Supplemental Figure Legends

Figure S1. CID MS/MS fragmentation spectra of lanosterol, T-MAS, zymosterol and ergosterol standards.

Figure S2. Effect of PSZ and FLU on the proliferation of *L. donovani* promastigotes. The growth of *L. donovani* LV82 promastigotes was determined in the presence or absence of PSZ or FLU for 72 h as outlined in the Methods section. Data shown represent biological replicates for PSZ (n=6 determinations) and FLU (n=4 determinations). The PSZ data was fitted using the four parameter equation $y = m1 + (m2 – m1)/[1 + (x/m3)^{m4}]$ from GraphPad Prism (v. 9.3.1 San Diego, CA) and then the absolute IC_{50} value was determined as in Joice *et al.*[30] to be 2.8 μM. The IC_{50} value for FLU could not be ascertained as growth inhibition did not reach 50% at the highest FLU concentration tested, indicating an IC_{50} > 100 μM. For both PSZ and FLU, error bars represent the standard error of the biological replicates.

Figure S3. Effect of PSZ and FLU on the proliferation of *L. tarentolae* promastigotes. The growth of *L. tarentolae* UC strain promastigotes was determined in the presence or absence of PSZ or FLU for 72 h as outlined in the Methods section. Symbols and error bars represent the mean and standard error of biological triplicates. Data were fitted using the four parameter equation $y = m1 + (m2 – m1)/[1 + (x/m3)^{m4}]$ from GraphPad Prism (v. 9.3.1 San Diego, CA). The PSZ data did not fit the four parameter equation above ($R^2 < 0.5$); the absolute IC_{50} value for FLU was determined as in Joice *et al.*[30] to be 170 μM.

Figure S4. ^{1}H NMR spectra of (A) lanosterol standard and (B) the purified unknown intermediate sterol 4,14-dimethylzymosterol.

Figure S5. ^{13}C NMR spectra of (A-C) lanosterol standard and (D-F) the purified unknown intermediate sterol 4,14-dimethylzymosterol.

Figure S6. HMBC NMR spectra of (A-C) lanosterol standard and HMBC (D-E) and HSQC (F) NMR spectra the purified unknown intermediate sterol 4,14-dimethylzymosterol.

Figure S1. CID MS/MS fragmentation spectra of lanosterol, T-MAS, zymosterol and ergosterol standards.

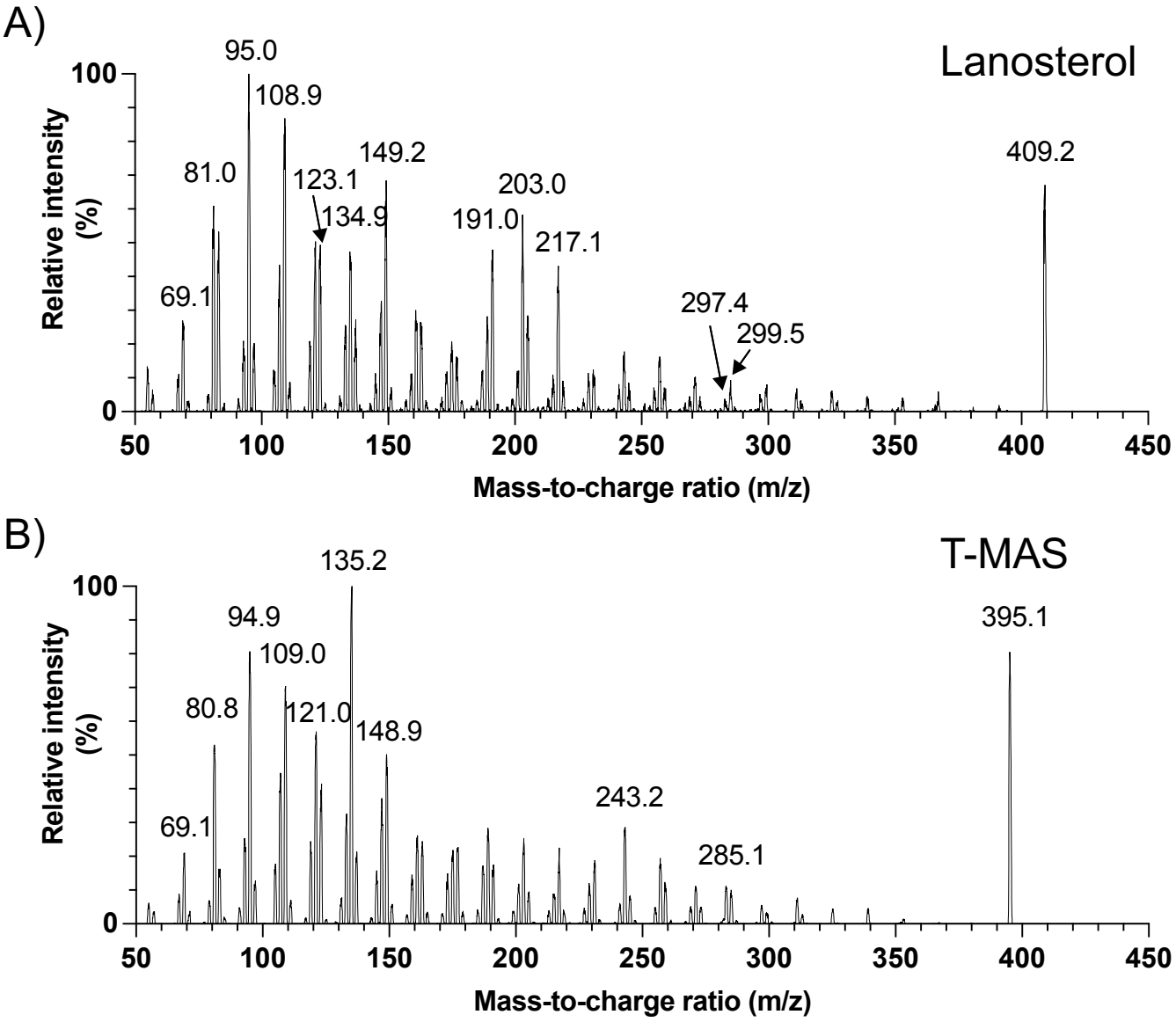


Figure 1S - Cont'd

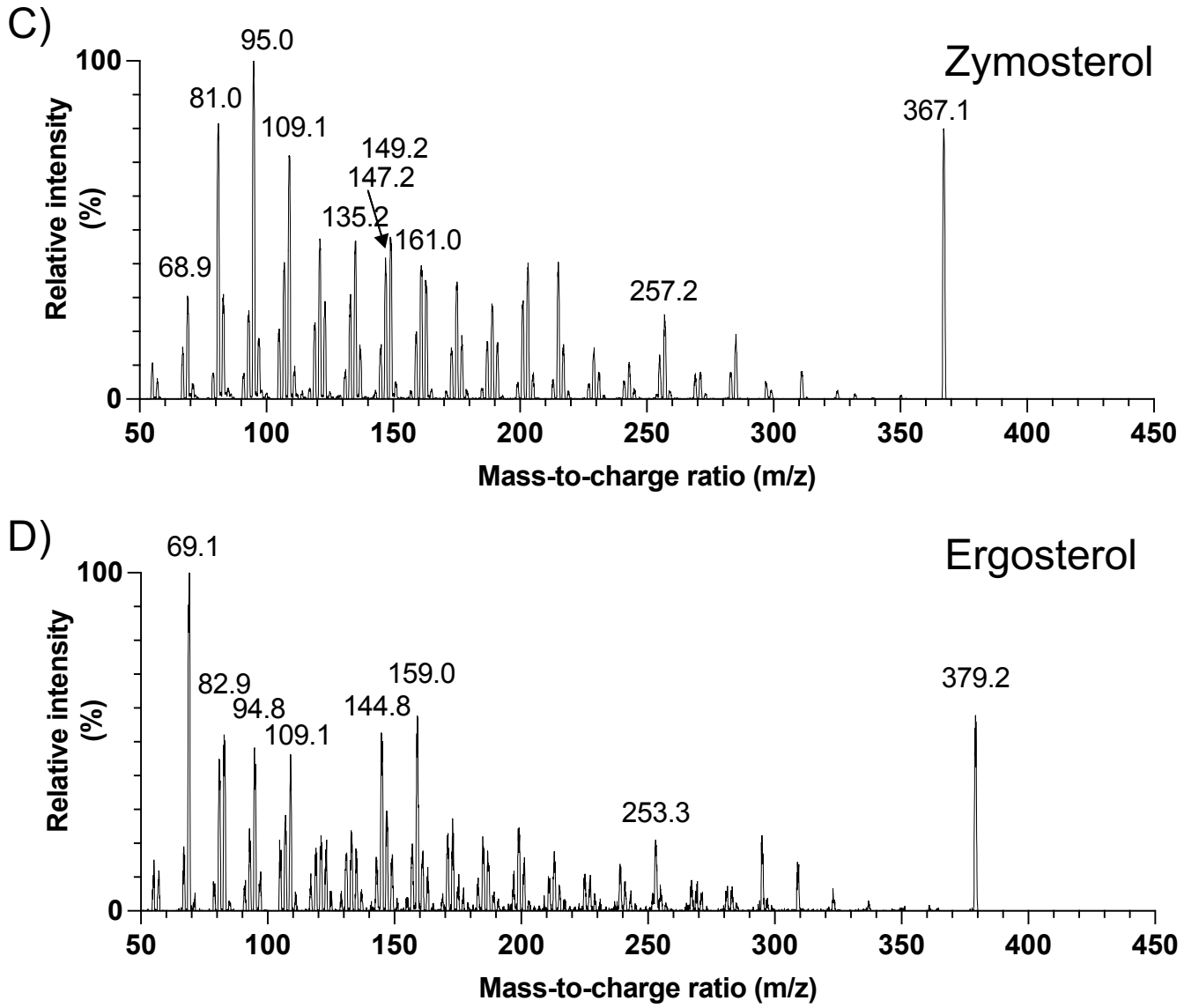


Figure S2. Effect of PSZ and FLU on the proliferation of *L. donovani* promastigotes.

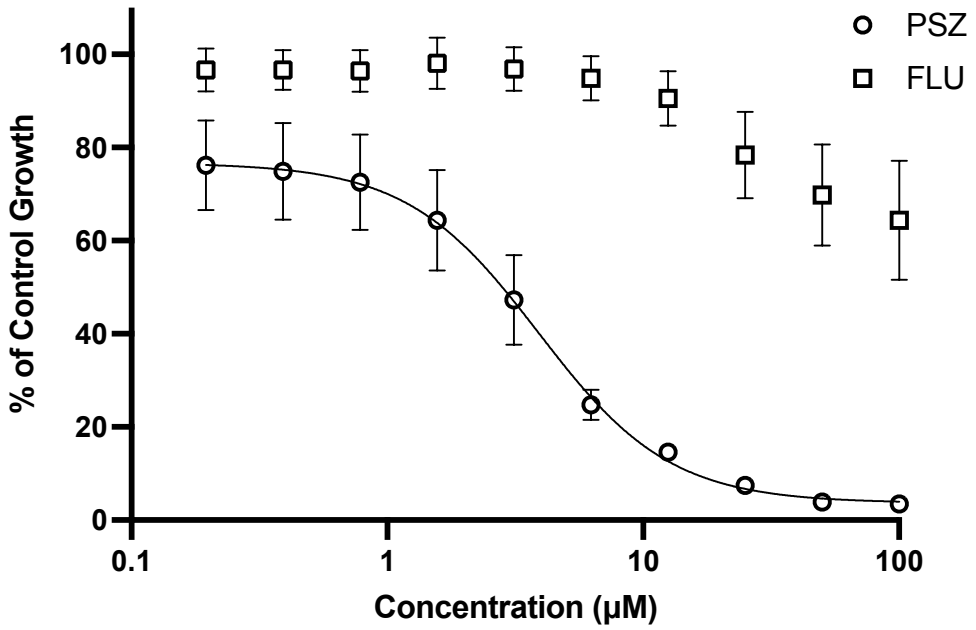


Figure S3. Effect of PSZ and FLU on the proliferation of *L.* tarentolae promastigotes.

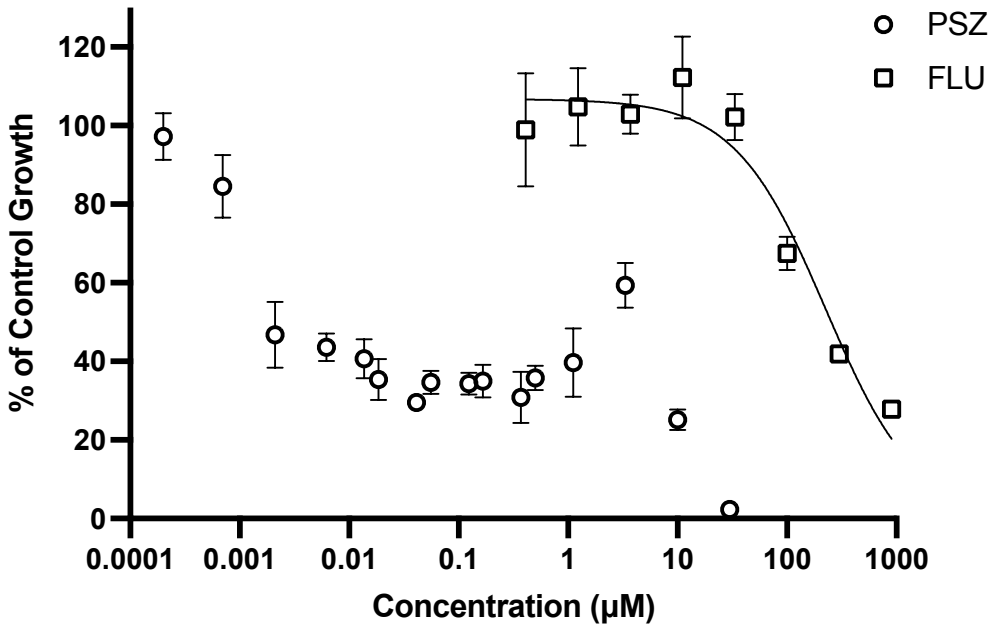


Figure S4. ^{1}H NMR spectra of (A) lanosterol standard and (B) the purified unknown intermediate sterol 4,14-dimethylzymosterol.

A) Lanosterol ^{1}H

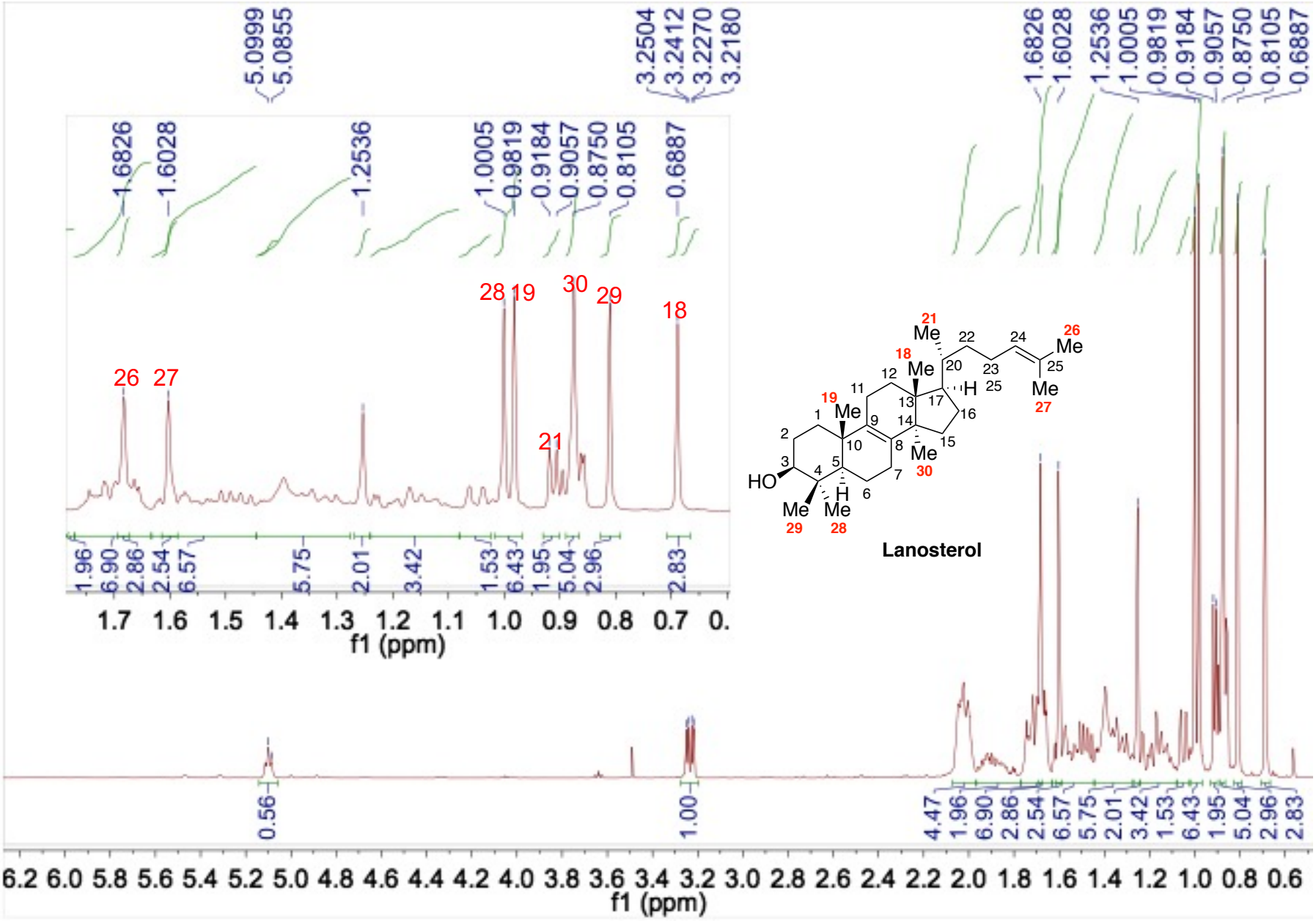


Figure S4. cont'd

B) Unknown ^{1}H

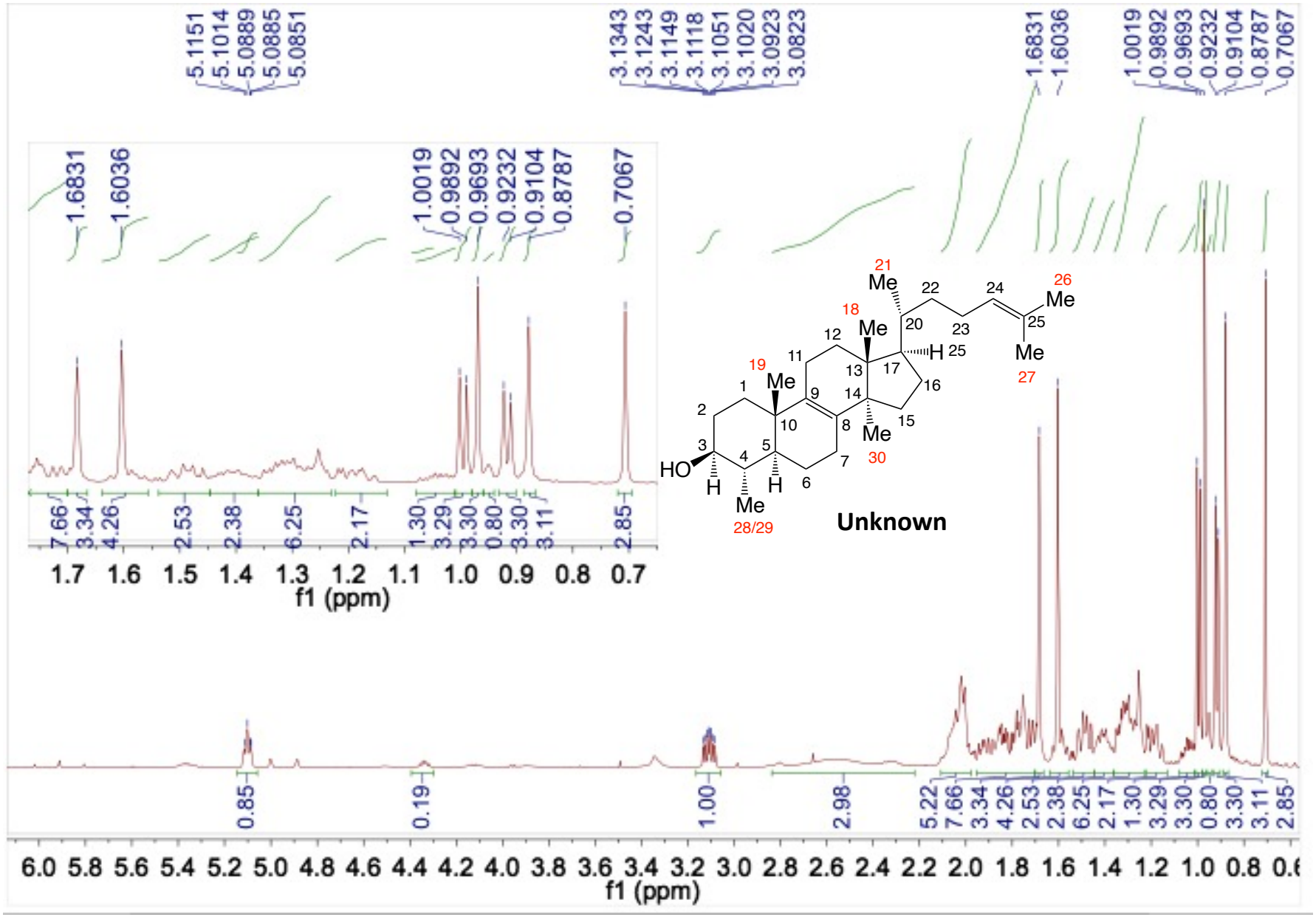
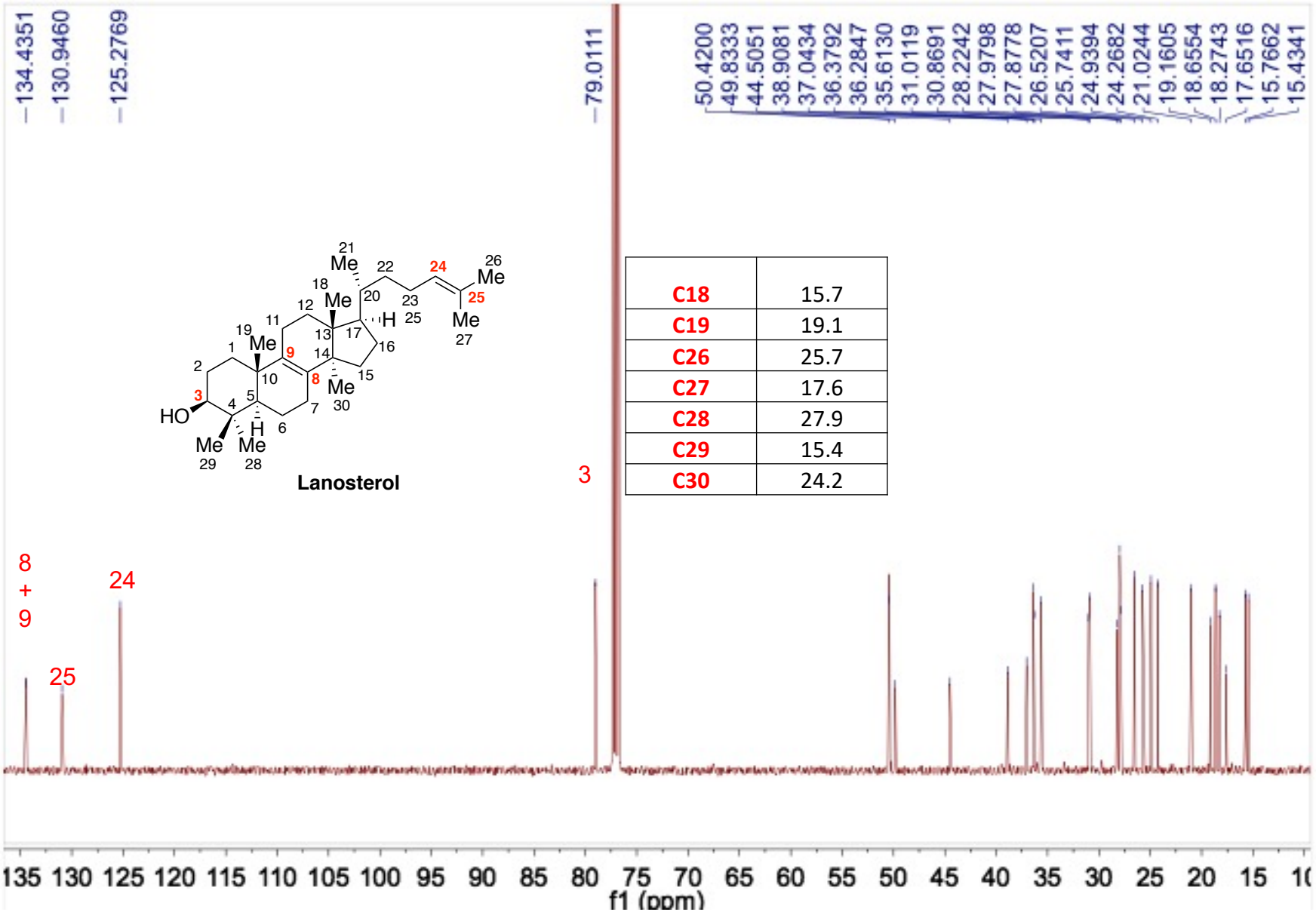


Figure S5. ^{13}C NMR spectra of (A-C) lanosterol standard and (D-F) the purified unknown intermediate sterol 4,14-dimethylzymosterol.

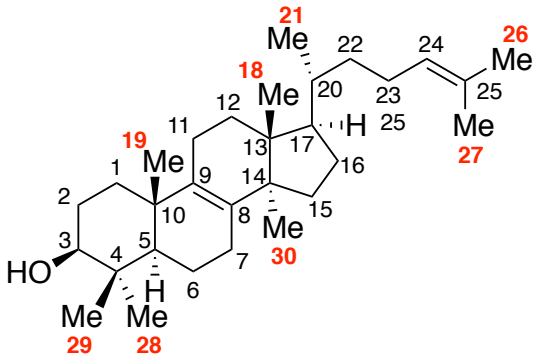
A) Lanosterol ^{13}C



C18	15.7
C19	19.1
C26	25.7
C27	17.6
C28	27.9
C29	15.4
C30	24.2

Figure S5. cont'd

B) Lanosterol DEPT vs. ^{13}C

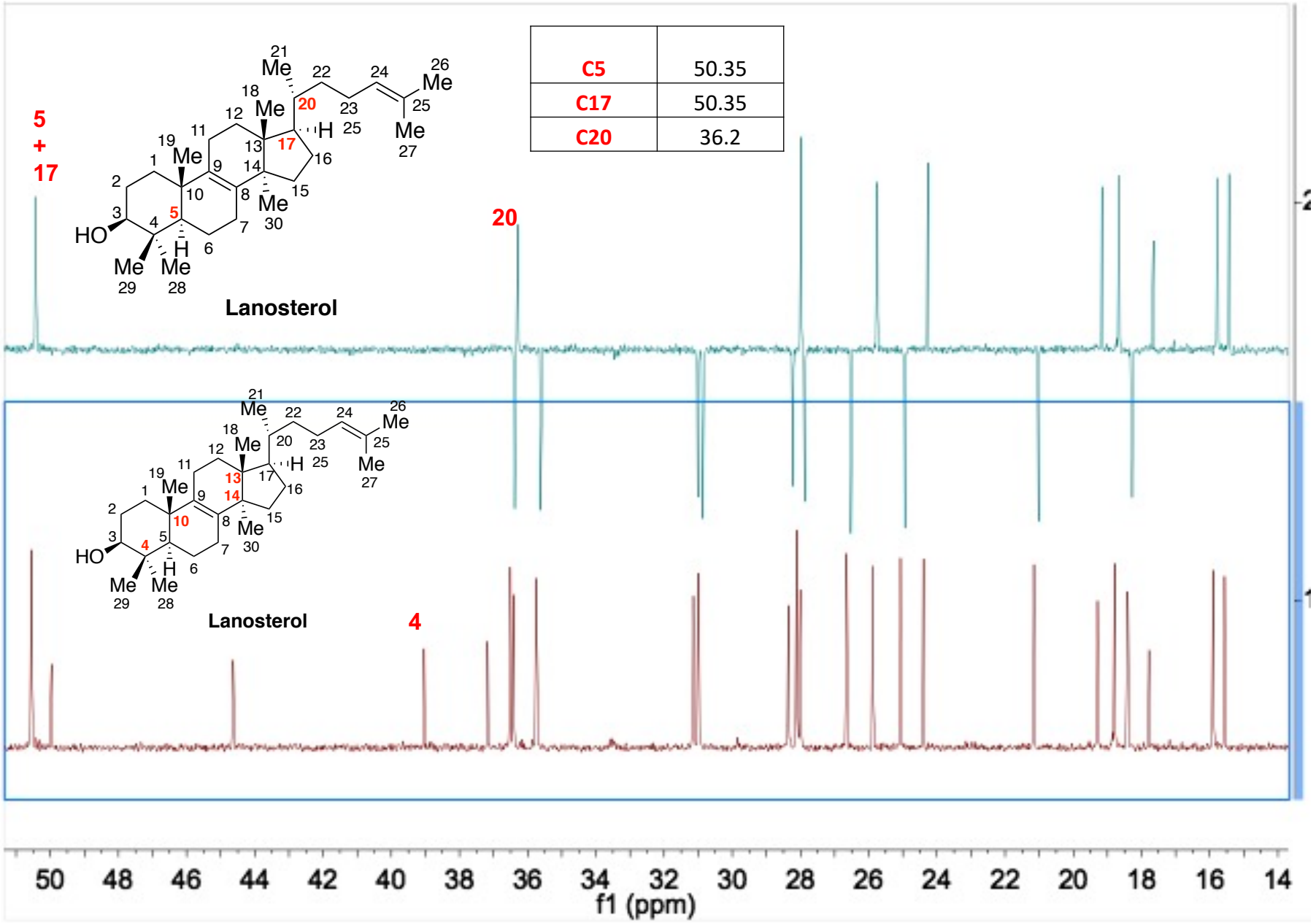


Lanosterol

C18	15.7
C19	19.1
C21	18.6
C26	25.7
C27	17.6
C28	27.9
C29	15.4
C30	24.2

Figure S5. cont'd

C) Lanosterol DEPT vs. ^{13}C



C5	50.35
C17	50.35
C20	36.2

Figure S5. cont'd

D) Unknown ^{13}C

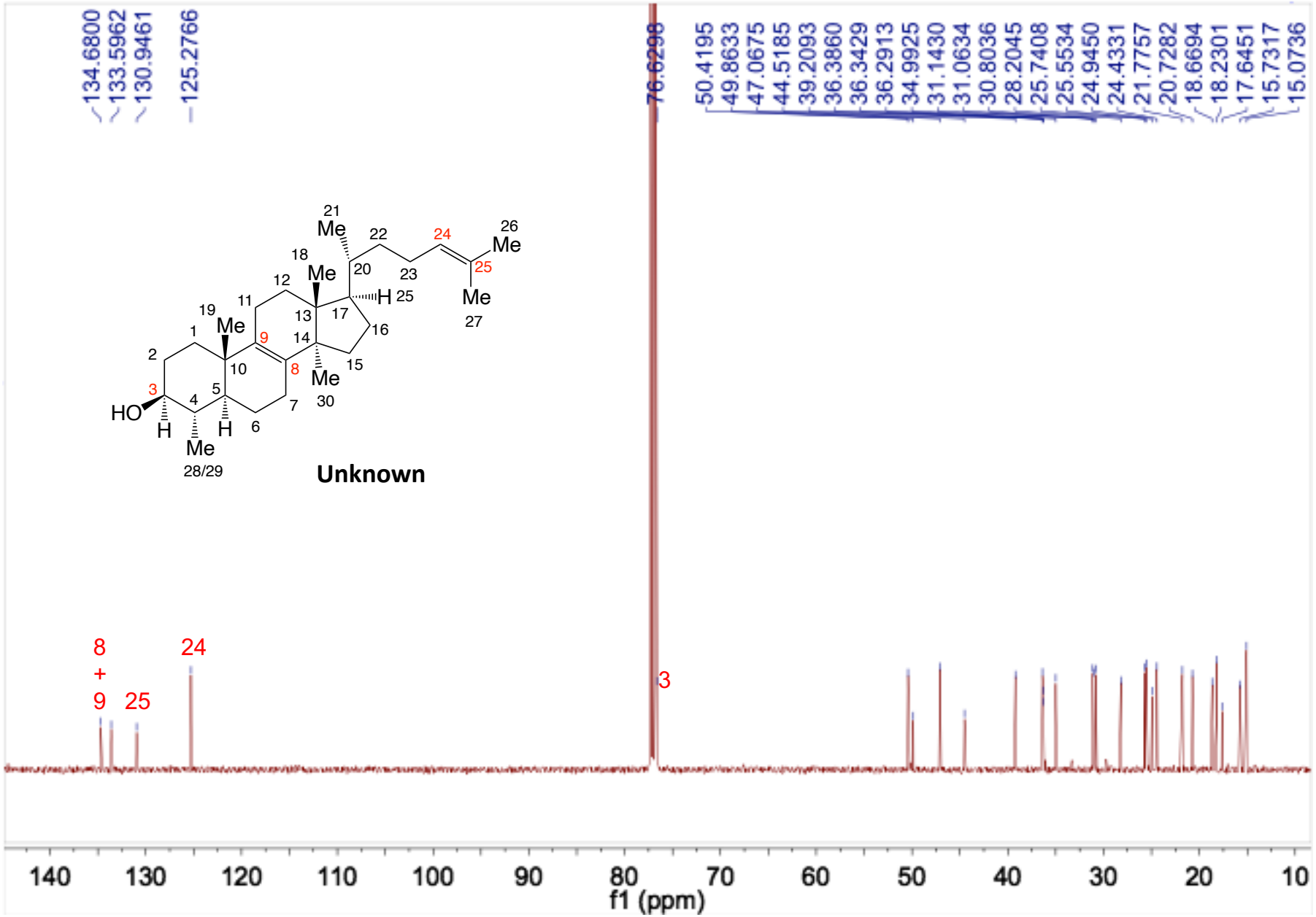


Figure S5. cont'd

E) Unknown DEPT

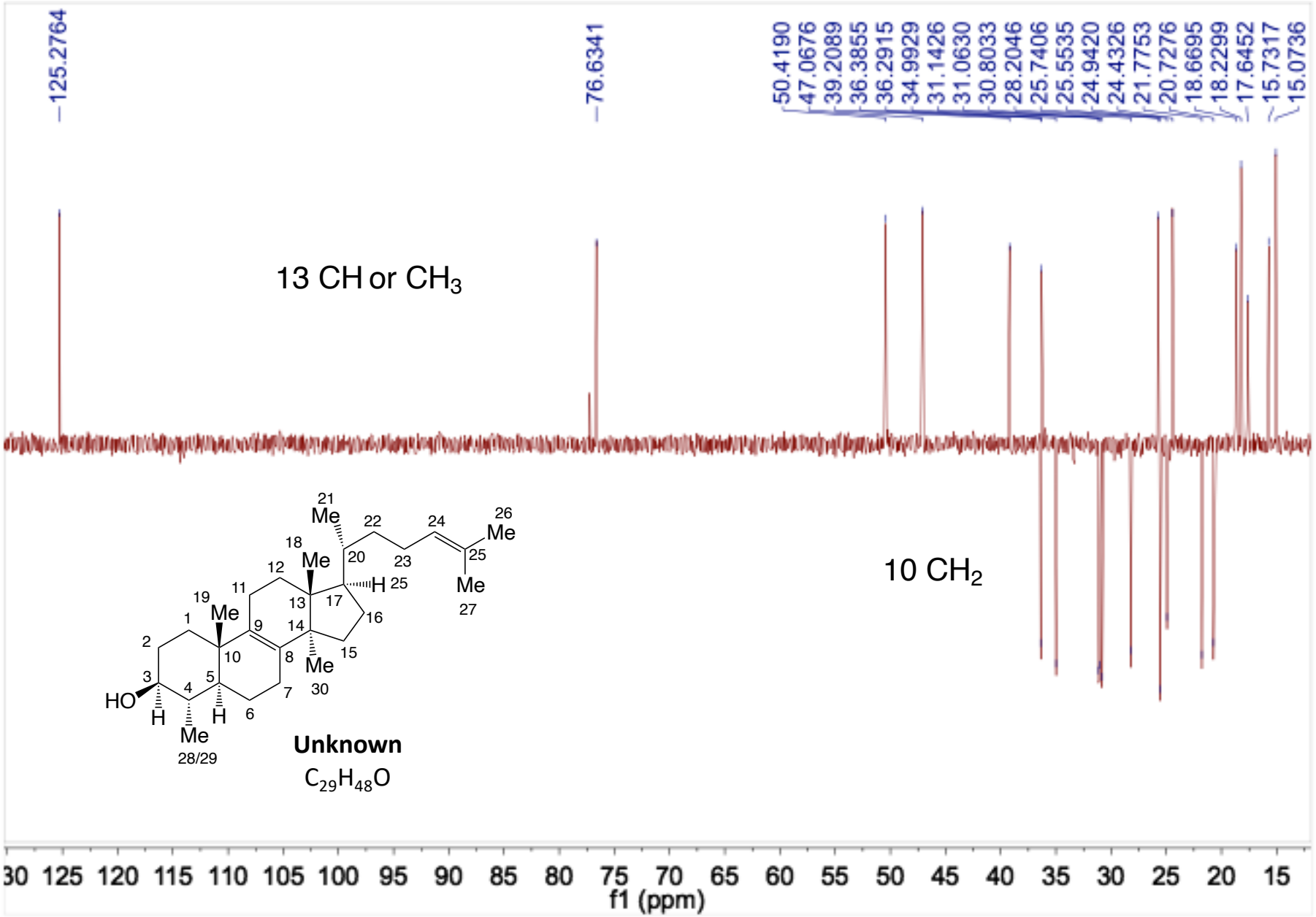


Figure S5. cont'd

F) Unknown DEPT vs. ^{13}C

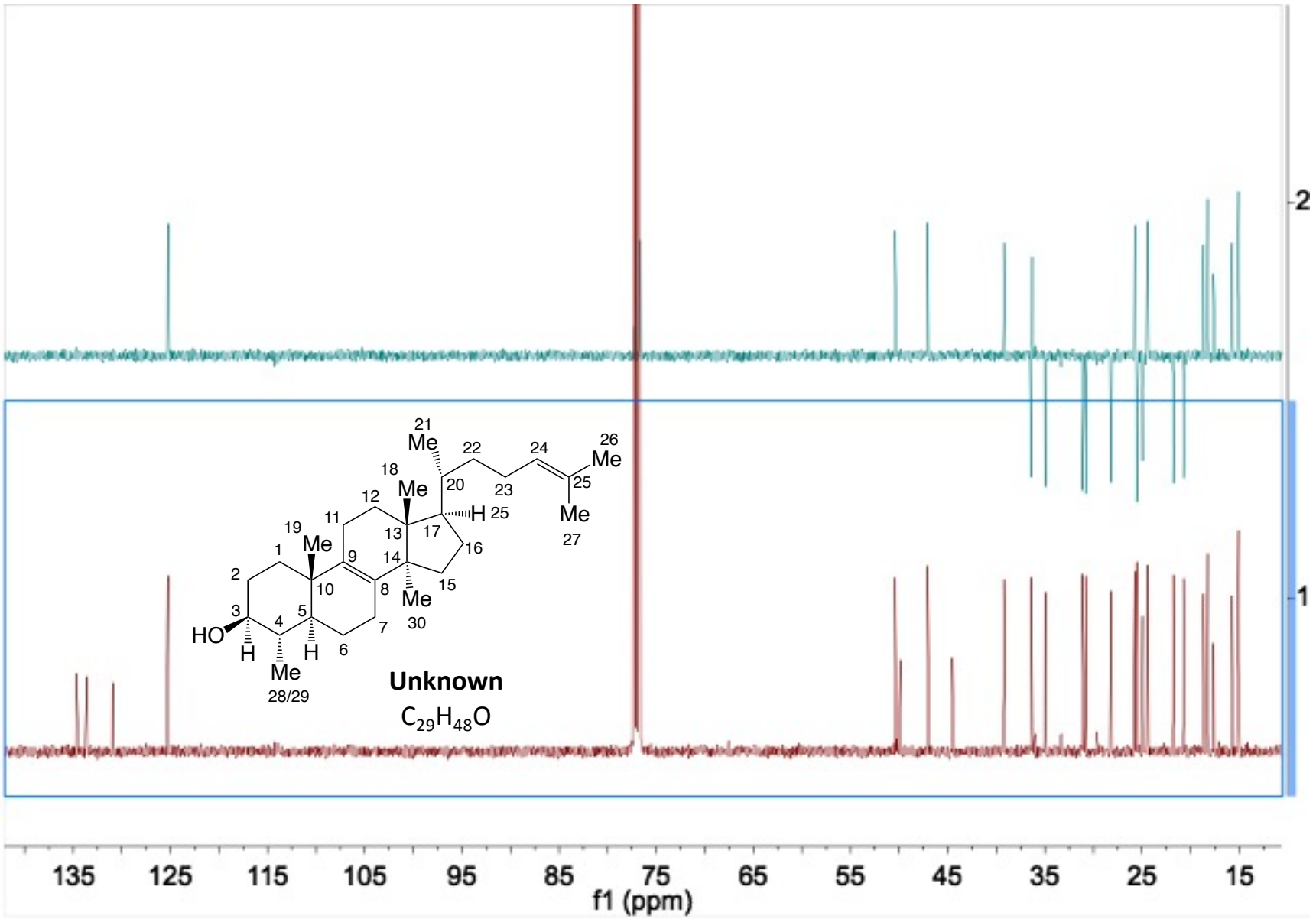


Figure S6. HMBC NMR spectra of lanosterol standard and the purified unknown intermediate sterol 4,14-dimethylzymosterol.

A) Lanosterol HMBC – H3 near C4

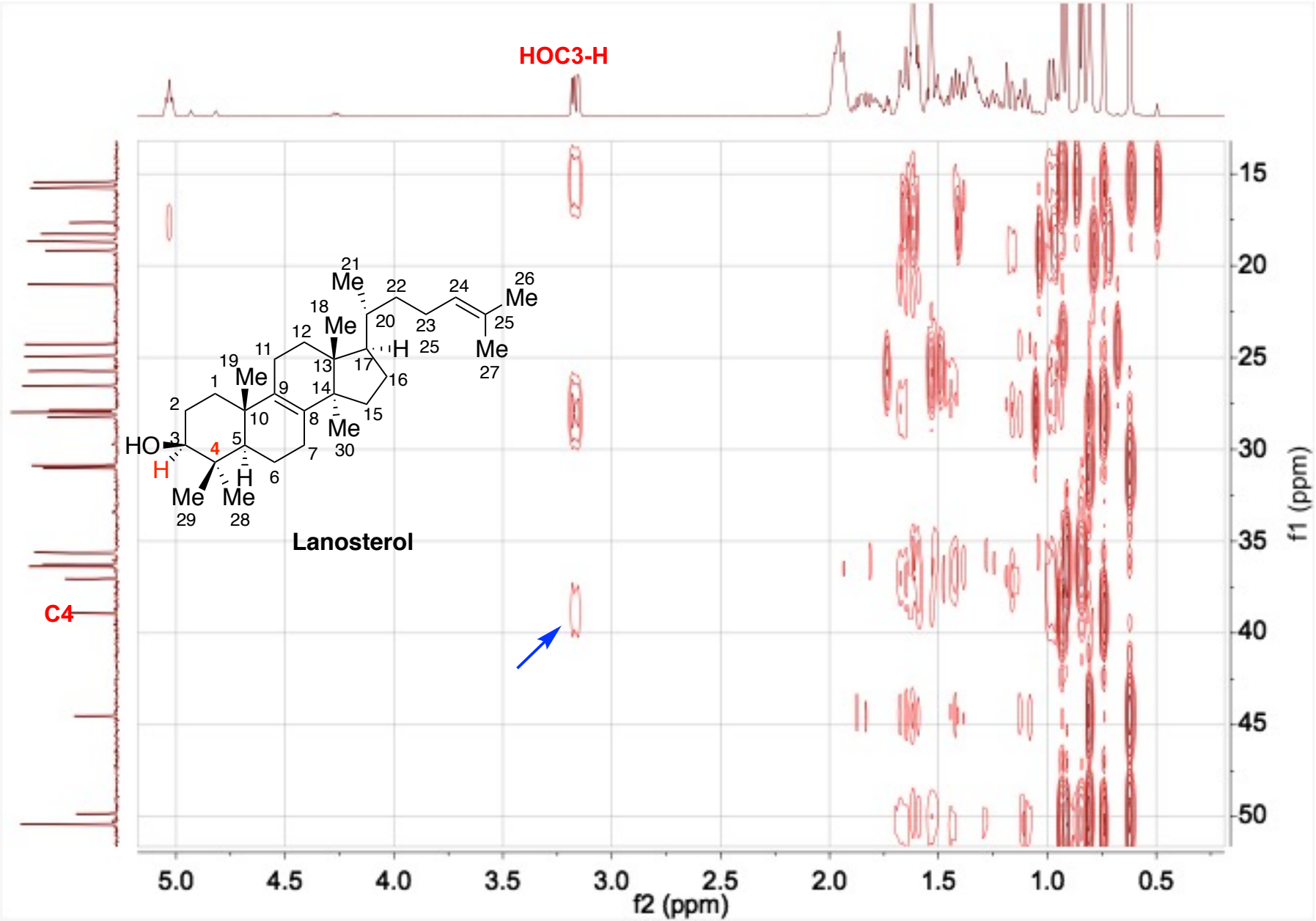


Figure S6. cont'd

B) Lanosterol HMBC

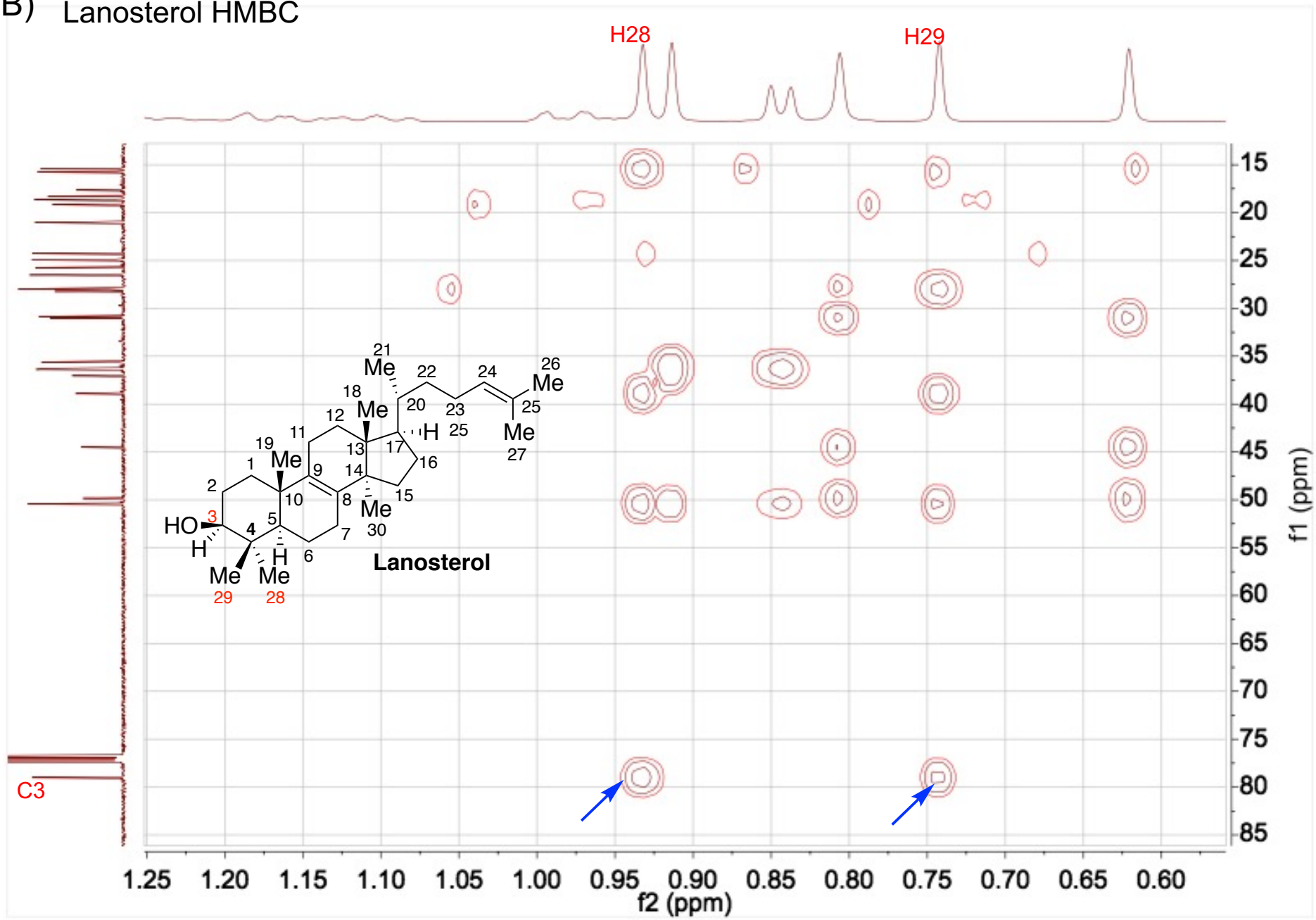


Figure S6. cont'd

C) Lanosterol HMBC

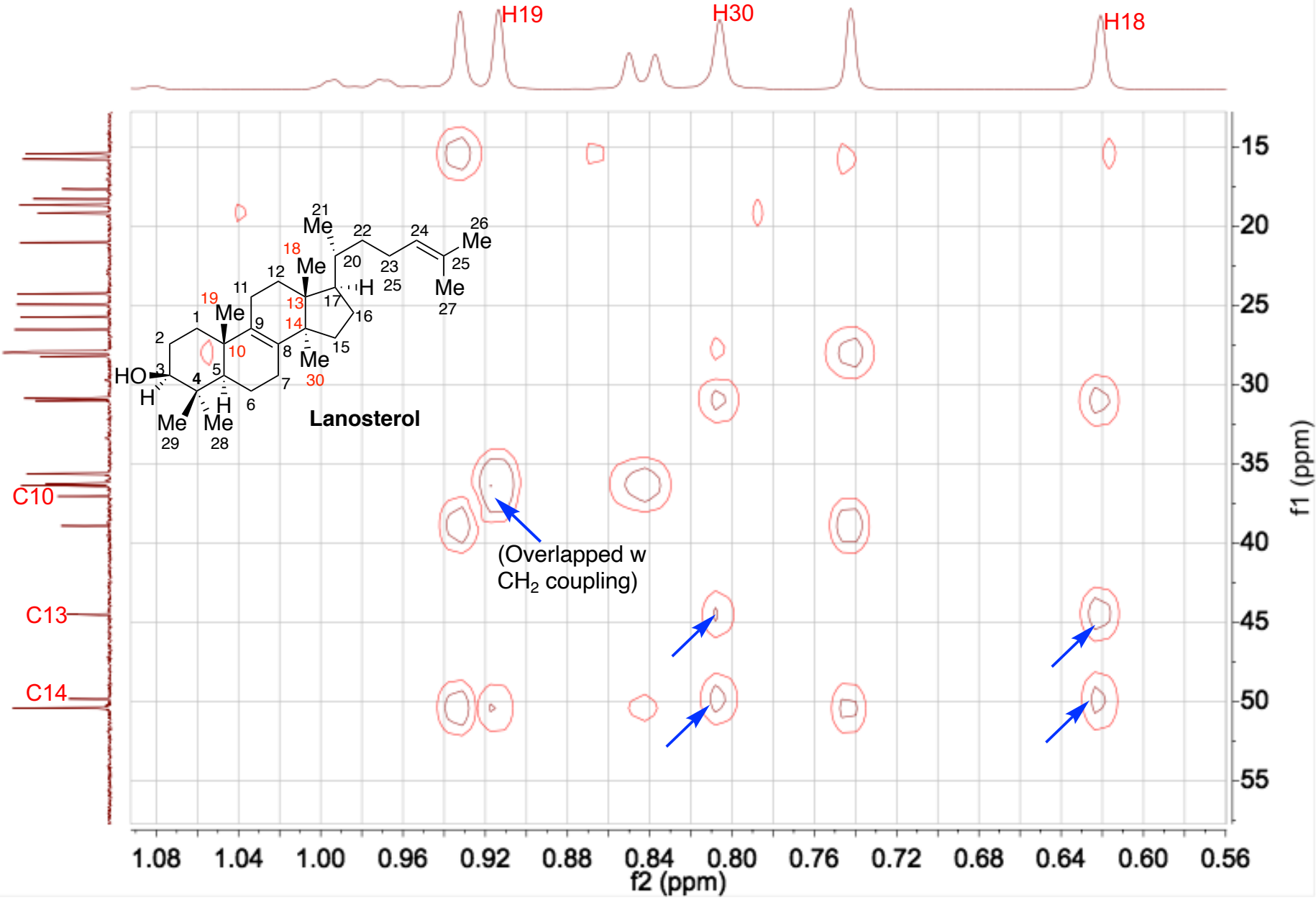


Figure S6. cont'd

D) Unknown HMBC

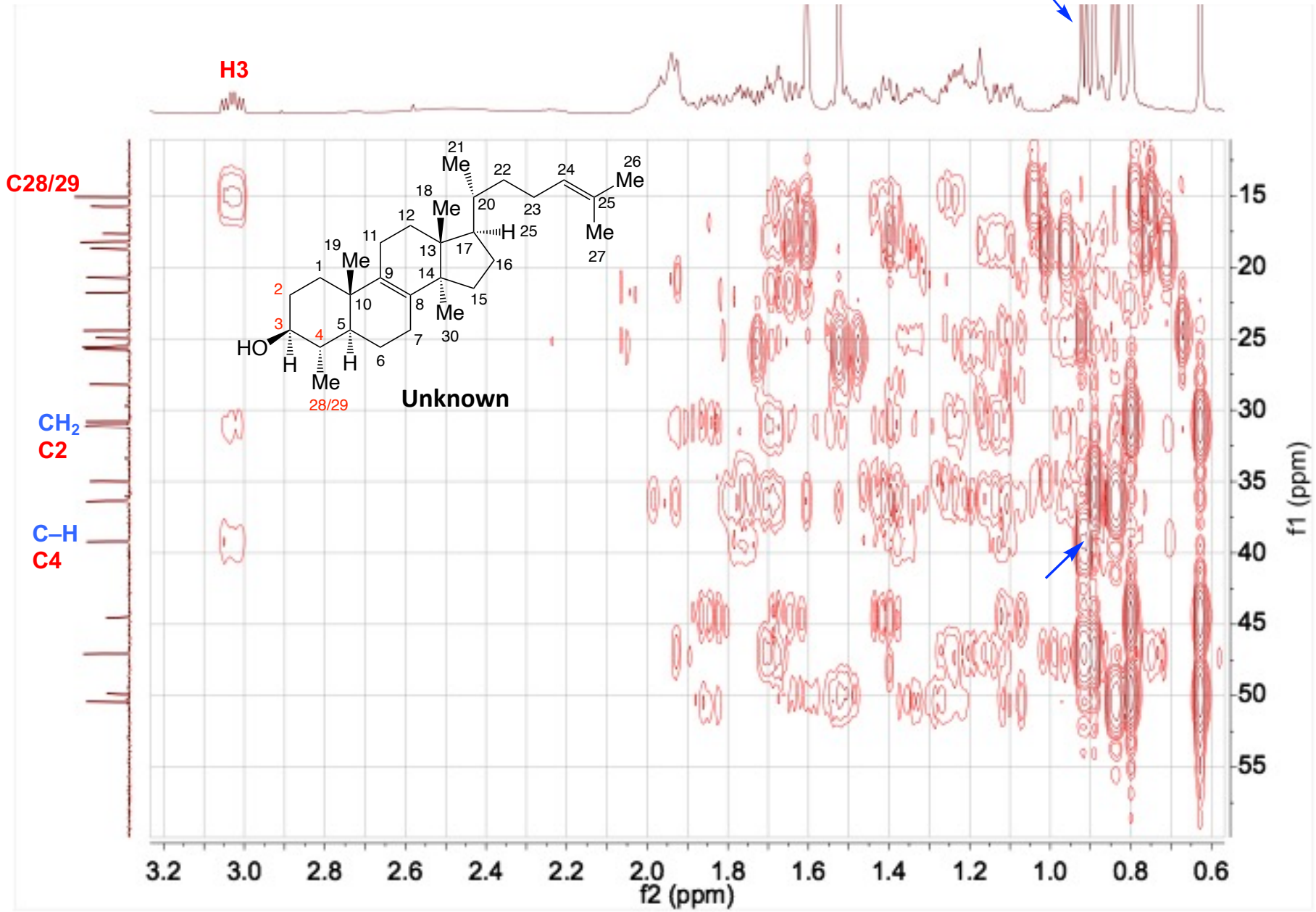


Figure S6. cont'd

E) Unknown HMBC - Establishing methyl's 18, 19, 30

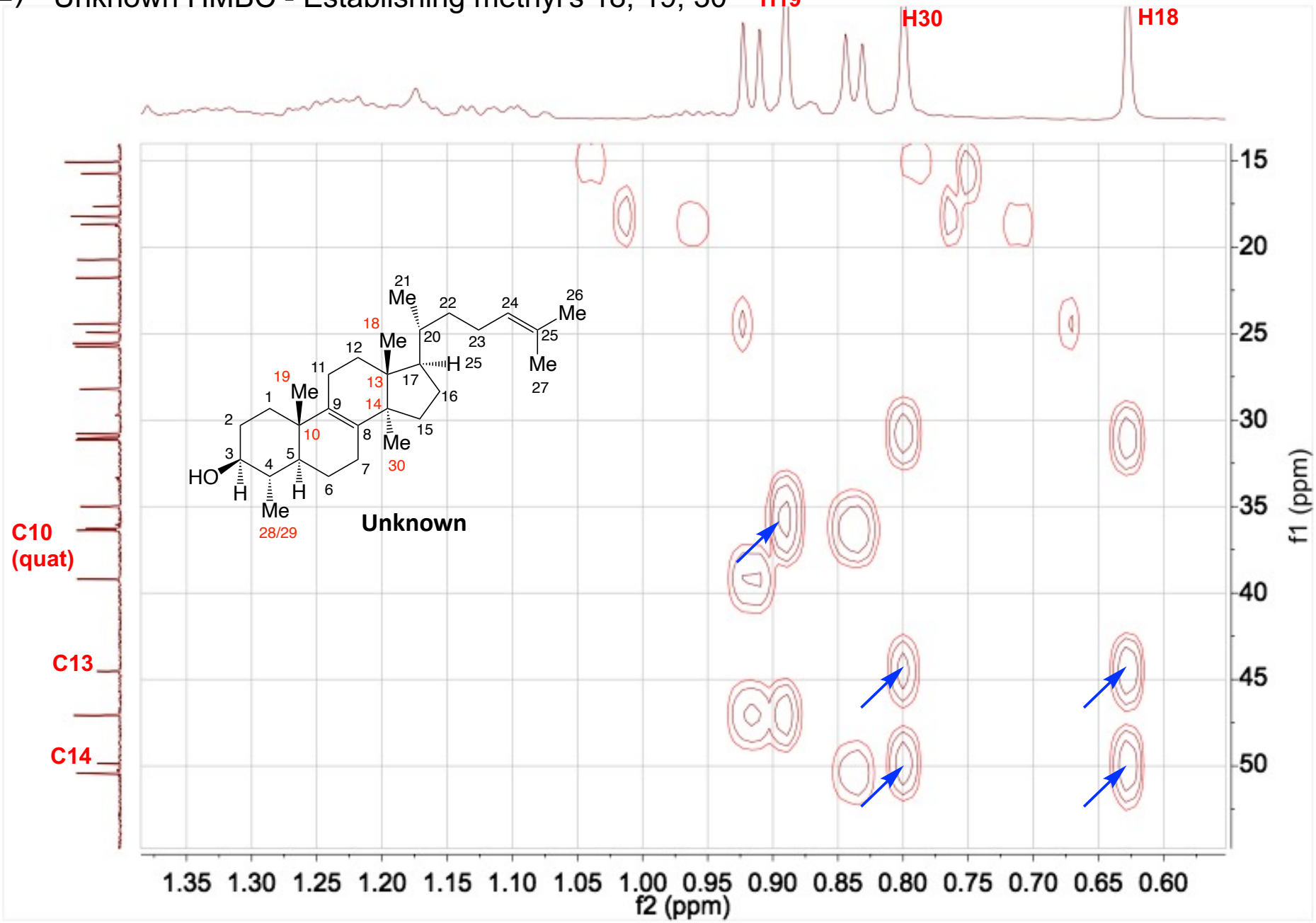


Figure S6. cont'd

F) Unknown HSQC - Locating H4 & H5 and assignment of corresponding ^{13}C peaks