RESEARCH PAPER

Effect of Packaging Materials and Storage on Quality Attributes of Freeze Dried Pineapple *Lassi* Powder for Defence Applications

V.K. Shiby[*], V. Jayashree, S. Sanchita, and M.C. Pandey

Freeze Drying and Animal Products Technology Division, Defence Food Research Laboratory, Mysuru - 570 011, India
[*]E-mail: shibyk@gmail.com

ABSTRACT

The study evaluates the effect of two different packaging materials on storage stability of and instant fruit-dairy functional beverage mix. Freeze dried pineapple *Lassi* (PL) powder was packaged in aluminium laminated polyethylene (ALP) and metallised polyester (MP). Quality changes at elevated temperature conditions (38±1 °C, 33 per cent relative humidity) were evaluated. Shelf life was calculated to be 44 days and 62 days in MP and ALP pouches, respectively. Moisture, water activity, Vitamin C, Lactococci, TBA, colour change and browning index were studied in both packaging materials. ALP was found to be better than MP as samples kept in ALP preserved most of the antioxidant properties. The kinetics of Vitamin C degradation was of first order (K = -0.017, R^2 = 0.99). Moisture content increased and *lactococci* count decreased during storage.

Keywords: Freeze drying; Pineapple; Fermented; Storage; Quality

NOMENCLATURE

PL	Pineapple *lassi*
ALP	Aluminium laminated polyethylene
MP	Metallised polyester
TBARS	Thiobarbituric acid reaction substances
RH	Relative humidity
$dw/d\theta_p$	Slope of the linear plot between the storage time (days) and weight increase of silica in respective packaging material
A_P	Area of packaging surface exposed to storage environment (m^2),
P^*	Saturation vapour pressure of water at temperature of storage (37 °C)
L_1, a_1, and b_1	Initial colour values
$L_2 \, a_2$, and b_2	Colour values after storage
M_0	Monolayer moisture content (db)
M	EMC (db)
C, K	Constants
ANOVA	Analysis of variance
K_0	Zero order reaction constant
K_1	First order reaction constant
C_0	Initial concentration
C	Concentration

1. INTRODUCTION

Fermented milk beverages should be light, low calorie, refreshing (thirst quenching), and should have good mouth feel, taste and smooth texture. Hybrid dairy products, such as 'juiceceuticals' containing fruit and dairy ingredients offer health, flavour and convenience[1]. *Lassi* (Indian Yoghurt drink)

is a popular and traditional dahi-based drink containing mixed mesophilic cultures of, *Lactococcus lactis subsp. cremoris, Lactococcus lactis subsp diacetylactis Lactococcus lactis subsp. lactis* along with flavour producing *Leuconostoc* species. A blend of *Lassi* with pineapple *(Ananas cosmosus)* juice has increased nutrient content including minerals and vitamins and also serves as a healthier product combining phytonutrients with dairy protein and calcium.

Freeze drying technology takes care of retention of antioxidants from fruit components and healthful lactic acid bacteria from the fermented dairy product and makes this product suitable for consumption in high altitude regions and desert areas where soldiers are deployed.

Keeping quality of dehydrated food powders is influenced by water activity. During storage, maillard reaction causes colour changes such as browning and also leads to development of an off flavours. Several insoluble compounds may be formed and lump formation occurs which reduce the acceptability of powders. Several authors have studied the retention of starter organisms, vitamins, antioxidants, occurrence of colour and oxidative changes in food powders during storage[3,4,5]. Secondary oxidation products are quantified in terms of Thiobarbituric acid reactive substances (TBARS).

In the present study, the stability of pineapple *Lassi* (PL) powder packaged in two different packaging materials viz. metallised polyester (MP) and high barrier aluminium foil laminated polyethylene (ALP) pouches and stored under elevated temperature conditions is discussed. As high RH condition of 90 per cent RH resulted in caking of the freeze dried product within a week, the conditions selected for accelerated storage studies in the present work are 38 ± 1 °C, 33 ± 1 per cent relative humidity.

2. MATERIALS AND METHODS

2.1 Preparation of Pineapple *Lassi* Powder

Pasteurised Pineapple juice (18% total solids), curd (8.2% total solids) and sugar were blended. Details of the methodology for the preparation of PL and its blend formulation and optimization have been published elsewhere[6]. Freeze drying (16 h) of fruit *Lassi* beverages was carried out in a freeze dryer (Martin Christ, Germany) maintaining a vacuum of 0.1 mm of Hg, fruit *Lassi* flakes were ground in a mill and sieved (40-mesh) to obtain uniform particle size. It was then packed in MP and ALP pouches for further studies.

2.2 Permeability of Packaging Material

Moisture gain data of silica gel in respective packaging material was collected during 7 days and water vapour Permeability of packaging material was computed by Eqn (1).

$$K = \frac{dw/d\theta p}{A_p P^*} \tag{1}$$

where $dw/d\theta_p$ =slope of the linear plot between storage time and weight increase of silica gel, A_P= Package area exposed to storage environment (m^2), and P^*=saturation vapour pressure of water at the temperature of the environment in which the sample is stored.

2.3 Determination of Changes during Storage

Moisture content was determined by gravimetric method. Thiobarbituric acid (TBA) value was expressed as OD measured at 532 nm as per the method[7]. A browning index (BI) was measured as absorbance at 420 nm. Instrumental colour of PL samples was measured with a Hunter Lab colorimeter. The colour values were monitored periodically during storage at elevated temperature of colour change (ΔE_c) occurred during storage was calculated using Eqn. (2)

$$\Delta E = \sqrt{(L1 - L2)^2 + (b1 - b2)^2 + (a1 - a2)^2} \tag{2}$$

$a1$, and $b1$ are the initial colour values for the *Lassi* powder samples and $L2$, $a2$, and $b2$ are the corresponding values after storage. Enumeration of *lactococci* was done using M17 Agar procured from Hi Media Laboratories Limited, Mumbai.

2.4 Shelf Life of PL Powder

Adsorption isotherms for PL powder at 40 °C were obtained by determining Equillibrium moisture content at varying RH. The GAB model (Eqn 3) was used for fitting the experimental aw and EMC data.

$$M_e = \frac{M_0 \ Cka_w}{(1 - ka_w + Cka_w)} \tag{3}$$

M_0 = Monolayer moisture content (db), M = EMC (db) and, C and K are model constants.

Parameters of the non linear equation were calculated using Sigma Plot 13 software (Starcom, Bengaluru) 20 g *Lassi* powder samples were packed in MP and ALP pouches (11.5 cm × 12 cm) . The packets were heat sealed carefully to make them leakproof and airtight. They were kept at 33 ± 1 per cent RH using a dessicator containing saturated Magnesium chloride solution. The dessicators were thermostatically maintained at 38 ± 1 °C in an incubator. It was ensured that pouches were held individually and are exposed to the same environment.

In weekly intervals, upto 42 days, one packet each was taken out from dessicator and analysed for all parameters.

The shelf life was calculated using the following Eqn (4)

$$\int d\theta = \frac{W_s}{P^* kA} \left(\int_{x1}^{x2} \frac{dx}{RH - a_w} \right) \tag{4}$$

where θ = shelf life (days), W_s = weight of the dry solids (g), P^* = saturated vapour pressure of water at ambient temperature (Pa), k = permeability of the packaging material (kg m^{-2} day^{-1} Pa^{-1}), A = area of the package (m^2), RH = relative humidity of the environment in which the package is placed (%), a_w = water activity of the product, X_i = initial moisture content (%, db) and X_c = critical moisture content (%, db).

2.5 Statistical Analysis

The statistical significance of the variation between fresh and stored samples were stabled using analysis of varience (ANOVA). Effect of packaging material and storage period were evaluated and the corresponding models were obtained. The interaction effects of the main factors were also considered.

Linear regression analysis was carried out to fit zero order and first order reaction kinetics for degradation of Vitamin C and other quality parameters.

3. RESULTS AND DISCUSSION

The moisture uptake in different packaging materials (Fig. 1) was analysed periodically and the variance of the data (Table 1) showed that both storage period and the packaging material had significant effect on the moisture gained during storage. Comparing the permeability of the materials used, it can be observed that MP has higher permeability (2.01×10^{-7} Kgm^{-2} day^{-1}Pa^{-1}) than ALP (1.8×10^{-8} Kg m^{-2}day^{-1}Pa^{-1}). During storage, caking was observed beyond a moisture content of 5.8% (db), leading to lump formation. The moisture gain in MP pouches was more significant ($P<0.01$) than in ALP pouches. The data generated with respect to moisture content of the
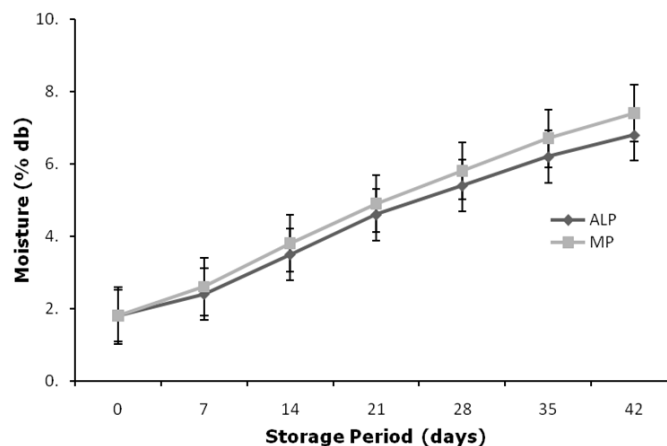

Figure 1. Variation in moisture content of PL powder packed in MP and ALP pouches during storage at 38 ± 1 °C and 33 ± 1 per cent RH.

Table 1. Analysis of Variance data for physico-chemical properties and culture counts of PL powder during storage under accelerated condition (38± 1 °C and 33± 1 per cent RH).

Source of variation	df	Moisture content	Vitamin C	TBA	Browning index	Colour change (ΔE)	*Lactococci*
Storage period (SP)	6	4.32**	456.91**	0.009*	2125.3**	20[ns]	268.2**
Packaging material (PM)	1	1.75**	50.2**	0.003*	532*	15[ns]	10052**
SP	6	0.008**	2.06[ns]	0.0001[ns]	48**	124.18**	68.34*
Error	15	1.78×10^{-5}	0.23	4.6×10^{-5}	2	1.4	1.5

** Mean significant at $P<0.01$. *$P<0.05$, **$P<0.01$

powder is important in determining its handling properties also. Water exerts a plasticising effect in food powders and most of the freeze dried powders liquefy at high relative humidity conditions. Liquefaction was not observed in Freeze dried pineapple *Lassi* powder subjected to accelerated storage study. After 6 weeks of storage, he percentage moisture reached 6.4 per cent on dry basis in MP, where as the corresponding value was only 5.8 per cent on dry basis in ALP. The loss of free flowness of the powder was considered as a limiting factor determining its shelf life. Hence the moisture in the range 5.8-6.2 per cent was found to be critical during the storage of freeze dried pineapple *Lassi* powder, beyond which, powder exhibited lump formation and caking behaviour.

3.1 Quality Changes in PL Powder during Storage at Elevated Temperature

The initial acidity of PL powder was 3.1 per cent lactic acid. The change in acidity in PL powder during accelerated storage is shown in Fig. 2. The acidity increased to 3.9 per cent and 3.7 per cent after 42 days of storage in MP and ALP, respectively.

Free fatty acids liberated due to breakdown of fat muscles result in hydrolytic rancidity of the product. As more and more moisture got absorbed, there was an increase in acidity of the powder. This leads to sour taste in the fermented beverage mix which can reduce its sensory acceptability. When moisture was above 6 per cent (db), the increase in acidity was too high. Such changes are accelerated under elevated temperatures compared to room temperature storage. At 33 per cent RH and 37 °C storage, both packaging material and storage time had a significant effect on acidity value which was correlated with the moisture uptake.
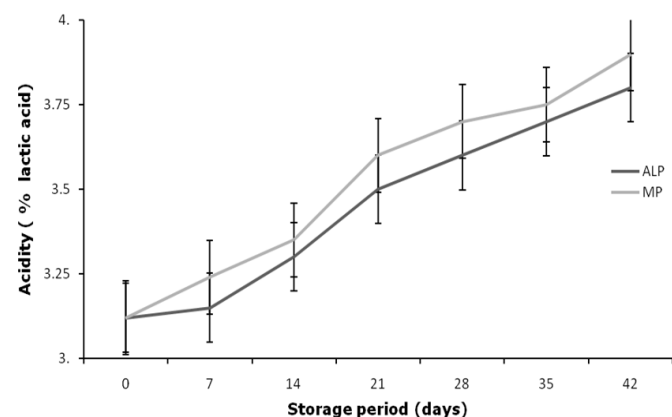
TBA value expressed as optical density at 532 nm (Table 1) increased to 0.23 and 0.2 at the end of storage in MP and ALP respectively. This implies an adverse sensory impact due to lipid oxidation which proceeds to form secondary oxidation products like carbonyls. Accelerated storage condition aggravates these deteriorative changes so that the research can predict the shelf life within a short period of experimentation compared to room temperature of food powders.

Several authors have quantified TBA values during storage of food powders. But it is extremely difficult to prescribe a cut off value for TBA which is applicable to all food products. It varies from product to product at what value of TBA that the rancidity becomes perceptible in a particular product. Hence it is important to correlate TBA values with sensory data to make a conclusive statement regarding this change in Freeze dried pineapple *Lassi* powder. The results indicate that both the type of packaging material used and the advance in storage time have significant effects on quality of powder in terms of its susceptibility to oxidative deterioration. (Table 1). As TBA increases, increased autoxidation rate and Maillard reaction can lead to protein polymerisation in food powders containing dairy ingredients, in effect decreasing the powder solubility during storage.

Non enzymatic browning is dependent on the packaging material used. With the lapse of time under accelerated condition, there observed significant changes in browning index expressed as absorbance at 420 nm. Hydroxy methyl furfural (HMF) is considered as an index of browning in whole milk powder and related products. An increase of HMF has been reported by a factor of 3 in whole milk powder and this increase is also reported in partially skimmed milk powder containing
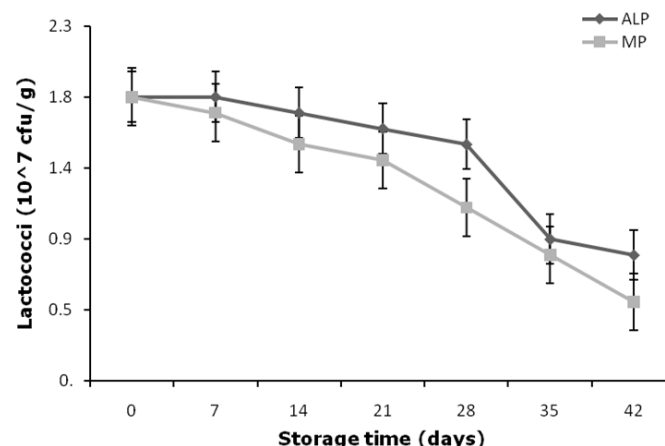

Figure 2. Changes in acidity of PL powder in MP and ALP pouches during storage at 38±1 °C and 33 ± 1% RH.

Figure 3. Culture count in PL powder in MP and ALP pouches during storage at 38±1 °C and 33 ± 1 % RH.

50 per cent total solids from skim milk[8]. Approximately 10 fold increase in HMF content of whey protein concentrate under accelerated storage conditions (37 °C and 75 per cent RH. 42 days) has also been reported in previous studies[9]. Changes during storage of wholemilk powder have been quanitified[10.]

Initially, *Lactococci* count in PL powder was 1.8×10^7 cfu/g, respectively .The viability of useful organisms is significantly affected by type of packaging and storage period. (Table 1) The therapeutic minimum for cultured product is 10^6cfu/ml. In the present study, *lactococci* count decreased to 7×10^3 and 9.8×10^4 cfu/g after 6 weeks of storage in MP and ALP respectively. The *Lactococci* count in FD- pineapple *Lassi* powder decreased below the therapeutic minimum suggested for processed foods, after 6 weeks of storage. For yogurt bacteria the optimum water activity values[11] is reported to be 0.2 and they survive better during chilled storage than at room temperature. Based on viability of lactococci the powder stability was limited to 35 days in MP and 42 days in ALP, respectively. Studies on effect of additives on the survival during freeze-drying and stability during storage of four lactic acid bacteria revealed that additives like gelatin significantly improve viability of *Lactococcus lactis* ssp. *lactis* RO58, following 12 months at 4 °C, were higher than controls. Overall, the additives showed a detrimental effect on the stability of *Streptococcus thermophilus* during storage at 20 °C.

3.2 Shelf Life of PL Powder

The moisture content increased from 1.98 per cent (db) to 58.8 per cent - 6.2 per cent when the powder lost its free flowing ability. Critical moisture content was determined based on free flowness and the corresponding water activity was obtained using constant GAB equation. M_O, C, K values were found to be 3.2 per cent (db), 4.201 and 0.977, respectively.

The area of packaging material exposed was 0.0288 m², in case of both materials (2 x 0.12 m x 0.12 m), as both the sides of the pouches are exposed to the storage environment. Weight of dry solids was 0.0192 kg in 0.02 kg *Lassi* powder. Integrating between the limits initial (0.031 kg/kg dry solid) and critical moisture (0.063 kg/kg dry solid) and applying the values for all often terms W dry solids (Ws), saturated vapour pressure (P^*), permeability (K) and area exposed (AP). Freeze dried pineapple *Lassi* powder was stable upto 44.3 and 52.9 days in two types of packaging, MP and ALP, under the storage conditions applied (33 per cent RH, 37 °C). The deterioration in terms of colour change was more in MP than in ALP. The barrier property of aluminium foil against light must have contributed towards the better colour stability of powder in ALP.

3.3 Changes in Colour During Storage

Colour values of pineapple *Lassi* powder changed during storage, (Fig. 4), a* values showed increase while no specific trend was observed for b* values (Table 2). The deterioration in terms of colour change was more in MP than in ALP. The barrier properties of Al. foil in ALP laminate against light must have contributed towards the better colour stability in ALP ANOVA revealed (Table 1) that the effect of both packaging material and storage time on colour change were significant. After storage of 40 days at 38 ± 1°C, 33 per cent RH there was
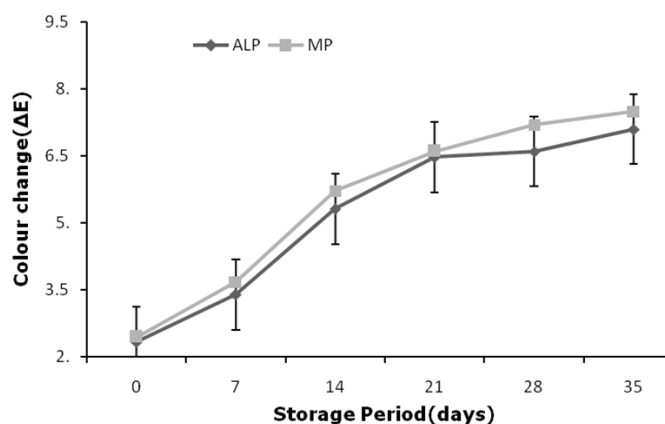

Figure 4. **Colour change in PL powder in MP and ALP pouches during storage at 38±1 °C and 33 ± 1% RH.**

Table 2. **Changes in Hunter lab colour values during storage of PL Powder in different packaging materials**

Storage period (days)	Packaging material					
	Metallised polyester (MP)			Aluminium laminated polyethylene (ALP)		
	L*	a*	b*	L*	a*	b*
0	75.78	0.32	34.5	75.78	0.32	34.5
7	76.81	0.33	33.9	776.1	0.35	33.25
14	76.85	0.42	35.2	77.25	0.81	34.2
21	77.26	0.52	32.4	78.99	0.85	30.98
28	78.05	0.82	31.1	78.77	0.81	29.38
35	78.22	0.92	29.7	79.00	1.58	29.90
42	79.1	0.93	29.5	79.45	1.93	30.21

greater colour change in PL powder packaged in MP than in ALP and this value correlated well with the Browning index values explained in previous section. The colour change of aloe vera powder during storage followed first order reaction kinetics with K value of 0.0444 per day for AF, 0.075 per day for BOPP and 0.0498 per day for PP[12]. Dehydrated paprika samples at degraded significantly rapidly at high temperature and RH, compared to samples room temperature storage.[13] Vacuum drying characteristics of coconut press cake have also been studied with reference to colour change[14].

3.4 Kinetics of Quality Changes in PL Powder during Storage

The functionality of the product depend on antioxidant activity, culture count and hence these parameters were modelled during storage of PL powder. Zero (Eqn. (5)) and first order (Eqn. (6)) degradation reaction kinetics were used for modelling the changes

$$C = C_0 + K_0 t \tag{5}$$

$$lnC = lnC_0 + K_1 t \tag{6}$$

where (+) and (-) indicate formation and degradation of quality parameters, respectively. The changes in vitamin C and antioxidant activity during storage are represented in Figs. 5 and 6. To determine the degradation kinetics of

Table 3. **Kinetics of Vitamin C degradation and culture count reduction during storage in different packaging materials**

Packaging material	Parameter	Zero order			First order		
		Ko	Co	R	K1	Co	R
MP	Vitamin C	-0.03	73.58	0.9511	-0.032 (s⁻¹)	4.2934	0.9978
	Culture count	-4.09	175.02	0.9947	-0.287 per day	20.67	0.9547
ALP	Vitamin C	-0.0190	73.58	0.9421	-0.017 (s⁻¹)	4.2834	0.9978
	Culture count	-2.9608	175.88	0.9938	-0.2117 per day	19.968	0.9611
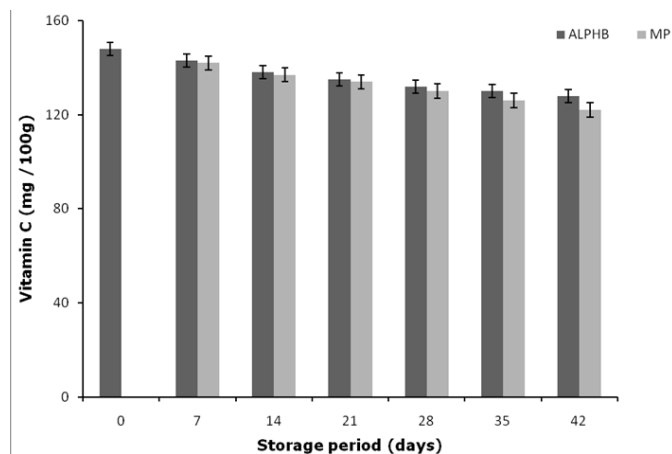
Figure 5. **Changes in vitamin C content of freeze dried pineapple *lassi* powder during accelerated storage in different packaging materials.**
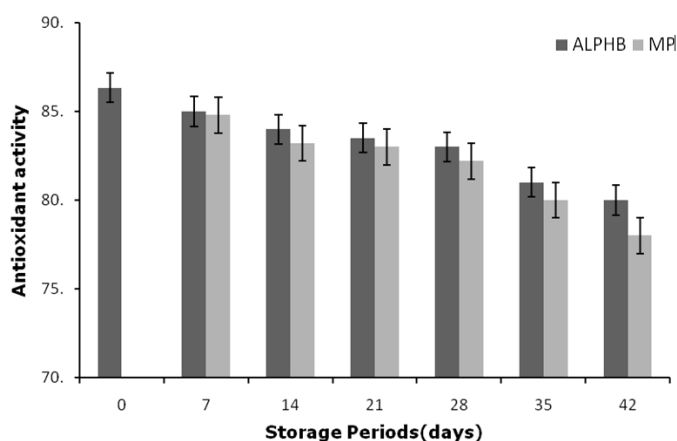
Figure 6. **Antioxidant activity of freeze dried pineapple *lassi* powder during accelerated storage in different packaging materials.**

vitamin C and culture count, zero and these parameters were investigated for first order reaction kinetics. Table 3 shows the kinetic parameters obtained. It can be observed on the basis of correlation coefficient (R) that culture count follow zero order reaction where as vitamin C degradation was better fitted by first order kinetics. Zero order kinetics was also reported by Kumar and Mishra[3] for *S. thermophilus* and *L. bulgaricus* count. The high values of rate constant for all parameters for MP pouches in comparison to these for ALP pouches means that more changes take place in powder when packaged in MP than in ALP (Table 3). Heat treatment, water activity and storage temperature exert effect on stability of food powders[15].

Aerobic conditions lead to significant increase in browning, moisture and water activities in iron-fortified formulas, while anaerobic packaging is more protective against such changes.

4.   CONCLUSIONS

The major antioxidant vitamin C in freeze dried pineapple *Lassi* powder and its moisture level are largely dependent on the characteristics of the packaging material used as well as the storage conditions. During accelerated storage, the fruit *Lassi* powder samples kept in Aluminium laminated pouches of high barrier preserved most of the antioxidant properties, suggesting that this type of film was better suited to maintain its stability and functionality. Our results indicate that moisture uptake is the limiting factor of the shelf-life of this dry powder containing fruit and dairy ingredients. The predicted shelf life of PL powder was 44.3 days and 61.9 days in MP and ALP pouches, respectively. Beyond this, the viability of *lactococci* and the free flowness of the powder were lost. Non enzymatic browning and lipid oxidation increased more in MP than in ALP and a corresponding change in Browning index and TBA were observed. The survival of starter culture was affected by moisture content as well as water activity of the PL powder. L* values increased during storage. Viability changes during storage of PL powder follow zero order kinetics and vitamin C degradation follows first order kinetics. Kinetic constants for quality changes were higher for PL powder packaged in MP than in ALP pouches. Hence, it can be concluded that ALP is more protective packaging material for Freeze dried Pineapple *Lassi* powder intended for defence food applications in extreme conditions.

REFERENCES

1. Khurana, H.K. & Kanawjia, S.K. Recent trends in development of fermented milks. *Curr. Nutrition Food Sci.*, 2007, **3** 91–108.
   doi: 10.2174/1573401310703010091
2. Van den, Berg & Bruin, S. Water activity and its estimation in food systems:theoretical aspects In. *Water activity: influences on food quality* ,1981, (1-61). Rockland L B Stewart G F (ed). Academic press New York
3. Kumar, P. & Mishra, H.N. Yoghurt powder: A review of process technology, storage and utilization. *Food Bioproducts Process.*, 2004, **82**(c2), 133–142.
   doi: 10.1205/0960308041614918
4. Jaya, S. & Das, H. Accelerated storage, shelf life and color of mango powder. *J. Food Process. Preservation*, 2005, **29** 45–62.
   doi: 10.1111/j.1745-4549.2005.00012.x
5. Shiby, V.K.; Radhakrishna, K. & Bawa, A. S. Development of whey fruit based energy drink mixes using Doptimal mixture design. *Int. J. Food Sci. Technol.,* 2013, **48** 742–748.
    doi: 10.1111/ijfs.12022
6. Varghese, S. K. & Pandey, M. C. Development of Freeze Dried Fruit *Lassi* (Indian Fruit Yoghurt) Powders Using

D-Optimal Mixture Design. *MOJ Food Processing Technology* ,2015, **1,** 00015-00021.
doi: 10.15406/mojfpt.2015.01.00015

7. King, R.L. Oxidation of milk fat globule membrane material. I.Thiobarbituric acid reaction as a measure of oxidized flavour in milk and model systems. *J. Dairy Sci.*, 1962, **45** 1165 – 1171.
doi: 10.3168/jds.S0022-0302(62)89590-3

8. Golubovic, Z.; Nenic, M. & Karabasevic, S. Ispitivanje mogucnosti iznalazenja optimalne koncentracije evaporiranog mleka radi proizvodnje mleka u prahu na spray postrojenju,s osvrtom na Maillard-ov tip potamnjivanja. *Mljekarstvo* ,1983, **33**(8), 231–236.

9. Li-Chan, E. Heat induced changes in the protein of whey protein concentrate. *J. Food Sci.,* 1983, **48 .**

10. Baldwin, A.J. & Ackland, J.D. Effect of preheat treatment and storage on the properties of whole milk powder. Changes in physical and chemical properties. *Netherland Milk Dairy J.*, 1991, **45** 169–181.

11. Nikolova,N.. The properties of lyophilized cultures of lactic acid bacteria in sour milk. *Dairy Sci. Abs.*, 1975, **37**, 414- 415.

12. Ramachandra, C. T. & Rao, P. S. Shelf-life and colour change kinetics of Aloe vera gel powder under accelerated storage in three different packaging materials. *J. Food Sci. Technol.*, 2013, **50** 747-754.
doi:10.1007/s13197-011-0398-9.

13. Addala, R.; Vasavada, M., Dong, J. & Subramanian, S. Effect of Storage Conditions on Rate of Color Degradation of Paprika based Products. *J. Food Process Technol.,* 2015, **6** , 423-426.
doi:10.4172/2157-7110.1000423

14. Jena, S. & Das, H. Modelling of vacuum drying characteristics of coconut presscake. *J. Food Eng.*, 2007, **79,** 92-99.
doi: 10.1016/j.jfoodeng.2006.01.032

15. Stapelfeldt, H.; Nielsen, B.R.; & Skibsted, L.H. Effect of heat treatment, water activity and storage temperature on the oxidative stability of whole milk powder. *Int. Dairy J.*, 1997, **7** 331–339.
doi: 10.1016/S0958-6946(97)00016-2

CONTRIBUTORS

Dr V.K. Shiby received her PhD in Agricultural and Food Engineering. Currently working as a Scientist 'D' at DFRL, has a research experience of 15 years in the area of food product development and process modelling. She has published about 35 papers in Journals. She is a recipient of *DRDO Laboratory Scientist Award-2013.*
She has initiated the product development and designed the experiments, she contributed towards the interpretation of results, drafting and revision of manuscript

Ms V. Jayashree is MSc project student, DFRL Mysuru.
Contributed significantly in carrying out the chemical analyses periodically and in tabulation of results.

Ms S. Sanchita is MSc project student, DFRL, Mysuru.
Contributed significantly in carrying out the microbiological analyses periodically and in tabulation of results.

Dr M.C. Pandey received his PhD in Agricultural Engineering. Currently working as a Scientist 'G' and Project coordinator at DFRL, Mysuru. He has a research experience of over 20 years in the area of agriculture and food engineering. He has published over 70 papers in Journals.
He provided help in statistical analysis of data and compilation of results, correction of manuscript at various stages with scientific inputs