Supplementary Material

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Additonal Calculations

Born-Onsager calculations. We are interested in the contribution of ΔG_{els} to $\Delta H_{n^{th}}$, which we construct as $\Delta H_{\text{els},n^{th}} = \Delta G_{\text{els}}(\text{P}_n) - \Delta G_{\text{els}}(\text{P}_{n-1}) - \Delta G_{\text{els}}(\text{P}_1)$, using the electrostatic solvation energy of the n-mer, $(n-1)$-mer, and monomer, respectively. The molecular volume of the oligomers studied was linearly proportional ($R^2 = 0.999$) to the number of peptides comprising the oligomer (n), and, as the oligomers were roughly globular, we can use the volume of a sphere to express the effective solute radius (a) as a function of n: $V = bn = 4\pi a^3/3$ implies that $a = cn^{1/3}$, where b and c are proportionality constants (linear regression of oligomer molecular volume against n yielded $c = 5.88$ Å). Substituting $cn^{1/3}$ for a into the Born-Onsager relation we determine $\Delta H_{\text{els},n^{th}}$ as a function of n for a few different scenarios.

For peptides with zero net charge, $q = 0$ and we need only consider the dipole term in the Born-Onsager equation. For the formation of parallel and antiparallel β-sheets, we will assume that each β-strand has an equal electric dipole moment of μ and that the dipoles of adjacent β-strands are aligned either perfectly parallel or antiparallel, respectively. The net dipole moment of a parallel sheet of n strands is then simply $n\mu$ as the dipoles all add up constructively; for an antiparallel sheet the net dipole is μ for odd values of n and zero for even values of n as adjacent dipoles cancel each other out in a destructive fashion. (In the case of twisted β-sheets the angle between adjacent strands can be as large as 15° to 20° so that the dipoles are not perfectly aligned, and thus the assumptions used here should be regarded as constituting a first approximation.) For parallel β-sheets it turns out that there is no electrostatic solvation contribution to $\Delta H_{n^{th}}$:

$$\Delta H_{\mathrm{els},n^{th}} = \frac{-(n\mu)^2}{2c^3 n} - \frac{-[\mu(n-1)]^2}{2c^3(n-1)} - \frac{-\mu^2}{2c^3} = 0. \quad \textbf{[1]}$$

Whereas for antiparallel β-sheets we have

$$\Delta H_{\mathrm{els},n^{th}} = \begin{cases} -\frac{-\mu^2}{2c^3} - \frac{-\mu^2}{2c^3(n-1)} = \frac{\mu^2}{2c^3}\left(1 + \frac{1}{n-1}\right), & n \text{ even} \\ \frac{-\mu^2}{2c^3 n} - \frac{-\mu^2}{2c^3} = \frac{\mu^2}{2c^3}\left(1 - \frac{1}{n}\right), & n \text{ odd.} \end{cases} \quad \textbf{[2]}$$

In **Eq. 2** $\Delta H_{\mathrm{els},n^{th}}$ is positive for $n \geq 2$ and undergoes damped oscillations about $\mu^2/2c^3$, meaning that electrostatic solvation in this case disfavors aggregation. Using c = 5.88 Å and μ = 6.4 Debye (the average dipole moment for STVIYE β-strands in the octameric starting structure), we can obtain an estimate of the limiting value of $\Delta H_{\mathrm{els},n^{th}}$, namely $\mu^2/2c^3$ = 1.4 kcal/mol. (To obtain a more rigorous value one would have to use a value for c parameterized to give good agreement with experiment rather than using a value based on considerations of the molecular volume.) For the aggregation of STVIYE peptides in random orientations to form amorphous oligomers the dipoles will not be perfectly aligned either constructively or destructively, and hence one would expect $\Delta H_{\mathrm{els},n^{th}}$ in this case to lie somewhere intermediate between the values for parallel and antiparallel sheets (*i.e.*, zero and $\mu^2/2c^3$). It therefore seems reasonable that the destabilization of antiparallel STVIYE β-sheets relative to the amorphous oligomers is of the same order of magnitude. Indeed, for STVIYE the β-sheet $\Delta H_{n^{th}}$ values were on average 1.2 kcal/mol weaker than the amorphous values (**Fig. S2B**).

In the case of charged peptides, $q \neq 0$ and the monopole term in the Born-Onsager equation dominates since it scales as the inverse radius while the dipole term scales as the

inverse radius cubed. For the binding of the n^{th} monomer (of net charge q) in any orientation to an oligomer of $n-1$ peptides to form an oligomer of n peptides we have,

$$\begin{aligned}\Delta H_{\mathrm{els},n^{th}} &= \frac{-(nq)^2}{2cn^{1/3}} - \frac{-\left[q(n-1)\right]^2}{2c(n-1)^{1/3}} - \frac{-q^2}{2c} \\ &= \frac{q^2}{2c}\left[1+(n-1)^{5/3}-n^{5/3}\right].\end{aligned} \qquad \textbf{[3]}$$

The expression in **Eq. 3** is a negative and monotonically decreasing function of n for $n \geq 2$, meaning that there is a favorable contribution to aggregation and that there is cooperativity in this contribution. At large n one would intuitively expect this contribution to level off, however, rather than keep decreasing monotonically. To get an estimate of how much electrostatic solvation would contribute to $\Delta H_{n^{th}}$ in the case of STVIYE$^{(-)}$, we computed $\Delta H_{\mathrm{els},n^{th}}$ for n = 2 to 8 using q = –1 and c = 5.88 Å (**Fig. S4**). We used Coulomb's law to compute the contribution to $\Delta H_{n^{th}}$ coming from the electrostatic repulsion of having point charges of q = –1 at the ends of antiparallel β-strands in the structures in **Fig. 2**, which we denote $\Delta H_{\mathrm{elr},n^{th}}$ (for this we considered each β-sheet as consisting of a planar antiparallel array of 20 Å straight line segments placed 4.7 Å apart, the second sheet being displaced 10 Å above and antiparallel with respect to the first, and 1 ε_0 was used for the dielectric constant). A high degree of cooperativity was observed in this repulsive component for n = 2 to 8 (**Fig. S4**), and, as before, one would expect this curve to level off in the limit of large n. The near correspondence of the two curves in **Fig. S4** suggests that electrostatic solvation could provide just the right energy needed to overcome charge repulsion during the aggregation of terminally charged peptides such as STVIYE$^{(-)}$ into antiparallel β-sheet networks.

Entropy calculations. Here we consider the rotational, translational, and conformational contributions to $\Delta S_{n^{th}}$, the entropy change for the reaction

$$\mathrm{P}_1 + \mathrm{P}_{n-1} \rightleftharpoons \mathrm{P}_n . \qquad \textbf{[4]}$$

We can use the following relation from classical statistical mechanics for the rotational entropy of an asymmetric polyatomic molecule in terms of its principal moments of inertia I_{xx}, I_{yy}, and I_{zz}:

$$S_{rot} = \tfrac{1}{2} R \ln\left[e^3 \pi^7 \left(8 k_B T\right)^3 h^{-6} I_{xx} I_{yy} I_{zz} \right] \qquad \textbf{[5]}$$

where R is the ideal gas constant. The classical expression for translational entropy, the Sackur-Tetrode equation, is inappropriate, however, as it overestimates the translational entropy of solutes in solution by as much as 20% to 40%.[1] Recently, Siebert and Amzel constructed an extension of free volume theory to predict the translational entropy of organic solutes.[2] Using their formalism we have for the translational entropy of N_ξ solutes immersed in N_w solvent molecules

$$S_\xi^{trsl} = k_B N_\xi \ln \frac{\left(N_w + N_\xi\right) \upsilon_{f,\xi} e^{5/2}}{N_\xi \Lambda_\xi^{\,3}}, \qquad \Lambda_\xi = \left(2\pi m_\xi k_B T\right)^{-1/2} h \qquad \textbf{[6]}$$

where m_ξ is the solute molecular mass and υ_f is the "free volume", defined as the volume accessible to the solute center of mass in the presence of solvent.

Due to the logarithmic dependence of the entropies on the principal moments and the mass, it turns out that $S_{\mathrm{rot}}(\mathrm{P}_{n-1}) \approx S_{\mathrm{rot}}(\mathrm{P}_n)$ and $S_{\mathrm{trsl}}(\mathrm{P}_{n-1}) \approx S_{\mathrm{trsl}}(\mathrm{P}_n)$. Thus, assuming the residual rotational and translational entropy of P_1 within the newly formed complex is negligible, to a good approximation $\Delta S_{\mathrm{rot},n^{th}} = -S_{\mathrm{rot}}(\mathrm{P}_1)$, $\Delta S_{\mathrm{trsl},n^{th}} = -S_{\mathrm{trsl}}(\mathrm{P}_1)$, and the two contributions are independent of n. **Eq. 5** was evaluated for monomeric STVIYE using the moments of inertia of the final snapshot of the 4 ns monomer simulation, yielding

$T\Delta S_{\text{rot},n^{th}}$ = –11.8 kcal/mol (at 300 K). For the evaluation of **Eq. 6** we assumed a concentration of 1 mM, the concentration at which the peptide is observed to form fibrils experimentally[3] (note that entropies, and consequently free energies, reported in this work do not represent standard state values). Using υ_f = 0.03 Å^3, the free volume Siebert and Amzel postulate for macromolecules such as polypeptides, we obtain for STVIYE (m_{ξ} = 751 Daltons) $T\Delta S_{\text{trsl},n^{th}}$ = –11.8 kcal/mol, which seems reasonable in light of the fact that it is 25% less than the value that is obtained if the Sackur-Tetrode equation is used.

There is one additional consideration in store in light of the above calculations. The enthalpy of an ideal, asymmetric polyatomic gas due to its rotational and translational motion is given by $H_{\text{rot+trsl}} = 4RT$;[4] we therefore lose $4RT$ (2.4 kcal/mol) in enthalpy for the reaction in **Eq. 4** due to this contribution. The enthalpy values reported in this work were calculated from MD trajectories using only the potential energy, so to take this contribution into account we simply add it into our estimation of $T\Delta S_{n^{th}}$.

Finally, we consider conformational contributions to $\Delta S_{n^{th}}$. Recent simulations of small peptides[5] indicate that the polypeptide backbone has on average 1.51 cal/(mol-K) of conformational entropy per residue in the random coil state at 300 K; similarly, the conformational entropy due to sidechain motions in the random coil state is shown to be on average 2.17 cal/(mol-K) per nontrivial sidechain torsion angle. Using these numbers, if we assume that when P_1 binds P_{n-1} it loses all of its conformational entropy without affecting the conformational entropy of P_{n-1}, we then have for STVIYE: $T\Delta S_{\text{conf},n^{th}} = -TS_{\text{conf}}(P_1)$ = –9.2 kcal/mol. Quasiharmonic analysis[6] was performed in Cartesian coordinates in CHARMM in order to validate that $\Delta S_{\text{conf},n^{th}}$ was indeed independent of n. The conformational entropy of each amorphous n-mer (for n = 1 to 8) was calculated by

analyzing each snapshot in the last 3 ns of simulation. More precisely, $S_{conf}(P_n)$ was obtained by taking the sum of the entropies computed in CHARMM for each peptide in a given *n*-mer. The conformational component of $\Delta S_{n^{th}}$ was then computed as $\Delta S_{conf,n^{th}} = S_{conf}(P_n) - S_{conf}(P_{n-1}) - S_{conf}(P_1)$ for STVIYE$^{(-)}$ and STVIYE (**Fig. S5**). Intuitively, one might expect $\Delta S_{conf,n^{th}}$ to decrease with increasing *n* as a monomer is more likely to lose all of its conformational entropy when binding to larger oligomers, but the data in **Fig. S5** does not support this.

Adding up all the contributions to $\Delta S_{n^{th}}$ discussed above we have, finally, $T\Delta S_{n^{th}} = T\Delta S_{rot,n^{th}} + T\Delta S_{trsl,n^{th}} + T\Delta S_{conf,n^{th}} + 4RT$ = (–11.8 – 11.8 – 9.2 + 2.4) kcal/mol = –30.4 kcal/mol, which is in reasonable agreement with the value of –28.5 kcal/mol we arrived at using **Fig. S6**.

Supplementary Figures

Fig. S1.

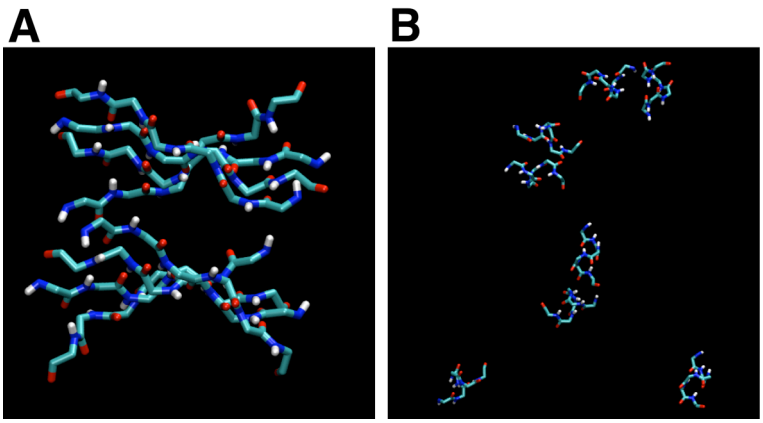

Fig. S1. Starting structures used in REMD. Sidechain atoms are omitted for clarity. **(A)** Equilibrated β-sheet octamer used to generate starting structures for β-sheet oligomer simulations. **(B)** Random configuration of eight peptide monomers used to generate starting structures for amorphous aggregate simulations.

Fig. S2.

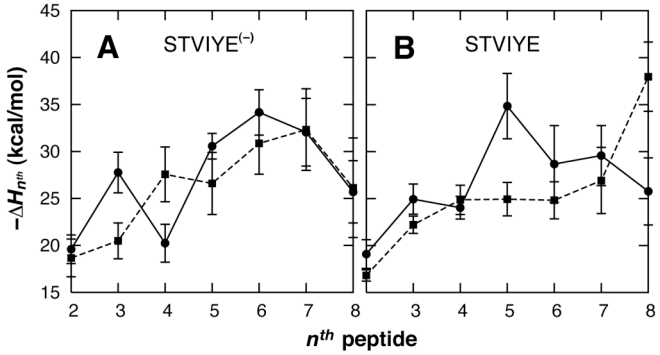

Fig. S2. Association enthalpies. The $\Delta H_{n^{th}}$ values in **Fig. 3** are plotted here for STVIYE$^{(-)}$ **(A)** and STVIYE **(B)** simulations, this time comparing amorphous aggregates (circles) with β-sheet oligomers (squares).

Fig. S3.

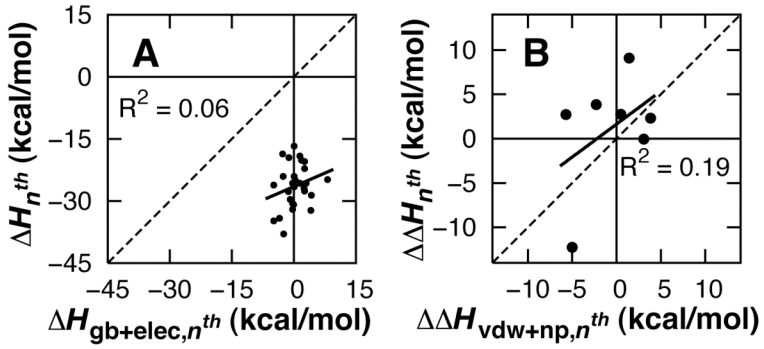

Fig. S3. Correlations of the $\Delta H_{n^{th}}$ and $\Delta\Delta H_{n^{th}}$ values in **Fig. 4** with alternative energy components; regression lines (solid) are contrasted with the identity line $y = x$ (dashed). **(A)** The electrostatic/polar solvation contribution to $\Delta H_{n^{th}}$, $\Delta H_{\mathrm{gb+elec},n^{th}}$, fluctuated about zero and was an order of magnitude smaller than $\Delta H_{n^{th}}$, having minor effects on the magnitude of the association enthalpy. **(B)** Differences between the β-sheet and amorphous enthalpies for STVIYE ($\Delta\Delta H_{n^{th}}$) were not highly correlated with shifts in the hydrophobic component of the enthalpy ($\Delta\Delta H_{\mathrm{vdw+np},n^{th}}$).

Fig. S4.

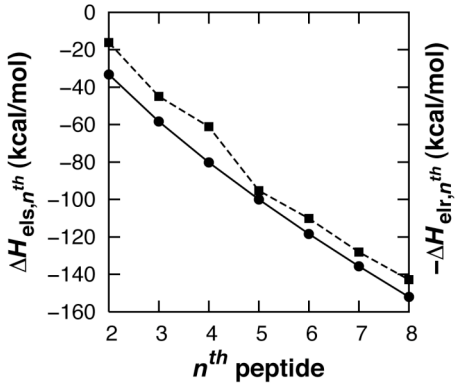

Fig. S4. Charged peptide aggregation. For STVIYE$^{(-)}$, the predicted contribution of electrostatic solvation to $\Delta H_{n^{th}}$, $\Delta H_{\mathrm{els},n^{th}}$ (circles), is seen to overcome the electrostatic repulsion between charged termini in an antiparallel sheet structure, $\Delta H_{\mathrm{elr},n^{th}}$ (squares).

Fig. S5.

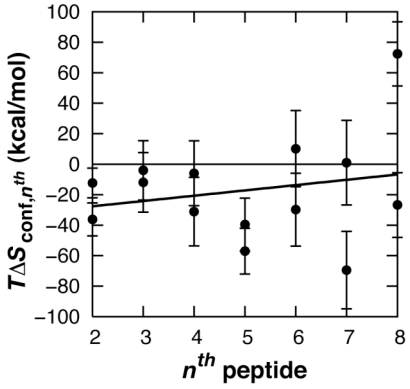

Fig. S5. Conformational entropy calculations. The $\Delta S_{\mathrm{conf},n^{th}}$ values for the amorphous oligomers of STVIYE$^{(-)}$ and STVIYE were not observed to become more negative with increasing n, as evidenced by the upward sloping regression line.

Fig. S6.

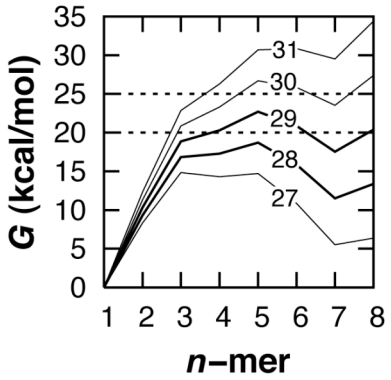

Fig. S6. Oligomer free energies expressed relative to the monomeric state for data set I. Five profiles are given for $T\Delta S_{n^{th}} = -27$ to -31 kcal/mol; 28 or 29 kcal/mol (emboldened) is our best estimate of $-T\Delta S_{n^{th}}$.

Fig. S7.

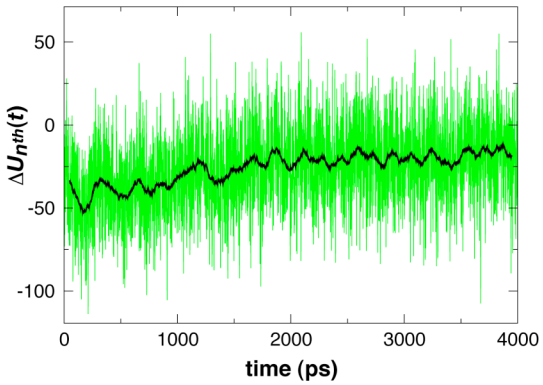

Fig. S7. Instantaneous potential energy difference (green) for the reaction in **Eq. 4** during the course of the amorphous $n = 4$ simulation of STVIYE$^{(-)}$. The enthalpy plateaus after 2 ns, as is more easily seen from the 100-point running average (black).

References

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