Effects of Different Amino Acids on Physicochemical Properties of Rice Starch

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Abstract. In this study, we investigated the effects of different amino acids (Glycine acid (Gly), lysine (Lys), arginine (Arg), aspartic acid (Asp) and glutamic acid (Glu)) and different levels of amino acids (1 %, 5 %, 10 %) on the physicochemical properties (transparency, supernatant, solubility, swelling, freeze-thaw stability) of rice starch. The results showed that Lys and Arg significantly increased the swelling power of rice starch, but Asp and Glu had the opposite effect. All charge-carrying amino acids reduced the transparency of rice starch, while increased the settling property and the syneresis of freeze-thaw treatment process. Gly had no significant effect on the properties of rice starch.

1. Introduction
Starch is the main component of rice, accounting for more than 80% of its dry mass. Rice starch has the characteristics of easy digestion, small granules, white, and good freeze-thaw stability. It is an important factor affecting the development and application of starch.

Gelatinization refers to that when starch granules are heated together with water, they absorb water and expand rapidly, and when the temperature is over 50 °C, the intra- and inter-molecular hydrogen bonds collapsed and the crystal structure becomes stable[1]. Along with this, the viscosity of the starch solution increased sharply [2]. This plays an important role in starch-based foods.

Many substances, such as proteins and polysaccharides, can significantly affect starch gelatinization and retrogradation behavior[3]. Rice starch is the main ingredient in people's daily diet, but little research has been done on the effect of amino acids on rice starch. Therefore, the purpose of this study was to investigate the effects of different amino acids (glycine acid (Gly), lysine (Lys), arginine (Arg), aspartic acid (Asp) and glutamine acids (Glu)) and different levels (1 %, 5 %, 10 %) on the physicochemical properties of rice starch.

2. Materials and Methods

2.1 Materials
Rice starch was provided by Jinnong Biological Technology Company, (Wu Xi, China). Amino acids (Lys, Arg, Asp, Glu and Gly) were provided by Changhua Biological Technology Company (Tian Jin, China).
2.2 Swelling Power and Solubility
Appropriate modifications were made to measure the swelling power and solubility of starch[4]. Put an aqueous suspension of rice starch (1 %, w / w) with different amino acid levels (1 %, 5 %, 10 %) into a beaker, heated in a water bath at different temperatures (65 °C, 75 °C, 85 °C, 95 °C) for 30 minutes with manual stirring. The control group was rice starch without added amino acid. All samples were then cooled to room temperature and centrifuged at a speed of 4000r/min for 20 minutes at 20 °C. The supernatant was poured into an aluminum box of known mass and constant weight. Put the box into a constant temperature oven and dried at 105 °C for 24 hours. Weighted the dried supernatant (Ws) and the precipitate (Wp). The weight of rice starch was 0.25g (W). The swelling power and solubility was calculated according to the following formula:

Swelling Power (g/g) = \( \frac{W_p}{W(100-S)} \times 100\% \) \hspace{1cm} (1)

Solubility(%) = \( \frac{W_s}{W} \times 100\% \) \hspace{1cm} (2)

2.3 Transparency
Put an aqueous suspension of rice starch (1%, w / w) with different amino acid levels (1%, 5%, 10%) into a beaker, heated in water bath at 95 °C and stirred manually for 30 minutes. The starch suspension was then cooled to room temperature. UV / VIS spectrometer (XinshijiT6, Beijing General Analysis General Instrument Corp, China) was used to measure the transmittance (% T) of the sample at 620 nm (with distilled water as a blank control).

2.4 Supernatant
Put an aqueous suspension of rice starch (2%, w / w) with different amino acid levels (1%, 5%, 10%) into a beaker, heated in a boiling water bath (95 °C) and stirred manually for 30 minutes. Then cooled to room temperature. 10 mL of starch paste was taken and placed in a stoppered test tube. The sedimentation volume of the lower layer was observed every 1 hour. After the sedimentation volume was stable, calculated the sedimentation volume ratio.

2.5 Syneresis after freeze-thaw
Rice starch paste (10%, w / w) with different amino acids and levels (1%, 5%, 10%) were put into a beaker, heated in a water bath at 95 °C for 20 minutes with manual stirring. The paste was then accurately weighed (4 g) into a centrifuge tube and stored in the refrigerator at -20 °C for 22 h. After that, thawed the frozen paste at room temperature for 2 hours. Then the paste was centrifuged at 4000r/min for 20 min. Repeated above operation 5 times, and measured the percentage of separated water, and calculated the water separation rate (%) according to the formula[5].

\[ R = \left( \frac{m_1 - m_2}{m_1 - m_0} \right) \times 100\% \] \hspace{1cm} (3)

In the formula: \( m_0 \) is the weight of the centrifuge tube (g); \( m_1 \) is the total weight of the centrifuge tube and the sample after thawing (g); \( m_2 \) is the total weight of the centrifuge tube and starch paste after the supernatant is discarded after centrifugation (g).

2.6 Data analysis
All experiments were performed in triplicate. Data processing and graphing were performed by using Microsoft Excel. Data analysis was performed by using SPSS 26.0. Differences between the mean values were processed by using Duncan's multiple range test (p <0.05).

3. Results and analysis

3.1 Swelling power and solubility
Figure 1. Effect of amino acids on the swelling power of rice starch.

As shown in Figure 1, as the temperature continued to increase, the swelling power of rice starch continued to increase. In the range of 65 °C-85 °C, the starch swelling power increased rapidly. This may be related to the starting gelatinization temperature of rice starch, which is about 60 °C-70 °C. In the case of sufficient water, when the temperature exceeded the gelatinization temperature, the starch absorbed water quickly and gelatinizes. When the temperature exceeded 85 °C, the starch absorbed water and is substantially saturated. Compared with charged amino acids, uncharged amino acids (Gly) had no significant effect on the swelling power of rice starch (P > 0.05). Adding Lys and Arg (positively charged) increased the swelling power of rice starch, and its swelling power gradually increased with increasing concentration. Interestingly, the addition of Asp and Glu (negatively charged) reduced the swelling power of rice starch. As the concentration increased, its swelling power gradually decreased. The swelling behavior of starch is mainly determined by amylopectin, while amylase will inhibit starch swelling. That is to say, amylase can limit the degree of swelling of the starch and maintain the integrity of the starch granules. Therefore, charged amino acids may inhibit the leaching of amylase from starch granules, thereby reducing starch swelling.

Table 1. Effects of amino acid on the solubility of rice starch.

| Levels/% | 65°C  | 75°C  | 85°C  | 95°C  |
|---------|-------|-------|-------|-------|
| Starch  |       |       |       |       |
| 1       | 2.42±0.01a | 5.93±0.73b | 6.84±0.32a | 17.27±0.03b |
| 5       | 4.40±0.82a | 7.96±0.65c | 6.87±0.06a | 18.34±0.17c |
| 10      | 5.97±0.48b | 8.86±0.83d | 8.89±0.32b | 19.34±0.52e |
| Glu     |       |       |       |       |
| 1       | 9.65±1.19b | 7.13±0.12c | 6.99±0.63a | 19.54±0.04e |
| 5       | 7.81±0.60a | 10.78±0.51e | 12.39±0.36d | 16.42±0.31a |
| 10      | 11.61±1.64c | 13.46±0.17g | 15.11±1.22e | 18.69±0.47d |
| Asp     |       |       |       |       |
| 1       | 8.23±2.59a | 5.81±0.10b | 8.95±0.01b | 18.75±0.10d |
| Lys     |       |       |       |       |
8.53±1.42b 11.34±0.46f 11.42±0.06c 22.027±0.07f
10.67±1.34d 13.44±0.06a 15.0567±0.10e 24.78±0.66g

Arg
7.64±3.19a 4.82±0.23a 7.03±0.04a 18.19±0.23c
5.82±0.38c 11.25±1.23f 11.30±0.35c 26.42±0.33g
10.77±0.38c 11.25±1.23f 11.57±0.62c 26.42±0.33g

Gly
8.26±1.47a 8.30±0.35d 8.82±0.27b 17.27±0.45b
8.97±1.08b 10.82±0.02e 12.46±0.03d 24.84±0.02g
10.56±0.24c 11.9±0.56f 12.62±0.26d 29.22±0.06h

All values are the mean ± standard deviation of three replicates. Different letters following the values in the same column indicate significant differences (p <0.05).

It can be seen from Table 1 that, like its swelling capacity, the solubility of rice starch increased with the increase of the charge-carrying amino acid concentration as well (Figure 1). However, uncharged Gly had no significant effect on the solubility of starch (P> 0.05). The solubility is related to the amylese content leaked out during the swelling process. The more amylese exudated, the greater the solubility Added Asp and Glu (negatively charged) were the best, probably because the swelling of starch granules was suppressed, which increased the amylese spillage.

3.2 transparency

Light transmittance can reflect the transparency of starch under light, which depends on the swelling and unswelling particles. The light transmittance (%) of rice starch added with different amino acids is significantly lower than that of natural rice starch (Figure 2). As the amount of amino acid added increases, the transmittance value becomes lower. This is because the transmittance value of starch paste decreases with the increase of the degree of cross-linking, which prevents the dissociation of the starch chains and thus reduces the swelling of the particles[6]. When rice starch was heated, it interacted with water during the heating process, and the starch particles will swell, causing more light to pass through the particles without being reflected, thereby increased light transmission. The higher the charged amino acid concentration, the lower the value of the transmittance. This indicates that charged amino acids may reduce the interaction between starch particles and water, thereby reducing swelling of starch particles. Therefore, the photometric value decreases.
3.3 Supernatant

![Figure 3. Effect of amino acids on the supernatant of rice starch. Different letters indicate significant differences in mean value (p <0.05).](image)

The settling property can cause great changes in the mechanical properties of starch products, and then affect the appearance, taste, nutritional value and processing characteristics of food. In the rapid sedimentation experiment, the sedimentation volume after 48 h is shown in Figure 3. Compared with the original starch, the addition of amino acids significantly enhanced the starch's sedimentation volume ratio. With the increase of the amount of amino acids, the retrogradation was enhanced, and the charged amino acids could increase the retrogradation of rice starch more than the uncharged amino acids. It may be that the charged amino acids inhibit the swelling of rice starch. Therefore, reducing the ability of the starch granules to bind to water leads to an increase in the retrogradation of starch

3.4 syneresis after freeze-thaw

![Figure 4. Effect of amino acids on syneresis of rice starch. Different letters indicate significant differences in mean value (p <0.05).](image)

Water analysis rate is an important indicator to evaluate starch degradation during freeze-thaw treatment of starch. Due to the extrusion of the freeze-thaw crystals, the starch particles collide and aggregate. This in turn causes a retrogradation of the starch and produces a hard texture. As shown in Figure 4, the addition of Gly had no significant effect on the dehydration yield of rice starch paste during the freeze-thaw process. However, when charged amino acids were added to starch, the percentage of syneresis increased significantly. The addition of Asp and Glu significantly increased the water dehydration rate of rice starch. The high concentration of lysine significantly reduced the gel stability of rice starch. This may be after freezing and thawing at low temperatures, the molecular associations between starch chains increased, leading to syneresis and increased starch regeneration. Water is not included in the paste. The result showed that charge-carrying amino acids can enhance this association in starch paste.

4. Conclusion

The physical and chemical properties of rice starch in the presence of amino acids with different levels were studied based on swelling power, solubility, transparency, Supernatant, and syneresis. Charged
amino acids Lys, Arg have increased swelling, and Asp and Glu have reduced swelling. All charged amino acids can play a significant role in increasing solubility, supernatant, syneresis. However, glycine with a neutral charge has no significant effect on these properties of rice starch. The results showed that charged amino acids had more influence on the physicochemical properties of rice starch than neutral amino acids.

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References
[1] Tester, R. F., & Morrison, W. R. (1990). Swelling and gelatinization of cereal starches. I. Effects of amylopectin, amylose, and lipids. Cereal Chemistry, 67(6), 551–557.
[2] Yang, W., & Rao, M. (1998). Complex viscosity-temperature master curve of corn starch dispersion during gelatinization. Journal of Food Process Engineering, 21(3), 191–207.
[3] An, H., & King, J. (2009). Using ozonation and amino acids to change pasting properties of rice starch. Journal of Food Science, 74(3), C278–C283.
[4] Collado, L. S., & Corke, H. (1997). Properties of starch noodle as affected by sweetpotato genotype. Cereal Chemistry, 74(2), 182–187.
[5] Chen X L, Guan J, Mei X, et al. Effects of NaCl, sucrose and NaCO3 on pasting properties of starch from seeds of euryale ferox salisb[J]. Food Science, 2017,38(17):60-65
[6] Garg, S., & Jana, A. K. (2011). Characterization and evaluation of acylated starch with different acyl groups and degrees of substitution. Carbohydrate Polymers, 83(4), 1623–1630.