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RESEARCH ON OBTAINING, CHARACTERIZATION AND USE OF EDIBLE FILMS IN FOOD INDUSTRY

Floricel CERCEL[1], Mariana STROIU[1], Daniela IANIȚCHI[2], Petru ALEXE[1]

[1] "Dunarea de Jos" University of Galati, Faculty of Food Science and Engineering,
111 Domneasca Street, 800201, Galati, Romania
[2]University of Agronomic Sciences and Veterinary Medicine of Bucharest,
59 Mărăști Blvd., District 1, 011464, Bucharest, Romania

Corresponding author email: dianitchi@yahoo.com

Abstract

This review study aimed to give information about the use of plant extracts in meat product processing as antimicrobial and antioxidant agent. Microbial spoilage and lipid oxidation are the major causes of the deterioration and reduction of shelf-life in meat products. Lipid oxidation in meat products results in formation of off-flavors and undesirable chemical compounds such as aldehydes, ketones, alcohols and hydrocarbons. Growth of microorganisms in meat products causes not only microbial spoilage but also development off oodborne diseases. To inhibit lipid oxidation and growth of microorganisms, especially pathogenic microorganisms in meat products, several preservation techniques, such as pasteurization, reduction of water activity (salting, drying, freezing etc.), acidification, fermentation, synthetic and natural antimicrobial and antioxidant additives have been used in meat industry. Many synthetic and natural food additives such as butylated hydroxytoluene (BHT), butylated hydroxyanisole (BHA), propyl gallate, α-tocopherol, nisinand organic acidsare commonly used in the meat industry to inhibit or delay the oxidation process and reduce the microbial growth. In recent years, consumer demands for natural food additives have increased because of negative and toxic effects of synthetic food additives on human health. Herbs, spices, fruits and vegetables, and their powders, oils and extracts have been reported to be a good source of various phenolic compounds, such as flavonoids, terpenoids, carotenoids, could therefore be incorporated in meat products as a source of natural antioxidants and antimicrobials to extend shelf-life and safety of meat products.

Key words*: edible films, myofibrillar proteins, water vapor permeability, moisture content, color of the films*.

INTRODUCTION

The films are products obtained from food biopolymers and additives having food purity. Film-forming biopolymers can be proteins, polysaccharides and lipids.

The preparation of edible films and edible membranes, based on muscle protein, can be accomplished using:

a) *myofibrillar protein concentrates*:
- fish surimi (Cuq et al., 1995, 1997; Monterrey-Q, 1988);
- beef surimi (Souza et al., 1997);
- surimi from mechanically deboned poultry, heart muscle (Ionescu et al., 2008). The following categories of surimi can be used: wet surimi, frozen or thawed surimi, surimi dried by lyophilisation (Monterrey-Q, 1998) or surimi dried in the air (Cuq et al., 1997d);

b) *sarcoplsamatic proteins* (Iwata, 2001; Tanaka et al., 2001). Sarcoplasmatic proteins, unlike the myofibrillar proteins, are globular proteins that require an initial heat denaturation to form a continuous matrix (Iwata et al., 2000). By the heat treatment, the globular protein structure is changed, causing an exposure of the -SH groups, and concequently -S-S- links are produced between the adjacent protein chains and also hydrophobic interactions are occuring (Perez-Gogo and Krochta, 2001; Sobral et al., 2004, 2005; Garcia and Sobral, 2005);

c) *fish muscle* that contains both myofibrillar and sarcoplasmatic proteins (Nile Tilapia, Paschoalich et al., 2003).

MATERIALS AND METHODS

Bighead carp was procured fresh from the local fish store.

The fish was transported to the laboratory in a cool bag and then stored at 4°C until processing.

Determining the approximate chemical composition

The contents of water, protein, fat and ash were determined using standard method of analysis (AOAC, 1990; Ionescu et al., 1992). Also, moisture was determined by fast drying to constant weight using the thermobalance "Precisa XM 60" Total nitrogen was determined by Kjeldahl semimicro method, mineralization being performed in the "Trade Raypa" facility. Total proteins were calculated by multiplying the total nitrogen content by a factor of 6.25. All chemical analyzes were carried out in duplicate.

The pH was measured potentiometrically, using the pH meter type "Hanna" using protein dispersions with a concentration of 10% (G/V)), at a temperature of 22 ± 1^{0}C.

Samples were ran in duplicate.

The formation of biodegradable/edible films

In order to obtain the protein films, two methods are used:

- *the solvent process* involves the protein dispersion or solubilization in the film-forming solution. This procedure has been extensively studied and applied to produce edible / biodegradable films and membranes from diferent proteins and, in particular myofibrillar proteins (Cuq et al., 1995, 1998; Monterrey-Q, 1988);
- *dry process* is based on the thermoplasmatic properties of the proteins to a low water content (Hernandez-Izquierdo et al., 2008; De Graaf, 2000).

Properties of edible films and coatings based on proteins

Films solubility

The proteins with high molecular weight are generally insoluble in water and thus have a high potential to form water-resistant films (Cuq et al., 1998). Protein films do not lose integrity after 24 hours of immersion in water (Cuq et al., 1998b). Plasticizers used (sorbitol, glycerin or sucrose) in the manufacture of protein-based films increase the content of dry substance soluble in water. In general, hydrophilic plasticizers improve the solubility, which increases when the levels of added plasticizers is increased. Monterey-Q (1998) reported that a significant part of glycerol remains insoluble in water, suggesting the production of protein-glycine interactions. Protein monomers and low molecular weight peptides, formed during the conditioning of film forming solutions and immobilized in the network, may be water-soluble protein components (Cuq et al., 1995).

Water vapor permeability

The values of water vapor permeability of films based on fish myofibrillar proteins are bigger (3.8-3.9x10^{-12}mol.m/m^2sPa) compared to synthetic films: cellulose acetate (0.28 - 0.90x10^{-12}mol.m/m^2sPa), high density polyethylene (0.014 x 10^{-12} mol.m/m^2sPa) and low density polyethylene (0.04-0.054 x 10^{-12} mol.m/m^2sPa), but lower than in case of films based on corn zein (6.5 x 10^{-12} mol.m/m^2sPa) or soybean proteins (194x10^{-12} mol.m/m^2sPa). Water vapor permeability of protein based films is limited due to the inherent hydrophobicity of the proteins. Hydrophilic plasticizers, such as glycerin, facilitate the transfer of water vapor through protein based films.

RESULTS AND DISCUSSIONS

In our experiment, edible / biodegradable films were created using bighead carp myofibrillar proteins.

Bighead carp myofibrillar proteins were obtained by the conventional procedure of repeated washing with cold water of the minced meat, followed by centrifugation and refining to remove water, water-soluble substances, sarcoplasmatic proteins, lipids, skin residues and bones (surimi procedure).

Table 1. Film-forming solutions composition

Film	pH solution	Protein, g%	Glycerine, %	Gelatine, g%	Cyclodextrin, g%
a	2.7	1.0	50	-	-
b	2.7	1.5	50	-	-
c	2.7	2.0	50	-	-
d	2.7	2.5	50	-	-
e	2.7	2.0	30	-	-
f	2.7	2.0	70	-	-
g	2.7	1.0	30	2.0	-
h	2.7	1.5	40	-	1.0

Several film types were made using different percentages of fish myofibrillar protein, different levels of glycerin in strongly acidic medium. In addition, composite films were obtained by adding gelatin or cyclodextrins, together with the basic constituents (protein, glycerine, water). The compositions of film

forming solutions (FFS) are indicated in Table 1. Film types were designated with letters.

The approximate composition of bighead carp myofibrillar proteins

The approximate chemical composition of myofibrillar fish proteins is shown in Table 2. According to Sikorski (1981), the protein isolate obtained by the conventional procedure contains myosin as main protein, which represents 50-60% of the myofibrillar proteins.

Table 2. Approximate composition of bighead myofibrillar proteins

Constituent	Quantity	
	Wet weight, g%	Dry substance, g/100 g s.u.
Water content	83.82829	
Total protein	14.68942	90.83406
Lipids	0.1951	1.206428
Ash	0.1698	1.049982

Films appearance

All the films we obtained were transparent, flexible and uniform. The films had smooth surfaces, without pores or cracks visible to the naked eye. When gelatine and cyclodextrin were used for film formulations, they showed a slightly yellowish color compared to the films based only on fish proteins.

The appearance of the two parts of the film was slightly different for all films made. The lower part of the film that came into contact with the casting plate was brighter, while the top part of the film was dull, possibly due to phase separation that occurs in solution during the drying process.

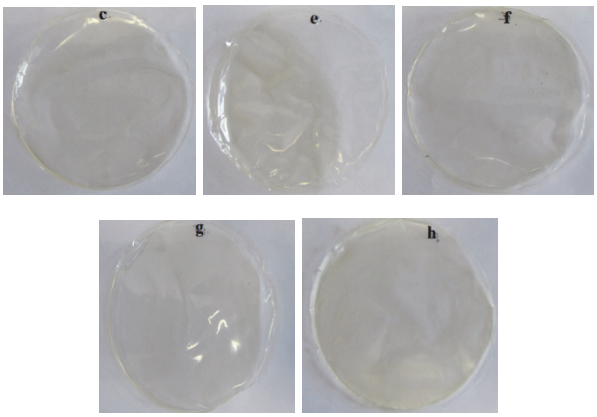

Figure 1. Edible/biodegradable films

All kinds of films we obtained were easily detached manually from the pouring plates, except for the films with a higher addition of glycerol (70 g per 100 g protein), these being slightly sticky.

Film thickness

The film thickness was measured using the micrometer in 10 randomly chosen areas. Film thickness measurement accuracy was ±5%. For each type of film, there were made 8 films, their thickness being measured after drying and conditioning. The average thickness and standard errors of the films made for this paper are shown in Table 3. In case of film-forming solutions at pH 2.7, in Figures 2-5 are shown the average thickness variations based on the type of film, on the region of measurement, on the protein concentration of the film forming solution, on the level of glycerin added and on the addition of gelatin or cyclodextrin. As it can be seen, the films showed some nonuniformity of thickness, depending on the areas where the measurements were made, the thickness differences were not statistically significant ($p<0.05$) (Figure 2). The average thickness of the films increased from 0.030±0.001 mm to 0.067±0.002 mm with the increase of fish myofibrillar protein level of from 1% to 2.5%, in the presence of 50% glycerol per 100 g protein (Figure 3). The average thickness of the films was strongly positively correlated with the levels of fish myofibrillar protein from the film forming solutions, Pearson correlation coefficient is 0.94.

Table 3. Thickness of the films basen on bighead carp myofibrillar proteins

Films	Film thickness, mm	Films	Film thickness, mm
a	0.030±0.001	i	0.066±0.03
b	0.052±0.001	j	0.098±0.09
c	0.066±0.002	k	0.100±0.04
d	0.067±0.002	l	0.061±0.03
e	0.066±0.002	m	0.088±0.04
f	0.075±.002	n	0.041±0.04
g	0.055±0.001	o	0.082±0.04
h	0.061±0.003	p	0.105±0.05

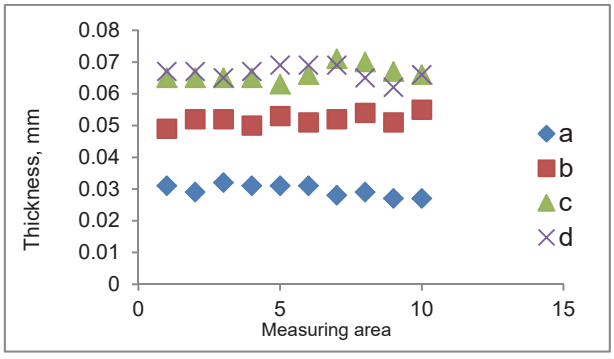

Figure 2. The variation of film thickness based on bighead carp proteins depending on the measuring location

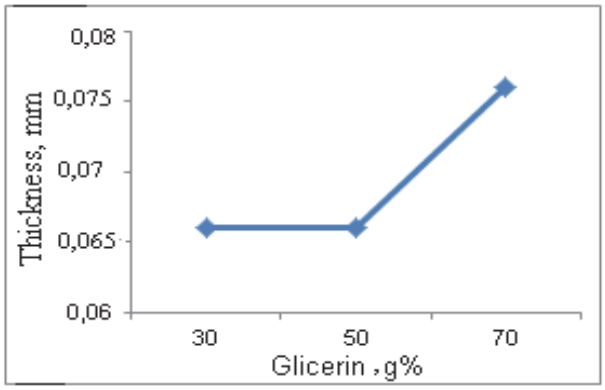

Figure 3. The influence of the protein concentration on the film thickness (pH 2.7)

In case of using various levels of glycerol, the average thickness of the 8 films depending on the measurement areas, varied within the limits of 0.061 to 0.078 mm (Figure 4). The highest average thickness was found in films prepared with an addition of 70 g glycerin / 100 g protein (0.075 ± 0.002) mm. At a level of 2% of protein there ware no differences in thickness depending on the level of glycerol, when it was within the range 30-50% compared with the protein (Figure 5).

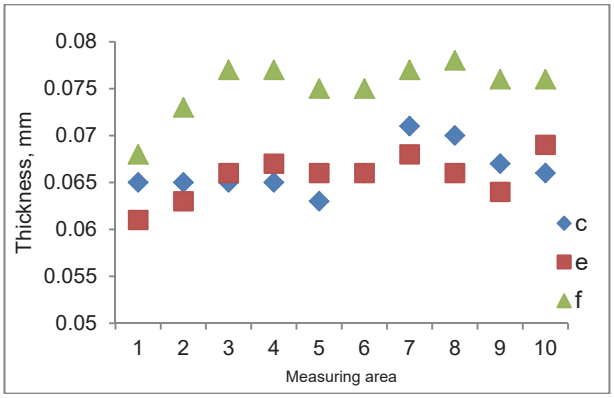

Figure 4. Film thickness depending on the addition of glyceroland on the measuring area (pH 2.7)

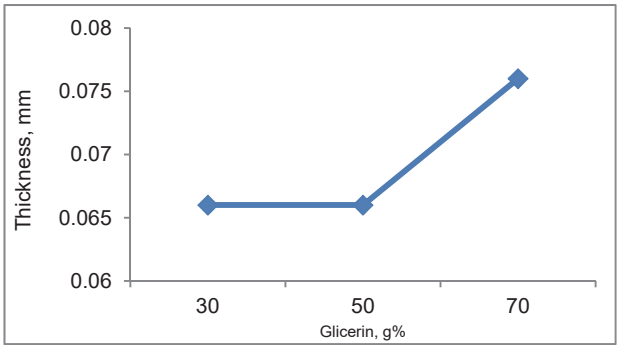

Figure 5. Film thickness variation depending on the level of glycerin (pH 2.7)

The water content, water solubility and water vapor permeability of the films

The water content and water solubility

The contents of water and the water solubility of the films based on fish myofibrillar proteins (FMP) shown in Table 4, varied depending on the type of film, within the limits of 19.94 ± 0.09 and 30.80 ± 0.09%, respectively 12.65 ± 0.08 and 24.58 ± 0.06%. The lowest water content was found on the film with 1% cyclodextrin addition (film h), and the biggest on the film with 2.5% fish myofibrillar protein content (film d). The lowest solubility in water was found in case of film d, and the highest solubility corresponded to the film with 2% protein, pH 2.7 and 30% glycerol / 100 g protein (film e). The water content and films solubility depended on the compositions of films forming solutions (FFS) (Figures 6, 7).

In the case of solutions with pH 2.7, the water content correlated poorly positive with the level of protein (r = 0.199), and negatively with the solubility (r = -0.465). We found a strong negative correlation between water solubility and moisture content of the films (r = -0.959). By increasing the protein content, polymer networks more dense and more resistant to water are formed. It is possible that at strongly acid pH, the fish myofibrillar proteins to undergo some structural changes that influence physical parameters of the films.

Table 4. The water content, water solubility and water vapor permeability of the films

Film type	Water content, g%	Solubility, g%	Permeability X 10^{-10}g m^{-1} s^{-1}Pa^{-1}
a	29.31±0.08	19.35±0.03	0.41±0.007
b	26.11±0.08	18.23±0.1	0.71±0.050
c	25.81±0.07	16.55±0.05	0.81±0.003
d	30.63±0.09	12.65±0.08	0.89±0.020
e	27.72±0.05	17.21±0.06	0.72±0.003
f	28.76±0.06	17.88±0.06	0.96±0.003
g	29.09±0.1	21.32±0.03	0.43±0.013
h	19.94±0.09	22.51±0.05	0.46±0.003

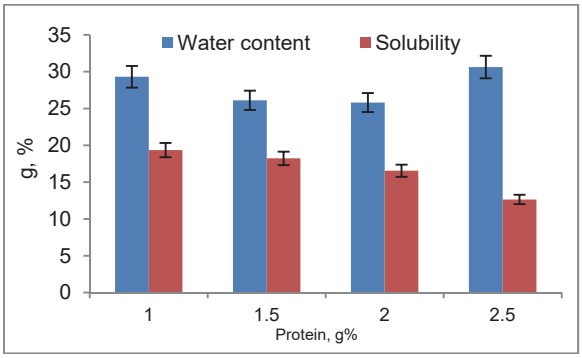

Figure 6. Content of the film forming solution with pH 2.7

The values obtained for moisture content (MC) and for the solubility in water (SW), depending on the level of glycerin and on the pH are shown graphically in Figure 7. For both values of pH, an increase of glycerin content from 30 to 70% (g/100 g protein) resulted in a significant increase in the moisture content and in the amount of solubilized dry substance, in particular for the addition of glycerin > 50%.

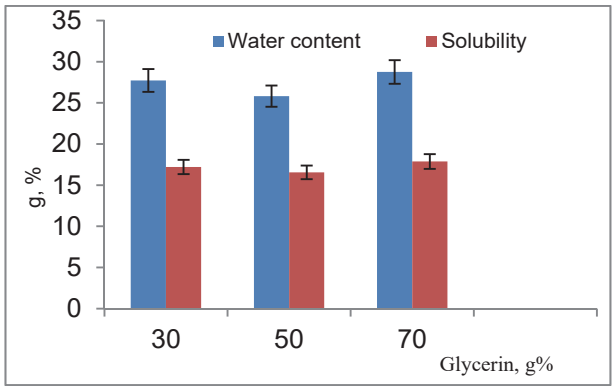

Figure 7. The variation of the water content and solubility depending on the level of glycerin (pH 2.7)

The solubility in water (SW) is regarded as an indicator of edible/biodegradable films resistance in water, which is an important factor in the prepackaging of food products because of the high water activity and the possibility of contamination in the presence of water (Bourtoom and Chinnan, 2005). In general, higher solubility indicates less resistance to water. The solubility in water for the films made by us, was similar to other research reports or lower. Thus, Wengo et al. (2007) reported for films based on Alaska Pollack surimi a solubility in water of 21%; Rostamzad et al. (2015) reported a solubility in water of 19.1% for films made from myofibrillar proteins from silver carp (*Hypophthalmichthys molitrix*); Tao et al. (2015) have found a value for solubility of 31.42 ± 0.89% for the films consisting of silver carp surimi. We consider, the same as Orliac et al. (2002) and Artharn et al. (2007) that cross linked proteins from the film were insoluble, whereas the most part of glycerol has been released into the water. The films based on fish myofibrillar proteins were stabilized by different links, which mainly include intermolecular disulphitic covalent links causing a lower solubility (Chinabhark et al., 2007).

Lower solubility in water for films made in strongly acidic medium can be justified by the fact that at pH <3.0 the protein degradation is more pronounced and Maillard reactions are favored, which would lead to the formation of strong cross links, stabilized by covalent links.
In conclusion, we can appreciate that the bighead carp myofibrillar proteins, obtained by the surimi procedure, led to the formation of stable networks and that only low molecular weight hydrophilic substances were soluble in water.

Water vapor permeability

Water vapor permeability (PVA) is another important and widely studied property of the biodegradable/edible flexible films. This property of the protective films covers their ability to preserve, as appropriate, a dry product (chips, pretzels, candies) or a wet product (cheese, muffins, chewing gum). Without proper protective packaging the products can lose or gain moisture until the relative equilibrium to the environment humidity is achieved, this resulting in products consistency and stability modification.
Water vapor permeability values for films based on fish muscle proteins listed in Table 4. shows the dependence of these values on several factors such as, the composition of film-forming solutions, the concentration of fish myofibrillar proteins, the level of added glycerin.The lowest value for permeability ($0.41\ gm^{-1}s^{-1}Pa^{-1}$) was found on the film containing 1% proteins, 30 g glycerin/100 g protein, and pH 2.7 (film a).
The variations of permeability for the films we tested over time are shown in Figures 8 and 9. Relatively high values of PVA indicate that films based on fish muscle proteins (bighead carp) are poor barriers to water vapor, due to the hydrophilic/hydrophobic nature of the polymer from the film matrix. As can be seen in Figure 9, film permeability values varied with the concentration of the protein, the highest values of PVA corresponding to the level of 2.5% proteins, both for acid environment (pH 2.7). This finding may be explained by the fact that the fish myofibrillar proteins contain significant levels of amino acids with polar nature, such as aspartic acid, glutamic acid, arginine and lysine (Shahidi,

1994; Paschoalick et al., 2003; Tongnuanchan et al., 2011).

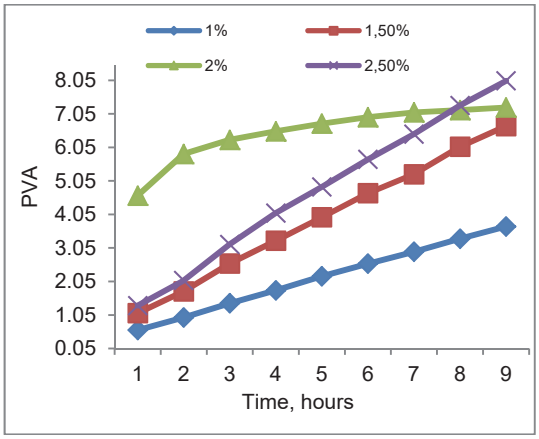

Figure 8. The variation of water vapor permeability of the films depending on the time and on the level of protein (pH 2.7)

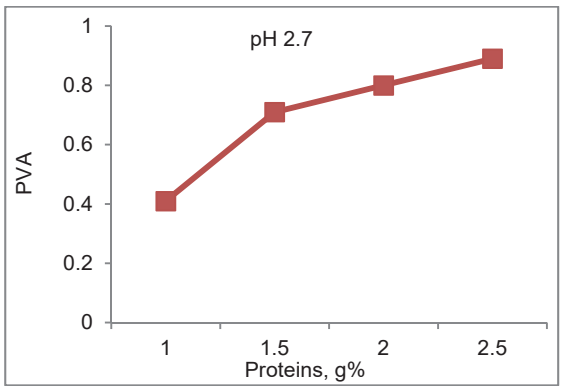

Figure 9. Water vapor permeability of the films depending on the level of myofibrillar proteins from the film forming solutions with pH 2.7

The properties of barrier to water vapor of the films we tested were affected by the level of incorporated plasticizer. The plasticizer agent used by us was glycerol, a hydrophilic polyol. Additions of 30 or 50 g glycerin/100 g protein caused insignificant increases for the PVA values of the films made in strongly acidic medium (Figure 10).

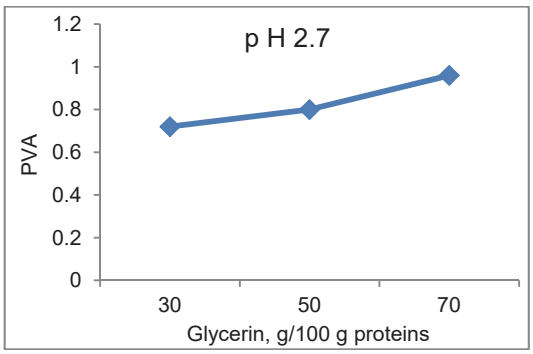

Figure 10. Water vapor permeability of the films depending on the addition of glycerin in the film forming solutions with pH 2.7

Protein films without plasticizer are often brittle and rigid, due to extensive interactions between the polymer molecules (Krochta, 2002). The addition of plasticizer affects not only the flexibility and other properties of the films, but additionally the film strength at water vapor and gases permeability (Sothornvit and Krochta, 2000 and 2001).

The variation in time of the water vapor transfer through comopozite films is shown in Figure 11, where you can observe greater resistance to water vapor for the films with gelatin addition.

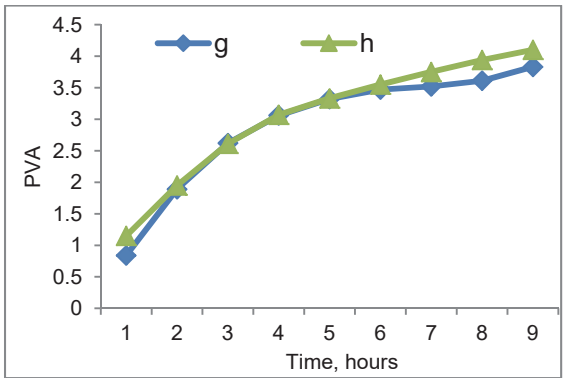

Figure 11. The permeability of the composite films according to the time

Increases in the permeability of films based on the level of glycerol were found as well by Nuthong et al. (2009) for the films based on porcine plasma (3%), by McHugh et al (1994) with films made from whey protein isolate, by Nemet et al. (2010) for the films obtained from myofibrillar proteins derived from chicken breast, who found values for the film permeability much lower than those obtained by us (0.21 to $0.29 \cdot 10^{-6} gm^{-1}s^{-1}Pa^{-1}$; myofibrillar protein concentration of 2%, pH 3.0, glycerin 25 to 65 g/100 g protein). Vanin et al. (2005) found no differences in the amounts of PVA on the gelatin based films where the levels of glycerin added to the film forming solutions varied between 10-30 g/100 g protein.

In our experimental conditions, the thickness of the films was relevant for PVA, in line with the increase of PVA for the films with greater thickness. The results listed in Table 4, indicate that PVA for edible protein films obtained by us are far greater than the values of PVA for plastic films [LDPE (low density polyethylene) $0.002 \cdot 10^{-10}$; high density polyethylene $0.008 \cdot 10^{-10}$; oriented

polypropylene $0.038 \cdot 10^{-10}$; polyester $0.198 \cdot 10^{-10}$; polyvinylidene chloride $0.002 \cdot 10^{-10}$ $gm^{-1}s^{-1}Pa^{-1}$] (Krochta et al., 1997; Fairley et al., 1997; Shiku et al., 2003; Garcia and Sobral, 2005).

Films color

Color is an important feature of the edible/ biodegradable films because it affects consumer acceptability of potential food applications. In our study, films color was assessed by determining the color parameters L* [(radiance, brightness, white/black; 100/0)], a* [red (+60)/ Green (-60)], b* [yellow (+60)/blue (-60) and ΔE] of food films which were prepared under both acidic and alkaline conditions.

Color parameters are registered in Table 5. Negative values of a* indicate, for the films made by us, a slightly green tint, which intensifies with the increase of protein content from the film forming solution. The same behavior has been reported by Sobral (1999) as well. For this specific parameter, slight differences were obseved between created acidic and alkaline conditions. The positive values of b* indicate yellow tones for films, the color intensity increasing along with the protein level increase from 1% to 2.5%, the increase being 39.4% for the film forming solution with pH 2.7.

Acidic conditions are inducing myofibrillar protein degradation which leads to the dismissal of amino groups for browning reactions during heat treatment and drying process (Prodpan and Benjakiti, 2005; Chinabhark et al., 2007). The values of L* parameter varied with the level of protein and with the addition of glycerin. The films have become less bright by increasing the protein concentration of the film forming solutions.

Table 5. Color parameters of edible films

Film	L*	a*	b*	ΔE
a	93.13±0.32	-1.22±0.05	10.19±0.46	93.69
b	92.95±0.39	-1.50±0.10	13.24±1.54	93.90
c	92.34±0.34	-1.73±0.03	15.97±0.33	93.73
d	89.95±0.47	-2.05±0.04	21.85±1.69	92.49
e	94.01±0.24	-0.94±0.05	7.47±0.39	94.30
f	93.57±0.29	-1.08±0.09	8.83±0.91	93.81
g	93.93±0.29	-1.03±0.08	8.59±0.53	94.33
h	93.66±0.21	-1.07±0.06	8.77±0.49	94.34

As shown in Table 5, parameter L* and b* were positively correlated with the level of protein from the film forming solutions, while a* parameters and color difference were negatively correlated, in all the cases the linear regression coefficients were greater than 0.975. Relatively weak correlations were recorded between L* and a * and between ΔE and a* parameters.

The increase of glycerin concentration causes the reduction of total color difference (ΔE) of the films, possibly due to dilution effect, basically in an independent manner of the fish myofibrillar protein concentration (Sobral et al., 2005); glycerin being a colorless substance.

The films obtained in this work showed, in general, colors comparable to films produced from muscle protein of Nile Tilapia (Paschoalick et al., 2003), but were more colorful than films based on myofibrillar proteins of Nile Tilapia (Sobral and et al., 2000), of egg albumen (Gennadios et al., 1996) and of pig skin gelatin (Sobral et al., 1999). Prodpran et al. (2005) have obtained films with the following color parameters: L = 86.1 ± 3; a* = 0.4 ± 0.1; b* = 39 ± 0.3 and 0.98 ± E = 86.18. Nuthong et al. (2009) have found the folowing values for films based on porcine plasma: L* = 87.12 ± 0.33; a* = - 2.37 ± 0.04; b* = 7.47 ± 0.13.

CONCLUSIONS

There were made 8 types of films based on fish myofibrillar proteins (bighead carp) by the solvent procedure by incorporation into the film forming solutions of different levels of protein, plasticizer and other additives, such as gelatin and cyclodextrin.

The films obrained by us were transparent, flexible and uniform, had smooth surfaces without pores or cracks visible to the naked eye. All kinds of films were easily detached manualy from the molding plates.

The thickness of the films varied depending on the measuring area, on the film forming solutions composition and on the environment characteristics. The increase of protein concentration and of the level of added glycerin caused an increase almost linear of the mean values of film thickness.

The increase of the glycerin level results in reduced color difference of the films due to the dilution effect of glycerin, regardless of the protein concentration. The transparency of the

films having higher protein concentration was lower in case of reduced concentration of glycerin, which becomes much more lower for high concentrations of plasticizer.

All the films studied in this work maintained their integrity after 24 hours of immersion in water, which indicates that the bighead carp myofibrillar proteins led to the formation of stable networks and that only low molecular weight hydrophilic substances were solubilized in water.

Water vapor barrier properties of the films were dependent on the levels of protein, of glycerin and of the addition of gelatin or cyclodextrin; PVA values and water content of the films increased with protein levels.

REFERENCES

Artharn A., Benjakul S., Prodpran T. and Tanaka M., 2007. Properties of a protein-based film from round scad (*Decapterus maruadsi*) as affected by muscle types and washing. Food Chemistry 103: p. 867-874.

Bourtoom T. and Chinnan M.S., 2008. Preparation and properties of rice starch - chitosan blend biodegradable film. Lebensm-Wiss. U-Technol., 2008, 41: p. 1633-1641.

Chinabhark K., Benjakul S., Prodpran T., 2007. Effect of pH on the properties of protein-basedfilm from bigeye snapper (*Priacanthus tayenus*) surimi. Bioresour.Technol., 98(1): p. 221-225.

Cuq B., Aymard C., Cuq J.L. and Guilbert S., 1995. Edible packaging films based on fish myofibrillar proteins: formulation and functional properties. Journal of Food Science, 60, p. 1369-1374.

Cuq B., Gontard N., Cuq J.L. and Guilbert S., 1997. Selected functional properties of fish myofibrillar protein-based film as affected by hydrophilic plasticizers. Journal of Agriculture and Food Chemistry 45: p. 622-626.

Fairley P., Krochta J.M. and German J.B., 1997. Interfacial interactions in edible emulsion films from whey protein isolate. Food hydrocolloids, 11: p. 245-252.

García F.T., Sobral P.J.A., 2005. Effect of the thermal treatment of the film-forming solution on the mechanical properties, color and opacity of films based on muscle proteins of two varieties of tilapia. LWT - Food Science and Technology, 38: p. 289-296.

Gennadios A., Weller C.L., Hanna M.A. and Froning G.W., 1996. Mechanical and barrier properties of edible wheat gluten-based films. J. Food Sci., 61: p. 585-589.

Hernandez-Munoz P., Villalobos R. and Chiralt A., 2004. Effect of cross-linking using aldehydes on properties of glutenin-rich films. Food Hydrocolloids 18: p. 403-411.

Ionescu A., Berza M., Banu C., 1992. Metode şi tehnici pentru controlul peştelui şi produselor din peşte. Editura Universităţii din Galaţi, p. 238.

Ionescu A., Aprodu I., Daraba A., Porneala L., 2008. The effects of transglutaminase on the functional properties of the myofibrillar protein concentrate obtained from beef heart. Meat Science, 79(2), p. 278-284.

Iwata K., Ishizaki S., Handa A., & Tanaka M., 2000. Preparation and characterization of edible films from fish-water soluble proteins. Fisheries Science, 66, p. 372-378.

Krochta M. and Johnston C.D., 1997. Edible and biodegradable polymer films: Challenges and opportunities. Food Technol., 51: p. 61-74.

Krochta J.M., Films, edible. 1997. In The Wiley Encyclopedia of Packaging Technology, 2nd Edition. A.L. Brody and K.S. Marsh (Eds.) John Wiley & Sons, Inc. New York.

Krochta J.M., 2002. Proteins as raw materials for films and coatings: Definitions, current status and opportunities. In: "Protein-based films and coatings". A. Gennadios (Ed.), p. 1, CRC Press Inc. Publishing Co., New York, CT, p. 1-39.

McHugh T.H. and Krochta J.M., 1994. Dispersed phase particle size effects on water vapor permeability of whey protein-beeswax edible emulsion films. J. Food Process. Preserv., 1994, 18(3): p. 173-188.

Nemet N.T., Šošo V.M. and Lazić V.L., 2010. Effect of glycerol content and pH value of film-forming solution on the functional properties of protein-based edible films. APTEFF, 41, p. 57-67.

Nuthong P., Benjakul S., Prodpran T., 2009. Effect of some factors and pretreatment on the properties of porcine plasma protein-based films. Food Science and Technology, 42: p. 1545-1552.

Orliac O., Rouilly A., Silvestre F. and Rigal L., 2002. Effects of additives on the mechanical properties, hydrophobicity and water uptake of thermo-moulded films produced from sunflower protein isolate. Polymer 43: p. 5417-5425.

Paschoalick T.M., Garcia F.T., Sobral P.J.A. and Habitante A.M.Q.B., 2003. Characterization of some functional properties of edible films based on muscle proteins of Nile tilapia. Food Hydrocolloids 17: p. 419-427.

Perez-Gago M.B. & Krochta J.M., 2001. Denaturation time and temperature effects on solubility, tensile properties, and oxygen permeability of whey protein edible films. Journal of Food Science, 66(5), p. 705-710.

Prodpran T. and Benjakul S., 2005. Acid and alkaline solubilization on properties of surimi based film. Songklanakarin. Journal of Science and Technology, 27(3): p. 563-574.

Rostamzad H., Paighambari S.Y., Shabanpour B. and Ojagh S.M., 2015. Characteristics of a biodegradable protein based films from Silver carp (Hypophthalmichthys molitrix) and their application in Silver carp fillets. International Food Research Journal 22(6): p. 2318-2326.

Shiku Y., Hamaguchi P.Y. & Tanaka M., 2003. Effect of pH on the preparation of edible films based on fish myofibrillar proteins. Fisheries Science, 69, p. 1026-1032.

Sikorski Z.E. and Noczk M., 1981. Modification of tehnological properties of fish proteins concetrates. Crit. Rev. Food Sci. Nutr., 1981, 14: p. 202-230.

Sothornvit R., Krochta J.M., 2000. Water vapor permeability and solubility of films from hydrolyzed whey protein. Journal of Food Science, Vol. 65, No. 4, p. 700-703.

Sothornvit R. and Krochta J.M., 2001. Plasticizer effect on mechanical properties of b-lactoglobulin films. Journal of Food Engineering, 2001, 50: p. 149-155.

Sobral P.J.A., 1999. Propriedades funcionais de biofilmesde gelatin em função da espessura. Ciência & Engenharia, 1999, 8(1): p. 60-67.

Sobral P.J.A., 2000. Influência da espessura sobre certas propriedade de biofilmes á base de proteínas miofibrilares. Pesquisa Agropecuária Brasileira, 35(6): p. 251-1259.

Sobral P.J.A., Santos J.S.S., García F.T., 2005. Effect of protein and plasticizer concentrations in film forming solutions on physical properties of edible films based on muscle proteins of a Thai Tilapia. Journal of Food Engineering, 70: p. 93-100.

Tanaka M., Shoichiro Ishizaki, Toru Suzuki and Rikuo Takai, 2001. Water Vapor Permeability of Edible Films Prepared from Fish Water Soluble Proteins as Affected by Lipid Type 1. Journal of Tokyo University of Fisheries, 87: p. 31-37.

Tao Z., Weng W.Y., Cao M.J., Liu G.M., Su W.J., Osako K. &Tanaka M., 2015. Effect of blend ratio and pH on the physical properties of edible composite films prepared from silver carp surimi and skin gelatin. J. Food Sci. Technol., 52(3): p. 1618-1625.

Tongnuanchan P., Benjakul S., Prodpran T., Songtipya P., 2011. Characteristics of film based on protein isolate from red tilapia muscle with negligible yellow discoloration. International Journal of Biological Macromolecules 48, p. 758-767.

Vanin F.M., Sobral P.J.A., Menegalli F.C., Carvalho R.A. and Habitante A.M.Q.B., 2005. Effects of plasticizers and their concentrations on thermal and functional properties of gelatin-based films. Food Hydrocolloids, 19: p. 899-907.

***AOAC, 1990. Moisture in Meat. Official Methods of Analysis 950.46, 11, 931.

***AOAC, 1990. Crude protein in Meat. Offcial Methods of Analysis 981.10, 11, 937.