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JOURNAL OF FOOD SCIENCE AND TECHNOLOGY

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RESEARCH PAPERS

Studies on Swelling and Hydration of Paddy by Hot Soaking

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Swelling and hydration of grains during hot soaking of paddy (or rough rice) have been studied as a function of time and temperature of soaking with four high-yielding varieties of Indian paddy. Temperature range covered was 50°-80°C and time was varied upto 12 hr. Swelling of grains is limited to a maximum of about 0.09 part per part initial volume of raw paddy upto moisture absorptions in the range of 0.425-0.500 g/g dry basis on soaking below galatinization temperature (around 65°C) but increases sharply with rapid hydration and bursting of grains on soaking at higher temperatures. The apparent degree of parboiling during soaking has been measured by visual observation of "white core" in the soaked grains after drying. It was observed that hot soaking of paddy at a temperature around 65°C gives considerable "degree of parboiling" of about 90% with a moisture content in the range of 0.50-0.55 g/g dry basis and without excessive swelling of grains.

Although sufficient work on hydration characteristics of paddy has been reported in the literature, proper attention has not been given to study the optimum processing factors for parboiling by hot soaking alone which is still practiced in home scale in India. The studies reported so far are by Bhattacharya *et al.*[1] and Ali[2]. According to Bhattacharya *et al.*,[1] parboiled rice can be obtained by hot soaking alone with milling quality and cooking quality of the rice approaching those of normal steam-parboiled rice, although appreciable gelatinization cannot be achieved without extensive leaching and deformity of the grain. Ali[2] studied some of the qualities of hot-soaked parboiled rice and corroborated the results of Bhattacharya *et al.*[1] So it appears that there is further scope to study parboiling of paddy by hot soaking only, and the extent of swelling and hydration of grains will be the major processing parameter which need to be investigated for this purpose. Some studies[2-5] have been reported on swelling during hot soaking, which, however, did not put forward any relation of the swelling of grains with the extent of hydration. The present investigation, has therefore, been undertaken to study swelling and hydration during soaking of some varieties of Indian paddy in water as a function of time and temperature of soaking. The objective was to study further the possibility of parboiling by mere hot soaking and to optimise different process variables in hot soaking method.

Materials and Methods

Paddy: All the varieties selected for the present investigation are high-yielding types and locally grown. These are *Jaya*, *Padma*, *Bala* and *Pankaj*. The samples for study were taken from stocks stored for six months or more to avoid variation in hydration characteristics due to physico-chemical changes that take place in freshly harvested paddy during storage. The samples were cleaned from chaff, dust, stones and other impurities in a paddy cleaner and, if necessary, with water.

Methods: The initial moisture content was determined, in duplicate, from the loss in weight of about 5 g whole paddy grain at 130±2°C in an air oven for 16 hr[6]. All the moisture contents are expressed as g/g dry basis.

Soaking of paddy samples (20 g each), contained in wire-gauze baskets, was done in a constant (±0.5°C) temperature water bath. After soaking for a given time, the samples were quickly withdrawn from water, superficially dried with blotting paper and immediately weighed to determine the moisture content of the soaked grains by calculation.

For determination of swelling, the true volume of soaked paddy samples was measured by the liquid displacement method according to Ghose *et al*[4,5]. The soaked sample immediately after weighing (for evaluation of moisture gain) was quickly transferred into a 50 ml burette containing water upto the 25 ml mark. The displaced volume of water indicated by a rise in the water level, read quickly, was taken as the true volume of the sample. The initial volume of the raw paddy (20 g) was also determined by the same method. The volume gain of soaked paddy was calculated from the above and expressed as swelling in part per part initial volume of raw paddy. Experiments were carried out at temperatures 50°, 60°, 65°, 70° and 80°C and time intervals ranging from 10 min to 12 hr.

The extent of parboiling of soaked grains during hot soaking was measured indirectly by visual observation

of "white core" or "opaque spot" present in the central region of the endosperm. For this a definite number of soaked paddy grains, after drying in shade, was cut into two halves and each half was examined for "opaque spot". The percentage of grains without opaque spots was calculated and reported as "degree of parboiling". However, whether the translucent kernels are fully parboiled or not is a matter to be studied by other properties of rice such as alkali degradation, water uptake, etc.[7,8] Therefore the degree of parboiling reported here may be called the "apparent degree of parboiling".

Results and Discussion

Swelling of grains: Fig. 1 shows the swelling of paddy grains as a function of time and temperature of soaking for the variety *Padma*. Two distinct trends are noted: (*i*) the rate of swelling of grain increases with increase of temperature, but at temperatures of 65°C and below, the rate decreases with time and ultimately becomes almost constant after prolonged soaking. (*ii*) at temperatures 70°C and above, the rate becomes much faster and goes on increasing sharply after a certain interval of time. Similar observation was noted in the other three varieties of paddy, which is also in agreement with that of Ghose *et al.*[4,5].

The curves of swelling are very much similar in nature with those of moisture content as a function of time and temperature of soaking presented by us in the earlier studies on soaking characteristics of paddy[9,10] as well as seen in the earlier literature[1,4]. This indicates that a direct relation exists between swelling and moisture content of paddy. Fig. 2 shows such a relation for the variety *Padma*. The moisture content data have been taken from the previous report[9]. The straight line plot embracing the data at all temperatures shows a sharp change in slope after a break at a moisture content of about 0.50 g/g dry basis. The break was observed as a common phenomenon in all the varieties of paddy in the moisture content range of 0.425-0.500 g/g dry basis, with corresponding values of swelling in the range of 0.08-0.11 part/part initial volume. Similar correlations were reported by Ghose *et al.*[4,5] and Ali[2,3] but their observations of the break in the swelling line were not elucidated in the light of hydration of starch granules.

The present study indicates that swelling of grains is not very much perceptible upto a grain moisture content in the above range corresponding to soaking temperatures of 60°-65°C and below, but is very rapid as the moisture level increased when soaking is done at higher temperatures. The maximum swelling of paddy observed in the lower range, viz. about 0.09, is in agreement with the value obtained by Bhattacharya *et al*[11] in their studies on changes in physical properties of rice and paddy with changing moisture content. These authors explained the phenomenon in the light of paradoxical behaviour of paddy of increasing in density with increase in moisture content. This, they explained, is due to the air space inside the husk

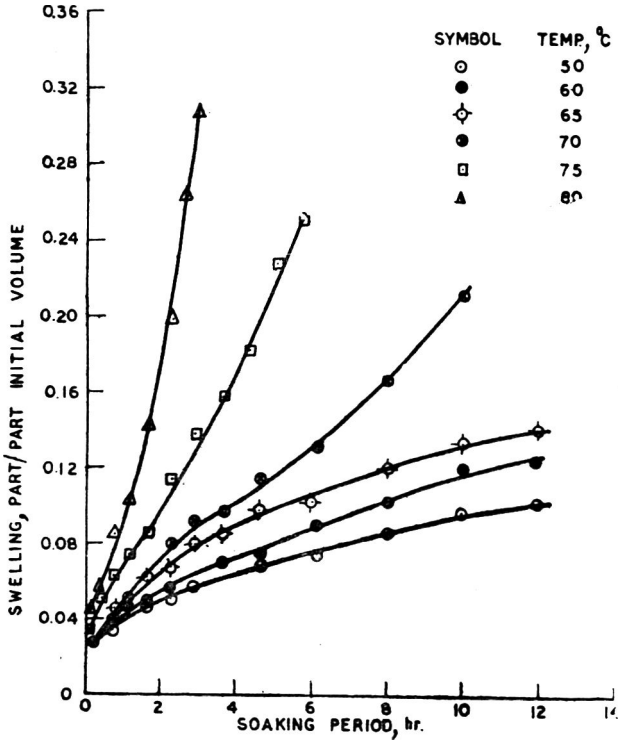

Fig. 1. Swelling of grain as a function of time and temperature of soaking. Variety: *Padma*.

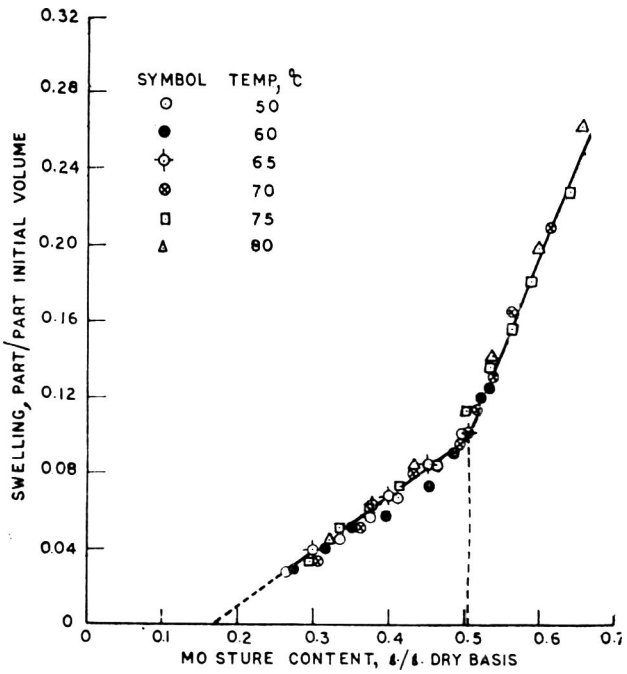

Fig. 2. Correlation between moisture content and swelling. Variety: *Padma*.

which enables the kernel to hydrate and expand without appreciable change in the external grain volume. Hence the very low swelling of the grain on soaking. However, further increase in moisture content above 0.50 g/g dry basis, obtained on soaking at a temperature higher than 60°-65°C, rendered the swelling perceptible, as reflected in the break in the swelling line (Fig. 2). This was evidently related to the splitting of husk that occurred in all the varieties at temperatures of 65°C and above and in the moisture content range 0.50-0.60 g/g dry basis, as observed by us[9,10] as well as earlier[1]. Evidently once the expanding internal kernel fills the husk container, any further expansion caused by further hydration forces the husk to split, and thereafter swelling proceeds unhindered. So the splitting of husk is the crucial point of departure, and thereby correlates the break with swelling and moisture content of paddy.

A further corroboration of this result is obtained from the earlier analysis of soaking data by the authors[9,10]. The analysis was based on the diffusion equation proposed by Ghose *et al*,[4,5] viz. $x - x_o = k_m \sqrt{\theta}$, where $k_m = \frac{2}{\sqrt{\pi}} (x_s - x_o) \left(\frac{S}{V} \right) \sqrt{D_m}$ is a temperature dependent function. The break in the Arrhenius plot of log k_m *vs* the reciprocal of absolute temperature, around 62°-69°C, took place in the vicinity of gelatinization temperature of starch and indicated the predominant existence of rapid hydration process due to gelatinization with higher activation energy (25.0-31.7 Kcal/mole). A typical plot is shown in Fig. 3. So the onset of rapid hydration due to splitting which takes place above

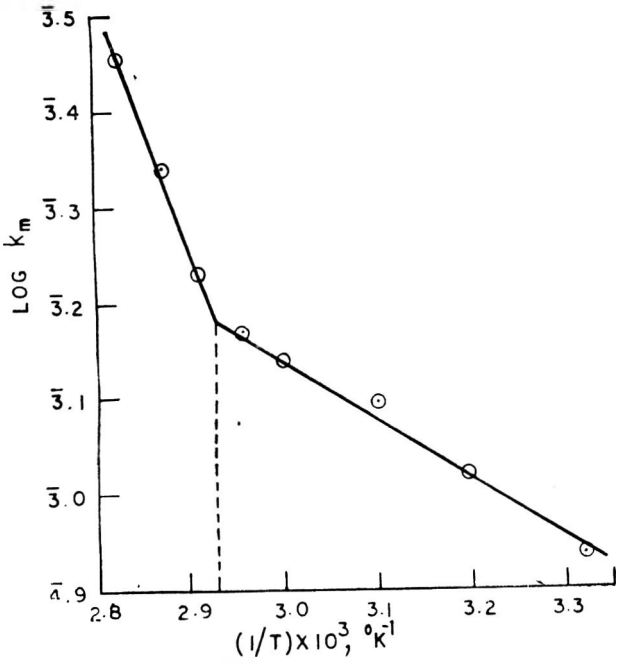

Fig. 3. Log k as a function of the reciprocal of the absolute temparature. Variety: *Pankaj*.

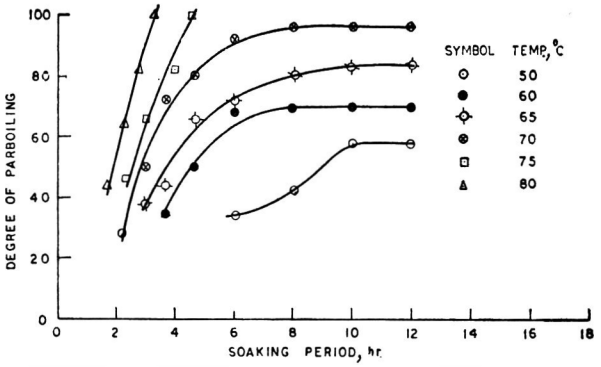

Fig. 4 Degree of parboiling of grains as a function of time and temperature of soaking. Variety: *Padma*

gelatinization temperature thereby correlates the gelatinization temperature, swelling, splitting and moisture absorption.

Degree of parboiling: The apparent degree of parboiling measured as percentage of grains without opaque spot is plotted in Fig. 4 as a function of time and temperature of soaking for the variety *Padma*, in the temperature range 50°-80°C. Here also two distinct trends similar in nature to the swelling curves are noted. (*i*) The apparent degree of parboiling increases with increase in temperature, but upto 70°C rate of increase is comparatively slow, and it reaches a constant value and does not attain 100 per cent. (*ii*) Above 70°C the rate is very much rapid. This is almost true for the other varieties. However, at 75°C the apparent degree of parboiling becomes 100 per cent within a very short duration of soaking in *Jaya* and *Padma*, but not in *Bala* and *Pankaj*, where it reaches 80-90 per cent. At 80°C a 100 per cent value is obtained within a very short duration of soaking for all the varieties.

Fig. 5 shows liner correlation between the apparent degree of parboiling and moisture content for the two varieties *Padma* and *Pankaj*. The data of the *Padma* variety in the range 50°-70°C are fitting to one straight line, whereas the data at 75° and 80°C yield different

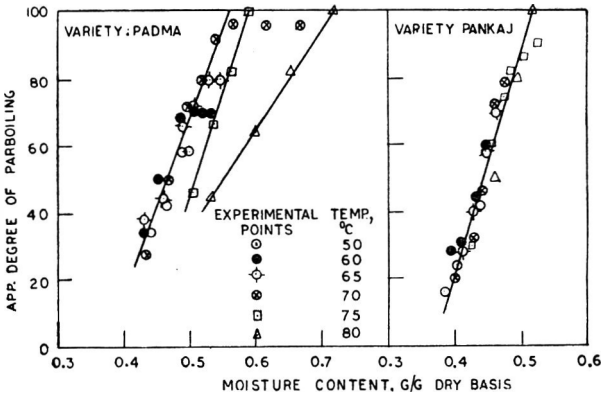

Fig. 5 Correlation between apparent degree of parboiling and moisture content of paddy grains

lines. Similar is the case with the variety *Jaya*, but in *Bala* and *Pankaj* varieties, all the data in the range 50°-80°C, yield one line only. There are some significant deviations in the correlations. The data on apparent degree of parboiling which reach a constant value at 70°C and below in case of *Padma* as shown in Fig. 4 have deviated from the line. It means that in this temperature region the grains have attained almost a finite amount of moisture[9,10] which, as well as the temperature, might have been insufficient for complete gelatinization of starch granules. The data at 75° and 80°C each showing separate linear plots may also be considered as deviations in the sense that these data were collected during the stages of "rapid hydration" at a temperature much higher than the gelatinization temperature. In case of *Bala* and *Pankaj* data collected at 75° and 80°C indicated "rapid hydration" at much later stages of soaking, showing deviations in the linear plot.

The important finding of the relation between swelling apparent degree of parboiling and moisture content is that a moisture content of 0.50-0.55 g/g dry basis can effect a considerable "degree of parboiling" of about 90 per cent which may be adequate for the soaked grain to be processed into finished rice provided breakage is minimum during milling. This is achieved without undesirable swelling and hydration of grains leading to bursting and around a temperature of 65°C which is in the range of gelatinization temperature of rice starch. The result evidently indicates the possibility of adopting mere hot soaking (without steaming) as a means of parboiling and thereby corroborates the findings of Bhattacharya *et al*[1]. and Ali[2].

Acknowledgement

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Nomenclature

D_m diffusion coefficient, cm^2/sec

S exposed surface area of a solid (paddy grain), cm^2

V volume of a solid (paddy grain), cm^3

x_o initial, uniform moisture content, g/g dry basis

x_s moisture content at the bounding surface, g/g dry basis.

$\bar{x}$ average moisture content, g/g dry basis

θ time, sec.

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Factors Influencing the Malting Quality of Indian Barley

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Two commercial varieties of Indian barley, 'C 138' and 'C 164' were investigated for malting as influenced by germination at 20°C for 3, 5, 7 and 9, days with and without the application of gibberellic acid. The diastatic power, alpha-amylase and proteolytic activities of the malts of both the varieties increased markedly by gibberellic acid. The effect on 'C 138' variety was more pronounced in case of 3-day germinated malts. The enzyme activity more or less levelled off in 7-and 9-day germinated malts.

The Institute of Brewing, London, conducted investigations into the malting quality of Indian barley for brewing during 1936-38 under a project of the Imperial (now Indian) Council of Agricultural Research[1]. Very little work has been reported since then regarding the potential of Indian barley for malting. In the intervening years, the technology of malting has made considerable advances leading to several effective malting systems[2]. As information on factors such as variety, environment, duration of germination, and response to gibberellic acid (GA) in relation to the overall malting quality of indigenous 6-rowed barley was lacking, this investigation was carried out. The results describe the beneficial effect of using gibberellic acid for producing malt of improved quality.

Materials and Methods

Two commercial varieties of barley 'C 138' and 'C 164', crop, of 1972 were obtained from local sources. The variety 'C 138' is grown commercially in the Punjab state and 'C 164' variety in the Haryana state. The samples were cleaned and stored in air tight containers and withdrawn for various tests.

Hectoliter weight: This was determined on the Griener's balance and expressed as kg/100 litres.

1000-*kernel weight:* The kernels (1000) were counted and weighed on an analytical balance.

Germination capacity: This was determined according to the method described by Cook[3]. Kernels (200) were steeped in hydrogen peroxide solution (0.7 per cent) at 20°C. At 24 hr intervals, the number of kernels which showed germination were counted and removed.

Water-sensitivity: This was determined by observing the germination of 100 kernels of each variety on filter papers in petridishes wetted with 4 ml and 8 ml of water, respectively[4]. The petridishes were kept in the dark at 20°C. The difference in the germination at the lower and higher moisture environment in the petridishes served as an index of water-sensitivity of the sample.

Husk: This was determined according to the procedure described by Cook[3]. A weighed sample of 1000 kernels of barley with the husk was digested at room temperature with sulphuric acid (50 per cent) to remove the husk.

Starch: This was determined polarimetrically according to the procedure described by Fraser *et al*[5]., Fraser and Holmes[6] and Fraser and Hoodless[7].

Moisture, ash and protein contents were determined according to the AOAC methods[8].

Malting procedure: Weighed sample (1 kg) of each variety of barley was steeped in distilled water at 20°C. The water was changed every two hours for the first twelve hours of steeping and afterwards at six hourly intervals until the average moisture content was about 42 to 45 per cent. During the final stage of steeping, gibberellic acid (GA) was added to the system allowing a pick up of 0.1 ppm GA by the steeped barley. The excess water was drained prior to germination in wooden boxes having wire mesh screen bottoms to facilitate aeration. The boxes were placed in the Harrington daylight germinator, maintained at 20°C and 95 per cent RH. Test samples of each lot were germinated for 3, 5, 7 and 9 days, respectively. The malts were dried at 45°C in an air flow drier for 24 hr to reduce the moisture content from 42 to 6-8 per cent. The withered rootlets were gently brushed off and malt weighed. The yield is expressed on dry basis. The malts were preserved in air tight containers for evaluation of quality.

Acrospire length: This was determined according to the method of Rahim and Bains[9] by staining with cupric acetate reagent. The length of stained acrospire was measured and expressed in mm.

Diastatic power (DP)*:* This was determined according to the AOAC method[10]. The results are expressed as °*L*.

Alpha-amylase activity: This was determined according to the ICC method described by Perten[11] and values expressed as SKB units/100 g.

Proteolytic activity: This was determined according to the procedure described by Ayre and Anderson[12]. The results are expressed as mg N/100 g.

Yield of extract and wort quality: The mashing of test malts was done according to theAOAC[10] procedure and yield of extract was determined by the EBC[13] method. Observations were made regarding the conversion time (min), colour, speed of filtration and soluble nitrogen contents to find out the degree of modification expressed as Kolbach index(soluble N/malt N×100) of the malt.

The results are expressed on moisture free basis.

Results and Discussion

The results of test weights, ash, protein, husk and starch contents of the varieties of barley obtained from two different locations are given in Table 1 and of germination capacity and water-sensitivity in Table 2.

The variety, 'C-164', showed higher hectoliter and 1000-kernel weights than the variety 'C 138'. The kernels of 'C 164' variety looked plumper as also attested by its higher test weights (45.7-47.7 g) compared with those of 'C 138'. Standridge *et al.*[14] reported a value of 33.2 g for six rowed barley which approximated the 1000-kernel weight of 'C 138'. The ash (2.4 and 3.0 per cent) and protein (12.3 and 12 6 per cent) contents, of 'C 138' variety of barley grown in two locations were higher than those of 'C 164' variety of barley. The higher protein content of 'C 138' variety would be disadvantageous for brewing compared with 'C 164' variety which contained less protein. Kleber[15] malted high nitrogen barley and reported greater loss of malting due to greater root growth which was ascribed to the higher nitrogen contents of the cultivars. However, there was more starch in 'C 164' variety as compared to that of 'C 138' variety.

The range in germination capacity and the water-sensitivity of 'C 164' variety (Table 2) were: 96–99 per cent, and 9–1 per cent compared with 92–95 per cent and 21–8 per cent, respectively of 'C 138' variety. Ward and Briggs[16] reported a germination capacity of 97 per cent and water-sensitivity of 4 per cent, in their studies of barley cultivars. The variety 'C 164' is superior to that of 'C 138', the latter being more water-sensitive.

Yield of malt: Considerably higher yields of malts were obtained in respect of GA treatment of 'C 164' variety (87.4 and 90.9 per cent) compared with 80.0 and 88.4 per cent of the malt of 'C 138,' when germinated for 3 and 9 days respectively. (Table 3). Rinke[17] reported 78.5 to 82.5 per cent yield of malt which is less than the yield of malt obtained from both the varieties of Indian barley.

The hectoliter and 1000-kernel weights of the malts of both the varieties were lower than their initial values which further decreased with the extension of germination to 9 days. That was attributed to the greater malting losses on prolonged germination as was also observed by Ward and Briggs[16]. There was definite indication of accelerated malting caused by GA as judged by the faster growth of acrospires compared with those of control samples. The acrospire length, an indication of malting efficiency, was increased by GA treatment specially in case of 'C 164' variety malted for 5 and 7 days. This trend, in case of 'C 138' malt was revealed when the germination was for 7 and 9 days.

The values for diastatic power of 'C 138' malt germinated for 3 to 7 days without GA were 76° and 125°L respectively compared with corresponding values of 114° and 166°L respectively when germinated in the presence of GA (Table 4). These values were considerably higher than corresponding diastatic values of 'C 164 'variety of barley. Sandegren and Beling[18] reported a 20 per cent rise in the diastatic activity of malt using GA during malting. Fiser *et al.*[19] reported maximum diastatic activity after the germination for seven days using GA during malting. The higher activity of malts of 'C 138' barley may be attributed to the higher protein content.

The alpha-amylase activity was found higher in malts from 'C 138' variety than the malts of 'C 164' variety. The activity increased considerably in case of GA treatment of the grain. Sandegren and Beling[18] reported

TABLE 1. QUALITY CHARACTERISTICS OF BARLEY VARIETIES

Variety	Location	Hectoliter wt. (kg)	1000-kernel wt. (g)	Ash (%)	Protein (%)	Husk (%)	Starch (%)
C 138	I	60.0	33.5	2.4	12.6	21.1	58.0
	II	56.3	29.8	3.0	12.3	20.9	59.4
C 164	I	62.0	47.7	2.3	10.4	24.1	57.5
	II	59.2	45.7	2.5	10.1	23.3	60.4

TABLE 2. GERMINATION CAPACITY AND WATER-SENSITIVITY OF BARLEY VARIETIES

Variety	Location	Germination (%)	Water-sensitivity (%)
C 138	I	92	21
	II	95	8
C 164	I	96	9
	II	99	1

TABLE 3. EFFECT OF GIBBERELLIC ACID (GA) AND GERMINATION TIME ON THE YIELD AND CHARACTERISTICS OF MALTS

Variety	Germination (days)	Treatment	Hectoliter wt. (kg)		1000-kernel wt. (g)		Acrospire length (cm)		Malt yield (%)	
C 138			I	II	I	II	I	II	I	II
	3	Control	42.4	48.8	27.4	27.1	0.68	0.65	84.7	87.2
		(+) GA	43.4	46.9	27.8	28.0	0.69	0.65	85.9	88.4
	5	Control	39.5	45.5	26.3	26.0	0.81	0.70	80.9	83.7
		(+) GA	40.8	44.7	26.6	26.5	0.81	0.71	83.1	85.2
	7	Control	37.8	41.0	23.6	24.6	0.84	0.71	76.0	81.4
		(+) GA	36.1	41.8	25.4	25.6	1.04	0.77	79.0	82.1
	9	Control	37.3	40.7	24.4	24.7	0.87	0.76	75.9	80.6
		(+) GA	34.5	37.4	25.9	25.1	1.09	0.94	80.0	80.6
C 164	3	Control	49.5	48.9	43.6	42.9	0.45	0.46	88.3	88.8
		(+) GA	49.7	48.8	45.4	43.0	0.45	0.51	90.5	90.9
	5	Control	49.4	47.7	43.8	42.9	0.47	0.51	88.0	88.4
		(+) GA	47.7	47.6	42.4	42.8	0.59	0.61	88.9	88.0
	7	Control	46.0	45.9	40.3	40.7	0.55	0.53	87.4	87.3
		(+) GA	45.4	46.0	40.9	41.0	0.64	0.67	88.0	89.2
	9	Control	—	44.3	—	39.4	—	0.71	—	84.8
		(+) GA	—	41.7	—	39.8	—	0.96	—	87.4

TABLE 4. EFFECT OF GIBBERELLIC ACID AND GERMINATION TIME ON THE ENZYME ACTIVITY OF MALTS

Variety	Germination (days)	Treatment	Diastatic power (°L)		Alpha-amylase (SKB/100g)		Proteolytic activity (mg N/100g)	
			I	II	I	II	I	II
C 138	3	Control	80	78	39.3	27.8	139.6	139.7
		(+) GA	114	129	68.7	72.0	203.9	230.6
	5	Control	116	106	42.3	30.2	146.8	155.1
		(+) GA	127	151	79.8	82.6	210.6	239.0
	7	Control	125	115	49.0	30.3	174.5	173.5
		(+) GA	125	166	127.2	114.6	186.9	172.6
	9	Control	100	103	39.6	37.5	172.7	163.5
		(+) GA	118	165	114.9	118.4	221.9	266.8
C 164	3	Control	70	81	21.0	22.2	133.4	94.8
		(+) GA	97	99	36.2	66.7	186.2	170.3
	5	Control	76	91	23.7	22.4	124.8	108.8
		(+) GA	99	116	50.6	67.1	187.0	180.5
	7	Control	76	94	28.7	28.9	140.5	126.0
		(+) GA	99	119	56.4	71.4	234.0	149.5
	9	Control	—	91	—	30.5	—	110.5
		(+) GA	—	119	—	82.3	—	180.7

TABLE 5. EFFECT OF GIBBERELLIC ACID AND GERMINATION TIME ON THE YIELD OF COLD AND HOT WATER EXTRACTS OF MALTS

Germination (days)	Treatment	Extract yield (%) Cold water C 138 I	C 138 II	C 164 I	C 164 II	Hot water C 138 I	C 138 II	C 164 I	C 164 II
3	Control	19.2	18.7	16.6	17.3	71.0	73.3	76.6	76.9
	(+) GA	27.8	25.1	21.4	24.1	72.6	71.9	78.5	—
5	Control	21.4	20.0	17.2	20.3	70.6	72.3	76.4	75.3
	(+) GA	33.1	30.0	24.3	29.3	74.7	76.3	79.4	78.9
7	Control	22.9	21.1	18.2	21.1	71.3	74.0	77.2	77.2
	(+) GA	36.1	34.9	27.7	30.7	74.9	76.6	79.3	78.8
9	Control	24.8	21.1	—	21.9	70.4	72.2	—	76.5
	(+) GA	37.4	38.9	—	33.2	72.3	72.2	—	76.6

GA during malting. The alpha-amylase activity of the malts of 'C 138' barley ranged from 27.8 to 49.0 SKB units/100g (control) compared with 68.7 to 127.2 units as a result of using GA.

The proteolytic activity of malts of 'C 164' barley was less than that of 'C 138' malts. The values for 'C 138' ranged from 94.8 to 140.5 mg N (control) and from 149.5 to 234.0 mg N (GA) of malt for 'C138', respectively. Weith[20] reported that the addition of GA increased the formation of proteolytic enzymes, whereas Sandegren and Beling[18] reported 10 to 15 per cent increase in the proteolytic activity using GA in malting. In the present investigation the activity was found to increase upto seventh day, but decreased on the nineth day of germination in the control samples. There was a decrease in proteolytic activity on the seventh day of germination in the GA treated barley.

Extract yields: The GA treated malts of both the varieties showed uniformly higher cold water extracts than the controls. This effect was more pronounced than that of the length of germination. The hot water extract yields were also enhanced by the GA application compared with the control (Table 5). The yields of the extracts were considerably higher in case of 'C 164' variety than that of 'C 138' variety.

There was invariably more soluble nitrogen in the GA malts than the controls. By suitably adjusting the dosage of GA, it should be possible to obtain malts of desired degree of modification for brewing. Ward and Briggs[16], Paul and Denade[21], and Pomeranz and Shandr[22] obtained similarly higher yields of extract in GA treated malts. Ward and Briggs[16] reported decreased hot water extract in the five-day germinated malts with and without GA treatment. The maximum yield of

TABLE 6. EFFECT OF GIBBERELLIC ACID AND GERMINATION TIME ON THE QUALITY OF WORTS

Germination (days)	Treatment	Conversion time (min) C 138 I	C 138 II	C 164 I	C 164 II	Wort colour (EBC units) C 138 I	C 138 II	C 164 I	C 164 II	Nitrogen (%) C 138 I	C 138 II	C 164 I	C 164 II
3	Control	15	15	15	18	2.75	2.00	5.25	2.75	0.63	0.63	0.56	0.51
	(+) GA	10	10	12	9	2.75	4.25	4.25	3.25	0.77	0.87	0.77	0.69
5	Control	15	15	12	15	3.75	2.25	3.75	3.25	0.66	0.70	0.54	0.53
	(+) GA	5	6	10	8	4.75	4.75	3.50	3.25	0.78	0.89	0.78	0.71
7	Control	15	13	12	13	3.75	2.50	4.25	3.75	0.73	0.71	0.60	0.55
	(+) GA	<5	6	8	<5	5.75	6.75	3.50	3.75	0.76	0.71	0.84	0.66
9	Control	15	12	—	12	4.00	2.75	—	3.75	0.72	0.67	—	0.53
	(+) GA	6	<5	—	<5	6.75	8.25	—	5.00	0.81	1.00	—	0.72

TABLE 7. EFFECT OF GIBBERELLIC ACID AND GERMINATION TIME ON THE MODIFICATION (KOLBACH INDEX) OF MALTS

Germination (days)	Treatment	Soluble N as % of total nitrogen of malt			
		C 138		C 164	
		I	II	I	II
3	Control	36.5	36.2	35.4	33.1
	(+) GA	—	45.9	44.5	45.8
5	Control	38.3	40.7	36.0	32.3
	(+) GA	38.7	47.2	48.4	47.3
7	Control	42.4	42.7	41.0	33.6
	(+) GA	—	—	53.2	40.6
9	Control	41.6	36.1	—	33.0
	(+) GA	42.6	50.9	—	47.2

extract was given by the 5-day germinated sample treated with GA whereas 7–day germination was essential for the control to reach a same increase in the yield of extract.

Wort quality: The conversion time of 'C 138' malts ranged between 12 and 15 min which was reduced to between 5 and 10 min when GA was used in the malting process (Table 6). Under similar conditions of malting and mashing, 'C 164' malts (control) showed higher range of conversion times i.e. 12 to 18 min compared with 5 to 10 min for GA malts.

The worts of 'C 164' malts contained lower percentage of soluble nitrogen than those of 'C 138'. The extent of modification as shown by the Kolbach index values[23] was greater in malts of 'C 138' variety than those of 'C 164' malts (Table 7). The values ranged from 36.5 to 42.7 per cent of the malts for 'C 138' without GA treatment. The degree of modification was notably increased by applying GA (Table 7). Schilfarth *et al.*[23] reported that the higher Kolbach index was associated with shorter germination and preferred malts of high Kolbach index values due to the higher contents of amylolytic and proteolytic enzymes.

From the foregoing results, it is seen that the application of GA considerably improved the quality of malts of both the indigenous varieties of barley, which by themselves had lower amylase and proteolytic activities when germinated for same periods. The time of germination might be reduced with advantage during malt production by using judicious amount of GA.

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Studies on Osmotic Dehydration of Banana

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A method of partial dehydration of fruits (bananas) by osmosis in sugar syrup of 70% concentration is described. The fruit is reduced to about 50% of its original weight by the process of osmosis, after which it is drained, washed and vacuum dried which gives best results. Flavour, colour, appearance and texture attributes are maximally retained in osmotically dried products. The dehydrated products can be preserved up to one year or more, depending upon the packaging material used and the storage conditions.

Most dried fruits are produced today by much the same methods used for hundreds of years. The fresh fruit is placed on trays in the sun, and the action of heat and light produces a commodity that has been relished by man in many areas of the world. Artificially dried fruit by use of conventional tray dryers or vacuum dryers are wholesome, nutritious and palatable in its own right but it has not in general found popular acceptance because it does not have the flavour, colour and texture of the original fruit even after rehydration. Freeze drying of fruits results in good quality of dried fruits and of long storage stability but cost of processing is very high. Hence a new method of drying on the basis of osmosis, in which partial dehydration of fruits either whole or in sliced form is brought about by dipping them in sugar solution and then vacuum drying[1,2]. This is claimed to result in products with better flavour than freeze dried fruits at comparatively less cost than freeze drying[1,2].

A continuing interest in dried banana products in tropics has led to work on dehydration by different methods. Bananas have been sun dried, drum dried, spray dried and dried in a cabinet dryer[3]. Apparently, the banana powder produced by drum drying has attracted more attention from commercial food processors than have other dried banana products[4]. However, a high quality dried banana, whole or in the form of chips or slices, would command even greater commercial appeal for the simple reason that the powder will have less culinary applicability than the product which has retained its form. Air and vacuum drying unfortunately have failed consistently in giving a good banana product[5]. It was, therefore, thought of applying for banana the osmotic drying[1,6,7] which, has proved its merit as a good substitute for the excessively expensive freeze drying[8].

In the experiments reported here, optimum conditions for osmotic dehydration of banana have been established, quality changes during storage have been determined and commercial feasibility of the process has been indicated based on the results of semi-pilot plant scale operations. The paper also contains a discussion on design and fabrication aspects of some of the equipment used in the scaled up process.

Materials and Methods

Table ripe cavendish bananas were used for the experiment. Cane sugar used was of commercial grade (grade C) and sodium meta bisulphite is of technical grade. Polycell (700 gauge) polyethylene (700 gauge) and aluminium laminate pouches (paper/polyethylene/aluminium/polyethylene (2.4/1.0/0.5/1.5 mil) were used as packaging materials for storage studies.

Moisture content: Moisture content of fresh and dried bananas was determined using AOAC method[9]. Samples (2 g), in triplicate, were dried in a vacuum oven at 70 to 75°C under pressure of 29 in. Hg for 8 hr. The loss in weight was calculated as per cent moisture.

Rehydration: Rehydration of dehydrated material was done by boiling on a hot plate 10 g of sample in a covered beaker (600 ml capacity) with 150 ml of distilled water upto 3 and 5 min. The contents were transferred to a 75 mm Buchner funnel fitted with Whatman filter paper No. 1. Gentle suction was applied till the drip from funnel stopped. Samples were then removed from the funnel and weighed. Rehydration of sample was expressed in terms of per cent of water in the rehydrated material using results from ten samples.

Sensory evaluation: Sensory evaluation of osmotically dehydrated bananas was carried out by a taste panel consisting of 12 members from the scientific staff using 9 point hedonic scale. Scoring was done for appearance, odour, flavour, texture and overall acceptance. Average scores of 4 tests are given.

Colour: Colour of banana slices was measured on a reflectance accessory attached to a Beckman DB spectrophotometer using duplicate samples contained in cells. The samples were not removed from their individual cells during the whole storage period; the cells were oriented in the same position for each reading by a calibration mark. In this manner, the same pieces of fruit slices were used for colour measurement over the entire storage period.

Preparation of material for dehydration: Ripened bananas were peeled and cut by hand longitudinally or into chips of 8-10 mm thickness. The weighed cut slices, or chips were washed and subjected to sulphiting by dipping in 0.25 per cent sodium metabisulphite solution for 15 min, drained and subjected to osmosis under varied conditions.

Osmosis in dry sugar and sugar syrup: To study the effect of dry sugar for osmosis, known amount of the sulphited slices were placed in aluminium trays. Weighed quantity of sugar was spread on them.

Rates of loss of weight under different ratio of sugar: Fruit (1:1 to 3:1) were recorded. To study the effect of sugar syrup and its quantity and concentration on the osmotic rate weighed quantity of banana slices were dipped in syrups of 60, 70, 80 and 82 strength (W/W). Percentage losses in weight of bananas were recorded after 1, 2, 3 and 4 hr of dipping.

To study the effect of quantity of sugar syrup on the osmotic rate banana slices were dipped in sugar syrup in fruit to syrup ratio of 1:1 to 1:3.

The effect of temperature on the osmotic rate: For studying the effect of temperature on osmosis, banana slices were dipped in 70 per cent sugar syrup for $3\frac{1}{2}$ hr in 4 separate containers maintained at 60, 50, 40 and 27°C, using electrical hot plate having electronic thermostat control for regulation of temperature during heating. Percentage loss in weight of banana slices after every 30 min was recorded.

The effect of agitation on the osmotic rate: Sugar syrup maintained at the above temperatures but kept agitating by means of a small stirrer were used in these experiments. Agitation will provide a uniform distribution of water removed, throughout the syrup and hence better results can be expected.

Reuse of sugar syrup after osmosis: To reuse the sugar syrup after osmosis, it was filtered using glass wool in a funnel, concentrated by evaporation, mixed with activated charcoal to remove brown colour and filtered again.

Final drying: Banana slices after osmosis were washed in running tap water and spread on aluminium trays surfaced with aluminium foil to prevent sticking. The trays were loaded either in standard tray dryer or vacuum dryer for further drying. Temperature of the dryer was maintained at 60°C and drying was carried out to a moisture level of 3 to 5 per cent. Only vacuum drying of samples was carried out for comparative purpose. In order to establish optimum conditions for discolouration, samples of osmotically dried and conventionally vacuum dried bananas were moistened and stored at elevated temperatures. Two sets of storage conditions were used at 32°C at 10 per cent moisture and 37°C at 20 per cent moisture. For storage stability tests, dehydrated samples were packed in polycell, polyethylene bags (700 gauge) and aluminium laminate pouches (paper/polyethylene/aluminium/polyethylene) (2.4/1.0 0.5/1.5 mil) and stored for different periods at room temperature and then evaluated for quality at intervals.

Results and Discussion

Dehydration by osmosis is essentially brought about by a process of maintaining a proper gradient in solute concentration which in this case is sucrose although glucose could also be an effective osmotic agent.

Osmosis by dry sugar and syrup: Maximum rate of osmosis may be expected with dry sugar (sucrose), the ratio of sugar to fruit affecting the rate to a certain extent. At the optimum ratio, all the sugar gets dissolved in the moisture abstracted from the fruits. It is seen

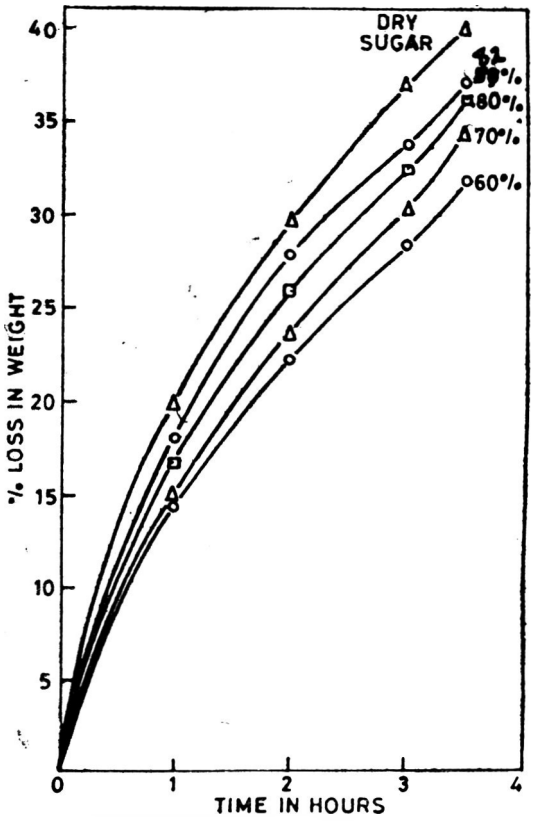


Fig. 1*a* Effect of sugar concentration on rate of osmotic dehydration of banana slices in syrup

TABLE 1. RATE OF LOSS OF WEIGHT UNDER DIFFERENT SUGAR TO FRUIT RATIO

Sugar : fruit ratio	Percent loss in wt during 2 hr	4 hr	20 hr	24 hr	Syrup condition
1 : 1	22.7	31.1	43.8	47.9	Sugar totally dissolved
2 : 1	28.7	40.3	55.4	56.0	Undissolved sugar
3 : 1	25.0	36.7	49.7	52.8	Undissolved sugar

from Fig. 1*a* that the extent and rate of osmosis is greater when dry sucrose powder is used than when the syrups are used. Table I shows that a ratio of 2 to 1 gives the optimum dehydration rate but large quantities of sugar remain undissolved, causing difficulties of draining the syrup from slices. Although with 1:1 ratio, the rate and extent of dehydration is slightly less, the sugar gets completely dissolved making it easier to drain the syrup. Hence from the points of view of convenience, handling and economy, a ratio of 1:1 seems to be suitable.

With the use of sugar syrup, the rate and extent of dehydration are increased with syrup strength (Fig. 1*a*.) However, since highly concentrated syrups are very viscous, they offer difficulties in handling compared to the minor benefits. A concentration of 70 per cent is suitable in actual practice. Further, for the sake of convenience, use of syrup is preferable than of dry sugar.

Fig. 1*c* shows that the rate of osmosis increases with the ratio of syrup to fruit but, as seen from Fig. 1*c*, the increase is small. Besides, larger ratios offer practical difficulties in handling the syrup-fruit mixture for processing. A ratio of 1.5:1 seems to be good enough for practical purposes.

Effect of temperature: Fig. 1*b* shows percentage loss in weight with time at temperature 27, 40, 50 and

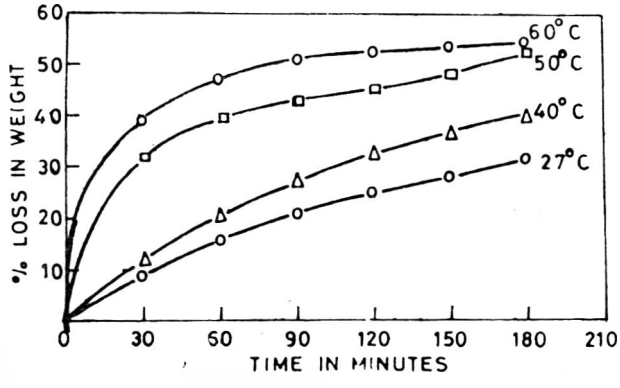


Fig. 1*b* Effect of temperature on rate of dehydration of banana pieces in sugar syrup

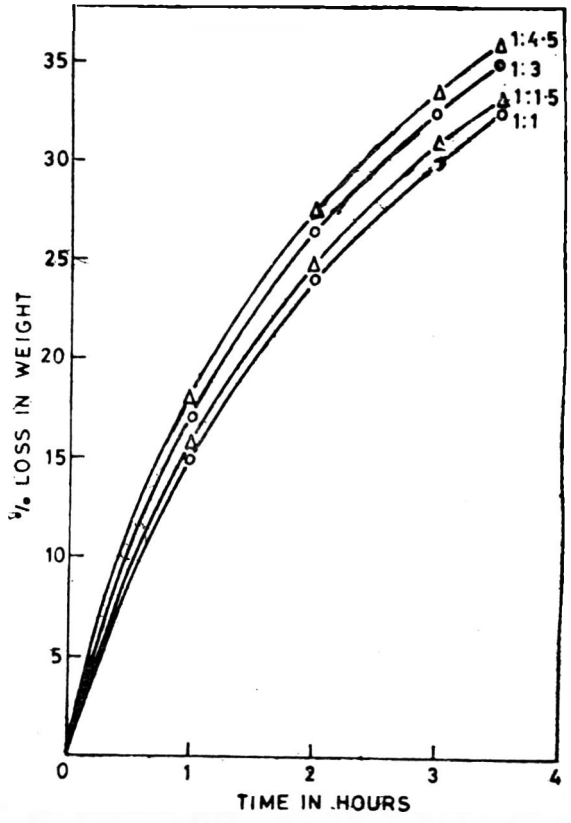


Fig. 1*c* Relation of fruit to sugar syrup ratio on osmotic dehydration of bananas

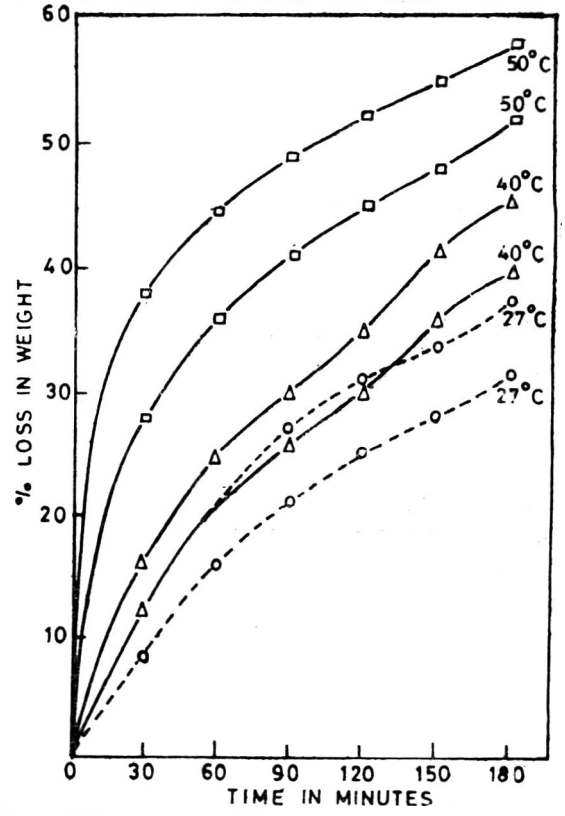


Fig. 1*d* Effect of temperature and agitation on rate of dehydration of banana pieces in sugar syrup

60°C when 70 per cent syrup is used in 1.5:1 ratio. The rate of osmosis as seen from the Fig. 1*b* is markedly affected by temperature. Since viscosity decreases with increase in temperature, it may be expected that temperature will favour osmosis[10]. Predictably, the rate of osmosis increases considerably at above 40°C. However, too high a temperature will have adverse effects on flavour and texture of the fruit. Hence an optimum temperature at which there is negligible flavour and texture loss needs to be chosen. Accordingly 50°C was found to be the most suitable temperature at which 50 per cent loss in weight would occur within three hours without any damage to the quality.

Effect of agitation: Fig. 1*d* shows weight loss with time at 27, 40, 50°C in 70 per cent sugar syrup (ratio 1.5:1) with and without agitation or shaking. The results clearly indicate that agitation or circulation of syrup reduces the time of osmosis.

Rehydration characteristics: Rehydration of osmotically dried bananas is slightly less than that of unsugared conventional dried bananas. Its reconstitution is about 55 to 60 per cent as compared to 62 to 65 per cent for air dried bananas as shown in Fig. 2, because part of the solids is a coating of sugar. The product however is more suitable as a ready-to-eat snack item. The product gets affected by moisture and light.

Sensory evaluation of osmotically dried banana: The average scores for freshly prepared samples and samples stored for 10 and 15 months at room temperature conditions were 8, 8 and 7, respectively, indicating the overall quality of the product in very good to good range of acceptance. A comparative evaluation of the products obtained by conventional air drying and by vacuum drying with osmotically dried product showed that the osmotically dried products retained more flavour, odour, puffiness and crisp texture than the others. In case of air dried samples, colour, flavour and texture were poor while with vacuum dried sample puffiness and flavour were lost.

Colour: Fig. 3 shows changes in colour of the product during storage at 32°C at 10 per cent moisture level and at 37°C at 20 per cent moisture level. Average Rd

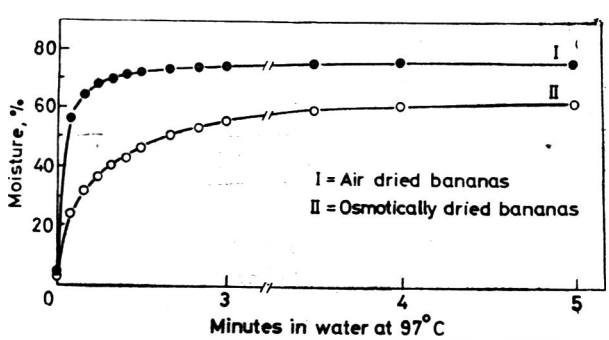


Fig. 2 Rate of rehydration of osmotically and air dried bananas

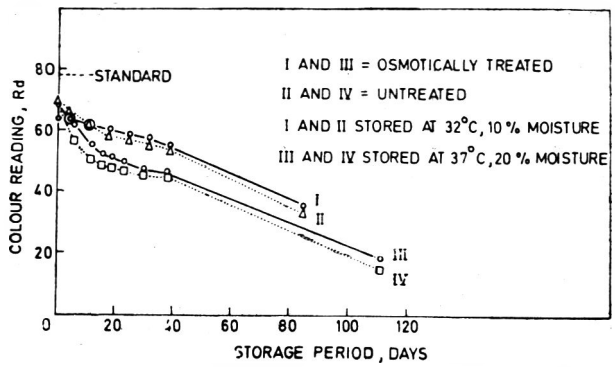


Fig. 3 Decrease in colour reflectance of banana slices at elevated temperature

values of duplicate samples show that there is little difference in colour stability between osmotically dried and conventional vacuum dried bananas, although the colour of osmotically dried bananas was more stable during the earlier storage periods. The average Rd values in this study indicate the lightness or darkness of the product.

Flavour: In general, flavour stability closely parallels colour stability. However, a phenomenon not previously noticed is the development of rancid flavour in osmotically dried bananas after a storage of $1\frac{1}{2}$ years. The cause of this flavour loss, although not determined, could be the flavour oils which probably are retained in greater amount in osmotically dried bananas than in bananas dried by other methods. This problem however could be overcome by use of an antioxidant in packaging. Further, in view of the finding[11] that walnut kernels were more stable to rancidity at a certain optimum moisture content than at either a higher or lower one, it may be desirable to store osmotically dried bananas at such an optimum moisture.

Storage time estimates: Considering these limits of colour and flavour, an estimate of the storage life of the products was made. At 25-28°C, vacuum dried samples stored satisfactorily in respect of flavour for 2 months, osmotically dried for 1 yr. Air dried bananas held at same temperature were satisfactory for only 2 weeks. A rough interpolation of data indicates that osmotically dried bananas processed by the above mentioned processes were 10 times as stable as the air dried bananas.

Packaging: As osmotically dried fruits are having moisture content in the range of 3 to 5 per cent, proper packaging is very essential. Amongst the various packaging materials used for storage of osmotically dried bananas, aluminium laminate pouches (paper/polyethylene/aluminium/polyethylene) (2.4/1.0/0.5/1.5 mil) were found to be the best. In case of polyethylene and polycell bags of various gauges, moisture absorption to a certain extent did occur resulting in spoilage of texture of the product.

Cardboard cartons were used as secondary packaging material to prevent crushing due to impacts during transportation of osmotically dehydrated products. Use of in-package desiccant like 'drierite' (alumina) or silica gel may help in extending shelf life.

Reuse of sugar syrup after osmosis: During osmosis, some volatile flavour component diffuses through the cell walls into the syrup[12]. Hence, the syrup after osmosis has a particular fruit flavour. This syrup can be utilized in the canning industry for fruit canning or as a fruit syrup. If required, it can be reutilized after proper concentration. After osmosis concentration of syrup reduced to 65 per cent which is to be increased to 70 per cent before reuse. For this, the syrup is filtered through a filter press to remove fruit fibres and then evaporated either under vacuum or in open atmospheric pressure. In the latter case, there is development of yellow brown colouration which can be removed by absorption on activated charcoal followed by filtration. Loss during handling in each cycle does not exceed 2-3 per cent syrup. Thus, the same syrup can be reused again and again after making to volume.

Applicability of the process for different fruits: The method is found to be applicable to fruits like mangoes, strawberries, sapotas, pineapple, apples, custard apple, etc. In case of mangoes the colour, flavour and texture of the fruit is maintained to an acceptable limit for a storage period of one year. In case of soft and sponge fruits like bananas and strawberries, during vacuum drying, the fruits do not collapse and give a puffy product with a crisp, honeycomb like texture[13]. The sourness of strawberries due to acids gets reduced because of leaching by syrup and deposition of traces of sugar on the outer surface of the fruit. The moisture level in such products is always kept about 2 per cent in order to retain crispness and puffiness. There is scope for use of this method for dehydration of tropical fruits like guava and papaya.

Based on these studies, a flow diagram for osmotic dehydration of banana chips or slices on semi pilot scale studies as shown in Fig. 4 was used.[13]

Semi pilot scale studies: To find out the feasibility of the process on large scale, semi pilot scale studies were carried out using 10 kg of peeled bananas. The process is semicontinuous because of the operation of vacuum drying after osmosis. The process arrived at on the basis of laboratory scale experience (Fig. 4) consists of many discrete operations. As large scale handling of material is involved, it was necessary to design and fabricate some equipment for better results. The designed and fabri-

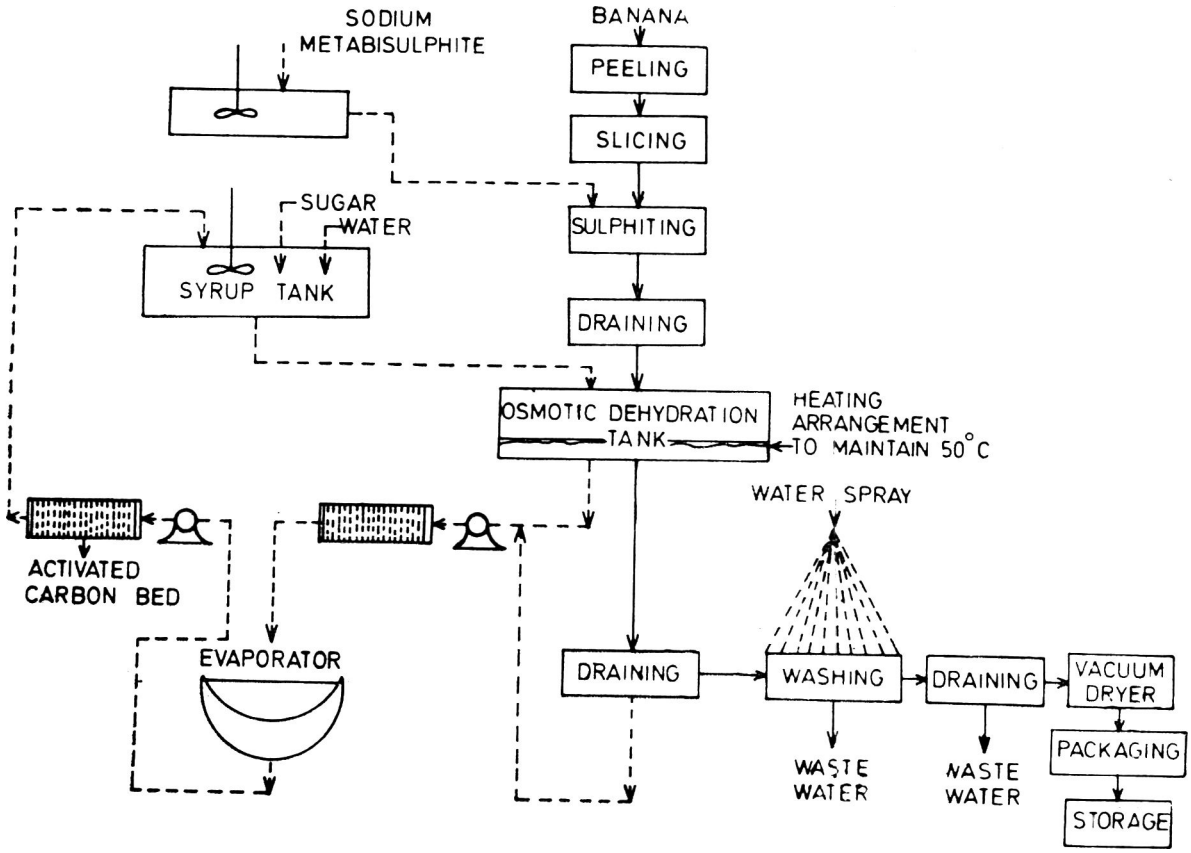
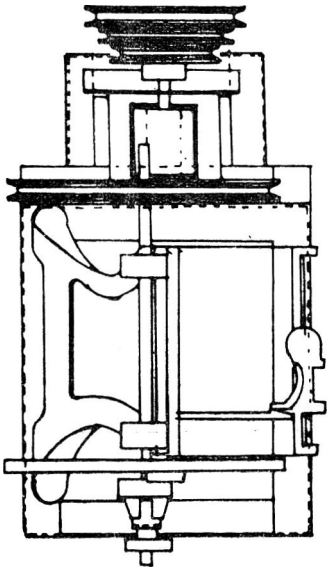


Fig. 4 Flow diagram for osmotic dehydration process

cated equipment included (*i*) fruit slicing machine, (*ii*) sulphiting tank, (*iii*) osmotic dehydration tank, (*iv*) sugar syrup preparation tank, (*v*) sodium metabisulphite preparation tank, (*vi*) product trays, and (*vii*) bracket for lifting the tray. The descriptions of the designed and fabricated equipment for osmotic dehydration of bananas are given below.



Fruit slicing machine: As shown in Fig. 5, the machine consists of (1) fruit cutter, (2) stand, (3) motor, and (4) product tray. The design features of the machine are such that (1) variable thickness of the fruit slices during cutting is possible and (2) variable cutting speeds could be obtained as a result of which throughput can be varied, and (3) the same machine could also be used for cutting vegetables and fish with little alteration. The fruit cutter is made of stainless steel and is half round in shape. The cutter is installed on a stand with its shaft, fruit holder assembly and pulley for its drive arrangement. A 1/4 H.P. single phase A.C. supply, 250 volts, 1400 rpm, class E insulation and continuous rating electrical motor was used for driving this cutter. The motor shaft is fitted with a variable speed pulley drive arrangement. The motor, cutter and the whole assembly is supported on a 6 mm thick mild steel angle stand. The stand is fabricated with grooves as shown in Fig. 5 for holding product trays of aluminium. The fruit holder assembly as shown in Fig. 5 is fabricated from stainless steel 316. The fruit to be cut is held tight in

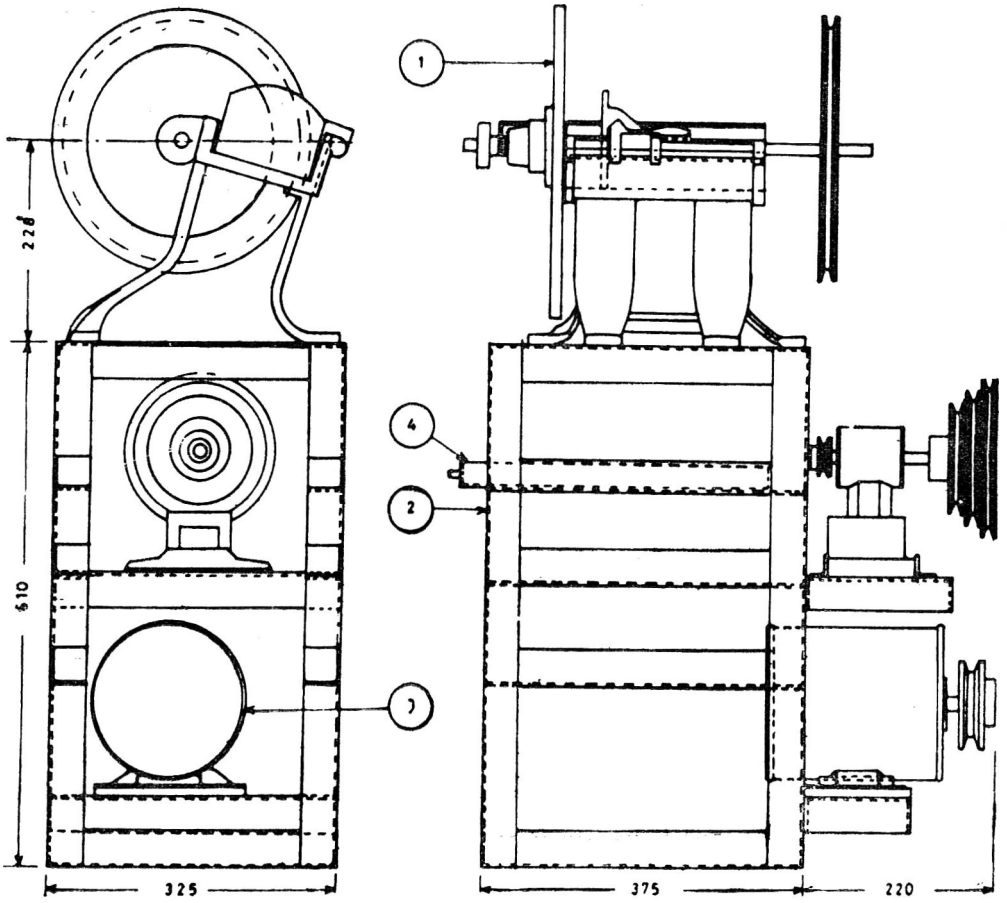


Fig. 5 Fruit slicer

1. Fruit cutter; 2. Stand; 3. Motor; 4. Product tray

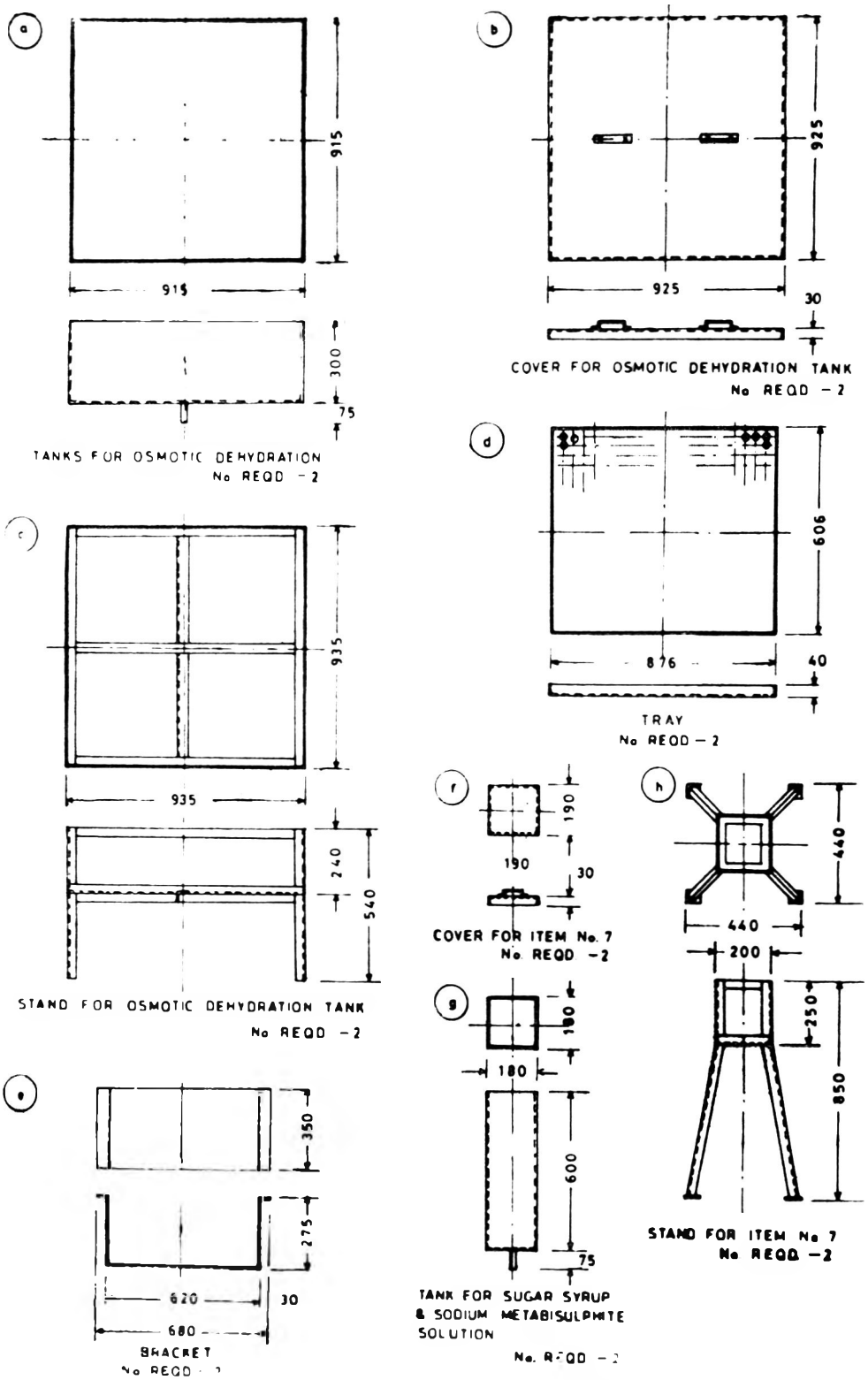


Fig. 6 Equipment for osmotic dehydration

position with an adjustable clamp below the cutter portion in this assembly. As the cutter starts rotating, the fruit surface held tight in the fruit holder assembly below the cutter gets separated into fruit slices and the slices are collected in the product trays. The product trays could then be emptied as and when needed. About 40 kg of peeled bananas could be cut into slices of 8 to 10 mm thickness per hr using this machine.

Osmotic dehydration tank: The tank made of aluminium sheet 0.3 cm thick is very similar in size, shape and other aspects to sulphiting tank, as shown in Fig. 6a. The tank was provided with two electrical heaters each of 1 KW capacity complete with electronic thermostat controls for maintaining temperature (50°C) and an electrically operated standard pump (10 lit capacity) for gently circulating sugar syrup to produce some agitation during osmosis. Similarly sugar syrup and sodium metabisulphite preparation tanks as shown in Fig. 6g were fabricated.

Product trays: For carrying out the operation of sulphitting and osmosis conveniently, product trays of nearly same dimensions as that of the sulphiting and osmotic dehydration tanks are fabricated Fig. 6*d*. The trays (of aluminium) are fabricated in such a way that an adjustable perforated wire mesh cover can be slipped in the groove at the top of the tray. As a result, product kept in the tray will not float when the trays with products are dipped in sodium metabisulphite or sugar syrup. About 25 dozen sliced bananas could be spread on each tray.

Bracket for lifting the tray: This is made of aluminium (680 mm width, 275 mm height). Its main purpose is to lift or lower the product tray in sulphiting or osmotic dehydration tank. The bracket can be inverted and used during washing operation.

A 10 plate (9″ ×9″ size) and 12 frame filter press assembly having about 5000 sq. cm. filtering area, was used for filtration of sugar syrup after osmosis. The concentrator was used for increasing the concentration of sugar syrup from 65 to 70 per cent by evaporating the water from diluted sugar syrup after osmosis. Vacuum chamber dryer was used for final drying of fruit slices to a moisture content of 3 to 5 per cent at 60°C, 29 in. vacuum, for 8 hr.

Observations on the semi pilot scale studies: Sensory evaluation studies on the product obtained in the scaled up process were in agreement with those on laboratory scale. The process can be made semi-continuous by carrying out osmosis and vacuum drying, simultaneously as shown in Fig. 7. Only initial hold-up time of about 12 hr will be required to start the process, but then the continuity can be maintained.

These studies demonstrated the potential design and operation problems of a commercial osmotic drying process. A major problem is the large difference in specific gravity of bananas (0.8 to 0.9) and concentrated sugar syrup (1.35). The fruit tends to float, particularly at high temperatures viz. 50°C when entrapped air in the banana tissue expands. When slices are submerged in the syrup using pressure, the packed bed formed of

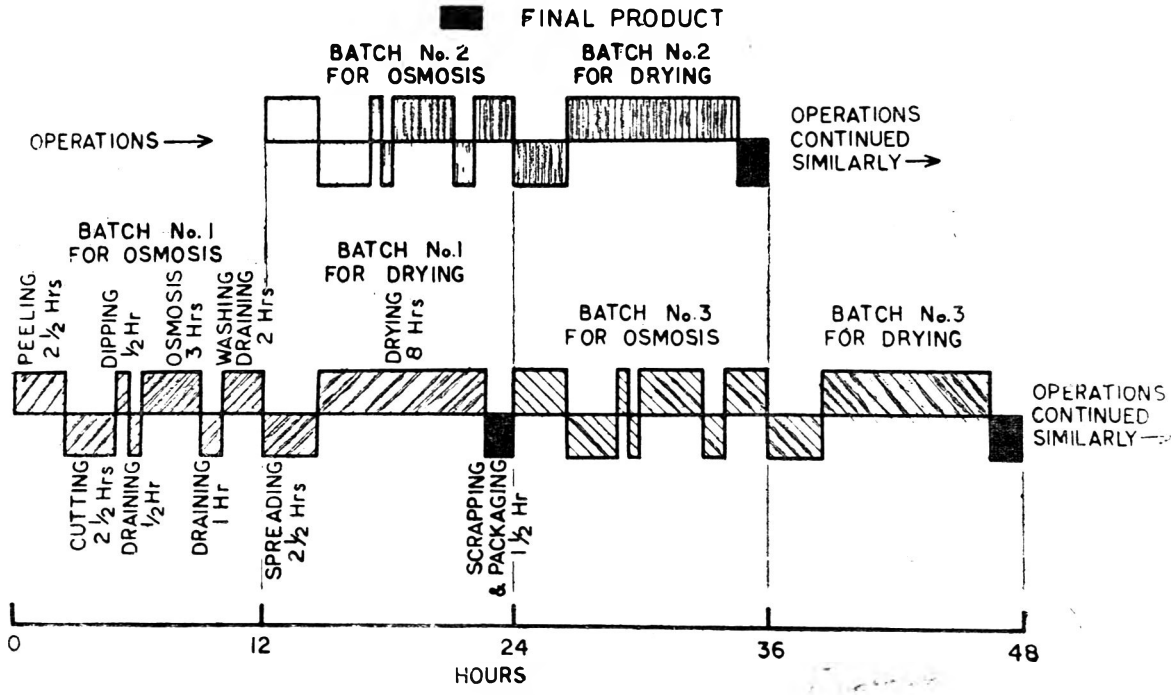


Fig. 7. Operation schedule for osmotic dehydration of bananas on semi—continuous basis

banana slices reduces syrup circulation. Tumbling beds in a rotary unit with internal syrup recirculation might also succeed if the fruit can withstand the mechanical forces.

Washing of banana slices after osmosis must be performed quickly and positively to prevent residual heat from further softening the fruit. For this, water spraying method seems to be the best. Reuse of sugar syrup after filtration, concentration, and mixing with activated charcoal and then filtering was found to be adequate for six times. If the sugar syrup after concentration is stored at 0°C, it can be reused 15 times or more. The material containing extracted banana essence and solids made a premium table syrup.

The feasibility of the process: Laboratory and semi-pilot scale studies demonstrated the potential design and operating problems of a commercial osmotic drying process.

As the process involves two steps viz. osmosis and vacuum drying, it is expected to be costlier than other traditional processes. But it produces high quality products and costs much less than freeze dried products. The main advantage of the process is storage stability, flavour and colour retention to a high degree.

For producing osmotic dried fruits, several avenues are open. Choice depends upon combination of economic factors and on planned use for the product. Snack items to eat out of hand might be dried in air to 15 per cent moisture, or vacuum dried to 2 per cent.

In laboratory production, dry sucrose and syrups can be used in static or agitated conditions at different temperature ranges. When syrup is used, it can be recirculated and reconcentrated in conventional evaporator or open pan kettle. As a result, there will be a saving in the cost of processing.

Syrup concentration is not critical but from practical point of view 70 per cent concentration was found to be optimum. Invert sugar can be used in whole or in part. It maintains high osmotic pressure without having the sucrose crystallizing out. It has been observed that flavour deterioration begins to take place above about 52°C. At 50°C, 50 per cent weight reduction is accomplished in from $2\frac{1}{2}$ to 3 hr. Thus, there is considerable saving of time in the process of osmosis, as a result, production could be increased.

Flavoured syrup is produced when either dry or liquid sugar are used. Excess syrup could be used in concurrent canning operations; packaged as a fruit flavoured syrup; or packaged with 50 per cent weight reduction fruit for dehydro-freezing. From all these considerations, it may be seen that the process could be made economical by judicious selection of optimum conditions and reuse of osmotic agents.

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Determination of Thermal Process for Canned Mandarin Orange Segments

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Peroxidase and pectinmethylesterase (PME) are the heat resistant enzymes present in mandarin orange. Thermal process has been evolved on the basis of the inactivation of the PME which is more heat resistant than the peroxidase. Procedure for determining the correction to be applied for the heating lag in the determination of thermal inactivation time (TIT) and decimal reduction time (D) is given. The concept of decimal reduction has been applied for process evaluation. The TIT of PME in homogenate containing lye peeled segments and sugar syrup in the ratio of 11.6:6 and soluble solids of 20 per cent, was $F_{203.5}^{20.4} = 1.00$ and $F_{206.5}^{21.6} = 1.00$ at pH 3.6 and 4.0 respectively. The F values were equivalent to 3.72 D and 4.67 D respectively. In commercial canning, a 5 D process at pH 3.6 and 6 D process at pH 4.0 is recommended. The time required at different initial and processing temperatures are given.

World over, oranges are mostly processed in the form of juice or concentrate for subsequent use as juice or in the preparation of beverages. Mandarin oranges (*Citrus reticulata* Blanco) are canned as segments in syrup in Japan and Taiwan. In India, juice, concentrate and segments are being commercially manufactured from mandarin oranges.

Orange contains enzyme systems such as peroxidase and pectinmethylesterase (PME) which are heat resistant. Unless inactivated, peroxidase causes undesirable flavour changes and PME causes loss of cloud stability and gelation in frozen concentrates.[1,2] Generally, the pH of orange is lower than 4.0 and spoilage is caused by non-spore forming bacteria like *Lactobacilli* or yeasts which have lower heat resistance than PME.[3-5]

The heat treatment necessary to inactivate the PME in juice varies with the variety, pH, etc.[6-9] The effectiveness of heat treatment is assessed either by the quantitative estimation of the residual enzyme activity[6] or by qualitative jel-test.[9] The heating times and temperatures reported are for the commercial pasteurization conditions of juice and not for the actual thermal inactivation time (TIT) of the enzyme which can be used in process evaluation.

According to Hirano[10], in Japan, the filled cans of orange segments in syrup are sealed in a vacuum sealing machine and heat processed in a continuous agitating cooker and cooler. The heating time for No. 5 cans (ca. 301 × 303) at 180°F is about 13 min. In India, in one of the processing factories, the filled A $2\frac{1}{2}$ cans (401 × 411) are passed through a steam exhaust box and then processed at 190°F for 30 min in a rocking pasteurizer and in another plant, cold filled, mechanically vacuumized 11 oz (ca. 301 × 304) cans are processed in a rocking pasteurizer at 180°F for 18 min.

Process time for canned fruits in syrup have been evolved on the basis of thermal inactivation of the enzymes[11-14]. Investigations carried out to determine the TIT of the PME and the peroxidase, and calculation of the safe thermal process for segments in syrup from mandarin orange form the subject matter of this paper.

Materials and Methods

1. *Preparation of the fruit for the TIT studies:* Mandarin oranges, purchased from the local market, were peeled and segments separated. A cold lye peeling procedure found most suitable was adopted to peel the segments.[15] After hand peeling of fruit, the segments were soaked in 1.5 per cent HCl for 60 min at room temperature (25°C) washed in water, dipped again in 1 per cent sodium hydroxide for 25 min at room temperature and re-washed thoroughly. The seeds were removed, and the peeled segments were blended. The resulting pulp was filled into polyethylene bags, quick-frozen in a plate freezer at –40°F and stored at 0°F until use. The syrup homogenate was prepared by blending the thawed pulp and sucrose syrup in the ratio 11.6:6 so as to get a final product having 20° Brix, similar to the cut-out Brix of the final product and the pH was adjusted to 3.6 and 4.0 using citric acid.

2. *TIT of PME:* The technique used was almost similar to that of Bigelow and Esty[16] for determination of the thermal death time (TDT) of bacteria.

(*i*) *Determination:* Pyrex glass tubes (here-in-after referred to as TIT tubes) of 13.5 cm length, 1.6 cm OD and 1.4 cm ID were used. Ten ml of the sample were

pipetted into each of the tubes, heated in a thermostatic water bath for known times and cooled immediately by plunging the tubes into chilled water. Two tubes were used for each time and temperature, and the experiment was performed in triplicate. Tubes of larger diameter than that recommended for TDT had to be used, because 10 ml of the sample was required to test the activity.

The come-up time was determined using a 10.5 cm long needle-type thermocouple fitted with spacers to position the thermocouple in the centre of the tube. The temperature at time intervals was noted with a manually operated Leeds and Northrup potentiometer calibrated in terms of degree Fahrenheit. Experiment was done in triplicate for each temperature.

(ii) Correction for heating lag during the come-up time: The extent of inactivation of the PME (analogous to lethality of microorganisms) during the time-lag in heating and cooling was calculated by applying the general method of Bigelow *et al.*[17] for process calculation as suggested by Perkins *et al.*[18] in the case of *Cl. botulinum*. TIT values as determined without corrections for the come-up time were plotted as shown in Fig. 1. Heat penetration rate into the TIT tubes during the come-up time was plotted as shown in Fig. 2. The TIT corresponding to temperature at time intervals during the come-up time was found from the TIT curve (Fig. 1) and the inactivation rate was calculated using the expression: $\frac{1}{\text{TIT}}$. Area under the thermal inactivation curve (analogous to lethality curve of microorganisms) QRSQ represented the extent of inactivation contributed during the come-up time, and the area under OPRSO represented total inactivation, if the temperature rise had been instantaneous. The effectiveness and the corrected TIT were calculated using the following expressions:

$$\text{Effectiveness} = \frac{\text{Area under QRSQ}}{\text{Area under OPRSO}}$$

$$\text{Correction} = \text{Come-up time (1 – effectiveness)}$$

$$\text{Corrected TIT} = \text{Experimental TIT – correction.}$$

Since the cooling was very fast, and the inactivation contributed by it was negligible, it was not considered.

(iii) Test for PME activity: The jelly-set procedure of Patron and Swinzow[19] as modified by Rothschild *et al.*[9] was further modified to determine the time required to inactivate the PME. Ten ml of 0.5 per cent pectin (6 per cent methoxyl) solution was added to 10 ml of the sample, pH adjusted to 7.0 with 0.1 N NaOH and the volume made to 60 ml with distilled water. To this, 1 ml of 1 N $CaCl_2$ solution was added, mixed, transferred to 100 ml stoppered conical flask and incubated at 34°C for 48 hr after adding a few drops of toluene. The

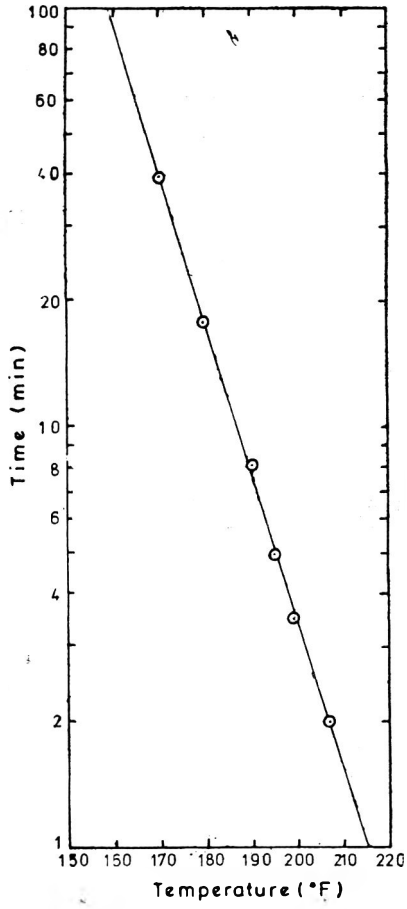

Fig. 1 Uncorrected TIT curve of PME in syrup homogenate at pH 3.6.

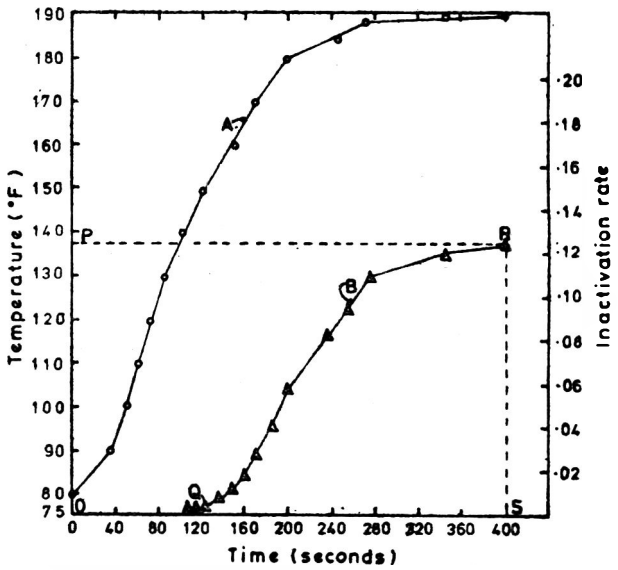

Fig. 2 Heat penetration curve (A) into TIT tubes and the corresponding thermal inactivation rate curve (B) at pH 3.6 during the come-up time.

control sample having no enzymic activity was prepared in the same way, but using a sample heated for 10 min in boiling water. After incubation, viscosity of the samples was measured using Brookfield viscometer (spindle No. 1 and speed 60 rpm) at 26.7°C. Any increase in the viscosity of the sample as compared to that of the control indicated the presence of the active enzyme. The minimum time at a temperature when the sample showed no activity was taken as TIT.

3. *Calculation of decimal reduction time:* Ten ml of sample was heated in TIT tubes described earlier for varying periods at different temperatures as in the TIT determination. The heating times selected were above the come-up time and well below the TIT of the enzyme. The residual PME activity was determined by the method of MacDonnel *et al.*[20] as modified by Rouse and Atkins[6] and the activity was expressed as PE.u. per ml $\times 10^4$. Correction to be applied to the actual heating time for the lag during come-up time was determined according to the procedure given under TIT determination.

Time required at a particular temperature to reduce the enzyme activity to one-tenth (D) was calculated using the formula suggested by Stumbo[21]

$$D = \frac{U}{\log a - \log b}$$

where '*a*' is the initial activity and '*b*' is the activity which survived the heating time of 'U' in min. The D values calculated for different heating times at one temperature were averaged.

4. *TIT of peroxidase:* The procedure used in heat inactivation studies was essentially that of Nanjundaswamy *et al.*[14] As the samples were cloudy because of pulp, the colour formed with guaiacol was measured in Bausch and Lomb Spectronic-20 colorimeter with reflectance attachment at 520 nm. the wavelength of minimum reflectance. The sample was heated for 5 min in boiling water, and the reagents added served as control to set the instrument to 100 per cent reflectance. The minimum heating time at a temperature at which the reflectance was 100 per cent was taken as the inactivation time. The correction for heating lag was determined as in the case of PME.

5. *Quantitative estimation of peroxidase activity:* The activity was estimated by the ascorbic acid oxidation method of Joslyn and Zuegg.[22]

6. *TIT and thermal resistance curves:* These curves were plotted on semilog paper by applying the least square analysis.[23]

7. *Heat penetration studies:* Orange segments were prepared by cold lye peeling procedure as described in the preparation of the fruit for the TIT studies. Seeds were removed carefully with bamboo sticks to avoid breakage. Unbroken segments (580 g) were filled into A $2\frac{1}{2}$ (401 $\times$411) cans and covered with (300 g) sucrose syrup (about 45° Brix). In one set of studies, the syrup was filled hot (160°F) and the cans were exhausted for 7 min and sealed. In the other set, syrup was filled at room temperature. The sealed cans were processed at 190°, 207° and 212°F and cooled in water. Six cans were used for each run.

Heat penetration rate into the cans was measured using Ecklund's non-projecing, plug-in, needle-type thermocouples. Heating was found to be by rapid convection initially, followed by slow convection or conduction later. Heat penetration studies carried out by positioning the thermocouple at different positions indicated the heating to be the slowest at the geometric centre. Therefore, the thermocouples were positioned at the geometric centre of the can in subsequent studies. Lead wires from thermocouples were connected through selector switch to a manually operated Leeds and Northrup potentiometer.

8. *Process calculation:* Processing required for canned segments in syrup at different pH was calculated by the improved graphical method of Ball and Olson.[24] Inactivation rates at various temperatures of the heat penetration curve during heating and cooling were calculated using the expression:

$$I = \log^{-1} \frac{T - T_x}{z}$$

where I is the inactivation rate at temperature T, T_x is the temperature at which F is 1 min and *z* is the number of °F required for the TIT curve to traverse one log cycle.

Thermal inactivation rate curve was drawn by plotting inactivation rate against time. The F value was calculated by cutting the area beneath the inactivation rate curve and dividing its weight with the weight of an area equivalent to unit inactivation from the same paper. To calculate the processing time having the desired F value, cooling curves at different points were drawn parallel to the original cooling curve. Areas under these curves were found and the corresponding F values were calculated. The actual processing time was determined by graphical interpolation.

Results and Discussion

In the canned fruits and beverages, thermal process given should be sufficient to prevent spoilage by the micro-organisms and to inactivate the enzymes. As already stated, PME has been found to have higher heat resistance than the spoilage microorganisms in orange products. Hence, PME as well as peroxidase were used as test enzymes for process evaluation. Activity of these enzymes as also the total soluble solids (TSS), acidity and pH of the edible portion varied considerably (Table 1). Lower the pH, shorter is the pro-

TABLE 1. TOTAL SOLUBLE SOLIDS, ACIDITY, PH AND ENZYME ACTIVITY IN ORANGE

Particulars	Edible portion[a]	Syrup homogenate for TIT studies[b]	
TSS (%)	7-12	20	
Acidity as anhydrous citric acid (%)	0.59 – 1.15	0.63	0.58
pH	3.0 – 4.0	3.6	4.0
PME activity (PE.u. × 10^4/ml)	20.6 – 52.0	10.6 – 18.0	
Peroxidase activity (k_1/g)	0.19 – 0.5		

[a]Prepared by hand peeling of fresh orange segments and blending
[b]Prepared by lye peeling of segments and blending with sucrose syrup in a blender.

cess which may be used with safety. The pH of the fruit varied from 3.0 in early rainy season crop of June-September to 4.0 in late main summer crop of November to February. Generally, summer crop is used for processing, during which period, the pH ranges from 3.6 to 4.0. Hence, thermal inactivation studies and process evaluation were carried out at pH 3.6 and 4.0. The TSS of syrup homogenate was adjusted to 20 per cent to simulate the cut-out Brix of canned segments in syrup.

F and D values of PME

Correction for heating lag during come-up time in TIT determination: TIT, as determined, needs correction to account for the necessary delay in bringing the contents of the tube to the temperature of the water bath. This correction depends upon the sterilizing value of the time lag in heating and cooling. In the determination of TIT of enzymes, Kaplan *et al.*[11] applied an arbitrary lag correction of 1 min chosen as an approximation of the time having no lethal action, while Nebesky *et al.*[12] used a correction of 1.5 min. Only 42 per cent of the retort come-up time has been found to contribute to lethality during the processing of the cans.[24] Dastur *et al.*[13] and Nanjundaswamy *et al.*[14] used this correction factor in determining the TIT of the enzyme.

In literature, though reference is made for determination of lethality of the come-up time by the general method of Bigelow *et al.*[17] for process calculation, the actual procedure of evaluation has not been given by the earlier workers. The procedure described under the experimental portion provides a basis.

At pH 3.6 and 4.0, 45.06 and 42.86 per cent respectively of the come-up time was effective. Hence, the correction required decreases slightly with the decrease in pH (Table 2).

Corrected TIT: Kaplan *et al.*[11] found the TIT curve of the enzyme, when plotted on a semilog paper, to be a straight line as in the case of micro-organisms. Therefore, the terms F (the time required to inactivate the enzyme at a particular temperature) and *z* (number of °F required for the TIT curve to traverse one log cycle) are used to denote the TIT curves as in the case of the thermal death time (TDT) curves. TIT curves of PME at different pH are given in Fig. 3. The temperature

TABLE 2. CORRECTIONS FOR COME-UP TIME IN THE DETERMINATION OF TIT OF PECTINMETHYLESTERASE IN SYRUP HOMOGENATE

pH	*z* (°F)	Temp. (°F)	Come-up time (min)	Effectiveness (%)	Correction* (min)
3.6	28.36	180	6.4	43.80	3.63
		190	6.6	48.10	3.43
		195	6.0	50.10	2.99
		199	6.2	38.20	3.83
Mean			6.3	45.06	3.46
4.0	28.04	180	6.4	41.17	3.76
		190	6.6	45.70	3.58
		195	6.0	48.42	3.96
		199	6.2	36.13	3.96
Mean			6.3	42.86	3.60

*This is to be subtracted

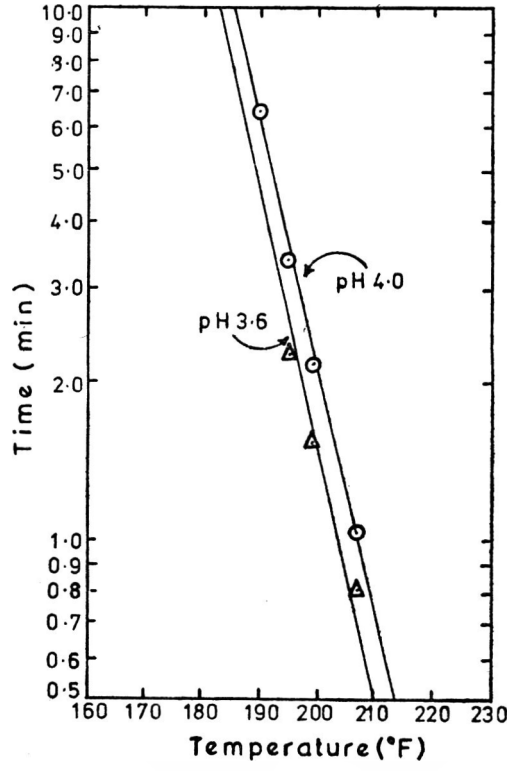

Fig. 3 TIT curve of PME in orange syrup homogenate at pH 3.6 and 4.0.

TABLE 3. F, D AND z VALUES OF PME AND F/D VALUE FOR PROCESS CALCULATIONS

pH	From TIT curve		From thermal resistance curve		
	Temp at which F = 1.00 (°F)	z (°F)	D value at temp at which F = 1.00 (min)	z (°F)	F/D
3.6	203.5	20.4	0.269	21.5	3.72
4.0	206.5	21.6	0.214	20.0	4.67

at which the time required to inactivate the enzyme is 1 min and the z values are given in Table 3. The inactivation time for PME decreased as the pH lowered.

Decimal reduction time of PME: In some of the canned orange segments, wherein the sterilization value (F) of the process was equal to the F value of PME in syrup homogenate, the enzyme had not been completely inactivated (Table 4). Similar observations have been made by Kaplan *et al.*[11] and Nebesky *et al.*[12] in other canned products. The enzyme activity in fruits considerably varies (Table 1). The F value of the enzyme depends upon the initial activity. When the enzyme activity in the sample canned is higher than that used for TIT studies, the calculated process time would not be sufficient for complete inactivation of the enzyme. As in the case of micro-organisms, the rate of the inactivation of the enzymes is logarithmic and is independent of the initial activity.[25] Hence, the time required to reduce the enzyme activity to one-tenth (D value) at different temperatures were determined to calculate the process time based on decimal reduction. The D values found are given in Table 3. The thermal resistance curves (similar to the "Phantom" TDT curve of bacteria) for PME were plotted and the z values determined (Fig. 4). The z value from the thermal resistance curves was higher by 1.1 at pH 3.6 and was lower by 1.6 at pH 4.0 as compared to the values obtained from TIT curves (Table 3). These slight variations may be attributed to the different procedures used in the determination of the TIT and the decimal reduction time.

At pH 3.6 and 203.5°F, F and D values were 1 min and 0.295 min respectively. For almost complete inactivation of PME, 3.72 decimal reductions (which is given by F/D ratio) are required (Table 3). Unlike micro-organisms, the enzymes do not multiply, but their initial activity may vary. Hence, a slight increase in the decimal reduction process should ensure inactivation of the PME. Accordingly, the F values equivalent to slightly higher decimal reductions in the canning of orange segments are given in Table 4. The decimal reductions (F/D) required for inactivation of the enzyme increased as the pH increased (Table 3).

F value of peroxidase: TIT of peroxidase was determined at pH 3.6 and 4.0. Come-up time was 2 min and 22 sec of which 44 per cent was effective. The F values of peroxidase were 1 min at 186.5°F with a z value of

TABLE 4. PROCESS TIME FOR ORANGE SEGMENTS CANNED IN SYRUP IN 401×411 (A 2½) CANS AT DIFFERENT FILLING AND PROCESSING TEMPERATURES

pH	F value for process calculations	Process equivalence in decimal reductions	Initial temp (°F)	Processing time (min) at 190°F	207°F	212°F
3.6	$F_{203.5}^{20.4} = 1.00$	3.72 D	81	34.0	30.0	10.0
	,, = 1.08	4.00 D		35.0	31.2	10.4
	,, = 1.48	5.00 D		38.5	32.6	11.4
	,, = 1.00	3.72 D	160	19.0	14.0[a]	6.0
	,, = 1.08	4.00 D		20.0	15.0	6.2
	,, = 1.48	5.00 D		23.0	16.2	6.6
4.0	$F_{206.5}^{21.6} = 1.00$	4.67 D	81	35.5	31.5	10.8
	,, = 1.28	6.00 D		39.0	33.0	11.4
	,, = 1.50	7.00 D		41.5	34.5	12.6
	,, = 1.00	4.67 D	160	21.0	16.0[a]	6.4
	,, = 1.28	6.00 D		24.0	17.3	7.0
	,, = 1.50	7.00 D		26.0	18.5	7.4

[a]PME had not been completely inactivated in these samples.

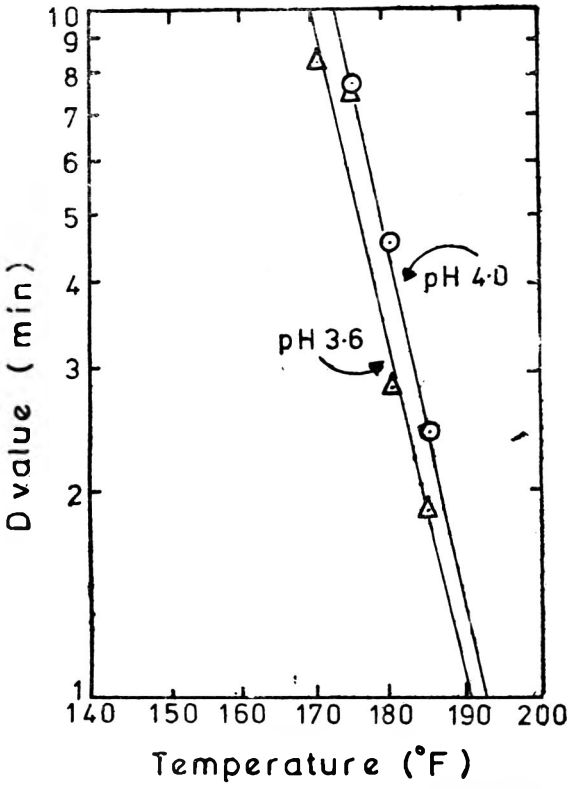

Fig. 4 Thermal resistance curve of PME in orange syrup homogenate at pH 3.6 and 4.0.

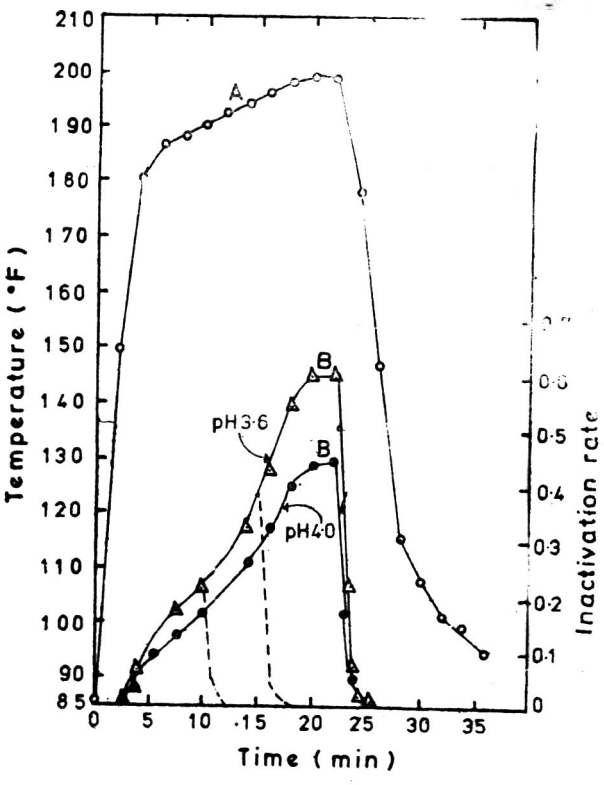

Fig. 6 Heat penetration curve of canned orange segments in syrup in A 2½ (401×411) cans with initial temperature of 81°F and processed at 212°F and the corresponding thermal inactivation rate curves at pH 3.6 and 4.0.

A, Heat penetration

B, Inactivation rate

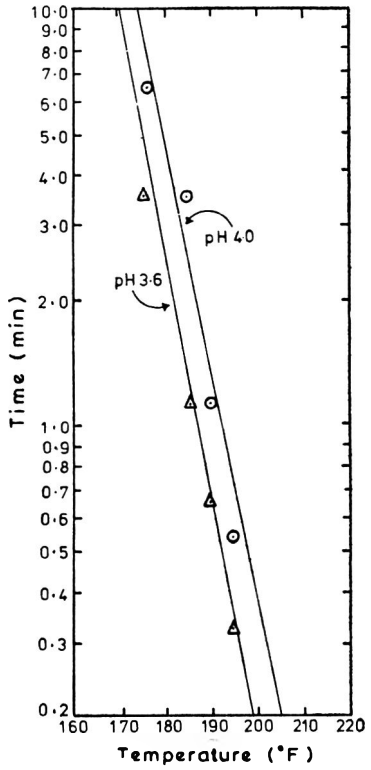

Fig. 5 TIT curve of peroxidase in orange syrup homogenate at pH 3.6 and 4.0.

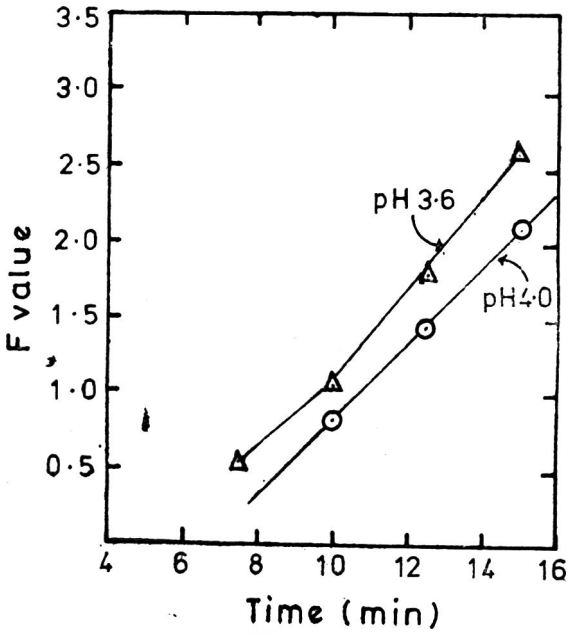

Fig. 7 F value *vs* process time plot for orange segments canned in A 2½ (401×411) cans with initial temperature of 81°F and processed at 212°F.

17.2 at pH 3.6, and 1 min at 192°F with a z value of 18.2, at pH 4.0 (Fig. 5). Since these values were lower than those of PME ($F_{203.5}^{20.4} = 1.00$ at pH 3.6 and $F_{206.5}^{21.6} = 1.00$ at pH 4.0), the process evaluation was based on the inactivation of PME.

Process calculation: The heat penetration curve of canned orange segments filled cold and processed at 212°F is given in Fig. 6. From the TIT curve of PME (Fig. 3), it is evident that the time required to inactivate the enzyme at pH 3.6 is 1 min at 203.5°F, and at pH 4.0, 1 min at 206.5°F. Hence these temperatures were chosen as the base temperature for calculation of process time.

The thermal inactivation rate curve are given in Fig. 6 and the graphical interpolation curves to find the process time corresponding to the desired F values in Fig. 7. The process times calculated are given in Table 4. Similar studies were carried out either with or without exhausting the cans prior to sealing, and at processing temperatures of 190°, 207° and 212°F. The process times calculated are given in Table 4.

Quality of the canned product: Examination of the canned product for the calculated periods either immediately after processing or after two and ten months of storage at room temperature (25-30°C) showed no peroxidase or PME activity. The segments had retained normal colour and texture, and the flavour was full. The breakage of the segments was negligible and the syrup was clear.

Recommendation for commercial processing: In the canning of orange segments, the F values of 1 min at 203.5°F and pH 3.6, and 1 min at 206.5°F and pH 4.0 were equal to 3.72 D and 4.67 D respectively (Table 3). At pH 3.6 and 4.0, the time required for processing A $2\frac{1}{2}$ (401 × 411) cans at 190°F was 34.0 min and 35.5 min respectively when the initial temperature was 81°F (Table 4). When the initial temperature was 160°F, the time required was 19.0 min and 21.0 min respectively. In the commercial processing, it would be advisable to use a process equivalent to 5 D and 6 D instead of 3.72 D and 4.67 D process at pH 3.6 and 4.0 respectively which would ensure not only the inactivation of enzyme but also other variables like fruit to syrup ratio, strength of the covering syrup, initial temperature of the can, the rate of cooling, etc. encountered in the canning process. The corresponding increase in the processing time is very little which decreases with the increase in the processing temperature. The process time required to achieve this at the different processing temperatures are given in Table 4.

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Studies on the Chromatographic Deterpenation of Lemon and Lime Oils

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Deterpenation of cold pressed lemon and lime oils was carried out by the column chromatographic method and the various parameters affecting the chromatographic separation of terpenes and oxygenated compounds have been studied in some detail so as to achieve the conditions of maximum economy consistent with reasonable efficiency. The results showed the necessity for using a silicagel oil ratio of 1 :1 for both lemon and lime oils and silicagel activity of I / IIa (Brockman scale) for efficient and economic deterpenation of these oils. Distilled oils are less preferred for deterpenation as they are poor in their oxygenated compounds and flavour value.

Citrus essential oils are much valued for their flavouring properties and are widely used in processed foods, beverages, pharmaceuticals and perfumery. The terpenes present in the essential oils deteriorate rapidly and develop off-flavours as a result of their oxidative and photochemical transformations. Partial or complete removal of the terpenes is therefore necessary to get deterpenated or terpeneless oils, which are valued for their enhanced stability, improved solubility and increased flavour strength.

Deterpenation can be carried out by different methods like fractional distillation, counter-current extraction, column chromatography and hydrotropic method.[1-13] Different workers[8-11] have tried to improve the column chromatographic method mainly with respect to the operating costs. Recently, deterpenation of some of the Indian orange oils was done by the chromatographic method.[15] The various parameters affecting the chromatographic separation of the terpenes and the oxygenated compounds were studied so as to achieve the conditions of maximum economy consistent with reasonable efficiency. The column chromatographic method was found to have some distinct advantages with respect to the simplicity of the equipment, the ease of operation and the complete separation of the terpenes and the oxygenated constituents (terpeneless oil).

The work reported herein relates to deterpenation of citrus essential oils. The data regarding the various aspects of chromatographic deterpenation of lemon and lime oils are presented here.

Materials and Methods

Oils used: The physico-chemical constants of the lemon and lime oils used in these experiments are given in Table 7. The various types of lemon and lime oils collected during the investigation were analysed for their quality characteristics according to the standard method.[1,12,13]

Silica gels: For column and thin layer chromatography (TLC) silica gels of 100-200 mesh and silica gel-G were used respectively. Silica gels for column chromatography were heated in hot air oven and their activity was determined by the Hernandez procedure.[14] Deactivation of silica gel was done by adding required quantity of water to the activated silica gel. Deactivation was done to get silica gels of different activities.

Solvents: Solvents of chemically pure grade were used.

Deterpenation: The method of deterpenation was similar to that used for orange oil.[13] The set up for the deterpenation of lemon and lime oils consisted of a glass column (90 cm length and appropriate diameter) fitted with a stopcock adapter and a receiver assembly. The receiver assembly comprised of a test tube with a side tube for collecting the eluate under any desired vacuum. A cotton plug and a filter paper disc were kept at the bottom of the vertically clamped column and silica gel was poured through a funnel into the column with stopcock open. The column was tapped gently to get a packing of moderate tightness. Another filter paper disc and a glass wool plug were kept on top of the column and a weighed quantity of oil was added. The oil was allowed to percolate down the column under mild vacuum (as indicated by the manometer in the water pump assembly). About 50 to 60 per cent of the terpenes could be eluted out without the need for a solvent and collected in tared receivers, which were changed by closing the stopcock, disconnecting the suction at the side tube and detaching the outer test tube. The residual terpenes, still left on the column

were eluted with a non-polar solvent (about 1.5 column volumes), and checked by TLC to ensure recovery. The terpene-free material was finally eluted with a polar solvent. The terpenes and the terpeneless fractions were recovered by distilling off the solvents on a water bath, and removing the last traces of solvent *in vacuo*.

Results and Discussion

The main factors which govern the efficiency of separation in the chromatographic process are (*a*) nature and particle size of adsorbent; (*b*) activity of silica gel; (*c*) adsorbent to adsorbate ratio; (*d*) mode and degree of packing; (*e*) height of adsorbent and elution rate; (*f*) eluting solvents; and (*g*) chemical inertness of adsorbent. With a view to achieving maximum economy without undue sacrifice of efficiency, the above variables were examined in some detail, with only column grades of silica gel instead of alumina which is known to promote undesirable chemical transformations.

Activity of adsorbent: Data in Table 1 indicate typical results for cold pressed lemon and lime oils showing the influence of the activity of the adsorbent on the economy of deterpenation. As expected the stronger the activity, the higher is the adsorptive capacity of the silica gel, particularly for the oxygenated (terpeneless) fraction. More oil can be deterpenated with silica gel of higher activity. The reduction in (percentage) yield at lower activity is indicative of incomplete separation, suggesting that active adsorbent always lowers the cost. The yield of terpeneless lemon oil obtained was 11.78 per cent for I*a* activity and 9.63 per cent for II*b* whereas the percentage yield of terpeneless lime oil was 20.16 for I and II*a* and 15.84 for>III*b* activity. It can be seen that the time taken for elution decreased with the decrease in the activity of the adsorbent used.

Adsorbent-oil ratio: The ratio of adsorbent to oil used for deterpenation is critical with respect to the yield and the cost. A high silica gel to oil ratio ensures complete separation of the terpenes from the terpeneless fraction and hence results in higher yields of the latter, but the process becomes uneconomical. Too low a ratio, on the other hand, leads to poor separation and consequent loss of the valuable oxygenated materials which are partly eluted with the terpenes.

Representative results obtained for the cold processed lemon and lime oils are presented in Table 2. The silica gel to essential oil ratio used in the experiments ranged from 10:1 to 1:2 whereas the percentage yield of oxygenated compounds for lemon oil ranged from 17.84 to 7.4 while it ranged between 27.97 and 10.35 in case of lime oils. Besides this the total time taken for elution increased with increase in silica gel to oil ratio. Next to activity of the adsorbent, the silica gel oil ratio is the most important factor affecting the economy and yield of (oxygenated compounds) terpeneless oils.

Mode and degree of packing: The dry packing of the column was preferred to the conventional packing in a non-polar solvent slurry from the standpoint of (*a*) economy, (*b*) greater separative efficiency reported for dry column chromatography, (*c*) convenience, and (*d*) lower operational time. The columns were packed to a moderate degree of tightness as tight packing although gives better resolution decreases flow rate in narrow columns.

It can be noticed from the Table 3 that no significant difference existed between the wet and dry methods of packing with regard to the yield of terpeneless fraction. Nevertheless, the dry method was found to be superior with respect to convenience and economics.

Diameter and height of column: Even though a long column of adsorbent is likely to have better resolving power with a lower silica gel to oil ratio, and a lower cost a slower elution rate from long columns would be undesirable. In Table 5 the percentage yield of terpene-

TABLE 1. EFFECT OF SILICA GEL-ACTIVITY ON THE YIELD OF TERPENELESS LEMON AND LIME OILS*

	Cold pressed lemon oil					Cold pressed lime oil			
Activity of silica gel	I*a*	I & II*a*	II*a*	II*b*	III*a*	I & II*a*	II*b*	III*a*	III*b*
Oil used (g)	20.05	20.05	02.04	20.03	20.06	25.06	25.07	25.19	25.08
Terpenes obtained (g)	17.25	17.50	17.50	17.80	17.80	19.51	19.55	20.08	21.71
Terpeneless oil (g)	2.36	2.08	2.03	1.93	1.62	5.05	4.72	4.61	3.97
Terpeneless oil (%)	11.78	10.40	10.10	9.63	8.08	20.16	18.84	18.31	15.84
Elution time (hr)	2.50	2.25	2.15	2.15	2.00	3.00	3.00	2.75	2.50

*A glass column 1.4×90 cm was packed with 50g of silicagel and operated at a pressure of 585 mm Hg, 250 ml each of polar and non-polar solvent were used for elution. The recovery of polar and nonpolar solvents in the various experiments ranged between 76 and 88 per cent.

TABLE 2. EFFECT OF SILICA GEL-OIL RATIO ON THE DETERPENATION OF LEMON AND LIME OIL*

	Cold pressed lemon oil								Cold pressed lime oil					
Silicagel to oil ratio	1:2	10:1	5:1	3.33:1	2.5:1	2:1	1:1	1:1.4	10:1	5:1	3.33:1	2:1	1:1	1:2
Oil used (g)	100.20	5.07	10.10	15.24	20.05	25.09	50.24	69.36	5.19	10.09	15.00	25.13	50.17	100.26
Terpenes (g)	91.60	4.00	8.37	13.20	17.25	22.20	43.90	61.57	3.37	7.72	11.42	19.52	41.01	89.09
Oxyg. compds. (g)	7.55	0.90	1.43	1.89	2.36	2.78	5.25	6.79	1.45	2.37	3.08	5.11	8.36	10.37
Oxyg. compds (%)	7.54	17.84	14.17	12.39	11.78	11.07	10.45	9.79	27.96	23.54	20.55	20.35	16.67	10.35
Elution time (hr)	3.50	2.50	2.50	2.75	2.75	3.00	3.00	3.25	2.50	2.25	2.50	2.75	3.25	3.75

*Experimental conditions same as in Table 1.

less fraction decreases as the diameter of the column increases—thus indicating the importance of keeping a proper height or diameter of the column for a given silica gel to oil ratio. With increased column height (60 cm) mild suction was necessary for ensuring a reasonable elution rate. The loss of volatiles under such conditions was practically nil as shown by the negligible condensate collected in a chilled (−15°C) trap interposed between receiver and the pump. Too high a suction however may cause a greater loss of volatiles, poorer separation due to coning of bands and eventual slower elution due to further packing.

It can be seen in Table 4 that the time of elution decreased with increase in diameter of the column used. Further, no vacuum was necessary for columns of diameter 3.25 cm. The diameter and height of the columns for a given silica gel-oil ratio, however, are not as important as the activity of adsorbent or the silica gel to oil ratio. For example, the percentage yield of terpeneless lime oil did not change much with the change in diameter and height of the column.

Efficiency of solvents: The cost of deterpenation depends to a significant extent on the solvents required for elution and on the extent to which they can be economically recovered and reused. By dry packing of active silica gel to the required height and application of mild suction, about 50-60 per cent of the terpenes filter through without the use of an eluting solvent. This itself represents a substantial saving of the solvent. The residual terpenes can be eluted off the column in about 2 to 3 column volumes of a non-polar solvent like cyclohexane, petroleum ether, or carbon tetrachloride which do not appear to differ much in relative efficiency. However, recovery of carbon tetrachloride and removal of its last traces presented some difficulties and its use is not favoured if the product is to be used in food flavours. Among the polar solvents methanol was found to be equally efficient but the removal of last traces is difficult and its presence in the final product is objectionable. High volatility, inflammability and ready tendency of peroxide formation are drawbacks which make ether less favoured in large scale applications and hence it

TABLE 3. MAXIMUM CONTENT OF TERPENES AND TERPENELESS (OXYGENATED) FRACTIONS IN LEMON AND LIME OILS*

	Cold pressed				Distilled	
	Lemon oil-A	Lemon oil-A	Lime oil B	Lime oil B	Lime oil-C	Lime oil-D
Experiments	1	2	3	4	5	6
Method used	Wet	Dry	Wet	Dry	Wet	Dry
Oil used (g)	10.11	5.07	10.02	10.02	10.03	10.09
Elution time (hr)	3.0	2.5	4.5	2.5	2.5	2.5
Terpenes obtained (g)	7.78	4.00	6.67	6.72	7.23	7.95
Oxyg. fraction (g)	1.83	0.90	2.82	2.84	2.52	1.98
Oxyg. fraction (%)	18.13	17.84	28.18	28.43	25.00	9.80

*In all these experiments silicagel of activity I & II*a* was used (100 g in expt. 1, 4, 5 and 6 and 50 g in 2 and 120 g in 3. The volume of polar solvent used was 540 ml in expt. 1, 250 ml in 2 and 500 ml in others and the percentage recovery was about 75%. The nonpolar solvent used was 770 ml in expt. 1, 200 ml in 2 and 500 ml in others and the percentage recovery was 90%. No vacuum was used for expts. 1& 3 while 585 mm Hg was used for others. The dia. of the column was 1.4 cm in expt. 2 while it was 2.25 cm for others.

TABLE 4. EFFECT OF COLUMN DIAMETER AND HEIGHT ON DETERPENATION OF LEMON AND LIME OILS*

	Cold pressed lemon oil						Cold pressed lime oil				
Diameter (cm)	1.4	1.4	1.7	2.5	3.25	3.25	1.4	1.7	2.5	3.25	1.7
Column ht (cm)	60	60	43	18	12	12	59	42	17	12	73
Oil used (g)	50.06	118.54	50.03	50.09	50.14	118.81	25.25	25.18	24.41	25.08	100.24
Terpenes (g)	43.96	110.36	43.50	43.75	43.50	111.00	19.53	19.59	19.54	20.01	81.14
Oxyg. compds (g)	5.20	7.18	5.10	5.10	5.06	6.81	5.12	5.09	5.03	4.72	19.10
Oxyg. compds (%)	10.40	6.06	10.18	10.16	10.11	5.74	20.27	19.97	19.82	18.82	19.05
Elution time (hr)	2.50	3.50	2.00	1.75	1.50	1.75	2.50	2.50	1.75	1.50	2.75

*50 g silicagel was used in all the experiments. 250 ml of polar and 250 ml of non-polar solvents were used for elution and their percentage recovery ranged between 75 and 85. No pressure was used for column dia. 3.25 cm while a pressure of 585 mm Hg was used for columns of other diameters.

was not included in these experiments. The recoveries of the solvents are fairly satisfactory and these can repeatedly be used for deterpenation. The final choice of the solvents will depend on relative costs and toxicity.

During deterpenation, cyclohexane, carbon tetrachloride and petroleum ether were used to elute the hydrocarbons while the oxygenated compounds were eluted with polar solvents like ethanol, methanol, acetone, or ethyl acetate. About 200 ml of polar solvent and 200 ml of non-polar solvents were necessary to deterpenate 50 g of either lemon or lime oil.

Efficiency of commercially available adsorbents: Experiments were carried out under similar conditions using 50-g lots of activated silica gels and alumina manufactured by five different firms to determine their efficiency. The efficiency can be judged from the percentage of the deterpenated material (Table 5). There was considerable difference in the total time taken for elution which ranged from 1.5 to 3.00 hr. These differences in efficiency observed may possibly be due to their particle size and/or the mode of manufacture.

The silica gel used for the deterpenation of lemon and lime oils was recovered and reused for deterpenation and the results showed (Table 6) that the recovered silica gel was as good as the fresh silica gel for this purpose. The used silica gel was recovered by boiling first with 1 per cent NaOH solution; washed with water, and again boiled with 5 per cent acetic acid and washed with water till free of acid, and finally activated by heating to 120°C.

Compounds containing certain labile or reactive functional groups are known to undergo chemical transformation in contact with certain highly activated adsorbents. It is important to ensure, therefore, inertness of the adsorbent used for deterpenation towards the components of the oil, otherwise off-flavours will

TABLE 5. DETERPENATION EFFICIENCY OF COMMERCIALLY AVAILABLE ADSORBENTS*

	Cold pressed lemon oil						Cold pressed lime oil				
Adsorbent used	SG-A	SG-B	SG-C	SG-D	SG-E	Alumina	SG-A	SG-B	SG-C	SG-D	SG-E
Column ht (cm)	60	62	52	55	68	34.50	40	43	38	39	46
Oil used (g)	50.24	50.00	50.00	50.00	50.02	50.02	50.13	50.17	50.27	50.27	50.36
Terpenes (g)	43.90	43.92	44.16	44.33	44.69	46.67	41.92	41.02	43.05	42.04	43.39
Oxyg. compds (g)	5.25	4.90	4.76	4.76	4.71	1.88	7.71	8.55	6.42	7.73	6.46
Oxyg. compds (%)	10.45	9.80	9.52	9.52	9.41	3.75	15.39	17.05	12.77	15.38	12.83
Elution time (hr)	3.00	3.00	2.00	1.50	1.50	1.00	2.50	2.50	2.50	1.75	1.25

*50 g. of adsorbent was used in all expts. and the activity of the silicagels was I & II*a* while alumina was I. A column dia. of 1.4 cm was used for lemon oil while 1.7 cm was used for lime oil. A pressure of 540 mm Hg was used for lemon oil while 585 mm Hg was used for lime oil. 250 ml each of polar and non-polar solvents were used for lime oil and 200 ml each of polar and non-polar solvents were used for lemon oil. The percentage recovery of the solvents ranged between 75 and 85.

SG—Silicagel from different commercial sources—A, B, C, D, E,

develop in the product. In the present experiments terpeneless products prepared on silica gel columns, indicated no objectionable aroma/flavour.

Alumina even with activity I was found inferior to silica gels with regard to the yield of terpeneless lemon and lime oils. The flavour and colour of the deterpenated oils from columns of alumina were found to differ very much from that of the silica gel columns. Deterpenated oils from alumina are poor in their flavour value and hence its use for this purpose is very much limited.

Analysis and flavour quality of lemon and lime oils: The results of analysis of lemon and lime oils are presented in Table 7. The maximum content of terpeneless fractions (oxygenated compounds) in lemon and lime oils used for this investigation can be seen in Table 3. As seen from the data the specific gravity, aldehydes, esters, acid number and evaporation residue of the lemon and lime oils increase considerably with increase in their terpeneless fraction (oxygenated compounds) whereas the optical rotation decreases with increase in their oxygenated compounds. Cold pressed oils contain certain quantity of waxes whereas the distilled oils are almost wax-free. Cold pressed lime oil used here contained 10.36 per cent wax and its terpeneless fraction contained about 37 per cent wax. The cold pressed lemon oil used for the experiments was almost free of waxes showing that it might have been winterized very well. Distilled oils are almost colourless whereas the cold pressed oils were light green, yellow or brown in colour. The aroma of the distilled oils was however poor when compared to the cold pressed oils.

The reasons for the variations in the analytical and flavour value of the lemon and lime oils can be attributed to the difference in their variety, season of harvest, maturity of the crop, methods of extraction, storage time and conditions, adulteration of the oils, etc.

Sensory evaluation of lemon and lime oils showed wide variations in their flavour quality. Distilled oils were the least preferred and cold pressed fresh oils were found extremely good in their flavour quality. By sensory evaluation it was difficult to differentiate between the cold pressed oils and their terpeneless counterparts when they were tested in comparable dilutions. For example, 0.2 ml of cold pressed lime oil, 0.05 ml of pure terpeneless lime oil and 0.1 ml of twice deterpenated lime oils were found to be more or less same in their flavour strength when they were tasted with 400 ml of

TABLE 6. DETERPENATION OF LEMON AND LIME OILS USING FRESH AND RECOVERED SILICA GELS*

	Fresh sillica gel		Recovered sillica gel	
	Lemon oil	Lime oil	Lemon oil	Lime oil
Oil wt. (g)	50.24	50.50	50.50	50.50
Terpenes (g)	43.90	42.27	43.70	42.17
Oxyg. compds (g)	5.25	7.14	5.31	7.14
Oxyg. compds (%)	10.45	14.14	10.50	14.14
Elution time (hr)	2.30	2.50	2.25	2.50

*Silica gel of activity I & II*a* was used in 50g. lots and a pressure of 585 mm Hg was used for these experiments. 250 ml each of polar and non-polar solvents were used for elution. The recovery of the solvents varied from 75 to 85%.

TABLE 7. PHYSICOCHEMICAL CONSTANTS OF LEMON AND LIME OILS ANALYSED AT 24 TO 26°C

	Sp. gr.	Opt. rotn	Ref. index	Aldehydes as citral (%)	Esters (%)	Acid No.	Evap. res. (%)	Solubility**	Oil colour***
Cold pressed lemon oil-A	0.8553	+72.71	1.4720	2.76	2.40	0.8202	2.35	Clear	LY
2 X-lemon oil from A	0.8716	+64.98	1.4740	11.98	4.87	2.776	6.04	Turbid	LY
Terpeneless lemon oil from A	0.9301	+ 1.472	1.4805	42.37	16.90	6.510	22.15	,,	Y
Terpeneless lemon oil with 5% cold pressed oil	0.9239	+ 5.44	1.4785	41.55	12.21	7.818	23.22	,,	Y
Lime oil-B	0.8646	+38.84	1.4750	1.37	1.25	0.7216	2.53	Clear	LY
Terpeneless-lime oil from B	0.9592	- 0.900	1.4825	6.09	5.42	5.648	19.36	,,	Y
Cold pressed lime oil-C	0.8818	+53.60	1.4840	4.85	4.72*	1.296	14.17	,,	Y
Terpeneless lime oil from C (dewaxed)	0.9750	Nil	1.5100	23.90	14.83	6.343	54.68	,,	DB
50% deterpenated lime oil from C	0.9315	+22.26	1.4980	9.879	7.80	3.784	33.81	,,	LB
Terpeneless lime oil from C (with wax)	1.0210	- 0.9149	1.5260	19.98	17.06	7.991	66.07	,,	DB

*difficult end point

**solubility tests were done at 1:3 dilution in 95% alcohol.

***Y = Yellow, LY = Light yellow, DB = Dark brown, LB = Light brown.

12 per cent sugar solution containing 0.2 per cent citric acid. This shows that the terpeneless oils have highly concentrated flavour strength and can be used in smaller quantities.

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RESEARCH NOTES

MINOR SEED OILS. XIII. EXAMINATION OF SEED OILS RICH IN LINOLEIC ACID

Five seed oils from *Artobotrys odoratissimus* (N. O. Annonaceae), *Lagascea mollis* (N. O. Compositae), *Mimosa pudica* (N. O. Leguminosae), *Solanum trilobatum* and *Solanum verbascifolium* (N. O. Solanaceae) were examined by reversed phase partition column chromatography for their component fatty acids. The content of linoleic acid ranges from 42.2 to 60.1%, and of oleic acid from 10.9 to 27.8%, while palmitic (12.9-17.6%) and stearic (3.2-11.9%) acids are the major component saturated acids. *M. pudica* contains 5.7% behenic acid.

Artobotrys odoratissimus,[1] R. Br. is a shrub, often scandent, distributed in South India, Ceylon, Java and China. The seeds are oblong, a little flattened, deeply grooved on one side and more than 13 mm long. The yellow flowers are acrid, bitter, heating, useful in vomiting, biliousness, diseases of the blood and the heart, leucoderma, headache, etc. A decoction of the leaves is given for cholera in some of the islands of the Malaya Archipelago.

Lagascea mollis,[2] Linn. is a genus of about seven shrubs or herbs native to Mexico and Central America. *L. mollis* is a velvety annual, not particularly valuable horticulturally. Though an exotic plant, once introduced, it has established itself permanently as an important constituent of the local flora.

Mimosa pudica,[3] Linn (English: Touch-me-not) is a diffuse under-shrub, the stems and branches being sparingly prickly and clothed with long weak bristles, and the leaves sensitive. The root is bitter and acrid, cooling and vulnerary, and cures biliousness, leprosy, dysentery, etc. The seeds are used as an emetic. The leaves are prescribed in cases of piles and fistula. An infusion of the leaves is given as a bitter tonic.

Solanum trilobatum,[3] Linn. is a much-branched climbing shrub commonly found in the Deccan peninsula. The flowers are pretty, purple-blue or violet purple, the berries globose, red or scarlet and the seeds smooth or slightly pitted. The bitter roots are used for consumption in the form of an electuary, decoction or powder. The berries and flowers are administered for treatment of cough. The decoction of the various parts of the plant is used in chronic bronchitis. Leaves are cooked and eaten as a vegetable. The steroidal alkaloid solasodine is present in the fruits and leaves.

Solanum verbascifolium,[5] Linn. is a shrub or small tree frequently met with throughout tropical and subtropical India and the Andamans. It is also cultivated in South India for its fruits which are eaten in curries. Roots of the plant are reported to be used in Malaya as a compound decoction for curing violent pains all over the body, accompanied by discomfort after meals. Leaves are given to women suffering from vaginal discharge. The glyco-alkaloid solasonine is present in the fruits and leaves of the plant.

The seeds were collected from the botanical gardens of the Karnatak University, Dharwar and College of of Agriculture, University of Agricultural Sciences, Dharwar. The oil was soxhlet extracted from the crushed seeds using light petroleum. The fatty oil, unsaponifiable matter and the mixed acids were obtained as described earlier.[6]

The mixed fatty acids as such, after hydrogenation and after oxidation[7] were each examined by reversed phase column chromatography as described earlier.[8] The presence of oleic and linoleic acids was confirmed by preparing their corresponding hydroxy and bromo derivatives from the acids isolated from the appropriate chromatographic fractions.[9]

TABLE 1. CHARACTERISTICS OF THE OIL

Seed species	Oil content (%)	Unsap. matter (%)	Iodine value* Obs.	Iodine value* Calc.	Sap. eqv.* Obs.	Sap. eqv.* Calc.	Refr. index at 30°C	Protein**
Artobotrys odoratissimus	7.3	1.4	113.5	112.3	271.5	273.9	1.4676	—
Lagascea mollis	22.1	0.8	116.2	116.8	276.5	276.2	1.4701	17.10
Mimosa pudica	2.2	1.8	103.8	104.6	275.3	274.8	1.4658	28.61
Solanum trilobatum	18.5	0.6	111.8	112.2	275.4	275.7	1.4699	13.81
Solanum verbascifolium	21.0	0.8	105.9	105.5	275.1	275.4	1.4662	14.43

*Values measured on mixed acids free from unsaponifiable matter and calculated from the composition determined in this investigation.
**In seeds on dry basis; multiplication factor 6.25.

TABLE 2. CHROMATOGRAPHY RESULTS (MOLE %) ON PARAFFIN COLUMNS

Seed species	Treatment of mixed acids*	Load mg	Recovery %	Eluting solvents (% aqueous acetone)							
				35	43	53	62	67	73	78	83
	H.A.	13.7	88.1	—	2.2	1.5	2.0	14.2	77.6	1.6	0.9
A. odoratissimus	M.A.	18.7	83.1	—	2.8	1.3	55.4	30.2	7.7	1.6	1.0
	O.A.	95.2	—	—	—	—	1.5	14.1	7.8	1.3	0.8
	H.A.	14.2	99.4	—	1.7	0.8	1.0	13.9	78.8	2.4	1.4
L. mollis	M.A.	19.7	100.0	—	1.2	0.6	60.2	24.6	11.6	1.2	0.6
	O.A.	92.4	—	—	—	0.4	0.8	13.6	11.7	1.2	0.8
	H.A.	19.6	99.5	0.9	1.1	1.2	1.3	18.7	71.2	2.4	3.2
M. pudica	M.A.	20.8	98.7	1.7	1.3	1.5	51.8	33.3	3.9	1.9	4.6
	O.A.	88.2	—	—	—	—	1.1	18.7	3.6	1.3	3.1
	H.A.	14.8	97.2	—	—	0.5	0.9	21.2	76.4	0.4	0.6
S. trilobatum	M.A.	19.8	99.4	—	—	0.6	49.7	46.0	3.1	0.2	0.4
	O.A.	83.2	—	—	—	0.6	1.2	18.9	5.1	0.3	0.4
	H.A.	18.2	98.0	0.7	0.5	1.0	1.3	15.2	79.9	0.7	0.7
S. verbascifolium	M.A.	20.6	97.8	0.8	0.4	1.2	47.6	39.3	9.2	0.6	0.9
	O.A.	91.5	—	—	—	2.0	1.6	16.1	7.9	1.0	1.0

*H.A.—Hydrogenated acids; M.A.—Mixed acids without any treatment; O.A.—Oxidised acids.

TABLE 3. COMPONENT FATTY ACIDS (WEIGHT %)

Seed species	Fatty acids									
	8:0*	10:0	12:0	14:0	16:0	18:0	20:0	22:0	18:1	18:2
A. odoratissimus	—	1.8	0.9	1.2	13.2	8.0	1.8	1.3	16.6	55.2
L. mollis	—	0.8	0.5	0.8	12.9	11.9	1.4	0.7	10.9	60.1
M. pudica	0.9	0.7	0.9	1.1	17.5	4.0	2.7	5.7	15.0	51.5
S. trilobatum	—	—	0.4	1.0	17.6	3.2	0.2	0.5	27.8	49.3
S. verbascifolium	0.4	0.2	0.9	1.0	14.2	9.5	0.8	1.1	24.7	47.2

*These figures indicate the number of carbon atoms and the number of double bonds respectively, in the acids.

TABLE 4. SEED OILS OF MIMOSA SPECIES

Fatty acids	*M. pudica* present work	*M. pudica* Aggarwal *et al.*[10]	*M. invisa* (var. intermis) Gunstone *et al.*[11]
Caprylic	0.9	—	—
Capric	0.7		—
Lauric	0.9	—	—
Myristic	1.1	—	—
Palmitic	17.5	8.7	16.0
Stearic	4.0	8.9	2.0
Arachidic	2.7	—	—
Behenic	5.7	—	—
Oleic	15.0	31.0	13.0
Linoleic	51.5	51.0	68.0
Linolenic	—	0.4	—

Seeds of *Lagascea mollis*, *Solanum trilobatum* and and *S. verbascifolium* are fairly rich in oil, 22.1, 18.5 and 21.0 per cent respectively. Protein content in the seeds varies from 13.8 to 28.6 per cent. All these oils have an iodine value of more than 100.

Linoleic acid is the major component acid in all these seed oils; it ranges from 47.2 to 60.1 per cent, *S. verbascifolium* carrying the least and *L. mollis* the highest. Oleic acid ranges from 10.9 per cent for *L. mollis* to 27.8 per cent for *S. trilobatum*. Palmitic (12.9–17.6 per cent) and stearic (3.2–11.9 per cent) acids form the major saturated component acids. *S. trilobatum* and *L. mollis* contain highest amount of palmitic and stearic acids respectively. *Mimosa pudica* contains, along with other acids, about 2.7 per cent arachidic and 5.7 per cent

behenic acids. Small amounts of lower saturated acids (8:0 to 12:0) were also found in some of the above oils.

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GINGER FROM HILLY REGIONS OF INDIA

Ginger varieties grown in the hilly regions of India are abnormally plumpy, containing over 85 per cent moisture. When dried, however, they give normal yields of oil and oleoresin.

The rhizome of the ginger plant, *Zingiber officinale* Roscoe, is generally used in the green (fresh) form for culinary purposes in India. Even those varieties which are specially grown for making dry ginger are sometimes sold in green form, when the price is more attractive. However, depending on the demand from abroad, some six to seven thousand tons of dry ginger are regularly produced in India, chiefly in Kerala. A few varieties which give high yield of dry ginger (23 to 28 per cent on weight of green ginger), with bold attractive 'fingers' are used for drying.

Ginger is grown in most of the states in India and we have had occasion to examine samples from different regions, in connection with a project on production of ginger oil and oleoresin.[1-3] We were particularly struck by the giant size of the rhizomes grown in hilly regions like Kalimpong, Sikkim and Arunachala Pradesh. To some extent this is true of the *Rio-de-Janeiro* variety also grown in Nilgiri region of South India.

These rhizomes appear abnormally plumpy and generally contain over 85 per cent water. They are evidently not suited for drying, since the yield of dry material is low, and the product is shrivelled and very unattractive in appearance. They are also often high in fibre content. The harvested material is sold as green ginger, but its keeping quality is poor.

It is possible that the giant size and high water content of such ginger is due to genetic and agro-climatic reasons.

Since the material is perishable, and the prices are often very unattractive, we have examined the possibility of using dried ginger from these regions for production of ginger oil and oleoresin. Two to three samples of each variety have been analysed by us. Typical analytical data for some varieties are given in Table 1. The values for 'Wynad', a variety specially used for drying in Kerala, are also given for comparison. Volatile oil was determined by the Clevenger distillation method.[3] Oleoresin yield was determined by cold percolation extraction of the ground spice in small glass columns, using ethylene dichloride as solvent.[5]

The yields of oil and oleoresin from the dried material, as also their quality were good. If the green ginger is available at a sufficiently cheap cost from those hilly regions, it would be economical to use them for production of oil or oleoresin.

TABLE 1. ANALYSIS OF HILL GINGER VARIETIES

Region	Yield of dry ginger (%)	Dry ginger	
		Volatile oil (%)	Oleoresin (%)
Sikkim	19	2.4	5.1
Arunachal Pradesh	20	1.4	5.0
Garo Hills	17	0.9	4.2
Manipur	20	2.1	4.8
Rio-de-Janeiro (Nilgiris)	17	2.2	5.3
Kalimpong	18	2.5	5.0
Wynad	28	1.5	4.9

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A MODIFIED GRAPHICAL INTERPOLATION METHOD FOR RAPID DETERMINATION OF WATER ACTIVITY IN FOODS

The water activity (a_w) of intermediate and low moisture foods can be determined by exposing the samples in desiccators containing saturated salt solutions of different relative humidities and by graphical interpolation. The point where the curve cuts the datum line represents the a_w of the sample, and the value obtained at the end of 2 hr compares well with the value obtained by exposure for long periods or Wink's weight equilibrium method.

Intermediate moisture (IM) foods have been gaining importance in recent times. The primary problem in their preservation is to maintain water activity (a_w) at levels low enough to prevent the growth of bacteria and other food spoilage organisms. During our work on the development of IM fruits and vegetables for defence use, we felt the need for a rapid and inexpensive method of measuring water activity.

Of the various methods, the weight equilibration method of Wink[1] gives satisfactory results but is time consuming. In the graphical interpolation method proposed by Landrock and Proctor[2], although rapid and simple, the normality of sulphuric acid used to provide atmospheres of different relative humidities (RH) must be maintained rigorously. The use of saturated salt solutions employed by Wink[1] instead of sulphuric acid and the graphical interpolation technique of Landrock and Proctor[2] eliminated the drawbacks of both the methods and combined their advantages which resulted in a reliable and rapid method for use in the determination of a_w.

IM fruit and vegetable pieces[3-6] and low moisture foods such as instant tomato and chicken soup powders[7] and omelette mix[8] used in the present study were prepared according to the methods reported earlier. Ten gram lots of the comminuted/powdered sample of known initial moisture content were spread uniformly in tared petri dishes, weighed accurately and exposed to different RHs ranging from 0 to 98 per cent at room temperature (25–30°C) in desiccators (or glass bowls with air tight ground glass lids) containing saturated salt solutions having definite relative humidities. The gain or loss in weight of the samples after 2, 4 and 6 hr, and thereafter, at intervals of 24 hr till it reached equilibrium (no further loss or gain) was recorded. The equilibrium moisture content of the sample at each RH was also determined by estimating its moisture content after equilibration.

The loss or gain in weight of the sample at the end of 2, 4 and 6 hr was plotted against RH by the graphical interpolation method as shown in Fig. 1. The point where the curve intersects the zero line represents the equilibrium relative humidity (ERH) of the sample.

TABLE 1. WATER ACTIVITY OF SOME INTERMEDIATE AND LOW MOISTURE FOODS AS DETERMINED BY THE MODIFIED GRAPHICAL INTERPOLATION METHOD AND WINK'S WEIGHT EQUILIBRATION METHOD

Sample	Moisture (%)	Water activity (a_w)					
		Modified graphical interpolation method					Wink's wt. equilibration method
		2 hr.	4 hr.	6 hr.	24 hr.	48 hr.	
Intermediate moisture foods							
Mango	33.0	0.775	0.775	0.775	0.780	0.780	0.775
Banana	35.0	0.800	0.800	0.800	0.790	0.790	0.800
Carrot	40.0	0.755	0.755	0.760	0.750	0.750	0.760
Low moisture foods							
Tomato soup powder	3.5	0.240	0.240	0.230	0.240	0.250	0.240
Chicken soup powder	3.8	0.180	0.180	0.180	0.190	0.180	0.180
Omelette mix	2.1	0.255	0.260	0.260	0.260	0.260	0.260

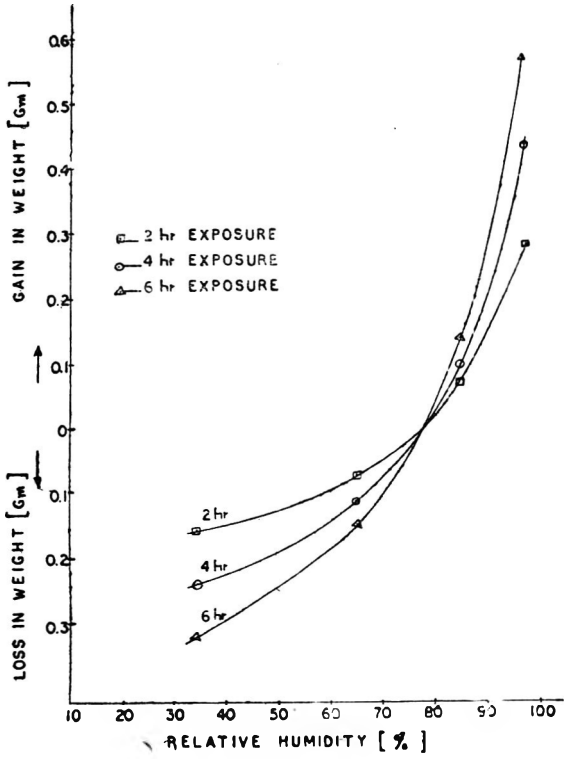

Fig. 1. Graphical interpolation isotherm for IM mango at room temperature for different times of exposure by modified graphical interpolation method.

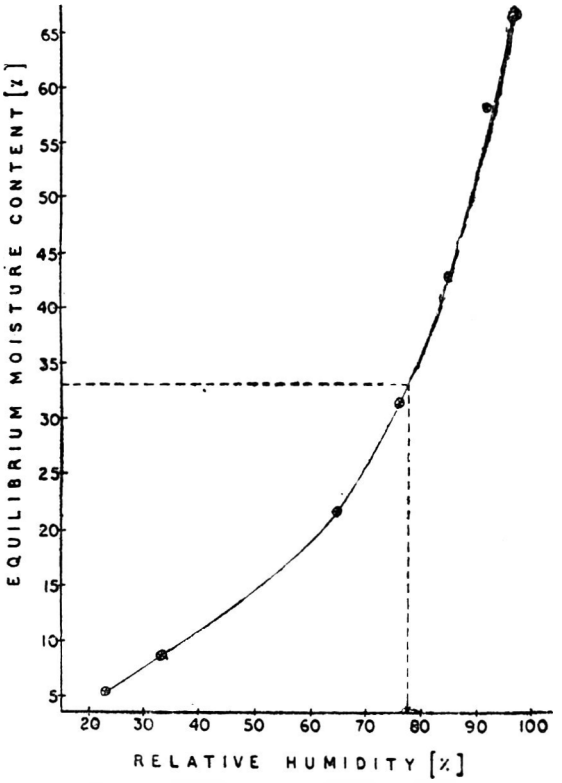

Fig. 2. Humidity equilibrium curve for IM mango at room temperature by Wink's weight equilibration method

From Fig. 1 and data in Table 1, it is obvious that irrespective of the time, the graphical interpolation curves cut the zero line at the same point. The ERH so obtained compares well with the values from the weight equilibrium method (Fig. 2). From the result, it is obvious that the a_w (i.e., ERH/100) of IM and low moisture foods can be determined by exposure for 2 hr by the modified graphical interpolation method outlined in the present study.

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ORGANOLEPTIC EVALUATION AND NUTRITIVE VALUE OF RECIPES OF SOYMILK AND SOYRESIDUE

Soymilk preparations received fairly high grading upon organoleptic evaluation. Pudding was rated almost at par with standard (cowmilk) pudding. Custard and milk shake prepared from soymilk were also very palatable. Use of soymilk in various recipes could effect estimated savings of upto 50% in the cost of preparation, compared to standard recipes. The solid material remaining after soymilk preparation (soyresidue) could also be effectively used for improving the nutritive value of traditional snacks like *Samosa*. Soyresidue snacks are 25-33% cheaper than the standard preparations.

Results of an investigation on standardization of soymilk preparation and its organoleptic properties and chemical composition have been reported in earlier communications[1,2] from this laboratory. Soyresidue contains protein of high biological value[3]. It was

considered worthwhile to evaluate the organoleptic properties, nutritive value and cost of preparation of popular snacks after incorporation of soymilk or soyresidue.

Soymilk with desirable organoleptic properties was prepared from *Bragg*, *Pb*-1 or *JS*-2 varieties of soybean using the improved method[1] in which the original hot grinding-cum-rapid hydration technique has been modified to ensure complete removal of the offensive beany flavour. Thus, soaking time was reduced to 3 hr; the soak water containing a considerable proportion of the soluble protein was used for homogenization, and the final volume was adjusted (W/V) in the ratio of 1 part soybean to 6 parts water. The average protein content (4 per cent) in soymilk so prepared compares favourably with that of cowmilk (3.7 per cent). The estimated cost of production was only 60 paise per litre, compared to Rs. 3 per litre for milk[2].

Soymilk was used in the preparation of pudding, ice cream, custard and milk shake, and the organoleptic (sensory) evaluation of soymilk recipes against standard cowmilk recipes was done by a panel of six judges familiar with the tasting techniques. Two replicates of each preparation were used. The ten-point scale, *viz.* ideal (9-10), excellent (8-8.9), very good (7-7.9), fairly good (5-5.9), acceptable (4-4.9) and not acceptable (<4) was used. Air dried samples of soyresidue—obtained as a byproduct of soymilk preparation—contained 52.5 per cent protein[3]. Organoleptic testing of soyresidue recipes was done by another panel of six judges, the criteria being appearance, taste, flavour and texture. The ten point scale used was, ideal (10), excellent (9-9.9), very good (8.8.9), good (7-7.9), fairly good (6-6.9), acceptable (5-5.9) and not acceptable (<5). Two replicates of each preparation were taken. The data were analysed as per 6×2 factorial design.

The results of organoleptic scoring of soymilk recipes *vs* standard cowmilk recipes have been summarised in Table 1 and organoleptic scores of soyresidue substituted recipes *vs* standard recipes in Table 2.

Soyflour is generally mixed with wheatflour for enrichment of bread in the U.S.A. Kanthamani[5] used soybean and soyflour in several recipes but did not evaluate the organoleptic characteristics of these preparations against standard recipes. Results of an investigation on the effect of soyflour incorporation upon nutritive value and palatability of items of daily diet such as chapati, *dal*, *dhokla*, *usal*, etc. have been reported elsewhere[6].

In the present study both soymilk pudding and standard pudding were graded very good in all the characteristics. However, statistical analysis showed

TABLE 1. ORGANOLEPTIC SCORE† OF SOYMILK RECIPES *vs* STANDARD RECIPES

Recipes	Appearance	Consistency	Palatability	Flavour
Pudding				
Soymilk	8.01 VG	8.14 VG	8.34 CG	8.35 VG
Standard	8.91**VG	8.79**VG	8.75**VG	8.58^{NS}VG
SE	0.15	0.15	0.12	0.11
Ice cream				
Soymilk	7.38 G	7.88 G	7.84 G	7.64 G
Standard	9.45**E	9.33**E	9.58**E	9.44**E
SE	0.18	0.28	0.18	0.18
Custard				
Soymilk	8.12 VG	8.42 VG	8.35 VG	8.24 VG
Standard	9.29**E	8.79^{NS}VG	9.04* E	8.95* VG
SE	0.22	0.42	0.22	0.22
Milk shake				
Soymilk	7.60 G	8.01 VG	7.31 G	7.48 G
Standard	8.62**VG	8.70* VG	8.75**VG	8.66**VG
SE	0.23	0.23	0.29	0.27

NS Non-significant † Average of two replicates
E Excellent; VG Very good; G Good
**P < 0.01 *P < 0.05

TABLE 2. ORGANOLEPTIC SCORE† OF SOYRESIDUE RECIPES *vs* STANDARD RECIPES

Recipes	Appearance	Flavour	Palatability	Texture
Cake				
Soyresidue	7.03 G	7.92 G	8.06 VG	6.98 FG
Standard	8.57* VG	8.19* VG	9.12* E	8.58* VG
SE	0.24	0.57	0.59	0.84
Khurma				
Soyresidue	7.0 G	7.75 G	7.75 G	7.79 G
Standard	7.8 ^{NS}G	7.21^{NS}G	7.59^{NS}G	7.3 ^{NS}G
SE	0.1	0.65	0.05	0.06
Sev				
Soyresidue	4.74 A	5.66 A	6.38 FG	4.57 A
Standard	8.16**VG	8.00**VG	8.14**VG	8.25**VG
SE	1.04	0.47	0.57	0.34
Samosa				
Soyresidue	5.38 A	6.69 FG	5.63 A	7.16 G
Standard	8.50**VG	8.08**VG	8.45**VG	8.91**VG
SE	0.037	0.049	0.58	0.09

VG Very good; G Good; FG Fairly good; A Acceptable
** P < 0.01 * P < 0.05
NS Non-significant † Average of two replicates

the superiority of the latter (significant at 1 per cent level) in all the traits, except flavour (Table 1). Soymilk ice cream was rated good and standard ice cream, excellent. Soymilk custard was adjudged very good, and standard custard excellent. The differences were statistically significant except for consistency. Though rated good in all traits soymilk ice cream failed to give the smooth feel of well prepared standard ice cream because of formation of ice crystals. Unless remedied, this would come in the way of its popular acceptability. Possibly, use of blended milk (soymilk 4 parts and cowmilk 1 part) and addition of cream might improve the palatability of this product. The cost of blended milk (estimated at 72 paise per litre[2]) is considerably less than that of whole milk. Soymilk custard and shake have proved quite successful. These preparations could be used also by families with moderate income because of low cost and relative ease of preparation. It is pertinent to state that the electric blender could be replaced by the ordinary household stone grinder (*sil batta*) in soymilk preparation.

Soyresidue incorporated cake was found very good in palatability, whereas standard cake was rated as excellent. Soyresidue *samosa* was just acceptable, while standard *samosa* received the very good rating. Soyresidue substituted *sev* and standard *sev* obtained fairly good and very good grade, respectively. In all organoleptic traits soyresidue *khurma* closely approached standard *khurma*, both preparations having been rated as good.

Use of soymilk recipes in cafetaria and homes would be economic and help to relieve the pressure on milk, particularly during the lean summer months. It may be noted that soyresidue incorporation also increased the protein content of the resultant snacks markedly, two-fold in cake and *samosa* and four-fold in *halwa*.

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EFFECT OF 'ENSTAR', A JUVENILE HORMONE ANALOGUE ON THE LARVAE AND PUPAE OF RED FLOUR BEETLE *TRIBOLIUM CASTANEUM* (HERBST)

Effect of 'Enstar' (Prop—2—ynyl (2E, 4E)—3, 7, 11—trimethyl—2, 4—dodecadienoate) a new Zoecan insect growth regulator was studied on the last instar larvae and 24-48-72 hr old pupae of *Tribolium castaneum*. Larvae were treated indirectly by feeding them on the treated food and pupae were tested by topical application (μl/pupa) on the thorax. In addition to the supernumerary larval moults, toxicity was observed in the larvae about to pupate and in pupae freshly moulted from larvae.

The use of Juvenile Hormone Analogues (JHA) as a new approach to the control of insect populations had attracted considerable interest in the area. In recent times many such compounds mimicking the juvenile hormones are being reported. In the present communication 'Enstar' (Prop—2—ynyl (2E, 4E)—3, 7, 11—trimethyl—2, 4—dodecadienoate) a new Zoecan insect growth regulator* which is reported to be highly active against homopteran insects is used against the larvae and pupae of *Tribolium castaneum* Herbst.

In the present study about 60 last instar larvae were selected for each experiment. They were divided into 3 batches of 20 each and introduced on to 3 g of bread wheat flour treated with 1 ml of test solution. The test solutions were prepared by dissolving 0.5 mg and 1.0 mg of JHA in 1 ml of acetone. Control experiments were carried out with equal number of larvae by treating the same weight of flour with 1 ml of acetone solution. Simultaneously larvae were also maintained on stock media for the sake of comparision. Observations were made periodically at the end of 1st, 2nd, 3rd and 4th week as shown in Table 1. Pupae of 24, 48 and 72 hr were treated topically on the thorax with 0.005, 0.01, 0.025, 0.05 and 0.1 μg/μl of JHA dissolved in acetone. An agla microsyringe fitted with a micrometer was employed for this purpose to assure a constant appli-

*ZR 777, Zoecan Technical Bulletin., Palo Alto, California, U.S.A.

TABLE 1. EFFECT OF 'ENSTAR' (JHA) ON THE LAST INSTAR LARVAE OF *Tribolium castaneum*

		% of larvae/pupae/adults found after the indicated weeks			
		I	II	III	IV
JHA 0.5 mg/ml.	Larvae	82	80	80	80*
	Pupae	18	13	—	—
	Adults	—	—	—	—
JHA 1.0 mg/ml.	Larvae	95	95	55	50*
	Pupae	5	—	—	—
	Adults	—	—	—	—
Control (acetone)	Larvae	50	25	10	—
	Pupae	50	25	15	—
	Adults	—	50	75	100
Stock medium (diet)	Larvae	20	—	—	—
	Pupae	80	40	—	—
	Adults	—	60	100	—

*Supernumerary moults.

cation rate. About 20 pupae in triplicate were used for each concentration and 20 puapae were treated with equal quantity of acetone as control. Since the moulting of pupae into adults usually takes 6-8 days, results were recorded on the 7th day, the per cent mortality was calculated (Abbott)[1] and is presented in Table 2.

Extra larval instars and an indefinite postponement of the pupal moult were observed when the last instar larvae were fed on JHA treated food (Table 1). By the end of 1st week nearly 80 per cent in stock medium and 50 per cent in control had moulted into pupae while in the treated only 5-18 per cent moulted. By the 2nd week more than 50 per cent in control and stock media had emerged into adults while treated larvae remained unmoulted. By 4th week 50-80 per cent of the treated remained as larvae with an increase in the body size (0.15-0.2 cm more than the normal) indicating thereby supernumerary moulting; the rest were not found either in pupal or adult form. However, there was 100 per cent adult emergence in the control and in stock medium batches. It has been reported[2] that JHA may indirectly stimulate the prothoracic gland to secrete moulting hormone somewhat prematurely which affects normal development resulting in more larval instars. Present studies indicate that the larvae treated with JHA could not successfully moult into pupae and during moulting they died. Therefore no pupae and adults were found at any stage during 2nd to 4th week of the experiment. This was more pronounced with higher dose of JHA (0.1 mg/ml). Acetone was found to have no detrimental effect on the larvae.

TABLE 2. LETHAL EFFECTS OF THE JHA TO 24-48-72 HR. OLD PUPAE OF *Tribolium castaneum*

JHA concn (μg/μl/pupa)	Corrected % mortality after 24 hr	48 hr	72 hr
0.005 ..	1	—	—
0.01 ..	17	15	—
0.025 ..	78	36	10
0.05 ..	94	45	20
0.1 ..	100	68	30

Pupae of 24-48-72 hr when treated with different concentrations of JHA showed early pupae as having more of lethal effects than the Juvenoid effects. They were found completely shrunken and dried up. The 48-hr old pupae also showed mortality but to a lesser degree. However, 72-hr pupae were mostly unaffected and were found to be normal (Table 2). Thus a close relationship between the age of the pupae and the effectiveness of JHA was observed. No deformity or gaint forms were observed in the adults moulted from the last pupae. Such sensitivity of early pupae to JHA is attributed to the replication of DNA during that period[3,4] which is known as the critical phase of the life cycle.

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CONVENIENT DEVICES FOR HANDLING HOUSE FLIES FOR TOPICAL APPLICATION OF PESTICIDES

An easy to construct fly-holder and a modified vacuum pencil are described which simplify the otherwise cumbersome method of topical application of pesticides to houseflies. These devices permit a single operator to carry out the procedure in shorter time.

The technique of topical application using houseflies has been shown to be useful in assaying pesticide residues in or on food materials.[1] The method is unique, in which a known dose of toxicant is applied to individual insects, thus requiring fewer insects and replicates. However, a major difficulty encountered in the procedure is the handling and treatment of insects individually, which is time-consuming and requires a two-member team.[2] Capillary tubes used as vacuum pencils allow the pick-up of individual insects. Release of the treated insect from the vacuum pencil was achieved by manual pinching of the suction line[3]; a foot operated pedal[4] and a knee-operated valve or an extra air line reversing the suction pressure.[5] A narrow glass tube similar to that used by Kerr[3] to orient the head of the fly onto the vacuum pencil, allowed only 10-15 flies to be treated, and was found unsatisfactory due to the condensation of moisture within the tube resulting from the respiration of the flies.

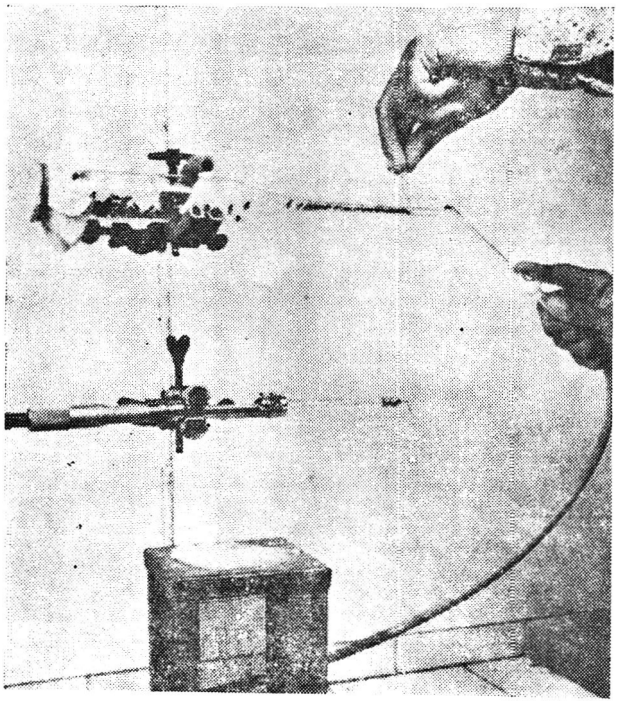

Fig. 1. Laboratory set-up of the devices with fly holder (above) and microsyringe (below) fitted to the stand. On the right is the vacuum pencil.

This communication describes a new design of a fly-holder and vacuum pencil. The fly-holder permits easy and continuous pick-up of 50-70 flies by a vacuum pencil, to be topically treated by a microsyringe and at the same time avoids moisture condensation. The vacuum pencil simplifies the release of the treated insect, without additional attachments to the suction line. These devices are very convenient and time saving, allowing a single operator to carry out the procedure.

The fly-holder is made by drawing one end of a glass tube 14 cm long and 29 mm internal diameter and fusing it with another 9 cm long tube and 5 mm internal diameter. Towards the free end of the narrower tube, a cross slit is made, into which a V-shaped plastic strip (thickness, 1-2 mm) is fitted to act as a gate. The length of the narrower tube behind the gate provides space for a single file of 8-10 flies, avoiding over crowding. This feature and the muslin cloth used to cover the open end of the holder, prevents the condensation of moisture inside.

The vacuum pencil is made by drawing out one end of a glass tube of 1.5 mm internal diameter to a bore of 1 mm and flame polished. The other end is fused to another tube of 6 mm internal diameter through a bulb of 15 mm internal diameter provided with a pressure releasing hole of 6 mm diameter. The vacuum pencil is connected to a water suction pump via a vacuum gauge and a stop cock regulator by means of which, the vacuum is regulated between 156 and 380 mm Hg. Higher vacuum was found to be injurious to the flies.

A laboratory set up of the device is shown in Fig. 1. The mounting of the fly-holder and the 'Agla' microsyringe on the same stand permits a single person to operate both the fly-holder and microsyringe with one hand, leaving the other hand free to manipulate the vacuum pencil. The fly-holder is directed towards a source of light so that the flies move into the narrower tube. The gate on the fly-holder is raised to release a single fly, which is held by the head capsule with the vacuum pencil by closing the pressure releasing hole with a finger. After treating the fly topically by the microsyringe, it is ejected into the respective cage by withdrawing the finger from the pressure releasing hole.

The devices described are simple, easy to construct and inexpensive. With the procedure outlined, a single operator can treat 50 flies in about 10 min.

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PROTEOLYTIC ACTIVITY OF TWO *LACTOBACILLUS* SPECIES AND THEIR GAMMA-RADIATION INDUCED MUTANTS

Two gamma radiation-induced mutants of *Lactobacilli*, selected on the basis of their pronounced proteolytic activity were examined for their ability to degrade different caseins and other protein substrates. Both the mutant and parent cultures degraded cow casein and crossbred cow casein much better than either buffalo or goat casein. However, the mutants degraded the above substrates much faster than their parents. Some variations were noted in the breakdown of other protein substrates by the cultures.

The role of *Lactobacilli* in the degradation of cow casein has been extensively studied.[1,2] The degradative ability of *Lactobacilli* on other casein species and on non-milk protein substrates has not been thoroughly investigated except for a few recent reports on buffaloe's[3,4] and soy milk.[5,6] Earlier studies in this laboratory[7] have dealt with the development of mutants of *Lactobacilli* with increased proteolytic activity after exposure of the cultures to chemical or physical mutagenic agents. The present report deals with the degradative ability of two such mutants and the respective parent cultures on different protein substrates including cow casein.

The cultures used were *Lactobacillus bulgaricus* 59 and *L. casei* RTS and one gamma radiation-induced mutant of each of these cultures, selected earlier on the basis of increased proteolytic activity. Different acid caseins were prepared from samples of skim milk drawn from cow (Red Sindhi) buffalo (Murrah), goat (Alpine) and crossbred cow (Karan Swiss) by isoelectric precipitation according to the method of Gupta and Ganguli.[8] Solutions of individual casein substrates were prepared by dissolving 1.0 g of the substrate in a minimum amount of 0.1 N NaOH and the pH in each case was adjusted to 7.0 with 0.1 N HCl. The final volume of each substrate was made up to 100 ml with citrate buffer (0.01M; pH 7.0).

The other protein substrates such as lactalbumin, bovine serum albumin, egg albumin, peanut protein and soyabean protein were examined in this study. Individual protein substrates were prepared by dissolv-

TABLE 1. DEGRADATION OF DIFFERENT TYPES OF CASEINS BY TWO LACTOBACILLUS SPECIES AND THEIR MUTANTS (mg. OF TYROSINE LIBERATED/g)**

Culture	Type of caseins*	Period of incubation (days) 0	3	5	10	15
Lactobacillus bulgaricus 59	Cc	0.06	0.11	0.15	0.22	0.26
	Bc	0.06	0.10	0.14	0.19	0.21
	Gc	0.05	0.08	0.10	0.15	0.17
	CBc	0.07	0.13	0.19	0.28	0.30
,, ,, G 57 (mutant)	Cc	0.06	0.12	0.17	0.28	0.32
	Bc	0.06	0.09	0.15	0.18	0.23
	Gc	0.05	0.08	0.12	0.16	0.18
	CBc	0.06	0.14	0.20	0.36	0.38
Lactobacillus casei RTS	Cc	0.06	0.12	0.16	0.25	0.27
	Bc	0.06	0.10	0.12	0.20	0.23
	Gc	0.05	0.09	0.11	0.15	0.17
	CBc	0.06	0.14	0.21	0.30	0.35
,, ,, G 1 (mutant)	Cc	0.06	0.12	0.18	0.31	0.34
	Bc	0.06	0.11	0.16	0.22	0.24
	Gc	0.05	0.09	0.14	0.20	0.21
	CBc	0.07	0.15	0.25	0.44	0.48

*Cc = Cow casein; Bc = Buffalo casein Gc = Goat casein; Cbc = Crossbred cow casein
**Each value is the mean of three trials.

TABLE 2. DEGRADATION OF DIFFERENT TYPES OF PROTEIN SUBSTRATES BY TWO LACTOBACILLUS SPECIES AND THEIR MUTANTS (mg. OF TYROSINE LIBERATED/g)**

Culture	Substrates*	Period of incubation (days)				
		0	3	5	10	15
Lactobacillus bulgaricus 59 (Parent)	Pnp	0.09	0.11	0.14	0.20	0.23
	Sb	0.08	0.12	0.16	0.20	0.22
	La	0.p9	0.09	0.14	0.18	0.19
	Ea	0.09	0.10	0.15	0.18	0.20
	Bsa	0.07	0.09	0.12	0.14	0.25
	Cc	0.07	0.10	0.14	0.20	0.25
" " G 57 (mutant)	Pnp	0.10	0.13	0.16	0.22	0.25
	Sb	0.10	0.12	0.18	0.26	0.29
	La	0.18	0.09	0.15	0.21	0.22
	Ea	0.10	0.10	0.16	0.19	0.21
	Bsa	0.08	0.09	0.15	0.22	0.26
	Cc	0.06	0.09	0.16	0 29	0.31
" " C 1 (mutant)	Pnp	0.10	0.14	0.15	0.21	0.23
	Sb	0.10	0.11	0.16	0.26	0.28
	La	0.09	0.10	0.18	0.21	0.21
	Ea	0.08	0.10	0.19	0.20	0.22
	Bsa	0.09	0.10	0.16	0.20	0.25
	Cc	0.07	0.12	0.19	0.32	0.35
Lactobacillus casei RTS (Parent)	Pnp	0.10	0.11	0.17	0.19	0.23
	Sb	0.08	0.09	0.16	0.18	0.20
	La	0.09	0.09	0.17	0.20	0.22
	Ea	0.08	0.10	0.18	0.20	0.22
	Bsa	0.08	0.08	0.16	0.19	0.20
	Cc	0.07	0.11	0.15	0.26	0.28

*Pnp = Peanut protein; Sb = Soyabean protein; La = Lactalbumin; Ea = Egg albumin; Bsa = Bovine serum albumin Cc = Cow casein.

**Each value is the mean of three trials.

TABLE 3. ANALYSIS OF VARIANCE OF PROTEOLYTIC OF *Lactobacilli* IN DIFFERENT CASEINS/PROTEIN SUBSTRATE

Source	Significance of F ratio			
	Caseins		Protein substrates	
	L. bulgaricus 59 & Lb/G-57	*L. casei* RTS & Lc/G-1	*L. bulgaricus* 59 & Lb/G-57	*L. casei* RTS & Lc/G-1
Cultures (*a*)	19.00** (1)	29.00** (1)	24.00** (1)	84.00** (1)
Incubation period	273.00** (4)	189.00** (4)	269.00** (4)	500.00** (4)
Caseins/Protein (*c*)	87.00** (3)	67.25** (3)	10.55** (3)	22.00** (3)
$a \times b$	4.5** (4)	4.75** (4)	1.50NS	14.00** (4)
$b \times c$	9.00** (12)	7.75** (12)	5.00** (12)	7.00** (12)
$a \times c$	2.50NS (3)	3.00NS (3)	3.00** (3)	5.00** (3)

**$P < 0.01$; NS. Not significant; Degress of freedom in parentheses

ing 1 g of the protein substrate in citrate buffer (0.01 N; pH 7.0) and the final volume was made upto 100 ml with the same buffer. The different substrates were individually inoculated with 10 per cent washed cell suspension (optical density=0.70) of the test culture. Sodium chloride (3 per cent) and merthiolate (0.025 mg/ml) were added to the inoculated substrates to inhibit the growth of undesirable microorganisms.[9] After incubation for 3, 5, 10 and 15 days, cells were centrifuged at 2000 × **g** for 15 min in each case. Trichloroacetic acid (12 per cent) was added to the supernatent and the precipitated protein was removed by filtration using Whatman filter paper No. 1. The liberated peptide in the filtrate was estimated by the method of Lowry *et al*[10] and proteolytic rate was expressed as mg tyrosine liberated per gram of the sample.

Data on the comparative proteolytic activity of one mutant each of *L. bulgaricus* 59 and *L. casei* RTS and the corresponding parent cultures on four different types of casein substrates are given in Table 1. Progressive enhancement in the proteolytic activity with increase in the incubation period was observed in all the cultures. At the end of 10 and 15 days of incubation, both cow and crossbred cow casein were degraded much better than either buffalo or goat casein. Although similar variations in proteolytic activity of lactic cultures has been recorded in milks of different species[4] very little information is available on the proteolytic action of *Lactobacilli* on different casein species.

In regard to the degradative ability of *Lactobacilli* and their mutants on protein substrates of non-dairy origin, a progressive increase in the rate of proteolysis was noted up to 10 days (Table 2). However, the two mutants show some characteristic variations in their degradative ability of these substrates. *L. bulgaricus* G-57 and *L. casei* G-1 mutants degraded both soyabean protein and bovine serum albumin much faster than the corresponding parent cultures. Statistical analysis was carried out using the conventional procedures of analysis of variance and critical difference. The statistical data given in Table 3 clearly reveal higher proteolytic activity (liberation of tyrosine) of the mutant as compared to the parent cultures. This fact is further confirmed by the differences in arithmetic mean values. Sources like soya and peanut milks for preparation of cultured milk products has already been suggested by several workers.[5,6,11] In view of the above reports, the results of the present study would be of interest on account of the faster degradation of soyabean protein by *Lactobacillus* mutants.

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B. RANGANATHAN

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BOOK REVIEWS

People and Food Tomorrow: Edited by Dorothy Hollingsworth and Elisabeth Morse. Applied Science Publishers Limited, London. 1976, pp. 173. Price ?

The book carries 15 papers read at a conference organised in April 1976 by the British Nutrition Foundation in an attempt to explore the scientific, economic, political and social factors that would affect food supplies by the turn of the century. These are grouped under four broad sections: food requirements, national policies, the provision of food, and constraints on meeting needs. Every paper is replete with information and insights, with factual data and imaginative interpretation.

As is to be expected, several of the papers centre on the United Kingdom, though many analogize more widely therefrom. This can sometimes lead to amusing results. H.C. Pereira proclaims that in the last two decades "the yield of our crops and livestock rose in a manner which, if extended on a world scale, would already have solved the problems of world food supply". They have not solved even the food needs of the UK, which still needs to import half its food. In a brilliant review J. M. Goldsmith points out that more damage has been done to the soil through intensive agriculture in the last 70 years than in the whole of human history, and that it takes 300 years even in undisturbed conditions to reproduce 25 mm of topsoil. Yet John McKenzie sees in the future a further continuation of such happy-go-lucky trends as foods for nourishment and foods for fun, and formal and casual meals.

R. G. Whitehead notes the low protein levels now recommended by international bodies but warns that long-term studies may uncover undesirable metabolic changes if the course is followed; in fact some recent studies at MIT and elsewhere indicate that at adequate energy levels, the recommended low protein levels did result in negative nitrogen balance. The British diet he feels could well do however with a greater proportion of cereals and potatoes and less meat and milk products than at present. A. S. Truswell outlines some micronutrients whose level may be critical in the UK.

Certain other papers are more relevant to our nutritional situation. C. Gopalan outlines the three basic differences between under-developed and developing countries as those relating to population growth, to labour-intensive against capital-incentive production technology (with the challenge of recycling inputs efficiently in the former) and to direct plant food consumption rather than through the animal route. Weighing the Indian situation, he comes out with the optimistic view that India will be able to feed her expected population in 2000 A.D.

W. Brass also takes an optimistic view of the future world population, choosing 2050 for his forecast rather than 2000 which he feels is too short a time-span. His main thesis is that whereas in the past it was socio-economic concomitants that motivated fertility control, today this will occur through the rapid communication of ideas; in consequence fertility reduction, once initiated, will take effect extremely rapidly. For example, the last decade has seen a 40 percent drop in fertility in Britain.

The paper on food in China by K. L. Blaxter is full of admiration for the means by which that country has provided its people with adequate food by collectivisation of 200 or more families to form a viable brigade. These production brigades sell one-tenth of their produce to the state at a fixed high price, using the rest themselves or selling it at a free fixed price in the open market. Decisions on norms and targets, and on investment in new machinery or in other ways, are likewise made at brigade level. Population increase has been effectively controlled through public education and raising the age of marriage. A paper from Norway describes how a socially-conscious country has laid down a conscious nutrition policy of self-sufficiency, and of moving towards a more rational diet that has less fat, more polyunsaturated fats, and less meat, while at the same time aligning its policies with the recommendations of the World Food Congress for greater food availability globally.

The book is a pleasure to read. One paper remarks that "few things are more dangerous than an efficient, energetic and well-equipped man who lacks wisdom". Granted its ethos, there is much here to peruse with profit.

K. T. ACHAYA
PROTEIN FOODS ASSOCIATION, BOMBAY.

International Colloquium on the Chemistry of Coffee: Association Scientifique Internationale du Cafe, June 1975, Hamburg.

The Seventh International Colloquium on the Chemistry of Coffee was held at Hamburg in June 1975 under the auspices of Association Scientifique Internationale du Cafe (ASIC) Paris. Representatives of twenty seven

countries participated. India was singularly absent. Over two hundred and fifty participants who took part in the deliberations, the major representation being from Germany. The papers discussed were covered broadly under Physiology (9), Chemical composition and analysis (27), Chemistry and Technology (9) and Agronomy (16).

Physiology: The Central effects of caffeine by EEG spectral analysis, effect of caffeine on metabolism, peroral application of caffeine causing significant increase of the free fatty acids and free glycerol of the serum, blood sugar and cholesterol being uninfluenced, depressing effect of caffeine on blood alcohol concentrations in rats, absence of teratogenic effect of caffeine, non-correlation of cardiovascular and cerebrovascular death with the use of coffee in a community study, no deleterious effect of coffee in normal consumption with other sclerosis and effect of caffeine on chemical diabetes are some of the findings reported.

Green coffee: Study of water soluble and non-protein components in green coffee using gelo-isolelectric focussing, identification of volatile compounds mainly heterocyclic compounds and associating methoxy pyranine with green coffee odour, potassium content of green coffee and its role in green coffee yield studies on soluble coffee, estimation of 26 elements in green coffee by thermic neutron activation with high resolution, spectrometry and presence of macromolecular substances and structures in coffee are aspects presented in this group. Presence of acid phosphatases and trypsin inhibitors are reported in green coffee.

Roasted coffee: Mechanism of formation of aroma components during roasting, pyrolytic studies on roasting using thermal differential analysis and selective ion monitoring device, studies on absorption of aroma via vapour phase in coffee extract, adsorption isotherm of CO_2 on four roasts of coffee, fluidized bed drying concept applied to roasting of coffee and design of packages with a value or use of CO_2 absorbing packaging material for roasted coffee have been presented in a number of papers.

Formation of nitrogenous compounds in the development of leaves, caffeine content of 130 strains of coffee in Madagascar and a semi-micro analytic method for caffeine estimation using pyro-hydrolysis and separation are detailed.

Analytical methods: Detection and elimination of over-fermented beans (stinkers) by fluorescence studies, establishment of the structure of 'Mascaroside' a bitter principle in species of coffee, estimation of deterpene compounds as ametabolite of astractyligenin in urine, estimation of trigonelline in coffee by N.M.R. studies, moisture determination in green coffee using distillation technique, comparative studies on the determination of total chlorogenic acid by Lehmann and Hausermann/ Branden burger methods are aspects covered under analytical procedures. Composition of fatty acids in coffee oil and wax have been determined.

Coffee pulp: Proteins of coffee pulp has been studied and reduction of tannins by tannase to make the pulp fit for animal feed has been suggested.

Sensory evaluation: Influence of individual components in coffee affecting taste sensations, sensory analysis of coffee and related products are covered in two papers. Role of micro-organism with change of moisture in affecting cuptesting has been pointed out.

Quality control: Differentiation between quality assurance and quality control and a systematic approach has been discussed in one contribution. As in earlier colloquium Dr. Smith has presented a review of the Chemistry and Technology work on coffee.

Agronomy: Coffee rust studies bave been reported in a number of papers. Metrological and genetic factors in the spread of coffee rust and coffee breeding for leaf rust resistance are presented.

Improvements of coffee by genetic manipulation, coffee genetics and inter-specific hybridization, studies on haploids in *C. arabica* hybridisation of *C. arabica* and tetraploid *C. canephora* to give a strong hybrid 'Arabusta' and contribution to the genetic studies in Mascareignes islands with reference to the absence or presence of traces of caffeine are the contributions in genetic studies.

Other studies include differentiation of arabica, robusta and 'arabusta' coffee, serology applied to the study of coffee varieties, relationship between percentage germination and quality of liquor and sequential control of the percentage of insect damaged coffee beans.

The proceedings of the Colliquium presents the current research in the various aspects of coffee and hence will be an useful publication in the library connected with coffee research. Most of the articles are in German or French and this may be a handicap to English and other language readers, but the organisers have rightly appendiced English summary at the end.

C. P. NATARAJAN
C F T R I, MYSORE.

Single Cell Proteins from Cellulose and Hydrocarbons: by Peter J. Rockwell; Noyes Data Corporation, Park Ridge, New Jersey, U.S.A., 1974; pp: 335; Price: $ 39.

In the foreward it is stated that 'this review contains carefully excerpted and collated data from diverse and difficult to locate sources and information is based on studies carried out by research teams; very often under the auspieces of various governmental agencies'. It is therefore in the form of a review and hence the double caption; (Chemical Technology Review No. 74; Food Technology Review No. 34) under which the material is compiled and brought out under one cover.

The short, precise and clear introduction crisply states that Part I of this book is concerned primarily with a process developed at Lousiana State University for production of single cell protein from bagasse as the carbon source. Part II of this book is an updated version of a previous book by Noyes Data Corporation 'Proteins from Hydrocarbons' by Sidney Gutcho, and it contains much additional recent process information.

Having thus set the sight; Peter J. Rockwell proceeds to arrange the text under two broad heads; Part I Cellulose, covers nearly 132 pages with the following subtitles; Sources and characteristics of cellulose, LSU (Lousiana State University) project description; Pilot Plant for LSU Project; Protein isolation from single-cell organisms; and Economic potential of single-cell protein. Part II on Hydrocarbons, covers nearly 200 pages, with the following sub-titles; Liquid hydrocarbon feed stocks, gaseous and solid hydrocarbon feed stocks, Special strains of micro-organisms, growth promoting techniques, Fermentation techniques and Recovery and processing techniques.

It is also stated that the chapter on "Special micro-organism strain" is a completely new section under Part II and that the last chapter contains a number of recent processes designed to eliminate the odor, taste and texture problems usually associated with single-cell protein (SCP) from hydrocarbons. Part I, under cellulose deals with details of raw material availability, physical and chemical characteristics of cellulose. The pioneering work carried out by the LSU group using bagasse to produce single cell protein is extensively reviewed. What is of greater interest is the chapter on "Economic potential of single-cell protein". The information given covers all aspects on the problem including comparative cost estimates. From the technological point of view, a breakthrough has been achieved by the LSU group by their studies on 'Symbiotic growth' using Cellulomonas and Alcalgenes in a mixed culture fermentation of treated bagasse. Part II under Hydrocarbon is more elaborate in its treatment as many countries and industrial units based on hydrocarbons are interested in the development of SCP from hydrocarbons. In an earlier review we have covered the main developments in the field of SCP from hydrocarbons. What is of additional interest is detailed in the chapter on "Recovery and Processing Techniques", which includes, such topics as texturization process, dealing with induced cell leakage, mixture of SCP and vegetable protein and chemical formation of intercellular bonds.

Primarily, for feed purposes, but ultimately for food supplementation also a world wide search is on for new sources of proteins. The two contending substrates are cellulose and hydrocarbons. During the past decade, a great deal of work was done on SCP from hydrocarbons, but more recently the emphasis has shifted to cellulose substrates. It is in this context of a global quest for proteins that the present publication on "Single Cell Proteins from Cellulose and Hydrocarbons" is welcome as an up-to-date and competent coverage of the entire field. The book brings under one cover all relevant information on the subject in a format which leaves nothing to be desired. The present publication is scientifically and technologically compelling and the publishers deserve our compliments. Because of the high cost this useful book may not be within the reach of many who would like to have it.

T. N. RAMACHANDRA RAO
CFTRI. MYSORE

Intermediate Moisture Foods: Edited by R. Davies, G. G. Birch, & K. J. Parker, Applied Science Publishers Ltd., London, 1976.

Intermediate moisture foods (IMF) have received widespread attention and popularity in recent years especially in Defence departments, pet food industry and space programmes on account of their stability at room temperature through lowering of water activity and at the same time retaining the wetness and other characteristics of the fresh food. This is not a new class of foods; familiar examples being cheese, candied fruits and jam, honey, etc but with imagination it could provide a wide range of new and interesting foods. Removal of water is minimum, the bound water is retained within the food so that changes in texture, structure and flavour are reduced or eliminated and the addition of humectants will permit relatively high moisture levels.

The present publication is the proceedings of an Industry-university Co-operation Symposium held at the National College of Food Technology, Weybridge under the auspices of the National College of Food Technology, University of Reading and is probably the first of its kind. It contains 19 papers by international authorities presented at the Symposium divided into 4 Sessions. However it is nowhere mentioned when the Symposium was held.

Chapter 1 is more by way of an introduction of the subject to the Symposium and welcome to the participants. Chapter 2 discusses at length the commercial potential for intermediate moisture foods an account of their shelf stability, convenience and ease of nutrient adjustment and stability but mentions that IMF has been slow to become a significant factor in human food systems primarily because of poor organoleptic acceptability. The chief areas requiring research and development are brought out.

Chapter 3 stresses the fact that for modern IMF for humans to be completely successful, products should be completely new and different from or demonstrably better than the traditional types. Savings in cost to the consumer are not by themselves sufficient to ensure commercial success.

The next chapter gives an account of the different types of IM pet foods which could be broadly divided into 2 types from manufacturing angle: (1) making a slurry and extruding to form the final shape; (2) taking whole chunks of meat or meat analogues and diffusing humectants into them. Only the first has been commercially exploited and a typical product manufactured is described at length.

Chapter 5 deals with the thermodynamic definition of water activity, pointing out some of its consequences and compares the thermodynamic water activity with the quantity often measured and labelled a_w. Next Chapter reviews the various concepts and methods used in the food industry to assess water binding, hydration, etc. Chapter 7 reviews the different instruments used to measure water activity and stresses the need to carry out frequent calibration checks on any commercial instrument using for example, a dew point hygrometer or a salt solution. Chapter 8 describes a method of determining the interaction between constituent water and the solid components of the food by a simple NMR technique. Next Chapter reviews the chemical changes that take place in the major and minor components of foods during processing and storage of IM foods.

Chapter 10 is on the stability of IMF with regard to microorganisms and shows that the inhibition and toxin production of microorganism in IMF does not solely depend on the a_w, but the pH, Eh, temperature, preservatives and the type and number of residual microflora. The next Chapter reviews sources and vectors of contamination, the occurrence and factors influencing growth of yeasts in IMF. It also gives the mechanism of osmotolerance, methods of prevention of yeast spoilage and isolation procedures.

Chapter 12 gives the results of comprehensive investigations into the factors affecting mould free shelf life of cakes and shows that it is governed largely by the ERH of the product. An attempt is made to indicate the minimum mould free shelf life that could be expected from IMFs of different initial ERH. The effect of storage temperatures and moisture loss during storage is also discussed.

The next chapter shows how the risk of *Staphylococci* and their enterotoxins exist in IMF and describes at length the detection procedures and recommends Baird-Parker's medium for recovering *Staphylococci* from IMF. Chapter 14 gives a survey of literature on *Clostridia* problems and indicates that if the product is is seriously abused with respect to a_w and temperature. the risk exists.

Chapter 15 deals with *Salmonella* problems and discusses how difficult it is to eliminate them from the final product by heating as their resistance to heating is greatly increased at these a_w values even though they cannot multiply. The importance of quality control and hygiene of the ingredients especially during manufacture is thus stressed.

Chapter 16 on mycotoxin hazards in these products mentions that although many toxigenic moulds may survive at low a_w values, the limiting level of a_w for toxin production is above that for growth and very little is known about it. Whatever little is known is only for aflatoxin, ochratoxin or penicillic acid and lot of research is needed in this area.

The next chapter on microbiological specifications by Mossel stresses the importance of Good Manufacturing Practices (GMPs) and gives an account of the methods useful in enumerating yeasts, moulds, *Staphylococci Streptococci* and *Enterobacteriaceae.*

Chapter 18 gives the role of humectants in IMF. They lower water activity, lower the available moisture content and also inhibit microbial growth. Temperature plays an important role and hence chilling could be employed to improve the bactericidal action of fatty acids in foods.

Chapter 19 discusses how the growth of microorganisms, the heat resistance of the spores, germinated spores and vegetative cells get modified in the changed atmosphere of IMF.

The coverage of literature in almost all chapters is large and up-to-date.

The book will be very useful for any research and development laboratory in food technology and also some of the industries intending to make such products.

RUGMINI SANKARAN
D F R L, MYSORE.

The Unsaturated and Polyunsaturated Fatty Acids in Health and Disease—by James Mead and Armand J. Fulco. A monograph in The Bannerstone division of American Lecture Series in Living Chemistry. Ed. by Neuton Kugelmass, Charles C. Thomas, Springfield, Illinois, U.S.A.

The authors briefly outline the historical aspects beginning with the production of essential fatty acid deficiency in rats (Burr & Burr, 1929) and proceed to provide a brief and succient account of the chemistry and occurrence of unsaturated fatty acids (UFA). Analytical methods for the determination of UFA in biological material are given in depth, including GLC-mass spectrometer methods and degradation methods such as ozonalysis for structural studies.

The metabolism and biosynthesis of unsaturated fatty acids in plants, protozoa and mammalian systems, enzymological aspects and the mechanism and specificity of unsaturation are presented vividly and discussed. Physiological aspects of UFA essential fatty acids as precursors of prostaglandens and the role of polyunsaturated fatty acids (PUFA) in biomembranes regulating the membrane fraction for proper transport processes and osmotic characteristics have been presented in a simple lucid style. The significance of UFA & PUFA in human diseases—infant malnutrition, atherosclerosis, cancer, multiple screlerosis and genetic diseases is briefly reviewed in Chapter 5.

The chapter on biochemical aspects of autoxidation of UFA provides very stimulating reading. Evidence for *in vivo* peroxidation is appearance of fluorescent pigments ceroid and lipofuscin. Lipofuscion formation in mammals appears to be related to ageing process and the authors speculate that if peroxidation could be inhibited pigment formation and perhaps ageing could be slowed (page 167).

This review is well conceived and written in a very lucid manner. This book is certainly of great value to all researchers in nutrition, biochemistry, medicine and physiology.

P. B. RAMA RAO
CFTRI, MYSORE.

Lipid Chromatographic Analysis Vol. 1: Revised and expanded—edited by Guido V. Marinetti, Publishers - Marcel Dekker, New York & Basel.

This revised comprehensive review on methodological aspects of analysis of lipids of biological origin has contributions from six leading expert groups in this area of research.

Thin layer chromatography (TLC) of phospholipids and glycolipids and a comparative assessment of TLC and paper chromatographic methods (Whatman SG-81 loaded papers) are elaborated and discussed in the first two chapters. Chromatographic analysis of alkyl, alk-1-enyl ether lipids followed by analytical aspect of phosphotides and glycolipids by chromatography of their partial hydrolysis products are presented in chapters 3 & 4. Additional notes on the practical aspects of many of the methods discussed, provide useful information for the research worker. Liquid-liquid partition (LCC), adsorption chromatography and analysis of neutral glycerodes and fatty acids are discussed in Chapter 5. The recent interest in LCC as a potential tool for lipid analysis and the development of instrumentation for this purpose has been pointed out.

Techniques such as high pressure liquid chromatography and the use of lipophilic sephadex have not been reviewed. It is hoped that in subsequent editions this would be added to this volume so that all the techniques developed for lipid analysis would be compiled into one such useful volume.

In the last chapter is described the gas chromatography methods for direct GLC analysis of neutral acylglycerols. This review is very extensive and it has been pointed out by the author (Dr. A. Kuksis) that in combination with mass spectrometry, the TLC-GLC methods can open new avenues for lipid metabolic studies and help in assessment of its role in the metabolism of the cell.

This revised review on lipid chromatographic analysis is well compiled and is of particular interest to lipid chemists and biologists and certainly a welcome addition to all libraries of lipid research groups/Institutes.

P. B RAMA RAO
CFTRI, MYSORE.

Principles of Food Chemistry: By John M. DeMan. The Avi Publishing Co., Inc., West Port, Connecticut, U.S.A. pp. xx×426, 1976, Price: $ 16.

The author states in the preface "The present book contains little of the material commonly covered in biochemistry texts and also does not cover food analysis or food processing. It is hoped that the material covered here will serve as a guide to the most important aspects of food chemistry." Those aspects of food chemistry which the author considers important are presented in 5 chapters dealing with proximate principles and one chapter each on odour, texture, flavour, vitamins, enzymes and additives. Treatment of chapters on water, lipids, proteins, carbohydrates, enzymes and

minerals are typical of undergraduate text books except that sections on the effect of water activity on food preservation and immobilized enzymes in food processing are welcome additions. Food flavours and texture are discussed concisely and the last chapter on food additives is well presented.

A certain amount of generalization and lack of precision are perhaps inevitable in such books but "Principles of Food Chemistry" appears to have more than fair share of misleading statements, errors and omissions. For example guinea pig is a primate (p.324) and lysozyme is an antibiotic (p. 123). Page 86 starts with the sentence "The proteins are polymers of 21 amino acids...." and end with "of the 20 amino acids, only 8 are essential for human nutrition." In the chapter on lipids, fairly extensive coverage is given to the unusual fatty acids in ruminant fats and fish oils but no mention is made of the common cyclopropene fatty acids in vegetable oils or of the significance of polyunsaturated fatty acids. Similarly two pages are devoted to the chemistry and biological activity of vitamin D but no mention is made of hydroxycalciferols. Surprisingly even the editing of this book, brought out by one of the best known publishing firms, appears to be somewhat lackadaisical. For example "Nonenzymatic browning or Maillard reactions *are* one of the most important factors causing food spoilage" (p. 26): "the starch would have to be *postdried*" (p. 30).

The text is well illustrated and is free of printing errors. For most part the book is very readable and should hold the interest of students to whom it is addressed. However, it can be reccmmended as a text book only with some reservation.

P. J. THOMAS
HINDUSTAN LEVER LTD., BOMBAY.

Cowpeas Home Preparation and Use in West Africa: Published by IDRC, Box 8500, Ottawa, Canada 1976 pp. 96, Price: $ 5.

"Cowpeas—Home Preparation and Use in West Africa", is useful information presented in a simple and straight forward manner. The book can be a valuable guide to home science and agriculture teachers and extension workers. The table indicating the nutritional value of cowpeas compared with other foods, available not only in Africa but also in India, and other reference tables, serve as "at a glance" dietary aids. Cowpeas are an excellent source of vegetable protein and when eaten in combination with cereal foods will enhance the dietary protein.

The cookery section is quite thoroughly and systemically developed giving the cooking time and cooked volume of different varieties of the legume as well as the processes and equipment used, for its preparation. Hints on purchasing, storage and different methods of cooking cowpeas are simply but adequately presented and illustrated.

In addition, 56 recipes portraying the Art of African Cookery have been included in the book. These recipes should help in promoting a wider use of this excellent source of vegetable protein in parts of the world where cowpeas are grown.

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ASSOCIATION NEWS

Hyderabad Chapter

Dr. G. Lakshminarayana of the Regional Research Laboratory, Hyderabad, delivered an interesting talk on Edible Oils and Detection of their Adulteration, on 14th May 1977.

Bangalore Chapter

Address by Mr. Gurunath of Modern Bakeries, at Bangalore delivered on 23rd Nov 1976 at Public Health Institute, Bangalore.

The Speaker in his brief talk outlined the marketing techniques adopted by the Modern Bakeries. He said that the shelf life of the Modern Bread was about 4 days and preservatives like calcium propionate and acetic acid were being added. Modern bread from Bangalore Unit was being marketed in far away places like Gulburga, Belgaum, Vasco, Mangalore, etc. Distribution system being direct, the time lag between production and reaching the retail outlet was minimum. Modern bread was distributed on all 7 days of the week unlike their competitors.

35,000 loaves of bread and 25,000 buns are produced per day. About 25,000 beneficiaries per day under the mid-day meal programme of Education Department and 60,000 beneficiaries per day in the age group of 1 and 4 under the special nutrition programme of women and child welfare department received Modern bread.

Food-Fair Held on 6th March 1977

The Fair held at Bowring Institute, was inaugurated by Smt. Vimala Rangachar, Vice-Chairman, Committee for Children's Welfare and Recreation. A variety of food stalls both by leading manufacturers and hotels were put up.

A sum of Rs. 3,000 collected at the fair together with other donations from individuals and industries will be handed over, towards Prof. V. Subrahmanyan Industrial Achievement Award Fund.

Annual General Body Meeting of the Chapter held on 15th March 1977.

The President Sri M. K. Panduranga Setty welcomed the members. He thanked the members for their cooperation. The services rendered by Sri M. R. Chandrasekhara for the chapter was placed on record.

Since no nominations were received for the offices, the nominations were called from the floor. The members unanimously re-elected the following office bearers.

President	Sri M. K. Panduranga Setty
Hon. Secretary	Miss Madura M. Chatrapathy
Treasurer	Sri S. M. Jambagi

The Hon. Secretary, Miss Madhura placed a brief report on the activities of the Chapter. She thanked the members for the cooperation and encouragement extended to the office bearers during the year.

Meeting held on Tuesday, the 21st Dec. 1976.

Dr. K. K. Iya, was requested to take the Chair. Mr. M. K. Panduranga Setty, President, introduced Dr Sulebele, Project Coordinator, Miltone Project, Bangalore to the members.

Dr. Sulebele, emphasised the need for sound quality control in the Dairy Industry. He described some aspects of research work done by his group on infant milk foods and milk based indigenous sweetmeats. Dr. Sulebele focussed attention on the need for a new look at the analytical methodology for the microbiological examination of dairy products. He pointed out that in foods which have been processed so that the coliforms are readily destroyed, there is a need for an alternative index of sanitary quality such as *Enterococci* which are reportedly more resistant to heating, freezing and frozen storage. While coliforms were extremely low in baby foods, *Enterococci* were found in almost all the samples examined. After discussing the microbiological quality aspects of *Pedha* he reported about the widespread occurrence of *Staphylococci* in the samples examined. Enterotoxigenic *Staphylococci* were found in some samples of *Pedha*. Studies on radiation sterilized model food systems simulating *Pedha* demonstrated clearly that *Pedha* does not support the growth of *Staphylococci* while cottage cheese provides an excellent opportunity for the growth of this pathogen to pose a potential health hazard.

He spoke of the advantages of a new medium developed for the detection and enumeration of *Staphylococci* in dairy products.

Southern Regional Branch

Seminar on 'Strategies for Meeting Food and Nutritional Needs of the Rural Population—the Role of Food Science & Technology', held on 23rd and 24th April 1977 *in Madras.*

The Seminar was inaugurated by His Excellency P. Govindan Nair, Acting Governor of Tamil Nadu, and was presided over by Dr. Amla, Director, Central Food Technological Research Institute, Mysore.

There were four technical sessions. The first Session dealing with Nutritional Needs of the Rural Population and Means of Fulfilling Them, was presided over by Dr. Rajammal P. Devadas, Principal, Sri Avinashalingam Home Science College, Coimbatore. The Second Session dealing with Technologies for Food Processing in Rural Areas was presided over by Shri V. Sankaran, Managing Director, Tamil Nadu Agro—industries Corporation. Dr. S. V. Pingale, Technical Advisor, Food Corporation of India, chaired the Third Session, which covered the Case Studies on Related Subjects. Prof. N. Viswanatham, Professor of Pharmacy, Andhra University, was the Chairman of the last Session dealing with Constraints in Setting Up Food Processing Units in Rural Areas.

About 30 papers were presented by food scientists and technologists from all over India covering the entire field of the nutritional needs and manner in which food scientists and technologists could help in solving these problems.

Based on the papers presented and the discussions held some of the important recommendations made in the Seminar are as follows:

1. Maternal nutrition and infant nutrition require priority attention. Nutritional needs of these vulnerable sections of the population living in rural areas require to be met locally. "Mahila Mandal" and village level workers will have to be involved in this endeavour.

2. The Seminar recognized the need for promoting the availability of low cost nutritious foods made out of locally available food materials, in the rural areas, to meet the problem of wide spread under-nutrition and malnutrition.

3. The subject of food and nutrition with particular relevance to the rural needs should form part of the school curriculum all over India as a matter of policy.

4. The export policy of the country should be reviewed with a view to making available nutrtious foods and food byproducts to meet the domestic needs of the population. In this context, export of groundnut cake, rice bran, fruits and vegetables, marine products, etc required to be reviewed.

5. Adequate processing facilities should be created to process the above food commodities and make these available to the rural population. This will also help in generating additional rural employment and increasing the purchasing power. In this regard the Seminar felt that labour intensive composite food processing units could easily be set up with low investment in rural areas.

6. The present food grain procurement and pricing policy requires careful examination in order to leave adequate stock of food grains with the rural holders throughout the year. Establishment of Rural Grain Banks will help in this regard.

7. It was realised that the present practices of food storage in rural areas are far from satisfactory. To overcome this it was recommended that the rural grain storage should be improved, based on modern technology. It was further recommended that simple and scientific storage facilities should be made available in the rural areas.

8. In view of the recognized advantages of higher yield, lesser breakages and important nutritional contents it was strongly recommended that parboiling of paddy needs to be popularised on a wider scale. Improved parboiling techniques have already been developed by various institutions to yield parboiled rice of acceptable quality. A number of small units should be established in rural areas.

9. Considering the large potential available to develop fisheries it was felt that preservation of fish in different forms to meet the needs of rural population should be developed. Large quantities of trash fish available during certain seasons could also be converted into fish cake and fish flour.

10. The professional bodies like the Association of Food Scientists & Technologists, who have adequate know-how and expertize should be consulted in implementing the recommendations of the Seminar.

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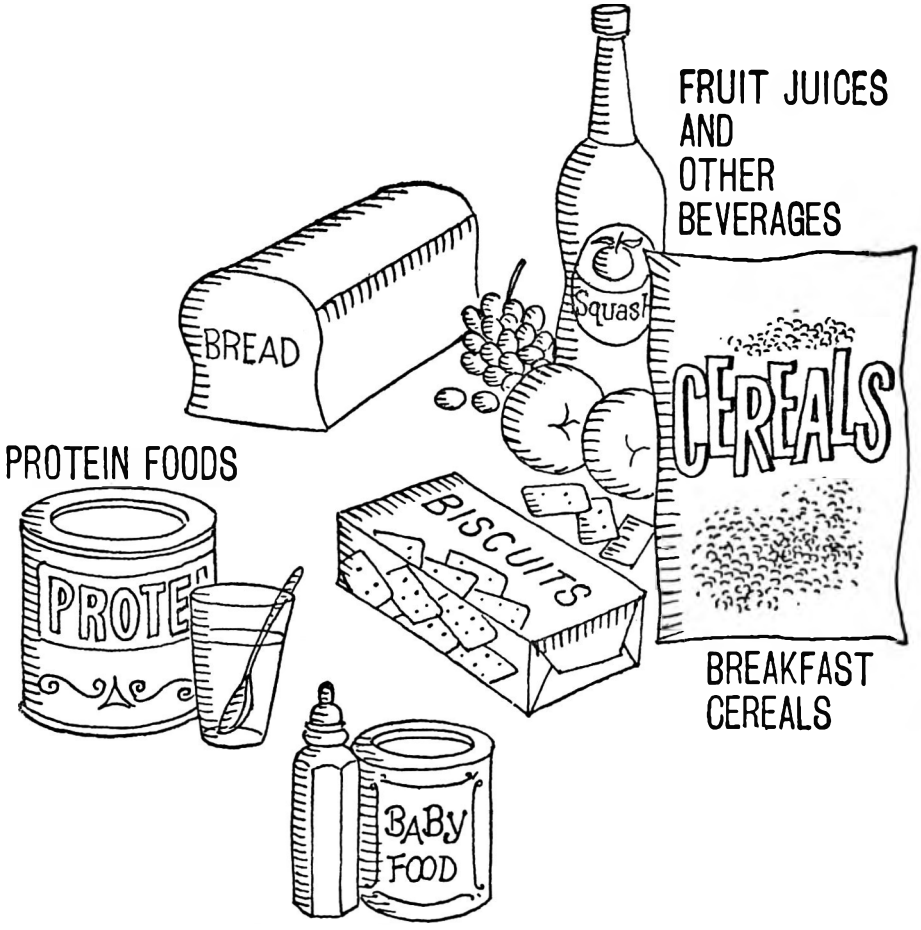

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   (*e*) *Thesis:* Sathyanarayan, Y., Phytosociological Studies on the Calcicolous Plants of Bombay, 1953, Ph.D. thesis, Bombay University.

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