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Supporting Information

for

Isolation of fungi using the diffusion chamber device FIND technology

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Genomic sequences of isolated fungi, data on bioactivity and halotolerance, spectroscopic data of compounds 1 and 2 from *Heydenia* cf. *alpina* strain 824

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Table S1: Salinity dependency of the growth of *Cladosporium allicinum* on agar plates

	colony diameter after 14 days [mm]					
Salinity	plate 1	plate 2	plate 3	$\bar{x}$	σ^2	σ
0‰	55	53	54	54	1	±1
7‰	74	71	71	72	2	±1
14‰	90	87	89	89	2	±1
21‰	94	92	89	92	4	±2
28‰	106	106	108	106	1	±1
35‰	107	108	111	109	3	±2

σ = standard deviation

Table S2: Salinity dependency of the growth of *Heydenia* cf. *alpina* on agar plates

	colony diameter after 14 days [mm]					
Salinity	plate 1	plate 2	plate 3	$\bar{x}$	σ^2	σ
0‰	68	66	67	67	1	±1
7‰	123	125	123	124	1	±1
14‰	130	130	130	130	0	±0
21‰	130	130	130	130	0	±0
28‰	130	130	130	130	0	±0
35‰	130	130	130	130	0	±0

σ = standard deviation

Biological activities

Both compounds (**1**,**2**) were tested for antimicrobial activities against *Staphylococcus aureus* 133, *Bacillus subtilis* 168, *Micrococcus luteus* 4698, *Arthrobacter crystallopoietes* DSM 20117, *Escherichia coli* I-11276b, and *Klebsiella pneumoniae* sp. *ozeanae* I-10910 using agar diffusion assays on Culture plates (5% sheep blood Columbia agar, BD) overlayed with growth suspension of the bacteria to be tested. None of the compounds showed antimicrobial effects at concentrations of up to 100 µM.

Both compounds were tested for cytotoxicity in HEK293 cells using CellTiter-Blue cell viability assay. None of the compounds showed cytotoxic effects at concentrations of up to 100 µM.

Table S3: Agar diffusion assay with extracts of fungal isolates from experiments 1–4

	Inhibition zone [mm]				
Extract/chemical 1mg/mL	*Escherichia coli*	*Bacillus megaterium*	*Eurotium rubrum*	*Microbotryum violaceum*	*Mycotypha microspora*
Alternaria armoraciae	n.n.	**2**	n.n.	n.n.	n.n.
Auxarthron umbrinum	n.n.	**1**	n.n.	n.n.	n.n.
Chaetomium globosum	n.n.	**1,5**	n.n.	n.n.	n.n.
Chrysosporium sp.	n.n.	**1**	n.n.	n.n.	n.n.
Cladosporium allicinum	n.n.	**4**	n.n.	n.n.	n.n.
Clonostachys rosea	n.n.	**5**	n.n.	n.n.	n.n.
Cadophora sp.	n.n.	n.n	n.n.	n.n.	n.n.
Heydenia cf. alpina	n.n.	**5**	n.n.	n.n.	n.n.
Ilyonectria europaea	n.n.	**3**	n.n.	**3**	n.n.
Leucothecium sp.	n.n.	n.n	n.n.	n.n.	n.n.
Metarhizium carneum	n.n.	n.n	n.n.	n.n.	n.n.
Scopulariopsis brevicaulis	n.n.	n.n	n.n.	n.n.	n.n.
Benzylpenicillin	6	n.n.	n.n.	n.n.	n.n.
Streptomycin	n.n.	10	n.n.	n.n.	n.n.
Miconazole	n.n.	n.n	10	10	15

n.n. = no inhibition

Cytotoxicity assay

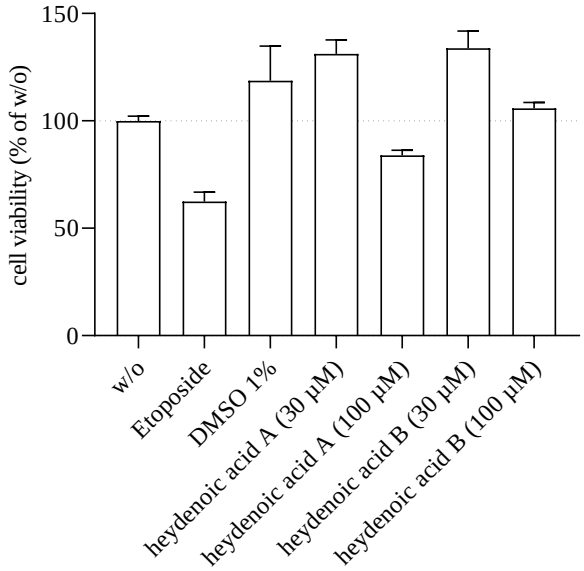

Figure S1: CellTiter-Blue cell viability assay

ITS sequences

Alternaia armoraciae **No. 830**
>Z1828-4418AI

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ACCCGCTGAACTTAAGC

>Z1828-4418AEFCL

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GCGCGCCGCCATTTCTGGCTTATCGCGATGAGGGGCATTTTTGGGTGGTGGGGTTGTGCGAA
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CAATTTCCCCAATTCGGCCACAGCTAACAAGCCTCACAGGAAGCCGCCGAACTCGGTAAGG

>Z1828-4418BI

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GCCTTAAAGTCATTGGCAGCCGGCCTACTGGTTTCGGAGCGCAGCACAAGTCGCGCTCTTCA
TCCAGCCAAGGTCAGCGTCCAGCAAGCCTTTTTTCAACCTTTGACCTCGGATCAGGTAGGGAT
ACCCGCTGAACTTAAGC

***Auxarthron cf. umbrinum* No. 825**
>Z1828-4420I

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***Cadophora* sp. No. 829**
>Z1876-4467I

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>Z1876-4467VS

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TGAACG

***Chaetomium globosum* No. 827**
>Z1828-4417BT

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>Z1828-4417RPB2

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***Chrysosporium sp.* No. 831**
>Z1876-4466I

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***Cladosporium allicinum* No. 823**
>Z1378-2864EF

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>Z1378-2864ITS

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***Clonostachys rosea* No. 821**
>Z2328-2172 ITS

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CCCAACTTTTCCAAGGT

***Heydenia* cf. *alpina* No. 824**
>Z2259-3333ITS

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***Ilyonectria europaea* No. 822**
>Z2328-2173BL ITS

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***Leucothecium* sp. No. 828**
>Z1876-4468I

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***Metarhizium carneum* No. 826**
>Z1876-4469I

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***Scopulariopsis brevicaulis* No. 832**
>Z1828-4419I

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>Z1828-4419EF2

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GGCTCAGGTCATCGTCCTCAACCACCCTGGTCAGATCGGCGCCGGCTACGCGCCCGTTCTTGA
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Spectroscopic data of compounds 1 and 2

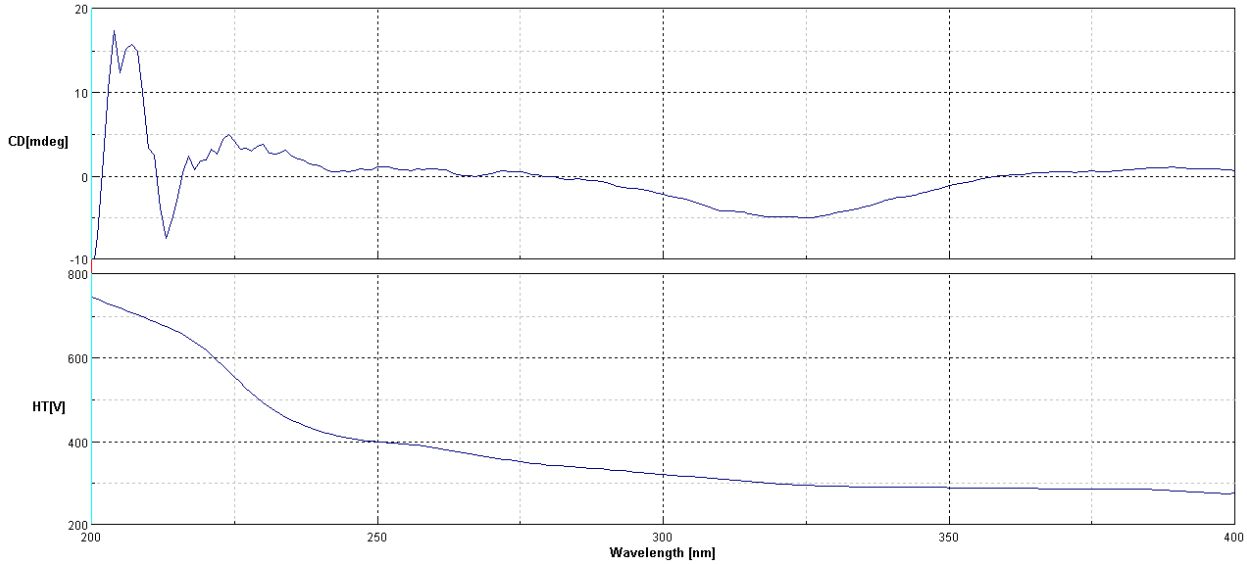

Figure S2: CD spectrum of heydenoic acid A (**1**) in MeOH

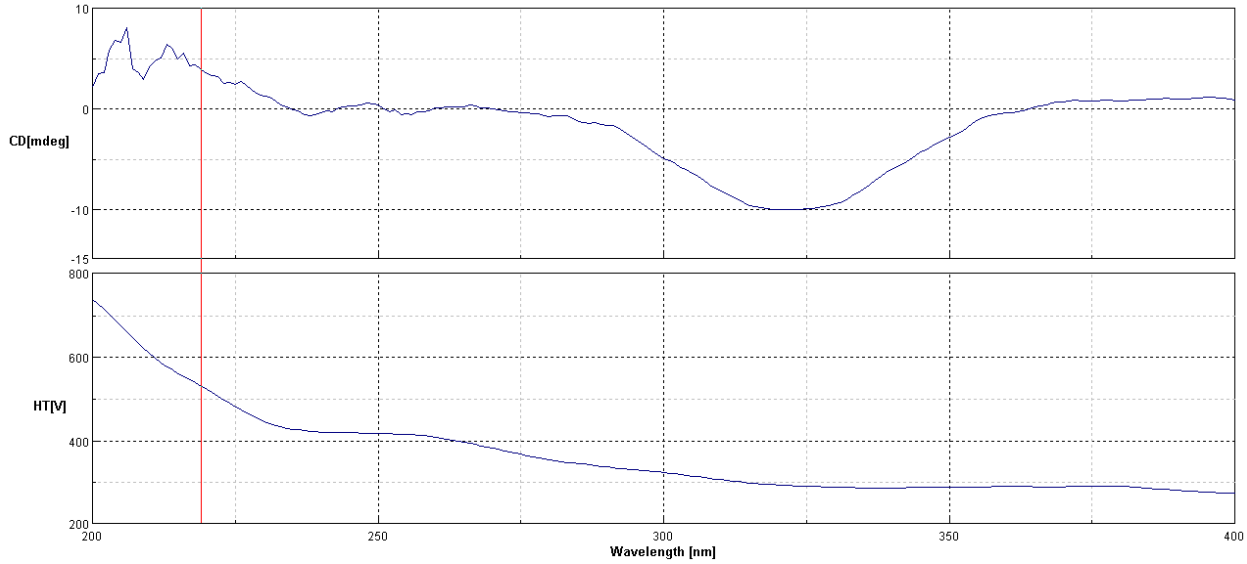

Figure S3: CD spectrum of heydenoic acid B (**2**) in MeOH

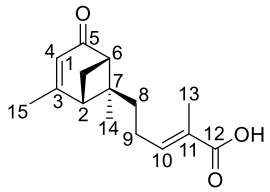

Table S4: ^{13}C NMR (75 MHz) and ^{1}H NMR (300 MHz) data of heydenoic acid A (**1**) in MeOD

C	δ_H	δ_C	COSY	HMBC	NOE
1	a: 2.12, m	41.9, CH_2	1b	2, 3, 5, 6, 7, 14	1b
	b: 2.93, dt (10.0, 6.0)		1a, 2, 6	2, 3, 5, 6, 7	1a, 2, 6, 8
2	2.65, t (6.0)	49.8, CH	1b, 4, 6	3, 4, 6, 7, 15	1b, 8, 14, 15
3		174.1, qC			
4	5.78, q (1.3)	122.1, CH	2, 6, 15	2, 6, 15	15
5		206.6, qC			
6	2.73, t (6.0)	57.2, CH	1b, 4	2, 4, 5, 7	1b, 8, 14
7		58.6, qC			
8	2.07, m	38.2, CH_2	9	7, 9, 14	1b, 6, 14
9	2.32, m	25.0, CH_2	8, 10	7, 8, 10, 11	13, 14
10	6.86, t (7.6)	143.0, CH	9, 13	12, 13	8, 9
11		129.5, qC			
12		177.8, qC			
13	1.89, s	12.5, CH_3	10	10, 11, 12	9
14	1.07, s	19.2, CH_3		2, 6, 7, 8	8, 15
15	2.10, d (1.3)	23.6, CH_3	4	2, 3, 4	2, 4, 14

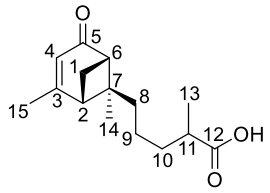

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Table S5: ^{13}C NMR (75 MHz) and ^{1}H NMR (300 MHz) data of heydenoic acid B (**2)** in MeOD

C	δ_H, (J in Hz)	δ_C, mult	COSY	HMBC	NOE
1	a: 2.10, m	41.9, CH_2	1b	2, 3, 5, 6, 7, 14	1b
	b: 2.89, dt (9.0, 5.6)		1a, 2, 6	2, 3, 5, 6, 7	1a, 2, 6, 8
2	2.59, t (5.6)	49.7, CH	1b, 4, 6	1, 3, 4, 6, 7, 8, 15	1b, 8, 14, 15
3		174.2, qC			
4	5.77, q (1.3)	122.0, CH	2, 6, 15	2, 6, 15	15
5		206.9, qC			
6	2.67, t (5.6)	57.3, CH	1b, 2, 4	1, 2, 4, 5, 7, 8	1b, 8, 14
7		58.8, qC			
8	1.94, m	39.4, CH_2	9	2, 6, 7, 9, 10, 14	1b, 2, 6, 14
9	1.37, m	23.4, CH_2	8, 10	7, 8, 10	
10	a: 1.49, m	35.5, CH_2	9, 10b, 11	9, 12	10b, 13
	b: 1.73, m		9, 10a, 11	9, 11, 13	10a
11	2.50, m	40.7, CH	10a, 10b, 13	9, 10, 12, 13	10a, 13
12		180.8, qC			
13	1.21, d (7.0)	17.8, CH_3	11	10, 11, 12	10a, 11
14	1.01, s	19.3, CH_3		2, 6, 7, 8	8
15	2.09, d (1.3)	23.6, CH_3	4	2, 3, 4	2, 4, 14

1D and 2D NMR spectra of heydenoic acid A (1)

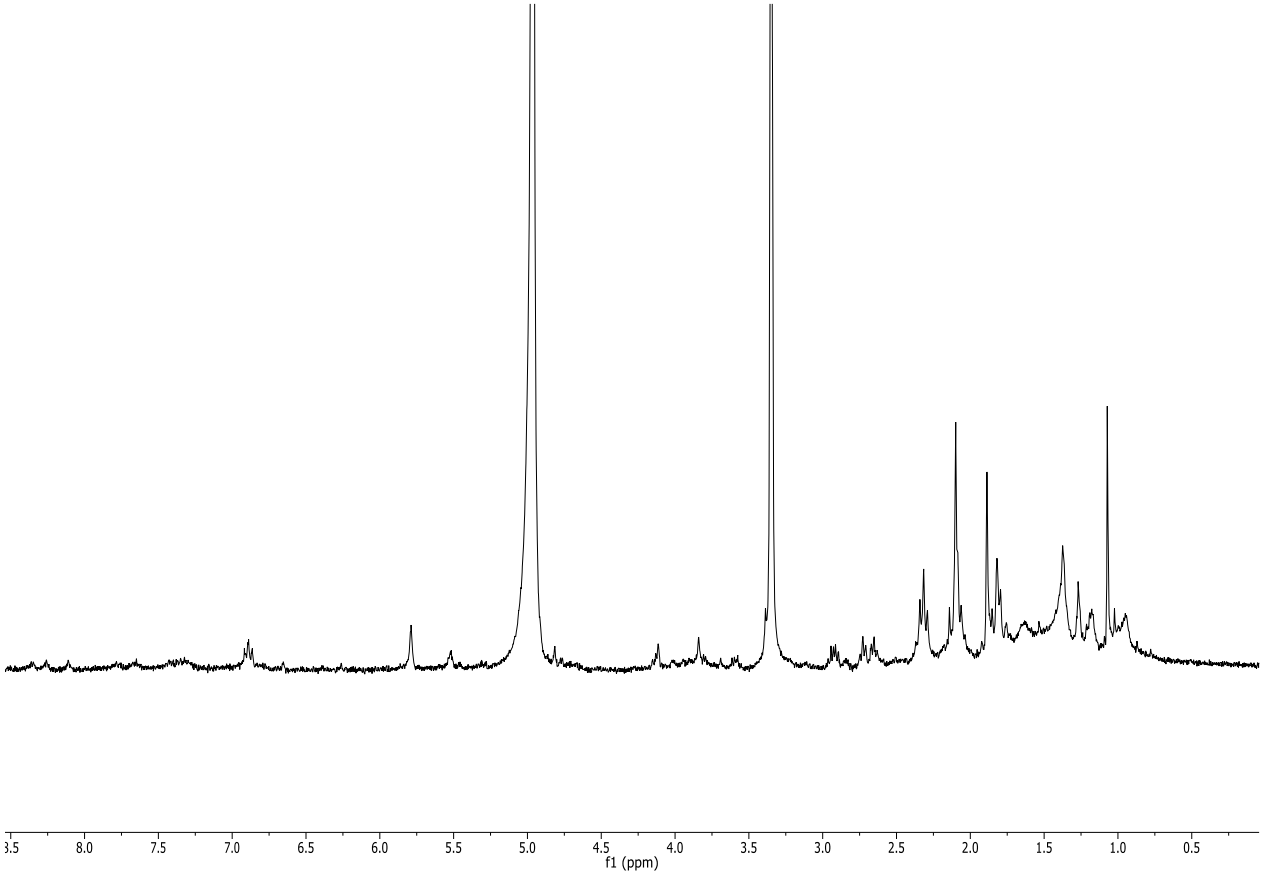

Figure S4: ^{1}H NMR of heydenoic acid A (1) (300 MHz, in MeOD)

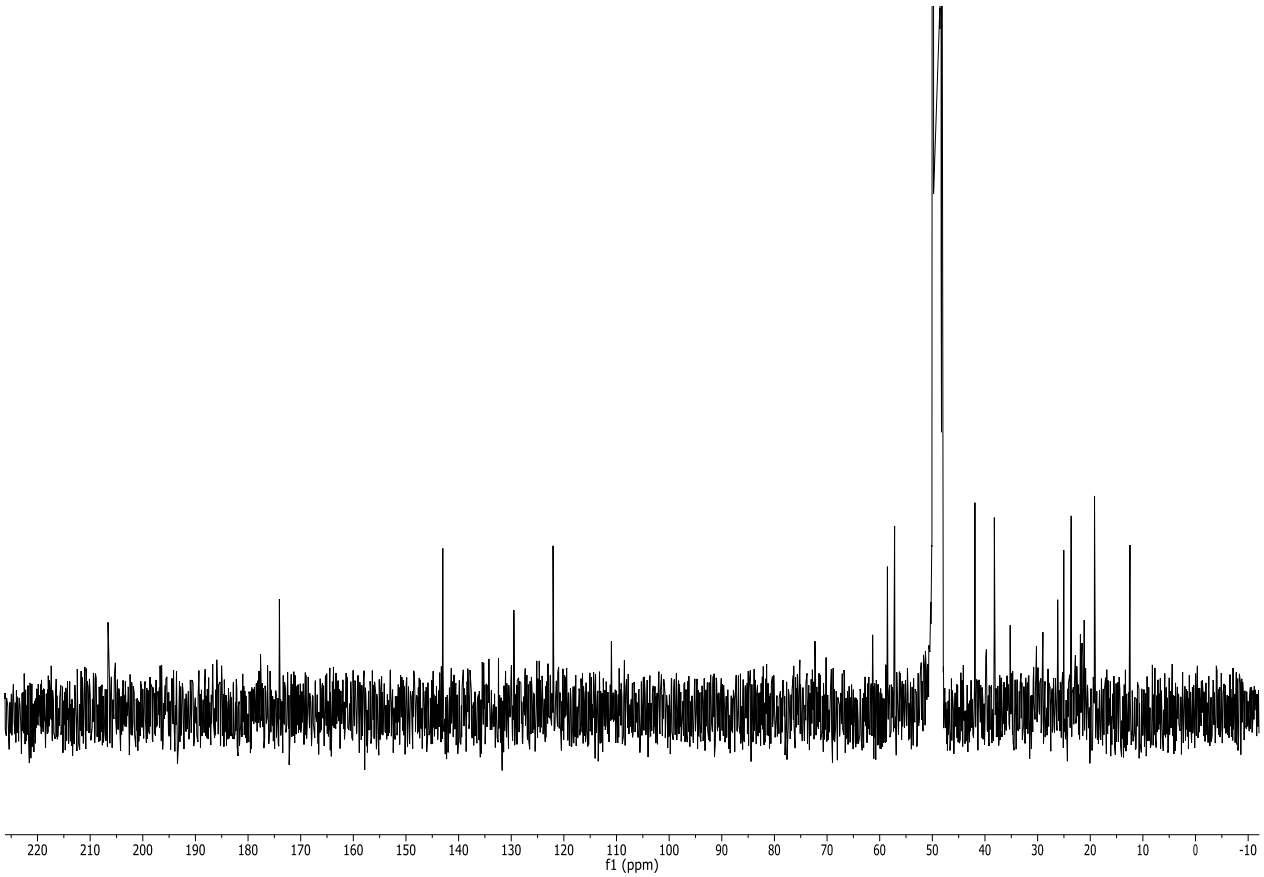

Figure S5: ^{13}C NMR of heydenoic acid A (**1**) (75 MHz, in MeOD)

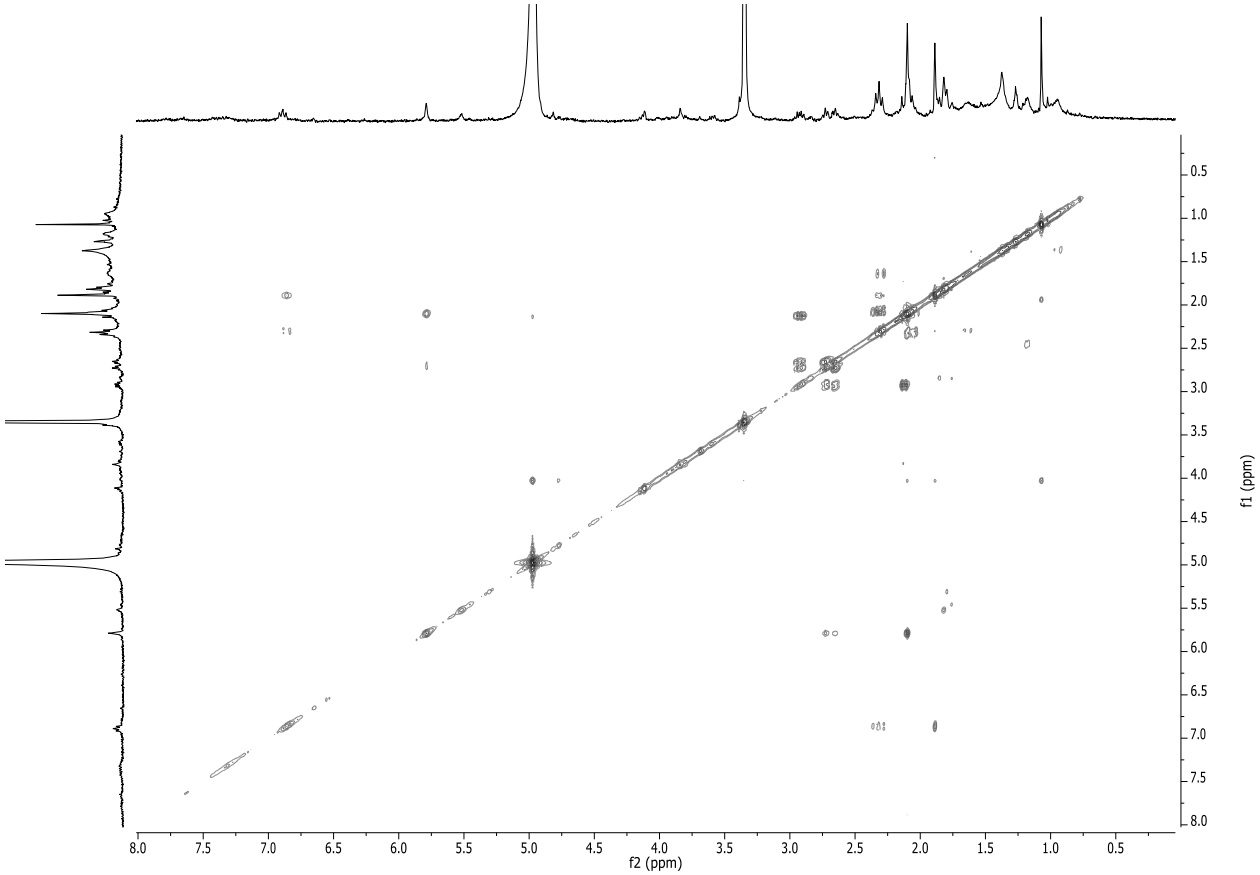

Figure S6: $^{1}H,^{1}H$-COSY spectrum for heydenoic acid A (**1**) in MeOD

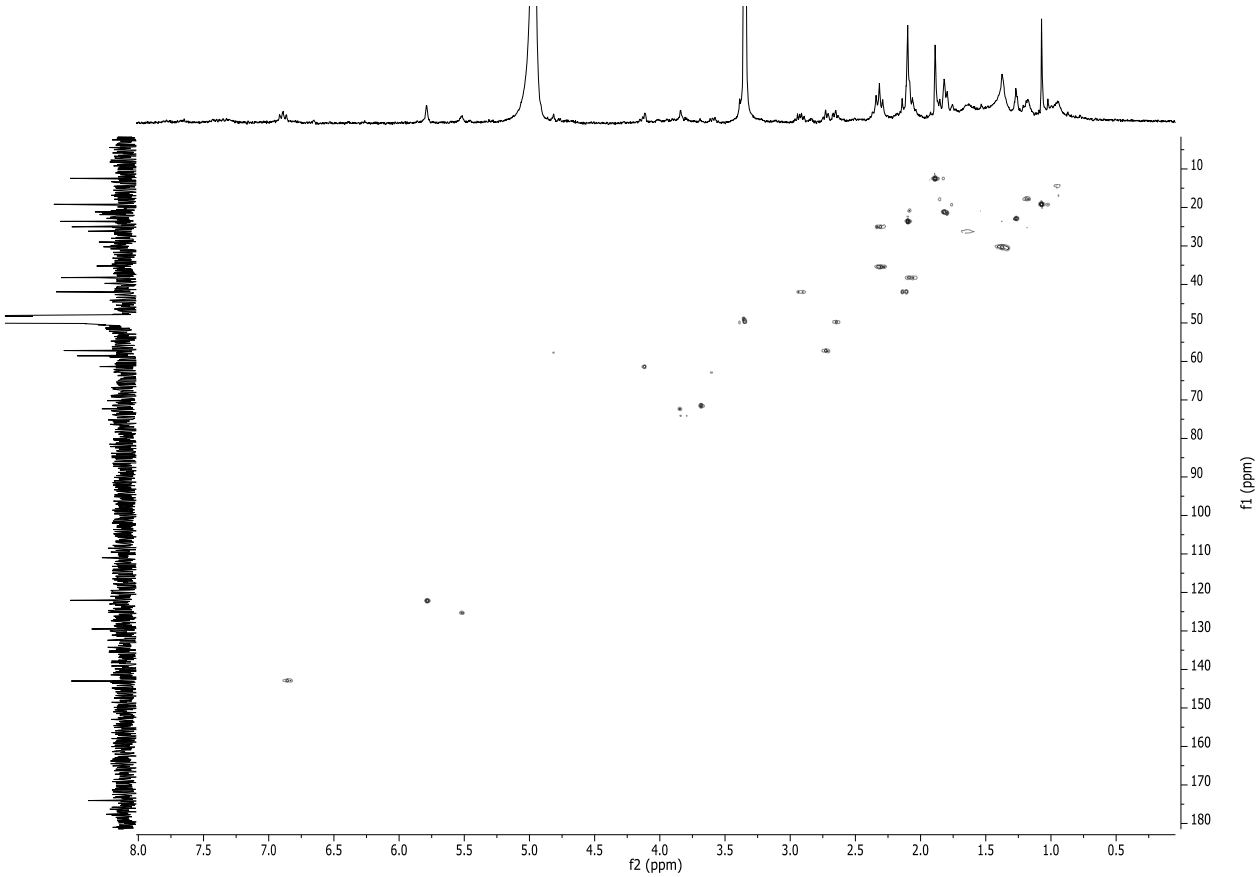

Figure S7: HSQC spectrum for heydenoic acid A (**1**) in MeOD

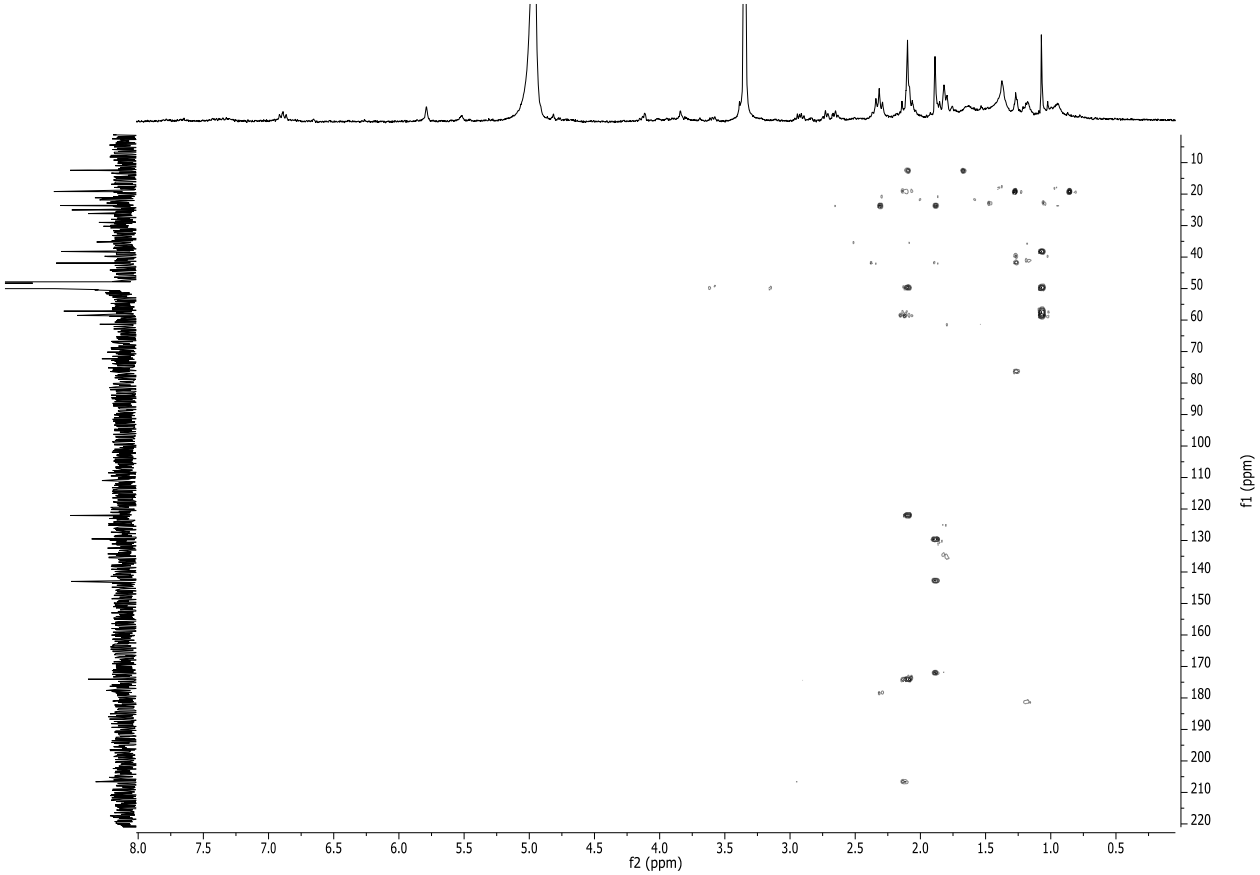

Figure S8: HMBC spectrum for heydenoic acid A (**1**) in MeOD

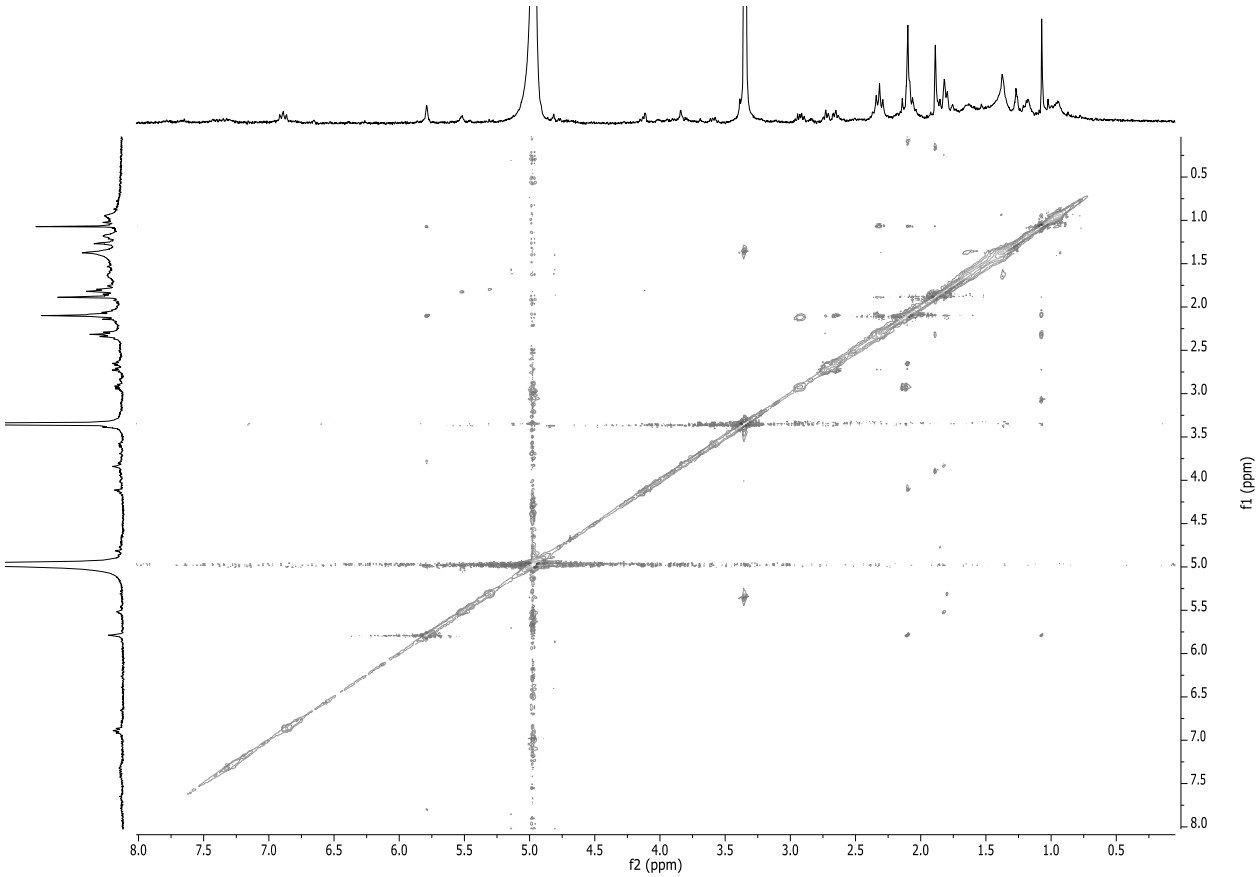

Figure S9: NOESY spectrum for heydenoic acid A (**1**) in MeOD

1D and 2D NMR spectra of heydenoic acid B (2)

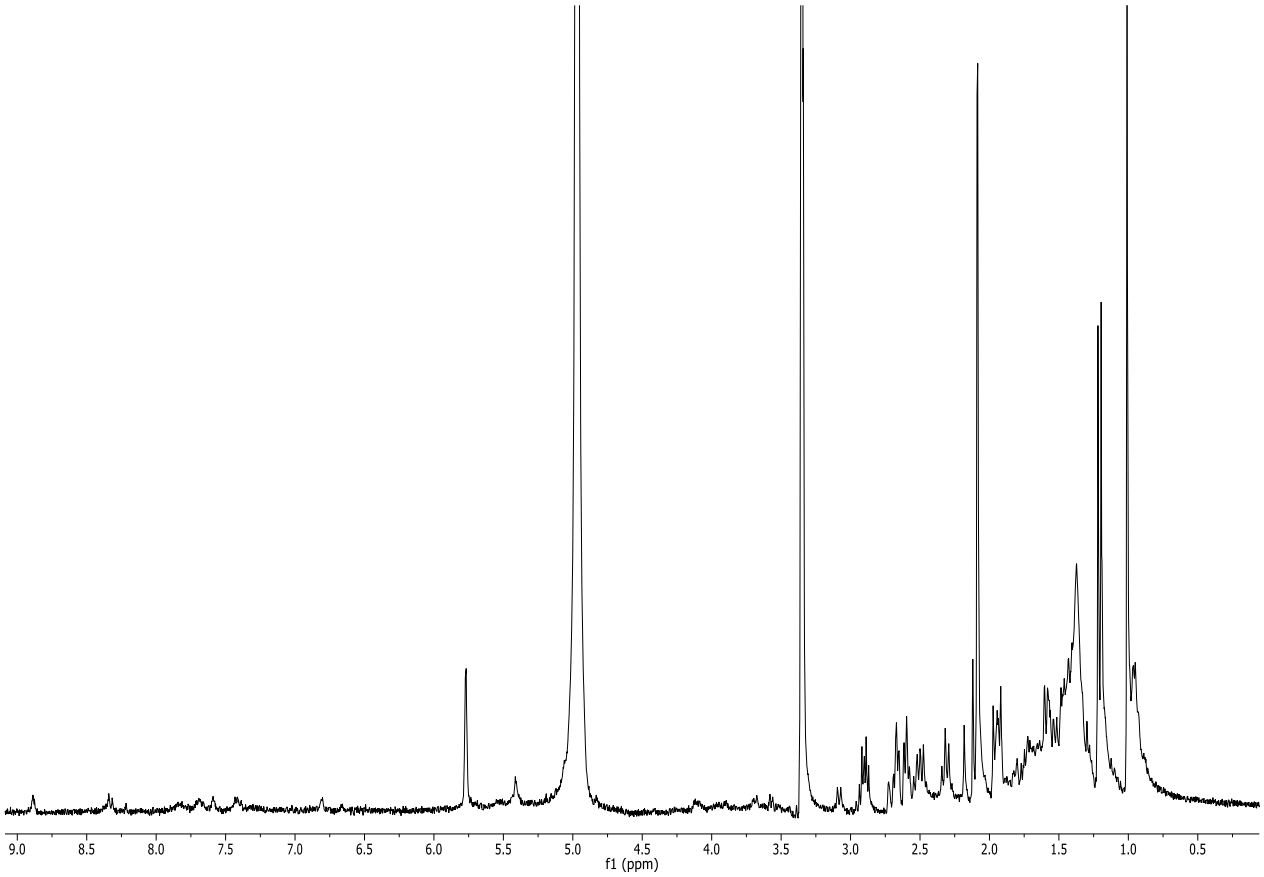

Figure S10: ^{1}H NMR of heydenoic acid B (**2**) (300 MHz, in MeOD)

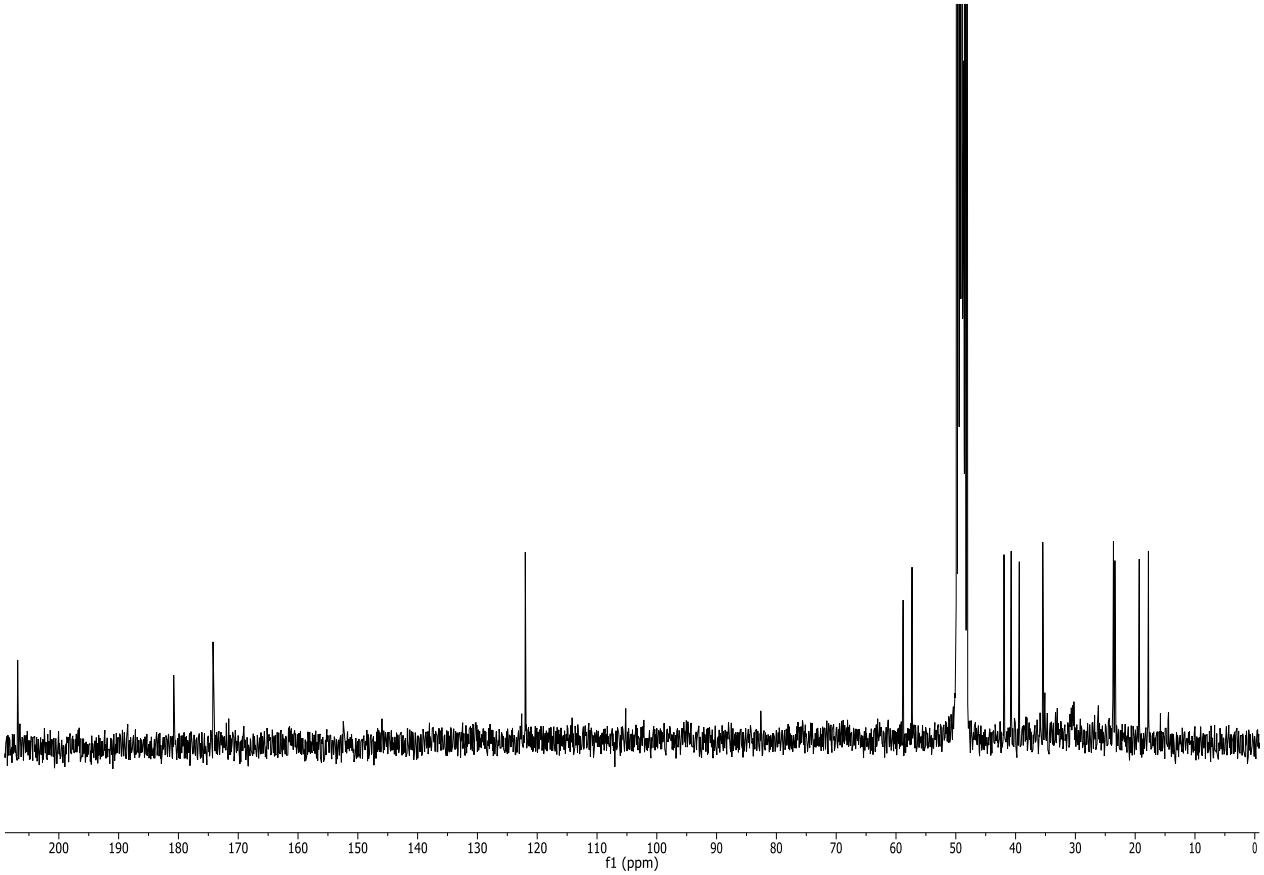

Figure S11: ^{13}C NMR of heydenoic acid B (**2**) (75 MHz, in MeOD)

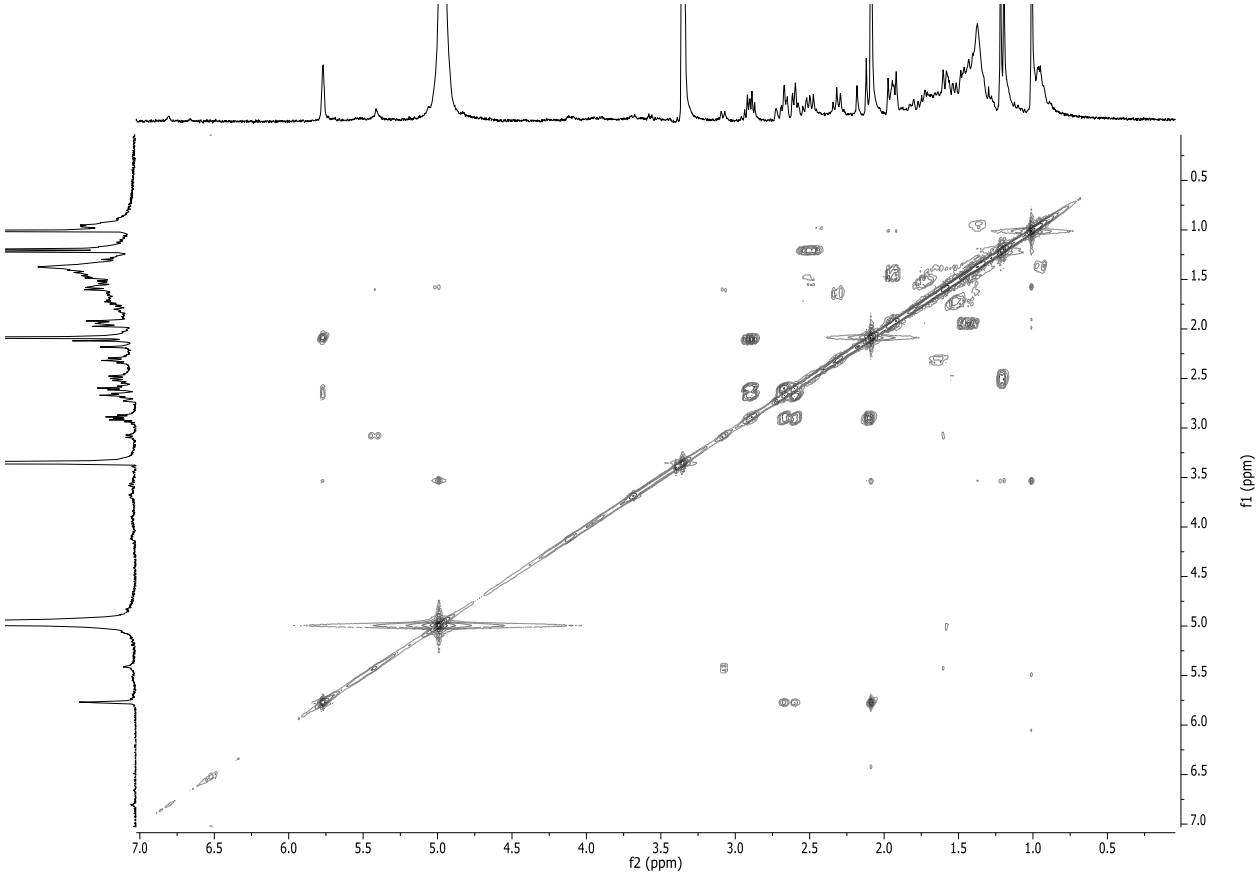

Figure S12: ^{1}H,^{1}H-COSY spectrum for heydenoic acid B (**2**) in MeOD

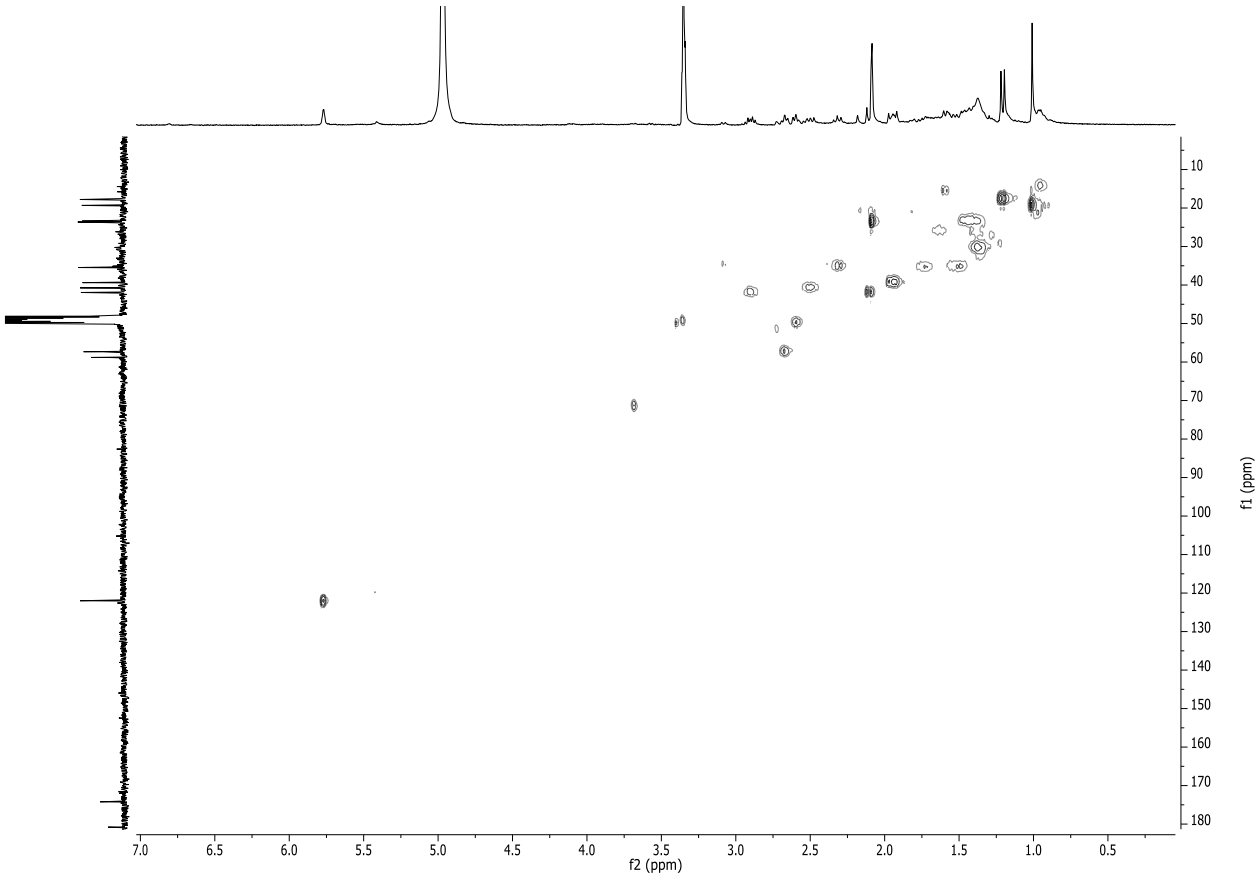

Figure S13: HSQC spectrum for heydenoic acid B (**2**) in MeOD

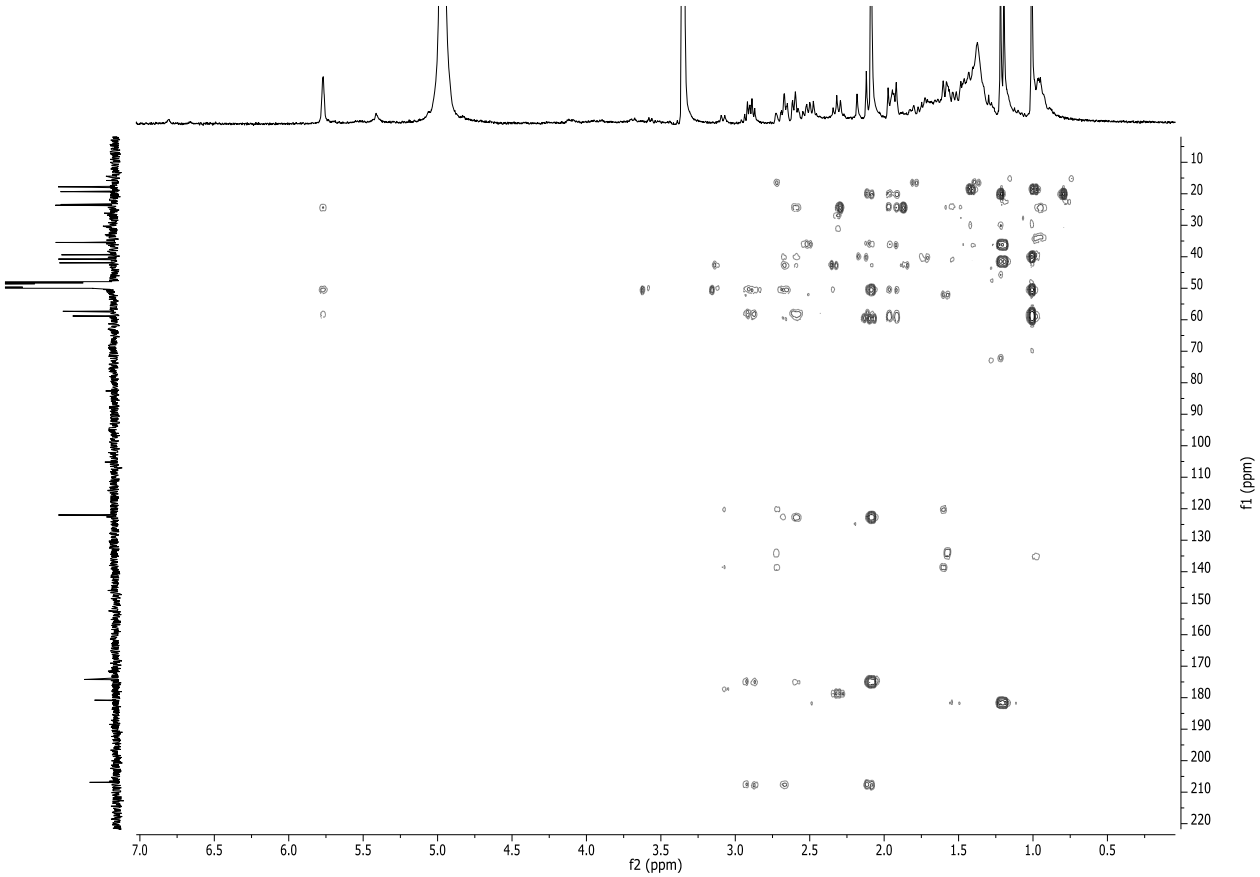

Figure S14: HMBC spectrum for heydenoic acid B (**2**) in MeOD

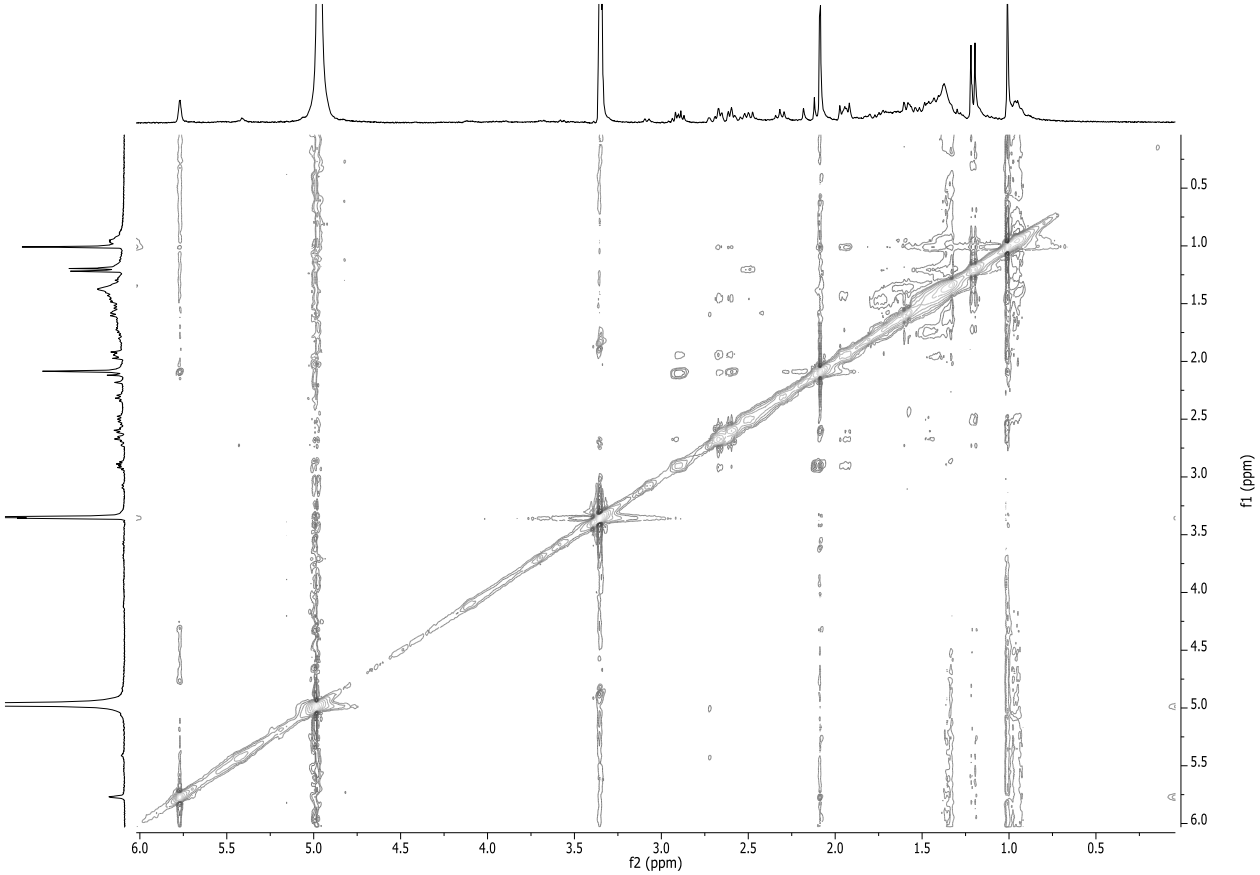

Figure S15: NOESY spectrum for heydenoic acid B (**2**) in MeOD