Research Article

Effect of Coating Formulation and Ambient Storage Conditions on Physical Properties of Litchi

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Abstract

The concept of coating only to prevent browning of the pericarp is old, and recently vitamins have been added to the coating material to enrich the final product and extend shelf life. Physiological weight loss of coated litchi fruits was significantly affected by the coating and packaging process when stored under ambient conditions and ranged from 5.23 to 8.81%. The firmness is an important factor, which decides market acceptance and it is related to water content and metabolic changes in litchi after harvest. The firmness of the coated litchi fruit ranged from 15.52 to 21.47 N at ambient temperature. The synergistic effect of coating and packaging could be seen under ambient storage.

Keywords: Litchi, α-Tocopherol, Salicylic acid, Perforation, Physical

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Introduction

Litchi (*Litchi chinensis Sonn*.) is a subtropical fruit of high commercial value because of its white, translucent aril and attractive red colour. The fruit is highly valuable due to its characteristics pleasant flavour and attractive colour. Litchi is highly perishable due to its delicate nature. Its shelf life under ambient conditions does not exceeds 24-72 hours. Postharvest decay was identified as major problems restricting expansion of the industry in litchi-exporting countries. Postharvest losses in litchi are estimated to be 20–30% of the harvested fruit and can even be as high as 50% prior to consumption [1].

Postharvest treatments, such as sulphur fumigation and acid dip can effectively increase shelf life of litchi fruit [2]. The aim of this work was to study the effect of coatings formulations to maintain its physical properties under ambient conditions to expand its market.

Material and Methods

Materials

In the experiment, cultivar Rose Scented of litchi was harvested at ripe stage (90 – 100% of the peel exhibiting red colour). Immediately after harvesting, fruits were taken to the laboratory and fruits of uniform size, shape and colour as well as free from diseases or blemishes were selected for the experiment. The α-Tocopherol, chitosan and salicylic acid, Tween-20 and other chemicals required for experiment were purchased from Sigma Aldrich Chemicals Pvt. Ltd. Mumbai (Maharashtra).

After selecting uniform fruits, they were treated with aqueous solutions of α-Tocopherol, chitosan and salicylic acid, in combination according to the treatment details. Treatments were performed by dipping the fruits in 5 litre treatment solutions containing Tween-20 (2 g L^{-1}) as surfactant, at 25ºC for 5 min. Whereas, control fruits were dipped in distilled water. Following treatment application, fruits were air-dried at room temperature, then packaged in perforated polyethylene bags and stored at room temperature (25^0C) in plastic basket. After 3, 6, 9, 12 and 15 days of storage, fruits from each treatment were sampled at random for analysis of various physical parameters.

Preparation of coating formulation

The aqueous solutions of α-Tocopherol, chitosan and salicylic acid, in combination according to the treatment details. Chitosan is insoluble in water but, soluble in slight acidic (< 6.5) solution, the acidity is increased by addition of salicylic acid into solution during formation solution along with high speed stirrer.

Coating application and storage

Treatments were performed by dipping the fruits in 5 litre treatment solutions containing Tween-20 (2 g L^{-1}) as surfactant, at 25ºC for 5 min. Whereas, control fruits were dipped in distilled water. Following treatment application, fruits were air-dried at room temperature, then packaged in perforated polyethylene bags and stored at room temperature (25^0C) in plastic basket.

Experimental plan

The variables that have greatest influence on the response could be selected as operating parameters of final experiments [3].

Table 1 Selected independent, dependent and constant parameters and their values

Sr. No	Parameters	Levels	Range	Responses
Independent variables				
1	α-Tocopherol (%)	5	0.1,0.2,0.3,0.4,0.5	Physiological loss
2	Chitosan (%)	5	0.5,1.0,1.5,2.0,2.5	in weight
3	Salicylic acid (mM)	5	0.5,1.0,1.5,2.0,2.5	Firmness
4	Perforation (%)	5	0.1,0.2,0.3,0.4,0.5	

Physiological loss in Weight

To determine the physiological loss in weight of the litchi fruit during postharvest storage, both treated and control fruits were weighed at 3 days sampling intervals with the help of an electronic balance. Then weight loss was calculated by using the following formula and data were communicated in percentage.

$$\text{Physiological loss in weight (\%)} = \frac{\text{Initial weight} - \text{Weight after known storage} \times 100}{\text{Initial weight}}$$

Firmness (N)

The pericarp of litchi turn out to be dry throughout storage due to moisture loss and firmness upsurges. The firmness of litchi was measured using texture analyzer (Stable Microsystems, Model TA-xt 2i) using a P/2 cylindrical probe, test speed of 1mm/s and distance of 3mm. A curve was plotted between force and time and maximum force was noted. The data were presented a means of 10 independent measurements.

Statistical analysis

Experiments were designed using Central Composite Rotatable design (CCRD) method for satisfying the principle of randomization of Response Surface Methodology (RSM). For vanishing the effect of residuals during Response surface analysis, levels of the independent variables have been expressed in coded form. **Table 2** shows the actual and coded values of different levels of each independent variable considered during study.

Table 2 Experimental variables (independent) for enhancement of shelf life of Litchi (coded and actual values)

Independent variables		**Coded Levels**				
Name	**Code**	**-2**	**-1**	**0**	**1**	**2**
		Actual Levels				
α-Tocopherol (%)	A	0.1	0.2	0.3	0.4	0.5
Chitosan (%)	B	0.5	1.0	1.5	2.0	2.5
Salicylic acid (mM)	C	0.5	1.0	1.5	2.0	2.5
Perforation (%)	D	0.1	0.2	0.3	0.4	0.5

Fitting of Second Order Model

To determine the relationship between the dependent and independent variables, the obtained experimental data were subjected to the multiple regression analysis. Regression analysis was done by using Design Expert 8.0.6 software. Each response was represented by a second order polynomial equation. A second order response function for four independent variables had the following general form:

$$Y = \beta_0 + \sum_{i=1}^{4}\beta_i A_i + \sum_{i=1}^{2}\sum_{j=i+1}^{4}\beta_{ij}A_i B_j + \sum_{i=1}^{4}\beta_{ii}A_i^2 \qquad (1)$$

Where, β_0, β_i, β_{ii}, β_{ij} are constants; A, B are variables (coded).

The adequately qualified model with respect to known criteria (i.e., High R^2, $F_{cal}>F_{tab}$ and non-significant lack of fit) was selected to describe the behaviour of response, otherwise, transformation of the dependent variable was performed [3].

Statistical Analysis and Optimization of Variables

Multivariate optimization and data analysis (ANOVA) of independent and dependent variables were carried out through Response Surface methodology (RSM) using Design Expert 8.0.6**.** 3-D surface plot were drawn using Sigma plot-10 software to visualize the individual and combined effect of each independent variable by keeping another two variables at their respective optimum points.

Results and Discussions

The significant effect of coating formulations was found on physical properties of litchi. The result and discussion for ambient storage conditions were depicted in **Table 3**.

Table 3 Experimental data on optimization of coating and packaging perforation of litchi for ambient storage condition (15th Day)

Treatments	α- Tocopherol (%)	Chitosan (%)	Salicylic acid (mM)	Perforation (%)	Physiological loss in weight (%)	Firmness (N)
1	0.2	2	1	0.2	8.12	19.78
2	0.5	1.5	1.5	0.3	5.43	15.51
3	0.3	1.5	1.5	0.3	6.35	16.55
4	0.1	1	2	0.4	6.43	16.43
5	0.4	2	1	0.2	7.49	16.95
6	0.4	1	1	0.2	7.94	18.48
7	0.4	2	2	0.2	6.27	16.44
8	0.3	1.5	1.5	0.3	6.38	16.37
9	0.3	1.5	0.5	0.3	7.58	16.58
10	0.3	1.5	1.5	0.3	6.33	16.35
11	0.4	2	1	0.4	6.52	16.26
12	0.3	1.5	1.5	0.3	6.37	16.17
13	0.2	1	1	0.2	8.81	21.26
14	0.2	1	1	0.4	8.79	21.46
15	0.4	2	2	0.4	5.23	15.73
16	0.2	2.5	1	0.4	7.16	17.25
17	0.3	2.5	1.5	0.3	5.56	16.12
18	0.3	1.5	1.5	0.3	6.41	16.42
19	0.4	1	2	0.2	7.59	17.29
20	0.3	0.5	1.5	0.3	7.13	17.39
21	0.4	1	1	0.4	7.68	18.57
22	0.2	2	2	0.3	8.45	17.56
23	0.5	1.5	1.5	0.3	7.35	17.66
24	0.3	1.5	1.5	0.5	5.73	21.89
25	0.3	1.5	1.5	0.3	6.32	16.28
26	0.3	1.5	1.5	0.3	5.57	16.18
27	0.3	1.5	1.5	0.1	7.78	17.82
28	0.2	0.5	1.5	0.2	8.19	20.74
29	0.2	0.5	1.5	0.4	6.92	17.59
30	0.2	2	1.5	0.4	6.71	16.88

Effect of independent variables on physiological loss in weight

Physiological loss in weight of coated litchi fruits stored at ambient storage condition varied in the range of 5.23 to 8.81%. The maximum value obtained at experiment number 13 with experimental conditions of α-Tocopherol 0.2% (A=-1), 1% chitosan (B= -1), salicylic acid 1 mM (C= -1) and perforation 0.2% (D=-1) while minimum (5.23) at experimental conditions of α-Tocopherol 0.4%(A=1), 2% chitosan (B=1), salicylic acid 2 mM (C=1) and perforation percentage of 0.4% (D=1) at experiment no. 9 (**Table 4)**.

Table 4 Effect of treatments on physiological loss in weight under ambient storage conditions

Source	SS	Df	MS	F-value	P-value
Model	24.13	14	1.72	5.10	0.0016***
A-α-Tocopherol	5.84	1	5.84	17.30	0.0008***
B-Chitosan	3.79	1	3.79	11.23	0.0043***
C-salicylic acid	4.80	1	4.80	14.24	0.0018***
D-perforation	5.52	1	5.52	16.38	0.0010***
AB	0.2162	1	0.2162	0.6407	0.4359
AC	0.1406	1	0.1406	0.4167	0.5283
AD	0.0196	1	0.0196	0.0580	0.8128
BC	0.1332	1	0.1332	0.3947	0.5392
BD	0.25	1	0.25	0.7408	0.4029
CD	0.5625	1	0.5625	1.66	0.2162
A^2	0.6483	1	0.6483	1.92	0.1859
B^2	0.5569	1	0.5569	1.65	0.2183
C^2	1.0971	1	1.097	3.25	0.0914
D^2	1.6464	1	1.64	4.87	0.0431**
Residual	5.06	15	0.3375		
Lack of Fit	NS				
R^2	0.8267				
Adj R^2	0.6649				

*** 1% level of significance, **5% level of significance

Physiological weight loss data obtained from coated litchi fruits stored under ambient storage condition was fitted to full second-order mathematical model equation (1) and the result of regression analysis was represented by equation (2). The coefficient of determination (R^2) for the fitted regression model was 82.67% and adj R^2 was 66.49%, which implies that the model could account for 82.67% data. The model was found to be significant at 1% level of significance with non-significant lack of fit. Therefore, the second-order model was considered to be adequate for describing the change in physiological weight loss with the specified values of independent parameters.

$$\text{Physiological weight loss} = 6.36 - 0.49A - 0.39B - 0.44C - 0.48D - 0.12AB - 0.093AC + 0.035AD + 0.091BC - 0.12BD - 0.18CD + 0.15A^2 + 0.14B^2 + 0.2\,C^2 + 0.24D^2 \quad (2)$$

Where, A is α-Tocopherol, B is chitosan, C salicylic acid and D is perforation percentage (all in coded form).

Table 4 expresses the individual effect of each term in second-order quadratic equation fitted to the experimental data. It is revealed that at linear levels all independent variables affected significantly with 1% of the level of significance while at quadratic levels only perforation percentage of packaging material had a significant effect ($P<0.05$) on the weight loss. There was non-significant effect in interactive terms of independent variables. The results were in accordance with [4]. Simplified second order equation of physiological weight loss becomes,

$$\text{Physiological weight loss} = 6.36 - 0.49A - 0.39B - 0.44C - 0.48D + 0.24D^2 \quad (3)$$

Response surface plots as shown in **Figure 1** confirmed the physiological weight loss of the treated fruit decreased rapidly at the initial increase in tocopherol percentage, at the same time with increase in perforation percentage beyond its central value, it was increased. The α-Tocopherol significantly affected the physiological loss in weight, it prevents the respiration rate initially and prevents catalase enzymes activity which is responsible for breaking down of the complex structure into simple, which can be utilized the during respiration and one of the major cause of loss in weight. The minute particle size of α-Tocopherol is easily absorbed in the pericarp and can play avital role in the prevention of oxidative enzymes activity, results partially confirm those of [5].

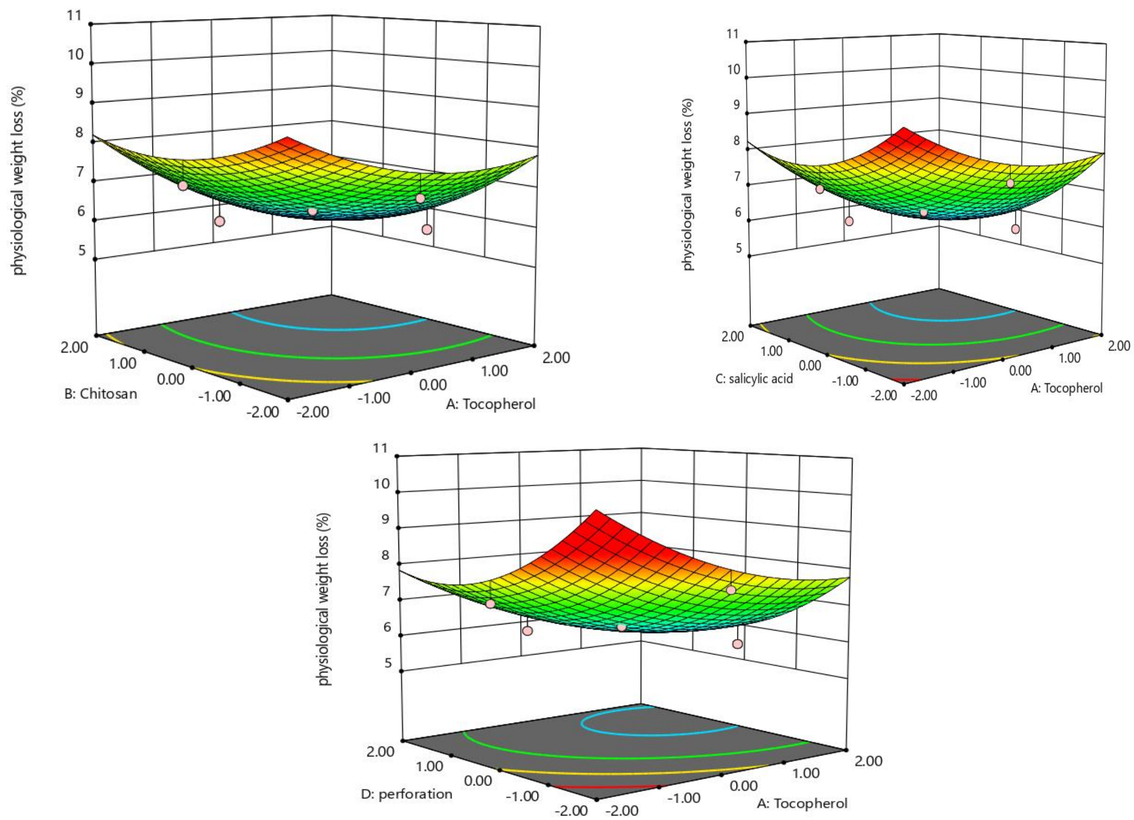

Figure 1 Effect of independent variables on physiological weight loss under ambient storage condition

The chitosan forms a thin layer around coated fruits which, maintained humidity around the fruits by preventing its exposure to the external factors like temperature, oxygen which, influences rates of respiration and evapo-transpiration i.e. direct loss in weight. The chitosan has definite moisture absorption and retention qualities in response to that the coated litchi produced less bio-heat [6].

The salicylic acid maintained membrane integrity by maintaining the activities of three main cell wall degrading enzymes namely cellulose, polygalacturonase (PG) and xylanase, which tends to less electrolyte leakage.

Weight loss increases with increase in perforation percentage as the availability of oxygen increases cause higher oxidative enzymes activity and more utilization of substrate for enzymatic activity. The similar results were reported by [7], the loss in weight due to a major loss of water from pericarp and mesocarp, packaging modified atmosphere around fruits increases CO_2 concentration acts as an inhibitor of oxidative enzyme activity. The significant effect of perforation percentage was recorded in a loss in physiological weight of packaged coated litchi fruits, [8] reported that plastic films affected weight loss of litchi during storage. The unwrapped fruits had the higher moisture loss and high enzymatic activity utilizes more substrates are one of the major cause of physiological loss in weight. Increases in weight loss over storage was also recorded in litchi by [9].

Effect of independent variables on firmness

The loss of water from pericarp results into minute cracks on it and causes disruption of compartmentation, leads to more water loss and desiccation of pericarp. There is the absence of conducting tissues between pericarp and mesocarp in litchi, which fails the pericarp to replenish water.

The range of firmness of coated litchi fruits were varied between 15.52 to 21.47 N. The maximum firmness recorded in experiment number 13 with experimental conditions of LD α-Tocopherol 0.2%(A=-1), 1% chitosan (B= -1), salicylic acid 1 mM (C= -1) and perforation 0.2% (D=-1) whereas the minimum (15.52) with experimental conditions of LD α-Tocopherol 0.4% (A=1), 2% chitosan (B=1), salicylic acid 2 mM (C=1) and perforation percentage of0.4% (D=1)at experiment no. 9 (Table 3).

The data of firmness recorded from packaged coated litchi fruits stored under ambient storage condition was fitted into full second-order mathematical model equation (1) and the result of regression analysis was expressed by equation (4). The coefficient of determination (R^2) for the regression model for this parameter was 73.20% and adjusted R^2 was 48.19%, which insinuate that the model could account for 73.20% data. The model was found to be significant at 1% level of significance with non- significant lack of fit. Therefore, the second-order model was considered to be capable of describing the change in firmness with the specified values of independent parameters.

$$Firmness = 16.98-0.86\ A-0.75\ B-0.44\ C-0.59\ D + 0.24AB + 0.20AC + 0.16\ AD + 0.18\ BC+ 0.15\ BD - 0.08CD + 0.11A^2 + 0.15B^2 + 0.06\ C^2 + 0.26\ D^2 \quad (4)$$

Where, A is LD α-Tocopherol, B is chitosan, C salicylic acid and D is perforation percentage (all in coded form).

Table 5 expresses the individual effect of each term in second-order quadratic equation fitted to the experimental data.

Table 5 Effect of treatments on firmness under ambient storage conditions

Source	SS	Df	MS	F-value	P-value
Model	50.42	14	3.60	2.93	0.0238**
A-α-Tocopherol	17.88	1	17.88	14.53	0.0017***
B-Chitosan	13.69	1	13.69	11.13	0.0045***
C-Salicylic acid	4.65	1	4.65	3.78	0.0708
D-perforation	8.59	1	8.59	6.98	0.0185**
AB	0.9945	1	0.9945	0.8083	0.3828
AC	72.00	1	0.6669	0.5420	0.4729
AD	5.47	1	0.4181	0.3398	0.5686
BC	15.77	1	0.5602	0.4553	0.5101
BD	14.45	1	0.3887	0.3159	0.5824
CD	495.70	1	0.1104	0.0897	0.7687
A^2	0.3907	1	0.3907	0.3175	0.5814
B^2	0.6829	1	0.6829	0.5550	0.4678
C^2	0.1248	1	0.1248	0.1015	0.7545
D^2	1.94	1	1.94	1.58	0.2280
Residual	18.45	15	1.23		
Lack of Fit	NS				
R^2	0.7320				
Adj R^2	0.4819				

*** 1% level of significance, **5% level of significance

It is revealed that at linear levels two independent variables α-Tocopherol and chitosan were affected significantly with 1% of the level of significance, whereas the perforation percentage showed significance at 5% level of significance. Amongst the linear levels, salicylic acid had no significant effect on the firmness of coated litchi fruits. It might be due to the prevention of water loss from pericarp by chitosan and α-Tocopherol (source of vitamin E) which prevents enzymatic activity responsible for drying of fruit pericarp. There was no any significant effect of independent variables on the firmness of coated litchi fruits at interactive and quadratic levels. The results were in accordance with [10]. Therefore, simplified second order equation of firmness becomes,

$$Firmness =16.98 - 0.86A - 0.75B - 0.59D \quad (5)$$

Response surface plots as shown in **Figure 2** accustomed the effect of selected independent variables on the firmness of packaged coated litchi fruits stored at ambient condition. The graph clearly showed that firmness of the treated fruit decreased slightly at the initial increase in tocopherol percentage. It was also found that the increase in chitosan decreases the firmnessup to its central value and became stable thereafter. The α-Tocopherol and chitosan in combination preventwater loss from pericarp and replenish pericarp externally by maintaining high humidity around coated fruits [11]. The α-Tocopherol bioactive compound might prevent enzymes which degrade the cell wall and affect firmness, in apple [12]. The increase in perforation percentage up to its central value tends to decrease in firmness, while it was nearly constant thereafter. The combined effect of antimicrobial agents with the packaging of perforated bioriented polypropylene litchi fruits stored for 18 days under 2^0C and 95% relative humidity, maintained the firmness of fruits [13].

Optimization of independent variables

The firmness was maintained up to 15 days in treated fruits like fresh fruits, showed its suitability and an appropriate proportion of α-Tocopherol, chitosan and salicylic acid along with proper perforation percentage on packaging polyethylene.

Among all the optimized solutions, the best optimized solution for optimum values of independent variables was selected on the basis of the criteria that the optimum values should be close to viable values at higher desirability. To optimize the process parameters, the goal was fixed to be maximize or minimize **Table 6**.

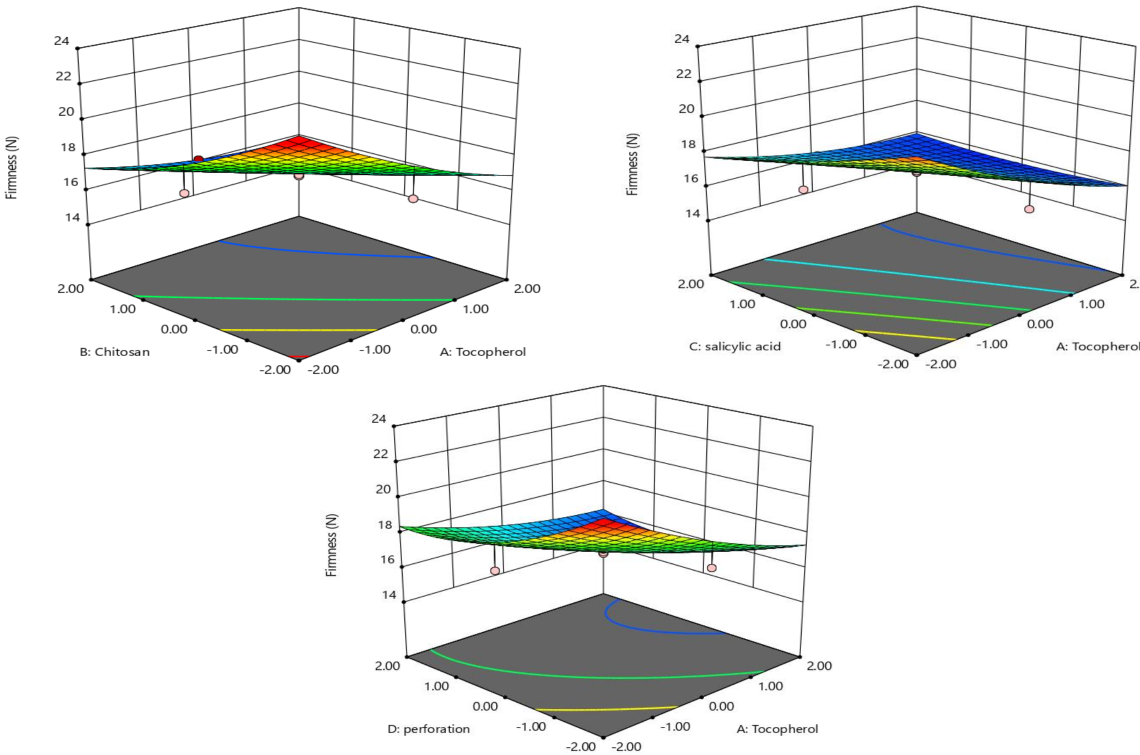

Figure 2 Effect of independent variables on firmness under ambient storage condition

All the responses and independent variables were given similar (+++) importance. If the response selected to be maximum, upper limit was taken as maximum value of the response and lower limit was taken as the 25% less of the upper limit of response. If the response required is minimum, lower limit was considered as minimum value of response and upper limit was 25% more of the lower limit.

Based on mentioned criteria, the optimization was carried out. During optimization, 28 solutions were obtained in case of coating of the litchi fruit and its storage at ambient condition, out of which the one that suited the criteria most was selected. The most suitable optimum point is given in the **Table 7**.

Table 6 Constraints for optimization coating formulation for ambient storage of litchi

Name	Goal	Lower limit	Upper limit
α-Tocopherol	is in range	-2	2
Chitosan	is in range	-2	2
salicylic acid	is in range	-2	2
perforation	is in range	-2	2
Physiological weight loss	minimize	5.23	8.81
Firmness	minimize	15.51	21.89

Table 7 Optimum value of parameters for coating formulation applied on litchi fruit

Value	α-Tocopherol, % (X_1)	Chitosan, % (X_2)	Salicylic acid, mM (X_3)	Perforation, % (X_4)
Coded	1.0	1.0	1.0	1.0
Actual	0.4	2	2	0.4
Observations recorded at optimum levels of independent parameters during ambient storage for 15 days				
Physiological weight loss (%)			5.23	
Firmness			15.73	

The optimum results of coating formulation applied on litchi fruit includes 0.4% of α-Tocopherol, 2% of chitosan, 2 mM of salicylic acid and 0.4% of perforation on packaging material. Whereas the fruits under control condition starts losing its quality from second day onwards on the day 3rd most of the fruits were not suitable for marketing and becomes brown.

Harvested litchi fruits of cv. Rose scented

Coated litchi fruits of cv. Rose scented

Control fruits at 3rd day of storage under ambient condition

0.4 % α-Tocopherol + 2.0 % Chitosan + 2.0 mM Salicylic acid + 0.4% Perforation at 15th day of storage under ambient condition

Figure 3. Representative photos of the experiments.

Conclusion

The study includes optimization of coating and packaging of litchi for better shelf life under ambient conditions. The litchi fruits maintained sufficient fruit weight and firmness at 15th day of storage, during experiment. Whereas the control fruits were stored under ambient conditions starts decaying after 2 days. The coating formulation along with packaging extends shelf life of litchi by 12 days under ambient conditions.

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