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## Gamma Irradiation of Rice Grains

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Rice grains of the variety, 'Pusa-33', at 12.0% moisture, were irradiated with doses of 0-150 kGy. The crystallinity of starch, soluble amylose and yellowness of treated grains increased with increment in the dose of radiation but water absorption and volume expansion on cooking decreased. Irradiation at doses of 3-5 kGy increased imperceptibly the hardening of rice cooled after cooking, but had no effect on edibility. The off-aroma in irradiated grains was perceptible at doses higher than 5 kGy. The changes in colour and aroma persisted also on cooking. Upto a dose of 5 kGy, the sensory scores of rice, both cooked and uncooked, were at or above acceptable limit of score (5.5). The doses of 3 and 5 kGy were highly effective in reducing fungal population in irradiated grains, but in view of the changes in colour and cooking qualities, 3 kGy is the preferred dose-limit of irradiation.

A recent study<sup>1</sup> with an Australian variety of rice at 16.3 and 25.1 per cent moisture indicated that gamma irradiation at a dose above 1 kGy (100 krad) burns off rice, the product becoming unacceptable and unconsumable. This suggested that rice cannot be irradiated with doses above 1 kGy, which are required for reduction or elimination of fungi or other microbes. Consumer acceptability is the success of irradiation processing. However, Wang *et al.*<sup>2</sup> in Taiwan reported that irradiation at a dose below 3 kGy did not cause significant changes in cooking quality as judged by water uptake during cooking, volume expansion, pH of rice soup and sensory evaluation for colour and flavour of cooked rice. Japanese studies<sup>3</sup> also brought out the importance of moisture content in irradiation preservation of rice. Studies in Egypt<sup>4</sup> showed that 1.0 kGy is the acceptability limit for brown rice and 2.5 kGy for milled rice. These findings necessitated further research in this area. The objectives of the present work were to study the quality changes, determine acceptability limit of irradiation, reduce fungal load and add to the available information on these aspects of rice irradiation.

### Materials and Methods

The rice grains (dehusked paddy) of the variety, 'Pusa-33', used in the study, contained as determined with a Marconic moisture meter, 12.0 per cent moisture. The grains in 10 (for mould counts) 20 and 250 g lots, gamma (<sup>60</sup>Co) irradiated in sealed polythene bags, with doses of 1.0, 3.0, 5.0, 10.0, 50.0 and 150.0 kGy at a dose rate of 0.08 kGy/min, were stored at ambient temperature (30±8°C) during the study. The X-ray diffraction patterns of the powdered (powdered after irradiation) rice grains (250 μm) were recorded within a few days after irradiation by using Philips (Model PW 1729-PW1710) X-ray diffractometer operated at 35 KV and

30 mA to obtain nickel-filtered cuK radiation. The width of both the scatter and divergence slit was 3 mm and that of the receiving slit was 0.2 mm. The samples of unirradiated and treated rice powder were scanned between 5°-30° 2θ at a scanning speed of 2° 2θ /min. The relative crystallinity of single sample was calculated<sup>5</sup> by dividing the area of the peaks of irradiated samples with the corresponding peak area of the control sample. Whole grains were used to study colour changes in 5 replicates with a Hunterlab colorimeter by standardised method. Soluble amylose was determined in 4 replicates by a modified starch iodine blue method<sup>6</sup>. The water uptake during cooking was determined in duplicate by autoclaving for 25 min at 100°C, 20 g of rice taken in 100 ml water in 250 ml beakers. The rice so cooked was strained and quickly rolled over a filter paper to drain off water<sup>7</sup>. The water uptake was calculated by dividing the increased weight after cooking by the original weight of rice grains used. The volume expansion per unit volume of uncooked rice was determined in duplicate on the basis of difference in the volume of water displaced by uncooked and cooked grains<sup>8</sup>. For sensory ratings of both cooked and uncooked rice by 9 panelists, a 9-point Hedonic scale was used. The scores of 5.5 or more were considered acceptable. The texture of cooked rice and the after-cooking hardening was judged by pressing between the fingers and by crushing with a glass rod<sup>1</sup>. To study the effect of irradiation on hardness of uncooked rice, the averages of pressures in lb/square inch required just to crush 11 grains per treatment were determined with the aid of a fruit texturometer. Six evenly placed grains were dipped in 1.7 per cent KOH in small petri dishes in triplicate kept at 30 ± 1°C for 16 hr to study alkali digestibility using a 7-point scale<sup>9</sup>. One hundred grains from each treatment and the untreated control from

sealed 10 g lots were placed on malt salt agar containing 2.0 per cent malt extract, 7.5 per cent sodium chloride and 2.0 per cent agar agar, 6½ and 1 month after irradiation in separate experiments to examine the reduction as a result of irradiation<sup>10</sup> in fungal population arising from natural infection of grain.

### Results and Discussion

The X-ray diffractograms in Fig.1 showed A-type diffraction pattern characteristic of rice starch at all levels of irradiation and are in good agreement with other studies on rice<sup>1</sup>. The shape of the peak at 5.87 Å is apparently not affected by irradiation of the grains thereby showing no change in polymorphic form as a result of irradiation. In the region of 5.22-4.80 Å four peaks are discernible in the control and in the grains treated at 1 kGy; but on irradiation at 3 kGy and above only three distinct peaks are discernible in the region indicating a change in the atomic ordering<sup>11</sup>. The next major peak in the region of 3.87-3.76 Å is a broad one in the control. On irradiation at 1 kGy, it develops a shoulder at the higher side of 2θ angle and on irradiation at still higher doses of 3, 5 and 150 kGy it splits into two distinct peaks suggesting improvement in the ordered arrangement of atoms.

The relative crystallinity data in Table 1 show small increases with increments in the dose of irradiation when calculated on the basis of the area of the peak marked B in Fig.1. Whereas, in contrast, this increase is large when the area under peak A in Fig.1 is used. However, at the high dose of 150 kGy, these increases are not sustained probably because of charring effect of irradiation on the grains. Studies with irradiated rice and wheat<sup>12</sup> starches also reported such increases in relative crystallinity based only on peak area but did not report peak delineation or peak sharpening as found in the study. These latter characteristics further lend support to the fact of increased crystallinity.

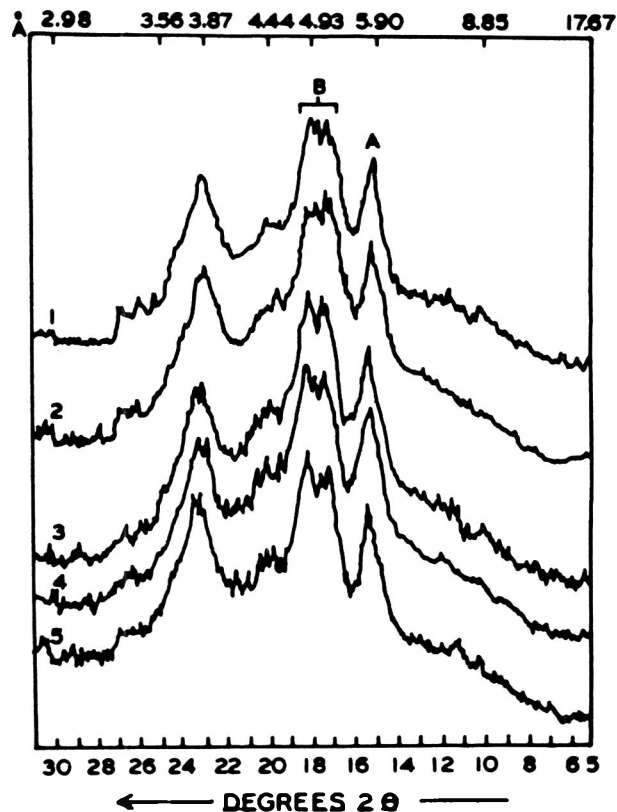


Fig.1. X-ray diffraction pattern of powdered rice (irradiated).

1 : Control; 2 : 1 kGy; 3 : 3 kGy; 4 : 5 kGy and 5 : 150 kGy

The Hunterlab L and b values in Table 1 show that as the irradiation dose increased the L-values, compared with the control, decreased significantly, but the b-values at all the doses increased significantly except at 1.0 kGy as also the yellowness of the treated grains. Such colour changes with parallel descending L and ascending b-values have also been reported in other studies<sup>1,4</sup>. Melanoidins formed due to oxidation of phenols or Maillard reactions between carbonyl compounds and free amino groups have been suggested as probable causes of yellowing in irradiated rice<sup>1</sup>. Water

TABLE 1. COLOUR, WATER UPTAKE DURING COOKING, SOLUBLE AMYLOSE AND RELATIVE CRYSTALLINITY OF IRRADIATED RICE

Irradiation dose (kGy)	Mean Hunterlab values		Av. water uptake*	Av. soluble amylose (%)	Av. relative crystallinity <sup>†</sup>	
	L	b			A	B
0	58.62	12.11	3.19	23.8	100	100
1	57.27	13.08	2.42	24.3	120	106
3	56.35	13.80	1.99	26.0	130	107
5	56.03	14.37	1.84	27.7	128	111
10	56.52	16.09	1.30	28.5	—	—
50	54.60	18.48	0.62	31.5	—	—
150	49.26	21.24	0.22	29.4	108	106
C.D. at 5%	0.40	1.21	0.13	0.33	—	—

(—) Not done

\*Wt after cooking/wt before cooking

<sup>†</sup>A and B as in Fig 1

TABLE 2. COLOUR, AROMA, TEXTURE AND CRUSHING PRESSURE OF IRRADIATED RICE

Irradiation dose (kGy)	Uncooked rice			Cooked rice			
	Colour	Aroma	Texture	Av. crushing pressure/grain (Psi)	Colour	Aroma	Texture
0	7.7	7.1	6.4	20.5	7.1	6.6	6.4
1	7.6	6.4	7.1	22.0	6.6	6.4	6.0
3	6.0	5.9	6.4	22.3	6.6	6.2	6.2
5	6.0	5.9	7.0	25.0	5.5	6.4	6.0
C.D. at 5%	1.33	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.

N.S. – Not significant

uptake (Table 1) decreased significantly with increments in dose of irradiation and is in good agreement with other results<sup>1</sup>. Soluble amylose (Table 1) considered as the most important determinant of cooking<sup>13</sup> and eating<sup>14</sup> quality of rice increased significantly with irradiation probably due to breakdown of amylopectin<sup>1</sup> and solubilisation of starch caused by changes in its structure. The direct relationship that increase in soluble amylose increases the water uptake, holds good as is generally known, in a different situation i.e. in un-irradiated rice. Besides, negative relationship between these two factors of cooking quality has also been reported<sup>9</sup>.

The sensory evaluation scores for colour and aroma of cooked rice (Table 2) decreased with irradiation indicating significant change in colour above 1 kGy and loss of aroma (significant). The scores for texture suggested slight increase in hardness but was not statistically significant. The increases in pressure required to crush irradiated grains were also not significantly different. Nevertheless, upto the dose of 5 kGy the sensory characteristics scores of both cooked and uncooked rice were at or above the acceptability limit of 5.5. The acceptable dose is, therefore, much above 1 kGy as suggested in the Australian studies which were carried out with rice that was cold-stored (5°C) after treatment and had high moisture content.

The data in Table 3 show that irradiation at the doses of 3 to 5 kGy reduced the fungal population by 83 to 94 per cent in grain after 1 month of storage. But this dose could reduce the fungal counts after 6½ months of storage by about 78.0 per cent (Table 4). This dose range of 3-5 kGy could not eliminate moulds completely. A few propagules survive massive radiation assault perhaps because of reversibility of

TABLE 3. FUNGAL COUNTS (NUMBER OF COLONIES/100 GRAINS OF IRRADIATED RICE) AFTER ONE MONTH OF STORAGE

Radiation dose (kGy)	Fungal counts	% reduction in fungal counts (over control)
0	37	—
3	6	83
5	2	94

growth inhibition<sup>15,16</sup> by irradiation with passage of time, or due to mechanisms of damage repair operative in fungi<sup>17</sup>. The reversible reactions and mechanisms of damage repair, however, require further research for complete explanation. Wang *et al.*<sup>2</sup> reported that the dose of 3 kGy could reduce microbial population by about 91 to 95 per cent while 10 kGy could bring down the population by about 99 per cent during 12 months of storage period. Japanese workers<sup>3</sup> showed that an increase in moisture content of rice by about 2-3 per cent (from 14-17 per cent) doubled the irradiation dose from 2 kGy to 4 kGy for reducing microbial load and extending storage life of rice. The efficacy of irradiation in reducing fungal load as obtained in this study is in complete agreement with that of other studies<sup>2,3</sup>.

TABLE 4. FUNGAL COUNTS (NUMBER OF COLONIES/100 GRAINS OF IRRADIATED RICE) AFTER 6½ MONTHS OF STORAGE

Fungi	0 kGy	3 kGy	5 kGy
<i>Aspergillus flavus</i>	8	2	1
<i>Aspergillus niger</i>	3	—	1
<i>Aspergillus amstelodami</i>	—	—	1
<i>Aspergillus sp.</i>	7	1	1
<i>Alternaria alternata</i>	—	1	—
Total	18	4	4
% reduction over 0 kGy		78	78

TABLE 5. ALKALI – DIGESTIBILITY, VOLUME EXPANSION AND AFTER COOKING HARDENING OF IRRADIATED RICE

Radiation dose (kGy)	Av. alkali digestion scores*	Av. vol. expansion/unit vol. of rice	Av. score on hardening of cooked rice on cooling
0	5.7	5.0	8.0
1	5.2	4.7	7.9
3	4.0	4.2	7.8
5	4.7	3.5	7.8
Significance		**	N.S.
C.D. at 5%		0.36	

\*\*Significant at 5% level.

N.S. – not significant

Irradiation increased (Table 5) imperceptibly the hardening of rice cooled after cooking at ambient temperature. The alkali-digestibility scores showed only small and insignificant effect of irradiation on grains. The volume expansion decreased significantly on irradiation at doses above 1 kGy. The effects, however, did not disqualify the grains for acceptability or edibility. A dose of  $3.5$  kGy is necessary for effective reduction in microbial population but a dose much lower than 1 kGy is considered good enough when the problem is to kill insects in storage<sup>13</sup>.

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# Kinetics of Liquid Water Absorption by Evacuated Paddy Grains

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An empirical equation correlating moisture content to duration of soaking in water at 30–80°C was developed for evacuated paddy. The equation was based on generalised mathematical equation developed for direct soaking of paddy. The predicted and experimental values of moisture content were non-significant for entire range of this study at 0.5 per cent level of significance. The developed equation therefore can be directly used for prediction of moisture content of paddy soaked in water, after evacuation, in the temperature range of 30–70°C for soaking duration upto 6 hr and 2 hr for 80°C.

Soaking is an important step in paddy processing. The soaking time depends upon soaking temperature, initial moisture content of grain and level of moisture content desired. It is, therefore, desirable to study the kinetics of moisture absorption for prediction of moisture content for optimising the process variables. Some studies<sup>1–3</sup> are reported but the information on complicated variables such as grain surface area, diffusion co-efficient and grain volume is required for solving those equations. It, thus, restricts the use of equations with information on soaking conditions alone. A mathematical expression was therefore, developed<sup>4,5</sup> assuming that soaking is analogous to drying<sup>6</sup> and correlating moisture content and duration for direct soaking of paddy. Since the evacuated paddy soaking speeds up the moisture absorption<sup>7</sup> and gives the rice with improved product quality<sup>8</sup>, an empirical equation for evacuated paddy soaking was developed.

**Review of equation:** The equation was developed with the following assumptions; Paddy is an isotropic and hydrophilic material. The process of soaking is limited upto the gelatinization of paddy starch and does not include cooking. The higher values of vapour pressure of water in liquid or vapour form in the surrounding medium increase the rate of absorption of water vapour into the grain, and the pressure of water vapour is directly proportional to the absolute temperature of soak water. At soaking temperatures less than or equal to the gelatinization temperature, paddy attains a state of equilibrium, when it is soaked for relatively longer duration. Moisture content at equilibrium is designated as saturation moisture content (Ms).

From the assumptions it is obvious that, rate of water absorption is proportional to difference in vapour pressure of surrounding medium and vapour pressure in the grain. The rate of water uptake ( $\frac{dM}{d\theta}$ ) by the paddy is directly

proportional to the difference of saturation moisture content (Ms) which remains same for any duration and temperature of soaking<sup>9</sup>, and moisture content of grain (M) at any given soaking duration ( $\theta$ ).

$$\text{i.e. } \frac{dM}{d\theta} \propto (M_s - M) \text{ or } \frac{dM}{d\theta} = \beta (M_s - M) \dots\dots(1)$$

Where,

$\theta$  = soaking time, min

$\beta$  = soaking constant,  $\text{min}^{-1}$

$\frac{dM}{d\theta}$  = rate of moisture absorption, per cent d.b./min.

$M_s$  = saturation moisture content of grain, per cent db

$M$  = moisture content of grain at any duration,  $\theta$ , per cent d.b.

The soaking constant ( $\beta$ ) is a constant of proportionality which has the dimension ( $\text{min}^{-1}$ ) and its value should remain same for any given soaking temperature.

Integration of equation 1 for  $\theta = 0$  to  $\infty$  and  $M = M_0$  to  $M_s$  as boundary conditions yield

$$\bar{M} = \frac{M_s - M}{M_s - M_0} = e^{-\beta\theta} \text{ or } \bar{M} = e^{-\beta\theta} \dots\dots(2) \text{ at } \alpha = 1$$

Where,

$\bar{M}$  = moisture ratio

$M_0$  = initial moisture content of grain, per cent d.b.

$\alpha$  = dimensionless constant

The constant ( $\alpha$ ) represents the moisture ratio ( $\bar{M}$ ) in equation 2 at  $\theta = 0$ . Theoretically, its value is unity as  $M = M_0$  at  $\theta = 0$  in equation 2.

## Materials and Methods

The raw paddy 'Jaya', at 13.2 per cent d.b. samples (150 g) were subjected to vacuum ( $-76$  cm of Hg as indicated by compound gauge) for 5 min before soaking. Hot water (225 ml) was then released into the flask containing sample.

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Vacuum was released after thorough mixing of water and paddy, and sample was then soaked. Samples were soaked for 9 durations viz 0.25, 0.50, 0.75, 1, 2, 3, 4, 5 and 6 hr and at 6 soak water temperatures viz 30, 40, 50, 60, 70 and 80°C in a preset hot water bath ( $\pm 0.5^\circ\text{C}$ ). The moisture content was calculated after weighing<sup>4</sup>. The data were analysed for development of empirical equation for prediction of moisture content within the range of the study.

**Results and Discussion**

**Moisture absorption:** Fig 1 indicates sudden rise in the moisture content upto 35 per cent d.b. Initial moisture gain was reported to be more in evacuated paddy soaking than direct soaking and thus reduced soaking time for achieving particular level of moisture content. The significant rise in moisture uptake is seen after soaking at 80°C for 1.5 to 2 hr (Fig 1). It is due to the bursting and subsequent cooking of grains during soaking. Therefore, the empirical equation has been developed for 2 hr soaking duration upto which the soaking continued, instead of 6 hr soaking duration at 80°C (Fig 2).

**Empirical equation:** The soaking test data upto certain moisture gain at which the husk splitting took place (45-50 per cent d.b.) were analysed (Table 1) using equation 2 and the average Ms value for all soaking duration. However, the

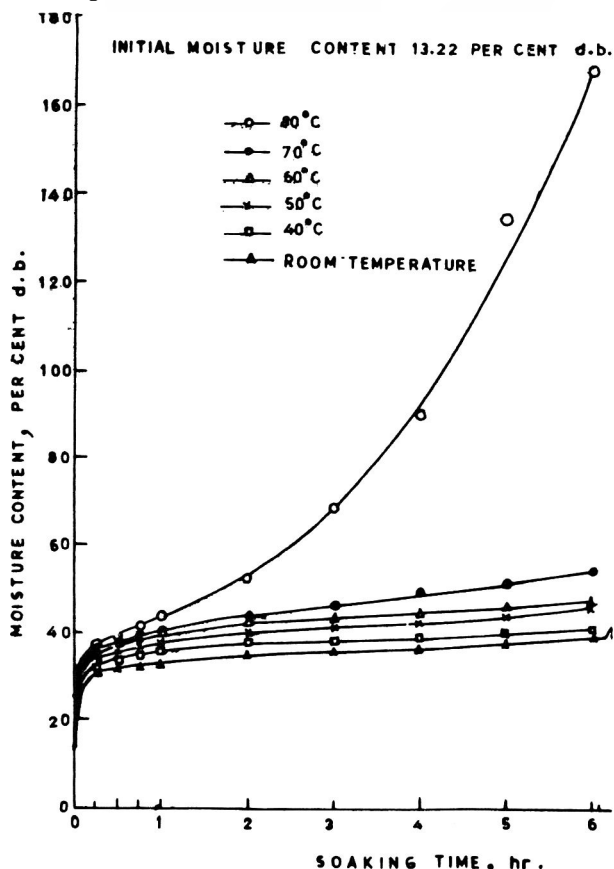


Fig. 1. Variation in moisture content of paddy soaked after evacuation with soaking time.

Ms value ranged<sup>9</sup> from 69-73 per cent d.b for different soaking temperatures. Value of  $\bar{M}$  calculated from data for any given  $\theta$  indicated the available potential for moisture absorption. When  $\bar{M}$  was plotted against  $\theta$  for different soaking temperature gave straight line relationship (Fig 2).

**Variation of  $\alpha$  with soaking temperature:** Theoretically (equation 2) the value of  $\alpha$  should be unity. But since the developed equation does not account for initial uptake of moisture in the constant rate period,  $\alpha$  can not obviously be unity if the equation is to be used for entire period of soaking. Value of  $\alpha$  was always less than unity (Table 1) and did not show any definite relationship with soaking temperature. Therefore, an average value ( $\alpha = 0.6394$ ) was assumed to be constant and used in generalised equation.

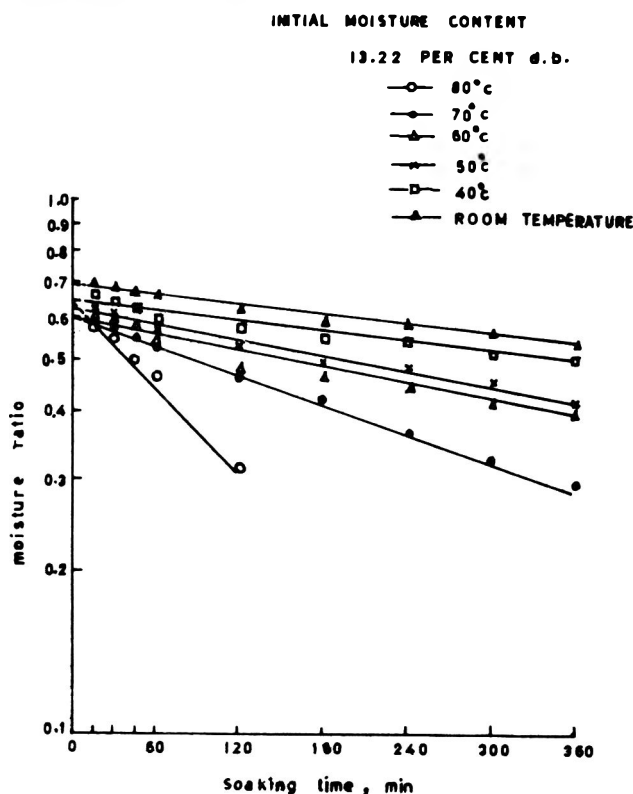


Fig. 2. Variation of moisture ratio ( $\bar{M}$ ) with soaking time.

TABLE 1. VALUES OF CONSTANTS  $\alpha$ ,  $\beta$ , AND CORRELATION CO-EFFICIENT 'r' FOR REGRESSION EQUATION  $\bar{M} = \alpha e^{-\beta}$  FOR EVACUATED PADDY SOAKING AT DIFFERENT TEMPERATURES.

Soaking temp (°K)	$\alpha$	B	r
303	0.6972	(-) 0.0007	(-) 0.9848
313	0.6484	(-) 0.0007	(-) 0.9701
323	0.6209	(-) 0.0011	(-) 0.9874
333	0.5997	(-) 0.0012	(-) 0.9722
343	0.6136	(-) 0.0022	(-) 0.9989
353	0.6535	(-) 0.0066	(-) 0.9989

**Variation of  $\beta$  with soaking temperature:** It is clear (Table 1) that  $\beta$  increased with soaking temperature. The analysis of variation of  $\beta$  with absolute temperature (T,°K) of soaking was found to yield best correlation for the relationship of the form

$$Y = a \times b^x, \text{ or } \beta = a \left(\frac{1}{T}\right)^b \text{ i.e.}$$

$$\beta = 4.157 \times 10^{-36} \left(\frac{1}{T}\right)^{-12.9326} \quad (r = -0.9313) \dots\dots(3)$$

and gave a straight line plotted on log-log scale (Fig. 3).

**Generalized soaking equation:** An empirical equation of the form (eqn. 4) relating  $\bar{M}$  with  $\theta$  and T was developed using average value of  $\alpha$ , equations 2 and 3 for the temperature range of 30-70°C and soaking duration upto 6 hours and up to 2 hours for 80°C

$$\bar{M} = 0.6394 e^{-[4.157 \times 10^{-36} \left(\frac{1}{T}\right)^{-12.9326}] \theta} \dots\dots(4)$$

From this study, it was established that the absorption and desorption are similar phenomena and confirm the generalised drying equation<sup>6</sup> of the form  $\bar{M} = e^{-k\theta}$ . Equation 4 was predicting the moisture content of paddy soaked after evacuation and used for, within the range of this study. The

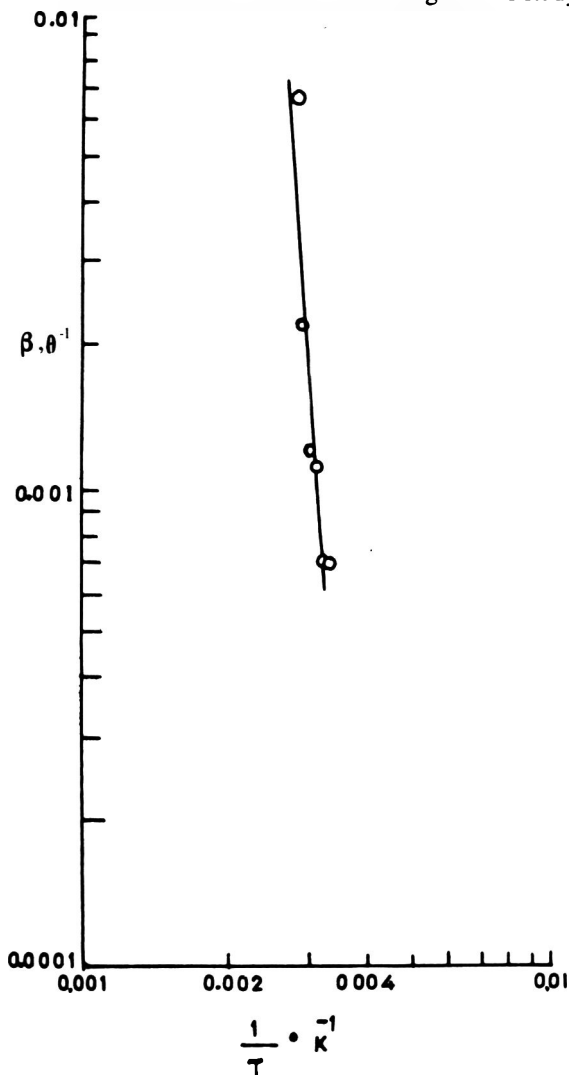


Fig.3. Relationship between soaking constant with soaking temperature.

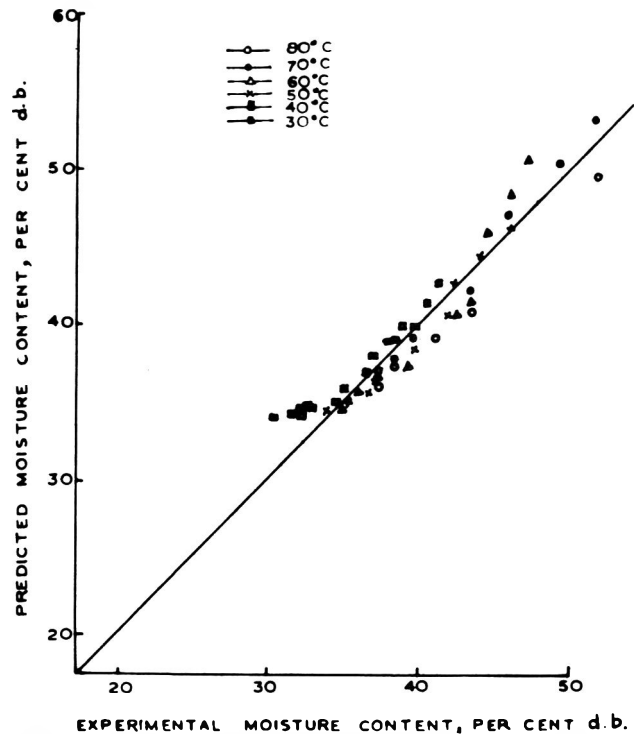


Fig.4. Variation in predicted and experimental values of moisture content of paddy soaked after evacuation.

differences between the observed and predicted values were found to be non-significant (Fig 4) at 0.5 per cent level of significance by the chi square ( $\chi^2$ ) test. Fig 5 indicates the trend of experimental and predicted moisture content values at 40,50 and 70°C. The data were further analysed by the chi square test for soaking duration (upto 1 hr), and all the soaking temperatures. The difference between the observed and predicted values was found to be non-significant (Table

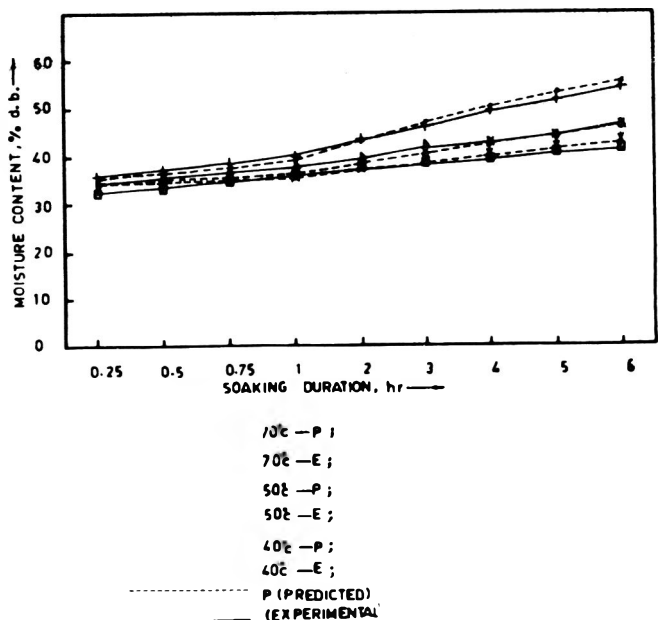


Fig.5. Variation of predicted (P) and experimental (E) moisture content with soaking duration.

TABLE 2. DETAILS OF CHI SQUARE TEST ANALYSIS FOR DIFFERENT SOAKING DURATIONS AND TEMPERATURES

Soaking duration upto (hr)	D.F. (n-1)	$\chi^2$ (0.99)	
		Calculated	Table value
0.25	5	0.507	0.55
0.50	11	0.800	3.10
0.75	17	1.140	6.40
1	23	1.670	10.20
6	49	3.120	22.60

\*Soaking temperatures 30-80°C with 10°C interval and soaking duration 15 min to 6 hr for all temperatures except 80°C (up to 2 hr only).

2) at 1 per cent level of significance in all the cases, confirming the validity of the developed equation for initial period of soaking for all the temperatures and also for entire range of study i.e. 15 min to 6 hr soaking duration and 30 to 70°C soaking temperature and 2 hr for 80°C. However, this equation is not expected to be valid for a constant rate period which might fall anywhere up to 15 min soaking duration which is otherwise outside the range of this study.

This study clearly indicates the validity of developed empirical equation for prediction of moisture content of evacuated Jaya paddy soaked at any given soaking duration

upto 6 hr and temperature range of 30-70°C and up to 2 hr for soaking at 80°C. The wider application of this equation is possible as the only information required for prediction of moisture content after evacuation of paddy is the duration and temperature of soaking.

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## Effect of Varieties and Processing Methods on Phytic Acid and Protein Digestibility of Groundnut (*Arachis hypogaea* L.)

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Processed groundnuts (cultivar 'Florunner', 'US No. 1') by boiling, water- and steam- blanching and roasting, and other varieties (viz. 'TP 171-2', 'TP 172-2', 'TP 175-3', 'TP 175-6', 'TP 178-1', 'TP 18-3', 'TX AG-3', and 'RMP-12') were analyzed for protein, phytic acid, total phosphorous, nitrogen solubility and *in vitro* protein digestibility (IVPD). Phytic acid of nine varieties varied from 2.89 to 3.96 mg/g indicating significant differences among varieties. The IVPD values of these varieties ranged from 66.8 to 77.5% with mean being 70.9%. There was a significant negative correlation between phytic acid and IVPD of these varieties. Removal of seed coat in the present study did not reveal noticeable differences in phytic acid, whereas it considerably influenced nitrogen solubility and IVPD values. Processing methods reduced the phytic acid of groundnut and effect was more pronounced in boiling process (wet-heating) followed by blanching processes. Dry-heating (roasting) considerably reduced IVPD of groundnut, whereas it did not show any noticeable effect on phytic acid.

Over the past years, major attention has been directed to the uses of oilseeds as cheaper yet adequate protein foods. In addition to being a good source of oil, peanut also called groundnut (*Arachis hypogaea* L.) is used as human food in various forms. Boiling, blanching and roasting processes are commonly employed for converting raw groundnut into consumable form. Nutritional quality of groundnut proteins has been the subject of several studies in the past and this subject has been periodically reviewed<sup>1</sup>. Raw and heat processed groundnut flours were found to have higher trypsin inhibitors and lectins than similarly processed soy flour<sup>4</sup>.

When consumed in excess, phytic acid can function as an anti-nutrient. Major concern is over the bioavailability of minerals such as zinc, calcium, and iron which are not readily absorbed when insolubilised as calcium phytate<sup>5</sup>. Phytic acid has also been linked to the inhibition of digestive enzymes such as protease<sup>6</sup>, lipase<sup>7</sup>, and alpha-amylases<sup>8</sup>. Complexing between phytate and proteins has been reported for several proteins of cereals and legumes including groundnut and this might affect the protein digestibility and bioavailability<sup>9,11</sup>. Information on the phytic acid content of groundnut is scanty. Therefore, the objectives of this study were: 1) to examine the variability in phytic acid content of groundnut varieties, 2) to study the effect of processing methods on removal of phytic acid, and 3) to study the relationship between phytic acid, nitrogen solubility and *in vitro* protein digestibility (IVPD) of groundnut.

### Materials and Methods

Seed samples of nine varieties ('TP 171-2', 'TP 172-2', 'TP 175-3', 'TP 175-6', 'TP 178-1', 'TP 178-3', 'TXAG-3', 'RMP-12' and 'Florunner', 'US No 1') were obtained from Yoakum Experimental Research Station, Texas, USA. These varieties were grown in 1988 and the 'Florunner' ('US No. 1') was used as a control. Seed lots were cleaned and stored in a cold room at 4°C until used. To study the effect of processing methods, the variety 'Florunner' was used. For boiling process, about 50 g seed material was boiled in 200 ml distilled water for 30 min. Water was brought to a boiling point and then seed material was transferred to the boiling water and boiling continued for 30 min. After boiling, excess water was discarded and seeds were dried in the oven at 50°C overnight. For water-blanching, seeds were dipped in hot distilled water at 90°C for 2 min according to Ukuku *et al.*<sup>12</sup>. Excess water was discarded and material was dried as above. The steam-blanching was carried out at 100°C for 2 min<sup>12</sup>. The water- and steam-blanched samples were dried at 50°C overnight. The roasting was carried out at 165°C for 8 min in a cabinet drier (Proctor and Schwartz Inc. Horsham, PA). All processed samples were decorticated by removing the seed coat manually. Raw, processed and decorticated samples were ground using a Wiley Mill (Arthur H. Thomas Company, Philadelphia).

Moisture, protein, fat, and ash contents in the ground samples were determined using standard AOAC methods<sup>13</sup>.

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Protein content was calculated by using the nitrogen to protein conversion factor of 5.46. Phytic acid was determined according to the method described by Wheeler and Ferrel<sup>14</sup>. Phytate content was calculated from the iron concentration by assuming a constant Fe: P molecular ratio of 4:6 in the precipitate. A colorimetric method using ammonium molybdate and amino naphthol-sulphuric acid reagent was used for the determination of phosphorus<sup>15</sup>.

The nitrogen solubility was determined by employing AACC method<sup>16</sup> with minor modifications.

For determination of *in vitro* protein digestibility (IVPD)<sup>17</sup>, an amount of defatted sample containing  $5.0 \pm 0.1$  mg N was used.

### Results and Discussion

Protein content of these varieties ranged from 23.2 to 29.5 per cent showing a large variation (Table 1). There was also a large variation in 100-seed weight which ranged from 50.4 to 65.0 g. Fat content did not show much variation, as it ranged from 42.4 and 48.9 per cent with a mean of 46.1 per cent.

Phytic acid content ranged from 2.89 ('T P 178-3') to 3.96 mg/g ('T P 172-2'). Phytic acid constituted from 61.2 to 76.0 per cent of the total phosphorus (Table 1). Nitrogen solubility of these varieties ranged between 49.7 and 60.5 per cent and *in vitro* protein digestibility (IVPD) between 66.8 and 77.5 per cent, with mean being 70.9 per cent. The IVPD values are considerably lower than those reported by Anurag and Geervani<sup>18</sup>. The highest protein digestibility was observed in 'T P 178-3' which contained the lowest amount of phytic acid. There was a negative and significant correlation ( $r = -0.353^{**}$ ), although the magnitude of correlation was low, between phytic acid and IVPD values (Table 2). Phytic acid is reported to form a complex with proteins rendering them less soluble<sup>10</sup>. The results of the present study do not appear to lend support to this observation. The ability of phytic acid to complex with

TABLE 2. CORRELATION COEFFICIENT BETWEEN PHYTIC ACID, NITROGEN SOLUBILITY AND *IN VITRO* PROTEIN DIGESTIBILITY (IVPD).

	Phytic acid	PA/P	N solubility	IVPD
Phosphorus (P)	0.116	-0.027	-0.192	-0.175
Phytic acid (PA)	—	0.251*	-0.071	-0.353**
PA/P	—	—	0.041	-0.235*
N Solubility	—	—	—	0.589**

\*Significant at 5% level.

\*\*Significant at 1% level.

proteins and inhibit enzyme activity has been reported by earlier workers<sup>6</sup>. It appeared that phytic acid reduced the protein digestibility by interfering with protease enzymes. The formation of a complex with protein did not appear to be a strong factor in the present study as there was no noticeable negative correlation between nitrogen solubility (as an index of protein solubility) and phytic acid (Table 2). However, the present results suggest that phytic acid possibly inhibits the enzyme activity.

The processing methods studied reduced the protein content to variable extents, maximum reduction being noticed in case of boiling process. This might have been due to the solubility of proteins in boiling water (Table 3). Removal of seed coat in the present study did not reveal noticeable differences in phytic acid, whereas it considerably influenced nitrogen solubility and IVPD values (Table 3). This contradicts the previously reported results on phytic acid which was significantly increased due to dehulling in dry beans<sup>19</sup>. Boiling resulted in a considerable (15 per cent) reduction in phytic acid. Phytic acid reduction due to roasting was less (1.2 per cent). It has been reported that 30-min autoclaving reduced the phytate content of cereals by less than 10 per cent<sup>20</sup>. The boiling of groundnut did not change the nitrogen

TABLE 1. MOISTURE, 100-SEED WEIGHT, FAT, PROTEIN CONTENTS, PHOSPHORUS, PHYTIC ACID, NITROGEN SOLUBILITY, *IN VITRO* PROTEIN DIGESTIBILITY (IVPD) OF GROUNDNUT

Cultivar	Moisture (%)	100-seed wt (g)	Fat (%)	Protein (%)	Phosphorus (mg/g)	Phytic acid (mg/g)	Phytic acid as % of P	N solubility (%)	IVPD (%)
TP 171-2	4.9	60.5	44.6	24.9	4.8	3.3	68.1	60.5	73.0
TP 172-2	4.8	62.3	46.3	23.2	5.4	4.0	73.9	50.0	68.5
TP 175-3	5.2	59.9	48.9	24.1	4.8	3.1	65.0	58.3	72.5
TP 175-6	5.1	59.5	46.3	24.2	4.5	3.2	71.1	52.6	72.7
TP 178-1	5.3	53.0	46.2	25.0	5.0	3.4	67.9	55.4	70.4
TP 178-3	4.8	50.4	45.3	24.7	4.6	2.9	63.4	51.5	77.5
TX AG-3	4.9	65.0	47.7	28.7	4.9	3.5	72.0	49.7	69.6
RMP-12	4.9	56.0	47.6	29.3	4.9	3.3	66.7	51.4	67.0
Florunner	4.9	50.9	42.4	29.5	5.1	3.9	76.0	50.6	66.8
Mean	5.0	57.5	46.1	26.0	4.9	3.4	69.3	53.3	70.9
SD $\pm$	0.38	1.32	0.98	0.81	0.32	0.27	1.52	1.26	2.04

1. Means of two independent determinations.

TABLE 3. EFFECT OF PROCESSING METHODS ON PROTEIN, PHYTIC ACID, NITROGEN SOLUBILITY, AND *IN VITRO* PROTEIN DIGESTIBILITY (IVPD) OF GROUNDNUT<sup>1</sup>

Processing method	Protein (%)		Phytic acid (mg/g)		Nitrogen solubility (%)		IVPD (%)	
	a	b	a	b	a	b	a	b
	Raw	29.5	30.6	3.4	3.5	50.6	53.7	66.8
Boiling	25.4	26.0	2.9	2.9	50.8	58.5	70.0	74.7
Water-blanching	27.9	28.5	3.1	3.1	50.4	52.3	69.4	77.5
Steam-blanching	29.0	29.5	3.2	3.2	49.6	50.5	68.4	72.0
Roasting	28.5	29.3	3.4	3.4	45.5	48.0	60.7	65.8
Mean	28.1	28.8	3.2	3.2	49.4	52.6	67.1	72.1
SD ±	0.35	0.32	0.18	0.19	1.34	1.08	1.32	1.45

1. Means of two independent determinations.

a. With testa (seed coat)

b. Without testa

solubility, whereas it improved *in vitro* protein digestibility. McWatters and Cherry<sup>3</sup> reported that heat processing of groundnut flour reduced protein solubility. But in the present study, roasting decreased both nitrogen solubility and protein digestibility whereas boiling increased the protein digestibility. The observation partly disagrees with those of Anurag and Geervani<sup>18</sup> who reported that roasting, boiling, and frying improved the *in vitro* protein digestibility of groundnut.

To conclude, it may be mentioned that large variability existed in IVPD and phytic acid of groundnut varieties. Nitrogen solubility also showed noticeable variation among varieties. There was a significant and negative correlation between phytic acid and IVPD of groundnut implying that phytic acid would adversely influence the protein quality of groundnut. The boiling process considerably decreased the phytic acid and this might have improved the *in vitro* digestibility of groundnut.

### Acknowledgements

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## Dehydration Characteristics of Ten Onion Cultivars

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Onion varieties were dehydrated as 5 mm thick rings in an air cabinet drier ( $65 \pm 2^\circ\text{C}$ , 7 hr). Dehydration reduced pungency (as pyruvic acid) and ascorbic acid levels, and induced browning; the extent of these changes depended upon variety. Colour of dehydrated product showed significant positive correlation with flesh colour but not the peel colour. Varieties were ranked on the basis of quality of their dehydrated rings. 'VL-1' was found to be excellent, 'Pusa Red' was very good, and 'Punjab Red' and 'Sel-102-1' were good for dehydration.

Demands for fresh and dehydrated onions have considerably increased over the last two decades<sup>1</sup>. To meet this challenge many new varieties possessing superior field characteristics have been developed. But their dehydration characteristics differ markedly and the Indian varieties generally have low solids, pungency/shelf-life and are pigmented, which limit their suitability for dehydration<sup>1-5</sup>. The objective of this study was to evaluate some of the onion varieties for their dehydration characteristics.

### Materials and Methods

Mature and cured bulbs of ten varieties ('Agrifound Light Red', 'Arka Niketan', 'Pusa White Round', 'Pusa White Flat', 'Pusa Red', 'Punjab Red', 'Punjab Red Round', 'Sel-102-1', 'VL-1' and 'VL-3') under field trials at Horticultural Research Centre of this University were stored under ambient conditions (April-October,  $30 \pm 3^\circ\text{C}$ , RH 40-80 per cent) in a well ventilated room on open shelves. Among these varieties, 'Pusa Red' is under commercial cultivation while others are new for the Tarai region.

**Physical analysis:** Average weight, diameter, neck diameter, and height of 10 randomly selected whole bulbs of each variety were determined separately. Their visual surface colour was also noted.

**Chemical analysis:** Peels, roots and tops were removed, and 3-4 bulbs of each variety were ground to paste in a blender. The total soluble solids content (TSS) was determined with a pocket refractometer. Vacuum oven method<sup>6</sup> for moisture, AOAC method<sup>7</sup> for alcohol insoluble solids (AIS), Lane and Eynon method<sup>8</sup> for total and reducing sugars, dye-titration method<sup>8</sup> for ascorbic acid (AA), and method of Schwimmer and Guadagni<sup>9</sup> for pyruvic acid (PA)

were followed. Peel or flesh colour was estimated by the method of Moore *et al.*<sup>10</sup> using acidified methanol as solvent. OD of extract was measured at 540 nm on a Spectronic-20 D colorimeter and from these values, OD per g sample/OD per g solids were calculated. OD in these units were used to express and compare the colour of fresh and dehydrated samples.

**Drying characteristics:** Bulbs were peeled, trimmed and sliced manually into 5 mm thick rings. 'Pusa Red' variety was given different treatments but in the second set of studies the varieties were dried without any pre-treatment. Rings were spread thinly on trays (tray loading  $4.2 \text{ kg/m}^2$ ) and dried in an air cabinet drier ( $65 \pm 2^\circ\text{C}$ , about 7 hr) to 5-7 per cent moisture. Dehydrated rings were packed quickly in polyethylene bags and stored in desiccators for further analysis.

Rings of 'Pusa Red' variety were dipped in 5 per cent NaCl solution for 30 min or warm water ( $70^\circ\text{C}$ ) for 1 min<sup>11</sup>, 0.5 per cent L-cysteine for 10 min<sup>12</sup> or 5 per cent starch solution for 30 min<sup>13</sup>. Dipped samples were drained for 5 min and dried.

**Evaluation of quality of dehydrated rings:** Shrinkage ratio (SR), dehydration ratio (DR), co-efficient of rehydration (CR) and extent of non-enzymatic browning (NEB) were estimated by the methods suggested by Ranganna<sup>8</sup>. Methods for estimating moisture, pigments, total and reducing sugars, and AA in dehydrated rings were similar to those for fresh onion. For PA estimation, dried samples were prepared by the method of Peleg *et al.*<sup>14</sup> and the analytical method was that used for fresh onion<sup>9</sup>.

Suitability of varieties for dehydration was assessed on the basis of the quality of their dehydrated products. Varieties

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TABLE 1. PARAMETERS WITH THEIR RANGE AND WEIGHTAGE USED FOR RANKING DEHYDRATED RINGS OF DIFFERENT VARIETIES

Quality parameter	Rating range with score			Weightage <sup>2</sup>
	+	++	+++	
	(4)	(6)	(10)	
Shrinkage ratio	> 9	8-9	< 8	2
Co-efficient of rehydration	< 0.6	0.6-0.7	> 0.7	2
Non-enzymatic browning (OD) <sup>1</sup>	> 0.22	0.13-0.22	> 0.13	1
Pyruvic acid retention (%) <sup>1</sup>	< 10	10-20	> 20	3
Ascorbic acid retention (%) <sup>1</sup>	< 20	20-30	> 30	1
Increase in flesh colour (%) <sup>1</sup>	> 30	10-30	< 10	1

1 On dry weight basis; 2 Weightage in a maximum score of 10

were categorised into appropriate range of 6 quality parameters (Table 1) and evaluated based on scores.

Samples were drawn by RBD technique and were analysed in triplicate. Significance of difference between values was estimated at  $p < 0.05$  by applying analysis of variance<sup>15</sup>. Inter-relationship between different parameters was evaluated by calculating correlation co-efficient ( $r$ )<sup>15</sup>.

## Results and Discussion

**Physico-chemical characteristics:** The varieties can be grouped into three categories on the basis of their surface colour-white ('Pusa White Round' and 'Pusa White Flat'),

light red ('Agrifound Light Red', 'Arka Niketan', 'Punjab Red', 'Sel-102-1' and 'VL-1'), and red ('Pusa Red', 'Punjab Red Round' and 'VL-3'). Their sizes (diameter x height) and weights differed significantly (Table 2) and were larger than those reported by Karla *et al.*<sup>4</sup> and Siva Kumar<sup>5</sup> but smaller than those of Singh and Kumar<sup>3</sup>. Their bulb diameter (4.3-6.3 cm) was more than minimum desirable diameter (4.0 cm) for dehydration. Neck diameter (0.56-1.23 cm) of bulbs differed significantly (CD: 0.17 cm) at  $p < 0.05$  but they were in the range of 0.69-1.30 cm reported for other varieties<sup>1</sup>.

All the ten varieties were analysed for 9 chemical constituents (Table 2) which have been reported to influence the quality of dehydrated rings<sup>5</sup>. Their flesh moisture (84.9-88.8 per cent) were in the reported range of 83.5 to 88.7 per cent<sup>1,5</sup>. 'Pusa Red' and 'VL-1' had significantly high ( $> 15.0$  per cent) total solids. Sugars (total and reducing) were within the reported range,<sup>1,5</sup> but reducing sugars content did not follow the pattern of total sugars. Reducing sugar contents of 'Pusa White Round', 'Pusa Red', 'Punjab Red Round' and 'VL-1' were within the range ( $< 20$  per cent, dw) desirable for dehydration (Table 2). Alcohol insoluble solids (AIS) of all the ten varieties were above 1 per cent (fresh weight basis), the level reported to yield product with superior rehydration characteristics.<sup>2</sup> Mature bulbs are poor sources of ascorbic acid (AA) because of their low AA content (44.0-80.9 mg/100g of solids) and low consumption levels.

Pungency was estimated in terms of pyruvic acid (PA). It ranged from 2.9 to 12.8 mole/ml. juice (Table 1). 'VL-3'

TABLE 2. PHYSICO-CHEMICAL CONSTITUTENTS OF TEN ONION VARIETIES

Variety	Size <sup>1</sup>	Wt (g)	Neck dia. (cm)	Moisture (%)	TSS (%)	Sugars (% d.w.)		AIS	AA	PA (Mole)		Colour (OD/g)	
						Total	Reducing (% d.w.)			per ml per g TSS	Flesh	Peel	
Agrifound Light Red	6.2×6.0	86.1	0.86	87.2	10.3	68.6	20.6	17.6	64.7	5.2	50.2	3.2	48.4
Arka Niketan	5.8×5.8	81.3	1.01	88.5	11.2	66.4	23.5	22.8	80.9	11.0	97.9	1.9	48.8
Pusa White Round	4.3×4.4	34.7	0.76	87.1	9.7	54.4	18.0	24.1	55.1	5.2	53.3	0.5	2.1
Pusa White Flat	4.4×4.7	38.5	0.77	87.6	12.1	71.3	20.6	20.7	62.7	7.5	62.3	1.1	2.1
Pusa Red	5.0×4.8	63.9	0.56	85.0	13.3	64.5	11.4	24.8	44.0	7.4	55.6	3.1	93.0
Punjab Red	5.4×5.5	62.6	0.62	87.4	12.0	73.5	24.5	23.9	52.4	3.5	29.2	3.4	30.2
Punjab Red Round	5.8×5.7	96.3	0.55	88.8	8.9	56.0	18.7	23.2	58.9	6.8	76.0	5.7	71.9
Sel-102-1	5.5×5.2	86.4	1.23	86.0	13.0	50.7	18.1	19.4	49.7	2.9	22.3	4.5	62.7
VL-1	6.3×5.0	75.6	1.05	84.9	13.4	60.8	19.5	22.4	45.6	4.2	31.1	1.9	35.8
VL-3	5.9×6.6	91.6	0.88	88.6	9.2	50.4	18.5	23.5	63.4	12.8	139.5	2.1	135.2
Fc	7.95* 6.73*	8.19*	14.16*	18.79*	273.59*	19.88*	51.41*	97.25*	21.11*	87.28*	138.50*	445.60*	230.61*
C D	0.69 0.72	21.07	0.17	0.96	0.30	5.60	1.46	0.68	9.60	1.01	8.90	0.22	10.73

<sup>1</sup>Size: Diameter (cm) x Height (cm); AA: Ascorbic acid in mg/100g solids; PA: Pyruvic acid; Table F value at  $p = 0.05$  was 1.98 for size, weight and neck diameter, and 2.40 for others.

had significantly higher PA content and was followed by 'Arka Niketan'. These values are close to the values reported by Schwimmer and Guadagni<sup>9</sup>. Some of the varieties (viz., 'Agrifound' 'Light Red' and 'Pusa White Round' or 'Pusa White Flat' and 'Pusa Red') may have the same PA content on fresh weight basis but their sensory perception and pungency of varietal dehydrated rings may be different due to varietal variations in TSS and total solids. PA contents were, therefore, expressed on the basis of TSS and they changed their relative pungency rating based on PA content.<sup>16</sup> 'Arka Niketan' and 'VL-3' can be said to be of intermediate pungency; 'Pusa Red', 'Punjab Red Round' and 'Pusa White Flat' to be of moderately weak pungency and the rest 5 varieties to be of weak pungency. None were strongly and very strongly pungent.

Peel OD was much higher than the respective flesh OD (Table 2) because peels were darker than flesh. Peel OD correlated better with visual surface colour of whole bulbs. But peels are removed before dehydration and there was no significant correlation between the above two optical densities. Varieties should, therefore, be selected on the basis of flesh colour.

All the physico-chemical characteristics differed significantly within variety and no significant inter-relationship was observed between them, except for size-weight ( $r: 0.871, 0.792$ ) or moisture-TSS ( $r=-0.810$ ). These characteristics, thus depended upon variety only.

**Dehydration characteristics:** Browning, discolouration, loss of pungency and poor rehydration are the major problems of dehydrated onions. Some of the treatments<sup>11,13</sup> reported to overcome them were tried with 'Pusa Red', a commercial variety. Though the treatments reduced browning (NEB: 0.071-0.081 OD/g solids as against 0.275 OD/g solids for control), there was considerable loss of pungency (PA: 3.6-5.3  $\mu$  mole/g solids as against 7.2  $\mu$  moles/g solids for control

sample). Therefore, further studies were carried out without any pre-treatment.

Seven hours drying reduced the moisture content of onion rings to 4.3-7.6 per cent. Their shrinkage ratio (SR) was 7.2-11.0 (Table 3). Dehydration ratio (DR) found in this study (5.9-9.6) was close to the reported values<sup>3</sup> of 6.3-10.1. Their co-efficients of rehydration (CR) ranged from 0.53 to 0.74, CR of only 'Pusa Red', 'Sel-102-1' and 'VL-1' were above 0.70 and were significantly higher (Table 3). Moisture ( $r$  cal: -0.712), TSS ( $r$  cal: 0.859), SR ( $r$  cal: -0.712) and DR ( $r$  cal: 0.699) exhibited significant correlation with CR ( $r$  table: 0.632 at  $p < 0.05$  and 0.765 at  $p < 0.01$ ).

PA contents of dried onions were 2.1-7.2 moles/g solids as compared to the reported values of 3.97-6.83 moles/g samples<sup>2</sup>, 2.83-6.40 moles/g samples<sup>5</sup>, or 10.6 moles/g solids<sup>14</sup>. Though only few varieties were common to the above studies, range of residual PA was very narrow. A large percentage of PA is lost during dehydration and only 2.8-13.2 per cent of the original content was retained in the dehydrated product due to thermal or other degradative changes in pungency compounds, their volatilization<sup>17</sup> or due to both these factors.

Onions became darker upon drying and OD of methanolic extract of dried products were higher than those of corresponding fresh onion flesh (Tables 2 and 3). NEB of dehydrated pigmented varieties (excepting 'Pusa Red', 'Sel-102-1' and 'VL-3') were close to those of white varieties (Table 3). There was a significant positive correlation between colour of dried product and their NEB ( $r: 0.749$ ) or colour of fresh bulb flesh ( $r: 0.967$ ). This indicates that darker colour of the dried onions was due to the presence of pigment in fresh onion as well as due to NEB. Varieties with light red flesh like 'Arka Niketan' and 'VL-1' yielded light coloured dried products (Table 3).

TABLE 3. QUALITY CHARACTERISTICS OF DEHYDRATED RINGS OF TEN ONION VARIETIES

Variety	Shrinkage ratio	Dehydration ratio	Co-eff. of rehydration	PA	Colour	NEB (OD)	AA	RS	Ranking with score
Agrifound Light Red	8.5	7.5	0.60	3.0	3.49	0.147	13.2	20.4	M (5.4)
Arka Niketan	8.7	7.8	0.55	7.0	2.32	0.181	13.8	19.1	P (4.8)
Pusa White Round	8.5	7.3	0.59	5.3	0.98	0.184	18.5	16.5	M (5.1)
Pusa White Flat	9.3	8.3	0.60	3.5	1.77	0.123	14.1	18.0	M (5.2)
Pusa Red	7.2	5.9	0.73	7.2	3.60	0.275	17.5	9.9	V (7.8)
Punjab Red	7.4	6.6	0.60	2.3	3.55	0.170	14.0	20.8	G (6.6)
Punjab Red Round	11.0	9.6	0.55	2.1	5.82	0.345	15.7	17.2	P (4.8)
SEL-102-1	8.5	7.8	0.70	3.1	4.67	0.261	19.6	17.3	G (6.6)
VL-1	7.5	6.8	0.74	4.1	2.38	0.152	14.2	19.1	E (8.0)
VL-3	10.3	9.4	0.53	5.8	3.60	0.295	18.0	16.2	P (4.2)
Fcal	—	—	228*	158*	5.9*	853*	26.6*	459*	—
C D	—	—	0.015	0.44	1.71	0.008	1.33	0.42	—

F<sub>tab</sub> value at 5% level 1.98

PA: Pyruvic acid ( $\mu$  mole/g solid); Colour: Flesh colour (OD/g solids); AA: Ascorbic acid (mg/100 g solids); RS: Reducing sugar (g/100 g solids); E: Excellent (score 8.0); V: Very good (score 7.0-8.0); G: Good (score 6-7); M: Medium (score 5-6); P: Poor (score 5.0)

Drying reduced AA content by 60 to 80 per cent but reductions in corresponding percentages of reducing sugars and sugars ratio were slight (Tables 2 and 3). AA retention was comparatively higher (31.2-39.9 per cent) in 'Pusa Red', 'Sel-102-1', 'VL-1' and 'Pusa White Round' varieties. PA retentions in these four varieties were also higher (9.9-13.3 per cent) than the other six varieties (2.8-7.9 per cent). The first three varieties had high TSS of 13.0-13.4 per cent (Table 3).

Varieties were scored for quality of their dehydrated products. The scores ranged from 4.2 out of 10.0 for 'VL-3' to 8.0 for 'VL-1' (Table 3). They were ranked for their dehydration characteristics on the basis of these scores. On this basis, 'VL-1' can be termed as excellent for dehydration closely followed by 'Pusa Red' (score 7.8), and 'Punjab Red' and 'Sel-102-1' varieties (score 6.6) were good. White fleshed varieties, 'Pusa White Round' and 'Pusa White Flat', were of medium suitability.

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## Studies on the Preparation, Packaging and Storage of Inderse – An Indian Traditional Sweet

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**Inderse** – an Indian traditional deep-fat fried sweet is prepared from partially fermented rice flour and powdered sugar. Traditionally, it is prepared from rice flour soaked for 72 hr but the present studies revealed that the Inderse could be prepared from 24 hr soaked rice flour without affecting the quality/acceptability of the product. Attempts were made to prepare Inderse from *Lactobacillus plantarum* cultured rice flour. Equilibrium Relative Humidity (ERH) was determined to be 69 per cent. Samples stored well upto 45 days in LDPE (150 gauge), polypropylene (120 gauge), and friction top tins without affecting the quality characteristics significantly due probably to high ERH of the product. Moisture increase was well near the Equilibrium Moisture Content (EMC) levels at the ERH of the product. Free fatty acid content and Peroxide Value increased slightly in all the packagings without affecting the acceptability/quality of the product upto 45 days storage.

In India, a number of sweets are prepared from a variety of raw materials like Besan (Bengalgram flour), Urd dhal flour, Moong dhal flour, Maida (refined wheat flour) and rice. Studies have been carried out on some of the traditional sweets like Gulab Jamun<sup>1-3</sup>, Shrikhand<sup>4</sup>, Burfi and Pera<sup>5</sup>, Sohan Halwa<sup>6</sup>, and Rewari and Gajak<sup>7</sup>. Inderse is a deep-fat fried sweet, prepared from partially fermented rice flour and sugar and is consumed in the States of Uttar Pradesh, Rajasthan, Madhya Pradesh as well as in parts of Pakistan<sup>8</sup>. But, very little scientific data and published information are available<sup>8</sup> on this sweet. Hence, systematic studies were carried out on the preparation, packaging and storage aspects of this sweet to ensure improvement in the quality and to extend shelf-life. The results of these studies are presented in this paper.

### Materials and Methods:

**Raw materials:** Rice (var. 'Parmal'), sugar, Vanaspati (hydrogenated fat) (Dalda brand) and poppy seeds (khas khas) were procured locally.

**Preparation of Inderse:** Cleaned rice was washed in tap water and soaked in excess water for optimum of 24 hr at room temperature and one sample was incubated with *Lactobacillus plantarum* culture for 12 hr. Excess water was drained off and soaked rice was then dabbed between the muslin cloth to remove adhering water. The soaked rice was ground in a waring blender to pass through 60 mesh sieve. Rice powder was mixed with dry powdered sugar (60 mesh) in the ratio of 1:1 on dry raw rice basis. The mix was made into balls and kept aside for 24 hr at room temperature

( $30 \pm 5^\circ\text{C}$ ). The fermented rice-sugar mix balls were broken and kneaded into dough with water (8 per cent on rice basis). The dough was made into small balls weighing 10-15 g. These balls were flattened into circular discs (about 5-6 cm diameter and 1 cm thick) on a wooden plate by pressing with fingers and dressed with poppy seeds (khas-khas) on one side. These discs, facing cressed side upward, were deep-fat fried at  $160^\circ\text{C}$  in vanaspati until turned into brown. The Inderse sweets thus prepared were cooled to room temperature ( $30^\circ\text{C}$ ) and packed for storage studies. Details are described in the Material balance sheet (Fig 1).

**Equilibrium relative humidity (ERH) studies:** The ERH studies were carried out by storing the Inderse in dessicators maintained at 11 to 92 per cent relative humidity at room temperature ( $\pm 0 \pm 5^\circ\text{C}$ ) as per the method described by Rockland<sup>9</sup>.

**Packaging and storage studies:** Inderse samples (100 g each) were packed in flexible pouches (LDPE 150 gauge, polypropylene 120 gauge) and friction-top tins and stored at room temperature ( $30 \pm 5^\circ\text{C}$ ).

**Physico-chemical analysis:** Inderse samples were analysed for unit weight, unit diameter, thickness, shape, moisture, crude fat, reducing sugars, total sugars, total ash, acid insoluble ash, free fatty acids (FFA) and peroxide value (PV). The moisture, FFA and PV were determined at intervals of 0, 15, 30, 45 days during storage.

**Sensory evaluation:** Inderse samples were evaluated by a panel of fellow scientists for their quality characteristics like colour, texture, aroma, taste and flavour and overall quality.

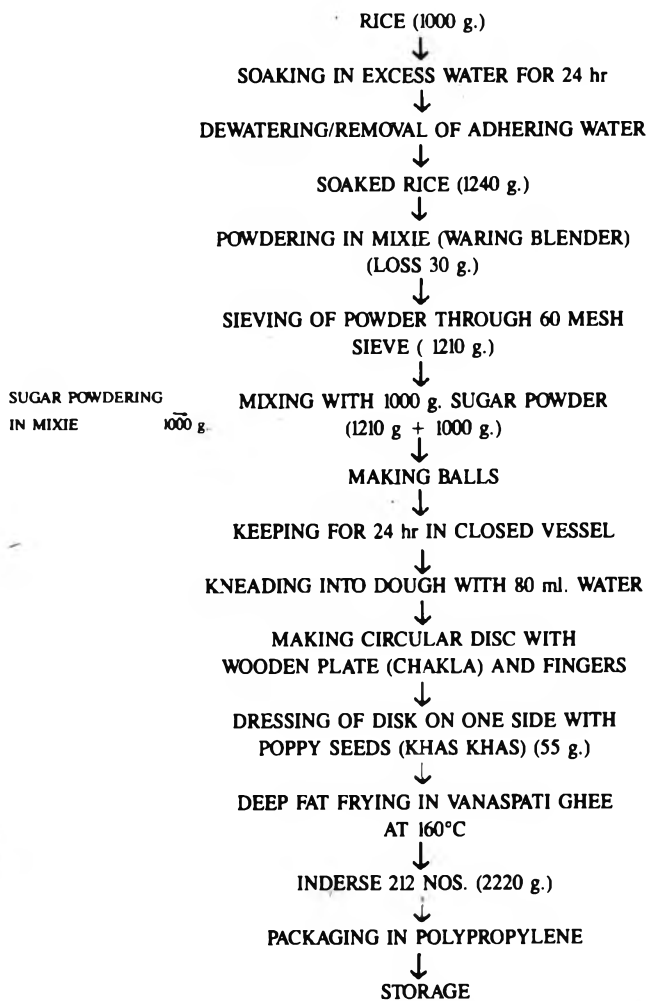


Fig. 1. Materials balance sheet on the preparation of inderse.

## Results and Discussion

The rice samples contained an initial moisture of 9.8 per cent which increased to 31.2 per cent upon soaking in water

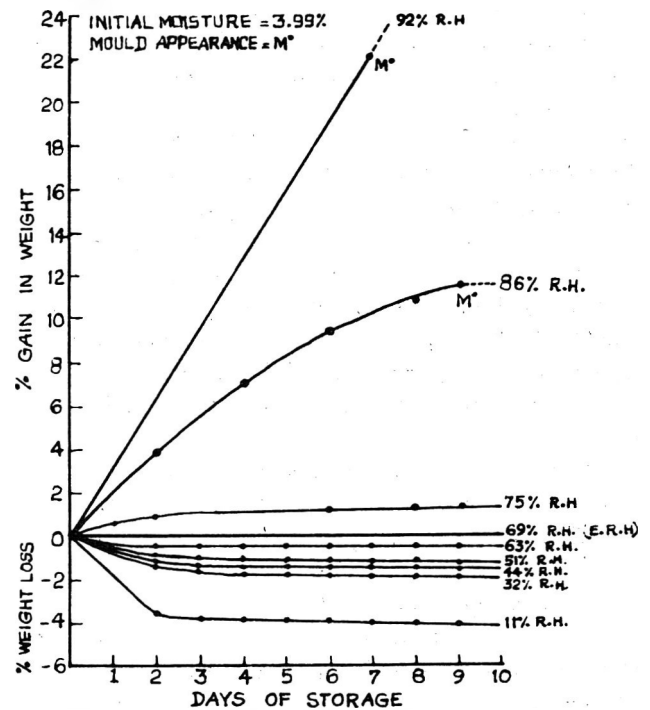


Fig. 2. Per cent change in weight of inderse at different relative humidities at R.T. (30±5°C).

for 24 hr but did not increase further beyond 24 hr soaking. Acidity increased from nil to 0.12, 0.18 and 0.20 per cent during soaking for 24, 48 and 72 hr respectively. The pH changed from 7.5 (initial) to 4.00 (final) during 48 hr soaking. Samples cultured with *L. plantarum* attained an acidity of 0.18 per cent during 24 hr. Slight off flavour/fermented flavour was noticed in all samples soaked beyond 24 hr.

From Table 1, it is seen that Inderse samples prepared from 24 (I) 48 (II), 72 (III) hr soaked rice and *L. plantarum* fermented rice (IV) had light-brown colour. Sample I was considered quite acceptable since it possessed soft, crisp and

TABLE 1. EFFECT OF SOAKING TIME ON THE QUALITY OF INDERSE

Soaking period (hr)	Colour	Texture	Aroma	Taste & flavour	Overall quality
24 (I)	Light brown, desirable	Chewy, soft, crisp. 80% honey-comb structure	Good	Good, peculiar of Inderse	Acceptable
48 (II)	Brownish, slightly undesirable	Chewy, soft, 80% honey-comb structure	Fermented flavour	Poor, off fermented flavour	Not acceptable
72 (III)	Brown, un-desirable	Hard, chewy 100% honey-comb structure	..	Poor, perceptible off fermented flavour	Not acceptable
<i>L. plantarum</i> fermentation for 12 hr (IV)	Light brown, desirable	Chewy, hard No honey comb structure cracked surface	Oily, aroma of lactic acid fermentation	Good taste	Off-flavour not acceptable

chewy texture, good flavour and taste typical of Inderse, and a good honey-comb structure (80 per cent). Other samples had peculiar undesirable fermented flavour and hence were not acceptable.

The physico-chemical analysis data for Inderse are given in Table 2. They were round/circular disc shaped with honey-comb structure, having a diameter ranging from 5 to 6.6 cm, thickness ranging from 0.85 to 1.46 cm and unit weight ranging from 10 to 16 g. It contained moisture 3.99 per cent, crude fat/ether extractives 22.39 per cent, total sugars 35.45 per cent and total ash 0.77 per cent. There was no acid insoluble ash as samples were prepared under laboratory conditions. The FFA content was 0.74 per cent and PV was 8.17 milli equiv. O<sub>2</sub>/kg oil.

Storage behaviour of Inderse under different humidities ranging from 11 to 92 per cent RH is presented in Fig 2 and 3. The ERH of Inderse was quite high (69 per cent RH) at which no loss or gain in moisture was noticed. The sensory quality of the samples did not change upto 75 per cent RH at equilibrium moisture content of 5.38 per cent. However, beyond this RH, the product became soggy and unacceptable. Mould appeared on the samples at 86 and 92 per cent RH at the EMC level of 15.34 and 25.97 per cent on 9th and 7th day, respectively.

The samples packed in LDPE, PP and friction-top-tins, remained quite acceptable upto 45 days of storage without showing any discernible change in chemical and organoleptic

TABLE 2. PHYSICO-CHEMICAL CHARACTERISTICS OF INDERSE PREPARED FROM 24 HR SOAKED RICE IN THE LABORATORY

Parameters	Values/characteristics
Unit wt (g)	15.0 (10-16)
Diameter (cm)	5.86 (5-6.6)
Thickness (cm)	1.38 (0.85-1.46)
Shape	Round/circular disc with honey-comb structure
Colour	Light brown
Texture	Soft, crisp, chewy
Aroma	Good
Taste & flavour	Good
General appearance	Desired honey-comb structure
Moisture (%)	3.99
Crude fat (%)	22.39
Reducing sugars (%)	Nil
Total sugars (%)	35.45
Total ash (%)	0.77
Acid insoluble ash (%)	Nil
Free fatty acid (as % oleic acid)	0.74
Peroxide value (milli equiv O <sub>2</sub> /kg oil)	8.17

Results of chemical analysis are the average of 2 replicates

Figures in parantheses indicate range values.

TABLE 3. CHANGES IN MOISTURE, FFA AND PV IN INDERSE SAMPLES STORED IN DIFFERENT CONTAINERS AT ROOM TEMPERATURE (30±5°C)

Storage period (days)	Moisture (%)	FFA (as % oleic acid)	PV (million equiv. O <sub>2</sub> /kg oil)
<b>Polythene pouch (150 gauge)</b>			
0	3.99	0.74	8.17
15	4.10	0.77	8.90
30	4.25	0.84	9.80
45	4.20	0.86	10.00
<b>Polypropylene pouch (120 gauge)</b>			
0	3.99	0.74	8.17
15	3.86	0.76	8.70
30	3.59	0.77	8.92
45	3.82	0.86	9.82
<b>Friction-top-tin</b>			
0	3.99	0.74	8.17
15	3.90	0.80	8.80
30	4.00	0.86	9.00
45	4.10	0.89	9.40

quality characteristics, which may be due to the high ERH of the product. The FFA content increased slightly from 0.74 to 0.89 per cent (maximum) in the product stored in friction-

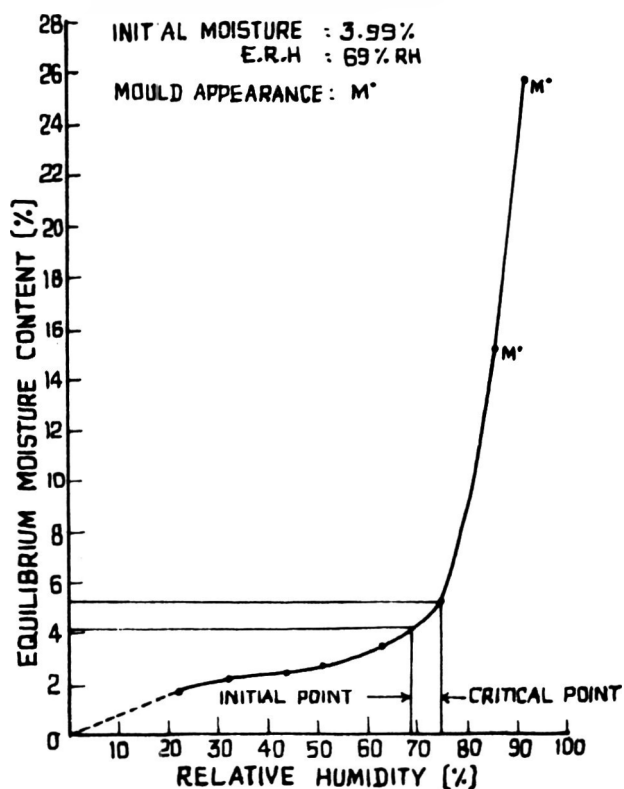


Fig. 3. Relationship between relative humidity (R.H.), equilibrium moisture content (E.M.C.) and number of days to attain equilibrium for inderse.

top tins, while PV increased from 8.17 to 10.00 milli equiv. O<sub>2</sub>/kg oil in the product stored in LDPE pouches. Nevertheless, the product remained quite acceptable without any off flavour/rancid flavour and taste (Table 3).

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## Energy Management in Baking Industry

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**Energy consumption pattern in bread manufacturing industry was studied by energy accounting system. The baking oven used 90% of the total energy input. The overall energy consumption in the production of 1405.2 kg bread/day was 44716.32 MJ. Several energy saving measures are suggested.**

During the last decade, the third world countries, including India have been severely hit by the increased cost of energy. Imported oil, in most countries, has caused a considerable drain on foreign exchange reserve. Energy management in industrial operation in these countries carries extra significance<sup>1</sup>.

The food industry is highly diversified with processing plant ranging from small to large industrial units and use all types of fuel sources besides electrical energy. A fundamental step in any energy management effort is the comparison of the actual energy consumption with the minimum energy requirement of the process in question.<sup>2</sup> There have been numerous reports on energy requirement associated with food industries of Western countries. The reports<sup>3,5</sup> clearly indicated the energy intensiveness of food processing industry.

Laukkanen<sup>3</sup> reported total energy consumption in bakery industry of Finland was about  $0.2 \times 10^4$  MJ or 0.5 per cent of the total consumption of energy of the Finland industry. One of the major energy consuming food industries in Sweden is the baking industry<sup>4</sup>. In the United States of America, energy consumption of 7.26 MJ/kg bread baked was reported in a baking industry which produced 35,000 kg bread/day<sup>5</sup>.

Literature related to the Indian food industry wide energy consumption survey is rather fragmentary. It calls for thorough survey on energy consumption pattern of Indian food industries, including baking industry which would provide the energy consumption cost data on which realistic energy demand forecasts can be based. Present investigations were undertaken with the following objectives: (1) to compute energy consumption data of each baking equipment and

process, (2) to compute energy efficiency of entire processing lines, and (3) to identify energy intensive operations in baking industry as a basis for establishing feasibility for reduction of energy requirement.

### Materials and Methods

The survey on energy utilization in bread manufacture was conducted in the baking industry (5000 kg bread/day) located in Jabalpur city. Following data collected in one shift of 8 hr were considered : 1. Power consumption, 2 fuel oil consumption, 3 coke consumption, 4 labour hr and 5 production. The data were collected for 30 days and average values are reported. The theoretical or anticipated energy consumption of each baking equipment was calculated based on input-output analysis of bread production and actual or practical energy consumption data of each process equipment was measured individually for electrical, fuel oil, coke fuel and man energy. The electrical energy was measured in terms of Kilo-Watt hr with the help of an energy meter. Fuel oil energy was measured in terms of volume of fuel whereas solid fuel (Coke) energy was accounted in terms of weight. The energy utilization efficiency of each process equipment used in baking was calculated in terms of energy use index (Ei) by the following expression<sup>6</sup>:

$$\text{Energy use index (Ei)} = \frac{\text{Actual consumption} - \text{Anticipated consumption}}{\text{Anticipated consumption}} \times 100$$

The energy consumption pattern of each unit operation and for entire process was presented in the form of an energy



TABLE 1. AVERAGE ENERGY CONSUMPTION DATA AND ENERGY-USE INDEX OF VARIOUS EQUIPMENTS USED IN BREAD MANUFACTURE.

Equipments	Electrical energy (MJ)			Man energy (MJ)		
	Anticipated	Actual	Ei (%)	Anticipated	Actual	Ei (%)
Screw conveyor	4.18	4.82	15.31	0.41	0.51	24.39
Flow shifter	3.94	4.75	20.56	0.39	0.51	30.76
Dough kneader	45.90	49.39	7.60	4.50	5.00	11.11
GMS solution	1.35	1.80	33.3	0.63	0.71	12.69
Plant	—	15.37	—	—	—	—
Divider	12.71	16.69	31.31	2.24	2.77	23.66
Rounder	11.23	11.52	2.58	1.12	1.63	45.53
Interprover table	11.23	11.73	4.45	1.12	1.59	41.96
Moulder	26.20	26.44	0.90	3.51	4.04	15.09
Slot conveyor (1)	8.29	9.14	10.25	2.43	2.95	21.39
Final prover trolley	—	—	—	1.02	1.16	13.72
Final prover	—	—	—	2.60	3.88	49.23
Oven	135.32	158.66	17.25	4.50	6.09	35.33
Slot conveyor (2)	8.10	8.60	6.17	2.43	2.62	7.82
Depaning table	8.10	8.31	2.59	1.59	2.18	37.10
C.T. trolley	—	—	—	0.52	0.88	69.23
Cooling tunnel	19.02	21.87	14.98	—	—	—
Slicing	46.64	52.05	11.59	6.03	6.40	6.14
Packaging	4.68	6.92	47.86	0.47	0.48	2.13
Water pump	—	1.96	—	—	—	—
Diesel pump	—	0.72	—	—	—	—
Pusher	12.67	14.22	12.23	—	—	—
Diffuser	63.57	64.80	1.93	—	—	—
Other	—	—	—	—	12.83	—
<b>Total</b>	<b>389.65</b>	<b>505.76</b>	<b>29.79</b>	<b>35.51</b>	<b>56.23</b>	<b>58.34</b>

accounting diagram (EAD) by following the method of Singh et al.<sup>1</sup>

**Results and Discussion**

The energy consumption pattern for manufacturing 1405.2 kg bread/day or one shift of 8 hr is given in Table 1 and energy

flow in the bread production is shown in the energy accounting diagram (EAD), (Fig. 1). It has been observed that there were three main energy intensive processing steps in bread making i.e. 1. mixing in which mechanical energy is produced by electricity, 2. fermentation where thermal energy in the form of steam from coke as well as electrical energy is employed and 3. baking-in which thermal energy is provided both by electricity and liquid fuel.

The proportions of various energy input in the bakery were 91.74, 6.99, 1.13 and 0.125 per cent from liquid fuel, coke, electricity and labour respectively. The baking oven used more than 90 per cent of the total energy input. It is obvious because oven used liquid fuel, electricity and labour energies. Next to baking oven, fermentation unit used both electrical and steam produced by solid fuel.

The anticipated and actual energy consumption data of electrical, man, solid and liquid fuel in the baking process were 389.65, 505.76, 35.54 and 56.25, 2289.26, 3127.38, 38507.10 and 41027.16 MJ, respectively. The anticipated energy consumption of each baking equipment was compared with the practical or actual energy consumption to find out the energy use index given in Table 1. The energy use index of total electrical energy in the baking was 29.76 per cent, which indicated that 70.21 per cent of the total electrical energy was utilized by various baking equipments. Similarly, solid fuel, liquid fuel and labour energy utilization in the process were 63.35, 93.46 and 43.77 per cent respectively.

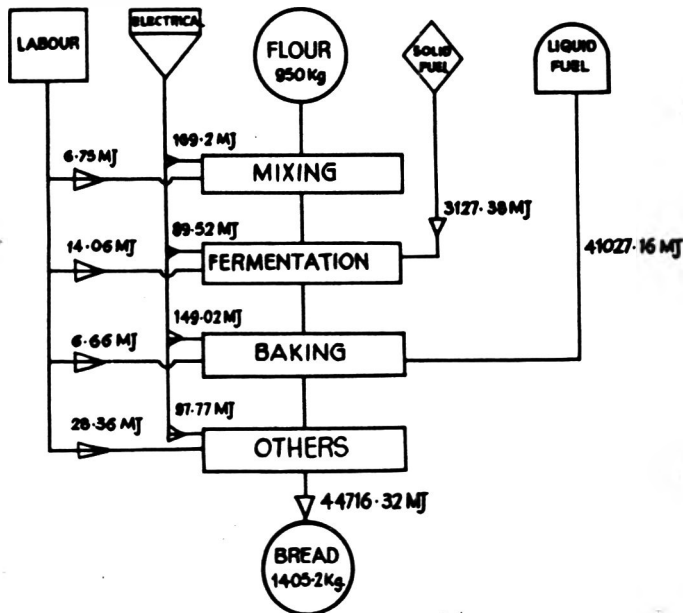


Fig.1. Energy accounting diagram (EAD) of bread production.

The overall energy consumption in the production of 1405.2 kg bread was 44716.32 MJ or 31.82 MJ/kg bread. Tragardh *et al.*<sup>7</sup> reported total energy consumption pattern of two Swedish bakeries were about 13.96 and 4.88 MJ/kg bread, respectively for a capacity of 2,50,000 and 3,50,000 kg bread Table 2 per year.

The energy cost data of bread manufacture (1405.2 kg bread/day) were calculated based on observed values of energy consumption (Table 2). It has been observed that in bread manufacture, highest money is spent on liquid fuel energy. The cost of bread manufacture calculated based on energy consumption data is about Re.0.67/kg bread.

Based on the above findings, following energy saving measures are suggested : Correct use of the equipments (time of use, correct control valves, use of automation, etc.) and regular service are the most important and best energy saving measures.

The most important source of waste of heat in bakery is the exhaust from the baking oven. Installation of waste energy (heat) recovery system could save the total energy waste to a great extent.

TABLE 2. ENERGY COST ANALYSIS FOR BREAD MANUFACTURE

Type of energy	Measurement unit	In MJ unit	Approx. energy cost (Rs Ps)
Electrical	505.76 H.P.hr.	505.76	5.05
Solid fuel	747103.85 Kcal	3127.38	157.75
Liquid fuel	109800.00 Kcal	41027.16	410.00
Labour	16.56 H.P.hr.	56.23	372.65
Total		44716.53	945.45

It may be stated in conclusion that suitable energy saving system should be introduced to conserve the energy waste during different processes identified in the baking industries, which not only save the energy for processing but also save money. If heat recovery measures are to be taken in the bakery, the designer should be in continuous close co-operation with the operating personnel and the effect of heat recovery measures on the process and *vice-versa* should not be ignored.

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## Production of *Narampak Sandesh* from Buffalo Milk

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**The technology of *narampak sandesh* production from buffalo milk with reference to its sugar level, fat content in milk and the method of heating during cooking was standardised. On the basis of sensory evaluation, best results were indicated with the use of 30% crystalline cane sugar and 4% fat in milk. For a good quality *sandesh* preparation, slow process of heating was essential. Addition of *chhana* in two instalments markedly improved the body and texture of the finished product. Chemical and microbiological results obtained would be useful in formulating the standards of *narampak sandesh*.**

*Sandesh*, a very popular and oldest *chhana*-based sweetmeat of Bengal<sup>1</sup> is gaining popularity in other regions of the country<sup>2</sup>. Traditionally, *sandesh* is prepared by *Halwais* on a small scale. Three kinds of *sandesh* are available in market viz., *karapak* (low moisture grade), *narampak* (medium moisture grade) and *kachhagolla* (high moisture grade)<sup>3,4</sup>, of which *narampak* is the most popular variety<sup>5</sup>. The National Commission on Agriculture (1976) had recommended that efforts should be made to standardise the production of various indigenous milk sweets which might be adaptable for industries<sup>6</sup>. *Sandesh* making has an old history<sup>7</sup>, with the art of making has remained in the hands of *Halwais*<sup>8,9</sup>. As a result of this, not much has been reported in literature about the technology of *sandesh*. In view of the above facts, an attempt has been made in this study to standardise the technology for the production of *narampak sandesh* from buffalo milk.

### Materials and Methods

**Production of *chhana*:** Fresh pooled buffalo milk was procured from the Experimental Dairy of the Institute. It was filtered through a muslin cloth and standardised to 2.0, 3.0, 4.0, 5.0 and 6.0 per cent fat with the help of buffalo skim milk as per requirement. The SNF content of milk ranged from 9.6 to 9.9 per cent with an average of 9.8 per cent. At a time, 50 l standardised milk was taken in a double jacketted steam kettle and *chhana* was made by the procedure of Kundu and De<sup>10</sup> with certain modifications. Here just after boiling the milk, 30 per cent potable water (v/v) in relation to the quantity of milk taken was added in the milk, cooled to 70°C, coagulated with 0.5 per cent citric acid solution, strained immediately for rapid expulsion of whey, and *chhana* was

pressed for 10 min to expedite further removal of whey. The moisture per cent of *chhana* ranged from 55 to 59. It was then ground to a smooth paste with the help of a power operated mechanical grinder.

**Production of *sandesh*:** On the basis of preliminary trials, 2.0 kg ground *chhana* was taken for each trial and this was further divided into two equal lots. Different proportions of cane sugar viz., 25,30,35 and 40 per cent (by weight of total *chhana*) was added separately with the first lot of *chhana* in a double jacketted steam kettle for ascertaining the optimum sugar level in the final product. Temperature of the *chhana* and sugar mixture was then gradually raised to about 70°C in 15 min by continuous stirring and scraping with the help of a wooden ladle. At this juncture, the mixture developed its initial pat stage and the remaining lot of *chhana* was added. Due to this addition, the temperature of the mixture suddenly fell to about 46°C and it was then slowly heated to about 60°C in 10 min with continuous stirring and scraping as before. At this point, the mixture exhibited its final pat stage. Heating was discontinued at this stage and the product is cooled slowly to 37°C in about 10 min, transferred it to rectangular trays and cut into desired sizes. The resultant *sandesh* samples were subjected to sensory, chemical and microbiological analyses.

**Time-temperature combination during *sandesh* production:** During the cooking process, temperature of the product was varied by regulating the steam inflow. The product temperature was recorded at a regular interval of time with a thermometer. This method provides a fair idea about the effect of various time-temperature combinations on the quality of the finished product.

**Analysis of milk:** Fat and SNF in milk were determined as per IS : 1223<sup>11</sup> and IS : 10083<sup>12</sup>, respectively.

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**Sensory evaluation of sandesh:** The sensory characteristics of *sandesh* samples were evaluated by a panel of 7 trained judges in terms of flavour, body and texture, colour and appearance, sweetness and overall acceptability using the 9-point Hedonic scale<sup>13</sup>. Scores of the judges were also statistically analysed to find out the extent of variations between samples, varieties, treatments, judges and interactions thereof.

**Chemical analysis of chhana and sandesh:** The moisture in *chhana* and the moisture, fat, protein and ash contents in *sandesh* were determined by the IS : 2785 methods<sup>14</sup>. Total sugar was estimated by the difference. The sum total of moisture, fat, protein and ash were subtracted from 100. Titratable acidity was calculated following the procedure of Sachdeva and Rajorhia<sup>15</sup> for *burfi*. The water soluble acidity was estimated as per the method of AOAC<sup>16</sup> described for *cheese*. Free fatty acidity (FFA) was determined as per IS : 3508<sup>17</sup> with some minor modifications as described by Sen and Rajorhia<sup>3</sup>. Free fat (FF) was analysed by the recommended procedure of Hall and Hedrick<sup>18</sup> originally developed for milk powder. The product was mashed thoroughly with a stainless steel grater before weighing. The para dimethyl amino benzaldehyde reactivity (p-DMAB) test was carried out according to the method of Kumar and Hansen<sup>19</sup> during the processing of *sandesh*. The pH was measured with a digital pH meter as described by Kosikowski<sup>20</sup> for *cheese*. The superficial density was calculated by weighing blocks of 4.0 cm<sup>3</sup> size and expressed as g/cm<sup>3</sup>. Hardness was measured with a cone penetrometer as per Sen and Rajorhia<sup>4</sup>.

**Microbiological analysis of sandesh:** The total viable counts (TVC), coliform counts and yeast and mould counts were determined by the method of APHA<sup>21</sup> using tryptone dextrose yeast agar, violet red bile agar and potato dextrose agar, respectively. Spore counts were enumerated following

the procedure described in BS : 4285<sup>22</sup> using tryptone dextrose yeast agar. The staphylococcus counts were determined as per the recommended method of APHA using staphylococcus medium S<sub>10</sub> agar<sup>23</sup>.

## Results and Discussion

**Optimisation of sugar level:** The results of sensory evaluation of *sandesh* samples made from the four different levels of sugar are presented in Table 1. From the Table, it could be seen that the highest scores in all the sensory attributes were awarded to the *sandesh* samples prepared with 30 per cent sugar. *Sandesh* samples made with 25 and 35 per cent sugar were considered slightly less and more sweet, respectively. Incorporation of 40 per cent sugar made the product excessively sweet and yielded the lowest sensory scores (Table 1). Variation in the level of sugar brought about minimum changes in the sensory scores of colour and appearance. Both under and over sweetening reduced the acceptability scores of *sandesh* significantly. This can be observed from the analysis of variance data (Table 2). Sugar levels significantly influenced the flavour, body and texture and overall acceptability of *sandesh*. The colour and appearance were not much affected by the variation in sugar content. Likewise, no significant variations were noted between replicates, judges and interactions between treatments and judges. Earlier workers<sup>15,24,25</sup> have also advocated the incorporation of 30 per cent sugar in *burfi*. The chemical analysis of *sandesh* (Table 3) showed that the addition of higher amount of sugar progressively reduced the fat, protein and ash contents in *sandesh* but the moisture content neither changed appreciably nor it followed any definite trend. In these experiments, *sandesh* samples were prepared from milk of 5.0 per cent fat as recommended by Kundu and De<sup>10</sup>. Dissolution of crystalline sugar in *sandesh* preparation was not a problem. Incorporation of 40 per cent sugar even did

TABLE 1. INFLUENCE OF CANE SUGAR ON THE SENSORY CHARACTERISTICS OF SANDESH

Sensory characteristics	% of cane sugar by wt of <i>chhana</i>			
	25.0	30.0	35.0	40.0
Flavour	6.5 - 8.0 (7.50 ± 0.09)	7.5 - 9.0 (8.43 ± 0.09)	7.0 - 9.0 (8.36 ± 0.11)	6.0 - 8.0 (7.25 ± 0.19)
Body and texture	7.0 - 8.5 (8.02 ± 0.08)	7.5 - 9.0 (8.46 ± 0.09)	7.0 - 9.0 (8.30 ± 0.10)	7.0 - 8.5 (7.84 ± 0.11)
Colour and appearance	7.0 - 8.5 (8.11 ± 0.07)	7.5 - 9.0 (8.27 ± 0.09)	7.5 - 9.0 (8.16 ± 0.08)	7.5 - 9.0 (8.05 ± 0.09)
Sweetness	6.0 - 8.0 (7.57 ± 0.10)	8.0 - 9.0 (8.55 ± 0.08)	7.5 - 9.0 (8.39 ± 0.08)	6.0 - 8.0 (7.11 ± 0.10)
Overall acceptability	6.5 - 8.0 (7.43 ± 0.08)	7.5 - 9.0 (8.36 ± 0.09)	7.5 - 9.0 (8.25 ± 0.10)	6.0 - 7.5 (7.05 ± 0.09)
Sensory comments	Less sweet	Optimum sweetness	Slightly higher sweetness	Highly sweet

Figures in the parentheses are the means with standard errors.

Values are in range

TABLE 2. ANOVA FOR LEVELS OF CANE SUGAR ON THE SENSORY QUALITY OF BUFFALO MILK SANDESH

Source of variation	df	Mean sum of squares				
		Flavour	Body and texture	Colour and appearance	Sweetness	Overall acceptability
Between replicates	3	0.26	0.33	0.04	0.36	0.07
Between treatments	3	9.98**	2.20**	0.23	13.13**	11.24**
Between judges	6	0.05	0.37	0.07	0.06	0.11
Between treatments × judges	18	0.04	0.38	0.09	0.05	0.10
Error	81	0.34	0.22	0.23	0.28	0.29
Total	III					

\*\*Significant at 1.0 per cent level.

TABLE 3. EFFECT OF CANE SUGAR ON THE CHEMICAL QUALITY OF SANDESH

Constituents	% of cane sugar by wt of <i>chhana</i>			
	25.0	30.0	35.0	40.0
Moisture (%)	24.82 – 26.47 (25.86 ± 0.38)	25.43 – 27.00 (26.07 ± 0.34)	25.97 – 27.25 (26.52 ± 0.27)	24.87 – 26.31 (25.63 ± 0.30)
Fat (%)	22.24 – 23.57 (23.04 ± 0.29)	19.70 – 20.52 (20.62 ± 0.19)	17.05 – 18.32 (17.73 ± 0.29)	15.00 – 16.23 (15.64 ± 0.27)
Proteins (%)	18.73 – 20.31 (19.51 ± 0.34)	16.98 – 17.75 (17.37 ± 0.16)	14.39 – 15.04 (14.85 ± 0.15)	12.82 – 14.03 (13.44 ± 0.25)
Total sugars (%)	23.93 – 30.12 (29.45 ± 0.25)	33.88 – 34.65 (34.35 ± 0.16)	38.87 – 39.87 (39.31 ± 0.24)	43.79 – 44.08 (43.92 ± 0.07)
Ash (%)	1.98 – 2.29 (2.13 ± 0.06)	1.78 – 2.03 (1.95 ± 0.06)	1.42 – 1.71 (1.59 ± 0.06)	1.16 – 1.48 (1.37 ± 0.07)

Figures in the parentheses are the means with standard errors.

Values are in range.

not leave any undissolved sugar granules. Moisture content in *chhana* and period of heating were enough to dissolve all the sugar crystals. These observations differed from the procedures described by earlier workers<sup>26-28</sup> who had recommended powdered sugar for *sandesh* making.

**Optimisation of fat level in milk:** The sensory scores and characteristics of *sandesh* samples prepared from milk of the five different fat levels are presented in Table 4. It may be observed that low fat milk resulted in a dry and weak body and coarse texture. The product also lacked the typical rich flavour. *Sandesh* prepared from buffalo milk with 4.0 per cent fat was rated as the best in all the sensory attributes. These scores were followed by the *sandesh* samples made from the milk containing 5.0, 6.0, 3.0 and 2.0 per cent fat, respectively. The analysis of variance data (Table 5) of the sensory scores of *sandesh* highlighted that the fat content in buffalo milk significantly affected the flavour, body and texture, colour and appearance and overall acceptability of *sandesh*. Changes due to replicates, judges and interactions between treatments and judges were insignificant. It can be seen from Table 6 that the free fat content in *sandesh* increased progressively with the increase in the fat per cent in milk. Free fat

proportion in *sandesh*, produced from 4.0 per cent milk fat was considerably lower (61.42) than the samples prepared from either 5.0 (66.27) or 6.0 (74.53) per cent milk fat. For finding out the desired fat level in milk, *sandesh* samples were prepared separately from the above five different fat levels of milk using 30 per cent cane sugar by weight of *chhana*. Obviously, the fat content in *sandesh* increased progressively with the increase in fat percentage in milk while the protein and ash contents reduced the moisture and sugar contents did not exhibit any specific trends. However, considering the results of sensory properties and free fat contents in *sandesh*, it may be recommended that the buffalo milk could be standardised to 4.0 per cent fat for *sandesh* production.

**Time-temperature combination during cooking of *sandesh* production:** In the preliminary experiments, when the mixture of entire amount of *chhana* and sugar was heated in the jacketed kettle at 100°C or above with simultaneous stirring and scraping, the product became extremely hard, brittle and chewy with very little cohesiveness in 5-10 min. It was, therefore, decided that the mixture should be heated at less than 100°C. Incorporation of *chhana* in two instalments

TABLE 4. EFFECT OF FAT CONTENT IN BUFFALO MILK ON THE SENSORY CHARACTERISTICS OF SANDESH

Sensory characteristics	Sensory quality at indicated Fat % in milk				
	2.0	3.0	4.0	5.0	6.0
Flavour	6.0 – 8.0 (6.61 ± 0.13)	7.0 – 9.0 (8.05 ± 0.09)	7.5 – 9.0 (8.43 ± 0.09)	8.0 – 9.0 (8.34 ± 0.07)	7.0 – 9.0 (8.27 ± 0.10)
Body and texture	6.0 – 8.0 (7.02 ± 0.10)	7.0 – 9.0 (8.32 ± 0.09)	8.0 – 9.0 (8.46 ± 0.08)	7.5 – 9.0 (8.34 ± 0.09)	7.5 – 9.0 (8.14 ± 0.08)
Colour and appearance	6.0 – 8.0 (7.16 ± 0.11)	7.0 – 9.0 (7.87 ± 0.09)	7.5 – 9.0 (8.52 ± 0.08)	7.5 – 9.0 (8.41 ± 0.09)	7.5 – 9.0 (8.18 ± 0.09)
Sweetness	7.0 – 8.0 (7.86 ± 0.09)	7.5 – 8.5 (8.09 ± 0.07)	8.0 – 9.0 (8.66 ± 0.07)	7.5 – 9.0 (8.48 ± 0.10)	7.0 – 9.0 (8.21 ± 0.11)
Overall acceptability	6.0 – 7.5 (6.70 ± 0.08)	7.0 – 8.5 (7.86 ± 0.10)	7.5 – 9.0 (8.43 ± 0.08)	7.5 – 9.0 (8.27 ± 0.08)	7.5 – 9.0 (8.16 ± 0.08)
Sensory comments	Dry, coarse, lacks richness	Weak, coarse	Most liked	Good, acceptable	Moderately good, excessive free fat

Figures in the parentheses are the means with standard errors.

Values are in ranges.

TABLE 5. ANOVA DATA FOR DIFFERENT LEVELS OF FAT ON THE SENSORY ATTRIBUTES OF BUFFALO MILK SANDESH

Source of variation	df	Mean sum of squares				
		Flavour	Body and texture	Colour and appearance	Sweetness	Overall acceptability
Between replicates	3	0.17	0.04	0.18	0.19	0.23
Between treatments	4	15.83**	9.82**	8.29**	2.82	13.52
Between judges	6	0.45	0.18	0.49	0.18	0.34
Between treatments x judges	24	0.25	0.07	0.34	0.27	0.11
Error	102	0.26	0.28	0.23	0.22	0.23
Total	139					

\*\*Significant at 1.0% level.

TABLE 6. INFLUENCE OF FAT CONTENT IN BUFFALO MILK ON THE CHEMICAL QUALITIES OF SANDESH

Constituents	Fat % in milk				
	2.0	3.0	4.0	5.0	6.0
Moisture (%)	26.43 – 28.27 (27.36 ± 0.40)	25.86 – 27.35 (26.49 ± 0.31)	25.45 – 27.01 (26.23 ± 0.34)	25.83 – 27.25 (26.74 ± 0.33)	24.91 – 26.54 (25.82 ± 0.34)
Fat (%)	12.09 – 14.15 (13.25 ± 0.45)	15.50 – 17.22 (16.14 ± 0.37)	17.68 – 18.42 (18.09 ± 0.17)	19.58 – 20.76 (20.08 ± 0.25)	21.16 – 22.49 (21.94 ± 0.28)
Proteins (%)	21.94 – 23.61 (22.83 ± 0.34)	20.53 – 22.03 (21.02 ± 0.34)	17.96 – 19.64 (18.84 ± 0.35)	17.38 – 18.45 (17.95 ± 0.23)	15.72 – 17.20 (16.71 ± 0.34)
Total sugars (%)	33.27 – 34.86 (34.21 ± 0.34)	33.07 – 34.85 (34.12 ± 0.42)	34.20 – 35.20 (34.83 ± 0.22)	32.68 – 34.18 (33.27 ± 0.34)	32.85 – 34.85 (33.74 ± 0.46)
Ash (%)	2.06 – 2.54 (2.35 ± 0.10)	1.99 – 2.34 (2.23 ± 0.08)	1.85 – 2.15 (2.01 ± 0.07)	1.86 – 2.07 (1.96 ± 0.05)	1.64 – 2.01 (1.79 ± 0.08)
Free fat (%)	49.66 – 51.30 (50.29 ± 0.41)	58.31 – 59.64 (58.76 ± 0.30)	60.90 – 62.01 (61.42 ± 0.25)	65.65 – 67.28 (66.27 ± 0.37)	73.92 – 75.02 (74.53 ± 0.25)

Figures in the parentheses are the means with standard errors

Values are in ranges.

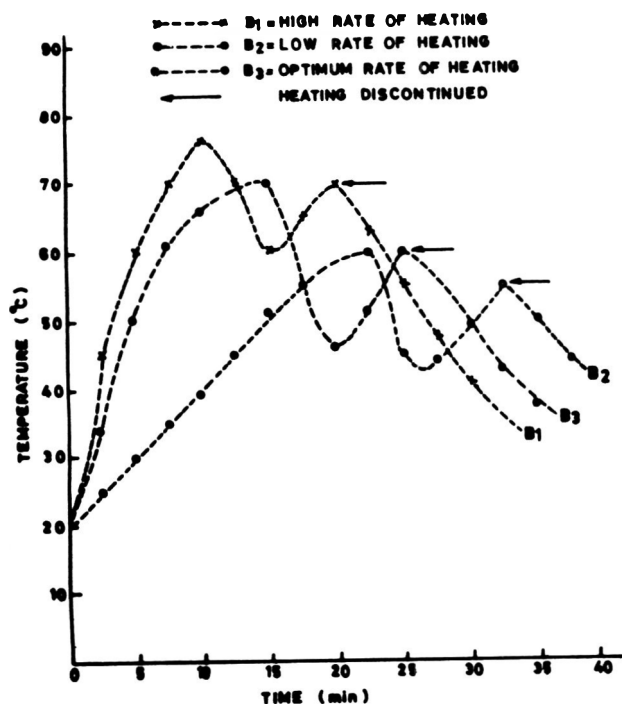


Fig 1. Time-temperature curves during preparation of buffalo milk sandesh

considerably improved the body and texture of the finished product. In subsequent trials, 50 per cent of the *chhana* was mixed with the entire sugar (30 per cent by weight of total *chhana*) and continued the scraping and stirring. When the mixture developed doughy consistency indicated the initial pat stage, the remaining lot of *chhana* was added with continuous stirring and scraping. These subsidiary processes were essential for attaining soft body and smooth texture in the finished product.

The initial and final temperatures for rapid heating of buffalo milk *chhana* with sugar were found to be 76° and 70°C in 10 and 20 min, respectively (Fig 1). Rapid heating caused hard, chewy and brittle body with an extremely coarse texture (Table 7). This indicated that rapid heating of buffalo milk *chhana* should be avoided (curve B<sub>1</sub>). The structural differences in casein micelles of cow and buffalo milk proteins

TABLE 7. EFFECT OF VARIOUS TIME-TEMPERATURE COMBINATIONS DURING PREPARATION, ON THE SENSORY QUALITY OF SANDESH.

Time-temp combination curve	Hedonic impression	Comments	
		Flavour	Body and texture
B <sub>1</sub>	Disliked very much	Characteristic cooked	Very hard, dry and chewy, extremely coarse
B <sub>2</sub>	Liked slightly	Undercooked/ raw <i>chhana</i> type	Very soft, smooth
B <sub>3</sub>	Liked very much	Slightly cooked	Soft, smooth

TABLE 8. MICROBIOLOGICAL QUALITY OF SANDESH MADE IN THE PLANT

Organisms	No. of samples +ve	Range (counts/g)	Mean ± SE (counts/g)
Total viable	10	2.5 × 10 <sup>2</sup> - 7.6 × 10 <sup>4</sup>	3.6 × 10 <sup>3</sup> ± 620.60
Coliforms	6	< 10 - 4.0 × 10 <sup>1</sup>	1.3 × 10 <sup>1</sup> ± 4.48
Spores	10	2.0 × 10 <sup>1</sup> - 1.2 × 10 <sup>2</sup>	4.6 × 10 <sup>1</sup> ± 8.97
Yeast and mould	9	< 10 <sup>1</sup> - 6.0 × 10 <sup>1</sup>	3.0 × 10 <sup>1</sup> ± 5.77
Staphylococci	10	2.0 × 10 <sup>1</sup> - 1.4 × 10 <sup>2</sup>	6.6 × 10 <sup>2</sup> ± 98.43

Average of 10 replicates  
SE = Standard error

might be the possible reason for it. At low rate of heating (curve B<sub>2</sub>) the highest temperature (60°C) and heating discontinued temperature (55°C) of *chhana* and sugar mixture were reached in about 22.5 and 32.5 min, respectively. Although *sandesh* samples obtained with this time temperature combination were undercooked with raw *chhana* type flavour (Table 7), the body and texture of the finished product improved considerably. However, on the whole none of the two curves (B<sub>1</sub> and B<sub>2</sub>) were suitable for *sandesh* production but indicated that the suitable time-temperature combination could exist in between the two curves (B<sub>1</sub> and B<sub>2</sub>). The time taken to reach the first pat formation stage was 15 min at 70°C (curve B<sub>3</sub>). The heating discontinued stage was reached in 25 min at 60°C (curve B<sub>3</sub>). Prepared

TABLE 9. QUALITY ASSESSMENT OF SANDESH

Quality attributes	Range	Mean ± SE
Moisture (%)	26.47 - 27.91	27.14 ± 0.35
Fat (%)	18.10 - 21.82	18.42 ± 0.27
Proteins (%)	18.59 - 18.83	18.71 ± 0.05
Total sugars (%)	33.12 - 33.92	33.83 ± 0.30
Ash (%)	1.70 - 2.09	1.90 ± 0.10
Free fat (as % of total fat)	60.62 - 64.31	61.81 ± 0.86
Titrate acidity (as % lactic acid)	0.67 - 0.85	0.74 ± 0.04
Water soluble acidity (as % lactic acid)	0.26 - 0.34	0.31 ± 0.02
Free fat acidity (as % oleic acid)	0.22 - 0.32	0.26 ± 0.02
p-DMAB reactivity absorbance	0.11 - 0.13	0.12 ± 0.005
pH at 20°C	5.94 - 5.99	5.97 ± 0.01
Density (g/cc at 20°C)	1.268 - 1.295	1.283 ± 0.006
Penetration value (0.1 mm at 30 ± 2°C)	142 - 160	148 ± 4.24
Yield (as % of <i>chhana</i> )	93.67 - 97.30	94.84 ± 0.83
Yield (as % of milk)	17.51 - 22.39	20.78 ± 1.11

SE = Standard errors  
Figures are average of 4 replicates.

*sandesh* samples with this time-temperature combination were liked very much by the judges (Table 7) and therefore, the same method was followed for subsequent *sandesh* preparations to assess the various microbiological and physico-chemical properties of *sandesh*.

**Microbiological quality of sandesh:** Results revealed that about 40 per cent of the *sandesh* samples were free from coliforms and about 10 per cent free from yeasts and moulds in  $10^1$  dilution (Table 8). It would seem difficult to produce *sandesh* free from bacterial sporeformers and staphylococci. The counts obtained in this study may be helpful to serve as guidelines while formulating the microbial standards for *sandesh* (Table 8).

**Quality assessment of sandesh:** The various quality attributes of *sandesh* samples are highlighted in Table 9. These values may serve as guidelines while formulating the chemical standards for *sandesh* made from buffalo milk. Yield of *sandesh* has also been calculated in terms of both, per cent of milk and *chhana* originally taken for *sandesh* production.

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## Study on Influence of Post-production Heat Treatment on Quality of Fresh Shrikhand

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**The post-production heat treatment of shrikhand had no adverse effect on its average composition, titratable acidity, pH, precipitated protein, syneresis, flavour, body and texture characteristics. There was slight improvement in the consistency of the product. It had a marked destructive effect on groups of microorganisms studied depending on severity of heat treatment and the type of microorganisms involved.**

Shrikhand is a popular indigenous sweetened fermented milk product. Being a summer dessert, it is in high demand during the hot summer months. Shrikhand has a fairly long shelf life of about 35 to 40 days at refrigeration temperature, but very limited shelf life of about 2 to 3 days at ambient temperature<sup>1</sup>. Hence, extending the shelf life of shrikhand under prevailing hot climatic conditions is of prime importance, as in India, refrigeration facility is not readily available in most of the households.

Use of post-production heat treatment has been found successful in extending the shelf life of cultured milk products like yoghurt and such treatment is being practised in the other countries<sup>2,4</sup>. Very limited work on post-production heat treatment for extending the shelf life of shrikhand has been reported in the literature<sup>5</sup>. Hence, a study was planned to evaluate the effects of post-production heat treatment on physical, chemical, microbial and sensory qualities of fresh shrikhand.

### Materials and Methods

**Preparation of shrikhand:** In the present study, eighteen batches of shrikhand, six for each replication were prepared. Heating fresh buffalo skim milk (9.33 per cent total solids) to 85°C for 15 min, curd and then chakka (shrikhand base) was prepared as per the method of Desai *et al.*<sup>6</sup> To adjust 6 per cent fat in shrikhand, pasteurized cream (72-74 per cent fat) was used. The chakka-cream mixture was made homogenous by passing it through a colloidal mill, to which the fine crystalline sugar and ground cardamom were added at the rate of 55 per cent (w/w of chakka-cream mixture) and 0.2 per cent (w/w of shrikhand), respectively. The whole mixture was blended for about 30 min in planetary mixture.

**Post-production heat treatment:** Shrikhand was subjected to heat treatment selected on the basis of preliminary trials,

viz., 55°C/30 min (T1), 60°C/20 min (T2), 65°C/10 min (T3), 70°C/5 min (T4) and 75°C/2 min (T5). Indirect heating in hot water was performed using jacketted kettle. Shrikhand was hot-packed in sanitized (200 p.p.m. available chlorine solution) polystyrene cups having snap-in-lid and cooled to 8-10°C. Unheated shrikhand served as control (C) and handled in similar way.

**Analyses:** Representative samples of shrikhand were analysed for moisture<sup>7</sup> (by Mojonnier tester) protein<sup>8</sup>, total sugar<sup>9</sup>, fat<sup>10</sup> (by modified Gerber method) and titratable acidity<sup>11</sup>. pH was determined by Systronic pH meter using 1 : 1 (shrikhand: distilled water) homogenate. Precipitated protein<sup>12</sup>, syneresis<sup>13</sup> and consistency<sup>14</sup> of shrikhand were measured using specified procedures. Sensory quality of shrikhand was evaluated by a panel of six judges for its appearance and colour (max. score, 3), body and texture (max. score, 5) and flavour (max. score, 10). Microbiological examination of shrikhand samples was performed by using appropriate dilutions and culture media for total viable counts<sup>15</sup>, acid producer counts<sup>16</sup>, proteolytic counts<sup>17</sup>, lipolytic counts<sup>18</sup>, yeast and mould counts<sup>16</sup> and coliform counts<sup>16</sup>.

**Statistical analysis:** The mean values of each attribute under study obtained from duplicate samples of three replications were analysed using factorial CRD<sup>19</sup>. The microbial counts were analysed after square-root transformation with additive one.

### Results and Discussion

**Composition and pH:** The data collated in Table 1 reveal that all the shrikhand samples studied had fat, protein, titratable acidity and pH within the range reported by various workers<sup>20,21</sup>. The moisture content of shrikhand was higher than the value specified under BIS specifications<sup>8</sup>, on

TABLE 1. EFFECT OF POST-PRODUCTION HEAT TREATMENT ON AVERAGE COMPOSITION, (PER CENT BY WEIGHT) TITRABLE ACIDITY AND pH OF FRESH SHRIKHAND

Treatment		Moisture	Fat	Proteins	Total sugars	Titratable acidity (%)	pH
Heating temp (°C)	Heating time (min)						
Unheated-Control (C)		49.74	5.93	7.38	35.98	1.22	4.34
55	30	49.37	5.96	7.45	36.05	1.24	4.34
60	20	49.21	6.07	7.49	36.05	1.24	4.33
65	10	49.09	6.00	7.58	36.07	1.25	4.34
70	5	48.90	6.03	7.59	36.08	1.26	4.30
75	2	48.82	6.03	7.63	36.10	1.26	4.30
SE ±		1.17	0.09	0.45	0.07	0.02	0.11
Significance		NS	NS	NS	NS	NS	NS
CV %		4.14	2.69	10.40	3.45	3.41	4.51

account of lower rate of sugar addition to chakka-cream mixture. However, the moisture values of the samples are well below the moisture range reported by Bhattacharya *et al.*<sup>27</sup>

From the compositional values depicted in the Table, it is evident that post-production heat treatment did not bring about any appreciable change in its composition, acidity and pH, except that it resulted in slight decrease in moisture content, with concomitant changes in other parameters studied. The above changes could be ascribed to evaporative losses of moisture during heating.

**Consistency, syneresis and protein-precipitation:** The tabulated values on consistency of shrikhand as influenced by post-production heat treatment (Table 2) reveal that heated samples were more firm as compared to unheated sample. This may be the combined effect of reduced moisture content and physico-chemical changes induced in shrikhand by heat

TABLE 2. INFLUENCE OF POST-PRODUCTION HEAT TREATMENT ON THE CONSISTENCY, SYNERESIS AND EXTENT OF PROTEIN-PRECIPIATION OF FRESH SHRIKHAND.

Treatment		Consistency (mm penetration at 10°C)	Syneresis* (g/70 g)	Precipitated protein (wet) (g/100 g)
Heating temp (°C)	Heating time (min)			
Unheated-Control (C)		30.70	5.80	2.07
55	30	24.40	5.70	2.00
60	20	22.63	5.90	1.98
65	10	22.27	5.77	2.02
70	5	22.13	5.73	2.07
75	2	21.83	5.93	2.23
SE		1.88	0.46	0.53
Significance		NS	NS	NS
CV %		13.62	13.85	45.20

\*Rate of syneresis measured at 30°C for 8 hr

treatment. However, this effect of post-production heat treatment on consistency was non-significant.

Post-production heat treatment of shrikhand had neither beneficial nor adverse influence on syneresis property. No published data on this aspect of study are available for comparison.

Post-production heat treatment of acidic product like shrikhand tends to bring about structural changes in protein and induce aggregation to impart grainy texture. To elucidate the effect of heating on protein stability, such protein aggregates were measured as wet protein precipitated in g per 100 g (Table 2). The observed differences among experimental samples, including control, were found to be non-significant, implying that post-production heat treatment did not promote much protein precipitation. Information on such aspects of study is not traceable in the literature, hence comparison of the results is not possible. However, Korolczuk *et al.*<sup>12</sup> reported 0.2, 0.4, and 1.5 per cent of the total milk protein precipitated during thermization (65°C for 30 min) at pH 3.5, 3.7 and 4.0 respectively in acid-curd cheeses such as quarg or tvaroh.

**Microbiology of shrikhand:** The microbial profile of shrikhand as influenced by the post-production heat treatment is presented in Table 3. All the samples of shrikhand studied had shown absence of coliforms. Considering count of different groups of microorganisms present in unheated shrikhand as 100 per cent, corresponding reduction of counts in heat treated shrikhand samples- T1, T2, T3, T4 and T5 were 61.92, 66.13, 74.13, 78.79 and 82.47 per cent for total viable

TABLE 3. MICROBIOLOGICAL PROFILE (COUNT PER GRAM)\* OF FRESH SHRIKHAND AS AFFECTED BY POST-PRODUCTION HEAT TREATMENT.

Treatment		Total viable	Acid producer	Proteolytic	Lipolytic	Yeast and mould
Heating temp (°C)	Heating time (min)					
Unheated-Control (C)		52.28	46.16	7.02	2.80	8.07
55	30	19.91 (61.92)	19.19 (58.42)	5.11 (27.21)	1.77 (64.29)	6.89 (14.62)
60	20	17.71 (66.13)	9.82 (78.73)	3.38 (51.85)	1.00 (100.00)	2.19 (72.86)
65	10	13.52 (74.13)	1.00 (100.00)	2.25 (67.95)	1.00 (100.00)	1.00 (100.00)
70	5	11.09 (78.79)	1.00 (100.00)	1.00 (100.00)	1.00 (100.00)	1.00 (100.00)
75	2	9.60 (82.47)	1.00 (100.00)	1.00 (100.00)	1.00 (100.00)	1.00 (100.00)
SE		0.71	1.09	0.58	0.80	0.56
CD (P < 0.05)		2.21	3.38	1.79	NS	1.72
CV %		6.01	14.58	30.65	96.97	28.87

\*Microbial values are square root transformed with additive value as one.

Figures in brackets indicate per cent reduction of microbial count.

**TABLE 4. EFFECT OF POST-PRODUCTION HEAT TREATMENT ON SENSORY CHARACTERISTICS OF FRESH SHRIKHAND.**

Treatment	Flavour*	Body and texture**	Total sensory score***
Heating temp (°C)	Heating time (min)		
Unheated-control (C)	9.08	4.09	16.17
55	30	9.14	4.32
60	20	9.17	4.41
65	10	9.27	4.38
70	5	9.23	4.41
75	2	9.22	4.42
SE ±	0.12	0.12	0.21
Signif.	NS	NS	Ns
CV %	2.38	4.68	2.19

\*Max. score is 10

\*\*Max. score is 5

\*\*\*Max. score is 18

counts, respectively. Acid producers were absent in T3, T4 and T5 while extent of their survival was 41.58 and 21.27 per cent in T1 and T2 respectively. Proteolytic count was reduced by 27.21, 51.85, 67.95, 100 and 100 per cent in T1, T2, T3, T4 and T5, respectively. Lipolytic count of shrikhand reduced by 100 per cent at and above 60°C, while yeast and mould counts were destroyed completely at and above 65°C.

The post-production heat treatment had marked destructive effect on the major groups of microorganisms commonly found in shrikhand. The effect was dependent on severity of heat treatment employed and types of microorganisms present in the product. Published information on microbial profile of shrikhand as affected by post-production heat treatment is not available for comparison. However, the results obtained in the present investigation are in line with those reported for heat treated yoghurt and cultured milk products.<sup>2,4,23,24</sup>

**Sensory quality:** The post-production heat treatment of shrikhand had no adverse effect on flavour, body and texture and total sensory score (Table 4). In fact, it resulted in slight improvement in its sensory attributes though not appreciably. Published data on sensory attributes of shrikhand as influenced by post-production heat treatment are not available for comparison. However, the values of sensory scores of experimental shrikhand observed in the present study are similar to those previously reported<sup>25</sup>.

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## Effect of Polyphosphate Chilling on the Quality of Quail Meat

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Spent Japanese quails (240) with equal number of males and females were dressed and chilled in 3 and 6% sodium tripolyphosphate (STPP) solution. The chilled carcasses were stored at  $4 \pm 1^\circ\text{C}$  for 6 days and at  $-18 \pm 1^\circ\text{C}$  for 60 days and evaluated microbiologically and for sensory qualities. Both the levels of STPP treatment retarded the growth of aerobic mesophilic counts and psychrophilic bacteria on the surface of quail carcasses during 60 days of frozen storage. STPP treatment has improved all the sensory characteristics. Chilling for 6 hr gave better scores for colour, appearance, and flavour. In general, fresh quail carcasses, females and breast muscle gave better sensory scores than their counterparts.

Polyphosphate chilling of chicken carcasses improves the quality of meat. Chicken carcasses treated with polyphosphate solution had reduced microbial counts compared to the untreated controls<sup>1,2</sup>. Improvement in the sensory characters of chicken carcasses treated with polyphosphate was reported<sup>3-5</sup>. As work on the influence of polyphosphates on quail carcasses is scanty, the present experiments were undertaken.

### Materials and Methods

Spent Japanese quails (*Coturnix coturnix Japonica*) aged over 25 weeks with 120 each males and females were dressed in four batches. Ready-to-cook carcasses were chilled in slush ice containing zero (control), 3 and 6 per cent sodium tripolyphosphate (STPP) solutions for 3 or 6 hr, packed in polyethylene bags and stored either under refrigeration ( $4 \pm 1^\circ\text{C}$ ) for 6 days or at frozen condition ( $-18 \pm 1^\circ\text{C}$ ) for 60 days. Both control and treated carcasses were evaluated for microbiological and sensory characters.

The swab technique of Malloman *et al.*<sup>6</sup> was used for determining the surface bacterial counts. The method was slightly modified by using sterile cotton swab along with a sterile metal template of 2 cm<sup>2</sup> size. The swabbing was done thoroughly on the breast region and then the swabs were suspended in 10 ml of sterile normal saline. After serial dilutions, suitable aliquots were used for inoculation. The inoculated plates were incubated at 37°C for 48 hr for total aerobic and at  $5 \pm 1^\circ\text{C}$  for 6 days for psychrophilic bacterial counts.

The carcasses were cooked at 1.1 kg pressure per cm<sup>2</sup> for 10 min. Uniform sized meat samples from both breast and thigh regions were cut and served to five member taste panel

for evaluating their colour and appearance, flavour, juiciness, tenderness and overall acceptability on a 9-point Hedonic scale. The data were statistically analysed as per Snedecor and Cochran<sup>7</sup>.

### Results and Discussion

**Total aerobic counts:** Both three and six per cent STPP treatments were effective in reducing the surface total aerobic counts of quail carcasses (Table 1). Similar reduction in bacterial counts of polyphosphate treated chicken carcasses were observed by Panda<sup>1</sup> and Susilkumar *et al.*<sup>8</sup>. On the other hand, Hoes *et al.*<sup>9</sup> and Sofes<sup>2</sup> observed that the addition of STPP to meat products did not delay the microbial growth. These differences in the results might be variation in the STPP concentration used and duration of storage followed by them. Duration of chilling did not have any influence on the surface aerobic counts of quail carcasses.

Frozen storage at  $-18 \pm 1^\circ\text{C}$  for 30 days had significantly reduced the total plate counts below the initial values but storage upto 60 days had significantly increased the total aerobic counts again (Table 1). Panda<sup>10</sup> Reddy and Varadarajulu<sup>11</sup> recorded significantly lower microbial counts in frozen stored carcasses than in fresh carcasses. In case of refrigerated storage at  $4 \pm 1^\circ\text{C}$ , the aerobic counts were significantly ( $P < 0.01$ ) increased steadily and continuously upto 3 and 164 days of storage.

**Psychrophilic counts:** As in the case of total aerobic counts, both 3 and 6 per cent STPP treatment has significantly restricted the growth of psychrophilic bacteria during storage. The antibacterial property of STPP might be due to its ability to reduce the water activity in the meat.

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TABLE 1. MEAN TOTAL AEROBIC AND PSYCHROPHILIC COUNTS ( $\times 10^7/cm^2$ ) OF QUAIL CARCASSES AS INFLUENCED BY STPP LEVELS, DURATION OF CHILLING AND STORAGE PERIODS.

STPP level (%)	Chilling time (hr)	Storage periods in days										Mean for STPP	
		0		3		6		30		60		Meso-philic	Psychro-philic
		Meso-philic	Psychro-philic	Meso-philic	Psychro-philic	Meso-philic	Psychro-philic	Meso-philic	Psychro-philic	Meso-philic	Psychro-philic		
0	3	3.63	1.16	15.86	4.68	26.92	9.77	1.11	1.05	12.88	10.47	6.92 <sup>b</sup>	3.98 <sup>b</sup>
	6	2.75	1.55	12.88	5.89	29.51	17.78	0.81	0.91	14.13	10.75		
3	3	2.40	0.85	13.49	2.88	19.06	7.08	0.87	0.74	9.12	7.94	5.25 <sup>a</sup>	2.88 <sup>a</sup>
	6	2.46	1.32	9.55		25.70		0.63		10.23			
6	3	1.91		13.49		18.20		0.58		9.12		4.90 <sup>a</sup>	1.95 <sup>a</sup>
	6	3.16		8.51		21.88		0.65		10.47			
Means for storage periods													
Mesophiles		2.57 <sup>b</sup>		12.02 <sup>c</sup>		22.39 <sup>d</sup>		0.76 <sup>a</sup>		10.72 <sup>c</sup>			
Psychrophiles		1.12 <sup>b</sup>		4.37 <sup>c</sup>		10.00 <sup>c</sup>		0.79 <sup>a</sup>		9.12 <sup>d</sup>			
Means for chilling trial													
Mesophiles		3 hr = 5.75 <sup>d</sup>		6 hr = 5.50 <sup>d</sup>									
Psychrophiles		3 hr = 2.82 <sup>c</sup>		6 hr = 3.72 <sup>b</sup>									

Mean values within each factor bearing at least one superscript do not differ significantly ( $P < 0.05$ ).

Chilling for 6 hr had resulted in higher counts than chilling for 3 hr regardless of STPP levels and storage periods (Table 1).

The multiplication of psychrophiles was significantly controlled by frozen storage at  $-18 \pm 1^\circ C$  for 30 days but further storage upto 60 days resulted in higher counts. Probably lower temperature may be needed for effective prolonged storage of quail carcasses. Similar conclusions were made by Panda<sup>10</sup>, Reddy and Varadarajulu<sup>11</sup> in broiler carcasses.

Refrigerated storage at  $4 \pm 1^\circ C$  was not effective in controlling the growth of psychrophilic organisms. The

psychrophilic load of carcasses was progressively and significantly increased during the course of refrigeration of 6 day period (Table 1). Similar results were recorded by Reddy and Varadarajulu<sup>11</sup> and Panda<sup>1</sup> in broiler carcasses.

Despite an increase in the microbial counts during storage, the counts did not reach the levels of spoilage and rejection by the rate panel members.

*Sensory characteristics:* All the sensory characteristics evaluated namely, colour and appearance, flavour, juiciness, tenderness and overall acceptance of spent Japanese quail meat were significantly improved by the STPP treatment (Table 2). Similar improvements in the above characteristics

TABLE 2. MEAN VALUES OF SENSORY SCORES OF QUAIL CARCASSES AS INFLUENCED BY THE INTERACTION OF STPP LEVELS, DURATION OF CHILLING, STORAGE PERIODS, SEX AND SOURCE OF MUSCLE

Parameter		Colour and appearance	Flavour	Juiciness	Tenderness	Overall acceptance
STPP level (%)	0	5.28 <sup>a</sup>	5.31 <sup>a</sup>	5.48 <sup>a</sup>	5.61 <sup>a</sup>	5.49 <sup>a</sup>
	3	5.63 <sup>b</sup>	5.62 <sup>b</sup>	5.78 <sup>b</sup>	5.88 <sup>a</sup>	5.81 <sup>b</sup>
	6	5.63 <sup>b</sup>	5.66 <sup>b</sup>	5.80 <sup>b</sup>	5.92 <sup>b</sup>	5.79 <sup>b</sup>
Chilling period (h)	3	5.42 <sup>a</sup>	5.46 <sup>a</sup>	5.65 <sup>a</sup>	5.78 <sup>a</sup>	5.65 <sup>a</sup>
	6	5.60 <sup>b</sup>	5.60 <sup>b</sup>	5.72 <sup>a</sup>	5.82 <sup>a</sup>	5.74 <sup>a</sup>
	0	5.97 <sup>c</sup>	6.07 <sup>b</sup>	6.13 <sup>c</sup>	6.16 <sup>c</sup>	6.09 <sup>d</sup>
Storage period (d)	3h	5.80 <sup>a</sup>	5.77 <sup>a</sup>	5.92 <sup>a</sup>	5.98 <sup>a</sup>	5.95 <sup>c</sup>
	6h	5.39 <sup>b</sup>	5.30 <sup>b</sup>	5.55 <sup>b</sup>	5.69 <sup>b</sup>	5.49 <sup>b</sup>
	30	5.44 <sup>a</sup>	5.56 <sup>a</sup>	5.56 <sup>b</sup>	5.78 <sup>b</sup>	5.72 <sup>c</sup>
	60	4.95 <sup>a</sup>	4.99 <sup>a</sup>	5.19 <sup>a</sup>	5.42 <sup>a</sup>	5.72 <sup>c</sup>
Sex	Male	5.36 <sup>a</sup>	5.42 <sup>a</sup>	5.59 <sup>a</sup>	5.70 <sup>a</sup>	5.61 <sup>a</sup>
	Female	5.64 <sup>b</sup>	5.64 <sup>b</sup>	5.77 <sup>b</sup>	5.91 <sup>b</sup>	5.80 <sup>a</sup>
Muscle source	Breast	5.68 <sup>a</sup>	5.63 <sup>a</sup>	5.74 <sup>a</sup>	5.88 <sup>a</sup>	5.76 <sup>b</sup>
	Thigh	5.32 <sup>b</sup>	5.44 <sup>b</sup>	5.62 <sup>b</sup>	5.73 <sup>b</sup>	5.65 <sup>b</sup>
S.E. Range		(0.04-0.07)	(0.04-0.06)	(0.00-0.06)	(0.01-0.06)	(0.03-0.05)

Mean values for each factor bearing same superscript within a row do not differ significantly ( $P < 0.01$ ).

was observed in STPP - treated poultry meats<sup>3-5</sup>. Contrary to these observations Dawson and Simson<sup>12</sup> observed no significant improvement in juiciness, tenderness, flavour and general acceptability scores of chicken parts marinated in polyphosphate solution.

**Effect of chilling time:** Chilling for 6 hr in STPP solution has given better scores of colours and appearance and flavour than for 3 hr period. But no significant variations were noticed between the two chilling periods of quail carcasses with respect to juiciness, tenderness and overall acceptance scores (Table 2). Whereas Pandey *et al*<sup>13</sup> observed preference of 3 hr chilled quail carcasses over the others.

**Effect of storage period:** Fresh quail meat recorded the highest panel scores for all the organoleptic traits and preferred by the taste panelists over the stored meats except for a higher juiciness scores in 30 days frozen stored quail carcass. This was especially, true in case of untreated meat than the STPP treated one (Table 2). Similar inferences of lower panel scores for stored meats than the fresh meat were drawn by Landes<sup>3</sup>. On contrary, Koushik<sup>14</sup> observed significant improvement in tenderness, juiciness and flavour scores of chicken meat after frozen storage for 10 days at  $-10^{\circ}\text{C}$  compared to the control.

**Effect of source of meat:** The breast meat from spent Japanese quail carcasses had superior scores on all the sensory characteristics evaluated irrespective of other treatments in the present study. This finding concurs with the results of Baker and Darfler<sup>15</sup> in chicken carcasses.

Most of the sensory traits were in favour of the meat samples from female quail carcasses rather than from male quails irrespective of other treatments. Contrary to this, Ristic<sup>16</sup> observed better sensory quality in breast muscles of male broilers than females. Greater body size of female quail carcasses than the males unlike in chicken might be the reason for these variations.

Highly significant positive correlation between various sensory traits of breast and thigh meats of spent Japanese quail observed in this study were in agreement with the findings of Baker and Darfler<sup>15</sup>. Among the interrelationship between various sensory traits, the overall acceptance score was more positively correlated with tenderness and flavour scores suggesting that these two traits influence the overall acceptance of the meat by taste panelists to a greater extent than other traits.

Based on the overall results of this study, it may be concluded that chilling of spent Japanese quail carcasses of either sex for 3 hr in slush ice containing 3 per cent STPP is advisable for improving the micro-biological and sensory characteristics of quail meat.

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## Microbial Profile of Tropical Prawn *Metapenaeus dobsoni* During Frozen Storage

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Microbiological changes of Indian prawn, *Metapenaeus dobsoni* frozen as whole (W) headless-shellon (HL) and peeled and deveined (PD) were investigated. Exposure to freezing temperatures caused mortality of major bacteria present in prawn, 83.67%, 79.48% and 52.90% of cells being destroyed in W, HL and PD types. Frozen storage for a period one month caused less destruction (13.58%, 16.90% and 9.78%). After two months, death was very slow and gradual. The results point to a difference in the survival rate of bacteria present on PD prawn. Qualitatively, various genera were affected differently by freezing and frozen storage. *Vibrio* spp. were the most sensitive and *Micrococcus* spp. the most resistant.

Major part of the prawn landings in India are frozen for export. When prawn is frozen to  $-40^{\circ}\text{C}$ , the lethal action of temperature causes elimination of bacteria which are very susceptible to low temperature. Further reduction may occur during prolonged frozen storage. The survivors of the freezing process will be the most resistant groups of initial bacterial load. A succession of bacterial genera is thus found to occur in such cases<sup>1</sup>.

The bacteriological profiles of the shrimp from temperate waters have been reported<sup>2-5</sup>. Numerous publications are also available on the bacteriology of tropical prawn with emphasis on bacteria of public health significance<sup>6-12</sup>.

The present work summarizes the observations on the microbiology of frozen prawn, *Metapenaeus dobsoni*.

### Materials and Methods

The tropical prawn, *Metapenaeus dobsoni* was used in the study. The raw material was collected from the fishing vessels operating off Kochi on its arrival at the landing centre and consisted of fresh clean prawn in the count range of 100-200/kg. The maximum time that could have elapsed between the catch and collection of sample was 4-6 hr.

The material was frozen as whole (W) headless-shellon (HL) and peeled and deveined (PD). Generally, the practices followed for handling and packaging were outlined in IS: 4303<sup>13</sup>.

Freezing was carried out in horizontal plate freezer at  $-40^{\circ}\text{C}$ . The material was packed in one kg lots in duplex cartons lined inside with 100 gauge polythene sheets. Freezing was completed in 2-3 hr. After glazing with chilled potable water, blocks were transferred to deep-freezer maintained at  $-18 \pm 2^{\circ}\text{C}$  and stored upto one year. Samples were drawn at

regular intervals and subjected to chemical, sensory and bacteriological analysis.

About 100 g of the material was cooked in 2 per cent brine for 15 min and assessed by a 6 member taste panel using scalar system of scoring. The scoring rate was as 5-good, 4-good to fair, 3-fair, 2-fair to poor, 1-poor and 0-off.

The total plate counts (TPC) and the counts of faecal indicator organisms were determined<sup>14</sup>. The medium used for TPC was tryptone glucose extract agar. Appropriate dilutions were surface plated and plates incubated at  $5^{\circ}\text{C}$ , room temperature ( $29 \pm 1^{\circ}\text{C}$ ) and  $37^{\circ}$  for 21 days and 2 days. The plates incubated at  $5^{\circ}\text{C}$ , after completion of the incubation at  $5^{\circ}\text{C}$  were kept for an additional 24 hr at RT and counts (5/30) taken. The bacterial count per g of muscle was calculated.

About 30 to 50 colonies were picked randomly from plates of tryptone glucose extract agar and identified up to generic level<sup>15-17</sup>.

The data were analysed by simple regression analysis<sup>18</sup> taking months of storage as X and logarithm of bacterial count as Y. In the case of *Pseudomonas*, *Moraxella*, *Acinetobacter* and *Micrococcus* groups, the percentage of each to the total count was taken as Y. The significance of the regression co-efficient 'b' of months of storage on bacterial count was tested using 't' test, where 'sb' is the standard error of 'b'.

$$t = \frac{b}{sb}$$

The degrees of freedom of 't' is (n-2) where 'n' is the number of months of storage.

TABLE 1. CHANGES IN THE TOTAL PLATE COUNT AT 5°C AND RT OF PRAWN, *M. DOBSONI* DURING FREEZING AND FROZEN STORAGE

Period of frozen storage (months)	Whole				Bacterial count on headless shellor				Peeled and deveined			
	Temp. of incubation		Ratio of		Temp. of incubation		Ratio of		Temp. of incubation		Ratio of	
	5°C	RT	5/30	5/5/30	5°C	RT	5/30	5/5/30	5°C	RT	5/30	5/5/30
Initial	1.03×10 <sup>4</sup>	2.45×10 <sup>3</sup>	2.35×10 <sup>3</sup>	0.04	9.40×10 <sup>3</sup>	1.90×10 <sup>3</sup>	1.60×10 <sup>3</sup>	0.06	2.60×10 <sup>3</sup>	2.15×10 <sup>3</sup>	2.00×10 <sup>3</sup>	0.01
0	2.70×10 <sup>3</sup>	4.00×10 <sup>4</sup>	3.40×10 <sup>4</sup>	0.08	8.40×10 <sup>2</sup>	3.90×10 <sup>4</sup>	2.63×10 <sup>4</sup>	0.03	9.20×10 <sup>3</sup>	1.03×10 <sup>5</sup>	6.40×10 <sup>4</sup>	0.14
1	9.60×10 <sup>2</sup>	6.50×10 <sup>3</sup>	4.60×10 <sup>3</sup>	0.20	5.70×10 <sup>2</sup>	6.90×10 <sup>3</sup>	6.00×10 <sup>3</sup>	0.09	2.00×10 <sup>3</sup>	8.20×10 <sup>4</sup>	4.90×10 <sup>4</sup>	0.40
2	8.00×10 <sup>2</sup>	1.35×10 <sup>3</sup>	1.02×10 <sup>3</sup>	0.78	1.60×10 <sup>3</sup>	3.90×10 <sup>3</sup>	3.80×10 <sup>2</sup>	0.42	3.20×10 <sup>3</sup>	6.00×10 <sup>4</sup>	5.90×10 <sup>4</sup>	0.54
6	4.60×10 <sup>2</sup>	6.50×10 <sup>2</sup>	6.30×10 <sup>2</sup>	0.73	8.70×10 <sup>2</sup>	1.01×10 <sup>3</sup>	9.79×10 <sup>2</sup>	0.90	8.80×10 <sup>3</sup>	1.10×10 <sup>4</sup>	1.10×10 <sup>4</sup>	0.88
10	3.00×10 <sup>2</sup>	3.80×10 <sup>2</sup>	3.10×10 <sup>2</sup>	0.98	4.80×10 <sup>2</sup>	5.10×10 <sup>2</sup>	4.90×10 <sup>2</sup>	0.98	7.70×10 <sup>3</sup>	7.80×10 <sup>3</sup>	7.60×10 <sup>3</sup>	1.01
12	2.00×10 <sup>2</sup>	2.80×10 <sup>2</sup>	2.70×10 <sup>2</sup>	0.74	3.20×10 <sup>2</sup>	3.70×10 <sup>2</sup>	3.60×10 <sup>2</sup>	0.89	7.40×10 <sup>3</sup>	7.80×10 <sup>3</sup>	7.90×10 <sup>3</sup>	0.94

\*Values in the parentheses represent the % of the surviving microorganisms.

## Results and Discussion

Changes in the TPC of prawn *M. dobsoni* are presented in Table 1. The initial value noted in the Table represents the bacterial count of the raw material which has been subjected to handling and hence not identical for the three types, W, HL and PD. On freezing, the TPC at 5°C and RT of all the three types decreased. This was accompanied by a further decrease on storage at -18°C. The count at 5°C was lower compared to that at RT. However, this difference in TPC at the three incubation temperatures disappeared on storage after six months.

When the plates incubated at 5°C were reincubated at RT for one day, an increase in TPC was observed in the beginning of storage period. On prolonged storage, the count at 5°C increased. The corresponding increase in 5/30 count was low with the result the two counts became comparable. In this regard, the behaviour of the three varieties was very much identical.

The percentage of surviving population was determined for each variety (Table 1). Freezing caused major mortality in bacteria, 86.78, 79.48 and 53.01 per cent of cells being killed in the process of freezing of W, HL and PD respectively. After frozen storage for one month at -18°C, the percentage reduction in bacterial count rose to 97.24, 96.37 and 61.87 per cent respectively. Thereafter, the decrease was negligible for W and HL types although PD continued to register a fall in count. The results clearly point to a difference in survival rate in W, HL and PD prawns.

The sensory evaluation is given in Table 2. The sensory qualities of the samples were acceptable till 4 to 6 months storage. But the major changes in the bacterial flora occurred in the early period of storage.

The changes in the various bacterial genera are presented in Table 3. The initial flora of the raw material W and HL differed from the PD having a predominance of Gram

TABLE 2. CHANGES IN THE SENSORY CHARACTERISTICS OF PRAWN *M. DOBSONI* AFTER FREEZING AND FROZEN STORAGE

Period of frozen storage (months)	Whole prawn	Headless prawn	Peeled and deveined prawn
0	Excellent (9)	Excellent (9)	Excellent (9)
1	Very good (8)	Very good (8)	Very good (9)
2	Good (7)	Good (7)	Good (7)
4	Good (7)	Good (7)	Good (7)
6	Good slightly tough (5)	Good slightly tough (5)	Good (6)
10	Poor, tough (4)	Poor (4)	Poor (4)
12	Poor, very tough (3)	Poor, very tough (3)	Poor, very tough (3)

Figures in the parenthesis indicate the score numbers.

positives as a result of greater handling during dressing. The faecal indicator organisms were, not detected in this study.

After freezing, there occurred a fall in the number of Gram negatives especially *Pseudomonas*, *Vibrio* and *Flavobacterium* spp. The percentage of *Micrococcus* spp., on the other hand, showed a gradual rise. The same trend was noted during the storage period also.

Regression analysis of the data indicated that the *Pseudomonas* species decreased during freezing and storage and the *Micrococcus* group increased (Table 4). *Moraxella* and *Acinetobacter* species were not much affected by freezing. The *Vibrio* species were completely eliminated from the system showing its high sensitivity to freezing. Hence, this was not included in statistical analysis.

The immediate effect of freezing was a drastic reduction in the total microbial population in W, HL and PD varieties of frozen prawn. The decline however, was much less during frozen storage. Most of the bacterial death occurring on storage could be accounted in the early storage period upto 2 months. Hence, it could be concluded that during freezing



TABLE 3. \*QUALITATIVE CHANGES IN THE MICROBIAL FLORA OF WHOLE, HEADLESS AND PEELED PRAWN DURING FROZEN STORAGE

Storage period (months)	Type	Pseudo- monas	Acineto- bacter	Mora- xella	Vibrio	Fl.cyto- phaga	Micro- coccus	Bacillus	Arthro- bacter	Others	Total isolates
BF	W	20 <sup>+</sup>	16	17	14	6	20	1	2	4	75
	HL	20	17	14	15	3	24	2	3	2	75
	PD	8	11	10	10	4	46	3	4	4	72
0	W	14	18	20	5	1	37	1	4	0	77
	HL	15	21	22	5	1	35	1	1	1	74
	PD	2	17	14	5	2	50	2	8	0	70
1	W	13	13	19	0	0	52	1	1	1	84
	HL	13	17	21	0	0	50	0	1	0	81
	PD	8	20	16	0	2	50	0	2	2	84
2	W	12	15	20	0	0	53	0	0	0	73
	HL	10	15	24	0	0	51	0	0	0	77
	PD	2	22	20	0	0	52	1	3	0	79
6	W	9	10	18	0	0	63	0	0	0	92
	HL	6	16	17	0	0	60	1	0	0	94
	PD	2	13	15	0	0	64	0	0	0	63
10	W	4	14	15	0	0	67	0	0	0	91
	HL	0	15	17	0	0	68	0	0	0	88
	PD	3	10	12	0	0	71	0	2	2	86
12	W	0	15	16	0	0	69	0	0	0	93
	HL	0	15	14	0	0	71	0	0	0	92
	PD	0	12	8	0	0	76	0	0	4	73

\* Based on the bacterial count at RT

+ Expressed as %

BF = Before freezing.

TABLE 4. REGRESSION ANALYSIS OF BACTERIAL COUNT

Log bacterial count		xy	x <sup>2</sup>	y <sup>2</sup>	n	b	sy-x	sb	t	df	P
Log bacterial count	W	-32.2485	159.3934	9.3934	9	-0.2023	0.9654	0.0765	2.632	7	0.05
	HL	-28.2948	159.3934	6.4313	9	-0.1775	0.4486	0.0355	4.996	7	0.01
	PD	-33.7446	159.3934	10.2664	9	-0.2117	0.6677	0.0529	4.003	7	0.01
<i>Pseudomonas</i>	W	-126.5423	159.3934	133.5556	9	-0.7939	2.1743	0.1722	4.610	7	0.01
	HL	-255.7283	159.3934	466.2222	9	-1.6044	2.8268	0.2239	7.166	7	0.001
	PD	-77.1013	159.3934	82.8889	9	-0.4837	2.5521	0.2021	2.393	7	0.05
<i>Moraxella</i>	W	30.4573	159.3934	103.5566	9	0.2941	3.7366	0.2960	0.994	7	NS
	HL	-67.8473	159.3934	174.2222	9	0.4257	4.5567	0.3609	1.179	7	NS
	PD	168.6103	159.3934	175.5556	9	-0.4304	4.5985	0.3642	1.182	7	NS
<i>Acinetobacter</i>	W	-70.7967	159.3934	68.8889	9	-0.4442	2.3128	0.1832	2.425	7	0.05
	HL	-14.9150	159.3934	46.0000	9	-0.0936	2.5243	0.1999	0.468	7	NS
	PD	-110.21110	159.3934	206.0000	9	-0.6918	4.3047	0.3410	2.029	7	NS
<i>Micrococcus</i>	W	362.5420	159.3934	966.0000	9	2.2745	4.4943	0.3560	6.389	7	0.001
	HL	491.9143	159.3934	1900.8889	9	3.0862	7.3946	0.5857	5.269	7	0.01
	PD	421.6437	159.3934	1202.8889	9	2.6453	3.5358	0.2801	9.444	7	0.001

b - regression co-efficient; n - sample size; sb - standard error of b; df - degree of freedom; p - level of significance; NS - not significant at 5% level

storage of prawn, the bacterial inactivation during freezing was more significant than that during storage of fish as reported earlier<sup>19</sup>.

Qualitatively as well as quantitatively bacterial inactivation profiles during freezing and frozen storage of whole and headless prawn were almost comparable. Contrary to this, the bacterial flora of PD prawn showed higher survival capacity as indicated by enhanced TPC counts. Fieger *et al*<sup>3</sup>

noted a greater reduction in bacteria in peeled than unpeeled shrimp. A higher TPC in frozen PD prawn was also noted previously for tropical prawn by Pillai *et al*<sup>10</sup>. They attributed this behaviour to the large meat surface area exposed to bacterial attack. The present study indicates that a shift in the bacterial flora pattern to more resistant types as a result of excessive human handling may be the decisive factor. The greater protection, afforded to bacteria of the

meat<sup>20</sup>, than those on shell surface may be an added advantage.

While increase in the ratio of 5 to 5/30 count is attributed to the presence of microcolonies<sup>21</sup> or to the presence of mesophiles which failed to grow at 5°C, the rise in count at 5°C could be attributed to the gradual physiological adaptation of the mesophiles to low temperature<sup>19</sup>. This clearly shows that the freeze resistant fraction is psychotropic also.

The major event in the qualitative studies was the gradual replacement of the Gram negative flora by the more resistant Gram positive types. Irrespective of their initial number in raw material, the Gram positive cocci rose to predominance. The greater resistance to low temperature of the Gram positive cocci has been reported earlier<sup>19,22</sup>. The present study confirms the finding.

The TPC values reported for frozen prawn is comparable to that reported for temperate shrimp<sup>4,6</sup>. There is very little information on the bacterial flora of frozen prawn whether it is of tropical or temperate origin<sup>6,11,12</sup>. The study of Kawabata *et al*<sup>11</sup> of the bacterial flora of tropical shrimp imported to Western countries bear much similarities to the PD prawn in this study.

The bacterial flora of frozen food has been suggested to comprise the most cold tolerant organisms<sup>19</sup>. This means that continued exposure of prawn meat to freezing temperature results in the concentration of selected groups depending on the response of the individual species to the lethal action of cold. It is, therefore, likely that the bacterial flora of frozen stored prawn from different areas may represent more or less similar flora pattern that are cold tolerant.

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## Potential of Some Rhizomes of Zingiberaceae Family as Grain Protectants Against Storage Insect Pests

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The comparative efficacy of indigenous plant species viz. *Alpinia galanga* Willd., *Curcuma amada* Roxb., *Curcuma longa* L., *Curcuma zedoaria* Rose. and *Zingiber officinale* Rose. all belonging to the family Zingiberaceae was evaluated against *Sitophilus oryzae* (L.) and *Corcyra cephalonica* (Staint.) infesting wheat *Triticum sativum* Lam. All the powdered rhizomes tested were found significantly superior to control in protecting the grain at the concentrations of 1 and 3% W/W in bringing about higher mortality of *S. oryzae* adults. The insect mortality was cent per cent in the treatments even at 1% level after 45 days. Besides, the treatments *C. amada* and *C. longa* were highly significantly effective in inhibiting the emergence of F<sub>1</sub> generation of the weevil followed by *A. galanga*, *C. zedoaria* and *Z. officinale*. While against *C. cephalonica* larvae, the treatment *A. galanga* was the best grain protectant even at 1% level after a period of 15 and 30 days. However, at 3% level there was no significant difference between *A. galanga*, *C. longa* and *Z. officinale* and they were superior in causing high larval mortality followed by *C. zedoaria* and *C. amada*.

The use of conventional insecticides for the control of insect pests at farm or storage level has created potential hazards due to toxic and persistent residues in food. Hence, all over the world there has been a revival of interest in natural products for their use in the integrated insect pest control programmes. Among the various avenues explored, insecticides of plant origin being safe to mammals, easily biodegradable and practically innocuous to non-target species indicate better solution. Besides offering great scope for environmental safety, natural products may give lead to the development of safer synthetic analogues<sup>1</sup>. Apart from the known botanical origin insecticides such as Pyrethrins, Rotenone and Nicotine, workers have been exploring the possibility of using various other plant products for the effective control of insect pests<sup>2,6</sup>. In the present investigation, five plants belonging to the family Zingiberaceae known for their bioactivity and medicinal properties<sup>7,8</sup> were evaluated for their effectiveness as protectants of wheat against the infestation of *Sitophilus oryzae* (L.) and *Corcyra cephalonica* (Staint.).

### Materials and Methods

The plant materials viz. rhizomes of the greater galangal *Alpinia galanga* Willd., turmeric *Curcuma longa* L., mango ginger *Curcuma amada* Roxb., round zedoary *Curcuma zedoaria* Rose. and ginger *Zingiber officinale* Rose. were

procured from the local Ayurvedic drug store, dried in a hot air drier at 40°C and ground in Raymond's Hammer mill and the powder was sieved to 200 mesh for uniform adherence to grain surface.

One week old rice weevil (*Sitophilus oryzae*) adults and third instar larvae of rice moth (*Corcyra cephalonica*) were obtained from the laboratory cultures maintained at 27 ± 2°C temperature and 70-75 per cent relative humidity. The experiments were conducted at the above mentioned temperature and relative humidity.

Wheat (*Triticum sativum* Lam.) samples were disinfested by keeping at -18°C for two weeks. The disinfested samples were brought to equilibrium moisture of 12-13 per cent by conditioning at 70 per cent relative humidity prior to use for experimental purpose.

Samples of 50 g uninfested wheat were taken in glass jars (150 ml) and mixed with powdered plant materials at the concentrations of 1 and 3 per cent. Four replicates were maintained for each treatment including control.

In an experiment with *S. oryzae*, 25 adult insects were introduced into each replicate. The efficacy of the plant powders was judged by observing the percentage mortality of adult insects. The mortality was recorded after 15, 30 and 45 days, and insects from all the treatments including control were discarded. The emergence of F<sub>1</sub> generation of the weevil was scored at every alternate day till there was no

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further emergence. The percentage data were subjected to angular transformation before carrying out statistical analysis which was done by ANOVA employing two-tailed test.

In another experiment, 25 third instar larvae of *C. cephalonica* were introduced in treated and control samples of wheat. Data on the mortality of larvae were taken at intervals of 15 and 30 days and were subjected to angular transformation before carrying out statistical analysis.

### Results and Discussion

The data presented in Table 1 indicate that all the rhizome materials tested against *S. oryzae* were found to be significantly superior to control in bringing about insect mortality even at 1 per cent level after 15 days. The treatment *C. amada* was superior and showed cent per cent insect mortality at 3 per cent concentration after 30 days, followed by the rhizomes of *C. longa*, *A. galanga*, *C. zedoaria* and *Z. officinale*. After 45 days, all treatments at both the concentrations showed cent per cent weevil mortality in wheat and were found superior to the control. Chander and Ahmed<sup>5</sup> however, have reported that *C. zedoaria* powder even at 5 per cent level was not very effective against *S. oryzae* adults infesting sorghum. The better results in the present investigations could be due to the morphological differences between the wheat and sorghum grains. Wheat having a brush or bristles at the distal end and a deep crease in the middle would retain the rhizome powders well, resulting better efficacy to test insects compared to sorghum with round and smoother surface.

The emergence of  $F_1$  generation of *S. oryzae* is presented in Table 2. All the treatments were significantly superior to control in reducing the emergence of  $F_1$  generation. However, *C. amada* and *C. longa* were highly superior to other treatments showing an average emergence of 1 and 3  $F_1$  adults. The low emergence of the  $F_1$  generation in treated wheat could be attributed to the early adult mortality in those treatments.

TABLE 1. EFFICACY OF VARIOUS RHIZOMES ON THE MORTALITY OF *SITOPHILUS ORYZAE* L. ADULTS IN WHEAT AT DIFFERENT CONCENTRATIONS.

Plant materials	% mortality after indicated days and concen.					
	15 days		30 days		45 days	
	1%	3%	1%	3%	1%	3%
<i>Curcuma amada</i>	34.40	66.94	41.55	90.00	90.00	90.00
<i>Curcuma longa</i>	39.19	53.27	41.53	66.77	90.00	90.00
<i>Alpinia galanga</i>	29.23	43.84	41.55	58.17	90.00	90.00
<i>Curcuma zedoaria</i>	34.40	39.19	48.46	58.08	90.00	90.00
<i>Zingiber officinale</i>	29.00	39.19	41.55	55.57	90.00	90.00
Control	16.17	16.17	29.17	29.17	58.51	58.51
S.Em	+2.23	+2.61	+1.42	+1.75	+1.60	+1.60
CD at 5%	4.68	5.48	2.98	3.68	3.36	3.36

TABLE 2. EFFECT OF VARIOUS RHIZOME POWDERS ON THE EMERGENCE OF  $F_1$  GENERATION OF *SITOPHILUS ORYZAE*, L.

Plant material	Emergence of $F_1$ generation
<i>Curcuma amada</i>	1.00
<i>Curcuma longa</i>	3.00
<i>Alpinia galanga</i>	39.00
<i>Curcuma zedoaria</i>	34.00
<i>Zingiber officinale</i>	55.00
Control	142.00
S.Em	+2.52
C D at 5%	5.29

The per cent mortality of *C. cephalonica* larvae in wheat treated with various rhizome powders is furnished in Table 3. After 15 days of observation, all the treatments were found significantly superior to control at both the concentrations tested. However, at 3 per cent level the treatments *A. galanga* and *C. longa* were superior in showing larval mortality compared to other treatments. Observation after 30 days showed that at 3 per cent concentration, there was no significant difference between the treatments *A. galanga*, *C. longa* and *Z. officinale* and they were superior in causing high larval mortality followed by *C. zedoaria* and *C. amada*. As the larvae are the active feeding stage, the mortality could be attributed to the injection of plant material causing stomach toxicity. Chander and Ahmed<sup>6</sup> have also reported that plant powders such as powdered rhizomes of *Acorus calamus*, powdered leaves of *Clerodendron inerme*, *Tylophora asthmatica*, *Justicia betonica* and *Cestrum nocturnum* at 2 and 5 per cent levels were highly effective against the development of *C. cephalonica* larvae in treated wheat. Rice grains coated with oil of *C. longa* at 100 p.p.m. level has also been reported<sup>9</sup> to be a strong repellent against red flour beetle (*Tribolium castaneum*).

The rhizomes of the plants selected in the present investigation have medicinal properties<sup>7,8</sup> and some are

TABLE 3. PERCENTAGE MORTALITY OF *CORCYRA CEPHALONICA*, STAINT LARVAE IN WHEAT TREATED WITH VARIOUS RHIZOMES AT DIFFERENT CONCENTRATIONS

Plant material	% mortality after indicated days and concen.			
	15 days		30 days	
	1%	3%	1%	3%
<i>Alpinia galanga</i>	50.57	52.95	62.89	65.98
<i>Curcuma longa</i>	36.84	50.93	48.48	62.83
<i>Zingiber officinale</i>	37.15	49.61	49.08	61.52
<i>Curcuma zedoaria</i>	42.62	41.68	58.82	59.39
<i>Curcuma amada</i>	37.76	46.01	46.15	56.20
Control	19.78	19.78	29.61	29.61
S.Em	+1.47	+1.44	+2.42	+2.28
CD at 5%	3.08	3.03	5.09	4.79

regularly used as spices. They are safe from mammalian toxicity point of view, and therefore, the plants can safely be incorporated in cereals during storage for the prolonged protection against insect infestation.

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## EVALUATION STUDIES ON HIMACHAL GALGAL (*CITRUS PSEUDOLIMON* TAN.) FOR PROCESSING INDUSTRY

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Galgals from two geographic locations viz., Palampur and Nurpur in the Kangra valley of Himachal Pradesh were evaluated for physico-chemical characteristics of fruits with special reference to recovery of juice as well as yield and quality of oil and pectin from their processing waste. Yield and quality of oil and pectin were reasonably good from the waste of fruits from both the locations. Pectin yield from rags was much higher than from peel. Methoxyl content, degree of esterification and jelly grade of pectin extracted from rags were higher than those from other fruit components. Alcohol precipitation method gave higher pectin yield of good quality than aluminium chloride precipitation. A good quality oil was obtained by all the methods used for extraction though the cold press method gave the lowest yield. Fruit from Palampur gave higher oil yield.

Annual citrus production in Himachal Pradesh, at present, is 10,870 tonnes grown in an area of 31,276 hectares<sup>1</sup>. Among major citrus fruits, galgal (*Citrus pseudolimon* Tan.) an indigenous variety of lemon is growing well in low and mid hills comprising the belt of Hamirpur, Una and Kangra districts, Dhaulakuan area of Sirmour district, some parts of Mandi and Bilaspur districts of Himachal Pradesh. It is a very hardy plant and thrives well in rainfed areas and bears heavy crop. Fruits are available from early October to end of March. On an average, single galgal tree bears 300 kg of fruits and yield per acre is roughly estimated to 30,000 kg<sup>2</sup>. There is no industry in private or Government sector to utilize this fruit fully. Himachal Pradesh Horticulture Department had left galgal fruits worth Rs.12 lakhs unused during 1986 owing to inadequate processing facilities. However, a small quantity is being utilized for pickles and squashes in homes or small scale factories. This is the time to set up processing industries in galgal growing areas for utilizing the fruits including the waste which otherwise as fresh has low market value. After juice extraction, galgal waste accounts for 55-60 per cent which should be used for making useful by-products like oil and pectin which are well known products of commercial importance. Pectin yield of lemon on dry weight basis has been reported in the range of 21.7 to 33.6 per cent<sup>3</sup>. Keeping in view the large availability of this fruit in the region, oil

and pectin extracted from the waste of this fruit will be sufficient to meet our requirements.

Present investigations have therefore, been made to recover oil and pectin from processing waste of galgal grown in Himachal Pradesh by the standard methods followed by evaluation of these products for yield and quality. Also, physico-chemical characteristics of galgal fruits were determined for two most important citrus growing areas in Himachal Pradesh.

Fruits were harvested at ripe stage from Palampur about 1300 metres above mean sea level and Nurpur about 700 metres above mean sea level in the Kangra valley of Himachal Pradesh during mid November, 1988 after the colour of fruits changed from green to lemon yellow. Fruits were randomly selected for the determination of physico-chemical characteristics from each location and a sample of 5 fruits was taken for each observation with 3 replications. The total soluble solids were determined with an Erma hand refractometer (0 to 32° Brix). The total and reducing sugars were estimated by titration method<sup>4</sup>. Quantitative determination of ascorbic acid was done by using 2,6-dichlorophenol indophenol dye<sup>5</sup>. The pH was determined by pH meter Model-3030.

Peel oil from fresh peel (flavedo) was extracted using samples of 500g at each time for each method, i.e. steam distillation method, cold-press method<sup>6</sup> and solvent extraction method<sup>7</sup>. In solvent extraction method, fresh peel soaked for 72 hours in n-hexane released the oil from oil cells present in peel. Then, oil-in-hexane mixture was taken and subsequently hexane was distilled at 37°C from the mixture to recover the oil. Specific gravity, relative viscosity, saponification value, iodine value and acid value of oil were determined according to the method recommended<sup>8</sup>. Refractive index of oil was determined with Atago Digital Refractometer Model RX-I at 25°C.

Homogenous samples in three different groups of blanched and dried peel, rags and peel plus rags (200g each) were used for extraction of pectin by alcohol precipitation method<sup>8</sup> and aluminium chloride precipitation method<sup>9</sup>. Moisture and ash contents of pectin were determined by the recommended methods<sup>8</sup>. Methoxyl content, equivalent weight and anhydrogalacturonic acid content of pectin were also estimated<sup>10</sup>. Degree of esterification of pectin was calculated by using the formula given by Schultz<sup>11</sup>. Jelly grade and jelly setting time of pectin were determined by the standard methods<sup>8</sup>.

It is evident from Table 1 that the percentage of peel (albedo and flavedo), rags, juice and seeds showed negligible differences on the location basis. The contents of total soluble solids (T.S.S.), acidity, pH, sugars (total and reducing) and ascorbic acid were good and can be efficiently used for

preparation of various fruit products. Lower oil yield was recorded in cold-press method (Table 2) which may be attributed to the handling loss of oil during pressing the peel

and centrifuging the oil<sup>7</sup>. In general, oil yield of galgal was higher in the peel waste of fruits from Palampur as compared to Nurpur using different methods. Data also indicate that the specific gravity, refractive index, relative viscosity, saponification value, iodine value and acid value of oil showed the desired values in all the methods of extraction. Different locations also showed slight differences in the values of different quality parameters of oil. These slight variations in the quality of peel oil may be due to the fact that quality of oil depends upon locality of production, weather conditions and method of extraction<sup>12</sup>.

Pectin yield from rags was much higher than other fruit components (Table 3). It is also evident from the data that alcohol precipitation method gave slightly higher pectin yield than aluminium chloride precipitation method, in all the components of fruit used for pectin extraction. Pectin from rags had better quality than other fruit components used for

TABLE 1 PHYSICO-CHEMICAL CHARACTERISTICS OF HIMACHAL GALGAL

Particulars	Palampur	Nurpur
Albedo (%)	18.97	18.35
Flabedo (%)	7.09	6.88
Rags (%)	20.69	20.73
Juice (%)	48.90	49.33
Seed (%)	1.83	2.09
Total soluble solids (°B)	7.03	7.23
Acidity (% citric acid)	5.71	5.60
pH	1.48	1.61
Total sugars (%)	1.56	1.69
Reducing sugars (%)	1.23	1.36
Ascorbic acid (mg/100g)	18.46	18.37

TABLE 2. EFFECT OF METHODS OF EXTRACTION ON RECOVERY OF GALGAL OIL AND ITS QUALITY

	Palampur			Nurpur		
	Steam dist method	Solvent extr. method	Cold press method	Steam dist. method	Solvent extr. method	Cold press method
Oil yield (%)	1.22	1.33	0.62	1.04	1.16	0.46
Sp. gr. (25°C)	0.886	0.808	0.851	0.876	0.790	0.840
Refr. index (25°C)	1.471	1.4759	1.4714	1.4686	1.4748	1.4701
Relative viscosity (25°C)	1.127	1.188	1.169	1.109	1.178	1.154
Sap. value	194.09	200.61	207.99	180.69	185.48	188.99
Iodine value	16.32	16.83	17.96	15.25	15.66	16.70
Acid value (% oleic acid)	2.40	3.69	2.71	1.50	3.26	2.40

TABLE 3 EFFECT OF METHODS OF EXTRACTION ON RECOVERY OF GALGAL PECTIN AND ITS QUALITY

Characteristics	Location	Alcohol precipitation method			Aluminium chloride precipitation method		
		Peel	Rags	Peel + Rags	Peel	Rags	Peel + Rags
Pectin yield (%)	Palampur	17.46	21.08	19.13	16.49	19.14	17.80
	Nurpur	16.02	18.01	17.25	15.31	17.96	16.90
Quality of pectin							
	Moisture content (%)	Palampur	6.52	8.40	7.33	7.28	9.36
	Nurpur	6.29	8.00	7.02	7.04	8.97	8.09
Ash content (%)	Palampur	0.52	0.40	0.45	0.90	0.72	0.80
	Nurpur	0.40	0.35	0.40	0.77	0.63	0.70
Equivalent wt	Palampur	855	890	867	828	860	834
	Nurpur	832	868	852	805	840	822
Methoxyl content (%)	Palampur	8.80	9.53	8.67	8.50	9.12	8.55
	Nurpur	8.65	9.40	8.53	8.15	9.05	8.20
Anhydrogalacturonic acid (%)	Palampur	65.20	58.20	55.70	58.40	45.40	50.80
	Nurpur	55.00	52.30	50.30	50.50	42.20	43.30
Degree of esterification (%)	Palampur	76.63	92.97	88.37	82.63	114.05	95.55
	Nurpur	89.29	102.04	96.27	91.63	121.75	107.52
Jelly grade	Palampur	225	250	235	230	240	230
	Nurpur	210	225	230	200	220	220
Jelly setting time (min)	Palampur	13.00	5.00	9.00	14.00	6.00	10.50
	Nurpur	16.00	5.50	10.00	14.50	6.50	11.50

pectin extraction. In general, pectin obtained by alcohol precipitation method had better quality parameters. Different geographical locations also had influence on the quality of pectin.

The above studies suggest that from such good quality galgal fruit, the juice (40-45 per cent) should be utilized efficiently for preparation of citrus products. The left over peel and pomace (55-60 per cent) can be used for recovering the good quality peel oil and pectin of high grade.

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## A COMPARATIVE EVALUATION OF MEDIA FOR ENUMERATION OF ENTEROTOXIGENIC STAPHYLOCOCCI BY SELECTIVE ENRICHMENT TECHNIQUE

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Two enrichment media namely, Giolitti Cantoni's Tellurite broth (TEB) and Raj's Mannitol Salt Sorbic Acid (MSSA) broth were compared from selective isolation of enterotoxigenic Staphylococci from food samples. With the limited number of samples screened, the present findings indicate that Raj's MSSA medium has a distinct advantage for dairy products over TEB medium.

Resuscitation of Staphylococci from processed foods by selective and enrichment technique has been studied by several investigators<sup>1,5</sup>. The selective media employed for the isolation and enumeration of Staphylococci by direct plating techniques have been found to be inhibitory to the growth of these organisms<sup>1,2,6</sup>. It is difficult to achieve staphylococcal recoveries better than 70 per cent and in case of low numbers and debilitated cells, this becomes a critical factor<sup>2,3</sup>. Selective media are unsuitable due to the toxic effect of their ingredients in recovery of damaged cells<sup>7,8</sup>. In view of these factors, it has been found necessary to adopt enrichment methods for the detection and enumeration of Staphylococci especially when present in low numbers or in impaired state due to some form of food processing operations.

In the present work, a comparative study on the effective recovery of sub-lethally damaged Staphylococcal cells between two media namely TEB<sup>9</sup> and MSSA<sup>10</sup> has been attempted in food samples which when directly plated on Baird-Parker's ETGPA medium indicated total absence of Staphylococci.

Samples of different food commodities collected from retail outlets in Bombay were placed in sterile containers and were taken up for investigation within two hours. The dairy based products were emulsified by suspending 10 g of the samples in 90 ml sterile 1.25 per cent sodium citrate solution, by shaking with glass beads on a rotary shaker at 220 r.p.m. for 15 to 30 min so as to get a homogenous mixture. In case of meat, the samples were cut into small pieces, the tissue twitched with sterile scalpels and 10 g quantities were emulsified with 90 ml of sterile 1.25 per cent sodium citrate solution in a blender under aseptic conditions. The slurry was then transferred to a sterile conical flask and shaken at 220 r.p.m. with glass beads for 15 - 20 min.

*Enumeration of Staphylococci:* 0.1 ml from each sample was surface seeded on Baird-Parker's ETGPA media plates and incubated at 37°C upto 48 hr. The most probable number (MPN) of Staphylococci was noted by the 5 tube MPN enrichment method employing TEB<sup>9</sup> and MSSA<sup>10</sup> broth. (Tryptone, meat extract and yeast extract were purchased from Oxoid, England; Mannitol from Difco lab, U.S.A.; Glycine and Sodium pyruvate from E. Merck and Co., Germany; Sodium chloride from B.D.H. India and Cystine from B.D.H. England; Sorbic acid from Nutritional Biochemical Corporation, U.S.A.; Lithium chloride from Riedel De Haen AG, Germany and Thioglycollic acid from Eastman Kodak, U.S.A.) The tubes were incubated at 37°C for 48 hr. Tubes showing positive growth were streaked on ETGPA medium and incubated at 37°C upto 48 hr for further studies. Organisms from representative colonies developed on the plates were identified on the basis of morphological and biochemical characteristics. Enterotoxigenicity of the isolates was examined by PAG electrophoresis using partially purified enterotoxins A, B, C, D and E as reference standards<sup>11</sup>.

Both the media i.e. TEB and MSSA broth are partially selective for the growth of Staphylococci. Although Giolitti and Cantoni<sup>9</sup> reported that only Staphylococci produced a black precipitate in TEB medium, reduction of potassium tellurite was observed in all the positive TEB tubes. The results are similar to those reported by Baer *et al.*<sup>12</sup> who have observed 96 per cent tellurite reduction in growth tubes of which Staphylococcal growth was only in 41 per cent.

Consistently lower recoveries of Staphylococci were obtained by enrichment on TEB medium in frozen foods examined (ice creams). This suggested that Staphylococci subjected to a low temperature shock are not recoverable as completely on TEB as on MSSA broth. Microbiological standards for frozen prawns have been specified by Indian Standards Institution (ISI)<sup>13</sup>. Since the maximum limit for Staphylococci tolerated in such foods is 100/g, one has to have recourse to enrichment methods for enumeration. In view of the present data, there is a *prima facie* evidence to suggest that enrichment in MSSA broth would be more suitable than the TEB medium for enumeration of Staphylococci from frozen foods.

There is a remarkable variation in the media components of TEB and MSSA. In TEB, the selective enrichment of *Staphylococci* is brought about by addition of mannitol, glycine and sodium pyruvate whereas K-tellurite and lithium chloride are the inhibitory agents (K. tellurite inhibits Gram positive bacteria whereas lithium chloride inhibits Gram negative, lactose fermenting organisms). MSSA medium however, contains only mannitol as the selective component whereas sodium chloride and sorbic acid as the inhibitory agents. In terms of routine work, MSSA medium is convenient

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TABLE 1. CHARACTERISTICS OF *STAPHYLOCOCCI* ISOLATED FROM FOOD SAMPLES BY ENRICHMENT TECHNIQUE

Isolate number*	FTGPA medium	Chromogenesis <sup>1</sup>	Haemolysis of blood			Coagulase activity	Mannitol fermtn.		Gelatin liquifan.	Enterotoxigenicity
			Rabbit	Horse	Human		Aerobic	Anaerobic		
2	-	W	+	-	+	-	-	-	-	
3	-	W	+	+	+	-	+	+	-	
5	+	GY	+	+	+	+	-	-	+	
6	+	GY	+	+	+	+	+	+	+	
7	-	W	-	-	+	-	+	-	-	
8	-	W	+	-	+	-	+	-	-	
9	-	W	+	-	+	-	+	+	-	
11	+	GY	+	+	+	+	-	-	+	
14x	+	GY	+	+	+	+	-	-	+	
14y	-	W	-	-	+	-	+	+	-	

\*Isolate numbers are with reference to Table 1. 14x and 14y indicate that in sample number 14, two isolates were found.

1 : Chromogenesis on nutrient agar slants

GY : Golden Yellow

W : White

to prepare and handle whereas TEB medium involves use of Seitz filter for sterilization of potassium tellurite.

The characteristics of Staphylococci isolated from these foods have been tabulated (Table 1). The only common characteristic of all the isolates was to haemolyse human red blood corpuscles. About 40 per cent of the isolates produced golden yellow pigments. Among the isolates studied, complete correlation was observed between zone of clearance on Baird Parker's ETGPA medium, chromogenesis, coagulase activity and enterotoxigenicity.

Enrichment media that use MPN technique have been developed and tested for recovery of stressed *S. aureus* cells<sup>14-16</sup>. Heat injured Staphylococci show several damage sites. Magnesium and D-alanine are lost from the cell. Cell membrane damage is manifested by loss of salt tolerance and H<sub>2</sub>O<sub>2</sub> accumulation due to loss of catalase activity. Taking into consideration these facts, many of the enrichment media make use of catalase or sodium pyruvate incorporation. One of the most common MPN procedures is that of the AOAC which uses trypticase soy broth with 10 per cent NaCl. Although NaCl is a selective agent for Staphylococci, sub-lethally injured cells exhibit inhibitory effect of NaCl in both broth as well as agar. Lancette *et al.*<sup>16</sup> have reported that decrease in the inhibitory effect of NaCl with the addition of 1 per cent pyruvate MSSA enrichment broth has been found to be advantageous for MPN method in various ways. (a) The technique is more efficient for enumeration of low numbers of organisms (Fig. 1). (b) The medium is good for the foods that contain high numbers of other competing microflora. (c) Ten per cent NaCl is very selective for *S. aureus*. However, many of the sub-lethally damaged cells of Staphylococci do not retain their salt tolerance and hence might not grow in the medium.

Van Dorne *et al.*<sup>14,15</sup> have used a liquid modification of Baird-Parker's medium in which pyruvate incorporation helps in the recovery of sub-lethally damaged cells during anaerobic

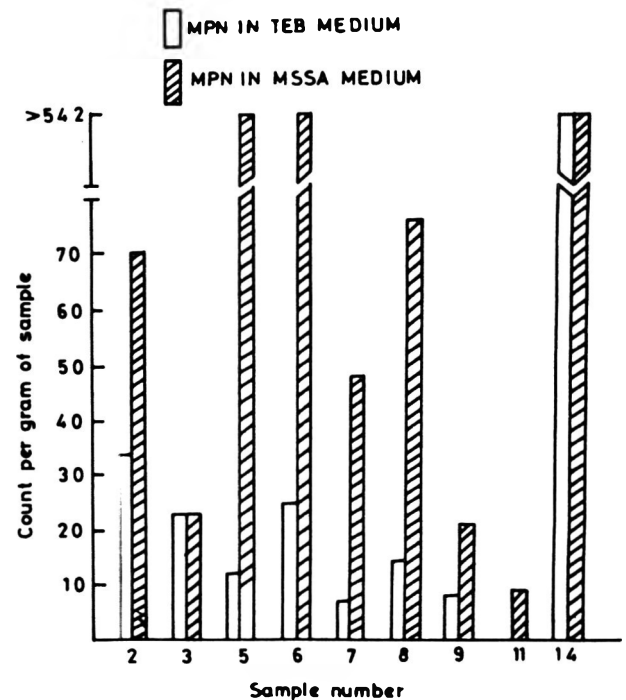


Fig. 1. Comparative evaluation of TEB and MSSA broth for the selective enumeration of Staphylococci.

incubation at 37°C for 48 hr. This medium also has shown certain disadvantages. (a) Addition of tellurite is to be done in each tube before inoculation. (b) Positive liquid B.P. medium tubes must be streaked because the broth is not selective. (c) A plug of liquid paraffin must be incorporated in each tube after inoculation and (d) Competing bacteria may outgrow low numbers of *S. aureus*.

Although TEB medium and Raj's MSSA medium were developed at about the same time, a critical evaluation of the latter medium by food microbiologists was in demand. With the limited number of food samples screened, the present findings indicate that Raj's MSSA medium has a distinct

TABLE 2. MPN OF MICROORGANISMS PER GRAM OF THE FOOD SAMPLES ESTIMATED BY TEB AND MSSA MEDIUM

Sample No.	Commodity	Isolate No.	MPN of TEB	Staphylococci MSSA
1	Ice cream	—	Nil	Nil
2	Ice cream	2	34	70
3	Ice cream	3	23	23
4	Ice cream	—	Nil	Nil
5	Kulfi*	5	12	542
6	Kulfi*	6	25	542
7	Kulfi*	7	7	48
8	Kulfi*	8	14	76
9	Cream	9	8	21
10	Cream	—	Nil	Nil
11	Cottage cheese	11	Nil	9
12	Cream	—	Nil	Nil
13	Sausages	—	Nil	Nil
14	Sausages	14x,14y	542	542
15	Sausages	—	Nil	Nil

\*Kulfi - A local ice cream recipe prepared from condensed milk evaporated in an open pan.

advantage over Giolitti and Cantoni's TEB medium (Table 2 and Fig. 1).

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## PARBOILING OF RICE: EFFECT ON PHYSICO-CHEMICAL, MILLING AND COOKING PROPERTIES

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Parboiling treatments of paddy varieties HM-95, Jaya, Basmati-370, PR-103, PR-106 and PR-107 improved head rice recoveries and pressure parboiling (steaming for 20 min at 15 lb/square in. pressure) proved to be more beneficial in increasing the head rice yield and reducing the free fatty acid content. Varieties and treatments had a significant effect on mean length/breadth ratio of milled rice kernels. Parboiling brought about increase in amylose content as compared to control. Traditionally parboiled rice required less time for cooking than pressure parboiled rice. Parboiling treatment increased elongation ratio and reduced the losses of solids and gruel upon cooking.

Parboiling of paddy by soaking in hot water has been practised since long and efforts were made to overcome certain disadvantages associated with traditional method. To cope up with the demand to produce better quality parboiled rice, Ali and Bhattacharya<sup>1</sup> introduced pressure parboiling in which paddy was wetted to desired moisture level before pressure cooking. Review of literature shows only limited work in Punjab where comparison of parboiling methods has been made in relation to physical, chemical and cooking properties of rice. So, the present study was conducted to find out the effect of traditional as well as pressure parboiling treatments on some high yielding varieties of Punjab rice.

Paddy of six high yielding varieties namely, 'HM-95', 'Jaya', 'Basmati-370', 'PR-103', 'PR-106' and 'PR-107' were procured from the Regional Rice Research Station, Kapurthala of Punjab Agricultural University, Ludhiana. The samples were cleaned and kept for two months as a prelude to treatments. Treatments given to paddy included (i) soaking paddy at 30°C until it attained 18 per cent moisture followed by steaming for 10 min at 15 lb per square in. pressure, (ii) soaking paddy at 30°C until paddy attained 18 per square in. pressure and (iii) soaking paddy at 70 ± 1°C for 2.5 - 3.5 hr until 35 per cent moisture was attained and steaming for 15 min under atmospheric pressure. A part of the paddy was not given any treatment which served as control. The samples were dried under shade to 14 per cent moisture and tempered for uniform moisture distribution.

All the paddy samples were shelled in the laboratory Satake Rice Sheller equipped with rubber rolls and milled on the Kett polisher (Type TP-2) to 5 degree of polish. Yields of brown rice, head rice and broken were calculated. Moisture, 1000 grain weight, grain dimensions and free fatty acids were determined by AACC<sup>2</sup> procedures. Amylose was estimated by the method of Juliano<sup>3</sup>. Cooking quality of intact kernels was determined using standard methods<sup>4</sup> for minimum cooking time, elongation ratio and solid loss in gruel. The summarized and statistically analysed data giving variation with parboiling treatments are given in Table 1.

Pressure steaming as well as traditional parboiling resulted in significant increase in the head rice yields of all the varieties. However, variations between treatments for head rice was non-significant. The results corroborate with those obtained earlier<sup>5</sup>. The increased yields have been attributed to reduced breakage of parboiled rice during milling due to hard gelatinized endosperm<sup>6</sup>. Traditional parboiling significantly decreased the length/breadth ratio of milled rice

TABLE 1. EFFECT OF PARBOILING TREATMENTS ON PHYSICO-CHEMICAL, MILLING AND COOKING PROPERTIES OF RICE

Treatment	Head rice recovery (%)	L/B ratio of rice	FFA (as % oleic acid, DM basis)	Amylose (%) (DM basis)	Min. cooking time (min)	Elongation ratio	Gruel solids (%)
Control	77.00 <sup>a</sup>	3.66 <sup>b</sup>	0.66 <sup>c</sup>	21.90 <sup>a</sup>	13.20 <sup>a</sup>	1.39 <sup>a</sup>	3.95 <sup>a</sup>
Steaming (10 min 15 lb/sq. in.)	90.90 <sup>b</sup>	3.62 <sup>b</sup>	0.26 <sup>b</sup>	22.90 <sup>b</sup>	19.30 <sup>b</sup>	1.43 <sup>b</sup>	3.11 <sup>b</sup>
Steaming (20 min. 15 lb/sq. in.)	94.70 <sup>b</sup>	3.61 <sup>b</sup>	0.19 <sup>a</sup>	23.40 <sup>c</sup>	20.50 <sup>c</sup>	1.44 <sup>b</sup>	2.78 <sup>c</sup>
Traditional parboiling	93.10 <sup>b</sup>	3.55 <sup>a</sup>	0.23 <sup>a</sup>	22.80 <sup>b</sup>	17.90 <sup>b</sup>	1.45 <sup>b</sup>	3.22 <sup>b</sup>
L.S.D. at 5%	4.07	0.05	0.06	0.47	1.77	0.03	0.25

Each value is mean of three replicates and representing six varieties. Means with different superscripts in the same column differ significantly. DM = Dry matter basis.

kernels than the control as well as pressure parboiled rice. A significant decrease in free fatty acids content was found with parboiling treatments. Steaming inactivates lipases and as a result free fatty acid content of bran as well as milled rice decreases. Parboiling treatments brought about significant increase in the amylose content over control. Among the different parboiling methods, pressure steaming for 20 min produced greatest effect. This might have been due to the fact that the fraction of total soluble amylose extracted by water was higher in treated samples than raw rice.

Parboiled rice required more cooking time than control. Among the treatments, pressure parboiling by steaming for 20 min required longest cooking time. This might have been due to increased kernel hardness during the process. Highly significant variations were observed for kernel elongation ratio between raw milled rice and parboiled rice, the ratio being more for parboiled rice. However, the differences due to traditional and pressure parboiling were non-significant. The increase in elongation ratio might have been due to increased grain length brought about by parboiling of paddy<sup>7</sup>. Parboiling had significant effect in decreasing the solids loss in gruel with respect to raw milled rice. However, the pressure parboiling was more effective than traditional one. The lower leaching of solids in parboiled rice was attributed to the increased resistance of starch in parboiled rice due to swelling and solubilisation in hot water.

It was observed that parboiling could be achieved even at a very low grain moisture content of 12-20 per cent by proper

pressure steaming and time combinations. Pressure parboiling has several advantages over traditional parboiling including saving of time in soaking. Secondly, the moisture content of pressure parboiled rice is only 22-24 per cent as compared to 32-35 per cent in traditional method, so drying time and energy is saved. Further leaching losses are eliminated resulting in increased per cent recovery and nutritional quality. The data obtained show that pressure steaming for 20 min at 15 lb per square inch pressure produced better results.

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## EXTENSION OF SHELF LIFE OF PANEER BY SORBIC ACID AND IRRADIATION

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**Paneer, an acid and heat coagulated milk product, is highly perishable because of high moisture (58-60%) and low acidity (pH 5.6-5.8). The product had 430 total plate count, 120 proteolytic bacteria, 170 lipolytic bacteria and 40 fungi/g of sample with a shelf life of few hr. Treatment with sorbic acid and/or gamma irradiation reduced the microbial load. Combination treatment of 0.10% sorbic acid in milk and irradiation of the product at 2.5 kGy preserved the paneer for 30 days at ambient temperature (25-35°C) with good acceptance.**

Cheese has not become popular in India because of the peculiar food habits and religious sentiments. In its place, an acid and heat coagulated milk product called 'paneer' is widely used. It is sometimes also referred to as cottage cheese. The product is extensively used as an ingredient in the preparation of vegetable curry and holds a prestigious position in the preparation of large number of culinary dishes. Paneer has outstanding nutritional qualities since it normally contains all the essential amino acids present in milk and also appreciable amounts of minerals and vitamins excepting vitamin B and vitamin C which are mostly lost in processing. Such nutrient rich products have good potential as an ingredient in the rations of service personnel who are posted at distant borders under inhospitable climatic conditions. However, due to high moisture content and low acidity, the product is highly perishable and its shelf life in climatic conditions of this country can be only a few hours.

Some work has already been done for the preservation of paneer, by low temperature<sup>1,2</sup>, drying<sup>3</sup> or frying<sup>4</sup>. Systematic work with special reference to its microbiological quality in relation to spoilage is scant. A feasibility study was, therefore, undertaken by making use of sorbic acid and  $\gamma$ -irradiation at a dose not exceeding 1 Mrad which has been unconditionally approved by the Joint Committee of FAO/IAEA/WHO<sup>5</sup> for all types of food.

Standardised and pasteurised milk from local dairy (Karnataka Milk Federation, Mysore) with 4.5 per cent fat and 8.5 per cent solids-not-fat (SNF) was used. For paneer preparation, the method described by Bhattacharya *et al.*<sup>2</sup> was followed. The pressed paneer was cut into small pieces weighing 50 g each, and packed in 400 gauge high density polyethylene pouches. One per cent solution of potassium sorbate (E. Merck) was heated to 70°C. The required amount of sorbic acid solution was added to hot milk (70°C), before

the addition of citric acid to get a final concentration of 0.05, 0.10 and 0.15 g per 100 ml of milk. The samples without sorbic acid were used as control. The packed paneer samples were irradiated at the rate of 0.5376 kGy/hr at a local irradiation plant situated at the Central Sericulture Research and Training Institute, Mysore. The plant having 2L capacity used Co<sup>60</sup> as a source of gamma rays. The treatment dose has been 0, 2.5, 5.0 and 7.5 kGy at ambient conditions. All treated and untreated samples were stored at 25-30°C (room temperature).

All samples were analysed initially as well as at intervals during storage for moisture<sup>6</sup>, pH and microbial load. Sorbic acid content was monitored using steam distillation method<sup>7</sup>. The sensory evaluation of samples was conducted by a team of seven panelists. The observations were recorded on a 9-point Hedonic scale<sup>8</sup>. Ten g of the well homogenised sample was used for microbiological analysis with quarter strength Ringer's solution as diluent. Samples were analysed for total plate count, proteolytic bacteria, lipolytic bacteria, coliforms and yeasts and moulds following standard methods<sup>9,10</sup>. Dextrose tryptone agar (DTA) was used for total plate counts and violet red bile agar (VRBA) for coliforms. For proteolytic bacteria, skim milk agar (SMA) was used. Lipolytic bacteria were enumerated on tributyrin agar (TBA). Yeasts and moulds were counted on potato dextrose agar (PDA) which was supplemented with 100 p.p.m. chloramphenicol to inhibit bacterial growth<sup>11</sup>.

Paneer prepared from standardised milk (4.5 per cent fat and 8.5 per cent SNF) had 58 to 60 per cent moisture and pH varied from 5.6 to 5.8. The control samples showed to harbour 430 total plate count (TPC), 120 proteolytic bacteria, 170 lipolytic bacteria and 40 yeasts and moulds per g, initially. Addition of sorbic acid to milk led to the reduction in microbial load in all groups. Addition of sorbic acid at a level of 0.05 per cent did not contribute much to the reduction of microbial load. However, addition of 0.15 per cent sorbic acid reduced counts to 140 TPC, 60 proteolytic bacteria, 80 lipolytic bacteria and 10 yeasts and moulds per g of paneer. The coliforms were not present in any of the paneer sample. The absence of coliforms can be attributed to their extreme sensitivity to heat during pasteurisation of milk as well as during paneer preparation.

During storage at ambient temperature (25 - 35°C), the samples added with 0.05 per cent sorbic acid spoiled within 24 hr. In 0.10 per cent sorbic acid treated samples, the bacterial population showed a continuous multiplication with time of storage till the end of 21 days and then it started decreasing. The increase in sorbic acid to 0.15 per cent level could not check the bacterial multiplication. However, the growth was slightly retarded and maximum numbers were recorded at the end of 30 days. Although sorbic acid at 0.10 - 0.15 per cent could not check bacterial population, it could



TABLE 3. SHELF LIFE IN DAYS OF SORBIC ACID ADDED AND IRRADIATED PANEER

Sorbic acid (%)	0 kGy	2.5 kGy	5.0 kGy	7.5 kGy
0	1	1	1	1
0.05	1	21	30	30
0.10	7	30	30	30
0.15	7	30	30	30

treatments. Individual treatment of either sorbic acid or irradiation gave only limited improvement in shelf life of the product. Although the recommended treatment (0.1 per cent sorbic acid in milk + 2.5 kGy gamma irradiation) resulted in residual level of preservative slightly more than the permitted level<sup>14</sup>, the study shows the possibility of storing a perishable product like paneer at ambient temperature. Some more studies are required to reduce the level of preservative further.

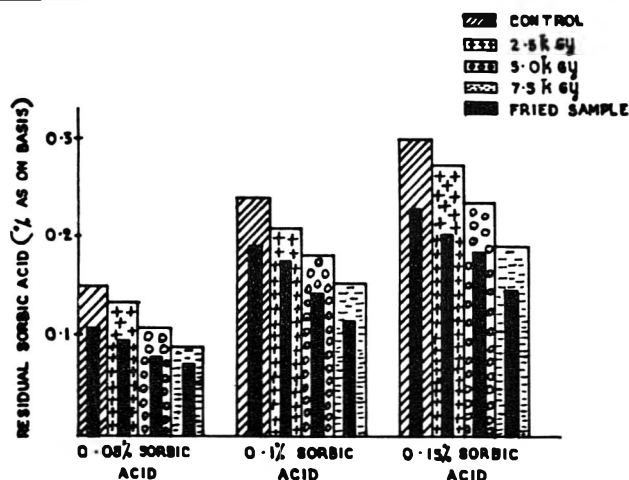


Fig. 1. Effect of irradiation on residual sorbic acid in paneer.

four treatments i.e., 0.05 per cent sorbic acid with 5.0 and 7.5 kGy, 0.1 per cent sorbic acid with 2.5 kGy and 0.15 per cent sorbic acid with 2.5 kGy gave acceptable product when scored on 9-point Hedonic scale. Out of these treatments, a combination of 0.1 per cent sorbic acid in milk and subsequent treatment of the product with 2.5 kGy irradiation was found to be the best. On some other milk based products, Bongirwar and Kumta<sup>11</sup> observed that combination treatment is able to store the product for 20 - 27 days at 35-40°C, although higher doses of irradiation seemed to decrease the acceptability of the product.

From these findings, it is concluded that in India where the ambient temperature is high, paneer can be successfully stored for a period of one month by the combination of

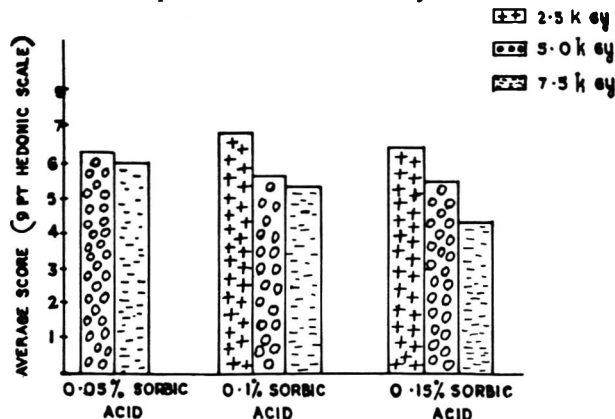


Fig. 2. Sensory evaluation of 30 days old paneer

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## STANDARDISATION OF MANUFACTURE OF HIGH FAT YOGHURT WITH NATURAL FRUIT PULP

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**Yoghurt made from milk with 8% sugar and homogenised at 200 bar pressure was found to be of good quality. Addition of mango pulp upto 4% of milk marginally improved the flavour characteristics. Addition of mango pulp higher than 4% affected the delicate yoghurt flavour and also the body and texture characteristics irrespective of homogenisation pressure. Homogenisation at higher pressure was observed to reduce the whey separation and also resulted in smoother consistency. Addition of mango pulp increased the softness and syneresis of yoghurt. Homogenisation at 200 bar pressure was observed to improve the consistency, water holding capacity and also the sensory characteristics. The Penetration Value (PV) and syneresis were observed to decrease with the increase in the homogenisation pressure.**

Yoghurt is a popular fermented product consumed in many parts of the world. It is produced in different forms like whole milk yoghurt, skim milk yoghurt, cream yoghurt, fruit yoghurt and drinking yoghurt. In India, *Dahi* is prepared commonly and yoghurt differs from *dahi* in the type of culture used for its manufacture. In the present paper, results of a study made to standardise the manufacture of high fat dessert type yoghurt with added fruit pulp are presented.

The milk was obtained from the experimental dairy of the Institute. In the trials, only cow's milk (fat=3.5 to 4.0 per cent and SNF=8.5 to 8.7 per cent) was used. The milk was separated into skim milk and cream. Skim milk was added to cream to obtain a fat content of 10 per cent. The corresponding SNF content ranged from 7.2 to 7.4 per cent.

**Fruit pulp:** Fruit pulps of mango and pineapple supplied by M/s WIMCO limited, Bangalore was used at different levels for the preparation of set yoghurt. Banana pulp was prepared in the laboratory by using bananas (Dwarf Cavendish variety) obtained from the local market.

Commercially available sugar was used at 8 per cent level. This level was arrived at by conducting the preliminary trials.

**Culture:** Yoghurt culture 709 containing a mixture of *Lactobacillus bulgaricus* (21 per cent) and *Streptococcus thermophilus* (79 per cent) supplied by M/s Laboratorium, Weisby GmbH, West Germany was used for the preparation of yoghurt.

**Preparation of yoghurt:** The milk standardised to 10 per cent fat was heated (85°C/15 min) and homogenised at 100,

150 and 200 bar pressures using 2 stage Gaulin homogeniser of USA. The temperature of the milk coming out of the homogeniser ranged from 70 to 73°C. This was subsequently cooled to 45°C and mixed with preboiled, cooled sugar syrup (prepared by mixing one part of water to 2 parts of sugar) along with calculated amount of fruit pulp. After the addition of pulp and culture, the inoculated milk was filled in cups (3.5cm height X 7.5cm dia.) and incubated at 42 ± 1°C till the pH dropped to 4.6 and then was stored in the refrigerator (8-10°C) for atleast 24 hr before being evaluated for sensory and physical characteristics. For preliminary investigations, mango (total solids (TS) content of 24 to 25 per cent), concentrated pineapple juice (TS of 8 to 10 per cent) and banana pulp (TS of 24 per cent) were used to assess the suitability of the pulp for the manufacture of dessert yoghurt. The preliminary studies indicated that the mango pulp was most acceptable and hence in all subsequent trials only mango pulp was used for the preparation of yoghurt.

**Fat and total solids (TS):** The fat in milk and TS in milk, yoghurt and pulp were determined by following the procedures described in IS: 1479<sup>1</sup>. pH of yoghurt was measured by using Elico digital pH meter.

**Penetration value (PV):** The PV was measured by using a penetrometer (M/s Associated Instrument Manufacturers (India) Ltd.). Rubber cone (28.5g) was specially designed at the institute to measure the hardness of the yoghurt. All the observations were recorded at 8—10°C after taking out from the refrigerator. The PVs obtained were inversely proportional to the firmness of the product and were expressed as  $\text{mm} \times 10^{-1}$ .

**Whey separation (syneresis):** The whey separation was measured by centrifugal method. The yoghurt samples were agitated by using mechanical stirrer operated at 2500 r.p.m. for 20 sec. Ten g of homogeneous sample was weighed in a graduated centrifuge tube and centrifuged at 5000 r.p.m. for 15 min. The whey separated from curd was then directly read on the graduated scale of the tube.

**Sensory evaluation:** The yoghurt samples were evaluated by a selected panel of judges for flavour, body and texture on a 10-point scale where 1 represented the most unacceptable and 10 represented most acceptable, while the appearance was evaluated on a 5-point scale where 1 represented most undesirable and 5 represented most desirable.

The sensory results of yoghurt prepared by using different levels of added mango pulp under different pressures of homogenisation are presented in Table 1. The yoghurt with 4 per cent mango pulp had the highest mean flavour, body and texture and appearance scores compared to those in yoghurts with other levels of pulp. The body and texture score of yoghurt prepared by addition of 6 per cent mango pulp was distinctly inferior which perhaps was due to the weak and granular body and texture characteristics and visible whey

TABLE 1. MEAN SENSORY SCORES FOR YOGHURT PREPARED WITH DIFFERENT LEVELS OF MANGO PULP.

Homogenisation pressures (bar)	% mango pulp added				
	0	3	4	5	6
<b>Flavour</b>					
100	8.4	8.8	8.5	8.5	7.8
150	9.0	9.0	8.8	7.6	7.0
200	8.8	8.5	8.8	7.5	7.2
<b>Body and Texture</b>					
100	7.9	8.2	8.0	7.0	5.0
150	8.2	8.2	8.1	6.9	5.4
200	8.6	8.4	8.4	7.0	5.6
<b>Appearance</b>					
100	4.6	4.2	4.3	3.5	3.4
150	4.7	4.5	4.3	3.6	3.0
200	5.0	4.6	4.6	3.5	3.4

separation. This could be ascribed to localised coagulation resulting in improper gel formation in yoghurt<sup>2</sup>. Since the flavour, body and texture scores of yoghurt with 4 per cent mango pulp were superior, this level of pulp incorporation was retained for the study of the syneresis and penetration value.

Homogenisation was observed to have favourable effect on the physical characteristics of yoghurt. Yoghurt homogenised at 200 bar pressure was observed to be firmer than the yoghurt prepared from milk homogenised at 100 and 150 bar pressures (Table 2). Addition of pulp resulted in softer yoghurt which could be due to the marginal dilution effect and due to the lower pH of pulp (3.8 to 4.0). The whey separation was also observed to decrease with the increase in the homogenisation pressure and it was minimum at 200 bar pressure. The syneresis was higher in yoghurt with mango pulp which could be due to the softer body characteristics of yoghurts prepared

TABLE 2. EFFECT OF ADDITION OF MANGO PULP AND HOMOGENISATION ON THE SYNERESIS AND CONSISTENCY OF YOGHURT (WITH 4% PULP)

	Homogenisation pressures (bar)		
	100	150	200
<i>Syneresis (10 g)</i>			
Plain (ml)	4.0	3.4	2.4
Pulp added (ml)	4.3	3.9	2.7
<i>Penetration value (mm × 10<sup>-1</sup>)</i>			
Plain	211.7	164.6	159.3
Pulp added	218.8	188.0	174.3

with added mango pulp. It was observed that the addition of mango pulp before fermentation lowered the pH of the mix from 6.6 to 6.1. This perhaps affected the gel structure compared to the yoghurt prepared without the added mango pulp. The homogenisation resulted in better gel structure because of the better distribution of fat globules<sup>3</sup>. The addition of mango pulp marginally weakened the gel structure which could be due to dilution effect or the particles weakening the gel strength by sitting in between casein micelles. This apart from reducing the gel strength also reduced the water holding capacity due to the weakened strength which was reflected in the form of increased whey separation.

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## HEAT RESISTANCE OF ENTEROTOXIGENIC *S. AUREUS* IN MILK, RECONSTITUTED INFANT FOOD AND CREAM

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Heat treatment of cells of *Staphylococcus aureus* strains 234, 834, 1151M, 790, S<sub>6</sub> and non-enterotoxigenic (NT) in different menstrua at 50°, 55°, 62.5°C for different time intervals resulted in their non-logarithmic death and hence F-values were determined. *S. aureus* 234 was the most heat resistant in both cow and buffalo milks as well as reconstituted infant food as evidenced by an F value of more than 120 min at 62.5°C. It was followed by *S. aureus* S<sub>6</sub>, 834, 1151M and 790. Buffalo milk conferred maximal protection to all the strains. The maximal recovery of *S. aureus* after heat treatment was recorded with soyabean-casein-digest-agar as compared with Baird Parker's agar. Both *S. aureus* 234 and S<sub>6</sub> survived at 71°C in pasteurized cream upto 120 min. The typical colony characteristics of the survivors from the cream on Baird Parker's medium were altered.

*Staphylococcus aureus*, the most common food poisoning organism capable of producing highly heat stable enterotoxins, viz., A, B, C, D and E, has been encountered in dairy products<sup>1</sup>. The residual staphylococci which escape sub-lethal processing treatments in the final product may endanger the health of the consumers by virtue of their ability to produce highly potent enterotoxins during faulty storage conditions prevalent in some tropical countries. In this connection, behaviour of enterotoxigenic strains of *S. aureus* in processed foods becomes highly significant as has been reported in our previous study<sup>2</sup>. Hence, this investigation has been undertaken to determine the survival of some of the selected enterotoxigenic strains of *S. aureus* subjected to defined laboratory heat treatments simulating commercial milk processing conditions.

Of the five enterotoxigenic strains of *S. aureus* used, four strains namely 234, 834, 1151M and 790 were procured from Food and Drug Administration, U.S.A. while one strain S<sub>6</sub> and non-enterotoxigenic strain NT were obtained from N.C.D.C., NDRI, Karnal. All cultures were propagated for 18 hr in brain heart infusion (BHI) broth at 37°C, and centrifuged at 4,000 x g for 15 min. The cell pellet was washed twice by re-suspending in phosphate buffered saline (PBS), pH 7.3, followed by centrifugation after each washing. Finally, the cell pellet was suspended in the same buffer and stirred thoroughly before adjusting to 0.30 O.D. at 540 nm. One ml of this standardized cell suspension when added to 99 ml of

each heating menstruum gave a final population of approximately  $1 \times 10^7$  viable cells per ml. The heating menstrua used in the present study were sterilized PBS (pH 7.3), sterilized full cream cow's milk (4.0 per cent fat), sterilized full cream buffalo milk (7.0 per cent fat), 11 per cent sterilized reconstituted infant food (Glaxo, spray dried) and pasteurized cream (60 per cent fat). The ampoule method<sup>3</sup> was used for heat resistance studies at 50°, 55°, 62.5°C in the heating menstrua for 0, 5, 10, 20, 30, 40, 50, 60, 70, 80, 90, 100, 110 and 120 min. However, in case of cream, heat treatment was given at 71°C for different time intervals.

The viable cells after the heat treatment were determined by the spread plate method using Baird Parker's Agar (BPA) and soybean casein digest media (SCD) (Hi-Media, India) as the recovery media. After spreading, the plates were incubated at 37°C for 48 hr and the typical colonies that developed on the respective media were counted. In order to confirm that the survivors belonged to *S. aureus*, a few colonies surviving heat treatment were randomly picked from the recovery media plates and subjected to microscopic examination and a battery of biochemical tests as described previously<sup>2</sup>. Based on these observations, the colonies appeared to be typical *S. aureus*. The survivor curves were drawn by plotting time in min on the X-axis and log number of survivors on the Y-axis. F-values were calculated according to the method of Banwart<sup>4</sup>. All the trials were conducted in triplicate and their average values were calculated.

The order of death of *S. aureus* strains examined during this study was non-logarithmic on the basis of their thermal death curves. Hence, D and Z values could not be determined. Instead, thermal death point measurements, i.e., F-values were determined as they give direct measurements of times required for a given probability of a kill at a given temperature. All the strains of *S. aureus* used in this investigation had different F-values in a particular menstruum at a particular temperature (Table 1). On the basis of their F-values, the strains can be arranged in the following descending order of their heat resistance -*S. aureus* 234, S<sub>6</sub>, 834, 1151M, 790 and NT. The differences in F-values of the different strains may presumably be due to variation in heat resistance characteristics of individual strains. The variations in the efficiency of recoveries of the survivors in the present study resulted in different F-values on the two recovery media. This finding is in agreement with the observations of Reiter *et al.*<sup>5</sup>, Baird-Parker and Davenport<sup>6</sup> and Allwood and Russel<sup>7</sup> also observed similar variations in the recovery of heat stressed cells on the different recovery media. By and large, the SCD medium recorded maximum number of survivors (higher F-values) as compared to BPA irrespective of the strain and the heating menstruum. Ibrahim<sup>8</sup> also found SCD as a better recovery medium for stressed *S. aureus*

TABLE 1. HEAT RESISTANCE OF *S. AUREUS* IN TERMS OF F-VALUES (IN MIN.) IN DIFFERENT MENSTRUA

Temp. (°C)	<i>S. aureus</i> S <sub>6</sub>		<i>S. aureus</i> 234		<i>S. aureus</i> 834		<i>S. aureus</i> 1151M		<i>S. aureus</i> 790		<i>S. aureus</i> NT*	
	BPA	SCD	BPA	SCD	BPA	SCD	BPA	SCD	BPA	SCD	BPA	SCD
<b>Phosphate buffered saline</b>												
50	> 120	> 120	> 120	> 120	> 120	> 120	> 120	> 120	> 120	> 120	> 120	> 120
55	> 120	> 120	> 120	> 120	80	100	40	80	30	50	12	22
62.5	40	50	80	100	40	60	23	30	10	10	2	5
<b>Cow milk</b>												
50	> 120	> 20	> 120	> 120	> 120	> 120	> 120	> 120	> 120	> 120	> 120	> 120
55	> 120	> 120	> 120	> 120	100	> 120	80	100	50	60	21	42
62.5	60	80	> 120	> 120	60	80	40	50	20	30	11	20
<b>Buffalo milk</b>												
50	> 120	> 120	> 120	> 120	> 120	> 120	> 120	> 120	> 120	> 120	> 120	> 120
55	> 120	> 120	> 120	> 120	120	120	100	120	70	20	30	40
62.5	80	120	> 120	> 120	70	100	70	90	30	40	13	20
<b>Reconstituted infant food</b>												
50	> 120	> 120	> 120	> 120	> 120	> 120	> 120	> 120	> 120	> 120	> 120	> 120
55	> 120	> 120	> 120	> 120	100	> 120	70	90	40	50	15	20
62.5	90	110	100	120	50	70	30	40	10	19	5	12

\**S. aureus* NT, a non-enterotoxigenic strain served as a control BPA: Baird - Parker's Agar; SCD : Soybean Casein Digest.

cells. In case of all the five strains of *S. aureus* used in this study, F-values were higher in buffalo milk followed by cow's milk, reconstituted infant foods and phosphate buffered saline. Higher F-values in buffalo milk may be due to higher solid content which may protect the organism against heat treatment as has been suggested by others<sup>9,10</sup>. On the other hand, when reconstituted infant food was used as the heating menstruum, *S. aureus* 234 was not destroyed at 50 and 55°C even after 120 min but was completely inactivated within the same period at 62.5°C (F 120). Reconstituted infant food offered lesser protection as compared to cow's and buffalo milk. This may presumably be due to lower total solids in reconstituted infant foods.

The data pertaining to heat resistance of *S. aureus* strains in pasteurized cream at 71°C have been plotted in Fig. 1. Again, the thermal death curves were non-logarithmic and hence F-values were determined. F-values were considerably higher in cream as compared to other heating menstrua. Both *S. aureus* 234 and S<sub>6</sub> exhibited higher heat resistance in cream as is evidenced by higher F-values ( 120 min). They were followed by *S. aureus* 834, 1151M and 790 (120, 110 and 90 min). The higher heat resistance of *S. aureus* in cream compared to other menstrua might be due to the higher fat content which could offer additional protection to the organism against heat even at higher temperature. The striking observation in case of survivors withstanding heat treatment in cream was the absence of opalescence zones around the colonies recorded on Baird-Parker's medium in almost all the strains. Some of the colonies exhibited lobed edges as compared to entire margins of their unheated counterparts

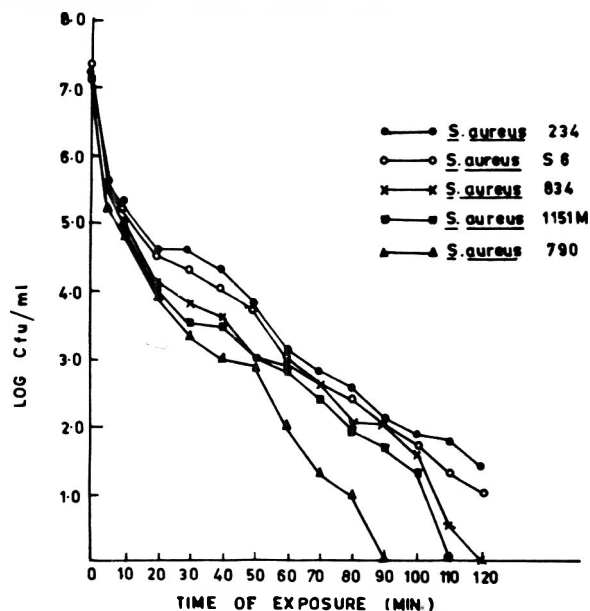


Fig. 1. Survivor curves of *S. aureus* strains in pasteurized cream at 71°C after different time intervals.

indicating that the heat treatment in cream can also alter the colony characteristics of the culture. The variations in the biochemical characteristics of *S. aureus* after heat treatments were also recorded during this study and the same have been described elsewhere<sup>2</sup>.

From the foregoing study, it can be concluded that enterotoxigenic *S. aureus* can survive heat treatment during the processing of milk. Higher total solids and fat contents seem to confer greater protection during heat treatments. The

importance of using suitable recovery media to allow heat stressed cells to form normal colonies is indicated.

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## EFFECT OF SUPPLEMENTING POULTRY MEAT WITH TEXTURED SOY ON THE QUALITY OF LOAVES

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The chicken meat loaves were prepared from culled hens and broiler breeder males using minced meat in combination with various levels of textured soy (TS). The TS was incorporated at levels of 10, 20, 30, 40 and 50% of the meat. The sensory evaluation revealed that loaves containing TS upto 30% level were acceptable. The broiler samples were preferred to hens but the preference was not significantly different. The loaves had a shelf life of about 12 days at refrigeration temperature.

The laying hens and broiler breeder males at the end of their active life period are considered a by-product of the egg and broiler industry<sup>1</sup>. The meat from such birds is tough and not readily acceptable to the consumer and fetches lower price to the producer. The addition of textured soy or soy protein isolates, increases binding of fat and water, improves adhesion/cohesion and reduces weight loss during cooking of sausage type products<sup>2</sup>. Poultry sausages have been successfully produced by blending chicken with vegetables<sup>3</sup> and binder such as dried skim milk, semolina, corn starch and pasteurized egg white<sup>4</sup> and textured soy flour<sup>5</sup>.

Meat loaves are a delicacy and traditionally prepared from pork. With this in view, the present study was undertaken to investigate the effect of supplementing poultry meat from culled layers and broiler breeder males, with textured soy on the quality of loaves.

Approximately 72-75 week old culled white leg-horn hens and broiler breeder males were procured from the Department of Animal Sciences at PAU, Ludhiana. The birds were slaughtered and dressed using standard procedure. The dressed birds were chilled and packaged in 150 gauge polyethylene bags and kept in deep-freezer at -10°C till the deboning was completed. The deboned meat was minced using manually operated meat mincer and packaged in polyethylene bags for further use.

Textured soy (Nutrella) nuggets procured from the local market were rehydrated in warm water. Extra water was drained and the nuggets were minced in the manual meat mincer. The minced material was used to supplement chicken meat at different levels in the recipe (Table 1), for preparation of loaves. Twelve loaves weighing about 2.5 kg each were prepared, five each for meat from culled layers and broiler

TABLE 1. RECIPE FOR CHICKEN LOAF PREPARATION

Material	per cent
Deboned chicken meat	75
Fat	7
Ginger (grated)	1
Garlic (grated)	1
Onion (grated)	2.5
Phosphate	0.4
Sodium nitrite	0.01
Sodium nitrate	0.03
Black pepper	0.2
Coriander	0.2
Cumin	0.3
Cinnamon	0.1
Salt	1.8
Sugar	1.3
Water	9

breeder males containing Nutrella at 10, 20, 30, 40 and 50 per cent levels and one each as control.

All the ingredients were mixed, passed through manual mincer (2-3 times) to obtain a fine paste. The resulting batter was stuffed into Ham moulds (M/s Gardner Corpn., New Delhi) and compressed. The moulds were then placed in water at 70-72°C for 3½ hrs (till the temperature at the centre of the loaves was 71°C). The moulds were drained, cooled and the loaves were removed from the moulds. The loaves were sliced, packed in polyethylene bags and stored in refrigerator. The stored samples were evaluated for sensory qualities by a semi-trained panel using a semi-structured scale<sup>6</sup> and proximate composition was determined by Standard AOAC methods<sup>7</sup> at an interval of two days. The whole experiment was repeated twice.

The proximate composition showed that meat from broiler breeder males contained more moisture (73.2 per cent) and fat (9.8 per cent) than that from hens. The proximate composition of loaves from either type of meat (Table 2) revealed that the moisture contents of loaves increased with the increase in the level of total solids (TS) in both fresh as well as 14 days old samples, while the fat content decreased. The storage resulted in a decrease in the moisture content, which was more in case of control and 10 per cent TS containing samples as compared to those containing higher levels. The decrease in moisture during storage in case of 10 per cent TS samples can be attributed to exudation in stored samples on thawing. Absence of exudate on thawing of samples containing higher levels of TS may be due to improved water binding.

There was no significant change in sensory panel scores upto 12th day of storage. However, the subsequent storage resulted in a decrease when compared to fresh samples. The supplementation of chicken meat with 10, 20 and 30 per cent TS improved the texture of loaves as compared to control.

TABLE 2. PROXIMATE COMPOSITION OF CHICKEN LOAVES (N = 3)

Textured soy added (%)	Hen						Broiler breeder males					
	0 day			14 days			0 day			14 days		
	Moisture (%)	Fat (%)	Protein (%)	Moisture (%)	Fat (%)	Protein (%)	Moisture (%)	Fat (%)	Protein (%)	Moisture (%)	Fat (%)	Protein (%)
0	62.3	14.2	15.5	60.1	14.1	17.8	62.9	12.0	18.2	60.5	12.1	19.8
10	64.4	11.2	17.2	63.2	11.4	18.1	64.8	10.6	19.3	62.5	10.5	19.9
20	64.8	9.6	19.5	63.5	9.5	18.8	65.2	8.9	18.5	63.8	9.0	20.5
30	64.8	9.0	20.2	63.4	9.1	21.0	65.4	8.5	18.8	63.6	8.5	20.7
40	66.6	8.7	17.5	66.3	8.8	18.8	66.9	7.8	18.2	66.5	7.9	19.2
50	67.9	8.0	17.6	67.8	8.2	17.3	67.8	7.3	18.0	67.7	7.5	18.6
S.D.	+0.47	+0.57	+0.43	+0.66	+0.57	+0.30	+0.43	+0.47	+0.12	+0.64	+0.45	+0.18

The evaluation of sensory panel scores of loaves indicated an improvement in sensory attributes upto 30 per cent level of TS. The samples containing 40 per cent TS were fairly acceptable while those containing 50 per cent were unacceptable. The source of meat did not have a marked effect on the sensory scores of meat loaves for any of the attributes.

The meat loaves can be prepared with meat from either the culled layers or broiler breeder males. The textured soy (TS) can be incorporated into poultry loaves upto 30 per cent level of supplementation without having a marked effect on the acceptability. The loaves thus produced have a shelf life of about twelve days in polyethylene bags at refrigeration temperature.

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## EVALUATION OF SENSORY ATTRIBUTES AND SOME QUALITY INDICES OF IRRADIATED SPICES

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Ground pepper, chilli, coriander, cumin, turmeric and a spice mix (sambar powder) exposed to an absorbed dose of 10 kGy of gamma irradiation were studied for microbial and sensory quality changes, if any, on storage for 3 months. No significant differences between the irradiated and non-irradiated samples could be observed in their sensory qualities. While no increase in microbial load in sealed irradiated samples could be observed, those that were irradiated but opened repeatedly for daily use did show a marginal increase in microbial load.

The efficacy of gamma irradiation in the decontamination of spices has been established during the past few years<sup>1,2</sup>. Irradiation doses of 3-5 kGy were able to take care of thermophilic spore formers, mesophilic spore formers, yeast, moulds and coliforms. But the total plate counts required 10 kGy for their complete elimination<sup>3</sup>, and this dosage was effective in destroying pests as well<sup>4</sup>.

An important aspect of irradiation technology is the consumer acceptance of irradiated spices. Though objective analyses of essential oils and other chemical and microbiological attributes have been extensively reported<sup>5,6</sup>, reports relating to sensory qualities and acceptability of irradiated spices are rather limited<sup>6</sup>. The present study was, therefore, focussed on colour, pungency, microbial count and sensory evaluation of irradiated and non-irradiated spices.

Samples of ground spices (black pepper, chilli, coriander, cumin, turmeric) and a spice mix (sambar powder) of Khamkar brand were procured from a local consumer co-

operative store and exposed to an absorbed dose of 10 kGy of gamma irradiation. The unexposed samples from the same lot served as control. The irradiated and control samples were stored under (a) sealed and (b) repeatedly - opened conditions. The spices were evaluated for microbiological and sensory quality during storage upto three months. The total plate count was determined by the procedure described by ASTA<sup>7</sup> and standardised at BARC, Bombay. The curcumin present in turmeric was determined and the colour intensity was calculated therefrom<sup>8</sup>. Pungency in chilli-capsicum was tested by the dilution at which the burn sensation was identified<sup>8</sup> in 30 sec after swallowing a few drops of the sample.

A triode test was conducted to determine the quality of raw spice by a trained panel of 10 members who were found to have the capacity to distinguish the different degrees of concentration of the basic tastes and odours.

The cooked samples were evaluated for acceptability on a modified ISI Hedonic scale<sup>8</sup> for the sensory attributes like colour, appearance, odour, pungency, texture and taste. The scores represented: Excellent = 5, Good = 4, Fair = 3, Poor = 2 and Inedible = 1. The standardised recipes that incorporated different spices were: Pepper rasam, spicy chilli potatoes, coriander potato curry, cumin rice, sambar and lime rice (with turmeric).

All the spices except cumin powder showed a high total plate count (TPC) at start on purchase in sealed packs ( $1.5 \times 10^3$  cfu/g to  $6 \times 10^3$  cfu/g). After irradiation, however, no colony forming units were seen in any of the spice mixes, showing that 100 per cent elimination of microbial contamination in spices is possible with irradiation (Table 1).

The extent of recontamination during storage under home conditions, where the samples were opened and closed repeatedly for daily use, was found to be much lower in irradiated samples as compared to the non-irradiated ones. On storage for one month even in sealed conditions, the TPC in non-irradiated samples was high, but the samples that were opened and closed repeatedly showed a still higher TPC. The increase in contamination of the non-irradiated spice was

TABLE I. PLATE COUNTS IN IRRADIATED AND NON-IRRADIATED SPICES OVER A PERIOD OF 3 MONTHS

Spice powder	Plate counts (cfu/g × 10) <sup>3</sup> at indicated months of storage															
	Non-irradiated (sealed)				Non-irradiated (simulated to home conditions)				Irradiated (sealed)				Irradiated (simulated to home conditions)			
	0	1	2	3	0	1	2	3	0	1	2	3	0	1	2	3
Chilli	2.56	10.19	12.60	13.20	2.56	14.79	16.60	17.80	0	0	0	0	0	0	0.58	0.84
Turmeric	0.06	15.43	13.12	14.40	6.06	17.85	15.58	14.43	0	0	0	0	0	0	0	0.06
Black pepper	5.39	11.50	14.69	15.21	5.39	16.21	34.97	32.36	0	0	0	0	0	1.08	1.49	2.15
Coriander	1.45	2.50	4.64	5.20	1.45	1.43	10.15	10.81	0	0	0	0	0	0	1.60	2.10
Sambar	4.57	3.37	5.06	6.01	4.57	7.56	47.8	41.80	0	0	0	0	0	0	0.93	0.91
Cumin	0	3.86	2.50	2.10	0	3.26	5.77	5.12	0	0	0	0	0	0	0	0



16 times that of irradiated samples during the first month of storage. The increase in microbial growth was much less during the second month of storage. Pepper was found to be the most easily contaminated spice. Thus, 1 dose of kGy eliminated the microflora in all the spice samples and there was only a marginal increase in microbial load on storage in semi-sealed condition.

Colour intensity of turmeric as percentage curcumin was found to be fairly stable during storage, in both irradiated and non-irradiated samples though there was a slight difference between irradiated and non-irradiated samples even before storage. This slight increase in intensity of colour in irradiated turmeric could be due to improved curing and colour extractability<sup>3</sup>.

The pungency of irradiated samples again was found to be higher than the non-irradiated samples. This increase in the pungency of irradiated chilli powder could be due to the possible enhancement of the volatile oils during the irradiation process and needs further study<sup>6</sup>. While the pungency units remained the same during storage of non-irradiated spices, there was an increase in the irradiated samples during the first month of storage, remaining constant thereafter.

The sensory evaluation of the raw as well as cooked spices showed no difference in the overall acceptability either between the irradiated and non-irradiated spices or between the sealed and stored samples and repeatedly opened and closed samples. While no difference in appearance, texture and taste was observed in irradiated and non-irradiated spices there was a slight but non-significant decrease in the scores assigned for sambar prepared from irradiated spice mix.

Similarly, there was a slight enhancement in odour and pungency of chilli powder and pepper powder on irradiation. The slight increase in odour and pungency in pepper powder could be due to the oxidation occurring during irradiation and production of pepric acid along with other carboxylic acids resulting in an increase in volatile components<sup>6</sup>. Thus, neither irradiation nor storage was found to affect the sensory attributes of spices.

It is concluded that irradiation improved the quality of spices without altering their sensory quality and acceptability.

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### NEW PUBLICATION FROM CFTRI

The Central Food Technological Research Institute, Mysore has just brought out under the Industrial Monograph Series, new Monograph entitled "LITCHI IN INDIA - Production, Preservation and Processing". Beautifully brought out, and scholarly presented, this Monograph covers in a nutshell the following aspects: Propagation, cultivation, pests and diseases, physico-chemical composition, yield and harvesting, physiological disorders, grading, packaging, transportation and storage, physico-chemical changes during ripening and processing. An exhaustive list of references and an appendix of equipment/machinery (with suppliers' names and addresses) required for manufacture of Litchi products, are useful additions. It is Priced at Rs. 40/- (Postage extra).

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## A REPORT OF MYCOTOXIN CONTAMINATION IN BHUTANESE CHEESE

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Nine dominant fungi were isolated from 19 samples of cheese obtained from Bhutan. The incidence of *Aspergillus ochraceus* was maximum (74%) followed by *Penicillium citrinum* (68%). Twelve of the 19 cheese samples were found to be contaminated with different levels of mycotoxins. Citrinin was detected in maximum number of cheese samples (6), either alone or as co-contaminant with ochratoxin A. Three samples of cheese were also contaminated with aflatoxins.

Mycotoxins are secondary metabolites, representing a wide diversity of chemical species, mainly produced by certain toxigenic strains of *Aspergillus*, *Fusarium* and *Penicillium* species during their development in food and feed. These fungi contaminate a variety of food and feed items<sup>1-3</sup>. Mycotoxin contamination of cheese and dairy products has also been reported<sup>4,6</sup>.

On an occasional visit to Thimpu, the capital of Bhutan, it was noticed that a variety of cheese, sold in the market, were very commonly used by the local people. It was observed that the mouldy growth of brown and green colony was evident on the cheese. Owing to this, a study was undertaken to record the dominant mycoflora and their consequent problem of toxin contamination. The variety of cheese described in the current study is called *chuku* and *chhurpi* in Bhutanese and Nepalese languages, respectively.

During September-October, altogether 19 samples (250 g/sample) were obtained from different shops at Thimpu. In the laboratory, all the samples were kept in refrigerator (-4°C) to check further fungal growth. Ten g cheese samples from each lot were cut into small pieces and were shaken in 20 ml sterilised distilled water, which was further decanted and centrifuged at 1000 r.p.m. for 10 min. The supernatant was aseptically placed in petri dishes containing potato dextrose agar medium. These plates were incubated at 28 ± 2°C, and the incidence of fungi was recorded after 3-4 days.

For the analysis of toxins, the samples were dried at 60°C for 3-4 days and were ground to fine powder. Fifty g powder from each sample was further extracted for mycotoxins following the method of Roberts and Patterson<sup>7</sup>. Qualitative assay of different mycotoxins were done by TLC. The solvent system toluene:ethylacetate:formic acid (6:3:1) was used for separation of toxins. Chromatoplate after irrigation was

observed under long wave UV-light for comparison of sample with standard toxins (obtained from Sigma Chemical Co., St. Louis, U.S.A.). Chemical confirmation of aflatoxin B<sub>1</sub> was performed by spraying aqueous sulphuric acid (25 per cent). The presence of ochratoxin A and citrinin was confirmed by treatment with ammonia vapour<sup>8</sup>. The extracts positive to toxins were re-examined on high performance thin layer chromatographic (HPTLC) plate, as before. Quantification was done fluorodensitometrically with the help of CAMAG TLC Scanner-II, using D<sub>2</sub> Lamp and K-400 filter. At specific retention, the peak due to sample spot and standard toxin spot were achieved through SKLAR integrator. The quantity of toxins was calculated as per the following formula (standardized in the laboratory):

$$\text{Amount of toxin present in } 1 \mu\text{l of sample extract (W)} = \frac{V \times W}{A \times v} \times a$$

Where,

A = area of standard spot peak; a = area of sample spot peak; V = volume of standard spotted; U = volume of sample extract spotted and W = amount of standard solution in  $\mu\text{l}$ .

During the visit to Thimpu, the climatic conditions were moderately cold in the night. The day was sunny (temperature 26-30°C) with occasional rains.

Altogether 6 fungi belonging to 4 genera were recorded as dominant fungi (Table 1). *Aspergillus ochraceus* was isolated from maximum number of samples (74 per cent) followed by *Penicillium citrinum* (68 per cent) and *A. flavus* (47 per cent). The number of fungal colonies varied in different samples. *A. ochraceus* was in the range of 6-10 colonies/sample, while the number of colonies of *A. flavus* and *P. citrinum* was between 3 and 7 and 3 and 9 respectively.

Of the 19 cheese samples, 12 were contaminated with different mycotoxins (Table 2). Citrinin was the most prevalent, which alone was detected in 4 samples and as co-contaminant with ochratoxin A in two samples. The amount

TABLE 1. FUNGI ISOLATED FROM CHEESE SAMPLES

Dominant fungi	% samples infected	No. of colonies in positive sample (range)
<i>Aspergillus flavus</i>	47	3-7
<i>A. niger</i>	21	5-8
<i>A. nidulance</i>	37	2-3
<i>A. ochraceus</i>	74	6-10
<i>Aspergillus</i> spp.	37	4-9
<i>Penicillium citrinum</i>	68	4-9
<i>Penicillium</i> spp.	63	3-8
<i>Rhizopus</i> sp.	42	2-6
<i>Mucor</i> sp.	32	3-4

Mean of 5 plates/sample

TABLE 2. MYCOTOXINS IN CHEESE SAMPLES

Contaminated sample	Mycotoxins detected	Amount ( $\mu$ g/kg)
1	Ochratoxin A	80
	Citrinin	143
2	Aflatoxin B <sub>1</sub>	188
	Aflatoxin B <sub>2</sub>	56
3	Citrinin	74
4	Aflatoxin B <sub>1</sub>	212
5	Ochratoxin A	42
6	Ochratoxin A	74
7	Citrinin	92
	Ochratoxin A	109
8	Aflatoxin B <sub>1</sub>	68
9	Citrinin	128
10	Citrinin	224
11	Citrinin	88
12	Ochratoxin A	116

of citrinin was estimated to be between 74 and 224  $\mu$ g/kg. Of the five samples contaminated with ochratoxin A, the level was in the range of 42-116  $\mu$ g/kg. Three samples contained aflatoxin.

Occurrence of mycotoxigenic fungi and mycotoxins may be attributed to unhygienic system of storage. In a spot study, it was observed that the pieces of cheese are tagged together with a jute thread and hung in the shops for selling. The

exposed cheese gets the fungal contamination from the surrounding environment. Being a substrate, cheese forms a good base for the growth and development of toxigenic moulds leading to the formation and accumulation of toxins.

We are grateful to Professor K.S. Bilgrami, Head, University Department of Botany, Bhagalpur University, for necessary facilities.

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## IFCON-93

III International Food Convention (IFCON-93) will be organised by AFST (I) during January/February 1993 at CFTRI, Mysore. Contact has also been made with the International Union of Food Scientists and Technologists (IUFoST) Secretariat to arrange for a regional seminar of one day duration during the period. The theme of IFCON-93 Convention is:

### FOOD TECHNOLOGY FOR BETTER HEALTH

A Food Exhibition is also being planned as part of the Convention. Look for further announcement.

*Nutrition: Proteins and Amino Acids*: Edited by Yoshida, Akira, Naito, Hiroshi, Niiyama, Yoshiaki, Suzuki, and Takeshi, Japan Scientific Societies Press, Tokyo, 1990; pp: 327; Price: DM 148.00.

This is a compilation of the research findings of the Research Committee of Essential Amino Acids (Japan) over a period of about 30 years. Contributors have been scientists of long standing, actively engaged in the field of proteins and amino acids.

Historical outlines of studies on amino acid nutrition in Japan provided important information on trends of net food supply, per capita per day in calories, physique of school children and their daily food nutrient requirements, protein sources, their utilization etc., Protein synthesis and gene expression have been written well and have contributed very good information. Ribosome metabolism in association viz. 5 S-RNA-protein particle, ribosomal protein interaction with m-RNA and synthesis of ribosomal proteins are very well presented and provide useful information on eucaryotic ribosomes and their stability. For intracellular protein degradation, lysosomal and non-lysosomal pathways have been emphasized. Various enzymes, factors and hormones in different pathways have been reviewed with specific references to environmental changes and nutritional conditions. Mechanism of regulation of animal growth and development, and peptide and intermediates excretion in urine and accumulation have also been reviewed.

Kinetic techniques for body protein turnover, dietary protein requirement, body nitrogen balance and body protein pools are also reviewed by taking different organs of rat or mice. Gene expression during development and carcinogenesis and proteins and amino acid metabolism in cultured hepatocytes have been studied.

Nutritive value of proteins in man by taking egg protein, wheat gluten as protein sources indicated that about 0.2 g/Kg of protein upto maintenance showed a rectilinear relationship between protein intake and nitrogen balance, and if intake is restricted below 0.2 g/Kg, the ingested protein was utilized fully regardless the quality of protein. Amino acid requirements of domestic animals, fishes and rats have been reviewed exhaustively with literature. Studies on Papua, New Guinea highlanders suggest an mechanism of adaptation to low protein intake which maintains their nitrogen balance and healthy physical activity due to their small physique, small obligatory nitrogen loss and reutilization of urea nitrogen for protein synthesis. Factors of protein-energy malnutrition in Ghana, West Africa listed were non-availability of nutrient dense foods for children, infections and infestations, measles and diarrhoea, and prolonged breast-feeding. Self selection

of amino acid and protein and its control mechanism in rats was studied. Dietary protein and aging did not show any correlation. In addition, difference in level and source of protein or energy diets pointed to age related diseases. In a review of studies on protein metabolism in physical training and its physiological role, positive balance of nitrogen during training period was observed. RBC destruction, sports anaemia and lipid metabolism for hard physical exercise have been discussed.

Nutrition and metabolism of methionine and cystein are very well documented and their utility for life is also stressed. Amino acid composition of Japanese food, dietary protein and cholesterol metabolism, casein-phospho peptide and calcium bio-availability are also reviewed. Production of amino acids is also an important chapter in the book.

The book is attractively presented with the information on the subject compiled excellently and expressed in simple language. It is a book that a teacher and a researcher in nutrition should possess.

D.S. WAGLE

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*Yeast Strain Selection*: Chandra J. Panchal (Ed) Pub: Marcel Dekker, 1991; pp:349; Price: US\$ 125/=(US & Canada), \$150/(all other Countries)

The book is an excellent selection of articles by leading yeast scientists and would be useful to a beginner as well as to an advanced person in this area of research.

Chapter 1 gives a list of major yeasts culture collections from where yeast strain can be obtained. Although the list is incomplete with regard to yeast resources in other countries, for a newcomer to yeast, this list is adequate. The Chapter also has some of the technologies which are used in classifying yeast routinely. The 2nd chapter deals with habitat of yeasts which would be especially useful for those looking for specific types of yeasts. Chapter 3 is about yeast strain selection. Although it is not complete, it deals with most of the technologies available to improve yeasts. The Chapters 4 to 6 on selection of yeasts for brewing of wine and for bakers' yeast production, and Chapter 8 for fuel ethanol production have not only the information with regard to the selection of yeast type but also selective techniques that can be used. These chapters will be of immense use for those working in these areas and are upto date. Yeasts are not known for producing extra cellular enzymes. The quantity of enzymes produced by yeast is rather small, although a variety of them is produced. Chapter 7 deals primarily with regard

to some of the enzymes that yeast produces and have industrial application. Chapter 9 is a useful section on the technologies available and the status of improving yeasts other than *Saccharomyces*. A variety of techniques is available for improving non-*Saccharomyces* yeasts but of special importance to those who would like to work with the genetics and biochemistry of yeasts other than *Saccharomyces*.

The problems with regard to heterologous protein production by yeasts have been dealt in Chapter 10 and some of the suggestions in maintenance of foreign genetic material in yeasts have been dealt and remedies have been suggested. The book could be an excellent addition to every library and specially for those who work with yeasts, although it is highly priced.

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*Control of Fish Quality*: by J.J. Connell, Fishing News Books, Blackwell Scientific Publications Ltd., Osney Mead, Oxford, OX2 0EL, England; 1990, pp. 228. Price = £17.95.

The arduous task of protecting the quality of flesh foods is further compounded in the case of fish by a multiplicity of factors like species variations, influence of habitat, seasonal changes in body composition, and above all, the total lack of pre-harvest control over the commodity. Naturally, a major share of scientific literature on fish as food has been claimed by topics related to spoilage, assessment of quality and techniques of preservation.

The book under review, however, stands out among this vast spread of published matter, for more than one reason. The author, Dr. J.J. Connell, is a former Director of Torry Research Station of the U.K., Ministry of Agriculture, Fisheries and Food. Quality of fish has been his main interest during most part of his long association with this institution. As such, Dr. Connell is in a unique position to deal with the various facets of fish preservation, and expectedly, the manner in which he has gone about this task bears the stamp of authority.

The opening chapter is an attempt to define quality, especially the ramifications as it applies to the various agencies concerned - producer, middleman, consumer and of course the Government. Chapter 2 dwells on the intrinsic aspects of quality of fish, viz., species, size, sex, 'condition', composition, biotoxins, pollutants, micro-organisms and parasites. The various ways in which quality deteriorated in

fish and fish products, and how these changes could be retarded or prevented, are discussed in Chapters 3 and 4. In the fifth Chapter, the author elaborates further on microbiological aspects and nutritional implications of spoilage, and also briefly touches upon the role and effect of additives. A carefully selected account of sensory and objective methods available for assessment of quality is provided in Chapter 6. The following three Chapters highlight the essentials of organized quality control programmes, including standards, specifications and codes of practice. Also quoted here are the systems currently in vogue in a few countries. Model standards for selected commodities chosen from India, Ireland and the U.S.A., have been illustrated in the appendices at the end of the text. The latter also contains some useful information on recommended International Standards for microbiological limits for seafoods. Finally, there is a section-wise bibliography pertaining to the preceding Chapters.

The treatment, on the whole, is refreshingly simple and forthright. The sections dealing with commodities are generally structured in three frames: (i) deterioration (ii) causes and effects and (iii) practical remedies. Mere documentation of information has not been the objective as reflected in the style of presentation. The writing, for the author is primarily a means of direct communication to the actual user. Naturally, the stress is on practical problems faced during handling, processing, storage and distribution. Causes of deterioration are analysed, quality determinants identified and remedies suggested, bringing into play the deep understanding and vast experience of the author.

The volume has run into the third edition within a span of fifteen years, proving its continued validity and relevance. There is visible improvement over the previous editions in both form and content. The numerous illustrations serve to hold the interest alive for the reader, apart from providing useful support to the text.

R. B. NAIR  
CFTRI, MYSORE.

## AFST(I) News

In the series on technical lectures on various topics in Food Science and Technology, Mr. R.B. Nair, Scientist, Animal Products Technology Division, CFTRI, Mysore, spoke on 'Technological Advances in Meat Production and Processing' at CFTRI on 24th October 1991, followed by a technical discussion.

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