Xylose oligosaccharides preparation from agricultural waste catalyzed by carrier-free immobilized enzymes

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Introduction

XOS is a new green additive, which can be used in medicine, agriculture, cosmetics and other fields. The content of agricultural wastes such as rice cob, bran, mulberry branch and soybean straw was higher in xylan. But most of them have been wasted. Commonly used methods are chemical catalysis and biological catalysis. Chemical reaction conditions are harsh and polluting, while enzymatic method is expensive, enzyme stability and cannot be reused. In this study, xylanase-polymer conjugates were prepared to enable xylanase to maintain high stability and reuse times with high catalytic efficiency. It is innovative and economic to hydrolyze agricultural wastes

Methods

In this paper, the xylanases was used to synthesize polymer graft XYL containing different alkyl side chains by ATRP technology, and on this basis, the effect of polymer grafting on the molecular structure of XYL was systematically investigated.

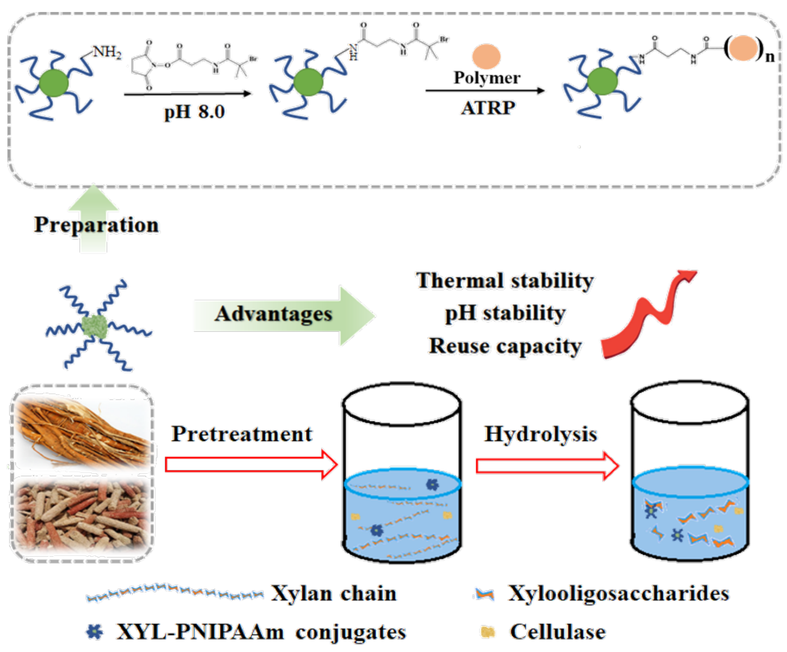

Fig.1 Enzyme-grafted polymer catalyzed hydrolysis

Results & Discussion

The understanding of the mechanism of regulating the XYL enzyme activity mechanism of polymer molecular side chain structure regulation was deepened by the determination of the activity parameters and the stability of polymer graft XYL enzyme.

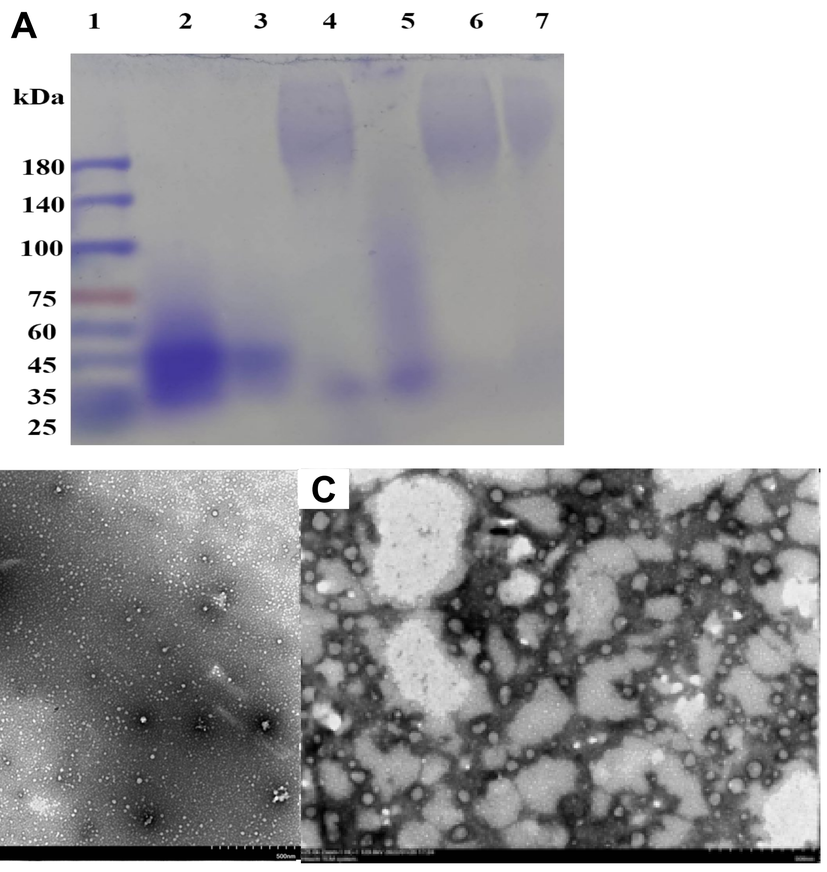

Fig. 2 (A)SDS-PAGE profile of xylanase and xylanase-polymerLine 1: Maker, Line 2: XYL, Line 3: XYL-MI, Line 4: XYL-GMA, Line 5: XYL-HEMA; Line 6: XYL-pMA; Line7: XYL-BMA SDS-PAGE; (B) TEM diagram of XYL, scale bar is 500nm (20000×); (C) TEM diagram of XYL-Polymer conjugates, scale bar is 200nm (50000×).

The SDS-PAGE results show the probably molecular weight of free XYL, XYL MI and conjugate.Conjugates were observed as broad bands at high molecular weight. As showed by the TEM micrographs in Fig.2(B)and (C), free lipase could be easily solubilized in water as dispersed particles and conjugated XYL in the form of protein clusters.Characterization results proved the successful prepared of XYL- Polymer conjugates.

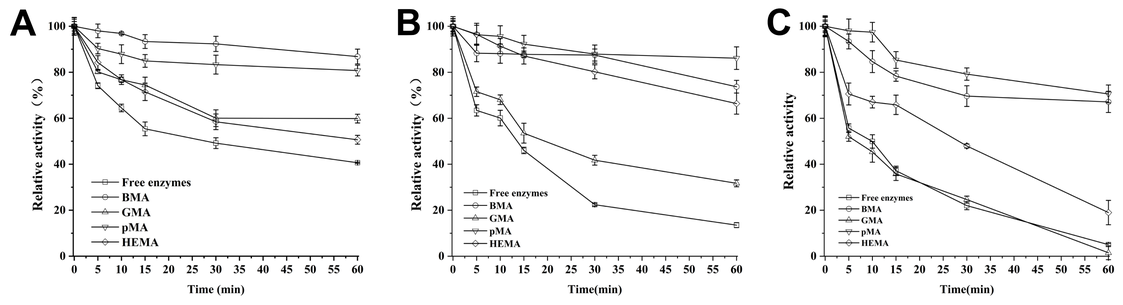

Fig. 3 (A) Temperature stability of free XYL, XYL-BMA, XYL-GMA, XYL-pMA and XYL-HEMA in 50℃; (B) Temperature stability of free XYL, XYL-BMA, XYL-GMA, XYL-pMA and XYL-HEMA in 80℃; (C) Temperature stability of free XYL, XYL-BMA, XYL-GMA, XYL-pMA and XYL-HEMA in 90℃.

Fig 3 shows polymer modification improves the thermal stability of the conjugates. Moreover, The molecular weight of polymer directly affects the properties of coupling compounds. It can be shown in Fig 3 (B) and (C) that the remain activity of XYL-pMA can keep over 80% in 80℃ and 70% in 90℃ after incubation 60 min.

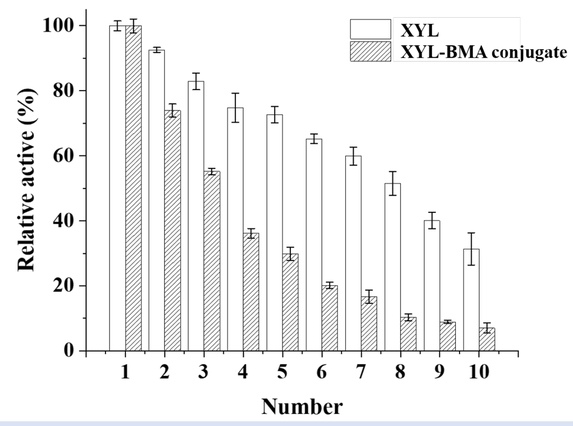

Fig. 4 Activities of free XYL and XYL-BMA conjugate according to the number of reuses. The reusability was determined by incubating both free XYL and XYL-BMA conjugate at 50 °C.

Fig.4 shows reuse times of conjugates far exceed free enzyme. The reactive activity of conjugates was 60.0% after repeated use for 7 times, while free XYL can only be used repeatedly for 2 times with the same enzyme activity.

Conclusions

- Successfully prepared of XYL- Polymer conjugates in the study.
- The optimum temperature and pH of XYL- Polymer conjugates have changed.Compared with the free enzyme, its optimum temperature changed to 60°C, and its optimum pH reached 6.
- Polymer grafted xylanase can increase the stability of enzyme.The remain activity of XYL-pMA can keep over 80% in 80℃ and 70% in 90℃ after incubation 60 min.
- Reuse times of conjugates far exceed free enzyme. The reactive activity of conjugates was 60.0% after repeated use for 7 times, while free XYL can only be used repeatedly for 2 times with the same enzyme activity.

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