List of Figures

Figures	Title	Page
1.1	Schematic representation of the protein aggregation process. Main objectives of the current research work are highlighted in blue.	1
2.1	Schematic representation of protein folding and misfolding process.	5
2.2	Insoluble higher order deposits are associated with diseases.	7
2.3	Amyloid formation leads to lethal diseases.	7
2.4	Schematic representation of nucleation process of protein aggregation as a function of time. Where, N, is the native conformation of a protein; O, is the oligomeric state of it; and A, is the aggregated mature higher order structures.	8
2.5	Therapeutic strategies against amyloidogenic diseases (Skovronsky et al., 2006).	9
3.1	Coaggregation studies of mixed monomers of different globular proteins in PBS at ~70°C. (a) BSA and Insulin coaggregation: (II) BSA at ~4.9 μ M; (A) insulin at ~45.4 μ M; (◆) BSA (~2.9 μ M) + insulin (~ 27.5 μ M). (b) Insulin and Iysozyme coaggregation: (●) Lysozyme at ~27.9 μ M; (A) Insulin at ~45.3 μ M; (♦) Insulin (~27.5 μ M) + Iysozyme (~12.5 μ M). (c) BSA and Iysozyme coaggregation: (●) Lysozyme at ~25.9 μ M; (II) BSA at ~ 4.9 μ M; (♦) BSA (~2.9 μ M) + Iysozyme (~12.5 μ M). (d)BSA, Iysozyme and Insulin coaggregation: (II) BSA at~4.9 μ M; (A) Insulin at ~40.2 μ M; (O) Lysozyme at ~18.9 μ M; (II) BSA (~1.9 μ M) + Iysozyme (~8.4 μ M) + insulin (~18.4 μ M). (e) Coaggregation of mixed monomers of BSA and cytochrome <i>c</i> . Visualization of the absorbance profile of cytochrome <i>c</i> monomers at different time points: o hour (olive curve), 6 hours (blue curve) and 20 hours (red curve). Inset shows the progress of amyloid formation by Thioflavin T readings. (f) Stabilities of the amyloid fibrils against chemical denaturant. GnHCL	13
3.2	Structural conformational analysis of mature coaggregates of proteins. Electron micrographs of amyloid fibrils: (a) Lysozyme; (b) BSA; (c) Insulin; (d) and (e) are the mature fibrils obtained from coaggregation of BSA + Insulin + Lysozyme sample. Scale bar, \sim 100 nm. (f) Circular dichroism spectra of coaggregation of mixed monomers of BSA, lysozyme and cytochrome c at different time points. Due to precipitation of aggressive coaggregation later time points could not be obtained.	14
3.3	Control aggregation experiments of globular proteins at ~70 °C at pH7.4. (a) Aggregation kinetics of individual reactions of BSA at ~10.5 μ M (\blacksquare), lysozyme at ~48.9 μ M (\bigcirc) and BSA (~2.9 μ M) + lysozyme (~12.5 μ M) mixed sample (\blacktriangle). (b) Change in cytochrome c concentration as a function of time. Cytochrome C (at 24.53 μ M), ThT measurements at different time points of cytochrome c sample are as follows: ohrs = 0, 7 hrs = 0, and 48 hrs = 7. This aggregation kinetics of cytochrome c alone was taken as control experiment for both the experiments mentioned in Figure 3.1e and Figure 3.4d.	15
3.4	Cross-seeding of proteins during amyloid formation (a) BSA at ~7.5 μ M (\blacksquare); lysozyme at ~ 27.9 μ M (\bullet); Insulin at ~52.3 μ M (\blacktriangle); BSA + BSA seed (\diamond); lysozyme + BSA seed (\bigtriangledown); Insulin + BSA seed (\triangleleft). (b) BSA at ~6.0 μ M (\blacksquare); lysozyme at ~27.9 μ M (\bullet); Insulin at ~52.3 μ M (\blacktriangle); BSA + lysozyme seed (\triangleleft); lysozyme + lysozyme seed (\diamond); Insulin + lysozyme seed (\checkmark); lysozyme + lysozyme seed (\diamond); Insulin + lysozyme seed (\checkmark); lysozyme at ~18.9 μ M (\bullet); Insulin at ~40.2 μ M (\blacktriangle); BSA + Insulin seed (\triangleleft); lysozyme + Insulin seed (\checkmark); lysozyme + Insulin seed (\checkmark). (d) Aggregation of cytochrome c monomers (at ~24.5 μ M) in the presence of BSA amyloids (seeds). Inset shows the Thioflavin T readings.	16
3.5	Coaggregation and cross seeding experiments between lysozyme, BSA and Insulin at 37° C in PBS. (a) BSA only at 10.5µM (I); Lysozyme only at 34.9µM (O) and Insulin only at 104.7µM (A); BSA+ BSA seed (V); Lysozyme + lysozyme seed (\diamond); Insulin +insulin seed (\triangleleft). (b) BSA monomers (10.5µM) + Lysozyme monomer (34.9µM) mixed sample (I); Lysozyme monomers (34.9µM) + BSA seeds (\diamond). The seed concentration was ~15% weight/weight. All these data are average values of at least two independent experiments. Thioflavin T readings were also measured for these samples after two weeks; however no change of Thioflavin T signal was detected.	17

Figures Title

- 3.6 Amyloid formation of lysozyme in presence of collagen. (a) Inhibition of spontaneous aggregation lysozyme by type I collagen. Lysozyme aggregation at 10 mg/ml was studied in presence of different concentrations of type I collagen: lysozyme+0 μM collagen (■), lysozyme +1 μM collagen (●), lysozyme +2 μM collagen (●) and lysozyme +3 μM collagen (●). (b) Inhibition of seed-induced amyloid formation of lysozyme in presence of collagen: lysozyme+0 μM collagen (■), lysozyme+0 μM collagen (■), lysozyme +5% (w/w) seeds (○), lysozyme + 5% (w/w) seeds + 3 μM collagen (▲). Seed is the preformed lysozyme fibrils. All the data showed here are the average values of at least three independent measurements.
- 3.7 (a) Intrinsic tryptophan fluorescence spectra of lysozyme: lysozyme only (–), lysozyme + 5 M GnHCl (–), lysozyme + collagen + 5 M GnHCl (–). (b) Biological activity of lysozyme in the presence of collagen. Catalytic activity of lysozyme both in the presence and absence of collagen was performed at pH 2.0 (upper panel) and at pH 6.5 (lower panel). All the measurements were obtained at 25 °C and the ratio of lysozyme to collagen was maintained at 100:1. Each plot shown from both fluorescence and activity measurements represents the average value of two to three independent measurements.
- 3.8 Effect of collagen on lysozyme conformation and stability by circular dichroism (CD). (a) CD spectra of lysozyme samples: lysozyme monomers before aggregation (—), lysozyme amyloid fibrils after aggregation (—), inhibited lysozyme sample in the presence of collagen (—). (b) Thermal unfolding of hen egg white lysozyme in the absence (\bigcirc) and the presence (\Box) of type I collagen. Temperature scan rate for the thermal unfolding experiments was maintained at 2°C/min. The value of transition temperature (T_m) of lysozyme was 62.3 °C and in the presence of collagen the T_m was increased to 67.4 °C.
- 3.9 (a) Amino acid composition of α1 chain of type I collagen. (b) Amino acid composition of α2 chain of type I collagen. (c) Effect of Iysozyme on collagen fibril formation at pH 7.3, 37 °C. Type I collagen (from Rat Tail Tendon) (~0.2mg/ml) was dissolved in 5% acetic acid solution and an appropriate volume of 10X PBS was added to adjust the sample buffer to pH ~7.3. The process of fibril formation was monitored by recording the change in the optical density of the sample at 313 nm using a spectrophotometer. Details of the symbols are as follows: collagen only (■), collagen + lysozyme (●). The molar ratio of collagen to lysozyme was 1:100.
- 3.10 Schematic representation of rapid coaggregation among globular proteins during amyloid formation.
- 3.11 Schematic representation of inhibition of lysozyme amyloid formation by type I collagen
- Inhibition of insulin and BSA amyloid formation in the presence of eugenol. (a) Effect of 4.1 eugenol on spontaneous aggregation of insulin ($\sim 43 \,\mu$ M) at different molar ratios; 1:0 (\blacksquare), 1:35 (●), 1:70 (▲), 1:100 (▼) and 1: 150 (■). (b) Seed-induced aggregation of insulin (at ~ 43 μ M): Insulin alone (\blacksquare), insulin + seed (\bullet); insulin + seed + eugenol at 1:100 molar ratio of protein:inhibitor (\blacktriangle). (c) Effect of eugenol on spontaneous aggregation of BSA (at ~ 6.5 μ M): BSA only (\blacksquare); BSA+eugenol at 1:250 molar ratio of protein: inhibitor (\blacktriangle); BSA+eugenol at 1:800 molar ratio of protein: inhibitor (•). (d) Seed induced aggregation of BSA (~ 3μ M); BSA only (\blacksquare), BSA + seeds (\bigcirc), and BSA + seeds + eugenol (1:400 molar ratio) (\blacktriangle). (e) Effect of eugenol on coaggregation of BSA, insulin and lysozyme: ~ 2 μ M BSA (●), ~ 19 μM Insulin (▲), ~ 8.4 μM Iysozyme (▼), [2 μM BSA+ 19 μM Insulin+ 8.4 μM $[y_{sozyme}] (\bullet), [2 \mu M BSA+ 19 \mu M Insulin+ 8.4 \mu M Iy_{sozyme} + 3 m M eugenol] (<). (f) Effect$ of eugenol on the disassembly process of matured amyloid fibrils of insulin and BSA; Insulin amyloids (■), insulin amyloids + eugenol (1:100 molar ratio of protein:inhibitor) (●), BSA amyloids (▲), BSA amyloids + eugenol (1:400 molar ratio of protein:inhibitor) (▼). Until ~300 hr of observation, no indication of dissociation of amyloid fibrils was observed. Seed implies 15% (w/w) preformed mature fibrils of the protein sample.
- 4.2 Control experiments for aggregation and CD experiments. (a) Kinetics of amyloid formation of insulin measured through ThT assay. (b) CD spectra of eugenol at ~0.4 mM (—) and water (—).
- 4.3 AFM images of Insulin and BSA aggregates formed in presence of eugenol. (a) Mature Insulin amyloids after 72 h incubation. (b) Spheroidal oligomers of insulin observed after ~72 h incubation. (c) Mature amyloids of BSA, after ~72 H incubation. (d) Protofibrilar assemblies obtained in BSA sample incubated for ~72 h. Scale Bar, ~100 nm.

Page 19

20

21

23

23

24 27

28

29

Figures Title

- AFM images from 5 hours incubated sample of Insulin aggregates, formed in the presence 4.4 and in the absence of eugenol. (a) Insulin aggregates in the absence of eugenol. (b) Oligomers turning to protofibrils in the presence of eugenol. Scale Bar, ~ 100 nm.
- Structural studies on eugenol-protein interaction. (a) CD spectra of BSA undergoing 4.5 aggregation in the presence and in the absence of eugenol (molar ratio of protein:ligand was 1:400): BSA only at 0 h (---); BSA + eugenol at 0 h (---); BSA only at 90 h (---); BSA + eugenol at 90 h (-). (b) Native gel-electrophoresis of the BSA (-5μ M) undergoing amyloid formation in the presence and absence of eugenol (at $\sim3mM$): (1) soluble BSA;(2) mature BSA aggregates; (3) Sample taken from an aggregation reaction of BSA in the absence of eugenol at 5 h; (4) Sample taken from an aggregation reaction of BSA in the presence of eugenol at 5h. (c) CD spectra of insulin undergoing aggregation in the presence and in the absence of eugenol (molar ratio of protein:ligand was 1:100): insulin only at 0 h (---); insulin + eugenol at 0 h (---); insulin only at 90 h (---); insulin + eugenol at 90 h (---). Insets shown in the panel A and panel B show Thioflavin data for the respective CD samples. (d) ATR FTIR second derivative spectra of final aggregates: 1) insulin aggregates in eugenol (molar ratio of protein: ligand is 1:100); 2) insulin aggregates in the absence of eugenol; 3) BSA aggregates in eugenol (molar ratio of protein: ligand is 1:400); 4) BSA aggregates in the absence of eugenol.
- (a) Fluorescence spectra of eugenol sample (at 1.65 μ M) with increasing concentrations of 4.6 insulin. Excitation wavelength was 262 nm. Concentrations of insulin were: 1) 0 μ M; 2) 1 μ M; 3) 2 μ M; 4) 7 μ M; 5) 11 μ M. The inset shows the baseline corrected emission curves of eugenol. (b) The curve of log ((Fo-F)/F) versus log ([Bt]-(1/n)[Dt] (Fo-F)/Fo) for eugenol binding with insulin. (c) Stern-Volmer plot of eugenol and insulin. All the fluorescence measurements were carried out at room temperature. (d) Sedimentation assay for determining the affinity of eugenol to amyloid fibril of BSA and Insulin.
- (a)Docked complex of insulin (PDB ID: 415Z) with eugenol represents five interactions: four 4.7 hydrogen bonds (B:ARG22:HE - eugenol:O2, B:ARG22:HH21 - eugenol:O2, eugenol:H24 -B:CYS19:O, and B:GLY20:HA1 -eugenol:O1) and one pi-alkyl interaction (eugenol - B:ARG22). (b) Docked complex of BSA (PDB ID: 4F5S) with eugenol represents seven interactions: one hydrogen bond (eugenol:H24 - A:THR578:OG1), three alkyl interactions (A:ALA527 eugenol:C12, Eugenol:C12 - A:LYS524, eugenol:C12 - A:LEU528), and three pi-alkyl interactions (A:PHE550 - eugenol:C12, eugenol - A:LEU531, eugenol - A:VAL546). The detailed information about the protein-ligand interactions is given in the Table 4.1 and 4.2.
- Effect of capsaicin on type I collagen fibril formation and fibril dissociation. (a) Kinetics of 4.8 collagen fibril formation was monitored in the presence and in the absence of capsaicin, at different molar rations: Collagen alone (1:0, —), Collagen+capsaicin (1:10, —), Collagen+capsaicin (1:25, —), Collagen+capsaicin (1:50, —), and Collagen+capsaicin (1:100, —). Concentration of the collagen was kept at $\sim 1 \mu M$ in all the samples and the measurements were recorded in PBS buffer (pH 7.4) at 37°C. AFM images of final collagen fibrils of collagen in the absence and in the presence of capsaicin were also shown. Scale bar, ~100 nm. (b) Effect of capsaicin on disassembly of collagen fibrils. Molar ratio of the collagen to capsaicin was 1:100 and the measurements were recorded at 25°C as a function of time. (c) Effect of dihydro-capsaicin on collagen fibril formation in PBS at 37°C. Collagen concentration was ~0.1mg/ml and the molar ratio of collagen to dihydro-capsaicin was 1:100.
- (a) Protective effect of capsaicin against enzymatic degradation of rat tail tendons (RTT). 4.9 35 The percentage of degradation of collagen tendons by collagenase enzyme was determined in the absence and in the presence of capsaicin (10 μ M, 25 μ M, 50 μ M, and 100μ M). The collagen to collagenase ratio was maintained at 50:1 and the samples were incubated at 37°C for 96 hrs. All the measurements were repeated at least two times. (b) Correlation between the increase in shrinkage temperature and % protection against collagenase. R^2 for the linear regression fit is 0.76.
- CD spectroscopic studies of collagen-capsaicin interactions. (a) CD spectra of molecular 4.10 collagen in the presence and in the absence of capsaicin, recorded at different molar ratios. (b) Thermal unfolding of type-1 collagen was measured in the presence and in the absence of capsaicin. The change in the CD signal at 222 nm for the triple-helix was monitored as the temperature of the sample was increased from 20 to 60° C at a rate of 1 °C min⁻¹. All the CD scans were averages from three accumulations.

Page 29

30

31

32

34

4.11	Docking of capsaicin with Gly-Pro-Hyp triplet of the triple-helical collagen like peptide (PDB ID: 1CAG). (a) Capsaicin docked with the collagen triple helix, the triple-helix surface of which is rendered as its electron-density map. (b) Molecular interaction of capsaicin with the collagen triple helix. Hydrogen bonds are shown with <i>gray dotted lines</i> , whereas the electrostatic interaction is shown with a <i>light purple dashed line</i> . The docking studies were performed by the use of Discovery Studio 4.0 (see Annexure A). (c) Capsaicin docking region was highlighted in the collagen triple helical peptide.	38
4.12	Representing a decoy of capsaicin; ZINC35317807, that showed interaction with GPO regions of collagen peptide (PDB ID: 1CAG). This study was performed to confirm the role of functional groups of capsaicin that were identified to be involved in making interactions with GPO triplets of 1CAG peptide, 10 capsaicin decoys (See Annexure A) were identified by the use of ZINC database.	38
4.13	Docking of capsaicin with GER triplet was carried out using a triple-helical collagen like peptide (PDB ID: 1Q7D). (a) Capsaicin docking region was highlighted in the collagen triple helical peptide. (b) Interaction of capsaicin with the triple helix. Hydrogen bonds are shown with <i>gray dotted lines</i> , whereas the summary of H-bond interactions taking place was also mentioned.	39
4.14	Docking of capsaicin with LOG triplet was carried out using a triple-helical collagen like peptide (PDB ID: 2DRT). (a) Capsaicin docking region was highlighted in the collagen triple helical peptide. (b) Interaction of capsaicin with the triple helix. Hydrogen bonds are shown with <i>gray dotted lines</i> , whereas the summary of H-bond interactions taking place was also mentioned.	39
4.15	Docking of capsaicin with KEG triplet was carried out using a triple-helical collagen like peptide (PDB ID: 1QSU). (a) Capsaicin docking region was highlighted in the collagen triple helical peptide. (b) Interaction of capsaicin with the triple helix. Hydrogen bonds are shown with <i>gray dotted lines</i> , whereas the summary of H-bond interactions taking place was also mentioned.	40
4.16	Docking of capsaicin with GFO triplet was carried out using a triple-helical collagen like peptide (PDB ID: 1Q7D). (a) Capsaicin docking region was highlighted in the collagen triple helical peptide. (b) Interaction of capsaicin with the triple helix. Hydrogen bonds are shown with <i>gray dotted lines</i> , whereas the summary of H-bond interactions taking place was also mentioned.	40
4.17	Analysis of the presence of (Gly-Pro-Hyp) regions in the sequence of type I collagen. In type I rat collagen there are two a-1 chains (NCBI Accession ID: AAI33729 & UniProt ID: P02454) and one a-2 chain(NCBI Accession ID AAD41775 & UniProt ID: P02466, Organism- <i>Rattus</i> norvegicus).	41
5.1	Characterization of tyrosine and tryptophan cappeed nanoparticles. (a) Schematic representing tyrosine (AuNPs ^{Tyr} and AgNPs ^{Tyr}) and tryptophan (AuNPs ^{Trp}) coated nanoparticles. The orientation of the attached tyrosine residues over the silver nanoparticles (AgNPs ^{Tyr}) was kept reversed as compared to AuNPs ^{Tyr} . (b) UV-visible spectra of the nanoparticle samples: () AuNPs ^{Tyr} ; () AuNPs ^{Tyr} ; (b) UV-visible spectra of nanoparticles samples: () AgNPs ^{Tyr} ; () AuNPs ^{Tyr} ; () Tyrosine; () AuNPs ^{Trp} ; and () Tryptophan. (d) Transmission electron microscopic images of AuNPs ^{Tyr} , AuNPs ^{Trp} and AgNPs ^{Tyr} . Scale bar, 100 nm. The average diameter of the nanoparticles was observed arround ~10-30 nm. (e) Particles size histograms corresponding to tyrosine and tryptophan coated AuNPs ^{Tyr} , AuNPs ^{Trp} and AgNPs ^{Tyr} nanoparticles. This analysis was done by using ImageJ software.	44
5.2	Inhibition of insulin amyloid formation by tyrosine and tryptophan coated nanoparticles. (a) Effect of AuNPs ^{Tyr} on aggregation of ~40 µM insulin sample at different values of molar ratio: (•) Insulin; (•) Insulin + AuNPs ^{Tyr} at 4:1; (•) Insulin + AuNPs ^{Tyr} at 2:1; (•) Insulin + AuNPs ^{Tyr} at 2:1; (•) Insulin + AuNPs ^{Tyr} at 1:1. (b) Effect of AuNPs ^{Trp} on spontaneous aggregation of ~40 µM insulin at different molar ratios: (•) Insulin only; (•) Insulin + AuNPs ^{Trp} at 1:1 molar ratio; (•) Insulin + AuNPs ^{Trp} at 1:3 molar ratio; (•) Insulin + AuNPs ^{Trp} at 1:5 molar ratio. (c) Effect of AuNPs ^{Tyr} at 1:1 molar ratio; (•) Insulin + AuNPs ^{Tyr} at 1:1 molar ratio. (c) Effect of AuNPs ^{Tyr} at 1:1 molar ratio; (•) Insulin + 15% (w/w) seed; (•) Insulin+seed+AuNPs ^{Tyr} at 1:1 molar ratio; (•) Insulin+15% (w/w) seed; (•) Insulin+seed+AuNPs ^{Tyr} at 1:1 molar ratio; (•) Insulin+15% molar ratio. (d) Effect of isolated tyrosine, tryptophan and phenylalanine residues on aggregation of insulin: (•) Insulin only; (•) Insulin+Tyr at 1:1; (•) Insulin+Trp at 1:5; (•)	45

Page

Figures

Title

Insulin+Phe at 1:5 molar ratio.

Figures Title

- (a) Thermal unfolding of insulin by circular dichroism by monitoring change of CD signal at ~222nm: (○) insulin only; (□) insulin+ AuNPs^{Tyr} at 1:1 ratio; (▷) insulin+ AuNPs^{Tyr} at 1:5 molar ratio. (b) CD spectra of insulin in the presence and absence of Trp and Tyr coated nanoparticles: (·····) insulin only; (─) insulin+ AuNPs^{Tyr} at 1:1 molar ratio; (─) insulin+ AuNPs^{Tyr} at 1:5 molar ratio. (c) CD spectra of insulin undergoing aggregation in the presence and in the absence of nanoparticles: insulin only at 0 hr; (─) and 9 hr (─); insulin+ AuNPs^{Tyr} at 1:1 molar ratio at 0 hr (─) and at 9 hr (─); insulin+ AuNPs^{Tyr} at 1:1 molar ratio at 0 hr (─) and at 9 hr (─); insulin+ AuNPs^{Tyr} at 1:1 molar ratio at 0 hr (─) and at 9 hr (─); insulin at 0 hr (─) and at 9 hr (─); insulin mature amyloid fibrils. (e) Comparison of the effect of AuNPs^{Tyr} and AgNPs^{Tyr} at 1:1 molar ratio; (■) insulin + AuNPs^{Tyr} at 1:1 molar ratio.(f) Effect of tyrosine coated nanoparticles on disassembly of mature amyloid fibrils of insulin: (■) insulin: (■) insulin fibrils; (●) insulin fibrils + AuNPs^{Tyr} at 1:1 molar ratio of insulin: tyrosine.
- (a) Molecular docking studies of a tyrosine molecule with insulin. The complex shows seven interactions comprising of four hydrogen bonds (B:ARG22:HE—L-tyrosine:O14, B:A RG22:HH21—L-tyrosine:O14, B:GLU21:OE1—L-tyrosine:H21, B:VAL18:O—L-tyrosine:H24), two pi interactions (B:GLU21:HN—L-tyrosine, B:GLY20:HA1—L-tyrosine) and one electrostatic bond (B:GLU21:OE2—L-tyrosine:N13). (b) Analysis of a crystal structure of insulin dimer to visualize the binding partners of tyrosine residue of one insulin molecule with different functional groups of another insulin molecule at the interface region. Discovery Studio 4.0 was used for the docking studies.
- 5.5 Molecular dynamics studies on (◆) insulin-alanine, (◆) insulin-tryptophan and (◆) insulin-tyrosine docked complexes. (a) Energy vs time graph showing the stability of complexes over 5000 picoseconds, where tyrosine-insulin complex was found to be more stable. (b)
 RMSD vs time graph of the docked complexes. Discovery Studio 4.0 was used for the molecular docking studies.
- 5.6 Molecular docking studies, represents interaction complexes of: (a) alanine-insulin (CDocker energy= -23.4018 and Interaction energy= -14.0875 Kcal/mol); and (b) tryptophaninsulin (CDocker energy= -23.93 and Interaction energy= -17.0 Kcal/mol). Histogram represents fraction of participation of all the protein residues involved while making the interactions with ligand. Fraction of participation was calculated for all the amino acids participating in bond formation, when best 50 poses of ligand is docked with the insulin molecule.
- 5.7 Analyzing molecular interactions of insulin dimer (PDB ID: 2A3G). (a) Represents interface 50 of insulin dimer, where two insulin monomers are interacting by their B-chains via H-bonds.
 (b) Represents all the amino acid residues that are taking part in making H-bonds to stabilize the insulin dimer. (c) Represents all the details of the molecular interaction taking place such as, atoms that are taking part in bond formation, residues taking part, chain of insulin involved, bond length and type of bond.
- 5.8 Analyzing molecular interactions of insulin dimer (PDB ID: 2INS). (a) Represents interface of insulin dimer, where two insulin monomers are interacting by their B-chains via H-bonds.
 (b) Represents all the amino acid residues that are taking part in making H-bonds to stabilize the insulin dimer. (c) Represents all the details of the molecular interaction taking place such as, atoms that are taking part in bond formation, residues taking part, chain of insulin involved, bond length and type of bond.
- 5.9 Analyzing molecular interactions of insulin dimer (PDB ID: 2ZP6). (a) Represents interface 52 of insulin dimer, where two insulin monomers are interacting by their B-chains via H-bonds.
 (b) Represents all the amino acid residues that are taking part in making H-bonds to stabilize the insulin dimer. (c) Represents all the details of the molecular interaction taking place such as, atoms that are taking part in bond formation, residues taking part, chain of insulin involved, bond length and type of bond.
- 5.10 Analyzing molecular interactions of insulin dimer (PDB ID: 4IDW). (a) Represents interface 53 of insulin dimer, where two insulin monomers are interacting by their B-chains via H-bonds.
 (b) Represents all the amino acid residues that are taking part in making H-bonds to stabilize the insulin dimer. (c) Represents all the details of the molecular interaction taking place such as, atoms that are taking part in bond formation, residues taking part, chain of

Page 47

insulin involved, bond length and type of bond.

- Figures Title
- 5.11 Statistical analysis of crystallographic structures of selected insulin dimers (2A3G, 2INS, 2ZP6 and 4IWD), reported in PDB. (a) Fraction of participation of amino acids, involved in interface interactions among insulin dimers. (b) Represents Fraction of participation of interacting amino acid pairs, involved in interface interactions among insulin dimers. Fraction of participation was calculated by taking an average of all the interaction taken place in all four dimers.

•••

5.12 Schematics representation of insulin amyloid inhibition by Trp and Tyr coated gold NPs.