) 160 140 120 100 80 60 40 11.338 20 Ω 6 8 10 12 14 16 min _____ Area Percent Report _____ Sorted By : Signal Multiplier: : 1.0000 : Dilution: 1.0000 Use Multiplier & Dilution Factor with ISTDs Signal 1: MWD1 B, Sig=254,4 Ref=off Signal has been modified after loading from rawdata file! Peak RetTime Type Width Area Height Area % # [mAU*s] [min] [min] [mAU] ----|-----|----|-----|-----|-----| 1 9.491 BB 0.0580 713.53564 181.14104 90.6033 12.10154 2 9.871 BV 0.0470 3.91669 1.5366

Data File D:\DATA\2023\BURKART\BURKART-HPLC-01042023-A 2023-01-04 13-04-53\A5-B.D Sample Name: A5: 4.0 ug loaded

Peak	RetTime	Туре	Width	Area	Height	Area	
#	[min]		[min]	[mAU*s]	[mAU]	00	
3	9.957	VB	0.0572	45.88954	11.85467	5.8270	
4	10.140	BB	0.0497	8.67712	2.61813	1.1018	
5	11.338	BB	0.0553	7.33431	1.97690	0.9313	
Total	s:			787.53816	201.50742		
=====	========	=====	========				
				*** End of	Report ***		

Data File D:\DATA\2023\BURKART\BURKART-HPLC-01042023-A 2023-01-04 13-04-53\B5-B.D Sample Name: B5: 4.0 ug loaded ______ Seq. Line : 14 Acq. Operator : Y. Su Acq. Instrument : Instrument 1 Location : Vial 3 Injection Date : 1/4/2023 5:54:49 PM Inj : 1 Inj Volume : 20.0 µl Different Inj Volume from Sequence ! Actual Inj Volume : 40.0 µl : D:\DATA\2023\BURKART\BURKART-HPLC-01042023-A 2023-01-04 13-04-53\HPLC-UV-MS-Acq. Method 2021-A.M Last changed : 1/4/2023 12:59:41 PM by Y. Su Analysis Method : C:\CHEM32\1\METHODS\HPLC-DATA-ANALYSIS-2022-A.M Last changed : 1/4/2023 1:51:37 PM by Y. Su (modified after loading) *MWD1 B, Sig=254,4 Ref=off (D:\DATA\2023\BURKART\BURKART-HPLC-01042023-A 2023-01-04 13-04-53\B5-B.D - D:\DATA\2023\BURK mAU 160 -140 10R,11R-FD-895 (1c) 120 100 80 60 40 9.216 20 0 6 8 10 12 14 16 min _____ Area Percent Report _____ Sorted By : Signal Multiplier: : 1.0000 : Dilution: 1.0000 Use Multiplier & Dilution Factor with ISTDs Signal 1: MWD1 B, Sig=254,4 Ref=off Signal has been modified after loading from rawdata file! Peak RetTime Type Width Area Height Area # [mAU*s] % [min] [min] [mAU] ----|-----|----|-----|-----|-----| 1 9.216 BV 0.0488 28.92623 9.16686 3.4638 9.327 VV 2 0.0629 721.26758 165.39879 86.3688

Data File D:\DATA\2023\BURKART\BURKART-HPLC-01042023-A 2023-01-04 13-04-53\B5-B.D Sample Name: B5: 4.0 ug loaded

Peak	RetTime	Туре	Width	Area	Height	Area	
#	[min]		[min]	[mAU*s]	[mAU]	00	
3	9.512	VV	0.0502	32.86232	9.53437	3.9351	
4	9.572	VV	0.0548	27.23235	7.25556	3.2610	
5	9.677	VB	0.0658	24.81337	5.28304	2.9713	
Total	ls :			835.10185	196.63863		
=====		=====					
				*** End of	Report ***		

Data File D:\DATA\2023\BURKART\BURKART-HPLC-01042023-A 2023-01-04 13-04-53\E5.D Sample Name: E5



Data File D:\DATA\2023\BURKART\BURKART-HPLC-01042023-A 2023-01-04 13-04-53\E5.D Sample Name: E5

Peak	RetTime	Туре	Width	Area	Height	Area	
#	[min]		[min]	[mAU*s]	[mAU]	00	
1	9.269	BV	0.0456	41.34902	13.96243	3.3074	
2	9.365	VB	0.0600	1025.93884	249.21840	82.0618	
3	9.749	BV	0.0542	79.59547	21.48303	6.3666	
4	9.839	VB	0.0637	72.47417	16.04055	5.7970	
5	10.027	BB	0.0585	30.84453	7.73407	2.4672	
Tota	ls :			1250.20203	308.43848		
=====	=========	=====		================		==========	

Data File D:\DATA\2023\BURKART\BURKART-HPLC-01042023-A 2023-01-04 13-04-53\C5.D Sample Name: C5: 4.0 ug loaded ______ Acq. Operator : Y. Su Seq. Line : 4 Acq. Instrument : Instrument 1 Location : Vial 4 Injection Date : 1/4/2023 2:11:00 PM Inj : 1 Inj Volume : 20.0 µl Different Inj Volume from Sequence ! Actual Inj Volume : 40.0 µl : D:\DATA\2023\BURKART\BURKART-HPLC-01042023-A 2023-01-04 13-04-53\HPLC-UV-MS-Acq. Method 2021-A.M Last changed : 1/4/2023 12:59:41 PM by Y. Su Analysis Method : C:\CHEM32\1\METHODS\HPLC-DATA-ANALYSIS-2022-A.M Last changed : 1/5/2023 10:05:03 AM by Y. Su (modified after loading) *MWD1 B, Sig=254,4 Ref=off (D:\DATA\2023\BURKART\BURKART-HPLC-01042023-A 2023-01-04 13-04-53\C5.D - D:\DATA\2023\BURKAR mAU 250 17-methoxy-FD-895 (1e) 200 150 100 50 11.766 11.258 0 6 8 10 12 14 16 min _____ Area Percent Report _____ Sorted By : Signal Multiplier: : 1.0000 : Dilution: 1.0000 Use Multiplier & Dilution Factor with ISTDs Signal 1: MWD1 B, Sig=254,4 Ref=off Signal has been modified after loading from rawdata file! Peak RetTime Type Width Area Height Area # [min] [min] [mAU*s] % [mAU] 1 11.258 BB 0.0491 8.02855 2.52526 0.7448 2 11.436 BB 0.0568 954.69629 254.49214 88.5691

Data File D:\DATA\2023\BURKART\BURKART-HPLC-01042023-A 2023-01-04 13-04-53\C5.D Sample Name: C5: 4.0 ug loaded

Peak	RetTime	Туре	Width	Area	Height	Area	
#	[min]		[min]	[mAU*s]	[mAU]	00	
3	11.766	BB	0.0779	69.58083	12.89894	6.4552	
4	13.833	BV	0.0566	7.35602	2.01246	0.6824	
5	13.987	VB	0.0542	38.24908	11.09269	3.5484	
Total	ls :			1077.91076	283.02150		
=====		=====					
				*** End of	Report ***		

Data File D:\DATA\2023\BURKART\BURKART-HPLC-01042023-A 2023-01-04 13-04-53\K5.D Sample Name: K5



Data File D:\DATA\2023\BURKART\BURKART-HPLC-01042023-A 2023-01-04 13-04-53\K5.D Sample Name: K5

Peak	RetTime	Туре	Width	Area	Height	Area	
#	[min]		[min]	[mAU*s]	[mAU]	00	
1	11.442	BB	0.0548	138.16333	37.68406	90.4594	
2	11.769	MM	0.0844	9.07993	1.79201	5.9449	
3	13.965	BB	0.0539	5.49186	1.60504	3.5957	
Total	s:			152.73512	41.08111		
=====	========	======					
				*** End of	Report ***		

Data File D:\DATA\2023\BURKART\BURKART-HPLC-01042023-A 2023-01-04 13-04-53\G5.D Sample Name: G5



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Data File D:\DATA\2023\BURKART\BURKART-HPLC-01042023-A 2023-01-04 13-04-53\G5.D Sample Name: G5

Peak	RetTime	Туре	Width	Area	Height	Area
#	[min]		[min]	[mAU*s]	[mAU]	olo
3	9.604	BV	0.0622	2360.77832	548.17621	84.4495
4	9.801	VB	0.0510	49.38311	14.06137	1.7665
5	9.951	BB	0.0628	103.31221	23.73966	3.6957
6	10.258	BV	0.0593	220.40396	54.38601	7.8843
7	10.401	VB	0.0600	26.77395	6.51367	0.9578
8	10.616	BB	0.0833	16.69579	2.77030	0.5972
Tota	ls :			2795.49041	654.72459	

Data File D:\DATA\2023\BURKART\BURKART-HPLC-01042023-A 2023-01-04 13-04-53\F5.D Sample Name: F5



Data File D:\DATA\2023\BURKART\BURKART-HPLC-01042023-A 2023-01-04 13-04-53\F5.D Sample Name: F5

Peak	RetTime	Туре	Width	Area	Height	Area
#	[min]		[min]	[mAU*s]	[mAU]	00
3	9.914	VV	0.0573	1611.82349	406.07657	91.9243
4	10.126	VV	0.0614	26.76514	6.19754	1.5264
5	10.274	VB	0.0596	16.39960	4.01876	0.9353
6	10.586	BB	0.0528	63.69561	17.76866	3.6326
7	10.980	BB	0.0548	5.99510	1.67153	0.3419
Total	ls :			1753.42422	443.48721	

Data File D:\DATA\2023\BURKART\BURKART-HPLC-01042023-A 2023-01-04 13-04-53\I5.D Sample Name: I5



S99

Data File D:\DATA\2023\BURKART\BURKART-HPLC-01042023-A 2023-01-04 13-04-53\I5.D Sample Name: I5

Peak	RetTime	Туре	Width	Area	Height	Area	
#	[min]		[min]	[mAU*s]	[mAU]	00	
3	9.804	BB	0.0621	4840.58594	1105.59180	95.6789	
4	10.157	BB	0.0709	42.31945	8.52066	0.8365	
5	10.500	BB	0.0547	32.17272	8.79540	0.6359	
6	11.067	BB	0.0506	19.46455	5.72739	0.3847	
7	12.547	BB	0.0548	4.25495	1.24645	0.0841	
Tota	ls :			5059.19702	1155.20261		
=====	========	=====	========				

Data File D:\DATA\2023\BURKART\BURKART-HPLC-01042023-A 2023-01-04 13-04-53\H5.D Sample Name: H5



Data File D:\DATA\2023\BURKART\BURKART-HPLC-01042023-A 2023-01-04 13-04-53\H5.D Sample Name: H5

Peak	RetTime	Туре	Width	Area	Height	Area	
#	[min]		[min]	[mAU*s]	[mAU]	00	
3	9.829	VV	0.0586	784.32239	196.46071	95.5690	
4	10.056	VV	0.0557	12.70005	3.38903	1.5475	
5	10.171	VB	0.0646	9.44691	2.13526	1.1511	
Total	s:			820.68738	206.17768		
=====	========	=====		============			
				*** End of	Report ***		