UDC 612.323: 612.822.2

THE EFFECT OF GASTRIC HYDROLYSIS OF PROTEINS ON THE FORMATION OF PROTEIN-POLYSACCHARIDE COMPLEXES

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✓ ***Resume***

The effect of gastric hydrolysis of proteins on the formation of protein-polysaccharide complexes was studied. The study was conducted in vitro, gastric juice, starch, casein, albumin solutions, as well as casein and albumin hydrolysates were used in the work. It is concluded that the use of a mixture of starch and casein or albumin contributes to a significant decrease in light transmission, which is the inverse of the increase in turbidity of the solution. Simultaneously, an increase in turbidity may be a consequence of the formation of starch-casein or starch-albumin complexes. Simultaneously, the use of a mixture of starch and casein hydrolysate or albumin hydrolysate contributes to higher light transmission or lower turbidity of the solution, which may be a consequence of the low formation of starch complexes and protein hydrolysates. The obtained results also demonstrate that the hydrolysis of proteins by pepsin of the stomach contributes to a decrease in the number of protein-polysaccharide complexes.

ВЛИЯНИЕ ЖЕЛУДОЧНОГО ГИДРОЛИЗА БЕЛКОВ НА ОБРАЗОВАНИЕ БЕЛКОВО-ПОЛИСАХАРИДНЫХ КОМПЛЕКСОВ

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✓ ***Резюме***

Изучили влияние желудочного гидролиза белков на образование белково-полисахаридных комплексов. Исследование проводили in vitro, в работе использовали желудочный сок, растворы крахмала, казеина, альбумина, а также гидролизаты казеина, альбумина. Сделано заключение, что использование смеси крахмала и казеина или альбумина способствует значительному снижению светопропускания, которое является обратной величиной увеличения помутнения раствора. При этом увеличение помутнения может являться следствием образования крахмально-казеиновых или крахмально-альбуминовых комплексов. В тоже время применение смеси крахмала и гидролизата казеина или гидролизата альбумина способствует более высокому светопропусканию или понижению помутнения раствора, что может являться следствием низкого образования комплексов крахмала и гидролизатов белков. Полученные результаты также демонстрируют, что гидролиз белков пепсинами желудка способствует снижению количества белково-полисахаридных комплексов.

ПРОТЕИН-ПОЛИСАККАРИД КОМПЛЕКСЛАРИНИНГ ШАКЛЛАНИШИГА ОШҚОЗОН ГИДРОЛИЗИНИНГ ОҚСИЛЛАРНИ ТАЪСИРИ

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✓ ***Резюме***

Оқсилларнинг меъда гидролизининг оқсил-полисахарид мажмуаларининг ҳосил бўлишига таъсири ўрганилди. Тадқиқот ўтказилди in vitro, меъда шираси, крахмал, казеин, албумин эритмалари, шунингдек казеин ва албумин гидролизатларидан фойдаланилган. Бундан шундай хулоса келиб чиқадики, крахмал ва казеин ёки албумин аралашмасидан фойдаланиш ёруғлик узатишнинг сезиларли пасайишига ёрдам беради, бу эса эритманинг лойқалиги ортишига тескари пропорционалдир. Шу билан бирга, лойқалик ортиши крахмал-казеин ёки крахмал-албумин комплекслари шаклланиши натижасида бўлиши мумкин. Шу билан бирга, крахмал ва казеин гидролизат ёки албумин гидролизат аралашмасидан фойдаланиш крахмал комплекслари ва оқсил гидролизатларининг паст шаклланиши натижасида бўлиши мумкин юқори ёруғлик узатиш ёки эритма паст лойқалик ҳисса қўшади. Олинган натижалар, шунингдек, ошқозон пепсинлари томонидан оқсилларнинг гидролизи оқсил-полисахарид комплекслари сонининг камайишига ёрдам беради.

Relevance

Food proteins and polysaccharides are two key structural elements of food substances. As a rule, polysaccharides can form various types of physical complexes with proteins, depending on the pH, ionic strength, and the charge distribution of the biopolymer. The formation of such complexes presupposes the predominance of short forces of attraction. Protein-polysaccharide interactions can be strong (long-lasting) or weak (reversible). Physical and chemical factors, such as pH, ionic strength, protein-to-polysaccharide ratio, polysaccharide, and protein charge, as well as molecular weight, affect the formation and stability of such complexes [5]. Precipitation is a constant problem for the increasingly widespread use of protein-polysaccharide complexes, but the relationship between the two types of phase separation is not fully understood. It was found that the formation of complexes and precipitation are, in fact, different phenomena that react to different factors, but their simultaneity (for example, with a change in pH) can be confused with transitions from one state to another. The formation of complexes does not turn into a precipitate, but rather that both the complexes and precipitate are in equilibrium with a free protein and a polysaccharide so that the dissolution of one and the formation of the other can overlap in time [2].

The pH factor is of great importance for the implementation of many intermolecular interactions since it affects the ionisation of some functional groups of biopolymer compounds [1].

Proteins are usually sensitive to hydrolysis under the action of gastric pepsin, but when they interact with polysaccharides, they may decompose to a lesser extent than free proteins [4,6].

The complex formation of protein and polysaccharides causes the formation of insoluble and soluble complexes that can resist the degradation of pepsin to varying degrees [3].

The aim of the study: To study the effect of gastric hydrolysis of proteins on the formation of protein-polysaccharide complexes.

Material and methods

The effect of casein and egg albumin proteins (albumin) and their gastric hydrolysates on interactions with starch and the formation of protein-polysaccharide complexes was studied in vitro. The degree of formation of protein-polysaccharide complexes was studied by changing the light transmission at pH 2–7 and 520 nm corresponding to the inverse value of the turbidity of the liquid, separately starch (0.2%), casein (1.0%), and albumin (1.0%), as well as a mixture of starch (0.2%) + casein (1.0%), starch (0.2%) + albumin (1.0%). Additionally, the light transmission of a joint mixture of starch (0.2%) + pepsin hydrolysate obtained after a 30-minute pre-incubation of casein (1.0%) with gastric juice was studied. Also, starch (0.2%) + pepsin hydrolysate was obtained after 30 min of preincubation of albumin (1.0%) with gastric juice. Mixtures of starch with proteins or with protein hydrolysates were used to determine the light transmission after a preliminary joint 30-minute incubation. The light transmission at 520 nm was determined as a percentage of the light transmission of water.

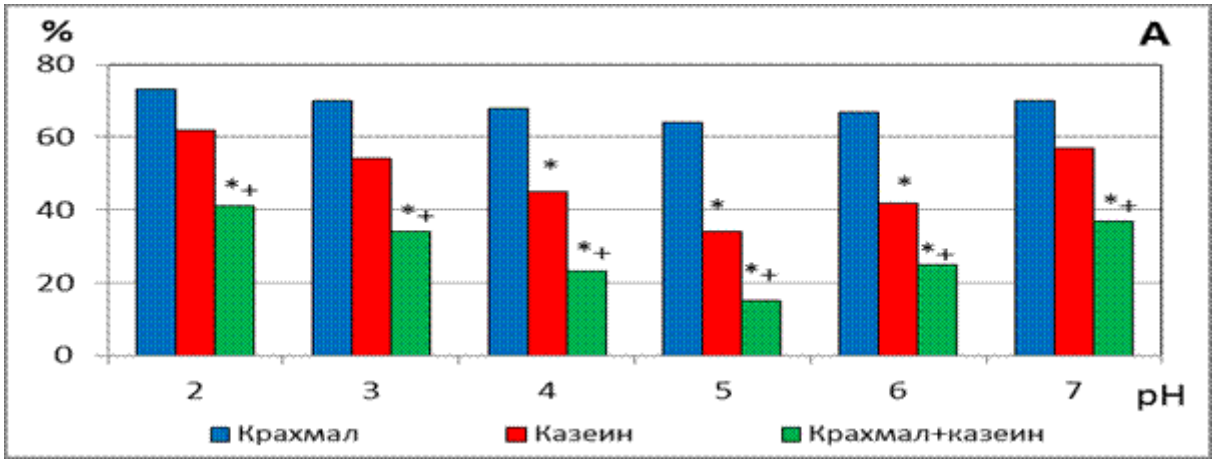
Result and discussion

Based on the obtained results of changes in the level of starch light transmission, it was found that this indicator at pH 2 was 73±6.9%. With an increase in pH, the level of starch light transmission decreased slightly, and at pH 5 it reached the minimum values and was 64±6.2%, which was unreliably lower than the result at pH 2. With a further increase in pH, an unreliable

increase in the degree of light transmission was noted, and at pH 7 this indicator was 70±6.5% (Fig. 1A).

Simultaneously, the level of light transmission of casein at pH 2 was 62±5.7%, which was not significantly lower than the results of starch at pH 2. A further increase in pH contributed to a decrease in the level of light transmission, and at pH 4 this level was 45±4.1%, which was significantly lower than the similar result of starch at pH 4. At the same time, at pH 5, the value of casein light transmission reached the minimum values and was 45±4.1%, which was significantly lower than similar starch data. The subsequent increase in pH 6 increased the light transmission of casein, but this indicator was significantly lower than the similar result of starch. At pH 7, the light transmission of casein also increased and was unreliable for the same result of starch (Fig. 1A).

According to the results of studying the light transmission value of a mixture of starch and casein, at pH 2, the light transmission index was 41±3.9%, which was significantly less than both starch and protein separately. A further increase in pH contributed to a further significant decrease in the light transmission of the starch and casein mixture relative to the starch and protein mixture separately. Simultaneously, at pH 5, the minimum light transmission was detected, which was 15±1.6%. The subsequent increase in pH contributed to a significant increase in the light transmission of the starch and casein mixture, and at pH 7 this indicator was 37±3.9%, but this result was significantly lower than similar starch data (Fig. 1A).



The degree of light transmission at pH 2 of the casein hydrolysate was 68±7.1%, which was slightly less than the similar results of starch. With an increase in pH, the level of light transmission of casein hydrolysate decreased and was unreliably lower than similar starch results and at pH 5 it reached a minimum value and was 52±5.1%, but also unreliable in relation to similar starch results. With a further increase in pH, an unreliable increase in the light transmission of casein hydrolysate indicators was noted, and these results were unreliable to those of starch, and at pH 7, the indicator was 64±5.9% (Fig. 1B).

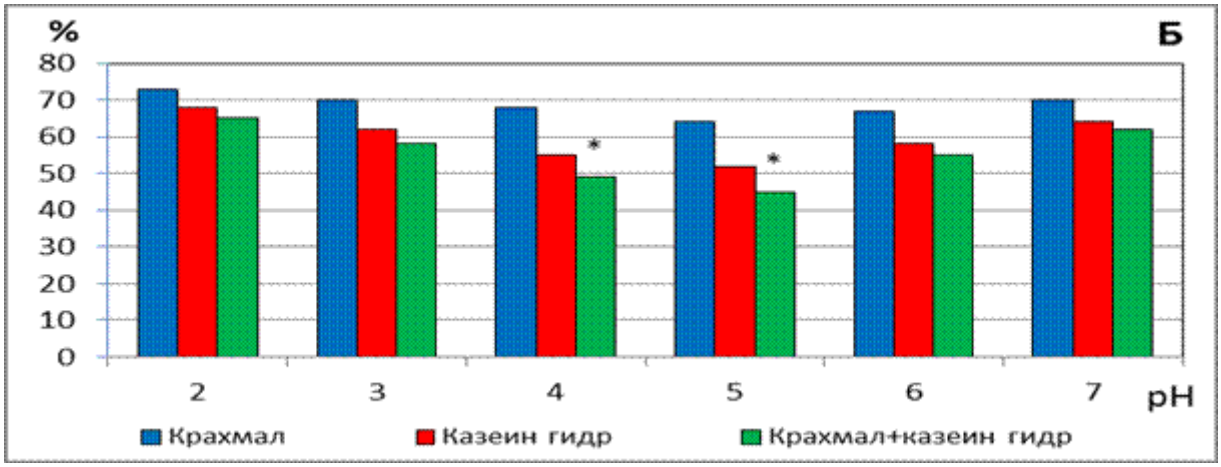


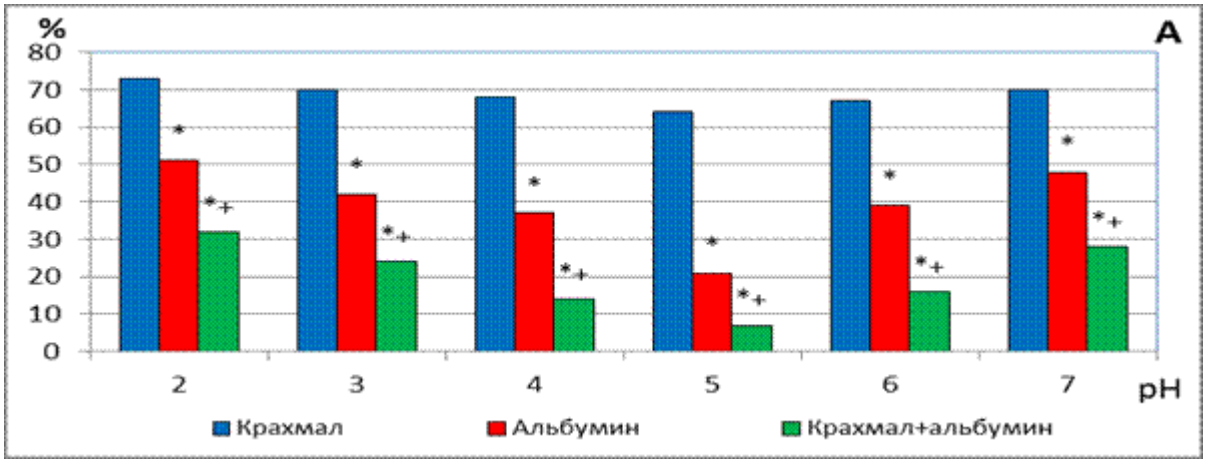
Figure 1. Study of light transmission at different pH values. A-a solution of starch, casein and a mixture of starch + casein. B-a solution of starch, casein hydrolysate and a mixture of starch + casein hydrolysate.

* - significantly different values of light transmission in relation to starch.

\+ - significantly different values of light transmission in relation to casein or casein hydrolysate.

When measuring the level of light transmission of a mixture of starch and casein hydrolysate, it was found that at pH 2, slightly less light transmission was observed (65±6.3%), compared with similar indicators of starch. With an increase in pH, the value of light transmission of a mixture of starch and casein hydrolysate in relation to similar starch results decreased significantly at pH 4 (49±4.5%), and reached reliable minimum values at pH 5 (49±4.5%). With a further increase in pH, an unreliable increase in the light transmission of a mixture of starch and casein hydrolysate was noted in relation to similar starch data (Fig. 1B).

The result of the study of albumin light transmission at pH 2 was 51±4.9%, which was significantly lower than similar starch results. Simultaneously, an increase in pH contributed to a decrease in light transmission indicators to pH 5, when this indicator reached a minimum and was 21±2.4%, simultaneously, this result was significantly and significantly lower in relation to starch indicators. A further increase in pH contributed to an increase in albumin light transmission, but these results were significantly lower than those of starch, and at pH 7 they were 48±5.1% (Fig. 2A).



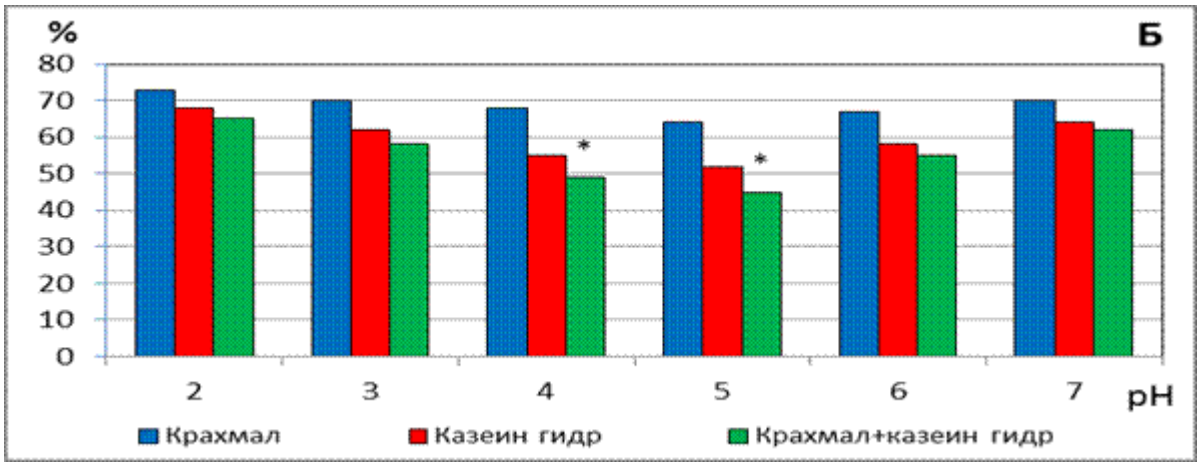


Figure 2. Investigation of light transmission at different pH values. A-a solution of starch, albumin and a mixture of starch + albumin. B-a solution of starch, albumin hydrolysate and a mixture of starch + albumin hydrolysate.

* - significantly different values of light transmission in relation to starch.

\+ - significantly different values of light transmission in relation to albumin or albumin hydrolysate.

When studying the degree of light transmission of a mixture of starch and albumin, at pH 2, the light transmission level was 32±3.5%, which was significantly less than both starch and protein separately. A further increase in pH contributed to an additional significant decrease in the light transmission of the starch and albumin mixture in relation to starch and protein separately. Simultaneously, at pH 5, the minimum light transmission was detected, which was 7±0.6%. The subsequent increase in pH contributed to a significant increase in the light transmission of the starch and albumin mixture, and at pH 7 this indicator was 28±3.2%. Which was also significantly lower separately for both starch and protein (Fig. 2A).

From the obtained data, it was found that at pH 2, the light transmission of albumin hydrolysate was 65±6.8% and was slightly less than the similar results of starch. With an increase in pH, the level of light transmission of albumin hydrolysate decreased and was unreliably lower than similar results of starch. And at pH 5, it reached the minimum value and was 47±5.0%, but it was reliable in relation to similar starch results. With further increase in pH was observed insignificant increase in light transmission hydrolysate of albumin, and at pH 7 was 64±5,9%, which was inaccurate to such data starch (Fig. 2B).

The results of the study of light transmission level of the mixture of starch and albumin hydrolysate at pH 2 were observed slightly lower transmittance compared to with similar indicators of starch, which was 60±6.1 percent. The value of the light transmission of the mixture of starch and albumin hydrolysate decreased with increasing pH and was significantly lower with respect to similar starch results at pH 4 and pH 5. Simultaneously, at pH 5, it reached reliable minimum values and was 36±3.2%. A further increase in pH caused a significant increase in the light transmission of a mixture of starch and albumin hydrolysate at pH 6 and an unreliable increase at pH 7 with respect to similar starch data (Fig. 2B).

Discussion of the results

The obtained results of these studies showed that the level of starch light transmission decreased slightly with increasing pH, and reached a minimum at pH 5, and then increased to a maximum at pH 7. Simultaneously, the amount of light transmission of casein had significant changes, decreasing with increasing acidity from pH 2 and reaching minimum values at pH 5. These changes in light transmission were lower than similar starch data, and at pH 4 and pH 5, there were significantly less starch results. With the subsequent increase in pH, an increase in light transmission was observed at pH 6 significantly, and at pH 7 it was not significantly lower than those of starch. When studying the light transmission of a mixture of casein and starch that can form protein-polysaccharide complexes, a significant change was also noted, which was manifested by similar dynamics of casein changes with an increase from pH 2 to pH 7. Simultaneously, there was a decrease in light transmission indicators in the range from pH 2 to pH 5, and with a further increase to pH 7, an increase in these indicators was observed. Nevertheless, all the results from pH 2 to pH 7 were significantly and significantly lower, both for starch and casein.

The conducted studies also showed a less pronounced decrease in the light transmission of casein hydrolysate, in relation to those results of starch, as well as less pronounced compared with similar results of casein. Simultaneously, there was a less pronounced decrease in the light transmission of a mixture of starch and casein hydrolysate, which can form protein-polysaccharide complexes in relation to those results of starch, and also less pronounced compared with those results of a mixture of starch and casein.

Additionally, from the data obtained, it was found that the level of albumin light transmission had significantly pronounced changes, while there was a decrease in light transmission from pH 2, which reached the minimum values at pH 5. All these changes in light transmission were also significantly and significantly lower than the results of starch, as well as less than those of casein. With a further increase to pH 7, an increase in light transmission was observed, and these increase indicators were significantly lower than those of starch data. Simultaneously, there was a significant change in the light transmission of a mixture of albumin and starch, which can form protein-polysaccharide complexes, which was manifested by similar dynamics of albumin changes with an increase from pH 2 to pH 7. Simultaneously, there was a decrease in light transmission indicators in the range from pH 2 to pH 5, and with a further increase to pH 7, an increase in these indicators was observed. Nevertheless, all the results of the mixture of albumin and starch from pH 2 to pH 7 were significantly and significantly lower than both the indicators of starch and the mixture of starch and casein.

Also, the obtained results showed a slight decrease in the light transmission of albumin hydrolysate, compared with similar results of

starch, and lower than similar results of casein hydrolysate. Additionally, there was a less pronounced decrease in the light transmission of a mixture of starch and albumin hydrolysate, which can form protein-polysaccharide complexes, in relation to those results of starch, as well as less pronounced compared with those results of a mixture of starch and casein.

Thus, the use of a mixture of starch and casein or albumin, which can form protein-polysaccharide complexes, contributes to a significant decrease in light transmission, which is the inverse of the increase in turbidity of the solution. Simultaneously, we can talk about an increase in turbidity, which may be a consequence of the formation of starch-casein or starch-albumin complexes. Simultaneously, the use of a mixture of starch and casein hydrolysate or albumin hydrolysate contributes to higher light transmission or reduced turbidity of the solution, which may be due to the low formation of starch complexes and protein hydrolysates. The obtained results also demonstrate that the hydrolysis of proteins by pepsin of the stomach reduces the number of protein-polysaccharide complexes that can interfere with the hydrolysis of proteins under the influence of pepsin in the stomach.

Conclusions

The use of a mixture of starch and casein or albumin contributes to a significant decrease in light transmission, which is the inverse of the increase in turbidity of the solution. Simultaneously, an increase in turbidity may be a consequence of the formation of starch-casein or starch-albumin complexes. Simultaneously, the use of a mixture of starch and casein hydrolysate or albumin hydrolysate contributes to higher light transmission or reduced turbidity of the solution, which may be due to the low formation of starch complexes and protein hydrolysates. The obtained results also demonstrate that the hydrolysis of proteins by pepsin of the stomach contributes to a decrease in the number of protein-polysaccharide complexes.

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Entered 09.08.2021