

Graft Copolymerization of Granular Allyl Starch with Carboxyl-containing Vinyl Monomers for Enhancing Grafting Efficiency

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Abstract: Graft copolymerization of granular allyl starch with carboxyl-containing vinyl monomers using $\text{H}_2\text{O}_2/\text{Fe}^{2+}$ initiator in aqueous dispersion was investigated for enhancing grafting efficiency of the copolymerization. The graft copolymerization was evaluated in terms of grafting efficiency, grafting ratio, and conversion of monomer to polymer. Influences of both allyl etherification of starch and structures of vinyl monomers used on the copolymerization were revealed. Variables such as molar ratio of Fe^{2+} to H_2O_2 , initiator concentration, monomer concentration, polymerization temperature, and time of the graft copolymerization were also studied. It was found that allyl etherification of starch enhanced the grafting efficiency and grafting ratio evidently. Acrylic acid exhibited the greatest grafting efficiency and ratio for the copolymerization after starch was allyl-etherified. The copolymerization should be carried out under the protection of nitrogen gas at 30-35 °C for 3 h by using $\text{H}_2\text{O}_2/\text{Fe}(\text{NH}_4)_2(\text{SO}_4)_2$ as initiator. Preferred molar ratio of $\text{H}_2\text{O}_2/\text{Fe}(\text{NH}_4)_2(\text{SO}_4)_2/\text{anhydroglucose}$ was in a range of 20/1/1000 to 60/3/2000.

Keywords: Allyl starch, Starch graft copolymer, Graft copolymerization, Grafting efficiency, Grafting ratio

Introduction

Nowadays, starch graft copolymers are widely used in water treatment [1], paper-making [2], printing [3], finishing [4], and sizing [5,6]. The copolymers were prepared through the graft copolymerization of starch with various vinyl monomers. It has been demonstrated that the copolymerization is an effective method for improving the end-use ability of starch [7,8]. Apparently, grafting efficiency and grafting ratio are the two essential parameters and directly determine the performance of the copolymers [9]. Generally, the greater the grafting efficiency, the higher the grafting ratio when the monomer used is constant. In this case, better performance could be achieved. For this reason, some studies paid much attention to the efficiency. The investigators tried to enhance the grafting efficiency through exploring suitable initiators [10], pre-gelatinizing starch before the copolymerization [11], and choosing suitable medium for the copolymerization (e.g., toluene [12], 1-allyl-3-methylimidazolium chloride [13], and *N,N*-dimethylacetamide [14]).

It should be noted that the graft copolymerization of starch with vinyl monomers is always accompanied with homopolymerization of vinyl monomers. The homopolymerization that reduces grafting efficiency is a side reaction of graft copolymerization. It can be imagined that, by introducing double bonds onto starch molecules before graft copolymerization, the double bonds can be also polymerized together with vinyl monomers when the bonds meet propagating activated chains in the graft copolymerization. In this case, part of the homopolymers would be converted to graft copolymers and the grafting efficiency would be expected to enhance. For

this reason, using allyl starch is one of the effective ways that increase the efficiency. The effectiveness of allyl starch used in the graft copolymerization for enhancing the efficiency has been demonstrated through our previous study [15].

Up to the present time, many investigators have explored suitable condition for the copolymerization of natural starch with vinyl monomers and obtained some interesting results [16,17]. However, no investigation took the condition required for the copolymerization of allyl starch with vinyl monomers into serious consideration. Only imitating the graft copolymerization condition of natural starch for allyl one is inappropriate since double bonds exist in allyl starch. Moreover, monomer structure also influences the copolymerization and grafting efficiency [18]. Naturally, it is necessary to investigate suitable condition required for the graft copolymerization of allyl starch with carboxyl-containing vinyl monomers.

In present study, by choosing Fenton's initiator, the reaction variables for the graft copolymerization of allyl starch with carboxyl-containing vinyl monomers are investigated in order to achieve greater grafting efficiency. The effect of allyl etherification of starch on the copolymerization is revealed. Then the variables, i.e., the molar ratio of Fe^{2+} to H_2O_2 , initiator concentration, monomer concentration, polymerization temperature and time, were studied for increasing the grafting efficiency and grafting ratio. In this way, desired condition could be obtained for improving the performance of the copolymers.

Experimental

Materials and Reagents

Commercial cornstarch, with a moisture content of 13.8 %

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and an apparent viscosity of 52 mPa·s (6 %, 95 °C), was supplied by Yixing Starch Factory (Jiangsu Province, China). The cornstarch was refined to eliminate protein [19] and hydrolyzed to reduce excessive viscosity [20] to 16 mPa·s before use. Allyl chloride, acrylic acid (AA), methacrylic acid (MAA), and itaconic acid (IA) were chemically pure reagents and offered by Sinopharm Chemical Reagent Co. Ltd. (Shanghai, China). The other reagents were all analytically pure.

Allyl Etherification of Starch

80 g (dry weight) of hydrolyzed cornstarch were mixed with distilled water-isopropyl alcohol (20/80, w/w) to form a 30 % (w/w) suspension. After 1 g of sodium hydroxide dissolved in 25 ml of distilled water was added, the suspension was heated to 30 °C and stirred with mechanical stirrer. 30 min later, allyl chloride-isopropanol solution was charged into the flask and the reaction was continued at 30 °C for 24 h. The starch was filtered, re-dispersed in distilled water-alcohol solution (18:82, v/v), and neutralized with dilute hydrochloric acid (4 mol/l) to pH 6-7. Finally, the product was filtered, washed thoroughly with water, dried at 50 °C, pulverized, and sieved for passing through a 100-mesh sieve.

Graft Copolymerization

60 g (dry weight) of allyl starch and 120 ml of distilled water were mixed, and the mixture was adjusted to pH 3-4 with dilute sulfuric acid and transferred into a 250 ml four-neck flask. The flask was maintained in a thermostatically controlled water bath at required temperature. After nitrogen gas was charged into the flask for 30 min, a certain amount of monomers and Fenton's initiator solution were added continuously through dropping funnels and the addition was completed uniformly in 20-30 min, respectively. The graft copolymerization was carried out under vigorous stirring and the protection of nitrogen gas for predetermined hours. After adding 1.6 ml of 2 % paradioxybenzene solution to terminate the copolymerization, the product was neutralized to pH 6-7, filtered, washed thoroughly with distilled water, dried at 50 °C, pulverized, and sieved using a 100-meshed sieve.

Characterization of Allyl Starch

The amount of the allyl groups introduced was measured by titrimetric analysis. The double bonds of allyl groups reacted with bromine and the residual bromine then reacted with iodine. The excessive iodine was titrated with sodium hyposulfite [21]. The degree of substitution (DS) was calculated using equations (1) and (2).

$$DS = \frac{162x}{79.904 \times (2 - 0.5006x)} \quad (1)$$

$$x = \frac{(V_1 - V_2) \times C \times 0.0799}{W} \times 100 \quad (2)$$

where x (%) was the bromine value; V_1 (ml) and V_2 (ml) were the volumes of the standard sodium hyposulfite solution consumed in blank and sample titration, respectively; C (mol·l⁻¹) was the concentration of the standard sodium hyposulfite solution, and W (g) was the dry weight of starch sample.

Measurement on Grafting Parameters

Purification was applied to remove the homopolymers from the products of the graft copolymerization. The samples were washed with distilled water [22] and the homopolymer in filtrate was examined in accordance with Bayazeed's method [23].

Conversion of monomer to polymer (%) was determined by measuring the amount of residual monomers through titrating the double bonds of residual monomers in reaction filtrate [21]. Conversion of monomer to polymer was calculated using equation (3).

$$\text{Conversion of monomer to polymer (\%)} = \frac{W_1 - W_2}{W_1} \times 100 \quad (3)$$

where W_1 and W_2 denoted the weights of charged and residual monomers, respectively.

Grafting ratio was used to indicate the amount of grafted branches on starch molecules. The ratio was measured by the titration of carboxylic acid groups in grafted branches. Sodium carboxylate in grafted branches was initially converted to carboxylic acid with dilute HNO₃ solution. Then excessive sodium hydroxide standard solution was added to neutralize the carboxylic acid. Finally, the residual sodium hydroxide was titrated with hydrochloric acid standard solution. The titration method and calculation were described by Zhang [24]. The grafting ratio and efficiency were calculated as follows:

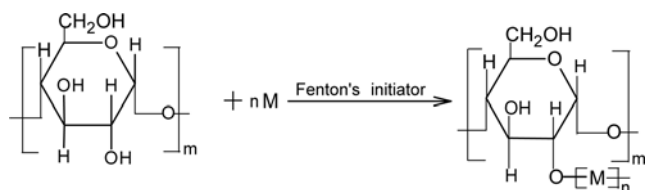
$$\text{Grafting ratio (\%)} = \frac{W_3}{W_0} \times 100 \quad (4)$$

$$\text{Grafting efficiency (\%)} = \frac{W_3}{W_1 - W_2} \times 100 \quad (5)$$

where W_3 and W_0 were the weights of grafted branches onto starch molecules and original starch, respectively.

Results and Discussion

Scheme 1 and 2 indicate the formation of the grafted branches in the copolymerization of granular allyl starch with vinyl monomers using H₂O₂/Fe²⁺ initiator. When allyl starch, instead of native starch, is used, the grafted branches produced will connect with the starch backbones not only through direct linkage with oxygen [25] via abstraction of hydrogen atom from starch hydroxyls as shown in Scheme 1, but also through the incorporation of the double bonds into the grafted branches [15] as shown in Scheme 2.



Scheme 1. Formation of the grafted branches connected with oxygen in hydroxyls by Fenton's initiator. M denotes the vinyl monomers.



Scheme 2. Formation of the grafted branches through incorporating double bonds into the grafted branches by Fenton's initiator. M denotes the vinyl monomers.

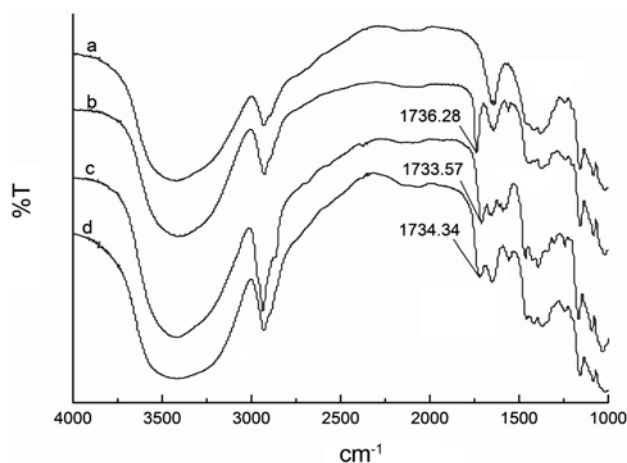


Figure 1. FTIR spectra of (a) pure allyl starch, (b) purified allyl starch-g-poly(acrylic acid), (c) purified allyl starch-g-poly(methacrylic acid), and (d) purified allyl starch-g-poly(itaconic acid).

Evidence of Grafting

FTIR spectra of purified allyl starch, allyl starch-g-poly(acrylic acid), allyl starch-g-poly(methacrylic acid), and allyl starch-g-poly(itaconic acid) are illustrated in Figure 1. A new peak appearing near 1735 cm^{-1} in the spectra of purified starch graft copolymers is corresponding to the carbonyl stretching vibration. This peak demonstrates the existence of carboxylic acid groups which are formed in the neutralization of products at the end of graft copolymerization.

Effect of Starch Etherification on the Copolymerization

It is observed from Table 1 that allyl etherification of

Table 1. Effect of starch etherification and monomer structure on the graft copolymerization^a

Starches	Monomers	Grafting parameters of starch graft copolymers		
		Conversion of monomer to polymer (%)	Grafting efficiency (%)	Grafting ratio (%)
Natural starch	Acrylic acid	98.0	53.2	5.21
	Methacrylic acid	96.3	51.8	4.99
	Itaconic acid	95.5	42.9	4.10
Allyl starch	Acrylic acid	99.6	64.9	6.46
	Methacrylic acid	98.1	62.4	6.12
	Itaconic acid	96.4	52.5	5.06

^aDS value of allyl starch 0.012; weight ratio of starch/liquor 3:7; weight ratio of monomer/starch 1:10; molar ratio of $\text{Fe}^{2+}/\text{H}_2\text{O}_2/\text{AGU}$ 1:20:1000; polymerization temperature $30\text{ }^\circ\text{C}$; polymerization time 3 h.

starch shows evident effect on the graft copolymerization. The etherification is able to increase the grafting efficiency and ratio while conversion of monomer to polymer does not vary significantly. Apparently, when the graft copolymerization of allyl starch with vinyl monomers occurs, propagating chain radicals might collide with the double bonds of allyl group on starch backbones. The double bonds will be incorporated into the propagating chains, thereby forming starch graft copolymers. The formation of extra grafted branches can be shown in Scheme 2. At the same time, another type of the formation of grafted branches shown in Scheme 1 also occurs. Obviously, the incorporation is capable of converting homopolymerization into copolymerization. And the double bonds on starch backbones can partly convert homopolymerization into graft copolymerization. Correspondingly, part of homopolymers would be transformed to copolymers. Therefore, both grafting efficiency and ratio are enhanced.

Structural effect of carboxyl-containing vinyl monomers on the graft copolymerization is shown in Table 1. According to grafting efficiency and ratio, the monomers used are ranged in the order: AA>MAA>IA. Based on the principle of graft copolymerization [25], grafted branches are formed through the bonding of starch radical with monomers. As is well known, α -methyl group in MAA occupies larger space than hydrogen atom in AA. When monomers are in the vicinity of allyl starch radicals, the graft copolymerization of MAA with the starch occurs in lower probability than that of AA does. It can be speculated that steric hindrance impedes the graft copolymerization. The greater the steric hindrance, the lower the grafting efficiency. Thus carboxymethyl in IA takes up larger space than α -methyl in MAA, greater steric hindrance of the substituent lowers grafting efficiency and ratio.

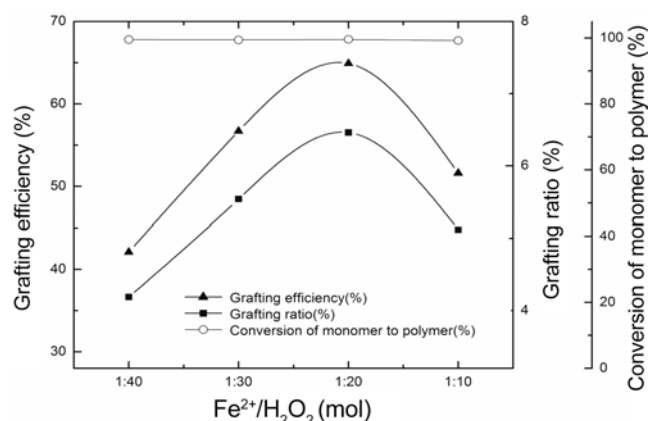
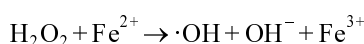


Figure 2. Effect of molar ratio of Fe²⁺/H₂O₂ on the graft copolymerization. DS value of allyl starch 0.012; weight ratio of starch/liquor 3:7; monomer acrylic acid; weight ratio of monomer/starch 1:10; molar ratio of H₂O₂/AGU 20:1000; polymerization temperature 30 °C; polymerization time 3 h.

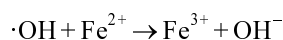
Effect of Molar Ratio of Fe²⁺/H₂O₂

Figure 2 shows the effect of molar ratio of Fe²⁺/H₂O₂ on the graft copolymerization of AA with granular allyl starch. Obviously, the grafting efficiency and ratio are significantly increased with increasing the molar ratio of Fe²⁺/H₂O₂ from 1:40 to 1:20. However, further increasing the molar ratio induces a decrease in grafting efficiency and ratio. In addition, the conversion of monomer to polymer does not show much sensitivity to the molar ratio of Fe²⁺/H₂O₂.

The presence of Fe²⁺ in the graft copolymerization contributes to the generation of hydroxyl radicals. The reaction occurs as follows:



With the increase in the molar ratio of Fe²⁺/H₂O₂ from 1:40 to 1:20, the yield of hydroxyl radicals increases and the number of allyl starch radicals is enhanced. This favors enhancing grafting efficiency and ratio. However, when the molar ratio of Fe²⁺/H₂O₂ is excessive, the probability of the reaction between ferrous ions and hydroxyl radicals increase markedly. The reaction [26] can be described as follows:



Obviously, the decrease in the number of effective hydroxyl radicals depresses the graft copolymerization and reduces the grafting efficiency and ratio. Based on the experimental results, it is preferred to apply 1:20 for the molar ratio of Fe²⁺/H₂O₂.

Effect of Molar Ratio of H₂O₂/Anhydroglucose

Initiator concentration is expressed with molar ratio of H₂O₂/anhydroglucose (AGU). Effect of the molar ratio on

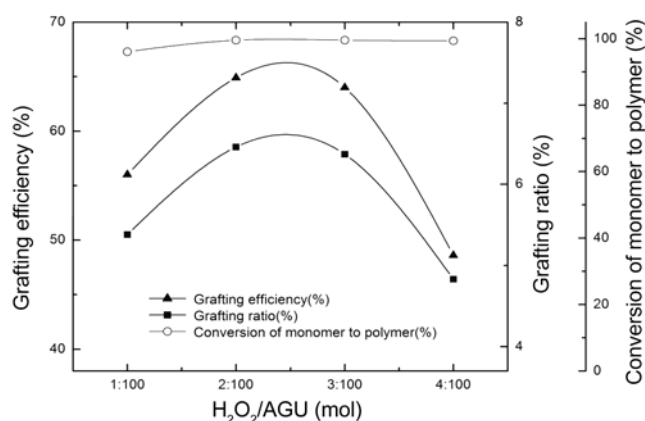


Figure 3. Effect of initiator concentration on the graft copolymerization. DS value of allyl starch 0.012; weight ratio of starch/liquor 3:7; monomer acrylic acid; weight ratio of monomer/starch 1:10; molar ratio of Fe²⁺/H₂O₂ 1:20; polymerization temperature 30 °C; polymerization time 3 h.

the graft copolymerization is described in Figure 3. Apparently, initiator concentration shows significant impact on the grafting efficiency and ratio, while the conversion of monomer to polymer enhances very slowly with the increase in initiator concentration. With the increase in the molar ratio of H₂O₂/AGU, the grafting efficiency and ratio increase initially, reach their maximum in a range of 2:100-3:100, and then decrease.

Initial increases in grafting efficiency and ratio are ascribed to the formation of increased amount of the radicals on the allyl starch backbone onto which the monomers can be grafted, thereby promoting the graft copolymerization. However, the grafting efficiency and ratio start to decrease when the molar ratio of H₂O₂/AGU exceeds 3:100. There are two major reasons for the decreases in the two parameters at excessively high initiator concentration: (1) the hydroxyl radicals can recombine among themselves, and (2) the excessively high initiator concentration causes an increased probability in both the termination of growing chain radicals and the initiation of homopolymerization of AA. Generally, the higher concentration of initiator will generate more hydroxyl radicals. The radicals are more likely to participate in the termination of growing chain radicals and to initiate homopolymerization. Experimental results indicated that maximal values both in grafting efficiency and ratio can be achieved simultaneously if the molar ratio of H₂O₂/AGU is in a range of 2:100-3:100.

Effect of Polymerization Temperature

As illustrated in Figure 4, the grafting efficiency and ratio depend on polymerization temperature, but no significant change in conversion of monomer to polymer is observed. The grafting efficiency and ratio initially increase when the temperature varies from 20 to 35 °C and then decrease with

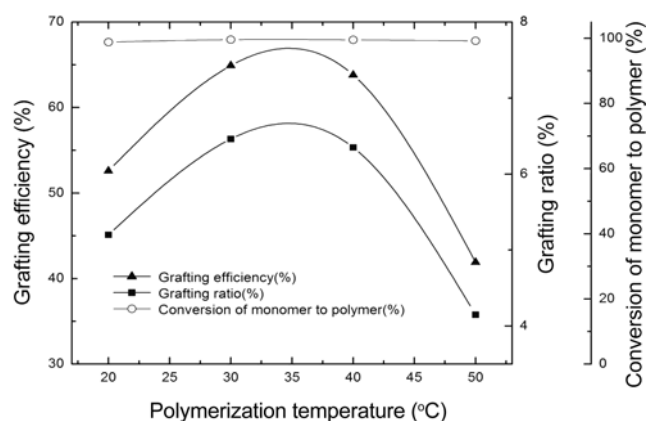


Figure 4. Effect of polymerization temperature on the graft copolymerization. DS value of allyl starch 0.012; weight ratio of starch/liquor 3:7; monomer acrylic acid; weight ratio of monomer/starch 1:10; molar ratio of $\text{Fe}^{2+}/\text{H}_2\text{O}_2/\text{AGU}$ 1:20:1000; polymerization time 3 h.

the further rise in the temperature.

With the increase in the temperature, swelling of starch granules increases. The increase favors the diffusion of initiators and monomers into starch granules. Moreover, the mobility of starch radicals is accelerated, which enhances the probability of their collision. Therefore, the grafting efficiency and ratio enhance initially. However, if the temperature is excessive, the rate of homopolymerization will possibly turn faster than that of graft copolymerization [16] and thus more homopolymers will be formed. In addition, the growing chains are also deactivated greatly by termination reaction. Therefore, the grafting efficiency and ratio decrease. Based on the experiment, the temperature of the graft copolymerization should be in the range of 30-35 °C.

Effect of Polymerization Time

The influence of polymerization time on the graft copolymerization is shown in Figure 5. The conversion of monomer to polymer, grafting efficiency, and grafting ratio are observed to reach their maximum at 3 h. After 3 h, the three parameters level off. This means that nearly all the monomers charged have been polymerized and the conversion of monomer to polymer arrives at nearly 100 % after 3 h of polymerization. The depletion of the monomers available for graft copolymerization causes grafting efficiency and ratio to level off. Therefore, the polymerization time should be 3 h.

Effect of AA Amount

The dependence of grafting efficiency and ratio on AA amount is shown in Figure 6. It can be observed that with the increase in AA amount, the grafting efficiency increases initially, reaches its maximum when AA amount is 10 % (w/w) and then decreases markedly. On the other hand, the grafting ratio increases continuously. The conversion of monomer to

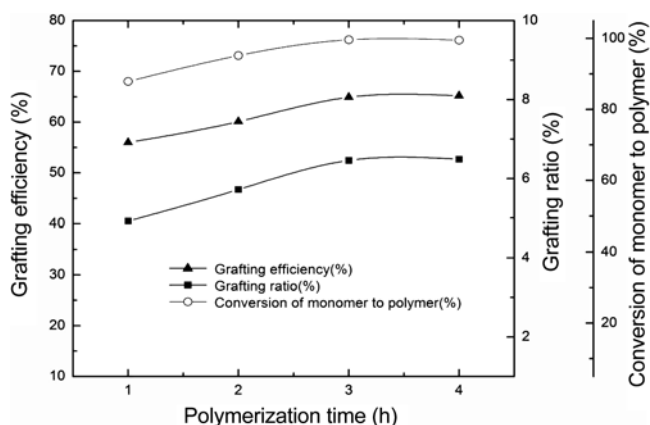


Figure 5. Effect of polymerization time on the graft copolymerization. DS value of allyl starch 0.012; weight ratio of starch/liquor 3:7; monomer acrylic acid; weight ratio of monomer/starch 1:10; molar ratio of $\text{Fe}^{2+}/\text{H}_2\text{O}_2/\text{AGU}$ 1:20:1000; polymerization temperature 30 °C.

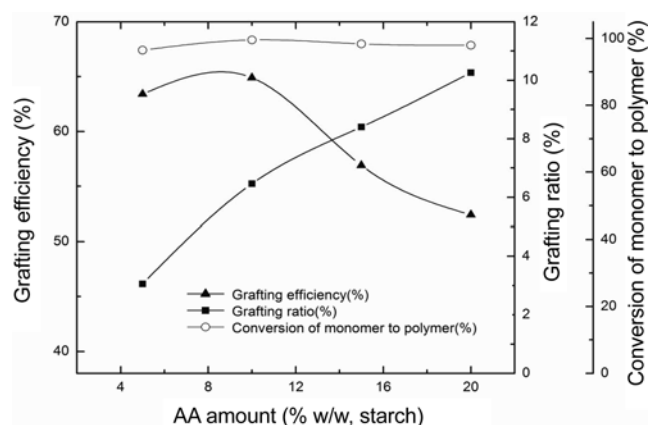


Figure 6. Effect of AA amount on the graft copolymerization. DS value of allyl starch 0.012; weight ratio of starch/liquor 3:7; monomer acrylic acid; molar ratio of $\text{Fe}^{2+}/\text{H}_2\text{O}_2/\text{AGU}$ 1:20:1000; polymerization temperature 30 °C; polymerization time 3 h.

polymer exhibits no marked change with the increase in AA amount.

The enhancement in the grafting ratio may be attributed to greater availability of AA molecules in the proximity of allyl starch radicals at higher AA concentration. It is understandable that the reaction of the radicals with monomers depends mainly on the concentration of monomer in the vicinity of the radicals since allyl starch macromolecule radicals lack mobility. With the increase in AA concentration, the monomers in the vicinity of allyl starch radicals and in the reaction medium are all more available. As a result, the probability of effective collision increases. The increase results in an increased amount of polymers. As the polymerization proceeds, the radical sites on the starch backbone are gradually occupied by the graft branches. And the residual

monomers in the reaction medium would be polymerized mainly through homopolymerization. For this reason, the grafting efficiency is then reduced due to excessive monomer concentration.

Conclusion

Allyl etherification of starch can enhance the grafting efficiency and ratio of graft copolymerization of starch with carboxyl-containing vinyl monomers when the copolymerization is carried out in aqueous dispersion using $\text{H}_2\text{O}_2/\text{Fe}^{2+}$ initiator. Moreover, the structure of the vinyl monomers also shows evident influence on the graft copolymerization. Steric hindrance of the monomers constrains the increases of grafting efficiency and ratio. Therefore, using AA is capable of increasing the efficiency and ratio.

Reaction variables such as molar ratio of Fe^{2+} to H_2O_2 , initiator concentration, polymerization temperature and time show evident effects on the graft copolymerization. The grafting efficiency and ratio increase as molar ratio of Fe^{2+} to H_2O_2 , initiator concentration, and polymerization temperature initially increase, after their maximum values are reached, the efficiency and ratio decrease with further increasing these variables. By extending the period of the graft copolymerization, the grafting efficiency and ratio increase continuously and then level off. In order to achieve higher grafting efficiency and ratio, the graft copolymerization of AA onto granular allyl starch should be carried out in aqueous dispersion under the protection of nitrogen gas at 30-35 °C for 3 h by using Fenton's initiator. Reasonable molar ratio of $\text{H}_2\text{O}_2/\text{Fe}(\text{NH}_4)_2(\text{SO}_4)_2/\text{anhydroglucose}$ was recommended from 20/1/1000 to 60/3/2000.

References

1. G. Gamze and K. Sibel, *J. Appl. Polym. Sci.*, **106**, 2422 (2007).
2. S. J. Lu, S. B. Lin, and K. D. Yao, *Starch*, **56**, 138 (2004).
3. R. N. Ghosh, T. Jana, B. C. Ray, and B. Adhikari, *Polym. Int.*, **53**, 339 (2004).
4. A. Hebeish, A. A. Aly, A. M. El-Shafei, and S. Zaghloul, *Starch*, **60**, 97 (2008).
5. X. D. Zhang, W. Y. Li, and X. Liu, *J. Appl. Polym. Sci.*, **88**, 1563 (2003).
6. K. M. Mostafa and A. A. El-Sanabary, *Polym. Degrad. Stab.*, **55**, 181 (1997).
7. S. Kalia, B. S. Kaith, S. Sharma, and B. Bhardwaj, *Fibers Polym.*, **9**, 416 (2008).
8. Z. Jia and S. Y. Du, *Fibers. Polym.*, **7**, 235 (2006).
9. B. Zhang and Y. Y. Zhou, *J. Donghua Univ. Nat. Sci. Ed.*, **31**, 86 (2005). (in Chinese)
10. J. L. Willett and V. L. Finkenstadt, *J. Appl. Polym. Sci.*, **99**, 52 (2006).
11. C. C. Nguyen, V. J. Martin, and E. P. Pauley, *U. S. Patent*, 5003022 (1991).
12. L. Chen, Y. Ni, X. Bian, X. Qiu, X. Zhuang, X. Chen, and X. Jing, *Carbohydr. Polym.*, **60**, 103 (2005).
13. Q. Xu, Q. R. Wang, and L. J. Liu, *J. Appl. Polym. Sci.*, **107**, 2704 (2008).
14. Y. C. You, C. Y. Zhu, J. L. Jiao, and X. Shen, *Acta Polym. Sinica*, **6**, 746 (2000). (in Chinese)
15. Z. F. Zhu, M. L. Li, and E. Q. Jin, *J. Appl. Polym. Sci.*, **112**, 2822 (2009).
16. B. Zhang and Y. Y. Zhou, *Polym. Compos.*, **29**, 506 (2008).
17. A. Hebeish, M. K. Beliakova, and A. Bayazeed, *J. Appl. Polym. Sci.*, **68**, 1709 (1998).
18. R. X. Zhuo, L. Huang, and Z. F. Zhu, *J. Wuhan Univ. Nat. Sci. Ed.*, **44**, 163 (1998). (in Chinese)
19. Z. F. Zhu and R. X. Zhuo, *Eur. Polym. J.*, **37**, 1913 (2001).
20. Z. F. Zhu and P. H. Chen, *J. Appl. Polym. Sci.*, **106**, 2763 (2007).
21. Z. F. Zhu, Z. Y. Qiao, C. Z. Kang, and Y. H. Li, *J. Appl. Polym. Sci.*, **91**, 3016 (2004).
22. V. D. Athawale and V. Lele, *Carbohydr. Polym.*, **35**, 21 (1998).
23. A. Bayazeed, M. R. Elzairy, and A. Hebeish, *Starch*, **41**, 233 (1989).
24. Y. P. Zhang, "Production and Application of Modified Starch", pp.323-324, Chemical Industry Press, Beijing, 2004. (in Chinese)
25. M. W. Meshram, V. V. Patil, S. T. Mhaske, and B. N. Thorat, *Carbohydr. Polym.*, **75**, 71 (2009).
26. K. D. Asim, K. D. Binay, and B. Sekhar, *Environ. Prog.*, **25**, 64 (2006).