Preparation of Hemoglobin-Loaded Polymer Micelles as Artificial Oxygen Carrier

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Introduction

The clinical application of cell-free Hb as artificial oxygen carrier is severely limited by the dissociation which causes serious renal toxicity, short circulation halftime and entrapped by NO to induce vasoconstriction and hypertension[1].

Great progress has been achieved for amphiphilic copolymers in physical encapsulation or chemical conjugation of hemoglobin (Fig.1)[2].They hint three advantages of self-assembly technique. 1)To prolong the circulation time. 2)To facilitate the inclusion of enzyme systems. 3)To avoid direct exposure to plasma[3].

Here, we report the amphiphilic copolymer micelles with high efficiency in encapsulating Hb, which have potential to be applied as an artificial oxygen carrier.

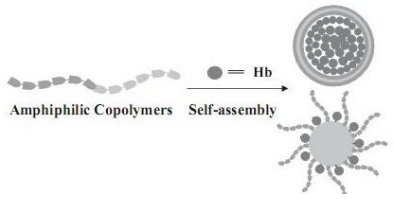
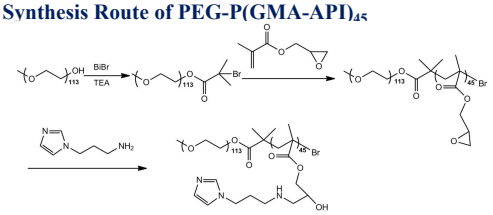


Fig. 1. Amphiphilic copolymers in physical encapsulation or chemical conjugation of Hb

Experiment

➢ Synthesis Route of PEG-P(GMA-API)$_{45}$



Scheme 1 the Synthesis Route of PEG-P(GMA-API)$_{45}$

➢ Encapsulation of Hemoglobin

Results and Discussions

Table 1. Molecular Characteristics

Samples	Mn[a]	Mw[a]	Mw/Mn[a]	Mn[b]
PEG-Br	9040	10153	1.12	5153
PEG-b-P(GMA)$_{45}$	11562	14260	1.23	11390
PEG-P(GMA-API)$_{45}$	16078	17465	1.08	17175

➢ Micelles Characterization

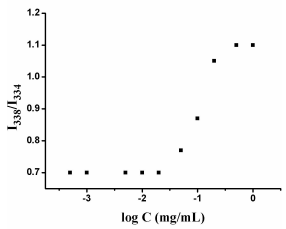


Fig. 2. Plots of I_{338}/I_{334}-log c, CMC=0.04 g/ml.

➢ Zeta Potential

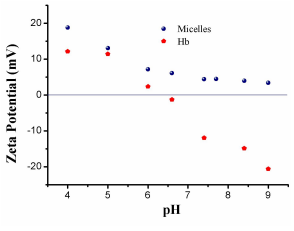


Fig.3. Zeta Potential of Hb and micelles in PBS with different pH

➢ Encapsulation of Hb in PEG-P(GMA-API)$_{45}$ Micelles

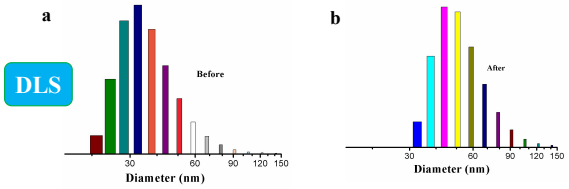


Fig. 4. DLS measurements. Size distributions of micelles before (**a**) and after Hb encapsulation (**b**).

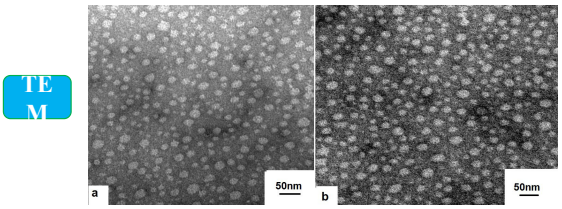


Fig. 5. TEM images of micelles before (**a**) and after Hb encapsulation (**b**).

- ✓ Hb could be encapsulated into micelles by electrostatic interaction and hydrogen-bonding interaction (pH7.4, 0.01M). The encapsulation efficiency is 32.4%.
- ✓ Micelles encapsulated Hb had larger particle size on average than blank micelles, and they had similar morphology.

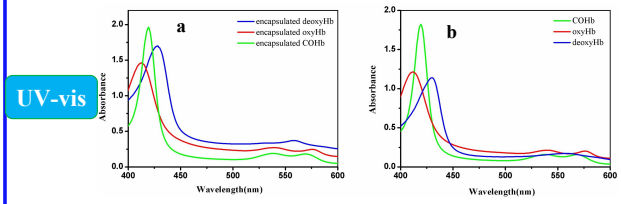


Fig. 6. UV-vis spectra of encapsulated Hb (**a**) and free Hb (**b**) in different gas-binding states

- ✓ The encapsulated Hb retained its biological activity, which is similar with free Hb.

Conclusions

- ☐ The amphiphilic copolymer micelles exhibit an outstanding performance in encapsulation of Hb in terms of encapsulation efficiency and biological activity of encapsulated Hb.
- ☐ They showed a promising future to be used as an artficial oxygen carrier.

References

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Acknowledgement

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