

Effects of Soy protein concentrate and Xanthan gum on physical properties of Silver carp (*Hypophthalmichthys molitrix*) surimi

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Abstract

Studies were conducted to evaluate the effects of soy protein concentrate (SPC) and Xanthan gum (X) on physical properties of surimi prepared from Silver carp (*Hypophthalmichthys molitrix*) using various amount of soy protein concentrate (0, 5 and 10%) and xanthan gum (0, 0.25 and 0.5%) in fish paste during 3 months of frozen storage at -18°C. Results obtained from folding test showed that the gel-forming ability of the gel treated with commercial blend (4% sorbitol+4% sucrose) was higher than the gels containing soy protein concentrate and xanthan gum ($P < 0.05$). Xanthan gum had a harmful effect on gel formation of surimi. The lowest expressible moisture values were reported for the gels containing 10% SPC and commercial blend (4% sorbitol+4% sucrose) that indicated the gels matrix had high water holding capacity ($P < 0.05$). During 3 months of frozen storage at -18°C, gel-forming ability and water holding capacity of surimi were markedly decreased ($P < 0.05$). Results of this studies demonstrated that the samples consisting commercial blend (4% sorbitol+4% sucrose) and 10 % SPC were more acceptable from the viewpoint of water holding capacity and gel forming ability, respectively.

Keywords: Surimi, Soy protein concentrate, Xanthan gum, Gel-forming ability, Expressible moisture

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Introduction

Surimi is the stabilized myofibrillar proteins of fish muscle, that fish was filleted, mechanically deboned, washed to eliminate most of the lipids, blood, enzymes and sarcoplasmic proteins and then stabilized by cryoprotectants for prolong shelf-life (Park, 2005). Surimi has some main functional properties such as gel-forming ability and water holding capacity (WHC) (Benjakul et al., 2003; Li, 2003). The prime point in surimi production is to maintain functionality of the myofibrillar protein to forming an elastic gel. Freezing is too used in surimi industries for processing and prolong preservation. Nevertheless, the denaturation of myofibrillar protein during frozen storage is caused to reduce the functional properties of surimi (Park, 2005). The denaturation results in aggregation of the myofibrillar proteins in which hydrogen bonds, ionic bonds, hydrophobic bonds and possibly disulphide bonds are formed (Hall and Ahmad, 1992). Water has central role in these changes. As the water is frozen solutes are concentrated, that changes the ionic strength and pH in the microenvironment surrounding the proteins, resulting in dehydration and conformational changes (Hall and Ahmad, 1992). To prevent such an undesirable change, or to improve the functionality of frozen meat, various denaturation-inhibiting materials, such as amino acids, beef plasma protein, soybean protein, sucrose, polyphosphate, sorbitol and starch have been used for surimi production (Park and Lanier, 1987; Hossain et al., 2003). Non-meat proteins such as soy protein

isolate can interact directly with meat proteins, which occupy the interstitial spaces in a gel matrix (Lanier, 1991). Soy protein concentrate contains 70% or more protein on a moisture-free basis. Regardless of the process used, soy protein concentrate have fat and water holding properties (Singh et al., 2007). Soy protein isolate has been found to have protective effect against proteolysis (Rawdkuen et al., 2007), Xanthan gum is an extracellular polysaccharide produced from the micro-organism *Xanthomonas campestris*. Xanthan gum is soluble in cold and warm water and solutions exhibit pseudo plastic flow and this gum compared to other gums is the best improvement in water-binding ability of low-fat meat emulsions (Lin and Mei, 2000; Phillips and Williams, 2000;). Perez-Mateos and Montero (2000) studied the effects of locust bean gum, guar gum, xanthan gum, iota-carrageenan, kappa-carrageenan, carboxymethylcellulose and alginate on the rheological properties of blue whiting muscle mince. They reported that the treatment containing xanthan had lower breaking force and hardness and higher elasticity than other treatments. Interactions between protein and polysaccharide play an important role in the structure and stability of many processed foods. Gel-forming ability and water holding capacity of proteins are affected by their interactions with polysaccharides (Tolstoguzov, 1997). More studies are needed for fish protein-hydrocolloid interactions. The most current hydrocolloid-myofibrillar protein interaction studies have been the

interaction of carrageenan and alginate gums with meat products because of the interest in expanding low fat products (Defreitas et al., 1997). There is a lack of data about the effect of xanthan on gelling properties of myofibrillar protein. Studies also indicated that the addition of polysaccharide gums could increase the water binding capacity of soy proteins (Sanchez et al., 1995). Therefore objectives of this study were to investigate the ability of soy protein concentrate and xanthan gum to stabilize Silver carp surimi during 3 months of frozen storage and to compare their effectiveness with that of the commercial blend (4% sorbitol and 4% sucrose).

Materials and methods

Sorbitol crystalline powder was obtained from Merk (Darmstadt, Germany). Xanthan gum (X) was obtained from Provisco AG (Hauptwil, Switzerland). Sucrose (food grade) was purchased in the local department store. Soy protein concentrate (SPC) was obtained from Atame Pars factory. Silver carp (*Hypophthalmichthys molitrix*) were purchased 4-5 h after catching from the local fish market of Anzali port in the southern Caspian Sea. The fish had an average weight of 700g. They were ice packed for up to 8 h before processing (immediately after purchase). Then transported to the laboratory at the

National Fish Processing Technology Research Center at Anzali port in Iran. The fish were beheaded, gutted, washed and then filleted. Skin and bones were removed by deboning machine (Sepmatic Deboner, Germany). The minced meat was then washed with cold fresh water (5°C) containing 0.25 % common salt (NaCl) at a mince/water ratio of 1: 3 (w/w). The mixture was stirred gently for 5 minutes and washed minced meat was squeezed manually with silk mesh. The washing process was repeated three times and the washed minced meat was added with soy protein concentrate (0, 5 and 10 %), xanthan gum (0, 0.25 and 0.5 %) and 8 % sorbitol / sucrose (1:1) in a cold room. According to the last performed studies in this theme we choose these values for soy protein concentrate and xanthan gum (Perez-Mateos and Montero, 2000; Lonescu et al., 2003; Luo et al., 2004). Surimi without cryoprotectant was used as the control. Treatments are shown in Table 1.

After mixing, each treatment was packed into polyethylene bags (100 g), frozen down to -18 °C and stored at the same temperature for 3 months. Similar to the conditions observed in the researches of Carvajal et al. (1999) and Sych et al. (1990a) on frozen storage of Alaska Pollock and Ling cod surimi, which were based on a time period of 3 months at -20 °C.

Table 1: Cryoprotectant used in this study

treatment ^a	
1	Control
2	Sorbitol 4% / Sucrose 4%
3	SPC 5%
4	SPC 10%
5	X 0.25%
6	X 0.5 %
7	SPC 5% + X 0.25%
8	SPC 5% + X 0.5 %
9	SPC 10% + X 0.25%
10	SPC 10% + X 0.5 %

^a Treatment 1 was the control formulation containing no cryoprotectant and treatment 2 contained commercial mix cryoprotectant. Treatments 3-10 were alone or blends formulated of soy protein concentrate and xanthan gum.

Every one-month intervals for 3 months, the samples were removed, thawed 4-5 h before analyses in the cold room at 4°C and analyses were carried out for folding test and expressible moisture. Moisture, crude protein, fat and ash contents were determined according to the method described by Association of Official Analytical Chemists, AOAC (1990). The crude protein and lipid contents were determined by the Kjeldhal and Soxhlet methods, respectively. Frozen surimi (50 g) was tempered at 5°C for 1 h, cut into small pieces and then 3 % NaCl and 30% chilled water was added in a mixer. Paste was stuffed into the polyethylene tube was heated at 40°C for

15 minutes (step-1) prior to heating at 85°C for 15 minutes (step-2) in the water bath. Then the gels were cooled immediately in ice water and were stored at 4°C until assessment of gel properties.

The gel strength of surimi was determined according to the folding test by Poon et al. (1981). The gel was removed from the tube and subjected to folding test for physical measurements.

For folding test a spherical disc of 1mm thick gel was cut off and held with thumb and index fingers while folding it first into 2 halves then quarter. The gel was graded using scores presented in Table 2 as suggested by Poon et al. (1981).

Table 2: Grade system used in the folding test of the gel

Grade	Results on folding
AA	No crack visible when disc is folded into quarter
A	No crack when disc is folded into half but one or more cracks or breaks are visible when folded into quarter
B	One or more cracks are visible when disc is folded into half
C	Breaks, but does not split into half
D	Splits into halves when folded into half
O	Sample too soft to evaluate

Expressible moisture was measured according to the method of Ng (1987). Gel samples were cut into a thickness of 5 mm, weighed (X) and placed between two pieces of filter paper at the bottom and one piece on the top of the sample. The standard weight (5kg) was placed at the top and held for 2 minutes. The samples weighed again (Y) after removing from the papers. Expressible moisture was calculated using the following equation:

Expressible moisture (%) = $[(X - Y) / X] \times 100$. The experiment was repeated three

times. Data were analyzed by One-way ANOVA analysis of variance and Duncan's multiple range tests to compare the differences among means. Significance was defined at $P < 0.05$. SPSS version 16.0 was used for statistical analysis.

Results

The moisture, cured protein, fat and ash contents of all treatments are shown in Table 3. The moisture content of the surimi without any added cryoprotectant (control) was 84.6%.

Table 3: Moisture, crude protein, fat and ash contents of surimi

Treatments	Moisture (%)	Crude Protein (%)	Fat (%)	Ash (%)
Control	84.6 ± 0.707 ^d	12.7 ± 0.282 ^{ab}	1.25 ± 0.0707 ^{ab}	0.4 ± 0.000 ^a
Sorbitol 4% / Sucrose 4%	79.05 ± 1.202 ^{abc}	13.25 ± 0.070 ^b	1.7 ± 0.000 ^d	0.4 ± 0.000 ^a
SPC 5%	81.4 ± 1.272 ^c	16.85 ± 0.494 ^c	1.45 ± 0.0707 ^c	0.55 ± 0.0707 ^b
SPC 10%	78.55 ± 0.919 ^{ab}	15.9 ± 0.424 ^c	1.45 ± 0.0707 ^c	0.75 ± 0.0707 ^d
X 0.25%	84.6 ± 1.272 ^d	11.45 ± 0.212 ^a	1.15 ± 0.0707 ^a	0.45 ± 0.0707 ^a
X 0.5 %	84.45 ± 1.202 ^d	12.95 ± 0.777 ^{ab}	1.15 ± 0.0707 ^a	0.4 ± 0.000 ^a
SPC 5% + X 0.25%	81.15 ± 1.202 ^{bc}	16.55 ± 1.343 ^c	1.35 ± 0.0707 ^{bc}	0.6 ± 0.000 ^c
SPC 5% + X 0.5 %	81.25 ± 1.202 ^c	17.1 ± 1.310 ^c	1.45 ± 0.0707 ^c	0.6 ± 0.000 ^c
SPC 10% + X 0.25%	78.45 ± 1.060 ^a	15.95 ± 0.070 ^c	1.45 ± 0.0707 ^c	0.75 ± 0.0707 ^d
SPC 10% + X 0.5 %	78.15 ± 1.060 ^a	16.15 ± 0.070 ^c	1.45 ± 0.0707 ^c	0.75 ± 0.0707 ^d

Different superscripts within a column indicate significant difference ($P < 0.05$)

The moisture content of treatments containing 0.25% X, 0.5 % X was close to the control moisture level of 84.6 % and statistical analysis showed that there was significant difference ($P < 0.05$) between

these treatments with others. The lowest moisture contents were reported for the treatments containing 4% sorbitol+ 4% sucrose, 10% SPC, 10 % SPC + 0.25 % X and 10 % SPC + 0.5 % X ($P < 0.05$). SPC

has high protein content (72.75%) so the protein contents of the treatments containing SPC were higher than the control and other treatments ($P < 0.05$). The results indicated that moisture content decreased by increasing of protein content. Soy protein concentrate increased the ash content of surimi ($P < 0.05$) because of the ash content of SPC in which was higher than the surimi. The surimi treated with 4% sorbitol+4% sucrose had the highest fat content ($P < 0.05$).

Effect of SPC and xanthan gum on gel-forming ability of surimi

The results of folding test are shown in Table 4. The highest folding test grade AA found with the gel treated with 4% sorbitol+4% sucrose. The folding test

grades of the treatments containing 5 % SPC and control in the first, second and third months were B, C and D, respectively and grades of the surimi treated with 10% SPC were close to the 5% SPC and control in the second and the third months but in the first month was lower than the grades of these treatments. Xanthan produced very soft gel when used alone. Gel-forming ability and gel strength decreased as a result of the increase of xanthan amount in the X/SPC ratio, as the treatments containing 0.5% X+5% SPC produced soft gel in 3 months, 0.25% X+5% SPC and 0.5% X+10% SPC produced soft gel in the second and the third months and 0.25% X+10% SPC produced soft gel just in the third month.

Table 4: Effect of different levels of soy protein concentrate and xanthan gum on gel-forming ability of surimi

Folding test			
Treatment	First month	Second month	Third month
Control	B	C	D
Sorbitol 4% / Sucrose 4%	AA	AA	A
SPC 5%	B	C	D
SPC 10%	C	C	D
X 0.25%	O	O	O
X 0.5 %	O	O	O
SPC 5% + X 0.25%	D	O	O
SPC 5% + X 0.5 %	O	O	O
SPC 10% + X 0.25%	D	D	O
SPC 10% + X 0.5 %	D	O	O

During 3 months of frozen storage at -18°C , gel-forming ability decreased markedly ($P < 0.05$).

Effect of SPC and xanthan gum on expressible moisture of surimi

Expressible moistures of surimi gels added with SPC and xanthan gum at different levels are shown in Table 5.

Table 5: Effect of different levels of soy protein concentrate and xanthan gum on the expressible moisture of the surimi

Expressible moisture			
Treatment	First month	Second month	Third month
Control	15.090 ± 0.575 ^e	25.417 ± 1.116 ^e	40.555 ± 5.538 ^c
Sorbitol 4% / Sucrose 4%	5.913 ± 0.845 ^a	7.053 ± 0.390 ^a	25.076 ± 1.406 ^a
SPC 5%	10.260 ± 0.396 ^d	22.763 ± 1.191 ^d	34.589 ± 1.471 ^b
SPC 10%	5.191 ± 0.129 ^a	14.884 ± 0.700 ^c	23.845 ± 1.148 ^a
X 0.25%	n.d.	n.d.	n.d.
X 0.5 %	n.d.	n.d.	n.d.
SPC 5% + X 0.25%	9.798 ± 0.091 ^{cd}	n.d.	n.d.
SPC 5% + X 0.5 %	n.d.	n.d.	n.d.
SPC 10% + X 0.25%	7.597 ± 0.127 ^b	10.820 ± 1.098 ^b	n.d.
SPC 10% + X 0.5 %	9.428 ± 0.323 ^c	n.d.	n.d.

Different superscripts within a column indicate significant difference ($P < 0.05$)

n.d. = not determined because of gel was very soft.

The treatments contain 0.5% X+5% SPC could produce soft gel in 3 months, 0.25% X+5% SPC and 0.5% X+10% SPC produced soft gel in the second and the third months and 0.25% X+10% SPC produced soft gel just in the third month. So we could not measure the expressible moisture of these treatments.

In the first, second and the third months of storage expressible moisture of the treatments containing 5 and 10% SPC were lower than the control significantly ($P < 0.05$), because the free water decreased in the gel by increasing of protein content thereupon expressible moisture decreased and reduction of the expressible moisture of the treatment containing 10% SPC significantly was higher than the 5% SPC.

The treatment containing 5% SPC had higher expressible moisture than the treatment containing 4% sorbitol+4% sucrose but in the first and the third months of storage the expressible moisture of the treatment containing 10% SPC didn't differ with the treatment containing 4% sorbitol+4% sucrose.

Thereupon the lowest expressible moisture values were reported for the gels containing and 10% SPC ($P < 0.05$) and the surimi without a cryoprotectant had the highest expressible moisture that indicated the water holding capacity of gel matrix was low ($P < 0.05$). The effects of time storage on the expressible moisture of the surimi containing SPC and xanthan gum are shown in Table 6.

Table 6: Effect of time storage on the expressible moisture of the surimi containing soy protein concentrate and xanthan gum

Expressible moisture			
Treatment	First month	Second month	Third month
Control	15.090 ± 0.575 ^a	25.417 ± 1.116 ^b	40.555 ± 5.538 ^c
Sorbitol 4% / Sucrose 4%	5.913 ± 0.845 ^a	7.053 ± 0.390 ^a	25.076 ± 1.406 ^b
SPC 5%	10.26 ± 0.396 ^a	22.763 ± 1.191 ^b	34.589 ± 1.471 ^c
SPC 10%	5.191 ± 0.129 ^a	14.884 ± 0.700 ^b	23.845 ± 1.148 ^c
X 0.25%	n.d.	n.d.	n.d.
X 0.5 %	n.d.	n.d.	n.d.
SPC 5% + X 0.25%	9.798 ± 0.091 ^{cd}	n.d.	n.d.
SPC 5% + X 0.5 %	n.d.	n.d.	n.d.
SPC 10% + X 0.25%	7.597 ± 0.127 ^b	10.820 ± 1.098 ^b	n.d.
SPC 10% + X 0.5 %	9.428 ± 0.323 ^c	n.d.	n.d.

Different superscripts within a column indicate significant difference ($P < 0.05$) n.d. = not determined because of gel was very soft.

During 3 months of frozen storage at -18 °C, expressible moisture increased significantly ($P < 0.05$). The significant point is that increasing expressible moisture procedure of the treatments containing 5 and 10% SPC was equal in the 3 months of storage but expressible moisture of the treatment containing 4% sorbitol+ 4% sucrose was sharply increased from the second month to the third month.

Discussion

The moisture content of the surimi without any added cryoprotectant (control) was 84.6% that suggesting good dewatering properties and meeting the standard that industrial surimi before the using of the cryoprotectants have less than 85% moisture (Lee, 1985; Sych et al., 1990b).

The moisture content is one of the critical factors in the surimi production, because it increases with increasing of matrix free water, so protein denaturation increased by ice crystallization (Uddin et al., 2006). The results indicated that xanthan (0.25% and 0.5%) wasn't suitable cryoprotectant for surimi prepared from Silver carp. According to Rawdkuen et al. (2007), proteolysis of Sardine surimi can be deferred by addition of soy protein isolate (1, 2 and 3%), leading to increased gel forming ability. Luo et al. (2004) reported that the addition of 10% soy protein isolate increased the breaking force and the distance of bighead surimi but values of these factors decreased when the soy protein isolate was increased from 10% to 40%. But in this study, soy protein concentrate didn't affect the gel forming

ability of surimi. The obtained results have shown that xanthan gum alone has a negative effect on the gel strength of surimi gel and produces very soft gel. Xanthan gum is an anionic polysaccharide, which has been reported to able to interact with proteins have a cationic charge (below their isoelectric point). Electrostatic interaction is considered responsible of the protein-hydrocolloid interactions under these conditions (Miller, 1994). The negative effect of xanthan gum on the gel strength of surimi gel might be attributed to its anionic nature. Myofibrillar proteins are above the isoelectric point when processed as surimi. Therefore, myofibrillar proteins of surimi charged negatively. Both macromolecules being of anionic nature, a repulsive force could responsible for antagonistic effect. Xiong and Blachard (1993) expressed that the breaking force of the gel made from soluble chicken proteins decreased by addition of xanthan gum, they derived that it was because of the polysaccharides interfered with the protein gel structure (Perez- Mateos and Montero, 2000). Various studies reported that during frozen storage the myofibrillar proteins of fish muscle aggregated into heavy polymers (Yoon and Lee, 1990). During storage at -18 °C , several changes befall in fish muscle proteins such as denaturation, ice crystallization, dehydration and changes in intermolecular conformation, such as salt-soluble protein, pH, ionic-strength (Park, 1994). Gel-forming ability, water holding capacity of surimi decreased

sharply when proteins deteriorated during frozen storage (Hsu, 1990; Yoon and Lee, 1990). The results from this test have shown that Gel-forming ability decreased during frozen storage because of myofibrillar protein denaturation.

The results of folding test indicated that the combination blend (4% sorbitol+4% sucrose) was more successful than SPC and xanthan to decreased denaturation of surimi prepared from Silver carp. The increased expressible moisture of the treatments is suggesting the decreased water holding capacity of gel matrix. Nature of actin and myosin changed by increasing of frozen storage period, leading to increase the free water in matrix. So water holding capacity declined and when expressible moisture measured much water leaved from matrix (Rawdkuen et al., 2009). There is a relation between expressible moisture and protease activity, as the expressible moisture increased by increasing of protease activity. Bigelow and Lee (2007) investigated the cryoprotective effect of SPI (5%, 10%) and alginate (0.75%, 1.5%) in combination with sorbitol and sodium tripolyphosphate on frozen Red hake muscle. They found that the muscle comprising of 10% SPI and 1.5% alginate had the lowest expressible moisture after 2 months of frozen storage. Pietrasik et al. (2007) expressed that non-meat proteins such as soy protein isolate reduced the percentage of water out go from gels after pressing the gel samples, indicating that they improved water holding capacity of

pork. In this research, soy protein concentrate decreased the expressible moisture, especially in 10% value.

A number of studies have been reported that expressible moisture of surimi decreased during frozen storage (Siah et al., 1998; Siddaiah et al., 2001). A decreasing trend was also observed in this study because of the protein denaturation during frozen storage. Several scientists reported that soy protein isolate increase gel strength in low fat products that was associated to its gel-forming and water retention ability and high resistant to denaturation (more than meat proteins) (Lin and Mei, 2000; Shand, 2000).

Luo et al. (2004) indicated that the addition of SPI to Bighead carp surimi decreased the protein concentrate by increasing SPI concentration, and retarded the hydrophobic interactions and disulfide bridge formation among the myofibrillar proteins. Thereupon the development of modori phenomena of Bighead carp surimi was reduced. Ionescu et al. (2003) reported that xanthan gum and starch improved the texture properties of the myofibrillar proteins concentrate when the protein gels are formed. The results from the expressible moisture test have shown that the treatments containing 4% sorbitol+4% sucrose and 10% SPC were better than other treatments because of their water holding capacity were higher than others.

From the present study, it can be concluded that the surimi prepared from Silver carp using 10% SPC has better water holding capacity than the control and

when using 4% sorbitol+4% sucrose. So we can use the surimi containing 10% SPC in the fishery products such as fish burger, fish finger, sausage and etc. The gel forming ability of surimi containing SPC and xanthan was lower than commercial blend (4% sorbitol+4% sucrose). Xanthan gum showed a harmful effect on gel formation of surimi.

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بررسی اثر کنسانتره پروتئین سویا و صمغ زانتان بر خواص فیزیکی سوریمی تهیه شده از ماهی کپور نقره‌ای (*Hypophthalmichthys molitrix*)

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چکیده

در این تحقیق اثر کنسانتره پروتئین سویا و صمغ زانتان بر خواص فیزیکی سوریمی تهیه شده از ماهی کپور نقره‌ای مورد بررسی قرار گرفت. کنسانتره پروتئین سویا به میزان (۰، ۵ و ۱۰ درصد) و صمغ زانتان به میزان (۰، ۲۵ و ۵۰ درصد) به سوریمی افزوده شد، سپس به مدت ۳ ماه در سردخانه ۱۸- درجه سانتی گراد نگهداری شد و در هر ماه آزمون‌های تا شدن و رطوبت تحت فشار انجام گرفتند. نتایج به دست آمده از آزمون تا شدن نشان داد که صمغ زانتان اثر منفی بر قابلیت تشکیل ژل سوریمی دارد و قابلیت تشکیل ژل حاوی سوربیتول/ ساکارز که به صورت متداول در تولید سوریمی استفاده می‌شود، به طور معنی داری بیشتر از نمونه حاوی کنسانتره پروتئین سویا بود. نمونه حاوی ۱۰ درصد کنسانتره پروتئین سویا و نمونه حاوی سوربیتول/ ساکارز به طور معنی داری کمترین رطوبت تحت فشار را داشتند که بیانگر بالا بودن ظرفیت نگهداری آب ژل می‌باشد. در تمام تیمارها کیفیت بافت سوریمی در مدت سه ماه نگهداری در ۱۸- درجه سانتی گراد به طور معنی داری کاهش یافت. در کل از نظر قابلیت تشکیل ژل نمونه حاوی سوربیتول/ ساکارز و از نظر رطوبت تحت فشار نمونه حاوی ۱۰ درصد کنسانتره پروتئین سویا مطلوب‌ترین تیمارها بودند.

واژگان کلیدی: سوریمی، کنسانتره پروتئین سویا، صمغ زانتان، قابلیت تشکیل ژل، ظرفیت نگهداری آب

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