International Journal of Research Studies in Biosciences (IJRSB)
Volume 3, Issue 8, August 2015, PP 91-97
ISSN 2349-0357 (Print) & ISSN 2349-0365 (Online)
www.arcjournals.org

Study of Mechanical and Water Barrier Properties of Chitosan Based Edible Films Affected By Process Parameters by Using Response Surface Methodology

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Abstract: *Response Surface methodology (RSM) was used to optimize the pH (4.0, 5.5, and 6.0), drying temperature (40, 50, 60 ^{0}C) for the development of chitosan based edible films. The optimization was done on the basis of different responses viz. water transmission, tensile strength and elongation at break. WTR showed the quadratic model with independent variables (pH and drying temperature). WTR was observed to be highly significant ($P<0.001$) with B (temperature). TS showed the linear relationship with pH and drying temperature and TS was observed to be highly significant ($P<0.001$) with A (pH) and B (Temperature). EB showed the quadratic relationship between the independent variables pH and drying temperature. EB was observed to be highly significant ($P<0.001$) with B (Temperature). All the response variables were in favourable range.*

1. INTRODUCTION

The continuously increasing demand of consumers in the quality, nutrition and safety of food has encouraged research into edible films. Edible films are thin films prepared from edible material that acts as a barrier to control the moisture, oxygen, carbon dioxide, flavor and aroma exchange between the food components and the atmosphere surrounding them. It is also used to enhance the shelf life, quality and reduce the microbial load of fresh produce [1].

Edible film provides the application of a layer of any edible material on the surface of food by providing modified atmosphere, reducing moisture, retarding gas transfer and improving the general appearance of the product. These films consist of biopolymer sources including polysaccharides, proteins and lipids that can function as barriers to water vapour, gases and other solutes and at the same time carriers for ingredients like antimicrobial, antioxidant agents thus extending shelf life and enhancing quality of fresh and processed food [2]. Edible films made from carbohydrates they have lower moisture barrier tendency due to their hydrophilicity [3]. Various materials were explored and chitosan exhibited a film forming property [4]. Moreover there is a growing interest in chitosan to develop newer packaging material because it is nontoxic, biodegradable, bio functional, biocompatible in addition to having antimicrobial characteristics [5]. Chitosan films also act as effective carriers of many functional ingredients like antimicrobial agents and antioxidants to extend storage quality of food [6][7].

The important properties of edible films are their barrier properties and mechanical properties. The mechanical properties provide the information regarding strength of the film and water barrier properties during its application, distribution and handling of the food [8]. Important factor affecting the functional property of edible films is pH and drying temperature. It has been reported that the pH and temperature induce physical and chemical changes in edible films that cause the structural changes in films resulting to alteration in the barrier and mechanical properties of the films.

However, information regarding the effect of pH and drying temperature on water vapour transmission rate and mechanical properties of chitosan based edible films is lacking in literature. Such information is important for handling, distribution and storage of chitosan based edible films. The objective of the study was to determine the effect of pH and drying temperature on water vapour transmission rate and mechanical properties of Chitosan based edible film.

2. MATERIALS AND METHODS

2.1. Preparation of Chitosan Based Edible Films

The chitosan biopolymer films were prepared using the process described by [9] with some modifications. Biopolymer films from chitosan were developed using casting technique on levelled trays. The effects of pH (4-7) and heating temperature (40-60°C) were then varied at different levels selected on the basis of the present investigation trial and previous studies [10][11][12]. The chitosan solution (1%w/v) was prepared by dissolving chitosan in (1%v/v) acetic acid under constant stirring at 300 rpm using a magnetic stirrer at room temperature for about 12h. One percent of plasticizer (w/w chitosan) was then added into the chitosan solution and stirring was continued at room temperature for about 1h. After mixing, the solution was centrifuged for 15min at 12,000 rpm by a refrigerated centrifuge to remove the undissolved impurities and bubbles in the solution. The solution (20g) was then poured on a polystyrene plate with dimension 40×40×8 mmH to cast a chitosan film. The films were dried at ambient temperature of about 40°C in a hot air oven.

3. PROCESSING PARAMETERS

3.1. Thickness

Film Thickness measured by the help of micrometer, with an accuracy of ±2µm. Five to ten thickness measurements were carried out on each film, from which an average was obtained [11].

3.2. Mechanical Properties Measurement

The measurement of mechanical properties of biopolymer film was measured by using a Texture Analyzer System. After conditioning chitosan films were cut into 10 cm strips and tested for tensile strength and percent elongation according to the [13]. Initial grip separation and crosshead speed were set at 50mm and 50 mm/min, respectively. Tensile strength was calculated by dividing the maximum load for breaking the film by its cross-sectional area. Percent elongation was determines by dividing the film elongation at rupture by the initial grip separation. All tests were performed in triplicate and the average values were reported.

3.3. Water Vapour Transmission Rate (WVTR)

WVTR was determined by gravimetric method using modified [14]. The test cup was filled with 15 ml of distilled water to produce 100% RH below the film . The biopolymer film was mounted to the top of the cup . The cup was placed in a dessicator containing magnesium nitrate solution at 25°C. Fans were operated in the chamber, thus circulating the air continuously over the surface of the film to remove the permeating water vapour. The mass of the cup was recorded 6 times at 2h interval after steady state was reached. Mass loss was plotted versus time and a straight line was obtained in duplicate. Linear regression was then used to first estimate the slope of the line to calculate the water vapour transmission rate (WVTR) as follows:

$$\mathrm{WVTR} = \frac{\mathrm{Slope}}{\mathrm{A}}$$

Where, WVTR is the water vapour transmission rate (g/m^2 day), Slope is the slope of the mass loss versus time curve (g/day), and A is the test cup mouth area (m^2).

3.4. Experimental Design and Statistical Analysis

Response Surface Methodology (RSM) was used to generate the experimental designs, statistical analysis and regression model with the help of Design Expert Software Version 8.0.5 (Stat ease Inc).The Central Composite Design (CCRD) with a quadratic model was employed [15]. Two independent variables namely pH (x_1) and Temperature (x_2) were selected. Each independent variable had 3 levels which were low, medium and high. These levels were denoted by -1, 0 and +1.

The design and level of factors are shown in Table 3.1. The main advantage of using experimental design approach was the ability to reduce the number of experimental runs required to provide sufficient information for statistically acceptable results. The experiments were conducted for two variables using Central Composite Design (CCRD) involving 13 combinations. The runs were randomized in order to minimize the effects of unexplained variability in the observed responses due to extraneous factors.

After the selection of model suggested, analysis of variance was done .To evaluate the fitness of model, F-value were determined.

Table1. *Levels of examined processing variables according to central composite RSM design*

Symbol	Experimental Factor	Coded value			Actual value		
A	pH	-1	0	+1	4	7	5.5
B	Temperature	-1	0	+1	40	60	50

4. RESULTS AND DISCUSSION

Response surface methodology (RSM) using central composite rotatable design (CCRD) with a quadratic model and linear model was employed. The level of 2 factors (processing variables) and experimental responses are presented in Table2. The data was analysed employing multiple regression technique to develop a response surface model.

Table2. *The Central Composite Experimental Design Employed for Preparation of Edible Film*

Run	**Factor1 pH**	**Factor 2 Temp ˚C**	**Response 1 WTR g/m^2/day**	**Response 2 TS%**	**Response 3 EB%**
1	7.62132	50	1.16	49.31	6.4
2	5.5	50	1.14	28.14	5.8
3	5.5	50	1.15	26.65	7.7
4	5.5	50	1.14	28.34	4.6
5	3.378	50	1.17	16.42	8.6
6	7	60	1.11	48.42	28.2
7	5.5	50	1.16	34.62	12.24
8	5.5	35.85	1.55	28.42	6.2
9	5.5	50	1.22	32.82	16.47
10	7	40	1.45	39.45	6.4
11	4	60	1.09	25.82	30.4
12	5.5	64.14	1.06	35.04	45.48
13	4	40	1.43	18.11	3.6

4.1. Effect of pH and Drying Temperature on Water Vapour Barrier Properties and Mechanical Properties

Water Transmission Rate (WTR)

WTR is the most important property of edible films mainly because water is important in deteriorative reactions in foods and made to impede moisture transfer between food and surrounding atmosphere [16]. High WTR of edible film is not desirable from usage point of you.

The equation given in Table 4 showed a quadratic equation which depicts a quadratic relationship between the independent variables (A, B) and the dependent variable WTR. A quadratic model (R^2=0.97) fitted for WTR was observed to be highly significant ($P<0.001$) with B (temperature) and B^2 as significant model terms (Table 3). The R^2 value (0.90) given in Table 4, being a measure of the goodness of fit of the model, indicated that 90% of the total variation was explained by the model. WTR varied with the temperature as compared to pH (Fig 1a). The WTR values decreases with increase in temperature (1.06-1.55 g/m^2/day) but no effect of pH on edible films. Our results are in agreement with Jangchud and Chinnan [10] who concluded that the reduction in WTR values with increased temperature may be due to greater cross linking, resulting in a tight and compact network structure. Similarly Bourtoom et al [17] also reflected the same trend in water soluble fish protein surimi edible films, which stated that increasing of thermal energy at higher temperature resulted in lower WTR due to the greater cross link between protein-protein chain resulting in tighter and compact structure. On the contrary Singh et al, [18] stated that the there is an increase WTR with increase in temperature. Bajpai et al, [16] observed that permeation of water vapour through films enhanced with the increasing in temperature. Gontard et al, [19] stated that the increase in WTR might be related to the hydrophilicity of all tested plasticizers. It is well known that the presence of plasticizers increases the concentration of polar residues in hydrocolloid based film

Table3. *Analysis of variance for different Responses studied*

Source [a]	WTR	TS	EB
	F value		
Model	53.67***	72.76***	24.96***
A	0.080 NS	134.38***	0.052 NS
B	225.41***	11.14**	90.26***
AB	0.00 NS	-	0.42 NS
A^2	0.70 NS	-	0.30 NS
B^2	42.86**	-	32.36***
Lack of Fit	0.84 NS	0.42 NS	0.11 NS
R^2	0.974	0.935	0.946
Adjusted R^2	0.956	0.922	0.908

** Significant at P <0.05; ** significant at P <0.01; *** significant at P <0.001.*

A= pH, B= Temperature, WTR= water Transmission Rate, TS= Tensile Strength, EB=Elongation at Break

NS= not significant

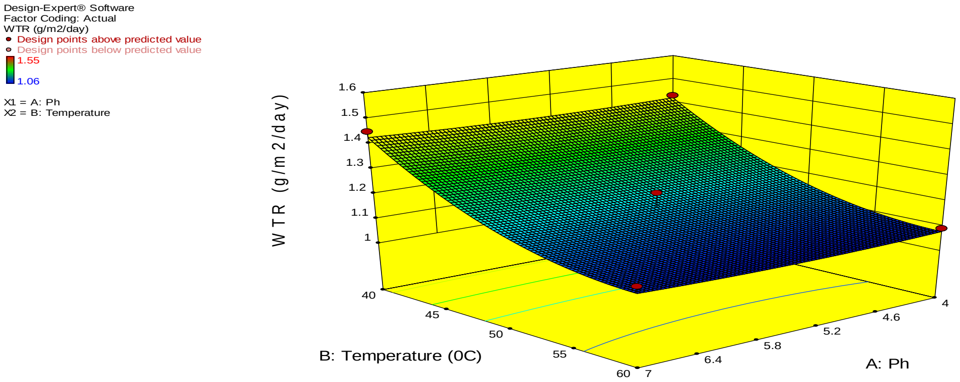
Table4. *Predicted Equation obtained for different responses using regression analysis*

Predicted equation for the responses in terms of coded factors [a]	R^2
$WTR= 1.16+3.23\times10^{-3}A-0.17\,B-8.37\times10^{-17}\,AB+0.01\,A^2 + 0.08\,B^2$	0.90
$TS= 31.66+ 11.31\,A+ 3.26\,B$	0.90
$EB= 9.36-0.31\,A+13.02\,B-1.25\,AB-0.81\,A^2+8.36\,B^2$	0.89

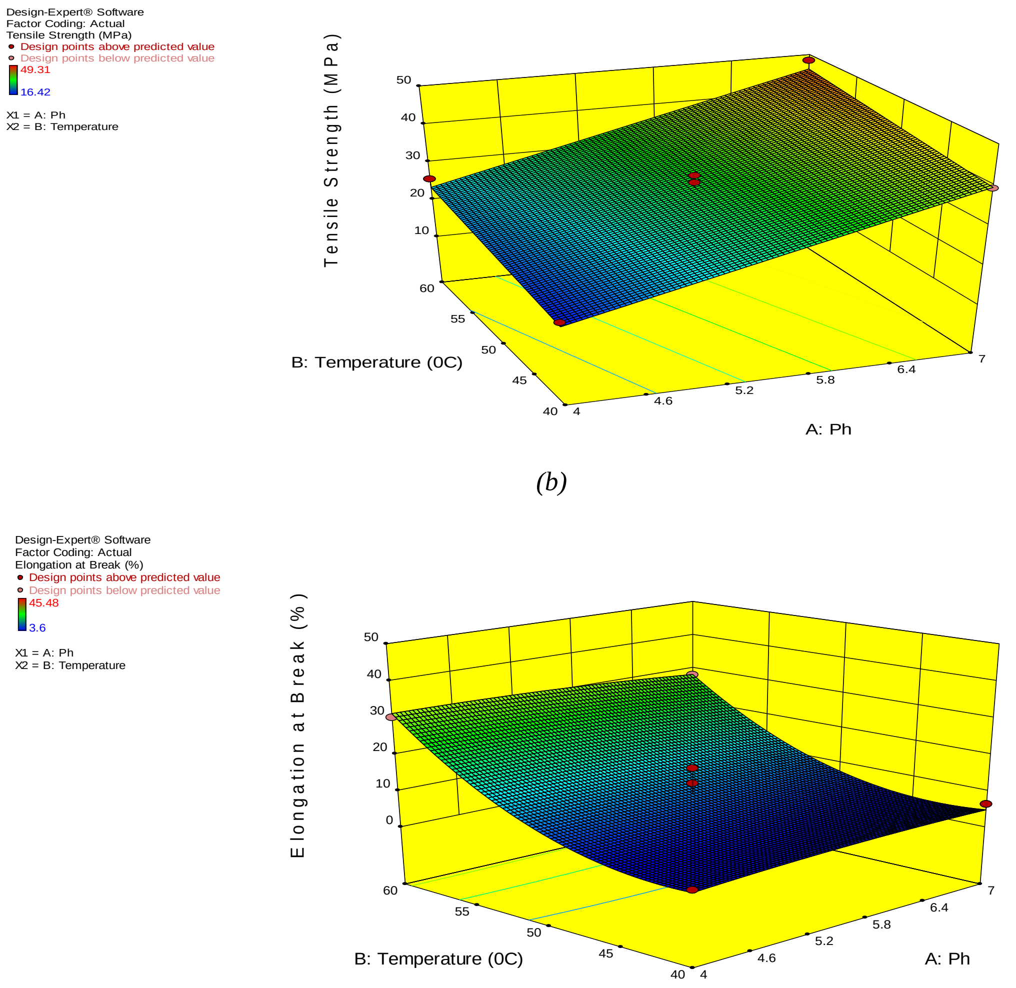
A= pH, B= Temperature, WTR= water Transmission Rate, TS= Tensile Strength, EB=Elongation at Break

Tensile Strength (TS)

The influence of experimental conditions T and pH, on the tensile strength using statistical model RSM, to determine the simultaneous effect of both T and pH and to optimize the experimental conditions in order to optimize the experimental conditions in order to obtain film with best mechanical properties. The regression equations for the relationship between factors (T and pH) investigated is presented in Table 4. It shows the linear relationship between the independent variables (A, B) and the dependent variable TS. A linear model (R^2=0.93) fitted for TS was observed to be highly significant (P<0.001) with A (pH) and B (Temperature) as significant model terms (Table 3). The R^2 value (0.90) given in Table 4, being a measure of the goodness of fit of the model, indicated that 90% of the total variation was explained by the model. The pH and temperature of the film solution are the most important factors affecting the mechanical properties, while heating time had very lesser effect. Comparing within the same heating temperature of film solutions, the results demonstrated that, TS increased as pH of film solutions increased (Fig1 b). Our result similar with the findings of Bourtoom et al, [17] who stated that at higher pH of film solutions induced formation of resistant films. However when pH was increased over 10, TS decreased. Similarly Popovic *et al*,[12] reported that the TS of PuOC films increased with both increases of pH and T.In contrast to data of Nandane and Jain, [15] concluded that TS decreased with the increased in pH because at alkaline pH away from the isoelectric point, denaturation of proteins was promoted and resulted in unfolding and solubilising of the proteins.



(a)



(b)

(c)

Fig1. *Response Surface for (a) WTR (b) Tensile Strength (c) Elongation at Break of Edible Film as a Function of pH and Drying Temperature*

Elongation at Break

EB is also a very important mechanical property of films. It is an indicator of the flexibility. The regression equations for the relationship between factors (T and pH) investigated is presented in Table 4. It shows the quadratic relationship between the independent variables (A, B) and the dependent variable EB. A linear model (R^2=0.94) fitted for EB was observed to be highly significant ($P<0.001$) with B (Temperature) and B^2 as significant model terms (Table 3). The R^2 value (0.89) given in Table 4, being a measure of the goodness of fit of the model, indicated that 89% of the total variation was explained by the model. EB was varied with the temperature (Fig 1c). EB increases with increase in temperature. Similar findings were observed by Bourtoom et al [17] which indicate that water soluble surimi films exhibited the greater EB values when temperature increases it was due to an increased number of interactions disulfide (SS bonds) [20].Prolonged heating resulted in the increase of elongation at break. The experiments showed that TS and EB films were almost inversely related.

5. CONCLUSION

It can be concluded from the present investigation that the mechanical properties and water vapour transmission rate of edible films prepared from chitosan based edible film were pH and drying temperature dependent. The WTR value decreases as temperature increases, EB value increases with increase in temperature. pH had no effect on WTR and EB, but TS increases with increases in pH. Response surface methodology was found to be an effective technique to optimize the process development of chitosan based edible film as a function of pH 5.5 and 50^0C for 48h. Chitosan based edible films useful for the food packaging industy for its use on commercial level.

REFERENCES

[1] Suyatma, E.N., Copinet, A., Tighzert, L. and Coma, V., Mechanical and Barrier Properties of Biodegradable Films Made from Chitosan and Poly (Lactic Acid) Blends. J Poly and the Envi. 12,1-6. (2004)

[2] Arvanitoyannis E, Psomiadoub E, Nakayama A., Edible films made from sodium caseinate, starches, sugars or glycerol. Part I Carbohydr Polymer31:179-192. (1996).

[3] Hiemenz, P. C. and Rajagopalan, R.,Surface Tension and Contact Angle, 3rd edn. Marcel Dekker, Inc., New York, (1997). pp 248-255.

[4] Dutta, P. K., Tripathi, S., Mehrotra, G. K., and Dutta, J., Perspectives for Chitosan based antimicrobial films in food applications. Food Chem, **114(**4), 1173-1182. (2009)

[5] Jayakumar, R., New, N. T., Tokura, S. and Tamura, H., Sulfated chitin and Chitosan as novel biomaterials. I. J of Biol Macromol, **40**, 175-181. (2007).

[6] Tripathi, S., Mehrotra, G. K. and Dutta, P. K. Chitosan Based Antimicrobial Films for Food Applications. E-Polym, 93, 1-7. (2008).

[7] Park S and Zhao Y., Incorporation of a high concentration of mineral or vitamin into chitosan-based films. Journal Agric Food Chem 52:1933- 1939.(2004)

[8] Bourtoom, T., Chinnan, M. S., Preparation and properties of rice starch-chitosan blend biodegradable film. LWT- Food Sci and Technol, **41**: 1633-1641. (2008).

[9] Mayachiew P, Devahastin S., Comparative evaluation of physical properties of edible chitosan films prepared by different drying methods. Dry Technol, **26**, 176–185. (2008).

[10] Jangchud, A. and Chinnan, M. S., Peanut protein film as affected by drying temperature and pH of film-forming solution. J. Food Sci, 64,153-157. (1999).

[11] Hamaguchi, P. Y., Yin, W. and Tanaka, M., Effect of pH on the formation of edible films made from the muscle protein of blue marlin (Makaira mazara). Food Chem,**100**,914-920. (2007).

[12] Popovic, S., Pericin, D., Vastag, Z., Popovic, L. and Lazic V., Evaluation of edible film- forming ability of pumpkin oil cake; effect of pH and temperature. Food Hydrocoll, 25, 470-476.(2011)

[13] American Society for Testing and Materials. Standard test methods for tensile properties of thin plastic sheeting. In Annual Book of ASTM Standards; ASTM: Philadelphia, 1995, 182-190.

[14] American Society for Testing and Materials. Standard test methods for water vapor transmission of materials. In Annual Book of ASTM Standards; ASTM: Philadelphia, 1995, 697-704

[15] Nandane, A.S. and Jain, R.K., Study of mechanical properties of soy protein based edible film as affected by its composition and process parameters by using RSM. J Food Sci Technol, 52(6), 3645-3650.(2015)

[16] Bajpai SK, Navin C, Ruchi L., Water vapour permeation and antimicrobialproperties of sago starch based films formed via microwave irradiation .Int Food Res J 18:417-426. (2011)

[17] Bourtoom, T., Chinnan, M. S., Jantawat, P. and Sanguandeekul, R., Effect of select parameters on the properties of edible film from water –soluble proteins in surimi wash-water. LWT- Food Sci Technol, **39**(4), 406-419. (2006).

[18] Singh, T. P., Chatli, M. K. and Sahoo, J., Development of Chitosan based edible films: process optimization using response surface methodology. J Food Sci Technol 52(5):2530-43. (2014).

[19] Gontard N, Guilbert S, Cuq J.L., Water and glycerol as plasticizers affect mechanical and water vapor barrier properties of an edible wheat gluten film. J. Food Sci. 58:206-211. (1993).

[20] Shimada, K and Cheftel, J. C.,Texture characteristics, protein solubility and sulfhydryl group/disulfide contents of heat-induced gels of whey protein isolate. Journal Agric Food Chem, 36, 1018-1025. (1998).

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