Mechanism of forming trimer, self-assembling nano-particle and inhibiting tumor growth of small molecule CIPPCT

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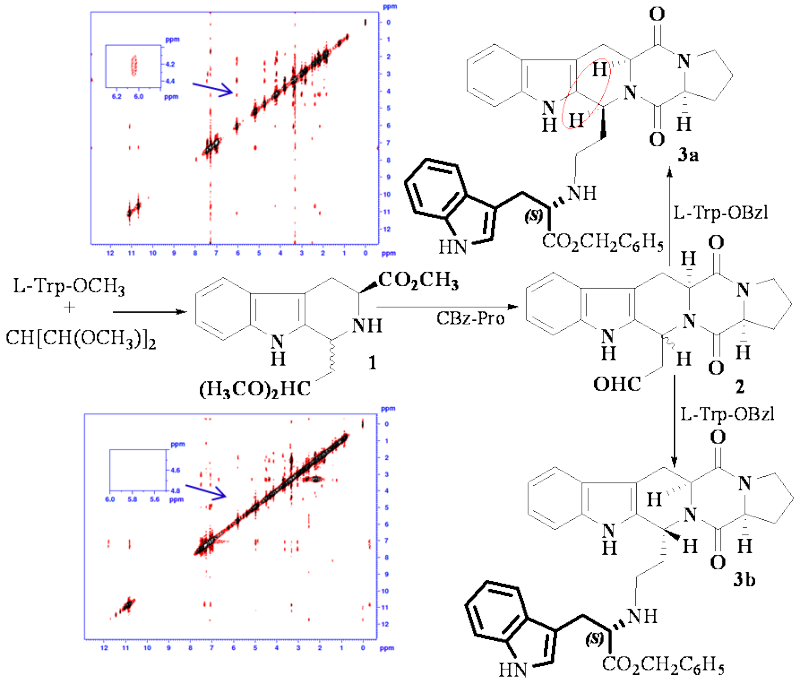
Supporting Information

Synthetic route

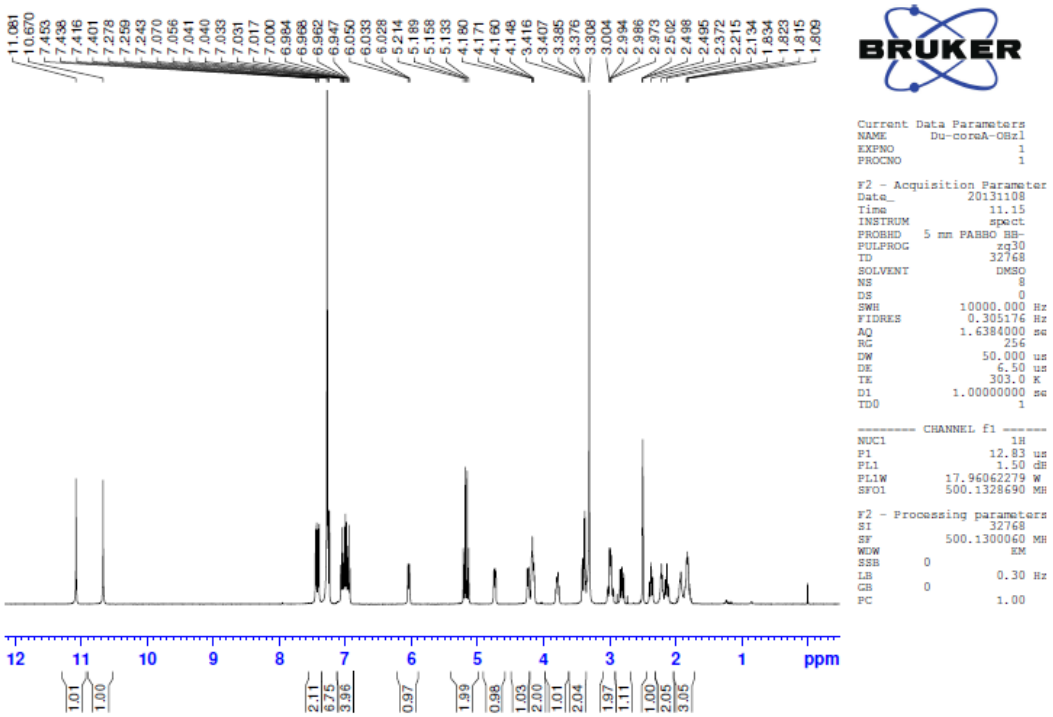
To conveniently obtain CIPPCT a 5-step reaction sequence of Scheme 2 was used, which can be divided into steps 1-3 for preparing **3a**, and steps 4,5 for preparing CIPPCT. In step 1 and 2 the cyclizations and hydrolysis of methanolacetal were performed. In brief, L-Trp-OMe (25 mmol) and 1,1,3,3-tetramethoxypropane (24 mmol) in CH_3OH was adjusted to pH 2 with F_3CCO_2H and stirred at 66 °C for 48 h to provide methyl 1-(2,2-dimethoxyethyl)-1,2,3,4-tetrahydrocarboline-3-carboxylate (**1,** ESI-MS: m/e 318, M^+) in 65% yield. Successively treating **1** with CBz-L-Pro (17 mmol)/oxalyl chloride (5 ml) for 5 h, with diisopropylamine for 24 h and with glacial acetic acid (24 ml) for 1 h to form aldehyde **2** (ESI-MS: m/e 337, M^+) in 80% yield. In step 3 the reductive alkylation of

aldehyde **2** with L-Trp-OBzl/triethylamine (7 mmol/1 mL) for 0.5 h, with sodium cyanoborohydride (10 mmol) for 2 h and the product was separated on silica gel column chromatography to provide **3a** and **3b** 34% and 17% yield, respectively. The 12C of **3a** was assigned S configuration due to a positive NOE signal between 5aS-H and 12-H occurring in its ROESY 2D NMR spectrum, and the 12C of **3b** was assigned R configuration due to no such a positive NOE signal between 5aS-H and 12-H occurring in its ROESY 2D NMR spectrum. Steps 1-3 are shown in Scheme 1. In step 4 the hydrolysis of **3a** provided CIPPC. In step 5 the coupling reaction of Thr-OBzl and CIPPC provided CICCPT.

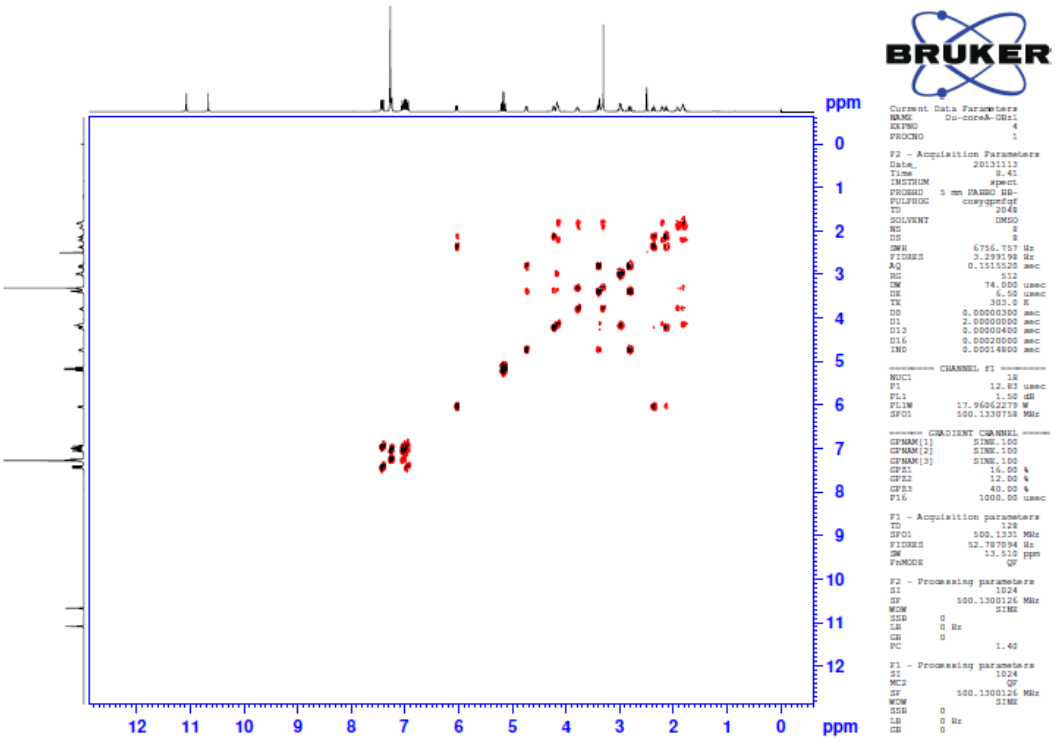


Scheme 1 Synthetic route and NOE between 5aS-H and 12-H of **3a**, (5aS,12S,14aS)-5,14-dioxo-12-(2-tryptophanbenzylester-N-ylethyl-1-yl)-1,2,3,5,5a,6,11,12,14,14a-decahydro-5H,14H-pyrolo[1,2:4,5]pyrazino[1,2:1,6]pyrido[3,4-b]indole. For **3b**, (5aS,

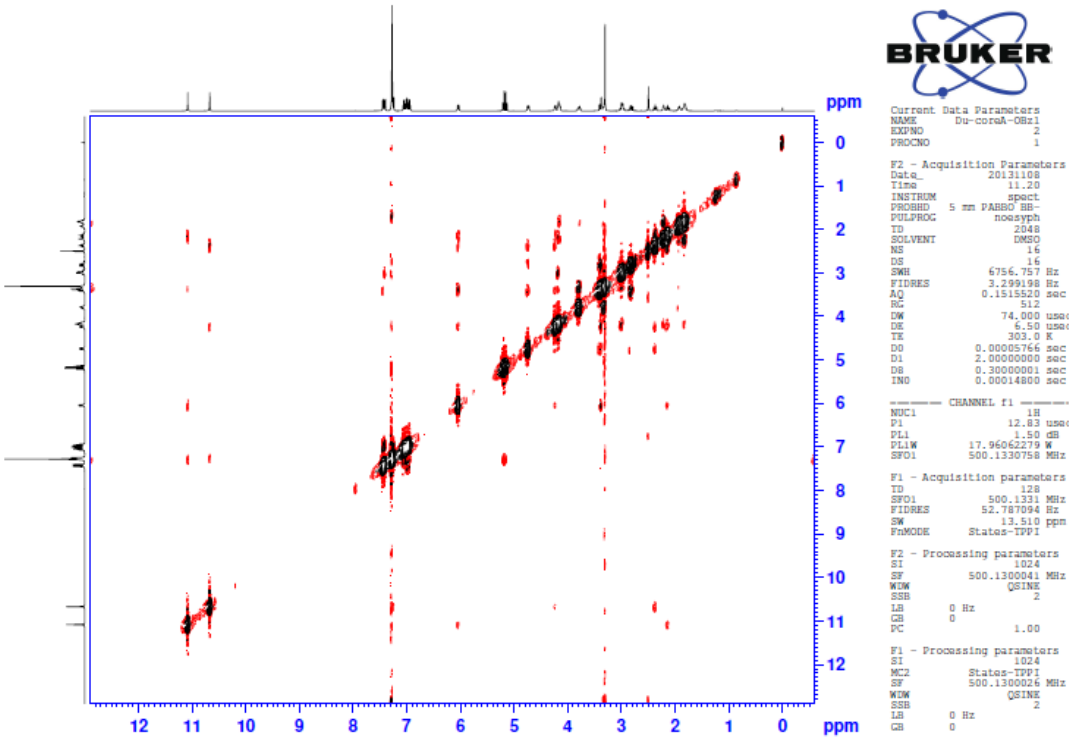
12R,14aS)-5,14-dioxo-12-(2-tryptophanbenzylester-N-ylethyl-1-yl)-1,2,3,5,5a,6,11,12,14,14a-decahydro-5H,14H-pyrolo[1,2:4,5]pyrazino[1,2:1,6]pyrido[3,4-b]indole, no NOE between 5aS-H and 12-H was observed.



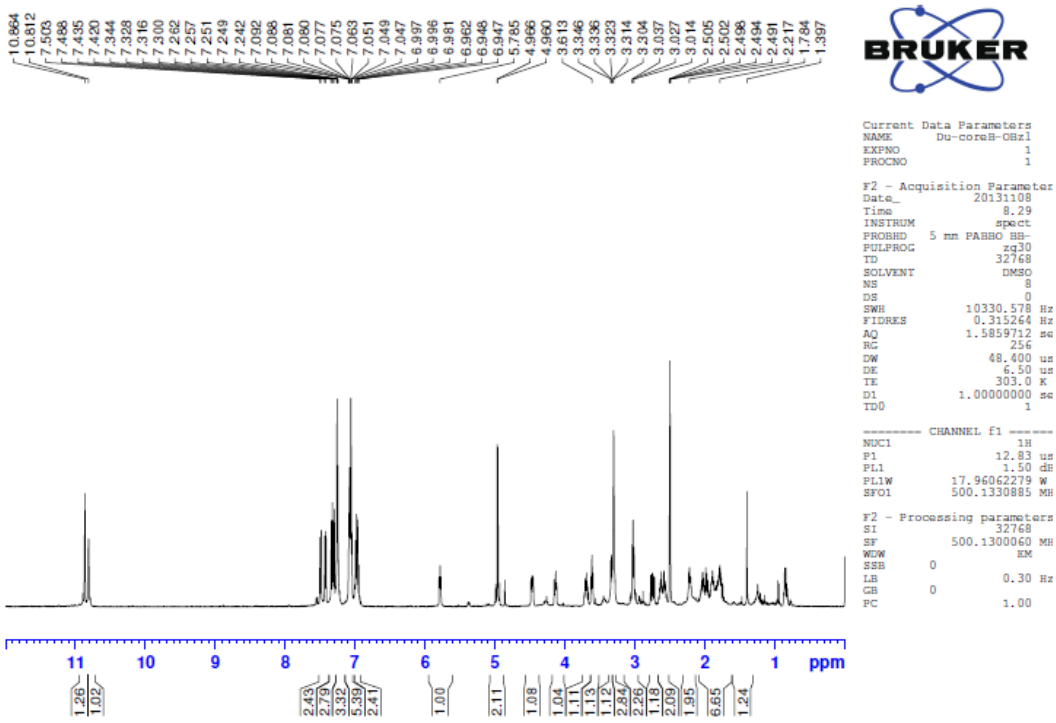
FigureS1. [1]H NMR of **3a**



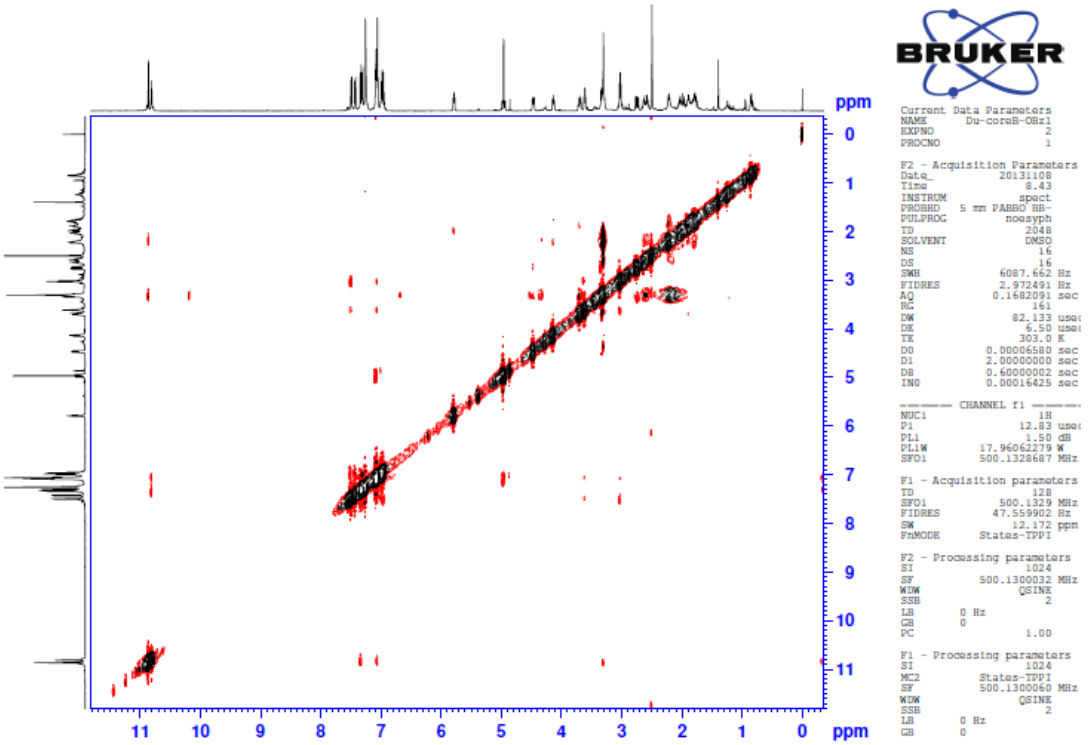
FigureS2. Cosy of **3a**



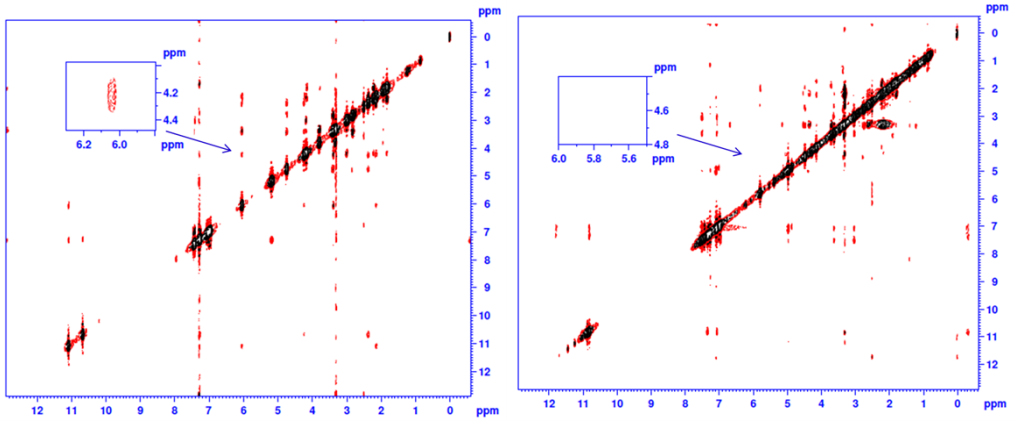
FigureS3. Noesy of **3a**



FigureS4. [1]H NMR of **3b**



FigureS5. Noesy of **3b**



FigureS6. Comparison of noesy spectra of **3a** and **3b**

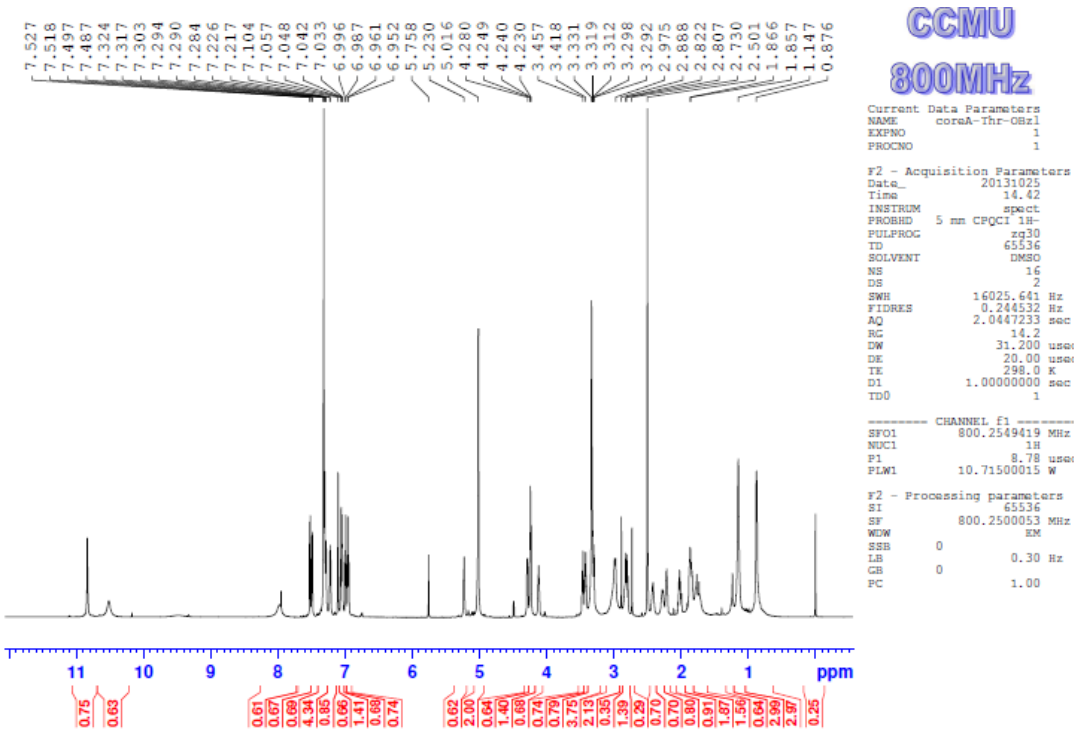


Figure S7. H^1NMR of CIPPCT

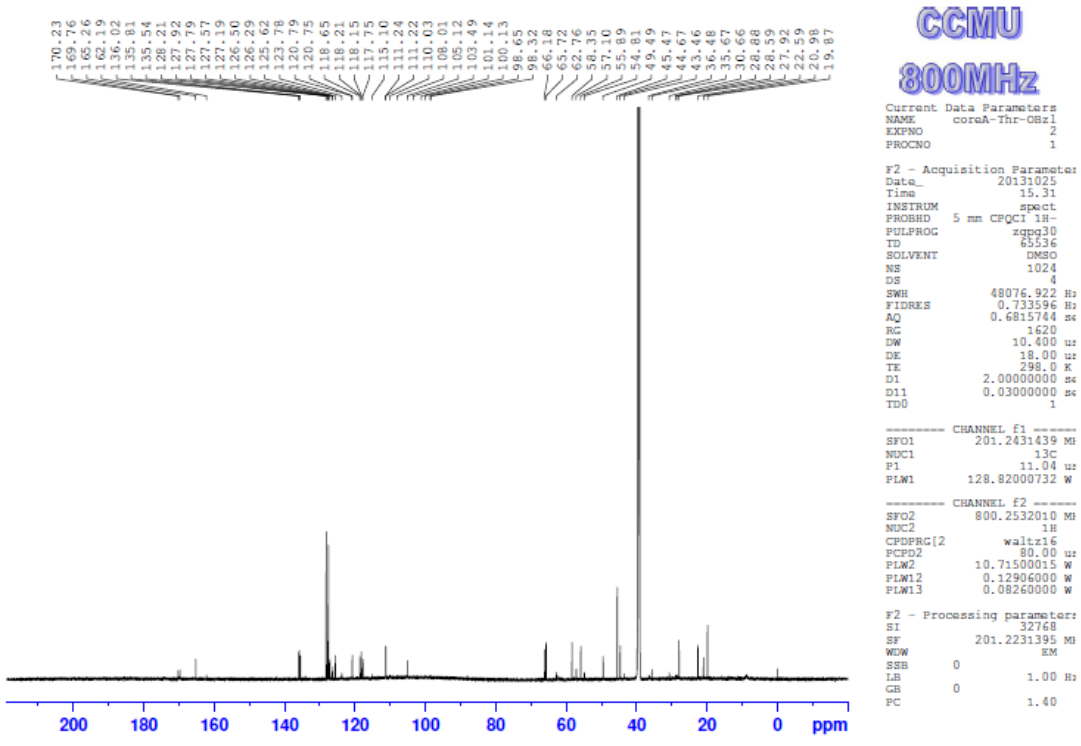


Figure S8. C^{13} NMR of CIPPCT

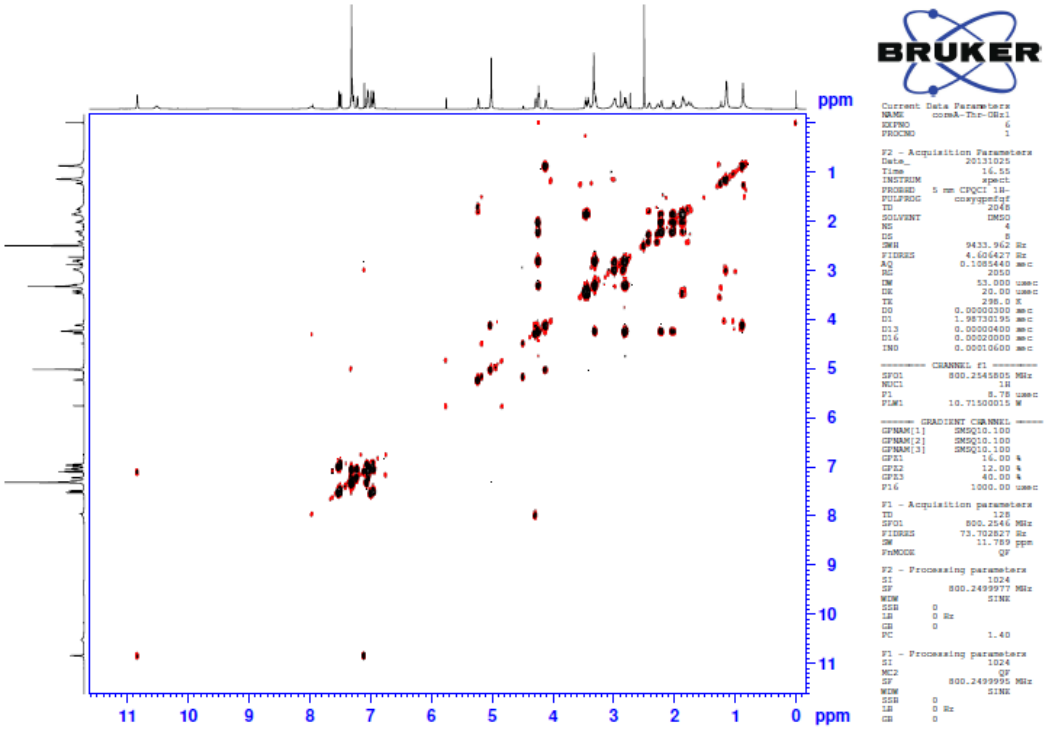


Figure S9. Cosy of CIPPCT

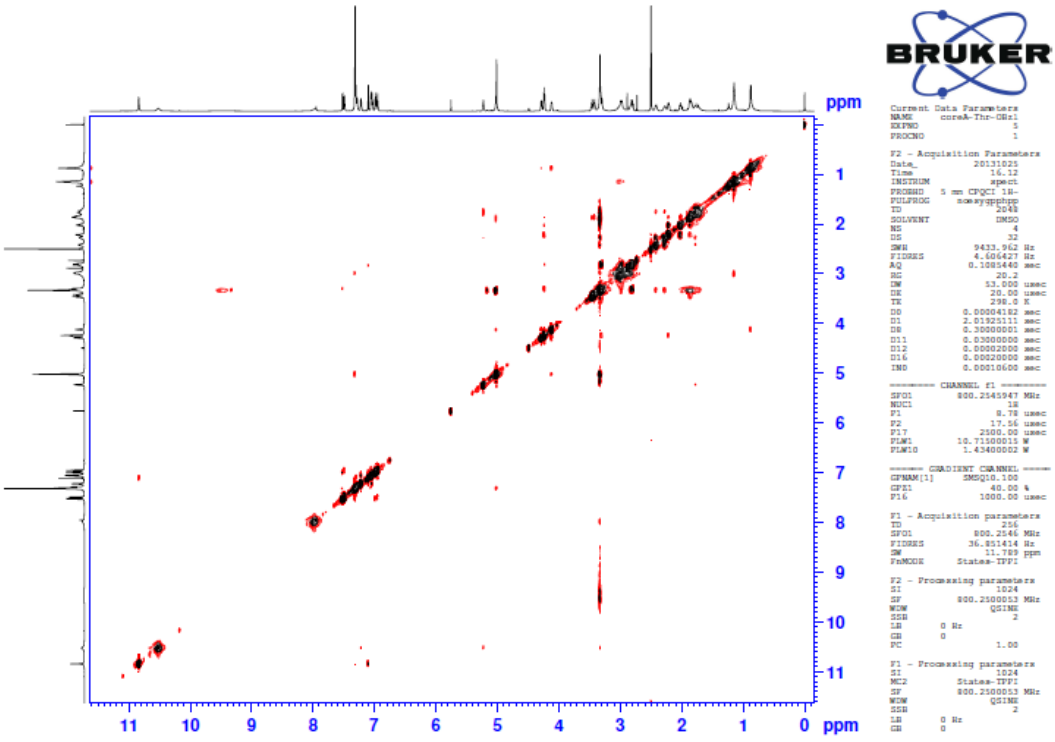


Figure S10. Noesy of CIPPCT