

Supporting Information

Self-assembly of Alanine-Isoleucine and Isoleucine-Isoleucine Dipeptides through Atomistic Simulations and Experiments

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Size and Internal Conformations of Dipeptides

The mean size of the four dipeptides is quantified through the calculation of their radius of gyration: $\langle R_g \rangle = \sqrt{\left\langle \frac{\sum_i m_i (r_i - R_{cm})^2}{\sum_i m_i} \right\rangle}$. The corresponding values in water are: Phe-Phe $0.34 \pm 0.001 \text{ nm}$, Ala-Ile $0.273 \pm 0.001 \text{ nm}$, Ala-Ala $0.26 \pm 0.001 \text{ nm}$, Ile-Ile $0.32 \pm 0.001 \text{ nm}$. Phe-Phe and Ile-Ile are slightly more extended compared to the rest two systems.

The radius of gyration for the different systems in methanol are: Phe-Phe $0.397 \pm 0.001 \text{ nm}$, Ala-Ile $0.275 \pm 0.0001 \text{ nm}$, Ala-Ala $0.25 \pm 0.00003 \text{ nm}$, Ile-Ile $0.314 \pm 0.0001 \text{ nm}$. Phe-Phe and Ile-Ile in methanol are also slightly more extended compared to the rest two systems.

Furthermore the distribution of the backbone dihedral angle which connects the two peptides (i.e., between the carbon atoms where side groups are bonded) is calculated from the crystal structures (i.e., initial configurations) and is presented in Figure SI-1 for the three dipeptides of “Val-Ala” class.

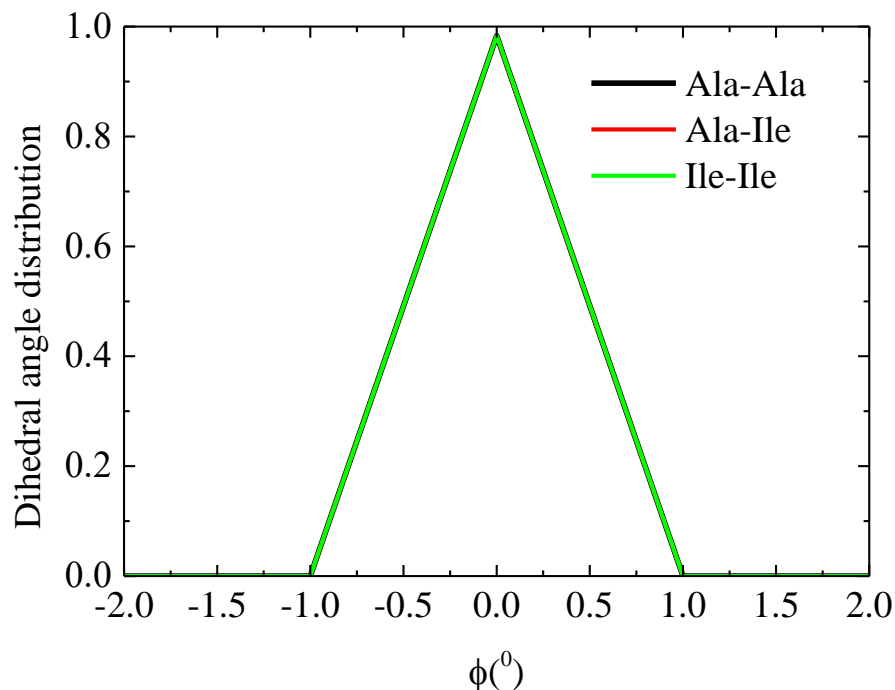


Figure SI-1. Distribution of backbone dihedral angle for Ala-Ala, Ala-Ile and Ile-Ile dipeptides.

Orientation of dipeptides

Antiparallel orientation of Phe-Phe and Ala-Ala dipeptides at short distances (i.e., $[0.4-0.5]nm$) in water has been observed both in our previous work¹ and other simulation studies.²⁻³ This preference is driven by the electrostatic interactions between the charged end groups. A similar analysis has been performed for Ala-Ile and Ile-Ile systems at center of mass – center of mass distances (Dr) between dipeptides in water in the range $[0.5-0.8]nm$. The preferable orientation of molecules is quantified by the dot product of the end to end vectors of the two molecules. The probability distribution of θ – value, $P(\theta)$, for the two systems, is presented in Figure SI-2. For both dipeptides an antiparallel orientation is favoured at short distances (i.e. $[0.5-0.6] nm$), where the distributions peaks around $\sim 160^\circ$. At longer distances (i.e., $0.8nm$) distribution curves become broad and orientation is almost randomized.

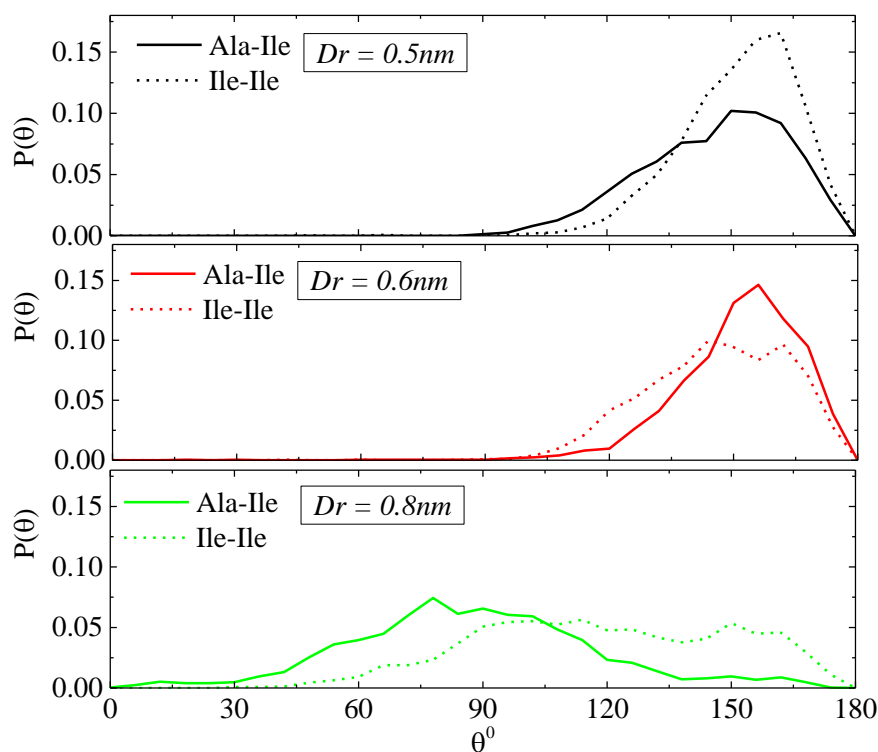


Figure SI-2. Probable orientations between a pair of dipeptides in water at different cm–cm constant distances (Dr).

Electrostatics Interactions

The strength of total electrostatic interactions (i.e., intramolecular and intermolecular), as it is quantified through the Coulomb energy, is presented in Figure SI-3a and follows the order: Phe-Phe > Ile-Ile > Ala-Ile > Ala-Ala. Comparison between the two solvents is shown in Figure SI-3b for Ala-Ile system.

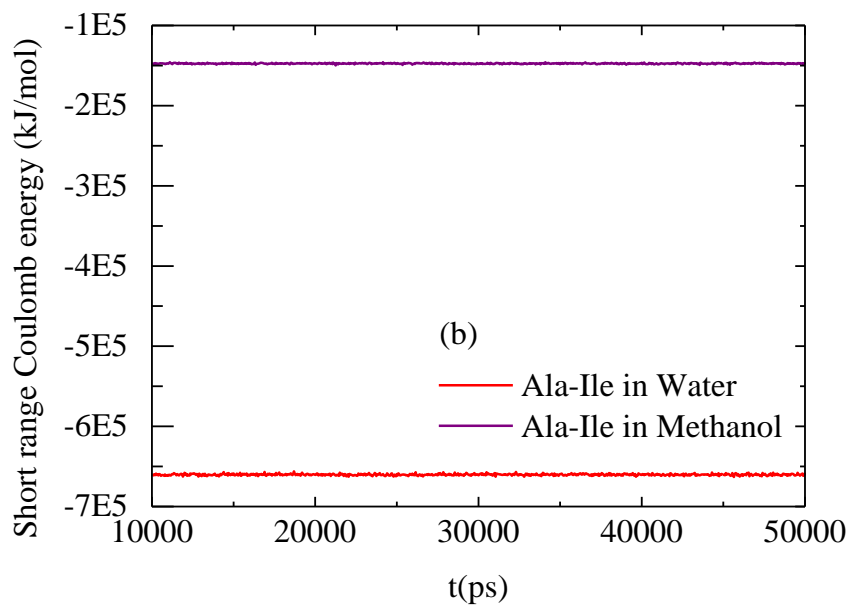
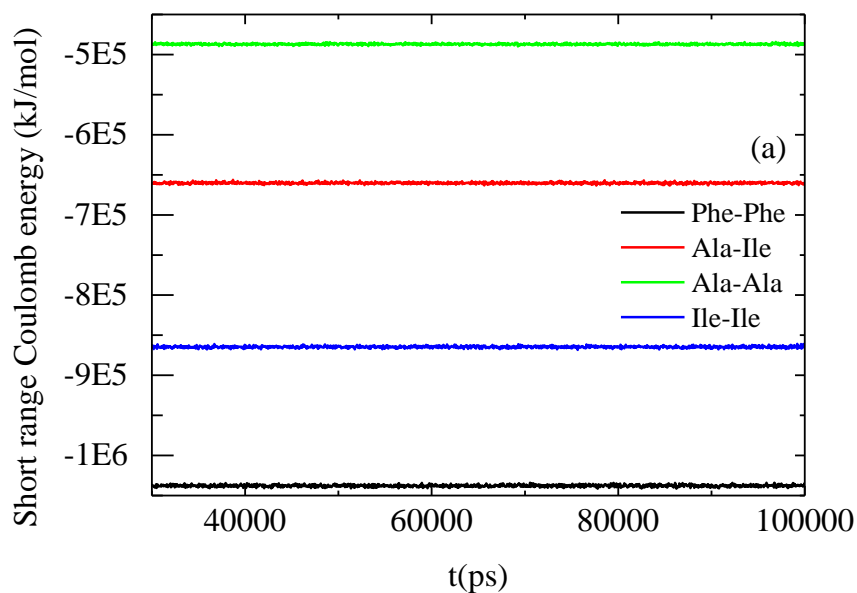


Figure SI-3. (a) The short range Coulomb energy as a function of time for the four dipeptide systems in water at 300K. (b) The short range Coulomb energy as a function of time for Ala-Ile dipeptide in water and in methanol at 300K

Kinetics of Self-Assembly

Supporting information on the above observations, is the way that the number of water molecules around each dipeptide changes with time. For this purpose we count the number of water molecules (represented by their center of mass) within a predefined region around each dipeptide molecule. In more details, this counting is performed for all water molecules which are within a distance $\leq 1nm$ from each individual atom of dipeptide and the final number is the sum over all water molecules correspond to each atom. This is a detailed analysis which follows the conformation (shape) of dipeptide, since it is based on each atom. The number of water molecules is expected to be a decreasing function of time when self-assembly takes place in the solution, but it will be almost constant for well dispersed molecules. Results for all four systems are presented in Figure SI-4 and are in agreement with all previous findings. Curves of Figure SI-4 are restricted to short times (i.e., $\sim 1ns$) before an almost constant value is attained. All curves are normalized with their initial value for each system correspondingly. A fast decrease of water molecules (i.e., $>50\%$) is observed. This is more pronounced for Phe-Phe, Ala-Ile follows with a weaker reduction, while for the two other systems not any decrease is observed; the number of water molecules fluctuates around a constant value. Moreover this calculation provides a rough quantification of the time beyond which formed aggregates have attained their average size (i.e., $\sim 600ps$ for Phe-Phe and $\sim 100ps$ for Ala-Ile). Length and time scales of atomistic simulations do not allow the observation of the macroscopic structures however, they allow a quantitative comparison for the dynamics of self-assembly propensity of the different dipeptides in aqueous solutions.

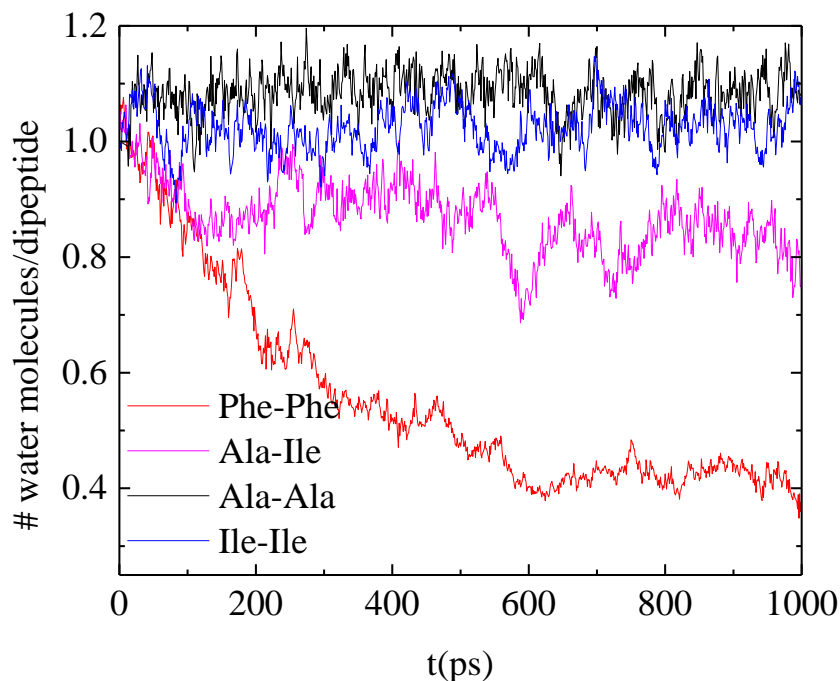


Figure SI-4. The number of water molecules within $1nm$ distance around each dipeptide as a function of time for the four dipeptide systems in water at 300K. Curves are normalized with the initial value for each system correspondingly.

The way that the total number of hydrogen bonds / atom, which are formed in Ala-Ile system changes with time is presented in Figure SI-5a and 5b, for water and methanol solutions correspondingly. Almost double hydrogen bonds are formed in water compared to methanol. In addition larger increase of their value, with respect to the initial configuration, is observed in the aqueous solution, indicating bigger deviation from the uniformly distribution of dipeptides in the solvent (i.e., structure formation).

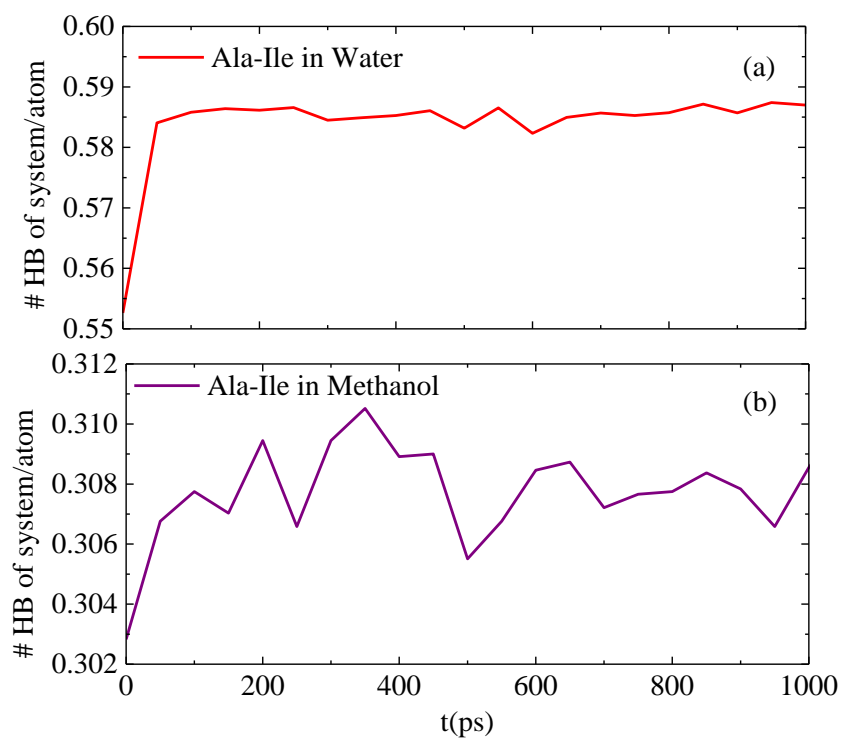


Figure SI-5. The total number of hydrogen bonds per atom in Ala-Ile system at 300K (a) in water; (b) in methanol.

Concentration Effect

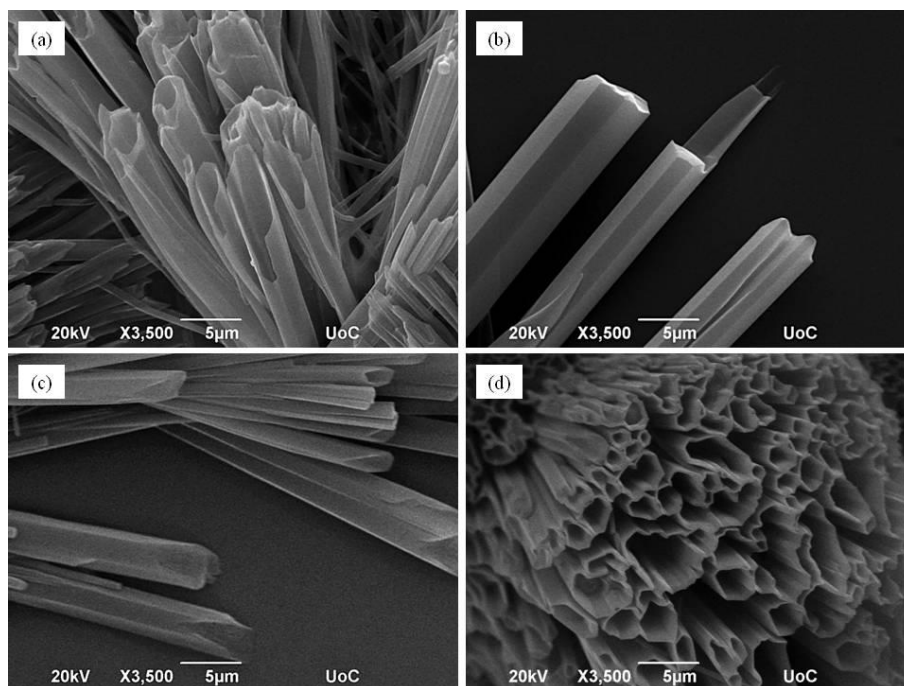


Figure SI-6. SEM pictures of microstructures taken following evaporation at room temperature of an Ala-Ile aqueous solution at concentration of 0.020 g/cm^3 at the following time points: **(a)** 0 min, **(b)** 1 hour **(c)** 2 hours and **(d)** 3 hours.

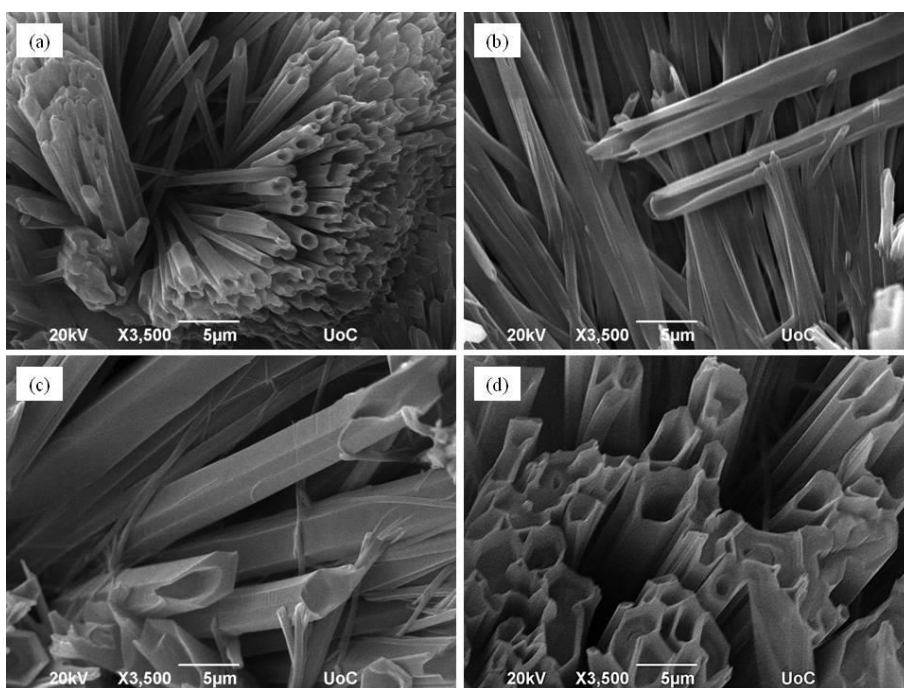


Figure SI-7. SEM pictures of microstructures taken following evaporation at room temperature of an Ala-Ile aqueous solution at concentration of 0.030 g/cm^3 at the following time points: **(a)** 0 min, **(b)** 1 hour **(c)** 2 hours and **(d)** 3 hours.

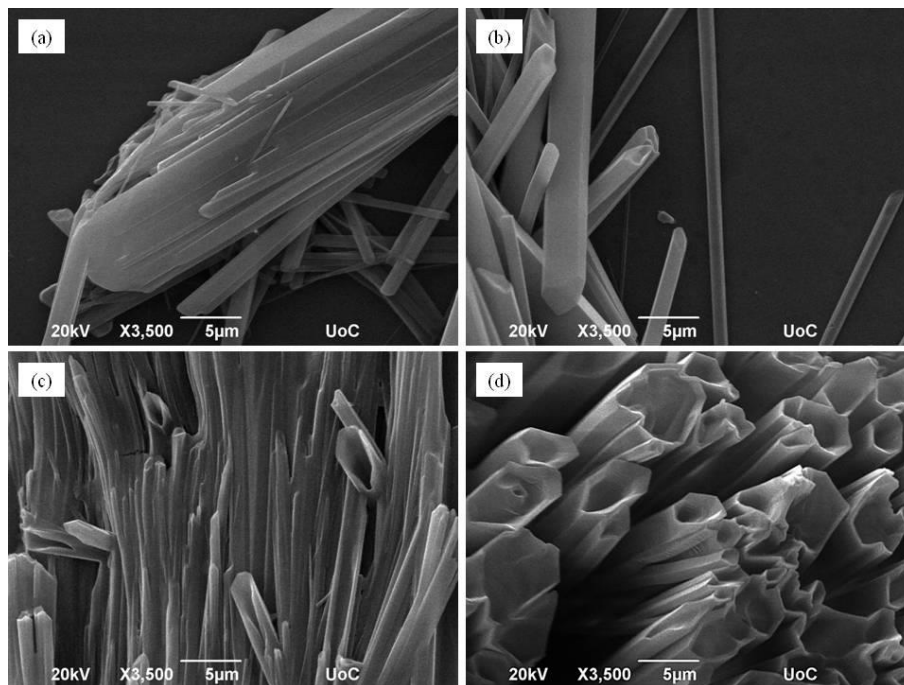


Figure SI-8. SEM pictures of microstructures taken following evaporation at room temperature of an Ala-Ile aqueous solution at concentration of 0.050 g/cm^3 at the following time points: **(a)** 0 min, **(b)** 1 hour **(c)** 2 hours and **(d)** 3 hours.

Temperature Effect

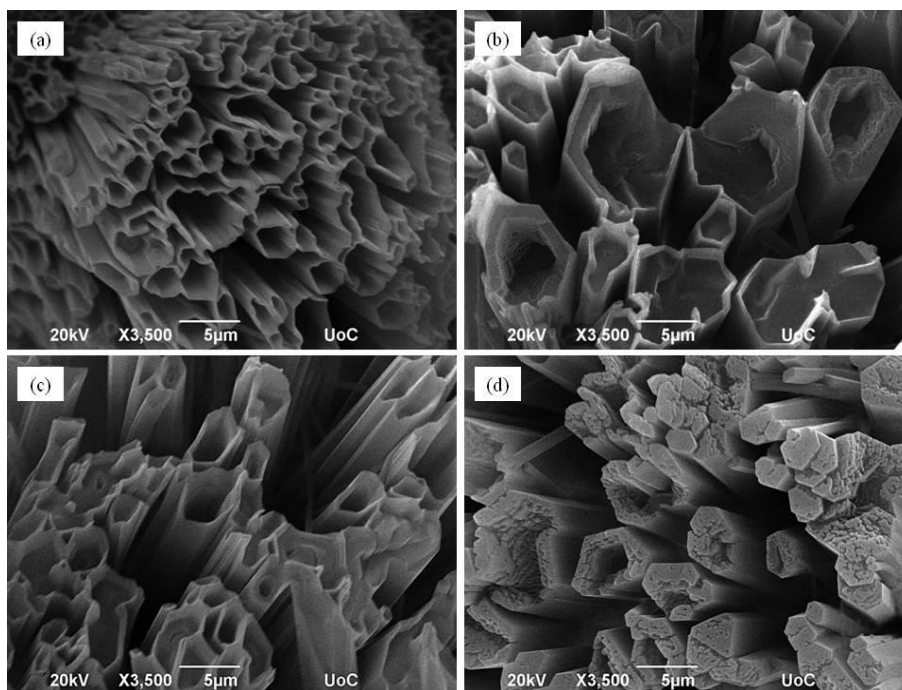


Figure SI-9 : SEM pictures of microstructures taken following evaporation of a 0.020 g/cm^3 Ala-Ile solution in water at room temperature **(a)** and 278K **(b)** and a 0.030 g/cm^3 solution_at room temperature **(c)** and 278K **(d)** .

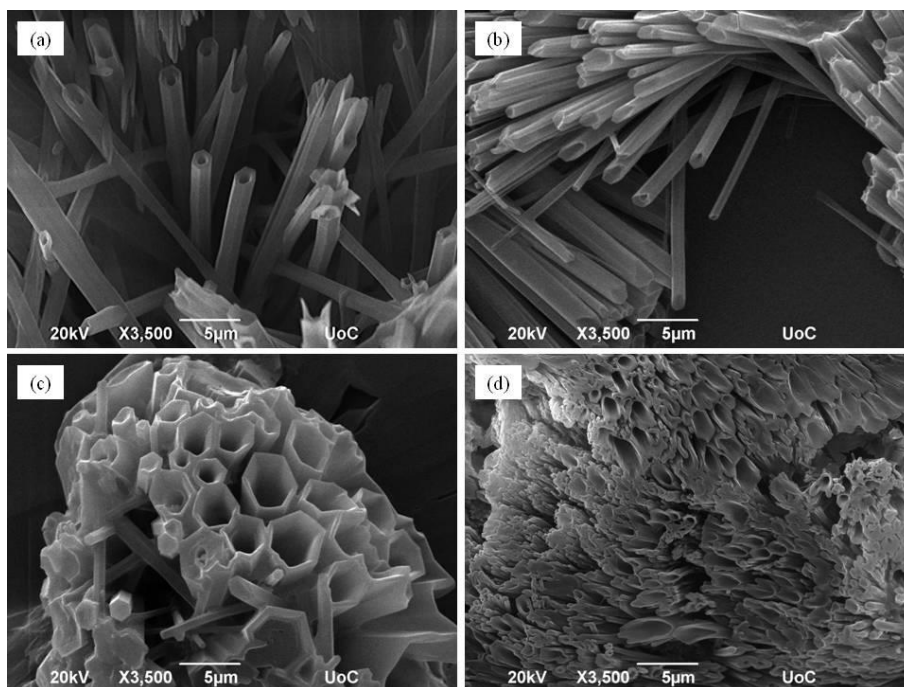


Figure SI-10. SEM pictures of microstructures taken following evaporation at 278K of an Ala-Ile aqueous solution at concentration of 0.002 g/cm^3 at the following time points: (a) 0 min, (b) 1 hour (c) 2 hours and (d) 3 hours.

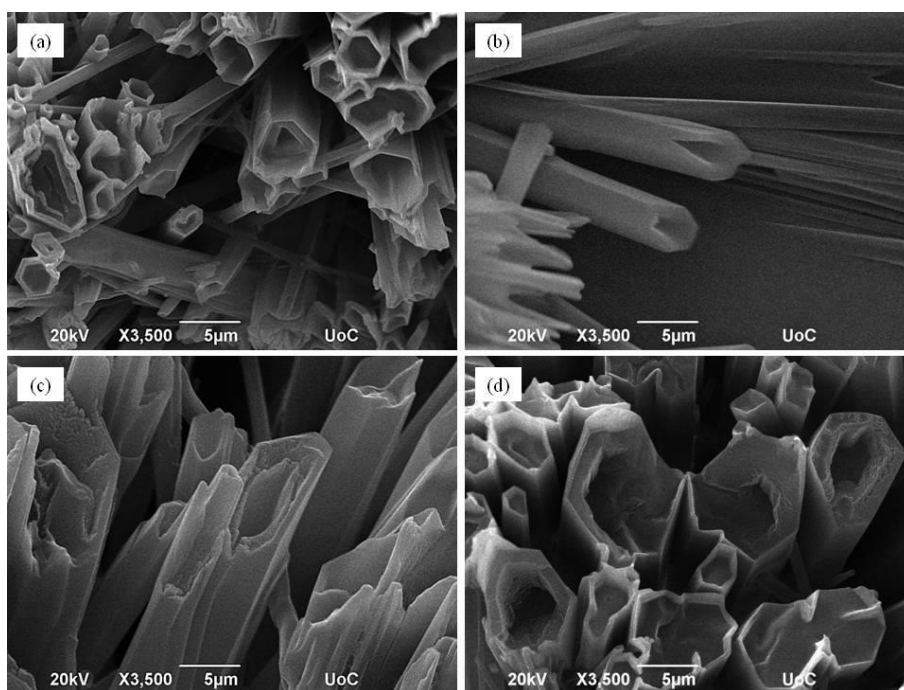


Figure SI-11. SEM pictures of microstructures taken following evaporation at 278K of an Ala-Ile aqueous solution at concentration of 0.020 g/cm^3 at the following time points: (a) 0 min, (b) 1 hour (c) 2 hours and (d) 3 hours.

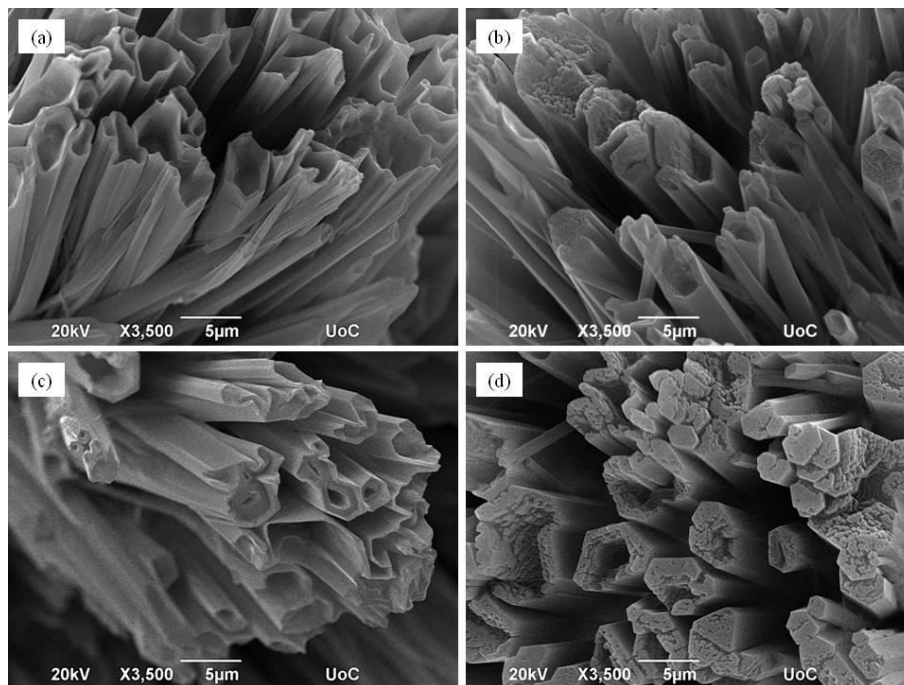


Figure SI-12. SEM pictures of microstructures taken following evaporation at 278K of an Ala-Ile aqueous solution at concentration of 0.030 g/cm^3 at the following time points: (a) 0 min, (b) 1 hour (c) 2 hours and (d) 3 hours.

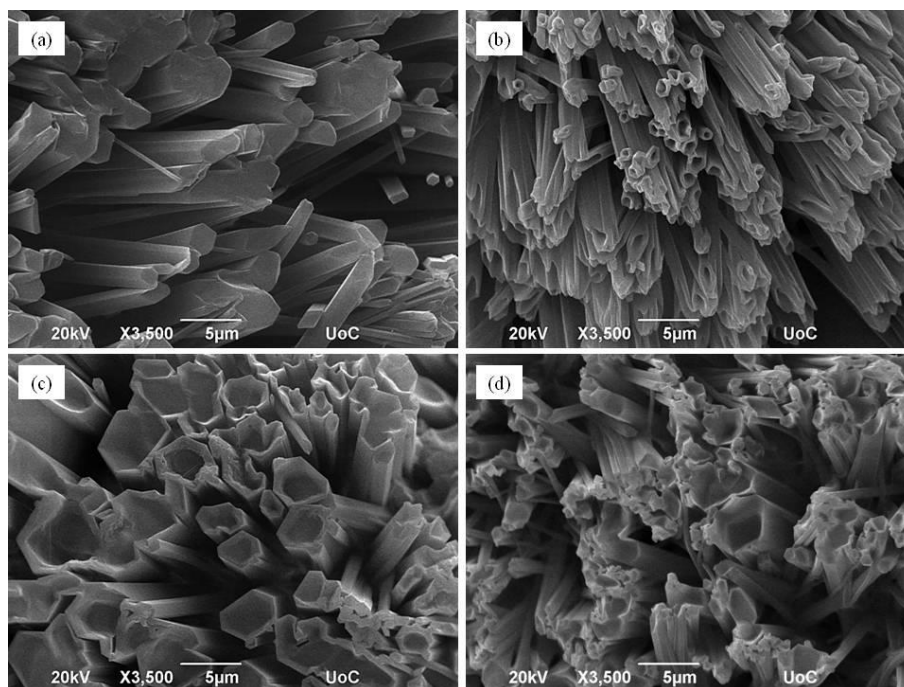


Figure SI-13. SEM pictures of microstructures taken following evaporation at 278K of an Ala-Ile aqueous solution at concentration of 0.050 g/cm³ at the following time points: (a) 0 min, (b) 1 hour (c) 2 hours and (d) 3 hours.

Aqueous vs. Organic Solvent

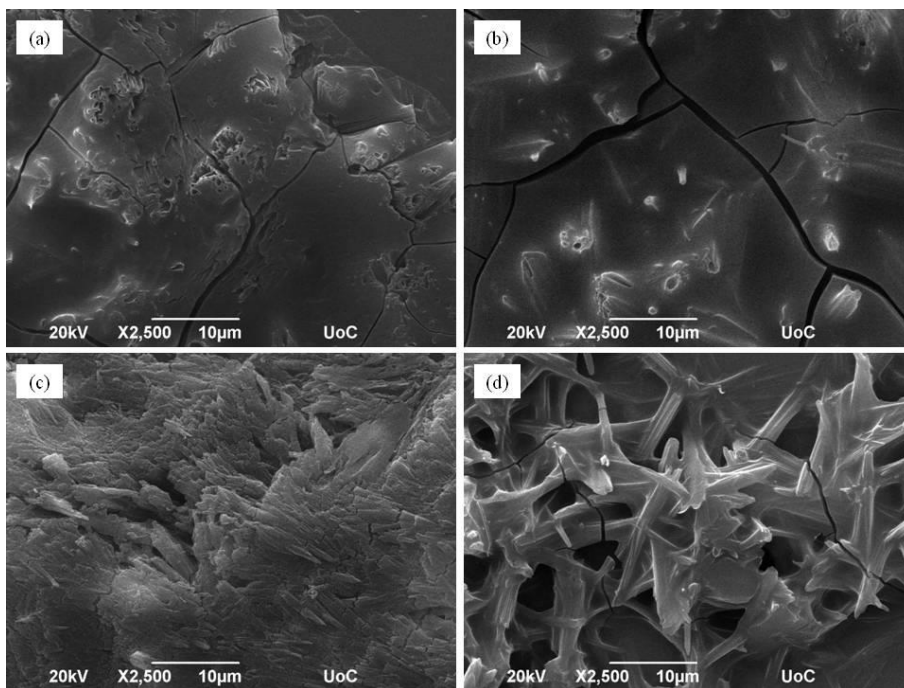


Figure SI-14: SEM pictures of microstructures taken following evaporation of a 0.002 g/cm³ Ile-Ile solution in methanol following incubation at room temperature for 0 min (**a**) and 24 hours (**b**) and at 278K for 0 min (**c**) and 24 h (**d**).

1. Rissanou, A. N.; Georgilis, E.; Kasotakis, E.; Mitraki, A.; Harmandaris, V., Effect of Solvent on the Self-Assembly of Dialanine and Diphenylalanine Peptides. *Journal of Physical Chemistry B* **2013**, *117* (15), 3962-3975.
2. Villa, A.; Peter, C.; van der Vegt, N. F. A., Self-assembling dipeptides: conformational sampling in solvent-free coarse-grained simulation. *Physical Chemistry Chemical Physics* **2009**, *11* (12), 2077.
3. Villa, A.; van der Vegt, N. F. A.; Peter, C., Self-assembling dipeptides: including solvent degrees of freedom in a coarse-grained model. *Physical Chemistry Chemical Physics* **2009**, *11* (12), 2068.