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Meta - Analysis of Self - Assembly of Aromatic Amino Acids: A Molecular Dynamics Study

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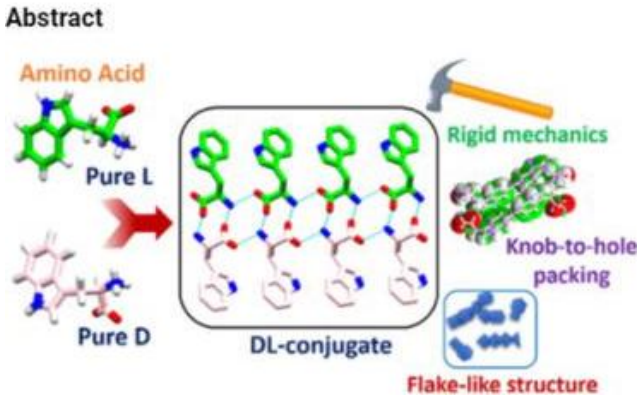
Abstract: ***Most natural biomolecules may exist in either of two enantiomeric forms. Although in nature, amino acid biopolymers are characterized by l-type homochirality, incorporation of d-amino acids in the design of self-assembling peptide motifs has been shown to significantly alter enzyme stability, conformation, self-assembly behavior, cytotoxicity, and even therapeutic activity. However, while functional metabolite assemblies are ubiquitous throughout nature and play numerous important roles including physiological, structural, or catalytic functions, the effect of chirality on the self-assembly nature and function of single amino acids is not yet explored. Herein, we investigated the self-assembly mechanism of amyloid-like structure formation by two aromatic amino acids, phenylalanine (Phe) and tryptophan (Trp), both previously found as extremely important for the nucleation and self-assembly of aggregation-prone peptide regions into functional structures. Employing d-enantiomers, we demonstrate the critical role that amino acid chirality plays in their self-assembly process. The kinetics and morphology of pure enantiomers is completely altered upon their coassembly, allowing to fabricate different nanostructures that are mechanically more robust. Using diverse experimental techniques, we reveal the different molecular arrangement and self-assembly mechanism of the dl-racemic mixtures that resulted in the formation of advanced supramolecular materials. This study provides a simple yet sophisticated engineering model for the fabrication of attractive materials with bionanotechnological applications.***

Keywords: functional metabolite, amino acids, chirality, self-assembly, nanomaterials, bionanotechnology

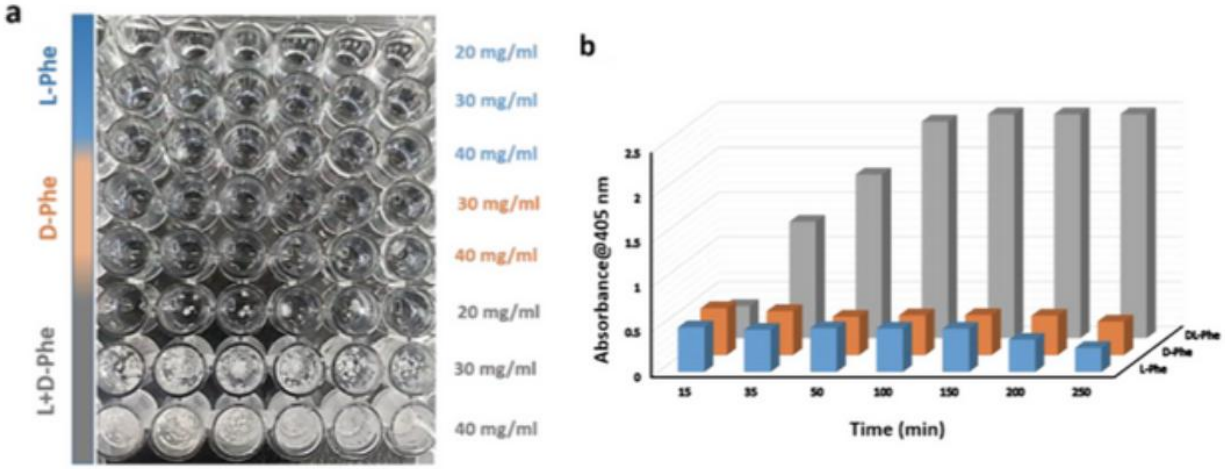
1. Background

In nature, metabolites self-assemble to form nanostructures. Science is trying to mimic this property for advances in material science. Amino acid biopolymers in nature have L-type homochirality. Effects of introducing D-Amino acids alter a lot of properties. In this experiment, Phe and Trp were used in both enantiomeric forms to derive the self-assembly kinetics and the mechanism of structure formation data. The role of chirality in the self-assembly and function of the amino acids and the effect of chirality on the recognition of amino acids was studied.



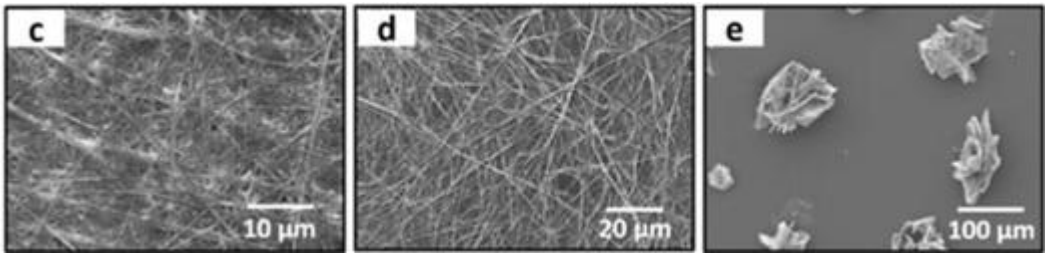
2. Methodology, Results, and Discussion

Self assembly of Phe



Faster rate of aggregation for DL-Phe system

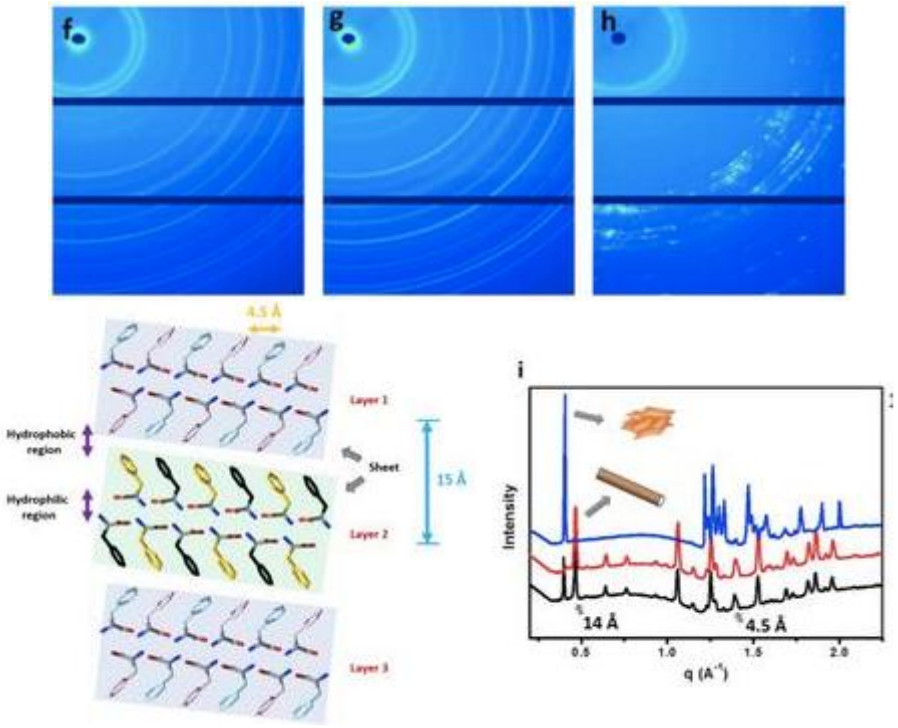
1) Phase behaviour analysis:
L-Phe, D-Phe, and DL-Phe(1:1) were dissolved in double distilled water > Heating to 90˚C > Cooling to room temperature. L-Phe and D-Phe gave clear solutions, and DL-Phe gave a turbid solution. Flake-like structures precipitated. OD analysis was done. High-Resolution Scanning Electron Microscopy was done.



L-Phe Fibers D-Phe Fibers DL-Phe Flake like crystalline structures

2) Structural characterisation

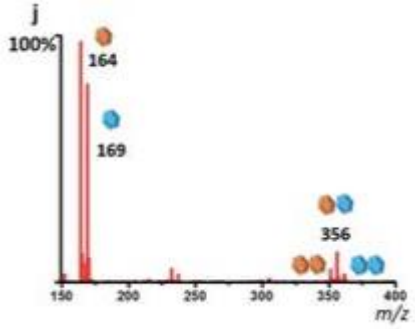
L-Phe and D-Phe fibers and DL-Phe aggregates dried on Kapton film. X-ray scattering experiments were performed. D and L-Phe showed isotropic 2D powder diffraction indicating the random orientation of structures in the plane of the film. DL showed peculiar diffraction, indicating flakes were preferentially oriented in certain directions. The corresponding spectrum showed an altered arrangement of D- and L-Phe molecules in the mixture.

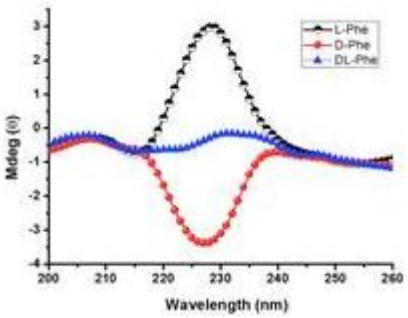


Azimuthally integrated spectra

3) Structural characterization

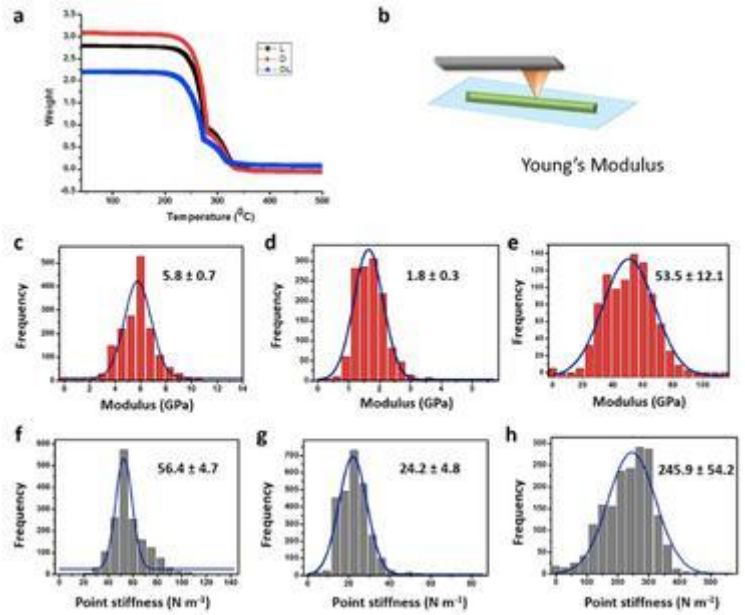
Mass spectroscopy was conducted to show the presence of both the enantiomers in the crystalline aggregates. A mixture of deuterium level L-Phe(d5) with D-Phe (1:1) was taken. m/z peaks at 164 and 169 corresponding to pure D-Phe and L-Phe(d5), a strong signal was observed at 356 emanating from their composite materials. This shows the formation of an enantiomeric conjugate in the mixed system. Coassembly formation was further confirmed by circular dichroism (CD) spectroscopy.





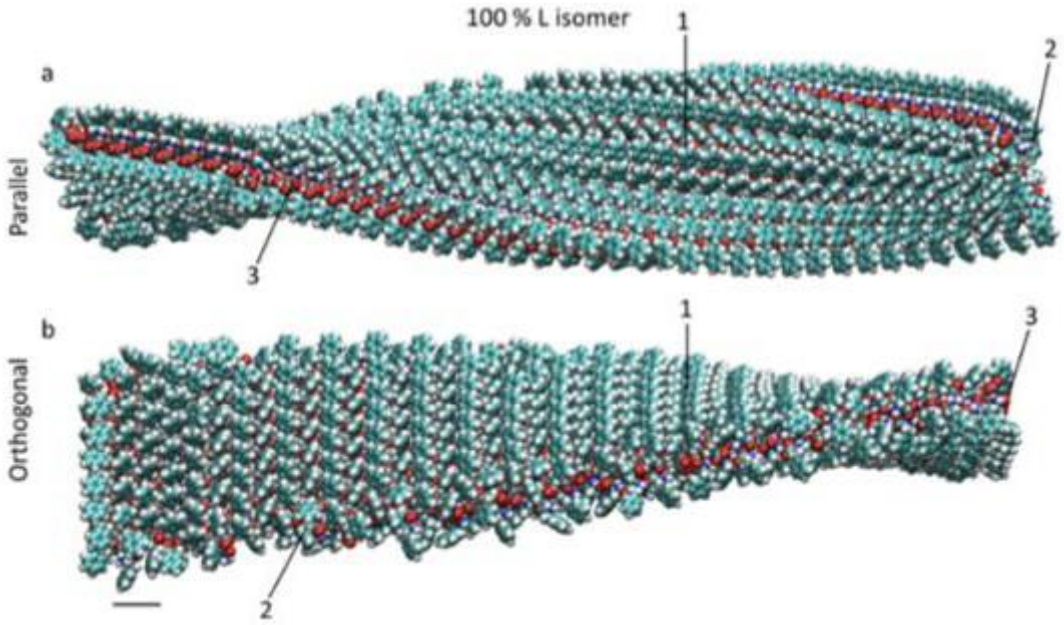
4) Material properties analysis

For thermal stability analysis, thermal gravimetric analysis was done. To describe the mechanical rigidity of the crystals, Young's modulus and point stiffness were calculated using nanoindentation through AFM. Results say that all structures have similar thermal stability, while the mechanical strength of DL-Phe is very high, indicating a strong molecular packing in the DL-mixed system.



5) Modeling Phe crystals

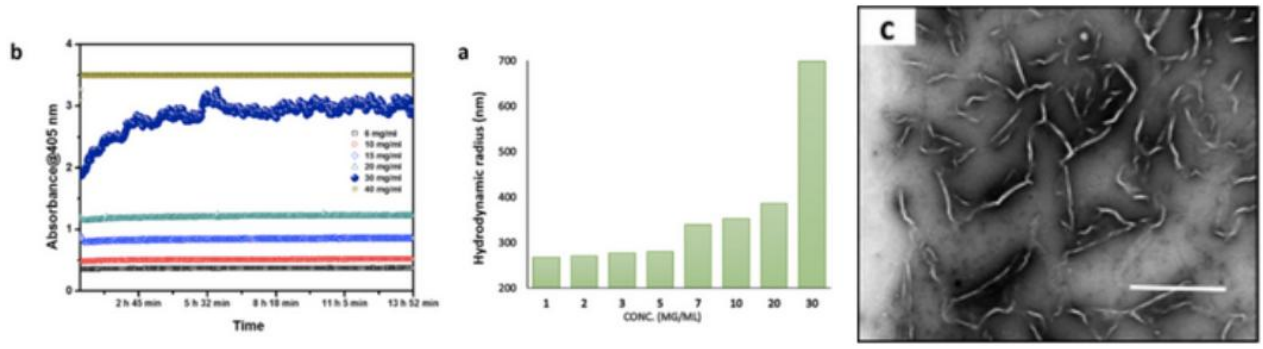
Atomistic molecular dynamics (MD) simulations to model Phe crystals. Only pure enantiomers were modeled because the structure of DL-Phe was unknown. These elongated crystals were cut either along the well-visible aromatic zipper or orthogonal to it. Results show that growth on the facet parallel to the aromatic zipper is more likely to continue and produce twisted 1D crystals. The D-isomer is only a mirror image of the L-isomer.



6) Self assembly of Trp

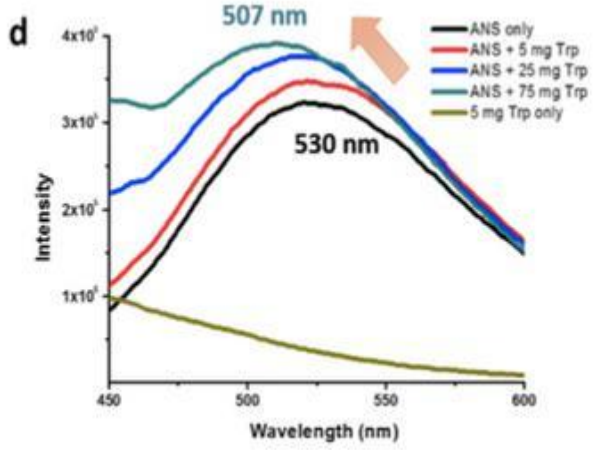
a) Self assembly nature analysis of pure L-Trp

Dynamic light scattering (DLS) was used. The size of the resultant nanostructures at increasing concentrations was measured. Size abruptly increased at a concentration of 30 mg/mL indicating an extensive high aggregation. Self-assembled morphology was studied by transmission electron microscopy (TEM). Molecules stacked into a tubular structure with a 4-fold symmetry and folded into extremely well-organized fibrillar structures were found where hydrophilic termini were positioned internally and the hydrophobic rings resided externally, exposed to the solvent.



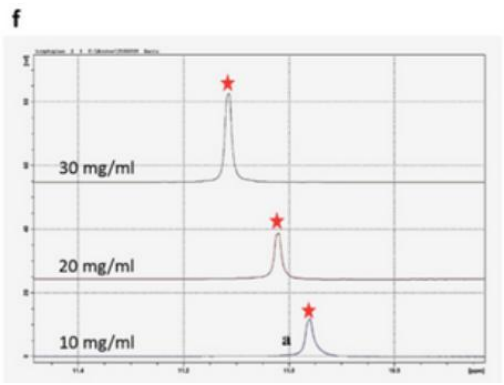
b) Role of the aromatic ring in the self-assembly

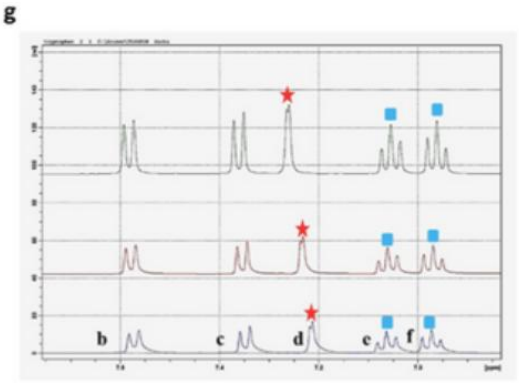
ANS (8-aniline-1-naphthalene sulfonic acid) binding assay was used. Elevated concentrations of L-Trp, showed an increase in the fluorescence intensity of ANS. This indicates a change in the environment toward higher hydrophobicity, probably due to aromatic interactions.

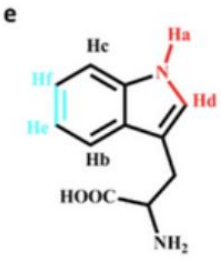


c) Role of the aromatic protons in the self-assembly

Concentration-dependent 1H NMR spectroscopy was performed. Increasing the concentration resulted in an upfield shift of the aryl proton, He, and Hf, indicating their screening due to π−π interactions between aromatic rings. The indole N−H proton (Ha) and the nearby proton(Hd) shifted toward a higher ppm value, which specified the involvement of the N−Ha proton in hydrogen bonding at higher concentrations.

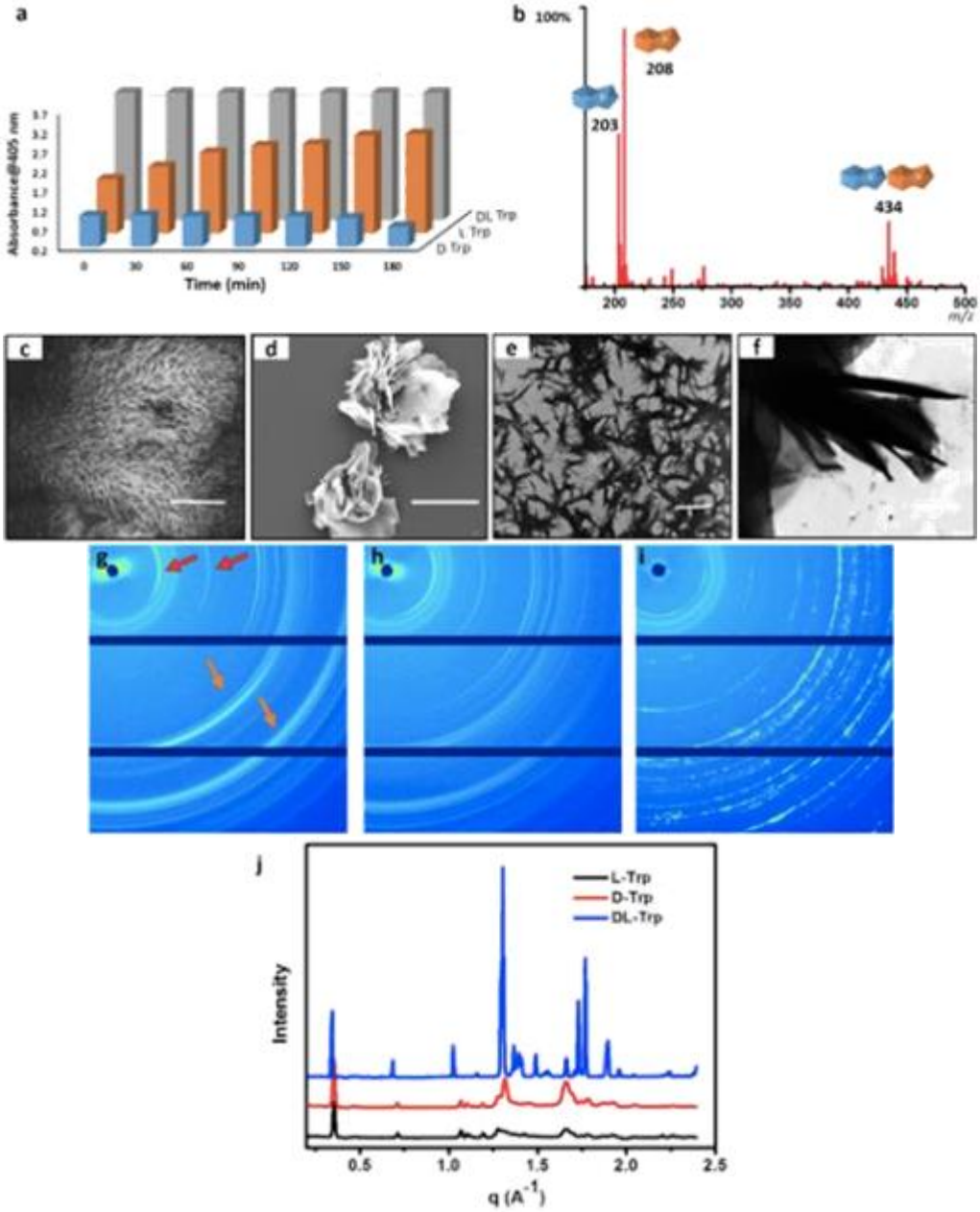






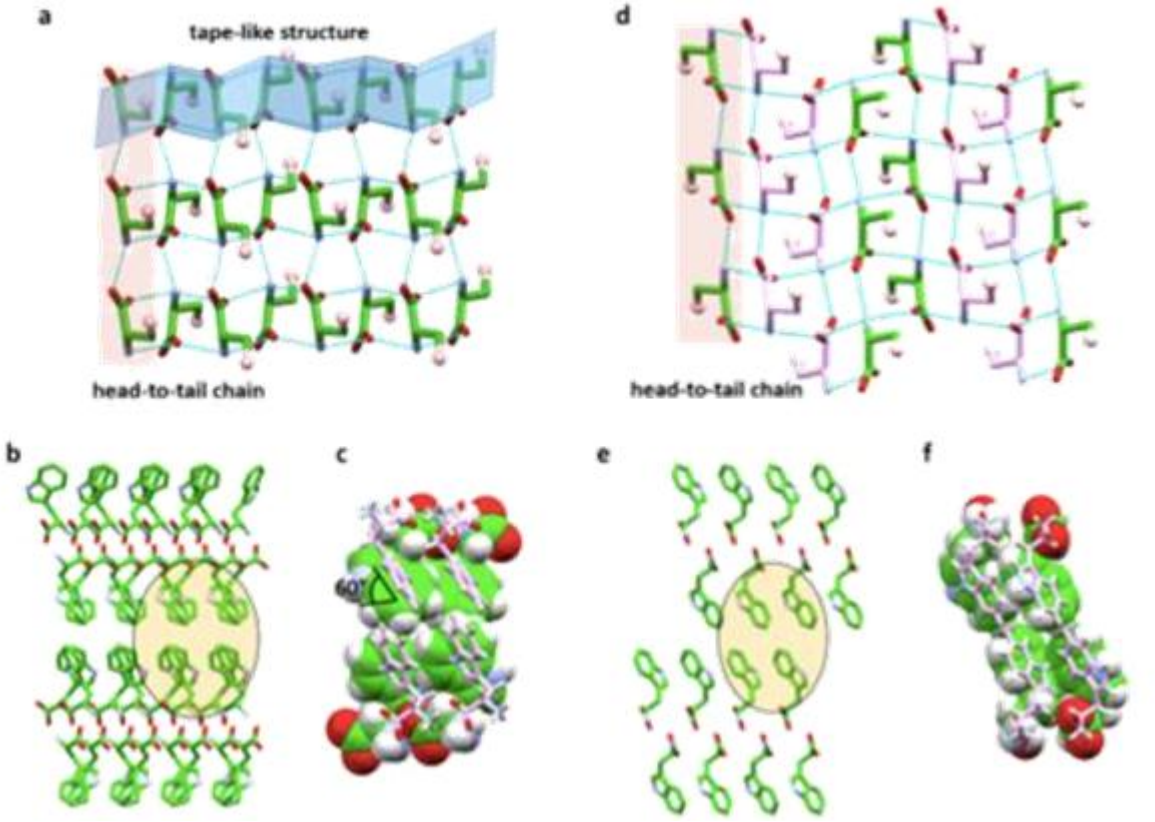
d) Effect of chirality on self-assembly

Phase behavior of pure D- Trp was different from L-Trp. It did not show a change in turbidity over time but formed structures at higher concentrations. ANS binding assay using D-Trp showed a similar change of environment from hydrophilic to hydrophobic, Concentration-dependent NMR study also demonstrated a shift toward lower ppm values due to the screening of the aromatic proton as a result of π−π stacking Turbidity of the DL-Trp solution increased very rapidly during cooling indicating a higher aggregation rate. An equimolar mixture of the L- and D-enantiomers was further confirmed by Trp mass spectrometry and CD Spectra. DL-conjugate fabricated nanostructures with a crystalline flake-like morphology. The details of the molecular arrangement was studied by WAXS



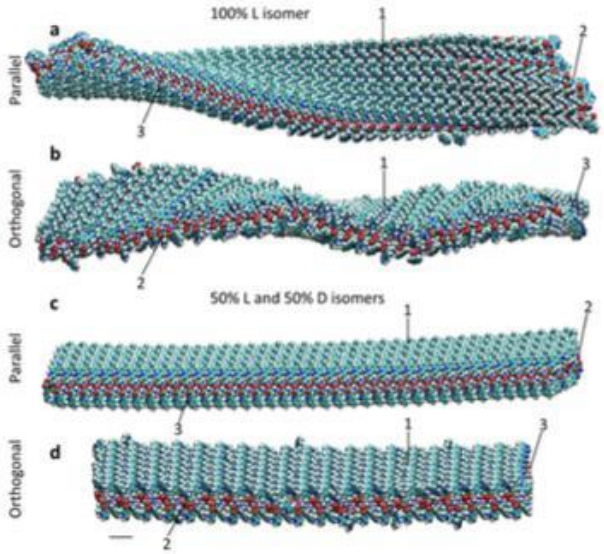
e) Packing structure ananlysis

Molecules were connected through head-to-tail H-bonding and fabricated into a single molecular chain. Along the a-axis, the molecules were connected in a sideways fashion through two H-bonds on each side and thus produced a tape-like arrangement. In the higher-order packing, L-Trp is arranged in a layer-by-layer structure resembling a supramolecular β-sheet structure. Structure stabilized by edge-to-face π–π stacking interactions. For DL-Trp, head-to-tail chain formation through H-bonding was observed, and centrosymmetric dimer formation of two adjacent chains. Perfect "knobs-in-holes" fitting of aromatic surfaces through face-to-face π–π interaction.



f) Modeling

L-Trp crystal has a tendency to fold in a direction parallel to the aromatic zipper and might grow along the aromatic zipper. DL-Trp crystals stay on average flat. In DL-Trp crystal, the facet orthogonal to the aromatic zipper is more stable, while the facet parallel to the zipper is less stable.



Discussion

The experimental evidence presented here suggests different self-assembly kinetics and mechanisms for DL-composites of aromatic amino acids, allowing the fabrication of interesting materials with exciting physical properties compared to the pure enantiomers.

The single-crystal structure analysis clearly demonstrated a favorable knob-to-hole packing of aromatic rings in the DL- mixture, which induced the easy growth of racemic crystals and the fabrication of self-assembled rigid materials.

References

[1] Aizen, R.; Tao, K.; Rencus-Lazar, S.; Gazit, E. Functional Metabolite Assemblies-A Review. J. Nanopart. Res. 2018, 20, 125, DOI: 10.1007/s11051-018-4217-3 [Crossref], [PubMed], Google Scholar

[2] Tadepalli, S.; Slocik, J.; Gupta, M.; Naik, R. R.; Singamaneni, S. Bio-Optics and Bio-Inspired Optical Materials. Chem. Rev. 2017, 117, 12705– 12763, DOI: 10.1021/acs.chemrev.7b00153 [ACS Full Text], [CAS], Google Scholar

[3] Knowles, T. P. J.; Buehler, M. J. Nanomechanics of Functional and Pathological Amyloid Materials. Nat. Nanotechnol. 2011, 6, 469– 479, DOI: 10.1038/nnano.2011.102 [Crossref], [PubMed], [CAS], Google Scholar

[4] Ménard-Moyon, C.; Venkatesh, V.; Krishna, K. V.; Bonachera, F.; Verma, S.; Bianco, A. Self-Assembly of Tyrosine into Controlled Supramolecular Nanostructures. Chem. - Eur. J. 2015, 21, 11681– 11686, DOI: 10.1002/chem.201502076 [Crossref], [PubMed], [CAS], Google Scholar

[5] Babar, D. G.; Sarkar, S. Self-Assembled Nanotubes from Single Fluorescent Amino Acid. Appl. Nanosci. 2017, 7, 101– 107, DOI: 10.1007/s13204-017-0551-5 [Crossref], [CAS], Google Scholar

[6] Singh, P.; Brar, S. K.; Bajaj, M.; Narang, N.; Mithu, V. S.; Katare, O. P.; Wangoo, N.; Sharma, R. K. Self-Assembly of Aromatic α-Amino Acids into Amyloid Inspired Nano/Micro Scaled Architects. Mater. Sci. Eng., C 2017, 72, 590– 600, DOI:

10.1016/j.msec.2016.11.117 [Crossref], [PubMed], [CAS], Google Scholar

[7] Guerin, S.; Stapleton, A.; Chovan, D.; Mouras, R.; Gleeson, M.; McKeown, C.; Noor, M. R.; Silien, C.; Rhen, F. M. F.; Kholkin, A. L.; Liu, N.; Soulimane, T.; Tofail, S. A. M.; Thompson, D. Control of Piezoelectricity in Amino Acids by Supramolecular Packing. Nat. Mater. 2018, 17, 180– 186, DOI: 10.1038/nmat5045 [Crossref], [PubMed], [CAS], Google Scholar

[8] Piperno, S.; Mirzadeh, E.; Mishuk, E.; Ehre, D.; Cohen, S.; Eisenstein, M.; Lahav, M.; Lubomirsky, I. Water-Induced Pyroelectricity from Nonpolar Crystals of Amino Acids. Angew. Chem., Int. Ed. 2013, 52, 6513– 6516, DOI: 10.1002/anie.201301836 [Crossref], [CAS], Google Scholar

[9] Meirzadeh, E.; Sapir, L.; Cohen, H.; Cohen, S. R.; Ehre, D.; Harries, D.; Lahav, M.; Lubomirsky, I. Nonclassical Crystal Growth as Explanation for the Riddle of Polarity in Centrosymmetric Glycine Crystals. J. Am. Chem. Soc. 2016, 138, 14756– 14763, DOI: 10.1021/jacs.6b09190 [ACS Full Text], [CAS], Google Scholar

[10] Shaham-Niv, S.; Arnon, Z. A.; Sade, D.; Lichtenstein, A.; Shirshin, E. A.; Kolusheva, S.; Gazit, E. Intrinsic Fluorescence of Metabolite Amyloids Allows Label-Free Monitoring of Their Formation and Dynamics in Live Cells. Angew. Chem., Int. Ed. 2018, 57, 12444– 12447, DOI: 10.1002/anie.201806565 [Crossref], [CAS], Google Scholar

[11] Jiang, S.; Chekini, M.; Qu, Z. B.; Wang, Y.; Yeltik, A.; Liu, Y.; Kotlyar, A.; Zhang, T.; Li, B.; Demir, H. V.; Kotov, N. A. Chiral Ceramic Nanoparticles and Peptide Catalysis. J. Am. Chem. Soc. 2017, 139, 13701– 13712, DOI: 10.1021/jacs.7b01445 [ACS Full Text], [CAS], Google Scholar

[12] Yeom, J.; Yeom, B.; Chan, H.; Smith, K. W.; Dominguez-Medina, S.; Bahng, J. H.; Zhao, G.; Chang, W. S.; Chang, S. J.; Chuvilin, A.; Melnikau, D.; Rogach, A. L.; Zhang, P.; Link, S.; Král, P.; Kotov, N. A. Chiral Templating of Self-Assembling Nanostructures by Circularly Polarized Light. Nat. Mater. 2015, 14, 66– 72, DOI: 10.1038/nmat4125 [Crossref], [PubMed], [CAS], Google Scholar

[13] Yang, G.; Zhang, S.; Hu, J.; Fujiki, M.; Zou, G. The Chirality Induction and Modulation of Polymers by Circularly Polarized Light. Symmetry 2019, 11, 474, DOI: 10.3390/sym11040474 [Crossref], [CAS], Google Scholar

[14] Mishuk, E.; Weissbuch, I.; Lahav, M.; Lubomirsky, I. Pyroelectricity in Nonpolar Directions in Crystals: Enantiomeric Disorder and Surface Wetting in Racemic α-Amino-

[15] Acids. Cryst. Growth Des. 2014, 14, 3839– 3848, DOI: 10.1021/cg5003644 [ACS Full Text], [CAS], Google Scholar

[16] Gomes, E. de M.; Viseu, T.; Belsley, M.; Almeida, B.; Costa, M. M. R.; Rodrigues, V. H.; Isakov, D. Piezoelectric and Pyroelectric Properties of DL-Alanine and L-Lysine Amino-Acid Polymer Nanofibers. Mater. Res. Express 2018, 5, 045049 DOI: 10.1088/2053-1591/aabd1c [Crossref], Google Scholar

[17] Meirzadeh, E.; Weissbuch, I.; Ehre, D.; Lahav, M.; Lubomirsky, I. Polar Imperfections in Amino Acid Crystals: Design, Structure, and Emerging Functionalities. Acc. Chem. Res. 2018, 51, 1238– 1248, DOI: 10.1021/acs.accounts.8b00054 [ACS Full Text], [CAS], Google Scholar

[18] Guerin, S.; O'Donnell, J.; Haq, E.; McKeown, C.; Silien, C.; Rhen, F. M. F.; Soulimane, T.; Tofail, S. A. M.; Thompson, D. Racemic Amino Acid Piezoelectric Transducer. Phys. Rev. Lett. 2019, 122, 047701 DOI: 10.1103/PhysRevLett.122.047701 [Crossref], [PubMed], [CAS], Google Scholar

[19] Smith, D. K. Lost in Translation? Chirality Effects in the Self-Assembly of Nanostructured Gel-Phase Materials. Chem. Soc. Rev. 2009, 38, 684– 694, DOI: 10.1039/b800409a [Crossref], [PubMed], [CAS], Google Scholar

[20] Moriuchi, T.; Hirao, T. Design of Ferrocene-Dipeptide Bioorganometallic Conjugates to Induce Chirality-Organized Structures. Acc. Chem. Res. 2010, 43, 1040– 1051, DOI: 10.1021/ar100022n [ACS Full Text], [CAS], Google Scholar

[21] Liu, M.; Zhang, L.; Wang, T. Supramolecular Chirality in Self-Assembled Systems. Chem. Rev. 2015, 115, 7304– 7397, DOI: 10.1021/cr500671p [ACS Full Text], [CAS], Google Scholar

[22] Sievers, S. A.; Karanicolas, J.; Chang, H. W.; Zhao, A.; Jiang, L.; Zirafi, O.; Stevens, J. T.; Münch, J.; Baker, D.; Eisenberg, D. Structure-Based Design of Non-Natural Amino-Acid Inhibitors of Amyloid Fibril Formation. Nature 2011, 475, 96– 100, DOI: 10.1038/nature10154 [Crossref], [PubMed], [CAS], Google Scholar

[23] Ridler, C. Alzheimer Disease: Misfolded Diabetes-Mellitus Peptide Seeds Amyloid-Beta Aggregation. Nat. Rev. Neurol. 2017, 13, 128, DOI: 10.1038/nrneurol.2017.5 [Crossref], [PubMed], [CAS], Google Scholar

[24] Flemming, H.-C.; Wingender, J.; Szewzyk, U.; Steinberg, P.; Rice, S. A.; Kjelleberg, S. Biofilms: An Emergent Form of Bacterial Life. Nat. Rev. Microbiol. 2016, 14, 563– 575, DOI: 10.1038/nrmicro.2016.94 [Crossref], [PubMed], [CAS], Google Scholar

[25] Garcia, A. M.; Iglesias, D.; Parisi, E.; Styan, K. E.; Waddington, L. J.; Deganutti, C.; Zorzi, R. D.; Grassi, M.; Melchionna, M.; Vargiu, A. V.; Marchesan, S. Chirality Effects on Peptide Self-Assembly Unraveled from Molecules to Materials. Chem. 2018, 4, 1– 15, DOI: 10.1016/j.chempr.2018.05.016

[26] Wang, M.; Zhou, P.; Wang, J.; Zhao, Y.; Ma, H.; Lu, J. R.; Xu, H. Left or Right: How Does Amino Acid Chirality Affect the Handedness of Nanostructures Self-Assembled from Short Amphiphilic Peptides?. J. Am. Chem. Soc. 2017, 139, 4185– 4194, DOI: 10.1021/jacs.7b00847

[27] Hu, K.; Jiang, Y.; Xiong, W.; Li, H.; Zhang, P. Y.; Yin, F.; Zhang, Q.; Geng, H.; Jiang, F.; Li, Z.; Wang, X.; Li, Z. Tuning Peptide Self-Assembly by an In-

Tether Chiral Center. Sci. Adv. 2018, 11, eaar5907 DOI: 10.1126/sciadv.aar5907

[28] Luo, Z.; Wang, S.; Zhang, S. Fabrication of Self-Assembling D-Form Peptide Nanofiber Scaffold D-EAK16 for Rapid Hemostasis. Biomaterials 2011, 32, 2013– 2020, DOI: 10.1016/j.biomaterials.2010.11.049

[29] Marchesan, S.; Easton, C. D.; Styan, K.; Waddington, L.; Kushkaki, K.; Goodall, L.; Mclean, K.; Forsythe, J. S.; Hartley, P. G. Chirality Effects at each Amino Acid Position on Tripeptide Self-Assembly into Hydrogel Biomaterials. Nanoscale 2014, 6, 5172– 5180, DOI: 10.1039/C3NR06752A

[30] Janek, K.; Rothemund, S.; Gast, K.; Beyermann, M.; Zipper, J.; Fabian, H.; Bienert, M.; Krause, E. Study of the Conformational Transition of Aβ(1–42) Using D-Amino Acid Replacement Analogues. Biochemistry 2001, 40, 5457– 5463, DOI: 10.1021/bi002005e [ACS Full Text], [CAS], Google Scholar

[31] Shi, J.; Du, X.; Yuan, D.; Zhou, J.; Zhou, N.; Huang, Y.; Xu, B. D-Amino Acid Modulate the Cellular Response of Enzymatic-Instructed Supramolecular Nanofibers of Small Peptides. Biomacromolecules 2014, 15, 3559– 3568, DOI: 10.1021/bm5010355 [ACS Full Text], [CAS], Google Scholar

[32] Zhao, Y.; Leman, L. J.; Search, D. J.; Garcia, R. A.; Gordon, D. A.; Maryanoff, B. E.; Ghadiri, M. R. Self-Assembling Cyclic D,L-α-Peptides as Modulators of Plasma HDL Function. A Supramolecular Approach toward Antiatherosclerotic Agents. ACS Cent. Sci. 2017, 3, 639– 646, DOI: 10.1021/acscentsci.7b00154 [ACS Full Text], [CAS], Google Scholar

[33] Ye, Z.; Zhu, X.; Acosta, S.; Kumar, D.; Sang, T.; Aparicio, C. Self-Assembly Dynamics and Antimicrobial Activity of All L- and D-Amino Acid Enantiomers of a Designer Peptide. Nanoscale 2019, 11, 266– 275, DOI: 10.1039/C8NR07334A [Crossref], [CAS], Google Scholar

[34] Fu, Y.; Li, B.; Huang, Z.; Li, Y.; Yang, Y. Terminal is Important for the Helicity of the Self-Assemblies of Dipeptides Derived from Alanine. Langmuir 2013, 29, 6013– 6017, DOI: 10.1021/la400910g [ACS Full Text], [CAS], Google Scholar

[35] Marchesan, S.; Styan, K. E.; Easton, C. D.; Waddington, L.; Vargiu, A. V. Higher and Lower Supramolecular Orders for the Design of Self-Assembled Heterochiral Tripeptide Hydrogel Biomaterials. J. Mater. Chem. B 2015, 3, 8123– 8132, DOI: 10.1039/C5TB00858A [Crossref], [PubMed], [CAS], Google Scholar

[36] Marchesan, S.; Easton, C. D.; Kushkaki, F.; Waddington, L.; Hartley, P. G. Tripeptide Self-Assembled Hydrogels: Unexpected Twists of Chirality. Chem. Commun. 2012, 48, 2195– 2197, DOI: 10.1039/C2CC16609G [Crossref], [PubMed], [CAS], Google Scholar

[37] Pappas, C. G.; Frederix, P. W. J. M.; Mutasa, T.; Fleming, S.; Abul-Haija, Y. M.; Kelly, S. M.; Gachagan, A.; Kalafatovic, D.; Trevino, J.; Ulijn, R. V. Alignment of Nanostructured Tripeptide Gels by Directional Ultrasonication. Chem. Commun. 2015, 51, 8465– 8468, DOI: 10.1039/C5CC02049B [Crossref], [PubMed], [CAS], Google Scholar

[38] Basak, S.; Singh, I.; Ferranco, A.; Syed, J.; Kraatz, H.-B. On the Role of Chirality in Guiding the Self-Assembly of Peptides. Angew. Chem., Int. Ed. 2017, 56, 13288– 13292, DOI: 10.1002/anie.201706162 [Crossref], [CAS], Google Scholar

[39] McAulay, K.; Dietrich, B.; Su, H.; Scott, M. T.; Rogers, S.; Al-Hilaly, Y. K.; Cui, de H.; Serpell, L. C.; Seddon, A. M.; Draper, E. R.; Adams, D. J. Using Chirality to Influence Supramolecular Gelation. Chem. Sci. 2019, 10, 7801– 7806, DOI: 10.1039/C9SC02239B [Crossref], [PubMed], [CAS], Google Scholar

[40] Tómasson, D. A.; Ghosh, D.; Kržišnik, Z.; Fasolin, L. H.; Vicente, A. A.; Martin, A. D.; Thordarson, P.; Damodaran, K. K. Enhanced Mechanical and Thermal Strength in Mixed-Enantiomers-Based Supramolecular Gel. Langmuir 2018, 34, 12957– 12967, DOI: 10.1021/acs.langmuir.8b02729 [ACS Full Text], [CAS], Google Scholar

[41] Nagy, K. J.; Giano, M. C.; Jin, A.; Pochan, D. J.; Schneider, J. P. Enhanced Mechanical Rigidity of Hydrogels Formed from Enantiomeric Peptide Assemblies. J. Am. Chem. Soc. 2011, 133, 14975– 14977, DOI: 10.1021/ja206742m [ACS Full Text], [CAS], Google Scholar

[42] Yeates, T. O.; Kent, S. B. H. Racemic Protein Crystallography. Annu. Rev. Biophys. 2012, 41, 41– 61, DOI: 10.1146/annurev-biophys-050511-102333 [Crossref], [PubMed], [CAS], Google Scholar

[43] Mandal, K.; Uppalapati, M.; Ault-Riché, D.; Kenney, J.; Lowitz, J.; Sidhu, S. S.; Kent, S. B. H. Chemical Synthesis and X-Ray Structure of a Heterochiral {D-Protein Antagonist Plus Vascular Endothelial Growth Factor} Protein Complex by Racemic Crystallography. Proc. Natl. Acad. Sci. U. S. A. 2012, 109, 14779– 14784, DOI: 10.1073/pnas.1210483109 [Crossref], [PubMed], [CAS], Google Scholar

[44] Singh, V.; Rai, R. K.; Arora, A.; Sinha, N.; Thakur, A. K. Therapeutic Implication of L-Phenylalanine Aggregation Mechanism and Its Modulation by D-Phenylalanine in Phenylketonuria. Sci. Rep. 2015, 4, 3875, DOI: 10.1038/srep03875 [Crossref], Google Scholar

[45] Reches, M.; Gazit, E. Casting Metal Nanowires within Discrete Self-Assembled Peptide Nanotubes. Science 2003, 300, 625– 627, DOI: 10.1126/science.1082387 [Crossref], [PubMed], [CAS], Google Scholar

[46] Pawar, A. P.; DuBay, K. F.; Zurdo, J.; Chiti, F.; Vendruscolo, M.; Dobson, C. M. Prediction of “Aggregation-Prone” and “Aggregation-Susceptible” Regions in Proteins Associated with Neurodegenerative Diseases. J. Mol. Biol. 2005, 350, 379– 392, DOI: 10.1016/j.jmb.2005.04.016 [Crossref], [PubMed], [CAS], Google Scholar

[47] Frederix, P. W. J. M.; Scott, G. G.; Abul-Haija, Y. M.; Kalafatovic, D.; Pappas, C. G.; Javid, N.; Hunt, N. T.; Ulijn, R. V.; Tuttle, T. Exploring the Sequence Space for (Tri-)Peptide Self-Assembly to Design and Discover New Hydrogels. Nat. Chem. 2015, 7, 30–

37, DOI: 10.1038/nchem.2122 [Crossref], [PubMed], [CAS], Google Scholar

[48] Shaham-Niv, S.; Adler-Abramovich, L.; Schnaider, L.; Gazit, E. Extension of the Generic Amyloid Hypothesis to Nonproteinaceous Metabolite Assemblies. Sci. Adv. 2015, 1, e1500137 DOI: 10.1126/sciadv.1500137 [Crossref], [PubMed], [CAS], Google Scholar

[49] Shaham-Niv, S.; Rehak, P.; Vuković, L.; Adler-Abramovich, L.; Král, P.; Gazit, E. Formation of Apoptosis-Inducing Amyloid Fibrils by Tryptophan. Isr. J. Chem. 2017, 57, 729– 737, DOI: 10.1002/ijch.201600076 [Crossref], [CAS], Google Scholar

[50] Banik, D.; Kundu, S.; Banerjee, P.; Dutta, R.; Sarkar, N. Investigation of Fibril Forming Mechanisms of L-Phenylalanine and L-Tyrosine: Microscopic Insight toward

[51] Phenylketonuria and Tyrosinemia Type II. J. Phys. Chem. B 2017, 121, 1533– 1543, DOI: 10.1021/acs.jpcb.6b12220 [ACS Full Text], [CAS], Google Scholar

[52] Do, T. D.; Kincannon, W. M.; Bowers, M. T. Phenylalanine Oligomers and Fibrils: The Mechanism of Assembly and the Importance of Tetramers and Counterions. J. Am. Chem. Soc. 2015, 137, 10080– 10083, DOI: 10.1021/jacs.5b05482 [ACS Full Text], [CAS], Google Scholar

[53] Perween, S.; Chandanshive, B.; Kotamarthi, H. C.; Khushalani, D. Single Amino Acid Based Self-Assembled Structure. Soft Matter 2013, 9, 10141– 10145, DOI: 10.1039/c3sm51054a [Crossref], [CAS], Google Scholar

[54] Bera, S.; Mondal, S.; Rencus-Lazar, S.; Gazit, E. Organization of Amino Acids into Layered Supramolecular Secondary Structures. Acc. Chem. Res. 2018, 51, 2187– 2197, DOI: 10.1021/acs.accounts.8b00131 [ACS Full Text], [CAS], Google Scholar

[55] Bera, S.; Mondal, S.; Tang, Y.; Jacoby, G.; Arad, E.; Guterman, T.; Jelinek, R.; Beck, R.; Wei, G.; Gazit, E. Deciphering the Rules for Amino Acid Co-Assembly Based on Interlayer Distances. ACS Nano 2019, 132, 1703– 1712, DOI: 10.1021/acsnano.8b07775 [ACS Full Text], Google Scholar

[56] De Luigi, A.; Mariani, A.; De Paola, M.; Re Depaolini, A.; Colombo, L.; Russo, L.; Rondelli, V.; Brocca, P.; Adler-Abramovich, L.; Gazit, E.; Del Favero, E.; Cantù, L.; Salmona, M. Doxycycline Hinders Phenylalanine Fibril Assemblies Revealing Apotential Novel Therapeutic Approach in Phenylketonuria. Sci. Rep. 2015, 5, 15902, DOI: 10.1038/srep15902 [Crossref], [PubMed], [CAS], Google Scholar

[57] Ihlefeldt, F. S.; Pettersen, F. B.; von Bonin, A.; Zawadzka, M.; Görbitz, C. H. The Polymorphs of L-Phenylalanine. Angew. Chem., Int. Ed. 2014, 53, 13600– 13604, DOI: 10.1002/anie.201406886 [Crossref], [CAS], Google Scholar

[58] Knowles, T. P. J.; Mezzenga, R. Amyloid Fibrils as Building Blocks for Natural and Artificial Functional Materials. Adv. Mater. 2016, 28, 6546– 6561, DOI: 10.1002/adma.201505961 [Crossref], [PubMed], [CAS], Google Scholar

[59] Do, T. D.; De Almeida, N. E. C.; LaPointe, N. E.; Chamas, A.; Feinstein, S. C.; Bowers, M. T. Amino Acid Metaclusters: Implications of Growth Trends on Peptide Self-

[60] Assembly and Structure. Anal. Chem. 2016, 88, 868– 876, DOI: 10.1021/acs.analchem.5b03454 [ACS Full Text], [CAS], Google Scholar

[61] Amdursky, N.; Stevens, M. M. Circular Dichroism of Amino Acids: Following the Structural Formation of Phenylalanine. ChemPhysChem 2015, 16, 2768– 2774, DOI: 10.1002/cphc.201500260 [Crossref], [PubMed], [CAS], Google Scholar

[62] Lee, C.; Wei, X. D.; Kysar, J. W.; Hone, J. Measurement of the Elastic Properties and Intrinsic Strength of Monolayer Graphene. Science 2008, 321, 385– 388, DOI: 10.1126/science.1157996 [Crossref], [PubMed], [CAS], Google Scholar

[63] Bera, S.; Mondal, S.; Xue, B.; Shimon, L. J. W.; Cao, Y.; Gazit, E. Rigid Helical-Like Assemblies from a Self-Aggregating Tripeptide. Nat. Mater. 2019, 18, 503– 509, DOI: 10.1038/s41563-019-0343-2 [Crossref], [PubMed], [CAS], Google Scholar

[64] Kol, N.; Adler-Abramovich, L.; Barlam, D.; Shneck, R. Z.; Gazit, E.; Rousso, I. Self-Assembled Peptide Nanotubes are Uniquely Rigid Bioinspired Supramolecular Structures. Nano Lett. 2005, 5, 1343– 1346, DOI: 10.1021/nl0505896 [ACS Full Text], [CAS], Google Scholar

[65] Smith, S. B.; Cui, Y.; Bustamante, C. Overstretching B-DNA: The Elastic Response of Individual Double-Stranded and Single-Stranded DNA Molecules. Science 1996, 271, 795– 799, DOI: 10.1126/science.271.5250.795 [Crossref], [PubMed], [CAS], Google Scholar

[66] Singh, R. P.; Blossey, R.; Cleri, F. Structure and Mechanical Characterization of DNA I-Motif Nanowires by Molecular Dynamics Simulation. Biophys. J. 2013, 105, 2820– 2831, DOI: 10.1016/j.bpj.2013.10.021 [Crossref], [PubMed], [CAS], Google Scholar

[67] Smith, J. F.; Knowles, T. P. J.; Dobson, C. M.; MacPhee, C. E.; Welland, M. E. Characterization of the Nanoscale Properties of Individual Amyloid Fibrils. Proc. Natl. Acad. Sci. U. S. A. 2006, 103, 15806– 15811, DOI: 10.1073/pnas.0604035103 [Crossref], [PubMed], [CAS], Google Scholar

[68] Uyaver, S.; Hernandez, H. W.; Habiboglu, M. G. Self-Assembly of Aromatic Amino Acids: A Molecular Dynamics Study. Phys. Chem. Chem. Phys. 2018, 20, 30525– 30536, DOI: 10.1039/C8CP06239K [Crossref], [PubMed], [CAS], Google Scholar

[69] Gasymov, O. K.; Glasgow, B. J. ANS Fluorescence: Potential to Augment the Identification of the External Binding Sites of Proteins. Biochim. Biophys. Acta, Proteins

[70] Proteomics 2007, 1774, 403– 411, DOI: 10.1016/j.bbapap.2007.01.002 [Crossref], [PubMed], [CAS], Google Scholar

[71] Sirangelo, I.; Malmo, C.; Casillo, M.; Irace, G. Resolution of Tryptophan–ANS Fluorescence Energy

Transfer in Apomyoglobin by Site-Directed Mutagenesis. Photochem. Photobiol. 2002, 76, 381–384, DOI: 10.1562/0031-8655(2002)076<0381:ROTAFE>2.0.CO;2 [Crossref], [PubMed], [CAS], Google Scholar

[72] Chaudhuri, A.; Haldar, S.; Sun, H.; Koeppe, R. E.; Chattopadhyay, A. Importance of Indole N-H Hydrogen Bonding in the Organization and Dynamics of Gramicidin Channels. Biochim. Biophys. Acta, Biomembr. 2014, 1838, 419– 428, DOI: 10.1016/j.bbamem.2013.10.011 [Crossref], [PubMed], [CAS], Google Scholar

[73] Fenwick, R. B.; Oyen, D.; Dyson, H. J.; Wright, P. E. Slow Dynamics of Tryptophan–Water Networks in Proteins. J. Am. Chem. Soc. 2018, 140, 675– 682, DOI: 10.1021/jacs.7b09974 [ACS Full Text], [CAS], Google Scholar

[74] Görbitz, C. H.; Törnroos, K. W.; Day, G. M. Single-Crystal Investigation of L-Tryptophan with Ź=16. Acta Crystallogr., Sect. B: Struct. Sci. 2012, B68, 549– 557, DOI: 10.1107/S0108768112033484 [Crossref], Google Scholar

[75] Li, Y.; Beck, R.; Huang, T.; Choi, M. C.; Divinagracia, M. Scatterless Hybrid Metal–Single-Crystal Slit for Small Angle X-Ray Scattering and High-Resolution X-Ray Diffraction. J. Appl. Crystallogr. 2008, 41, 1134– 1139, DOI: 10.1107/S0021889808031129 [Crossref], [CAS], Google Scholar

[76] Sheldrick, G. M. SHELXL-2013, Program for Crystal Structure Refinement; University of Göttingen: Göttingen, Germany, 2013. Google Scholar

[77] Phillips, J. C.; Braun, R.; Wang, W.; Tajkhorshid, E.; Villa, E.; Chipot, C.; Skeel, R. D.; Kale, L.; Schulten, K. Scalable Molecular Dynamics with NAMD. J. Comput. Chem. 2005, 26, 1781– 1802, DOI: 10.1002/jcc.20289 [Crossref], [PubMed], [CAS], Google Scholar

[78] MacKerell, A. D.; Bashford, D. All-Atom Empirical Potential for Molecular Modeling and Dynamics Studies of Proteins. J. Phys. Chem. B 1998, 102, 3586– 3616, DOI: 10.1021/jp973084f [ACS Full Text], [CAS], Google Scholar

[79] Darden, T.; York, D.; Pedersen, L. Particle Mes Ewald: An N log(N) Method for Ewald Sums in Large Systems. J. Chem. Phys. 1993, 98, 10089– 10092, DOI: 10.1063/1.464397 [Crossref], [CAS], Google Scholar

[80] Humphry, W.; Dalke, A.; Schulten, K. VMD – Visual Molecular Dynamics. J. Mol. Graphics 1996, 14 (1), 33– 38, DOI: 10.1016/0263-7855(96)00018-5 [Crossref], [PubMed], Google Scholar