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Effect of coating formulations and packaging perforations on physiological properties of Litchi under cold storage conditions

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Abstract
The application of coating and packaging methodologies exerted a substantial influence on the physiological weight reduction of litchi fruits subjected to various treatments, exhibiting a discernible range of 3.31% to 5.74%. Notably, the pivotal attribute of firmness, a determinant factor influencing market acceptance, was found to be intricately linked to the water content and post-harvest metabolic alterations in litchi specimens. Within the realm of cold storage conditions, the hardness of coated litchi fruits demonstrated a spectrum spanning from 15.51 to 21.89 N. Noteworthy is the observation that the minimum hardness (15.51 N) was achieved under specific experimental parameters, including α-Tocopherol at a concentration of 0.5%, Chitosan at 1.5%, salicylic acid at 1.5 mM, and a perforation percentage of 0.3%.

Keywords: Litchi, α-Tocopherol, salicylic acid, perforation, physical properties

Introduction

Litchi belong to the family of Sapindaceae, which includes certain cultivated fruit species such as longans and rambutans. It is a native of the China's Guangxi and Fujian regions, now cultivated in many parts of the world. In India, Nepal and Bangladesh, trees have been planted. Because of white, translucent arils and its attractive red colour *Litchi Chinensis* Sonn. is an extremely desirable subtropical fruit which can be exported in large quantities.

Litchi is highly perishable due to its delicate nature. Its shelf life under ambient conditions does not exceed 24-72 hours.Postharvest decay and loss of physiological weight have been identified as the main problems limiting the expansion of the industry in litchi exporting countries. Firmness is an important factor that determines market acceptance and is related to the water content and metabolic changes of litchi after harvest. The strength observed showed a continuous increase in storage due to the loss of moisture in the pericarp, which makes it dry and rigid.The aim of this work was to optimize the coatings formulations and packaging perforations to study physiological loss in weight and firmness of litchi fruits and to extend its shelf life under cold storage conditions for expansion of its market.

Materials and Methods

Experimental Materials

The fruit were collected from well managed trees of Litchi cv. Rose Scented planted at Horticultural Research Centre, Patharchatta, Department of Horticulture, G.B. Pant University of Agriculture and Technology, Pantnagar. The university located 28˚58'N to 79˚25'E or 28.97˚ N to 79.41˚ E in the Tarai region of Uttarakhand state of India.

Experimental Procedure

The litchi was harvested at ripe stage (90 – 100% of the peel exhibiting red colour), 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. After selecting uniform fruits based on size, shape, colour and free from diseases or blemishes they were treated with aqueous solutions of α-Tocopherol, Chitosan and salicylic acid, in combination according to the treatment details.

Treatments were executed by dipping the fruits in 5 liter treatment solutions containing Tween-20 (2 g L^{-1}) as surfactant, at 25 °C for 5 min. Whereas, control fruits were dipped in distilled water.

Experimental Plan

The variables that have greatest influence on the response could be selected as operating parameters of final experiments [1]. The variables standardized on the basis of preliminary experiments and also on the basis of review of literature. α-tocopherol, Chitosan, salicylic acid, and perforation percentage of packaging material were selected as the independent variables with five levels each (Table 1).

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 in weight Firmness
2	Chitosan (%)	5	0.5,1.0,1.5,2.0,2.5	
3	Salicylic acid (mM)	5	0.5,1.0,1.5,2.0,2.5	
4	Perforation (%)	5	0.1,0.2,0.3,0.4,0.5	

Measurement of Dependent Variables

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 different 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.

$$\text{Physiological loss in weight (\%)} = \frac{\text{Initial weight} - \text{Weight after known storage} \times 100}{\text{Initial weight}} \quad (1)$$

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 curvewas plotted between force and time and maximum force was noted. The data were presented a means of 10 independent measurements.

Experimental Design

Experiments were designed using Central Composite Rotatable design (CCRD) method for satisfying the principle of randomization of Response Surface Methodology (RSM).

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

The 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:

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

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

The regression coefficients of complete second order model and their significance are reported in respective response tables. 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 behavior of response, otherwise, transformation of the dependent variable was performed [1].

Results and Discussion

Effect of independent variables on physiological loss in weight

The major reason for weight loss in litchi fruit was the loss of moisture from pericarp causes minute cracks on pericarp and further moved inward up to parenchymatous tissues of fruit. Fruit coated with formulation recorded lower weight loss which might be due to coating formulations reduced the transpiration and respiration rates of litchi fruits by closing the stomata of the treated fruit.

The maximum physiological loss in weight obtained at experiment number 13 with experimental conditions of α-Tocopherol 0.2%, 1% chitosan, salicylic acid 1 mM and perforation 0.2% while minimum (3.31) with experimental conditions of α-Tocopherol 0.3%, 1.5% chitosan, salicylic acid 1.5 mM and perforation percentage of 0.3% at experiment no. 8 (Table 3).

Physiological weight loss data obtained from coated litchi fruit stored under cold storage condition was fitted into full second-order mathematical model equation (2) and the result of regression analysis was represented by equation (3). The coefficient of determination (R^2) for the regression model for this parameter was 76.72% and adj R^2 was 55.00%, which implies that the model could account for 76.72% data. The model was found to be significant at 5% 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.

Physiological loss in weight = 3.64 - 0.37A - 0.18B - 0.12C - 0.11D - 0.078AB - 0.014AC -0.011AD + 0.12BC +

$0.054BD - 0.016CD + 0.21A^2 + 0.16B^2 + 0.15C^2 + 0.19 D^2$ (3)

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

Table 4expresses the individual effect of each term in second-order quadratic equation fitted to the experimental data. It is revealed that at linear levels α-Tocopherol and chitosan affected significantly with 1% and 5% level of significance respectively. While at quadratic levels α-Tocopherol, chitosan and perforation percentage of packaging material had a significant effect on the weight loss at 5% level of significance. The results were in accordance with [2]. The simplified second order equation of physiological weight loss becomes,

$$\text{Physiological loss in weight} = 3.64 - 0.37A - 0.18B + 0.21A^2 + 0.16B^2 + 0.19 D^2 \quad (4)$$

Table 3: Experimental data on optimization of coating and packaging perforation of litchi for cold storage condition

Treatments	α- Tocopherol (%)	Chitosan (%)	Salicylic acid (mM)	Perforation (%)	Physiological loss in weight (%)	Firmness (N)
1	0.2	2	1	0.2	4.76	19.78
2	0.5	1.5	1.5	0.3	3.67	15.51
3	0.3	1.5	1.5	0.3	3.64	16.55
4	0.1	1	2	0.4	3.66	16.43
5	0.4	2	1	0.2	3.98	16.95
6	0.4	1	1	0.2	4.98	18.48
7	0.4	2	2	0.2	3.86	16.44
8	0.3	1.5	1.5	0.3	3.31	16.37
9	0.3	1.5	0.5	0.3	4.17	16.58
10	0.3	1.5	1.5	0.3	3.85	16.35
11	0.4	2	1	0.4	3.69	16.26
12	0.3	1.5	1.5	0.3	3.88	16.17
13	0.2	1	1	0.2	5.74	21.26
14	0.2	1	1	0.4	4.85	21.46
15	0.4	2	2	0.4	3.63	15.73
16	0.2	2.5	1	0.4	4.86	17.25
17	0.3	2.5	1.5	0.3	3.78	16.12
18	0.3	1.5	1.5	0.3	3.71	16.42
19	0.4	1	2	0.2	4.45	17.29
20	0.3	0.5	1.5	0.3	4.33	17.39
21	0.4	1	1	0.4	4.31	18.57
22	0.2	2	2	0.3	5.28	17.56
23	0.5	1.5	1.5	0.3	4.82	17.66
24	0.3	1.5	1.5	0.5	4.42	21.89
25	0.3	1.5	1.5	0.3	3.49	16.28
26	0.3	1.5	1.5	0.3	3.88	16.18
27	0.3	1.5	1.5	0.1	3.93	17.82
28	0.2	0.5	1.5	0.2	4.76	20.74
29	0.2	0.5	1.5	0.4	4.79	17.59
30	0.2	2	1.5	0.4	4.25	16.88

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

Source	SS	Df	MS	F-value	P-value
Model	7.85	14	0.5611	3.53	0.0104**
A-α-Tocopherol	3.40	1	3.40	21.39	0.0003***
B-Chitosan	0.7812	1	0.7812	4.92	0.0424**
C-salicylic acid	0.3927	1	0.3927	2.47	0.1367
D-perforation	0.3243	1	0.3243	2.04	0.1735
AB	0.0977	1	0.0977	0.6147	0.4452
AC	0.0033	1	0.0033	0.0208	0.8872
AD	0.0023	1	0.0023	0.0142	0.9067
BC	0.2377	1	0.2377	1.50	0.2402
BD	0.0473	1	0.0473	0.2978	0.5933
CD	0.0046	1	0.0046	0.0287	0.8678
A^2	1.22	1	1.22	7.65	0.0144**
B^2	0.7289	1	0.7289	4.59	0.0490**
C^2	0.6634	1	0.6634	4.18	0.0590
D^2	1.02	1	1.02	6.43	0.0228**
Residual	2.38	15	0.1589		
Lack of Fit	NS				
R^2	0.7672				
Adj R^2	0.5500				

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

It is clear from the graph Fig 1that physiological weight loss of the treated fruit decreased rapidly at the initial increase in tocopherol percentage, at the same time increase in chitosan showed similar effect on physiological weight loss. The salicylic acid and perforation percentage increase did not showed much fall in physiological loss in weight, but slight changes in curve can be seen.

The tocopherol significantly affected the physiological loss in weight [3]. It inhibits the respiration rate initially and inhibits catalase enzyme activity, which is responsible for breaking down the complex structure into simple, which can be used during respiration and one of the major causes of loss in weight. Similar tendencies were noted when edible films and coatings were applied to numerous items to regulate oxidation, gas exchange, and moisture transfer processes [4]. Chitosan, which forms a thin film around fruits that have been coated with it, maintains humidity around the fruit by keeping it from being exposed to outside elements like temperature, oxygen, etc., which can have an impact on the rate of respiration and evapo-transpiration, which results in a direct loss in weight.

Because of the chitosan's moisture absorption and retention properties, the coated litchi produced reduced bio-heat [5, 6], [7]. The organic acid (citric or tartaric) treatments in conjunction with 1% chitosan coating [8]. Tartaric acid and 1% chitosan treatment reduced fruit weight loss (1.4%) following 8 days of storage at 10 °C.
The lower temperature of 7 °C may superimpose the effect of salicylic acid as the rate of respiration, reduced along with enzymatic activities. Salicylic acid reduces the rate of respiration by reducing postharvest oxidative stress, which ultimately delayed senescence. The salicylic acid preserved membrane integrity by preserving the activity of three major cell wall-degrading enzymes: cellulose, polygalacturonase (PG), and xylanase [9]. This has the effect of reducing electrolyte leakage.
The weight loss from pericarp and mesocarp water loss, packaging modifications, and an increase in CO2 concentration in the environment around fruits all serve to inhibit the activity of oxidative enzymes [10]. The physiological weight of packaged coated litchi fruits was lost, which is a significant result of the perforation percentage. Unwrapped fruit lost more moisture, and high enzymatic activity, which uses more substrates, is one of the main factors contributing to physiological weight loss.

Effect of independent variables on firmness

The main reason for this is pericarp moisture loss. In litchi, the pericarp is unable to replenish water because there are no conducting tissues between the pericarp and mesocarp. Because of the pericarp's desiccation and hardening during storage, the firmness of litchi fruits increased.
The results of experimental data obtained by the response variables were shown in Table 3. Firmness of coated litchi fruits stored at cold storage condition increases with increase in storage time in the range of 15.51 to 21.89 N. The maximum firmness recorded in experiment number 24 with experimental conditions of α-Tocopherol 0.3%, 1.5% chitosan, salicylic acid 1.5 mM and perforation 0.5% whereas the minimum (15.51) with experimental conditions of α-Tocopherol 0.5%, 1.5% chitosan, salicylic acid 1.5 mMand perforation percentage of0.3% at experiment no. 2.
The data of firmness recorded from packaged coated litchi fruits stored under cold storage condition was fitted into full second-order mathematical model equation (2) and the result of regression analysis was expressed by equation (5). The coefficient of determination (R^2) for the regression model for this parameter was 73.94% and adjusted R^2 was 49.62%, which insinuate that the model could account for 73.94% data. The model was found to be significant at 5% 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.

$$Firmness = 16.35 - 0.86A - 0.72B - 0.50C - 0.007D + 0.26AB + 0.16AC + 0.24AD + 0.25BC - 0.055BD - 0.15CD + 0.16A^2 + 0.20B^2 + 0.11C^2 + 0.98D^2 \quad (5)$$

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

Table 5: Effect of treatments on firmness (N) under cold storage conditions

Source	SS	Df	MS	F-value	P-value
Model	67.38	14	4.81	3.04	0.0202**
A-α-Tocopherol	17.80	1	17.80	11.25	0.0044***
B-Chitosan	12.78	1	12.78	8.07	0.0124**
C-salicylic acid	6.15	1	6.15	3.89	0.0674
D-perforation	0.0015	1	0.0015	0.0010	0.9758
AB	1.10	1	1.10	0.6931	0.4182
AC	0.4258	1	0.4258	0.2689	0.6116
AD	0.9950	1	0.9950	0.6285	0.4403
BC	1.05	1	1.05	0.6604	0.4291
BD	0.0495	1	0.0495	0.0313	0.8620
CD	0.3813	1	0.3813	0.2409	0.6307
A^2	0.7477	1	0.7477	0.4723	0.5024
B^2	1.18	1	1.18	0.7468	0.4011
C^2	0.3556	1	0.3556	0.2246	0.6424
D^2	26.48	1	26.48	16.73	0.0010***
Residual	23.75	15	1.58		
Lack of Fit	NS				
R^2	0.7394				
Adj R^2	0.4962				

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

Table 5 expresses that at linear levels α-Tocopherol were affected significantly with 1% and while chitosan 5% of the level of significance respectively. Amongst the linear levels, salicylic acid and perforation percentage had no significant effect on the firmness of coated litchi fruits stored under cold storage condition. Therefore, simplified second order equation of firmness becomes,

$$Firmness = 16.35 - 0.86A - 0.72B + 0.98D^2 \quad (6)$$

The graph Fig. 2clearly showed that firmness of the treated fruit droppedacutly with the initial increase in tocopherol and chitosan percentage. The α-Tocopherol and chitosan in combination preventwater loss from pericarp and replenish pericarp externally by maintaining high humidity around coated fruits [4]. The α-Tocopherol bioactive compound might prevent enzymes which degrade the cell wall and affect firmness [11]. The combined effect of antimicrobial agents with the packaging of perforated bioriented polypropylene litchi fruits stored for 18 days under 2 °C and 95% relative humidity, maintained the firmness of fruits [12]. The equilibrium of O_2 and CO_2 maintained at certain perforation (0.3%), below or above it affects respiration rate of fruits [13].

Conclusion

In the cold storage the physiological loss in weight and firmness were better reduced up to 20 days in treated fruits like fresh fruits, showed its suitability and an appropriate proportion of α-Tocopherol, ChitosanandSalicylic acid along with proper perforation percentage on packaging polyethylene.

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