P2.212 - Designing self-assembling lipopeptide for tissue regeneration

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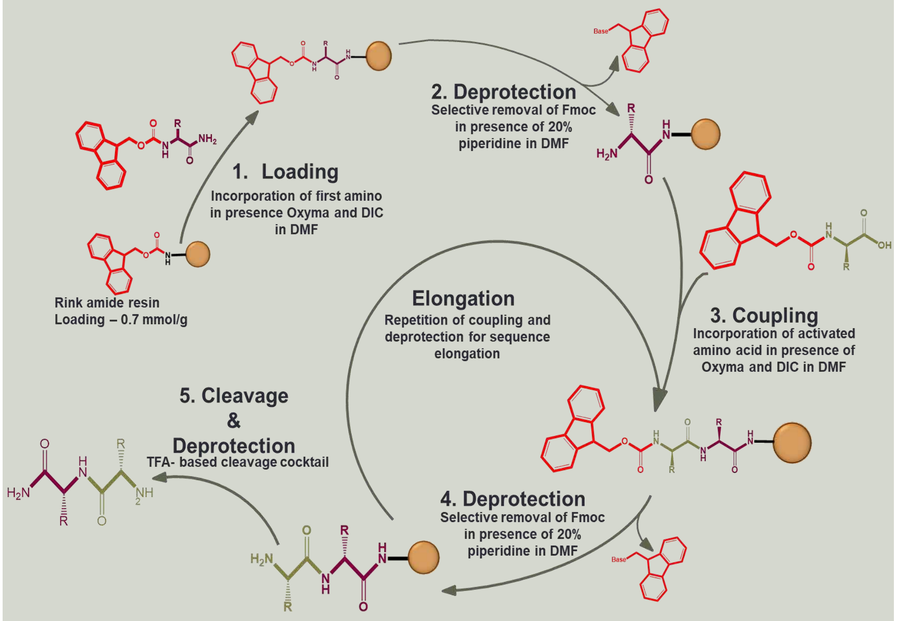
1. INTRODUCTION

Lipopetides are one class of biosurfactant materials, which have been widely studied in the past decade for tissue engineering applications. Their biocompatibility and flexibility make them ideal scaffold materials for tissue regeneration[1]. The hydrophobic lipid tail propels the self assembly and exposes the functional peptide sequences on the surface of the supramolecular structure. In our work, we've combined stearic acid with bioactive peptides which can imitate the structure and function of native ECM[2]. Here, the self-assembly propensity of these compounds are reported.

Bioactive peptides	Biological activity
IKVAV	Encourages vascularization and differentiation factors
REDV	Promoted endothelial cell adhesion and proliferation
GFOGER	Promotes cell adhesion, proliferation and differentiation of stem cells (hMSCs) (present in collagen type 1)

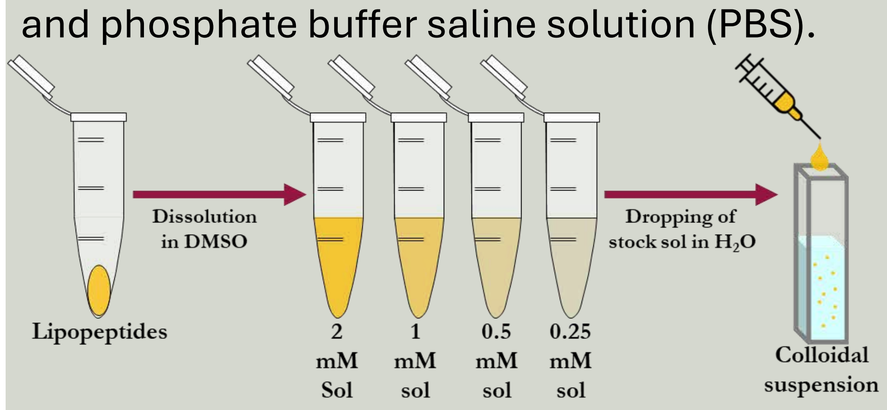
2. Methodology

The peptide sequences were *synthesized* using microwave assisted solid phase peptide synthesis technique on CEM liberty blue instrument.



The peptides were then purified by RP-HPLC and freeze dried.

The *self-assembly studies* were done using solvent displacement technique in pure water and phosphate buffer saline solution (PBS).



3. Results

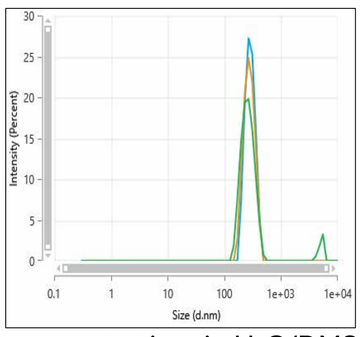
DLS results of colloidal suspensions of the lipopeptides in pure water at different peptide concentrations and solvent ratios are reported below

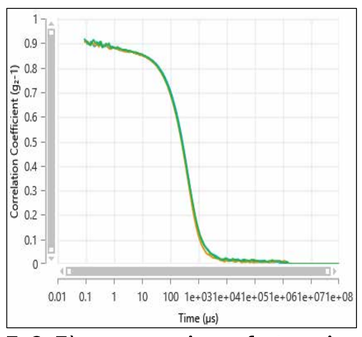
Peptide sequence	pH		9:1 (50 µM)	9.5:0.5 (50 µM)	9:1 (25 µM)	9.5:0.5 (25 µM)
Stearic acid-IKVAV-NH_2	6.93	PDI Size (nm)	0.4443±0.04 191±18.9	0.6717±0.21 870.1±147	0.7981±0.17 782.3±500	0.6277±0.62 383.2±382
Stearic acid-REDV-NH_2	7.29	PDI Size (nm)	0.2698±0.02 291±10.02	0.319±0.048 450.4±8.4	0.4018±0.03 497.1±12.4	0.3431±0.021 659.3±18
Stearic acid-GFOGER-NH_2	7.19	PDI Size (nm)	0.2625±0.02 1116±35.4	0.3017±0.06 1418±62.9	0.2661±0.008 414.2±12.4	0.1692±0.015 250±0.88

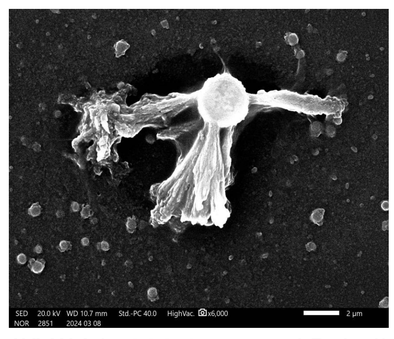
PDI- Polydispersity index

Below the DLS graphs and SEM images of the lipopeptides in H_2O/DMSO (9.5:0.5) are reported (for 25 µM concentration)

Stearic acid-IKVAV-NH_2

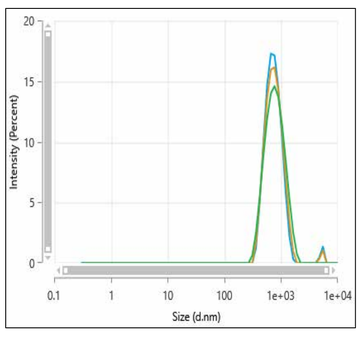


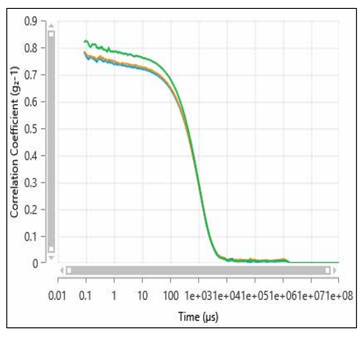


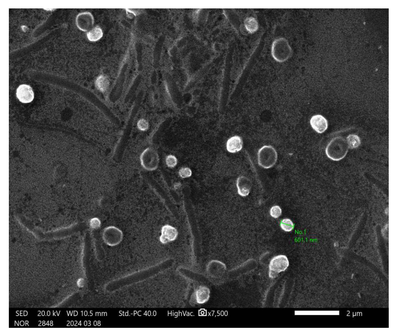


For 25 µM concentration, in H_2O/DMSO(9.5:0.5), mean size of stearic acid-IKVAV is 383.2 nm and Polydispersity Index is 0.6277

Stearic acid-REDV-NH_2

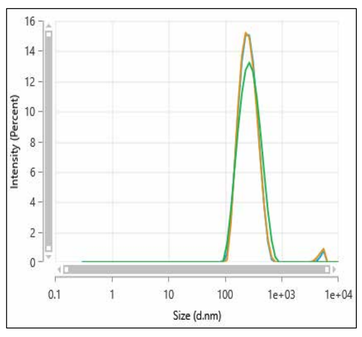


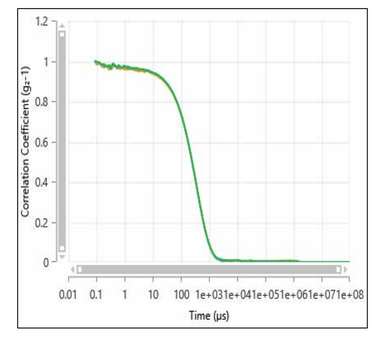


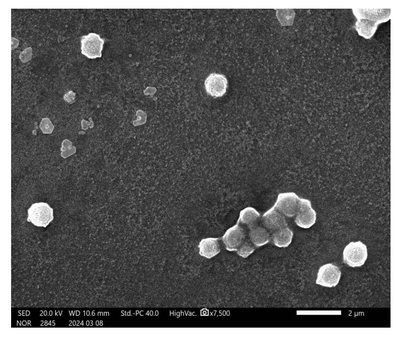


For 25 µM concentration, in H_2O/DMSO(9.5:0.5), mean size of stearic acid-REDV is 659.3 nm and Polydispersity Index is 0.3431

Stearic acid-GFOGER-NH_2







For 25 µM concentration, in H_2O/DMSO(9.5:0.5), mean size of stearic acid-GFOGER is 250 nm and Polydispersity Index is 0.1692

4. Conclusion

Our results showed that all lipopeptides self-assemble in aqueous environment. The best results were obtained with the higher percentage of water (95%) at the lowest concentration (25 µM). In PBS, an increase in the dimension was observed, probably due to the higher pH and salt concentration.

5. Future perspectives

The lipopeptides will be combined with low temperature gelling biopolymers like agarose to develop composite hydrogels which will then be used for 3D printing. The obtained scaffolds will be tested for their rheological properties and then seeded with L929 cells to evaluate their cytotoxicity and cell viability.

References

(1) Hutchinson, J. A.; Burholt, S.; Hamley, I. W. Peptide Hormones and Lipopeptides: From Self-Assembly to Therapeutic Applications. Journal of Peptide Science 2017, 23 (2), 82–94. https://doi.org/10.1002/psc.2954.

(2) Sun, W.; Gregory, D. A.; Zhao, X. Designed Peptide Amphiphiles as Scaffolds for Tissue Engineering. Advances in Colloid and Interface Science 2023, 314, 102866. https://doi.org/10.1016/j.cis.2023.102866.

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