

Supplemental Data

Cytochrome P450scc-dependent metabolism of 7-dehydrocholesterol in placenta and epidermal keratinocytes.

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Supplemental Figure S-1. Mass spectrum for 20,22-(OH)₂-7DHC.

Electrospray ionization (ESI) mass spectrum for 20,22-(OH)₂-7DHC.

Supplemental Figure S-2. ¹H-NMR spectrum for 20,22-(OH)₂-7DHC.

One- and two-dimensional NMR spectra for the metabolite was acquired using a 3 mm probe. Structures of the metabolites are labeled with each spectrum.

Supplemental Figure S-3. ¹H-¹H COSY NMR spectrum for 20,22-(OH)₂-7DHC.

Supplemental Figure S-4. ¹H-¹³C HSQC NMR spectrum for 20,22-(OH)₂-7DHC.

Supplemental Figure S-5. ¹H-¹H TOCSY NMR spectrum for 20,22-(OH)₂-7DHC.

Supplemental Figure S-6. ¹H-¹³C HMBC NMR spectrum for 20,22-(OH)₂-7DHC.

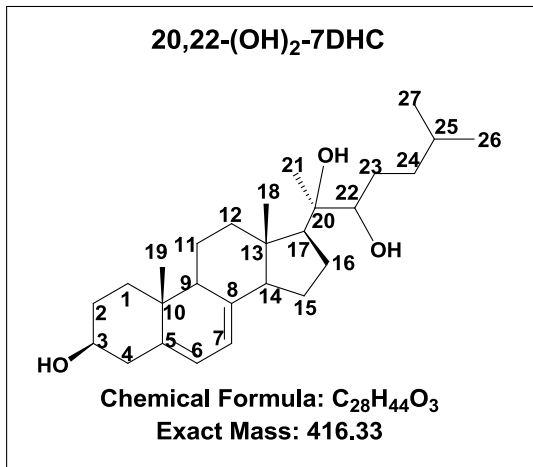
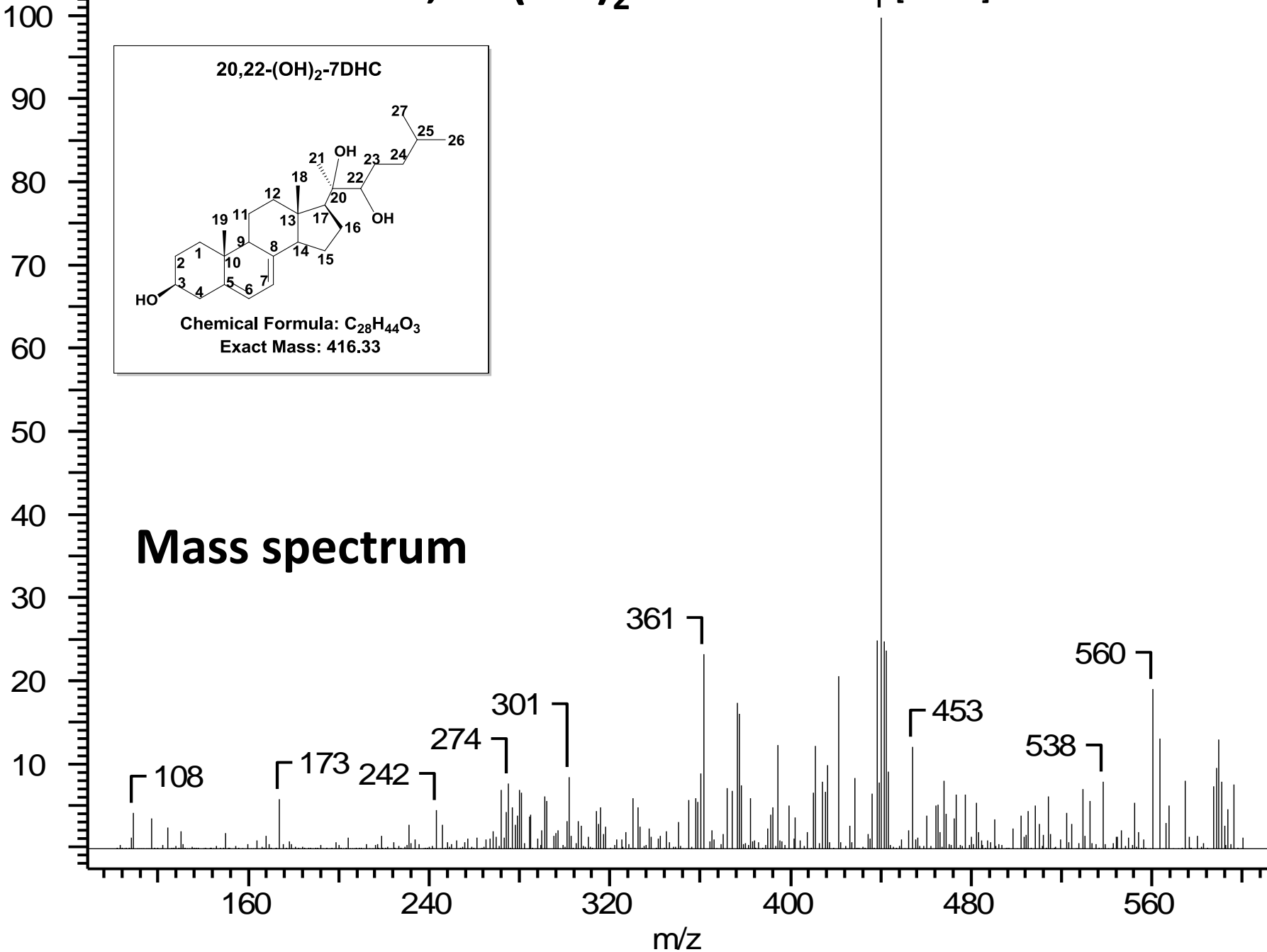
Supplemental Figure S-7. The position of 7DHC in the active site of human P450scc in relation to the heme group.

Supplemental Figure S-8. Docking scores of 7DHC, cholesterol and their hydroxyderivatives

Supplemental Figure S-9. Transformation of exogenously added 7DHC to 22(OH)7DHC, 20,22(OH)₂7DHC and 7DHP in HaCaT epidermal keratinocytes. HaCaT cells were incubated for 16 h with 50 μM 7DHC, extracted with methylene chloride and analyzed by RP- HPLC and LCMS as described in Materials and Methods and in (Slominski et al., 2012a). The HPLC chromatogram with UV detection does not show the product which were below the sensitivity level. Inserts are LCMS chromatograms for detection of the products analysed with ESI for 7DHP (SIM: m/z = 315) and APCI for 22(OH)7DHC (SIM: m/z = 401) and 20,22(OH)₂7DHC (SIM: m/z = 417).

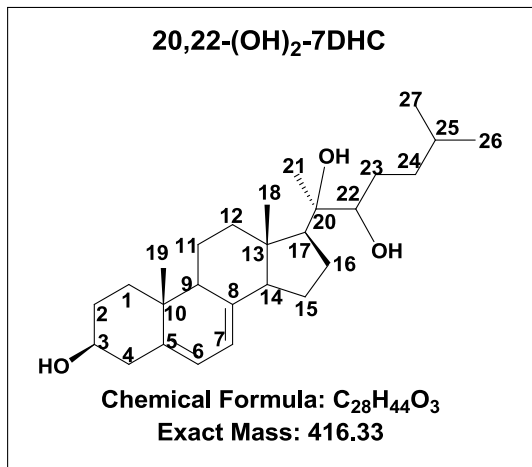
S-1**20,22-(OH)₂-7DHC** 439 [M+Na]⁺

Relative Intensity (%)

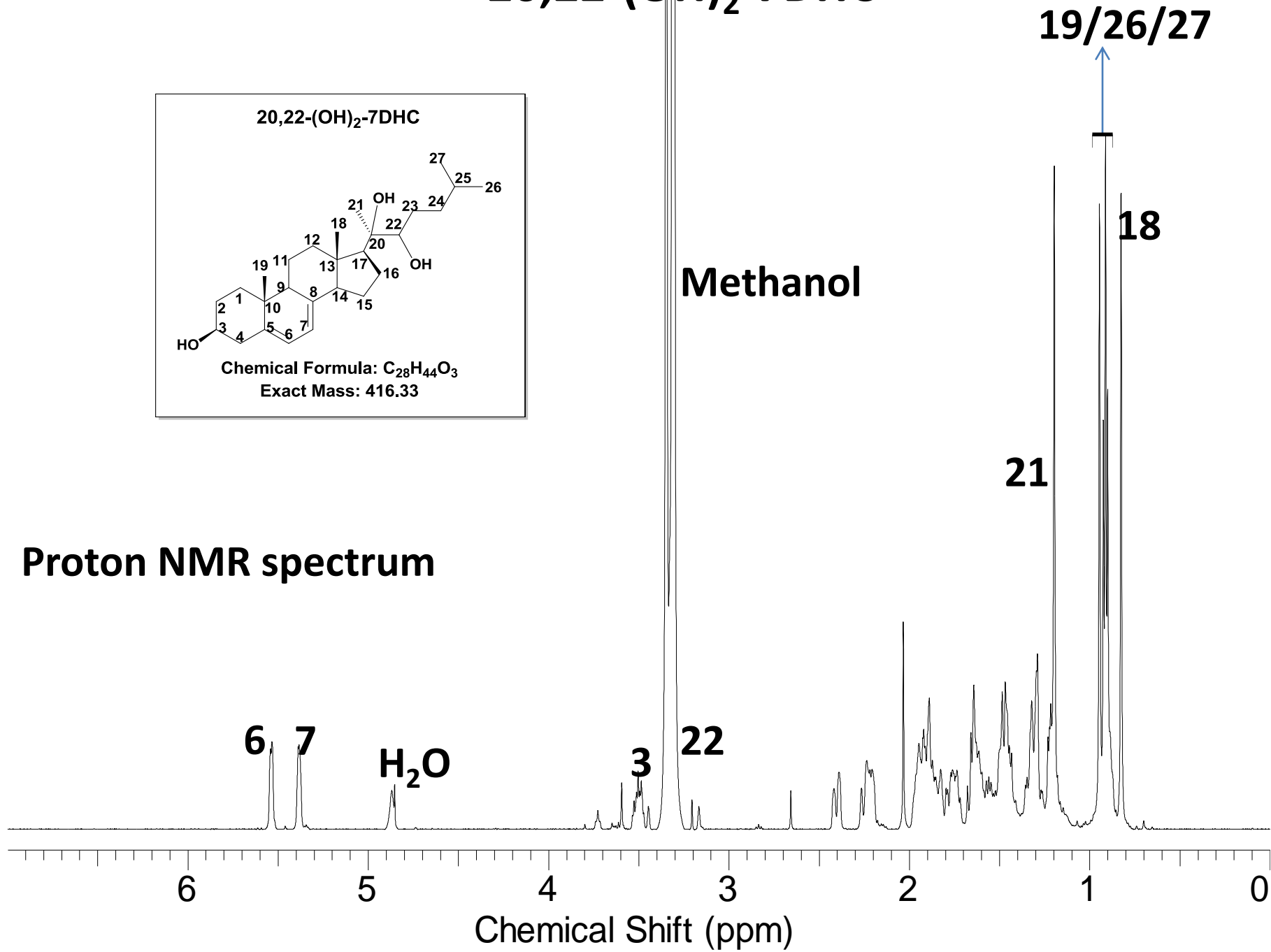
**Mass spectrum**

S-2

20,22-(OH)₂-7DHC

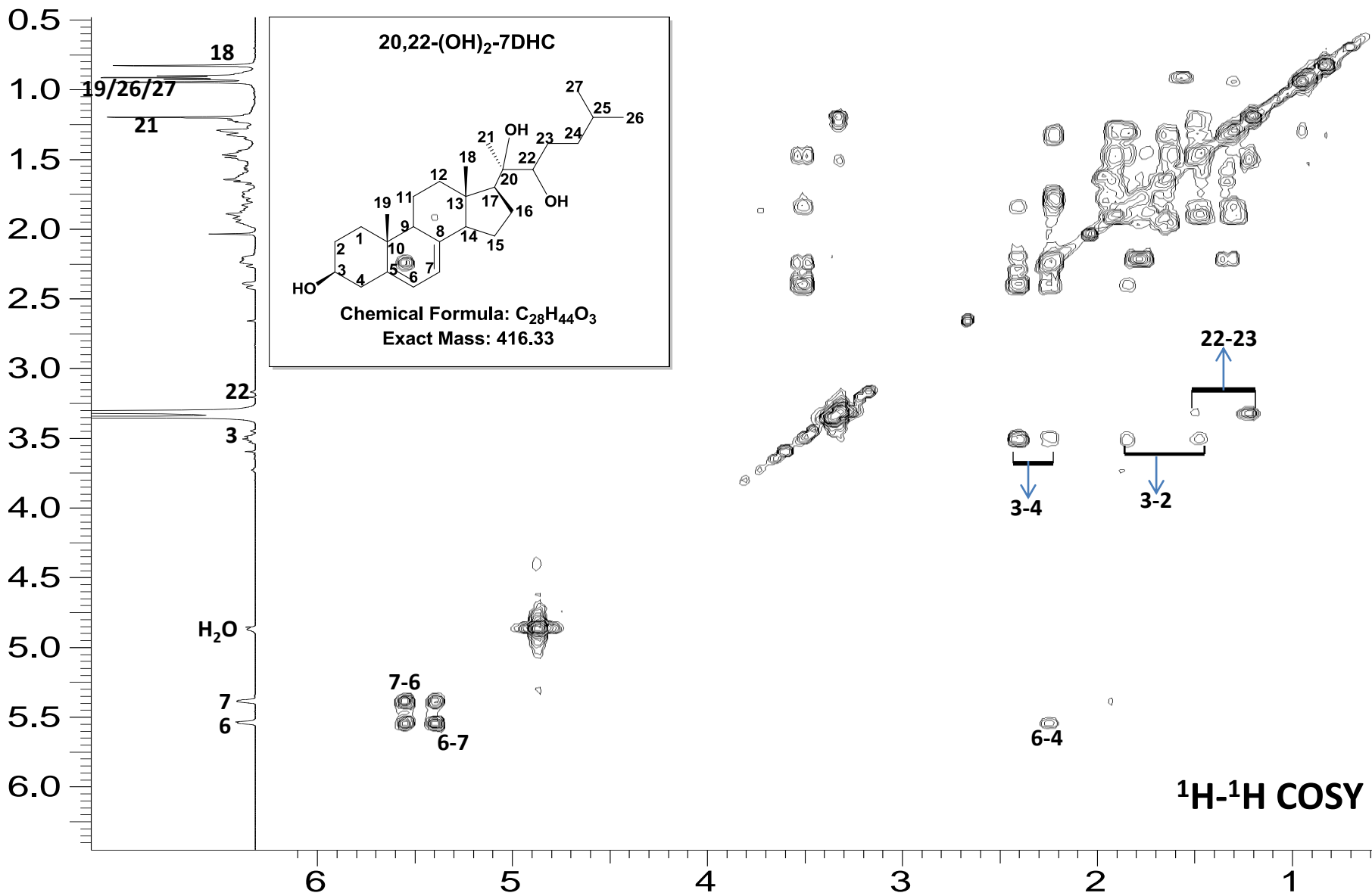


Proton NMR spectrum

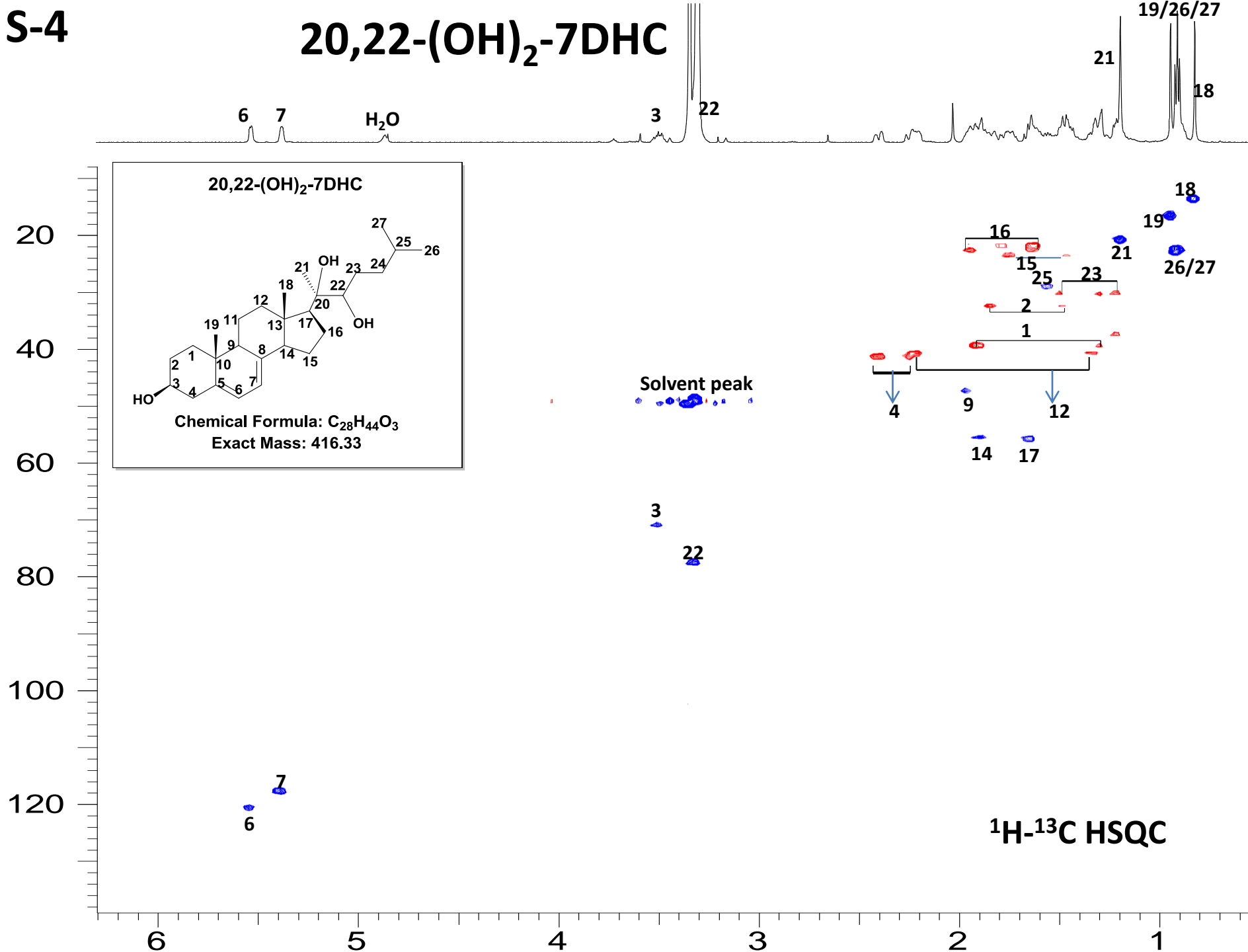


S-3

20,22-(OH)₂-7DHC

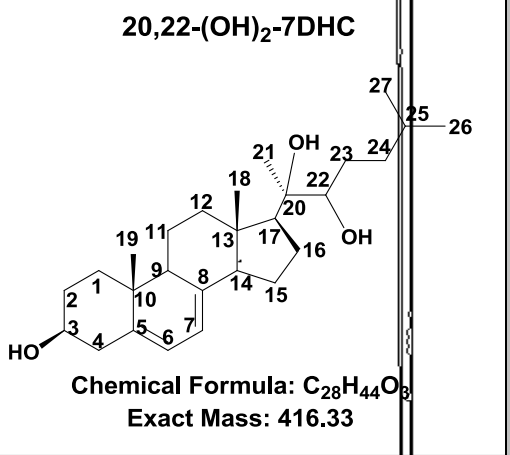
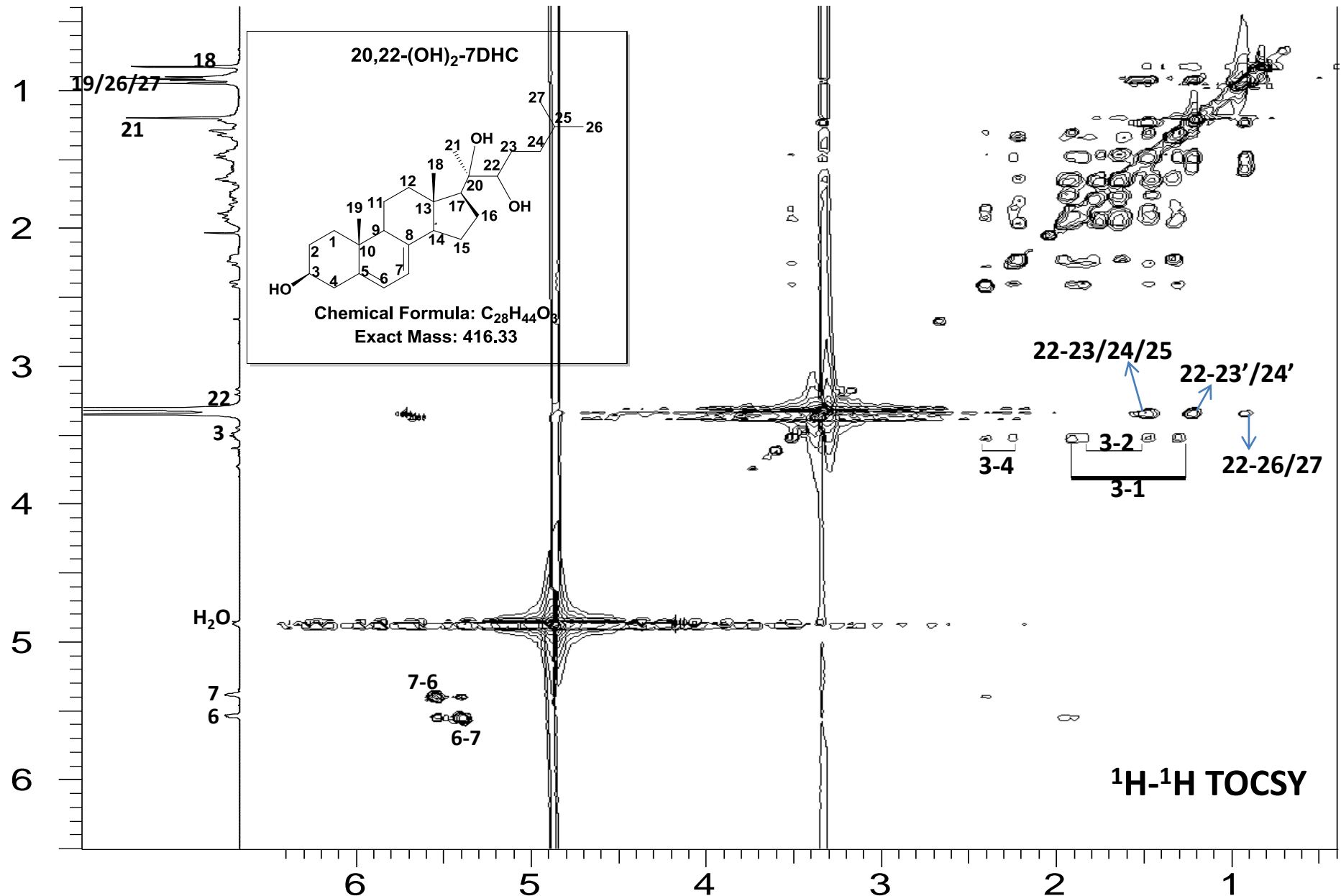


S-4

20,22-(OH)₂-7DHC

S-5

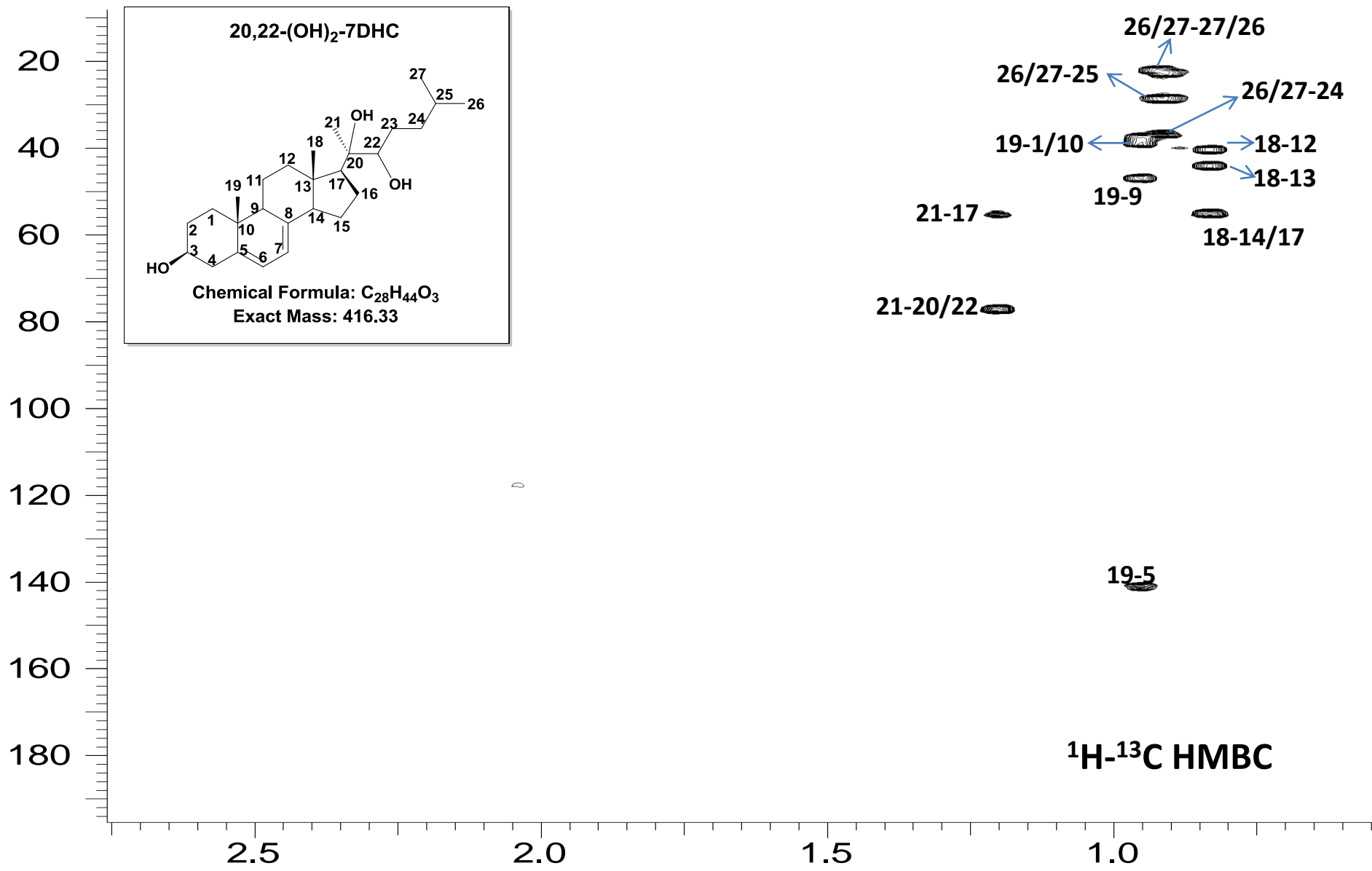
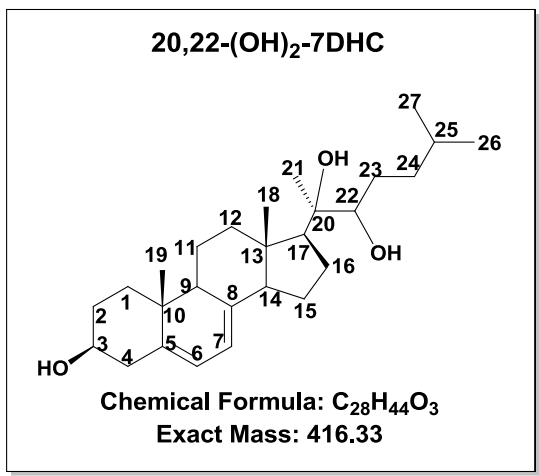
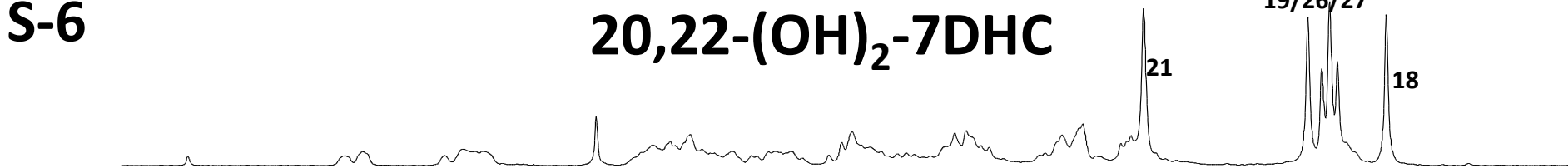
20,22-(OH)₂-7DHC



¹H-¹H TOCSY

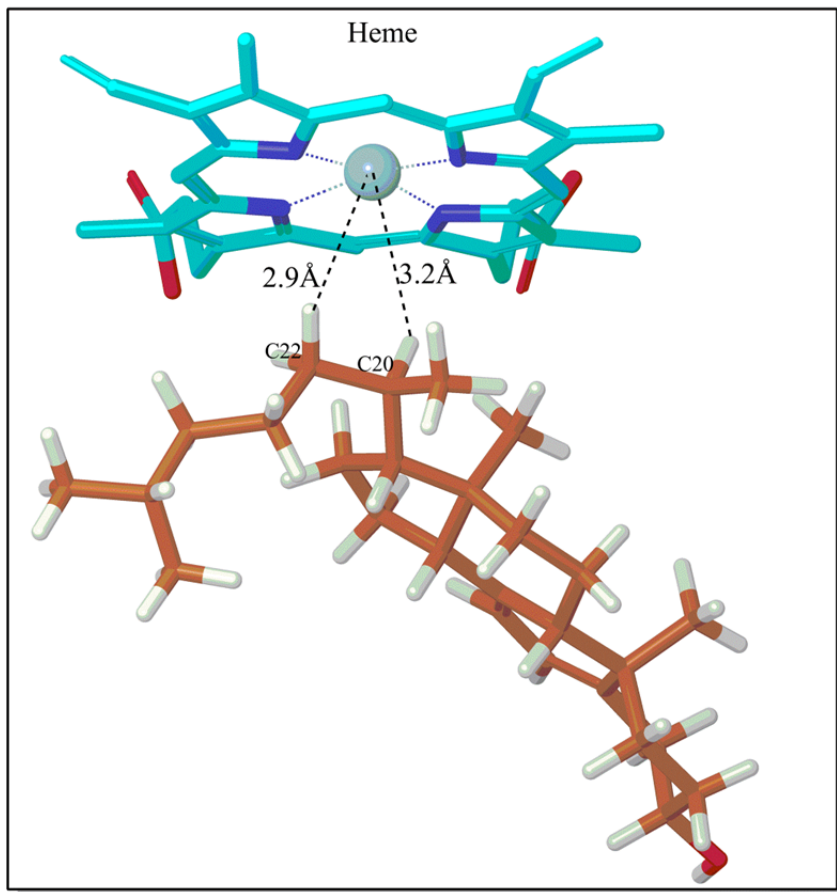
S-6

20,22-(OH)₂-7DHC

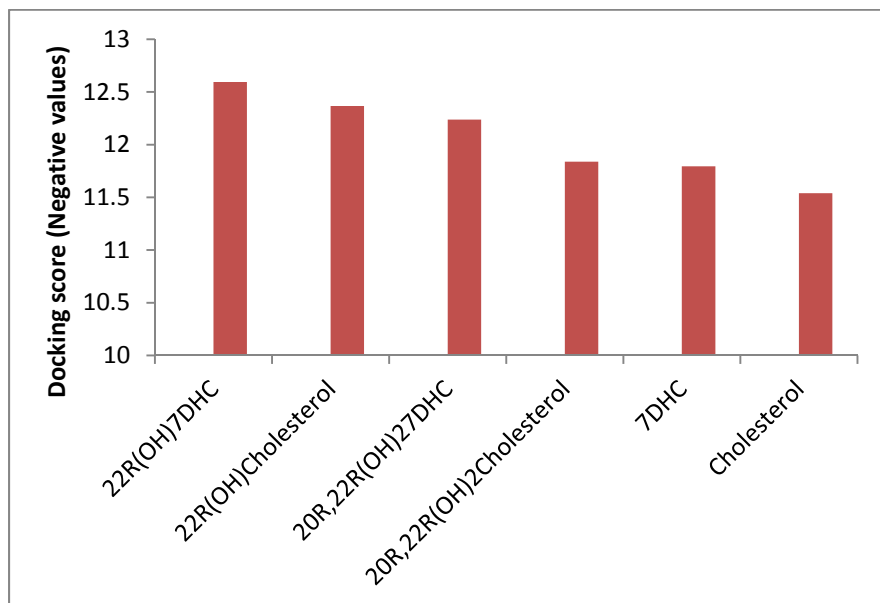


¹H-¹³C HMBC

S-7.



S-8.



S-9

