SUPPLEMENTAL DATA FOR

Breaking the regioselectivity of indole prenyltransferases: identification of regular C3-prenylated hexahydropyrrolo[2,3-b]indoles as side products of the regular C2-prenyltransferase FtmPT1

Beate Wollinsky[a], Ludwig Lena[a], Xiulan Xie[b], Shu-Ming Li[a*]

[a]Institut für Pharmazeutische Biologie und Biotechnologie, Philipps Universität Marburg, Deutschhausstrasse 17a, 35037 Marburg, Germany

[b]Fachbereich Chemie, Philipps Universität Marburg, Hans-Meerwein-Strasse, 35032 Marburg, Germany

**corresponding to: shuming.li@staff.uni-marburg.de; tel: 0049-64212822461; fax: 0049-64212825365*

Figure S15 HPLC chromatograms of incubation mixtures of **10a**, **10b** or **10c** in the presence and absence of FtmPT1.

Figure S16 HPLC chromatograms of incubation mixtures of **11a**, **11b** or **11c** in the presence and absence of FtmPT1.

Figure S17 HPLC chromatograms of incubation mixtures of **13a**, **13b** or **13c** in the presence and absence of FtmPT1.

Figure S18 Dependence of product formation of **b** and **c** series on different substrate concentrations of **1a**, **2a**, **5a**, **9a**, **10a**, **11a**, **13a** and **14a**.

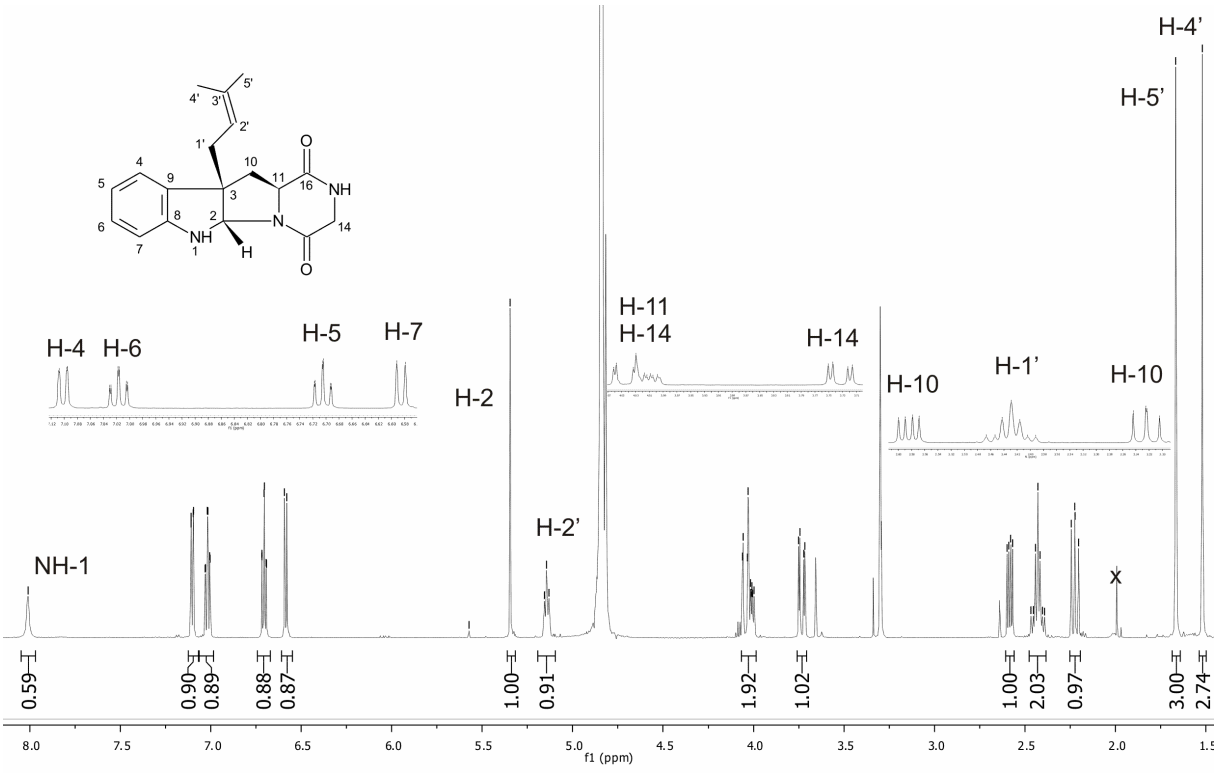

Figure S1.1 ^{1}H-NMR spectrum of *cyclo*-C3-dimethylallyl-L-Trp-Gly (**2c**) in CD_3OH (600 MHz)

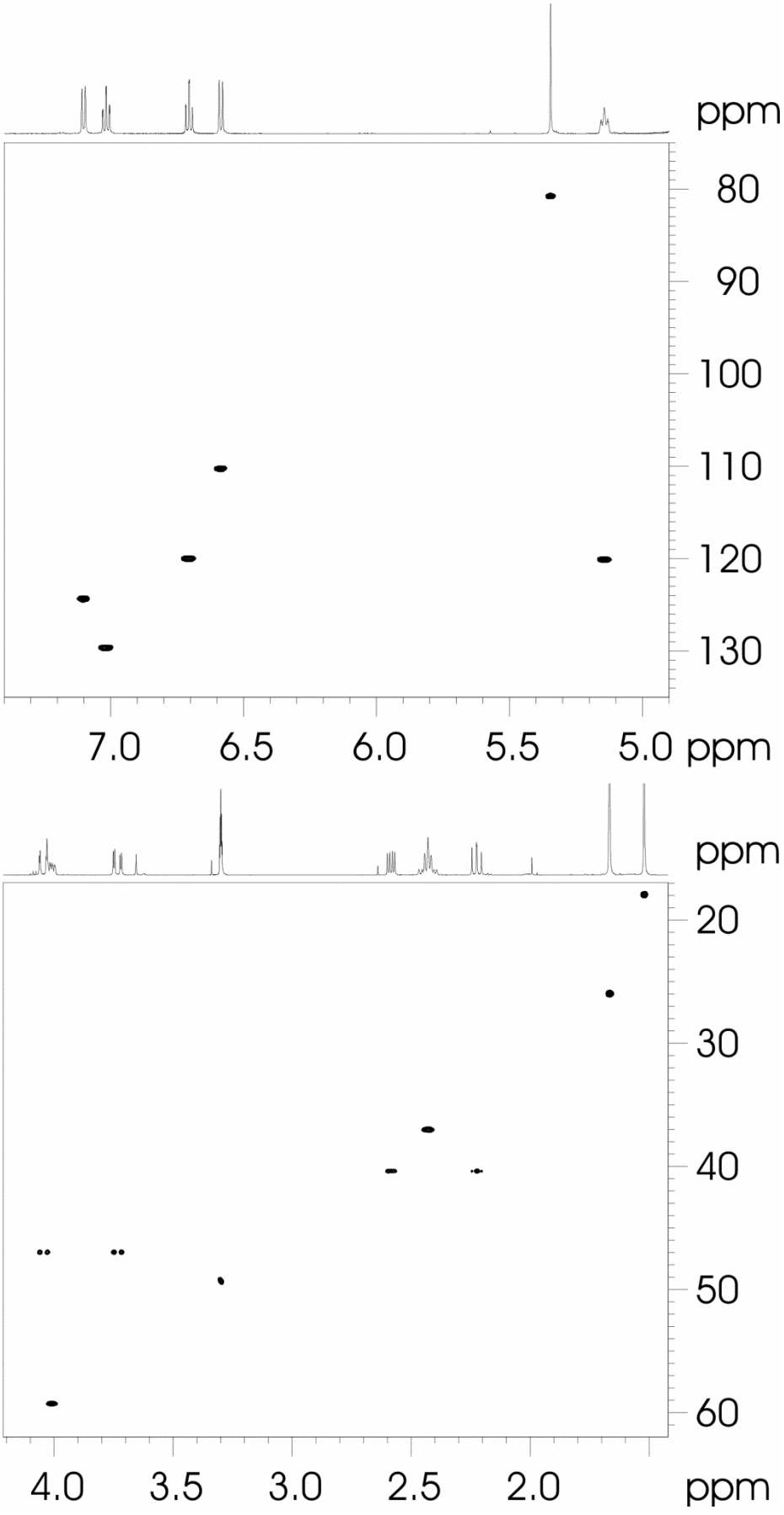

Figure S1.2 HSQC spectrum of *cyclo*-C3-dimethylallyl-L-Trp-Gly (**2c**) in CD_3OH (600 MHz)

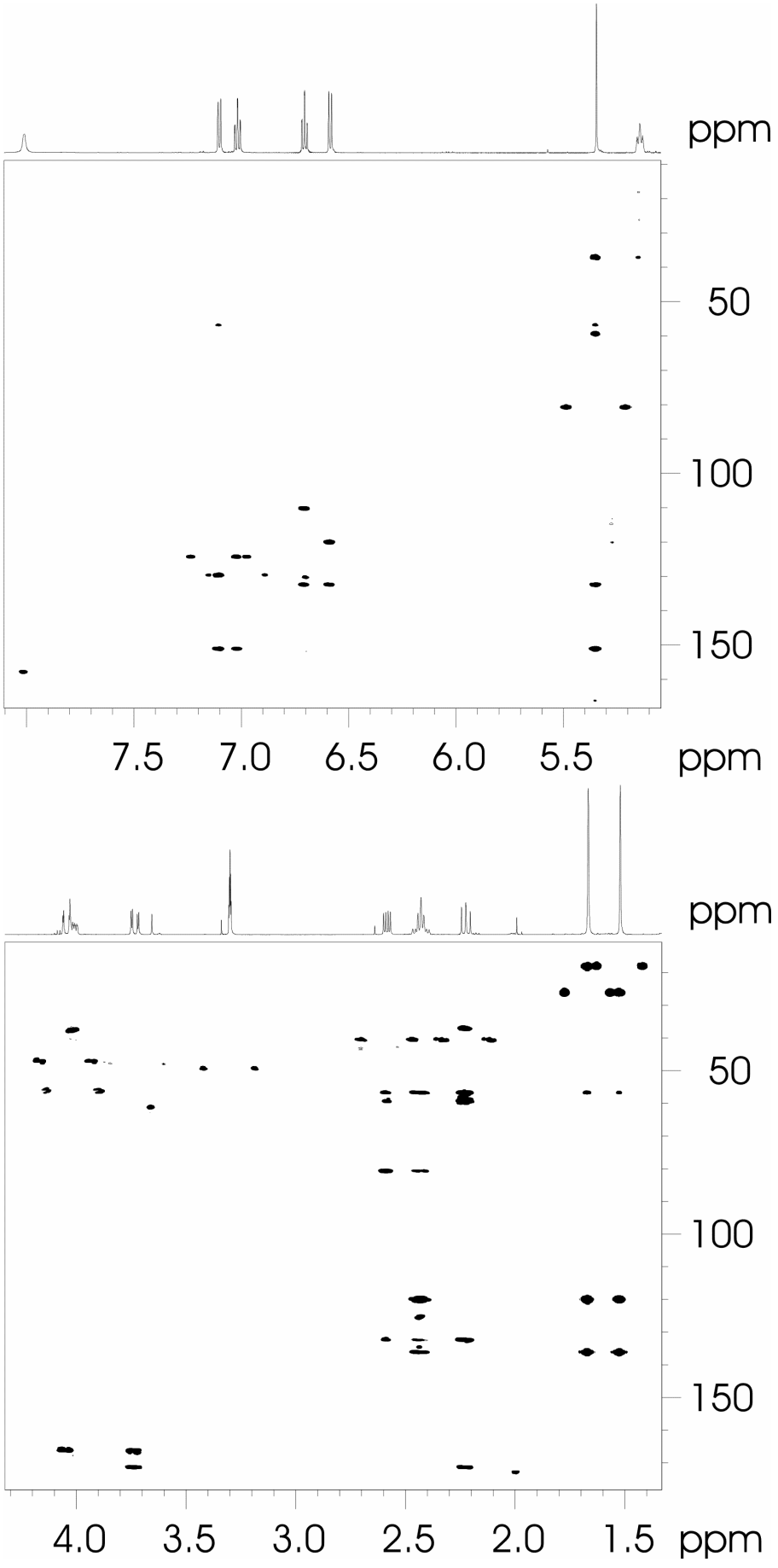

Figure S1.3 HMBC spectrum of *cyclo*-C3-dimethylallyl-L-Trp-Gly (**2c**) in CD_3OH (600 MHz)

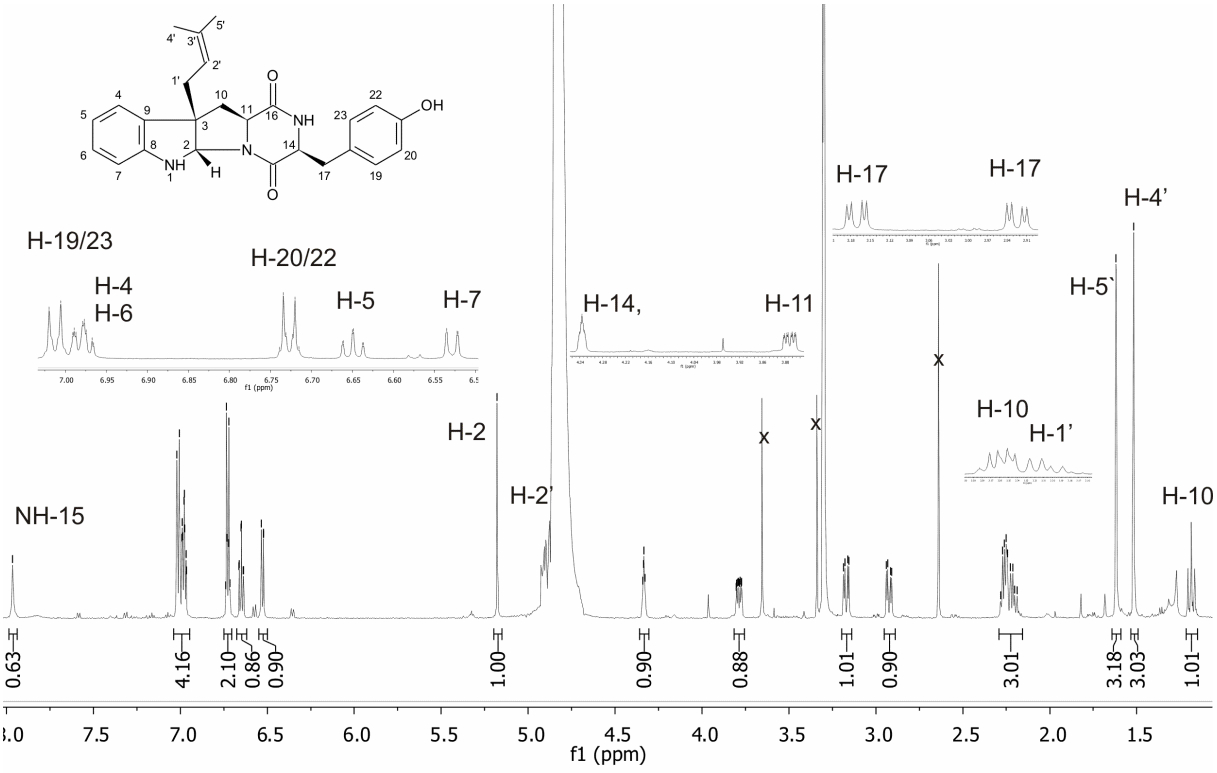

Figure S2.1 ^{1}H-NMR spectrum of *cyclo*-C3-dimethylallyl-L-Trp-L-Tyr (**5c**) in CD_3OH (600 MHz)

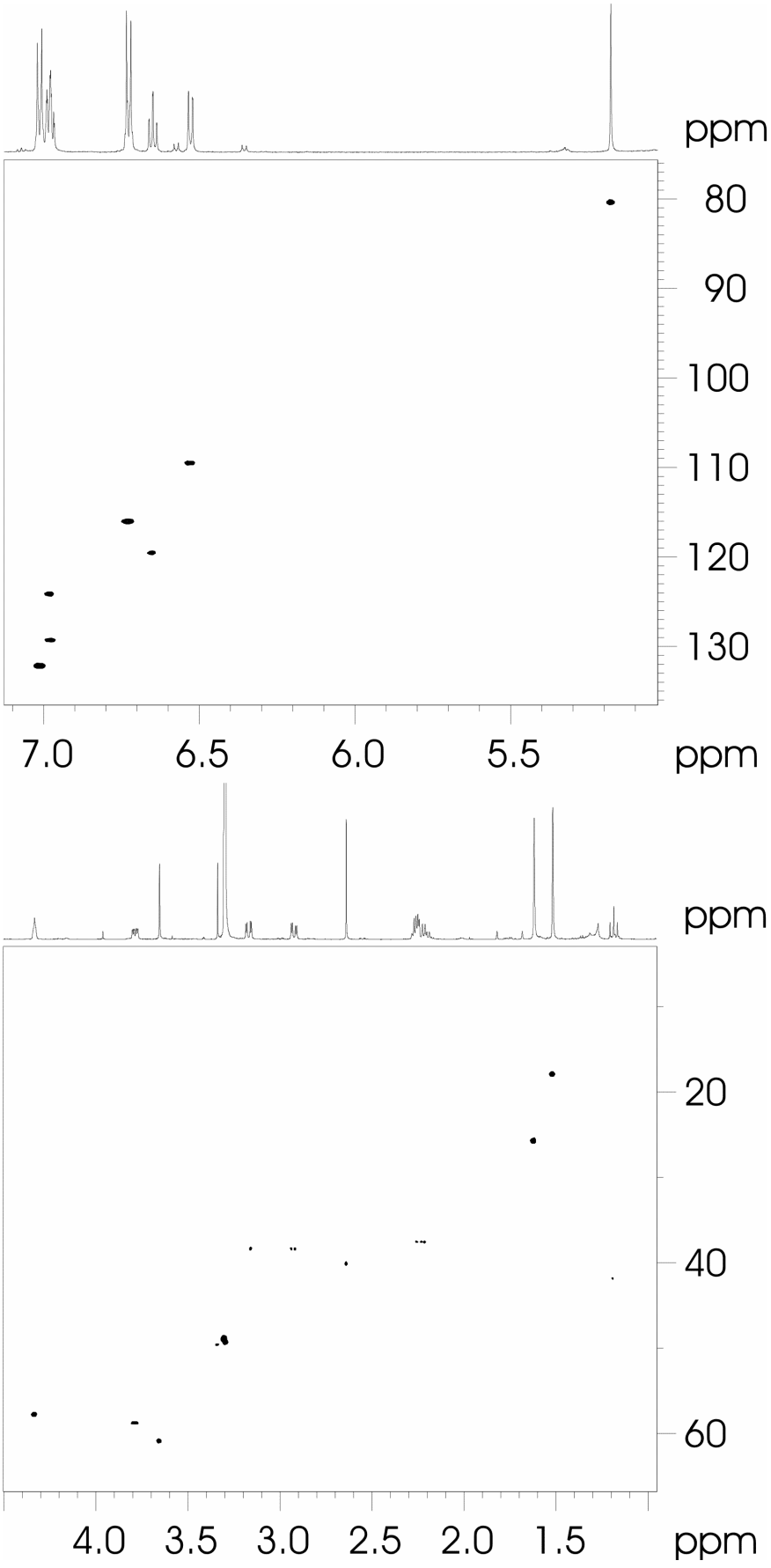

Figure S2.2 HSQC spectrum of *cyclo*-C3-dimethylallyl-L-Trp-L-Tyr (**5c**) in CD_3OH (600 MHz)

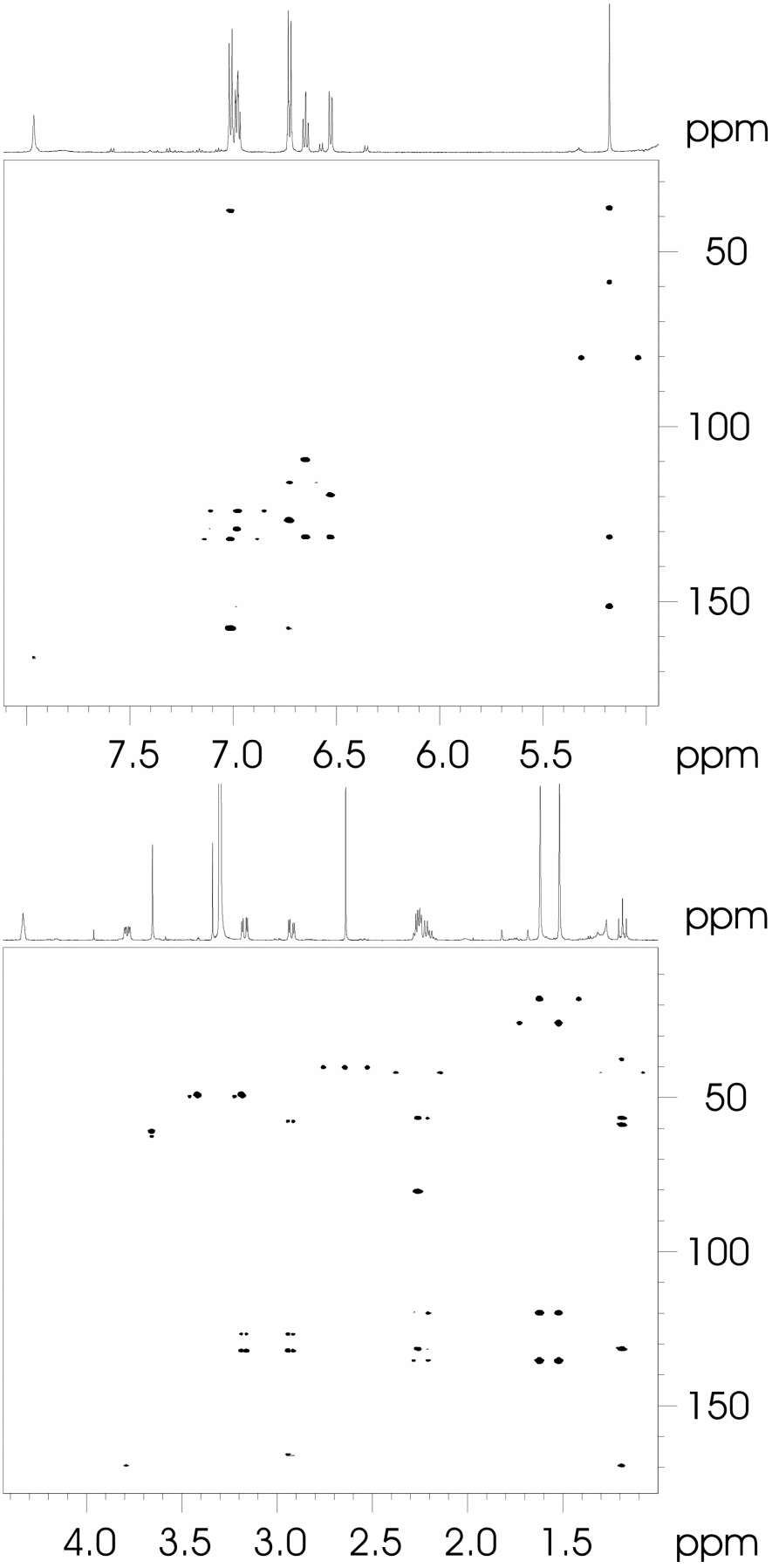

Figure S2.3 HMBC spectrum of *cyclo*-C3-dimethylallyl-L-Trp-L-Tyr (**5c**) in CD_3OH (600 MHz)

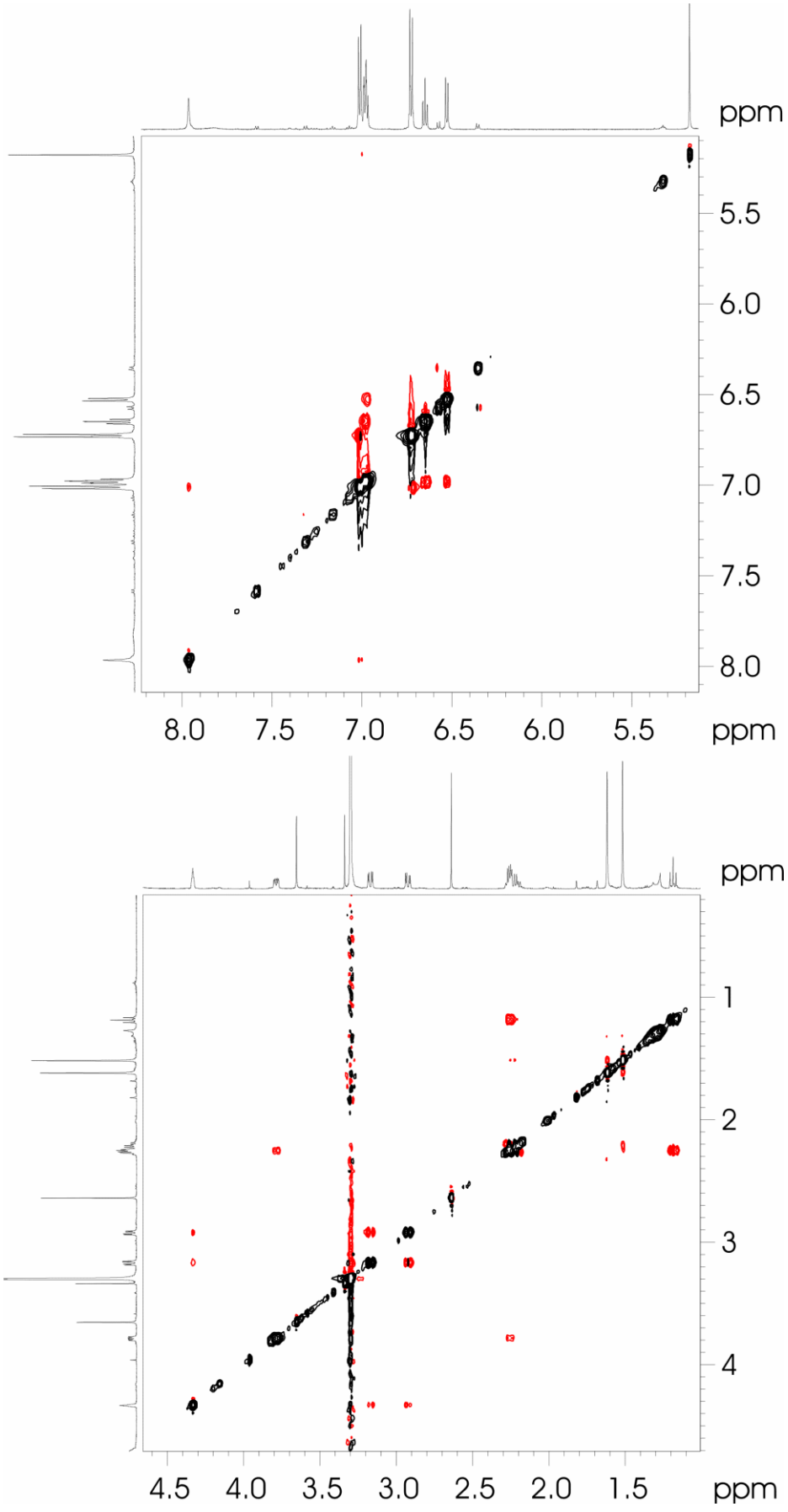

Figure S2.4 NOESY spectrum of *cyclo*-C3-dimethylallyl-L-Trp-L-Tyr (**5c**) in CD_3OH (600 MHz)

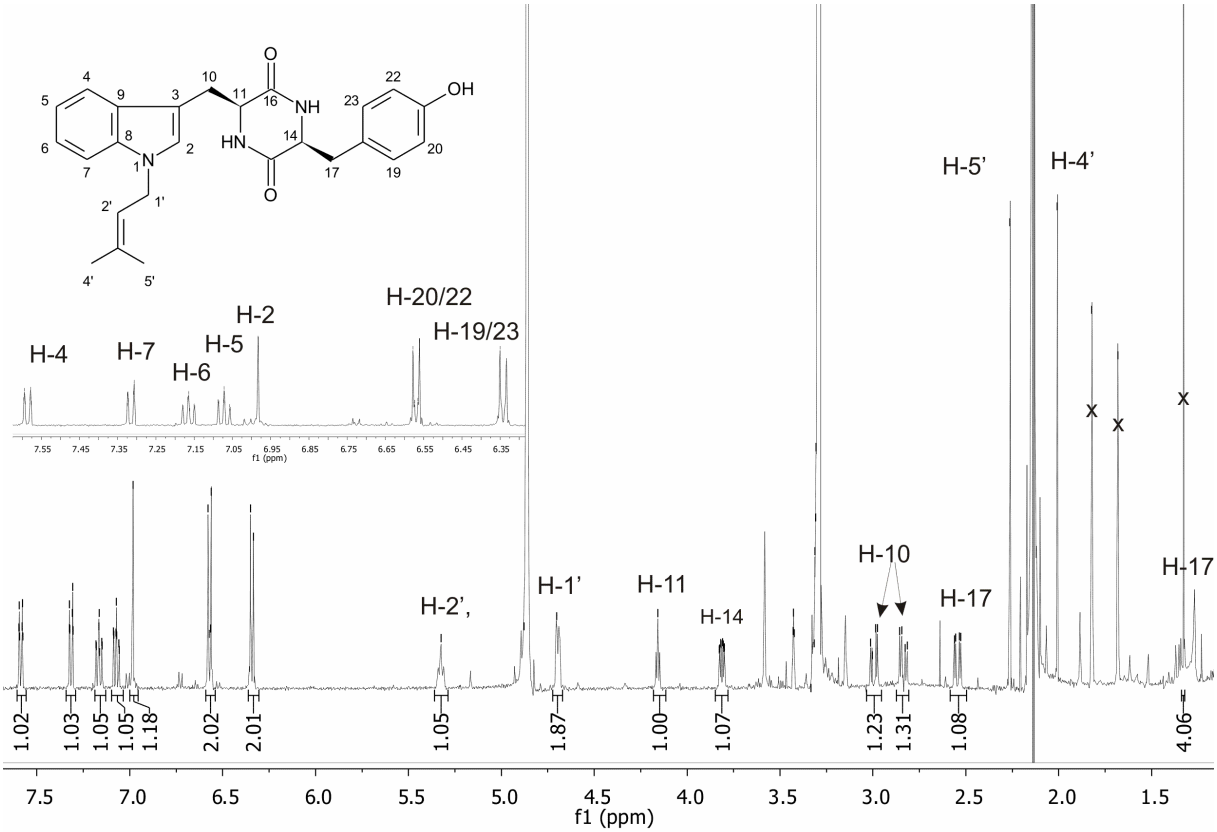

Figure S3.1 ^{1}H-NMR spectrum of *cyclo*-N1-dimethylallyl-L-Trp-L-Tyr (**5d**) in CD_3OD (500 MHz)

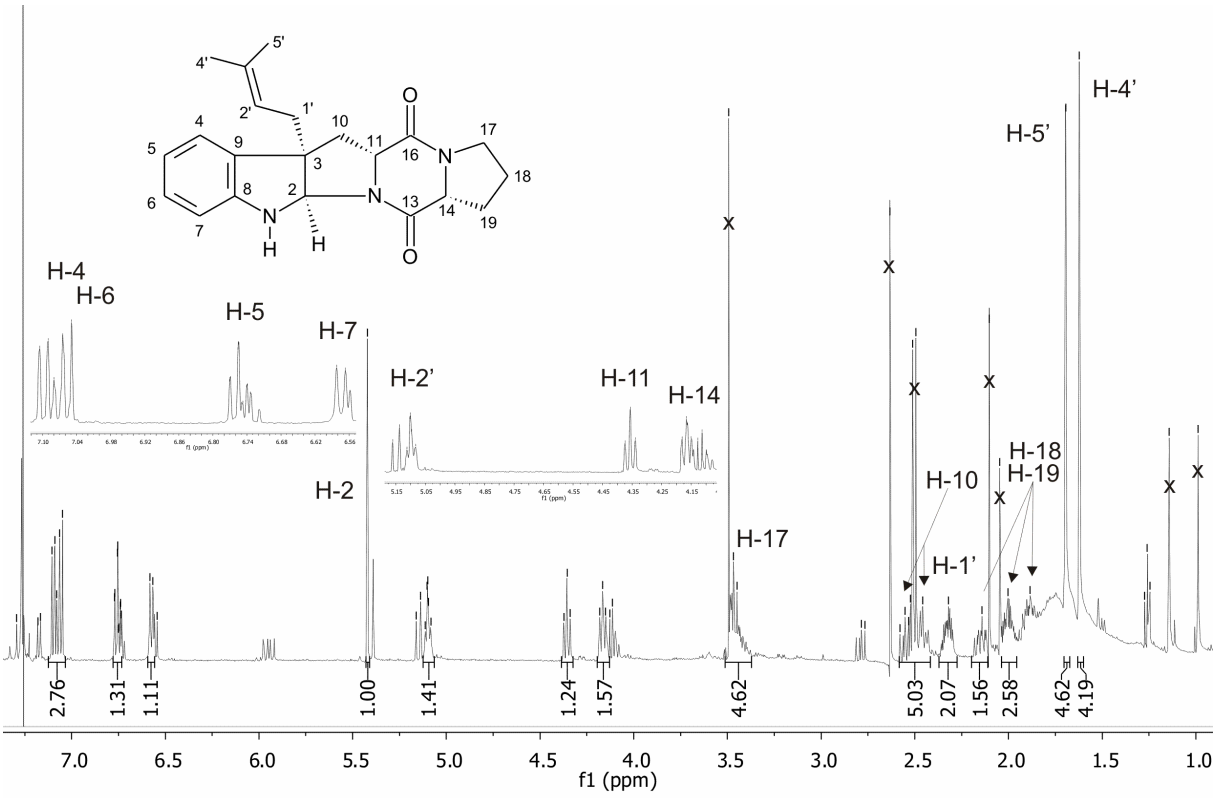

Figure S4.1 ^{1}H-NMR spectrum of *cyclo*-C3-dimethylallyl-D-Trp-D-Pro (**9c**) in $CDCl_3$ (500 MHz)

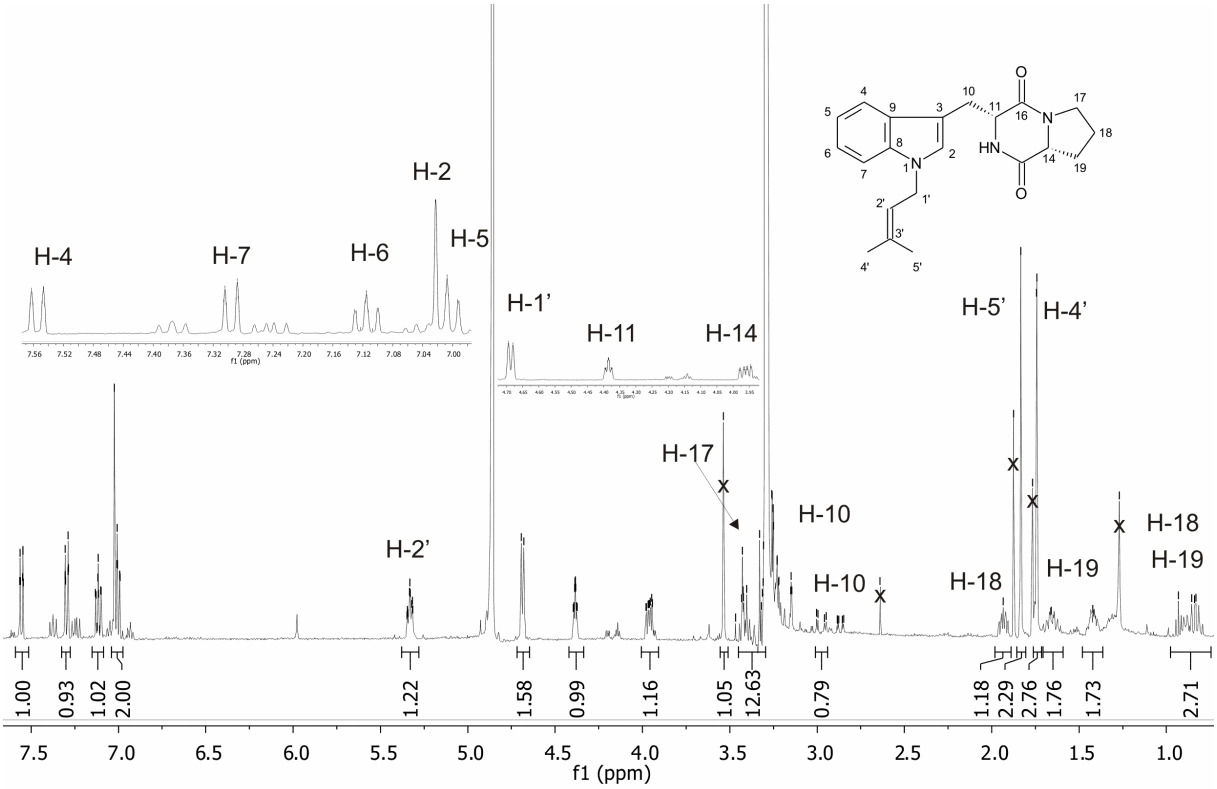

Figure S5.1 ^{1}H-NMR spectrum of *cyclo*-N1-dimethylallyl-D-Trp-D-Pro (**9d**) in CD_3OD (500 MHz)

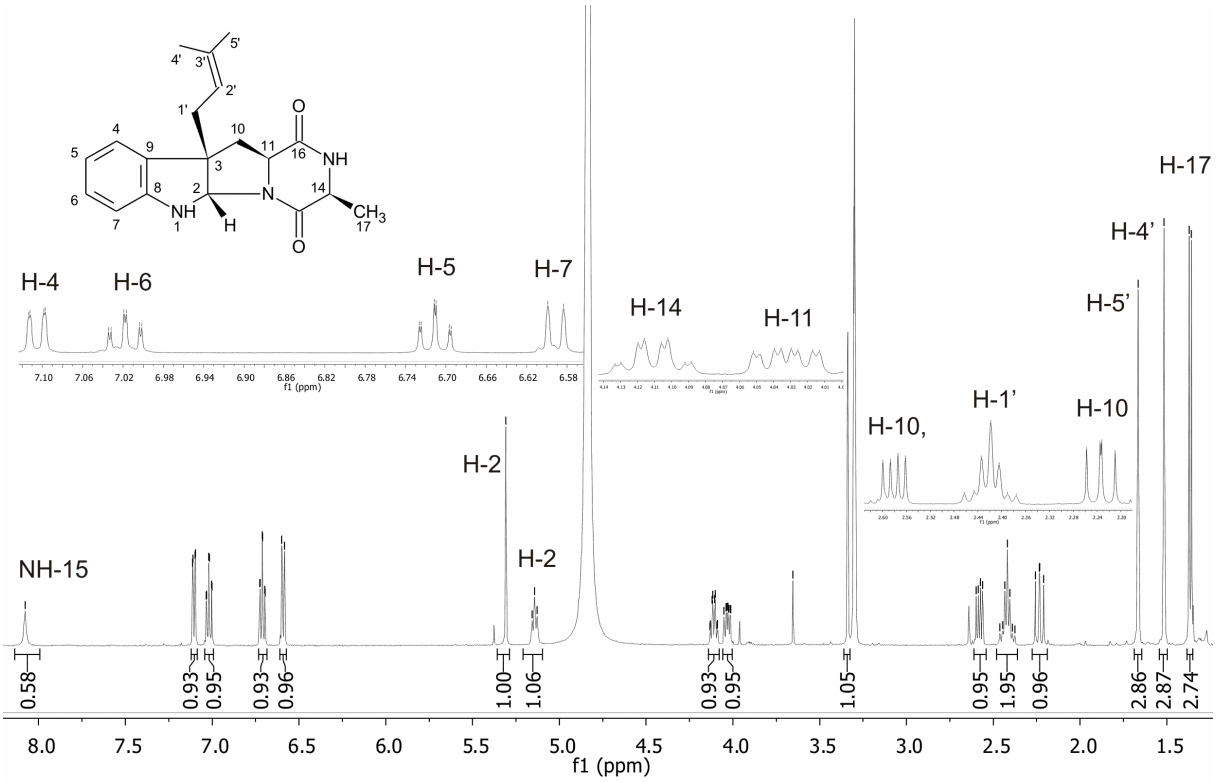

Figure S6.1 ^{1}H-NMR spectrum of *cyclo*-C3-dimethylallyl-L-Trp-L-Ala (**10c**) in CD_3OH (600 MHz)

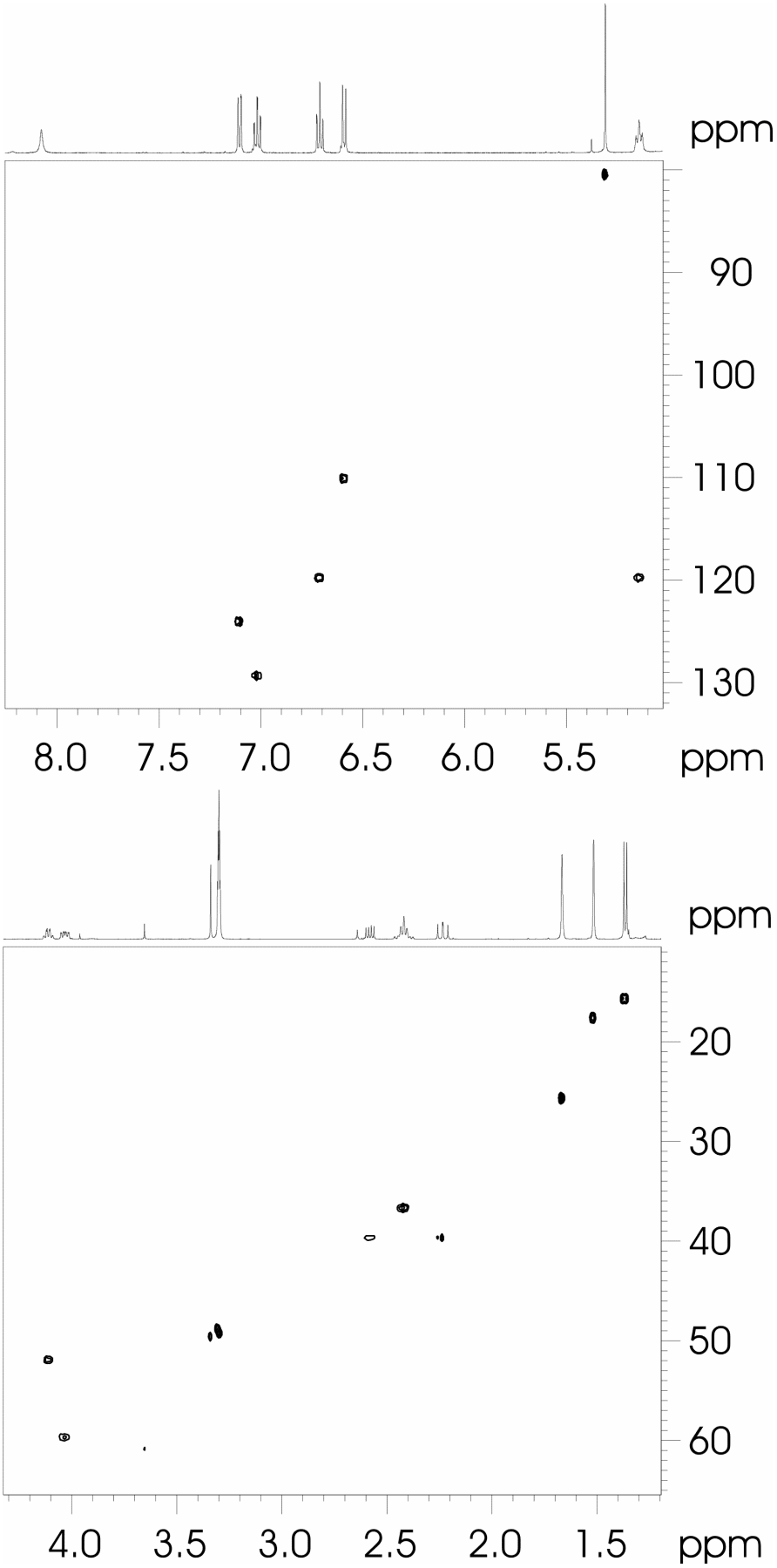

Figure S6.2 HSQC spectrum of *cyclo*-C3-dimethylallyl-L-Trp-L-Ala (**10c**) in CD_3OH (600MHz)

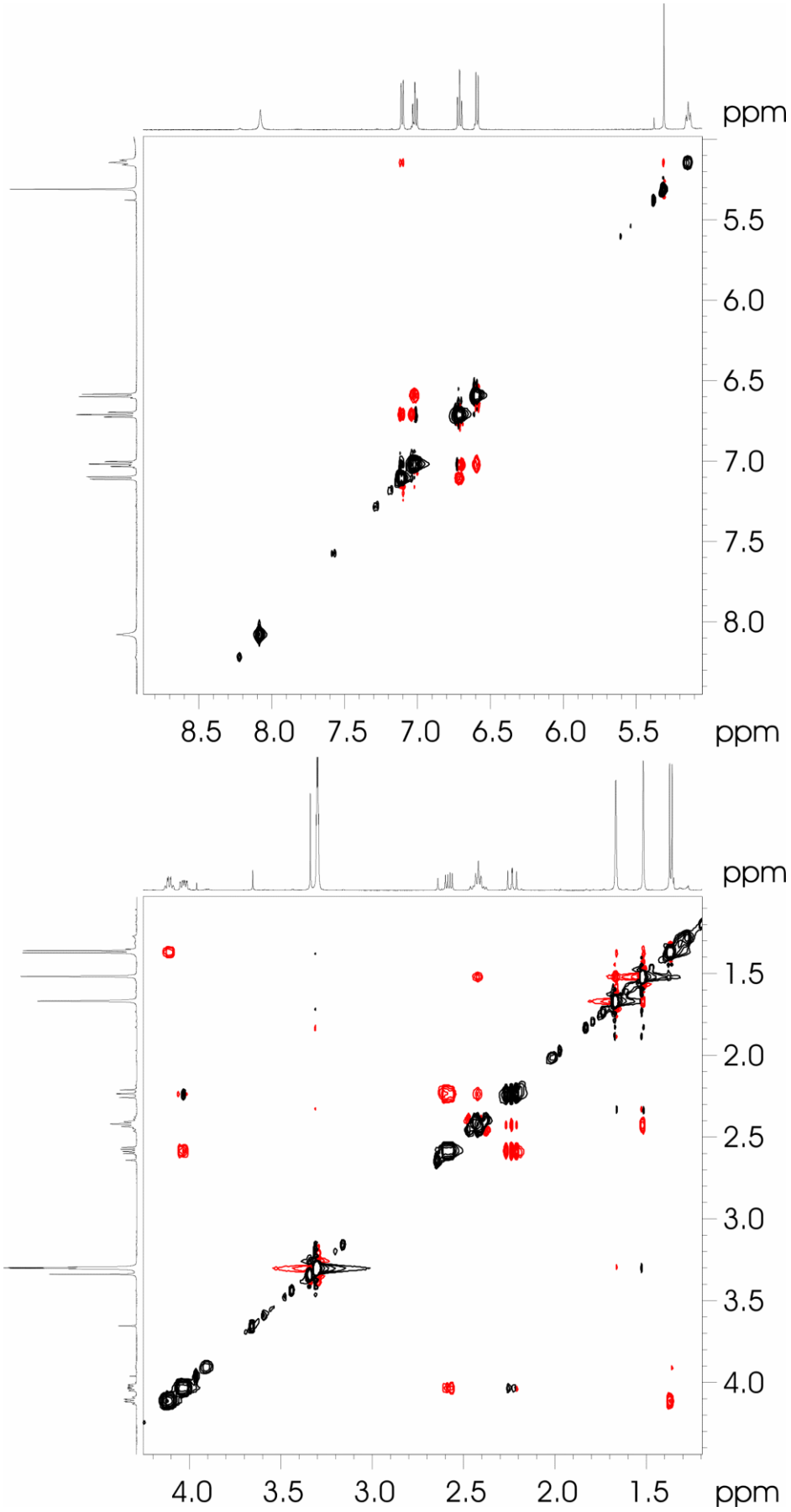

Figure S6.3 NOESY spectrum of *cyclo*-C3-dimethylallyl-L-Trp-L-Ala (**10c**) in CD_3OH (600MHz)

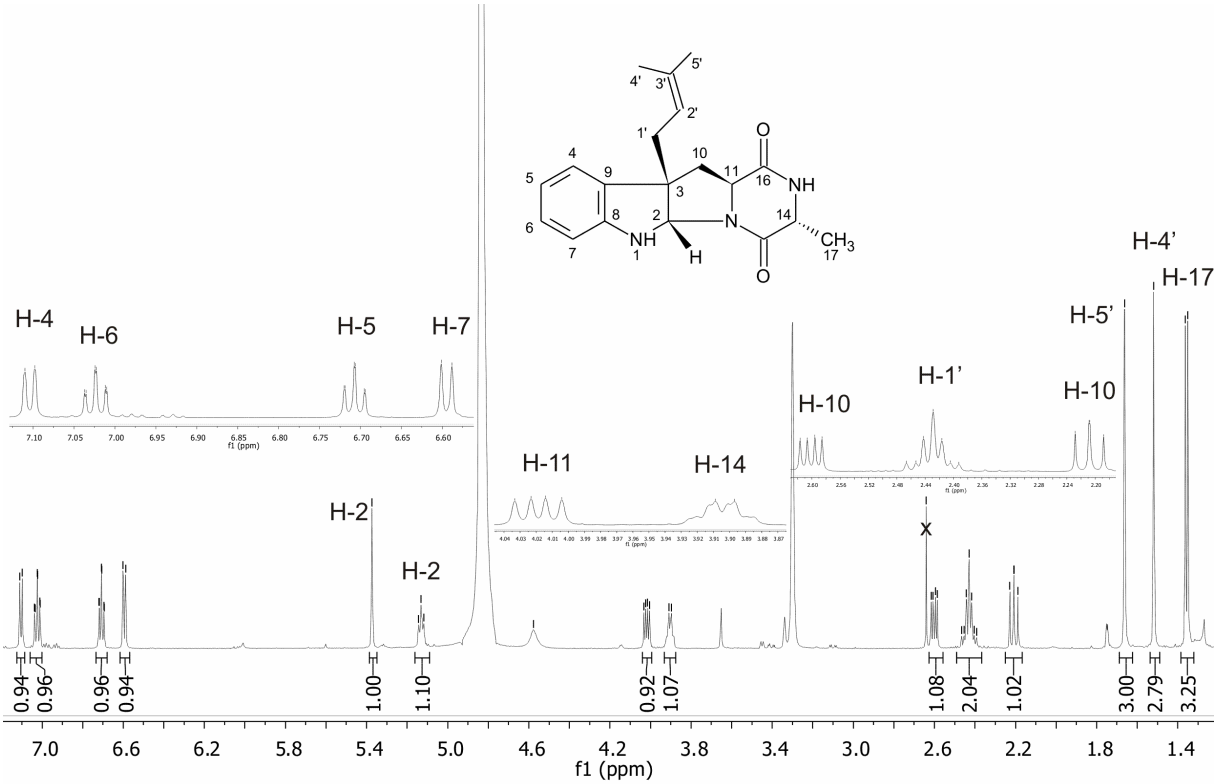

Figure S7.1 ^{1}H-NMR spectrum of *cyclo*-C3-dimethylallyl-L-Trp-D-Ala (**11c**) in CD_3OH (600 MHz)

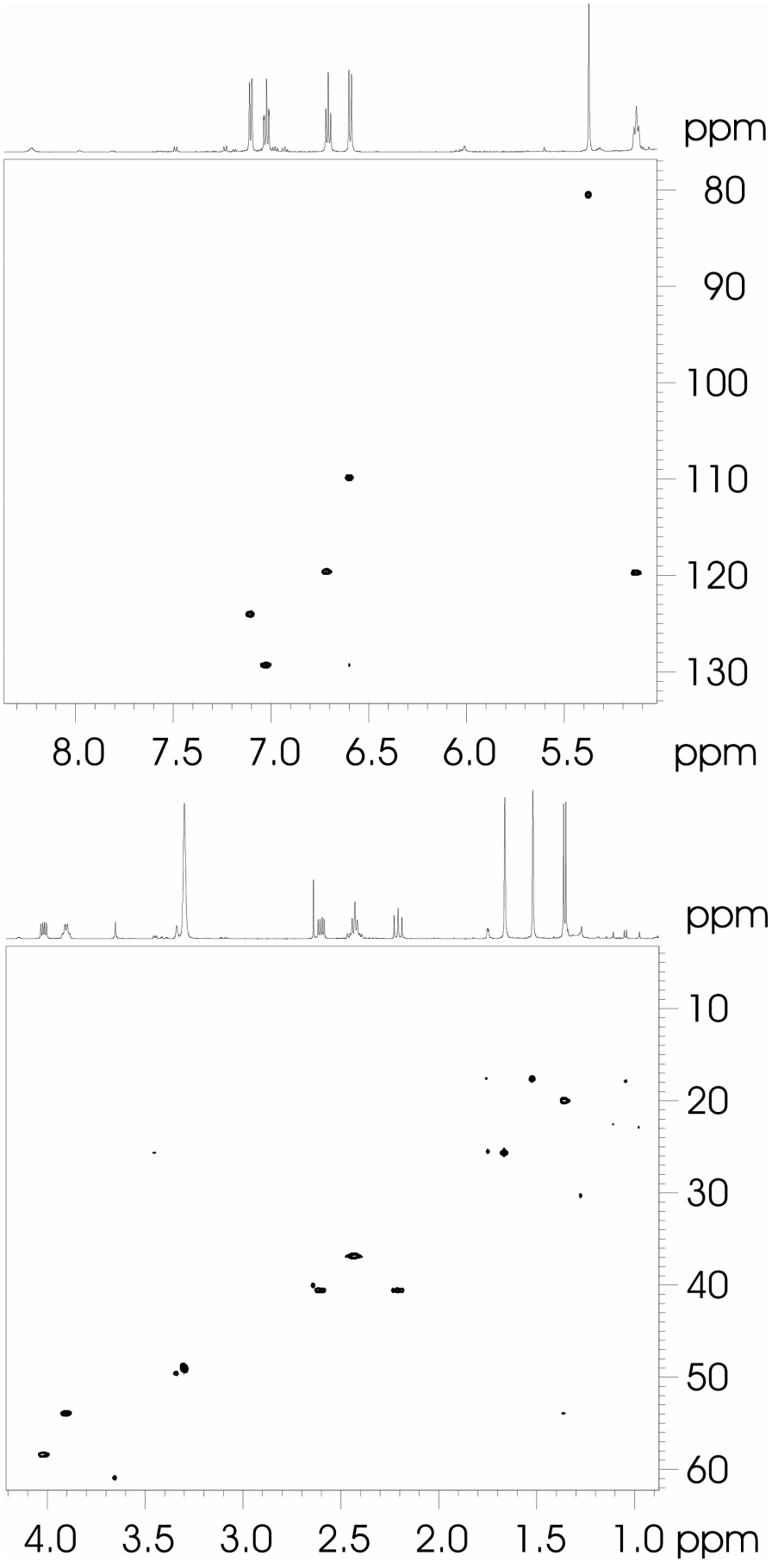

Figure S7.2 HSQC spectrum of *cyclo*-C3-dimethylallyl-L-Trp-D-Ala (**11c**) in CD_3OH (600 MHz)

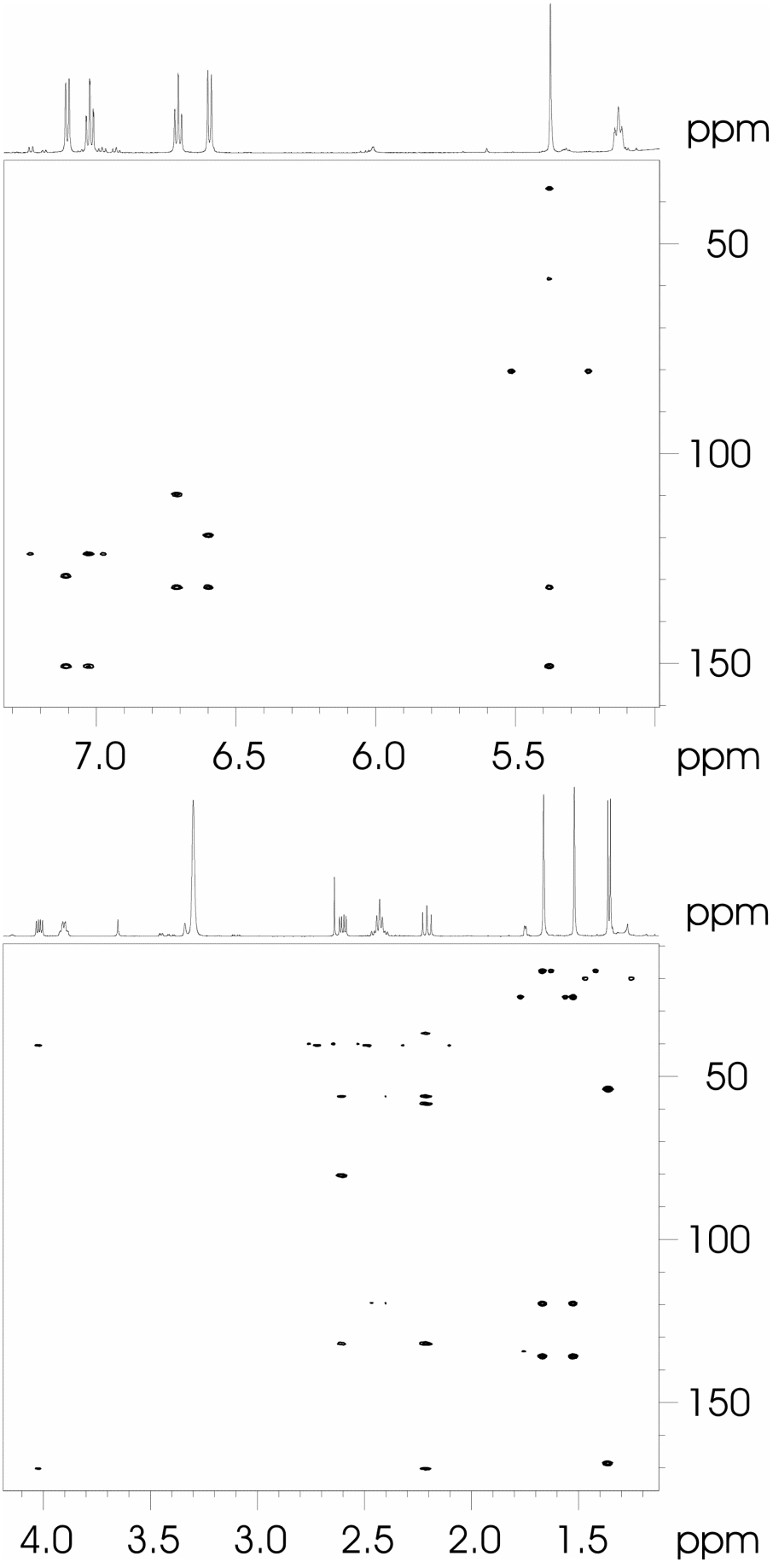

Figure S7.3 HMBC spectrum of *cyclo*-C3-dimethylallyl-L-Trp-D-Ala (**11c**) in CD_3OH (600 MHz)

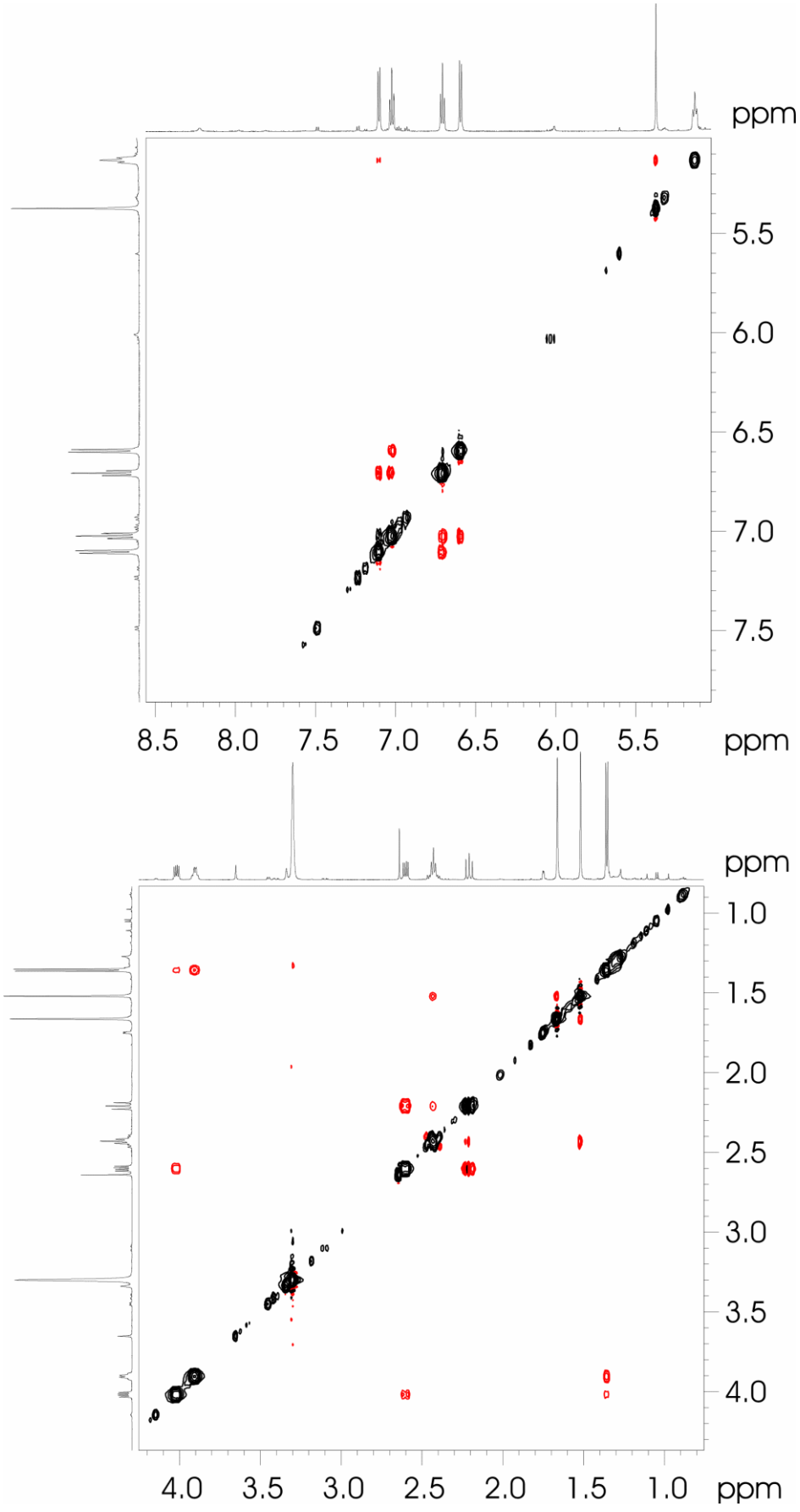

Figure S37.4 NOESY spectrum of *cyclo*-C3-dimethylallyl-L-Trp-D-Ala (**11c**) in CD_3OH (600 MHz)

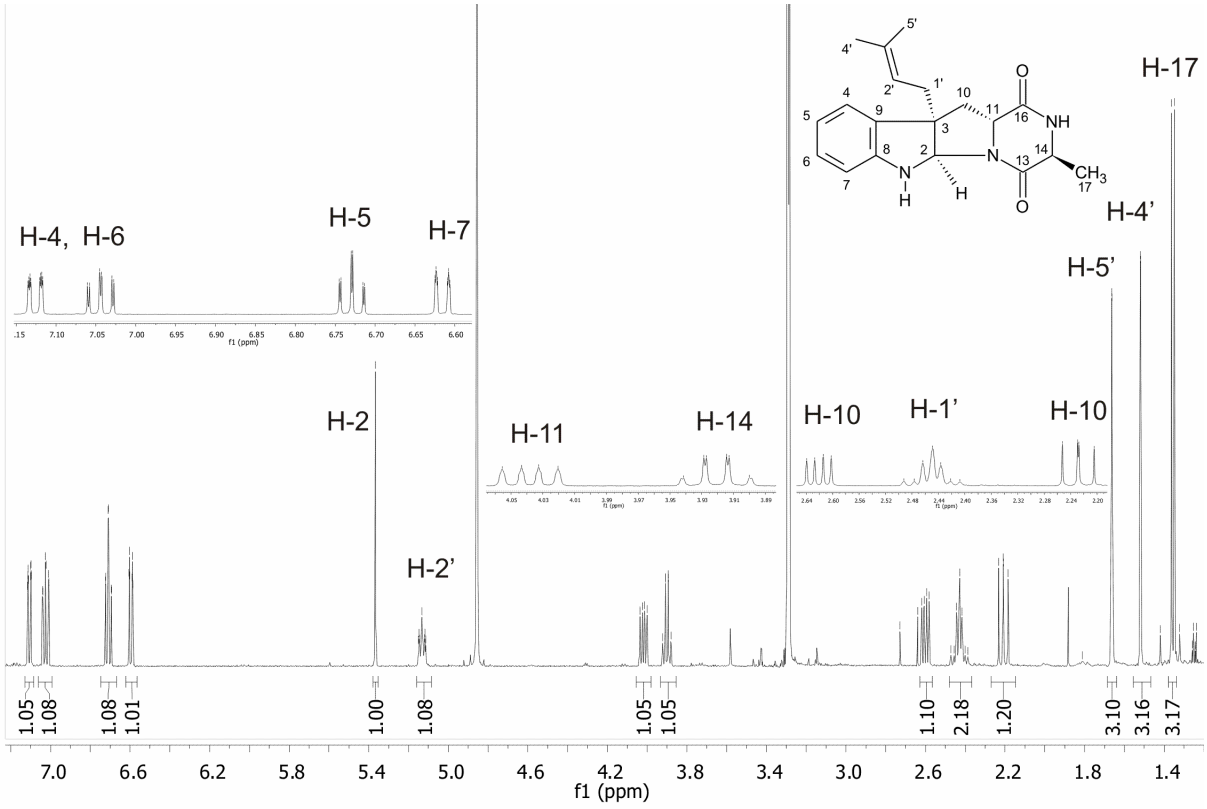

Figure S8.1 ^{1}H-NMR spectrum of *cyclo*-C3-dimethylallyl-D-Trp-L-Ala (**13c**) in CD_3OD (500 MHz)

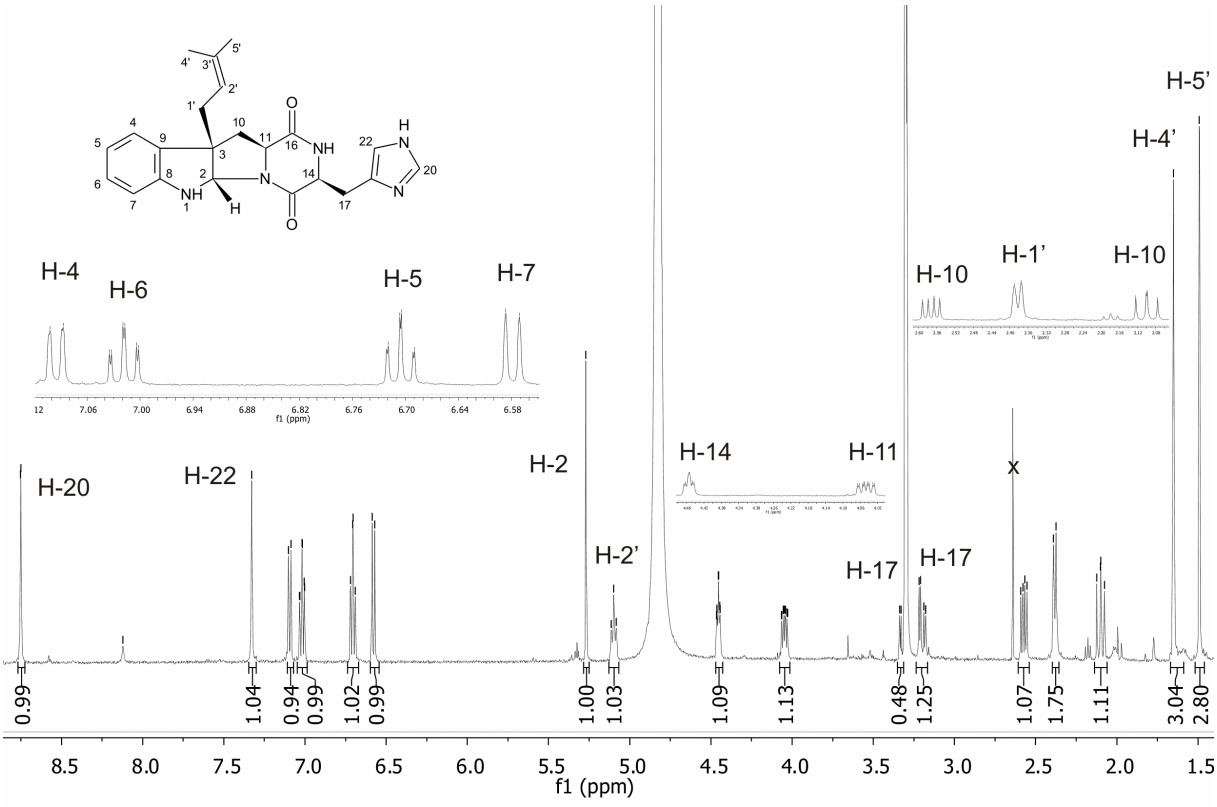

Figure S9.1 ^{1}H-NMR spectrum of *cyclo*-C3-dimethylallyl-L-Trp-L-His (**14c**) in CD_3OH (600 MHz)

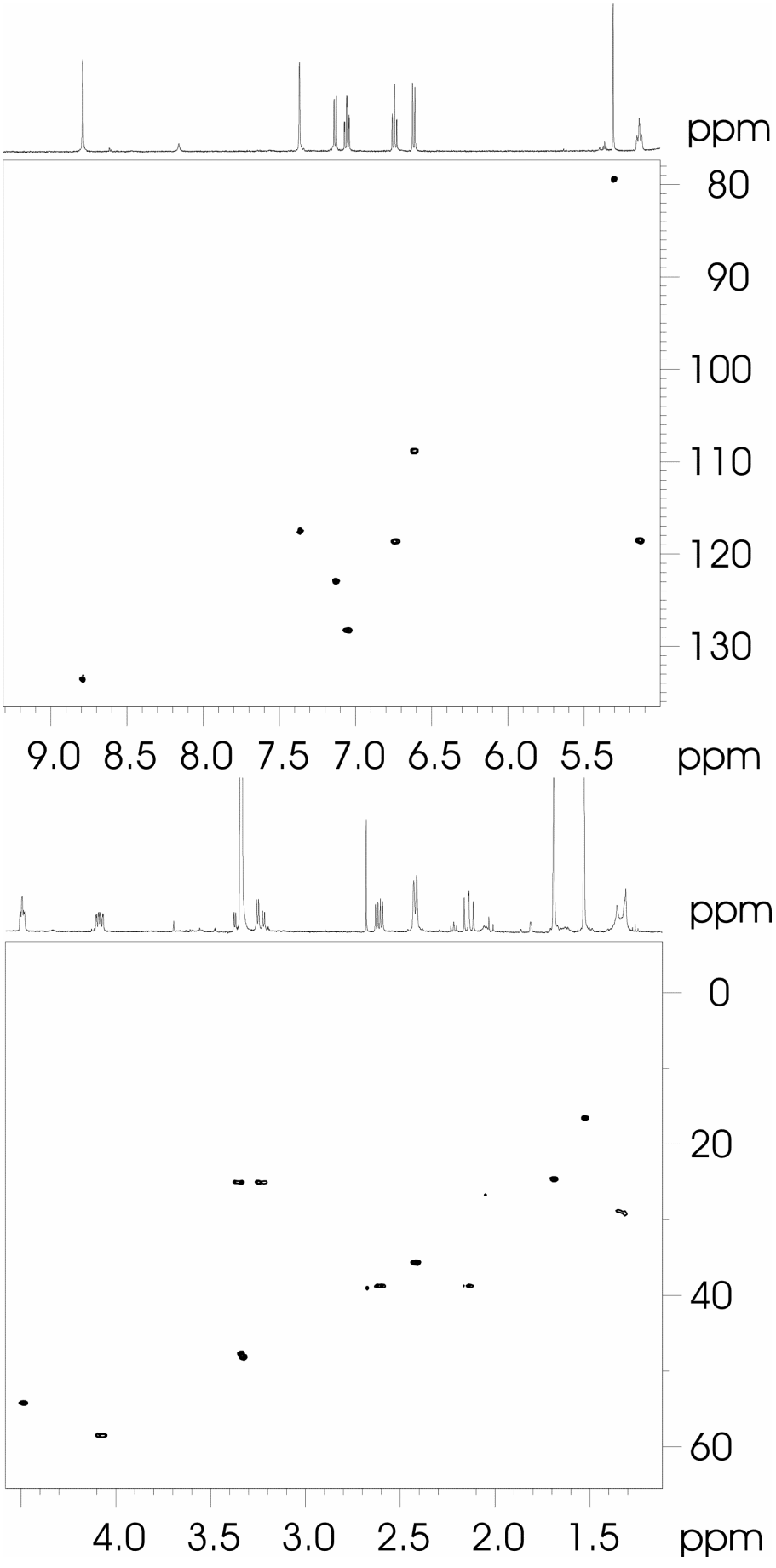

Figure S9.2 HSQC spectrum of *cyclo*-C3-dimethylallyl-L-Trp-L-His (**14c**) in CD_3OH (600 MHz)

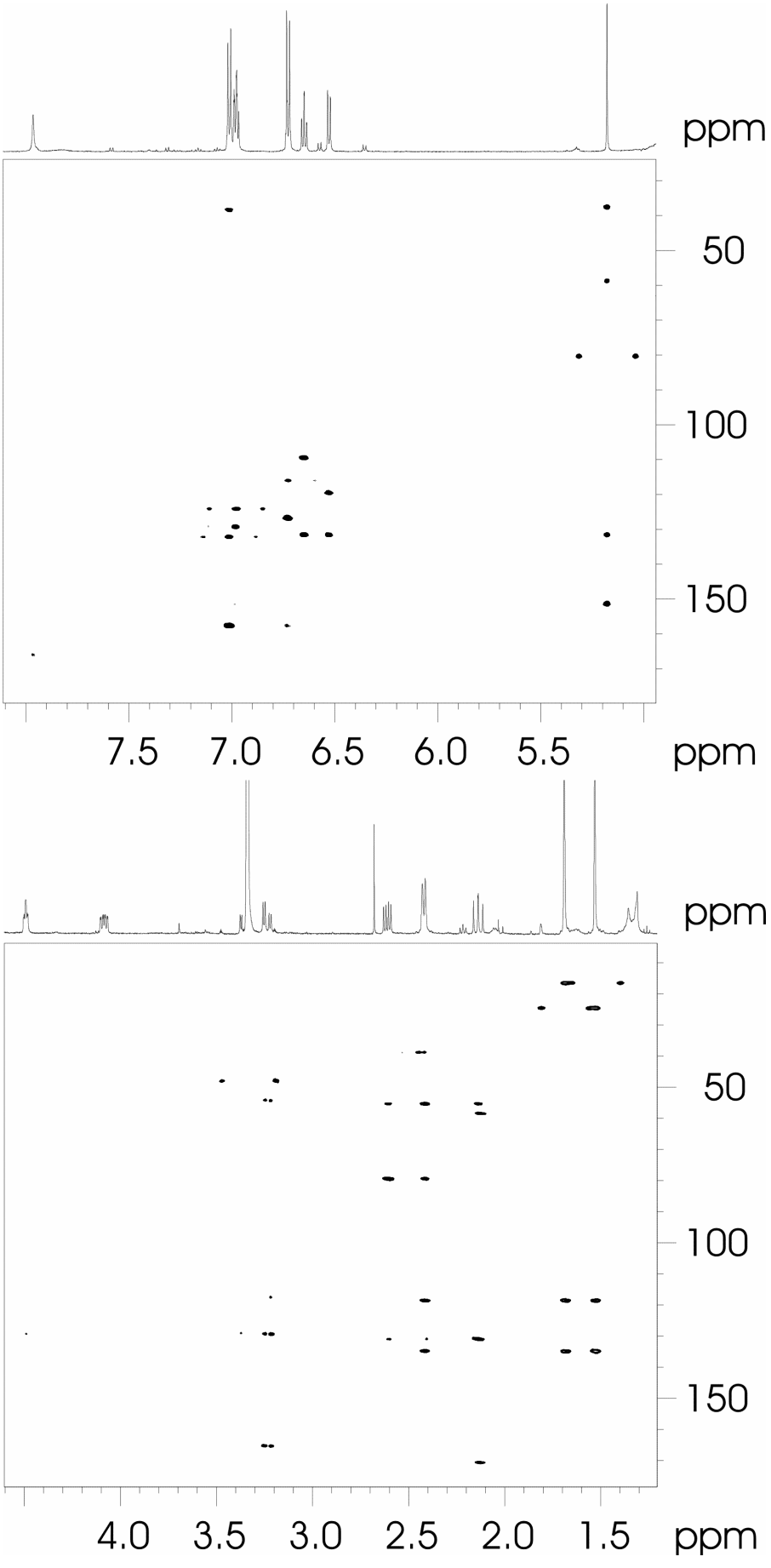

Figure S9.3 HMBC spectrum of *cyclo*-C3-dimethylallyl-L-Trp-L-His (**14c**) in CD_3OH (600 MHz)

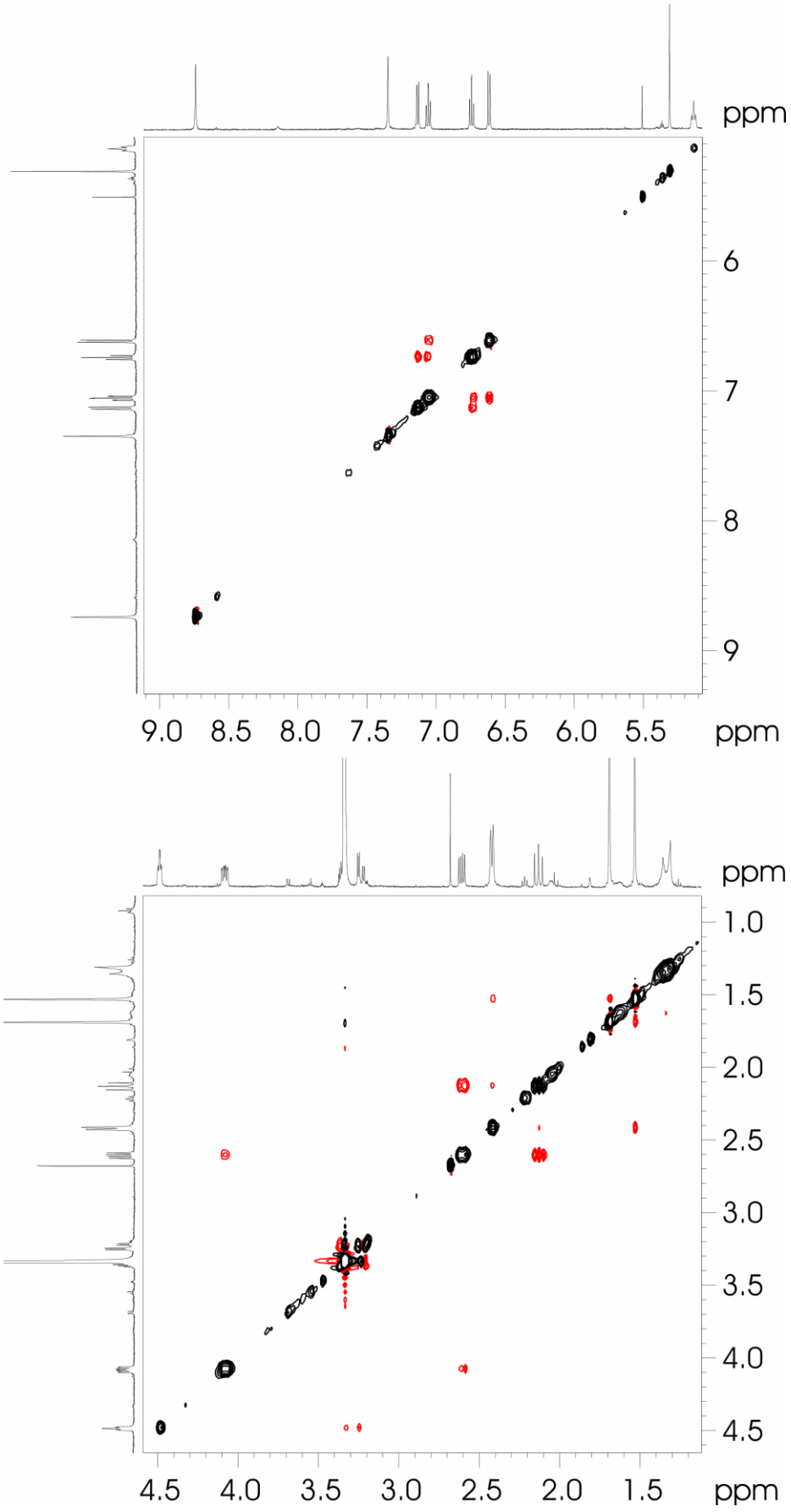

Figure S9.4 NOESY spectrum of *cyclo*-C3-dimethylallyl-L-Trp-L-His (**14c**) in CD_3OH (600 MHz)

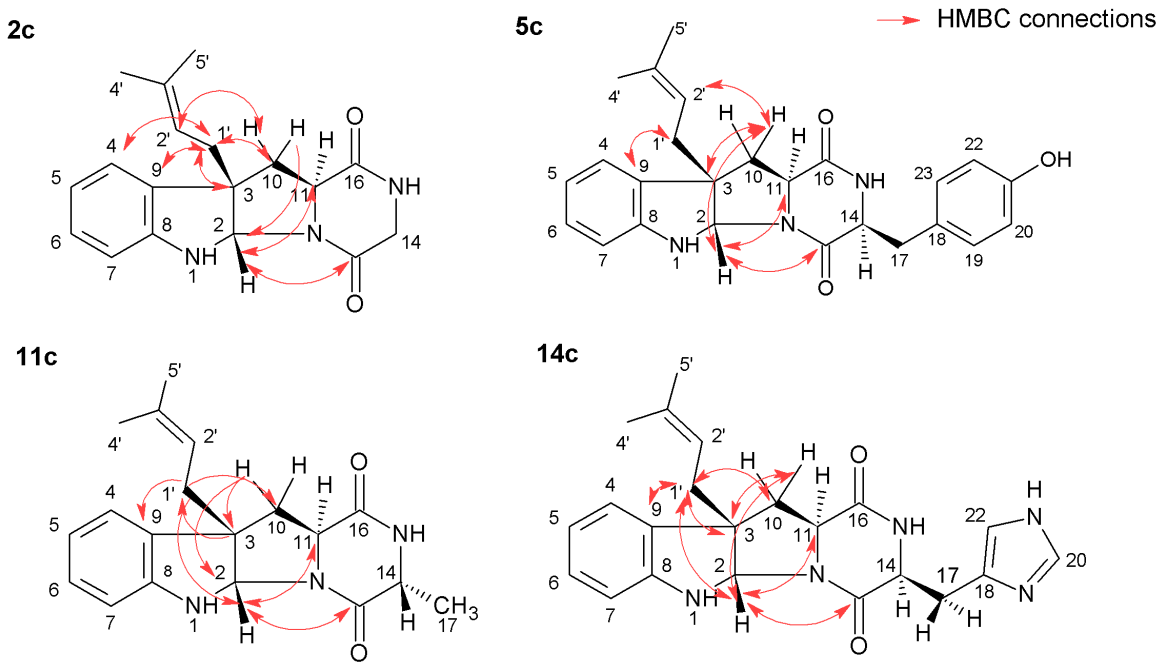

Figure S10 HMBC correlations of **2c**, **5c**, **11c**and **14c**

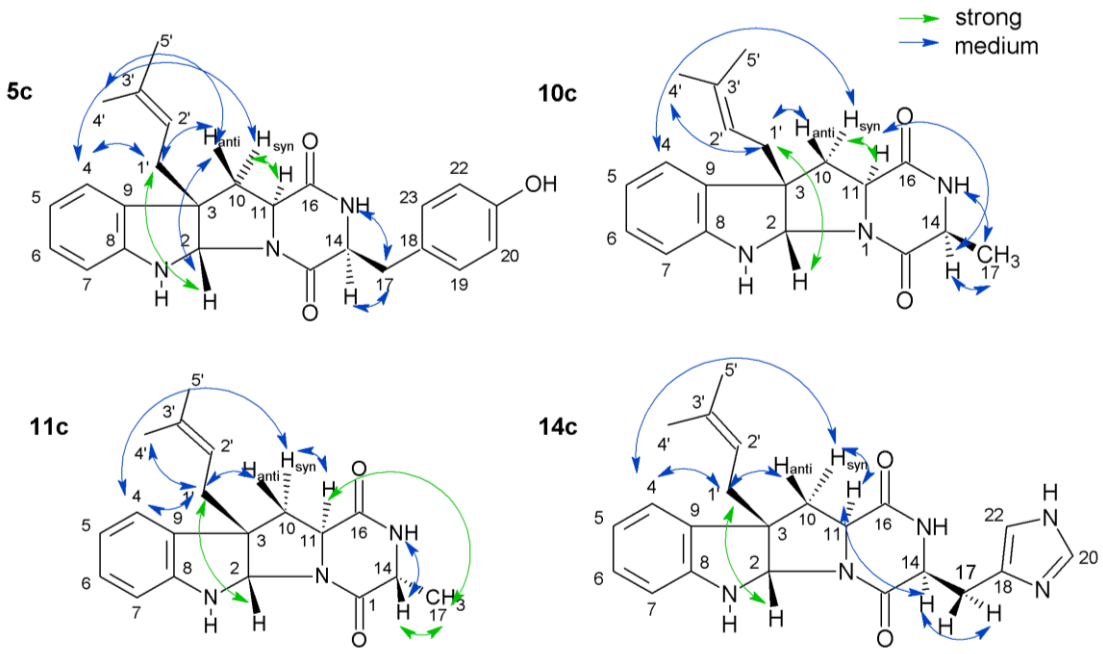

Figure S11 NOESY correlations of **5c**, **10c**, **11c** and **14c**

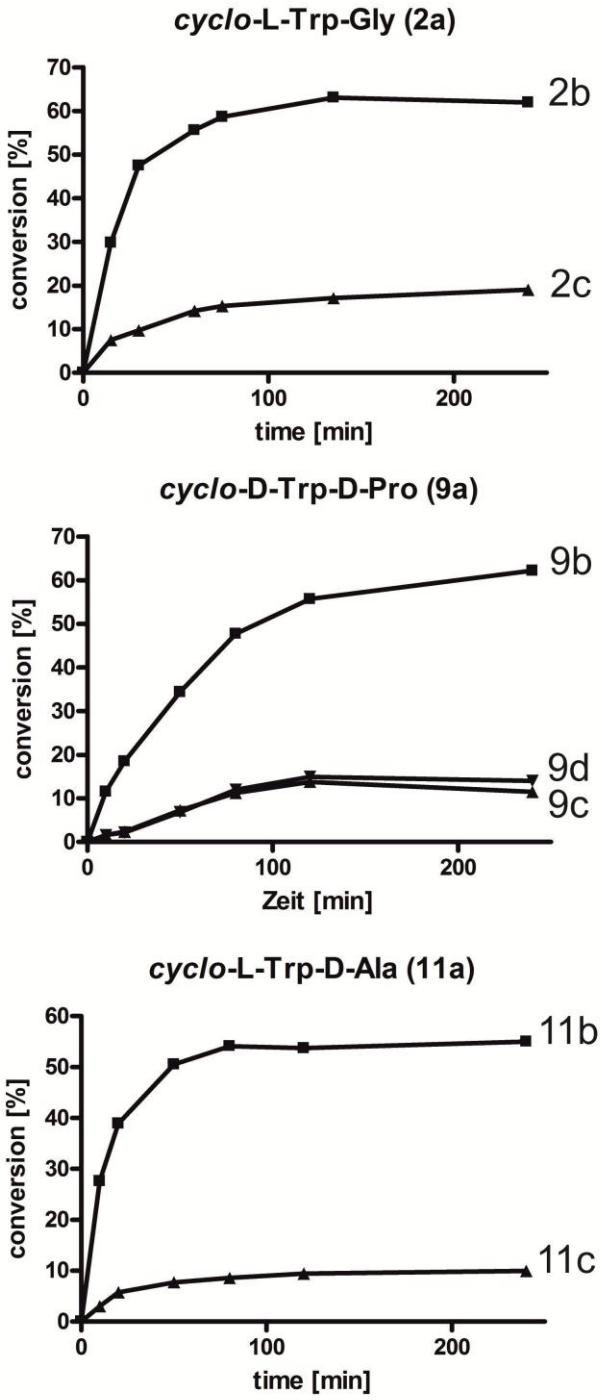

Figure S12 Time dependence of the product formation in the incubation mixtures of FtmPT1 with **2a**, **9a** and **11a**. The reaction mixtures contained 5 µg FtmPT1, 10 mM $CaCl_2$, 2 mM DMAPP and 1 mM cyclic dipeptide and were incubated at 37 °C. The substances were detected with a Diode Array detector and illustrated for absorption at 296 nm.

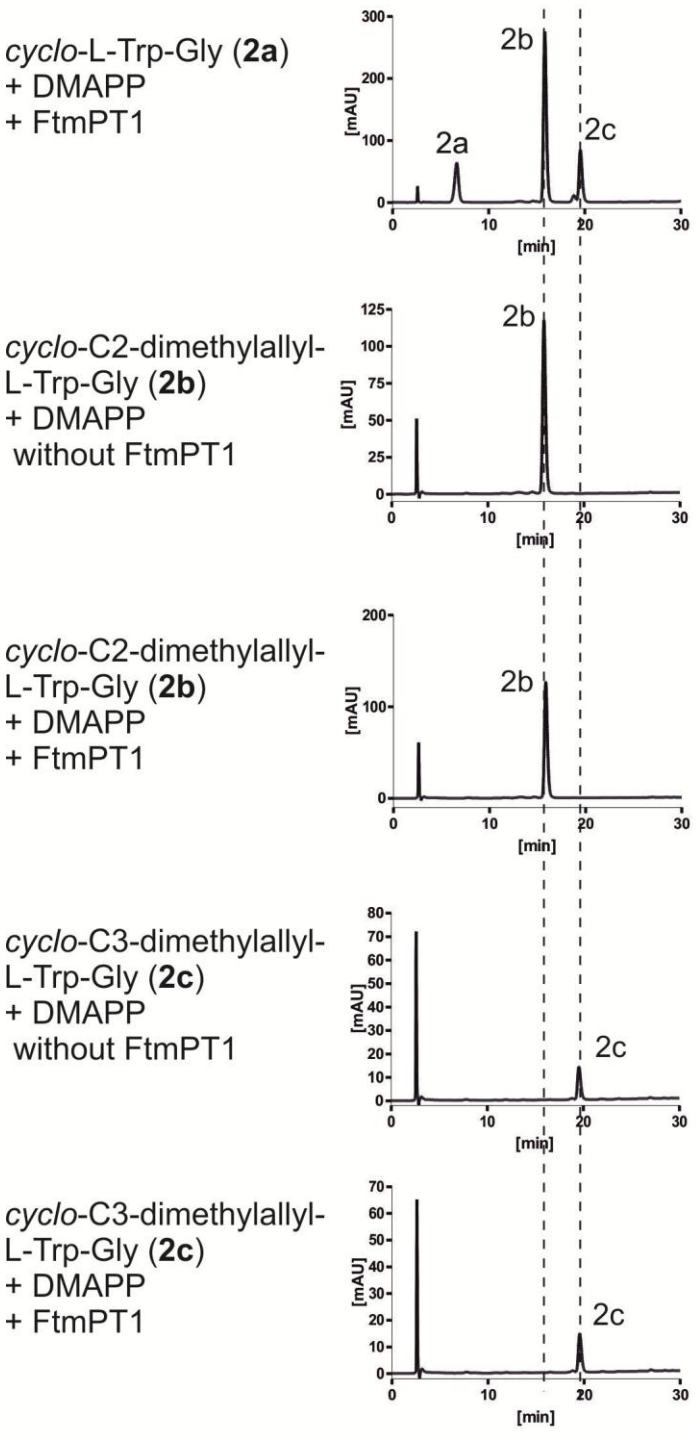

Figure S13 HPLC chromatograms of incubation mixtures of **2a**, **2b** or **2c** in the absence and presence of 5 µg FtmPT1. The reaction mixtures contained 10 mM $CaCl_2$, 1 mM aromatic substrate and 2 mM DMAPP and were incubated at 37°C for 2 h. The substances were detected with a Photo Diode Array detector and illustrated for absorption at 296 nm.

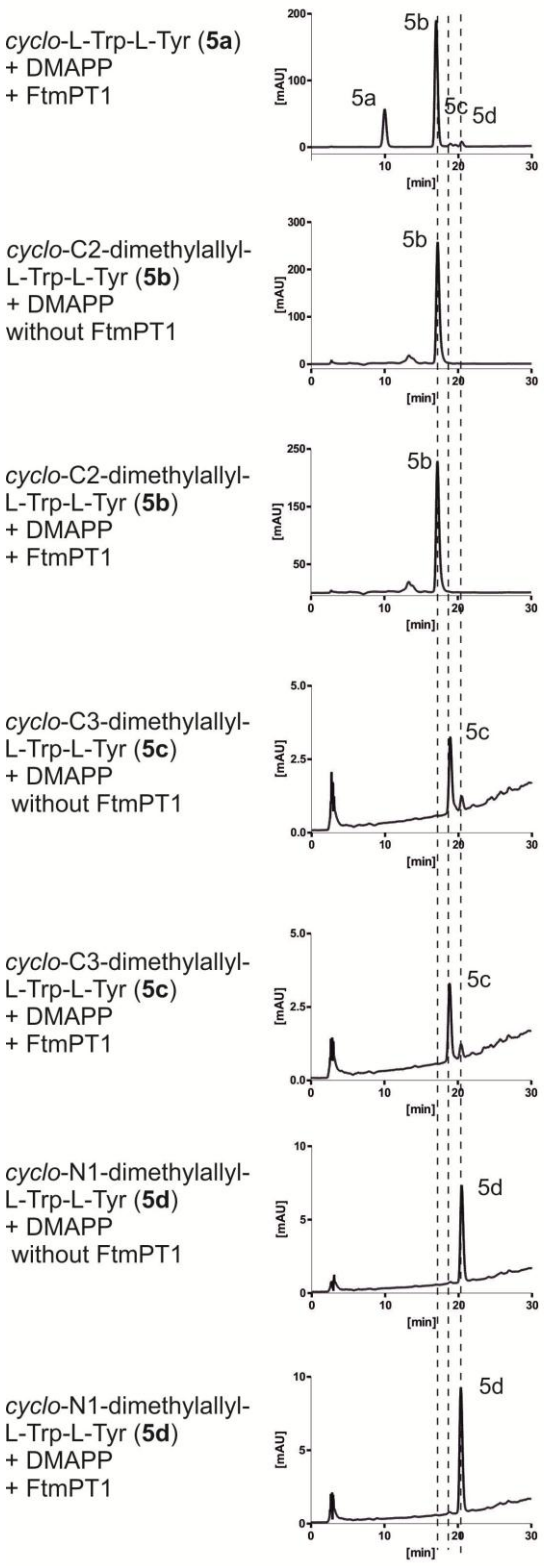

Figure S14 HPLC chromatograms of incubation mixtures with **5a**, **5b**, **5c** or **5d** in the absence and presence of 5 µg FtmPT1. The reaction mixtures contained 10 mM $CaCl_2$, 1 mM aromatic substrate and 2 mM DMAPP and were incubated at 37°C for 2 h. The substances were detected with a Photo Diode Array detector and illustrated for absorption at 296 nm.

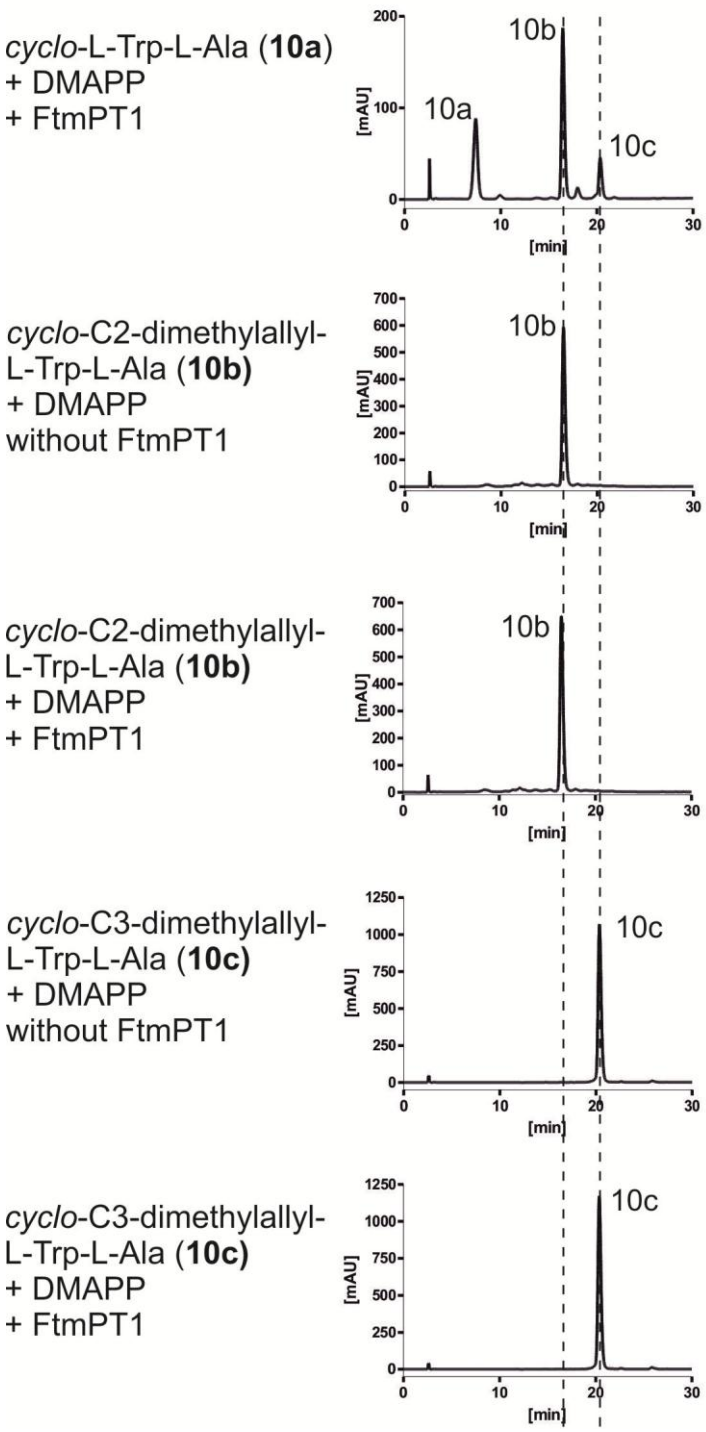

Figure S15 HPLC chromatograms of incubation mixtures with **10a**, **10b** or **10c** in the absence and presence of 5 µg FtmPT1. The reaction mixtures contained 10 mM $CaCl_2$, 1 mM aromatic substrate and 2 mM DMAPP and were incubated at 37°C for 2 h. The substances were detected with a Photo Diode Array detector and illustrated for absorption at 296 nm.

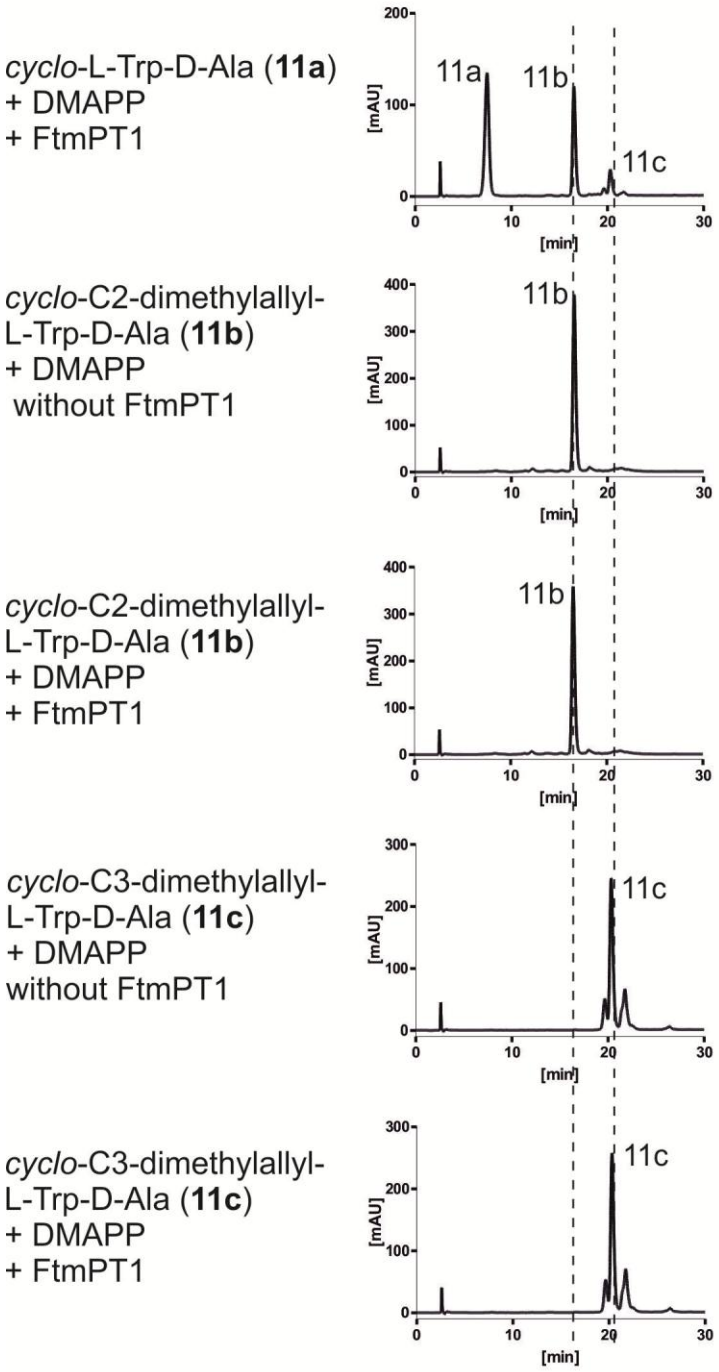

Figure S16 HPLC chromatograms of incubation mixtures with **11a**, **11b** or **11c** in the absence and presence of 5 µg FtmPT1. The reaction mixtures contained 10 mM $CaCl_2$, 1 mM aromatic substrate and 2 mM DMAPP and were incubated at 37°C for 2 h. The substances were detected with a Photo Diode Array detector and illustrated for absorption at 296 nm.

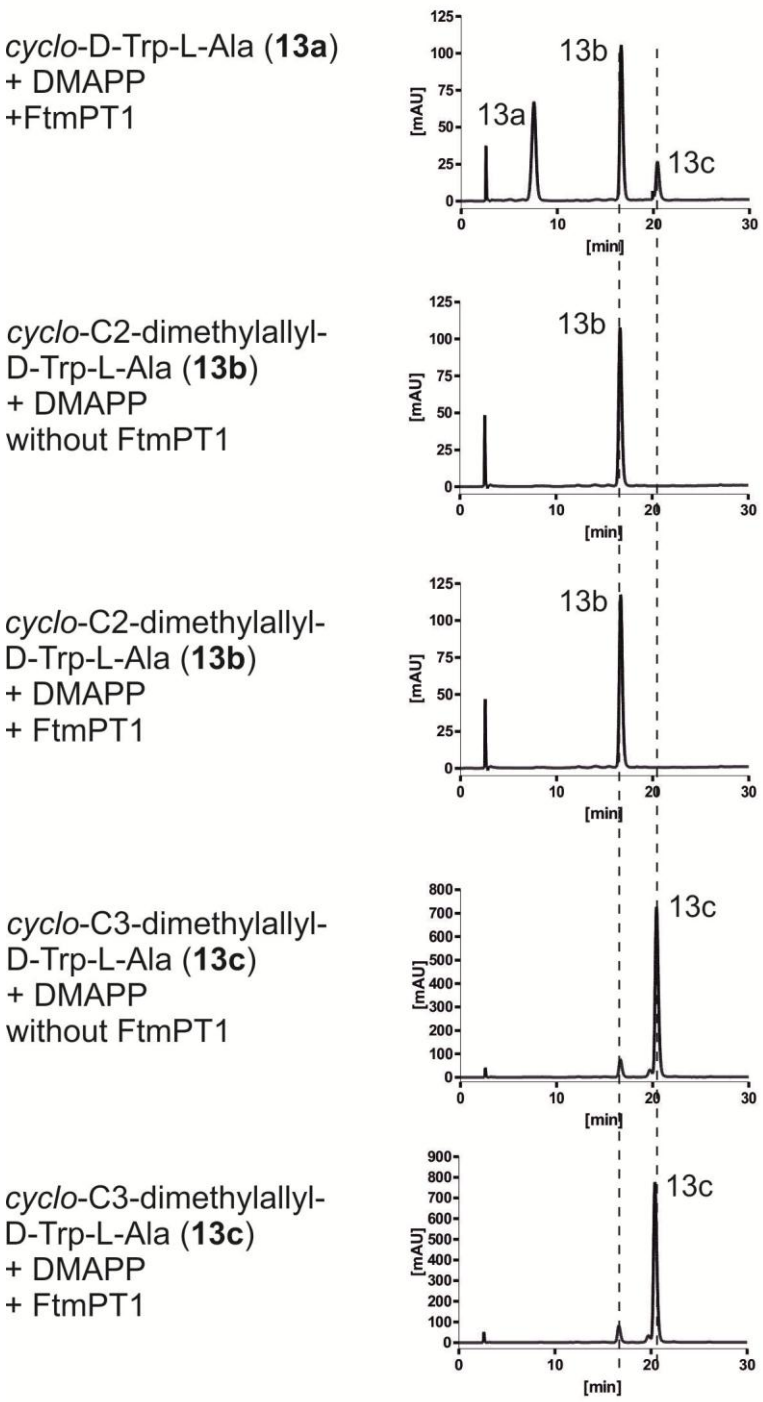

Figure S17 HPLC chromatograms of incubation mixtures with **13a**, **13b** or **13c** in the absence and presence of 5 µg FtmPT1. The reaction mixtures contained 10 mM $CaCl_2$, 1 mM aromatic substrate and 2 mM DMAPP and were incubated at 37°C for 2 h. The substances were detected with a Photo Diode Array detector and illustrated for absorption at 296 nm.

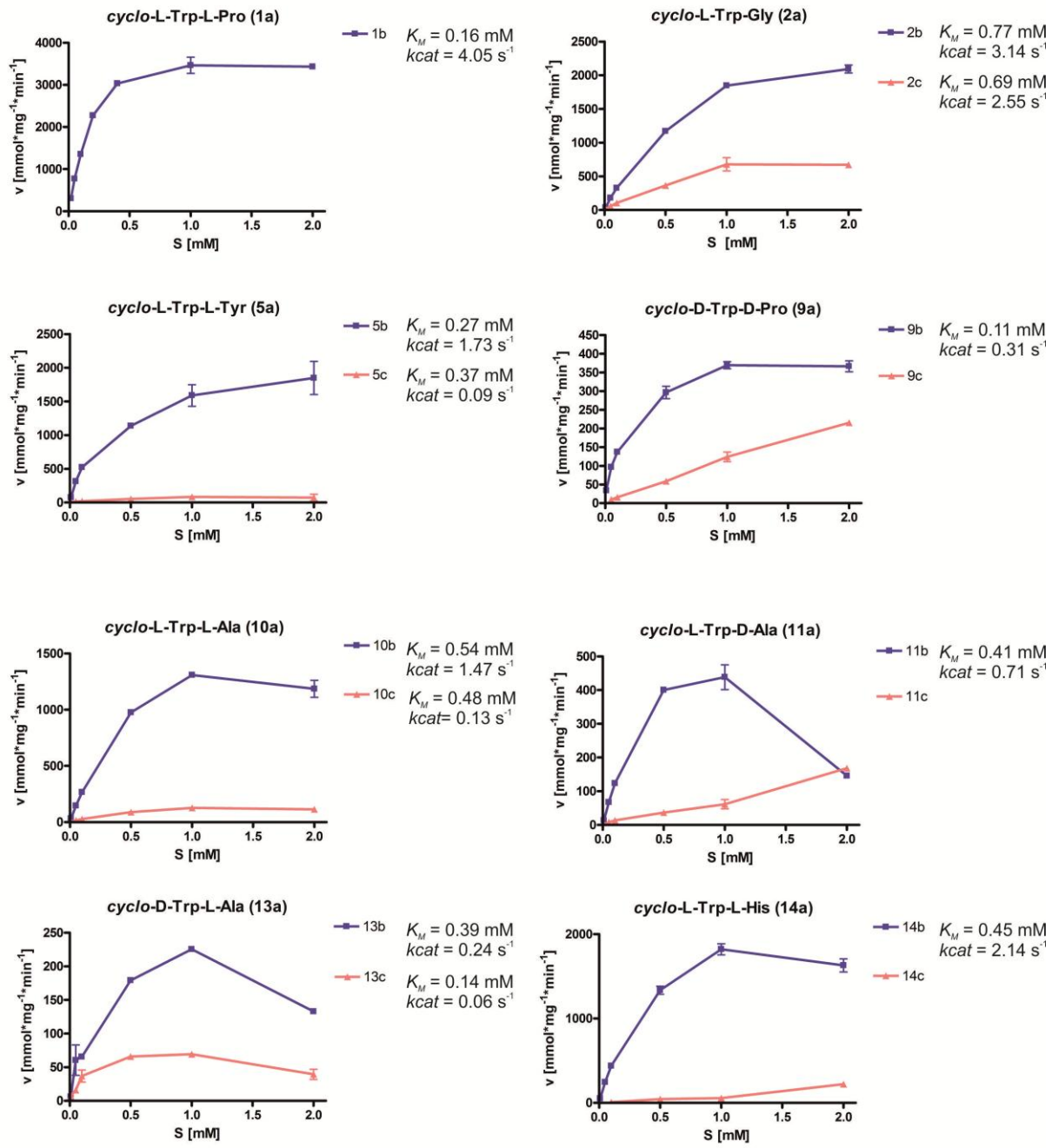

Figure S18 Dependence of product formation of **b** and **c** series on different substrate concentrations of **1a**, **2a**, **5a**, **9a**, **10a**, **11a**, **13a** and **14a**.