

Additional file 1

Optimization of the production process for the anticancer lead compound illudin M: downstream processing

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1. Comparison of analytical data from purified illudin M with published data

Table S 1 Chemical shifts of Illudin M in CDCl₃ (reported* and measured data)

Literature 1H	Measured 1H	Literature 13C	Measured 13C
0.43 (m, 1H)	0.40 (m, 1H)	200.5	200.6
0.83 (m, 1H)	0.83 (M, 1H)	146.6	146.8
0.96 (m, 1H)	0.94 (m, 1H)	138.8	138.9
1.11 (s, 3H)	1.09 (s, 3H)	134.5	134.6
1.15 (m, 1H)	1.11 (m, 1H)	132.9	133.1
1.17 (s, 3H)	1.19 (s, 3H)	78.9	79.0
1.37 (s, 3H)	1.35 (s, 3H)	75.9	76.1
1.55 (br s, 1H)	1.54 (br s, 1H)	49.1	49.2
1.69 (s, 3H)	1.67 (s, 3H)	31.6	31.8
3.55 (br s, 1H)	3.57 (br s, 1H)	27.3	27.4
4.41 (s 1H)	4.39 (s, 1H)	24.8	24.9
6.54 (s, 1H)	6.53 (s, 1H)	20.5	20.6
		14.2	14.3
		not reported	8.8
		not reported	6.1

* F.R. Kinder, K.W. Bair: **Total synthesis of (±) Illudin M**. J. Org. Chem. 1994, 59, 6965-6967

C₁₅H₂₁O₃: m/z [M+Na]⁺ calculated 271.1305, m/z [M+Na]⁺ measured 271.1307

m/z [M-OH]⁺ calculated 231.1385, m/z [M-OH]⁺ measured 231.1377

2. Estimation of purity using ^1H NMR data

To verify the purity of the crystalline material. A sample of 10 mg was dissolved in 500 μL CDCl_3 and a proton spectrum was recorded. The integration of the most dominant impurities in the spectrum (highlighted in Fig. S 1) indicates that the proportion non-illudin M to illudin M protons is smaller than 5%.

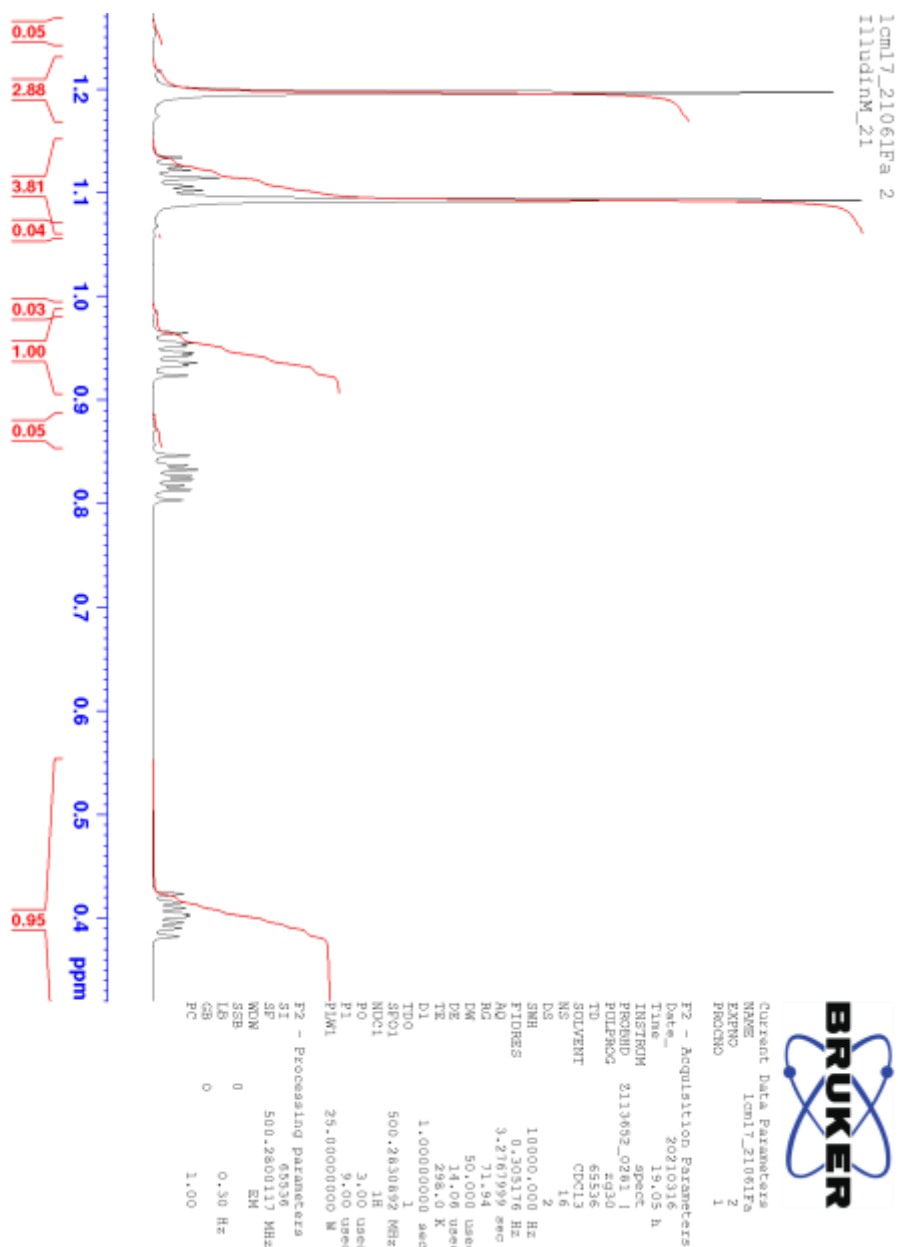


Fig. S 1 Enlargement of non-illudin M protons in the ^1H spectrum of illudin M crystals.

3. NMR spectra

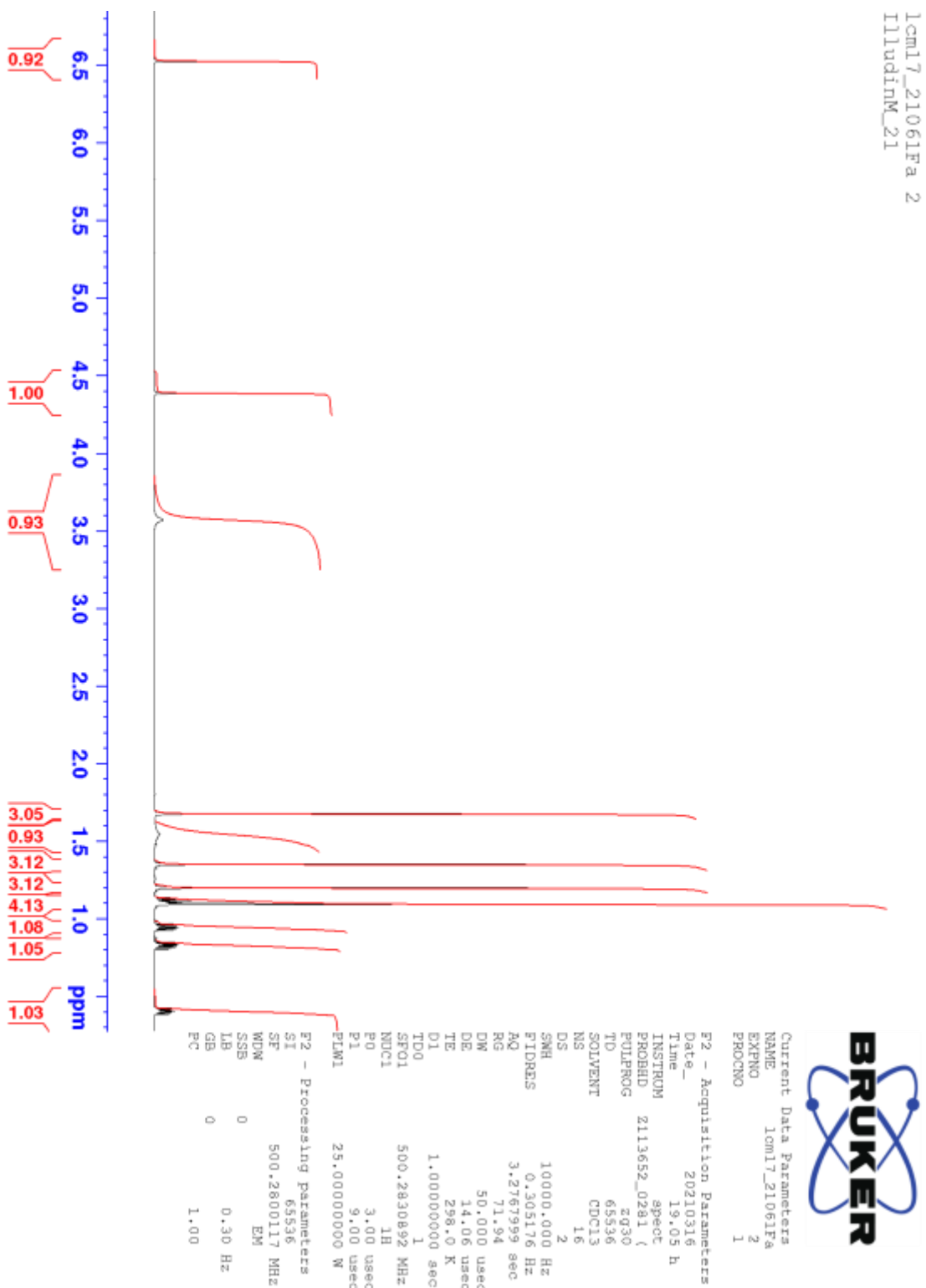


Fig. S 2 Proton NMR spectrum

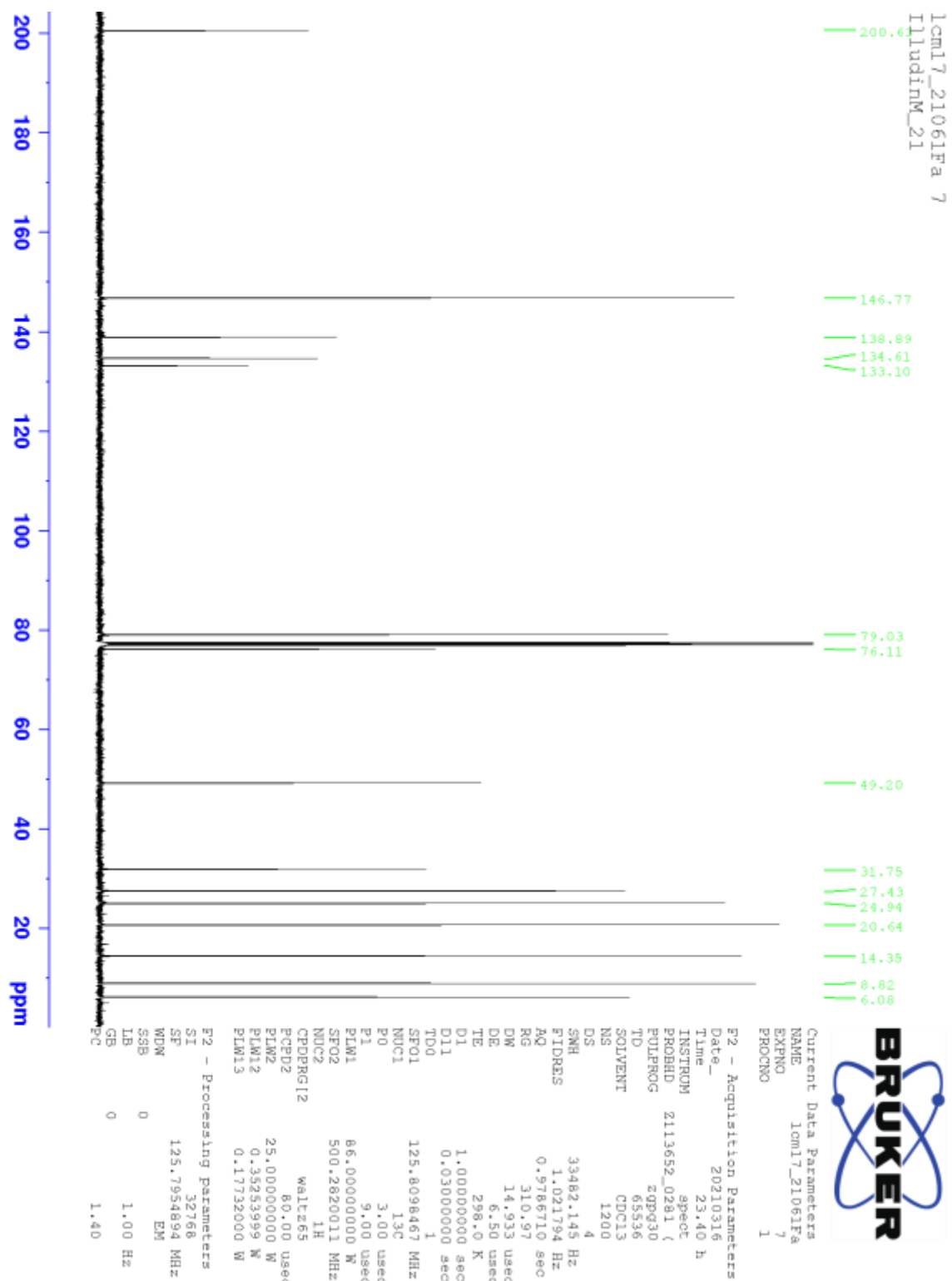
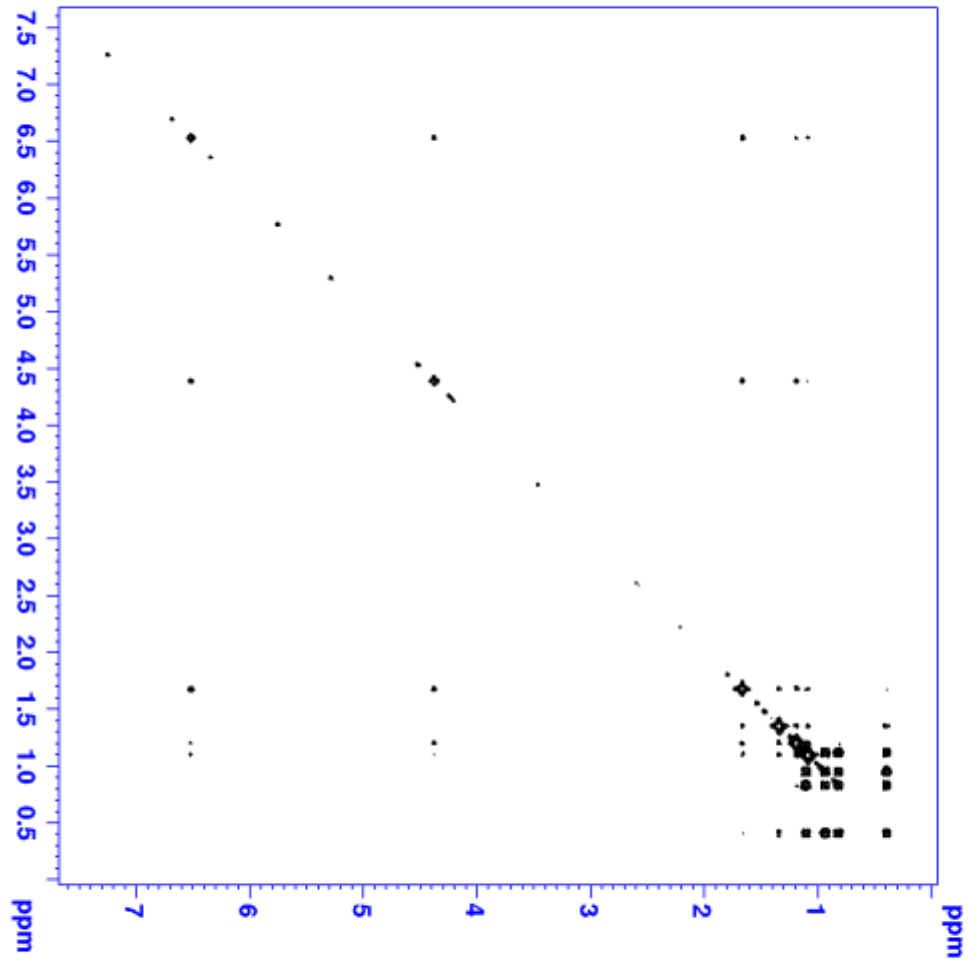


Fig. S 3 Carbon NMR spectrum

lcm17_21061Fa 3
 lludim_21
 COSY



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 DS 16
 SWH 3065.979 Hz
 FIDRES 3.775371 Hz
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 RG 184.28
 DW 129.333 usec
 DE 6.60 usec
 TE 298.0 K
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 D1 1.23999938 sec
 D11 0.03000000 sec
 D12 0.00000000 sec
 D13 0.00004000 sec
 D16 0.00020000 sec
 INO 0.00028600 sec
 TDav 1
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 NUC1 1H
 P0 9.00 usec
 P1 9.00 usec
 P17 2800.00 usec
 PL17 25.00300026 W
 PL10 2.20029933 W
 GRAM(1) SMO10.100
 GR21 10.00
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 FIDRES 30.210711 Hz
 SW 7.730 ppm
 FWHM 0.0
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 MON QSIINE
 SSB 0
 LB 0 Hz
 GB 0
 PC 1.40
 F1 - Processing Parameters
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 WDC 10.0
 SF 600.2800122 MHz
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 GB 0

Fig. S 4 COSY spectrum



BRUKER



1cm17_21061Fa 5
11ludim_21
HMBC

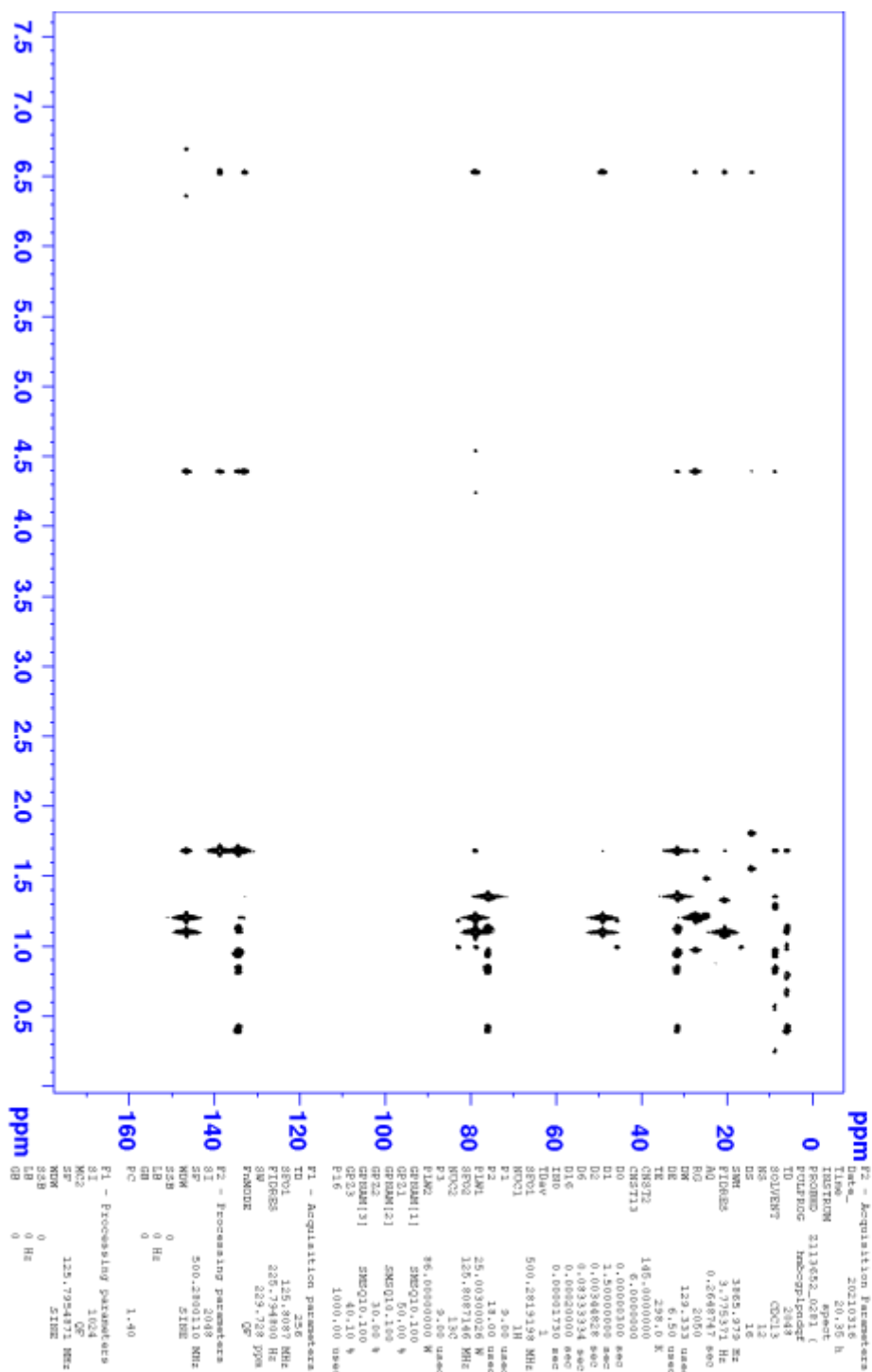


Fig. S 6 HMBC spectrum