CCS Chemistry

MINI REVIEW

Received: July 18, 2023 | Accepted: Sept. 19, 2023 | Published: Oct. 27, 2023

Oligopeptide Self-Assembly: Mechanisms, Stimuli-Responsiveness, and Biomedical Applications

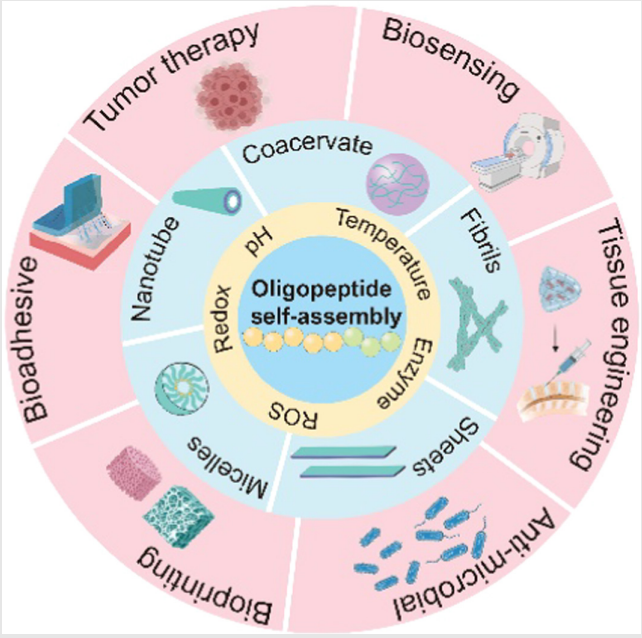
Lei Ge[1,2†], Hanyan Xu[1†], Xin Jiang[1] & Jing Yu[1,2*]

[1]*School of Materials Science and Engineering, Nanyang Technological University, Singapore 637553,* [2]*Institute for Digital Molecular Analytics and Science, Nanyang Technological University, Singapore 637553*

**Corresponding author: yujing@ntu.edu.sg;* [†]L. Ge and H. Xu contributed equally to this work.

Cite this: *CCS Chem*. **2024**, 6, 69–90
DOI: 10.31635/ccschem.023.202303209

Oligopeptide self-assembly materials have emerged as a promising class of biomaterials with diverse applications in biomedicine. This review highlights the recent progress in comprehending the self-assembly mechanisms intrinsic to oligopeptides and their behavior in response to specific stimuli. By methodically structuring the amino acid sequence and managing external stimuli such as pH levels, redox conditions, or enzymatic activity, we can exercise unprecedented control over the self-assembly process. By controlling the self-assembly process of oligopeptides, various structures with extraordinary versatility can be obtained, including micelles, nanofibers, and coacervate droplets, each possessing modifiable mechanical and chemical properties. Furthermore, these self-assembled constructs demonstrate immense potential within varied biomedical applications. The stimuli-sensitive nature of oligopeptide assembly materials facilitates timely encapsulation and release of therapeutic cargos, consequently eliciting desired cellular responses. This approach paves the way for more precise tumor targeting, personalized medicinal treatments, and well-regulated drug dispensation. Their innate biocompatibility and proficiency in replicating the extracellular matrix (ECM) render them ideally suited for applications such as tissue engineering, wound remediation, and regenerative medicine. In summary, oligopeptide self-assembling materials show tremendous potential as adaptable platforms for cutting-edge biomedical applications, thereby bridging the divide between fundamental research and practical clinical application.



Keywords: oligopeptides, self-assembly, nonequilibrium, stimuli-responsive behavior, biomedical applications

DOI: 10.31635/ccschem.023.202303209
Citation: *CCS Chem*. **2024**, 6, 69–90
Link to VoR: https://doi.org/10.31635/ccschem.023.202303209

Introduction

Oligopeptides, typically ranging from 2 to 20 amino acid residues in length, are integral components engaged in an array of biological processes, encompassing vital functions such as cellular signaling, enzymatic activities, and protein structure.[1] Their compact sizes allow for easier synthesis and modification, positioning them as optimal building blocks for the creation and synthesis of functional materials. Interestingly, oligopeptides can spontaneously arrange themselves into highly-defined nano or microstructures. These possess controlled sizes, shapes, and functionalities, thereby forming what we term oligopeptide self-assembly materials.[2–4]

The self-assembly process of oligopeptide self-assembly materials is predominantly directed by their primary and secondary structures.[5–7] The primary structure, defined by the distinct sequence of amino acids (AAs), lays the foundation of their self-assembly. Secondary structures, such as α-helices and β-sheets, emerge from the folding and interplay of the amino acid residues. Manipulating these primary and secondary structures allows for control over the self-assembly behaviors of oligopeptides, enabling the creation of functional materials suitable for an array of biomedical applications.[8,9] The permutations and combinations of diverse modular primary and secondary structures yield a broad spectrum of morphologies in oligopeptide self-assembly, including micelles,[10] wormlike micelles,[11] nanofibers,[12,13] nanotubes,[14] liquid droplets,[15] and more. Understanding the relationship between oligopeptide structure and morphology provides valuable insights for the development of novel biomaterials with enhanced functionality and performance.

The self-assembly of oligopeptides exhibits remarkable stimulus-responsive behavior, enhancing their versatility in biomedical applications. These materials are capable of structural alterations in response to a variety of external stimuli[16,17] such as pH, temperature, redox conditions, and enzymes. The inherent design and composition of oligopeptides allow for precise control over their responsiveness to specific triggers. By integrating specific functional groups or sequences into the oligopeptide structure, the self-assembling materials can exhibit reversible or irreversible transformations. These transformations result in changes in physical properties such as morphology, stability, and drug release kinetics.[18,19] Furthermore, the responsiveness of oligopeptide self-assembly materials can be engineered to echo physiological cues, enabling interactions with biological systems, including cell adhesion, migration, and tissue regeneration. The ability of these materials to adapt and respond to their environment makes them promising candidates for the development of advanced drug delivery systems, biosensors, and tissue engineering scaffolds with enhanced functionality and therapeutic efficacy.[20–22]

Compared to artificial materials governed by thermodynamic parameters, kinetic controls take precedence in oligopeptide self-assembly, providing the potential to create life-like, programmable, and "intelligent" materials.[23] These materials implement nonequilibrium concepts typically found in biological systems. Living systems exist out of equilibrium, experiencing formation and degradation processes driven by constant energy input. The input energy supports integrated reaction networks that, in dynamic behavior, uphold concentration gradients and facilitate an active transport of functional molecules over time and space.[24]

The oligopeptide self-assembling process demonstrates both thermodynamic and kinetic behaviors.[1,25] The energy and entropy contributions of the assembly determine the equilibrium status, while the kinetic aspects can complicate the assembly process, resulting in kinetic traps. These kinetically trapped states might relax into the global free-energy minimum within accessible timescales, although some might fail to do so throughout experimental observation time. Changes in solvent properties such as pH[26] and salt concentration[27] can permit kinetic controls as associated interactions and energy barriers are modified. Furthermore, differences in the preparative pathway can yield several distinct assembled architectures as kinetically trapped products.[28] Unlike isolated systems, the self-assembly kinetics of oligopeptides can be regulated by energy input and the rates of related dynamic reaction cycles.[29] Such kinetic control can orchestrate spatiotemporal organization over structures and properties through the tunable lifetime of intermediates in reaction cycles. Liquid–liquid phase separation (LLPS) mediated self-assembly represents a nonclassical nucleation and growth process. Its structural evolution is highly dynamic, and the associated energy barrier depends on the diffusion and relaxation of intermediates.[30] Understanding and reproducing the growth of fibrillar structures inside liquid condensate will aid in deciphering the pathological causes underlying neurodegenerative diseases.

Oligopeptide materials have been applied broadly in various areas of biomedicine, from drug delivery systems to biosensors and tissue engineering.[31,32] They have been utilized as drug delivery systems, where the self-assembled nanostructures encapsulate therapeutic agents and release them at specific sites or in response to specific stimuli. The structural changes associated with self-assembly can induce alterations in optical, electrical, or mechanical properties, providing a signal readout for analyte detection. The adaptability of these materials allows them to mimic the natural extracellular matrix (ECM), making them ideal for creating biomimetic environments for cell culture and tissue regeneration.

This review explores the multifaceted world of oligopeptide self-assembly materials, illuminating both the potential and challenges that lie ahead. We discuss their

DOI: 10.31635/ccschem.023.202303209
Citation: *CCS Chem.* **2024**, 6, 69–90
Link to VoR: https://doi.org/10.31635/ccschem.023.202303209

customized primary and secondary structures across various length scales, their stimuli-responsive behaviors, and dynamic transformations. Despite the vast applications and potential of these materials, we still lack a comprehensive understanding of the nonequilibrium self-assembly processes and structure-function relationships of oligopeptides. The introduction of machine learning offers promising opportunities for advancements in this field.

The self-assembly of oligopeptides with designed primary and secondary structures across various length scales

The self-assembly of oligopeptides can generate a multitude of structures across varying length scales, endowed with desired functionalities, utilizing a modest palette of just 20 AAs and simple motifs of secondary structures.[2] AAs, with their diverse physicochemical characteristics—polar or nonpolar, charged or noncharged—serve as minimal building blocks, enabling the design of peptide sequences in a programmable manner with predictable properties. Secondary structures in peptides such as β-sheet and α-helix can influence the assembly behavior kinetically and thermodynamically, further facilitating the formation of complex frameworks (Figure 1).

The delicate balance of noncovalent intermolecular interactions, including hydrogen bonds, hydrophobic interactions, π–π and cation-π interactions, and electrostatic interactions, plays a crucial role in guiding the collective self-organization in peptide assembly.[1,2] This interplay produces variations in the morphologies and dimensions of assembled structures (Table 1).

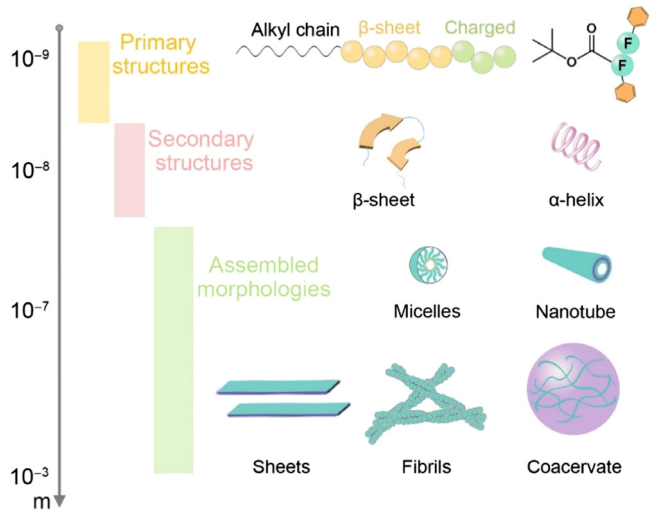


Figure 1 | *Length scales of the hierarchical complex structures involved in oligopeptide self-assembly.*

Among these interactions, hydrogen bonding, which originates from backbones and/or side chains, is the most pervasive.[33] The formation of secondary structures or disordered conformations arises from intramolecular hydrogen bonding between proximal AAs.[34] Hydrogen bonding is oriented and, with a strength of 10–40 kJ mol^{-1}, it is stronger than van der Waals forces.[21] Fibrillar assemblies created by peptide amphiphiles or amyloid-like peptides are associated with ordered β-sheet interactions.[35,36] The formation of LLPS can be influenced by extensive H-bonding.[15,37]

Hydrophobic interaction is another primary driving force, which encourages peptides to aggregate together and expel water molecules away. This interaction is typically long-range attractive, entropy-dominated, and generally less potent than hydrogen bonding. A specific form of hydrophobic interaction is π–π stacking, which is often involved with AAs with aromatic residues, particularly phenylalanine (F). Contrasting with typical hydrophobic interactions that are nonoriented, π–π stacking exhibits a high degree of organization and directionality in peptide assembly.[38] This characteristic contributes to the prevalence of fibril structures in certain short amyloid peptides and their analogues. Electrostatic interaction, specific to AAs with charged residues and highly dependent on solution environments like ionic strength and pH, is another essential long-range force that is easily regulated.[75]

In addition to primary forces, other interactions like cation-π interactions play a significant role. For instance, they are evident in the lysine (K)-3,4-dihydroxy-L-phenylalanine [(K)-DOPA] sequence found in mussel-inspired peptides.[37] Another example is the arginine (R)-tyrosine (Y) interactions in DNA/RNA-binding protein FUS,[76] which facilitate LLPS. Also, metal-coordination interactions are often present and applied in the assembly of higher-order structural frameworks through the spontaneous metal-coordinated crossings and folding of peptidyl linkers.[77,78]

The self-assembly of oligopeptides can generate a wide array of morphologies, including spherical micelles, worm-like micelles, fibrils, and nanosheets.[2,35,38] The scale of these assemblies can range from nanometers to submillimeters.[36,79] The principle of assembly shares similarities with that of conventional block copolymers, which can be determined using the packing parameter, $p = \frac{v}{l_c a_0}$, where v is the volume of the hydrophobic domain, O_a is the effective area of the hydrophilic head domain, c_l is the length of the hydrophilic segment.[75] When $p < 1/3$, spherical micelles are favored, whereas cylindrical micelles are preferable when $1/3 < p < 1/2$. When $1/2 < p < 1$, a transition to bilayer lamellae or vesicles occurs, and planar lamella is achieved at $p = 1$. For $p > 1$, inverted structures may occur.

However, the secondary structures folded during assembly can alter the expected assembly results. Peptide amphiphiles such as $C_{16}V_3A_3K_3$, due to the β-sheet

DOI: 10.31635/ccschem.023.202303209
Citation: *CCS Chem.* **2024**, 6, 69–90
Link to VoR: https://doi.org/10.31635/ccschem.023.202303209

DOI: 10.31635/ccschem.023.202303209
Citation: CCS Chem. 2024, 6, 69-90
Link to VoR: https://doi.org/10.31635/ccschem.023.202303209

Table 1 | *The Summary of Typical Sequence Designs Assembled Morphologies, Dominant Driving Forces, as well as Influenced Factors*

Morphologies	Sequences	Interactions	Factors	Ref.
Micelles	oSt(His)$_6$	Hydrophobic collapse, metal-ion coordination	Metal-ions, pH	39,40
	(Boc/Z-) FF	π–π stacking, β-sheet	Cosolvency, time	41,42
Fibrils	$C_{16}V_3A_3K_3$	β-sheet, electrostatic interactions, hydrophobic collapse	pH, salt, temperature, concentration	35,43–47
	Ac-KLVFFAL-NH_2	β-sheet	pH, enzyme	48–51
	(Fmoc-)FF	β-sheet, π–π stacking	pH, chirality, metal-ions, cosolvency, enzyme	52–57
	(NBD-)LLLL$_p$Y	α-helix, hydrophobic collapse	Enzyme	58,59
	EAK_{16}	β-sheet, electrostatic interactions	pH, salt, temperature, concentration	60
Nanotubes	C_{10}-FFVK	β-sheet, hydrophobic collapse	Time	61,62
	(Im/Ac-) KLVFFAL-NH_2	β-sheet		63–65
	Ac-FKFEFKFE-NH_2	β-sheet, electrostatic interactions	pH, temperature, time, chirality	66,67
	R_3L_{12}	α-helix, hydrophobic collapse	pH, chirality	59,68
Sheets	$C_{16}K_2$	Electrostatic interactions, hydrophobic collapse	pH, salt	69,70
	$E_2F_6C_{11}E$	π–π stacking, β-sheet, hydrophobic collapse	Cosolvency	71
Coacervates	FFssFF	π–π stacking (balance)	pH, salt, concentration	72,73
	GK-16*: GY*KGKY*Y*GKGKKY*Y*Y*K	H-bonding, electrostatic interactions	pH, salt, concentration	37,74

forming Val-Ala repeated domain show a strong propensity to form one-dimensional (1D) assemblies like those of fibers and nanoribbons.[35,43,80] Amyloid-forming oligopeptides such as Lys-Leu-Val-Phe-Phe-Ala-Leu (KLVFFAL), and the dipeptide FF also tend to form fibers and nanotubes due to the directional β-sheet patterns.[36,79,81] In contrast, surfactant-like peptide sequences, like $C_{16}K_n$ and $oStH_6$, can form a range of structures, including micelles and membranes (vesicles). These structures are achieved by simply adjusting the size of the head group due to the absence of folded conformation.[39,69]

While the self-assembly of β-sheet structures is well-established in the construction of various assembled structures, that of helical peptides has been far less explored, even though many native protein filaments comprise protomers that adopt an α-helical conformation.[3,82] This lack of study is due to several challenges: (1) the need for longer peptide sequences (20 or more AAs) to ensure stable helical folding, (2) the difficulties associated with de novo sequence design, and (3) the unpredictability of the self-association model.[83] Despite these challenges, peptides with helical structures can be precisely programmed into tertiary structures of cross-α architecture or/and quaternary structures of coiled-coils domains, which can further arrange into protofilaments and nanotubes.[84,85] Additionally, some surfactant-like oligopeptides and rod-coil diblock polypeptides based on a poly(L) helical motif have been designed.[68,86,87] The assembly of these structures can be adjusted through packing parameters, leading to the formation of fibrils, nanotubes, and vesicles.

In contrast to the organization and order found in structured proteins, intrinsically disordered proteins (IDPs) can exhibit LLPS in disordered phases. These IDPs usually have repeating motifs of low sequence complexity with multivalency, which allows for multiple weak and transient contacts among the repeated units.[15] Oligopeptide sequences derived or inspired from IDPs aim to recreate the phase separation and to understand the molecular-level mechanisms underlying coacervation. Examples include peptides derived from mussel foot proteins (GK-16*: GYKGKYYGKGKKYYYK), histidine-rich peptides from squid beak proteins (GY-23: GHGLY GAGFA GHGLH GFA GHGLY), and other designed oligopeptide motifs.[72,88] Phase separation is driven and/or regulated by electrostatic, hydrophobic, or cation-π interactions, or a combination thereof. In addition to LLPS triggered by a single peptide, the attraction of a peptide with other molecules into a phase can also occur. The propensity for phase separation can be tuned through the length and charge density, as well as the ionic strength, with typical examples, including polylysine/nucleotides[89] and polylysine/polyglutamate.[90]

Many functional proteins in biological systems fold into specific, ordered, and stable 3D structures, leading to macroscopic materials with hierarchical architecture that possess remarkable mechanical properties.[91] Current research on the self-assembly of oligopeptides is primarily focused on optimizing ordered structures such as fibers and their resulting functionalities to mimic these proteins. This approach is largely driven by thermodynamic considerations. However, IDPs, which lack specific secondary or tertiary structures but are highly flexible and adaptable play distinct roles in functional phenomena like allosteric regulation and enzymatic catalysis.[92] Recent research has leveraged the integration of order and disorder, applying kinetic control over the ordered structures and creating accessibility to systems with dynamic and temporal features.[93] The self-assembly of oligopeptides is now evolving towards dynamic responsiveness and adaptability to applied stimuli, enabling self-organization in a more kinetically controlled manner, which can be regulated in a precise space and a specific time frame.

Stimuli-responsive assembly of oligopeptides

The specific morphology of assembled peptides is a result of the balance and competition among various noncovalent interactions. External stimuli such as pH, salt, redox conditions, enzymes, and temperature can manipulate these self-assembled morphologies. pH and ionic strength have great impacts on electrostatic interactions. This is a reflection of the intricate interplay of driving forces being affected. pH and ionic strength significantly impact electrostatic interactions, while increasing temperature can enhance hydrophobic interactions and weaken hydrogen bonding between water and the amide carbonyls.[75] Thermal annealing can prompt a transition from nanofibers to nanosheets in specific peptide amphiphiles due to heightened hydrophobic interactions and stronger H-bonding among peptides, which results in more organized β-sheet arrangements.[28] Moreover, certain short peptide motifs undergo LLPS when the temperature rises, attributed to increased hydrophobic interactions.[15,72] The escalation in desolvation from elevated temperatures can further initiate their assembly into nanofibers or nanobelts.[88,94] Additionally, while covalent bond formation augments structural integrity,[44,95] it also acts as a chemical stimulus to catalyze the self-assembling process.[96,97] A recent example of this is the precursor peptide reacting with a hydrophobic tail to form a covalent oxime bond, producing a peptide amphiphile that subsequently assembles into fibers.[45]

Ionic AAs such as lysine (K), histidine (H), and glutamate (E) can undergo protonation and deprotonation in response to pH changes. For instance, protonation of glutamate residues at pH 6.8 can induce the assembly of a peptide amphiphile into nanofibers and hydrogelation due to the suppression of electrostatic repulsion at head groups. However, these structures cannot form at pH 7.4 (Figure 2a).[98]

DOI: 10.31635/ccschem.023.202303209
Citation: *CCS Chem.* **2024**, 6, 69–90
Link to VoR: https://doi.org/10.31635/ccschem.023.202303209

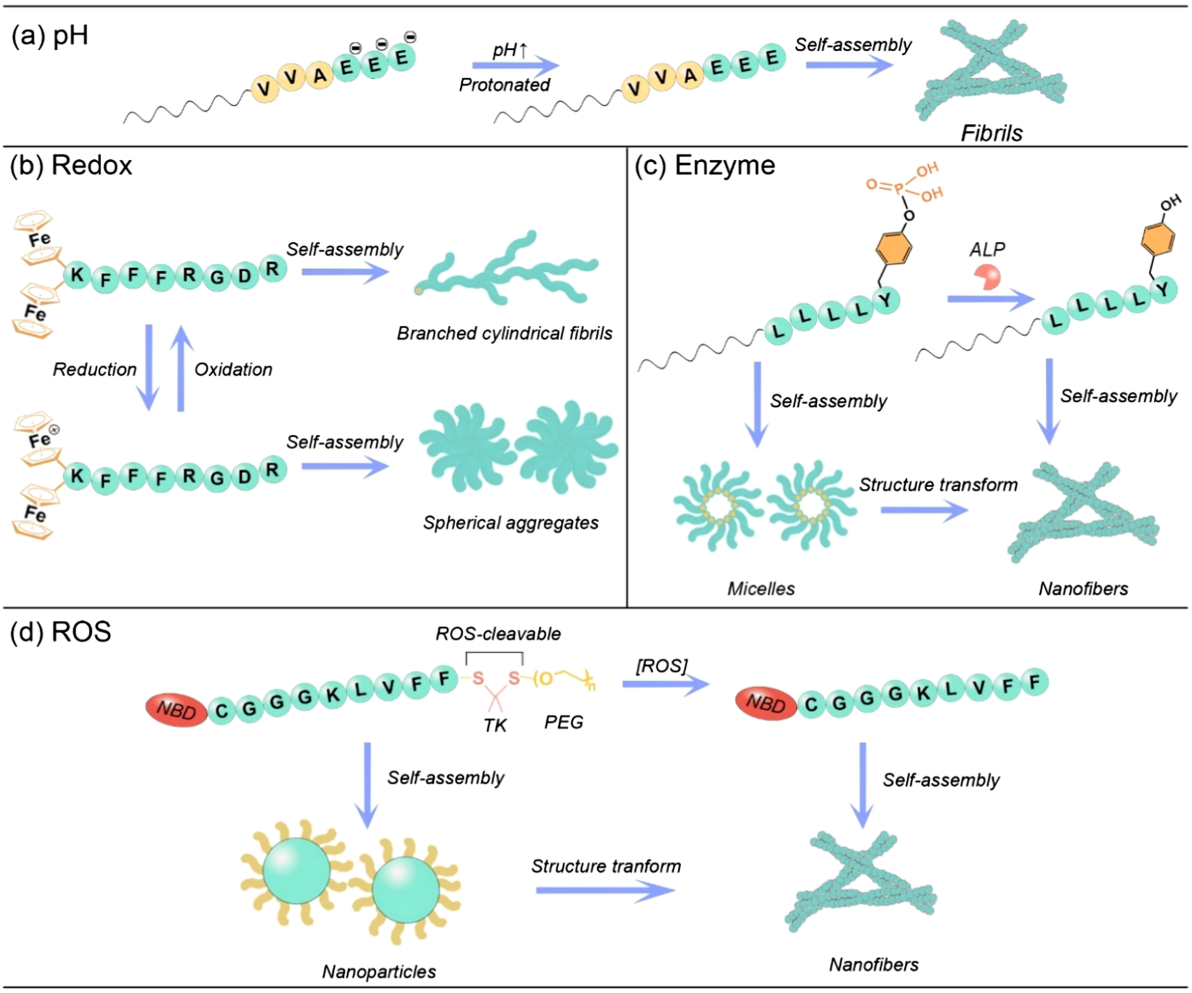


Figure 2 | *(a) pH-induced deprotonation or/and protonation of certain AAs (such as lysine, histidine, glutamate, etc.) which stimulates self-assembly of peptide amphiphiles. For example, the protonation of glutamate residues at pH 6.8 induces the assembly of an oligopeptide amphiphile into nanofibers and hydrogelation.[98] (b) Redox-controlled oligopeptide self-assembly. Reduction of Fc^+ to Fc by the reductant ascorbic acid leads to the increase of its hydrophobicity and aromaticity, thus leading to the formation of cylindrical fibrils. Meanwhile, oxidation into Fc^+ by the oxidant $Fe(ClO_4)_3$ with enhanced head charges induces the assembly of spherical micelles.[99] (c) Reactive oxygen species (ROS)-responsive oligopeptide self-assembling systems. Cleavage of a ROS-sensitive thioketal group induces fiber formation due to the loss of the hydrophilic PEG chain.[100] (d) Enzymes are mostly exploited for the intracellular transformation of synthetic oligopeptide precursors into activated monomers for self-assembly. The phosphopentapeptide, self-assembling to form micelles, which transforms into nanofibers upon the dephosphorylation catalyzed by ALP.[58]*

Redox-sensitive peptides can be triggered to change their morphology in oxidative-reductive environments. The redox moieties utilized include thiol-ether bonds, ferrocene (Fc), thioketal, and others.[101] The conversion of Fc to ferrocenium (Fc^+) through oxidation alters its hydrophobicity and aromaticity. Fc_2FKFFFRGDR with the Fc^+ terminals assembled into spherical micelles, but it formed fibrils with the reduction of Fc^+ into Fc (Figure 2b).[99] Redox conditions naturally occur in inter and intracellular environments, enabling spatiotemporal control over the formation of intracellular nanostructures and targeted functionalities. Glutathione (GSH), rich in eukaryotic cells, acts as a reducing agent to induce the cleavage of disulfide bonds.[102] Reactive oxygen species (ROS) are overproduced in abnormal cells and can promote tumorigenesis and proliferation. Thus, ROS-oxidation-driven self-assembly can specifically target cancer cells, creating advanced delivery systems and prodrugs that can distinguish between normal and abnormal cells or between intra- and extracellular compartments.[102] A polymer-peptide conjugate (PPC) was synthesized with β-sheet-forming peptide KLVFF that coupled with a hydrophilic poly(ethylene glycol) (mPEG) via thioketal moiety (Figure 2c).[100] The conjugate collapsed into

DOI: 10.31635/ccschem.023.202303209
Citation: *CCS Chem.* **2024**, 6, 69–90
Link to VoR: https://doi.org/10.31635/ccschem.023.202303209

nanoparticles at normal physiological conditions, whereas transformed into fibrillar nanostructures due to the cleavage of mPEG upon the oxidation of thioketal by the overgenerated ROS (measured in μM) in tumor cells.

Intracellular transformation of peptide precursors into self-assembling monomers, directed by enzymes, is specific to their cellular localization and substrate recognition motifs. The variety of intracellular enzymes allows for spatiotemporal tailoring of enzyme-triggered self-assembly.[103] A good example is the phosphopentapeptide NBD-LLLL$_p$Y, containing a leucine-rich segment and phosphotyrosine. The peptide can be catalyzed by the enzyme alkaline phosphatase (ALP) to undergo dephosphorylation (Figure 2d).[58] During this process, the surface charges decrease, which triggers a change in the conformation of the peptide to an α-helical structure. The change in conformation and surface charge promotes the transformation of the peptide's self-assembled structures from spherical micelles to more elongated structures such as fibrils or nanotubes.

The stimuli-responsiveness of peptide assembly in response to pH, redox conditions, and enzymes is particularly attractive for diagnostic and therapeutic applications. The transition from spherical aggregates to fibrillar structures triggered by specific intracellular cues can enhance their accumulation within tumors, inducing self-cytotoxicity. As a result, they serve as promising nanomedicines with sustained prodrug release and improved drug efficacy.

Nonequilibrium self-assembly of oligopeptides

Nonequilibrium self-assembly is essential in biological systems for the creation of dynamic, adaptable, and functional architectures. This mechanism allows biological systems to actively respond to stimuli, utilize energy, process information, and adapt to changing environments. While equilibrium self-assembly has its importance in generating stable structures, it often lacks the dynamic and functional capabilities observed in nonequilibrium systems. As such, there is a growing need to develop assembly systems with life-like properties such as dynamism and adaptability.[93]

By exploring the energy landscapes of assembled products, we can gain insight into the influence of thermodynamic and kinetic factors on molecular organization and aggregated structures. Peptide self-assembling systems can be categorized into four states: equilibrium, kinetically trapped, metastable, and dissipative states (Figure 3a).[25,104,105] The state of thermodynamic equilibrium is found at the lowest energy valley in the landscape. However, this state is not static, but enables the continuous exchange of substances at an equal rate during the formation and dismantling of assembled structures. Kinetically trapped and metastable assemblies are found at the local energy minimum, with the energy barrier of the kinetically trapped state being higher than the metastable one.[104] Kinetically trapped systems generally have a longer stability duration than the timescale of experimental observation, often requiring external triggers to escape from the trapped state. In contrast to these closed systems, dissipative nonequilibrium systems need a continuous energy flux to drive the assembly process. The assembly can only take place when energy fuel is presented, and the system will revert to the precursor when the fuel is used up[107] (Figure 3b).

Time-dependent assembling is a key characteristic of nonequilibrium assembly.[108] Rapid condensation of monomers may result in 'erroneous' binding due to nonspecific and weak noncovalent interactions, which lead the assembly into a kinetically trapped state. This eventually transitions into the most stable aggregates over the annealing period. This is observed in the transition of gels to crystals in diphenylalanine (FF) derivatives over time, as well as in assemblies that evolve from liquid condensates. The self-assembly of Z-FF evolved from the initial liquid droplet to several intermediates and finally to the thermodynamically stable nanofibrils (Figure 3c).[73] These instances of time-dependent evolution involve a rearrangement from random intermolecular interactions into more robust and long-ordered organizations.[30] Additionally, the association and growth of these aggregates into larger sizes and dimensions is also a time-dependent process. This has been observed in the Ostwald ripening of Boc-FF, in which a multistep nucleation and growth process was noted, including the structural transformation from soluble monomers to spherical assemblies, then to nanofibers, and ultimately to nanotubes.[41]

Solvation plays a crucial role in peptide folding and stabilization. Even trace amounts of osmolytes like urea can modify the stability and function of peptides, primarily through favorable interactions with protein backbones via hydrogen bonding.[87] The presence of a cosolvent during the assembly process can prompt the formation of kinetically-trapped intermediates. Moreover, variations in the sequence of sample preparation can lead to different morphologies. For example, the assembling of a peptide amphiphile (F_6C_{11}) transited from nanosheets to fibrils with increasing methanol (MeOH) concentration (Figure 3d).[71] The solvency of MeOH can reduce the polarity of the solvent, thereby weakening the hydrophobic interactions that occur during peptide association. Interestingly, the preformed nanosheets did not disassemble or transform into fibrils in over 2-month observation when the cosolvent solution was added. This highlights the stability of some kinetically-trapped assemblies and the dependency of the assembly process on specific pathways.

Biological systems regulate their structures and functions through chemical reaction cycles. A classic

DOI: 10.31635/ccschem.023.202303209
Citation: *CCS Chem.* **2024**, 6, 69–90
Link to VoR: https://doi.org/10.31635/ccschem.023.202303209

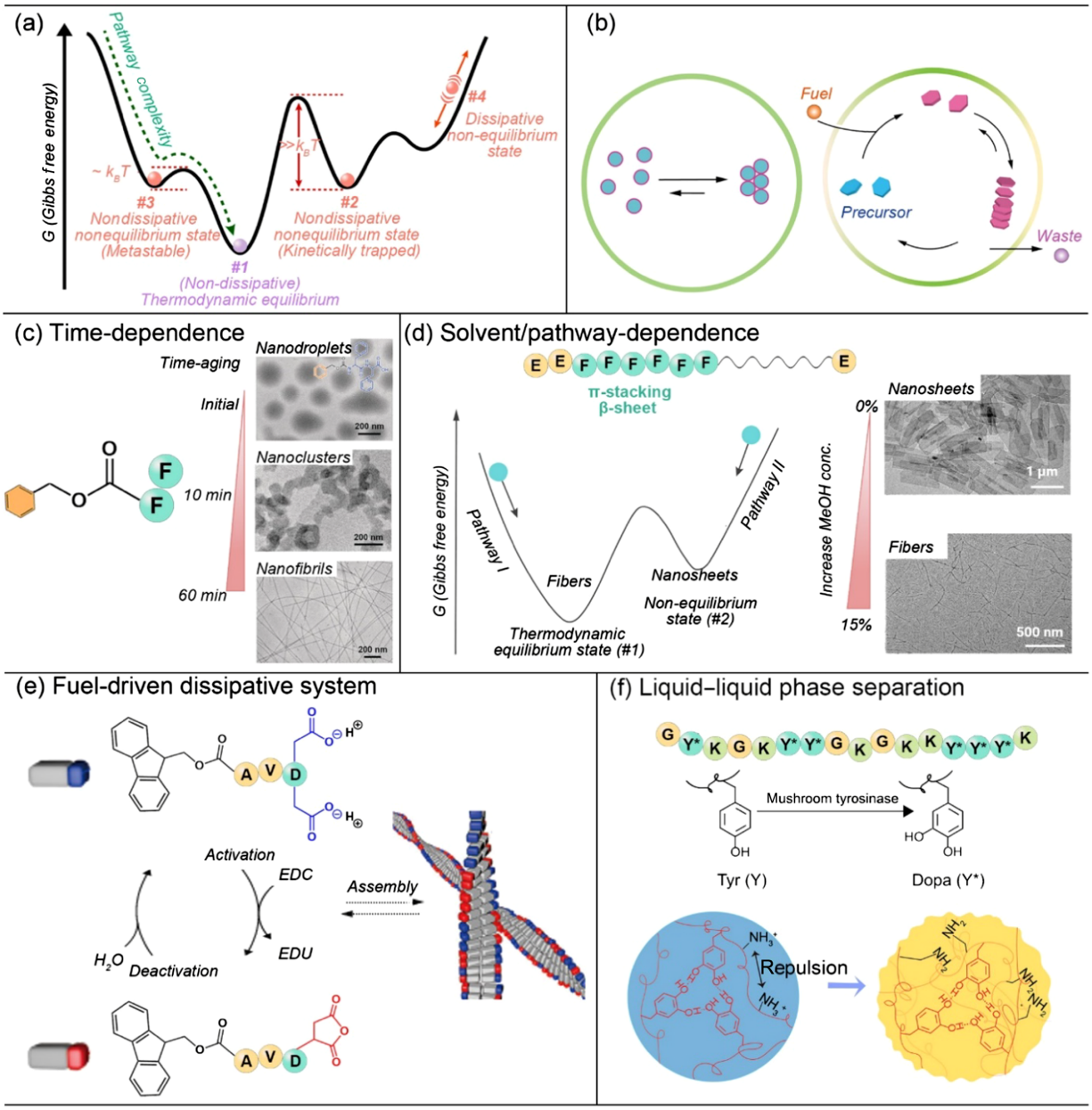


Figure 3 | *(a) Schematic illustration of Gibbs free energy landscape.[104] Reproduced with permission from ref 104. Copyright 2017 Royal Society of Chemistry. (b) Schematic illustration of a self-assembling system at equilibrium or in a kinetically trapped state (or metastable) and a chemical energy flux driving the equilibrium towards the aggregated state.[105] (c) Z-FF self-assembly with time-lapse. The structural evolution of Z-FF from the metastable liquid droplets at initial status to nanofibrils after 1 h time-aging.[73] Adapted with permission from ref 73. Copyright 2019 John Wiley & Sons, Inc. (d) The nanosheet-to-nanofibril (2D-to-1D) structural evolution occurs with an increased methanol concentration. A diagram showing the energy landscape of solvation-directed self-assembly, where pathway I underwent the thermodynamic equilibrium state with the formation of fibrils (state 1) and pathway II led to the formation of nanosheets (state 2) at a kinetically trapped state.[71] Adapted with permission from ref 71. Copyright 2019 ACS Publications. (e) The intramolecular anhydrides are formed when the building blocks (such as Fmoc-AVD) bearing two carboxylic groups are activated by fuel chemical EDC. The activated building blocks can assemble into fibrils while at the same time, it can be hydrolyzed back to its precursor with carboxylic acid by reacting with water.[106] Adapted with permission from ref 106. Copyright 2020 ACS Publications. (f) The pH-regulated electrostatic repulsion between Lys residues can tune the liquid-to-gel transition of the coacervate.[37] Adapted with permission from ref 37. Copyright 2022 Springer Nature Limited.*

DOI: 10.31635/ccschem.023.202303209
Citation: *CCS Chem.* **2024**, 6, 69–90
Link to VoR: https://doi.org/10.31635/ccschem.023.202303209

example of this is the cycle of catastrophe and rescue in tubulin, driven by reaction cycles between guanosine triphosphate (GTP) and guanosine diphosphate (GDP).[29] This process is intrinsically regulated by the free energy liberated from reaction conversions. To mimic such dynamic processes, oligopeptide systems that can facilitate chemically fueled self-assembly are created to exercise spatial and temporal control over material structures and properties.[109] The Boekhoven group has designed a series of carbodiimides-based reaction cycles in which carbodiimides act as fuels to drive peptide coupling. The intramolecular anhydrides at the sequence terminus (such as Fmoc-AVD) can be activated using 1-ethyl-3-(3-(dimethylamino)propyl) carbodiimide (EDC) (Figure 3e).[106] The deactivation process involves the hydrolyzation of the activated monomer back to its carboxylic acid form. These activated monomers, serving as intermediate products, can assemble into fibers. The kinetics of assembly and disassembly, as well as the lifespan of the building blocks, depend on the associated reaction rates, resulting in a dynamic assembly system.

Oligopeptides were also engineered to replicate the pathological states observed in neurodegenerative diseases, which are thought to be related to the transition of DNA/RNA-binding FUS proteins from a liquid droplet population into fibrillar structures.[110] The peptides first form LLPS, followed by the nucleation and growth of fibers. This research seeks to better understand the fundamental principles that govern order and disorder in a biological context. Minimalistic amphiphilic peptides, consisting of positively charged components from a mixture of LVFFAR9 and R9 and negatively charged counterparts such as adenosine triphosphate (ATP), were designed.[111] Interactions among R9/ATP triggered the formation of liquid droplets. The coassembly of these three components led to a local increase in the concentration of LVFFAR9, which further organized into thermodynamically stable fibers inside coacervates. The R9 terminal is proposed to stabilize the interface between the amyloid fibrils and the condensate. The kinetics of assembly can be regulated by the ratio of components, and the nucleation and growth of fibers occur over time.

Liquid-to-solid transition in the LLPS system can also be initiated by manipulating the interactions. The LLPS behavior formed by mussel-inspired GK-16* can be controlled through fine-tuning electrostatic interactions and DOPA-mediated H-bonding.[37] Suppressing electrostatic repulsion by increasing the pH from 3 to 7 triggers the solidification of liquid droplets (Figure 3f).

Oligopeptides have been engineered to construct complex supramolecular systems, incorporating multiple dynamic features such as molecular recognition, preorganization, induced fit, and spatiotemporal control. These advancements aim to develop active and adaptive life-like materials. Such systems are poised to revolutionize biomedical applications in terms of programmability, automation, and intelligence. By harnessing the principles of nonequilibrium self-assembly, researchers aim to design new functional materials, drug delivery systems, and artificial molecular machines, all with enhanced capabilities and adaptability.

Biomedical application of oligopeptide self-assembling materials

The biomedical field is seeing the emergence of an intriguing area driven by the use of self-assembling oligopeptide materials.[32] These impressive biomaterials, with their natural tendency towards spontaneous organization, hold immense promise for revolutionizing multiple aspects of medicine. Self-assembling oligopeptide materials have created new pathways for transformative interventions in various biomedical applications from targeted drug delivery and regenerative therapies to tissue engineering and bioimaging.[112] By harnessing their unique physicochemical properties and intricate self-assembly mechanisms, these materials offer unprecedented opportunities to address complex biomedical challenges in a sophisticated and precise manner.

Tumor therapy

Cancer has persisted as a significant threat to human health, necessitating the development of various therapeutic strategies such as chemotherapy, photodynamic therapy (PDT), and immunotherapy, among others.[113–117] Self-assembling oligopeptide materials have emerged as a prominent platform in cancer therapy, thanks to their inherent superior biocompatibility, biodegradability, and structural and functional versatility.[118] They show extraordinary potential in addressing the complexities of cancer treatment by facilitating precise and controlled interventions.

Chemotherapy remains a cornerstone in cancer treatment.[119,120] Self-assembling oligopeptide materials possess the ability not only to deliver chemotherapeutic drugs to tumors but also to leverage their responsive properties within the tumor microenvironments (TME) for effective drug release, thereby enabling targeted cancer treatment.[20,52,121,122] Hou et al.[123] designed an oligopeptide, named polysaccharide peptide (PSP), consisting of a hydrophobic *N*-terminal pentavaline (VVVVV) moiety, a pH-responsive segment (HH), and a hydrophilic *C*-terminal sequence (RGDC) (Figure 4a). By conjugating it to a D-peptide activator of tumor suppressor p53 termed ^{D}PMI (sequence: T^{D}A^{D}W^{D}Y^{D}A^{D}N^{D}F^{D}E^{D}A^{D}L^{D}L^{D}R), a self-assembling peptide with tumor microenvironment (TME) stimuli responsiveness named PSP-^{D}PMI were generated. Under physiological conditions (pH 7.4), PSP-^{D}PMI could self-assemble into nanoshells, facilitated by the presence of the PSP module. However, at pH 6.5,

DOI: 10.31635/ccschem.023.202303209
Citation: *CCS Chem.* **2024**, 6, 69–90
Link to VoR: https://doi.org/10.31635/ccschem.023.202303209

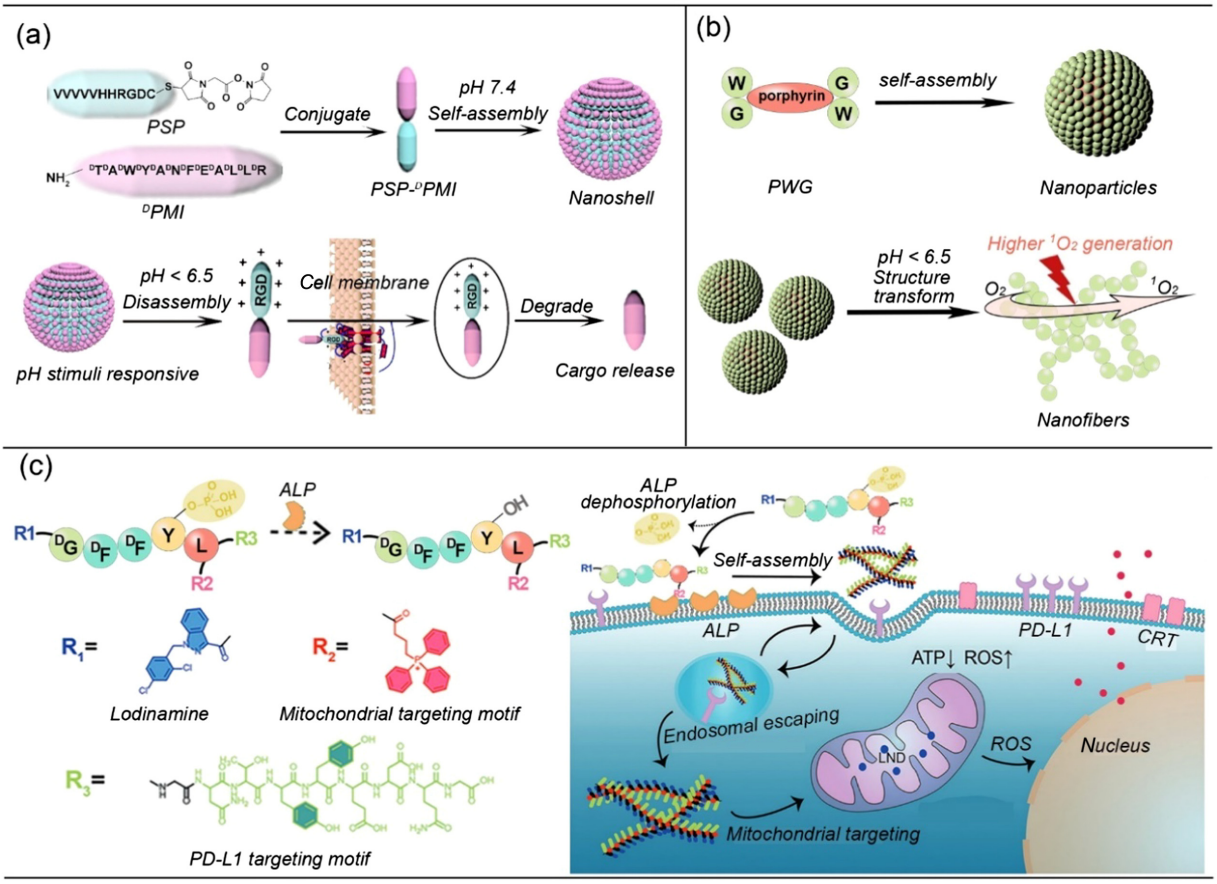


Figure 4 | *(a) Schematic illustration of the pH-responsive cargo release of PSP-^{D}PMI self-assembly spherical shells.[123] Adapted with permission from ref 123. Copyright 2018 ACS Publications. (b) Schematic illustration of self-assembly and fibrillar transformation of the acid-activated peptide-porphyrin (PWG) nanoparticles and their application in photodynamic therapy (PDT).[124] Adapted with permission from ref 124. Copyright 2020 John Wiley & Sons, Inc. (c) Schematic illustration of the cascade-targeted enzyme-instructed peptide and the proposed mechanism to boost immunogenic cell death (ICD).[125] Adapted with permission from ref 125. Copyright 2023 John Wiley & Sons, Inc.*

protonation of histidine (His) residues within the PSP triggered the disintegration of the nanoshells. After being internalized through RGD (Arg-Gly-Asp)-integrin-mediated endocytosis, the PSP peptide, functioning as a degradable accessory, was expected to undergo rapid degradation by endosomal/lysosomal peptidases. This degradation process would subsequently lead to the release of ^{D}PMI, enabling its activation of the p53 signaling pathway for anticancer therapy.

Moreover, oligopeptides have been employed to coassemble with photosensitizers utilizing multiple weak intermolecular interactions to construct self-assembled nanostructures for PDT.[44,126–130] PDT has attracted considerable attention in recent decades as an emerging therapeutic modality. It utilizes photosensitizers to produce cytotoxic ROS under specific wavelengths of light effectively killing cancer cells.[131,132] For example, a pH-responsive dipeptide, tryptophan-glycine (WG), was coupled to a hydrophobic photosensitizer, porphyrin (P), through the process of amidation, resulting in a porphyrin-tryptophan-glycine (PWG) oligopeptide-photosensitizer coassembled nanostructure for PDT (Figure 4b).[124] Under normal physiological conditions (pH = 7.4), PWG self-assembled into nanoparticles. Upon reaching the tumor tissue, the enhanced acidity of the TME facilitates the protonation of PWG, leading to the formation of intermolecular hydrogen bonds and subsequent transformation of the nanoparticles into nanofibers. Thanks to enhanced intersystem crossing, the fibrillar transformation of PWG nanostructures results in higher ROS production and improved PDT therapeutic efficacy.

In recent years, the utilization of oligopeptide self-assembled materials in cancer immunotherapy has witnessed a remarkable surge.[20,133–136] This innovative approach has shown significant success in treating diverse malignant tumors, particularly in the realms of adoptive T-cell immunotherapy and immune checkpoint therapy. Oligopeptide self-assembling materials offer several advantages for tumor immunotherapy: Firstly, they enhance the efficiency of immune checkpoint blockade by

DOI: 10.31635/ccschem.023.202303209
Citation: *CCS Chem.* **2024**, 6, 69–90
Link to VoR: https://doi.org/10.31635/ccschem.023.202303209

providing improved affinity and spatial hindrance through self-assembling peptide inhibitors. Secondly, when combined with immune checkpoint blockade or vaccines, oligopeptide self-assembling nanocarriers can address challenges such as poor immune response, high relapse rates, and limited target population; thus, improving overall therapeutic outcomes. A tetra-peptide self-assembling motif of -G^{D}F^{D}F^{D}Y-, as the core self-assembling skeleton, was conjugated with a mitochondria-specific drug lonidamine, a mitochondria-targeting motif triphenylphosphonium (TPP) and a programmed death ligand 1 (PD-L1)-specific sequence (NTYYEDQG) to construct the cascade-targeted enzyme-instructed peptide (Figure 4c).[125] Worthwhile, after dephosphorylation of -G^{D}F^{D}F^{D}Y- by overexpressed ALP in tumors, this peptide self-assembled into nanofibrils and precisely delivered lonidamine into mitochondria. The inclusion of the PD-L1-specific motif in the self-assembling peptides allowed for precise targeting and interaction with cancer cell membranes, effectively guiding their delivery to intracellular mitochondria. This self-assembling peptide leads to mitochondrial dysfunctions, triggering immunogenic cell death (ICD) responses and creating an immunogenic TME achieving favorable anticancer efficiency.

In addition to the above-mentioned peptide self-assembly materials that are indirectly involved in tumor therapy as therapeutic agent carriers, some oligopeptides can form supramolecular assemblies through self-assembly induced by enzymes and selectively inhibit tumor cells through various mechanisms.[103,137–139] For example, a carbohydrate phosphate derivative, reported by Pires et al.,[140] undergoes intracellular self-assembly to form nanofibers to selectively inhibit the metabolic activity of osteosarcoma (Saos-2) cells. Xu et al.[141] reported that intranuclear nanoribbons formed upon dephosphorylation of leucine-rich L- or D-phosphopeptide catalyzed by ALP to selectively kill osteosarcoma cells. One notable advantage of the enzyme-instructed self-assembly of oligopeptides for cancer cell eradication is its selective targeting ability, specifically aimed at cancer cells while sparing normal cells. Moreover, as peptide assemblies do not rely on traditional chemotherapeutic drugs, they circumvent the issue of drug resistance commonly encountered in conventional cancer treatments. The development of enzyme-responsive oligopeptide self-assembly systems holds significant promise for the advancement of precision cancer therapy and has the potential to revolutionize the field of oncology.

Bioimaging

Bioimaging plays a pivotal role in the field of biomedical research and clinical diagnostics by providing noninvasive visualization and characterization of biological structures and processes at various scales.[142] It encompasses a wide range of imaging techniques and modalities such as fluorescence imaging (FLI),[143–145] magnetic resonance imaging (MRI),[146] and photoacoustic imaging (PAI).[147,148] Oligopeptide self-assembling materials have emerged as promising candidates in the field of bioimaging due to their unique properties and versatile functionalities. By incorporating imaging agents such as fluorescent dyes or magnetic nanoparticles into the peptide assemblies, they can be transformed into functional nanoprobes for various bioimaging applications.

FLI is a powerful imaging technique that utilizes fluorescent probes to visualize and track specific molecules or biological processes in live organisms. It offers high sensitivity, real-time imaging capabilities, and excellent spatial resolution, allowing for noninvasive monitoring in vivo. Oligopeptide self-assembling materials could provide a stable and controlled environment for the encapsulation or conjugation of fluorescent dyes to protect the probes from degradation and enhance their photostability, enabling long-term and high-quality imaging.[149–151] More importantly, through the incorporation of stimuli-responsive oligopeptide sequences such as pH-sensitive or enzyme-cleavable motifs, the fluorescence emission of the probes can be modulated in response to specific microenvironmental changes. Nitroreductase (NTR) is an enzyme overexpressed in hypoxic regions such as solid tumors, capable of catalyzing the reduction of nitro groups to (hydroxy)amino groups.[152] Inspired by this, Yu et al.[153] prepared an NTR-responsive dye-functional oligopeptide based on histidine modified with nitroimidazole units (Figure 5a). Upon interaction with NTR, this oligopeptide underwent a transformative process, transitioning from its initial nanofiber structure to the formation of nanospheres. After the morphology-transformation process of the oligopeptide, the fluorescence of the conjugated dye, IR780, was recovered from the quenched state. These oligopeptide-based probes could penetrate solid tumors; thus, allowing for efficient FLI of hypoxic tumors. Apart from the direct incorporation of fluorescent molecules into oligopeptide self-assembly materials, the inherent photoluminescent phenomenon generated by the formation of nanoscale quantum confinement structures within the internal hydrogen bonding and aromatic interaction regions of these oligopeptide self-assembly materials can also find application in the field of bioimaging.[155,156] For example, Zhang et al.[157] drew inspiration from naturally occurring organized assemblies to create tryptophan-phenylalanine nanoparticles. These nanoparticles exhibited a remarkable ability to shift the peptide's intrinsic fluorescence signal from the ultraviolet to the visible range. This shift allowed the dipeptide nanoparticles (DNPs) to serve as versatile imaging and sensing probes.

With their remarkable properties and controllable assembly, oligopeptide self-assembling materials have exhibited exceptional promise in the realm of MRI. By ingeniously engineering these materials, one can

DOI: 10.31635/ccschem.023.202303209
Citation: *CCS Chem.* **2024**, 6, 69–90
Link to VoR: https://doi.org/10.31635/ccschem.023.202303209

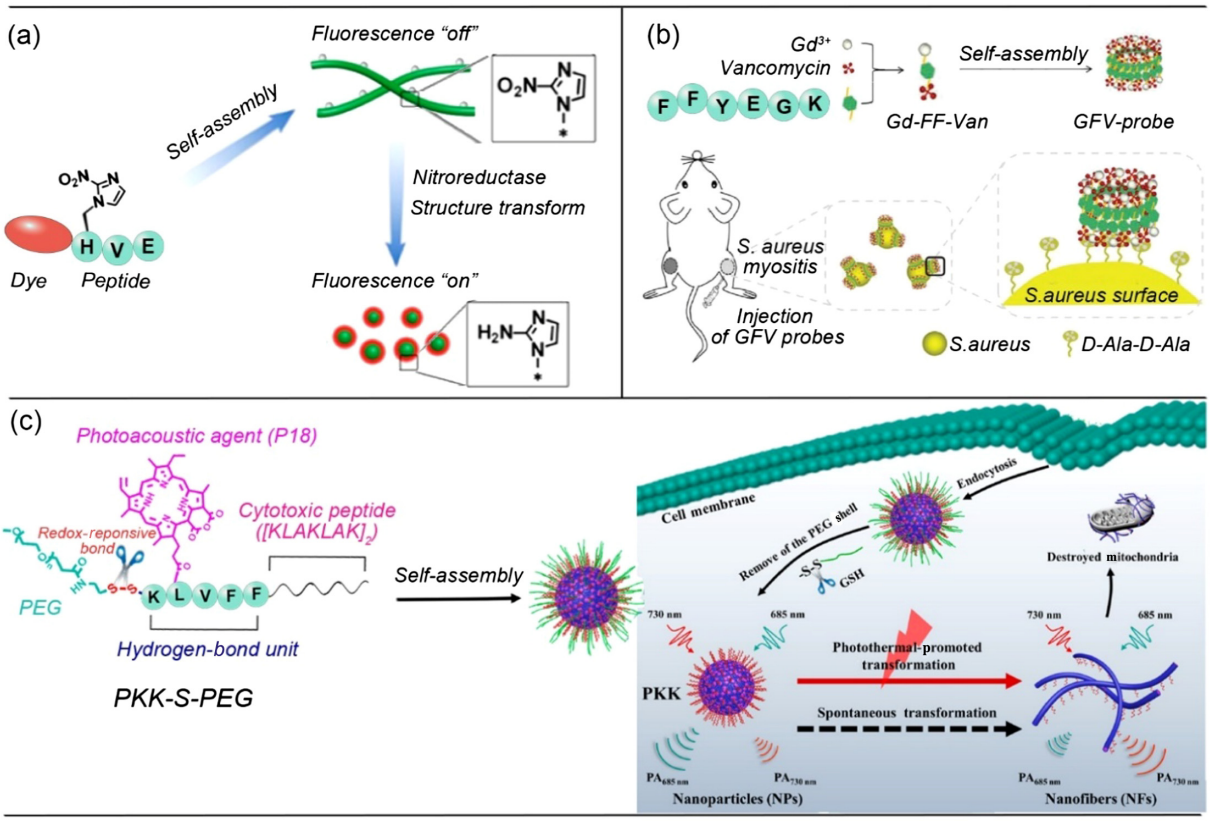


Figure 5 | *(a) Schematic illustration of the morphology-transformable supramolecular probes via coassembling dye and peptide, in which NTR-reduction led to the morphological transition of the nanostructures and recovery of the fluorescence of dye, respectively. The gray dots along the nanofibers and the red circles around the particles denote the quenched and luminescent dye, respectively.*[153] *Adapted with permission from ref 153. Copyright 2021 ACS Publications. (b) Schematics of the preparation of multifunctional GFV probe and bacterial infection imaging in vivo based on GFV probe.*[154] *Adapted with permission from ref 154. Copyright 2021 John Wiley & Sons, Inc. (c) Schematics of the structure of reduction responsive polymer–peptide conjugates and in vivo PAI.*[147] *Adapted with permission from ref 147. Copyright 2020 ACS Publications.*

embed paramagnetic or superparamagnetic agents like gadolinium-based complexes or iron oxide nanoparticles, which effectively amplify the contrast in MRI scans. This unique amalgamation of oligopeptides and the MRI technique offers an extraordinary avenue to acquire intricate and highly detailed images of the body's internal structures, harnessing the power of potent magnetic fields and contrast agents.[158,159] Through chelating with paramagnetic Gd^{3+} as T1 signaling unit and conjugating with *Vancomycin* as target ligand, the oligopeptide, FFYEGK (FF), could self-assemble into a nanoaggregate probe (named as GFV) due to the - stacking of "Phenylalanine–Phenylalanine" (Figure 5b).[154] Thanks to the nanostructure, the GFV probes had a high longitudinal relaxivity rate (r1). The GFV probe exhibited specific imaging of *Staphylococcus aureus* at the infection site by specifically binding to the cell walls of *S. aureus* through hydrogen-bonding interactions between *Vancomycin* and the D-alanyl-D-alanine dipeptide to distinguish between bacterial infections from sterile inflammations. Moreover, the GFV probe could monitor the efficacy of Daptomycin for *S. aureus*-infected mouse models, proving the potential clinical application of this oligopeptide self-assembling probe.

Excluding the aforementioned applications in FLI and MRI, oligopeptide self-assembly materials find extensive utility in other imaging modalities such as PAI and radiology imaging (RI).[160,161] For example, Wang et al.[147] introduced a novel strategy involving the use of photothermal molecules to accelerate the transformation process of transformable PPCs (Figure 5c). The designed reduction-responsive PPC, named as PKK-S-PEG, was composed of four key components: a therapeutic peptide targeting mitochondria, a hydrogen-bonding peptide motif, GSH-responsive disulfide bonds, and a photothermal/PA molecule (P18). Once inside the cancer cells, the disulfide bonds are cleaved by overexpressed GSH, leading to the transformation of NPs into nanofibers, driven by hydrogen bonding. Notably, the morphological transformation was controllably accelerated by the photothermal unit

DOI: 10.31635/ccschem.023.202303209
Citation: *CCS Chem.* **2024**, 6, 69–90
Link to VoR: https://doi.org/10.31635/ccschem.023.202303209

P18 under laser irradiation. The transformation process was monitored using PAI, enabling the evaluation of the nanofibers formation rate at the targeted site. Ultimately, this strategy enhanced the accumulation and retention in the tumor site, resulting in improved therapeutic efficacy, thereby offering a promising approach for tumor diagnosis and treatment through oligopeptide in vivo self-assembly.

Tissue engineering

Oligopeptide self-assembly materials, owing to their ability to mimic the ECM and offer precise molecular design, have emerged as a fascinating platform with immense potential in the field of tissue engineering.[2,162] For instance, researchers have successfully applied these materials in bone tissue engineering.[163,164] By designing oligopeptide sequences, it is possible to promote the adhesion and proliferation of bone cells, while providing the necessary cues for osteogenic differentiation. This results in the formation of tissue-engineered constructs that closely resemble native bone tissue.

Moreover, oligopeptide self-assembly materials hold significant promise for promoting axonal outgrowth, neuronal cell adhesion, and guidance.[165] One such example is the development of peptide-based scaffolds that mimic the composition and architecture of the ECM, providing a supportive environment for neuronal regeneration.[166,167] These scaffolds can enhance axonal outgrowth and guide the reconnection of damaged neural circuits. Another example is the incorporation of neurotrophic factors into oligopeptide self-assemblies. Neurotrophic factors such as nerve growth factor (NGF) or brain-derived neurotrophic factor (BDNF) play crucial roles in neuronal survival, growth, and differentiation.[168,169] By encapsulating or immobilizing these factors within self-assembled peptide structures, their controlled release can be achieved, promoting the survival and regeneration of neurons in damaged areas. Furthermore, oligopeptide self-assembly materials can be functionalized with cell-adhesive peptides or ligands such as RGD (arginine-glycine-aspartic acid) motifs to enhance neuronal cell adhesion and guidance.[170] These functionalized materials provide specific cues for cell attachment and direct neurite extension along desired pathways, enabling the directed growth of neurites and the establishment of functional neural networks.

Other biomedical applications

Furthermore, oligopeptide self-assembling materials still have other biomedical applications such as antimicrobial,[171] bioprinting,[172] bioadhesive,[37] and so on. These diversified applications are mainly attributed to the excellent biocompatibility of oligopeptide and its various self-assembling structures and forms.

Oligopeptides, with their intriguing ability to form self-assembling structures and their inherent antimicrobial properties, emerge as compelling contenders for combating microbial infections. These infections have long been acknowledged as a formidable menace to human health, wreaking havoc by instigating diseases like inflammation, sepsis, and impairment of vital physiological processes such as wound healing. Capitalizing on the remarkable attributes of oligopeptides and their propensity to organize into self-assembling configurations adds an extra layer of allure to their antimicrobial potential. This exceptional amalgamation of oligopeptides and their antimicrobial prowess offers a distinctive avenue for addressing microbial infections, safeguarding human well-being, and mitigating the detrimental impact of such infections on vital bodily functions.[173,174] The self-assembling nanostructures of oligopeptide could provide several advantages such as increased stability, enhanced drug delivery, and prolonged release of antimicrobial agents. Furthermore, the design flexibility of oligopeptide sequences allows for the incorporation of specific targeting motifs to selectively kill pathogens while minimizing harm to healthy cells or tissues. Enzyme-instructed self-assembly (EISA) is a unique approach that integrates enzyme catalysis with peptide self-assembly, enhancing antimicrobial efficiency and imparting additional physicochemical properties to antimicrobial peptide particles. Tyrosinase, an enzyme that is widely presented in the human skin, controls the production of melanin and provides the possibility of in situ enzymatic oxidation and self-assembly. An in situ tyrosinase-induced self-assembly antimicrobial oligopeptide powder with the sequence, WRWRWY, was prepared by Du et al.[175] to effectively heal the infected wound (Figure 6a). The hydrophobic tryptophan (W) within the antimicrobial peptide contains an indole ring that exhibits lipophilic properties, providing stability in bacterial membranes and facilitating its adherence to them. Upon spraying the aqueous oligopeptide solution at a wound site, the presence of tyrosinase resulted in the oxidation of antimicrobial peptide monomers, leading to their self-assembly into mWRWRWY nanoparticles due to the reduced solubility following oxidation. The mWRWRWY nanoparticles, in comparison with WRWRWY, demonstrated an amplified positive charge density on their surface, resulting in a superior antimicrobial effect.

Three-dimensional (3D) bioprinting is an innovative and rapidly evolving technology that has garnered significant attention in recent years. It has the potential to revolutionize the field of tissue engineering and regenerative medicine by enabling the precise fabrication of complex, functional, and biomimetic 3D structures. With its ability to create intricate architectures using bioinks composed of living cells, biomaterials, and growth factors, 3D bioprinting holds great promise for applications such as organ transplantation, drug screening, and

DOI: 10.31635/ccschem.023.202303209
Citation: *CCS Chem.* **2024**, 6, 69–90
Link to VoR: https://doi.org/10.31635/ccschem.023.202303209

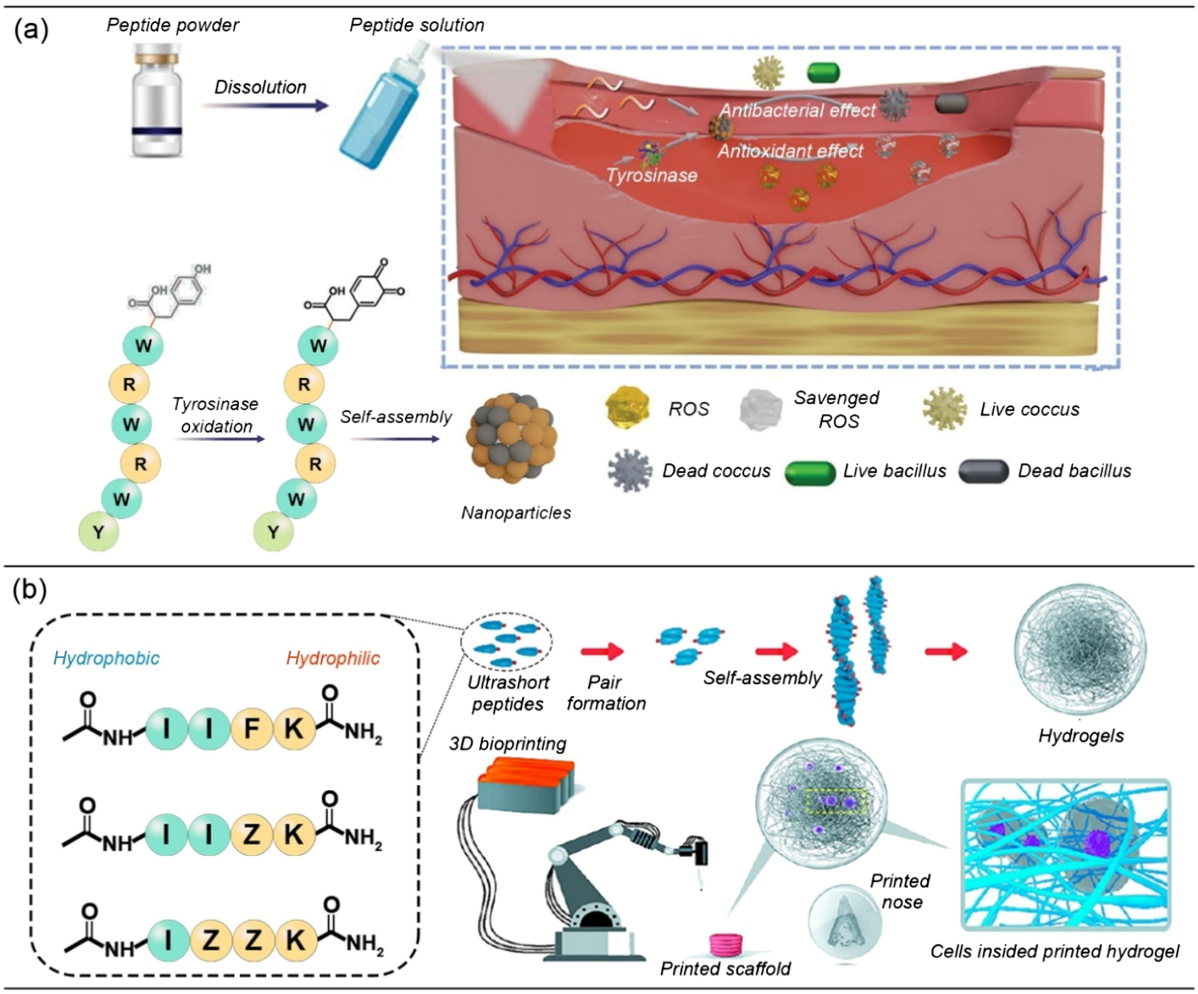


Figure 6 | *(a) Schematic illustration of in situ preparation of peptide nanoparticles to kill bacteria and promote wound healing.*[175] *Adapted with permission from ref 175. Copyright 2023 John Wiley & Sons, Inc. (b) Schematic illustration of self-assembling amphiphilic ultrashort peptides used for 3D bioprinting.*[176] *Adapted with permission from ref 176. Copyright 2021 ACS Publications.*

personalized medicine.[177] As programmable biomaterials, oligopeptide self-assembling materials can be utilized as the building blocks of bioinks, providing a structural framework for the 3D bioprinting process. The self-assembly behavior of oligopeptides allows precise control over the microenvironment within the bioink, influencing cell behavior and tissue development.[178,179] Three aromatic and nonaromatictetrapeptide amphiphiles sequences, including IIFK IIZK, and IZZK, were developed as potential bioinks for 3D bioprinting applications (Figure 6b).[176] All three peptides were able to form transparent hydrogels with high water content by self-assembling. The incorporation of these three oligopeptides into the bioink formulation enabled the creation of tissue scaffolds. The experimental findings demonstrated that the cell-containing bioinks effectively safeguarded the structural integrity of the cellular constructs throughout the demanding printing process, exhibiting long-term stability over several weeks. To address the challenges posed in improving the printing accuracy of peptide nanomaterials, fabricating multiscale scaffolds, and enhancing long-term biostability, researchers have employed several innovative strategies[180–182] such as optimized 3D bioprinting parameters, developed hybrid printing approach, surface modifications with bioactive molecules and growth factors, and so on.

Outlooks and Future Perspectives

Oligopeptide self-assembling materials show immense promise, with a wealth of untapped potential for future advancements. With the advent of novel design strategies and cutting-edge techniques, we find ourselves on the brink of uncharted territories in the realm of functional supramolecular assemblies. Researchers are delving deeper into the intricate molecular mechanisms governing self-assembly, setting the stage for

DOI: 10.31635/ccschem.023.202303209
Citation: *CCS Chem.* **2024**, 6, 69–90
Link to VoR: https://doi.org/10.31635/ccschem.023.202303209

revolutionary breakthroughs in diverse areas such as tissue engineering, drug delivery, biosensing, and more. By harnessing the synergistic interplay between structure, function, and stimuli responsiveness, oligopeptide self-assembling materials are poised to reshape the biomaterials landscape, offering unprecedented control, versatility, and tailor-made solutions to complex biomedical challenges. We anticipate a future where these materials pave the way for groundbreaking applications, ultimately improving human health and quality of life. Nonetheless, there are significant challenges and limitations that oligopeptide self-assembly materials must overcome.

A major limitation is our lack of comprehensive understanding of the nonequilibrium self-assembly mechanisms and structure-function relationships of oligopeptides. Despite significant progress in characterizing their assembly behaviors, there is a need for in-depth studies to elucidate the molecular interactions and kinetics involved in the nonequilibrium self-assembly process. This knowledge will facilitate the rational design of peptide sequences with improved assembly properties and tailored functionalities. Moreover, our incomplete understanding of the assembly mechanisms behind more complex methods such as multistage and gradient assembly complicates the precise control of these processes. The intricate interplay between molecular interactions, kinetics, and thermodynamics remains elusive in multistage and gradient assembly, impeding our ability to engineer complex and hierarchical structures with desired functionalities. The ability to precisely tune assembly parameters such as concentration, pH, temperature, and external stimuli is crucial for obtaining predictable and reproducible assembly outcomes.

Another significant challenge faced by the field of oligopeptide self-assembly materials is achieving scalability and reproducibility. The processes of synthesis and purification required to produce large quantities of high-quality oligopeptides can be intricate and expensive, thereby hindering the transition from laboratory-scale research to large-scale industrial production.[183] It is crucial to develop cost-effective and scalable manufacturing techniques that can satisfy the demand for these materials in clinical applications. Additionally, the stability and degradation properties of oligopeptide self-assemblies need to be thoroughly investigated. The long-term stability and biodegradability of these materials hold significant implications for their practical application in vivo.[184] A comprehensive understanding of the degradation kinetics, potential toxicity, and clearance pathways of oligopeptide assemblies is essential to ensure their safety and biocompatibility. Overcoming these challenges necessitates the use of cutting-edge analytical methods and advanced characterization techniques to evaluate the structural integrity and performance of oligopeptide assemblies. Novel strategies aimed at improving synthesis efficiency, enhancing reproducibility, and simplifying purification processes need to be explored. Moreover, the establishment of robust quality control measures and standardized protocols will be indispensable to ensure consistent and reliable production of oligopeptide self-assemblies on a large scale.[185]

Addressing these limitations and advancing the field of oligopeptide self-assembly will pave the way for their widespread use in biomedical applications. Successfully transitioning these materials from research laboratories to clinical practice has the potential to revolutionize drug delivery systems, tissue engineering, and regenerative medicine, ultimately leading to improved patient outcomes and quality of life.

Machine learning and artificial intelligence (AI) techniques can uncover hidden correlations and patterns by learning from extensive data. Such techniques present new opportunities and potential breakthroughs for oligopeptide self-assembly materials. Machine learning algorithms can analyze and predict the relationship between peptide sequences and assembly behavior.[186–188] Using existing experimental data, these models can identify specific patterns and structural features within peptide sequences, facilitating predictions about the assembly capabilities and modes of different sequences. Such AI tools provide researchers with a more efficient and precise method for designing and optimizing peptide materials with specific functionalities.

Additionally, AI techniques can expedite the screening and optimization processes for oligopeptide materials. By training on large-scale datasets, AI models can rapidly evaluate the assembly potential of various peptide sequences and predict material properties and functionalities.[189] This ability allows researchers to identify optimal peptide sequences and assembly conditions more quickly, thereby increasing the pace and efficiency of material development. For example, Li et al.[190] successfully developed a machine learning approach in 2019 to study the correlation between chemical features and the ability to form self-assembled dipeptide hydrogels. Sankaranarayanan et al.[191] reported an innovative machine learning workflow called AI-expert that integrated Monte Carlo tree search and random forest techniques with molecular dynamics simulations, enabling the development of a fully autonomous computational search engine for the exploration of peptide sequences with exceptional potential for self-assembly.

However, it is important to note that building and training AI models require abundant high-quality data and precise experimental measurements as inputs. Additionally, the interpretability of machine learning models can be a challenge, as they often rely on statistical correlations rather than fundamental physical principles.[192] Hence, it is crucial to combine machine learning results with experimental and theoretical considerations to assess their feasibility and reliability.

DOI: 10.31635/ccschem.023.202303209
Citation: *CCS Chem.* **2024**, 6, 69–90
Link to VoR: https://doi.org/10.31635/ccschem.023.202303209

With the continuing advancements in understanding the assembly mechanisms of oligopeptide self-assembly materials, exploring their diverse functionalities, and harnessing the power of computational approaches, we anticipate breakthroughs in designing and tailoring these materials for a broad range of applications. The integration of emerging technologies such as AI tools, bioinformatics, and nanotechnology will enhance our ability to engineer sophisticated peptide-based systems with unprecedented precision and control. By overcoming the current limitations and leveraging their unique properties, oligopeptide self-assembly materials are set to revolutionize fields such as biomedicine, nanotechnology, and materials science, heralding new frontiers for innovation and discovery.

Acknowledgments

The authors thank the Singapore National Research Fellowship (NRF-NRFF11-2019-0004) and the Singapore Ministry of Education (MOE) Tier 2 Grant (MOE-T2EP30220-0006) for their support.

References

1. Wang, J.; Liu, K.; Xing, R.; Yan, X. Peptide Self-Assembly: Thermodynamics and Kinetics. *Chem. Soc. Rev.* **2016**, *45*, 5589–5604.
2. Levin, A.; Hakala, T. A.; Schnaider, L.; Bernardes, G. J. L.; Gazit, E.; Knowles, T. P. J. Biomimetic Peptide Self-Assembly for Functional Materials. *Nat. Rev. Chem.* **2020**, *4*, 615–634.
3. Sinha, N. J.; Langenstein, M. G.; Pochan, D. J.; Kloxin, C. J.; Saven, J. G. Peptide Design and Self-Assembly into Targeted Nanostructure and Functional Materials. *Chem. Rev.* **2021**, *121*, 13915–13935.
4. De Santis, E.; Ryadnov, M. G. Peptide Self-Assembly for Nanomaterials: The Old New Kid on the Block. *Chem. Soc. Rev.* **2015**, *44*, 8288–8300.
5. Song, Z.; Chen, X.; You, X.; Huang, K.; Dhinakar, A.; Gu, Z.; Wu, J. Self-Assembly of Peptide Amphiphiles for Drug Delivery: The Role of Peptide Primary and Secondary Structures. *Biomater. Sci.* **2017**, *5*, 2369–2380.
6. Hu, K.; Xiong, W.; Sun, C.; Wang, C.; Li, J.; Yin, F.; Jiang, Y.; Zhang, M.-R.; Li, Z.; Wang, X.; Li, Z. Self-Assembly of Constrained Cyclic Peptides Controlled by Ring Size. *CCS Chem.* **2020**, *2*, 42–51.
7. Xing, R.; Yuan, C.; Li, S.; Song, J.; Li, J.; Yan, X. Charge-Induced Secondary Structure Transformation of Amyloid-Derived Dipeptide Assemblies from β-Sheet to α-Helix. *Angew. Chem. Int. Ed.* **2018**, *57*, 1537–1542.
8. Zhang, S. Fabrication of Novel Biomaterials Through Molecular Self-Assembly. *Nat. Biotechnol.* **2003**, *21*, 1171–1178.
9. Zhao, X.; Pan, F.; Xu, H.; Yaseen, M.; Shan, H.; Hauser, C. A. E.; Zhang, S.; Lu, J. R. Molecular Self-Assembly and Applications of Designer Peptide Amphiphiles. *Chem. Soc. Rev.* **2010**, *39*, 3480–3498.
10. Black, M.; Trent, A.; Kostenko, Y.; Lee, J. S.; Olive, C.; Tirrell, M. Self-Assembled Peptide Amphiphile Micelles Containing a Cytotoxic T-Cell Epitope Promote a Protective Immune Response In Vivo. *Adv. Mater.* **2012**, *24*, 3845–3849.
11. Shimada, T.; Sakamoto, N.; Motokawa, R.; Koizumi, S.; Tirrell, M. Self-Assembly Process of Peptide Amphiphile Worm-like Micelles. *J. Phys. Chem. B* **2012**, *116*, 240–243.
12. Moore, A. N.; Hartgerink, J. D. Self-Assembling Multidomain Peptide Nanofibers for Delivery of Bioactive Molecules and Tissue Regeneration. *Acc. Chem. Res.* **2017**, *50*, 714–722.
13. Zhao, L.; Li, S.; Liu, Y.; Xing, R.; Yan, X. Kinetically Controlled Self-Assembly of Phthalocyanine–Peptide Conjugate Nanofibrils Enabling Superlarge Redshifted Absorption. *CCS Chem.* **2019**, *1*, 173–180.
14. Hamley, I. W. Peptide Nanotubes. *Angew. Chem. Int. Ed.* **2014**, *53*, 6866–6881.
15. Abbas, M.; Lipiński, W. P.; Wang, J.; Spruijt, E. Peptide-Based Coacervates as Biomimetic Protocells. *Chem. Soc. Rev.* **2021**, *50*, 3690–3705.
16. Li, Z.; Zhu, Y.; Matson, J. B. pH-Responsive Self-Assembling Peptide-Based Biomaterials: Designs and Applications. *ACS Appl. Bio Mater.* **2022**, *5*, 4635–4651.
17. Li, S.; Zhang, W.; Xue, H.; Xing, R.; Yan, X. Tumor Microenvironment-Oriented Adaptive Nanodrugs Based on Peptide Self-Assembly. *Chem. Sci.* **2020**, *11*, 8644–8656.
18. Li, T.; Lu, X.-M.; Zhang, M.-R.; Hu, K.; Li, Z. Peptide-Based Nanomaterials: Self-Assembly, Properties and Applications. *Bioact. Mater.* **2022**, *11*, 268–282.
19. Fichman, G.; Gazit, E. Self-Assembly of Short Peptides to Form Hydrogels: Design of Building Blocks, Physical Properties and Technological Applications. *Acta Biomater.* **2014**, *10*, 1671–1682.
20. Delfi, M.; Sartorius, R.; Ashrafizadeh, M.; Sharifi, E.; Zhang, Y.; De Berardinis, P.; Zarrabi, A.; Varma, R. S.; Tay, F. R.; Smith, B. R.; Makvandi, P. Self-Assembled Peptide and Protein Nanostructures for Anti-Cancer Therapy: Targeted Delivery, Stimuli-Responsive Devices and Immunotherapy. *Nano Today* **2021**, *38*, 101119.
21. Mart, R. J.; Osborne, R. D.; Stevens, M. M.; Ulijn, R. V. Peptide-Based Stimuli-Responsive Biomaterials. *Soft Matter* **2006**, *2*, 822–835.
22. Tang, J. D.; Mura, C.; Lampe, K. J. Stimuli-Responsive, Pentapeptide, Nanofiber Hydrogel for Tissue Engineering. *J. Am. Chem. Soc.* **2019**, *141*, 4886–4899.
23. Adamski, P.; Eleveld, M.; Sood, A.; Kun, Á.; Szilágyi, A.; Czárán, T.; Szathmáry, E.; Otto, S. From Self-Replication to Replicator Systems En Route to De Novo Life. *Nat. Rev. Chem.* **2020**, *4*, 386–403.
24. Grzybowski, B. A.; Huck, W. T. S. The Nanotechnology of Life-Inspired Systems. *Nat. Nanotechnol.* **2016**, *11*, 585–592.
25. Tantakitti, F.; Boekhoven, J.; Wang, X.; Kazantsev, R. V.; Yu, T.; Li, J.; Zhuang, E.; Zandi, R.; Ortony, J. H.; Newcomb, C. J.; Palmer, L. C.; Shekhawat, G. S.; de la Cruz, M. O.; Schatz, G. C.; Stupp, S. I. Energy Landscapes and Functions of Supramolecular Systems. *Nat. Mater.* **2016**, *15*, 469–476.

DOI: 10.31635/ccschem.023.202303209
Citation: *CCS Chem.* **2024**, 6, 69–90
Link to VoR: https://doi.org/10.31635/ccschem.023.202303209

26. Ben Messaoud, G.; Le Griel, P.; Hermida-Merino, D.; Roelants, S. L. K. W.; Soetaert, W.; Stevens, C. V.; Baccile, N. pH-Controlled Self-Assembled Fibrillar Network Hydrogels: Evidence of Kinetic Control of the Mechanical Properties. *Chem. Mater.* **2019**, *31*, 4817–4830.
27. Ozbas, B.; Kretsinger, J.; Rajagopal, K.; Schneider, J. P.; Pochan, D. J. Salt-Triggered Peptide Folding and Consequent Self-Assembly into Hydrogels with Tunable Modulus. *Macromolecules* **2004**, *37*, 7331–7337.
28. Ghosh, G.; Barman, R.; Mukherjee, A.; Ghosh, U.; Ghosh, S.; Fernández, G. Control over Multiple Nano- and Secondary Structures in Peptide Self-Assembly. *Angew. Chem. Int. Ed.* **2022**, *61*, e202113403.
29. Chen, X.; Würbser, M. A.; Boekhoven, J. Chemically Fueled Supramolecular Materials. *Acc. Mater. Res.* **2023**, *4*, 416–426.
30. Yuan, C.; Li, Q.; Xing, R.; Li, J.; Yan, X. Peptide Self-Assembly through Liquid-Liquid Phase Separation. *Chem* **2023**, *9*, 2425–2445.
31. Chen, J.; Zou, X. Self-Assemble Peptide Biomaterials and Their Biomedical Applications. *Bioact. Mater.* **2019**, *4*, 120–131.
32. Qi, G.-B.; Gao, Y.-J.; Wang, L.; Wang, H. Self-Assembled Peptide-Based Nanomaterials for Biomedical Imaging and Therapy. *Adv. Mater.* **2018**, *30*, 1703444.
33. Paramonov, S. E.; Jun, H.-W.; Hartgerink, J. D. Self-Assembly of Peptide–Amphiphile Nanofibers: The Roles of Hydrogen Bonding and Amphiphilic Packing. *J. Am. Chem. Soc.* **2006**, *128*, 7291–7298.
34. Cheng, R. P.; Gellman, S. H.; DeGrado, W. F. β-Peptides: From Structure to Function. *Chem. Rev.* **2001**, *101*, 3219–3232.
35. Hendricks, M. P.; Sato, K.; Palmer, L. C.; Stupp, S. I. Supramolecular Assembly of Peptide Amphiphiles. *Acc. Chem. Res.* **2017**, *50*, 2440–2448.
36. Knowles, T. P. J.; Mezzenga, R. Amyloid Fibrils as Building Blocks for Natural and Artificial Functional Materials. *Adv. Mater.* **2016**, *28*, 6546–6561.
37. Guo, Q.; Zou, G.; Qian, X.; Chen, S.; Gao, H.; Yu, J. Hydrogen-Bonds Mediate Liquid-Liquid Phase Separation of Mussel Derived Adhesive Peptides. *Nat. Commun.* **2022**, *13*, 5771.
38. Tao, K.; Makam, P.; Aizen, R.; Gazit, E. Self-Assembling Peptide Semiconductors. *Science* **2017**, *358*, eaam9756.
39. Knight, A. S.; Larsson, J.; Ren, J. M.; Bou Zerdan, R.; Seguin, S.; Vrahas, R.; Liu, J.; Ren, G.; Hawker, C. J. Control of Amphiphile Self-Assembly via Bioinspired Metal Ion Coordination. *J. Am. Chem. Soc.* **2018**, *140*, 1409–1414.
40. Moyer, T. J.; Finbloom, J. A.; Chen, F.; Toft, D. J.; Cryns, V. L.; Stupp, S. I. pH and Amphiphilic Structure Direct Supramolecular Behavior in Biofunctional Assemblies. *J. Am. Chem. Soc.* **2014**, *136*, 14746–14752.
41. Levin, A.; Mason, T. O.; Adler-Abramovich, L.; Buell, A. K.; Meisl, G.; Galvagnion, C.; Bram, Y.; Stratford, S. A.; Dobson, C. M.; Knowles, T. P. J.; Gazit, E. Ostwald's Rule of Stages Governs Structural Transitions and Morphology of Dipeptide Supramolecular Polymers. *Nat. Commun.* **2014**, *5*, 5219.
42. Brown, N.; Lei, J.; Zhan, C.; Shimon, L. J. W.; Adler-Abramovich, L.; Wei, G.; Gazit, E. Structural Polymorphism in a Self-Assembled Tri-Aromatic Peptide System. *ACS Nano* **2018**, *12*, 3253–3262.
43. Qiu, R.; Sasselli, I. R.; Álvarez, Z.; Sai, H.; Ji, W.; Palmer, L. C.; Stupp, S. I. Supramolecular Copolymers of Peptides and Lipidated Peptides and Their Therapeutic Potential. *J. Am. Chem. Soc.* **2022**, *144*, 5562–5574.
44. Zhu, H.; Wang, H.; Shi, B.; Shangguan, L.; Tong, W.; Yu, G.; Mao, Z.; Huang, F. Supramolecular Peptide Constructed by Molecular Lego Allowing Programmable Self-Assembly for Photodynamic Therapy. *Nat. Commun.* **2019**, *10*, 2412.
45. Booth, R.; Insua, I.; Ahmed, S.; Rioboo, A.; Montenegro, J. Supramolecular Fibrillation of Peptide Amphiphiles Induces Environmental Responses in Aqueous Droplets. *Nat. Commun.* **2021**, *12*, 6421.
46. Castelletto, V.; Barnes, R. H.; Karatzas, K.-A.; Edwards-Gayle, C. J. C.; Greco, F.; Hamley, I. W.; Rambo, R.; Seitsonen, J.; Ruokolainen, J. Arginine-Containing Surfactant-Like Peptides: Interaction with Lipid Membranes and Antimicrobial Activity. *Biomacromolecules* **2018**, *19*, 2782–2794.
47. Li, J.; Wang, J.; Zhao, Y.; Zhou, P.; Carter, J.; Li, Z.; Waigh, T. A.; Lu, J. R.; Xu, H. Surfactant-like Peptides: From Molecular Design to Controllable Self-Assembly with Applications. *Coord. Chem. Rev.* **2020**, *421*, 213418.
48. Cheng, D.-B.; Wang, D.; Gao, Y.-J.; Wang, L.; Qiao, Z.-Y.; Wang, H. Autocatalytic Morphology Transformation Platform for Targeted Drug Accumulation. *J. Am. Chem. Soc.* **2019**, *141*, 4406–4411.
49. Bal, S.; Ghosh, C.; Ghosh, T.; Vijayaraghavan, R. K.; Das, D. Non-Equilibrium Polymerization of Cross-β Amyloid Peptides for Temporal Control of Electronic Properties. *Angew. Chem. Int. Ed.* **2020**, *59*, 13506–13510.
50. Zhang, K.; Yang, P.-P.; He, P.-P.; Wen, S.-F.; Zou, X.-R.; Fan, Y.; Chen, Z.-M.; Cao, H.; Yang, Z.; Yue, K.; Zhang, X.; Zhang, H.; Wang, L.; Wang, H. Peptide-Based Nanoparticles Mimic Fibrillogenesis of Laminin in Tumor Vessels for Precise Embolization. *ACS Nano* **2020**, *14*, 7170–7180.
51. Goswami, S.; Reja, A.; Pal, S.; Singh, A.; Das, D. Nonequilibrium Amyloid Polymers Exploit Dynamic Covalent Linkage to Temporally Control Charge-Selective Catalysis. *J. Am. Chem. Soc.* **2022**, *144*, 19248–19252.
52. Li, M.; Ning, Y.; Chen, J.; Duan, X.; Song, N.; Ding, D.; Su, X.; Yu, Z. Proline Isomerization-Regulated Tumor Microenvironment-Adaptable Self-Assembly of Peptides for Enhanced Therapeutic Efficacy. *Nano Lett.* **2019**, *19*, 7965–7976.
53. Smith, A. M.; Williams, R. J.; Tang, C.; Coppo, P.; Collins, R. F.; Turner, M. L.; Saiani, A.; Ulijn, R. V. Fmoc-Diphenylalanine Self Assembles to a Hydrogel via a Novel Architecture Based on π–π Interlocked β-Sheets. *Adv. Mater.* **2008**, *20*, 37–41.
54. Ji, W.; Yuan, C.; Zilberzwige-Tal, S.; Xing, R.; Chakraborty, P.; Tao, K.; Gilead, S.; Yan, X.; Gazit, E. Metal-Ion Modulated Structural Transformation of Amyloid-like Dipeptide Supramolecular Self-Assembly. *ACS Nano* **2019**, *13*, 7300–7309.

DOI: 10.31635/ccschem.023.202303209
Citation: *CCS Chem.* **2024**, 6, 69–90
Link to VoR: https://doi.org/10.31635/ccschem.023.202303209

55. 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.
56. Yang, L.; Peltier, R.; Zhang, M.; Song, D.; Huang, H.; Chen, G.; Chen, Y.; Zhou, F.; Hao, Q.; Bian, L.; He, M.-L.; Wang, Z.; Hu, Y.; Sun, H. Desuccinylation-Triggered Peptide Self-Assembly: Live Cell Imaging of SIRT5 Activity and Mitochondrial Activity Modulation. *J. Am. Chem. Soc.* **2020**, *142*, 18150–18159.
57. Song, Y.; Li, M.; Song, N.; Liu, X.; Wu, G.; Zhou, H.; Long, J.; Shi, L.; Yu, Z. Self-Amplifying Assembly of Peptides in Macrophages for Enhanced Inflammatory Treatment. *J. Am. Chem. Soc.* **2022**, *144*, 6907–6917.
58. Liu, S.; Zhang, Q.; Shy, A. N.; Yi, M.; He, H.; Lu, S.; Xu, B. Enzymatically Forming Intranuclear Peptide Assemblies for Selectively Killing Human Induced Pluripotent Stem Cells. *J. Am. Chem. Soc.* **2021**, *143*, 15852–15862.
59. Zhou, P.; Hu, X.; Li, J.; Wang, Y.; Yu, H.; Chen, Z.; Wang, D.; Zhao, Y.; King, S. M.; Rogers, S. E.; Wang, J.; Lu, J. R.; Xu, H. Peptide Self-Assemblies from Unusual α-Sheet Conformations Based on Alternation of d/l Amino Acids. *J. Am. Chem. Soc.* **2022**, *144*, 21544–21554.
60. Gelain, F.; Luo, Z.; Zhang, S. Self-Assembling Peptide EAK16 and RADA16 Nanofiber Scaffold Hydrogel. *Chem. Rev.* **2020**, *120*, 13434–13460.
61. Reja, A.; Afrose, S. P.; Das, D. Aldolase Cascade Facilitated by Self-Assembled Nanotubes from Short Peptide Amphiphiles. *Angew. Chem. Int. Ed.* **2020**, *59*, 4329–4334.
62. Kumar Mahato, A.; Pal, S.; Dey, K.; Reja, A.; Paul, S.; Shelke, A.; Ajithkumar, T. G.; Das, D.; Banerjee, R. Covalent Organic Framework Cladding on Peptide-Amphiphile-Based Biomimetic Catalysts. *J. Am. Chem. Soc.* **2023**, *145*, 12793–12801.
63. Omosun, T. O.; Hsieh, M.-C.; Childers, W. S.; Das, D.; Mehta, A. K.; Anthony, N. R.; Pan, T.; Grover, M. A.; Berland, K. M.; Lynn, D. G. Catalytic Diversity in Self-Propagating Peptide Assemblies. *Nat. Chem.* **2017**, *9*, 805–809.
64. Sarkhel, B.; Chatterjee, A.; Das, D. Covalent Catalysis by Cross β Amyloid Nanotubes. *J. Am. Chem. Soc.* **2020**, *142*, 4098–4103.
65. Li, S.; Mehta, A. K.; Sidorov, A. N.; Orlando, T. M.; Jiang, Z.; Anthony, N. R.; Lynn, D. G. Design of Asymmetric Peptide Bilayer Membranes. *J. Am. Chem. Soc.* **2016**, *138*, 3579–3586.
66. Clover, T. M.; O'Neill, C. L.; Appavu, R.; Lokhande, G.; Gaharwar, A. K.; Posey, A. E.; White, M. A.; Rudra, J. S. Self-Assembly of Block Heterochiral Peptides into Helical Tapes. *J. Am. Chem. Soc.* **2020**, *142*, 19809–19813.
67. Wang, F.; Gnewou, O.; Wang, S.; Osinski, T.; Zuo, X.; Egelman, E. H.; Conticello, V. P. Deterministic Chaos in the Self-Assembly of β Sheet Nanotubes from an Amphipathic Oligopeptide. *Matter* **2021**, *4*, 3217–3231.
68. Castelletto, V.; Seitsonen, J.; Ruokolainen, J.; Hamley, I. W. Alpha Helical Surfactant-Like Peptides Self-Assemble into pH-Dependent Nanostructures. *Soft Matter* **2021**, *17*, 3096–3104.
69. Gao, C.; Kewalramani, S.; Valencia, D. M.; Li, H.; McCourt, J. M.; Olvera de la Cruz, M.; Bedzyk, M. J. Electrostatic Shape Control of a Charged Molecular Membrane from Ribbon to Scroll. *Proc. Natl. Acad. Sci.* **2019**, *116*, 22030–22036.
70. Zaldivar, G.; Vemulapalli, S.; Udumula, V.; Conda-Sheridan, M.; Tagliazucchi, M. Self-Assembled Nanostructures of Peptide Amphiphiles: Charge Regulation by Size Regulation. *J. Phys. Chem. C* **2019**, *123*, 17606–17615.
71. Lin, Y.; Penna, M.; Thomas, M. R.; Wojciechowski, J. P.; Leonardo, V.; Wang, Y.; Pashuck, E. T.; Yarovsky, I.; Stevens, M. M. Residue-Specific Solvation-Directed Thermodynamic and Kinetic Control over Peptide Self-Assembly with 1D/2D Structure Selection. *ACS Nano* **2019**, *13*, 1900–1909.
72. Abbas, M.; Lipiński, W. P.; Nakashima, K. K.; Huck, W. T. S.; Spruijt, E. A Short Peptide Synthon for Liquid–Liquid Phase Separation. *Nat. Chem.* **2021**, *13*, 1046–1054.
73. Yuan, C.; Levin, A.; Chen, W.; Xing, R.; Zou, Q.; Herling, T. W.; Challa, P. K.; Knowles, T. P. J.; Yan, X. Nucleation and Growth of Amino Acid and Peptide Supramolecular Polymers Through Liquid–Liquid Phase Separation. *Angew. Chem. Int. Ed.* **2019**, *58*, 18116–18123.
74. Chen, S.; Guo, Q.; Yu, J. Bio-Inspired Functional Coacervates. *Aggregate* **2022**, *3*, e293.
75. Israelachvili, J. N. *Intermolecular and Surface Forces*, 3rd ed.; Elsevier Academic Press Inc.: San Diego, CA, **2011**.
76. Lim, J.; Kumar, A.; Low, K.; Verma, C. S.; Mu, Y.; Miserez, A.; Pervushin, K. Liquid–Liquid Phase Separation of Short Histidine- and Tyrosine-Rich Peptides: Sequence Specificity and Molecular Topology. *J. Phys. Chem. B* **2021**, *125*, 6776–6790.
77. Sawada, T.; Fujita, M. Folding and Assembly of Metal-Linked Peptidic Nanostructures. *Chem* **2020**, *6*, 1861–1876.
78. Dong, J.; Liu, Y.; Cui, Y. Artificial Metal–Peptide Assemblies: Bioinspired Assembly of Peptides and Metals Through Space and across Length Scales. *J. Am. Chem. Soc.* **2021**, *143*, 17316–17336.
79. Yuan, C.; Ji, W.; Xing, R.; Li, J.; Gazit, E.; Yan, X. Hierarchically Oriented Organization in Supramolecular Peptide Crystals. *Nat. Rev. Chem.* **2019**, *3*, 567–588.
80. Prince, E.; Kumacheva, E. Design and Applications of Man-Made Biomimetic Fibrillar Hydrogels. *Nat. Rev. Mater.* **2019**, *4*, 99–115.
81. Lampel, A.; Ulijn, R. V.; Tuttle, T. Guiding Principles for Peptide Nanotechnology Through Directed Discovery. *Chem. Soc. Rev.* **2018**, *47*, 3737–3758.
82. Kumar, P.; Paterson, N. G.; Clayden, J.; Woolfson, D. N. De Novo Design of Discrete, Stable 310-Helix Peptide Assemblies. *Nature* **2022**, *607*, 387–392.
83. Conticello, V. P. Peptide-Based Nanomaterials: Building Back Better & Beyond. *Curr. Opin. Solid State Mater. Sci.* **2023**, *27*, 101066.
84. Wang, F.; Gnewou, O.; Modlin, C.; Beltran, L. C.; Xu, C.; Su, Z.; Juneja, P.; Grigoryan, G.; Egelman, E. H.; Conticello, V. P. Structural Analysis of Cross α-Helical Nanotubes Provides Insight into the Designability of Filamentous Peptide Nanomaterials. *Nat. Commun.* **2021**, *12*, 407.
85. Beesley, J. L.; Woolfson, D. N. The De Novo Design of α-Helical Peptides for Supramolecular Self-Assembly. *Curr. Opin. Biotechnol.* **2019**, *58*, 175–182.

DOI: 10.31635/ccschem.023.202303209
Citation: *CCS Chem.* **2024**, 6, 69–90
Link to VoR: https://doi.org/10.31635/ccschem.023.202303209

86. Rasines Mazo, A.; Allison-Logan, S.; Karimi, F.; Chan, N. J.-A.; Qiu, W.; Duan, W.; O'Brien-Simpson, N. M.; Qiao, G. G. Ring Opening Polymerization of α-Amino Acids: Advances in Synthesis, Architecture and Applications of Polypeptides and Their Hybrids. *Chem. Soc. Rev.* **2020**, *49*, 4737–4834.
87. Nandakumar, A.; Ito, Y.; Ueda, M. Solvent Effects on the Self-Assembly of an Amphiphilic Polypeptide Incorporating α-Helical Hydrophobic Blocks. *J. Am. Chem. Soc.* **2020**, *142*, 20994–21003.
88. Yuan, C.; Xing, R.; Cui, J.; Fan, W.; Li, J.; Yan, X. Multistep Desolvation as a Fundamental Principle Governing Peptide Self-Assembly Through Liquid–Liquid Phase Separation. *CCS Chem.* **2023**. doi:https://doi.org/10.31635/ccschem.023.202302990
89. Cakmak, F. P.; Choi, S.; Meyer, M. O.; Bevilacqua, P. C.; Keating, C. D. Prebiotically-Relevant Low Polyion Multivalency Can Improve Functionality of Membraneless Compartments. *Nat. Commun.* **2020**, *11*, 5949.
90. Perry, S. L.; Leon, L.; Hoffmann, K. Q.; Kade, M. J.; Priftis, D.; Black, K. A.; Wong, D.; Klein, R. A.; Pierce, C. F.; Margossian, K. O.; Whitmer, J. K.; Qin, J.; de Pablo, J. J.; Tirrell, M. Chirality-Selected Phase Behaviour in Ionic Polypeptide Complexes. *Nat. Commun.* **2015**, *6*, 6052.
91. Ling, S.; Kaplan, D. L.; Buehler, M. J. Nanofibrils in Nature and Materials Engineering. *Nat. Rev. Mater.* **2018**, *3*, 18016.
92. van der Lee, R.; Buljan, M.; Lang, B.; Weatheritt, R. J.; Daughdrill, G. W.; Dunker, A. K.; Fuxreiter, M.; Gough, J.; Gsponer, J.; Jones, D. T.; Kim, P. M.; Kriwacki, R. W.; Oldfield, C. J.; Pappu, R. V.; Tompa, P.; Uversky, V. N.; Wright, P. E.; Babu, M. M. Classification of Intrinsically Disordered Regions and Proteins. *Chem. Rev.* **2014**, *114*, 6589–6631.
93. Sheehan, F.; Sementa, D.; Jain, A.; Kumar, M.; Tayarani-Najjaran, M.; Kroiss, D.; Ulijn, R. V. Peptide-Based Supramolecular Systems Chemistry. *Chem. Rev.* **2021**, *121*, 13869–13914.
94. Zhou, P.; Xing, R.; Li, Q.; Li, J.; Yuan, C.; Yan, X. Steering Phase-Separated Droplets to Control Fibrillar Network Evolution of Supramolecular Peptide Hydrogels. *Matter* **2023**, *6*, 1945–1963.
95. Sato, K.; Ji, W.; Palmer, L. C.; Weber, B.; Barz, M.; Stupp, S. I. Programmable Assembly of Peptide Amphiphile via Noncovalent-to-Covalent Bond Conversion. *J. Am. Chem. Soc.* **2017**, *139*, 8995–9000.
96. Monreal Santiago, G.; Liu, K.; Browne, W. R.; Otto, S. Emergence of Light-Driven Protometabolism on Recruitment of a Photocatalytic Cofactor by a Self-Replicator. *Nat. Chem.* **2020**, *12*, 603–607.
97. Morrow, S. M.; Colomer, I.; Fletcher, S. P. A Chemically Fuelled Self-Replicator. *Nat. Commun.* **2019**, *10*, 1011.
98. Yamamoto, S.; Nishimura, K.; Morita, K.; Kanemitsu, S.; Nishida, Y.; Morimoto, T.; Aoi, T.; Tamura, A.; Maruyama, T. Microenvironment pH-Induced Selective Cell Death for Potential Cancer Therapy Using Nanofibrous Self-Assembly of a Peptide Amphiphile. *Biomacromolecules* **2021**, *22*, 2524–2531.
99. Wang, Y.; Su, H.; Wang, Y.; Cui, H. Discovery of Y-Shaped Supramolecular Polymers in a Self-Assembling Peptide Amphiphile System. *ACS Macro Lett.* **2022**, *11*, 1355–1361.
100. Cheng, D.-B.; Zhang, X.-H.; Gao, Y.-J.; Ji, L.; Hou, D.; Wang, Z.; Xu, W.; Qiao, Z.-Y.; Wang, H. Endogenous Reactive Oxygen Species-Triggered Morphology Transformation for Enhanced Cooperative Interaction with Mitochondria. *J. Am. Chem. Soc.* **2019**, *141*, 7235–7239.
101. Hu, B.; Lian, Z.; Zhou, Z.; Shi, L.; Yu, Z. Reactive Oxygen Species-Responsive Adaptable Self-Assembly of Peptides Toward Advanced Biomaterials. *ACS Appl. Bio Mater.* **2020**, *3*, 5529–5551.
102. Chagri, S.; Ng, D. Y. W.; Weil, T. Designing Bioresponsive Nanomaterials for Intracellular Self-Assembly. *Nat. Rev. Chem.* **2022**, *6*, 320–338.
103. Wang, M.; Zhang, Q.; Jian, H.; Liu, S.; Li, J.; Wang, A.; Dong, Q.; Ren, P.; Li, X.; Bai, S. Role of Thermolysin in Catalytic-Controlled Self-Assembly of Fmoc-Dipeptides. *CCS Chem.* **2020**, *2*, 317–328.
104. Sorrenti, A.; Leira-Iglesias, J.; Markvoort, A. J.; de Greef, T. F. A.; Hermans, T. M. Non-Equilibrium Supramolecular Polymerization. *Chem. Soc. Rev.* **2017**, *46*, 5476–5490.
105. Ragazzon, G.; Prins, L. J. Energy Consumption in Chemical Fuel-Driven Self-Assembly. *Nat. Nanotechnol.* **2018**, *13*, 882–889.
106. Kriebisch, B. A. K.; Jussupow, A.; Bergmann, A. M.; Kohler, F.; Dietz, H.; Kaila, V. R. I.; Boekhoven, J. Reciprocal Coupling in Chemically Fueled Assembly: A Reaction Cycle Regulates Self-Assembly and Vice Versa. *J. Am. Chem. Soc.* **2020**, *142*, 20837–20844.
107. Wang, Q.; Qi, Z.; Chen, M.; Qu, D.-H. Out-of-Equilibrium Supramolecular Self-Assembling Systems Driven by Chemical Fuel. *Aggregate* **2021**, *2*, e110.
108. Wilson, C. J.; Bommarius, A. S.; Champion, J. A.; Chernoff, Y. O.; Lynn, D. G.; Paravastu, A. K.; Liang, C.; Hsieh, M.-C.; Heemstra, J. M. Biomolecular Assemblies: Moving from Observation to Predictive Design. *Chem. Rev.* **2018**, *118*, 11519–11574.
109. Rieß, B.; Grötsch, R. K.; Boekhoven, J. The Design of Dissipative Molecular Assemblies Driven by Chemical Reaction Cycles. *Chem* **2020**, *6*, 552–578.
110. Shin, Y.; Brangwynne, C. P. Liquid Phase Condensation in Cell Physiology and Disease. *Science* **2017**, *357*, eaaf4382.
111. Jain, A.; Kassem, S.; Fisher, R. S.; Wang, B.; Li, T.-D.; Wang, T.; He, Y.; Elbaum-Garfinkle, S.; Ulijn, R. V. Connected Peptide Modules Enable Controlled Co-Existence of Self-Assembled Fibers Inside Liquid Condensates. *J. Am. Chem. Soc.* **2022**, *144*, 15002–15007.
112. Wei, G.; Su, Z.; Reynolds, N. P.; Arosio, P.; Hamley, I. W.; Gazit, E.; Mezzenga, R. Self-Assembling Peptide and Protein Amyloids: From Structure to Tailored Function in Nanotechnology. *Chem. Soc. Rev.* **2017**, *46*, 4661–4708.
113. Behranvand, N.; Nasri, F.; Zolfaghari Emameh, R.; Khani, P.; Hosseini, A.; Garssen, J.; Falak, R. Chemotherapy: A Double-Edged Sword in Cancer Treatment. *Cancer Immunol. Immunother.* **2022**, *71*, 507–526.
114. Pham, T. C.; Nguyen, V.-N.; Choi, Y.; Lee, S.; Yoon, J. Recent Strategies to Develop Innovative Photosensitizers for Enhanced Photodynamic Therapy. *Chem. Rev.* **2021**, *121*, 13454–13619.

DOI: 10.31635/ccschem.023.202303209
Citation: *CCS Chem.* **2024**, 6, 69–90
Link to VoR: https://doi.org/10.31635/ccschem.023.202303209

115. Fan, W.; Huang, P.; Chen, X. Overcoming the Achilles' Heel of Photodynamic Therapy. *Chem. Soc. Rev.* **2016**, *45*, 6488–6519.
116. Qi, J.; Ou, H.; Liu, Q.; Ding, D. Gathering Brings Strength: How Organic Aggregates Boost Disease Phototheranostics. *Aggregate* **2021**, *2*, 95–113.
117. Sangro, B.; Sarobe, P.; Hervás-Stubbs, S.; Melero, I. Advances in Immunotherapy for Hepatocellular Carcinoma. *Nat. Rev. Gastroenterol. Hepatol.* **2021**, *18*, 525–543.
118. Wang, H.; Yan, Y.-Q.; Yi, Y.; Wei, Z.-Y.; Chen, H.; Xu, J.-F.; Wang, H.; Zhao, Y.; Zhang, X. Supramolecular Peptide Therapeutics: Host–Guest Interaction-Assisted Systemic Delivery of Anticancer Peptides. *CCS Chem.* **2020**, *2*, 739–748.
119. Chabner, B. A.; Roberts, T. G. Chemotherapy and the War on Cancer. *Nat. Rev. Cancer* **2005**, *5*, 65–72.
120. DeVita, V. T., Jr.; Chu, E. A History of Cancer Chemotherapy. *Cancer Res.* **2008**, *68*, 8643–8653.
121. Cao, M.; Lu, S.; Wang, N.; Xu, H.; Cox, H.; Li, R.; Waigh, T.; Han, Y.; Wang, Y.; Lu, J. R. Enzyme-Triggered Morphological Transition of Peptide Nanostructures for Tumor-Targeted Drug Delivery and Enhanced Cancer Therapy. *ACS Appl. Mater. Interfaces* **2019**, *11*, 16357–16366.
122. Wang, Z.; An, H.-W.; Hou, D.; Wang, M.; Zeng, X.; Zheng, R.; Wang, L.; Wang, K.; Wang, H.; Xu, W. Addressable Peptide Self-Assembly on the Cancer Cell Membrane for Sensitizing Chemotherapy of Renal Cell Carcinoma. *Adv. Mater.* **2019**, *31*, 1807175.
123. He, W.; Yan, J.; Jiang, W.; Li, S.; Qu, Y.; Niu, F.; Yan, Y.; Sui, F.; Wang, S.; Zhou, Y.; Jin, L.; Li, Y.; Ji, M.; Ma, P. X.; Liu, M.; Lu, W.; Hou, P. Peptide-Induced Self-Assembly of Therapeutics into a Well-Defined Nanoshell with Tumor-Triggered Shape and Charge Switch. *Chem. Mater.* **2018**, *30*, 7034–7046.
124. Sun, B.; Chang, R.; Cao, S.; Yuan, C.; Zhao, L.; Yang, H.; Li, J.; Yan, X.; van Hest, J. C. M. Acid-Activatable Transmorphic Peptide-Based Nanomaterials for Photodynamic Therapy. *Angew. Chem. Int. Ed.* **2020**, *59*, 20582–20588.
125. Xie, L.; Ding, Y.; Zhang, X.; Zhang, Z.; Zeng, S.; Wang, L.; Yang, Z.; Liu, Q.; Hu, Z.-W. A Cascade-Targeted Enzyme-Instructed Peptide Self-Assembly Strategy for Cancer Immunotherapy Through Boosting Immunogenic Cell Death. *Small Methods* **2023**, *7*, 2201416.
126. Li, Y.; Zhang, M.; Han, H.; Zhang, B.; Matson, J. B.; Chen, D.; Li, W.; Wang, Y. Peptide-Based Supramolecular Photodynamic Therapy Systems: From Rational Molecular Design to Effective Cancer Treatment. *Chem. Eng. J.* **2022**, *436*, 135240.
127. Cheng, Z.; Cheng, Y.; Chen, Q.; Li, M.; Wang, J.; Liu, H.; Li, M.; Ning, Y.; Yu, Z.; Wang, Y.; Wang, H. Self-Assembly of Pentapeptides into Morphology-Adaptable Nanomedicines for Enhanced Combinatorial Chemo-Photodynamic Therapy. *Nano Today* **2020**, *33*, 100878.
128. Luan, X.; Kong, H.; He, P.; Yang, G.; Zhu, D.; Guo, L.; Wei, G. Self-Assembled Peptide-Based Nanodrugs: Molecular Design, Synthesis, Functionalization, and Targeted Tumor Bioimaging and Biotherapy. *Small* **2023**, *19*, 2205787.
129. Liu, K.; Xing, R.; Zou, Q.; Ma, G.; Möhwald, H.; Yan, X. Simple Peptide-Tuned Self-Assembly of Photosensitizers Towards Anticancer Photodynamic Therapy. *Angew. Chem. Int. Ed.* **2016**, *55*, 3036–3039.
130. Li, J.; Wang, A.; Ren, P.; Yan, X.; Bai, S. One-Step Co-Assembly Method to Fabricate Photosensitive Peptide Nanoparticles for Two-Photon Photodynamic Therapy. *Chem. Commun.* **2019**, *55*, 3191–3194.
131. Ge, L.; Qiao, C.; Tang, Y.; Zhang, X.; Jiang, X. Light-Activated Hypoxia-Sensitive Covalent Organic Framework for Tandem-Responsive Drug Delivery. *Nano Lett.* **2021**, *21*, 3218–3224.
132. Ji, B.; Wei, M.; Yang, B. Recent Advances in Nanomedicines for Photodynamic Therapy (PDT)-Driven Cancer Immunotherapy. *Theranostics* **2022**, *12*, 434–458.
133. Chang, R.; Yan, X. Supramolecular Immunotherapy of Cancer Based on the Self-Assembling Peptide Design. *Small Struct.* **2020**, *1*, 2000068.
134. Froimchuk, E.; Carey, S. T.; Edwards, C.; Jewell, C. M. Self-Assembly as a Molecular Strategy to Improve Immunotherapy. *Acc. Chem. Res.* **2020**, *53*, 2534–2545.
135. Wang, M.-D.; Lv, G.-T.; An, H.-W.; Zhang, N.-Y.; Wang, H. In Situ Self-Assembly of Bispecific Peptide for Cancer Immunotherapy. *Angew. Chem. Int. Ed.* **2022**, *61*, e202113649.
136. Huang, Z.; Song, W.; Chen, X. Supramolecular Self-Assembled Nanostructures for Cancer Immunotherapy. *Front. Chem.* **2020**, *8*, 380.
137. Zhou, J.; Xu, B. Enzyme-Instructed Self-Assembly: A Multistep Process for Potential Cancer Therapy. *Bioconjug. Chem.* **2015**, *26*, 987–999.
138. Gao, J.; Zhan, J.; Yang, Z. Enzyme-Instructed Self-Assembly (EISA) and Hydrogelation of Peptides. *Adv. Mater.* **2020**, *32*, 1805798.
139. Kim, B. J.; Xu, B. Enzyme-Instructed Self-Assembly for Cancer Therapy and Imaging. *Bioconjug. Chem.* **2020**, *31*, 492–500.
140. Pires, R. A.; Abul-Haija, Y. M.; Costa, D. S.; Novoa-Carballal, R.; Reis, R. L.; Ulijn, R. V.; Pashkuleva, I. Controlling Cancer Cell Fate Using Localized Biocatalytic Self-Assembly of an Aromatic Carbohydrate Amphiphile. *J. Am. Chem. Soc.* **2015**, *137*, 576–579.
141. Liu, S.; Zhang, Q.; He, H.; Yi, M.; Tan, W.; Guo, J.; Xu, B. Intranuclear Nanoribbons for Selective Killing of Osteosarcoma Cells. *Angew. Chem. Int. Ed.* **2022**, *61*, e202210568.
142. Guo, Z.; Park, S.; Yoon, J.; Shin, I. Recent Progress in the Development of Near-Infrared Fluorescent Probes for Bioimaging Applications. *Chem. Soc. Rev.* **2014**, *43*, 16–29.
143. Zhou, S.; Jiang, L.; Li, C.; Mao, H.; Jiang, C.; Wang, Z.; Zheng, X.; Jiang, X. Acid and Hypoxia Tandem-Activatable Deep Near-Infrared Nanoprobe for Two-Step Signal Amplification and Early Detection of Cancer. *Adv. Mater.* **2023**, *35*, 2212231.
144. Zheng, X.; Wang, X.; Mao, H.; Wu, W.; Liu, B.; Jiang, X. Hypoxia-Specific Ultrasensitive Detection of Tumours and Cancer Cells In Vivo. *Nat. Commun.* **2015**, *6*, 5834.
145. Tan, W.; Zhang, Q.; Quiñones-Frías, M. C.; Hsu, A. Y.; Zhang, Y.; Rodal, A.; Hong, P.; Luo, H. R.; Xu, B. Enzyme-Responsive Peptide Thioesters for Targeting Golgi Apparatus. *J. Am. Chem. Soc.* **2022**, *144*, 6709–6713.

DOI: 10.31635/ccschem.023.202303209
Citation: *CCS Chem.* **2024**, 6, 69–90
Link to VoR: https://doi.org/10.31635/ccschem.023.202303209

146. Weiskopf, N.; Edwards, L. J.; Helms, G.; Mohammadi, S.; Kirilina, E. Quantitative Magnetic Resonance Imaging of Brain Anatomy and In Vivo Histology. *Nat. Rev. Phys.* **2021**, *3*, 570–588.
147. Zhang, X.-H.; Cheng, D.-B.; Ji, L.; An, H.-W.; Wang, D.; Yang, Z.-X.; Chen, H.; Qiao, Z.-Y.; Wang, H. Photothermal-Promoted Morphology Transformation in Vivo Monitored by Photoacoustic Imaging. *Nano Lett.* **2020**, *20*, 1286–1295.
148. Choi, W.; Park, B.; Choi, S.; Oh, D.; Kim, J.; Kim, C. Recent Advances in Contrast-Enhanced Photoacoustic Imaging: Overcoming the Physical and Practical Challenges. *Chem. Rev.* **2023**, *123*, 7379–7419.
149. Jiang, Q.; Liu, X.; Liang, G.; Sun, X. Self-Assembly of Peptide Nanofibers for Imaging Applications. *Nanoscale* **2021**, *13*, 15142–15150.
150. Deng, Y.; Zhan, W.; Liang, G. Intracellular Self-Assembly of Peptide Conjugates for Tumor Imaging and Therapy. *Adv. Healthc. Mater.* **2021**, *10*, 2001211.
151. Song, B.-L.; Zhang, X.-H.; Qiao, Z.-Y.; Wang, H. Peptide-Based AIEgens: From Molecular Design, Stimuli Responsiveness to Biomedical Application. *CCS Chem.* **2022**, *4*, 437–455.
152. Qin, W.; Xu, C.; Zhao, Y.; Yu, C.; Shen, S.; Li, L.; Huang, W. Recent Progress in Small Molecule Fluorescent Probes for Nitroreductase. *Chinese Chem. Lett.* **2018**, *29*, 1451–1455.
153. Hu, B.; Song, N.; Cao, Y.; Li, M.; Liu, X.; Zhou, Z.; Shi, L.; Yu, Z. Noncanonical Amino Acids for Hypoxia-Responsive Peptide Self-Assembly and Fluorescence. *J. Am. Chem. Soc.* **2021**, *143*, 13854–13864.
154. Li, L.; Gu, P.; Hao, M.; Xiang, X.; Feng, Y.; Zhu, X.; Song, Y.; Song, E. Bacteria-Targeted MRI Probe-Based Imaging Bacterial Infection and Monitoring Antimicrobial Therapy In Vivo. *Small* **2021**, *17*, 2103627.
155. Amdursky, N.; Molotskii, M.; Aronov, D.; Adler-Abramovich, L.; Gazit, E.; Rosenman, G. Blue Luminescence Based on Quantum Confinement at Peptide Nanotubes. *Nano Lett.* **2009**, *9*, 3111–3115.
156. Tao, K.; Fan, Z.; Sun, L.; Makam, P.; Tian, Z.; Ruegsegger, M.; Shaham-Niv, S.; Hansford, D.; Aizen, R.; Pan, Z.; Galster, S.; Ma, J.; Yuan, F.; Si, M.; Qu, S.; Zhang, M.; Gazit, E.; Li, J. Quantum Confined Peptide Assemblies with Tunable Visible to Near-Infrared Spectral Range. *Nat. Commun.* **2018**, *9*, 3217.
157. Fan, Z.; Sun, L.; Huang, Y.; Wang, Y.; Zhang, M. Bioinspired Fluorescent Dipeptide Nanoparticles for Targeted Cancer Cell Imaging and Real-Time Monitoring of Drug Release. *Nat. Nanotechnol.* **2016**, *11*, 388–394.
158. Zhou, Y.; Li, Q.; Wu, Y.; Li, X.; Zhou, Y.; Wang, Z.; Liang, H.; Ding, F.; Hong, S.; Steinmetz, N. F.; Cai, H. Molecularly Stimuli-Responsive Self-Assembled Peptide Nanoparticles for Targeted Imaging and Therapy. *ACS Nano* **2023**, *17*, 8004–8025.
159. Li, S.; Zou, Q.; Xing, R.; Govindaraju, T.; Fakhrullin, R.; Yan, X. Peptide-Modulated Self-Assembly as a Versatile Strategy for Tumor Supramolecular Nanotheranostics. *Theranostics* **2019**, *9*, 3249–3261.
160. Ren, C.; Wang, Z.; Wang, Q.; Yang, C.; Liu, J. Self-Assembled Peptide-Based Nanoprobes for Disease Theranostics and Disease-Related Molecular Imaging. *Small Methods* **2020**, *4*, 1900403.
161. Liu, Y.; Miao, Q.; Zou, P.; Liu, L.; Wang, X.; An, L.; Zhang, X.; Qian, X.; Luo, S.; Liang, G. Enzyme-Controlled Intracellular Self-Assembly of ^{18}F Nanoparticles for Enhanced MicroPET Imaging of Tumor. *Theranostics* **2015**, *5*, 1058–1067.
162. Loo, Y.; Goktas, M.; Tekinay, A. B.; Guler, M. O.; Hauser, C. A. E.; Mitraki, A. Self-Assembled Proteins and Peptides as Scaffolds for Tissue Regeneration. *Adv. Healthc. Mater.* **2015**, *4*, 2557–2586.
163. Hao, Z.; Li, H.; Wang, Y.; Hu, Y.; Chen, T.; Zhang, S.; Guo, X.; Cai, L.; Li, J. Supramolecular Peptide Nanofiber Hydrogels for Bone Tissue Engineering: From Multihierarchical Fabrications to Comprehensive Applications. *Adv. Sci.* **2022**, *9*, 2103820.
164. Zhu, M.; Zhong, W.; Cao, W.; Zhang, Q.; Wu, G. Chondroinductive/Chondroconductive Peptides and Their-Functionalized Biomaterials for Cartilage Tissue Engineering. *Bioact. Mater.* **2022**, *9*, 221–238.
165. He, P.-P.; Li, X.-D.; Fan, J.-Q.; Fan, Y.; Yang, P.-P.; Li, B.-N.; Cong, Y.; Yang, C.; Zhang, K.; Wang, Z.-Q.; Hou, D.-Y.; Wang, H.; Wang, L. Live Cells Process Exogenous Peptide as Fibronectin Fibrillogenesis In Vivo. *CCS Chem.* **2020**, *2*, 539–554.
166. Cheng, T.-Y.; Chen, M.-H.; Chang, W.-H.; Huang, M.-Y.; Wang, T.-W. Neural Stem Cells Encapsulated in a Functionalized Self-Assembling Peptide Hydrogel for Brain Tissue Engineering. *Biomaterials* **2013**, *34*, 2005–2016.
167. Lopez-Silva, T. L.; Cristobal, C. D.; Edwin Lai, C. S.; Leyva-Aranda, V.; Lee, H. K.; Hartgerink, J. D. Self-Assembling Multidomain Peptide Hydrogels Accelerate Peripheral Nerve Regeneration After Crush Injury. *Biomaterials* **2021**, *265*, 120401.
168. Rao, F.; Wang, Y.; Zhang, D.; Lu, C.; Cao, Z.; Sui, J.; Wu, M.; Zhang, Y.; Pi, W.; Wang, B.; Kou, Y.; Wang, X.; Zhang, P.; Jiang, B. Aligned Chitosan Nanofiber Hydrogel Grafted with Peptides Mimicking Bioactive Brain-Derived Neurotrophic Factor and Vascular Endothelial Growth Factor Repair Long-Distance Sciatic Nerve Defects in Rats. *Theranostics* **2020**, *10*, 1590–1603.
169. Lu, J.; Yan, X.; Sun, X.; Shen, X.; Yin, H.; Wang, C.; Liu, Y.; Lu, C.; Fu, H.; Yang, S.; Wang, Y.; Sun, X.; Zhao, L.; Lu, S.; Mikos, A. G.; Peng, J.; Wang, X. Synergistic Effects of Dual-Presenting VEGF- and BDNF-Mimetic Peptide Epitopes from Self-Assembling Peptide Hydrogels on Peripheral Nerve Regeneration. *Nanoscale* **2019**, *11*, 19943–19958.
170. Peressotti, S.; Koehl, G. E.; Goding, J. A.; Green, R. A. Self-Assembling Hydrogel Structures for Neural Tissue Repair. *ACS Biomater. Sci. Eng.* **2021**, *7*, 4136–4163.
171. Tian, X.; Sun, F.; Zhou, X.-R.; Luo, S.-Z.; Chen, L. Role of Peptide Self-Assembly in Antimicrobial Peptides. *J. Pept. Sci.* **2015**, *21*, 530–539.
172. Morgan, F. L. C.; Moroni, L.; Baker, M. B. Dynamic Bioinks to Advance Bioprinting. *Adv. Healthc. Mater.* **2020**, *9*, 1901798.
173. Lombardi, L.; Shi, Y.; Falanga, A.; Galdiero, E.; de Alteriis, E.; Franci, G.; Chourpa, I.; Azevedo, H. S.; Galdiero, S. Enhancing the Potency of Antimicrobial Peptides Through Molecular Engineering and Self-Assembly. *Biomacromolecules* **2019**, *20*, 1362–1374.

DOI: 10.31635/ccschem.023.202303209
Citation: *CCS Chem.* **2024**, 6, 69–90
Link to VoR: https://doi.org/10.31635/ccschem.023.202303209

174. Xie, X.; Gao, B.; Ma, Z.; Liu, J.; Zhang, J.; Liang, J.; Chen, Z.; Wu, L.; Li, W. Host–Guest Interaction Driven Peptide Assembly into Photoresponsive Two-Dimensional Nanosheets with Switchable Antibacterial Activity. *CCS Chem.* **2021**, *3*, 1949–1962.
175. Teng, R.; Yang, Y.; Zhang, Z.; Yang, K.; Sun, M.; Li, C.; Fan, Z.; Du, J. In Situ Enzyme-Induced Self-Assembly of Antimicrobial-Antioxidative Peptides to Promote Wound Healing. *Adv. Funct. Mater.* **2023**, *33*, 2214454.
176. Susapto, H. H.; Alhattab, D.; Abdelrahman, S.; Khan, Z.; Alshehri, S.; Kahin, K.; Ge, R.; Moretti, M.; Emwas, A.-H.; Hauser, C. A. E. Ultrashort Peptide Bioinks Support Automated Printing of Large-Scale Constructs Assuring Long-Term Survival of Printed Tissue Constructs. *Nano Lett.* **2021**, *21*, 2719–2729.
177. Murphy, S. V.; Atala, A. 3D Bioprinting of Tissues and Organs. *Nat. Biotechnol.* **2014**, *32*, 773–785.
178. Hedegaard, C. L.; Collin, E. C.; Redondo-Gómez, C.; Nguyen, L. T. H.; Ng, K. W.; Castrejón-Pita, A. A.; Castrejón-Pita, J. R.; Mata, A. Hydrodynamically Guided Hierarchical Self-Assembly of Peptide–Protein Bioinks. *Adv. Funct. Mater.* **2018**, *28*, 1703716.
179. Jian, H.; Wang, M.; Dong, Q.; Li, J.; Wang, A.; Li, X.; Ren, P.; Bai, S. Dipeptide Self-Assembled Hydrogels with Tunable Mechanical Properties and Degradability for 3D Bioprinting. *ACS Appl. Mater. Interfaces* **2019**, *11*, 46419–46426.
180. Cofiño, C.; Perez-Amodio, S.; Semino, C. E.; Engel, E.; Mateos-Timoneda, M. A. Development of a Self-Assembled Peptide/Methylcellulose-Based Bioink for 3D Bioprinting. *Macromol. Mater. Eng.* **2019**, *304*, 1900353.
181. Raphael, B.; Khalil, T.; Workman, V. L.; Smith, A.; Brown, C. P.; Streuli, C.; Saiani, A.; Domingos, M. 3D Cell Bioprinting of Self-Assembling Peptide-Based Hydrogels. *Mater. Lett.* **2017**, *190*, 103–106.
182. Lee, S. J.; Lee, D.; Yoon, T. R.; Kim, H. K.; Jo, H. H.; Park, J. S.; Lee, J. H.; Kim, W. D.; Kwon, I. K.; Park, S. A. Surface Modification of 3D-Printed Porous Scaffolds via Mussel-Inspired Polydopamine and Effective Immobilization of rhBMP-2 to Promote Osteogenic Differentiation for Bone Tissue Engineering. *Acta Biomater.* **2016**, *40*, 182–191.
183. Hajfathalian, M.; Ghelichi, S.; García-Moreno, P. J.; Moltke Sørensen, A.-D.; Jacobsen, C. Peptides: Production, Bioactivity, Functionality, and Applications. *Crit. Rev. Food Sci. Nutr.* **2018**, *58*, 3097–3129.
184. Shi, M.; McHugh, K. J. Strategies for Overcoming Protein and Peptide Instability in Biodegradable Drug Delivery Systems. *Adv. Drug Deliv. Rev.* **2023**, *199*, 114904.
185. Solano, C. M.; Wen, Y.; Han, H.; Collier, J. H. Practical Considerations in the Design and Use of Immunologically Active Fibrillar Peptide Assemblies. In *Peptide Self-Assembly: Methods and Protocols*;Nilsson, B. L., Doran, T. M., Eds.; Springer: New York, **2018**; pp 233–248.
186. Xu, T.; Wang, J.; Zhao, S.; Chen, D.; Zhang, H.; Fang, Y.; Kong, N.; Zhou, Z.; Li, W.; Wang, H. Accelerating the Prediction and Discovery of Peptide Hydrogels with Human-in-the-Loop. *Nat. Commun.* **2023**, *14*, 3880.
187. 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.
188. Cox, H.; Cao, M.; Xu, H.; Waigh, T. A.; Lu, J. R. Active Modulation of States of Prestress in Self-Assembled Short Peptide Gels. *Biomacromolecules* **2019**, *20*, 1719–1730.
189. Ferguson, A. L.; Tovar, J. D. Evolution of π-Peptide Self-Assembly: From Understanding to Prediction and Control. *Langmuir* **2022**, *38*, 15463–15475.
190. Li, F.; Han, J.; Cao, T.; Lam, W.; Fan, B.; Tang, W.; Chen, S.; Fok, K. L.; Li, L. Design of Self-Assembly Dipeptide Hydrogels and Machine Learning via Their Chemical Features. *Proc. Natl. Acad. Sci.* **2019**, *116*, 11259–11264.
191. Batra, R.; Loeffler, T. D.; Chan, H.; Srinivasan, S.; Cui, H.; Korendovych, I. V.; Nanda, V.; Palmer, L. C.; Solomon, L. A.; Fry, H. C.; Sankaranarayanan, S. K. R. S. Machine Learning Overcomes Human Bias in the Discovery of Self-Assembling Peptides. *Nat. Chem.* **2022**, *14*, 1427–1435.
192. Ramakrishnan, M.; van Teijlingen, A.; Tuttle, T.; Ulijn, R. V. Integrating Computation, Experiment, and Machine Learning in the Design of Peptide-Based Supramolecular Materials and Systems. *Angew. Chem. Int. Ed.* **2023**, *62*, e202218067.

DOI: 10.31635/ccschem.023.202303209
Citation: *CCS Chem.* **2024**, 6, 69–90
Link to VoR: https://doi.org/10.31635/ccschem.023.202303209