

## Supplementary data contents page

**Title:** Oxysterols from a Marine Sponge *Inflatella* sp. and their Action in 6-Hydroxydopamine-Induced Cell Model of Parkinson's Disease

**Authors:** Sophia A. Kolesnikova<sup>1,\*†</sup>, Ekaterina G. Lyakhova<sup>1,†</sup>, Anatoly I. Kalinovsky<sup>1</sup>, Roman S. Popov<sup>1</sup>, Ekaterina A. Yurchenko<sup>1</sup>, and Valentin A. Stonik<sup>1,2</sup>

**Address:** <sup>1</sup>*G.B. Elyakov Pacific Institute of Bioorganic Chemistry, Far Eastern Branch of Russian Academy of Sciences, Pr. 100-let Vladivostoku 159, 690022 Vladivostok, Russia*

<sup>2</sup>*School of Natural Science, Far Eastern Federal University, Sukhanova St., 8, Vladivostok 690000, Russia*

**Correspondence:** \*[sovin81@inbox.ru](mailto:sovin81@inbox.ru); Tel.: +7-423-231-1168

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- S23** HSQC NMR (500.13 MHz) spectrum of the (22*E*)-24-*nor*-cholesta-5,22-diene-3β,7β-diol (**4**) in CDCl<sub>3</sub>
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- S25** Viability of Neuro2a cells
- S26** ROS formation in Neuro2a cells
- S27** Viability of Neuro2a cells treated with 6-OHDA

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† These authors contributed equally to the work.

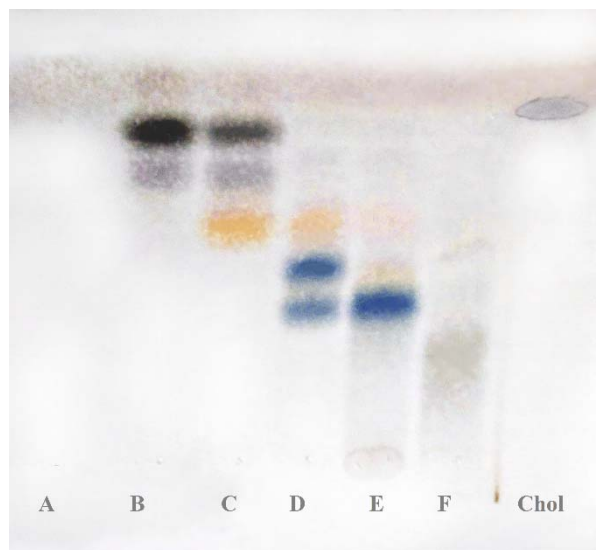
## S1 Experimental Section

### General experimental procedure:

$^1\text{H}$  NMR (500.13, 700.13 MHz) and  $^{13}\text{C}$  NMR (125.75 MHz, 176.04 MHz) spectra were recorded in  $\text{CDCl}_3$  on a Avance-III 700 and DRX-500 «Bruker». The  $^1\text{H}$  and  $^{13}\text{C}$  NMR chemical shifts were referenced to the TMS or to solvent peak for  $\text{CDCl}_3$  at  $\delta_{\text{H}}$  7.26 and  $\delta_{\text{C}}$  77.0.

HRESI-MS: Agilent 6510 Q-TOF LC/MS, 0.01 mg/ml ( $\text{CDCl}_3$ ),  $5\mu\text{l min}^{-1}$ . LSI-MS: AMD-604 (AMD Intectra), 1 mg/ml. HPLC: YMC-Pack ODS-A ( $5\mu$ ,  $250\times 4.6$  mm).

### TLC examination of fractions A-F :



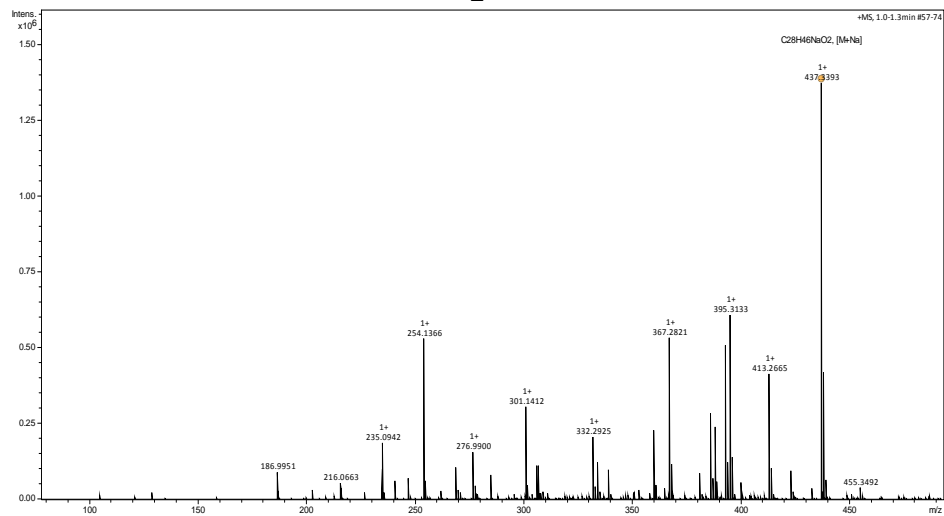
TLC was carried out on silica gel plates (CTX-1A, 5-17  $\mu\text{m}$ , Sorbfil, Russia),  $\text{CHCl}_3/\text{EtOH}$  (25:1).

### Statistical analysis

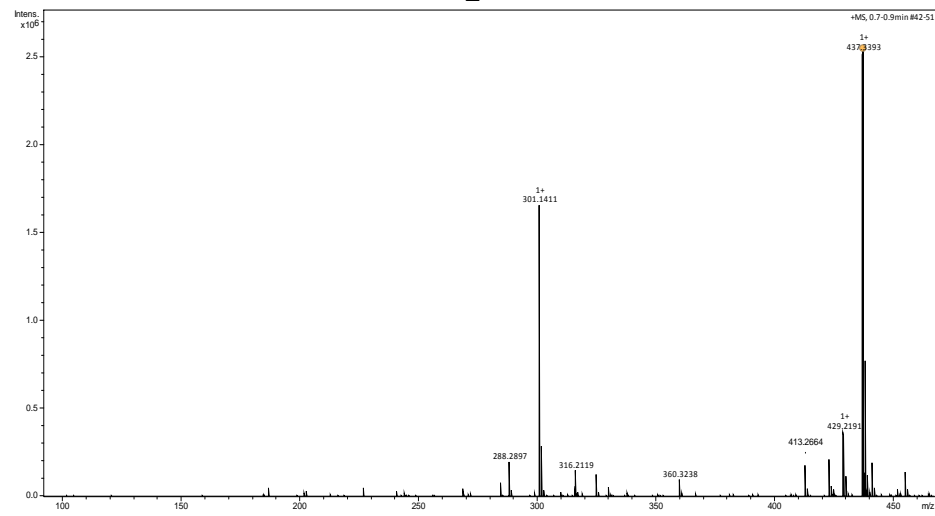
All assays were performed at least in triplicate. The results are expressed as the mean  $\pm$  standard deviation (SD). A Student's *t*-test was used to evaluate the data with the significance level of  $p < 0.05$ . The means and standard errors for each treatment were calculated and plotted using SigmaPlot 3.02 software (Jandel Scientific, San Rafael, CA).

# S2 HRESI MS Spectra (Positive Ion Mode) of compounds 1-4 in CDCl<sub>3</sub>

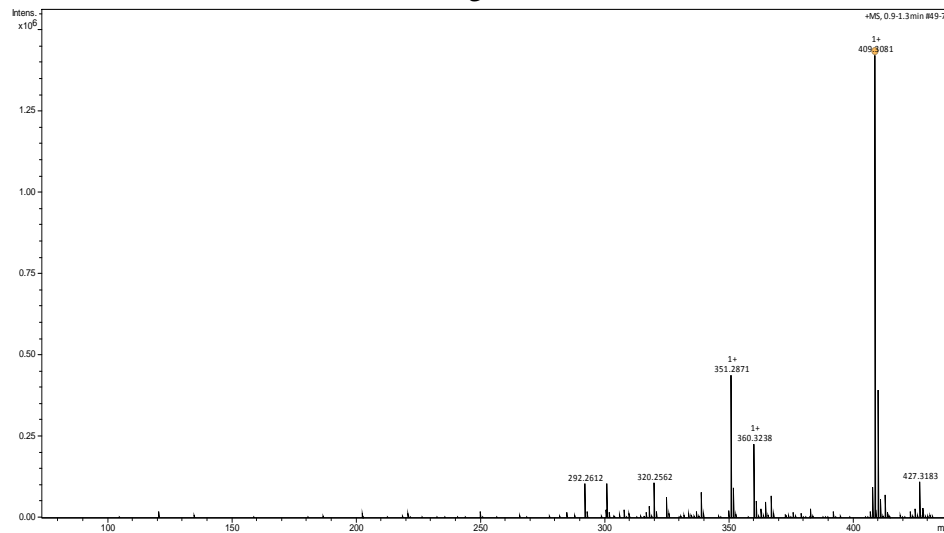
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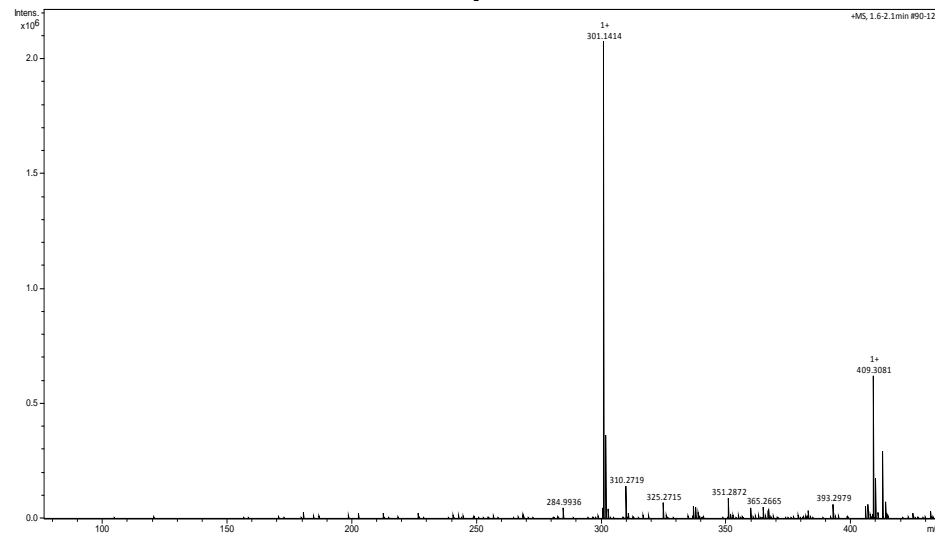
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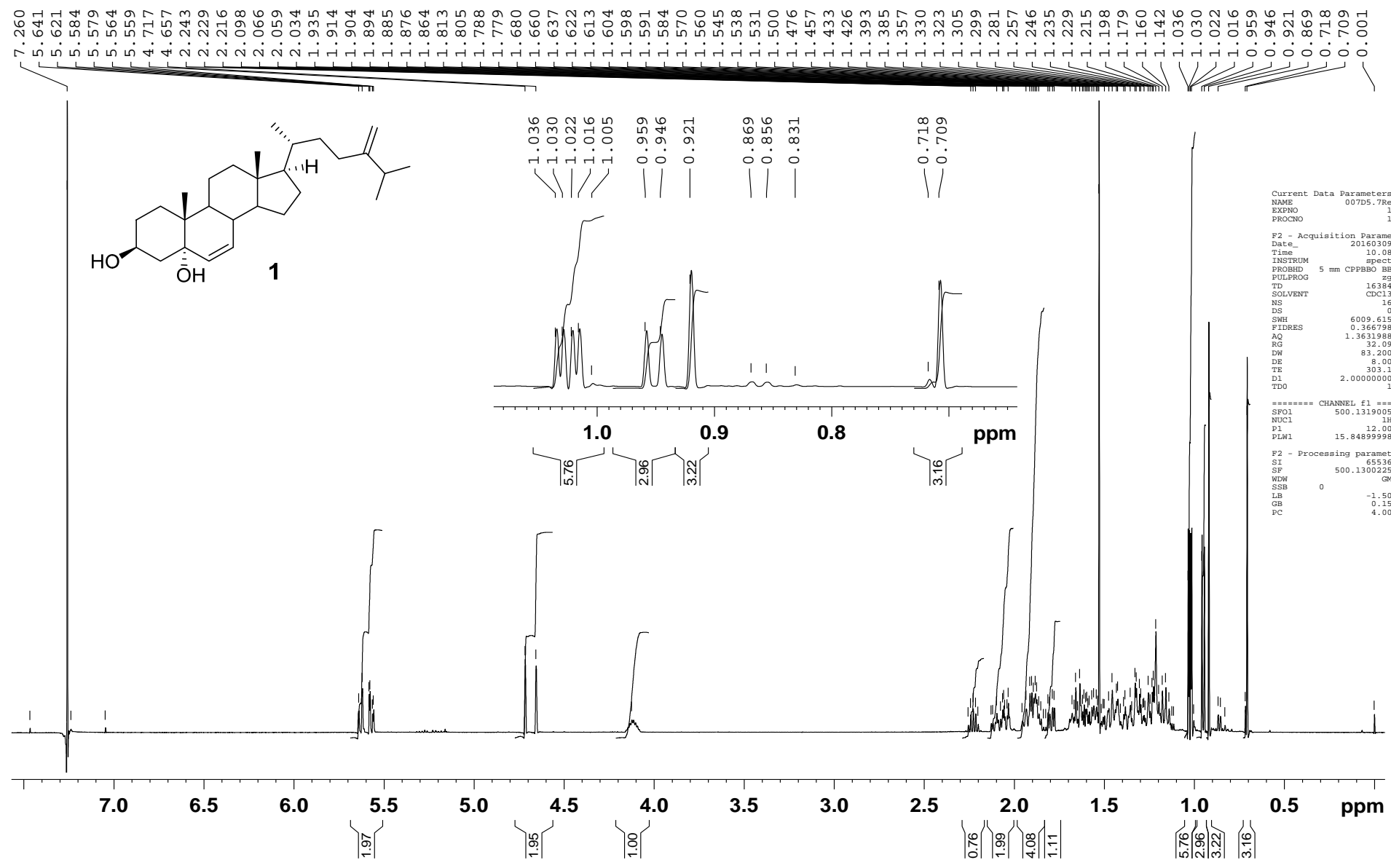
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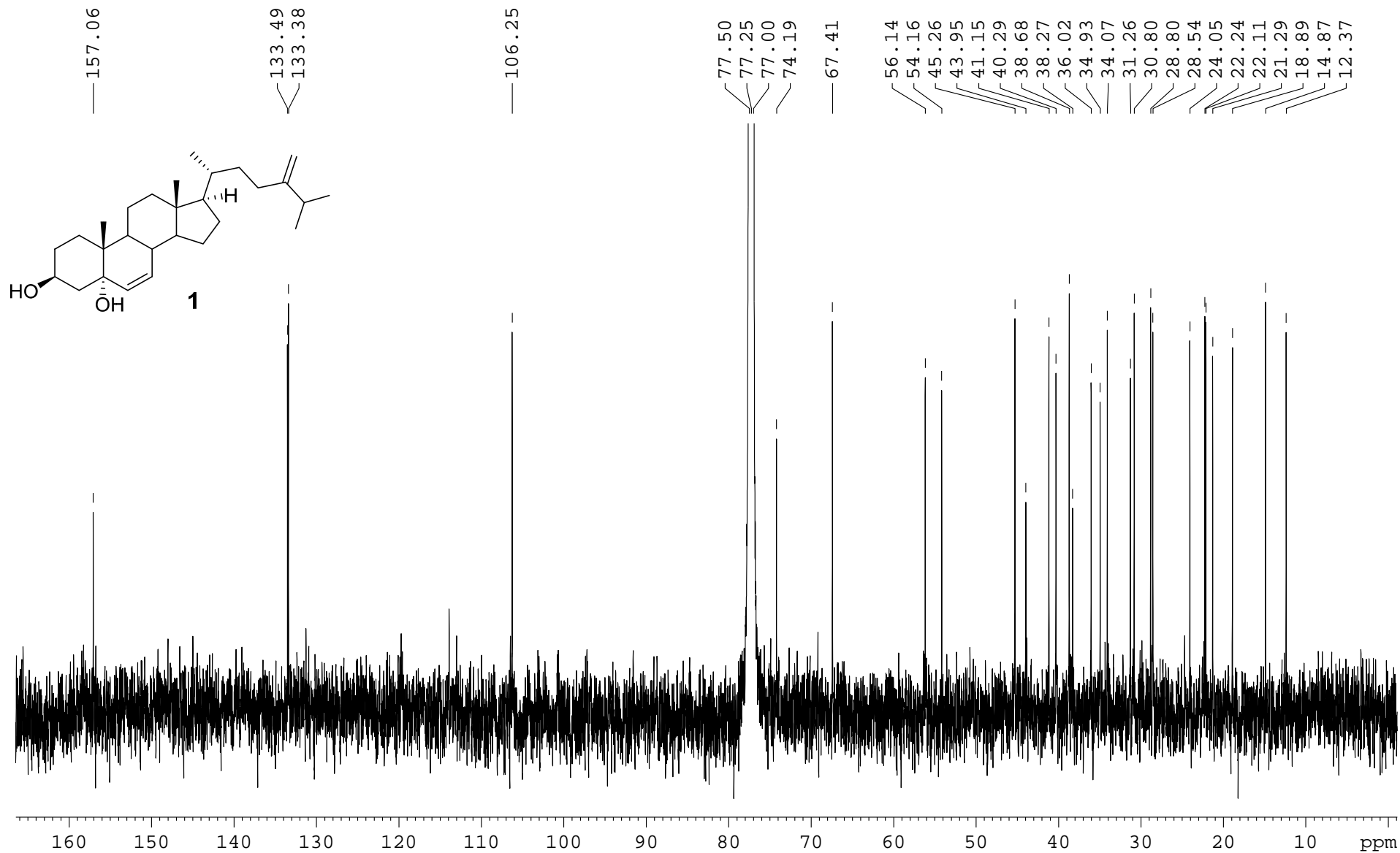
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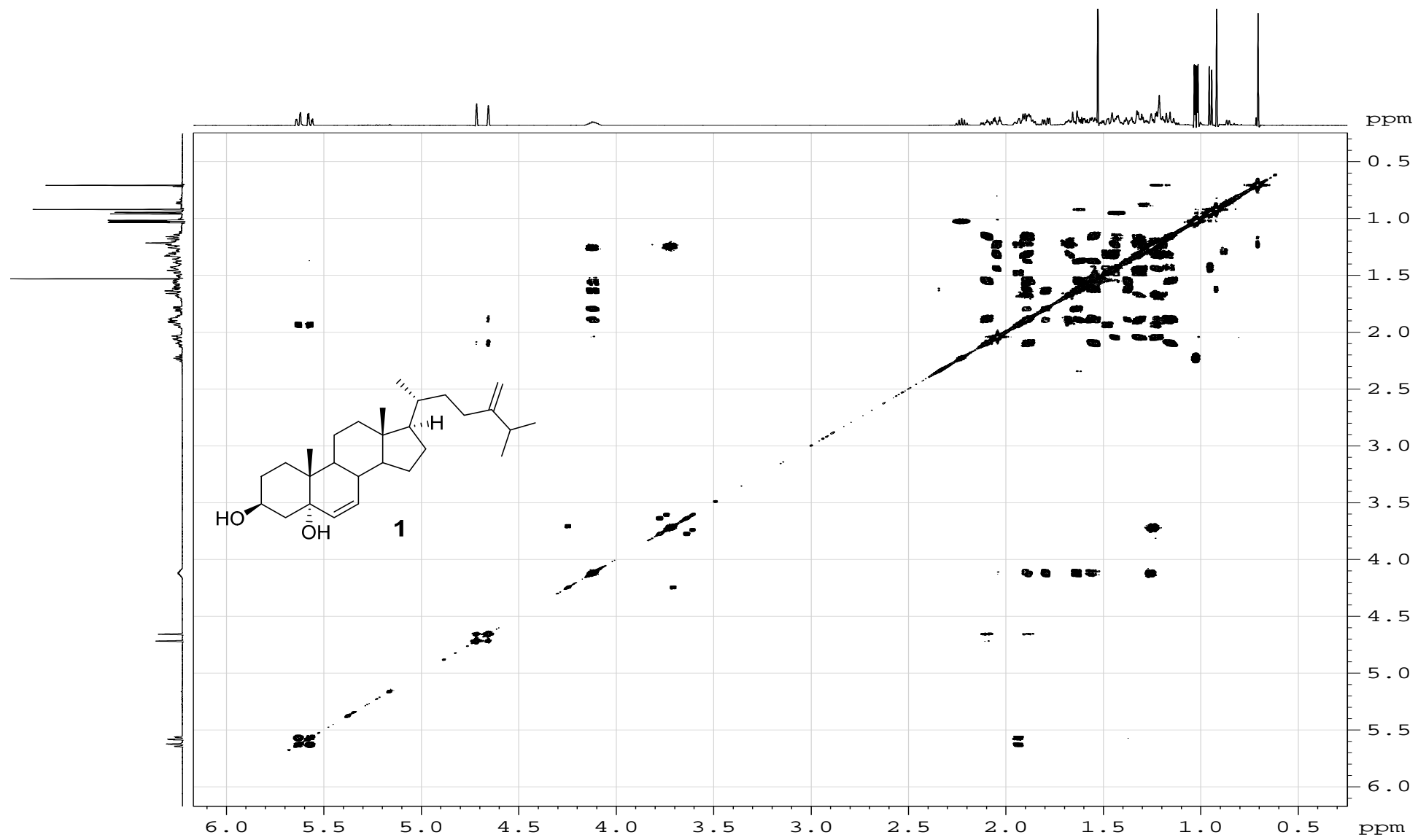
**S3** <sup>1</sup>H NMR (700.13 MHz) spectrum for the 24-methylcholesta-6,24(28)-diene-3β,5α-diol (**1**) in CDCl<sub>3</sub>



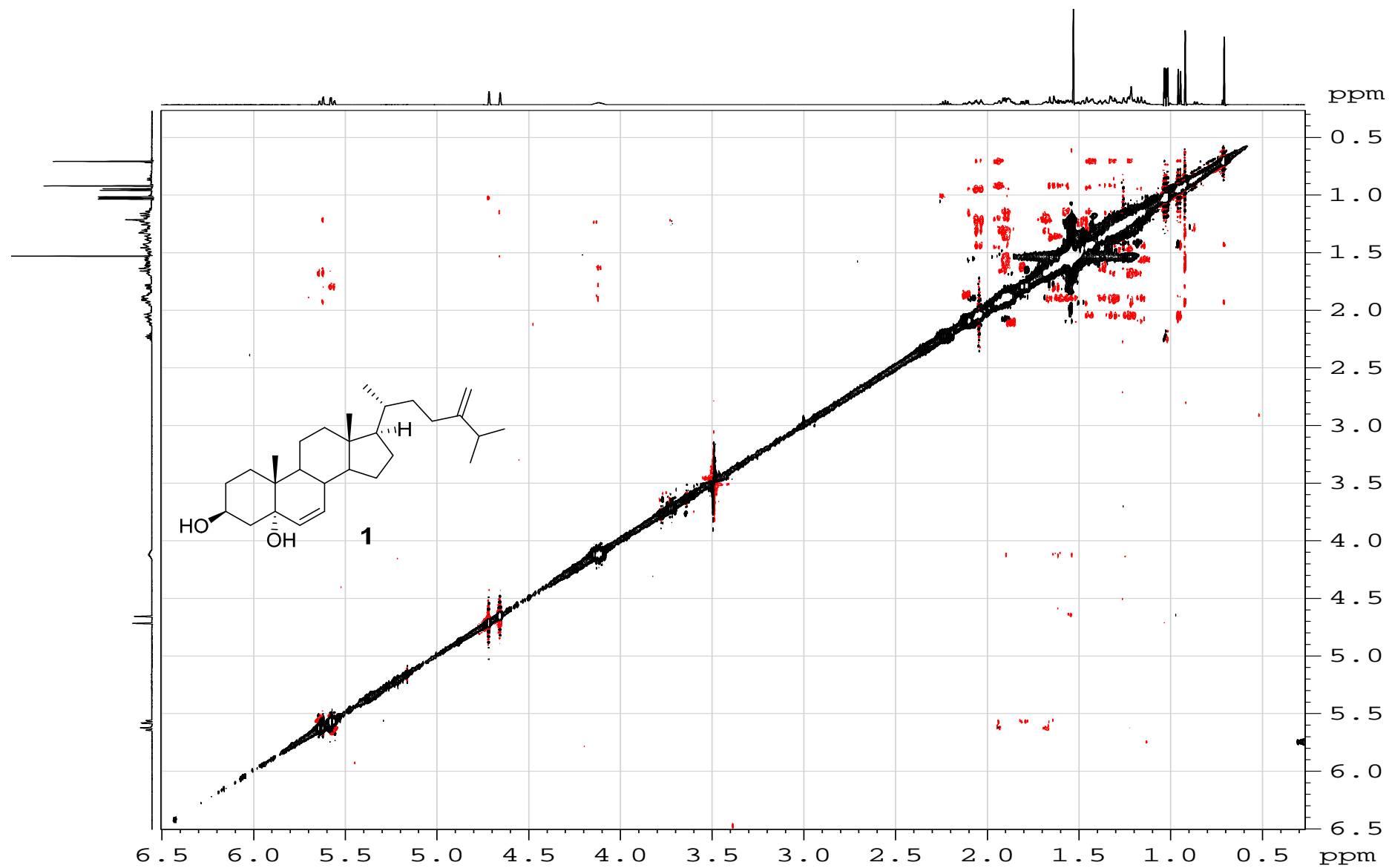
**S4**  $^{13}\text{C}$  NMR (176.04 MHz) spectrum for the 24-methylcholesta-6,24(28)-diene-3 $\beta$ ,5 $\alpha$ -diol (**1**) in  $\text{CDCl}_3$



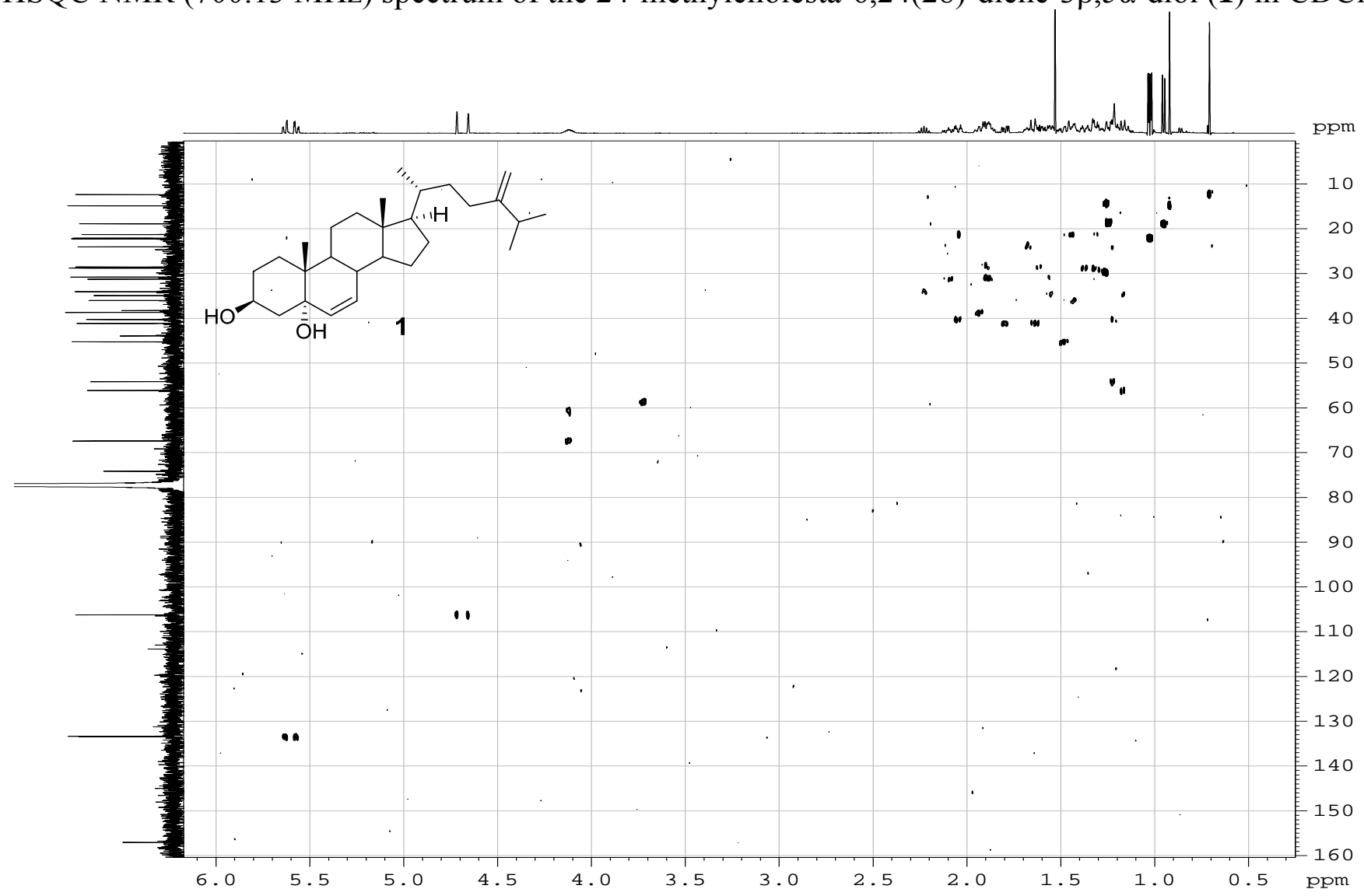
S5 COSY NMR (700.13 MHz) spectrum of the 24-methylcholesta-6,24(28)-diene-3 $\beta$ ,5 $\alpha$ -diol (**1**) in CDCl<sub>3</sub>



S6 ROESY NMR (500.13 MHz) spectrum of the 24-methylcholesta-6,24(28)-diene-3 $\beta$ ,5 $\alpha$ -diol (**1**) in CDCl<sub>3</sub>

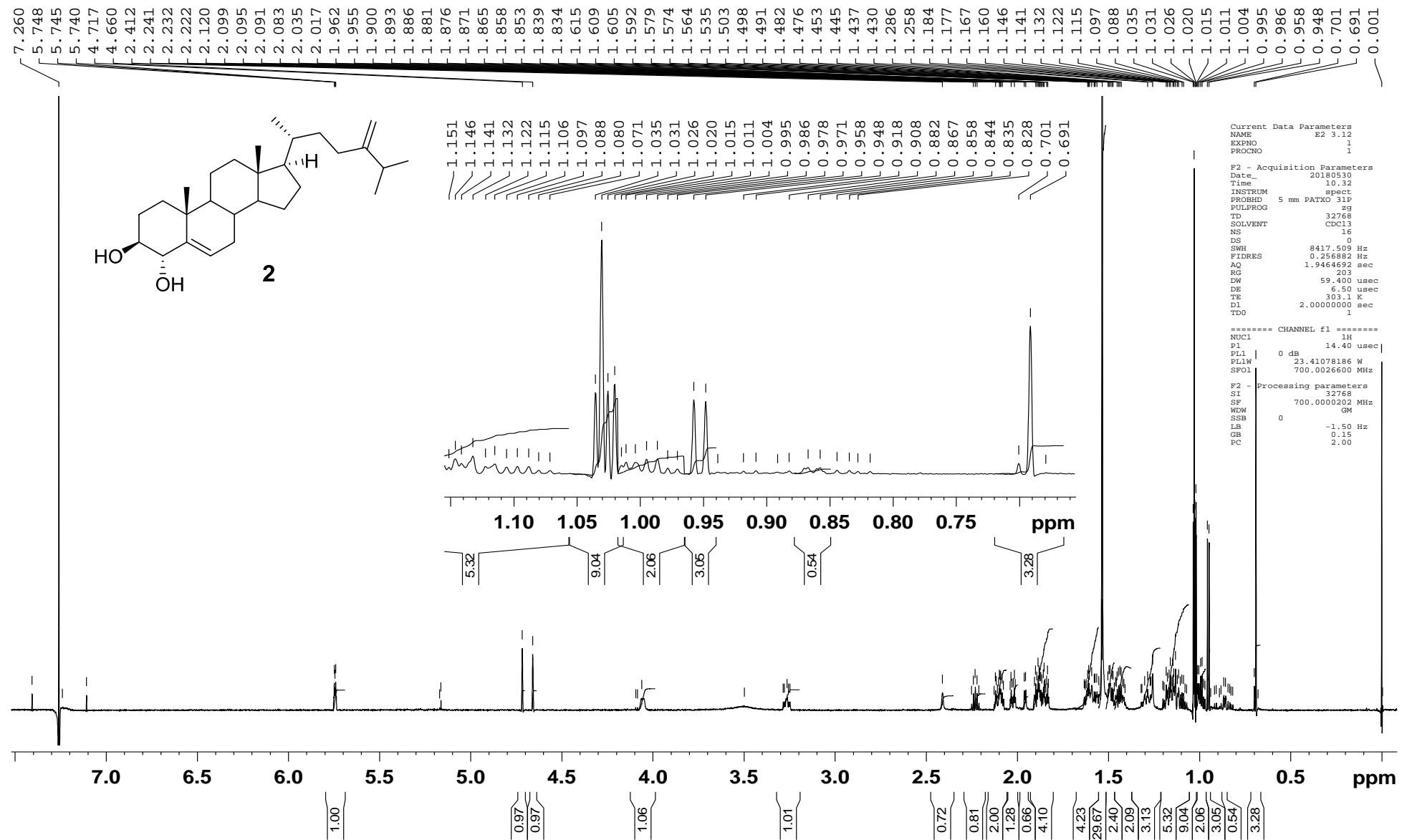


S7 HSQC NMR (700.13 MHz) spectrum of the 24-methylcholesta-6,24(28)-diene-3 $\beta$ ,5 $\alpha$ -diol (**1**) in CDCl<sub>3</sub>

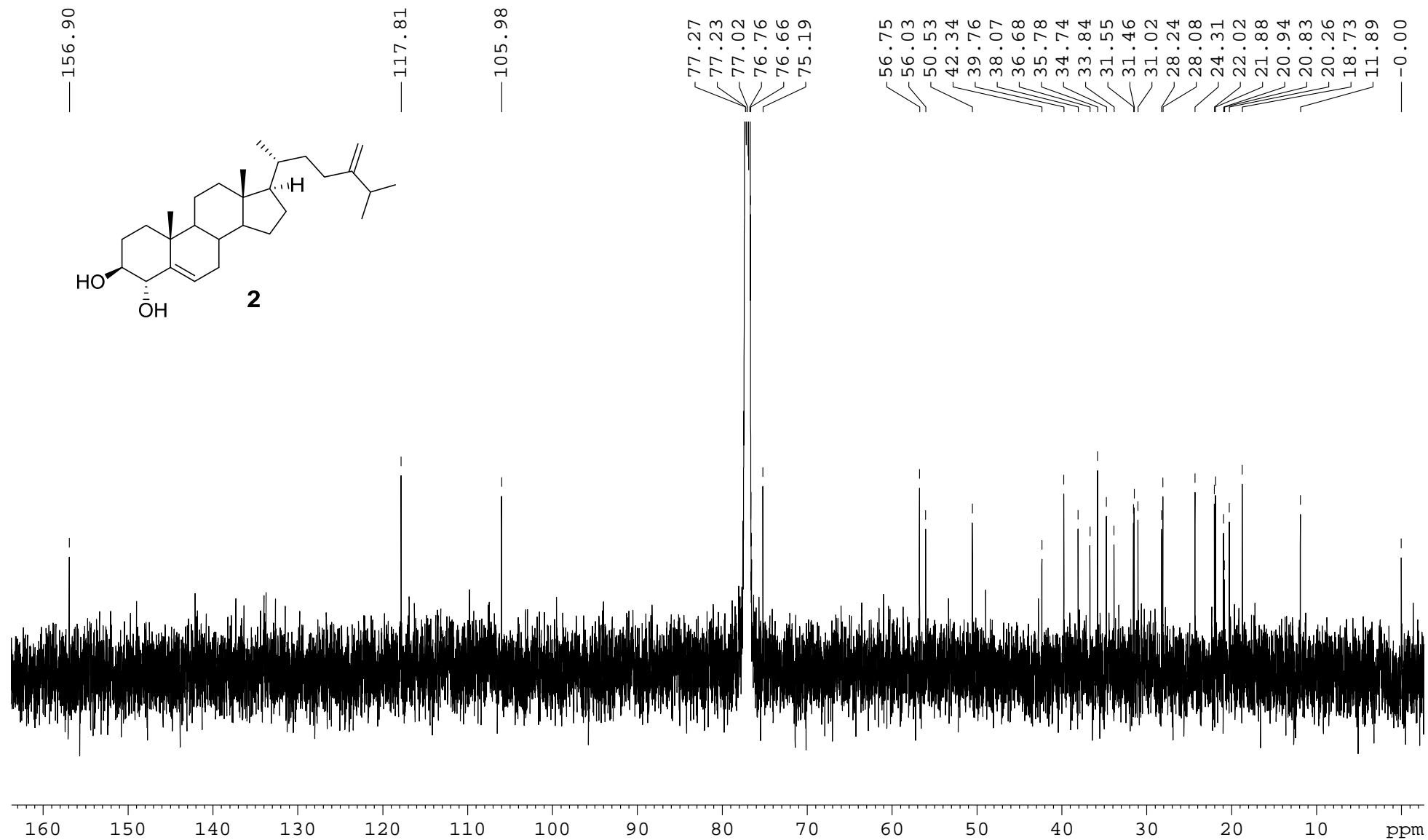




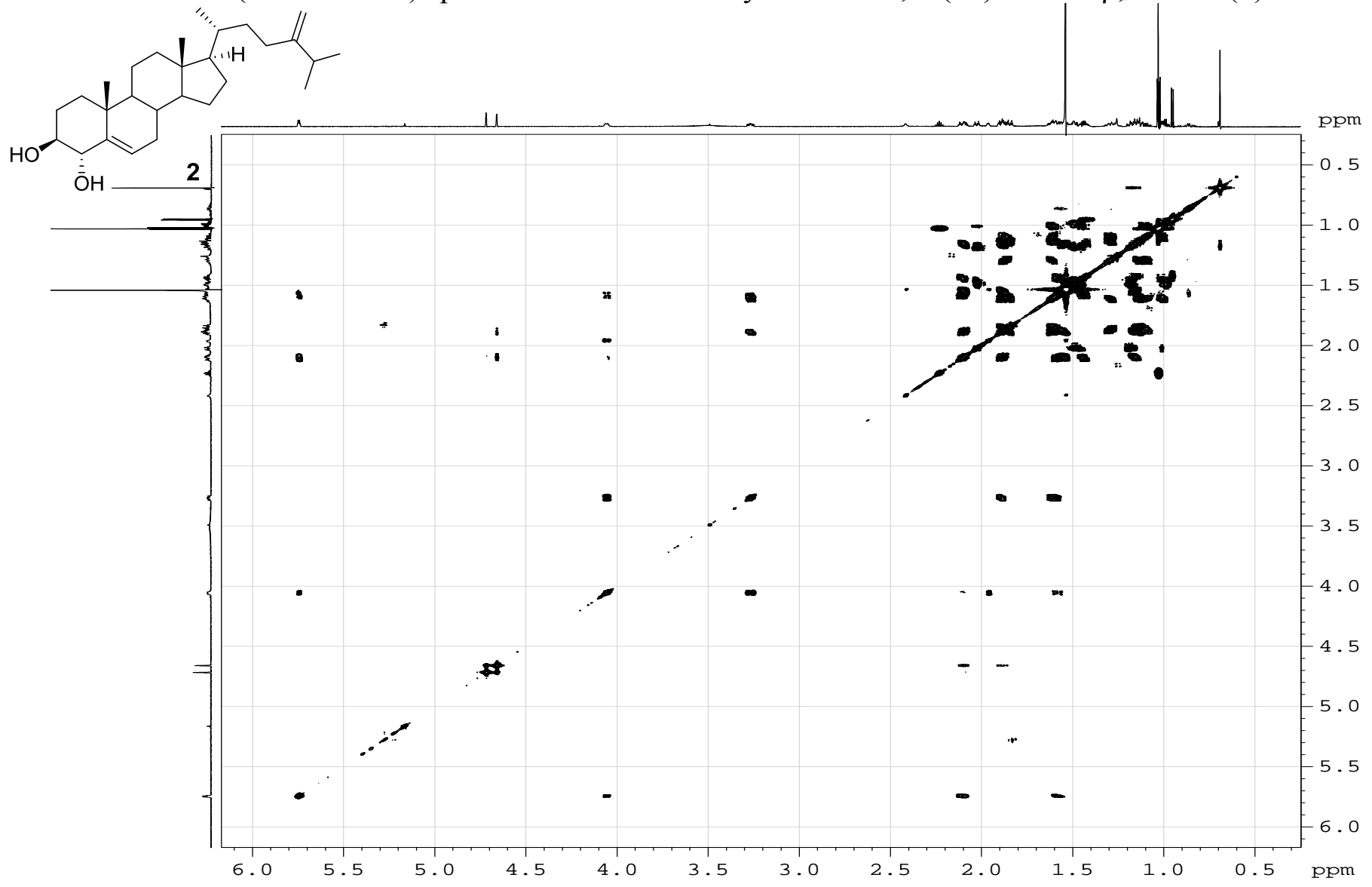
**S 8**  $^1\text{H}$  NMR (700.13 MHz) spectrum of the 24-methylcholesta-5,24(28)-diene-3 $\beta$ ,4 $\alpha$ -diol (**2**) in  $\text{CDCl}_3$



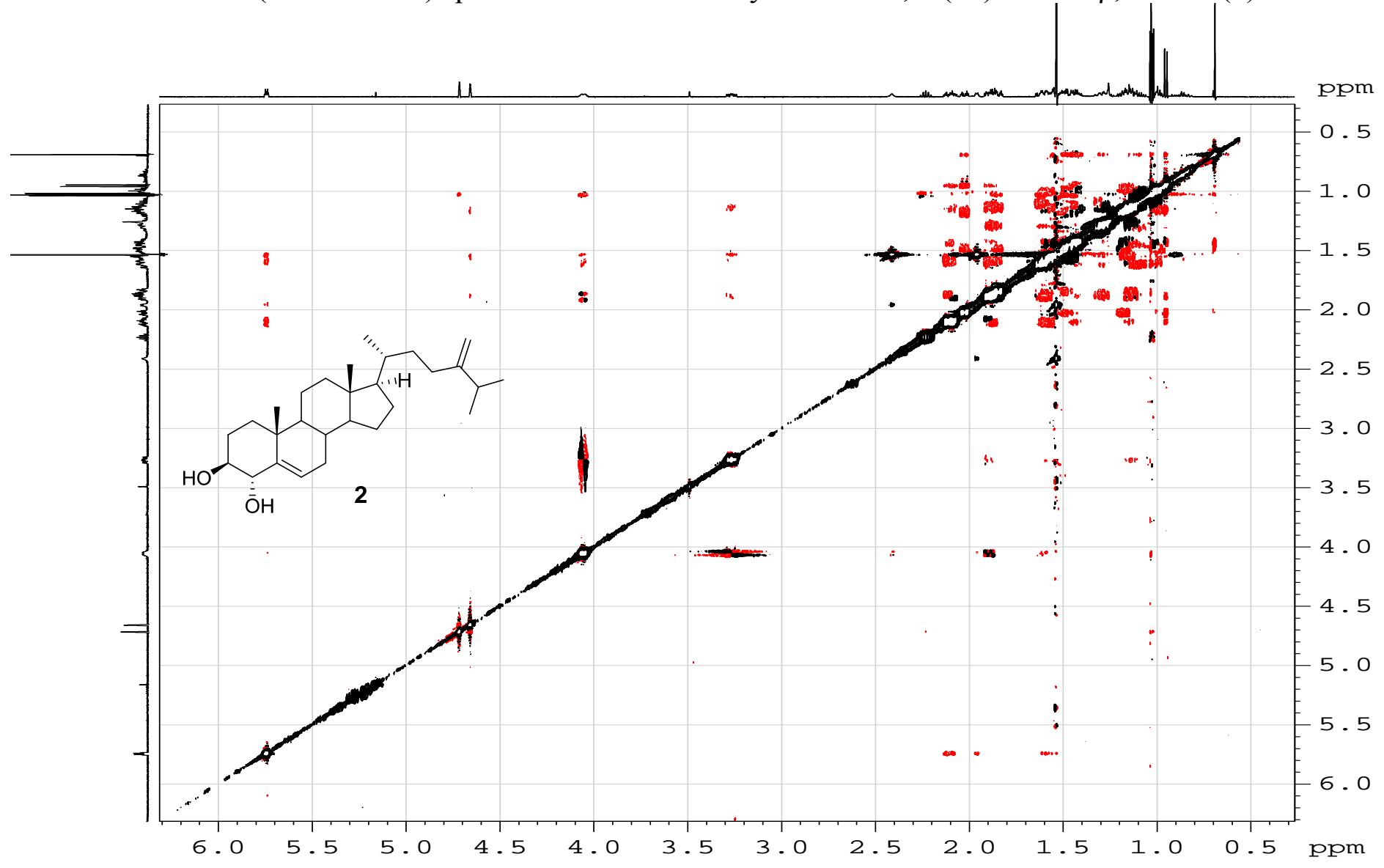
**S9**  $^{13}\text{C}$  NMR (176.04 MHz) spectrum of the 24-methylcholesta-5,24(28)-diene-3 $\beta$ ,4 $\alpha$ -diol (**2**) in  $\text{CDCl}_3$



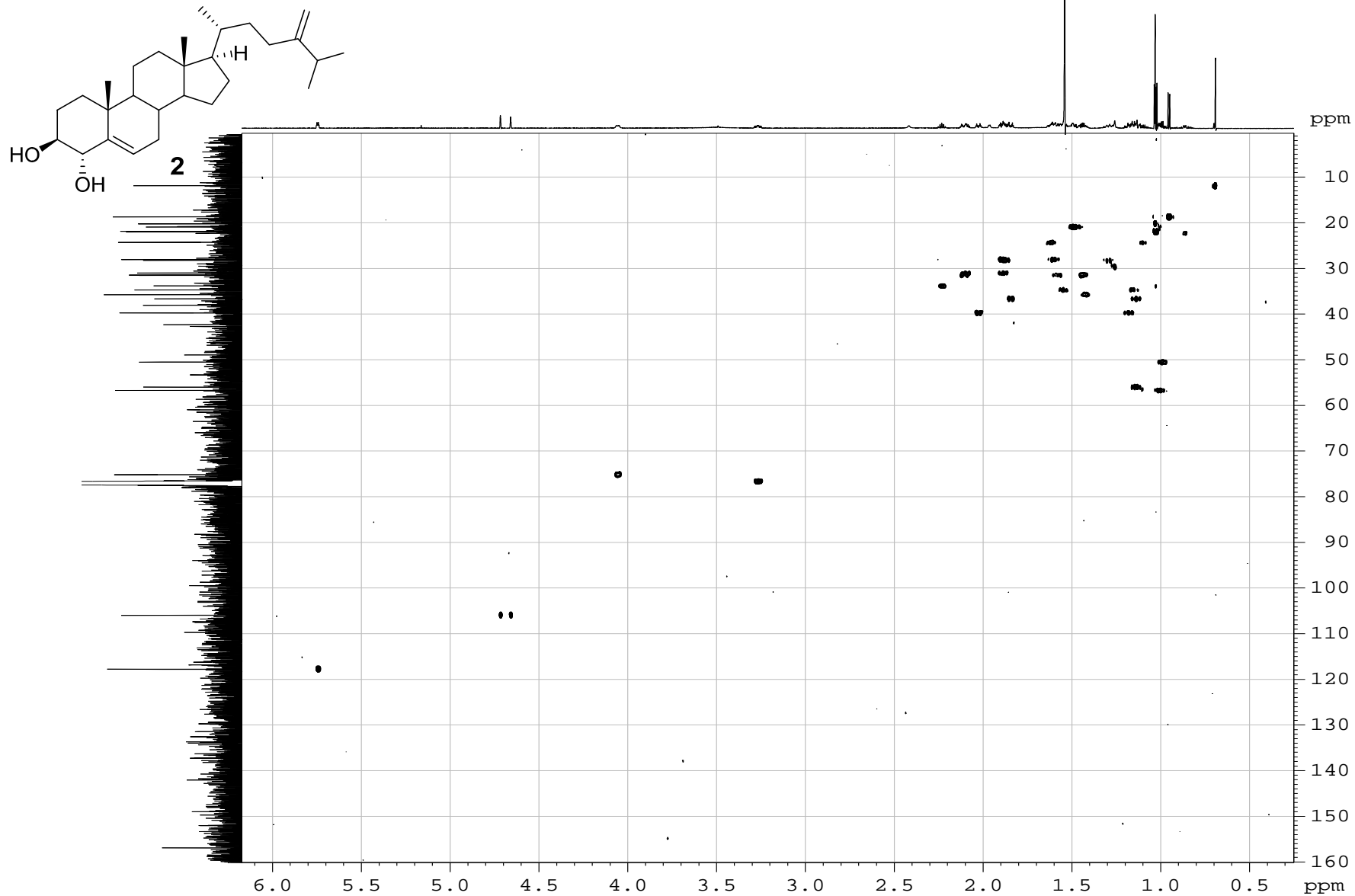
S10 COSY NMR (700.13 MHz) spectrum of the 24-methylcholesta-5,24(28)-diene-3 $\beta$ ,4 $\alpha$ -diol (**2**) in CDCl<sub>3</sub>



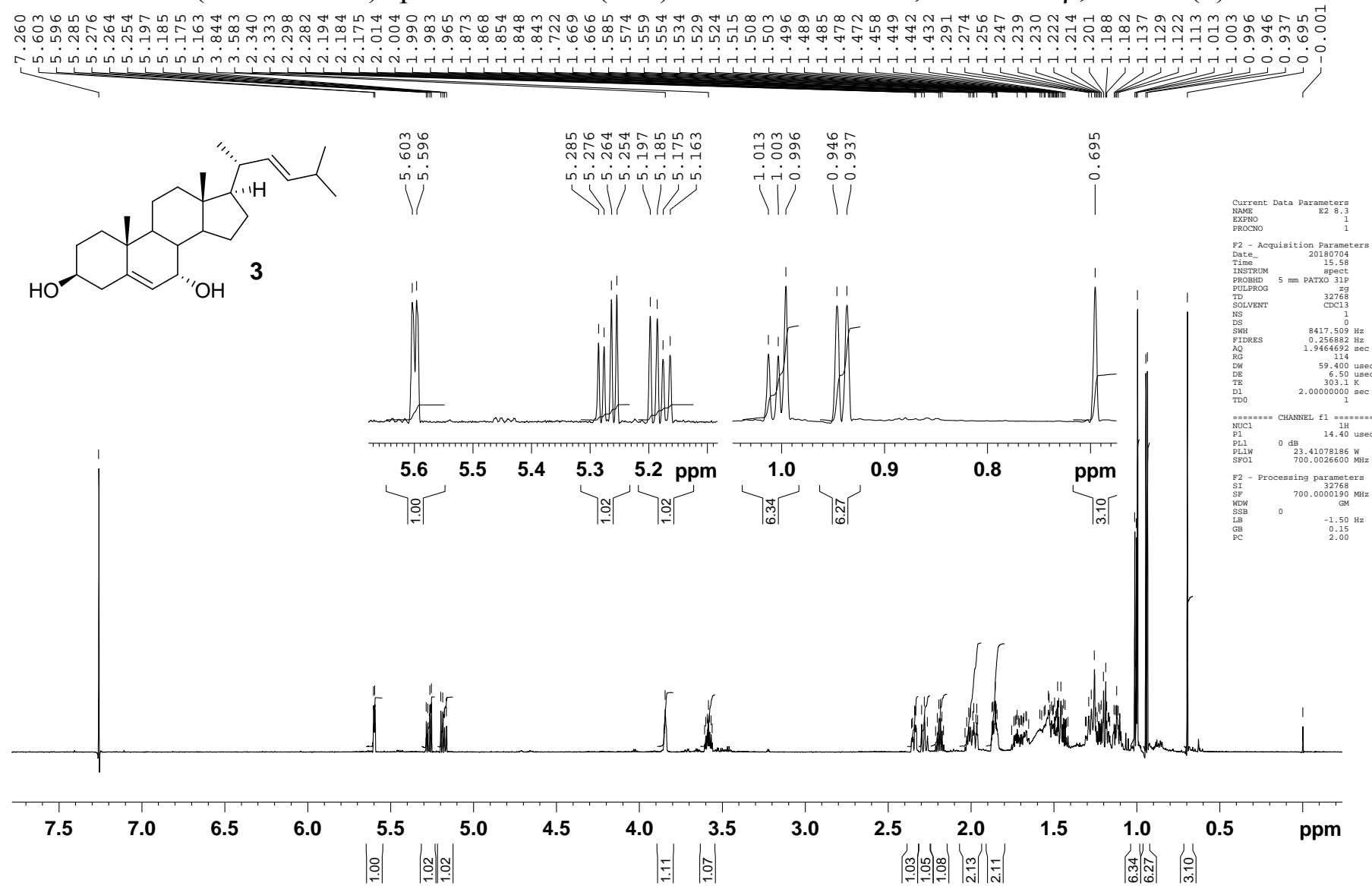
S11 ROESY NMR (500.13 MHz) spectrum of the 24-methylcholesta-5,24(28)-diene-3 $\beta$ ,4 $\alpha$ -diol (**2**) in CDCl<sub>3</sub>



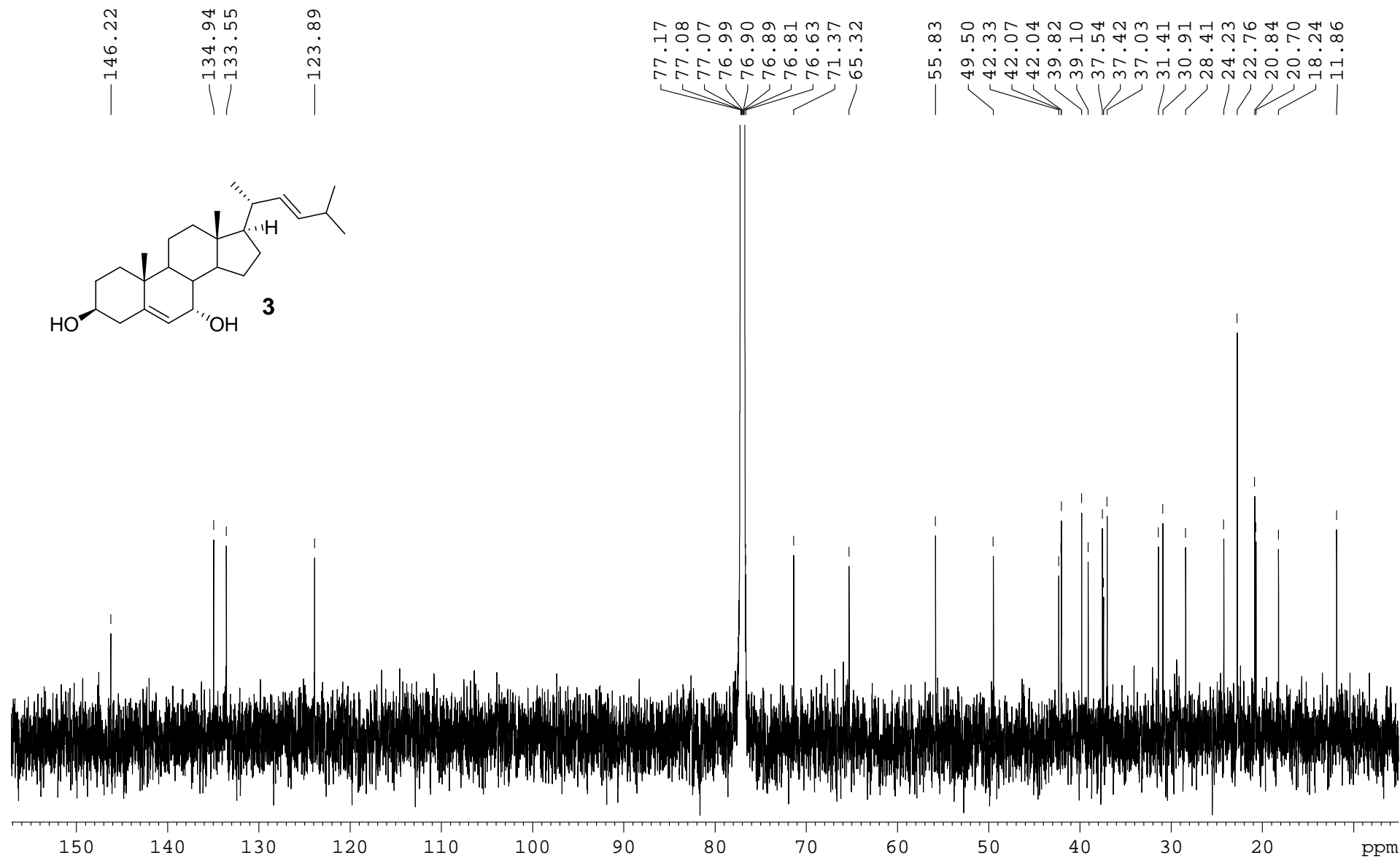
S12 HSQC NMR (700.13 MHz) spectrum for the 24-methylcholesta-5,24(28)-diene-3 $\beta$ ,4 $\alpha$ -diol (**2**) in CDCl<sub>3</sub>



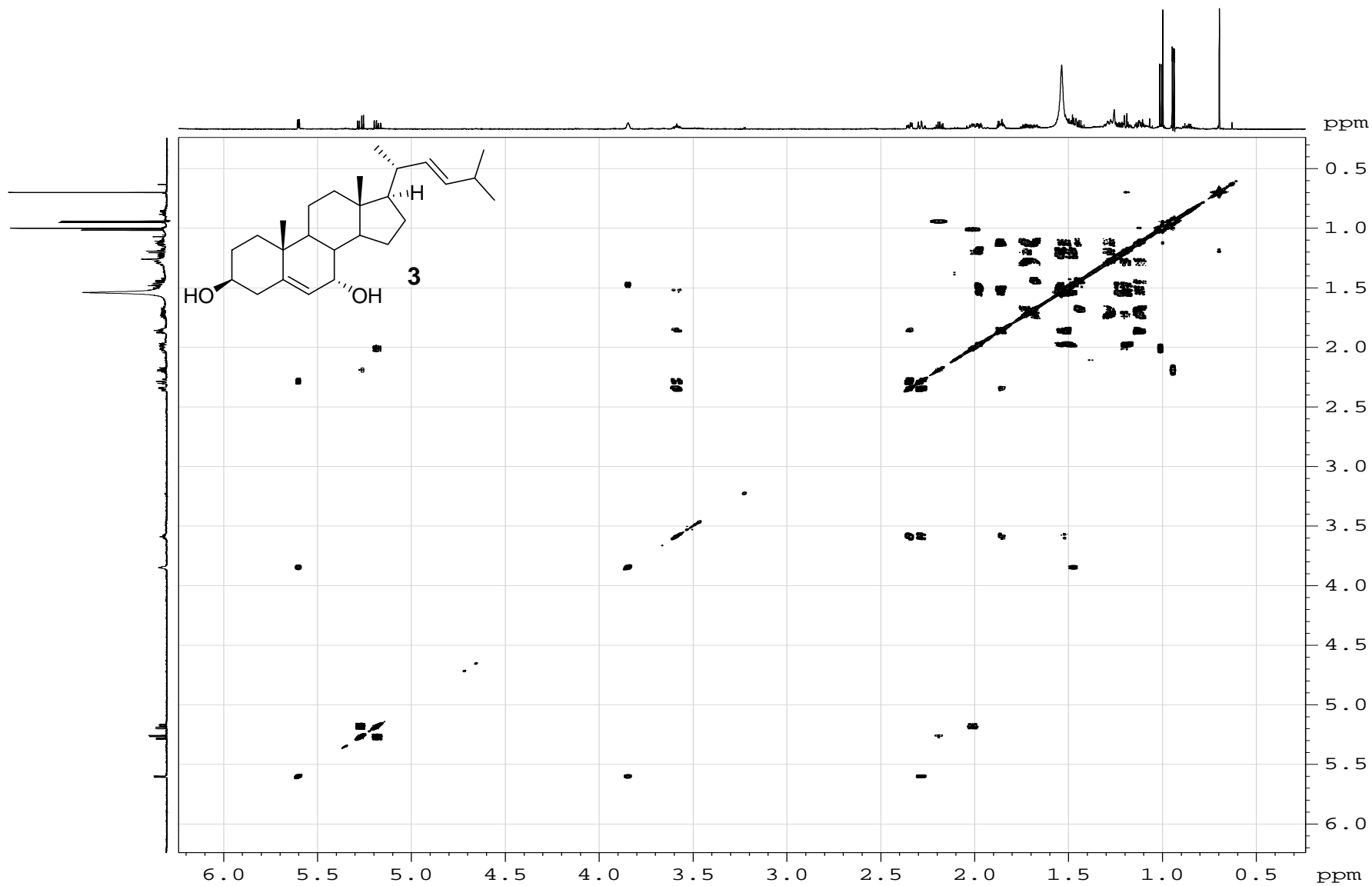
**S13** <sup>1</sup>H NMR (700.13 MHz) spectrum of the (22*E*)-24-*nor*-cholesta-5,22-diene-3 $\beta$ ,7 $\alpha$ -diol (**3**) in CDCl<sub>3</sub>



S14  $^{13}\text{C}$  NMR (176.04 MHz) spectrum of the (22E)-24-nor-cholesta-5,22-diene-3 $\beta$ ,7 $\alpha$ -diol (**3**) in  $\text{CDCl}_3$

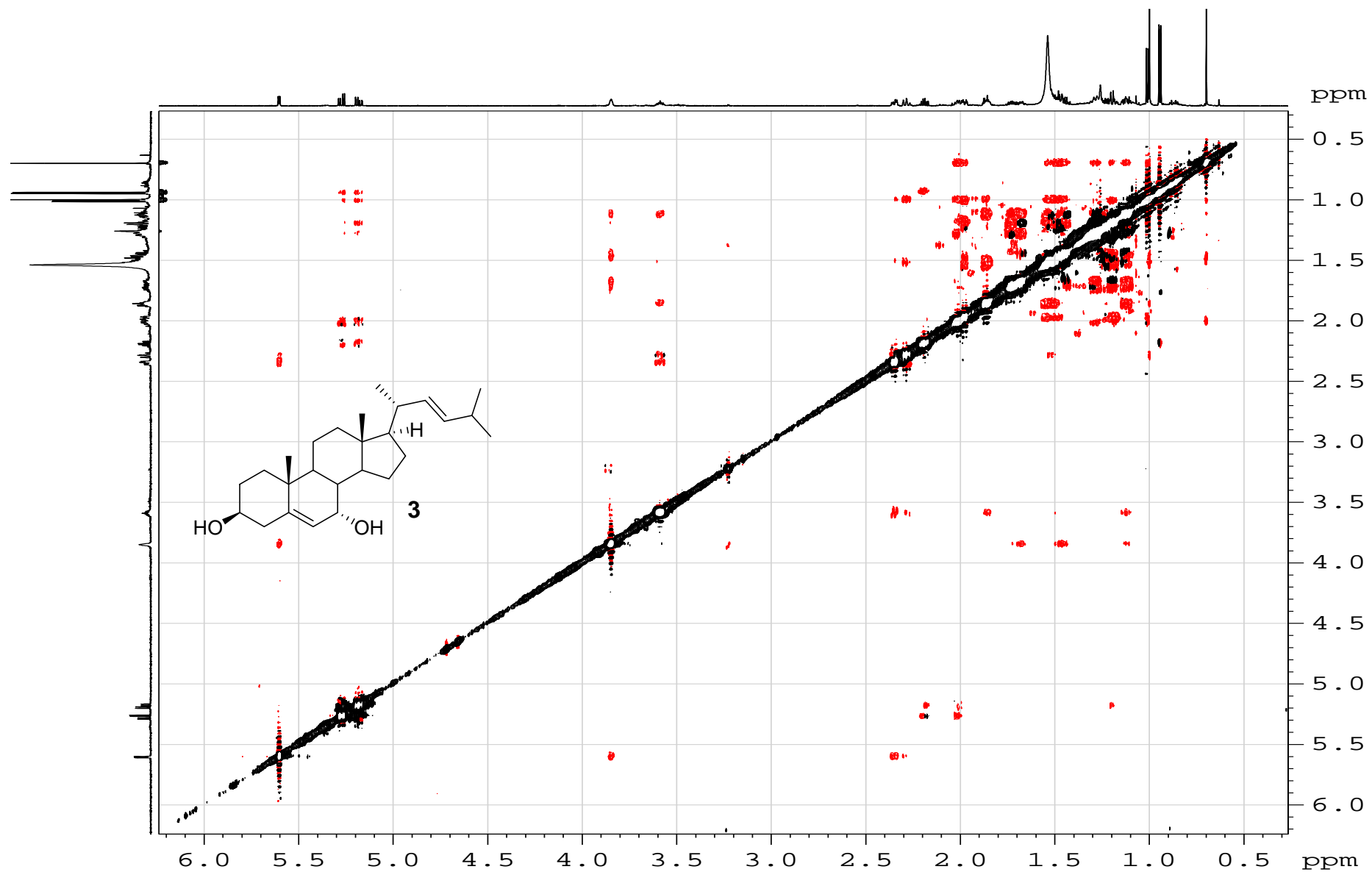


S15 COSY NMR (700.13 MHz) spectrum of the (22*E*)-24-*nor*-cholesta-5,22-diene-3 $\beta$ ,7 $\alpha$ -diol (**3**) in CDCl<sub>3</sub>

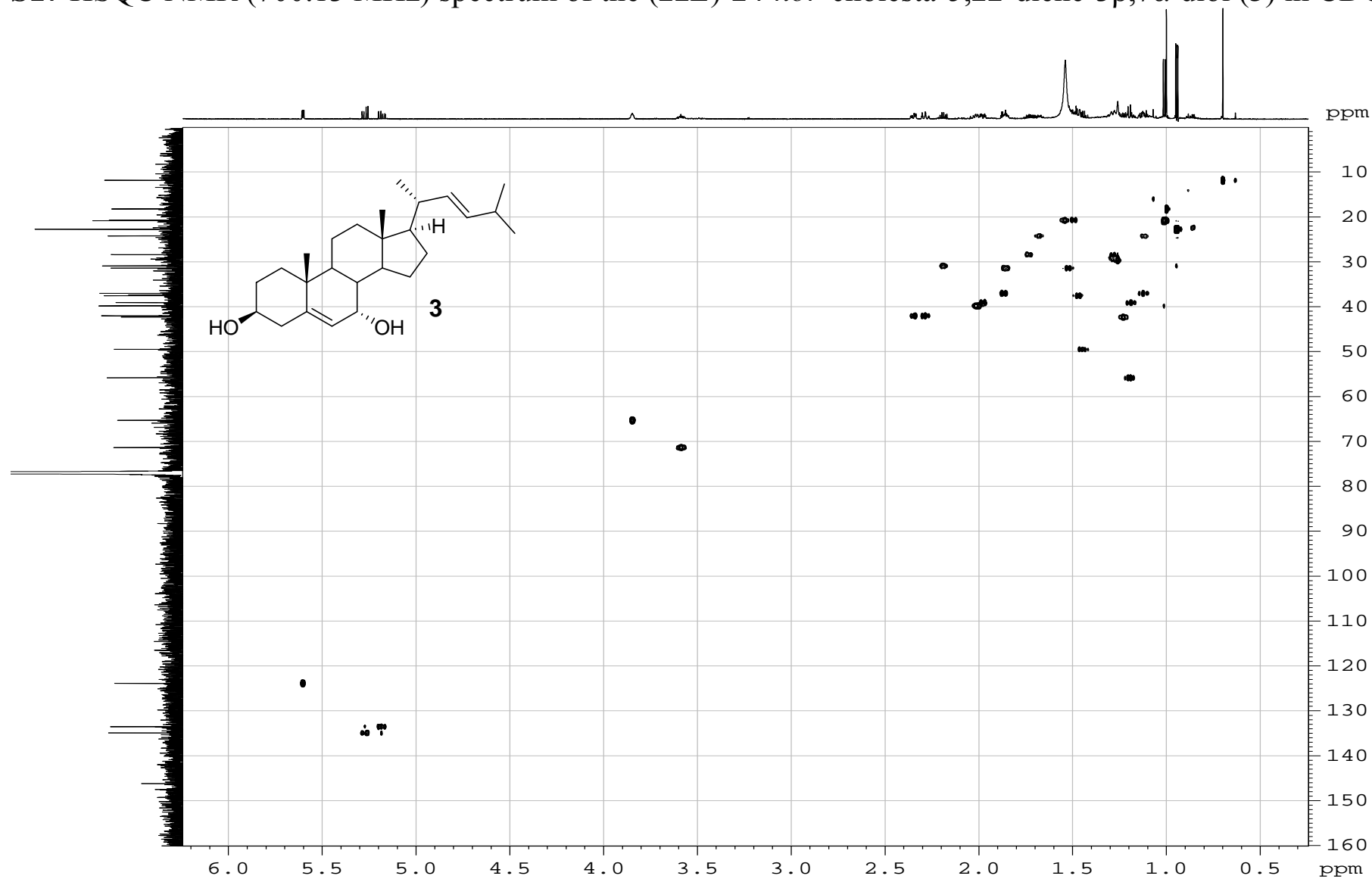




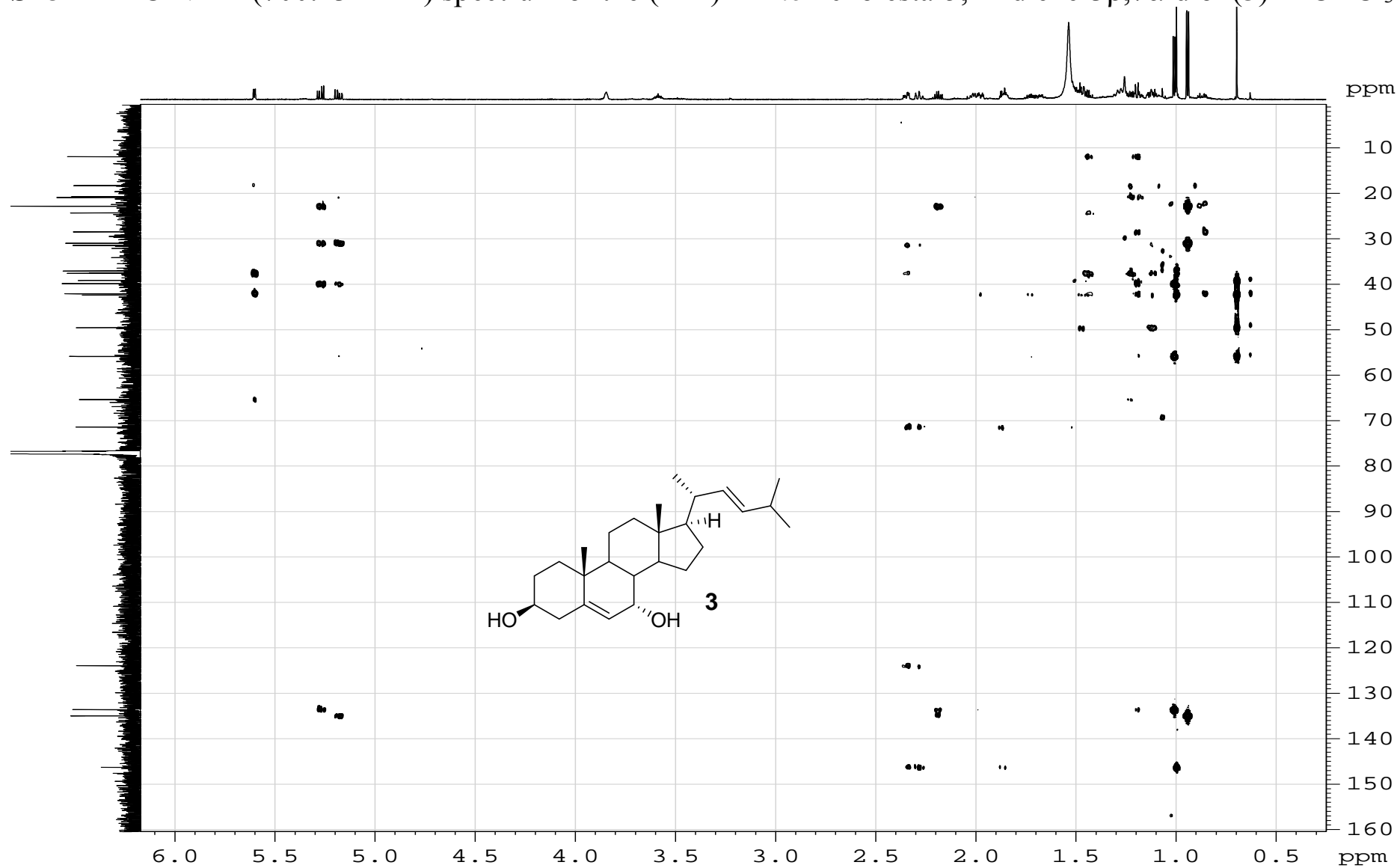
S16 ROESY NMR (700.13 MHz) spectrum of the (22E)-24-nor-cholesta-5,22-diene-3 $\beta$ ,7 $\alpha$ -diol (**3**) in CDCl<sub>3</sub>



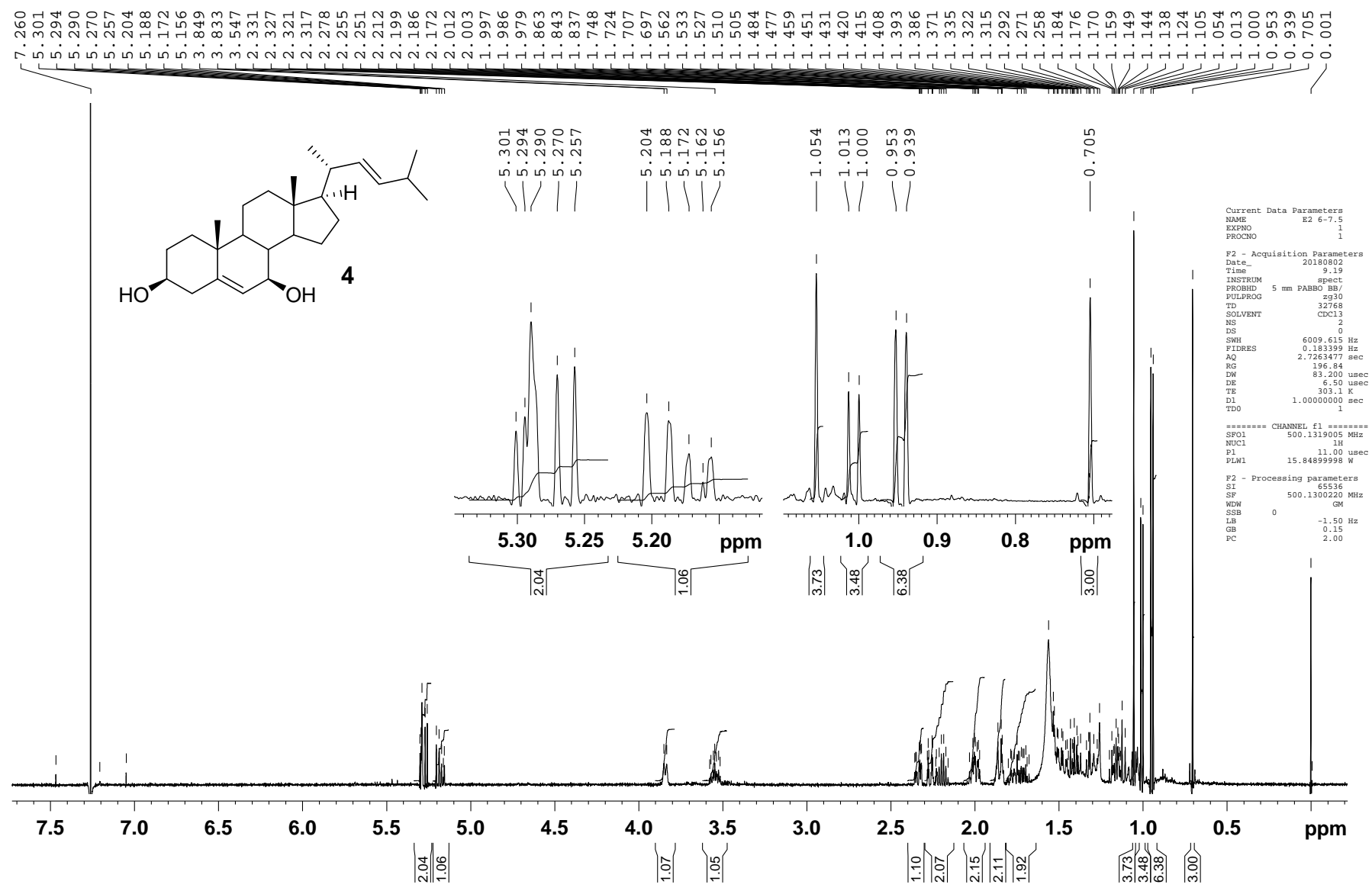
S17 HSQC NMR (700.13 MHz) spectrum of the (22E)-24-nor-cholesta-5,22-diene-3 $\beta$ ,7 $\alpha$ -diol (**3**) in CDCl<sub>3</sub>



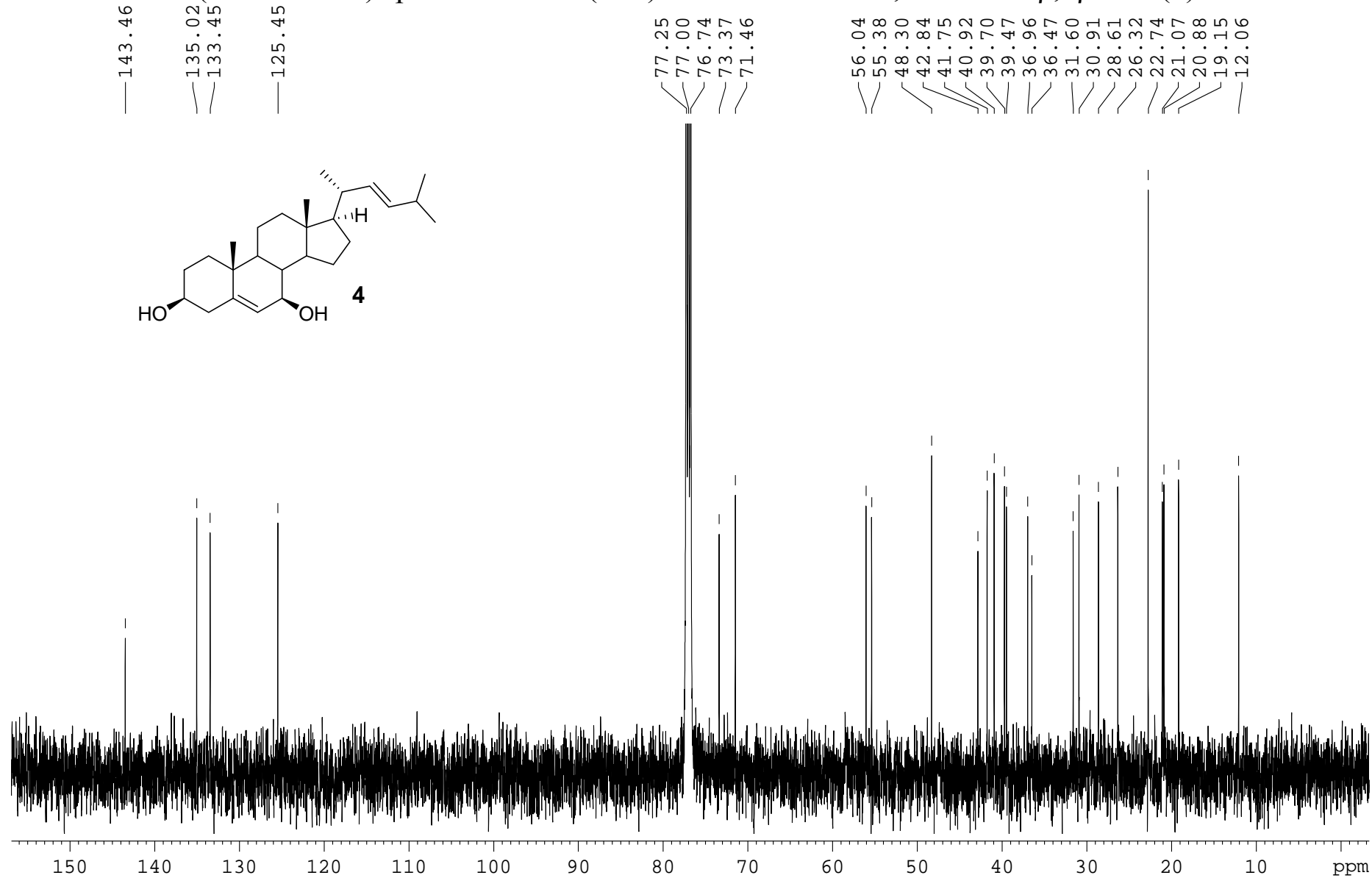
S18 HMBC NMR (700.13 MHz) spectrum of the (22E)-24-nor-cholesta-5,22-diene-3 $\beta$ ,7 $\alpha$ -diol (**3**) in CDCl<sub>3</sub>



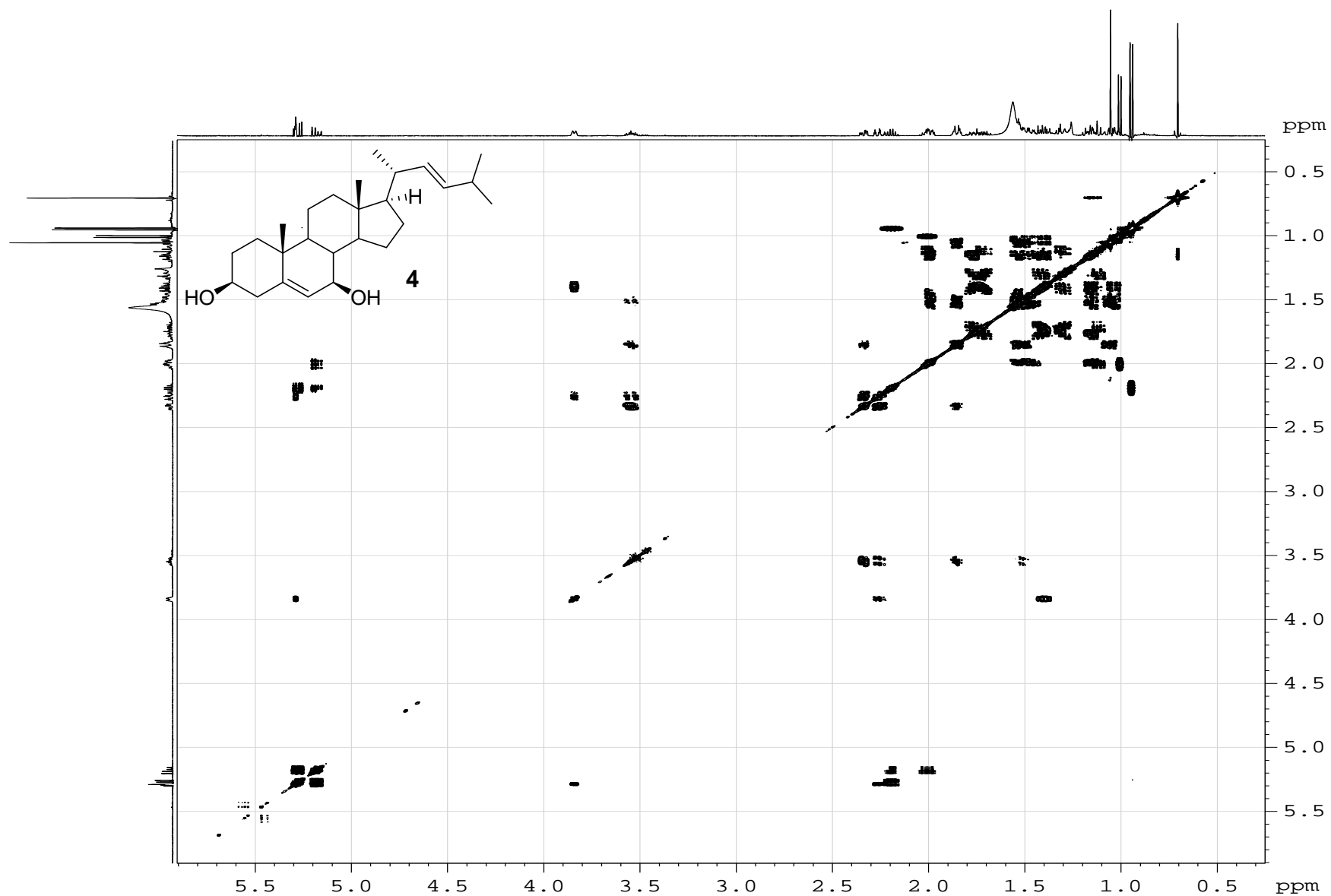
**S19** <sup>1</sup>H NMR (500.13 MHz) spectrum of the (22*E*)-24-*nor*-cholesta-5,22-diene-3 $\beta$ ,7 $\beta$ -diol (**4**) in CDCl<sub>3</sub>



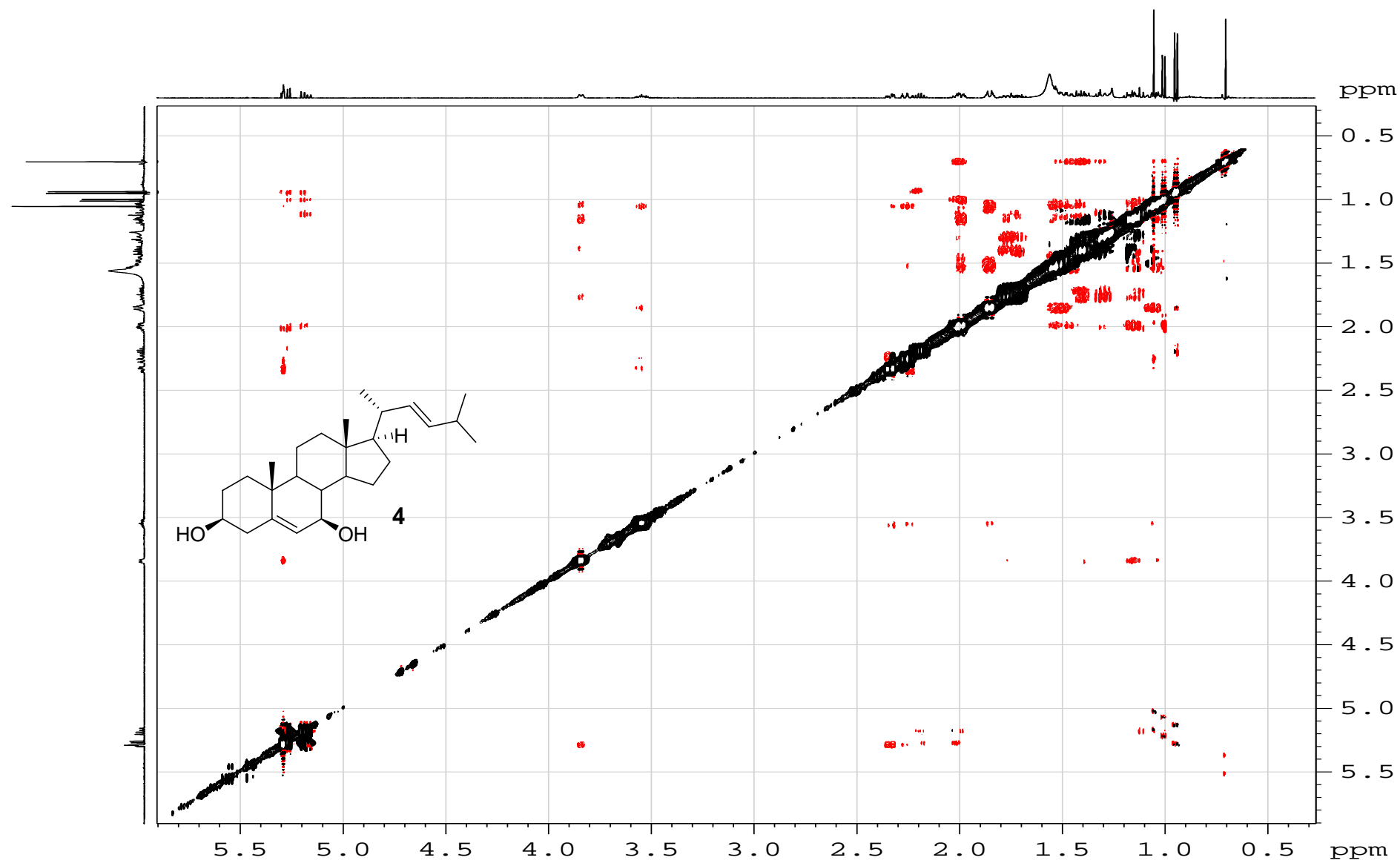
S20  $^{13}\text{C}$  NMR (125.76 MHz) spectrum of the (22E)-24-nor-cholesta-5,22-diene-3 $\beta$ ,7 $\beta$ -diol (**4**) in  $\text{CDCl}_3$



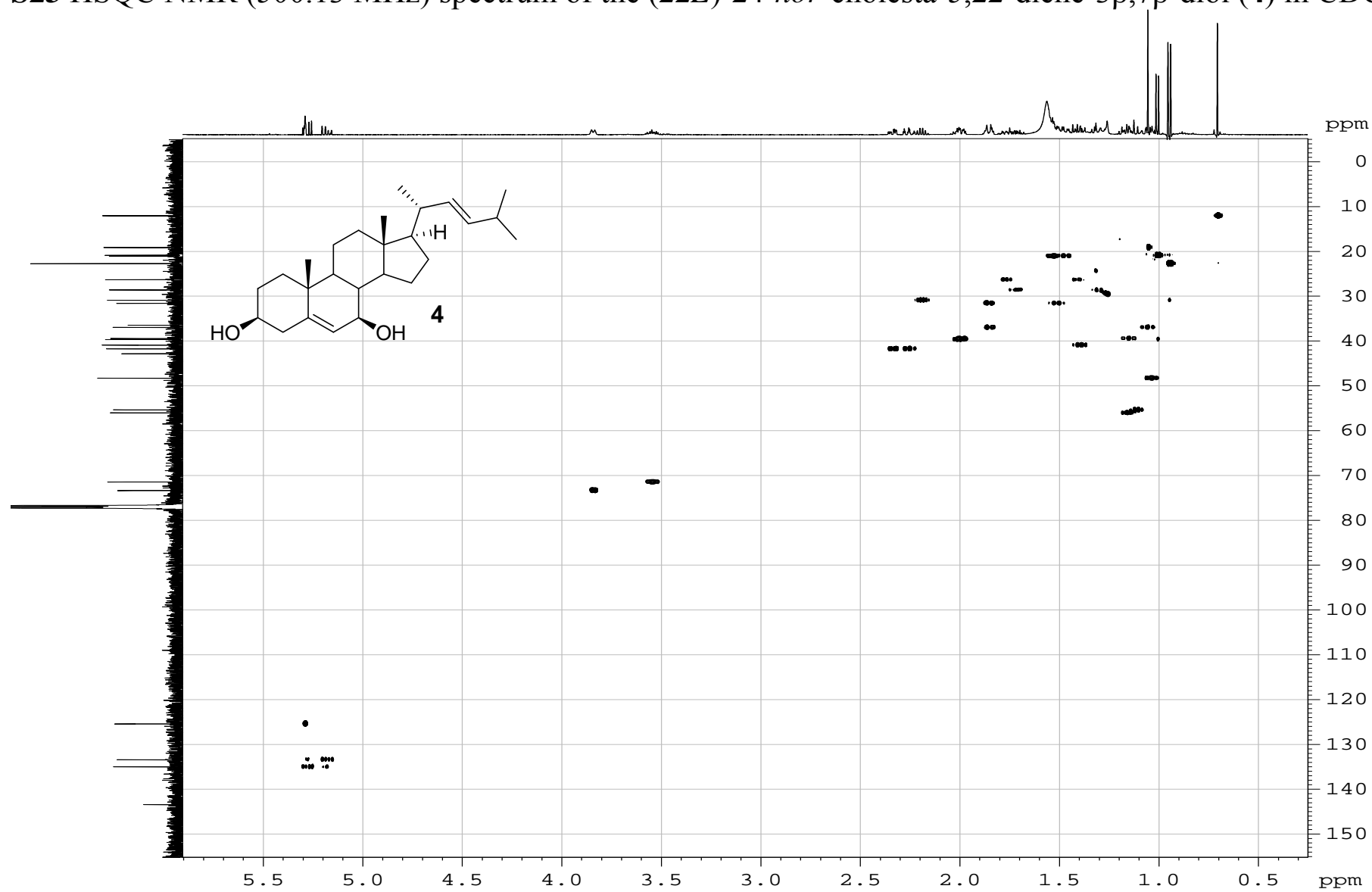
S21 COSY NMR (500.13 MHz) spectrum of the (22E)-24-nor-cholesta-5,22-diene-3 $\beta$ ,7 $\beta$ -diol (**4**) in CDCl<sub>3</sub>



S22 ROESY NMR (500.13 MHz) spectrum of the (22E)-24-nor-cholesta-5,22-diene-3 $\beta$ ,7 $\beta$ -diol (**4**) in CDCl<sub>3</sub>

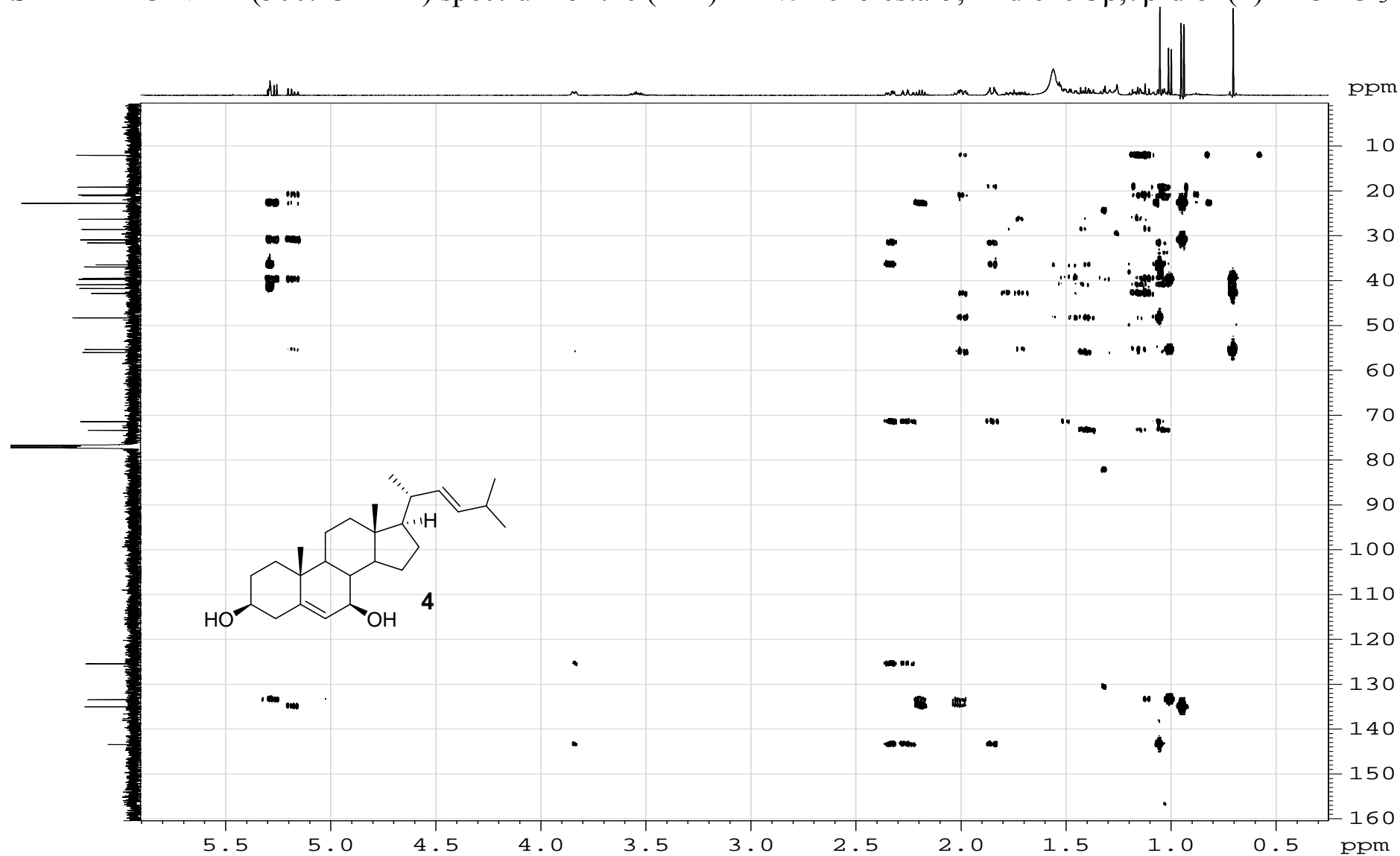


S23 HSQC NMR (500.13 MHz) spectrum of the (22*E*)-24-*nor*-cholesta-5,22-diene-3 $\beta$ ,7 $\beta$ -diol (**4**) in CDCl<sub>3</sub>

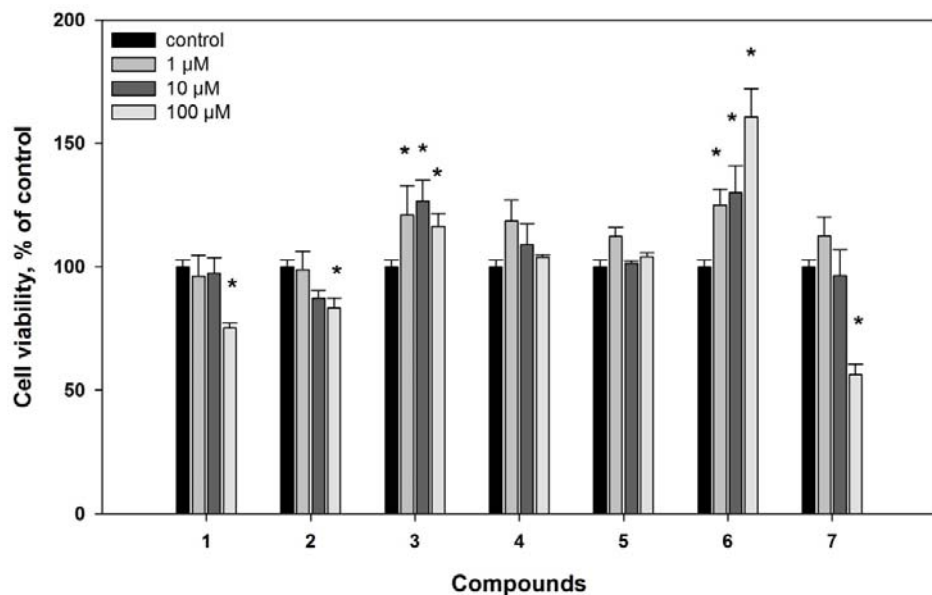




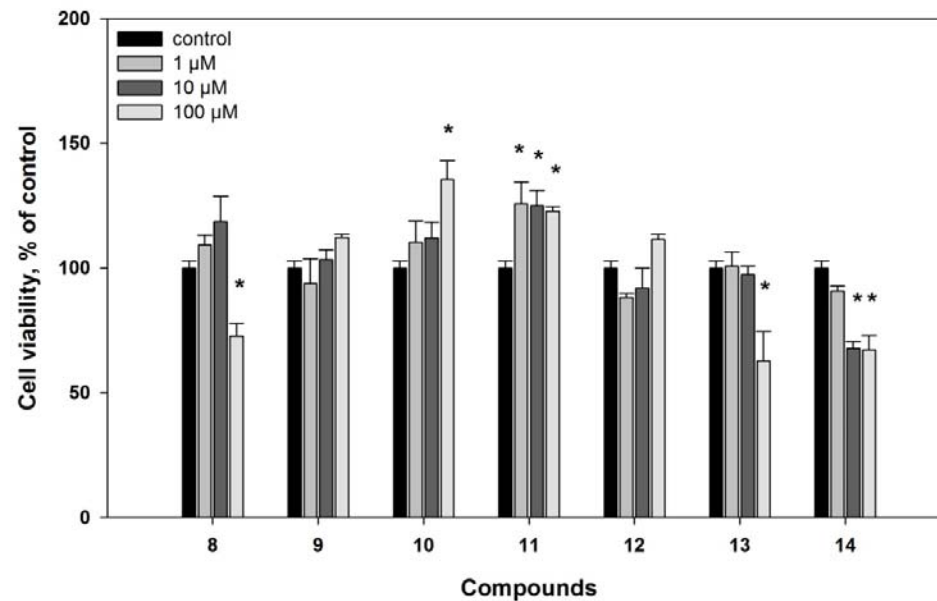
S24 HMBC NMR (500.13 MHz) spectrum of the (22E)-24-nor-cholesta-5,22-diene-3 $\beta$ ,7 $\beta$ -diol (**4**) in CDCl<sub>3</sub>



## S25 Viability of Neuro2a cells



(a)

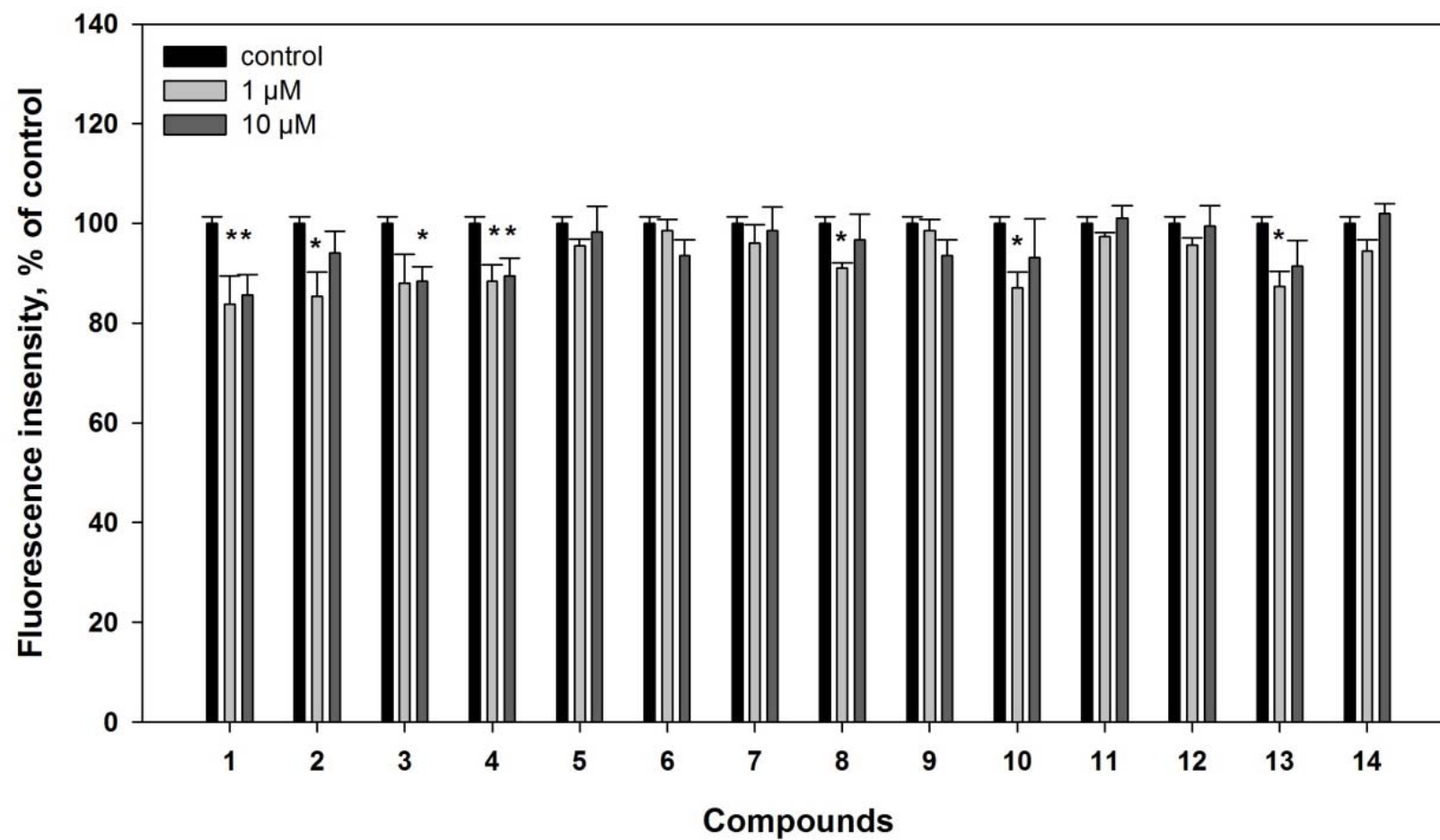


(b)

Influence of compounds 1-7 (a) and 8-14 (b) on viability of Neuro2a cells by MTT assay.

\* Statistically significant differences ( $p \leq 0.05$ ) between results for control cells and cells incubated with compounds.

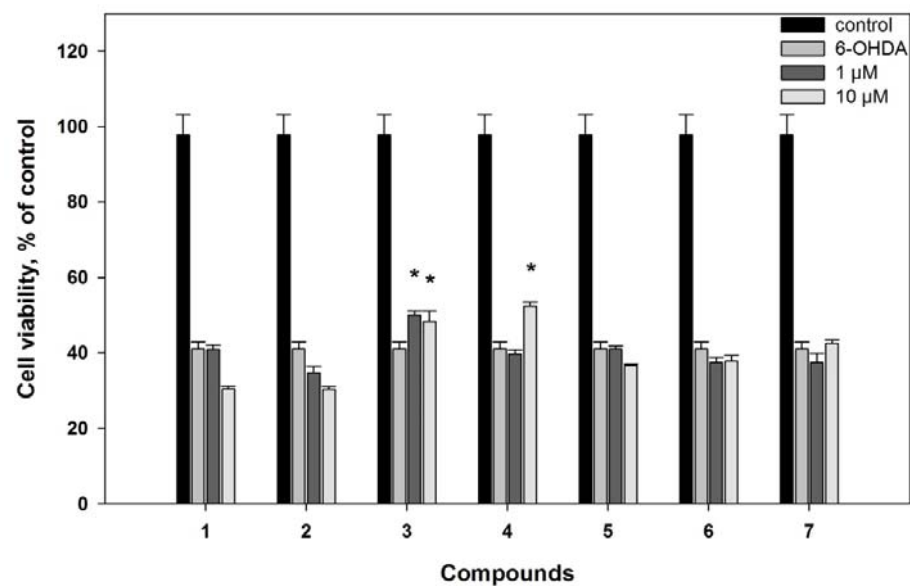
## S26 ROS formation in Neuro2a cells



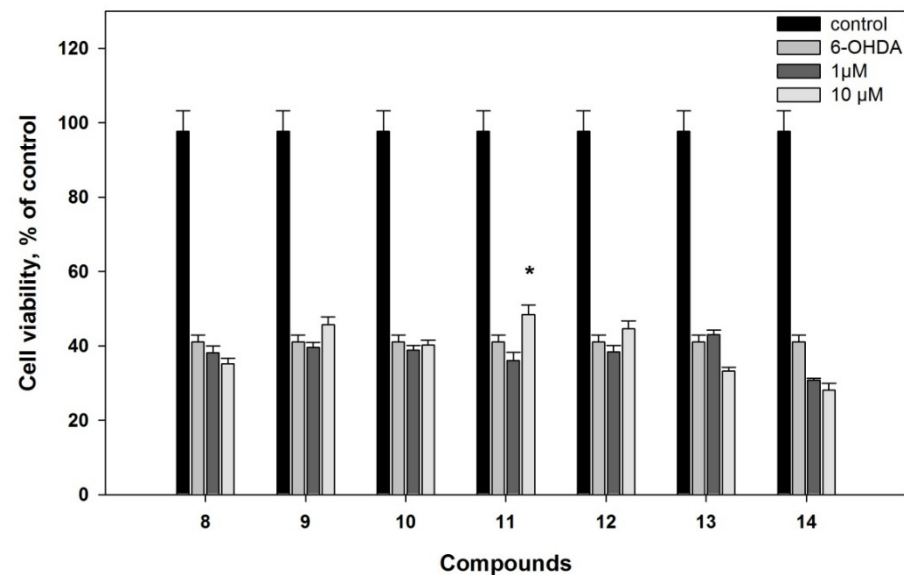
Influence of compounds 1-14 on ROS formation in Neuro2a cells.

\*Statistically significant differences ( $p \leq 0.05$ ) between results for control cells and cells incubated with compounds.

## S27 Viability of Neuro2a cells treated with 6-OHDA



(a)



(b)

Influence of compounds 1-7 (a) and 8-14 (b) on viability of Neuro2a cells treated with 6-OHDA (50 μM).

\* Statistically significant differences ( $p \leq 0.05$ ) between results for 6-OHDA-treated cells and cells incubated with compounds.