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JOURNAL OF FOOD SCIENCE AND TECHNOLOGY

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RESEARCH PAPERS

Biochemical Activity of *Staphylococci* Isolated During Bacteriological Survey of Catering Establishments

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Out of 132 strains of *Staphylococci* isolated during bacteriological survey of catering establishments, 43 (32.57%) were coagulase positive, which were evaluated for TNase activity, enterotoxigenicity, antibiotic sensitivity, phage typing, lysostaphin sensitivity, mannitol fermentation and phosphatase reaction. Thirtyseven isolates (86.04%) were positive for TNase activity; 18 out of the 40 coagulase positive isolates (45.00%) elaborated enterotoxin types A-E and AB, predominantly A type. Lysostaphin sensitivity and phosphatase reactions were 88.8 and 94.4% respectively among enterotoxigenic types. Majority of the typable isolates showed III and IV group phage reactions which are known to include enterotoxigenic types. Antibiotic sensitivity showed 92.67% resistance towards penicillin among the coagulase positive types and 98.87 and 17.97% resistance towards penicillin and lincomycin respectively among coagulase negative types. This observation clearly points to the indiscriminate use of antibiotics leading to the prevalence of drug resistant *Staphylococci* in catering establishments.

Staphylococcus aureus is a ubiquitous organism and is one of the most common flora of the body[1]. The nature of *Staphylococci* and their enterotoxins, epidemiology and significance as a major cause of food-borne intoxications have been well documented[2-5]. Relation between enterotoxin and thermonuclease production[6], biochemical characteristics and enterotoxigenicity[7], enterotoxin production[8] and enterotoxigenicity of staphylococcal strains[9] have been reported. The present communication deals for the first time, on enterotoxigenicity and some biochemical characteristics of isolates from catering establishments in the survey conducted[10].

Materials and Methods

Staphylococci used in the present study were isolated from crockery, drinking water and food handlers in fifty catering establishments surveyed in Mysore city.

All media and reagents were prepared according to compendium of methods for microbiological examination of foods[11], bacteriological analytical manual for foods[12] and microbiological specifications for foods[13]. The strains were subjected to morphological and biochemical examinations described below to determine their ability to produce enterotoxins, TNase, sensitivity to antibiotics and phages, coagulase production, mannitol fermentation, lysostaphin sensitivity and phosphatase reaction.

The primary isolations were carried out according to the method described by Seligman and Rosenbluth[14] using Vogel Johnson agar instead of mannitol salt agar.

The isolates were then subjected to oxidation-fermentation, catalase test and microscopic examination to make sure of avoiding the presence of *Micrococci* and *Streptococci* respectively.

Glucose utilization: The ability of the cultures to utilize glucose oxidatively or fermentatively was determined by Hugh and Liefson procedure[15]. Catalase test[16], phosphatase[17] and coagulase test[18,19] were also carried out.

Thermonuclease (TNase): TNase was qualitatively determined by microslide procedure. The cultures grown in Brain-heart infusion broth were heated in boiling water bath for 15 min and tested for TNase activity as described by Lachica *et al.*[20]

Production and detection of enterotoxins: Cellophane-over-agar technique as described by Robbins *et al.*[21] was employed for production of enterotoxins. For detection of enterotoxins optimum sensitivity plate method (OSP) was followed. *Staphylococcal* enterotoxins A, B, C, D and E together with their respective antisera were obtained through the courtesy of Prof. M. S. Bergdoll, Food Research Institute, University of Wisconsin, Madison, U.S.A.

Antibiotic sensitivity: The sensitivities of the isolates to different antibiotics generally used for gram-

positive bacteria were carried out as per Kirby Bauer's disc diffusion procedure[22].

Phage typing: The isolates were phage-typed through the courtesy of Prof. D. S. Agarwal, Maulana Azad, Medical College, New Delhi.

Results and Discussion

Identity of 132 isolates of *Staphylococci*, was confirmed by microscopy, gram's reaction, catalase reaction, mannitol fermentation and O/F utilization of glucose as given in Table 1.

As may be seen from the Table 1 total number of samples collected were 873 of which 470 were from food handlers and 403 were from other sources. Food handlers formed single largest source of *Staphylococci*. Nose swab rinse alone yielded 13.92 per cent positives followed by 3.90 per cent and 3-16 per cent of swabs from hands and finger nails respectively. The samples from other sources contributed much less positive indications. A total of 29.27 per cent of the samples were positive for *Staphylococci* of which 20.98 per cent were from food handlers. It has been often mentioned in the literature[23] that nearly 40 per cent normal healthy individuals carry *Staphylococci* on their body especially at finger nails and nasal region. In the present study the low frequency of 20.98 per cent may be due to better personal hygiene maintained by food handlers.

The toxic properties of *Staphylococcal* strains were considered to correlate well with thermonuclease (TNase) and coagulase production. Cultures of *Staphylococcus aureus* generally produce a heat stable nuclease which hydrolyze partially denatured DNA, RNA, and certain oligonucleotides. Data relating to correlation between TNase and coagulase are presented in Table 2. Out of the 132 isolates tested for coagulase reaction 43 (32.57 per cent) were coagulase positive. Majority of them (69.76 per cent) indicated 4+reaction and 27.90 and 2.32 per cent gave 3+ and 2+ reactions respectively: 93.30 and 75.00 per cent of 4+ and 3+ coagulase positive isolates were positive for TNase respectively. Among the coagulase positive isolates 86.04 per cent were also positive for TNase.

Coagulase positive isolates were subjected to enterotoxin assay to determine types of enterotoxins elaborated. The results obtained are presented in Table 3. Fifty per cent of the isolates derived from nose swab rinse of food handlers were found to produce different

TABLE 1. SOURCES OF *STAPHYLOCOCCI* ISOLATES USED IN THE STUDY

Source of samples	Samples tested (No.)	*Staphylococci* positive samples (No.)	(%)	Total isolates (No.)
Plates (Porcelain)	95	3	3.16	5
Coffee cups (Porcelain)	98	2	2.04	1
Spoons (Aluminium)	92	1	1.09	3
Drinking water tumblers (glass)	68	Nil	—	—
Drinking water	50	1	2.00	1
Nose*	158	22	13.92	114
Hands*	154	6	3.90	3
Finger nails*	158	5	3.16	5

*Swab rinses collected from food handlers.

TABLE 2. CORRELATION OF COAGULASE AND THERMOSTABLE NUCLEASE TESTS OF *STAPHYLOCOCCUS AUREUS*

Rabbit plasma coagulation	No. of strains	TNase	
		No.	%
0 (– ve)	89	—	—
1+(+ve)	—	—	Nil
2+(+ve)	1	—	Nil
3+(+ve)	12	9	75.00
4+(+ve)	30	28	93.30

TABLE 3. ENTEROTOXIGENIC TYPES OF *STAPHYLOCOCCUS AUREUS* IN RELATION TO SOURCES OF ISOLATION

Sources	Isolates tested (No.)	Isolates enterotoxigenic		Enterotoxin type						+ve to lysostaphin sensitivity		+ve to phosphatase activity	
		No.	%	A	B	C_1	D	E	AB	%	No.	No.	%
Nose*	22	11	50.00	3	3	1	1	1	2	11	100	11	100
Hands*	6	Nil	—	—	—	—	—	—	—	—	—	—	—
Finger nail cuttings*	5	2	40.00	1	1	—	—	—	—	2	100	2	100
Spoons (Aluminium)	3	2	66.60	2	—	—	—	—	—	2	100	2	100
Coffee cups (Porcelain)	2	2	100.00	1	—	—	—	—	—	1	50	1	50
Plates (Porcelain)	1	Nil	—	—	—	—	—	—	—	—	—	—	—
Drinking water	1	1	100.00	—	1	—	—	—	—	—	—	1	100
Total	40	18	45.00	7	5	1	1	1	2	16	88.8	17	94.4

*Food handlers swab rinses

types of enterotoxins, predominant type being 'A' and 'B'[7]. Strains producing enterotoxin Type 'E' are rare. In the present study, one isolate was producing 'E' type of enterotoxin but after sometime got cured, perhaps the enterotoxin synthesising ability was a plasmid borne character. Isolates derived from finger nails produced 'A' and 'B' type, while two isolates derived from spoons produced only 'A' type. Forty five per cent of the coagulase positive isolates were found to be enterotoxigenic, elaborating different types of enterotoxins A-E & AB. Niskanen *et al.*[6] in their studies, using isolates derived from mastitis cows and samples of food poisoning outbreaks, have reported 51 per cent of the strains to be enterotoxigenic.

Phage-typing was used to clarify the origin of (human or animal) *Staphylococci* and in the epidemiological studies of food poisoning outbreaks[23]. Enterotoxigenic *Staphylococci* strains isolated from nasal cavity of food handlers belonged to Phage Group III. Out of 43 coagulase-positive isolates submitted for phage-typing (Table 4) only 38 strains were used. Twenty nine strains (76.31 per cent) were typed with the phages available at the centre and 9 strains (23.38 per cent) were not typable. Using Routine Test Dilution 58.62 per cent strains were typable and 41.38 per cent were typable RTD $\times$100. The strains belonging to mixed groups comprised 24.14 per cent. Analysis of the results based on sources of isolates showed that all the phage groups were observed. Phage Groups III and IV were found to be most prevalent. Phage Groups I and II considered to be of human origin were found in isolates derived from food handlers.

TABLE 4. PATTERNS OF SENSITIVITY OF *STAPHYLOCOCCUS AUREUS* ISOLATES TO DIFFERENT PHAGES

Source	Isolates tested (No.)	Isolates giving +ve reactions (No.)	Phage No. and Group
Coffee cups (Porcelain)	2	1	6, 47, 53, 54, 75, 84, 85 (III)
		1	29, 80 (I) 47 (III) 94, 96 (IV)
Spoons (Aluminium)	2	1	6, 47, 53, 54, 75, 84, 85 (III)
		1	95 (IV)
Drinking water	1	1	29 (I) 47 (III)
Nose*		1	29 (I)
		3	3A, 3C, 55, 71 (II)
	17	7	42E, 47, 85 (III)
		3	94, 95, IV
		3	mixed groups (I) (II) and (III)
Hands*	1	1	29 (I)
		1	3C (II)
	4	1	47 (III)
		1	29 (I) 47, 75 (III)
Finger nail cuttings*	2	1	47 (III)
		1	95 (IV)

Figures in parentheses indicate phage group
*Swab rinse collected from food handlers.

Results of all the 132 isolates tested for antibiotic sensitivity are presented in Table 5. Data show

TABLE 5. STRAINS OF *STAPHYLOCOCCI* SENSITIVE TO ANTIBIOTICS

Antibiotics	Disc potency* Mgs	Coagulase+ve *Staphylococci***			Coagulase–ve *Staphylococci*+		
		Sensitive	Intermediate	Resistant	Sensitive	Intermediate	Resistant
Ampicillin	10	10(23.25)	5(11.62)	28(65.11)	89(100.0)	Nil	Nil
Chloramphenicol	30	43(100.0)	Nil	Nil	85(95.62)	2(2.24)	2(2.24)
Erythromycin	15	42(97.67)	1(2.32)	Nil	86(96.62)	3(3.37)	Nil
Gentamycin	10	43(100.0)	Nil	Nil	89(100.0)	Nil	Nil
Kanamycin	30	42(97.67)	1(2.32)	Nil	76(95.39)	8(8.98)	5(5.61)
Lincomycin	2*	39(90.69)	3(6.97)	1(2.32)	68(76.40)	5(5.61)	16(17.97)
Neomycin	30	43(100.0)	Nil	Nil	79(88.76)	9(10.11)	1(1.12)
Penicillin	10	Nil	1(2.32)	42(92.67)	Nil	1(1.12)	88(98.87)
Streptomycin	10	40(90.02)	1(2.32)	2(4.65)	75(84.26)	5(5.61)	9(10.11)
Tetracycline	30	41(95.34)	2(4.65)	Nil	77(86.51)	5(5.61)	7(7.86)

*Disc potency in terms of units
Figures in the parentheses indicate percentage
**Number of strains tested was 43 +Number of strains tested was 89.

100 per cent sensitivity of coagulase-positive *Staphylococci* against chloramphenicol, gentamycin and neomycin. There was no sensitivity to penicillin. Even coagulase-negative strains showed 98.87 per cent resistance towards penicillin with 100 per cent sensitivity to gentamycin and ampicillin. Resistance towards commonly used penicillin clearly points towards non-judicious use of antibiotics leading to drug resistance.

Lysostaphin produced by certain strains of *Staphylococci* (K-6-W_1), is a bacteriostatic agent for *Staphylococci*. It is an extracellular lytic factor. Not all *Staphylococci* are sensitive to it, a few of them are also resistant to its action. Lysostaphin, produced by certain strains of *Staphylococci*, is not effective on either viable or heat-killed cells of its own types. Eighteen isolates which are proved to be enterotoxigenic, TNase positive and coagulase positive, were tested for Lysostaphin sensitivity and 16 isolates (88.8 per cent) tested were found to be sensitive to it.

Barber and Kuper[17] observed high degree of correlation between phosphatase production and coagulase production as indicators of *Staphylococcal* pathogenicity - thus a *Staphylococcus* which will liberate phenolphthalein from phenolphthalein diphosphate will almost certainly coagulase blood plasma and be pathogenic to man. The same strains used for lysostaphin sensitivity were also tested for phosphatase activity and all but one of the isolates were phosphatase-positive.

In conclusion it may be stated that there is low frequency of positive indications for *Staphylococci* among food handlers. Coagulase positive isolates were also positive for TNase. Only 45 per cent of the coagulase positive TNase positive isolates were enterotoxigenic. Enterotoxigenicity of the isolates were further confirmed by phage reactions and were found to belong to Group III and IV. Majority of the coagulase positive isolates were sensitive to Lysostaphin and positives for phosphatase activity. Major portion of coagulase positive and coagulase negative isolates were resistant to penicillin.

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Application of Fungal Pectic Enzymes in Coffee Curing*

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Pectic enzyme concentrate (PEC) obtained from *Aspergillus carbonarius* (Bainier) Thom, was employed to break down the mucilage of pulped coffee berries of *Robusta* and *Arabica* varieties. Enzymic treatment reduced the processing time to one hour from 36-48 hr required for natural fermentation. Studies carried out over three successive harvest seasons revealed that coffee seeds could be washed free of mucilage, when crushed berries were steeped in 2.0-2.5% (W/V) PEC solution at ambient temperature (25-30°C). The resulting coffee seeds exhibited almost similar organoleptic quality as those obtained by the traditional method.

The principle grades of coffee produced, in India are *Plantation* and *Cherry*. In the former case, the ripe berries are strip-picked, crushed, heaped and allowed to ferment for 24-48 hr. During fermentation, the mucilaginous covering of the seed is degraded and the seed is washed. This process is beset with several disadvantages such as long period of fermentation (about 48 hr), increase in temperature of pulped beans during fermentation and enormous volume of water required for washing the seeds free of fermentation debris and smell. Some of these factors may lower the quality of seeds produced and cause environmental pollution. In the 'cherry' method, both ripe and unripe berries are dried and mechanically dehulled; the resultant seeds are of inferior quality. This process involves energy.

In natural fermentation the removal of mucilaginous layer is attributed to the activity of a variety of micro-organisms[1]. Johnston and Foote[2] have patented a process for the preparation of green coffee by the use of enzymes. In South America enzymatic treatment is being practiced. It has been reported that coffee mucilage contains 77.6 per cent anhydrogalacturonic acid and lesser quantity of hemicelluloses[3]. Agate and Bhat[4] employed pectinolytic yeasts to degrade the mucilaginous layer. Frank *et al*[5]. have employed strains of *Erwinia* for the same purpose. Rolz *et al.*[6] have described alternatives developed at ICAITI (Central American Research Institute for Industry). Coffee industry in India is on the lookout for simplifying the 'plantation' process. A pectic enzyme concentrate (PEC) developed in this laboratory was tried in coffee processing and was found to be advantageous in several ways. The results are presented in this paper.

Materials and Methods

Two varieties of coffee fruits namely, *Coffea arabica* L. var. 1934 ('Arabica') and *Coffea robusta* ('Robusta') procured from the substations of the Indian Coffee Research Institute located at Mysore and Chettahalli (Karnataka State, India) were used in the studies. Physical composition of the fruits are given in Table 1.

Seed coats (outer pericarp) of ripe fruits (wrongly referred to as berries) were removed by hand and the

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TABLE 1. COMPOSITION OF FRUITS*

	'Arabica'	'Robusta'
Moisture (%)	65–69	65–68
Skin (%)	40–51	38–41
Deskinned fruits (%)	49–60	59–62
Pectin in pulp (w/w)**	0.54–0.60	0.75–0.85

*Average of three cropping seasons.
**As % calcium pectate on wet basis.

deskinned fruit residue (pulp) was used in the experiments.

Pectic enzyme concentrate having pectin degrading activity of 3000 units/ml (the enzyme activity was expressed as per cent reduction in viscosity of 1 per cent pectin solution, at pH 4.0, at 40°C after 30 min incubation as described by White and Fabian[7] was prepared in the laboratory by utilizing a strain of *Aspergillus carbonarius* according to the procedures reported earlier[8,9].

Yeast strain *Saccharomyces cerevisiae* var. *ellipsoideus burgandy* was used for fermentation trials.

Pectin was estimated by the method of Carre and Haynes[10]. pH was recorded using "systronics" bench model pH meter. Reducing sugars were determined by either Lane and Eynon method[11] or by Shaffer and Hartman's procedure[12]. Total solids in the drained and spent enzyme liquid solution were estimated according to the procedure of Ruck[13].

Results and Discussion

Effect of PEC on dissolution of mucilage during coffee seed processing: Data obtained on the enzyme processing of coffe beans are presented in Table 2. Based on preliminary studies 5 kg of 'Robusta' and 2.5 kg of 'Arabica' ripe coffee berries were deskinned. 500 g lots of 'Robusta' and 300 g lots of 'Arabica' were soaked in 500 and 300 ml of PEC solutions of different concentrations respectively. After 90 min of contact, the spent liquids were decanted, the seeds were washed and dried. The results are given in Table 3.

From the Table 3 it is evident that at 3.0 per cent level almost all the mucilaginous material got solubilized. The material treated with 2.0 per cent liquid PEC solution was also free of mucilage and hence 2.5 per cent enzyme solution might be the optimum dose. The enzyme treatment also increased the total solids content and the reducing sugars in the run-off liquids. This may be due to the break down of carbohydrates by the enzyme. The pH of the steeped water decreased with the increase in enzyme level probably because of the pectin degradation products and hydrolysis of other components. The run-off water could be utilized as a substrate for fermentation after supplementation.

The physical appearance of treated and untreated coffee seeds after sun-drying is shown in Fig 1.

TABLE 2. ENZYMIC PROCESSING OF COFFEE

	'Arabica'	'Robusta'
Quantity of berries (g)	400	1000
Quantity of pulp (g)	270	620
Yield of dried seeds (g)	61	238
Parchment (g)	4.1	12.2

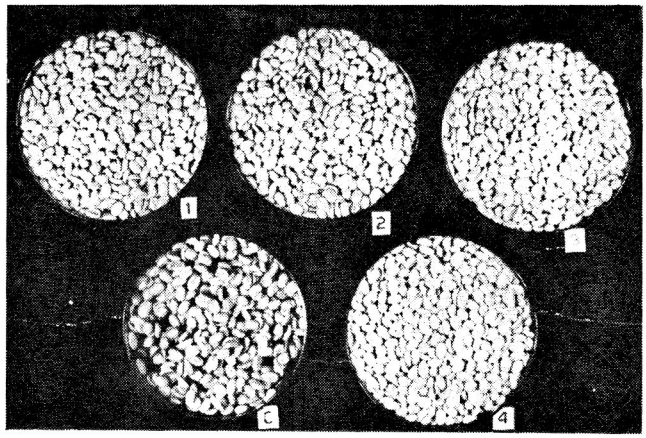

Fig. 1. Appearance of enzyme processed coffee seeds after sun-drying

C.Control; 1: 0.5% PEC; 2: 1.0% PEC; 3: 3.0% PEC; 4: 5.0% PEC

TABLE 3. COFFEE PULP TREATED WITH DIFFERENT CONCENTRATIONS OF PEC AND COMPOSITION OF RUN-OFF LIQUID

Characteristics	'Robusta'–Enzyme concn %						'Arabica'–Enzyme concn %					
	Control	0.5	1.0	2.0	3.0	5.0	Control	0.5	1.0	2.0	3.0	5.0
Wt. of processed seeds (with parchment) (g)	350	314	291	287	236	232	150	140	125	123	120	120
Vol. of run-off liquid I (ml)	505	515	525	535	550	550	140	330	365	370	370	375
pH (after treatment)	6.5	5.5	4.6	4.3	4.2	4.1	6.6	5.5	4.8	4.5	4.4	4.3
Total solids (%)	1.36	3.58	3.81	3.88	3.92	4.18	3.82	4.0	4.83	5.15	5.68	5.83
Free sugars (%)	Traces	1.93	2.03	2.41	2.47	2.82	1.14	1.70	2.16	2.46	3.64	4.10

TABLE 4. EFFICACY OF AGITATION ON THE REMOVAL OF MUCILAGE

	'Arabica'	'Robusta'
Ripe berries (kg)	2.40	2.50
Deskinned pulp (kg)	1.40	1.86
2.5% PEC solution (litres)	1.00	1.00
Run-off liquid I (ml)	1120	1120
Reducing sugars (%)	3.50	2.43
Total solids (%)	8.30	5.18
Run-off liquid II (ml)	800	800
Reducing sugars (%)	0.56	0.57
Total solids (%)	1.05	1.25
Yield of naked seeds (g)	375	382

Effect of agitation on the degradation of mucilage by PEC: Influence of periodical mixing of coffee seeds in steep solution was evaluated by immersing 1.4 kg of *C. arabica* 1934 and 1.80 kg of *C. robusta* (deskinned fruits) in 1.0 and 2.5 per cent PEC solution (just enough to cover the seeds) and stirring the contents every 10 min. It was observed that in 60 min the seeds were rendered free of mucilage. At the end of 60 min the steep water was decanted and the seeds were washed twice using 800 ml of water each time (the water needed would be around 1000 l/tonne of deskinned fruits). The seeds were then sun-dried. The relevant data are presented in Table 4.

The studies indicate that mucilage covering of coffee seeds can be completely degraded by immersing the pulp in 2.5 per cent PEC solution for 1 hr with periodical stirring. It was also noted that water required to process about 2.5 kg of berries was less than 3 l (about 10,000 l of water are required to process 10 tonnes).

The enzyme processed seeds were evaluated for quality at the Indian Coffee Board, Bangalore and was found satisfactory. The flow diagram of the process is given in Fig 2.

Suitability of liquid after steeping as fermentation substrate: The run-off liquid containing 3-4 per cent reducing sugars was pitched with *Saccharomyces cerevisiae* var. *elliposoideus burgandy* after fortification with nutrients. The fermentation was unsatisfactory. This may probably be due to mucilage which is made up of pectin and hemcelluloses[3], or probably the strain employed was unable to ferment the degraded products. Supplemented coffee pulp juice has been successfully employed for the production of fungal biomass employing *Aspergillus oryzae*, *Trichoderma harzianum*, *Penicillium crustosum* and *Gliocladium delequescems*, by Deleon *et al.*[14] who recommended this

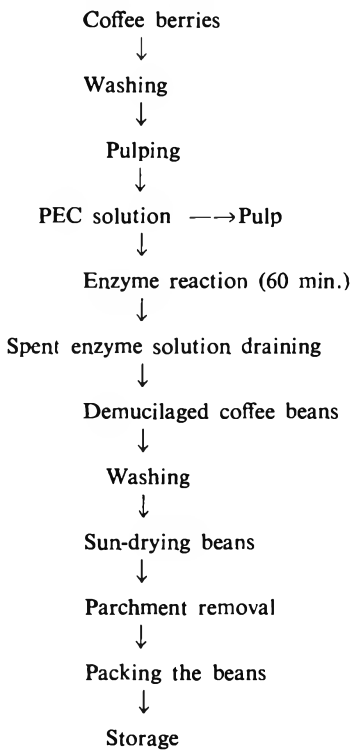

Fig. 2. Flow diagram of the enzyme processing of coffee beans.

procedure for controlling environmental pollution. A list of ten alternatives have also been suggested for use with advantage[6].

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Breakage of Rice During Milling. 5. Effect of Sheller, Pearler and Grain Types

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Paddy seeds harvested at two stages from three types (slender, medium and round) were milled using three kinds of shellers (rubber roller, centrifugal and metal–rubber roller) and three kinds of pearlers (emery roller, emery cone, metal roller). Rice breakage, in each case, originated mainly from defective (cracked and immature) grains, but breaking depended on the equipment and the grain type. Generally breakage of defective grains was less in rubber-roll than in other shellers, in abrasion-type than in friction-type pearlers, in round than in slender grain type, and in low than in high degrees of milling. All these factors interacted and the final product depended on the specific conditions employed.

Recently a critical review appeared on the factors responsible for breakage of rice during milling[1]. A comprehensive study on this problem is being carried out in this laboratory[2-5], and types of cracks in rice[2] and the effect of these cracked and immature grains on rice breakage as also the grain size and shape[3] were described earlier. The comparative performance of different types of sheller (dehusker) using a single variety of paddy (medium grain) was also studied[4]. Effect of different types of sheller as well as pearler (miller, whitener, polisher), their interaction with kernel defects and grain shape, is reported in the present paper.

Materials and Methods

Paddy seeds of 'MR 301' (slender) and 'Coimbatore Sanna' 'GEB 24' (medium) were procured from the University of Agricultural Sciences, Experiment Station, and *japonica* type (round) from Indo-Japanese Agricultural Extension Training Centre at Visweswaraya Canal Farm, Mandya, Karnataka. The seeds were harvested at two stages, one early and one late (to give relatively low and high degrees of grain cracking, respectively) dried, fumigated and stored in closed metal containers in the laboratory. They were milled about an year after harvest.

Analysis of defective grains: Defective kernels in manually shelled grains were detected with the help of a microbiological colony counter and expressed as per cent by weight[2] and grouped into the following catagori s:

Highly defective grains (HD) are grains with multiple transverse cracks+those with longitudinal cracks+immature grains,

Grains with a single transverse crack (STC) and Total defective (TD) grains are those with HD+STC,

Milling: The following three shellers (dehuskers) were used:

(*a*) Satake testing husker (type THU) - a laboratory rubber-roller sheller,

(*b*) McGill sample shellera metal-cum-rubber roller, and

(*c*) a small commercial model centrifugal sheller.

The following three laboratory rice pearling (milling, polishing) equipment were used:

(*a*) Modified[6] McGill miller no. l-a metal-roller, friction-type pearler,

(*b*) Satake grain testing mill-an emery abrasion-type, horizontal roller pearler and

(*c*) Minghetti cone-an emery cone polisher.

The general procedure adopted for milling and separation of broken grains has been reported earlier[3,4]. All shelling was 'full' (90-95 per cent grains shelled in one pass). Pearling, as before, was done to three degrees of milling (d. m.): viz., low, medium and high, representing approximately 3, 5 and 8 per cent d. m. (by weight) respectively. In the experiment on shellers, all pearling was done by the McGill miller; while experiment on pearlers, all shelling was done by the Satake rubber roll. Breakage (less than $\frac{1}{4}$ size) is expressed in per cent weight.

The breakage values were also expressed as relative breakage as discussed earlier[3]. This method helped to roughly visualize at a glance which grains generally broke at each stage, and also facilitated comparison between different grain types, equipment and kernel defects (Tables 2 and 3).

Results and Discussion

Characteristics of the paddy samples used in the experiments are shown in Table 1. There was no significant variation in the moisture content of different samples, therefore, it was presumed that it did not influence the results.

Effect of sheller type: After shelling in different units, pearling was carried out in a McGill (i.e. friction-type metal-roller) miller. Results presented in Table 2 generally reveal the following trend except in the case of CS (a) which exhibited some anomaly.

Breakage during shelling was exceedingly low with the rubber-roller sheller in all cases, accounting for only a small fraction of the respective HD grain count. It was higher in the other two shellers and nearly equalled the respective HD count. Relative breakage during shelling was greater in medium and slender varieties even in the rubber roll, although the breakage was small. Regarding the stage of harvest, relative as absolute breakages were more in late-than in early-harvested samples, suggesting that cracks in highly damaged paddy were more in number and severity than in less damaged paddy.

Breakage during pearling was highest in the brown rice obtained from the rubber-roll and the least in the one from the centrifugal sheller which are in conformity with the earlier findings[3,4]. As a result, the initial advantage of gentler shelling decreased progressively during pearling as the d.m. increased.

Total breakage after shelling and pearling varied with the type of grain, sheller, and the degree of milling (Table 2). The results can be summarised as follows:

Grain type	Sheller type	Final breakage
Round	Rubber roll	> HD grain count, but < TD grain
Round	Others	- do - count
Slender	Rubber roll	- do -
Slender	Others	Approaches or equals TD grain count

For pearling by friction-type metal pearler.

TABLE 1. CHARACTERISTICS OF PADDY VARIETIES

Code	Variety	Maturity	Moisture (wet basis, (%)	Grain parameters[a]			
				L (mm)	L/B	w (mg)	type[b]
Japon (a)	Japonica	Early	12.0	5.4	1.9	18	Round
Japon (b)	Japonica	Late	11.3				
CS (a)	Coimbatore Sanna	Early	12.2	5.6	2.6	14	Medium
CS (b)	Coimbatore Sanna	Late	11.2				
MR (a)	MR 301	Early	12.3	6.4	3.2	16	Slender
MR (b)	MR 301	Late	11.5				

[a]L=mean length, B=breadth, w=weight of milled kernel

[b]The grain type is based on L/B ratio

TABLE 2. EFFECT OF DIFFERENT SHELLERS ON RICE BREAKAGE DURING MILLING[a]

Sample	Defective kernels[b] (%)		d.m.[c]	Rubber-roll sheller			McGill sheller			Centrifugal sheller		
				Breakage (%)	Computed % of grains broken[b]		Breakage (%)	Computed % of grains broken[b]		Breakage (%)	Computed % of grains broken[b]	
	HD	TD			HD	STC		HD	STC		HD	STC
Japon (a)	23.8	48.2	O	2.5	11	0	13.5	57	0	23.7	100	0
			L	14.8	62	0	24.1	100	1	33.2	100	39
			M	22.7	95	0	27.4	100	15	35.6	100	48
			H	32.6	100	36	33.8	100	41	41.1	100	71
CS(a)	11.4	14.3	O	1.3	11	0	7.5	66	0	8.7	76	0
			L	5.9	52	0	7.8	68	0	12.1	100	24
			M	8.9	78	0	9.0	79	0	13.7	100	79
			H	13.3	100	66	10.7	94	0	14.5	100	100
MR(a)	25.2	36.9	O	4.2	17	0	20.4	81	0	28.2	100	26
			L	14.8	59	0	29.0	100	32	32.4	100	62
			M	20.3	81	0	34.5	100	79	34.5	100	79
			H	29.7	100	38	41.9	100	100(8)[d]	36.7	100	98
Japon(b)	62.6	86.0	O	13.2	21	0	39.6	63	O	55.0	88	0
			L	62.7	100	0	64.4	100	8	72.7	100	43
			M	66.3	100	16	69.8	100	31	75.4	100	55
			H	74.7	100	52	74.6	100	51	78.3	100	67
CS(b)	37.2	64.4	O	15.5	42	0	34.4	92	0	41.3	100	15
			L	40.0	100	10	46.3	100	33	51.9	100	54
			M	46.1	100	33	50.1	100	47	53.9	100	61
			H	53.2	100	59	60.9	100	87	56.6	100	71
MR(b)	33.7	55.5	0	12.0	36	0	31.0	92	0	43.7	100	46
			L	28.1	83	0	42.4	100	40	47.9	100	65
			M	35.1	100	6	45.9	100	56	51.2	100	80
			H	47.7	100	64	52.6	100	87	54.1	100	94

[a]All pearling by McGill miller No. 1

[b]HD=highly defective (i.e. multiple-cracked and immature) grains;
STC=grains with a single transverse crack;
TD=total defective grains (=HD+STC)

[c]0=zero (i.e. unmilled brown rice); L,M,H=low, medium and high degree of milling, respectively

[d]Figure in parenthesis represents computed percentage of sound grains broken.

When pearling is carried out by friction-type metal pearler, rubber-roll sheller does not give any special advantage in the case of round varieties but is advantageous for medium/slender varieties.

The initial difference in breakage among grain types at the shelling stage (breakage more in medium/slender than in round varieties) tends to disappear after pearling is followed by shelling in a rubber roll.

The results may be explained as follows: It has been reported earlier[3,4] that HD kernels of all grain types break easily both during shelling and pearling. Breakage in STC kernels is less, and is nil during pearl-

ing and in round varieties. With the result HD kernels in round varieties are exposed to complete breakage while the STC kernels escape, irrespective of the sheller type. HD kernels of medium and slender varieties break completely while STC kernels also are broken by inefficient shellers.

Effect of pearler type: After shelling the samples with rubber roll as in the previous experiments, efficacy of three types of pearlers were tested. Results are shown in Table 3.

There were striking differences in the performance of the three pearlers as in the case of shellers. Horizontal emery roller gave the best performance. It is also evident that the use of a good pearler is more advantageous than using a good sheller only. The best results can be obtained by using a rubber-roll sheller followed by a horizontal emery pearler.

In terms of defective grains, the total breakage at high d.m. (after rubber-roll shelling) was appreciably less than even the HD grains count with the emery

TABLE 3. EFFECT OF DIFFERENT PEARLERS ON BREAKAGE OF RICE DURING MILLING [a,b]

Sample	Defective kernels (%)		d.m.	Emery roller			Emery cone			Metal roller		
				Breakage (%)	Computed % of grains broken		Breakage (%)	Computed % of grains broken		Breakage (%)	Computed % of grains broken	
	HD	TD			HD	STC		HD	STC		HD	STC
Japon(a)	23.8	48.2	O	2.5	11	0	2.5	11	0	2.5	11	0
			L	6.8	29	0	8.7	37	0	14.8	62	0
			M	10.2	43	0	13.8	58	0	22.7	95	0
			H	13.0	55	0	17.7	74	0	32.6	100	36
CS(a)	11.4	14.3	O	1.3	11	0	1.3	11	0	1.3	11	0
			L	2.1	18	0	3.6	32	0	5.9	52	0
			M	2.5	22	0	5.4	47	0	8.9	78	0
			H	3.3	29	0	7.3	64	0	13.3	100	66
MR(a)	25.2	36.9	O	4.2	17	0	4.2	17	0	4.2	17	0
			L	7.6	30	0	11.0	44	0	14.8	59	0
			M	9.8	39	0	13.8	55	0	20.3	81	0
			H	12.2	48	0	17.4	69	0	29.7	100	38
Japon(b)	62.6	86.0	O	13.2	21	0	13.2	21	0	13.2	21	0
			L	37.9	61	0	42.0	67	0	62.7	100	0
			M	52.6	84	0	53.1	85	0	66.3	100	16
			H	58.4	93	0	61.5	98	0	74.7	100	52
CS(b)	37.2	64.4	O	15.5	42	0	15.5	42	0	15.5	42	0
			L	29.9	80	0	31.1	84	0	40.0	100	10
			M	34.6	93	0	35.8	96	0	46.1	100	33
			H	36.7	99	0	39.7	100	9	53.2	100	59
MR(b)	33.7	55.5	O	12.0	36	0	12.0	36	0	12.0	36	0
			L	18.7	55	0	21.4	64	0	28.1	83	0
			M	22.0	65	0	25.4	75	0	35.1	100	6
			H	25.2	75	0	31.5	93	0	47.7	100	64

[a]All shelling by rubber-roll sheller

[b]For abbreviations, see Table 2, footnotes b and c.

roller, and almost equalled that of emery vertical cone.

Among grain types, there was no significant difference in final relative breakage with any of the pearlers. This, might be due to the use of rubber-roll sheller. The relative breakage was higher in highly damaged (late-harvested) as compared to less damaged (early-harvested) paddy when emery pearlers were used while in the case of friction-type pearler the difference was small. This may be attributed to the protective action of the emery pearlers on the grains having mild cracks.

In conclusion, it can be stated that the rice grain breaks primarily due to preexisting defects in it and not due to the stress of milling *per se*, and that sound grains rarely, if ever, break. Also, all defective grains do not necessarily break during milling. Extent of breakage is a function of the grain type, the type of equipment, the degree of milling and their interaction. Machinery designed to exert mild pressure does not break round varieties having even defective grains; whereas in hard equipment, generally there is more breakage in slender varieties, the percentage increasing with the degrees of milling.

The results also indicate that the McGill sheller and miller, often used as standard laboratory equipment, are the least efficient (in terms of rice breakage). However, they can be used to detect the total defective kernels in a sample. In other words, this pair acts as an efficient 'defective kernel detector'.

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Studies on the Hydration of Starches, Flour and Semolina from Different Cereal Grains

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Comparative studies have been made on the cooking of sorghum semolina compared with that of maize and wheat. Sorghum semolina was more refractory to hydration than wheat or maize semolina while the isolated starch from sorghum absorbed much more water at 80°C than other starches studied. Treating sorghum semolina with a protein solvent and with papain increased water absorption generally more in sorghum than in wheat. Greater starch-protein interaction observed in sorghum semolina may represses the innate high water absorption capacity of sorghum starch.

Information on the comparative hydration and cooking characteristics of cereal flour and semolina from different grains is scanty. Lorenz[1] has reported water absorption and viscosity characteristics of wheat, maize and pearl millet flours and starches. Comparative cooking qualities and water absorption of semolina from different cereals have been reported from this laboratory[2] earlier wherein it was not possible to assess the reasons for the variations. The hydration properties of flour, isolated starch and semolina from different cereal grains have been worked out to seek explanation for the observed differences. Work relating to the effect of various treatments on water absorption and cooking times of wheat and sorghum semolina is reported.

Materials and Methods

Milling: Commercial varieties of wheat (*Triticum aestivum*), bajra (*Pennisetum typhoides*), ragi (*Eleusine coracana*), polished rice (*Oryza sativa*), sorghum (*Sorghum vulgare*), Italian millet (*Setaria italica*) and maize (*Zea mays*) were milled in a plate grinder to

pass through 44 mesh sieve. Semolina from wheat, maize and sorghum was made by incipient wetting with 3-4 per cent water, conditioning for 10-15 min and grinding in a Buhler plate mill, collecting −30+44 BSS mesh fractions from all samples

Chemical treatment: A part of the semolina was ground to pass through a 52 BSS mesh sieve for comparing the relative properties of semolina and flour. Part of the semolina was separately soaked in 70 per cent tertiary butanol (BDH) overnight at room temperature to extract the prolamines and were then dried in air. To study the hydration characteristics as affected by chemical treatments—30+44 mesh fractions were soaked at different temperatures in 0.1 per cent aqueous solutions of citric acid, tartaric acid, sodium carbonate, ammonium carbonate, magnesium chloride and potassium iodate, individually and the cooking time, as judged by the time taken for the opaque hard core of the semolina particle to disappear, were determined. Cooking time of semolina soaked for two hours at 50°C in water containing 0.1 per cent papain was also determined to evaluate the effect of protein on hydration. The water uptake of the treated samples was determined.

Water uptake: It was determined by soaking the samples in excess water for 3 hours, separating the solids by centrifuging at 3500 rpm for 15 min and determining the moisture by drying at 105°C for 16 hours. Starch was isolated by the method of Green[3], from these grains.

Results and Discussion

The water uptake of flour and starch isolated from individual samples are presented in Fig. 1. At 50 and 60°C (below the gelatinization temperatures of the starches) flours of all grains absorbed more water than corresponding starches, while at 70°C it was the reverse. This may be due to the fact that above gelatinization temperature (GT), hydration increases enormously in pure starch, while the nonstarchy constituents and the damaged starch modify the total swelling of flour especially below GT. Wheat and maize flours absorbed the highest amount of water while ragi and bajra the least at all temperatures (Fig. 1). On the other hand

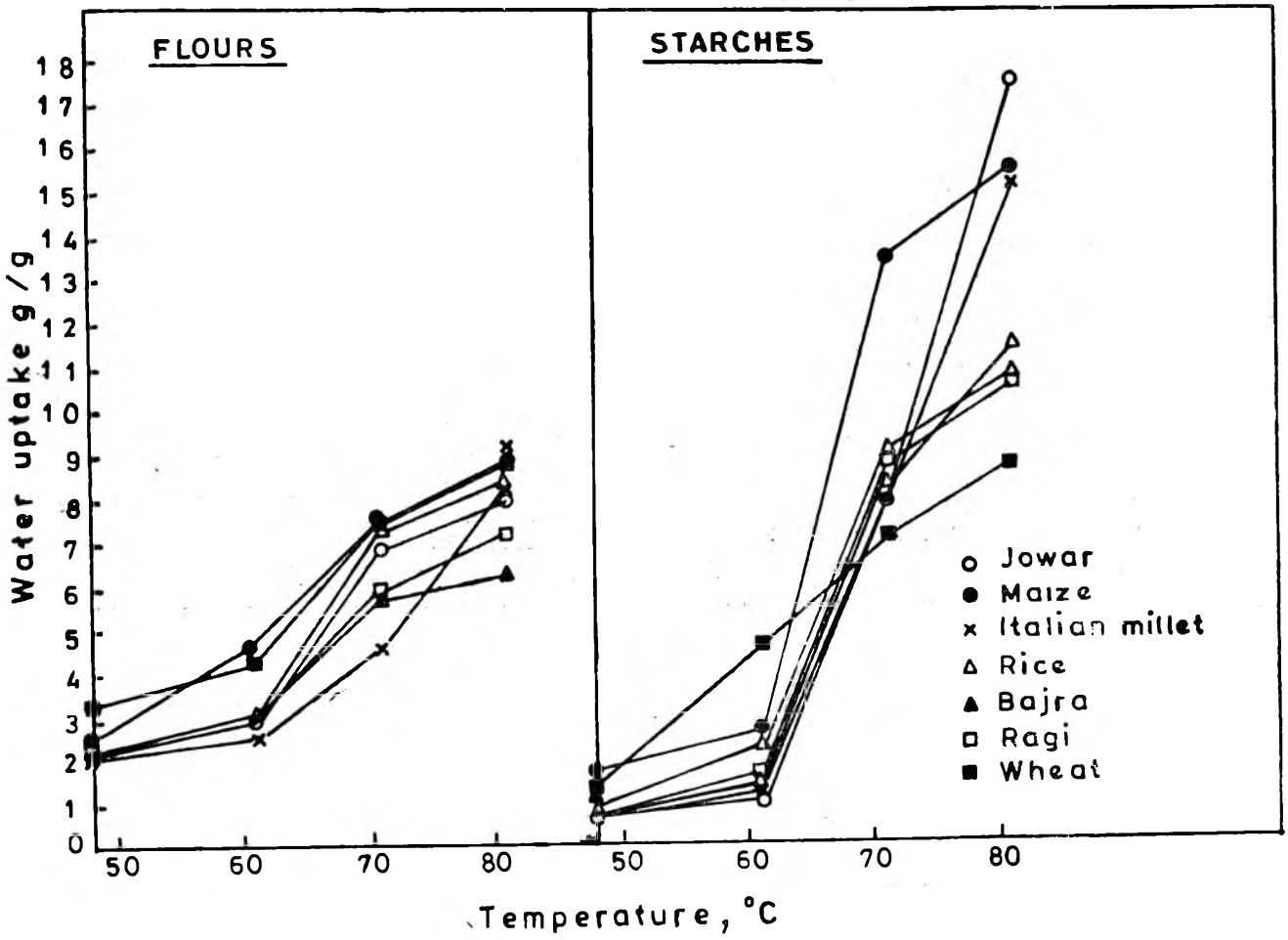

Fig. 1. Water uptake of flours and starches of different cereals at different temperatures

TABLE 1. WATER UPTAKE (G OF WATER PER G OF RESIDUE) BY SORGHUM AND WHEAT SEMOLINA AT DIFFERENT TEMPERATURES

	Sorghum				Wheat			
	50°C	60°C	70°C	80°C	50°C	60°C	70°C	80°C
Semolina	1.5	1.5	3.9	6.3	1.7	3.0	5.6	7.6
Semolina flour	1.7	2.3	5.8	8.3	2.9	4.9	7.0	8.0
Butanol extracted semolina	1.5	1.9	4.6	7.5	2.0	5.1	6.0	8.2

starch from wheat, bajra and ragi absorbed less water compared to those of sorghum, maize and italian millet, the highest being observed in sorghum. Though the reasons for these differences are not clear, one of the reasons may be the chain lengths of the various starch components. Biliaderis *et al.*[4] found a relationship between viscographic data and molecular weight distribution among various legumes studied by them.

The water uptake of wheat semolina was more than that of sorghum at all temperatures studied (Table 1). Grinding semolina to flour increased the water uptake in both the cereals narrowing the differences. Ter. butanol extraction increased the hydration of sorghum and wheat semolina as observed earlier[2]. The higher absorption of water by wheat semolina may be due to gluten[5].

Addition of organic acids (tartaric and citric) or alkaline chemicals (ammonium carbonate and sodium carbonate) decreased cooking time in both sorghum and wheat semolina. Magnesium chloride and potassium iodate had no effect (Table 2). However, changes in water uptake were observed mainly with sodium carbonate, butanol extraction and papain treatment.

One of the causes for the poor water absorption characteristics of sorghum seems to be its protein moiety. Extraction with tertiary butanol or enzymic digestion with papain decreased cooking time in sorghum. The changes were similar in wheat but to a much smaller extent. The results indicate that there may be starch protein interaction in sorghum semolina the nature of which needs further study.

TABLE 2. EFFECT OF VARIOUS CHEMICALS ON WATER UPTAKE AT 80°C AND COOKING TIME OF SEMOLINA FROM SORGHUM AND WHEAT

Chemicals	Water uptake (g/g) at 80°C		Cooking time (min)	
	Sorghum	Wheat	Sorghum	Wheat
Citric acid	7.6	—	10	—
Tartaric acid	7.7	8.2	10	4
Am. carbonate	6.7	—	6	—
Sod. carbonate	7.3	7.1	4	2
Pot. iodate	6.2	—	15	—
Mag. chloride	6.0	—	14	—
Ter. butanol (70%)	7.5	8.2	4	4
Papain	8.5	7.9	6	3
Control	6.3	7.6	16	6
	(6.9)	(7.4)	(12)	(4)

Figures in parentheses indicate water uptake by semolina soaked at 50°C for 2 hr in water before raising temperature to 80°C.

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Improved Indices for Dimensional Classification of Rice

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The surface area per unit weight indicated earlier as a suitable criterion for classifying rice into superfine, fine, common and coarse classes, although theoretically valid, has certain practical limitations. Three other criteria proved satisfactory: normalized grain weight (10w/L, mg/cm), grain weight per unit breadth-length (wB/L, mg) and wB (mg. mm). Grain breadth (B, mm) and grain weight (w, mg) also gave rough indices. Of these, the index 10w/L is simple and accurate. It is also a fundamental property of rice in as much as it (as well as the other four criteria) is highly significantly correlated with the water uptake by rice during cooking. L, L/B ratio and the amylose content are not correlated to water uptake.

Rice, especially in India, comes in a wide range of sizes and shapes. One way to classify it is to group it into four classes each as per grain length (L,mm), length to breadth ratio (L/B), and grain weight (w,mg), respectively[1]. The grain type can then be indicated by a code*. This system of classification is useful for engineering, processing and handling of rice. But it does not give a single, continuous, quantitative scale which would be useful for many purposes.

Rice is classified in India for marketing into superfine, fine, common (medium) and coarse classes, based on purely arbitrary criteria. Attempts made to rationalize this scheme—especially the Ramiah criteria (involving arbitrary combinations of L and L/B) and the Balasubramanian scale (based on L/B ratio alone)—, were unsuccessful[3]. This objective too needs a single, continuous, quantitative scale, embracing diverse sizes and shapes of rice.

The quantification scale suggested earlier[3] has now shown certain practical difficulties and hence other criteria have been sought.

Materials and Methods

The initial part of the analysis was carried out with dimensional data on 172 varieties of rice collected by us earlier[1]. Subsequently some additional work was carried out with 16 fresh varieties representing all the eight rice quality types[4], collected from the University of Agricultural Sciences Experiment Station at Mandya.

The 16 paddy samples were air dried, fumigated and stored in metal containers at 3-5°C . They were milled (to 8-10 per cent degree of milling) with laboratory McGill equipments by standard methods and brokens were separated using appropriate indented plates and discarded.

Amylose contents[4] and dimensional parameters[1] were determined by methods enumerated earlier. The 16 samples were classified into quality types based on the amylose data as discussed earlier[4]. Water uptake (g water absorbed by g milled rice) when rice was cooked at 96°C for 20 min was determined by earlier methods[5].

Results and Discussion

Applicability of surface area index: The three following formulae for calculating the surface area per unit weight of rice (cm^2/g), with successively lesser degrees of accuracy, were recommended in the earlier work[3]:

$$S = \tfrac{1}{2}.\pi.L.\sqrt{\frac{B^2+T^2}{2}}.\frac{10}{w}$$

$$S' = \frac{165\ L.}{7w}\sqrt{0.6458\ B^2 + 0.243\ B + 0.10125}$$

$$S'' = 20.LB/w$$

where

L,B,T = grain length, breadth and thickness (mm), respectively

and w = grain weight (mg).

*E.G., LsS indicates a long (6-7 mm), slender (L/B, >3), and small (12-18 mg) grain; MbB a medium (5-5.99 mm), bold (L/B, 2.0-2.39) and big (18.1-23 mg) grain; and so on.

TABLE 1. DIMENSIONS AND CERTAIN DIMENSIONAL PARAMETERS OF SIX VARIETIES OF RICE SIMILAR IN GRAIN SHAPE BUT DIFFERING IN GRAIN WEIGHT

Variety	Dimensions								
	L (mm)	B (mm)	W (mg)	L/B	S' (cm^2/g)	S'' (cm^2/g)	10w/L (mg/cm)	wB/L (mg)	wB (mg.mm)
Banaspatri	6.04	1.78	11.75	3.39	19.5	18.3	19.4	3.46	20.9
C.P. 231	6.64	1.85	14.02	3.59	18.6	17.5	21.1	3.91	25.9
Belle Patna	6.91	1.92	15.93	3.60	17.6	16.7	23.0	4.43	30.6
Blue Bonnet 50	6.78	2.05	17.64	3.31	16.5	15.8	26.0	5.33	36.2
Patnai 23	7.42	2.18	22.64	3.40	14.9	14.3	30.5	6.65	49.4
SR26B	7.89	2.30	25.78	3.43	14.6	14.1	32.7	7.52	59.3

The applicability of these indices was further tested with selected data taken from our earlier work[1]. Since the grain weight (w) and the grain shape (L/B) are the two primary criteria by which grain fineness/coarseness is perceived[3], the applicability of any index to indicate grain class can be best judged by examining a series of varieties with identical L/B ratio but varying grain weight, and *vice versa*.

Table 1 shows a set of varieties with relatively constant L/B ratio but increasing grain weight. It can be seen that both S' and S'' yielded successively decreasing numerical values with increasing grain weight, and thus gave a correct indication of the progressive coarseness of these varieties.

Table 2 shows another set of varieties where the grain weight was practically constant but the L/B ratio systematically decreased. The varieties thus became, relatively speaking, progressively coarser from the top to the bottom of the Table. While the parameter S no doubt brought out this progression reasonably well, unfortunately S'' in particular and S' in general failed to do so.

It is thus clear that surface area per unit weight as approximately calculated is not a foolproof index of the fineness/coarseness of rice. It could be a reasonably good index provided we use the criterion S (i.e. based on L, B, w as well as T), but that would make it too complicated and unsuitable for routine work. The parameter S' also could perhaps just serve the purpose, but it is again too complicated to calculate for routine work. The parameter S'' is clearly too approximate and unsatisfactory. It may be mentioned in passing that even S itself is based on several assumptions[3] and hence is not an absolute index of the surface area of rice.

Other criteria: Other criteria were therefore examined, viz., wB/L, 10w/L and wB. These parameters have certain theoretical validity.

The parameter wB/L (mg) expresses the grain weight per unit breadth-length, i.e. weight per such length

TABLE 2. DIMENSIONS AND CERTAIN DIMENSIONAL PARAMETERS OF SIX VARIETIES OF RICE SIMILAR IN GRAIN WEIGHT BUT DIFFERING IN GRAIN SHAPE

Variety	Dimensions					S	S' (cm^2/g)	S'' (cm^2/g)	10w/L (mg/cm)	wB/L (mg)	wB (mg.mm)
	L (mm)	B (mm)	T (mm)	w (mg)	L/B						
Slo16	6.00	1.69	1.42	10.91	3.55	20.2	19.9	18.6	18.2	3.07	18.4
Dehradoon Basmati	5.68	1.80	1.48	10.79	3.16	20.4	20.1	19.0	19.0	3.42	19.4
T26	5.60	1.86	1.49	11.03	3.01	20.2	20.0	18.9	19.7	3.66	20.5
Mahsuri	5.02	1.99	1.44	10.50	2.52	19.6	20.0	19.0	20.9	4.16	20.9
JS180	4.68	2.07	1.55	10.31	2.26	19.6	19.6	18.8	22.0	4.56	21.3
Ambemohar	4.32	2.14	1.50	10.41	2.02	18.1	18.5	17.8	24.1	5.16	22.3

as is equal to the grain breadth. Since coarse varieties have greater weight as well as greater breadth, their weight per unit breadth-length should be still larger, and conversely for fine varieties. The matter can be considered from another angle. In our intuitive perception, the greater the grain weight (w) and greater the B/L ratio (he inverse of L/B), the coarser should be the variety. A high value of their product should therefore indicate coarseness, and *vice versa*.

The parameter 10w/L (mg/cm) indicates the grain weight per unit length. In other words it indicates what the grain weights would be, had all the varieties been made 1 cm long but with their cross sections unchanged. The grain length being thus made identical in all varieties, the derived grain weight would be the sole indicator of its fineness/coarseness. Clearly a high value would indicate coarseness, and *vice versa*. The parameter may be called the 'normalized grain weight'.

The parameter wB (mg.mm) s a product of the grain weight and grain breadth, both of which are directly proportional to grain coarseness.

When applied to samples in Tables 1 and 2, all the three parameters gave excellent gradation in their values both in the case of samples with constant shape (L/B) but varying w (Table 1) and with constant w but varying shape (Table 2). It can, therefore, be concluded that all the three criteria would give excellent indices of fineness/coarseness of rice grain. The three parameters were well inter-correlated (Table 3). Among these the index 10w/L has the virtue of requiring the determination of only the two easier (w and L) of the three dimensions (L, B and w), and hence is the simplest for routine work.

Interestingly grain breadth (B) and grain weight (w) are also well correlated to the above three criteria (Table 3), suggesting that they could also be used as rough indices for classification of rice. Indeed the case of w has already been brought out earlier[3], and it may be noted from Tables 1 and 2 that B clearly increases as the rice becomes coarser.

Correlation with water uptake: Further evidence of the usefulness of these indices was sought. It has

TABLE 3. CORRELATION COEFFICIENTS AMONG VARIOUS RICE PARAMETERS[a]

	10w/L	wB	B	w
wB/L	0.981***	0.937***	0.963***	0.816***
10w/L		0.950***	0.909***	0.869***
wB			0.876***	0.963***
B				0.710***

a_n=172

TABLE 4. PROPERTIES OF SIXTEEN VARIETIES OF RICE

Quality type	Variety	Dimensions L (mm)	B (mm)	T (mm)	w (mg)	Amylose (%d.b.) Total	Insoluble	Water uptake (g/g)	10w/L (mg/cm)	wB/L (mg)	wB	Surface area (cm²/g) S	S'	S''
I	T(N)1	5.25	2.60	1.81	16.90	28.9	18.5	2.27	32.2	8.36	43.94	16.40	16.53	16.15
	Jaya	6.19	2.50	1.80	21.39	28.8	18.8	1.89	34.6	8.64	53.48	14.86	14.86	14.47
II	Mahsuri	5.05	2.10	1.48	11.60	27.4	16.3	2.82	23.0	4.82	24.36	18.64	19.09	18.28
	Sona	6.56	1.90	1.59	14.97	29.2	14.7	2.28	22.8	4.34	28.44	18.10	17.57	16.65
III	T141	5.61	2.20	1.66	14.68	29.6	13.4	2.41	26.2	5.76	32.30	17.55	17.47	16.81
	Adt8	3.85	2.40	1.72	11.15	29.3	13.0	2.23	29.0	6.95	26.76	16.99	17.08	16.57
	Unknown[a]	5.89	2.38	1.70	16.92	27.3	13.2	1.97	28.7	6.84	40.27	16.97	17.09	16.57
IV	Br9	4.00	1.90	1.44	8.38	25.9	11.2	3.36	21.0	3.98	15.92	18.97	19.14	18.14
	Basmati370	6.82	1.90	1.59	15.31	25.1	12.3	2.42	22.4	4.26	29.09	18.39	17.86	16.93
V	Co25	4.70	2.30	1.74	13.71	26.0	12.7	2.25	29.2	6.71	31.53	16.48	16.31	15.77
	Intan	6.32	2.18	1.66	17.53	26.1	11.8	2.14	27.7	6.05	38.22	16.46	16.35	15.72
VI	Benong130	6.10	2.58	1.89	21.82	23.1	9.7	1.76	35.8	9.23	56.30	14.90	14.77	14.42
	Sukanandi	4.95	2.70	1.96	18.51	25.0	10.5	1.84	37.4	10.10	49.98	14.87	14.74	14.44
VII	Changlei	6.35	2.35	1.72	18.58	17.5	8.0	1.99	29.2	6.88	43.66	16.59	16.58	16.06
VIII	Purple puttu	5.60	2.60	1.81	19.24	3.2	2.3	1.64	34.4	8.93	50.02	15.37	15.49	15.14
	Asm44	5.10	2.60	1.78	16.49	3.2	2.4	1.71	32.3	8.41	42.87	16.24	16.46	16.08

[a]The sample was labelled as IR24, a low-amylose variety. But the amylose data showed that it was evidently mislabelled.

TABLE 5. CORRELATION COEFFICIENTS OF WATER UPTAKE WITH OTHER PARAMETERS OF RICE ($n=16$)

Parameter	Correlation coefficient with water uptake
10w/L (mg/cm)	-0.818***
wB/L (mg)	-0.790***
wB (mg. mm)	-0.868***
w (mg)	-0.842***
B (mm)	-0.754***
L (mm)	-0.375
L/B	0.114
S (cm^2/g)	0.845***
S' (cm^2/g)	0.857***
S" (cm^2/g)	0.836***
Amylose (% d.b.)	0.492
Insoluble amylose (% d.b.)	0.418
Rice quality type	-0.467

been shown earlier[5] that the water uptake by rice during cooking is proportional to the grain surface area per unit weight, which latter is a function of the size and shape (geometry) of the grain. In view of this, the possible correlation of the water uptake with the above indices was examined with the 16 fresh varieties of rice. The basic data are shown in Table 4 and the correlation coefficients are shown in Table 5. Clearly all the five indices are highly correlated with the water uptake, the values being nearly as high as the correlation coefficients with surface area indices. This clearly proves that the above parameters are not only suitable as indices for dimensional classification of rice but also represent certain fundamental properties of the grain. The regression curve of the simplest among the three accurate indices, normalized grain weight (10w/L), against water uptake is shown in Fig. 1.

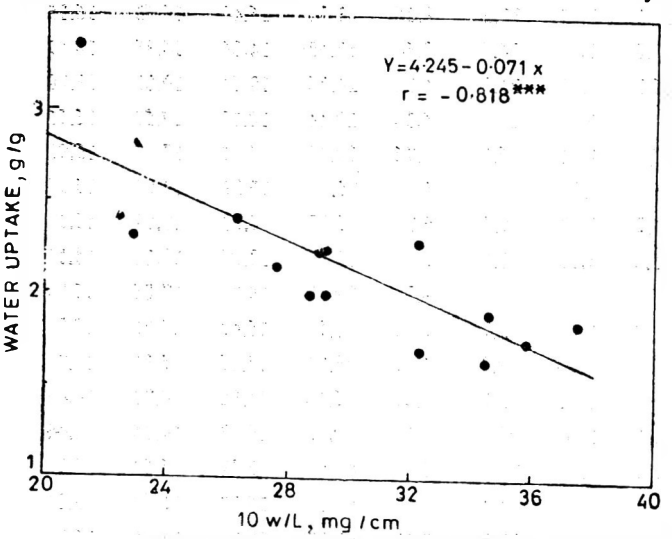

Fig. 1. Plot with regression curve of normalized grain weight (10w/L) of 16 rice varieties against their water uptake during cooking at 96°C for 20 min.

It may be noted that water uptake is not at all correlated either with L or L/B. This is further evidence of the purely arbitrary nature of the L/B value, which has been used as an index for classification of rice[3].

Interestingly, water uptake is not correlated either with the rice quality type or the total or insoluble amylose content (Table 5). This is despite the fact that the 16 samples represented the entire range of rice quality types (Type I to Type VIII) as well as the entire range of amylose contents (0-30 per cent). These results confirm the earlier finding obtained from 45 samples with a much narrower range of amylose contents[5], that hydration of rice during cooking is basically a physical phenomenon and is practically independent of any intrinsic quality characteristics of rice. Thus these data again demonstrate the error of the persistent belief that water absorption by rice during cooking is a resultant of its amylose content[6,7].

Classification of rice: Having fixed the above criteria, the cut-off points for the different rice classes had to be determined. For this the frequency distribution curves of 10w/L, wB/L and wB values (also B and w for comparison) for the 172 varieties studied in our earlier work[1] were plotted (not shown). Superfine, fine, common and coarse classes of rice were then derived from these figures by dividing them arbitrarily at 20, 50 and 80 per cent cumulative frequency values, as summarised in Table 6.

The suitability of the three new indices (as well as B and w as rough criteria) is shown with a number of popular Indian varieties in Table 7. Clearly the new indices give better results than the prevalent Balasubramanian and Ramiah criteria or even the surface area index (S") described earlier[3]. The index 10w/L, is the simplest and best.

TABLE 6. FIVE PROPOSED CRITERIA FOR CLASSIFYING RICE

Class	Classification as per 10w/L (mg/cm)	wB/L (mg)	wB (mg.mm)	B (mm)	w (mg)
Superfine	<23.0	<4.3	<23.0	<2.0	<13.0
Fine	23.0–27.0	4.3–6.1	23.0–34.5	2.00–2.25	13.0–17.0
Common	27.1–32.0	6.11–8.0	34.6–48.0	2.26–2.55	17.1–21.0
Coarse	>32.0	>8.0	>48.0	>2.55	>21.0

TABLE 7. APPLICABILITY OF DIFFERENT CLASS INDICES TO SEVERAL RICE VARIETIES

Variety	Dimensions			Classification[a] as per								
	L (mm)	B (mm)	w (mg)	L/B	10w/L	wB/L	wB	B	w	Bala-Subramanian[b]	Ramiah[c]	S'' (cm^2/g)
Br9	4.2	1.9	8.2	2.21	S	S	S	S	S	Cm	MS	S
Mahsuri	5.2	2.0	10.5	2.60	S	S	S	S	S	F	MS	S
Basmati370	6.1	1.8	13.1	3.38	S	S	F	S	F	S	LS	F
Jirasal180	4.7	2.1	10.3	2.24	S	S	S	F	S	Cm	SB	S
IR20	6.0	2.2	15.0	2.72	F	F	F	F	F	F	LB	F
RP5-3	6.4	2.0	16.1	3.20	F	F	F	F	F	S	LS	Cm
Slo18	5.6	2.3	14.6	2.44	F	F	F	F	F	Cm	SB	F
IR22	6.5	2.1	17.5	3.10	F	F	Cm	F	Cm	S	LS	Cm
Slo13	5.9	2.4	16.2	2.46	Cm	Cm	Cm	Cm	F	Cm	SB	F
Adt8	4.2	2.4	11.8	1.75	Cm	Cm	F	Cm	S	Cs	SB	F
Ptb10	5.6	2.0	18.4	2.80	Cm	Cm	Cm	F	Cm	F	MS	Cs
Patnai23	7.4	2.3	23.0	3.22	Cm	Cm	Cs	Cm	Cs	S	LS	Cm
Ptb9	5.7	2.6	18.3	2.19	Cs	Cs	Cm	Cs	Cm	Cm	SB	F
SR26B	7.7	2.3	25.4	3.34	Cs	Cm	Cs	Cm	Cs	S	LS	Cs
Jaya	6.4	2.5	22.8	2.56	Cs	Cs	Cs	Cm	Cs	F	LB	Cs
Ptb20	5.4	2.6	19.4	2.08	Cs	Cs	Cs	Cs	Cm	Cm	SB	Cm

[a]S, F, Cm, Cs=Superfine, fine, common and coarse, respectively

[b]Balasubramanian criteria (L/B): ≥ 3=S; 2.5 - <3=F; 2 - <2.5=Cm; <2=Cs

[c]Ramiah Committee criteria: LS=Long slender, MS=medium slender, SS=short slender, LB=long bold, SB=short bold.

The grain weight, w, is perhaps the most important factor in all the above formulations, which has been ignored in the Ramiah and the Balasubramanian criteria[3] of grain fineness, which is the prime reason of their failure.

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Minicolumn Chromatography for the Detection of Argemone Oil in Edible Oils

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Minicolumn chromatography has been used for the first time for the detection, confirmation and quantification of argemone oil in contaminated edible oils. Argemone oil at 0.01 percentage level could be detected.

Adulteration of edible oils with argemone oil, obtained from the seeds of a weed, *Argemone mexicana L.* is common. The toxicity of argemone oil has been attributed to an alkaloid, sanguinarine[1]. Several outbreaks of the toxic syndrome called epidemic dropsy in different parts of India have been reported[2]. Recognizing the need for the detection of argemone oil in edible oils a number of methods involving chemical tests[3-6], paper and thin layer chromatography[7,9,11], colorimetry[10] and spectro-photometry[9,11] have been developed in the past.

Glass as well as disposable polythene minicolumns were (devised for the detection of aflatoxins [12,13]) utilized for the rapid detection and quantification of argemone oil and the results are presented in this paper. Furthermore, a confirmatory test has been developed by using ferric chloride reagent, which renders the detection of argemone oil possible without UV source.

Materials and Methods

Preparation of glass and polythene columns: A glass minicolumn fabricated from 15 cm length, 6 mm outer diameter and 4 mm inner diameter borosilicate wall tubing plugged at one end with glass wool, and was packed with neutral alumina to a height of 3 cm and column chromatography silicagel (60-120 mesh, activated at 110°C for 1 hour) to a height of 6 cm. A cotton plug was put at the other end.

The polythene tubing (15×1.5 cm) prepared from an ordinary polythene sheet of 0.2 mm thickness was plugged with glass wool at one end. Neutral alumina and column chromatography silicagel (activated at 110°C for one hr) were added into the column to 2 cm and 6 cm heights respectively one above the other. A cotton plug was put at the top.

Extraction of sanguinarine: Sanguinarine from argemone oil mixed with pure refined groundnut oil and pure mustard oil to give 1 per cent contamination was extracted as per the procedure described by Shenolikar *et al.*[9]

Detection and quantification: The dry residue of the extract was dissolved in 0.5 ml of chloroform acidified with a drop of glacial acetic acid and transferred into a 5 ml test tube in which the packed column was placed. Complete solution was allowed to ascend through the column and then transferred into another 10 ml test tube containing a mixture of 3 ml of n-butanol, acetic acid and water (60:15:25, v/v) for the development. The solvent mixture was allowed to reach the top and the column was viewed under UV. A golden yellow band seen above the neutral alumina zone indicated the presence of sanguinarine. Similar process was repeated for mustard oil mixed with argemone oil (1 per cent).

Quantification was done by comparing the intensity of the fluorescent band of the column developed in sample with that of standard sanguinarine solution.

Confirmatory test: The developed column was placed in 5 ml of ferric chloride reagent (10 g in 100 ml of 10 per cent HCl in ethanol) taken in a test tube, and observed for brownish red in the place of golden yellow fluorescent band which could be seen with the naked eye.

Sensitivity test: Pure refined groundnut oil and pure mustard oil containing 1 ml argemone oil for every 100 ml, were suitably diluted to have 0.1, 0.05 and 0.01 per cent argemone oil. Two ml each of these were tested for the contamination.

Results and Discussion

Although the intensity of fluorescence changed

from bright golden yellow to pale yellow with the serial dilution, the fluorescent band was visible upto 0.01 per cent level (0.5 μg sanguinarine/ml). Ramasastri and Babu[14] have suggested that a level of 0.01 per cent contamination of argemone oil could be considered as permissible limit in other edible oils.

The entire operation of the technique with confirmatory test required one hour. This method is simple, economic and rapid when compared to conventional TLC method, is suitable for the public health analytical laboratories. Furthermore, the confirmatory test with ferric chloride reagent makes the technique more reliable and renders the detection of argemone oil possible even in the absence of UV light source.

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Suitability of Some Mango Varieties for Processing

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Nine table varieties, 4 juicy varieties and 5 hybrids of mangoes grown in Andhra Pradesh (India) were evaluated for canning as slices, juices and nectars. 'Baneshan', 'Suvarnarekha' and '5/5 Rajapuri × Langra' varieties were found suitable for canning as slices. 'Navaneetam', 'Baneshan', 'Goabunder', 'Royal special', 'Hydersaheb' and '9/4 Neelum Baneshan' varieties were found good for the preparation of juices, while 'Baneshan', 'Navaneetam', 'Goabunder', 'K.O.7' and 'Sharbatgadi' varieties were good for nectars. The effect of addition of calcium chloride as firming agent on the quality of slices is described.

Mango (*Mangifera indica L*), is grown in all parts of India and occupies about one million hectares and nine million tonnes of fruit are produced annually[1]. More than 1000 varieties with varied characteristics like fruit yield, size, taste and flavour of pulp are grown in the country[2]. Several new promising varieties of mangoes including many hybrids have been evolved with good characteristics with respect to yield, periodicity of bearing, disease and pest resistance, taste and keeping qualities. Suitability of some North Indian

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and South Indian mango varieties for the preparation of various products have been reported earlier[3-7]. Eighteen promising varities of mangoes grown in the Fruit Research Station, Sangareddy (A.P.) were selected to study their processing characteristics.

Materials and Methods

Nine table varieties ('Daseri', 'Goabunder', 'Baneshan', Totapuri', 'Suvarnarekha', 'Intemax', 'Kurnuli Mulgoa', 'K.0.7' and 'Jahangir'), 4 juicy varieties ('Royal Speical' 'Navaneetam', 'Sherbatgadi' and 'Hydersaheb') and 5 hybrids ('13/3 Rumani'×'Neelum', '5/5 Rajapuri× Langra', '1/4 Daseri×Mahmooda', '9/4 Neelum×Banganapalle' and '9/9 Samar-e-behist×Daseri') were procured from the Fruit Research Station, Sangareddy, Andhra Pradesh, India.

Fully matured and medium size fruits were collected in two consecutive seasons, kept at ambient temperature for 1 to 3 days for allowing all the fruits to ripen uniformly and observations recorded. Out of the total 10 kg of fruits collected for each variety, 3 fruits of big, medium and small size were selected from each variety for studying the physico-chemical characters such as shape, size, colour, weight, brix, acidity, etc.

Canning of slices: Peeled and sliced mangoes were packed net 220 g into each of 301×309 plain cans covered with 150 g of sugar syrup of 35° Brix, containing 0.3 per cent added citric acid. To six cans in each variety calcium chloride (at 0.02 and 0.04 per cent concentration of calcium on the net weight of the pack) was added as a firming agent. The cans were exhausted, sealed and processed in the usual way.

Canning of juice and nectar: The juice was collected by passing the pulp through a 30 mesh sieve. The pulp content of the final product was kept at 35 per cent and the brix and acidity were adjusted to 20° and 0.3 per cent respectively. The juice heated to 85°C, was filled full into plain 301×309 cans, quickly sealed and processed in the usual way. Similarly, the nectars were prepared keeping the pulp content at 20 per cent, the brix and acidity at 18° and 0.3 per cent respectively. All the products were stored at ambient temperature (16-40°C) for one year, cut out examination being conducted at intervals of 3 months.

Ascorbic acid in the fresh fruit was estimated by direct titration with 2:6 dichlorophenol indophenol[9]. The soluble solids were determined by a hand refractometer and acidity by direct titration against standard alkali. The total carotenoid pigments and β- carotene content were determined according to the methods reported in Vitamin Assay[9]. Volumetric procedure of A O A C was adopted for estimation of tin. The organoleptic evaluation was carried out immediately after canning and at intervals of 3 months during storage by a panel of 5 judges, using 5-point Hedonic scale on the basis of attributes like colour, texture, taste and flavour.

Results and Discussions

Physical characteristics of mango fruits studied are given in Table 1. 'K. Mulgoa', 'Baneshan', 'Intemax', 'Totapuri', 'Rumani×Neelum' and 'Daseri×Mahamooda' were bigger in size and weighed 320 to 415 g each, while 'Daseri' was the smallest weighing, 91 g. The stones of 'Daseri' variety were big (24.8 per cent) and its peel content was also high (19.8 per cent) as a result of which the yield of edible pulp was low (55.4 per cent). The yield of the pulp in other varieties varied from 52.5 ('Hydersaheb') to 79.4 per cent (Kurnali Mulgoa).

The physico-chemical characteristics of the mango pulp are given in Table 2. The soluble solids content was highest in 'Rajapuri×Langra' (27 per cent) and lowest in 'Totapuri' (15.8 per cent) while in other varieties values varied from 18 to 21 per cent. Maximum acidity was noticed in 'Intemax' (0.63 per cent). Ascorbic acid content was generally low (1.5-40 mg/100 g) in all the varieties examined, except in 'Royal special' (102.9 mg/100 g) and 'Sharbatgadi' (64 mg). All the varieties except 'Kurnali Mulgoa', 'Jehangir' and 'Rajapuri×Langra' had attractive bright yellow to orange yellow colour. 'Goabunder', 'Intemax', 'K.O.7', 'Royal Special', 'Navaneetam', 'Sherbatgadi', 'Hydersaheb', 'Neelam×Baneshan' fruits were more fibrous. The pulps of 'Royal Special', 'Navaneetam' and 'Neelum×Baneshan' were very thin, while others were thick. 'Daseri' variety had the highest β-carotene content, while 'Totapuri' had the lowest.

Slices of 'Baneshan', 'Suvarnarekha', 'Rajapuri× Langra' and 'Daseri×Mahamooda' had good taste, flavour and texture. Slices from 'K.Mulgoa' were good but had dull yellow colour, while those from 'Intemax', 'K.O.7' and 'Neelum×Baneshan' had become mashy during processing. 'Goabunder', 'Rumani×Neelum', 'Jehangir', 'Samar-e-bahist×Daseri' and 'Neelum× Baneshan' were fibrous and were not suitable for canning. 'Samar-e-bhaist×Daseri' developed off-flavour during processing. Slices from 'Jehangir' developed uneven white patches. Addition of calcium at 0.02 per cent concentration helped in retention of colour and the syrup was also clear. Use of more calcium (0.04 per cent concentration) altered the flavour of the product during storage. 'Baneshan' and 'Rajapuri×Langra' slices, stored for one year at ambient temperature, were

TABLE 1. PHYSICAL CHARACTERISTICS OF DIFFERENT VARIETIES OF MANGOES

Variety	Mean wt of fruits (g)	Peel (%)	Stone (%)	Edible pulp (by diff.) (%)	Yield of slices (%)
Table varieties					
Daseri	91	19.8	24.8	55.4	—
Goabunder	170	14.9	17.9	67.2	45.6
Baneshan	346	11.2	13.2	75.6	53.7
Totapuri	305	13.5	12.9	73.6	—
Suvarnarekha	225	15.0	16.6	68.4	40.0
Intemax	317	16.5	9.5	74.0	46.7
Kurnuli Mulgoa	415	9.3	11.3	79.4	48.8
K.O.7	288	21.4	18.0	60.6	44.6
Jehangir	230	15.1	13.7	71.2	42.6
Juicy varieties					
Royal Special	158	15.8	18.0	66.2	—
Navaneetam	170	12.6	22.8	64.6	—
Sherbatgadi	251	19.3	21.4	59.3	—
Hydersaheb	170	24.4	23.1	52.5	—
Hybrids					
9/9 Samar-e-behist×Daseri	171	12.3	17.5	70.2	47.2
13/3 Rumani×Neelam	323	9.2	13.1	77.7	57.7
5/5 Rajapuri×Langra	179	15.4	20.3	64.3	45.4
1/4 Daseri×Mahmooda	320	13.4	10.5	76.1	48.0
9/4 Neelam×Baneshan	287	18.6	15.2	66.2	44.0

found to retain their firm texture, colour, taste and flavour, 'Suvarnarekha', 'Goabunder' and 'Rumani × Neelum' retained their texture and flavour. 'Jehangir' slices became rather soft and colour was found to be uneven. 'Daseri × Mahmooda' and 'Samar-e-behist × Daseri' had developed off-flavour.

Canned Juices: Different varieties when assessed for consistency, colour, taste and flavour ranked in the following order; 'Baneshan', 'Navaneetam', 'Goabunder', 'Royal Special', 'Neelum × Baneshan' and Hydersaheb'. Canned juice from 'K. Mulgoa' and 'K.O.7' had good consistency, flavour and taste but was unattractive due to the dull yellow colour. Canned juice from 'Suvarnarekha', 'Daseri' and, 'Rumani × Neelam' had mild flavour. The other varieties were not found suitable for juice, as they developed disagreeable flavour after processing. Canned pulp from 'Navaneetam' variety retained maximum flavour and thick consistency, even after one year storage at ambient temperature, while the varieties 'Baneshan', 'Goabunder' 'Royal special' and 'Neelum × Baneshan' were found acceptable in that order.

Canned nectars: Nectars prepared from 'Baneshan', and 'Navaneetam' varieties retained their consistency, flavour and taste, even after one year storage at ambient temperature. 'Goabunder', 'K.O.7' and 'Daseri' varieties scored next in that order. Except for its dull colour, nectar from 'K. Mulgoa' had heavy consistency, pleasant flavour and taste. Nectars from 'Jehangir', 'Suvarnarekha', 'Royal Special', 'Intemax', 'Rumani × Neelum', 'Neelum × Baneshan', 'Samar-e-behist × Daseri' varieties had mild flavour. 'Totapuri' was not found suitable for making nectar as it developed unpleasant flavour during storage. The tin pick up in the canned nectars after one year storage at room temperature varied between 101 and 130 ppm, which was more than in canned juices (69-91 ppm).

TABLE 2. PHYSICO-CHEMICAL CHARACTERISTICS OF FRESH SLICES AND PULP OF DIFFERENT MANGO VARIETIES

Variety	°Brix	pH	Acidity (%)	Ascorbic acid (mg/100g)	Carotenoids (mg/100g)		Colour	Texture/ consistency of slices	Organoleptic quality
					Total	β-carotene			
Table Varieties									
Daseri	18.0	4.95	0.13	14.2	8.6	5.2	Orange red	Soft/Heavy	Sweet, mild flavour
Goabunder	18.6	3.75	0.13	14.2	3.4	—	Orange yellow	Firm and fibrous/Heavy	– do –
Baneshan	19.2	4.10	0.18	8.1	3.9	2.1	Orange Yellow	Firm/Heavy	Sweet, pleasant flavour
Totapuri	15.8	3.80	0.52	2.3	3.4	1.9	Light orange yellow	Firm/Heavy	Acidic, very mild flavour
Suvarnarekha	18.8	4.25	0.25	1.5	3.9	2.1	Orange yellow	Firm/Thin	Sweet, moderate flavour
Intemax	18.0	3.40	0.63	2.2	—	—	Yellowish orange	Firm/Heavy	Sour, mild flavour
Kurnuli Mulgoa	18.0	5.10	0.16	24.2	—	—	Creamy yellow	Firm/Heavy	Sweet, pleasant flavour
K.O.7	26.0	4.80	0.25	5.7	—	—	Light orange yellow	Firm/Heavy	Sweet, moderate flavour
Jehangir	21.1	3.80	0.40	2.0	—	—	Yellow	Firm/Heavy	Acidic, mild flavour
Juicy Varieties									
Royal Special	19.4	3.95	0.49	102.9	—	—	Orange yellow	Very soft	Acidic, terpenuous flavour
Navaneetam	18.0	4.60	0.25	3.0	4.1	2.6	,,	,,	Sweet, pleasant flavour
Sherbatgadi	17.6	4.35	0.31	64.1	—	—	,,	,,	Acidic, mild flavour
Hydersaheb	21.0	3.95	0.45	20.9	—	—	,,	,,	Acidc, moderate flavour
Hybrids									
9/9 Samer-e-behist × Daseri	20.0	4.55	0.20	40.6	—	—	,,	Firm/heavy	Insipid, very mild flavour
13/3 Rumani × Neelam	16.0	4.45	0.21	13.0	—	—	,,	,,	Sweet, mild flavour
5/5 Rajapuri × Langra	27.0	4.27	0.33	15.8	—	—	Yellowish	,,	Sweet pleasant aroma
1/4 Daseri × Mahmooda	21.0	4.55	0.21	23.5	—	—	Orange yellow	,,	Sweet, mild flavour
9/4 Neelam × Baneshan	19.0	5.60	0.12	6.3	—	—	,,	Very soft & stringy near peel	Sweet moderate flavour

Pulp was heavy consistency in all the table Varieties except in 'Suvarnarekha'.

In all juicy varieties the juice had thin consistency; In all hybrids juice had heavy consistency except in 9/4 'Neelam × Baneshan' where it had thin consistency.

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Effect of Post-harvest Fungicidal Treatment of Citrus Fruits Against the Control of Green Mold Rot

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Effectiveness of Benlate, Bavistin, Thiabendazole, Imazilil SP and Imazilil EC was evaluated for the control of green mold rot of artificially inoculated Coorg mandarin and Seedless lime. All fungicides showed a gradual loss of protective ability with time. Comparatively Imazilil (SP and EC formulations) was highly effective and showed a decay reduction index of 74.16 to 96.66, during a storage period of 36 days. Thiabendazole was found to be markedly ineffective in reducing the decay but its protective quality remarined for a short period.

The incidence of green mold rot of citrus incited by *Penicillium digitatum* Saccardo, in untreated fruits in storage is quite alarming and losses upto 20 per cent is common. This is particularly true in fruits harvested during monsoon season. It has been reported that more than 30 per cent of the monsoon crop is lost every year due to fungal rots and conventional handling methods[1]. Earlier studies in this laboratory, has indicated that the losses due to postharvest rots could be minimised by treating fruits before storage with 1000 ppm of Benomyl, Bavistin or Thiabendazole[2]. However, under commercial conditions of storage the chances of reinfection of fruits by *Penicillium* spp. cannot be ruled out even if a single decayed fruit is associated with the healthy fruits in store house[3,4]. So it becomes essential that fungicides used as postharvest treatment should be able to protect the fruits against a forthcoming infection.

The present sutdy was, therefore, undertaken to identify the effective duration of some fungicides used as postharvest protectants of green mold rot pathogen during storage.

Materials and Methods

Coorg mandarin (*Citrus reticulata* Blanco) and seedless lime (*Citrus latifolia* Tan.) of uniform maturity, free from blemishes and harvested during the monsoon fruiting season (July-August) were selected for the study. The fruits were separately treated for 5 min in 1000 ppm solution of Benlate [(Methyl 1-(butyl-carbomyl) -2-benzimidazole carbamate)], Bavistin [(2-(Methoxy-carbamyl)-benzimidazole], Thiabendazole [(2-(4-thiazolyl) benzimidazole)], Imazilil SP and EC formulations [(1-(2, 4-dichlorophenyl)-2-(2-propenyloxy) ethyl)-1 H-imidazole)]. Imazilil is a newly developed systemic fungicide and is slightly yellowish to brownish in colour (E.C. formulation being oily liquid). The acute toxicities in rats are 0.550 g/kg and 0.374 ml/Kg for SP and EC formulations respectively. The fungicide is known to be effective against benzimidazole-resistant strains of *Penicillium*. Fruits dipped in tap water served as control. Excess water or fungicidal solution from treated fruits was blotted with a sterile towel and fruits stored under ambient conditons (21-24°C, 85-90 per cent R. H.). Each treatment consisted of 400 fruits in four replicates. After 24 hr of treatment and subsequently at intervals of 7 days, 60 fruits (15 from each replication) from each treatment were artificially inoculated with *Penicillium digitatum*. Inoculation was accomplished by pricking at 4 points on blossom and stem end of fruits to a depth of 1-2 mm by pre-sterilised needles loaded

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with dry spores of the pathogen[4]. The inoculated fruits were incubated for 24 hr in polyethylene bags and later stored on a dry table under laboratory conditions. Percentage of fruits showing *Penicillium* infection was recorded separately for each treatment. The data were transformed into "Decay Reduction Index" (DRI) as suggested by Gutter[4,5] which is as follows:

$$DRI = \frac{\% \text{ decay in control} - \% \text{ decay in treatment}}{\% \text{ decay in control}} \times 100$$

Results and Discussions

The relative protective ability of different fungicides in protecting the fruits with a corresponding increase in time after treatment is presented in Table I and Fig. 1 and 2., in the form of decay reduction indices. All fungicides evaluated showed a gradual loss of protective ability to reduce decay with a corresponding increase in time after treatment. However, the relative performance showed that Imazilil, SP and Imazilil EC formulations, were highly effective, recording a very high decay reduction index of 74.16 to 96.66, depending on the formulation and citrus species tested. Imazilil EC proved much better than the Imazilil SP especially for Coorg mandarin, where the fruits showed no decay even when inoculated on the 29th day after treatment. Greater penetration and retention in the fruit may probably account for the high protective property of this fungicide. Imazilil SP and Imazilil EC have been reported to be effective agianst strains of *Penicillium* resistant to other fungicides like Benlate or Thiabendazole[6]. Thiabendazole, on the other hand

TABLE 1. EFFECTIVENESS OF FUNGICIDES IN THE CONTROL OF GREEN MOLD ROT OF CITRUS
(Average Decay Reduction Index)

Fungicides	Inoculation days after treatment						Av. for duration
	1	7	14	22	29	36	
			Coorg Mandarin				
Benlate	77.50 (61.50)	80.00 (67.50)	70.00 (56.94)	65.00 (54.00)	42.50 (40.39)	2.50 (4.61)	56.24
Bavistin	100.00 (90.00)	70.00 (56.94)	55.00 (47.94)	57.50 (37.72)	10.00 (13.28)	0.00 (0.00)	45.41
Thiabendazole	47.50 (43.60)	30.00 (33.50)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	12.91
Imazilil S. P.	100.00 (90.00)	97.50 (70.39)	82.50 (65.83)	80.00 (67.50)	62.50 (58.45)	47.50 (43.60)	78.33
Imazilil E.C.	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)	80.00 (67.50)	96.66
			Seedless Lime				
Benlate	100.00 (90.00)	100.00 (90.00)	80.00 (67.50)	55.00 (47.94)	32.50 (34.55)	5.00 (6.64)	62.83
Bavistin	100.00 (90.00)	92.50 (78.75)	65.00 (54.00)	22.50 (27.69)	10.00 (13.28)	0.00 (0.00)	48.33
Thiabendazole	100.00 (90.00)	67.50 (55.26)	45.00 (42.50)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	35.41
Imazilil S.P.	100.00 (90.00)	100.00 (90.00)	90.00 (71.56)	67.50 (55.26)	56.00 (46.50)	37.50 (37.22)	74.16
Imazilil E.C.	100.00 (90.00)	100.00 (90.00)	92.50 (78.75)	87.50 (67.50)	75.00 (60.27)	65.00 (54.00)	86.66
C.D. at 5%			Fungicides (F)		Duration (D)	F × D	
Coorg Mandarin			3.83		4.20	9.37	
Seedless Lime			3.38		3.71	8.29	

Value for inoculated control is zero for all days of inoculation for both the types of fruits. Figures in parentheses indicate Values after angular transfermation.

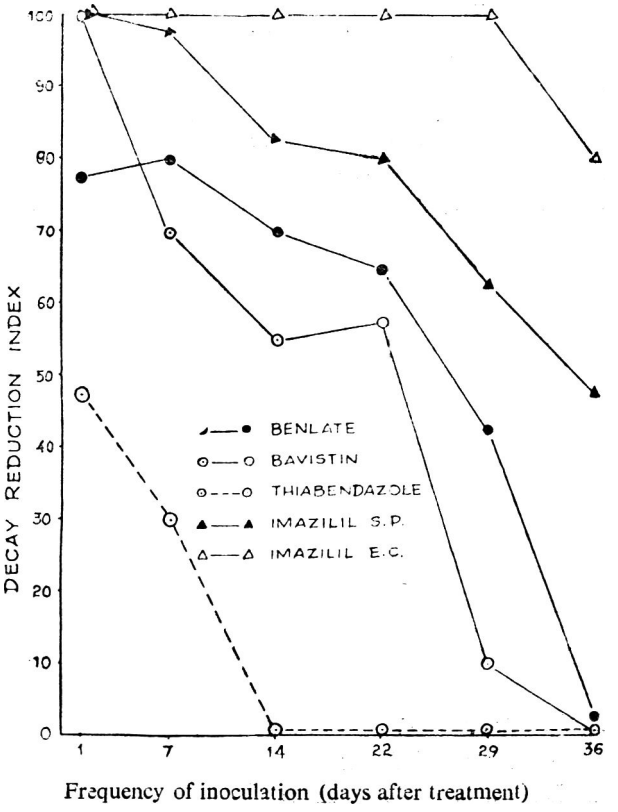

Fig. 1. Duration of effectiveness of fungicides in the control of green mold rot of Coorg orange.

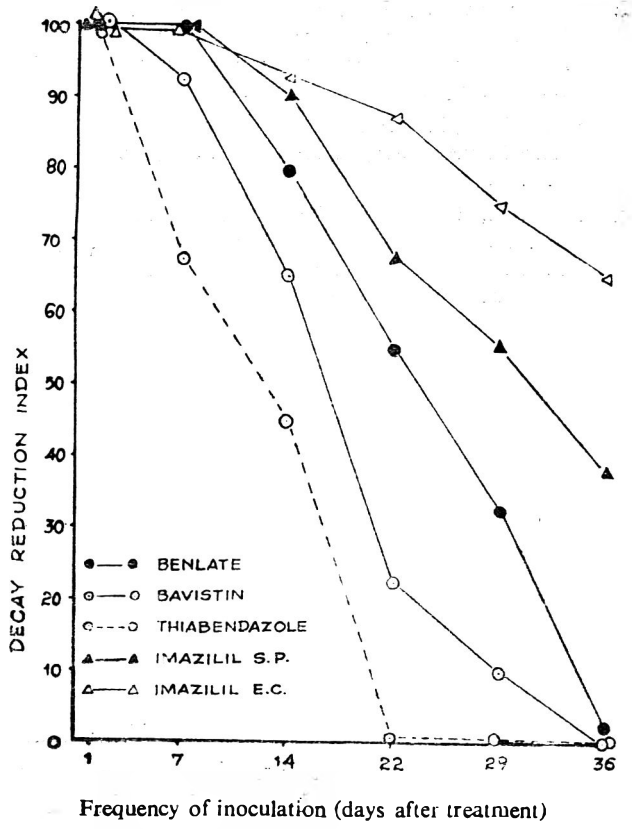

Fig. 2. Duration of effectiveness of fungicides in the control of green mold rot of Seedless lime.

was markedly ineffective, particularly for Coorg mandarin, where the average decay reduction index for the entire experimental duration was only 12.91. The fungicide failed to protect the fruits fully even for 24 hr after treatment and its protective ability was completely lost after 7 days in Coorg mandarin although some degree of protection was noticed in seedless lime upto 14 days.

The results obtained with the two citrus species in the present study also demonstrated some variation in their susceptibility to decay and differential permeability to fungicides. Seedless lime fruits treated with Benlate, Bavistin and Thiabendazole gave a higher decay reduction index when compared to Coorg mandarin when treated with the same fungicide. On the other hand Coorg mandarin fruits treated with Imazilil SP and Imazilil EC showed higher decay reduction index than Seedless lime treated with the same fungicides. This suggests that fruits of two citrus species apart from minor inherent variation in their susceptibility also demonstrate a differential permeability to fungicides.

The differential ability of the fungicides to protect the fruit agianst the infection depends mainly on the differential penetration and persistance of the fungicide in the fruit. The percentage of residue biologically active in the fruit has been shown to be directly related to the initial amount which penetrated and the region where it accumulates[7]. The greatest persistence of the fungicide usually occurs in the peel region of the fruit. Thiabendazole has been reported to be less mobile than Benomyl[7]. The lower penetration and poor stability of thiabendazole may be reasons for its poor performance in the present study as also has been observed by Gutter[5]. The relative differential ability of fungicides to penetrate and retain in fruits may also have practical implications where they are used as preharvest sprays to control postharvest decay.

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Effect of *Chhana* and *Rasogolla* Preparations on preformed *Staphylococcal* Enterotoxins and Thermostable Deoxyribonuclease in Milk*

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Raw cow's milk inoculated with enterotoxigenic strains of *Staphylococcus aureus* at levels of 1×10^6 CFU/ml of milk were incubated at 37°C for 10-12 hr. Incubated milk samples showed staphylococcal populations of 10^8-10^9 CFU/ml, thermostable deoxyribonuclease (TDNase) levels of 10-14 mm zone diameter and detectable levels of enterotoxins. Conversion of the incubated milk samples initially into *Chhana* and subsequently to *Rasogolla* showed the absence of *Staphylococci*, but presence of TDNase and enterotoxins. When *Chhana* and *Rasogolla* samples were enriched in 3.7% brain heart infusion (BHI) broth for 20 hr at 37°C, *Staphylococci* (10^4-10^9 CFU/ml) reappeared and increase in TDNase was observed.

Chhana and *Rasogolla* are milk preparations common in India. *Chhana* is the residue obtained after the liquid portion is drained off from boiled milk. It has a moisture content of 55-60 per cent. *Rasogolla* is prepared by boiling *Chhana* rolled into small balls in 40-60 per cent sugar syrup. High incidence of *Staphylococci*, including enterotoxigenic types, have been reported in Indian milk products[1-3].

Often, the raw milk from which these products are prepared is heavily contaminated with microorganisms including *Staphylococcus aureus* which elaborates thermostable enterotoxins under suitable conditions. Staphylococcal counts in the range of 2×10^3 to 3×10^7/ml have been reported in market samples of raw milk[4,5]. *Chhana* prepared from such a milk can carry over enterotoxins into the product. Experiments were carried out to find the carry over of contaminating *S. aureus* and preformed enterotoxins and thermostable deoxyribonuclease (TDNase) from milk to *Chhana* and *Rasogolla*.

Materials and Methods

Cultures: *Staphylococcus aureus* A_{100}, $B_{s\text{-}6}$, C_{137}, D_{472} and E_{326} elaborating enterotoxins A,B,C,D and E (SEA, SEB, SEC, SED and SEE), respectively were obtained from Food Research Institute, Univ. of Wisconsin, Madison, U.S.A., and used as test cultures.

Preparation of inoculum: Cultures were grown in 3.7 per cent brain heart infusion (BHI) broth for 24 hr at 37°C, centrifuged at 3,354 RPF for 20 min and the cells collected in sterile 0.9 per cent saline. This

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was diluted to give 50 per cent transmittance at 600 nm using sterile saline. Serial dilutions were prepared and surface plated on egg yolk-tellurite-glycine-pyruvate agar (ETGPA) of Baird-Parker[6] to determine the colony forming units (CFU) in different dilutions.

Inoculation of milk and preparation of Chhana *and* Rasogolla: Individual test culture (1×10^6 CFU/ml) was inoculated to one litre lot of cow's milk (raw) and samples were incubated at 37°C for 10-12 hr to enable *S. aureus* cells to grow and produce TDNase enterotoxins. These samples were then converted into *Chhana* according to the procedure of Soni *et al*[7]. initially and later to *Rasogolla* as per the method of Date *et al*[8].

During preparation care was taken to avoid external contamination and all materials used were previously sterilised.

With a view to activate microbial cells, in heat treated *Chhana* and *Rasogolla*, samples were enriched in 3.7 per cent broth. One gram of the sample was mixed with 99 ml of BHI broth and incubated for 24 hr at 37°C, and then staphylococcal counts were enumerated, and also tested for TDNase and enterotoxins.

Enumeration of Staphylococci: Appropriate dilutions of the samples were surface plated on ETGPA medium according to Baird-Parker[6]. One ml of 20 per cent sodium pyruvate solution filtered through Seitz funnel was added to a few serial dilutions of *Chhana* and *Rasogolla* samples before plating them on ETGPA medium. All the petri plates were incubated at 37°C for 24-48 hr and the colonies of *S. aureus* formed were counted and expressed as staphylococcal CFU per ml/g.

Extraction and detection of TDNase: Milk sample was acidified to pH 4.5 with 6N HCL and centrifuged at 3,354 RCF for 30 min. *Chhana* and *Rasogolla* were separately triturated in 2 per cent sodium citrate buffer (pH 7.2) and 10^{-1} dilutions of each was centrifuged at 3,354 RCF for 30 min. The supernatants were acidified to pH 4.5 by the addition of 6N HCL and recentrifuged at 3,354 RCF for 30 min. The supernatants from samples of milk, *Chhana* and *Rasogolla* were neutralised to pH 7.0 with 1N NaOH and then steamed at 100°C for 15 min. The final supernatants were examined for the presence of TDNase by the toluidine blue-deoxyribonucleic acid (TB-DNA) agar plate method[9].

Extraction and detection of enterotoxins: Enterotoxins were extracted from the samples according to the method of Read et al.[10,11], with some modifications. Dilutions of *Chhana* and *Rasogolla* 10^{-1}) made in 2 per cent sodium citrate buffer (pH 7.2) were centrifuged at 3,354 RCF. for 30 min. The resulting supernatants were acidified to pH 4.5 by adding 6N HCL and centrifuged at 3,354 RCF for 30 min. The final supernatants were examined for the presence of individual enterotoxins like SEA, SEB, SEC, SED and SEE by the optimal sensitivity plate method of Robbins *et al.*[12]

Results and Discussion

Effect on cell populations of S. aureus: The growth of *Staphylococci* in milk as well as in *Chhana* and *Rasogolla* is presented in the Table 1. All the five test cultures of *S. aureus* grew well in raw cow's milk as indicated by cell population densities of 10^8-10^9 CFU/ml after 10-12 hr of incubation at 37°C.

Staphylococci were absent in *Chhana* and *Rasogolla* prepared from milk containing high staphylococcal populations. In addition to sodium pyruvate already present in ETGPA medium, the addition of sterile sodium pyruvate solution before plating caused no appreciable change, except for the appearance of one

TABLE 1. *STAPHYLOCOCCUS AUREUS* STRAINS (COLONY FORMING UNITS PER MILLILITER PER GRAM) IN MILK, *CHHANA* AND *RASOGOLLA*

	A_{100}	B_{S-6}	C_{137} (CFU/ml/g)	D_{472}	E_{326}
Cow's milk (raw)	10^1	10^1	10^1	10^1	10^1
Incubated milk*	8×10^8	8×10^8	6×10^8	8×10^8	5×10^8
Chhana (enriched)**	22×10^5	27×10^9	20×10^5	Nil	Nil
Rasogolla (enriched)**	18×10^4	Nil	16×10^5	Nil	Nil

*Milk inoculated with test cultures (1×10^6 CFU/ml of milk) and incubated at 37°C for 10-12 hr.
Fresh *Chhana* and *Rasogolla* and with added sodium pyruvate were devoid of *S. aureus* strains
One milliliter of 20% Seitz filtered solution of sodium pyruvate to the serial dilutions of *Chhana* and *Rasogolla* before plating did not contain any of the strains of *S. aureus*.

**One gram sampel was mixed in 99 ml of BHI broth and incubated at 37°C for 24 hr
CFU: Colony forming units

colony on ETGPA plates which was not significant to record and the diameter of the colony after 72 hr of incubation was 1.0 mm.

It is well known that coagulated proteins afford protection to bacteria against heat treatment and perhaps this is true in the preparation of *Chhana* and *Rasogolla*. From the results obtained after plating it can be inferred that the staphylococcal cells have undergone heat injury. Studies[13] on the fate of *Staphylococci* in cheese revealed that during cooking of curd, these organisms became embedded between the curd particles, thereby escaping the effect of cooking temperature. However, earlier studies[14-18] have shown that *Staphylococci* are killed at or near pasteurisation temperature.

It was not significant to consider the appearance of only one colony on ETGPA plates in the case of samples, fortified with sodium pyruvate since contained all the requirements for the reactivation of heat-stressed cells[19]. In addition, this medium also contains a rapidly metabolisable energy source like pyruvate, which helps in the recovery of heat-stressed cells of *S. aureus*[20].

Enrichment of samples: Staphylococcal cells in the range of 10^4-10^9 CFU/ml were recovered from BHI-enriched samples of *Chhana* and *Rasogolla* prepared from samples of cow's milk inoculated with *S. aureus* A_{100}, B_{s-6} and C_{137}. However, *Staphylococci* were totally absent in the samples of *Chhana* and *Rasogolla* prepared from cow's milk inoculated with *S. aureus* D_{472} and E_{326} (Table 1) perhaps due to the irrecoverable injury caused to the cell.

In the case of BHI-enriched samples of *Chhana* and *Rasogolla* prepared from cow's milk inoculated with *S. aureus* A_{100} B_{s-6} and C_{137} respectively, the staphylococcal colonies which appeared on ETGPA plates after 72 hr were normal in appearance, except that the diameter of colonies were in the range of 0.5-1.0 mm. In the case of *S. aureus* C_{137}, each colony was surrounded by a hazy zone as well as a ring like structure which may be due to the resusscitation of the injured cells by BHI enrichment. Even here the staphylococcal colonies appeared on ETGPA plates after 72 hr of incubation at 37°C.

Effect on TDNase and enterotoxins: The levels of TDNase as evidenced in the zone diameters of 10-12 mm produced by *S. aureus* strains A_{100} and B_{s-6} after growth in cow's milk were carried over initially to *Chhana* and subsequently to *Rasogolla* (Table 2). However, with *S. aureus* C_{137}, D_{472} and E_{326} a reduction of only 2 mm from the initial level of 12-14 mm in the zone diameter of TDNase was observed in *Chhana* and *Rasogolla*. Enrichment in BHI of *Chhana* and *Rasogolla* prepared from milk inoculated with *S. aureus* A_{100} B_{s-6} and C_{137} resulted in an increased level of TDNase as evidenced in the zone diameters of 12-22 mm. No such changes were observed in *Chhana* and *Rasogolla* prepared from milk inoculated individually with *S. aureus* D_{472} and E_{326}. Similarly, detectable levels of preformed enterotoxins-SEA, SEB, SEC, SED and SEE produced by *S. aureus* test cultures were also carried over from inoculated cow's milk to *Chhana* and *Rasogolla* (Table 2).

The carry over of preformed TDNase and entero toxins from inoculated milk samples initially, into *Chhana* and subsequently to *Rasogolla* is not surprising, since both TDNase and enterotoxins are known to

TABLE 2. EFFECT OF CHHANA AND RASOGOLLA PREPARATIONS ON PREFORMED STAPHYLOCOCCOAL TDNASE ENTEROTOXINS IN MILK

	A_{100}		B_{S-6}		C_{137}		D_{472}		E_{326}	
	TDNase[+] (mm)	Toxin	TDNase (mm)	Toxin	TDNase (mm)	Toxin	TDNase (mm)	Toxin	TDNase (mm)	Toxin
Cow's milk (raw)	Nil	Nil	Nil	Nil	Nil	Nil	Nil	Nil	Nil	Nil
Incubated milk*	10	SEA	12	SEB	14	SEC	14	SED	12	SEE
Chhana (fresh)	10	SEA	12	SEB	12	SEC	12	SED	10	SEE
Rasogolla (fresh)	10	SEA	12	SEB	12	SEC	12	SED	10	SEE
Chhana (enriched)**	18	SEA	22	SEB	16	SEC	12	SED	10	SEE
Rasogolla (enriched)**	16	SEA	12	SEB	14	SEC	12	SED	10	SEE

*Milk inoculated with test cultures (1×10^6 CFU/ml of milk) and incubated at 37°C for 10-12 hr.

**One gram sample mixed in 99 ml of BHI broth and incubated at 37°C for 24 hr.

[+]Zone diameter includes 5 mm initial diamter of the agar well in TB-DNA agar plate.

be thermostable as well as resistant at pH range of 3.0 to 9.0[21-29]. The increase in levels of TDNase in BHI-enriched samples of *Chhana* and *Rasogolla* may have to be attributed to the resultant recovery and growth of heat injured cells.

The initial concentration of the enterotoxins, the duration of heating and the nature of substrates are some of the contributory factors.

Heat resistance of enterotoxins is better in a food menstruum than in a buffer, since the protein component of the food material offers protection to the enterotoxins against heat treatment.

The present study points out the potential public health hazards posed by *Chhana* and *Chhana*-based sweets, if these products are manufactured from raw milk initially contaminated heavily with enterotoxigenic strains of *S. aureus*.

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Comparative Study of Plant Gums, Gelatin and Sodium Alginate as Stabilizers in Buffalo Milk Ice Cream Mixes

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Guar gum, in low concentrations, produced pronounced effect on the viscosity of buffalo milk ice cream mix. The gum levels, consistent with ice cream quality on the basis of the organoleptic evaluation scores were; guar, 0.05%; *acacia* 0.50%; *karaya*, 0.10%; *dhak*, 0.25% as compared to 0.45% of gelatin and 0.30% sodium alginate. The ice cream body, texture and flavour scores were highly interrelated, the value of the coefficient of correlation being, $r = +0.88 \pm 0.07$.

The quality of ice cream depends upon the ingredients used in the mix among which selection of an appropriate stabilizer[1,2] is important. Gelatin and sodium alginate are widely used as stabilizers to improve the quality of ice cream. There has been a quest for stabilizers of plant origin as substitute for gelatin. According to Glicksman[3], hydrophillic plant gums in ice cream mixes reduces the amount of free water. Information on the use of gum *acacia*, *dhak*, *karaya* and guar in ice cream is scanty, hence this investigation was carried out to determine their suitability in buffalo milk ice cream.

Materials and Methods

Ice cream mix was constituted from composite full-fat buffalo milk of the Punjab Agricultural University herd. Alfa lavel separator was used for preparing cream. Skim milk solids of a commercial brand (*Verka* Punjab Dairy Development Corporation Ltd.) was used. Gums were obtained from local market. Edible quality guar gum was obtained from M/s Indian Gum Industries Ltd., Bombay. The stabilizers were incorporated in the ice cream mixes as solutions. Final levels were decided on the basis of preliminary tests based on the following:

Stabilizer		g/100g mix
Acacia (*Acacia arabica*)	—	0.25,0.50,0.75
Dhak (*Butea monosperma*)	—	0.25,0.50,0.75
Karaya (*Sterculia urens*)	—	0.10,0.20,0.30
Guar (*Cyamposis tetra-gonoloba*)	—	0.05,0.075,0.10
Sodium alginate	—	0.20,0.30,0.40
Gelatin	—	0.15,0.30,0.45

Ice cream: Plain ice cream mixes with the composition: fat 10 per cent, SNF 11 per cent, sugar 15 per cent, and varying quantities of stabilizers, were prepared by homogenizing the ingredients. To the warm (48.9°-54.5°C) cream and milk were added, mixed with skim milk solids, sugar and stabilizer, and the mixes were strained, pasteurized in a hot water bath at 71.1°C for 30 min, cooled and stored at 5°C. The mixes were aged in a refrigerator at 7.2°C for 4 to 12 hr prior to making ice cream from 1 kg mix in a batch type hand-freezer of 2 kg capacity. The frozen ice cream was kept for hardening in a deep freezer (-18°C) in waxed paper cups and withdrawn for evaluation of quality as required.

Viscosity of mixes: Viscosity of the mixes was determined using the Stormer viscometer (A. H. T. Co. USA) and expressed as time (sec) taken by 100 revolutions of the rotor in the mix at 20°C[4]. The viscosity was determined immediately after pasteurization and cooling and then after ageing for 4 and 12 hr, respectively.

Product evaluation: Sensory evaluation of ice cream samples was carried out by a panel of 10 judges, taking into account body, texture and flavour as the principle basis for assigning scores of 4, 3, 2 and 1 corresponding to excellent, very good, good and acceptable, respectively. The judges were served with four samples of ice cream at a time including the control. Sensory attributes data were examined statistically by the analysis of variance technique. Statistical significance of differences in mean quality scores due to various treatments was judged from the computed values of 'F' ratio (variance due to treatments/error variance) with the values from Senedecors tables[5]. Correlation

coefficients between body texture scores vs flavour scores were also calculated. Effect of storage on the body and texture was evaluated by transforming ranks into scores according to Fisher and Yates[6] and significances of differences in mean values by Duncan[7] multiple range test.

Results and Discussion

Effect of stabilizers and ageing on viscosity flow time of ice cream mixes: There was a profound effect of guar gum on the flow times of the ice cream mixes, even when incorporated at a level of 0.1 per cent as compared to the much higher levels of *karaya*, acacia, *dhak*, gelatin and sodium alginate stabilizers, respectively (Table 1). Even at a level of 0.075 per cent, guar gum produced significant increase in the body of the ice cream mix. Ageing of the ice cream mixes containing guar gum made negligible differences in the flow times. However, in the case of *Karaya*, *dhak*, gelatin and sodium alginate, ageing made a significant difference in the flow times of the mixes. In the case of *dhak* gum, the viscosity was maximum when the mix was aged for 12 hr as against 4 hr ageing reported by Dahle[8], Henning[9] and Mueller and Frandsen[10]. It is likely that, *karaya* gum used in the present investigation took longer time to hydrate when used in the buffalo milk ice cream mix. There was no significant effect of sodium alginate on the consistency of the mixes. Bendixen and Decker[11] also reported that sodium alginate produced mixes of lower viscosity than gelatin.

TABLE 1. EFFECT OF STABILIZERS AND AGEING ON THE RELATIVE VISCOSITIES OF ICE CREAM MIXES

Stabilizer level (g/100g)	Flow time (sec)[1] at the indicated ageing			'F' ratio	
	0 hr	4 hr	12 hr	Stabilizer**	Ageing*
			Guar		
0.00	21.8	23.5	22.2	12.65**	0.65NS
0.05	26.6	32.0	31.4		
0.075	34.9	39.6	36.6		
0.10	42.2	44.5	43.8		
			Karaya		
0.00	24.0	26.8	26.4	119.72**	8.64*
0.10	25.7	28.8	29.9		
0.20	30.4	32.4	27.5		
0.30	39.0	40.7	36.4		
			Acacia		
0.00	26.5	25.0	25.0	152.21**	3.59NS
0.25	27.5	27.8	27.7		
0.50	30.9	29.6	30.7		
0.75	35.4	33.7	34.9		
			Dhak		
0.00	23.6	23.5	23.9	109.53**	6.25*
0.25	24.1	24.0	24.6		
0.50	26.1	25.5	26.1		
0.75	27.4	27.3	28.7		
			Gelatin		
0.00	23.0	23.5	24.0	22.16**	6.73*
0.15	23.5	24.3	23.9		
0.30	23.6	24.0	24.8		
0.45	25.0	26.7	26.1		
			Na-alginate		
0.00	22.2	23.8	23.1	1.17NS	5.97*
0.20	22.8	23.8	24.4		
0.30	23.3	23.3	24.5		
0.40	23.2	23.2	24.4		

1 Time for 100 revolutions of stormer rotor
**Significant at P=0.01 } Variance due to treatment/error
*Significant at P=0.05 } variance
NS=Not significant

Effect of stabilizers on the quality of ice cream: Differences in the mean scores for body, texture and flavour showed statistically significant differences between the levels of the stabilizers used in the ice cream (Table 2). The concentrations of stabilizers which gave highest mean scores for body and texture were: guar gum, 0.05-0.075; *karaya*, 0.10-0.20; acacia, 0.5-0.75; *dhak*, 0.25; gelatin, 0.15-0.45 and sodium alginate, 0.30-0.40; per cent, respectively.

Interestingly, there was a good correspondence for the flavour scores, for similar concentration of the stabilizers in the ice cream mix. The results of mean scores, clearly showed that plant gums when used judiciously, are equally effective stabilizers as gelatin and sodium alginate. However, gum guar was more effective as a stabilizer at a much lower concentration than the rest of the gums. Acacia gum at a level of 0.5 per cent produced ice cream which obtained maximum scores where as *karaya*, at 0.1 per cent level, elicited highest mean flavour scores of 3.8 with equally high mean scores for body and texture. A level of 0.30 per cent sodium alginate produced better body, texture and flavour in ice cream. Dahle[12] recommended a similar level of sodium alginate for use in the ice cream from cow's milk. There was a high correlation between body and texture and flavour scores, the value of correlation coefficient being +0.88 which was statistically highly significant. The level of guar gum found suitable in the buffalo milk ice cream, was below the level of 0.4 per cent recommended by Steinsholt[13] for cows' milk ice cream. In respect of melt down properties at optimum levels of stabilizer, gelatin ice cream was more resistant to melting, followed by

TABLE 2. EFFECT OF STABILIZERS ON THE (MEAN SCORES) FOR QUALITY OF ICE CREAM

Stabilizer level (g/100g)	Body & texture[1]	'F' ratio	Flavour	'F' ratio	Stabilizer level (g/100g)	Body & texture[1]	'F' ratio	Flavour	'F' ratio
		Guar					**Dhak**		
0.00	2.1	7.21**	2.4	3.33**	0.00	2.1	5.42**	2.2	9.41**
0.05	3.4		3.3		0.25	3.4		3.4	
0.075	3.0		3.8		0.50	2.8		3.2	
0.10	2.1		2.7		0.75	2.0		2.3	
		Karaya					**Gelatin**		
0.00	2.6	1.80NS	2.6	4.78**	0.00	2.1	4.77**	2.4	6.42**
0.10	3.3		3.8		0.15	2.9		3.0	
0.20	3.0		3.1		0.30	2.8		3.4	
0.30	2.4		2.9		0.45	3.5		3.5	
		Acacia					**Na-alginate**		
0.00	2.1	5.10**	2.3	4.17**	0.00	2.0	11.34**	1.8	10.60**
0.25	2.7		3.2		0.20	2.6		2.8	
0.50	3.6		3.6		0.30	3.6		3.5	
0.75	2.9		3.2		0.40	3.1		2.7	

[1] 4-excellent 3-very good 2-good 1-acceptable

** significant at P=0.01 NS—not significant.

guar gum sample. In the present investigations, it has been observed that guar gum at the relatively higer levels produced ropiness and imparted a slimy feel to the product. Since ice cream was prepared in the hand-freezer, the over-runs were low, i.e. about 54 per cent as compared to higher over-runs achieved in mechanical freezers. However, guar gum seemed to depress the over-run more than the rest of the stabilizers.

Effect of stabilizers on the storage quality of ice cream: Body and texture and flavour scores of ice creams containing optimum levels of the stabilizers when stored for 35 days showed significant differences. Gelatin and gum acacia had better stabilizing effect on the body and texture of ice cream which stored equally well (Table 3). Ice cream having *dhak* gum was found to be coarser in texture due to crystal formation than others within 10 days of storage. Maximum deterioration in the texture was observed after 20 days storage of the control, *dhak* gum and sodium alginate ice creams unlike gelatin and gum acacia ice creams. Slight gumminess was observed in the stored guar gum ice cream. Gelatin has the property of absorbing water and forming a gel, thus avoiding the formation

TABLE 3. EFFECT OF STORAGE ON THE BODY AND TEXTURE OF ICE CREAM (MEAN SCORES) MADE WITH OPTIMIZED LEVELS OF STABILIZERS IN THE MIXES

Storage period (days)	Control	Acacia	Dhak	Gelatin	Guar	Karaya	Sodium-alginate	'F' ratio
0	–0.58	0.77	0.00	1.13	0.61	0.04	–0.07	14.64**
10	–0.32	0.70	–0.17	1.07	0.35	0.32	–0.06	7.41**
20	–0.27	0.99	–0.73	1.30	0.32	–0.11	–0.011	24.64**
35	–0.15	1.04	–0.45	1.11	0.12	0.10	–0.12	66.35**

**Significant at P=0.01

TABLE 4. EFFECT OF STORAGE ON THE FLAVOUR SCORE OF ICE CREAMS MADE WITH OPTIMIZED LEVELS OF STABILIZERS IN THE MIXES

Stabilizer	Stabilizer level (%)	Storage period (days)			
		0	10	20	35
Control		2.6	2.1	1.9	1.8
Guar	0.05	3.1	3.2	2.6	1.7
Karaya	0.10	3.1	3.0	2.3	2.3
Acacia	0.50	3.5	3.4	3.9	3.5
Dhak	0.25	3.0	2.1	1.6	1.6
Gelatin	0.45	3.9	3.3	3.8	3.2
Sodium alginate	0.30	2.8	2.6	2.2	1.7
'F' ratio	—	2.37*	9.32**	13.74**	10.16**

*P<0.05 4-Excellent; 3-Very good; 2-Good; 1-Acceptable
**P<0.01

of larger ice crystals in ice cream during storage[12], unlike in other samples where larger ice crystals were induced. There was no significant difference in the flavour of ice creams having gelatin and gum acacia, whereas significant difference were found in flavour scores of other ice creams (Table 4).

From the foregoing, it is evident that barring sodium alginate, the other gums, increased the consistency of the mixes, confirming the earlier results[14,15]. Ageing of mix prior to making ice cream was beneficial in most cases which was attributed to increased consistency of mixes. Gum acacia compared favourably with the customary stabilizer, gelatin for improving the quality of ice cream, but the quantity to be used (0.5 per cent) seemed high as compared to gum guar which was judged satisfactory at a much lower concentration of 0.05 per cent.

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RESEARCH NOTES

CHANGES INDUCED BY METANIL YELLOW AND BLUE VRS ON BONE MARROW CHROMOSOMES OF MICE

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Metanil Yellow and Blue VRS are commonly encountered non permitted dyes, in coloured foods in India. The effects of these dyes at the chromosomal as well as at the cytochemical levels were studied and found to be highly clastogenic *in vivo* in mice by both acute and chronic treatments.

A wide variety of dyes are used in the food, textile, printing industries and in chemical and biological laboratories[1], of which only eleven dyes are permitted in foods in India[2]. Blue VRS is now permitted only in Pakistan and Austria[3]. Recent study[4] indicates that about 70 per cent of coloured foods in Uttar-Pradesh contained non-permitted colours like Metanil Yellow and Blue VRS. Numerous publications are available about the toxicity of Metanil yellow and Blue VRS *in vivo* and *in vitro*[5-10]. But the effects of these dyes at the chromosomal level are meagre. Recent review by Combes and Haveland Smith[11] showed that the data on the effects of these dyes on the chromosomal level particularly in the *in vivo* system are lacking. Considering the serious health hazard problem due to the widespread use of these dyes in foods and industry the present investigation was undertaken to study the effects of these dyes on the bone marrow chromosome of mice by both acute and chronic treatment.

A laboratory bred strain of mouse *Mus musculus* of 75-day old was selected for this experiment. For acute treatment the dyes were dissolved in distilled water and were intraperitoneally injected to the five mice at the rate of 50 mg/kg body weight for 24 hr. Controls were injected with distilled water. For chronic treatment, dyes were forcibly fed to the five mice at the rate of 2 mg/kg of body weight for 30 days. Control mice were fed 0.1 ml of distilled water. After the treatment the animals were injected with 0.04 per cent colchicine at the rate of 1 ml/100 g body weight for 1½ hr before being sacrificed. For chromosome studies, flame dried preparations of bone marrow chromosomes were stained in Giemsa following the usual schedule.[12]

Chromosomal abnormalities induced by both Metanil Yellow and Blue VRS were grouped into three groups as follows:

Group I: Abnormalities principally involving spindle disturbances such as stickiness, C-mitosis, polyploidy, diplochromatid formation and centromeric stretching.

Group II: Those including alterations at the chromosomal level, such as breaks, gaps, deletion, centric fusion, dicentric formation and ring chromosome formation etc.

TABLE 1. PERCENTAGE OF CHROMOSOMAL AND MITOTIC ALTERATIONS INDUCED BY METANIL YELLOW AND BLUE VRS BOTH BY ACUTE AND CHRONIC TREATMENT*

Abnormality	Blue VRS				Metanil Yellow			
	Acute		Chronic		Acute		Chronic	
	Control	Treated	Control	Treated	Control	Treated	Control	Treated
Mitotic index	1.42±0.14	1.44±0.32	1.43±0.14	1.47±0.36	1.42±0.14	3.34±0.61**	1.43±0.14	1.16±0.25
Group I	2.67±0.91	11.52±1.49**	1.33±1.39	15.51±3.36**	2.67±0.91	9.30±1.24**	1.33±0.39	11.43±1.86**
Group II	2.33±0.91	2.72±2.83	2.33±0.01	3.61±3.80	2.33±0.91	8.87±2.51**	2.33±0.01	2.56±2.01
Group III	4.67±1.83	26.43±2.28**	4.33±1.90	16.04±5.29*	4.67±1.83	15.29±4.56*	4.33±1.90	28.04±4.90**

*300 metaphase scanned from 5 animal for each dose and for control.

Figures are mean±S.D.; Degrees of freedom=8.; *$P<0.01$ **$P<0.001$

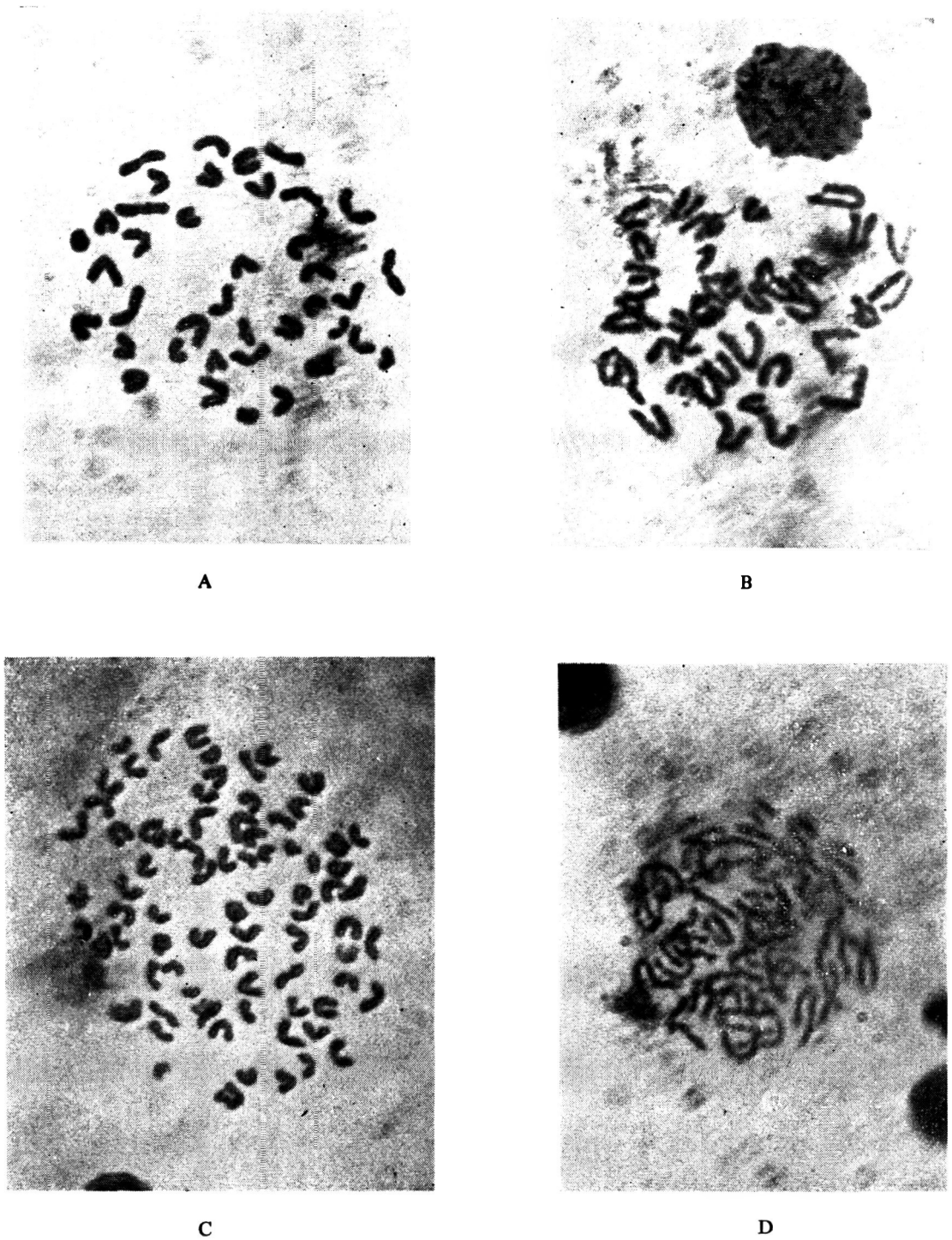

Fig. 1. Normal metaphase and some abnormalities induced by Metanil Yellow and Blue VRS.

A, Normal metaphase of *Mus musculus* showing 2n=40; B, Stickiness; C, Polyploid; D, Plate showing gross abnormalities.

Group III: Gross abnormalities i.e. when more than 10 chromosomes were affected and the major chromosomal changes where a number of chromosomal abnormalities could be observed in the same metaphase plate. These included fuzziness, pulverisation, erosion and translocation as well[13].

Acute and chronic study of Metanil Yellow and Blue VRS showed severe clastogenicity. In Metanil Yellow there was a significant increase in the mitotic index in case of acute treatment, but no such alterations were observed in chronic study (Table 1). In Blue VRS, mitotic index did not show any differences from the control by both acute and chronic treatment. So no conclusion could be drawn about the mitogenic or mitostatic nature of these dyes.

As regards the chromosomal changes in acute treatment there is a significant increase in the chromosomal abnormalities observed in the three groups of Metanil Yellow and Groups I and II by Blue VRS. In case of chronic treatment both the dyes showed a significant increase of the chromosomal abnormalities in Group I and Group III. No such alterations were observed in Group II in both the dyes. This confirms that both the dyes have a strong effect on the spindle function and the chromosome as a whole.

Hence this shows that Metanil Yellow and Blue VRS are highly toxic at the chromosomal and cytochemical levels. As these dyes are illegally used in food preparations, much attention should be paid to check the use of these dyes in the food.

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EFFECT OF SOME ANTIFUNGAL COMPOUNDS ON GROWTH OF *PENICILLIUM CITRINUM* AND CITRININ PRODUCTION

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The effect of antifungal compounds on the growth of *Penicillium citrinum* and the formation of citrinin was studied both in a liquid medium and on rice. Sorbic acid and butyl paraben were found to be the most effective inhibitors of citrinin synthesis in both synthetic medium and on rice. Calcium propionate, methyl paraben and propyl paraben also inhibited the synthesis of citrinin.

Extensive studies have been made in the past 20 years on aflatoxins and other mycotoxins with special reference to their formation in foods and feeds. Besides, the action of different chemicals, the solvents, insecticides and fungicides on the growth of mold and the effect of antifungal compounds like propionate, sodium acetate in controlling the formation of aflatoxin have been reported by a number of investigators[1-4].

Citrinin, potent toxin, inducing enlargement and tubular necrosis of the kidney has been detected in naturally contaminated grains and peanuts[5-7] of tropical region. The object of the reported study was to find out the effect of various antifungal compounds on the growth of *P. citrinum* and the formation of citrinin both in liquid medium and on rice.

The toxigenic strain of *P. citrinum* used, was isolated from stored groundnut cake[8]. The culture was maintained on potato dextrose agar slants. The synthetic medium used for the study consisted of (in per cent) maltose 4, sodium nitrate 0.2, KH_2PO_4 0.05, $MgSO_4$ 7 H_2O 0.02, $FeSO_4$ 0.00025, $ZnSO_4$ $7H_2O$ 0.00025, Na_2MoO_4 $2H_2O$ 0.00001, distilled water 100 ml. pH 5.8 ± 0.1. To this medium was added Sterilised antifungal compound (millipore 0.22 micron) in known concentration (Table 1) sterilized separately prior to inoculation. Fifty milliliter of the medium in a 250 ml Erlenmeyer flasks was inoculated with 0.1 ml spore suspension ($5\text{-}4\times10^8$ spores/ml) being collected

TABLE 1. EFFECT OF ANTIFUNGAL COMPOUNDS ON GROWTH AND CITRININ PRODUCTION BY P. CITRINUM IN A SYNTHETIC MEDIUM

Antifungal compounds Name	Concn. (mg/50ml)	Citrinin (μ g/ml)	Cell growth (g/l)
Control	—	410	10.1
Sorbic acid	0.5	120	5.1
	1.0	82	3.8
	2.0	51	2.7
	3.0	—	1.8
	5.0	—	—
	10.0	—	—
Calcium propionate	0.1	250	6.3
	0.5	150	5.1
	1.0	80	4.2
	5.0	39	2.3
	10.0	—	1.2
	20.0	—	—
Sodium bisulphite	0.5	200	6.5
	1.0	105	4.7
	5.0	98	4.4
	10.0	63	3.8
	15.0	45	3.5
	20.0	—	1.5
	30.0	—	—
	50.0	—	—
Sodium benzoate	100.0	255	8.4
	150.0	250	8.6
	200.0	235	7.5
	300.0	216	7.2
	500.0	195	5.9
Methyl paraben	0.1	215	7.7
	0.5	205	7.5
	1.0	175	7.6
	2.0	115	6.3
	3.0	97	5.4
	5.0	86	5.7
	10.0	41	4.3
	15.0	—	2.3
	20.0	—	0.6
	25.0	—	—
Propyl paraben	0.1	115	5.5
	0.5	88	5.1
	1.0	51	4.8
	3.0	49	4.8
	5.0	37	2.4
	10.0	—	1.5
	15.0	—	—
	20.0	—	—
Butyl paraben	0.1	58	4.2
	0.5	54	4.1
	1.0	47	3.5
	3.0	39	2.7
	5.0	—	1.0
	10.0	—	—
	15.0	—	—
Sodium sulphite	100	350	5.7
	150	320	5.0
	200	335	6.9
	400	285	5.8
	500	230	5.9
	800	198	4.2

TABLE 2. EFFECT OF INHIBITORS ON CITRININ PPODUCTION BY P. CITRINUM IN RICE

Antifungal compounds Name	Concn. (g/100 g)	Citrinin production on indicated days 5th day	7th day
Control	—	680	790
Sorbic acid	0.05	300	320
	0.1	90	130
	0.2	—	—
	0.3	—	—
Calcium propionate	0.05	280	310
	0.1	200	220
	0.2	190	200
	0.3	—	—
	0.4	—	—
Sodium bisulphite	0.05	580	690
	0.1	490	530
	0.2	300	340
	0.3	210	230
	0.4	110	150
	0.5	—	—
	0.6	—	—
Sodium sulphite	0.1	560	610
	0.3	480	490
	0.5	510	550
	0.8	490	500
	0.9	420	490
	1.0	390	410
Sodium benzoate	0.1	530	550
	0.3	540	490
	0.5	480	500
	0.8	450	480
	0.9	380	360
	1.0	370	360
Butyl paraben	0.05	220	230
	0.1	170	200
	0.15	30	80
	0.2	—	—
	0.3	—	—
Propyl paraben	0.05	440	530
	0.1	330	470
	0.15	190	210
	0.2	110	130
	0.3	30	40
	0.4	—	—
	0.5	—	—
Methyl paraben	0.05	340	380
	0.10	210	220
	0.15	200	210
	0.20	30	20
	0.30	—	—
	0.40	—	—

from a 7-day old culture slant. The flasks were agitated in a rotary shaker (120 rpm.) for 10 days maintaining the incubation temperature at 28 ± 1°C. The citrinin activity of the broth was determined by modified cup-plate method using *Bacillus subtilis* var. *mycoides* as the test organism[9]. Growth was determined after

centrifuging in the cold, washing thrice with water and drying at 60±5°C for 24 hr. The effect of antifungal compounds against the production of citrinin was also studied on rice, a good substrate, as follows: 10 g of polished broken rice with 50 per cent moisture taken in a 100 ml flask, was inoculated with 0.05 ml of spore suspension and incubated at 28°C in stationary condition. On the 5th and 7th day of fermentation citrinin was extracted with measured volume of 50 per cent alcohol, and combined extract was assayed[9]. A number of antifungal compounds including sorbic acid, calcium propionate, sodium sulphite, sodium bisulphite, sodium benzoate, methyl, propyl and butyl paraben were used (Table 1). Cell growth and citrinin produced on the 10th day in the liquid medium were recorded.

It is evident from Table 1 that out of eight compounds tested, sodium bisulphite, sorbic acid, calcium propionate, propyl and butyl parabens were potent inhibitors of cell growth and citrinin production by *P. citrinum*. Table 2 shows that sodium sulphite and sodium benzoate have some inhibitory effect on citrinin production by *P. citrinum* in rice, whereas sorbic acid and butyl paraben need to be added at 0.2 per cent for the inhibition of citrinin production. Methyl paraben and calcium propionate at 0.3 per cent and sodium bisulphite at 0.5 per cent completely inhibited citrinin production on rice. Of the various compounds tested sorbic acid and butyl paraben were found to be the most effective inhibitors of the synthesis of citrinin by *P. citrinum*.

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LIPID PROFILE AND FATTY ACID COMPOSITION OF FAT EXTRACTED FROM ARILS (MACE) OF *MYRISTICA FRAGRANS* AND SEEDS OF *ARTOCARPUS HIRSUTUS*

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The mace (*Myristica fragrans*) and the seed fat of wild jack fruit (*Artocarpus hirsutus*) contained 21.6 and 24.4% of total lipids (Chloroform-methanol, 2:1 V/V). Chloroform-soluble lipids constituted (as % weight) 83 and 88 made up of neutral lipids 60 and 63, glycolipids 27 and 21 and phospholipids 13 and 16 respectively. Neutral lipids were mostly glycerides, free fatty acids and hydrocarbons, with free sterols constituting 1.7 and 1.3% and sterol esters 0.9 and 5.6% in *M. fragrans* and *A. hirsutus* respectively. Palmitic acid is predominant, followed by Oleic acid, in all classes of mace lipids, and linoleic acid, is more followed by palmitic in *A. hirsutus* seed lipids.

Mace, the deep red, fleshy arils embracing the nut of *Myristica fragrans* (nutmeg), is used to flavour food products and liquors, and its oil to treat inflammation of the bladder and urinary tract[1]. Analytical characteristics of mace (aril) fat have been reported[2], but the class of lipid and its fatty acid composition have not been determined.

Wild jack (*Artocarpus hirsutus*), as well as the cultivated species, *Artocarpus integrifolia*, are grown in the western ghats. Wild jackfruit seeds are consumed, and the oil obtained from it is used for cooking. There is no report on the composition of seed fat.

Mace was obtained from the local market and the dehusked seeds of the wild jackfruit were collected from the forest. Both materials were dried, ground

fine and the lipids extracted with chloroform-methanol (2:1 V/V) mixture. The extracts were redissolved in the same solvent mixture, and passed through Sephadex G-25 column to remove non-lipid contaminants[3]. After weighing, the lipids were resolved into chloroform-soluble (the bulk) and chloroform-insoluble fractions. Characteristics of chloroform soluble fractions are reported here.

TABLE 1. LIPID CONTENT AND CLASSES OF MACE AND WILD JACK SEED

Lipids	Mace	Wild Jack seed
Total (%)	21.6	24.4
Chloroform-soluble (%)	17.8 (83)	21.4 (88)
Chloroform-insoluble (%)	3.8 (17)	3.0 (12)
Chloroform soluble		
Neutral	9.3 (60)	12.8 (63)
Glyco	4.2 (27)	4.7 (21)
Phospho	2.0 (13)	3.2 (16)
Neutral		
Glycerides, FFA, hydrocarbons	9.06 (97.4)	11.9 (93.1)
Free sterols	0.16 (1.7)	0.17 (1.3)
Sterol esters	0.08 (0.9)	0.83 (5.6)

Figures in parentheses indicate percentage

The chloroform-soluble lipids were resolved stepwise on a silicic acid column[4] into neutral lipids (Chloroform), glycolipids (acetone) and phospholipids (methanol). The neutral lipids were further analysed for total sterols as per Zlatkis *et al.*,[5] free sterols (digitonin precipitation) and sterol esters[6]. Fatty acid compositions of the total lipids and of each lipid class were determined after conversion to methyl esters (5 per cent methanolic HCl, 2 hr reflux)[7] on a CICC (Baroda) gas chromatograph using a 2M×3 mm column of 15 per cent DEGS on Chromosorb W at 180°C, N_2 gas and an FID. Peak area was calculated by triangulation without use of correction factors.

Mace (aril) fat: Lipids formed 21.6 per cent of total weight of the material, of this 83 per cent was soluble in chloroform (Table 1), which was made of 60 per cent neutral lipids 27 per cent, glycolipids and 13 per cent phospholipids. The neutral lipids contained 1.7 per cent free sterols and 0.9 per cent sterol esters, the remaining being glycerides, free fatty acids and hydrocarbons.

Palmitic acid was the predominant fatty acid in all the lipid classes (Table 2) constituting 37 per cent of the total fat. The GC graph showed two unidentified peaks after myristic acid (14:0) peak. Oleic acid stands next, contributing 26 per cent to the total lipids and 22 to 24 per cent to the constituent lipid classes. It is of interest that in the seed fats of other Myristica species different saturated acids are predominant; myristic acid (72 per cent) in *Myristica fragrans* (nut meg)[8], stearic acid (60 per cent) in *M. beddomei*[9] and

TABLE 2. FATTY ACID COMPOSITION (AS PER CENT WEIGHT) OF FATS FROM MACE AND WILD JACK SEED

Lipids	14:0	Peak I*	Peak II*	16:0	Peak III*	18:0	18:1	18:2	18:3
				Mace					
Total	2.0	2.0	2.3	37.0	—	6.5	26.2	9.4	14.6
Neutral	1.7	3.1	3.2	36.7	—	7.2	24.1	9.9	14.6
Glyco	24.1	Tr	Tr	23.3	—	6.3	24.3	Tr	22.0
Phospho	21.0	Tr	Tr	35.5	—	8.0	22.0	Tr	13.5
				Wild Jack Seed					
Total	3.0	—	—	20.8	3.9	5.0	11.0	56.3	Tr
Neutral	1.0	—	—	19.2	Tr	2.3	9.7	67.8	Tr
Glyco	8.6	—	—	39.9	15.0	8.1	13.8	14.6	Tr
Phospho	9.2	—	—	26.5	Tr	16.6	16.9	30.8	Tr

*Unidentified

Tr=Trace (<1%)

lauric acid (72 per cent) in *M. cannarica*[9]. In the aril fat of *M. fragrans*, palmitic acid is predominant.

Wild jackfruit seed fat: The seed contained 24.4 per cent total lipids 88 per cent being chloroform-soluble, and made up of neutral lipids (63 per cent) glycolipids (21 per cent) and phospholipids (16 per cent) and the neutral lipids containing 1.3 per cent free sterols and 5.6 per cent sterol esters, (Table 1). In the total lipids, neutral lipids, glycolipids and phospholipids, linolic acid forms 56, 68, 15 and 31 per cent and palmitic acid forms 21, 19, 40 and 27 per cent respectively, the rest being oleic, stearic and myristic acids.

The high percentage of total lipids in wild jackfruit seed is in contrast to the very small quantity of 0.4 per cent stated to occur in *Artocarpus heterophyllus*, the cultivated jackfruit seed. The high linoleic acid content of wild jackfruit seed fat is of some nutritional significance to those who consume the roasted seeds, but more so to those who use the oil for cooking.

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FATTY ACID COMPOSITION OF SEED OIL OF *TRIGONELLA CORNICULATA* LINN

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***Trigonella corniculata* contains 4.2% oil having a specific gravity of 0.9023 and iodine value of 105. Various fatty acids content of the seed oil has also been reported.**

Trigonella corniculata Linn (*Kasturi Methi*), a medicinal plant[1] has been reported to contain triacontane, sterols, alkaloids, steroideal saponins and flavonoids[1-6]. The characteristics and the fatty acid composition of the seed oil from Madhya Pradesh are reported here.

Oil extracted from the powdered *Trigonella corniculata* Linn seeds by petroleum ether (40-60) was studied. Methyl esters obtained by saponification, removal of unsaponifiable matter and esterification were studied by TLC, argentation TLC and GLC (Table 1). The particulars and operating conditions of GLC are: F & M Model 720; dual stainless steel columns (8'×3/16" O.D.); EGSS- × on chromosorb (40-60 mesh) carrier gas nitrogen; Injection port, columns and flame ionization detector block at 210, 300 and 300°C respectively; chart speed 15 in/hr.

TABLE 1. CHARACTERISTICS & FATTY ACID COMPOSITION OF *TRIGONELLA CORNICULATA* LINN SEED OIL

Yield	..	4.2%
Specific gravity at 30°C	..	0.9023
Refractive index at 20°C	..	1.4804
Saponification value	..	179
Iodine value	..	105
Palmitic (% w/w)	..	7.7
Stearic (% w/w)	..	1.9
Oleic (% w/w)	..	59.1
Linoleic (% w/w)	..	23.8
Linolenic (% w/w)	..	7.5

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COMPOSITION OF SWEET POTATO CULTIVARS AT HARVEST TIME

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Fifteen cultivars of freshly harvested sweet potatoes were analyzed for pH, total acidity, total solids, moisture content, carotenoid content, total phenols and orthodihydroxyphenols. Variations in mean measurements of the analyses were: pH 5.10-5.80, total acidity (as citric acid) 0.064-0.268%, total solids 24.0-34.6%, moisture content 65.3-75.8%, carotenoid 2.3-32.7 mg, total phenols 22.0-76.0 mg, and orthodihydroxyphenols 10.4-36.4 mg/100 g fresh tissue.

Sweet potato (*Ipomea batatas* (L) Lam.) is one of the world's major food staples which ranks high in commercial value and is also important as a vegetable crop. Knowledge of its chemical composition assumes real importance, and published literature deals with their sensitivity to cold temperature[1-3], infection fungi[4], starch compositon[5], protein and amino acid composition[6], phenolics[7,8], and carotenoid content[9]. Investigation was undertaken to determine the physical and chemical composition of fifteen sweet potato cultivars and the information collected is recorded here.

Fifteen cultivars of sweet potatoes, ('Centennial', 'Jasper', 'Jewel', 'LO-323', L3-243', 'L3-151', 'L4-62', 'L4-112', 'L4-131', 'Md-722', 'NC-345', 'NC-702', 'NC-718', 'NC-719' and 'V_2-237') harvested during the last week of September or the first week of October were obtained from Dr. T. P. Hernandez of Louisiana State University in Baton Rouge, Louisiana. A relative sample of each cultivar was immediately analyzed at the time of harvest. Three replicate samples (two roots each) were selected from each cultivar. All of the analyses performed were carried out in triplicates.

A 20 g portion of chopped sweet potato tissues was blended with 25 ml of distilled water and the pH determined using a Model 10 Corning pH meter[10]. Total solids determination consisted of weighing 10 g samples into tin boxes and drying at 70°C for 24 hr. Separate portions of sweet potato tissues were used to determine their moisture content. The methods as described by Constantin *et al.*[9] were used to determine the Carotenoid content of the sweet potatoes. Carotenoid contents were determined on 10 g samples of tissue, homogenized with 100 ml of hexane and then filtered. The optical density of the filtrate was determined at 400 nm. Carotenoid content was determined quantitatively by comparison with a beta-carotene standard curve. Ten grams of tissue were added to 50 ml of hot ethanol[11] ground for 2 min at high speed in a Virtis 45 homogenizer and filtered through ethanol-washed Whatman No. 2 filter paper and the total phenols were determined according to the methods of Simons and Ross[12]. The amount of orthodihydroxyphenol was estimated by the method of Johnson and Schaal[13] using a standard curve of chlorogenic acid. The total acidity was determined by extracting a known weight with water and titrating against 0.1 *N* NaOH to an endpoint of 8.2 on the pH meter[10] and expressing as per cent citric acid.

As shown in Table 1, there were considerable differences in the level of carotenoid, total solids, total phenols, orthodihydroxyphenols (0-DiOH phenols), ratio between total phenols and orthodihydroxyphenols and measurements for pH, moisture and total acidity in the 15 cultivars of sweet potatoes at the time of harvest. The pH of the cultivars varied from a minimum of 5.1 ('L4-131' and 'NC-145') to a maximum of 5.8 ('Centennial'). Total acidity was significantly high in 'Centennial' (0.268 per cent when compared to a low of 0.64 per cent in cultivar 'L3-151'. The highest and lowest moisture content were measured in cultivars 'L4-62' and 'NC-719' respectively. The carotenoid content ranged from a maximum of 32.7 mg in 'Jewel' to 2,3 mg in 'V_2-237'. The significantly high content of carotenoid in 'Jewel' may be due to the formation of more carotene molecules either from carotenoid pigment other than carotene or from precursors of some other type. The data would suggest that at harvest, as a source of provitamin A. 'Jewel' may be regarded as excellent while 'V_2-237' a poor source. 'NC-719' had the highest total solids. The highest content of total phenol was observed in 'LO-323' and the lowest in 'NC-719', while the orthodihydroxyphenol content was highest in 'LA-62' and the lowest

TABLE 1. CHEMICAL MEASUREMENTS ON SWEET POTATO CULTIVARS

Cultivar	pH	Total acidity[1] (%)	Total solids (%)	Moisture content (%)	Carotenoid (mg/100 g fresh tissue)	Total phenols[2] (mg/100 g fresh tissue)	0-DiOH phenols[3] (mg/100 g fresh tissue)	Ratio of total phenols 0-DiOH phenols
Centennial	5.80	0.268	29.7	70.2	20.2	74.9	23.6	3.17
Jasper	5.30	0.192	30.7	69.1	19.9	76.0	24.1	3.15
Jewel	5.40	0.198	33.3	66.7	32.7	39.0	15.6	2.50
L0-323	5.30	0.108	28.9	71.0	30.1	22.0	16.0	1.37
L3-243	5.40	0.121	30.0	70.2	21.6	32.0	23.5	1.36
L3-151	5.40	0.064	30.0	69.6	19.8	44.0	24.1	1.87
L4-62	5.50	0.096	24.0	75.8	12.8	48.0	36.4	1.31
L4-112	5.20	0.108	31.2	68.9	29.3	58.0	21.2	2.73
L4-131	5.10	0.128	30.7	69.1	14.9	49.0	26.7	1.83
MD-722	5.30	0.121	30.3	69.6	13.3	44.0	22.1	1.99
NC-345	5.10	0.121	33.0	66.9	10.7	49.0	13.6	3.60
NC-702	5.30	0.249	33.7	66.2	9.5	41.0	13.0	3.15
NC-718	5.30	0.256	32.4	67.5	16.2	22.0	16.7	1.31
NC-719	5.45	0.166	34.6	65.3	15.7	65.0	13.6	4.77
V_2-237	5.55	0.147	32.2	67.7	2.3	53.0	10.4	5.09

1,-Total acidity as citric $\frac{\text{ml base} \times N \times 0.64 \times 100}{\text{Sample (g)}}$

2,-Total phenols refer to the ortho—, meta-and the para isomers of the aromatic compounds.

3-Ortho dihydoxy phenols (0-DIH) refer only the ortho isomers of the aromatic compounds

'V_2-237'. The ratio between total phenols and orthodihydroxyphenols was 5.09 in 'V_2-237' and 1.31 in 'NC-718' and 'L4-62'.

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MICROFLORA OF *DAHI* PREPARED UNDER HOUSEHOLD CONDITIONS OF BANGALORE

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A majority of the samples of *dahi* collected from house holds and restaurants in Bangalore city contained both *Streptococci* and *Lactobacilli* in equal proportions while 35 per cent of them showed the presence of only *Lactobacilli*. A few samples contained coliform bacteria and spore formers while yeasts were present in all the samples. The titratable acidity of the samples ranged from 1.0 to 1.8 per cent. The average viable counts (per ml) of *Streptococci* and *Lactobacilli*, *Lactobacilli*, coliforms, sporeformers, yeasts and molds were 246×10^6, 132×10^6, 51, 17 and 2000 respectively. Among the 54 *Streptococci* isolates 46 were identified as *Str. thermophilus*, 4 as *Str. lactis* and 4 as *Str. cremoris*. The isolates of *Lactobacilli* were identified as *L. bulgaricus* (62), *L. casei* (8) and *L. helveticus* (4).

Dahi is a fermented milk extensively consumed in India. It is generally prepared domestically by traditional methods by inoculating boiled and cooled milk with a day old *dahi* and held at ambient temperature until it sets into a curd which may take 12 to 20 hr. During the preparation and handling of *dahi*, the product develops undesirable flavour and texture due to contamination by non-starter organisms. The type and number of starter bacteria and contaminants found in market samples of *dahi*, collected from different regions of the country have been reported earlier[1,2]. In this paper the number and types of lactic acid bacteria and non-lactic contaminants found in samples of *dahi* collected from private house holds and a few restaurants in Bangalore city have been reported.

Sixty samples of *dahi* collected during the months of September and October were examined for (*i*) organoleptic quality; (*ii*) titratable acidity; (*iii*) diacetyl content (qualitative method of Barritt,[3]); (*iv*) direct microscopic counts of *Streptococci* and *Lactobacilli*; (*v*) total viable counts on Elliker's lactic agar; and (*vi*) viable counts of *Lactobacilli* on Rogosa agar, coliforms on VRB agar, bacterial spores (surviving after heating *dahi* at 80°C/10min) on nutrient agar and yeasts and molds on malt agar[4]. Representative agar colonies of *Streptococci* and *Lactobacilli* were picked and transferred to yeast dextrose broth. The isolates after purification were examined for their cultural and physiological characteristics and their identity was established according to the descriptions given in Bergey's Manual[5].

TABLE 1. ANALYSIS OF HOUSEHOLD AND MARKET *DAHI* SAMPLES

Particulars	Min.	Max.	Average
Titratable acidity (% lactic acid)	1.0	1.80	1.33
Clump count/ml			
Streptococci ($\times10^6$)	0	1250	432
Lactobacilli ($\times10^6$)	74	1050	514
Viable count/ml			
Total count ($\times10^6$)	56	500	246
Lactobacilli ($\times10^6$)	11	350	132
Spores	0	30	17
Coliforms	0	110	51
Yeasts and moulds ($\times10^3$)	0.7	5	2.0

Note: Presence of diacetyl was indicated in 22 samples of *dahi*. 60 samples were tested.

It may be seen from Table 1 that the samples of *dahi* showed considerable variations in their physical consistency, flavour and acidity. The titratable acidity ranged from 1.0 to 1.8 per cent. Sixty per cent of the samples were very sour with titratable acidity exceeding 1.2 per cent and some of them also showed gassiness, curd floating and fruity flavour associated with the growth of yeasts. Only 22 samples gave the characteristic diacetyl flavour.

Microscopic examination revealed that 14 samples having more than 1.5 per cent acidity contained only *Lactobacilli* and in the remaining 46 samples both *Streptococci* and *Lactobacilli* more or less in equal proportions. Direct microscopic counts of *Streptococci* and *Lactobacilli* ranged from $0\text{-}1250\times10^6$/ml (average 432×10^6/ml) and from $74\text{—}1050\times10^6$/ml (average 514×10^6/ml) respectively. The total viable counts including *Streptococci* and *Lactobacilli* ranged from $56\text{—}500\times10^6$/ml (average 246×10^6/ml) while viable *Lactobacilli* counts of the samples ranged from $11\text{—}350\times10^6$/ml (average 132×10^6/ml). Viable counts of yeasts and molds, coliform bacteria, and spores in *dahi* samples were 700 - 5000, 0-110 and 0-30/ml respectively. Yeasts were present in all the samples while coliforms and spores were found in 20 and 12 samples respectively. Samples having more than 1.2 per cent of titratable acidity did not contain spore-formers. Yeasts are known to be common contaminants in

TABLE 2. ORGANISMS ISOLATED FROM *DAHI* SAMPLES

Organisms identified	Isolates (No.)
Str. thermophilus	46
Str. lactis	4
Str. cremoris	4
L. bulgaricus	62
L. helveticus	4
L. casei	8

sour milk products originating from the utensils and spoons used, hands of persons and atmosphere in the kitchen where *dahi* is prepared and handled. They may also be transferred along with the inoculum from the previous batch of curd. Spores may be present in milk surviving heat treatment. Presence of coliforms indicates contamination from utensils and hands of persons during the preparation and handling of *dahi*.

Out of 54 isolates of *Streptococci*, 46 were identified as *Str. thermophilus*, 4 as *Str. lactis* and 4 as *Str. cremoris*. Of the 74 isolates of *Lactobacilli* 62 were identified as *L. bulgaricus*, 8 as *L. casei* and 4 as *L. helveticus* (Table 2). Thus *Str. thermophilus* and *L. bulgaricus* constituted the predominant microflora of *dahi* samples collected in Bangalore which is in conformity with the observations of earlier workers[2]. The high acidity and sour taste of most of the samples may be attributed to the role of these two organisms.

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THE PRESERVATIVE EFFECT OF SOLID CARBON DIOXIDE AND CARBON DIOXIDE GAS ON QUALITY OF SALTED AND SWEETENED BUTTER MILK

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The shelf life and acceptability of salted and sweetened butter milk was studied for a period of 11 days at refrigeration temperature after the addition of solid carbon-dioxide or bubbling carbon dioxide. The titratable acidity increased rapidly in salted butter milk but it was slower in sweetened butter milk. The wheying off was found to be minimum in sweetened butter milk with solid carbon dioxide. Solid carbon dioxide was found to be more suitable for preserving salted butter milk whereas bubbling of carbon dioxide was best suited for sweetened butter milk in maintaining organoleptic quality.

Many of the organized dairies in India are marketing salted butter milk. Sweetened butter milk known as *Lassi* is marketed by some dairies. To help the production and marketing of butter milk, this investigation on preservative effect of solid carbon dioxide and carbon dioxide gas on the quality of salted and sweetened butter milk was carried out.

Fresh buffalo milk was modified to 1.5 per cent fat and 4.5 per cent solids non fat, divided into two portions and heated separately to 85°C for 30 min. To one part sugar was added at the rate of 10 per cent by weight of milk before heating when the temperature reached 65°C, gelatin at 0.25 per cent level was added for both the lots of milk in order to minimize the wheying off. Heated milk was cooled to 37°C, inoculated with starter culture DL[3] (obtained from NDRI, Karnal) at one per cent level and incubated at that temperature and the curd was churned to butter milk.

The sweetened butter milk was divided into 3 equal parts. One lot was bottled and carbonated with carbon dioxide gas (one minute at 5 lb pressure per square inch) and crown corked immeditaely. To the second portion pieces of dry ice was added (5 per cent by weight of butter milk), stirred well until the bubbling stopped, filled in bottles and crown corked. The third portion after bottling was used as control.

To the other half of butter milk, salt was added at 1 per cent level and similar treatments as stated above were given. All the butter milk bottles were stored at 5-8°C.

The butter milk samples-fresh as well as stored-were tested at periodic intervals for titratable acidity[1]. The evaluation of organoleptic quality was carried out according to the procedure of Harper and Hall[2] by a panel of five trained judges. The per cent wheying off was determined by measuring whey column height in butter milk.

Titratable acidity: The results given in Table 1 indicate that the acidity in salted as well as sweetened butter milk increased during storage in all the treatments. The rate of increase was more with carbon dioxide gas than with solid carbon dioxide. This increase in acidity may be due to protein bound carbon dioxide[3]. The lower acid content of sweetened butter milk, might be due to the sweetening agent[4].

Wheying off: Wheying off by sweetened butter

TABLE 1. MEAN TITRATABLE ACIDITY VALUES OF CULTURED BUTTER MILKS

Treatment	Titratable acidity (as % lactic acid) after indicated storage days					
	1st	3rd	5th	7th	9th	11th
			Salted butter milk			
Untreated	0.660	0.678	0.703	0.731	0.750	0.765
CO_2 bubbling	0.697	0.706	0.727	0.710	0.767	0.777
Solid CO_2 addition	0.686	0.696	0.716	0.742	0.759	0.773
			Sweetened butter milk			
Untreated	0.636	0.652	0.677	0.707	0.726	0.735
CO_2 bubbling	0.671	0.680	0.700	0.727	0.740	0.754
Solid CO_2 addition	0.660	0.671	0.692	0.716	0.738	0.750

TABLE 2. MEAN WHEYING OFF VALUES OF CULTURED BUTTER MILKS

Treatment	Wheying off values (%) after indicated storage days					
	1st	3rd	5th	7th	9th	11th
			Salted butter milk			
Untreated	0	11.82	15.10	17.04	18.79	20.17
CO_2 bubbling	0	9.76	13.19	16.49	17.70	18.35
Solid CO_2 addition	0	8.80	11.73	15.15	16.47	17.48
			Sweetened butter milk			
Untreated	0	3.28	6.79	10.46	11.93	14.12
CO_2 bubbling	0	2.21	4.90	8.47	10.09	11.89
Solid CO_2 addition	0	1.52	4.18	7.93	9.29	11.08

TABLE 3. MEAN ORGANOLEPTIC SCORE VALUES OF SALTED AND SWEETENED BUTTER MILKS AT DIFFERENT STORAGE PERIODS

Treatment	Organoleptic score after indicated days of Storage					
	1st	3rd	5th	7th	9th	11th
			Salted butter milk			
Untreated	98	95	92	90	86	83
CO_2 bubbling	97	97	95	94	92	90
Solid CO_2 addition	97	97	97	95	94	93
			Sweetened butter milk			
Untreated	99	97	94	90	88	86
CO_2 bubbling	98	97	96	94	93	92
Solid CO_2 addition	98	97	95	93	92	90

milk (Table 2) was less which might be due to the increase in total solids. The results also indicate that the preservatives used did influence wheying off, which was less in treated samples.

Organoleptic quality: The results in Table 3 indicate that addition of solid carbon dioxide preserves the organoleptic characteristics of salted butter milk whereas bubbling carbon dioxide is suitable for sweetened butter milk.

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YOGHURT FROM WHEY BASED RECONSTITUTED MILK

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Preparation of plain yoghurt containing 82 per cent fluid cheddar cheese whey as the only added diluent is described and the quality compared with that of yoghurt made from water based reconstituted milk and standardized cow milk with the same total solids content. The addition of fluid whey to the yoghurt mix instead of water during reconstitution resulted in a product which was rated excellent in flavour, body, texture and acceptance.

Milk whey contains soluble proteins, lactose, minerals, vitamins, etc., and is highly nutrtious[1]. Yoghurt, a fermented milk, has been reported to posses therapeutic and nutritional properties[2,3]. Studies were undertaken to fortify milk with whey for developing a more valuable fermented milk product.

In this study yoghurt manufactured by utilizing whey as a diluent was compared with that made from reconstituted milk using water and prepared from standard cow milk. Total solids level of the three samples were kept equal, but not the protein level as small variation does not influence the quality of yoghurt. The three yoghurt mixes used were:

Mix I (*T*1)*:* Cream with 30 per cent fat and non-fat-dry milk with 4 per cent moisture were compounded to give a mix of 3 per cent fat and 8.5 per cent solids non fat (SNF). Boiled (10 min) and filtered cheddar cheese whey was used to reconstitute the above, mix, the total solids of which increased to 16.5 per cent.

Mix II (*T*2)*:* Similar to (T1), but modified to give 3 per cent fat and 13.5 per cent SNF with water-based reconstituted milk.

Mix III (*T*3)*:* Fresh cow milk was standardized to 3 per cent fat and 13.5 per cent SNF by using non-fat dry milk.

The three mixes, were heated separately, to 85°C for 30 min and cooled to 45°C and inoculated with one per cent each of *Lactobacillus bulgaricus* and *Streptococcus thermophilus* culture. The inoculated mixes were filled in wax coated sanitized paper cups, incubated at 42±0.5°C until the formation of firm curd (3-3½ hr) and then stored in the cold (5-8°C).

TABLE 1. MEAN VALUES OF TITRATABLE ACIDITY, PH, PROTEIN AND LACTOSE OF YOGHURT MIX AND YOGHURT OF DIFFERENT TREATMENTS

Treatments	At manufacture		During storage (days)			
	BI	AI	1st	3rd	5th	7th
	Titratable Acidity (% lactic acid)					
T_1	0.27	0.88	1.13	1.29	1.40	1.59
T_2	0.19	0.89	1.16	1.32	1.54	1.65
T_3	0.28	0.97	1.11	1.30	1.48	1.57
	pH					
T_1	6.26	4.86	4.69	4.50	4.29	4.06
T_2	6.60	4.89	4.69	4.54	4.33	4.01
T_3	6.28	4.81	4.63	4.45	4.28	4.08
	Protein (%)					
T_1	4.11	4.43	4.46	4.53	4.65	4.85
T_2	5.07	5.28	5.34	5.49	5.64	5.94
T_3	5.21	5.47	5.50	5.55	5.71	5.86
	Lactose (%)					
T_1	8.92	7.10	6.67	6.46	6.06	5.94
T_2	7.08	6.38	6.00	5.63	5.48	5.38
T_3	7.56	6.79	6.42	6.33	6.27	5.66

T_1—Yoghurt from whey based reconstituted milk
T_2—Yoghurt from water based reconstituted milk
T_3—Yoghurt from standardized cow milk
Values are average of 8 replicates.
BI—Before inoculation (mix)
AI—After incubation (Yoghurt)

The three samples were tested for titratable acidity, pH, protein and lactose during processing and on alternate days of 7-days storage period as per ISI methods[4]. The organoleptic evaluation was done by 5 trained judges as per the procedure and score card recommended by Harper and Hall[5] for cultured dairy products. The results obtained are presented in Table 1.

Titratable acidity and pH: The average titratable acidities of T_1 and T_3 before inoculation were almost same but higher than T_2. After fermentation T3 showed higher acidity. The lactose content, prior to heat treatment of milk and protein levels might be responsible for the variation in acidity. The pH did not vary significantly during the manufacture and storage of yoghurt.

Protein level: There was an increase in the protein content in all the treatments which may be due to the reduction in the moisture content during storage and to some extent due to protein contributed by the lactic cultures[6].

Lactose: The lactose content decreased gradually in all the treatments during storage.

Organoleptic quality: The acid flavour was more pronounced in T1, followed by T3 and T2. The relative sweetness observed in T1 on first day increased on the 3rd and the 5th day. This may be due to the hydrolysis of lactose into monosaccharides. This might have also helped in the development of high acid flavour in T1, and resulted in acid flavour[7]. In T2 high acid flavour and bitterness were noticed right from the 3rd day, which might be due to high acidity, low pH and high protein content. The physical characteristics of T1, were rated excellent and the product was acceptable.

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A NOTE ON PROCESSING WEASANDS FROM MALE BUFFALO CALVES

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The esophagus from twenty slaughtered Murrah male buffalo calves of different body weights were collected, processed into weasands and their quality was assessed. Weasands obtained from 139.7±2.28 kg slaughter weight group were of Grade III quality, while those from 183.7±2.65kg slaughter weight could be grouped into Grade I (20%), Grade II (30%) and Grade III (50%). The entire *tunica mucosa* and part of the *tunica submucosa* of the esophagus form the weasand.

Various parts of the gastro-intestinal tract of the food animals are being used as sausage casings since time immemorial. Each country produces different types of sausages, making use of different varieties of casings. In India, sausage industry is still in its infancy. While intestines are converted into casings, the esophagus of cattle and buffaloes are not being fully utilised for making weasand. There is good market for properly processed quality weasand. No work has been reported so far from India regarding the production of weasand particularly from the esophagus of buffaloes.

The esophagus from twenty Murrah male buffalo calves slaughtered for meat production were collected. Ten calves (Group A) were slaughtered at an average slaughter weight of 139.7±2.28 kg. The other ten calves (Group B) were slaughtered at an average slaughter weight of 183.7±2.65 kg.

Following slaughter, flaying and evisceration, the esophagus was carefully collected, cleaned with tap water and the length was recorded. It has different layers viz. *tunica mucosa*, *tunica submucosa*, *tunica muscularis and tunica serosa*[1].

The esophagus was turned inside out to expose the mucous surface and was inspected for parasitic damage, blood spots, ulcers, cuts, etc. Those free from major damages were processed into weasands. One of the ends of the muscular part of the esophagus was hooked up and tied to a peg with a short thread and the other end held taut. With the help of a sharp knife, the loosely attached submucosa together with the mucous membrane was deflected from the tied up end and its attachement was carefully severed to the entire length of the esophagus. This constituted the weasand. It was stretched and the length was

recorded. Thus, a long, transluscent and thin tubular material was obtained leaving the muscular coats and the fibrous coat. The wet weasand was washed and tied at one end, inflated and tied at the other end, and allowed to dry in inflated condition in the shade at room temperature for 24 to 48 hr when it became dry. The dried weasand was a long, baloon-like structure. The knot at one end was cut off and flattened. Its length and diameter were recorded. The final inspection was done by viewing the weasand against a source of light, and graded into three grades[2].

The mean values of quality measurements of weasands in relation to weight of animals and length of esophagus are presented in Table I.

The weasands from Group A animals fell under Grade III eventhough a few of the weasands were reaching the length prescribed for Grade I, the diameters were less than 7.6 cm.

In Group B, 20 per cent of the weasands fell under Grade I, 30 per cent under Grade II and the remaining under Grade III.

The overall observation of the weasands revealed that they were free from blood spots or parasitic damages. There is good scope for the production of weasand of Grade I and II provided care is taken to harvest esophagus with longer lengths. This is possible if during slaughtering, the cutting of major blood vessels is done closer to the mandibular angle of the animal after extending the neck. Further, during evisceration, the esophagus should be cut close to its entry into the rumen. Care should be exercised while separating the weasand from muscular layers of the esophagus to prevent any damages.

According to Sisson[3], in medium-sized cattle, the esophagus measures about 90 to 105 cm which more or less equals to that of medium-sized buffaloes. About 80 to 90 cm dry weasand may be obtained from them with an average flat diameter of over 7.6 cm. Such weasands will be of Grade I quality provided they are free from grubs, blood spots, pin-holes, etc.

Mann[2], Brandly *et al.*[4] and Libby[5] stated that weasand is prepared from mucous lining of the esophagus, whereas Thornton and Gracey[6] observed that the "serous covering" of the esophagus is utilised as sausage casing. The entire *tunica mucosa* and part of the *tunica submucosa* together form the weasand and not the "serous covering" of the esophagus.

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TABLE 1. QUALITY PARAMETERS OF WEASAND IN RELATION TO WEIGHT OF ANIMALS AND LENGTH OF ESOPHAGUS

Particulars	Group A	Group B
Live wt (kg)	145.8±1.43	192.4±2.81
Slaughter wt (kg)	139.7±2.28	183.7±2.65
Length of esophagus (cm)	59.3±1.17	63.9±1.06
Length of wet weasand (cm)	57.7±1.40	63.0±1.09
Length of dry weasand (cm)	51.6±1.33	56.6±1.02
Diameter of dry weasand (cm)	6.4±2.94 (cm)	7.3±0.16

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SOLUBLE NITROGEN LOSSES IN SQUIDS (*LOLIGO DUVAUCELI*) DURING STORAGE IN SLUSH ICE

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Total water soluble and Non-protein Nitrogen fractions decreased considerably in dressed squid mantles stored in a system of crushed ice and meltwater after 8 hr. The Nitrogen fractions appeared in the meltwater immediately and their concentration increased with time, non-protein Nitrogen contributing the major part.

Dressed Squid mantles, referred as 'squid tubes' ('*tubes*') in the seafood industry are stored in crushed

ice and its meltwater in non-perforated containers, prior to freezing. It has been observed that the meltwater becomes turbid acquiring white colour. Preliminary observations indicated that the meltwater had maximum optical density (O. D.) with the blue filter (No.42 of a Klett-Summerson) in a colorimeter and also some precipitable substances with trichloroacetic acid. This experiment was carried out to determine the rate and quantum of this leaching from squid tubes in the light of their high water soluble nitrogen content[1].

Fresh Squids[3] (*Loligo duvauceli*) of medium size (8-12 cm mantle length) in uniced condition were procured from trawler landing centre of Veraval and immediately brought to the laboratory. Guts, skin, head, fins and pen were removed and the squid tubes were washed once in water. The 'tubes' were then mixed with ice and water in the ratio of 1:2:0.2 by weight, in an enamelled non-perforated container. The container was placed in an insulated aluminium ice-box (60×37×40 cm i. d. with 5 cm thick Thermocole insulation between the walls) packed with big ice pieces. Seven ml samples of meltwater were drawn from the bottom of the container periodically at 2 hourly intervals upto 8 hr. Quantity of water drawn was replaced with an equal volume of cold water. They were analysed for Total nitrogen (TN) and non-protein nitrogen (NPN) by micro-Kjeldahl method[2]. The weight of 'tubes', ice and water were recorded before and after storage. Moisture content, TN, NPN and water soluble nitrogen (WSN) of the squid 'tubes', before and after storage were also determined, following AOAC procedures[2]. Appropriate blanks were run in all cases.

The characteristics of meltwater in the squid tubes slush ice system are presented in Table 1.

The Nitrogenous material could be detected in the meltwater soonafter the 'tubes' were placed in ice and water indicating high solubility of these fractions. The TN content of the meltwater increased from 0.37 to 3.5 mg/ml after 8 hr of storage. The rate of increase was most rapid in the first 2 hr and later on gradually decreased with time. NPN increased from 0.23 to 3.05 mg/ml at the end of 8 hr and the rate of its increase was similar to that of TN. NPN comprised the major portion of the TN through out the storage period, increasing from about 62 per cent initially to about 86 per cent after 2 hr with a marginal increase there afterwards. Optical density of meltwater was found to decrease with time unless disturbed mechanically indicating the settling down of suspended particles.

The moisture content of 'tubes' increased from 78.33 to 83.08 per cent at the end of 8 hr of storage, but with an increase of only 81 g in weight. The weights of ice and water varied from 1770 & 177 to 840 & 1030 g respectively. About 28 g of solids amounting to 3 per cent of the fresh weight of squid mantles was lost during storage.

Changes which occurred in the nitrogen fractions of squid 'tubes' due to the storage are presented in Table 2.

Nitrogen fractions after storage, were seen to decrease in all the fractions, the greatest decrease being in the WSN fraction. There was not only quantitative decrease, but also in their relative proportions in the TN. Thus the proportions of WSN in TN decreased from 45 to 29 per cent, while that of NPN in TN from 23 to 17 per cent. The decrease observed in 8 hr were higher than those reported by Joseph *et al.*[3] in one day, stored squids in ice, draining the meltwater.

Thus the practice of storing squid 'tubes' in slush ice does not lead to a decrease in the physical yield of the product, but rather an increase. But as the loss in the solids leached out occurs predominantly in the NPN fraction, this could affect the organoleptic properties of the squids in general and its sweet taste in particular which is linked to the NPN fraction[4].

TABLE 1. CHARACTERISTICS OF MELTWATER IN THE SQUID-TUBE-SLUSH ICE SYSTEM

Storage period (hr)	T. N. (mg/ml)	N. P. N. (mg/ml)	N.P.N./T.N (%)
Initial	0.37	0.23	61.8
2	2.30	1.98	86.0
4	2.98	2.55	85.5
6	3.26	2.86	87.3
8	3.50	3.05	87.0

TABLE 2. NITROGEN FRACTIONS OF SQUID 'TUBES' BEFORE AND AFTER STORAGE IN SLUSH ICE

Storage	T. N. (%)	W. S. N. (%)	N. P. N. (%)
Before	3.11	1.41 (45.34)	0.72 (23.01)
After	2.48	0.72 (29.19)	0.43 (17.17)

Values on as is basis. Figures in parenthesis indicate as a % of the TN.

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LIPID COMPONENTS OF CLOVE

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Linoleic and palmitic acids were the main fatty acids of clove fat. An alkane present in the nonsaponifiable lipid fraction of clove was identified as hentriacontane

Clove (*Eugenia caryophyllata* Thunb) is found to contain 0.3 per cent of lipid materials of which the nonsaponifiable fraction (NSF) constitutes the major part. Narayanan and Natu[1] reported sitosterol along with triterpene acids in the spice. The presence of glucosides of sitosterol, stigmasterol and campesterol in clove was reported by Brieskorn and coworkers[2]. There is only one report[3] on the simple lipids while no information on the waxy materials present in clove is available. The present study describes the component fatty acids and a predominant waxy hydrocarbon of clove.

Dried clove procured from local gardens was steam distilled and the residue after drying was soxhlet extracted with petroleum ether (40-60°C) for 24 hr. One kilogram dried clove yielded 4.3g of extract which invariably contained some eugenol as impurity. The total extract was washed with dilute sodium bicarbonate solution to separate the free fatty acids (Traces) and then washed with dilute sodium hydroxide solution in the cold to remove the phenolic compounds. The remaining neutral lipid fraction (2.9g) was saponified by standard procedure[4] and the unsaponifiable fraction (2 g) was extracted with ethyl ether. The fatty acids liberated were converted into methyl esters (0.58 g) by refluxing with methanolic HCl (5 per cent) at 100°C for 2 hr. The methyl esters formed were extracted with petroleum ether (40-60°C) and the solvent was evaporated under nitrogen. The esters were then collected in chloroform and stored in refrigerator till gas chromatographic analysis. The nonsaponifiable fraction was chromatographed on alumina (Brockmaw Grade I) by elution with petroleum ether (40-60°C) (Fraction I) followed by ethyl ether (Fraction II). Fraction I was purified by preparatory silica gel TLC using petroleum ether as eluent and a compound with melting point 65.5°C was isolated. Analysis of Fraction II is in progress.

The fatty acid methylesters were analysed by a Hewlett Packard 5840 A gas chromatograph using a 6'×1/8" (i.d.) diethylene glycol succinate column with temperature programme from 100 to 190°C @ 5°C/min, nitrogen as carrier gas (20 ml/min), FID temperature at 300°C and injection temperature at 275°C. The retention times of the methyl esters were compared with those of authentic samples and their amounts were calculated as the average of triplicate analysis values of integrated area percentages. The compound isolated from the petroleum ether fraction of NSF (Fraction I) was analysed for I.R. (as KBr pellets using Perkin-Elmer 299 B Infrared Spectrophotometer) and mass spectra (using AEI MS 3074 double beam mass spectrometer with inlet temperature at 150°C and electron energy 70 eV).

The gas chromatographic analysis indicated the presence of the common fatty acids (C_6 to C_{20}) in clove fat. The data are given in Table 1. Linoleic ($C_{18:2}$) and Palmitic ($C_{16:0}$) acid esters were the

TABLE 1. FATTY ACID COMPOSITION OF CLOVE

Fatty acids	% Content
$C_{6:0}$	0.16
$C_{8:0}$	0.26
$C_{10:0}$	0.94
$C_{12:0}$	2.12
$C_{14:0}$	4.74
$C_{16:0}$	23.94
$C_{17:0}$	2.54
$C_{18:0}$	7.56
$C_{18:1}$	6.06
$C_{18:2}$	29.30
$C_{18:3}$	5.45
$C_{22:0}$	3.58

two main fatty acid esters among those identified. This is in contrast with the earlier report[3] where stearic acid ($C_{18:0}$) has been reported as the predominant fatty acid (89 per cent) in clove and the total saturated fatty acids as approximately 94 per cent. The present study gave the saturated to unsaturated fatty acids ratio of 1:0.89. The observations are fairly in agreement with literature[4] where as a general rule Myrtaceae family to which clove belongs is reported to possess C_{18} un-saturated fatty acids as the major component acids.

The waxy compound from Fraction I moved to the solvent front when chromatographed (TLC on silica gel/petroleum ether) indicating its nonpolar nature. IR spectra of the compound showed no functional groups and the molecular formula of the compound was found to be $C_{31}H_{64}$ by analysis. The fragmentation pattern of the compound (M^+ 436) showed prominent peaks at m/e 57,71,43 and 85 followed by series of peaks spaced 14 mass units apart corresponding to C_nH_{2n+1} ions indicating that it is a straight chain alkane, hentriacontane.

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HAEMATOLOGICAL CHANGES IN *SAROTHERODON MOSSAMBICUS* EXPOSED TO LINDANE

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Sublethal concentration (0.05 mg/1) of lindane caused a dramatic fall in oxygen consumption by fish exposed to lindane and in turn resulted in changes in haematological parameters. The degree of change increased with exposure time.

Of the large number of chemical pesticides developed for use in agriculture, veterinary and medical fields a number of them are known to be highly toxic to fish and other aquatic animals, although in practice most of them are not likely to contaminate fresh water in biologically significant quantities[1]. Lindane is a potent and widely used pesticide in India because of its high toxicity and relative high biodegradability[2]. The chronic effects of lindane on *Salvelinus frontalis*, *Lepomis macrochirus* and *Anabas testudineus* have been reported earlier[3,4]. Literature on toxic effects of lindane on haemopoietic system of a fresh water edible fish, *Sarotherodon mossambicus*, is sparce. Therefore, an attempt is made to find out the haemotological changes in fish exposed to Lindane and the results are incorporated here.

Fish weighing 10±2 g were caught from the local ponds around Tirupati, transferred to laboratory fish tanks and acclimatised to 27±2°C. They were fed with groundnut cake daily and frog leg muscle twice a week. After one week, they were exposed to different concentrations of lindane (99 per cent technical grade-supplied by Bharat Pulverising Private Limited, Bombay-20) according to their biomass ratio[5]. The stock solution was prepared by dissolving lindane in acetone. The LC_{50} value was found to be 0.15 mg/l[6]. One third of the LC_{50} concentration of lindane (0.05 mg/l) was used as a sublethal dose for experimental purpose. The pesticide medium was also changed daily by mixing the required amount of lindane stock solution to maintain the medium concentration. On 1, 2, 4, and 6 days of exposure, the blood samples were collected with hypodermic syringe for haematological analysis. The red blood corpuscles (RBC) count was made with Neubaur crystalline counting chamber[7], haemoglobin content (Hb) was estimated by acid haematin method[8] and packed cell volume (Pcv) by microhaematocrit method[9]. Mean corpuscular volume (Mcv), mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC) were calculated[10] after obtaining RBC, Hb and PCV values. The whole animal oxygen consumption was determined by Winkler's iodometric method[11].

Table 1 gives the haematological values of control and lindane exposed fish. The entire haematological profile of the exposed fish changed from the 2nd day onwards. The changes upto 24 hr were not significant. Table 2 indicates the fall in oxygen uptake by the treated fish. The fall in oxygen consumption of lindane exposed fish is proportionate with the increment in the haematological parameters.

In the present investigation the gradual increase in RBC, Hb, PCV content might be due to hypoxic conditions prevailed in the animals. The diminished uptake of oxygen from the environment by the

TABLE 1. HAEMOGRAM OF *S. MOSSAMBICUS* EXPOSED TO LINDANE

Component	Control	1 day	% change	2 day	% change	4 day	% change	6 day	% change
R.B.C. ($\times 10^6$ m/c mm)	2.196 ±0.135	2.228 ±0.096NS	+1.429	2.36 ±0.161*	+7.448	2.518 ±0.146**	+14.63	2.657 ±0.091**	+20.95
Haemoglobin (g/100 ml)	7.643 ±0.115	7.686 ±0.118*	+0.563	7.767 ±0.119*	+1.618	7.923 ±0.054**	+3.645	8.155 ±0.093**	+6.698
Packed cell volume (%)	25.23 ±0.176	25.57 ±0.204NS	+0.137	25.731 ±0.172*	+0.74	25.97 ±0.265**	+1.712	27.08 ±0.258**	+6.046
Mean corpuscular volume (c μ)	116.15 ±6.96	114.19 ±3.859NS	−1.698	109.38 ±6.876NS	−5.832	103.47 ±5.659**	−10.92	102.03 ±3.467*	−12.16
Mean corpuscular haemoglobin ($\mu\mu$ g)	34.89 ±2.042	34.32 ±1.569NS	−1.67	33.09 ±2.32NS	−5.162	31.56 ±1.858**	−9.55	30.73 ±1.31**	−11.95
Mean corpuscular haemoglobin concentration (%)	29.93 ±0.562	30.06 ±0.595NS	+0.43	30.19 ±0.737NS	+0.869	30.49 ±0.271NS	+1.865	30.119 ±0.593*	+0.621

Each value is the mean±S.D. of 6 individual observations. P='t' test
*Significant ($P<0.05$): **Highly significant ($P<0.001$)
N.S.: Not significant.

TABLE 2. WHOLE ANIMAL (TOTAL) RESPIRATION (mL O_2 CONSUMED/gFISH/HR) IN LINDANE EXPOSED FISH

	Lindane exposure period (in days)							
Control	1 day	% change	2 day	% change	4 day	% change	6 day	% change
0.234 ±0.032	0.199 ±0.01*	−14.96	0.170 ±0.016**	−27.35	0.154 ±0.021**	−34.18	0.141 ±0.019**	−39.74

Each value is the mean±S.D. of 6 individual observations. P='t' test.
*Significant ($P<0.02$); **Highly singnificant ($P<0.001$)

experimental fish might be due to the rupture of respiratory epithelium of the gill[6]. The physiological disfunctioning of haemopoietic system might have caused the alternatives in the haematological values[12]. Sublethal concentrations of some synthetic chemicals have been reported to increase the total haemogram of certain fishes[13-16]. It is obvious that the lindane intoxicated fish tries to cope with the adverse conditions by enhancing their respiratory capability through elevated RBC and Hb synthesis. Therefore, prolonged exposure of fishes to organochlorine compounds poses a great threat for their better survival.

The authors are grateful to Prof. R. Ramamurthi for providing fecilities and encouragement throughout the investigation. One of the authors, Madhu, Ch. is thankful to CSIR for providing SRF and also to M/s Bharat Pulversing Mills Private Limited, Bombay-20, for providing samples of technical grade lindane.

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OBITUARY

The Association of Food Scientists and Technologists (India) records its deep sorrow for the sad demise of Sri Dayanand, Ex-Director (Fruit & Vegetable products), Department of Food, Government of India on 28th January 1984. He was also the President of AFST (I) (1979–80).

We also record the sad demise of Sri B. A. Satyanarayana Rao, Scientist, CFTRI, on 2nd February 1984, who was a member of the Editorial Board of Indian Food Industry.

BOOK REVIEWS

Evaluation of certain food Additives and Contaminants: Twenty-seventh Report of the Joint FAO/WHO Expert Committee on Food Additives, Technical Report Series 696, World Health Organization, Geneva; 1983; pp. 49.

The Report of the Joint FAO/WHO Expert Committee on Food Additives which met in Geneva on 11-20 April, 1983 deals with the history and activities of the Expert Committee on Food Additives. In the report it has been emphasised that consumers in both developing and developed countries are demanding firm assurance that food additives were safe, which calls for their appropriate and thorough testing followed by sound and balanced evaluation. It has also been brought out that technological, nutritional and other public health considerations makes it all the more necessary to ensure the identity and purity of food additives. The report details the various tasks before the committee, viz.,

(*a*) to prepare specifications for the identity and purity of certain food additives and to carry out toxicological evaluations of them;

(*b*) to review specifications for selected food additives;

(*c*) to undertake toxicological evaluations and re-evaluations of certain food additives and contaminants; and

(*d*) to undertake the toxicological evaluation of xenobiotic anabolic compounds used in the rearing of live-stock intended for human consumption.

Specific food additives under the headings-Antioxidants. Extraction solvents, Flavouring agents, Food colours, Preservatives, Sweetening agents, Thickening agents, Miscellaneous food additives, besides dealing with contaminants such as metals and xenobiotic anabolic agents has been brought out with ADI & Specifications wherever finalised based on available data.

The report highlights the recommendations made by the Committee to FAO and WHO stressing the urgency of implementing the recommendations made in its 25th report that "a group of experts be convened, as soon as possible, to study the application of advances in methodology to the toxicological evaluation of food additives and contaminants and also of pesticide residues".

There are three Annexures-Annexure 1 dealing with the Report and other documents resulting from previous meetings of the Joint FAO/WHO Expert Committee on Food Additives; Annexure 2-Acceptable daily intakes and information on specifications; Annexure 3-Further toxicological studies and information required or desired by the committee.

This report will be a useful guideline for Technologists working in the Food Industries as well as Experts dealing with the Food Quality Assurance Programme in the country.

O. P. KAPUR
C.F.T.R.I., MYSORE

Principles and Practices for Harvesting and Handling Fruits and Nuts: Edited by M. O' brien, B. F. Cargill, and R. B. Fridley; AVI Publishing Company, Inc. Westport, Connecticut USA; 1983; PP 636; Price: $ 55.00; [$ 60.50 elsewhere].

This book gives an excellent and exhaustive information on the engineering principles and concepts used for mechanical harvest and handling of fruits.

It comprises of 4 main sections, each section with a number of chapters covering aspects like methods of harvesting both manual and mechanical, post harvest care, harvesting systems existing for specific fruits, nuts and its improvement.

Section 1 deals with a brief history on evolution of harvesting, steps involved in harvesting operations, effect of genetic characters. cultural practices like pruning, thinning, planting, fertilizer application, irrigation, harvest maturity on storage and marketing. The impact on social development and economic aspects of manual and mechanical harvests are also described.

Section 2 deals with the principles of harvesting, so as to make the harvest less labour intensive. Starting with man as the major fruit harvester it explains the various processes involved like hand picking,, collection of picked fruits, improvement of manual harvesting by use of mechanical aids for tree fruits and small fruits, comparison of manual and mechanical aids, appropriate devices to be adopted for detachment of fruits: for eg. detachement by combing and stripping in berries, cutting for grapes, pulling and twisting for apples and citrus. It also gives a vivid account of vibratory mechanisms adopted for tree shaking, application of tree shakers in high density orchards, pick up mechanisms and collectors for collecting the harvested produce.

In the 3rd Section on post harvest operations, authors give a list of operations to be followed either in the field or packing houses to protect the harvest quality of the produce; otherwise it loses its marketability incurring economic losses. This includes conveying operations using different types of conveyors, methods for removing nonedible portion of the fruit like husk, stem end, sorting by manual and mechancial means, washing, drying, waxing, heat treatment, fumigation, degreening, cooling to reduce the metabolic activity in the produce and shrivelling. Stress is laid on suitable packing for fresh market, devicing suitable containers for bulk handling and storage followed by the quality certification. Transportation in suitable packages and efficient handling during loading and unloading is very important as it minimises bruising of the produce.

Section 4 deals with the various types of harvesting systems adopted for bush, vine and tree crops. These systems are evolved by integration of several principles and practices suiting the biological and economic aspects of the crop. A vivid picture of the evolution of harvesting of different types of berries, change of cultural practices adopted for mechanical harvesting are discussed. In grapes different training systems suitable for mechanical harvest is indicated, especially mechanization adopted for raisin grape and wine grape harvest is very interesting. Under tree fruit harvesting systems, the authors discuss about the present status of harvesting system, types of harvesters used for almond, walnuts, macadamia nuts, pistachio, citrus, avocado, pineapple coffee, olives and others.

A big lacuna in harvesting of tropical fruits especially mango and banana is seen. Perhaps this void can be filled up by the agricultural engineers of India, after going through this book. Mechanised harvesting is essential because of two reasons, (*i*) increasing shortage of labour and (*ii*) because people are becoming more sophisticated and as a result they avoid onerous manual labour to seek more time for personal pleasure. Increased mechanization allows the increased production demanded by increased consumption.

The book is very informative, comprehensive, up-to-date supported by illustrations and excellent photographs of the equipments used. Literature summerization at the end of each chapter is very useful. Although containing the contributions of 22 authors the book is well organized and uniform in the presentation of information. It serves as a good reference book for researchers in harvesting and handling, food Scientists, economists and students in Agricultural engineering.

SHANTHA KRISHNAMURTHY
INDIAN INSTITUTE OF HORTICULTURAL RESEARCH, BANGALORE

ASSOCIATION NEWS

Poona Chapter

The Annual General Body Meeting was held on 5th January 1984 and the following Office-bearers were elected:

Presidrnt:—Dr. W. B. Date,
Vice-President:—Dr(Mrs) P. P. Kanekar,
Secretary:—Mrs. N. R. Joshi,
Treasurer:—Dr. S. B. Bhosale.

ASSOCIATION OF FOOD SCIENTISTS AND TECHNOLOGISTS (INDIA)

CFTRI CAMPUS, MYSORE—570 013.

FOURTH INDIAN CONVENTION OF FOOD SCIENTISTS AND TECHNOLOGISTS

The Fourth Indian Convention of Food Scientists and Technologists will be held between 7-9 June 1984 at CFTRI, Mysore with a focal theme on "Food Additives in Processed Foods"

A wide variety of food additives are being used in processed foods in recent times. There is not enough recorded information in the country on the different types of additives, their levels of use function and safety regulations. There is lack of information with regard to identity, purity and toxicological evaluation of some of the additives used in processed foods. The main objective of the Convention is to bring out an overview on the present status and focus the future of these additives in food industries as aids in processing of foods.

Individual food additives such as colours, flavours, preservatives, antioxidants, emulsifiers, stabilizers, non-conventional sweetners, extraction solvents, miscellaneous food additives and thickening agents will be discussed in detail along with regulatory aspects involved in the use of food additives. Recommendations emerging out of these will be useful to food technologists and regulatory agencies and officials in the country.

Special lectures on the emerging areas of topical interest such as 'Use of Plant Tissue Culture in Augmenting Food Resources' will be arranged. Poster Sessions on various aspects of Food Science and Technology will be held to make the Convention more comprehensive.

These deliberations will be held in the form of Symposia and Poster Sessions. Invited Speakers from Industries, Universities, Government and Private Institutions as well as Consumer protective agencies are expected to participate in the Convention.

AFST(I) requests your kind participation in this Convention which is of vital importance to the country.

O. P. Kapur
President
AFST(I)

S. C. Basappa
Hony. Exec. Secretary
AFST(I)

PUBLICATIONS

OF

THE ASSOCIATION OF FOOD SCIENTISTS & TECHNOLOGISTS (I)

(CENTRAL FOOD TECHNOLOGICAL RESEARCH INSTITUTE CAMPUS, MYSORE)

1. **REVIEWS IN FOOD SCIENCE AND TECHNOLOGY—VOL. IV**
   Royal 8vo, hard bound, P. 255.
   Price: India **Rs. 8/-**
   Abroad: **$5 Surface mail**
   $8 Air mail.

2. **PROCEEDINGS OF THE SYMPOSIUM ON FATS AND OILS IN RELATION TO FOOD PRODUCTS AND THEIR PREPARATIONS**
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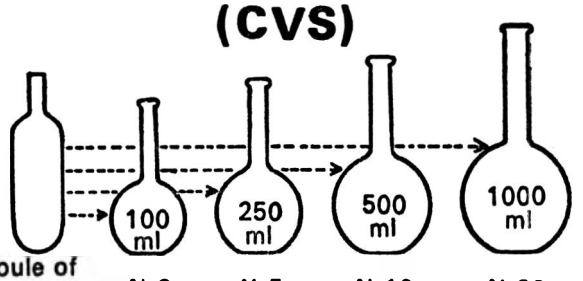

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2. Short communications in the nature of Research Notes should clearly indicate the scope of the investigation and the salient features of the results.

3. Names of chemical compounds and not their formulae should be used in the text. Superscripts and subscripts should be legibly and carefully placed. Foot notes especially for text should be avoided as far as possible.

4. **Abstract:** The abstract should indicate the principal findings of the paper. It should be about 200 words. It should be in such a form that abstracting periodicals can readily use it.

5. **Tables:** Tables as well as graphs, both representing the same set of data, should be avoided. Tables and figures should be numbered consecutively in Arabic numerals and should have brief titles. They should be typed on separate paper. Nil results should be indicated and distinguished clearly from absence of data, which is indicated by '—' sign. Tables should not have more than nine columns.

6. **Illustrations:** Graphs and other line drawings should be drawn in *Indian ink* on tracing paper or white drawing paper preferably art paper. The lettering should be in double the size of printed letters. For satisfactory reproduction, graphs and line drawings should be at least twice the printed size 16cms (ox axis) × 20cms (oy axis); photographs must be on glossy paper and must have good contrast; *three copies* should be sent.

7. Abbreviations of the titles of all scientific periodicals should strictly conform to those cited in the World List of Scientific Periodicals, Butterworths Scientific Publication, London, 1962.

8. **References:** Names of all the authors along with title of the paper should be cited completely in each reference. Abbreviations such as *et al.*, *ibid. idem*, should be avoided.

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   (*a*) *Research Paper:* Jadhav, S. S. and Kulkarni, P. R., Presser amines in foods, *J. Fd Sci. Technol.*, 1981, **18**, 156.

   (*b*) *Book:* Venkataraman, K., The Chemistry of Synthetic Dyes, Academic Press, Inc., New York, 1952, Vol. II, 966.

   (*c*) *References to article in a book:* Joshi, S. V., in the Chemistry of Synthetic Dyes, by Venkataraman, K., Academic Press, Inc., New York, 1952, Vol. II, 966.

   (*d*) *Proceedings, Conferences and Symposia Papers:* Nambudiri, E. S. and Lewis, Y. S., Cocoa in confectionery, *Proceedings of the Symposium on the Status and Prospects of the Confectionery Industry in India*, Mysore, May 1979, 27.

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

   (*f*) *Unpublished Work:* Rao, G., unpublished, Central Food Technological Research Institute Mysore, India.

9. Consult the latest copy of the *Journal* for guidance.

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Research Papers

Research Notes

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