Supporting Information for

Envisaging the Structural Elevation in the Early Event of Oligomerization of Disordered Amyloid Beta Peptide

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Supporting Information

Scheme S1. Schematic view of Raman measurements and related assembly status.

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Figure S1. Raman spectra (1150-1800 cm^{-1}) of Aβ40 of different assembly conditions.

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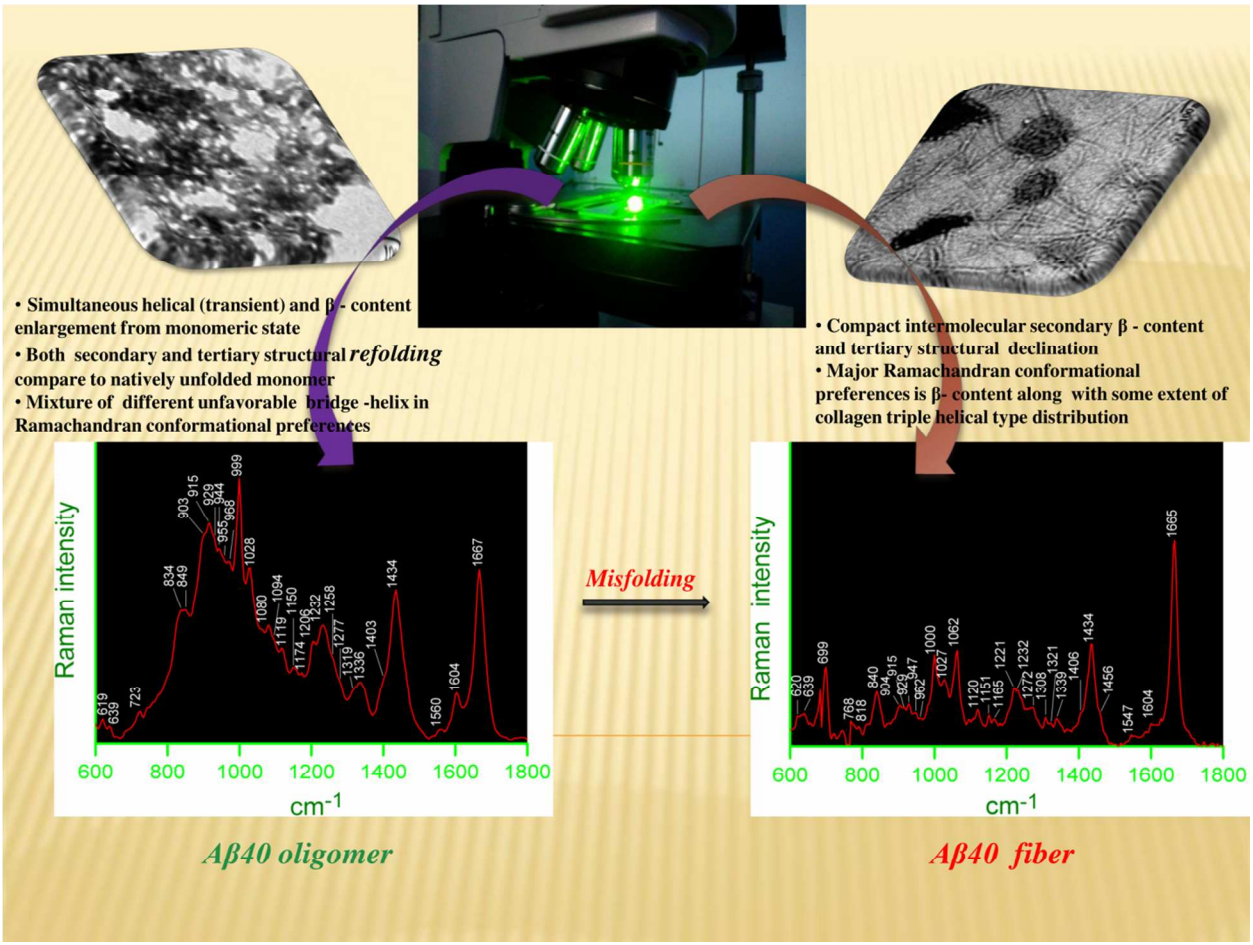
Figure S7. The residue specific average secondary structure analyses of Aβ40 dimer of MD simulation.

Figure S8. Contact map of Aβ40 dimer using OPLS-2005 force field.



Scheme S1. Schematic of Raman spectroscopic measurements of Aβ40 aggregates (oligomers and fibers) under micro Raman spectrometer. The sample was illuminated with green laser light at 532 nm wavelength. Left upper panel is the TEM images of Aβ40 oligomer and the bottom left corner shows its Raman spectrum as recorded under Raman microscope (center). Similarly the right hand side panels show the TEM image (upper right corner) of fibers and its Raman signature. Comments relavent to structural content are given in bullated form.

Table S1. Raman vibrational bands (cm^{-1}) of Aβ40 as monomer, oligomer and fiber states.

Monomer	oligomer	fiber	Origin of Raman vibration
-	619	620	Phe
-	639	639	Tyr
-	723	-	unknown
-	-	768	Ala
819	834	818	Tyr
843	849	840	Tyr
873	-	-	unknown
-	903	904	υCC
-	915	915	unknown
946	929,944	929,947	CH_2 symmetric rock+ Cα-C streching
-	955,968	962	CH_2 symmetric rock+ Cα-C streching
1006	999	1000	Phe
-	1028	1027	Phe
1072	1080, 1094	1062	υCC, υCN, υCO
1136	1119	1120	υCC, Val,Ile
-	1150	1151	Ile, υCC
1168	1174	1165	Try, Phe
1216	1206, 1232	1221, 1232	Amide III, β-turn, β-sheet
1245	-	-	Amide III, polyproline II
-	1258	-	Amide III, α-helix
1275	1277	1272	Amide III, 2.5_1-helix
1297	-	-	Amide III
-	-	1308	CH_2 twist/wag
1325	1319	1321	Amide III, CH_2 twist/wag
-	1336	1339	CH_2 twist or wag+Cα-H bending+ Cα-C streching
1362	-	-	unknown
-	1403	1406	Symmetric, υCO_2^-
1449	1434	1434, 1456	CH_2, CH_3 deformation, CH_2 scissoring
1573	1560	1547	Phe
-	1604	1604	Phe
1614	-	-	Tyr
-	1667	1665	Amide I, β-sheet
1677	-	-	Amide I, polyproline II, random β-space

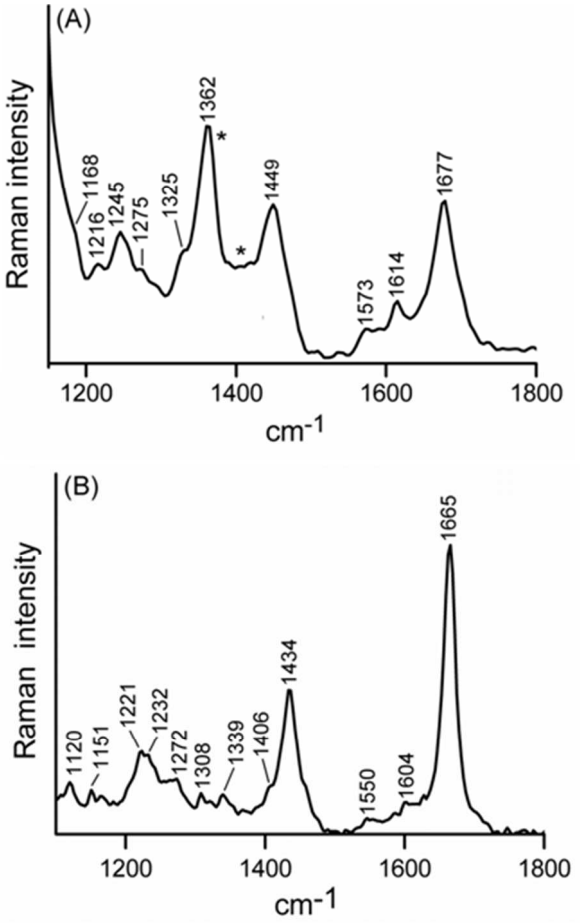


Figure S1. 532 nm excited Raman spectra (1150-1800 cm^{-1}) of Aβ40 of different assembly conditions**:** (A) Aβ40 monomer was prepared (1.5 mg/ml) dissolving in 10 mM phosphate buffer of pH 7.8, 10-30 μL solution was dropped onto a glass cover slip and (B) fiber was prepared 10 mM phosphate buffer of pH 7.2 incubation for 2 days at 25 ^{o}C, 10-30 μL solution was dropped onto a glass cover slip respectively. Laser power at source 30-35 mW, ~2 mW at the sample, recording time 10 ×15 sec. The represented figures obtained after baseline correction was done. The (*) marks indicate some of the contribution from phosphate in buffer.

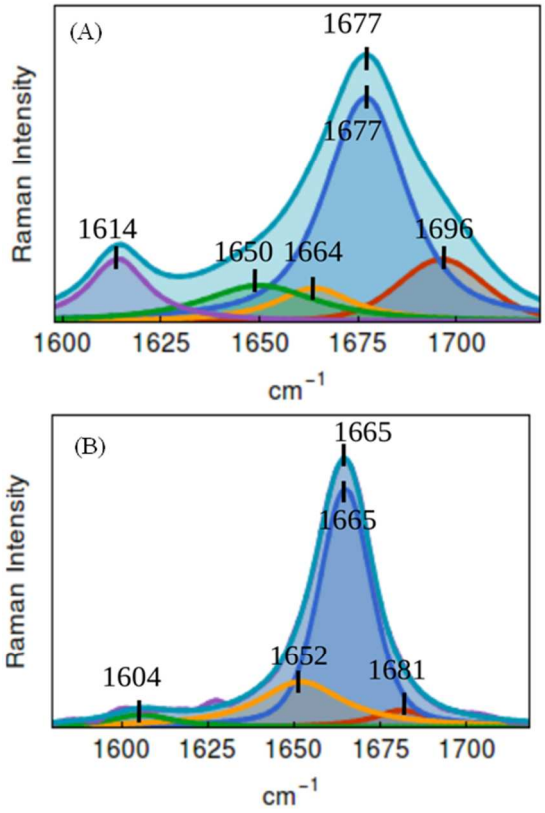


Figure S2. Curve fitting of the amide I region Raman spectra of Aβ40 of different assembly conditions: (A) monomer and (B) fiber. The other recoding conditions are same as Figure S1. Three component bands that represent total amide I bands were shown separately. The violet line was the original spectrum, red, blue,yellow, green, violet lines were the individual component bands and the cyan line was the sum of the bands.

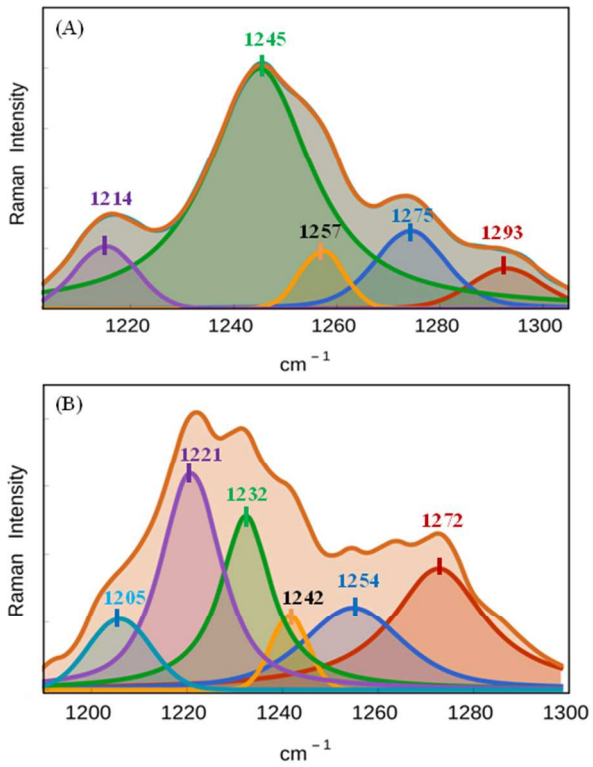


Figure S3. Band fitting of the amide III region of Raman spectra of Aβ40 monomer (A) and fiber (B), recpectively. The experimental conditions are similar to Figure S1. Amide III region (1200–1350 cm^{-1}) of oligomer, monomer and fiber (Figure 2 and Figure S3) also signified the results obtained from amide I band in the Raman spectrum. The major Raman band for monomer appeared at 1245 cm^{-1}, was a signature for PPII conformation. Weak bands at 1293 and 1319 cm^{-1} indicated presence of some residues in the helical space. The fiber showed major amide III signal at 1221cm^{-1} and 1232 cm^{-1} which corresponded to β-sheet structure. The band at 1232 cm^{-1} became much sharper compared to the similar band at 1228 cm^{-1} in the oligomeric state. The amide III band is primarily evolved from combination of C–N stretching and N–H bending and the mode of vibration is very sensitive to conformational state of the peptide bond. However, diversified contributions from vibrations of side chain residues may also contribute these regions.

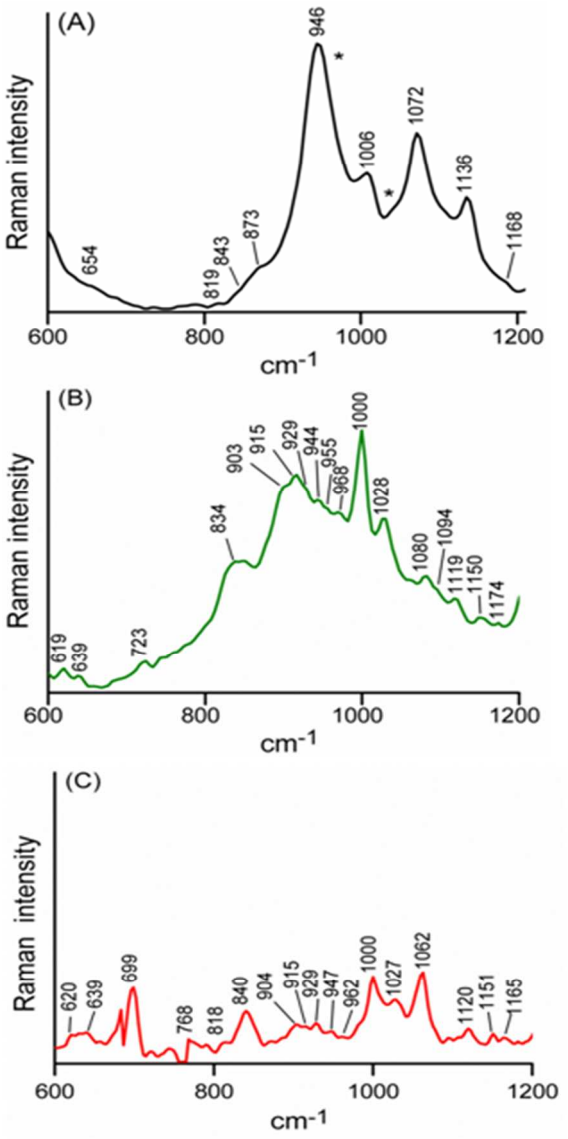


Figure S4. The 532 nm excited Raman spectra in the region of (600-1200 cm^{-1}) of Aβ40 as monomer (A), oligomer (B) and fiber (C) under Raman microscope respectively.

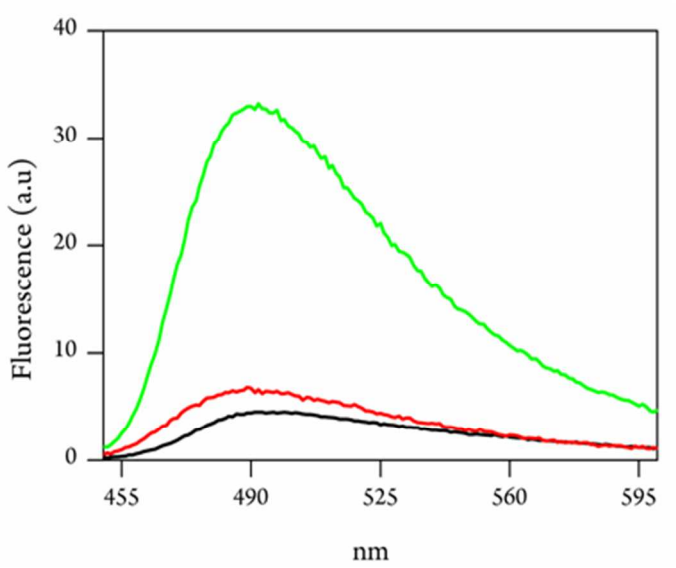


Figure S5. Thioflavin T fluorescence assay. The fluorescence spectra of Aβ40 monomer (black), oligomer (red) as obtained upon incubation at 25 ^{o}C for 3 h and the fibrillar condition (green), after incubation at 25 ^{o}C for 2 days.

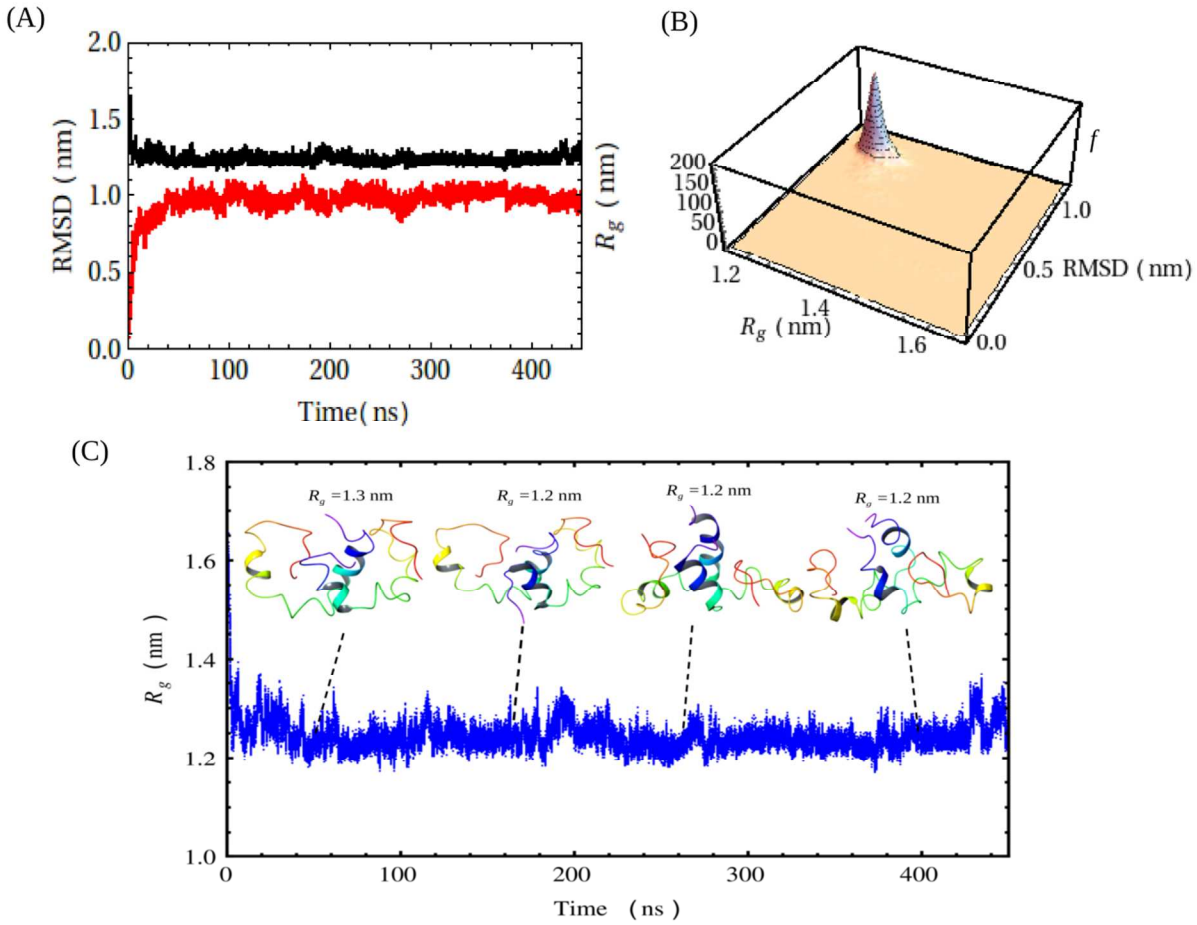


Figure S6. Root mean square deviation (RMSD) and radius of gyration (R_g) of the Aβ40 dimer as plotted against the simulation time. The MD simulation was carried out for 450 ns at 310 K. Panel A shows both the the RMSD (red) and R_g (black). Panel B displays the 3D histogram plot for clustering of different conformational states of the Aβ40 dimer by correlating the R_g with RMSD values and f is the frequency. Panel C depicts structures generated at different time interval of the simulation. The simulation time and the R_g values for each snapshort are given. The blue scattered plot at the bottom of the panel C shows the refinements in radius of gyration of Aβ40 backbone during the simulation processes.

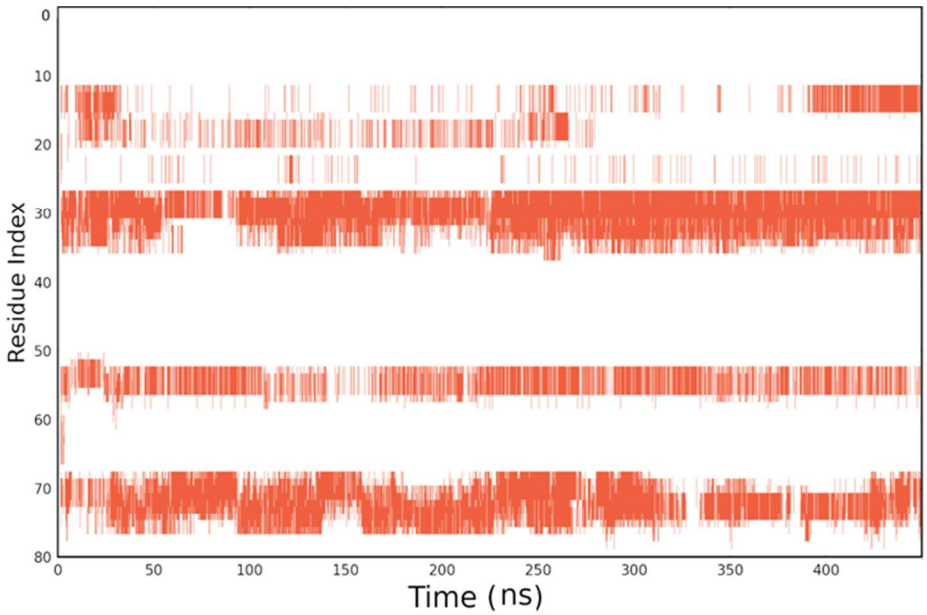


Figure S7. The residue specific average secondary structure analyses of Aβ40 dimer of MD simulation using desmond algorithm. The colour codes are as follows: white for loop, orange for helical conformation. Y axis gives the residue number of the peptide as residue index.

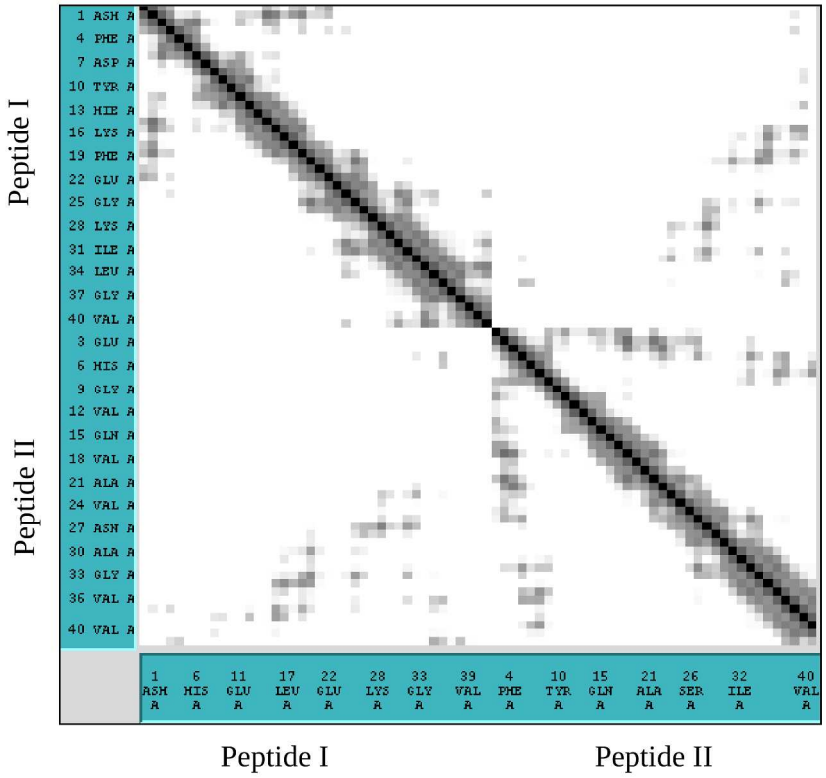


Figure S8. The Aβ40 dimer Contact map signify the α-Carbon to α-Carbon distance between all possible amino acid residues pairs of protein structure using a binary two-dimentional matrix plot. The contact distances are shows as a color-coded matrix where darker colors residues which are close to α-Carbon to α-Carbon and lighter colors show residue pairs which are far-away from α-Carbon to α-Carbon. linear square gray colour scale 10.0 Å, and white square colour is greater than 10.0 Å.