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2. To provide a forum for the exchange, discussion and dissemination of current developments in the field of Food Science, Technology and Engineering.
3. To promote the profession of Food Science, Technology and Engineering.

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Methods for Estimation of Starch: A Critical Appraisal

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Accurate analysis of starch in food and food products is of vital importance due to consequent implications in nutritional and functional aspects. This review analyses available estimation methods, factors affecting their reliability, merits and limitations. Among different methods, the enzymic method for starch estimation is most accurate. However, the literature survey indicated that the acid hydrolysis method, although less accurate in presence of non-starchy carbohydrates, is being widely used for starch estimation in various food products and complex food materials.

Keywords : Starch estimation methods, Direct methods, Indirect methods, Starch-iodine reaction, Acid hydrolysis, Enzymic hydrolysis, Glucose estimation, Merits, Limitations.

Starch, a polymer of glucose units with α 1-4 and α 1-6 linkages, is the major reserve polysaccharide in plants. It occurs in lower plants such as algae and in various parts of higher plants such as seeds, fruits, stems, leaves, tubers, bulbs and rhizomes in varying amounts. In studies of carbohydrate food reserves in plants, accurate estimation of starch content is of significant importance. Since the amount of stored starch in plants at any specific time is indicative of the physiological activity, it represents a valuable diagnostic tool in plant physiology, which, in turn, is used to determine the potential crop yields in perennial plants (Rasmussen and Henry 1990). The accurate analysis of starch content is also significant in determining the nutritional and functional qualities of food materials.

Although a number of methods are reported in the literature for the estimation of starch in materials such as tubers (Balagopalan et al. 1988), starchy substrates (AOAC 1990), cereals (AOAC 1990; Batey 1982), legumes (Dekker and Richards 1971), vegetables (McCready et al. 1950), plant tissues (Pucher et al. 1948; Carpita and Kanabus 1987), conifers (Ebell 1969) and fruits (Rasmussen and Henry 1990), each method has its own merits and demerits. The accurate determination of starch has hence posed several problems to analytical chemists. Although there are many comprehensive reviews on starch estimation methods (Hadorn and Doevelaar 1960; Southgate 1976; Mitchell 1990), there is hardly any review, wherein the advantages and disadvantages of each method are discussed. An attempt has been made here to evaluate various methods which are available for starch estimation in food and food products.

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Methods for starch estimation

These can be broadly classified into two categories, viz., direct or non-hydrolytic and indirect or hydrolytic methods. In the direct method of starch estimation, starch is first separated from other interfering materials such as proteins, fats and sugars, by using appropriate solvents, and with minimum starch degradation, based on either the physical or the physico-chemical properties of the polymer. In the indirect methods, the starch is first isolated from the material, hydrolysed into smaller oligosaccharides and the starch value is obtained after estimation of the hydrolysed products. Both these methods are discussed in the following sections :

Direct methods for starch analysis

These do not involve the need for starch hydrolysis. Instead, starch is analysed directly for quantification.

Gravimetric method : In this method, starch is first dispersed in a suitable solvent, precipitated with alcohol, filtered to recover the precipitate and quantitated gravimetrically by weighing the dried starch. Treatments with caustic soda, trichloroacetic acid, salicylic acid or lactic acid have been used for starch dispersion (Radley 1953). Gravimetric method of Rask (1927) for starch analysis in cotton seed meal, wheat flour, corn and potato was adopted by the Association of Official Agricultural Chemists (AOAC 1931). In this method, starch is dispersed in cold concentrated hydrochloric acid to form a clear and opalescent solution. The above solution is then freed of the acid after several

washings with increasing concentrations of ethanol and to form a precipitate which is collected and weighed. Balagopalan et al. (1988) proposed a gravimetric method for starch estimation in cassava tubers, based on dry matter content. Even though these methods are simple, these are not very accurate and hence are not used extensively for quantitative estimation of starch.

Optical method based on starch-iodine reaction : The starch-iodine reaction was first discovered by Colin and de Claubry in 1814 (Hollo and Szeitli 1968) and is based on the estimation of the blue-coloured complex formed by the reaction of amylose and iodine. The formation of the additive products between starch and iodine was used by Kaiser to estimate starch (Sullivan 1935). Iodine in potassium iodide containing aqueous solutions exhibits a very broad peak of absorption at 450 nm. When the starch-iodine complex is formed, a shift in the peak of absorption to 600-620 nm occurs. The values of the peaks, i.e., the absorption coefficients, depend on temperature and degree of polymerization of the starch molecule. Paloheimo and Paloheimo (1931) developed a iodocolorimetric method for estimation of starch in food products using the above principle. Recently, Magel (1991) suggested a qualitative and a quantitative method for starch estimation based on this principle. This procedure involves extraction of starch with HCL, complexing it with potassium iodide-iodine solution and measurement of absorbance at 605 and 530 nm, which corresponds to the absorption maxima of amylose and amylopectin, respectively. The starch value is calculated using the formula :

$$C_s = \frac{X_{(605\ nm)}}{\frac{A_{(a605\ nm)} - A_{(ap605\ nm)}}{100} \times P + A_{(ap605\ nm)}}$$

Where

C_s = starch concentraction in (µg/ml) of unknown solution

$X_{(605\ nm)}$ = absorption in OD units x cm^{-1} of unknown starch solution at 605 nm

$A_{(a605\ nm)}$ = absorption coefficient in OD units x ml x μg^{-1} x cm^{-1} for amylose at 605 nm

$A_{(ap605\ nm)}$ = absorption coefficient in OD units x ml x μg^{-1} x cm^{-1} for amylopection at 605 nm

P = amylose fraction in % of unknown starch solution, calculated according to the formula given below :

$$R = \frac{P \times C \times A_{(a605\ nm)} + (1-p) \times C \times A_{(ap605\ nm)}}{P \times C \times A_{(a530\ nm)} + (1-P) \times C \times A_{(ap530\ nm)}}$$

C = starch concentration in µg/ml; P = amylose %; a = amylose; ap = amylopectin; $A_{(605\ nm)}$ = 17.38; $A_{(a530\ nm)}$ = 11.44; $A_{(ap605\ nm)}$ = 6.76; $A_{(ap530\ nm)}$ = 8.98; R is the ratio of the absorbancies at 605 and 530 nm.

Kerr (1950) reported a iodine blue value method for starch determination which is a combination of the methods of Clenndenning (1942) and Kerr and Trubell (1943). It involves treatment of the ground starch sample with stannic chloride, before treating the aliquot of the solution with iodine-potassium iodide solution. The blue colour obtained is read at 600-620 nm. The unknown reading is extrapolated from the standard curve of starch in calcium chloride-stannic chloride with iodine. One of the assumptions of the method is that the starch does not undergo any modification during the whole process. Some of the advantages of the method include high sensitivity of the reaction of starch with iodine, non-hindrance of calcium chloride with starch-iodine complex formation and minimum degradation as well as easy solubility of starch in calcium chloride (Kerr 1950). The starch-iodine procedures for starch estimation have also been employed by Hadorn and Doevelaar (1960) and Kolbach and Rinke (1962), Hassid and Neufeld (1964), Blakelney and Matheson (1984). A number of similar methods, applied to cereal products, have since been reported.

The disadvantages of the above method are many. It is not applicable to materials containing dextrins, since there is significant contribution of the blue colour by dextrins. The particle sizes of the starch molecules influence the intensity of colour of starch-iodine complex (Paloheimo and Antilla 1931). The iodine colour of some starches and starch fractions changes with the concentration of starch and the amout of iodine added. Therefore, many of the starch-iodine complex colours recorded in the literature may not be suitable for comparative purposes (Watson and Whistler 1966). Sullivan (1935) suggested that this method is not suitable for estimation of starch in woody plants, since it resulted in problems during precipitation of the starch-iodine complexes.

Polarimetric methods : It is one of the most popular methods for starch determination, wherein the starch is dispersed in a suitable solvent and the starch content is quantitated from the clarified

solution by measuring the angle of rotation of the plane polarised light. Different types of solvents like benzoic acid, caustic potash solution, concentrated calcium chloride solution (Mannich and Lenz 1920; Radley 1953) have been used for this purpose. The most extensively used solvent for starch dipersion is a concentrated solution of calcium chloride. Fellenberg was the first to demonstrate the method of quantitative estimation of starch using this solvent (Radley 1953). Since then, a number of modifications were made by many workers (Mannich and Lenz 1920; Hopkins 1934; Clenndenning 1945; Kerr 1950). The modified method developed by Analytical Working Party of the Starch Experts Group of the European Starch Association (ESA 1987) is based on differential measurement of the optical rotation of the treated sample and an ethanol-extracted blank. This method involves solubilisation of starch in calcium chloride solution (33%) at neutral or slightly acidic pH values. As a result of high ionic strength, the starch molecule unfolds without the formation of oligosaccharides or glucose. The starch content is given by the difference between the two measurements, multiplied by a known factor $[\alpha]_D^E$ which is dependent on the nature of the starch. The $[\alpha]_D^E$ values for different types of starches, as reported by Kennedy et al. (1989) are given in Table 1.

TABLE 1. $[\alpha]_D^E$ VALUES FOR DIFFERENT STARCH TYPES*

Starch type	$[\alpha]_D^E$
Potato	185.7
Wheat	182.7
Rye	184.0
Barley	181.5
Oat	181.3
Rice	185.9
Corn	184.5
Other starches	184.0

*Data from Kennedy et al (1989)

The above method gives better reproducible results and is fast as well as simple to perform (Mitchel 1990). The whole process of estimation takes less than 2 h (Earle and Miller 1924). The only disadvantage of this mthod is the interference caused by hemicelluloses and other non-starchy polysaccharides, which affect the polarimetric measurements (Clenndenning 1948).

Indirect methods for starch estimation

These methods involve the analysis of the hydrolysed products of starch, obtained by different hydrolysis methods, namely, acid hydrolysis and enzymatic hydrolysis.

Acid hydrolysis method of Ewers : A polarimetric method involving acid hydrolysis of the starch sample and measurement of the optical rotation of the plane polarised light by the resultant reducing sugars was proposed by Ewers in 1908 (Kennedy et al. 1989). It is an official Enzyme Commission (EC) method, which is being used extensively for starch estimation and measurement of starch purity till today (Mitchell 1990). The method involves treatment of starch with glacial acetic acid, HCl and hot water. Clarification of the resultant solution is carried out by potassium ferrocyanide and the optical rotation of the treated sample is measured. Later, Ewers used 1.12% HCl for initial starch treatment and clarified the solution with sodium molybdate and phosphotungstic acid (Radley 1953). Subsequently, the above method has undergone several modifications, but the principle remained the same. The suitability of different clearing agents was examined by Dudas (1970), which revealed that phosphotungstic acid had lowest suitability, whereas ammonium and sodium acetate lowered the optical activity of the resulting solution. Moreover, the increase in the dissolution time of the sample with HCl for more than 15 min was found to result in lower values of the starch, since it increased starch solubility and consequently the difference in the optical measurements between the whole and the ethanol soluble fractions (Dudas 1971). Stirring of the samples during dissolution was also found to result in high starch values. However, the magnitude of this effect on starch value was lower as compared to that by alteration of the dissolution time (Dudas 1971). Kennedy et al (1989) reported that the concentration of HCl for starch dissolution is highly critical as higher concentration resulted in lower values of starch due to greater hydrolysis. Use of potassium ferrocyanide as clarifying reagent was found to be the best among all other reagents evaluated (Dudas 1970, 1972).

The method presently followed is carried out in two steps. In the first step, a suitable quantity of starch is heated with 0.3044 M HCl for 15 min at 100°C to gelatinize and hydrolyse the starch to smaller oligosaccharides. The hydrolysis is terminated by rapid cooling. The above solution is

clarified by potassium ferrocyanide (Carrez reagent), filtered and the angle of rotation of the filtrate (α^T) is measured.

In the second step, the sample is extracted with 40% ethanol and filtered after sufficient mixing. The filtrate obtained is acidified with HCl, treated with the clarifying reagent, followed by zinc sulphate treatment. After filtration, the angle of rotation of the plane polarised light of the alcoholic filtrate (α^S) is measured (Kennedy et al 1989; ESA 1987). The starch content is given by the difference between the two readings, multiplied by the factor $(\alpha)^E_D$, the value of which depends on starch type. The starch content (%) is given by the equation:

$$\% \text{ starch} = \frac{10{,}000\ (\alpha^T - \alpha^S)}{(\alpha)^E_D \times L \times c}$$

Where L is the path length, c = concentration of the sample in g/dl, while α^T and α^S are the angles of rotation of the plane polarised light by the whole samples and the alcoholic extracts, respectively.

Kennedy et al (1989) critically evaluated the above method for starch estimation and pointed out its demerits. They reported that the starch values obtained by the above method are different for a series of starches with different ratio of amylose and amylopectin. Higher values of starch are observed in samples containing high amylopectin. Hence, it was proposed that this method is not applicable to samples with varying amylose and amylopectin contents. Moreover, this method is not applicable to high amylose starches, due to their high gelation temperature (Mitchell 1990). Factors that affect the degree of solubilization of starch, before polarimetric measurements, such as HCl concentration, heating and cooling time are highly critical. Hydrolysis of starch by Ewers' method is known to depend on several factors which are critical in starch estimation. These involve the thickness of the flask, extent of agitation of the contents, thermal capacity of the water bath which, in turn, affect the heat transfer characteristics of the system (Kennedy et al. 1989). The clarifying reagent of Carrez, which is used to remove chiral molecules, was also found to adsorb malto-oligosaccharides, thereby leading to lower values of starch.

For Ewers' method of starch determination, a minimum purity of 97% starch on dry basis is required (Mitchell 1990). Hence, this method does not hold good for the starchy materials containing interfering substances. Sample containing high ash content is also known to result in incorrect values of starch (Mitchell 1990). In case of starch estimation in complex plant materials, the degradation of the polysaccharides interfere or rather modify the optical rotation value, thereby resulting in incorrect starch value (Thivend et al 1972; Libby 1970). Saunders et al. (1970) also observed that the values of starch are different for high fibre materials with the use of polarimetric and colorimetric methods.

Direct acid hydrolysis method of AOAC : This is one of the methods for starch estimation based on the hydrolysis of the starch into glucose. AOAC recommended this method for starch estimation in products like potatoes, raw starch, grains, wheat flour, confectionery and animal feeds (AOAC 1960). In this method, a known quantity of starch/starch material is hydrolysed with concentrated hydrochloric acid by refluxing at 100°C for 150 min. After neutralisation, the glucose obtained is estimated by any of the standard methods. The starch value is obtained by mulitplying the glucose value with a factor of 0.9 (AOAC 1960). This method, however, was discontinued for starch estimation in cereals from 1970 onwards (AOAC 1970), but is still recommended for starch estimation in confectionery products, animal feeds, roasted coffee, cocoa products, face powders, desserts and baking powders (AOAC 1990).

Pirt and Whelan (1951) performed experiments to replace HCl with other acids such as nitric, sulphuric and phosphoric acids. Their investigations indicated that the use of 1.5 N sulphuric acid leads to minium degradation of glucose, whereas HCl results in significant loss of glucose due to caramelization in the estimation of amylose and amylopectin of potato starch. MacRae and Armstrong (1968) also used this method for the estimation of starches in biological materials. This method is still being employed for starch estimation in various starchy substrates such as cassava (Rickard and Behn 1987; Padmanabhan and Lonsane 1992; Padmanabhan et al. 1992, 1993), composite foodstuffs (Boley and Burn 1990) and potato starches (Faithfull 1990). Rickard and Behn (1987) pointed out that there is no significant loss of glucose, when the above method is employed for starch estimation in cassava starch. Faithfull (1990) suggested that the acid hydrolysis method of AOAC is reliable, rapid, precise and is sufficiently accurate when employed for potato starch estimation. Recently, its erroneous nature has been established

beyond any doubt (Padmanabhan and Lonsane 1993).

Enzymatic method : Although the acid hydrolysis method has been widely used since a very long time for starch estimation, it has been reported to have several disadvantages, such as concomitant hydrolysis of non-starchy polysaccharides, which consequently leads to over-estimation of glucose and in turn, starch value (Rickard and Behn 1987; Padmanabhan and Lonsane 1993). Destruction of glucose during acid hydrolysis has also been observed in certain cases due to decomposition and caramelization, thereby resulting in lower values of starch during estimation (Pirt and Whelan 1951; Blake and Coveney 1978; Padmanabhan and Lonsane 1993). To overcome the above mentioned disadvantages, the acid hydrolysis method is being replaced by enzymatic hydrolysis for starch estimation (Thivend et al. 1972; Baur and Alexander 1979; Batey 1982; Karkalas 1985; Ostergard et al. 1988; Padmanabhan and Lonsane 1993). Different enzymes, such as malt and barley diastase, takadiastase, amyloglucosidase, thermostable alpha-amylase (Radley 1953; AACC 1962; Libby 1970) have been used for efficient hydrolysis of starch.

One of the basic requirements in these cases is the effective and complete degradation of starch molecules into the final product i.e., glucose, which depends on effective or complete solubilization of starch, types of enzymes used for hydrolysis, reaction time, and concentration of the enzymes (Batey 1982; Padmanabhan and Lonsane 1993; Holm et al. 1986). The first step for quantification of starch by enzymic method involves gelatinization/ solubilization of starch. Different methods have been employed for effective solubilization (Table 2). Thivend et al. (1965) gelatinized the starch in an autoclave and then hydrolysed the isolated starch by amyloglucosidase. This work enabled other researchers for a new approach for starch quantification. Dekkar and Richards (1971) solubilised starch in alkali, before its hydrolysis with amyloglucosidase and suggested that the solubilization of starch is incomplete, if alkali is not used for solubilization. However, solubilization of starch in alkali has been shown to result in formation of saccharinic acids which are not hydrolysed during starch hydrolysis to glucose (Boley and Burn 1990). Use of dimethly sulphoxide (DMSO) has also been suggested for complete solubilization of starch, prior to enzymatic treatment (Libby 1970). However, due to slow dissolution of canna and potato starches and toxic nature of DMSO, this method of solubilization is not widely practised (Karkalas 1985).

TABLE 2. DIFFERENT METHODS FOR STARCH SOLUBILISATION PRIOR TO ITS HYDROLYSIS.

Method employed	Demerits	Reference
Solubilisation using perchloric acid and estimation of glucose by anthrone method	Interference from hydrolysed products of glucose	Pucher et al (1948), McCready et al (1950) Faithfull (1990)
Solubilisation by autoclaving at 130°C	Incomplete gelatinization	Thivend et al (1965)
Solubilisation by boiling with water	High temperatures induce partial hydrolysis of amylose and amylopectin and degrade other polysaccharides	Dekker and Richards (1971), MacRae and Armstrong (1968), Haissig and Dickson (1979)
Solubilisation using dimethyl sulphoxide	Slow dissolution of potato and canna starches, toxic nature of DMSO	Libby (1970), Carpita and Kanabus (1987)
Solubilisation using acid	Concomitant hydrolysis of other compounds	Beutler (1978)
Solubilisation using alkali	Turbidity in solutions, formation of saccharinic acids which interfere with estimation of glucose	Donovan et al (1977), Boley and Burn (1990)
Solubilisation using alpha-glucosidase	Insolubility of starch in aqueous solutions and incomplete conversion of starch to glucose	Carpita and Kanabus (1987), Hassig and Dickson (1979)
Solubilisation using amylases other than bacterial thermostable enzyme	Hydrolysis of starch is incomplete	Batey (1982)
Solubilisation using thermostable alpha-amylase and amyloglucosidase	No disadvantage, since it ensures complete hydrolysis of starch to glucose	Batey (1982), Karkalas (1985), Padmanabhan and Lonsane (1993)

Batey (1982) employed a thermostable alpha-amylase for starch solubilization. A comparative study on the efficiencies of different methods of solubilization revealed that the solubilization of starch is complete when thermostable alpha-amylase was used as compared to the use of acids and alkalis (Batey 1982). These studies indicated valuable information. The use of a bacterial thermostable alpha-amylase for gelatinization of starch has been shown to prevent formation of retrograded starch, which is not susceptible to amyloglucosidase action. The thermostable alpha-amylase was also found to form a clear and non-viscous solution of gelatinised starch. Complexed amylose, and amylose–lipid molecule complex found in the native starch, are also susceptible to thermostable alpha-amylase attack and, thus, the above complex also undergoes hydrolysis during gelatinization, thereby allowing accurate estimation of starch. Consequently, a number of workers have followed this procedure for starch solubilization and estimation (Karkalas 1985; Holm et al. 1986; Franco et al. 1987; Henry et al. 1990; Padmanabhan and Lonsane 1993).

Thus, the steps involved in the estimation of starch by enzymatic method include a) gelatinization of starch using thermostable alpha-amylase, b) hydrolysis of the liquified starch to glucose by amyloglucosidase and c) estimation of the released glucose molecule, either by titrimetric, colorimetric or enzymatic method (Fig. 1). Multiplication of the glucose concentration by 0.9 gives the starch content in the sample (Padmanabhan and Lonsane 1993).

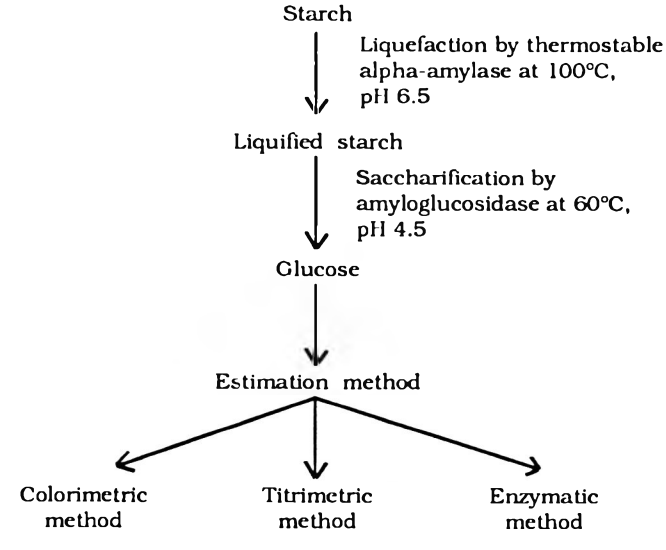

Fig. 1. Scheme for estimation of starch by enzymatic method.

Advantages of the enzymatic method over other methods for starch analysis : The greatest advantage of the enzymic method is its specificity. The use of enzymes for starch estimation leads to hydrolysis of the starch molecules only and there is absolutely no contribution of the reducing sugars from the hydrolysis of the non-starchy polysaccharides (Mitchell 1990; Padmanabhan and Lonsane 1993). Consequently, the method is most suitable for high-fibre products (Saunders et al. 1970). The higher temperature of incubation of starch and alpha-amylase also ensures rapid inactivation of the non-starch degrading enzymes, thereby increasing the specificity of the method for starch estimation (Rasmussen and Henry 1990). Recent work of Padmanabhan and Lonsane (1993) has revealed that the contribution of glucose from the hydrolysis of the non-starchy polysaccharides can be totally eliminated, if the enzymatic method is employed for starch estimation. The enzymatic method for starch estimation was found to give accurate estimations of starch in whole flours, cereals, grains, non-starchy polysaccharides containing starchy materials and impure as well as pure starches.

Factors affecting the reliability of the enzymic method : Few alpha-amylases and amyloglucosidase preparations are known to contain free glucose and starch (Padmanabhan and Lonsane 1993), which have been added as preservatives/stabilizing agents. Use of such enzyme preparations leads to incorrect values of glucose and consequently of the starch. Many commercial amyloglucosidases are also known to contain transglucosidase activity (Padmanabhan and Lonsane 1993). Since transglucosidase adds glucose to maltose, it can result in reduction in the glucose value estimated (Xavier et al. 1993). It is, therefore, of critical importance to use the enzyme preparations free from transglucosidase activity (Padmanabhan and Lonsane 1993). Englyst et al. (1992) reported the presence of hemicellulase and β-glucanase activity in some commercial preparations of amyloglucosidase. Use of such enzyme preparations results in contribution of glucose through concomitant hydrolysis of hemicellulose and β-glucan during starch hydrolysis. Harris et al (1984) observed that some amyloglucosidase preparations degraded guar galactomannan and locust bean and consequently, interfere with accurate estimation of glucose. Karkalas (1985) found the presence of high amount of sucrose in glucose-oxidase-peroxidase (GOD) reagent, which is used for glucose estimation. Ultimately, it affects the estimation of glucose.

In spite of the demerits of the acid hydrolysis method, it is still recommended by AOAC (1990) for estimation of starch in confectionery, roasted coffee, animal feed, dessert and baking powders and other complex food materials. Erroneous estimation of starch in these products by AOAC (1990) recommended acid hydrolysis method has been established recently (Padmanabhan and Lonsane 1993). A need exists to rectify the lacuna.

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Alpha-Amylase Inhibitor Activity in Sorghum Grains: Effects of Cooking and UV Radiation

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Sorghum (*Sorghum bicolor* M) seeds were analysed for amylase inhibitory activity and the effects of cooking on them were studied. Cooking and UV radiation decreased the amylase inhibitory activity. The loss of amylase inhibitory activity in pre-soaked sorghum seeds after these heat treatments was more drastic than in the heat-treated raw seeds. Amylase inhibitory activities in some commonly consumed sorghum products were tested. Popped sorghum and *idli* did not exhibit any amylase inhibitory activity, though it was present in the grains used for making these products.

Keywords : Alpha-amylase inhibitor, Sorghum, Cooking, UV radiation, Sorghum products.

Sorghum grain is the staple food of the people of the arid and semi-arid regions of the world. Many recipes are prepared from sorghum to suit wide diversity of food habits (Jadhav and Joglekar 1984). The most common preparation from sorghum is unleavened bread, dumpling and boiled rice-like products (Salunkhe et al. 1985). Also, *Dosa*, *roti* and vermicelli products are developed from sorghum (Raghavendra Rao et al. 1979). Sorghum grain has been shown to contain many antinutritional factors, and alpha-amylase inhibitor is one among them (Salunkhe et al. 1985). The results on the effect of cooking sorghum on alpha-amylase inhibitory activity, and also the levels of alpha-amylase inhibitory activity in different sorghum products are presented in this paper.

Materials and Methods

Sorghum grains were procured from Agricultural Research Station, Gulbarga, cleaned and stored till use. Human salivary amylase was purchased from Sigma (USA), while all the other chemicals were of analytical grade (Loba Chemicals, Bombay). A number of sorghum products were prepared. *Roti* (unleavened soft-thin, flat, circular shaped bread) was prepared from sorghum flour and baked on a concave pan. *Kichidi* was prepared from soaked sorghum grains, which were pressure-cooked, salted and seasoned with vegetable oil. *Idli* was prepared by mixing sorghum grit, *urad dhal* and rice in the ratio of 1:1:0.4, allowing the mixture to ferment and pressure-cooking the dough. *Thalipeeth* was prepared from sorghum flour mixed with wheat, roasted rice, pearl millet and Bengalgram flour in the ratio of 4:1: 0.5:0.5:0.2. The dough was made by mixing the flour with salt, turmeric and water. The dough was worked out into circles of 10-12 cm dia. and cooked on an oil-smeared pan by punching holes to allow self-steaming. *Sandige* was prepared out of cooked sorghum flour and passed through a press of required shape and dried. When fried in vegetable oil, it is eaten as a crisp snack. Popped sorghum was prepared by roasting sorghum grains in a very hot concave pan at a very high temperature. The popped sorghum then was salted, eaten as snack and tested for amylase inhibitory activity. *Sandige*, not fried in vegetable oil, served as raw *sandige*.

Sorghum meal was defatted before the extraction of the amylase inhibitor in the following way. Sorghum grains (10 g) were soaked overnight in 100 ml distilled water and then the soaked seeds were homogenized with 30 ml acetone for 30 min. It was air-dried rapidly under suction, and this dry meal was used as defatted meal. Amylase inhibitor extract was prepared by homogenizing 2 g defatted sorghum meal or 2 g freeze-dried samples (in case of sorghum products) with 20 ml of 0.2 M phosphate buffer (pH 6.9), containing 0.3 M NaCl. The mixture was stirred for 4 h at 0°C. The homogenate was centrifuged at 10,000 **g** for 20 min at 4°C, and the clear supernatant was dialysed against 0.01 M phosphate buffer (pH 6.9) for 8 h, before using for the assay of amylase inhibitory activity by the method of Bernfeld (1955). The method involved the incubation of the enzyme with an aliquot of inhibitor extract for 30 min before adding 1% starch. After incubation at 37°C for 5 min, the reaction was stopped by adding 1 ml of 3,5-dinitrosalicylic acid reagent, and the amount of reducing sugars liberated was estimated. One unit of inhibitory activity was defined as the amount of inhibition that suppressed one unit of enzyme activity under assay conditions.

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Raw sorghum grains and overnight soaked grains were cooked in an open pan containing water for various time intervals. Water was decanted and the grains were dried. The inhibitor was extracted by the above method. Similarly, the raw seeds, soaked seeds and defatted meal were exposed to UV radiation for different time intervals, and then amylase inhibitory activity was assayed. Amylase inhibitory activity was also determined in sorghum flour obtained by grinding the grains in manual *chakki* and an electric flour mill. The mechanical *chakki* is used by the rural people, and it consists of two circular stones which can be rotated. The grain, when passed in between the stones, gets converted into fine powder by the crushing action.

Results and Discussion

The effect of cooking sorghum grains in open pan on amylase inhibitory activity is presented in Table 1. It is observed that amylase inhibitory activity is lost on cooking sorghum grains in open pan. Amylase inhibitory activity decreased with the increase in cooking time. The loss in inhibitory activity was more drastic in soaked sorghum grains than in raw seeds. Similar results are reported for legume seeds (Shekib et al. 1988), and sorghum seeds (Obizoba and Atti 1991). These workers have observed that amylase inhibitor was heat labile, and the soaking of the seeds was beneficial for improving the nutritional quality. Similar loss in trypsin inhibitory activity on cooking dry cow peas has also been reported (Bakr and Gawish 1991).

TABLE 1. EFFECT OF COOKING SORGHUM GRAIN IN AN OPEN PAN ON AMYLASE INHIBITORY ACTIVITY

Cooking time, min	Amylase inhibitory activity, units/g	
	Raw grain	Overnight soaked grain
0	113.8	110.0
5	55.4	46.7
10	22.6	15.6
15	15.6	8.5
20	8.6	0.7
25	0.8	N.D.

Each value is an average of triplicate determination.

N.D. = Not detected

The effect of UV radiation on amylase inhibitory activity in sorghum is given in Table 2. Among the defatted sorghum meal, raw grains and overnight soaked sorghum grains, the latter showed maximum loss of amylase inhibitory activity on exposure to UV radiation for 60 min. Such a loss in amylase inhibitory activity has not yet been reported, though the loss in trypsin inhibitory activity on exposure to infra-red radiations has been reported by Kadam et al. (1987). It has been reported that the infra-red radiation treatment to winged seeds for 60 sec was effective in destroying most of the trypsin inhibitory activity. It is interesting to state that the UV radiation, which is employed as a method for preventing microbial deterioration of cereals, can be used to destroy amylase inhibitory activity. It may be possible that UV radiation affects the enzymic proteins present in sorghum grains.

TABLE 2. EFFECT OF UV RADIATION ON SORGHUM AMYLASE INHIBITORY ACTIVITY

Exposure time, min	Amylase inhibitory activity,units/g		
	Defatted meal	Raw grain	Soaked grain
0	109.2	112.0	110.5
15	98.5	102.2	100.1
30	46.0	84.5	78.0
45	40.0	68.4	40.0
60	10.7	40.0	10.5
90	N.D	10.8	N.D

Each value is an average of triplicate determination.

N.D. = Not detected

Amylase inhibitory activities in sorghum poducts are presented in Table 3. It is observed that amylase inhibitory activities were reduced in sorghum products, when they were heat-processed. Out of the seven products, *roti*, *sandige* (deep-fried) and *kichidi* had low amylase inhibitory activities. Two products, *idli* and popped sorghum did not exhibit any amylase inhibitory activities. *Thalipeeth* had the maximum inhibitory activity and this may

TABLE 3. AMYLASE INHIBITORY ACTIVITIES IN DIFFERENT SORGHUM PRODUCTS

Sorghum products	Amylase inhibitory activity, units/g	Loss in amylase inhibitory activities, %
Roti	20.6	78.4
Thalipeeth	78.0	29.4
Idli	0	100.0
Sandige (raw)	60.0	45.7
Sandige (fried)	12.6	88.7
Popped sorghum	0	100.0
Kichidi	22.8	79.4

Each value is an average of triplicate determination

be due to the presence of other constituents like rice, wheat, pearl millet and Bengalgram. Complete losses in amylase inhibitory activities in *idli* and popped sorghum are probably due to the fact that they are subjected to steaming in the former, and high temperature in the latter cases. Similar loss in amylase inhibitory activity in *idli* has been reported by Udupa et al. (1989). They have also found significant amylase inhibitory activities in fried foods like potato chips and peanuts. In terms of loss in inhibitory activity, *thalipeeth* and *kichidi* lost 29 and 79% activities, respectively. Similar losses in trypsin and chymotrypsin inhibitory activities in cooked products of sorghum have also been reported by Mulimani and Vadiraj (1991).

Amylase inhibitory activities in sorghum meal obtained by different types of milling are shown in Table 4. It is observed that there are lower amylase inhibitory activities in sorghum grains ground in an electric mill than the ones ground in a hand-operated *chakki*. Similar loss in amylase inhibitory activity in rice flour after roller milling has been reported by Snow and Kerin (1981). This loss in amylase inhibitory activity in sorghum grains ground in electric mill is because of the heat generated, while the grain is crushed between the stones.

TABLE 4. AMYLASE INHIBITORY ACTIVITIES IN SORGHUM FLOUR GROUND IN DIFFERENT GRINDERS

Sorghum variety	Amylase inhibitory activity, units/g	
	Chakki	Electric flour mill
'Nate Maldandi'	32.6	15.6
'5-4-1'	40.6	31.1
'Bili jola' (local variety)	54.3	15.6

Each value is an average of triplicate determination

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Changes in Free Fatty Acids and Insect Infestation During Storage of Brown Rice Obtained by Shelling Paddy in Rubber Roll and Disc Shellers

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Development of free fatty acids (FFA) in brown rice, obtained by shelling in rubber roll sheller and disc sheller, was monitored at various conditions of storage, viz., cold (4-6°C), room temperature (RT), 37°C- 62% relative humidity (RH), and at 37°C - 92% RH, with and without insects, upto 13 months. The samples were stored in cloth bags and bottles. FFA, which were initially less than 3%, increased with period of storage; the increase was almost double in disc-shelled rice compared to that in rubber roll shelled rice. High temperatures and relative humidity of the environment increased FFA. Increase in FFA was slightly more when stored in cloth bags than in glass bottles. Insects did not survive at 37°C-62% RH, but multiplied rapidly at RT. Accompanied by increase in FFA, the insects spoiled the rice in 3 months.

Keywords : Brown rice, Storage, Free fatty acids, Infestation, Bran, Brokens, Effect of shelling method.

Storing brown rice (after removing husk from paddy) offers considerable advantages in terms of handling lesser quantity and requirement of lesser space, as the husk contributes to about one-fourth of the weight and over one-third the volume of paddy (Houston 1972). However, the quality of brown or husked rice deteriorates during storage, mainly because of oxidative changes (Sowbhagya and Bhattacharya 1976) and lipolytic hydrolysis of about 3% oil present in it (Hunter et al. 1951). It has also been well established that during storage, there is a slight change in fatty acid levels, (Ramaratnam and Kulkarni 1983).

Several investigators have studied the development of free fatty acids in grains during storage and measurement of fat acidity has been recommended as a criterion of deterioration of the grains (Zeleny and Coleman 1938; Zeleny 1948; Yasumatsu and Moritaka 1964; Barber 1972). Hunter et al (1951) found that the free fatty acids in commercially dehulled 'Caloro' brown rice increased with increase in moisture levels of rice and temperatures of storage during a limited period of 22 weeks. However, the data for different milling processes are scarce. Although the effect of insect infestation on husked, hand-pounded and milled raw and parboiled rice has been studied in a limited context (Pingle et al. 1957), the free fatty acid development during storage of brown rice received little attention. To comprehend such changes, the present study was undertaken and the results are reported in this paper.

Materials and Methods

Materials : 'IR20' paddy, roughly 1 month after harvest, was divided into two portions and dehusked using rubber roll sheller and disc sheller, respectively, in a local commercial rice mill. The brokens in brown rice were separated using a trio cylinder. Head (whole) rice and brokens were fumigated in close drum for 6 days by using 1 aluminium phosphide tablet/300 kg brown rice (1 tablet contains 1 g phosphine). After fumigation, the drum was left open for 2 days, before using the fumigated grains in further experiments.

Storage conditions : Clean, wide mouth glass bottles (160 numbers), wrapped in black paper, were filled up to the neck with brown rice (about 450 g of rubber roll and disc - shelled rice in 80 bottles of each set). Into half of each set of bottles were introduced 20 insects (*Sitophilus oryzae*, 10 days old). The bottles were covered by cloth (*kora*) and tied over the mouth. These bottles were stored at (a) room temperature (23-34°C) and (b) 37°C - 62% RH (Cairns 1974). Another set was stored (without insects) at cold (4-6°C) and 37°C - 92% RH.

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In another trial, 10% and 20% brokens were added to brown rice and stored at RT and 37°C, (2 kg) without any RH control. Rubber roll shelled brown rice (2 kg) was also stored in cloth bags at cold, RT, 37°C-62% RH, and 37°C-92% RH. Brown rice was milled to 8% degree of polish and stored at RT in bottles, as control.

Methods : Samples were drawn in triplicate at regular intervals. Moisture was determined by drying 10 g sample in an oven at 105°C for 24 h and adding a correction factor of 0.5% to the values (Bhattacharya and Indudhara Swamy, unpublished). A portion of brown rice was ground in Raymond hammer mill to pass through 60 mesh for estimation of FFA and fat (AOCS 1973). The other portion of brown rice was milled in McGill laboratory miller to 8% degree of polish and 10 g bran sample was used for fat extraction (AOCS 1973) and FFA determination. Selected samples of milled rice, of which the corresponding brown rice samples (stored without insects for 13 months) showed very high FFA values, were ground and fat extracted by petroleum ether in Soxhlet for 24 h for estimating FFA.

Adult insects in 400 g material were counted using ethyl ether as anaesthetic and a 10 mesh standard (BSS) sieve. Kernel damage was determined by counting kernels having exit holes per hundred kernels. Frass % was estimated by sieving the rice on a 16 mesh (BSS) sieve, Mould count was carried out by the serial dilution method using Czepak's agar medium incubated at 30°C for 6 days. Uric acid content was also determined (Venkat Rao et al. 1959). The data were statistically tested by modified Duncan's new multiple range test (Harter 1960).

Results and Discussion

Changes in free fatty acids : The FFA, which initially was 3%, increased with increase in temperature and RH of the environment (Figs. 1 and 2). The decline phase in the curves of 37°C-92% RH samples may be due to the oxidation of liberated FFA; FFA being more prone to oxidation than corresponding esters (Sowbhagya and Bhattacharya 1976). The mean values of FFA showed significant ($P < 0.05$) differences for periods of storage and different conditions of storage (Table 1). Other workers (Hunter et al. 1951; Narayana Rao et al. 1954) also observed an increase in FFA with increase in temperature of stroage. High humidity of storage environment gave

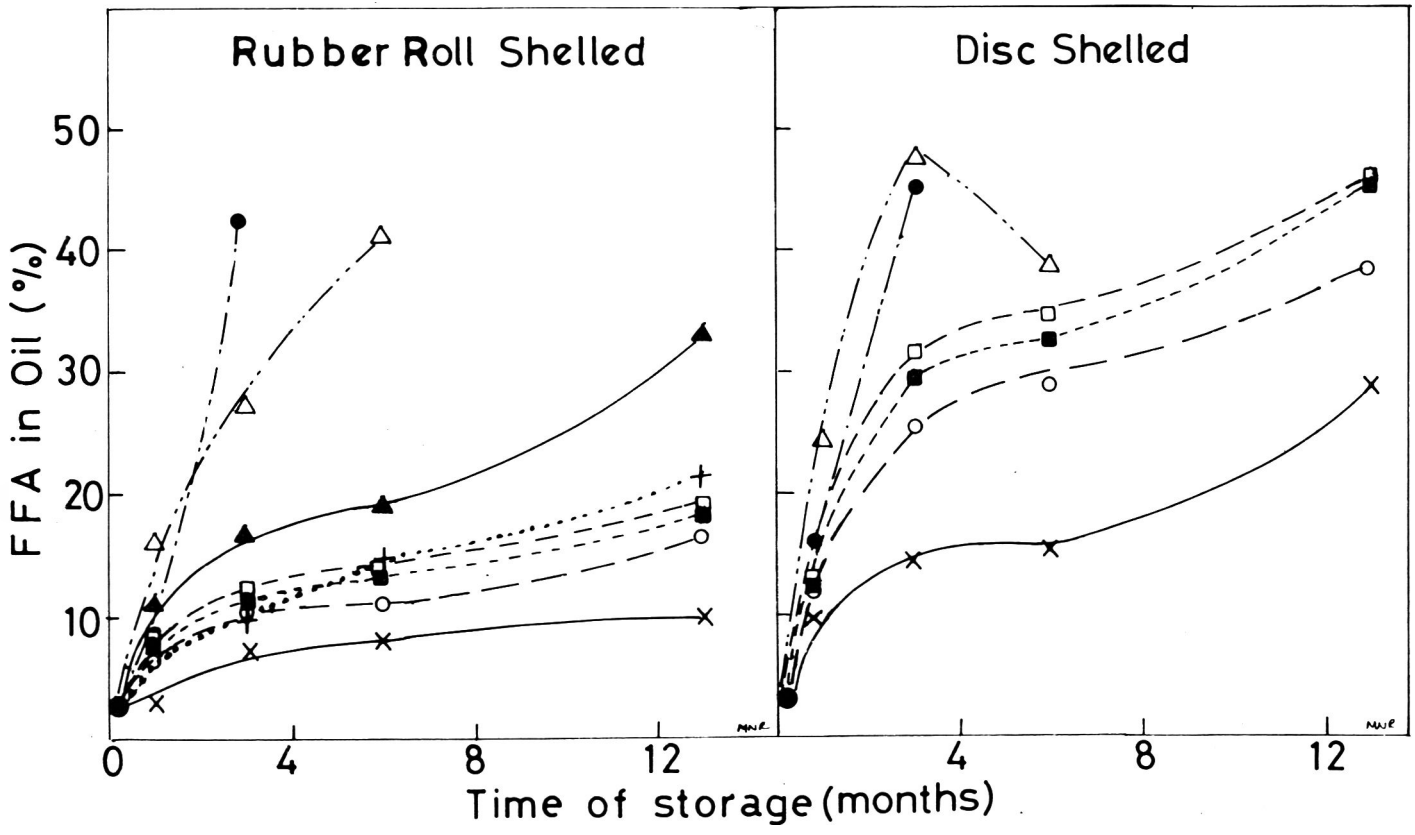

Fig. 1. Changes in free fatty acids in whole brown rice during storage at different conditions.
X : Cold; O : RT; ● : RT with insects; □ : 37°C (62% RH); ■ : 37°C (62% RH) with insects; Δ : 37°C (92% RH); ▲ : RT (milled rice); + : RT with 10% brokens.

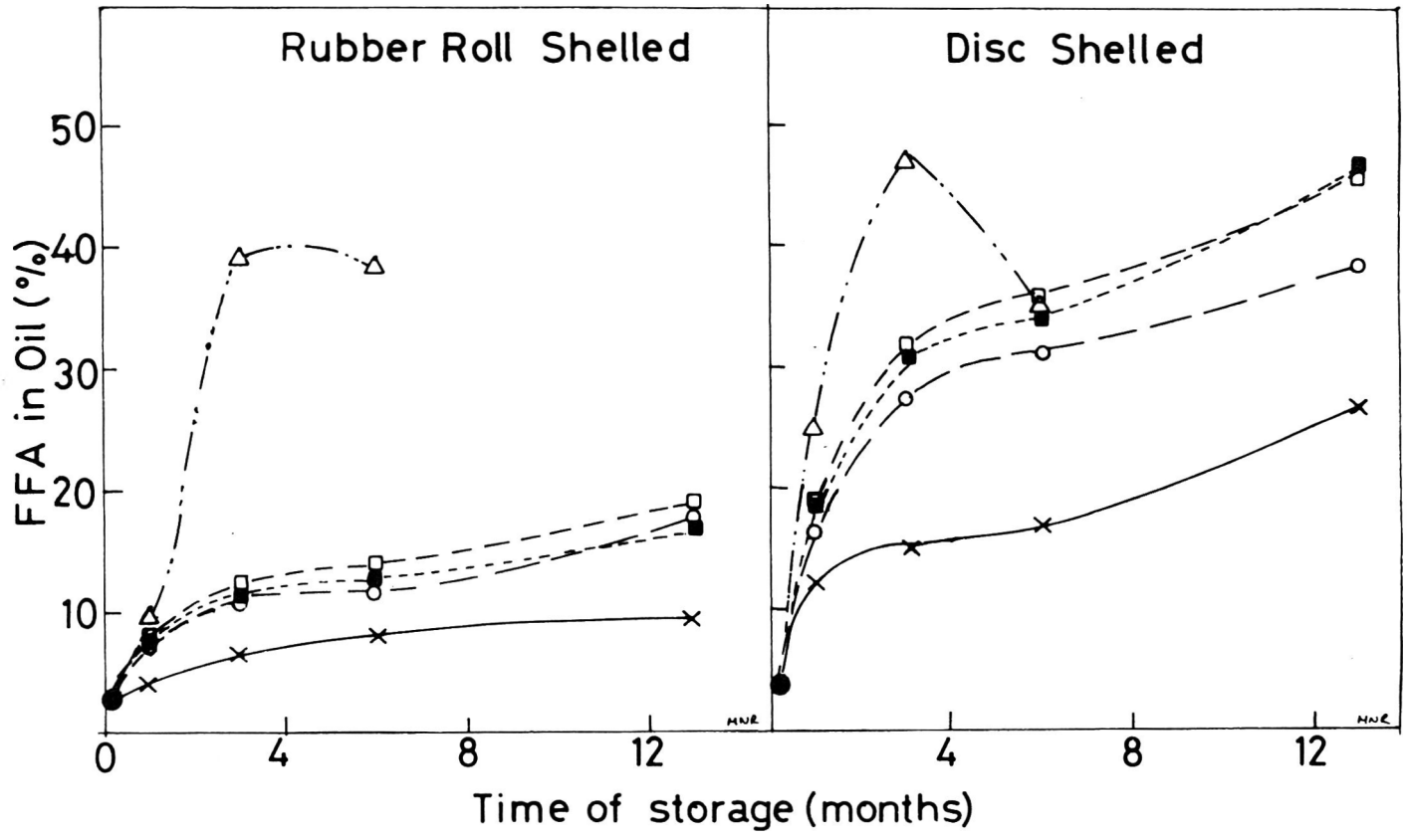

Fig.2. Changes in free fatty acids in bran of brown rice on storing at different conditions.
X : Cold; O : RT; □ : 37°C (62% RH); ■ : 37°C (62% RH) with insects; Δ : 37°C (92% RH).

TABLE 1. STATISTICAL SIGNIFICANCE OF FREE FATTY ACID CHANGES IN BROWN RICE AMONG DIFFERENT CONDITIONS OF STORAGE AND DIFFERENT STORAGE PERIODS[a]

Storage conditions	Storage period, months									
	0		1		3		6		13	
	R	D	R	D	R	D	R	D	R	D
RT	aA	aB	bA	bC	cA	cC	cA	dC	dA	eC
37°C-62% RH	aA	aB	bAB	bD	cB	cD	dB	dD	eB	eD
RT, with insects	aA	aB	bB	bD	cE	cF	-	-	-	-
37°C-62% RH, with insects	aA	aB	bAB	bD	cB	cG	dB	dE	eAB	eD
Cold	aA	aB	bA	bC	cA	cC	dA	cC	eA	dB
37°C-92% RH,	aA	aB	bB	bD	cB	cD	dB	dB	-	-
RT, with 10% brokens	aA	-	bE	-	cE	-	dC	-	eC	-
RT, with 20% brokens	aA	-	bE	-	cE	-	dC	-	eC	-

[a] Different small letters in each row show statistically significant differences ($P < 0.05$) among different periods at each condition of storage. Different capital letters in each column show statistically significant differences ($P < 0.05$) among different conditions of storage at each period. R: rubber roll sheller, D: disc sheller, RT: room temperature.

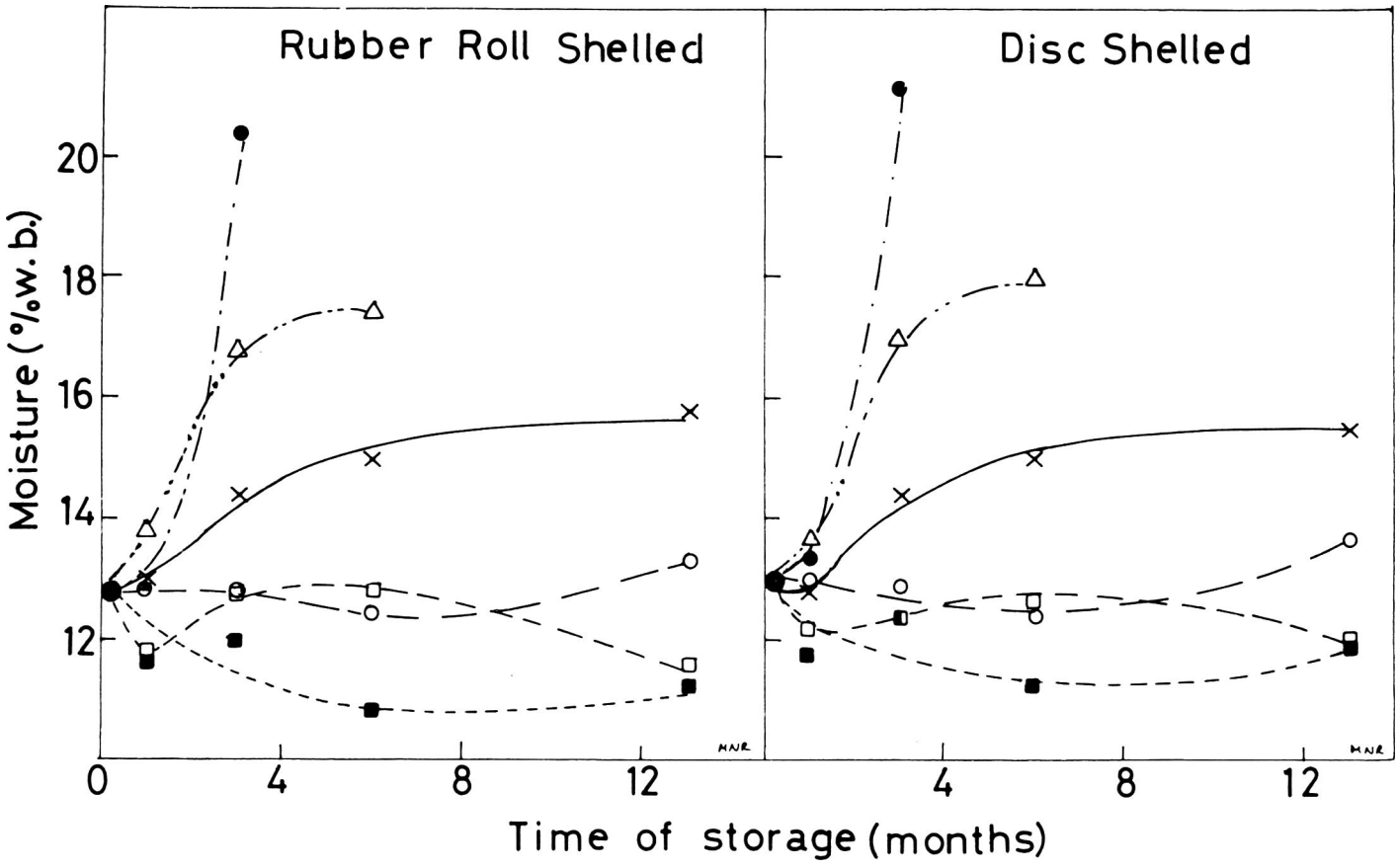

Fig.3. Changes in moisture content in brown rice during storage at different conditions.
X : Cold; O : RT; ● : RT with insects; □ : 37°C (62% RH); ■ : 37°C (62% RH) with insects; Δ : 37°C (92% RH).

rise to high moisture content in rice (Fig.3), which is conducive for enzymatic hydrolysis of fat (Sowbhagya and Bhattacharya 1976). Moisture content of about 15.5% in samples stored in cold explains the increased FFA values, which otherwise appears unexpected. In earlier studies also, FFA was found to increase with increase in moisture content not only in paddy (Houston et al. 1957), but also in steamed (which destroys enzyme activity to a great extent) rice on storage (Houston et al. 1951; Desikachar et al. 1969).

Rate of development of FFA was faster and of greater magnitute in brown rice obtained from disc sheller than in that from rubber roll sheller, the values of disc shelled samples being generally twice those of rubber roll shelled ones at same periods of storage (Fig. 1). Data indicate that bran layers are bruised to a greater extent on shelling paddy in disc sheller than on shelling in rubber roll sheller, which in turn, exposes the lipolytic enzymes for faster activity. Earlier, Hunter et al. (1951) predicted a similar possibility.

Addition of insects resulted in high infestation at RT and FFA values increased several fold as compared to samples without insects (Fig. 1). This was accompanied by high moisture content in rice samples (20.4 and 21.3% for rubber roll shelled and disc-shelled rice, respectively after 3 months). This explains the high FFA values and the rice spoilage as evidenced by the cake-like formation (Fig. 4). Pingle et al (1957) also reported that the husked rice got infected to a greater extent than milled raw and parboiled rice, and resulted in a non-appealing appearance. Futher, Pillaiyar (1979) reported an increase in FFA and insect infestation in stabilised and unstabilised parboiled and raw rice bran stored in gunny bags. Other workers (Adams 1977; Swaminathan 1977) also reported decrease in fat content and increase in FFA during insect development. However, the insects did not survive at higher temperature of storage (37°C - 62% RH) and the values of FFA did not show much variation between samples with and without insects.

Development of FFA in oil of bran fractions, obtained by milling the brown rice stored for different periods, showed a similar pattern as that of brown rice (Fig. 2); even the statistical difference of values among samples being the same (data not

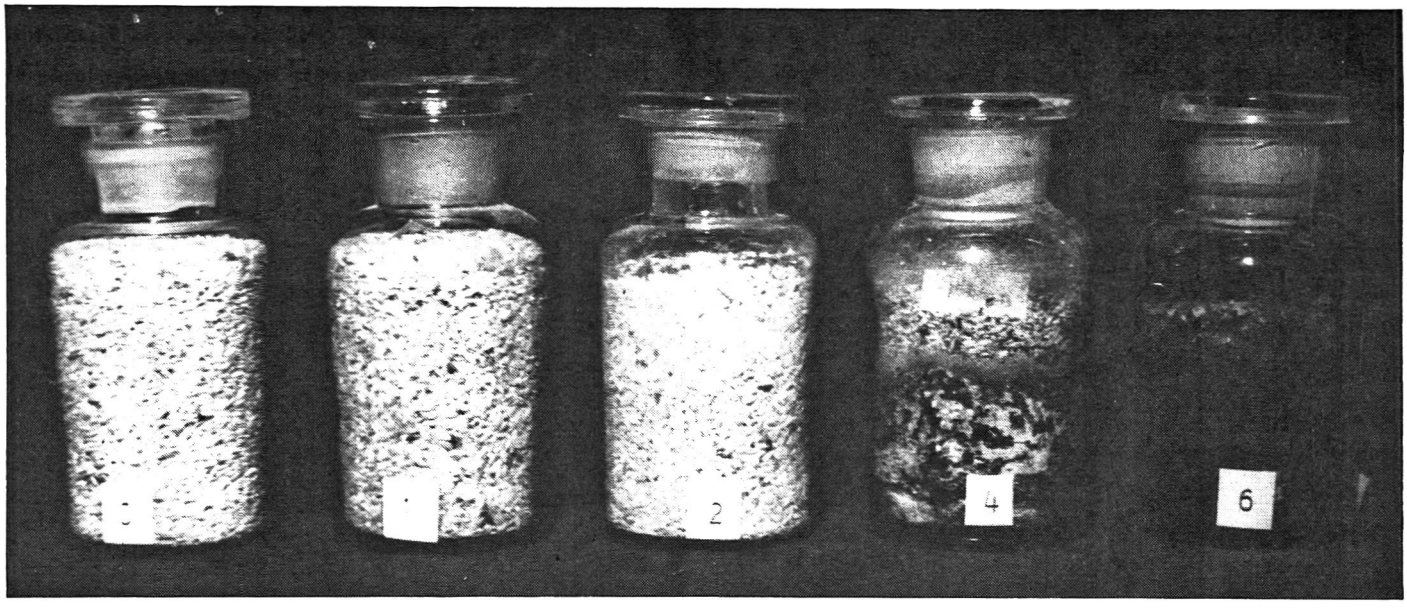

Fig.4. Rubber roll shelled brown rice stored at RT with insects. Labels indicate the period of storage in months.

presented). These data showed that the rice should not be stored as brown rice for periods longer than 3 months or 2 weeks, if shelled in rubber roll sheller and in disc sheller, respectively, when the bran from such rice is to be used for extracting edible oil. In both the cases, FFA increased to more than 10% in such short duration. As the major bulk of oil in rice kernel is distributed in the outer bran layers (Houston 1972; Houston et al. 1951), the actual content of FFA in the rice kernel endosperm (milled rice) cannot be very high, although a control sample of milled rice stored at RT showed FFA values, very similar to disc shelled rice in the same environment (Fig. 1). Data in Table 2 show that the FFA content (mg%) in milled rice, obtained from brown rice stored for 13 months, is not very high as compared to a control sample stored as milled rice. Hence, storing as brown rice does not affect the quality of milled rice substantially to give a dismal picture.

TABLE 2. CONTENT OF FREE FATTY ACIDS IN BROWN RICE, MILLED RICE, AND BRAN AFTER STORING FOR 13 MONTHS AS BROWN RICE

Storage conditions[a]	Free fatty acids in Brown rice (mg/100 g)		Milled rice (mg/100 g)		Bran[b] (mg/8 g)	
	R	D	R	D	R	D
Cold	183	516	30	41	141	437
RT	318	674	40	48	256	576
37°C-62% RH	363	813	57	62	281	691
37°C-92% RH	761	855	108	71	601	721

[a] Sample codes same as in Table 1.

[b] Values for 8 g bran given for easy comparison as rice is polished to 8% degree of polish.

Milled rice stored after polishing to 8% degree of polish gave 93 mg free fatty acids/100 g.

Presence of broken kernels in brown rice did not result in appreciable change in FFA values than the corresponding samples stored without brokens (Fig. 1). For example, there were no significant differences among values of samples after addition of 10 and 20% brokens (Table 1) and hence values only those of 10% brokens are shown in Fig. 1. Commercially dehulled brown rice samples (which normally contain brokens) could, thus, be stored with the same results as reported here.

Effect of storing in cloth bags : When brown rice was stored in cloth bags, the trend of changes in FFA with period of storage was similar to that of bottle storage at all conditions studied. However, the FFA values were slightly higher, as compared to those of bottle-stored samples, except at 37°C - 62% RH, where the values remained more or less same. Similar observations were made when stabilized bran was stored in bags and bottles, the latter being more suitable for storage at high humidity (Barber et al. 1974; Loeb et al. 1949).

Effect of addition of insects on other properties : Insect development was very rapid at RT and resulted in complete spoilage of brown rice within 3 months. There was no difference as to the development of infestation in rice between the two methods of shelling, except for slightly higher population after 3 months in disc - shelled rice. Insects not only did not breed, but also did not

TABLE 3. EFFECT OF ADDITION OF INSECTS ON STORABILITY OF BROWN RICE AT DIFFERENT CONDITIONS

Period of storage, months	Storage conditions*	Moisture, % w.b.	No. of living insects	Kernel damage, %	Frass, mg/100 g	Uric acid mg/100g	Mould count 10^5	Psocids
0	—	12.6	0	0	0	0	0.01	0
1	R-RT, with insects	12.9	19	0	0	2.2	0.037	0
	R-37°C - 62% RH, with insects	12.3	3	0	0	0.5	0.023	0
	D-RT, with insects	13.1	19	0	0	2.0	0.020	0
	D-37°C - 62% RH, with insects	12.3	3	0	0	1.0	0.023	0
2	R-RT, with insects	14.6	492	6	0.5	18.50	2.250	0
	R-37°C - 62% RH, with insects	12.0	2	0	0.1	2.25	2.000	10,000
	D-RT, with insects	15.5	431	7	0.6	17.50	2.250	0
	D-37°C - 62% RH, with insects	12.0	2	0	0.1	2.25	2.000	10,000
3	R-RT, with insects	20.4	2904	67	13.0	83.0	1100	0
	R-37°C - 62% RH, with insects	12.0	4	0	0	3.0	7.0	100,000
	D-RT, with insects	21.2	4547	72	13.5	97.0	1500	0
	D-37°C - 62% RH, with insects	11.8	4	0	0	6.5	8.5	100,000
	R-RT	12.6	0	0	0	0	0.03	0
	R-37°C - 62% RH	12.4	0	0	0	0	0.03	100
	D-RT	12.7	0	0	0	0	0.035	0
	D-37°C - 62% RH	12.5	0	0	0	0	0.03	0

*Sample codes same as in Table 1.

survive much at 37°C - 62% RH. However, there was a heavy psocid population in both rubber roll shelled and disc-shelled brown rice, which was absent in samples stored at RT. With increase in the insect activity, there was increase in kernel damage, frass, uric acid, mould count, and moisture content of the samples (Table 3). High respiratory activity was also evident in samples with higher insect population as indicated by rise in grain temperature by 6-7°C.

The following conclusions can be drawn from the present study : (a) Brown rice obtained by shelling paddy in rubber roll sheller can be stored upto 3 months at RT (23-34°C) and for one year in cold (4-6°C), as the FFA in either case reached only about 10% of the total fat, (b) The bran obtained from such stored rice can be used for extraction of oil for edible purpose, (c) Disc - shelled rice cannot be stored for more than 2 weeks even in cold as the FFA increased to more than 12% and (d) Milled rice obtained from the brown rice which stored for one year is better than rice stored as milled rice, as the former contained only half the amount of FFA.

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Storage Stability of Commercially Available Weaning Foods

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Equilibrium moisture content and water activity data obtained at 20°C, 30°C, 40°C, and 50°C for two commercially available weaning foods were analysed by isotherm models and isosteric heat values were calculated. The analysis revealed that a range of water activity exists at all temperatures, at which major moisture related changes take place in these foods. At 50°C, where worst possible storage conditions will be created, the storage stability of these foods is maximum at water activity of 0.275-0.315 and 0.285-0.350 corresponding to equilibrium moisture content of 0.026-0.028 g water/g dry solid and 0.047-0.055 g water/g soild. With increase in moisture content, the moisture binding energy decreases and moisture becomes more free. Isosteric heat decreases with an increase in moisture content at all four temperatures.

Keywords : Equilibrium moisture content, Local isotherm, Stability isotherm, Isosteric heat, Water activity, Monolayer moisture.

Processed foodstuffs are most stable in an optimum range of moisture content, above and below which, they are prone to rapid deterioration in quality and flavour. Several studies have been carried out on the storage stability of weaning foods based on pulses and ragi. The results have shown that these foods could be stored upto a period of 2-5 months depending on the moisture content of the food and the type of container (Malleshi et al. 1989; Malleshi and Desikachar (1982). The critical moisture content at which food material is at stable condition is governed by monolayer moisture value. Critical moisture contents such as bound water and moisture contents at change over points in the nature of moisture bindings in foods are of interest to processors. This paper deals with the local and stability isotherms and isosteric heat models.

Materials and Methods

Equilibrium moisture levels at different water activities were determined experimentally for both adsorption, and desorption processes at 20, 30, 40 50°C, using dynamic method. Based on the working principle described by Smith (1965), an apparatus was developed by the authors, similar to that used by Bandhyopadhyay et al. (1987). The working of the apparatus is that atmospheric air is compressed to a gauge pressure of 3 to 5.5 kPa, and is allowed to bubble through concentrated sulphuric acid, which results in partial dehydration. The air is fully dehydrated by allowing it to pass through two calcium chloride towers, arranged in series. The dehydrated air is then divided into two different streams, by means of a set of capillary tubes named as proportioning valve. One of the streams remains dry, and the other is saturated with water vapours by bubbling air through distilled water contained in three saturators. The proportioning valve consists of six capillary tubes, having length 20,10,10,4,4 and 4 cm, which divide 0.05, 0.10, 0.10, 0.25, 0.25 and 0.25 fractions of main air stream, respectively into either dry and wet line. By suitable combination of the capillary tubes, any fraction of the main air stream ranging from 0.05 to 0.95 with 0.05 interval can be directed either to the dry or the wet line. The dry and the wet saturated stream are mixed to give the mixed air stream at a certain desired relative humidity.

The commercially available brands of weaning foods A and B were used. Food A had the following composition (in %) : fat 9.0, proteins 15.5, moisture 2.5, carbohydrates 85.7, ash 3.2, dietary fibre 4.1, while food B had fat 1.0, proteins 6.0, moisture 4.0, carbohydrates 86.0, ash 0.7, and dietary fibre 2.7. Weaning food A is wheat-based, whereas food B is rice-based.

A local isotherm results, when the partial molar free energy, ΔF is plotted against the moisture content of the material. The value of ΔF is obtained by

$$\Delta F = -RT \ln a_w \quad \text{.....(1)}$$

The free energy required to transfer water molecules from the vapour state to solid surface is a measure of the affinity of the solid for the vapour. A combination of intersecting straight lines is usually obtained from such a plot. In the local isotherm plot, a change in the nature of moisture

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binding is revealed at moisture contents at each inflection point.

Stability isotherm is obtained, when the first derivative of the moisture, d_{Me}/da_w is plotted against the water activity (a_w). The value of d_{Me}/da_w is obtained by differentiating, and rearranging GAB equation with respect to a_w as described by Sawhney et al. (1989), and is equal to

$$d_{Me}/da_w = (C_1 + C_2 a_w + C_3 a_w) - a_w (C_1 + C_2 a_w + C_3 a_w) \cdot (C_2 + 2C_3 a_w) \quad \text{.....(2)}$$

Where $C_1 = 1/(Mg.\ Cg.\ Kg)$,

$C_2 = (1/Mg) \cdot [(Cg-2)/Cg]$, and

$C_3 = (Kg/Mg) \cdot [(1-Cg)/Cg]$

The minima of stability isotherm indicates the value of a_w, where the food shows the least inclination to moisture adsorption. This is a very stable state with regard to the change in the relative humidity of the atmosphere surrounding food.

During process of adsorption/desorption of water on to/from the food surface, some heat is released/adsorbed by the surface, which is termed as heat of adsorption and heat of desorption, respectively. Chung and Pfost (1967) described these heats as

$$\Delta Hst = R \frac{[T_1 - T_2]}{[T_1 \cdot T_2]} \ln (p_1/p_2) \quad \text{.....(3)}$$

The value of Hst is applicable for a temperature T, so that

$$1/T = 1/2\ [1/T_1 + 1/T_2] \quad \text{.....(4)}$$

The heat of adsorption or desorption indicates the binding energy or the intermolecular forces between the molecules of water vapour and the surface of adsorbent.

Results and Discussion

The water activity and equilibrium moisture content data obtained at 20, 30, 40 and 50°C were used to plot 'local' and 'stability' isotherms. These isotherms result mostly in combination of three straight lines with two change over points (inflection points). The points of discontinuity indicate a change in the nature of water binding. It is also observed that at low moisture content, change in free energy is very high (i.e. - F value is very high), which implies very tight binding of water molecules to the food. The change in free energy value decreases with an increase in moisture, indicating the moisture becoming more free or unbound in nature. The moisture content and water activity at inflection points on local isotherm for both the weaning foods are given in Table 1. The first discontinuity at 20°C occurred in case of weaning food A, at moisture content of 0.038 corresponding

TABLE 1. CRITICAL WATER ACTIVITY AND MOISTURE CONTENT VALUES OF WEANING FOODS

Temp °C		Minima of stability isotherm		Gibb's free energy change: First inflection point		Second inflection point	
		a_w	Me (% d.b.)	a_w	Mc (% d.b.)	a_w	Me (% d.b.)
				Weaning Food A			
20	ad	0.254	3.8	0.25	3.8	0.695	9.7
20	de	0.254	5.2	0.305	5.7	0.655	9.5
30	ad	0.258	3.6	0.290	3.9	0.695	8.2
30	de	0.258	4.3	0.310	4.7	0.650	8.8
40	ad	0.264	3.0	0.315	3.5	0.666	7.8
40	de	0.264	3.6	0.350	4.0	0.650	8.3
50	ad	0.275	2.6	0.315	2.8	0.705	7.5
50	de	0.275	3.1	0.305	3.4	0.660	7.5
				Weaning Food B			
20	ad	0.380	11.5	0.255	9.1	0.640	15.4
20	de	0.380	12.4	0.220	9.6	0.604	15.3
30	ad	0.520	13.1	0.320	9.3	0.660	15.3
30	de	0.520	13.6	0.405	11.8	0.615	15.1
40	ad	0.395	7.7	0.250	5.8	0.575	10.8
40	de	0.395	8.7	0.280	7.3	0.580	11.2
50	ad	0.295	5.1	0.285	4.7	0.665	10.3
50	de	0.295	5.9	0.310	6.3	0.645	10.3

ad - Adsorption, de - Desorption

to water activity 0.25 and is nearly equal to minima of the stability isotherm during moisture adsorption. Similarly, first discontinuity at 20°C for weaning food B occurred at moisture content of 0.091 corresponding to water activity of 0.255, and is little lower than minima of stability isotherm during adsorption. The values of water activity a_w, at the minima of stability isotherms for these foods and corresponding values of moisture content Me are also given in Table 1. It is observed that for weaning food B, minima of stability isotherms fall within the range of moisture contents, represented by second segment of local isotherm. It conforms to the view that second segment of local isotherm represents a moisture content in which food is more stable. But for weaning food A, minima of stability isotherms at higher temperatures (40°C and 50°C) fall in the first segment of local isotherm. Therefore, a shorter and more accurate range of water activity and moisture content can predict the most stable condition. Storage at 50°C, which may possibly generate worst storage conditions, the storage stability of weaning foods A and B is found to be maximum in water activity range of 0.275-0.315 and 0.285-0.350, corresponding to equilibrium moisture content of 0.026-0.028 g water/g dry solid and 0.047 - 0.055 g water/g dry soild, respectively.

Isosteric heat values found at moisture contents 0.06, 0.09, 0.12, 0.15 and 0.18 g water/g dry solid are shown in Fig.1. These values were found between temperatures 20°C and 50°C. The isosteric heat isotherms thus obtained, correspond to the isotherms at 34.25°C. It is observed that isosteric heat decreases exponentially with an increase in moisture content. For weaning food A, isosteric heat decreases from 5.57×10^7 to 4.53×10^7 Pa.m/kg. mol, as the moisture content increases from 0.03 to 0.18 g water/g dry solid during adsorption, and increases from 4.56×10^7 to 7.00×10^7 Pa.m/kg. mol. as moisture decreases from 0.18 to 0.03 g water/g dry solid during desorption. Isosteric heats are observed to be higher during desorption as compared to adsorption process, which means higher energy is required to attain equilibrium by losing moisture than by gaining it. A similar trend is observed for weaning food B. The isosteric heat values for both foods are found close to each other above moisture of 0.15 g water/g dry solid, and in the vicinity of latent heat of vaporization of pure water at isotherm temperature. This reveals that at higher moisture content, nature of moisture

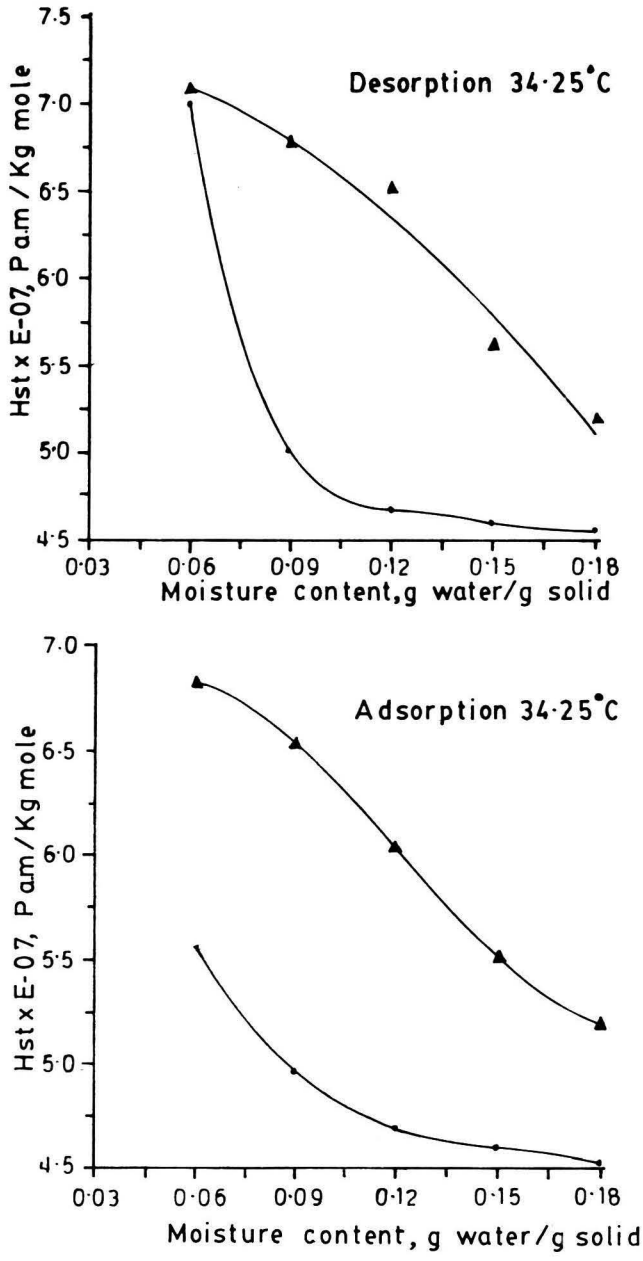

Fig.1. Isosteric heat curve for weaning foods.
● - Food A, ▲ : Food B

associated with foods approaches that of pure water.

Notations

a_w	=	Water activity, fraction
C_1, C_2, C_3	=	Constants of GAB equation
Cg	=	GAB sorption constant related to monolayer properties, g water/g dry solid.
Δ F	=	Free energy changes, Pa.m/kg. mole
Δ Hst	=	Isosteric heat, Pa.m/kg mole
kg	=	GAB sorption constant related to multilayer properties, g water/g dry solid.

Me = Equilibrium moisture content, g water/ g dry solid.

Mg = GAB monomolecular moisture content, g water/g dry solid.

P1, P2 = Vapour pressure at temperatures T_1 and T_2, respectively.

R = Universal gas constant, 8.314 x 10 Pa.m/kg mole K

T = Absolute temperature, K

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Spatial Dimensions of Soybeans and Their Dependence on Grain Moisture Conditions

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Spatial dimensions of soybean grains were determined for each lot at different moisture contents up to 114% d.b. A typical phenomenon of increase in dimensions was observed. Grain length increased by over 60%, whereas the changes in breadth and thickness were about 20 and 16%, respectively for increase in moisture content upto 114% d.b. In addition, about 18% decrease in grain sphericity, 26% increase in size and over 90% increase in 1000 grain mass were observed. These major changes in the longitudinal direction alone would lead to increase in aerodynamic properties of the material. The data, thus, indicate the need to design the individual system for each moisture content range of the soybeans.

Keywords : Soybean properties, Gravimetric properties, Spatial dimensions, Shape character, Grain moisture conditions.

Spatial dimensions of grains are of importance in the design of grading equipments, pneumatic conveying, and fluidized bed processing, especially due to the dependence of aerodynamic properties on these dimensions. It is well known that the spatial dimensions, like shape and size of the grain, change during grain processing steps such as hydration or drying (Mohsenin 1970). In turn, these alter the drag force (Mohsenin 1970). Likewise, cleaning and grading systems might need different sieve sets for handling grains with different moisture levels. Therefore, a need exists to appropriately design the specific processing system e.g., handling, pneumatic conveying etc., with provisions to take care of such changed aerodynamic properties. Earlier researchers (Mohsenin 1970; Shepherd and Bhardwaj 1986; Kulkarni et al. 1988) have consequently taken the cognizance of the importance of information on these grain properties. However, most of the studies have been confined to the existing grain moisture condition (Sharma et al. 1985), and narrow range of moisture content (Sreenarayanan et al. 1985; Dev et al. 1982; Dutta et al. 1988; Grover and Kumar 1985). Few reports are also available on wide range of moisture content (Kulkarni et al. 1988; Siripurapu and Jain 1985). It has been observed (Kulkarni et al. 1988) that the spatial dimensions of the grain, obtained after hydration (Siripurapu and Jain 1985), are different from those obtained after conditioning (Kulkarni et al. 1988). In addition, the moisture distribution is not uniform within the grain when grains are hydrated (Butcher and Stenvert 1973). It, thus, becomes imperative to indicate the grain moisture condition, while reporting the data (Anon 1980). To know the pattern of changes in the spatial dimensions of grains over the wide range of moisture levels, the present study was undertaken and the data have been analysed for various interactions and dependence.

Materials and Methods

Soybeans (variety 'JS-7244') were cleaned, and the moisture content was determined by exposing the sample to 105°C for 16 h (Roberts and Roberts 1972). Samples were conditioned by adding calculated quantity of water (equation 1) to attain desired moisture level in the range of 8 to 114% d. b., after equilibrating for 24 h.

$$W_1 (100 + M_2) = W_2 (100 + M_1) \quad \ldots\ldots (1)$$

Where, W_1, W_2 are initial and final mass (g) of the sample;

M_1, M_2 are initial and final moisture content of the sample; respectively.

Final moisture content of the conditioned sample was determined, before using the randomly selected grains for measurement of spatial dimensions. The size and sphericity were calculated using the formulae (Mohsenin 1970);

$$\text{Size, mm} = (a\ b\ c)^{1/3} \quad \ldots\ldots (2)$$

$$\text{Sphericity} = \frac{(a\ b\ c)^{1/3}}{a} \quad \ldots\ldots (3)$$

Where, a, b, c are length, breadth or width and thickness of grain in mm, respectively.

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The elongation and flatness ratios were calculated as follows (Kelly and Spottiswood 1982);

$$\text{Elongation ratio } (R_E) = \frac{\text{length}}{\text{breadth}} \quad \ldots\ldots (4)$$

$$\text{Flatness ratio } (R_F) = \frac{\text{breadth}}{\text{thickness}} \quad \ldots\ldots (5)$$

The 1000 grain mass was determined by counting randomly selected grains and weighing. In another set, soybeans were separated in three grades using Ro-tap sieve shaker with different sieves. The grain properties were determined to evaluate the variation with respect to grade and general lot. The variation of size, sphericity and other properties were studied for entire range of these experiments.

Results and Discussion

The grain dimensions measured, i.e., length, breadth and thickness, were found to vary with increase in moisture content (Table 1). A linear relationship was observed between the length and moisture content % d. b. (M), with acceptable correlation, e. g.,

Length, mm = 6.812 + 0.034 M (r = 0.962) ... (6)

Breadth of soybean varied with grain moisture content (% d.b.) with fair correlation :

Breadth, mm = 6.220 + 0.010 M (r = 0.681) ... (7)

Data in Table 1 show over 60% change in grain length, whereas breadth and thickness changes were in the range of 16-20% for about fourteen-fold change in grain moisture level i.e., from 7.80 to 114% d.b. Grain size is calculated using equation 2, increased with moisture content at fairly good correlation (r = 0.789) :

Size, mm = 6.020 + 0.014 M .....(8)

Over 26% increase was observed in all the cases (Table 1).

TABLE 1. VARIATION OF SOYBEAN GRAIN DIMENSIONS WITH MOISTURE CONTENT

Moisture content % d. b.	Length, mm	Breadth, mm	Thickness, mm
7.80	6.89	6.19	5.13
12.87	7.12	6.06	4.74
41.20	8.55	6.90	5.53
54.86	9.17	7.37	6.07
63.10	9.19	7.22	5.63
69.65	9.06	7.10	5.52
74.60	8.89	6.34	5.06
81.83	9.10	6.63	4.81
98.76	10.12	7.31	5.92
113.99	10.85	7.41	5.47
SD range	± 0.32 - 1.96	± 0.23 - 0.77	± 0.21 - 0.67

The coefficient of variation based on the means of respective values of length, breadth, and thickness varied from 4 to 18%. Such variations are common in biological materials, and the samples, which are randomly selected from general lot for determination of properties based on single grain determinations (Mohsenin 1970).

Sphericity of soybean decreased linearly with the increase in moisture content (Fig. 1). The reduction was about 20% for the entire range of experiments in the study :

Sphericity = 0.868 - 0.001 M (r = -0.935) ...(9)

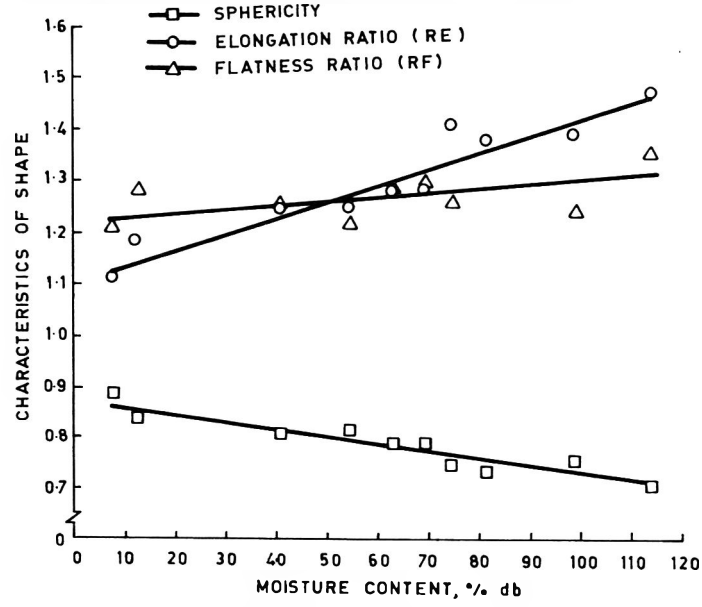

Fig. 1. Variation of sphericity, elongation ratio (RE) and flatness ratio (RF) with moisture content.

Elongation ratio (RE), which is a relative proportion of length (L) with breadth (B) at any given grain condition, was found to vary linearly (Fig. 1) with the grain moisture content (M) as :

RE = 1.105 + 0.003 M (r = 0.952) ..... (10)

Where RE = 1 for the condition where L = B. The equation 10 also shows near unity values at lower moisture levels, thereby reflecting an appreciable change in grain length over breadth. As the elongation ratio involves the length component, its unity value should contribute in improved grain sphericity. Higher values of elongation ratio should, therefore, represent poor sphericity of the material. This aspect is reflected in the following equation and Fig. 2.

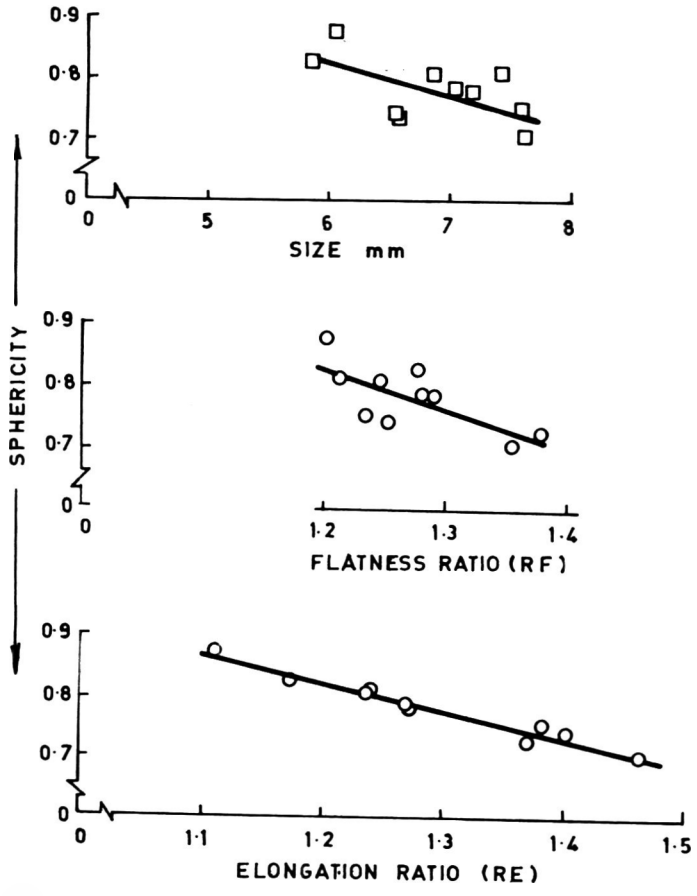

Fig.2. Variation of sphericity with average grain size, flatness ratio and elongation ratio of soybean.

Sphericity = 1.383 - 0.466 RE (r = 0.984) ... (11)

On the other hand, the flatness ratio (RF) did not show any direct correlation with increase in moisture content (r = 0.526). This may be due to insignificant and proportionate changes in the breadth and thickness of the grains as well as the random selection of the grain for determination of the property. The term flatness ratio implies that more the flatness ratio, more is the grain breadth or more flatter the grain or poor the grain sphericity (equations 3 and 5). Grain sphericity showed fairly good correlation in this regard (Fig. 2) :

Sphericity = 1.606 - 0.648 RF (r = 0.701) ... (12)

Poor correlation of size (r = 0.571) with sphericity again shows the significance of length change (equation 2) with grain moisture, and its effect on sphericity. The variations of sphericity with average grain size, flatness and elongation ratios are shown in Fig. 2.

One thousand grain mass increased linearly with the increase in moisture content :

1000 grain weight = 104.231 + 0.988 M (r =0.970) ...(13)

The information on properties of graded lots of same variety of soybean grains (Table 2), revealed variation in these properties from lot to lot. The data, thus, indicate the need for grading of grains before processing for better performance of a designed system, such as cleaning, pneumatic handling or any other unit operation.

Conclusion

The data reveal the changes in length, breadth, and one thousand grain mass thickness of soybean grains during moisture absorption from 8 to 114% d. b., thereby indicating the need for using graded material for efficient processing.

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TABLE 2. VARIATION IN DIMENSIONS OF SOYBEAN (MOISTURE CONTENT 7.8% d.b.) WITH SIZE GRADE

Size grade, mm	Mass, fraction	Length, mm	Breadth, mm	Thickness, mm	Elongation ratio	Flatness ratio	Size, mm	Sphericity
+ 4	0.53	7.10	6.18	5.44	1.11	1.19	6.32	0.28
- 4, + 3.35	0.44	6.65	5.42	4.82	1.12	1.23	5.74	0.87
- 3.35 + 1.7	0.03	5.87	5.07	3.90	1.16	1.30	4.88	0.83
General	1.00	6.89	6.19	5.13	1.11	1.21	6.03	0.88

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Quality Evaluation of 'Landrace' and 'Durum' Wheat Cultivars Grown in Jordan

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Ten 'Durum' wheat cultivars (including a 'Landrace', old and newly developed cultivars) have been evaluated for a number of quality parameters. The 'Landrace Horani-27, had the highest protein content (14.4%), but intermediate flour yield (43.7%). 'Laccsh', gave highest flour yield (57.5%), particle size index (PSI) (19.7%), sodium dodecyl sulphate sedimentation (SDS-SED) (52 ml), but was intermediate in protein content (9.7%). The old cultivars, 'Der Alla-2' and 'Der Alla-6' had no clear-cut patterns for the quality parameters studied. Proximate analysis showed that these cultivars were higher than average for fibre and ash contents. It was possible to cluster these cultivars into 3 groups each for flour yield, protein content, SDS-SED, PSI % and thousand kernel weight. The variability in the composition of each cluster allows for combining certain cultivars to complement each other for a better balance of these quality traits.

Keywords : Wheat cultivars, Quality evaluation, 'Landrace', 'Durum', Jordan, Cluster analysis

"Durum' wheat is the most important crop in Jordan in terms of acreage, and total gross value. The per capita consumption of wheat is about 170 kg/year, mostly as flat bread, and to a lesser extent, as *bulgur* (boiled wheat grits) and *farikah* (roasted wheat at hard dough stage). However, average yield is among the lowest in the world, being 740 kg/ha. Efforts are underway to improve wheat yield through breeding, and/or introducing high yielding cultivars (Jaradat 1988). Ten 'Durum' wheat cultivars including a 'Landrace', improved and newly introduced cultivars were compared for their physico-chemical properties, mineral composition, and gel electrophoretic patterns, to generate information on these vital aspects of quality.

Materials and Methods

Nine 'Durum' wheat cultivars and a 'Landrace' (Table 1) were planted in Maru Experimental Station (North Jordan), and the rainfall during growth season was 380 mm. Cultivars and 'Landrace' were planted in plots of 4 m^2 in a randomized complete block design with four replications. Plants were harvested at the dry stage and threshed. Sub-samples of 100 g for each treatment were milled on a Tecator 1093 cyclotec sample mill (Tecator, Sweden), the milled product was passed through a 0.40 mm screen, and used for chemical analysis. Other sub-samples of 500 g were tempered to 15.5% moisture content, milled into flour on a MLV 202 Buhler laboratory mill (Uzwil, Switzerland), and used for flour testing.

Proximate analysis : Moisture, protein (N x 5.7), crude fat, wet gluten, and ash were determined according to AACC (1982). Crude fibre was determined according to AOAC (1980). Nitrogen - free extract (NFE) was determined by difference. Particle size index (PSI %) was determined according to Williams et al (1988). Samples of 22 g of wheat were pulverized in a cyclotec sample mill (Tecator, Sweden) at the finest setting, followed by mixing and sieving 10 g of the meal for 10 min on a 200 mesh (74 μm) sieve and the amount of thrust was expressed as % particle size index (PSI). Protein and lysine contents in flour were determined by near infra-red reflectance spectroscopy, using FQC analyzer model 51A (Pacific Scientific, USA) according to Ereifej and Markakis (1983).

Mineral analysis : Mineral contents (Na^{+1}, K^{+1}, Ca^{+2}, Zn^{+2}, Mn^{+2}, Cu^{+2}, $Fe^{(+2,\ +3)}$ and Mg^{+2}) were determined by using an atomic absorption spectrophotometer (Pye Unicam, model Sp9, UK) by wet digestion with a mixture of nitric, sulphuric and perchloric acids (10 : 1 : 4). Phosphorus [$P^{(+3,\ +5)}$] was determined according to Watanabe and Olsen (1965), using a spectronic 21 spectrophotometer. Sodium dodecyl sulphate sedimentation test (SDS-SED) was performed according to Williams et al. (1988). Sodium dodecyl polyacrylamide gel electrophoresis (SDS-PAGE) of whole wheat kernel was done according to Laemmli (1970). A K- means cluster analysis procedure of Hartigan and Wong (1979) was run on mean values for individual traits, as executed by the cluster analysis program of systat according to Wilkinson (1988).

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TABLE 1. COLOUR, ONE THOUSAND KERNEL WEIGHT (TKWT) AND PROXIMATE ANALYSIS OF TEN JORDANIAN WHEAT CULTIVARS[a].

Wheat cultivar	Kernel colour	g/1000 kernel[c]	Protein %	Fat %	Fibre %	Ash %	Nitrogen-free extract %
'ACSAD-65'	Amber	40.7 ± 0.20[c]	10.9[e]*	4.6[a]	2.3[e]	2.0[c]	74.1[e]
'Amra'	Light yellow	33.4 ± 0.05[g]	10.8[e]	3.6[c]	2.4[de]	2.0[c]	75.0[c]
'Der Alla-6'	Yellow	39.1 ± 0.45[e]	10.1[f]	2.2[f]	3.1[a]	1.9[cd]	76.7[a]
'Der Alla-2'	Medium yellow	41.7 ± 0.45[a]	13.8[b]	3.2[d]	2.6[c]	1.7[e]	72.4[h]
'Veery'	Amber	32.0 ± 0.50[i]	11.5[d]	4.6[a]	2.8[b]	2.4[a]	72.6[h]
'Horani-27'[b]	Light yellow	38.1 ± 0.10[f]	14.4[a]	1.4[g]	3.2[a]	2.3[ab]	73.0[g]
'Korifla'	Yellow	41.7 ± 0.45[a]	11.8[c]	3.5[c]	2.5[cd]	2.0[c]	74.6[d]
'Lacesh'	Amber	33.0 ± 0.50[h]	11.4[d]	4.3[b]	2.2[f]	1.8[de]	74.1[e]
'Rabi-S'	Medium yellow	41.1 ± 0.55[b]	11.7[c]	2.9[e]	3.2[a]	2.2[b]	73.5[f]
'Sham-1'	Yellow	39.8 ± 0.10[d]	10.9[e]	2.8[e]	1.9[g]	1.9[cd]	76.3[b]

a: Average of three determinations, b: 'Landracc', c: Values are ± standard deviation.
* Means within columns having different letters are significantly different according to LSD at P ≤ 0.05.

Results and Discussion

Data on thousand kernel weights and proximate composition are given in Table 1. 'Horani-27' and 'Der Alla-2' showed the highest protein content. Among the Indian varieties, when compared to these varieties of Jordan, 'HD-2329' and 'PBW 65' had the highest protein contents (Hira et al. 1991). Protein values are comparable, while ash content is slightly higher than the values reported by Simmonds (1978). The nitrogen - free extract (NFE), an approximate measure of starch content, was slightly higher than the values reported by Simmonds (1978). Mineral analysis of the Jordanian wheats is presented in Table 2. Except Na and Ca, which were found to be higher than the values for other minerals, the values for K, Zn, Mn, Cu, Fe, Mg and P were comparable to values, reported previously by Watt and Merril (1963), Khatchadourian et al. (1985) and Pomeranz (1987).

Data on wheat flour are presented in Table 3. Lysine was highest in 'Amra' (3.4 g/100 g), but lower in 'Lacesh' and 'Horani-27' (2.9%). Generally, the 'Durum' wheats contain slightly higher lysine than 'hard red spring' wheat (2.82%) and 'Triticale' (2.77%) according to Simmonds (1978). In the Indian varieties, the available lysine contents are reported to range from 2.04 to 2.78 g/16 g N (Chopra and Hira 1986). Flour yield in 'Durum' wheats varied between 29.5 and 57.5%, while total flour lysine ranged between 2.9 and 3.4 mg/100 g.

TABLE 2. MINERAL CONTENT OF WHOLE WHEAT (mg/100 g)[a]

Wheat cultivar	Na	K	Ca	Zn	Mn	Cu	Fe	Mg	P
'ACSAD-65'	9.3[c]*	641.3[a]	66.5[a]	1.8[c]	4.3[d]	3.5[b]	8.5[d]	66.1[e]	217.4[d]
'Amra'	14.4[b]	554 [d]	41.5[b]	2.3[b]	5.3[c]	2.3[d]	6.5[f]	69.4[d]	201.1[g]
'Der Alla-6'	7.0[g]	449 [g]	27.5[f]	1.0[e]	3.8[e]	2.0[e]	10.8[a]	68.9[d]	186.4[h]
Der Alla-2'	7.1[g]	563 [c]	31.0[g]	2.5[a]	6.0[b]	2.0[e]	9.3[c]	71.4[c]	234.5[b]
Veery'	7.3[f]	514.3[f]	25.5[g]	1.8[c]	6.5[a]	1.0[f]	8.3[d]	59.2[g]	168.7[f]
'Horani-27'	7.9[d]	625 [b]	29.0[e]	2.5[a]	3.8[e]	2.3[d]	8.3[d]	85.1[a]	265.3[a]
'Korifla'	7.2[g]	622.8[b]	25.5[g]	1.0[e]	3.8[e]	0.8[g]	10.8[a]	72.3[b]	205.8[e]
'Lacesh'	16.1[a]	517 [f]	39.0[c]	1.8[c]	4.3[d]	3.8[a]	7.8[e]	61.9[f]	201.8[f]
'Rabi-S'	7.6[e]	541.5[e]	12.7[h]	1.5[d]	4.8[c]	2.5[c]	9.8[b]	58.2[h]	227.5[c]
'Sham-1'	9.3[c]	641.3[a]	66.5[a]	1.8[c]	4.3[d]	3.5[b]	8.5[d]	66.1[e]	217.4[d]

a: Average of three determinations on dry basis
* Means within columns having different letters are significantly different according to LSD at P ≤ 0.05.

TABLE 3. PROTEIN, GLUTEN, LYSINE CONTENTS AND MILLING PROPERTIES OF FLOUR OBTAINED FROM THE JORDANIAN WHEAT CULTIVARS*

Wheat cultivar	Protein %	Gluten %	Lysine mg/100g	Flour yield %	Bran %	Short %	Milling time, min	SDS, SED ml	PSI %	Hardness description
'ACSAD-65'	9.0^{h*}	23.3^{g}	3.3^{ab}	42.2^{c}	26.6^{d}	23.1^{c}	8.8^{c}	45^{f}	18.0^{e}	Medium hard
'Amra'	9.5^{ef}	28.4^{a}	3.4^{a}	40.8^{d}	34.1^{a}	20.1^{d}	8.1^{e}	47^{e}	17.1^{f}	Medium hard
'Der Alla-6'	9.3^{g}	25.6^{d}	3.3^{ab}	36.9^{e}	29.5^{b}	23.8^{d}	8.6^{d}	42^{g}	18.7^{c}	Medium hard
Der Alla-2'	10.7^{b}	25.6^{d}	3.0^{de}	36.0^{f}	29.7^{b}	28.6^{a}	7.4^{h}	39^{h}	10.3^{h}	Very hard
Veery'	10.2^{c}	27.1^{b}	2.9^{e}	29.5^{g}	26.3^{d}	5.8^{i}	8.0^{g}	56^{b}	20.3^{a}	Medium hard
'Horani-27'	11.2^{a}	26.5^{c}	2.9^{e}	43.7^{b}	26.5^{d}	24.9^{c}	6.8^{i}	38^{i}	17.3^{f}	Medium hard
'Korifla'	9.4^{fg}	23.8^{f}	3.1^{cd}	40.7^{d}	27.4^{c}	21.2^{f}	8.2^{f}	55^{c}	13.7^{g}	Hard
'Lacesh'	9.6^{e}	27.0^{b}	2.9^{e}	57.5^{a}	26.2^{d}	7.4^{i}	9.8^{b}	52^{d}	19.5^{b}	Medium hard
'Rabi-S'	9.8^{d}	20.2^{h}	3.0^{de}	43.7^{b}	19.1^{e}	8.5^{h}	10.1^{a}	59^{a}	18.3^{d}	Hard
'Sham-1'	9.4^{fg}	24.2^{e}	3.2^{bc}	43.4^{b}	26.5^{d}	26.5^{b}	6.8^{i}	45^{f}	18.0^{e}	Medium hard

a: Values are the average of four determinations. All values are calculated at 14% moisture content. Means within columns having different letters are significantly different at $P < 0.05$ level according to LSD. SDS - SED = Sodium dodecyl sulphate sedimentation, PSI = Particle size index.

The moisture content in flours ranged from 20.2% ('Rabi-S') to 28.4% ('Amra'). All the varieties tested had lower protein, and lower gluten than 'American bread' wheat. Generally, Jordanian wheats contain higher amounts of bran and shorts (19.1% in 'Rabi-S' to 934.1% in 'Amra'). Shorts varied from 5.8% ('Veery') to 28.6% ('Der Alla-2'). Data show that the bran and shorts content of these varieties are higher than those reported by Swanson (1938). SDS-SED, PSI% and hardness description as quality parameters of wheat flour are shown in Table 3. SDS-SED ranged from 38 ml ('Horani-27') to 59 ml ('Rabi-S'), thereby suggesting that these wheat varieties are all 'Durum' type wheats, which are suitable for flat bread making. The SDS-SED values were low because of the poor quality of their gluten according to Kitterman and Barmore (1969).

PSI was used to classify these wheats, according to their hardness using the relative hardness scale by Williams and Sobering (1986). The PSI ranged from 10.3 ('Der Alla-2') to 20.3 ('Veery'). The Jordanian wheats were found to be very hard ('Der Alla-2'), medium hard ('Sham-1', 'Lacesh', 'Amra', 'Der Alla-6' and 'Horani-27'), and hard ('ACSAD-65', 'Rabi-S' and 'Korifla'). SDS-PAGE for whole kernels revealed some differences in the banding patterns of these cultivars (Fig.1). Most cultivars showed thick bands in the 45-78 kDa region. These proteins are mostly α, β, and γ gliadins. The cultivars,

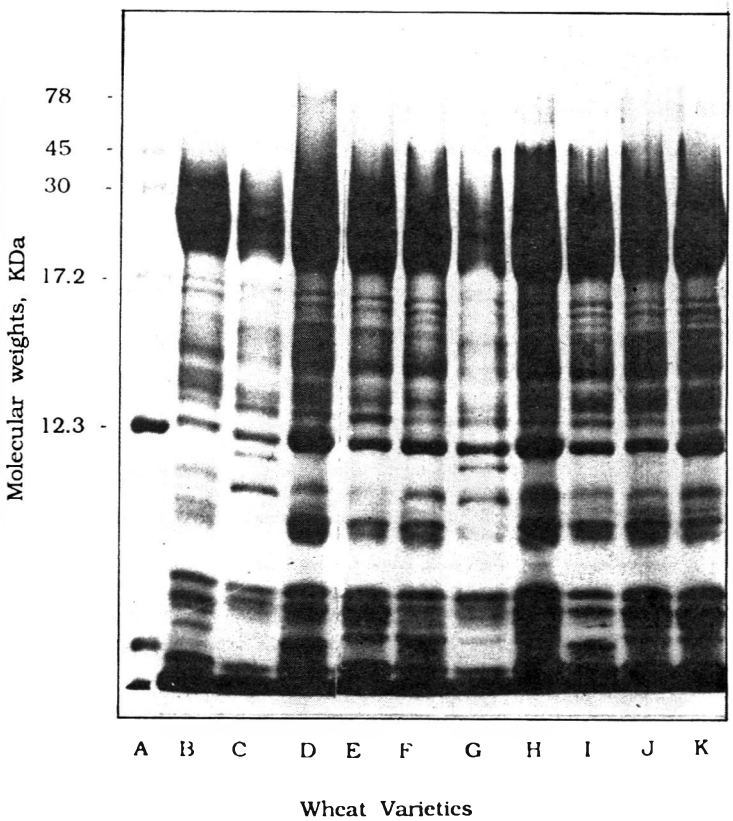

Fig.1. Sodium dodecyl sulphate polyacrylamide gel electrophoresis of whole kernel proteins of the 'Durum' wheats, extracted with 0.1M Tris buffer pH7. A : Molecular weight markers, ovotransferrin (78 kDa), albumin (45 kDa), ovalbumin (30 kDa), carbonic anhydrase (17.2 kDa) and myoglobin (12.3 kDa); B : 'Sham-1'; C : 'Rabi-S'; D: 'Lacesh'; E 'Korifla'; F : 'Horani-2' : G: 'Veery'; H : 'Der Alla-2'; I :'Der Alla-6'; J : 'Amra'; K : 'ACSAD-65'.

'Rabi-S', 'Horani-27' and 'Veery', showed relatively thin bands in this region, when compared with the rest.

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TABLE 4. CLUSTER ANALYSIS FOR FIVE QUALITY TRAITS IN 'DURUM' WHEATS GROWN IN JORDAN

Quality trait	Members of cluster	Mean	S.D.	F	P (0.05)
Flour yield	'Sham-1', 1,2,9,6,4,7	41.28	2.43	29.54	***
	'Lacesh'	57.46			
	'Fari'	29.47			
Grain proteins	'Sham-1', 8,2,1,9,5,7	112.29	3.8	44.23	***
	'Horani-27', 4	141.00			
	'Der Alla-6'	101.00			
SDS-SED	'Horani-27', 4, 3	3.95	0.18	45.60	***
	'Lacesh', 1,9,5,7	5.20	0.23		
	'Sham-1', 2	4.60	0.10		
% PSI	'Sham-1', 8,2,9,6,5,3	18.45	1.06	36.53	***
	'ACSAD-65', 7	13.40	0.28		
	'Der Alla-2'	10.30			
TKWT	'ACSAD-65', 9,4,7	41.30	0.42	138.74	***
	'Lacesh', 2,5	32.80	0.59		
	'Sham-1', 6,3	39.00	0.70		

1 = 'ACSAD-65', 2 = 'Amra', 3 = 'Der Alla-6', 4 = 'Der Alla-2', 5 = 'Veery', 6 = 'Horani-27', 7 = 'Korifla', 8 = 'Lacesh', 9 = 'Rabi -S', 10 = 'Sham-1'
SDS-SED = Sodium dodecyl sulphate sedimentation, PSI = Particle size index, TKWT = Thousand kernel weight

Data for individual quality traits were subjected to cluster analysis, according to Wilkinson (1988). The ten cultivars were separated into three clusters based on each of flour yield, grain proteins. SDS-SED, PSI% and TKWT. Differences between these clusters were highly significant (Table 4). 'Lacesh' was highest in flour yield (57.46%), while 'Veery' was lowest (29.4%). The remaining eight cultivars were intermediate (41.28 ± 2.43%). The local cultivars, 'Horani-27' and 'Der Alla-2', had the highest protein content (14%) and differed significantly from 'Der Alla-6' (10.1%) which had the lowest protein content.

Seven of the cultivars showed typical PSI% values for 'Durum' wheats (18.45 ± 1.06). The local cultivar 'Der Alla-2' showed the lowest value (10.3), while 'ACSAD-65' and 'Korifla' were intermediate (13.4 ± 0.28). SDS-SED was negatively correlated with grain proteins ($r = -0.447$, $P < 0.05$). However, the members of the first cluster for % PSI, except for 'Der Alla-6', had medium grain protein content (11.29 ± 0.38). PSI was negatively correlated with TKWT ($r = -0.66$, $P < 0.05$), which separated the cultivars into three clusters with significant differences between them.

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Application of Reverse Osmosis for Concentration of Buffalo Milk

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Buffalo milk was concentrated to 1.5X (25% TS) and 2.0X (33% TS) levels employing reverse osmosis process and the changes in its physico-chemical, microbial and sensory properties were examined. The average permeate flux was 30.4 and 23.8 l/m²-h for 1.5X and 2.0X, respectively. The lipids, proteins, minerals and lactose in concentrated milk increased by the same level as the concentration factor, thereby indicating their 100% rejection by the thin film composite reverse osmosis membrane. There were no milk solids in the permeate at any stage of operation. The reverse osmosis process did not exert any adverse effect on the titratable acidity, pH, free fatty acids, free fat and microbial load. The sensory quality of reverse osmosis processed milk was not different from that of fresh buffalo milk. The most notable effect of reverse osmosis process was noticed on the size of fat globules, which reduced to less than 2.5μ upon 2.0X concentration. Consequently, the skimming efficiency of reverse osmosis milk was extremely poor.

Keywords : Reverse osmosis, Buffalo milk, Flux, Physico-chemical properties, Fat globule size, Skimming efficiency.

Reverse osmosis (RO) is a relatively new process developed for concentration of aqueous foods. The feed stream under high pressure is allowed to flow across a semi-permeable membrane, which preferentially permits water to pass through it (Smith and MacBean 1978). According to an estimate, installed membrane area for RO in the dairy industry alone is 60,000 m² and its annual growth rate is around 20% (Maubois 1989). Concentration upto 18 to 25% total solids, depending on the feed stream could be done economically by RO, rather than by conventional evaporation processes (Pepper and Orchard 1982; Cheryan et al. 1987). Since the moisture is removed without changing its phase at relatively low operating temperatures, the changes in sensory quality, and chemical make up of the food would be minimum. In addition, RO can treat food plant waste streams to produce reusable water, and reduce effluent discharge to municipal waste treatment facilities (Cheryan et al. 1990).

In recent years, a considerable research work has been carried out on concentration of cow milk (Abbot et al. 1979; Hiddink et al. 1980; Versteeg 1985; Pal and Cheryan 1987), whey (Pepper and Pain 1987) and skim milk (Stabile 1983), employing RO process. The present study was undertaken to examine the feasibility of RO for concentration of buffalo whole milk. The results presented in this paper relate to the changes in physico-chemical characteristics of buffalo milk.

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Materials and Methods

RO plant : A pilot scale RO plant (Paterson Candy International Membrane Division, England) was used. It consisted of a feed tank, a triple plunger-type high pressure pump, a 0.6 m shell and tube heat exchanger, the membrane module and instrumentation, and controls necessary to regulate the operating pressure and temperature. A thin film composite membrane (AFC 99) with 99% retention limit for sodium chloride was used. A total of 18 membranes in tubular configuration with a total area of 0.9 m² were connected in series, and encased in a SS shell. The RO unit was operated at a pressure of 30 kg/cm² and flow rate of 18 l/min. The temperature of milk was maintained at 50°C.

Buffalo milk and its processing : Buffalo milk obtained from the Experimental Dairy of the Institute was standardized to 6% fat. It was then subjected to mild heat treatment (60°C without holding) for inactivation of lipase enzyme. After cooling to 50°C, it was transferred through a filter cloth into the feed tank of RO unit. Milk was recirculated through the RO module, till desired concentration of either 1.5X and 2.0X was achieved with respective volume reduction of 33% or 50%. The performance of the RO system is measured on the basis of the flux and rejection (Cheryan 1986). The flux (the rate of removal of permeate) was measured at a constant interval, during concentration of buffalo milk. Rejection, a measure of the membrane ability to

retain or separate a particular component of the milk, is expressed as :

$$R = (1 - Cp/Cr) \times 100$$

where, Cp and Cr are the concentrations in permeate and retentate, respectively. The retentate was measured for gross composition and the permeate for the total solids.

Analytical methods : The total solids, fat, titratable acidity, pH, alcohol test, methylene blue reduction test and total viable counts in milk and concentrate (retentate) were determined by standard methods (ISI 1961). A semi-microKjeldahl method suggested by Menefee and Overman (1940) was used for measuring the total proteins (N x 6.38). The ash content was determined by the AOAC (1980) method, and lactose by difference. For estimation of free fatty acids (FFA), the method recommended by Deeth et al. (1975) was adopted. An Elico pH meter (Model LI-122) with combined glass electrode was used for measuring the pH of milk and concentrates. Free fat was determined by the method of Hall and Hedrick (1971). The dynamic viscosity of milk and concentrates was measured at 20 ± 1°C with the help of a Hoppler falling ball viscometer (type BH manufactured by Veb Prufgeratewerk Medirgen/Dresden, Germany). The microscopic examination of fat globules for measuring their size and shape was done as per the standard method (Trout 1950). The flavour, which is the most important sensory attribute of fluid milk, was evaluated as per the method of Nelson and Trout (1964), using American Dairy Science Association's abridged score card. For measuring the skimming behaviour, RO concentrates as such and after dilution to the composition of normal buffalo milk (6% fat) were separated at 40°C in a laboratory cream separator (Model Felix 33, Cap 110 l/h, bowl speed 3500 rpm, Vulcan Laval, Poona, India). The fat test on skim milk and whole milk was done by Gerber method (ISI 1981).

For extrapolating the highest concentration that can be achieved in RO process, three mathematical models, namely linear, exponential and semi-log were tried. The semi-log form with the following equation fitted best in the present case.

$Y = 187.6043 - 51.3292 \log X$ (3.5655) $r^2 = 0.9801$

(value in the parenthesis indicates the SE of regression parameter)

Results and Discussion

Flux behaviour : Fig. 1 shows semi-logarithmic

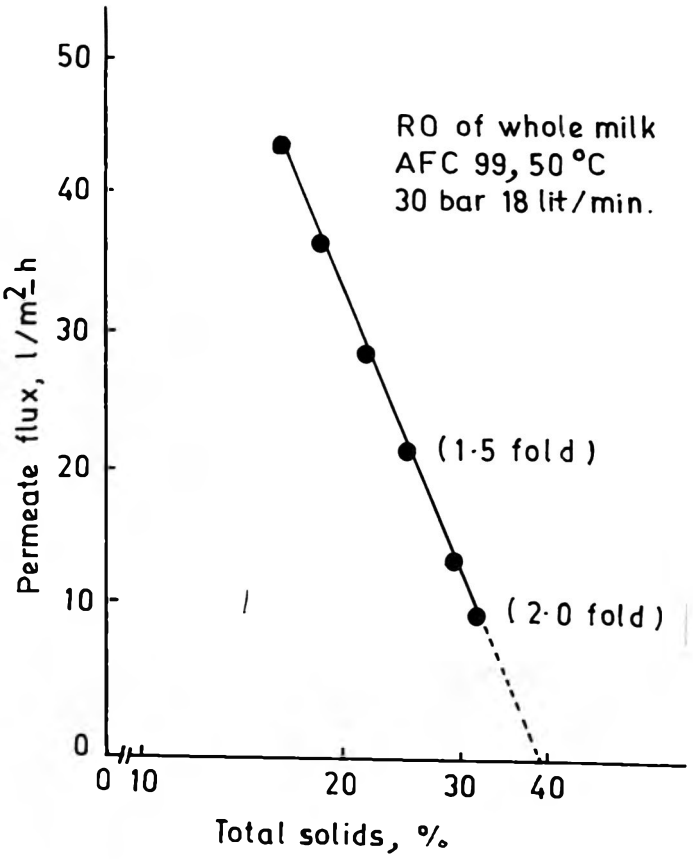

Fig. 1. Influence of concentration of the permeate flux during reverse osmosis processing of buffalo milk.

relationship between the permeate flux and total solids during processing of buffalo milk by RO. The initial flux for buffalo milk (about 16.5% total solids) was 41.6 l/m²-h. On concentration upto 25 (1.5 X) and 32.9% (2.0 X) total solids, the flux declined to 22.6 and 10.6 l/m²-h respectively, while the corresponding average flux was 30.4 and 23.8 l/m²-h. The decrease in flux during membrane processing of fluid foods is a normal phenomenon, and can be attributed to the increase in osmotic pressure of the retained solutes (Pal and Cheryan 1987), concentration polarization of solutes on the membrane, the consequent formation of associated boundary layer (Cheryan 1986), and fouling of the membrane due to precipitation of milk solids on the surface and within the pores of the membrane (Hiddink et al. 1960; Cheryan 1986). The average flux obtained in the present study was exactly the same as that reported by Cheryan et al (1990) for the same type of membrane, but significantly higher than that observed for spiral wound cellulose acetate (CA) membrane, which is operated at much lower temperature (Pal and Cheryan 1967). By extrapolation, it was found that maximum concentration of whole milk that could be achieved by RO process was 38.7% TS with

TABLE 1. GROSS CHEMICAL COMPOSITION OF BUFFALO MILK CONCENTRATED BY REVERSE OSMOSIS PROCESS

Constituents %	Buffalo whole milk	RO concentrated buffalo milks 1.5 X	2.0 X
Total solids	16.57 ± 0.13	24.98 ± 0.39	32.88 ± 0.18
Fat	6.05 ± 0.05	9.10 ± 0.05	12.05 ± 0.05
Proteins	4.60 ± 0.14	6.90 ± 0.15	9.12 ± 0.16
Ash	0.68 ± 0.05	1.04 ± 0.02	1.35 ± 0.02
Lactose	5.28 ± 0.10	7.85 ± 0.05	10.43 ± 0.05

Mean values ± SD based on three trials

X represents concentration factor, i.e. $\frac{\text{initial weight of milk}}{\text{weight of retentate}}$

confidence limits at 95%, varying between -61.23 and -41.43.

Rejection properties : The average composition of normal buffalo milk is presented in Table 1. During concentration by RO process, the individual constituents, i.e, total solids, fat, total proteins, ash and lactose, increased by about the same level as the concentration factor, i.e. 1.5 and 2.0. times. This can be translated into 100% rejection of all the major milk components by RO membrane, and no loss into the permeate. This trend was also confirmed by the analysis of permeate after different intervals. At no stage, the permeate was found to contain any solids. Cheryan et al (1980) and Marshall (1985) have also reported 100% rejection of major milk components by the composite RO membrane. Contrary to these, some other workers (Barbano et al. 1983; Pal and Cheryan 1987) observed the loss of lactose, ash and non-protein nitrogen upto 5% into permeate through the CA membrane, particularly at concentration factor of greater than 2.7. The maximum degree of concentration achieved in the present case was 2-fold. The thin film composite membrane has been recognized for better rejection properties as compared to CA membranes (Marshall 1985).

Effect of RO on the physico-chemical properties : Table 2 shows the effect of RO on titratable acidity, pH, alcohol test, clot-on-boiling (COB) test, viscosity, free fatty acids (FFA), free fat, and fat globule size. The titratable acidity increased and pH decreased for both 1.5X and 2.0X concentrated milk over that of normal buffalo milk. The increase in acidity was, however, not in the same proportion as the concentration factor, though all the major milk constituents, particularly those that contribute to the titratable acidity, increased in the same order

TABLE 2. INFLUENCE OF RO ON PHYSICO-CHEMICAL AND MICROBIOLOGICAL CHARACTERISTICS OF BUFFALO MILK

Characteristics %	Buffalo whole milk	RO concentrated milks 1.5 X	2.0 X
Titratable acidity % lactic acid	0.18 ± 0.05	0.34 ± 0.01	0.40 ± 0.01
pH	6.73 ± 0.05	6.58 ± 0.06	6.46 ± 0.06
Viscosity, cpu at 20°C	2.38 ± 0.06	6.49 ± 0.08	24.82 ± 0.40
Free fatty acids, µeq/g	1.82 ± 0.06	3.02 ± 0.06	3.20 ± 0.01
Free fat, % of total fat	6.78 ± 0.12	1.00 ± 0.01	0.60 ± 0.08
MBR test, h	3.50 ± 0.25	4.20 ± 0.25	4.18 ± 0.25
Cfu x 10^5/ml	18.54 ± 1.2	23.23 ± 0.8	24.71 ± 2.1
Fat globule size, µ	5.7 - 11.4	3.0 - 5.8	1.9 - 2.5
Flavour score #	43.72 ± 0.11	43.55 ± 0.28	43.22 ± 0.19

Alcohol and Clot-on-boiling test were negative in all cases
Mean values ± SD of three trials
Flavour score is based on 45 perfect score

as the concentration level. From these trends, it could be inferred that there was no development of acidity during the RO process, and some of the lactic acid would have passed into the permeate, particularly at higher levels of concentration. The alcohol test for normal milk and 1.5-fold concentrated milk was negative, indicating that the latter could be safely processed at high heat treatments. On the contrary, the 2-fold concentrated milk showed positive alcohol test. This is in keeping with other concentration processes. Concentration of milk to high solid levels by any other process also lowers its heat stability. These results, however, suggest that if RO concentrated buffalo milk (TS 30% and above) is to be used for high heat treatments, such as sterilization, its heat stability should be corrected. The COB test for control as well as both 1.5 and 2.0 - folds concentrates was found to be negative, indicating their suitability for pasteurization.

The viscosity of buffalo milk was 2.36 cpu at 20°C, which increased by almost 3 and 10 - folds in 1.5X and 2.0X concentrated milk, respectively. Such high increase in viscosity could be attributed to the combined effect of increased concentration of milk constituents, particularly proteins and lipids, and homogenization-like action during RO

processing. Versteeg (1985) has also reported similar effect of RO on the viscosity of milk.

Earlier workers (Abbot et al. 1979; Barbano et al. 1983; Pal and Cheryan 1987) had reported that RO processing of raw milk led to lipolysis and increase in FFA due to the action of lipase. To overcome this problem, the lipase was inactivated by pre-heating buffalo milk to 60°C before RO processing in the present study, and expected results were obtained. The FFA in buffalo milk was 1.8 µg/g, which increased to 3.02 and 3.20 on 1.5 X and 2.0 X levels of concentration, respectively. It means that the increase was even not of the same order as the concentration factor, probably due to some permeation of FFA through the membrane.

The microscopic examination of the fat globules revealed that there was homogenization-like action during RO processing of buffalo milk. The size of

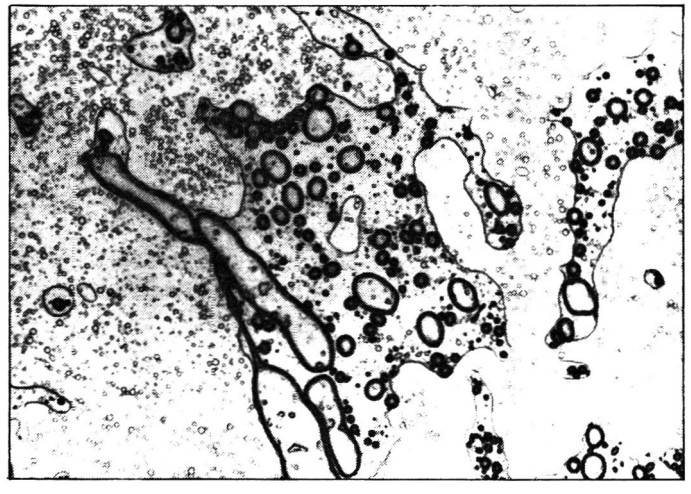

Fig 2 a. Fat globules of normal buffalo milk (Magnification 10 X)

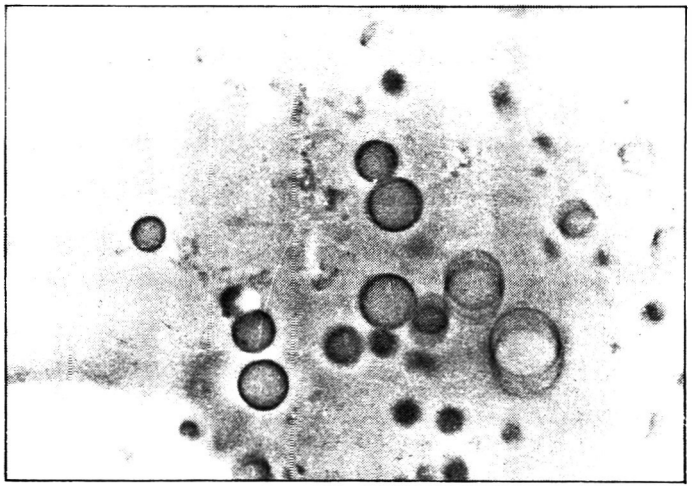

Fig.2 b. Fat globules of RO buffalo milk. (Magnification 100 X)

fat globules in normal buffalo milk ranged between 5.7 and 11.4 µ (Table 2). On RO processing, it reduced to less than 2.5 µ in 2.0 X RO concentrates. The RO process also altered the shape of the fat globules. The irregular-shaped fat globules (Fig. 2a) became uniform and spherical shaped after RO process (Fig. 2b). Contrary to these results, Versteeg (1985) did not find any reduction in the size of the fat globules after RO concentration. As far as shape was concerned, he observed both irregular and round-shaped fat globules. The free fat content also substantially reduced after RO processing. In normal buffalo mlik, the free fat content was 6.78% of total fat, which reduced to 1 and 0.6% in 1.5 X and 2.0 X RO milk (Table 2). The recirculation of buffalo milk for a long period at high operating pressure during RO process could be mainly responsible for decrease in size of fat globules and reduction in free fat content. The cooling of retentate to less than 10°C before its release from the pressure release valve has been reported to help minimizing fat damage (Barbano et al. 1983; Versteeg 1985). The RO unit used in present investigation did not have such provision.

Microbiological aspect of RO milk : The initial standard plate count in thermized buffalo milk was 18.54 x 10^5/ml. Since RO membranes are completely impermeable to microbial cells, theoretically the SPC should increase in the same proportion as the concentration factor. But, this was not so. The SPC (Table 2) in 1.5 x RO concentrates were 23.23 x 10^5/ml (1.3 - fold increase) and in 2.0 X RO 24.7 x 10^5/ml (1.4 - fold increase). It can be interpreted from these results that at a processing temperature of 50°C, there was decrease in bacterial population. This lethal effect increased with the increase of operational time as in case of 2.0 X RO milk. Drew and Manners (1985) also observed substantial decrease in bacterial population of RO concentrated milk with process temperature of 50-55°C, and ascribed this to the destruction of heat sensitive psychrotrophic Gram negative bacteria. Despite higher number of SPC in RO milk in comparison with the normal buffalo milk, higher MBR time was observed for the former, which could be due to lower bacterial activity in concentrated milk.

Sensory quality of RO milk : It is seen from Table 2 that the flavour scores of RO milks did not differ appreciably from those of normal buffalo milk. No abnormal or off-flavour was perceived in RO milks, but they were found to be sweeter in taste, obviously because of higher lactose content,

TABLE 3. EFFECT OF RO ON SKIMMING EFFICIENCY OF BUFFALO MILK

Type of milk	Fat percentage	
	Before separation (milk)	After separation (skim)
Buffalo milk	6.0	0.1
1.5 X RO milk	9.1	7.0
1.5 X diluted RO milk	6.0	3.8
2.0 X RO milk	12.0	11.4
2.0 X diluted RO milk	6.0	4.8

and hence some of the panelists awarded less score on that account. However, this difference could not persist on reconstitution.

Skimming efficiency of RO concentrates : The RO concentrates as such and after dilution to the composition of normal buffalo milk, were separated at 40°C in a cream separator. The fat percentage of milk before and after separation is given in Table 3. For efficient separation, skim milk should be completely devoid of fat content. This was not the case with the RO milks. The skimming efficiency of RO concentrates was very poor, because more than 90% of the total fat lost into skim milk. In case of diluted RO milks, the loss of fat in skim milk was more than 50%. The decrease in size of fat globules during RO processing could be entirely responsible for the poor skimming efficiency. Cream separation difficulties in homogenized milk have also been reported by Brunner (1974). But other workers (Zall 1984 Dixon 1985) did not face any difficulty in separation of RO milk, and they observed less than 0.1% fat losses in skim milk. As discussed earlier, they used different RO module and operating conditions, to minimize damage to the fat globules. Besides, the cream separator used in the present case had 3500 rpm as against the 7000 rpm of commercial cream separator used in the earlier studies. The homogenization effect during RO has its own advantages, such as no separation of fat during transportation and elimination of subsequent homogenization of fluid milk. However, its effect on the quality of dairy products made from such milk needs thorough investigation.

Conclusion

The application of reverse osmosis for the concentration of buffalo milk upto 2-fold level is technologically and economically viable. It retains all the milk constituents in the concentrate and does not alter the organoleptic properties of milk. The homogenization-like action exerted by reverse osmosis should be considered beneficial in fluid milk processing.

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Optimum Operating Time for Cleaning of Cane Sugar Juice Evaporators in Sugar Mills

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Studies were conducted for determining optimum operating time for cleaning of cane sugar juice evaporators in sugar mills. The overall heat transfer coefficient decreased with time of operation of evaporators and increase in incoming juice Brix. The overall heat transfer coefficient and time of operation gave a straightline relationship for both the evaporators under study. At the present operating conditions, the optimum operating time for maximum heat transfer in K_1 and K_3 evaporators was found to be 1800. It is sufficient enough to clean the evaporators only once during the entire cane crushing season of the mill. It is, thus, possible to save nearly 50 tonnes of bagasse and also detergents for cleaning the evaporators, in addition to the increase in the life of the evaporators.

Keywords : Cane sugar juice evaporators, Sugar mills, Cleaning duration, Overall heat transfer coefficients, Energy saving, Heat losses.

In sugar juice evaporators, heat losses occur due to radiation, convection, air leaks, poor vacuum and fouling of heating surfaces. The heat losses through radiation and convection can be reduced by providing proper insulation (Singh 1986). The energy loss due to poor vacuum and air leaks can be minimized by sealing joints. Fouling of evaporator tubes occurs due to deposition of suspended matter

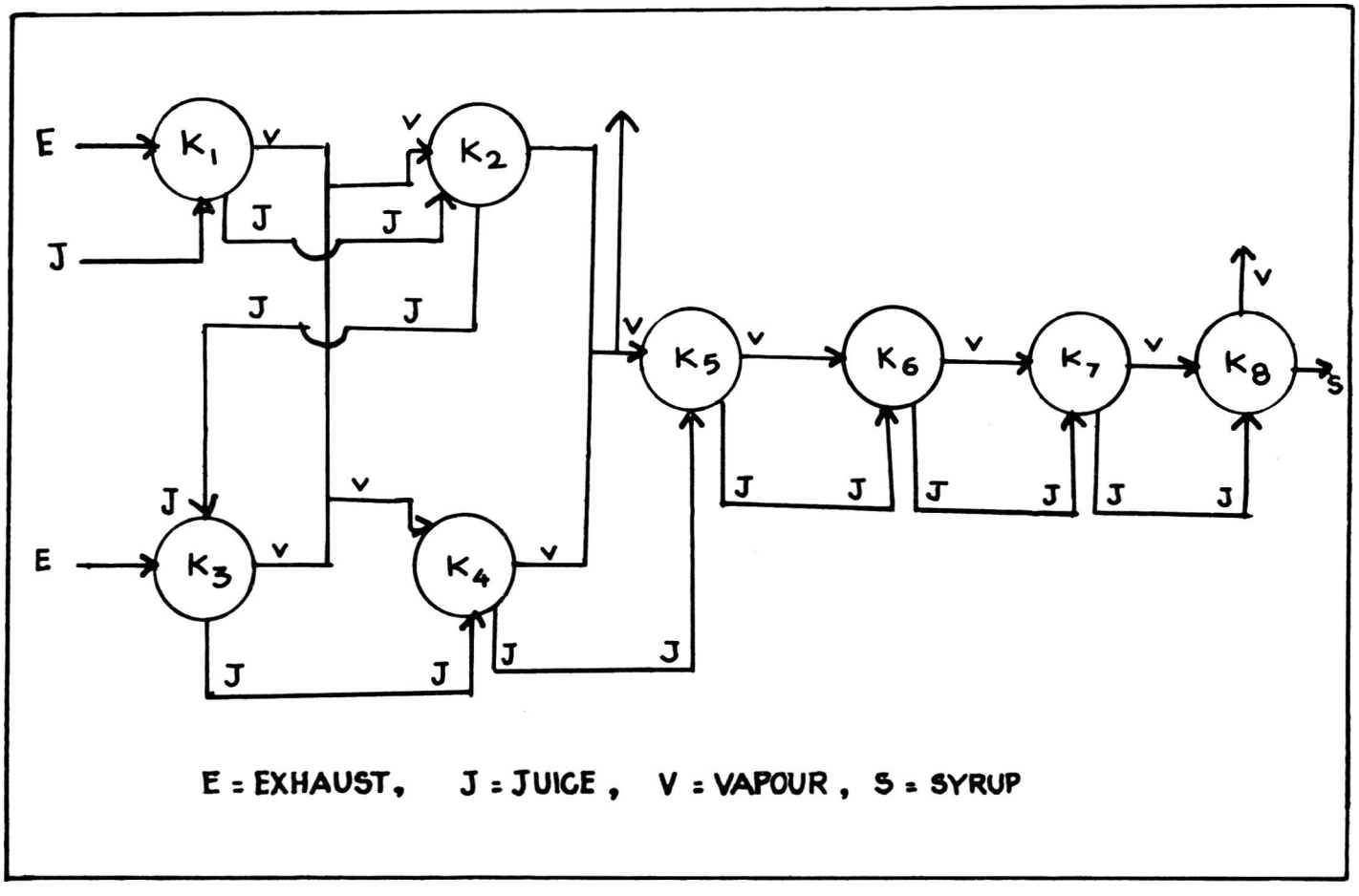

Fig 1. Material flow diagram of juice evaporation.

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on the surface of the tubes, reducing the heat transfer coefficient. It has been estimated that the scale formed in $2^1/_2$ days reduced overall heat transfer coefficient from 1200 to 730 w/m²/C° in multiple effect sugar juice evaporators (Angelletti and Moresi 1983). In sugar mills, it is the normal practice to clean the evaporators after certain number of hours of operation to improve their performance. The number of times the evaporators are cleaned vary between 2 and 4 during the entire crushing period of the mill. At the time of cleaning of evaporators, all other operations such as milling, pan boiling, steam production are also stopped and hence there will be no production of sugar. For restarting the mill after cleaning, the boilers are first started and considerable amount of firewood or bagasse are burnt to bring the steam to the desired pressure. For a given evaporator, the heat transfer rate depends on heat transfer coefficient of evaporator tubes which, in turn, depends upon cleanliness of the tubes, besides wall thickness and material of construction.

In the present study, data were collected in actual working of evaporators of a sugar mill to study the variation of overall heat transfer coefficient with operating time and subsequently, the number of times the evaporators have to be cleaned for obtaining maximum heat transfer was estimated.

Materials and Methods

The study was conducted at M/s. Dhampur Sugar Mills, Dhampur, Uttar Pradesh, India. The juice evaporation system and material flow are shown in Fig.1. The evaporation system is a hextuple effect with 8 evaporators having forward feed system. Evaporators K_1, K_2, K_3, K_4 and K_5 were Kestners and the remaining were rising film type. Exhaust steam from turbines at an average gauge pressure and temperature of 1.15 kg/cm² and 118°C respectively is fed to K_1 and K_2. Part of the vapour from these two evaporators is bled and fed to pans B and C and the balance to evaporators K_2 and K_4. Similarly, the vapour of K_2 and K_4 are bled at A pans before feeding to evaporator K_5.

Mass and energy balance : The mass and energy balance diagram of evaporators is shown in Fig.2. The following mass and energy balance equations were used for evaluating the performance of the evaporators. (Rumsey and Scott 1982).

Overall mass balance

$$L_i = V_i + L_{(i+1)} \quad \ldots (1)$$

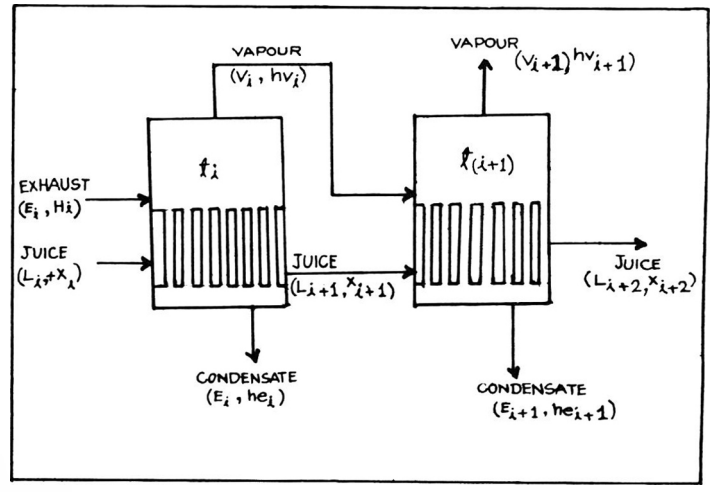

Fig. 2. Mass and energy balance around the evaporators.

Solid mass balance

$$L_i \; X_i = L_{(i+1)} \; . \; X_{(i+1)} \quad \ldots (2)$$

Steam side energy balance

$$E_i \; . \; H_{(i)} = Q_i + e_i . h_{(i)} \quad \ldots (3)$$

Product side energy balance

$$L_i . h_{(ti,\, xi)} + Q_i = V_i . H_{(Ti)} + L_{(i+1)} . h[(ti+1),\, (xi+1)] \quad \ldots (4)$$

Heat transferrate

$$Q_i = U_i \, A_i \; (T_i - t_i) \quad \ldots (5)$$

Where L_i = juice flow rate into ith effect, T/h; V_i = vapour flow from ith effect, T/h; X_i = Brix of juice entering ith effect, %. E_i = exhaust steam flow into ith effect, T/h.e_i = condensate flow from ith effect, T/h.H_i = enthalpy of exhaust from turbines, MJ/kg.; Q_i = heat transferred rate from exhaust steam to juice, MJ/h.; h_i = enthalpy of condensate, MJ/kg.; $h_{(t,x)}$ = enthalpy of juice, MJ/kg. U_i = overall heat transfer coefficient, MJ/m²/h/°C; A_i = area of heat transfer, m². ; T_i = temperature of exhaust steam, C°; and t_i = vapour space temperature, C°.

In equation (4), the enthalpy of juice ($h_{t,x}$) is calculated by equation.

$$h_{(t,x)} = C_{p(x)} \; [t_i - t_{ref.}] \quad \ldots (6)$$

Where

$C_{p(x)}$ = specific heat of juice, MJ/kg/°C.

t_i = temperature of juice, °C.

$t_{ref.}$ = reference temperature, °C

($t_{ref.}$ is taken as 0°C).

In equation (6), the specific heat of juice ($C_{p(x)}$) is calculated by the following equation (Hugot and Jumkins 1986).

$$C_{p(x)} = (1 - 0.006.\ x) \quad \ldots (7)$$

Where

x = Brix of the juice, %.

The temperature of the juice coming out of the evaporator is calculated by summing temperature of the vapour space to the boiling point rise of the juice. The boiling point rise of the juice was calculated by the equation (Angeletti 1983).

$$B_{pr} = 3.2.\ x - 2.42\ .\ x^2 + 14\ .\ x^3 \quad \ldots (8)$$

Where

x = Brix of the juice, %.

In the above equations (eq. 1 to 5), the variables which were measured and those calculated are shown in Table 1.

TABLE 1. MEASURED VARIABLES AND CALCULATED VARIABLES

	Measured	Calculated	Reference table
Juice flow in (T/h)	✓		
Brix of the entering juice (%)	✓		
Brix of the leaving juice (%)	✓		
Enthalpy of exhaust, (MJ/kg)			✓
Enthalpy of vapour, (MJ/kg)			✓
Heating surface (m^2)	✓		
Temperature and pressure of exhaust, (°C, kg/cm^2), respectively.	✓		
Vapour space pressure (kg/cm^2)	✓		
Vapour space temperature, °C		✓	
Vapour flow rate, T/h)		✓	
Juice flow out, (T/h)		✓	
Exhaust of vapour flown into the effect (T/h)		✓	
Heat transfer rate (MJ/h)		✓	
Overall heat transfer co-efficient $MJ/m^2/h/°C$		✓	

The measured variables listed in Table 1 are recorded for evaporators K_1 and K_3 at 1 h interval for a duration of 8 h in the period before the mill was stopped for first cleaning. The mill was stopped for first cleaning, after 6 weeks of operation (1010 h). By making use of weekly average values of measured variables, the calculated variables were obtained. Since K_1 and K_3 are the evaporators which were supplied with exhaust steam, the economy of this exhaust steam depends upon performance of these two bodies. Therefore, the measured variables were recorded only for these two evaporators.

Relationship between operating time of evaporators and overall heat transfer coefficient: A relationship was obtained between overall heat transfer coefficient and time of operation of evaporators for both K_1 and K_3 evaporators. The relationship so obtained was further used for estimating the total heat transferred during operating time of evaporators.

Estimation of total quantity of heat transferred : If Q represents the total amount of heat transferred in the operating time θ_b and A and Δt represent, respectively, the heat transfer area and temperature difference. The rate of heat transfer at any instant from exhaust steam to juice in the evaporators can be represented by the following equation.

$$\frac{dQ}{d\theta_b} = U.A.\ \Delta t \quad \ldots (9)$$

Where

$\frac{dQ}{d\theta_b}$ = heat transfer rate, MJ/h

U = overall heat transfer coefficient, $MJ/m^2/h/°C$

A = heat transfer area, m^2

Δt = temperature difference between exhaust steam and juice, °C

θ_b = juice boiling time, h.

The total heat transferred during any interval of time was obtained by substituting the value of U in terms of θ_b in equation (9), and integrating the resulting equation between the limits 0 and Q and 0 and θ_b

$$Q = \int_0^Q dQ = \int_0^{\theta_b} UA\ \Delta t\ d\theta_b \quad \ldots (10)$$

Substituting the value of U in terms of θ_b and integrating the solution of equation (10) result in equation of the following form :

$$Q = \frac{A\ \Delta t}{2} (S_1\ \theta_b - S_2\ \theta_b^2) \quad \ldots (11)$$

Where S_1 and S_2 are constants.

In the above equation, the values of A and Δt were constant for a particular evaporator throughout the operating period. A graph was drawn between variables Q and θ_b and the value of θ_b for maximum Q was selected frcm this graph.

Estimation of number of cleanings of evaporators : The number of times the evaporators have to be cleaned was calculated by the following formula (Perry and Dongreen 1984)

$$N_c = N_s - 1 \quad \ldots (12)$$

Where

N_c = number of cleanings.

N_s = number of cleaning cycles.

The number of cleaning cyles (N_s) was estimated by the following formula (Perry 1984)

$$N_s = \frac{H}{\theta_b + \theta_c} \quad \ldots (13)$$

Where

H = number of hours the mill was operated during the crushing season.

θ_b = boiling time for maximum heat transfer, h

θ_c = time required for emptying, cleaning and recharging of evaporator, h.

Results and Discussion

Tables 2 and 3 show the weekly average values of measured and calculated variables for evaporators K_1 and K_3. It is evident that the overall heat transfer coefficient decreased with time of operation. In spite of operating at the same temperature and pressure, the overall heat transfer coefficient was lower for K_3 evaporator than that for K_1 during the complete period of operation. This is due to increased concentration of juice entering into K_3 evaporator. The linear regression between these two variables for both the evaporators resulted in a straightline

TABLE 2. MEASURED VARIABLES FOR EVAPORATORS K_1 AND K_3

Evaporator	Variable	1st week (168 h)	2nd week (336 h)	3rd week (504 h)	4th week (672 h)	5th week (840 h)	6th week (1008 h)
K_1	Flow rate of incoming juice, kg/h	19.3 x 10^4	20.64 x 10^4	21.8 x 10^4	21.35 x 10^4	21.15 x 10^4	19.81 x 10^4
	Brix of incoming juice %	14.10	14.35	14.86	14.88	15.33	14.94
	Brix of outgoing juice %	18.70	18.55	18.70	18.75	18.81	18.67
	Pressure of exhaust steam kg/cm² (g)	1.62	1.66	1.72	1.75	1.79	1.75
	Temperature of exhaust steam, °C	113.2	113.81	114.89	115.55	116.20	115.58
	Vapour space pressure K/cm² (g)	1.43	1.45	1.45	1.45	1.44	1.45
	Vapour space temperature, °C	109.60	109.75	109.80	109.80	109.98	109.30
K_3	Flow rate of incoming juice, kg/h	11.78 x 10^4	13.17 x 10^4	14.26 x 10^4	14.05 x 10^4	14.38 x 10^4	13.04 x 10^4
	Brix of incoming juice, kg/h	23.1	22.5	22.74	22.60	22.50	22.8
	Brix of outgoing juice, %	28.0	30.7	28.50	28.45	28.75	28.3

Surface area of evaporator K_1 2044 m², Surface area of evaporator K_2 1607 M²

TABLE 3. CALCULATED VARIABLES FOR EVAPORATORS K_1 AND K_3

Evaporator	Variable	1st week (168 h)	2nd week (336 h)	3rd week (504 h)	4th week (672 h)	5th week (840 h)	6th week (1008 h)
K_1	Vapour flow rate, kg/h	4.7×10^4	4.67×10^4	4.48×10^4	4.40×10^4	3.91×10^4	3.90×10^4
	Outgoing juice flow rate, kg/h x 10^4	14.55	15.96	17.41	16.94	17.24	15.93
	Exhaust supply rate, kg/h x 10^5	49.10	49.25	48.98	42.90	43.30	41.36
	Heat transfer rate, MJ/h x 10^4	11.00	10.90	10.82	9.48	9.54	9.14
	Boiling point raise (°C)	0.605	0.605	0.605	0.605	0.605	0.605
	Overall heat transfer coefficient, MJ/hr/m²/°C	14.95	13.14	10.36	7.83	7.14	7.73
K_3	Vapour flow rate, kg/h	2.06×10^4	2.77	2.9×10^4	2.88×10^4	3.12×10^4	2.53×10^4
	Outgoing juice flow rate. kg/h x 10^4	9.72	10.40	11.36	11.16	11.25	13.04
	Exhaust supply rate, kg/h x 10^5	22.30	29.80	31.54	31.20	33.70	27.64
	Heat transfer rate, MJ/h x 10^4	7.40	6.60	6.97	6.89	7.45	6.10
	Boiling point raise, (°C)	1.01	1.01	1.01	1.01	1.01	1.01
	Overall heat transfer coefficient, MJ/h/m²/°C	12.01	10.1	8.48	7.49	7.09	6.57
	Enthalphy of exhaust steam,		= 2.69 MJ/kg				
	Enthalpy of vapour		= 0.476 MJ/kg				

with correlation coefficient of more than 0.96. The actual relationships obtained are as follows :

$$UK_1 = 16.53 - 0.0153\ \theta_{b1} \quad \ldots (14)$$

$$UK_2 = 11.01 - 0.0058\ \theta_{b2} \quad \ldots (15)$$

Where

Uk_1 , UK_2 = overall heat transfer coefficient, MJ/m²/°C/h

θ_{b1} and θ_{b2} = boiling time of juice in the evaporators, h. After substituting the values of UK_1 and UK_2, the basic heat transfer equation for these two evaporators can be written as follows :

$$\frac{dQ_1}{d\ \theta_{b1}} = A_1\ \Delta t_1\ (16.53 - 0.01533\ \theta_{b1}) \quad \ldots (16)$$

$$\frac{dQ_2}{d\ \theta_{b2}} = A_1\ \Delta t_2\ (11.01 - 0.0058\ \theta_{b2}) \quad \ldots (17)$$

The above equations (eq. 16 and eq.17) were integrated with the limits to find the total heat transferred during the operating time of evaporators.

$$\int_0^{Q_1} dQ_1 = A_1\ \Delta t_1 \int_0^{\theta_{b1}} (11.01 - 0.0058\ \theta_{b1})\ d\theta_{b1} \quad \ldots (18)$$

$$\int_0^{Q_2} dQ_2 = A_2\ \Delta t_2 \int_0^{\theta_{b2}} (16.53 - 0.01533\ \theta_{b2})\ d\theta_{b2} \quad \ldots (19)$$

The temperature difference Δt was constant for both the evaporators during the period of study. The total heat transferred was obtained by solving the above equations (eq. 18 and 19) and is as follows :

$$Q_1 = \frac{A_1\ \Delta t_1}{2} (33.06\ \theta_{b1} - 0.015\ \theta^2_{b1}) \quad \ldots (20)$$

$$Q_2 = \frac{A_2\ \Delta t_2}{2} (22.02\ \theta_{b2} - 0.006\ \theta^2_{b2}) \quad \ldots (21)$$

Fig.3 is the graph between θ and θb obtained by calculating the values of θ for different values of Ob, using equations 20 and 21. From the graph,

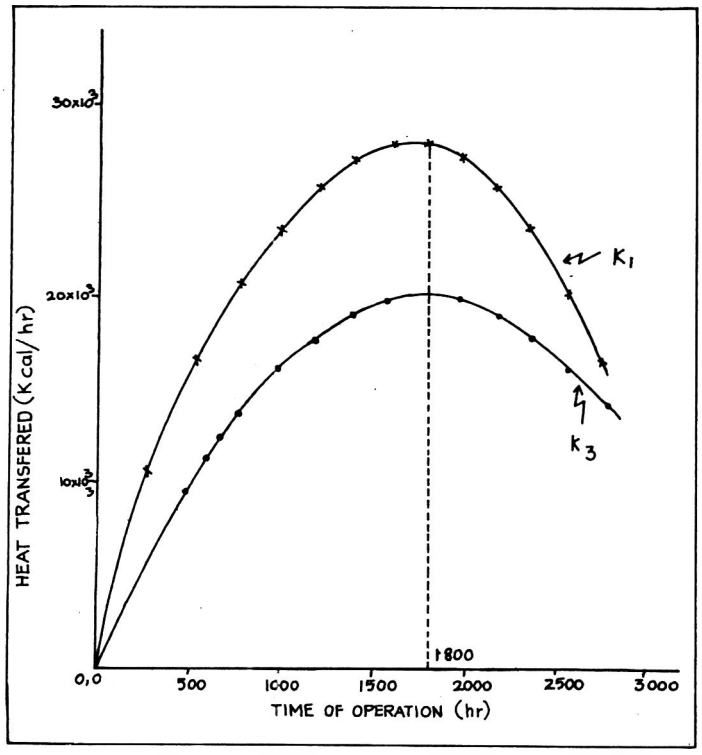

Fig. 3. Relationship between time of operation and total heat transferred

it is clear that the optimum time of operation of evaporators K_1 and K_3 for maximum heat transfer was 1800 h. The time required for emptying, cleaning and recharging was 36 h (Mill records). The mill worked for 3360 h during the entire cane crushing season. Therefore, the number of cycles was 3360/(1800 + 36) = 1.80. The number of times the evaporators has to be cleaned will be 1.80 – 1 =0.8. Since the figure (0.8) is less than 1.0. it is sufficient enough to clean the evaporators only once during the entire crushing season of the mill.

During the crushing season of 1989-90, the mill was stopped twice for cleaning, first time after 1010 h of operation and second time after 2100 h of operation. Whenever the mill was stopped for cleaning, it is a general practice to clean all the other machines including boilers, mills and pans at the same time. For restarting the boilers after each cleaning, it required, on an average, 50 tonnes of firewood or bagasse.

Since the above analysis of data collected has shown that it is sufficient to clean the evaporators only once, a saving of 50 tonnes of bagasse can be achieved. The detergents, chemicals etc., used in the cleaning of evaporators also can be saved. In addition, the life of the evaporator tubes can be increased due to reduced frequency of cleaning.

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Effect of Additives on Liquid - Solid Transformation in Coconut Oil

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The effect of admul, myverol and lecithin on the nucleation in coconut oil was studied through the measurement of nucleation temperature; light scattering method was employed for measurement of nucleation temperature. Lecithin and myverol were found to be effective in suppressing the nucleation to different levels. The mechanisms involved are discussed.

Keywords : Coconut oil, Additives, Nucleation, Light scattering, Inhibition, Molecular organization, Solidification.

Garti (1988) discussed the effect of several food emulsifiers on the transition kinetics of polymorphic modifications of stearic acid and triglycerides. The study revealed that the effectiveness to induce the crystal structure modifications depended on the structure of emulsifiers; surfactants with long chains and bulky hydrophilic groups were active in modifying the polymorphism.

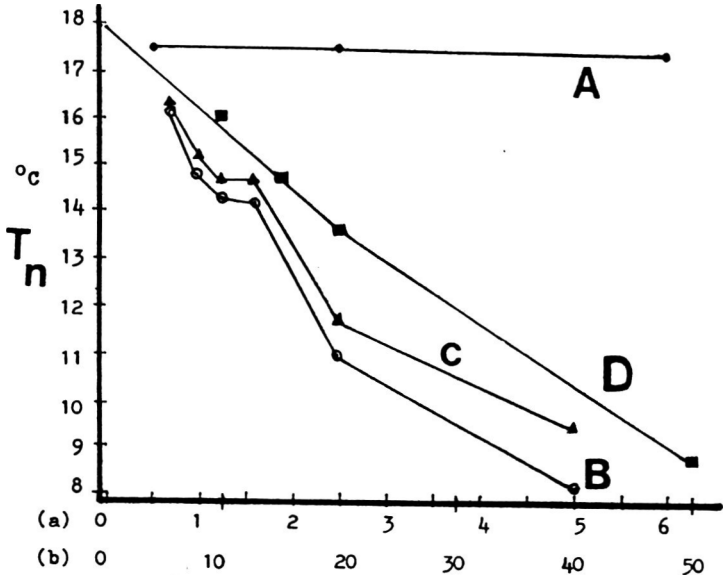
The present study was undertaken to examine the influence of certain additives (emulsifiers) on the solidification of coconut oil, via the measurement of nucleation temperature. This study is on the liquid-solid transformation and in contrast to some of the earlier studies on the polymorphic modifications in crystals present in solutions (Garti et al. 1982a; Garti and Sato 1986).

Coconut oil used had the solidification range 18.1-17.9°C, nucleation occurring at 18.1°C, and solidification getting completed at 17.9°C. Chief constituents of coconut oil (Rose and Rose 1961) are the glycerides of lauric acid, but with appreciable amounts of the glycerides of capric, myristic, palmitic and oleic acids. Cocks and Van Rede (1966) gave the following fatty acid composition (expressed as weight % of total fatty acids): C_{10} and lower : 12.5-13%. C_{12} : 47-50%. C_{14} : 19%. C_{16} : 7-8%. C_{18} : 2-3%. C_{20} and higher : 1%. C_{18} (oleic) : 5.5-8%. C_{18} (linoleic) : 1-1.5%.

High purity lecithin, admul and myverol were used as additives. Lecithin and monoglycerides are known for their activity as viscosity controllers and anti-bloom agents (Garti et al. 1982b).

The nucleation temperature of coconut oil in presence of additives was determined by employing a light scattering method (Prasad 1992). Light scattering is a unique technique for *in situ* studies of processes in a small volume (Steininger and Bilgram 1991). The present method consists of measuring the intensity of scattered light (by a liquid solution) at 90° to the incident beam. The intensity of the scattered light depends on the state of the liquid (Prasad 1992). Thus, a continuous recording of intensity of the scattered light versus temperature of the liquid gives characteristic curves. The nucleation temperature can be determined, in addition to the other data, from such curves.

Admul (polyglycerol ester of polyrecinoleate) : It is a viscous liquid at room temperature. The data shown in Fig.1 (curve A) indicate that admul has



(a) % of additive concentration.
(b) % of MEK in coconut oil.

Fig.1. Nucleation temperature (T_n) vs percentage of additives in cocount oil. A : admul, B : lecithin, C : Myverol, D: methyl ethyl ketone

inappreciable influence on the nucleation temperature (T_n) of coconut oil. For all proportions of admul employed, the suppression in nucleation temperature (T_{sup}) was about 0.5°C.

Lecithin (phospholipid derived from soya beans) : It has extremely low solubility in cocount oil. However, it has appreciable solubility in methyl ethyl ketone (MEK). Therefore, a standard solution of lecithin in (analytical grade) MEK was made (50 mg lecithin in 0.4 ml MEK), and coconut oil was added to 0.2 ml of solution, stepwise. For each step T_n was determined. The T_n of coconut oil-lecithin-MEK system is shown by curve B in Fig.1. It was observed that, with the ageing of the solution, lecithin coagulated and precipitated; such a situation could lead to different values of T_n.

Myverol : It is a monoglyceride which exists in solid form at room temperature and dissolves slowly in coconut oil. To make the comparison easy and rational, a solution of myverol in MEK (similar to lecithin) was used to test the influence of myverol on T_n. Curve C (Fig.1) shows the results.

The influence of MEK on T_n of coconut oil (due to dilution) was separately determined (curve D in Fig.1).

It may be noticed that curves B and C are below curve D, which is a clear indication that lecithin and myverol are able to suppress the T_n. Interestingly, the curves B and C converge almost to a single point at higher dilutions of additives.

The differences between curves D and B or curves D and C are assumed to give respective T_{sup} at corresponding additive concentration. It is seen that T_{sup} at 5% lecithin and 5% myverol are 2.2°C and 1°C, respectively.

The influence of an inhibitor can be either by way of interfering in the process of formation of nuclei or by slowing down the growth rate of nuclei into bigger particles (Nancollas and Zawacki 1984). From the molecular structure of the three additives that are used in the present study and their unequal influence on the T_n, it appears, in case of the present system, that primarily a greater similarity between additive and host molecule is essential. The situation is close to the case of solid solutions and solid solubility in organic systems, being discussed by Kitaigorodski (1984). The second aspect is the presence of a bulky group at one end of the additive molecule and such a bulky group essentially should be dissimilar to the host molecules. It appears that presence of such a bulky group destroys the possibility of a large number of molecules grouping together to help further growth of nucleus. Evidently, the van der Walls bonds can be formed only when a close approach is achieved between the molecules, leading to molecular organization.

The two criteria, mentioned above, match with the requirements reported by Garti (1988). It can, therefore, be stated that more or less, the same requirements are to be satisfied in case of inhibition of nucleation for liquid-solid transformation in case of oils and for the polymorphic transformations in case of fatty acid crystals in solution.

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Effect of Iron-Enriched Baked Products on Body Weights and Haemoglobin Levels of Slightly Anaemic and Normal Subjects

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Eleven varieties of iron-enriched baked products were developed by incorporating iron-rich foods, besides ferrous sulphate at the level of 7.5 mg of iron/100 g of product to provide 75% recommended dietary allowance upon use as supplement for food. Results of the supplementation of the enriched products in the diet of 35 each of anaemic and normal subjects for two months showed equal increase in body weight of these subjects. Greater increases in haemoglobin levels of anaemic subjects, than the normal subjects, were observed. All the enriched baked products were found acceptable to the normal and anaemic subjects.

Keywords : Baked products, Iron, Supplementation, Anaemia, Human study

Iron deficiency anaemia is a serious public health problem, having its impact on psychological and physical development, behaviour and work performance (Vijayalakshmi and Jayanthi 1986). Prevalence of iron deficiency anaemia in India is about 40-60% in school children and 25-40% in women in the reproductive age group (Narasinga Rao 1982). Some positive steps are necessary to cure the anaemic condition. In this context, the baked products enriched with iron can be an efficient mode, especially due to the current popularity of convenience foods.

Eleven enriched baked products were developed (Table1). These involved the use of iron-rich foods like *Shepu* (*Peucedanum graveolens*, local name *Subsige*) and raisins which contain 7.7 to 17.4 mg% iron, in addition to the use of ferrous sulphate tablets (Vaidehi et al. 1991). Nutrients analysed were proteins, fat, energy and iron. Analyses were conducted in duplicate. Protein and fat in enriched products were estimated by Micro-Kjeldahl and Soxhlet extraction methods, respectively (AOAC 1980). Gross energy was estimated by using Bomb calorimeter (Gopal Krishna and Ranjhan 1980). Iron was determined colorimetrically (Raghuramulu 1983). Body weight of the subjects were recorded using standard procedure (Jelliffe 1966). Haemoglobin levels were estimated using cyanmethaemoglobin method (Cannon 1958). Thirty five each of anaemic and normal subjects, in the age group 22 to 35 years, were selected and females were free from any physiological stress conditions. They were served 150 g of any two baked iron enriched products/day for a period of 20 days (Table 1). Student's test was applied and significant differences in the initial and final recordings of the parameters studied were compared (Snedecor and Cochran 1967).

TABLE 1. NUTRITIVE VALUE OF IRON-SUPPLEMENTED BAKED PRODUCTS AT ONE SERVING OF 150 G/ PERSON/DAY

Product	Protein, g	Fat, g	Carbohydrate, g	Energy, K.cal	Iron, mg
Experimental products					
Shepu bars	21.2	24.9	87.6	566	32.3
Ragi rings	24.9	22.8	83.7	597	25.8
Toffee bars	23.9	31.4	69.5	648	22.3
Jaggery cake	22.9	26.7	67.6	603	20.0
Ragi cake	20.9	31.7	71.8	642	22.1
Fruit cake	21.8	25.0	67.5	634	17.3
Pizza	17.5	13.8	49.8	465	22.9
Brown bread	21.7	15.4	64.4	597	20.7
Masala bun	21.2	14.7	69.4	505	25.4
Speciality egg	25.2	14.0	74.0	583	17.9
Enriched puff	20.3	42.5	70.0	686	21.9
Regular/commercial products					
Cookies	12.0	37.0	82.4	583	2.0
Cakes, sponge normal	9.0	18.6	60.0	576	2.4
Bread	9.8	2.5	60.6	600	NA
RDA (ICMR) per day*					
Man	60	20	-	3800	28.0
Woman	50	20	-	225	30.0
Children :					
Boys 16-18 yr	78	22	-	2640	50.0
Girls 16-18 yr	63	22	-	2060	30.0

NA - Not analysed, * - Recommended dietary allowances

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The data on proximate nutrients in 150 g enriched baked products (one serving/individual) indicated that the protein content ranged between 17.5 and 25.2 g/150 g product (Table 1). The fat and energy contents of puffs were the highest. Iron value was the highest in *Shepu* bars and lowest in fruit cake. The increases in weightes of anaemic males and females were higher than those in normal males and females. Student's test revealed no significant difference ($P \leq 0.05$) between normal and anaemic males. Similarly, the overall increases in haemoglobin in anaemic males and females were significantly ($P \leq 0.05$) higher than those in the normal subjects (Table 2). Study conducted on iron by Vagheti *et al* (1979) showed that adult females fed enriched-baked products for two months had a mean increase of 1.14 g, as against 1.07 g per ml increase in haemoglobin level of female subjects observed in the present study, thereby implying that the test products, are of dietetic importance. As most of the children are prone to protein and energy malnutrition and anaemia, the supplementation of these products is of importance in feeding programmes.

TABLE 2. OVERALL INCREASE IN BODY WEIGHT AND HAEMOGLOBIN LEVELS OF SUBJECTS FED IRON ENRICHED PRODUCTS

Subjects		Body weight kg.			Haemoglobin level g/100 ml.		
		Initial	Final	Overall increase	Initial	Final	Overall increase
Females							
Normal	(19)	52.42 ± 1.96	53.47 ± 2.12	1.05	13.15 ± 0.59	13.35 ± 0.48	0.20
Anaemic	(21)	40.45 ± 1.68	41.66 ± 1.87	1.21	10.87 ± 0.51	11.9 ± 0.76	1.07*
Males							
Normal	(19)	64.19 ± 2.40	65.28 ± 2.38	1.09	14.64 ± 0.35	14.86 ± 0.26	0.22
Anaemic	(14)	50.29 ± 2.14	51.71 ± 2.31	1.42*	12.74 ± 0.47	13.83 ± 0.52	1.09*
t-Values		Females	0.95 NS		Females	3.78 NS	
		Males	7.88*		Males	2.33*	

* Significant at 5% level

The data indicate that the iron-enriched baked products could be nutritious snacks. What in practice today, is of administering prophylactic iron capsules and tablets, which could become less practised in the long run. Further, the tablets and capsules provide only synthetic and specific nutrients, but these enriched products provide proteins, fat, calories and iron. Every one enjoys eating food rather than taking tablets. In view of these advantages, the enriched baked products will provide extra nutrients, and improve nutritional status, besides giving satisfaction for hunger.

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Isolation of Ampicillin Sensitive *Aeromonas* from Aquatic Foods Using Xylose-Lysine-Desoxycholate Agar and Ampicillin-Dextrin Agar

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In the present study, 96 fresh waterfishes, 37 marine fishes, 13 fresh water prawns, 13 marine shrimps and 26 molluscs have been screened for *Aeromonas* sp. by plating on xylose-lysine-desoxycholate agar (XLDA) and ampicillin-dextrin agar (ADA), after enrichment in alkaline broth. A total of 13 *Aeromonas* strains were isolated with the use of XLDA; 8 from fresh waterfishes, 4 from molluscs and one from marine shrimp. Among *Aeromonas* isolates, 11 were *A. sobria*, one each was *A. salmonicida* subsp. *salmonicida* and *A. salmonicida* subsp. *masoucida*. With the use of ADA, only 7 samples were positive for *Aeromonas*, while the undetected isolates were the ampicillin-sensitive strains. Among the isolates, 5 were sensitive to all the 10 antibiotics tested, while the sensitivity to other antibiotics varied widely. None of the isolates was resistant to nalidixic acid, oxytetracycline, chloramphenicol, norflox and doxycycline.

Keywords : Ampicillin-sensitive *Aeromonas*, Ampicillin-dextrin agar, Xylose-lysine-desoxycholate agar, *A. sobria*, Aquatic foods.

Aeromonas, a common psychrotroph of contaminated waters, has been reported to cause various diseases in aquatic fauna viz. fish, amphibians, snails and mussels (Bersani et al. 1980; Mead 1959; Rahim et al. 1984; Sanyal et al. 1975; Shott et al. 1972). *Aeromonas* was isolated from contaminated market seafoods, the commonest being *A. sobria*, followed by *A. hydrophila* and *A. salmonicida* (Okfer and Neaako 1985; Tompsett and Fricker 1987; Beri et al. 1989). Some plant extracts isolated from *Adenanthera pavonia* are known to inhibit the trimethylamine oxide production by *aeremonas* (Sachindra and Karunasagar 1989). In the present study, efforts were made to isolate *Aeromonas* spp. from aquatic food samples, available from commercial outlets in India. Xylose-lysine-desoxycholate agar (XLDA, Merck) and ampicillin-dextrin agar (ADA) were used as selective plating media for comparative isolation of *Aeromonas* spp.

Slime samples from gills and surface of various aquatic foods (Table 1), were collected using sterilized cotton swabs. These were immediately transferred to Cary and Blair (Difco) transport medium, stored under ice and brought to laboratory. After

TABLE 1. RESULTS OF ISOLATION OF *AEROMONAS* SP. FROM FISH AND AQUATIC FOODS ON DIFFERENT MEDIA

Area	Type of aquatic food		*Aeromonas* spp. isolated, No. of +ve samples on			
	No.of samples		XLDA Medium		ADA Medium	
Bareilly	Fresh water fish	(78)	*A. sobria*	(6)	*A. sobria*	(2)
	Molluscs	(26)	*A. sobria*	(3)	*A. sobria*	(1)
			A. salmonicida subsp *salmonicida*	(1)	*A. salmonicida* subsp *salmonicida*	(1)
Bijnor	Fresh water fish	(18)	*A. sobria*	(2)	*A. sobria*	(2)
Calcutta	Marine shrimp	(06)	*A. salmonicida* subsp *masoucida*	(1)	*A salmonicida* subsp *masoucida*	(1)

XLDA - Xylose-lysine desoxycholate agar; ADA-Ampicillin-dextrin agar.

Note : Irrespective of market, all the fresh water prawns (13) marine fishes (37), and shrimps (7) from Delhi and Bombay have not been found to be contaminated with *Aeromonas sp.*

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resuscitation in 5 ml of 0.1% peptone water at 37°C for 6-8 h, 1 ml of the fluid was transferred to alkaline enrichment broth (Bacto-peptone 1 g, NaCl 5 g, distilled water 1000 ml, pH 8), tubes and incubated at 37°C overnight. Ampicillin-dextrin agar (Havelaar and Vonk 1988) and XLDA medium (Merck) plates were, then, streaked and incubated at 37°C for 24 h. Large smooth yellow colonies from ADA plates, and oxidase positive yellow opaque colonies surrounded by yellow zone from XLDA plates, were picked up on triple sugar iron agar slant (Merck) and further characterized on the basis of morphological, cultural and biochemical characteristics (Cruickshank 1975). Drug resistance pattern of all isolates of *Aeromonas* was determined by paper disc method of WHO (1961) against streptomycin, ampicillin, nitrofurantoin, nalidixic acid, gentamycin, oxytetracycline, chloramphenicol, norflox, doxycycline and septran. These antibiotics were used at 10 mcg level except for the use of 100 mcg furadontine, 30 mcg each of nalidixic acid and chloramphenicol and 50 mcg of septran.

Out of 13 isolates of *Aeromonas* isolated, 11 belonged to *A. sobria* and one each to *A. salmonicida* subsp. *salmonicida* and *A. salmonicida* subsp. *masoucida* (Table 1). All of the *A. sobria* and *A. salmonicida* subsp *salmonicida* strains were isolated from the fresh water fishes. The single isolate of *A. salmonicida* subsp *masoucida* was from the sea shrimp sample. It is also evident that XLDA medium was superior to ADA for isolation of *Aeromonas*. It led to the isolation of *Aeromonas* from 13 samples as against only 7 from ADA medium.

Data indicated that six strains of *A. sobria* were sensitive to ampicillin and all other antibiotics, except the isolate from fresh waterfish, which was resistant to furadontine (Table 2). Remaining 5 isolates of *A. sobria*, one each of *A. salmonicida* subsp *salmonicida* and *A. salmonicida* subsp *masoucida* were resistant to ampicillin. Out of the 7 ampicillin-resistant isolates, three were resistant to streptomycin, and one each to septran and gentamycin. None of the isolates was resistant to norflox, oxytetracycline. chloramphenicol, nalidixic acid and doxycycline.

The isolation of 13 strains of 3 species of *Aeromonas* is of significance, as these organisms have been reported to cause diseases and death in aquatic fauna as well as the spoilage of stored foods (Rahim et al. 1984; Sanyal et al. 1987).

TABLE 2. ANTIBIOTIC SENSITIVITY PATTERN OF *AEROMONAS* ISOLATES FROM VARIOUS AQUATIC FOODS

Aeromonas sp.	Source (No. of isolated)		Sensitivity to drugs
A. sobria	Fresh water fish	(3)	A, C, DO, F, G, L, Na, NO, S, ST
	Fresh water fish	(1)	A, C, DO, G, L, Na, NO, S, ST
	Molluscs	(2)	A, C, DC, F, G, L, Na, NO, S, ST
	Molluscs	(1)	C, D, O, F, G, L, Na, NO, S,
	Fresh water fish	(2)	C, DO, F, G, L, Na, NO, S, ST
	Fresh water fish	(1)	C, CO, F, G, L, Na, NO, S, ST
	Fresh water fish	(1)	C, CO, F, L, Na, NO, S, ST
A. salmoncida subsp *salmoncida*	Marine shrimp	(1)	C, DO, F, L, Na, No, S
A. salmonicida subsp *masoucida*	Mollusc	(1)	C, DO, F, G, L, Na, NO, S

All the isolates were tested against the following 10 drugs

A - Ampicillin, C - Chloramphenicol, DO - Doxycycline, F - Furadontine, G - Gentamycin, L - Oxytetracycline, Na - Nalidixic acid, NO - Norflox, S - Septran, ST - Streptomycin

Aeromonas is also incriminated in diarrhoeal illness in humans. In USA, 7 outbreaks of *Aeromonas* food poisoning, due to consumption of molluscan foods, have been reported from 1978 to 1987 (Ahmad 1992). *Aeromonas salmonicida* has been reported to transfer drug resistance factor to other enterobacteria of human health significance, such as *E. coli* (Aoki and Egusa 1971; Dutta and Hedges 1973; Popoff and Davaine 1971). Therefore, isolation of streptomycin-resistant *A. salmonicida* subsp *salmonicida* from fresh water molluscs is of significance.

The other significant observation was the inability of ADA medium to allow the isolation of ampicillin-sensitive *A. sobria* from aquatic foods, although it has been reported to be a good medium for enumeration of *Aeromonas* spp. in water samples (Havelaar and Vonk 1988). *Aeromonas* forms non-specific yellow opaque colonies on XLDA plates like some other members of enterobacteriaceae, and could be differentiated by the oxidase positive reaction of *Aeromonas*. Therefore, XLDA can not be used for enumeration of *Aeromonas*, as is possible in case of ADA. However, XLDA has been used successfully for isolation of *Aeromonas*. The isolation of ampicillin-sensitive *A. sobria* from fresh water

fishes and molluscs indicated that these may be present in contaminated fresh waters. However, such water samples would escape from detection of *Aeromonas* spp., if ADA is used for isolation purposes. An elaborate study is, thus, needed to develop selective medium for enumeration of *Aeromonas* in food and water samples.

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Changes in the Quality of Turmeric Rhizomes During Storage

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Both cured and uncured dried rhizomes of four turmeric (*Curcuma longa* L.) cultivars were examined for changes in curcumin, essential oils, and oleoresins during 12 months storage at ambient temperature. Curcumin content continued to decline upto 10 months, but the change, thereafter, was minor. However, essential oil and oleoresins decreased throughout the storage period. The maximum losses in curcumin, essential oils and oleoresins were 23.4, 27.5 and 24.2%, respectively, after 12 months in cured rhizomes of cultivar 'EM-321' and uncured rhizomes of cultivar 'PCT-2'.

Keywords : *Curcuma longa*, Turmeric, Rhizomes, Storage, Curcumin, Essential oil, Oleoresins

Turmeric (*Curcuma longa* L.) is an important spice which is used widely in curry preparations as a natural colouring agent. Among the curcuminoid pigments responsible for the colour of turmeric, curcumin (diferuloyl methane) is the major pigment (Srinivasan 1953; Govindarajan 1980). It has been reported that processing and varieties affect the colour contents of turmeric (Krishnamurthy et al 1975). The medicinal properties of curcumin have also been reported (Ammon and Wahi 1991). Its volatile oil (essential oil) has been shown to be anti-inflammatory, anti-arithritic (Chandra and Gupta 1972) and anti-bacterial (Bhavani Shankar and Sreenivasa Murthy 1979) in nature. Hence, the quality of turmeric is mainly determined by curcumin and essential oil contents. There is, generally, a wide gap between the harvest time of turmeric and its consumption, and consequently curcumin, essential oil and oleoresins of turmeric rhizomes may undergo changes during this period. Hence, the rhizomes were stored for 12 months in the present study, and periodically evaluated for the quality characters.

Four turmeric cultivars viz., 'PCT-2', 'EM-321' 'ST-85' and 'ST-323 Y' were planted in randomized block design in three replications. The mature rhizomes were harvested (in the month of February) from each replication and pooled. The rhizomes (which constitute mother rhizomes as well as fingers) were washed thoroughly with water to remove any adherent. One half of the sample was cured (Govindarajan 1980) by boiling in water for 1.5 h so that the rhizomes became soft. The other half was kept as such, and referred to as uncured rhizomes. Both types of rhizomes were cut into slices (1 cm thick), dried at 65°C in hot air oven for 96 h to 5% moisture level, and stored in unsealed polythene bags at room temperature (10-35°C) and RH (23-95 %). The samples were drawn at 2 months interval for analysis.

Curcumin was estimated (in quadruplicate) as per ASTA (1968) method. Since the curcumin content in mother rhizome is always higher than fingers (Korla et al. 1992), a minimum of 50 g sample was taken to avoid any disproportion. The sample was ground to fine powder and 100 mg powder was used for curcumin estimation. The method of AOAC (1980) was followed for essential oil determination in triplicate. The oleoresins were extracted (in triplicate) with acetone by cold percolation method (Purseglove et al. 1981).

A gradual decrease in curcumin content was observed as the storage period increased (Table 1). After 6 months, the decrease in curcumin varied from 7 to 13.6% in the rhizomes of different cultivars. The curcumin level continued to decline upto 10 months. Thereafter, either there was no reduction in curcumin or it was very small, except in 'PCT-2', where a small decrease was noticed. At the end of 12 months, the minimum and maximum losses in curcumin were similar in uncured and cured rhizomes. Thus, the curing of rhizomes did not affect the overall % loss in curcumin during storage. The variations of % losses in different cultivars may, however, be due to the varietal differences. The decrease in curcumin during storage may be due to degradation of the pigment. According to Sampathu et al. (1988), the degradation may be due to the photosensitive nature of curcumin. The hydrolytic degradation (Tonneson and Karlsen (1985a) as well as chemical degradation of curcumin

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(Tonneson and Karlsen 1985b) has also been noticed under alkaline conditions. The comparison of cured and uncured rhizomes revealed a slightly lower value of curcumin in the latter. This is in spite of some loss in colour during curing. The decrease in other constituents of rhizome, especially starch, sugars and essential oils, appears to be responsible for the elevation of curcumin content in cured rhizomes. The process of boiling of rhizomes in water extracts free sugars and also solubilizes some starch.

The curing of rhizomes as well as their storage leads to reduction in essential oils (Table 1). The cured and uncured rhizomes of different cultivars

TABLE 1. CURCUMIN, ESSENTIAL OIL AND OLEORESIN CONTENT (% DRY MATTER) IN CURED AND UNCURED RHIZOMES OF TURMERIC AT DIFFERENT INTERVALS OF STORAGE

Cultivar	Rhizomes	Storage months							Overall
		0	2	4	6	8	10	12	mean
		Curcumin, % w/w							
'ST-85'	Cured	4.80	4.62	4.31	4.20	3.82	3.69	3.72	4.16
	Uncured	4.10	4.13	3.85	3.59	3.30	3.20	3.18	3.62
'PCT-2'	Cured	4.23	4.26	3.98	3.81	3.88	3.57	3.47	3.88
	Uncured	4.00	3.90	3.66	3.50	3.45	3.32	3.26	3.59
'ST-323 Y'	Cured	4.00	4.06	3.94	3.73	3.61	3.26	3.22	3.69
	Uncured	3.88	3.90	3.66	3.40	3.35	3.28	3.22	3.53
'EM-321'	Cured	2.73	2.58	2.40	2.46	2.21	2.06	2.09	2.34
	Uncured	2.50	2.51	2.28	2.16	2.06	1.96	1.93	2.17
Overal mean		3.76	3.72	3.51	3.35	3.21	3.04	3.01	
CD* (cultivar); 0.06, CD (curing/uncuring) : 0.04, CD (storage interval) : 0.08.									
		Essential oil, % v/w							
'ST-85'	Cured	7.50	7.05	6.72	6.30	6.08	5.87	5.65	6.45
	Uncured	8.00	7.88	7.08	6.75	6.33	6.20	6.00	6.89
'PCT-2'	Cured	6.40	6.00	5.72	5.28	5.00	4.88	4.75	5.43
	Uncured	6.75	6.42	6.05	5.78	5.38	5.22	4.90	5.79
'ST-323 Y'	Curred	5.67	5.50	5.08	4.78	4.45	4.37	4.20	4.86
	Uncured	6.17	5.83	5.57	5.28	4.87	4.70	4.55	5.28
'EM-321'	Cured	7.00	6.72	6.30	5.78	5.50	5.38	5.23	5.99
	Uncured	7.38	7.00	6.55	5.22	5.82	5.70	5.55	6.32
Overall mean		6.86	6.55	6.13	5.77	5.43	5.29	5.10	
CD* (cultivar) : 0.8, CD (curing/uncuring) : 0.05, CD (storage interval) : 0.10.									
		Oleoresins, % w/w							
'ST-85'	Cured	12.93	12.15	11.44	10.73	10.38	10.10	9.98	11.10
	Uncured	14.12	13.63	12.70	11.80	11.52	11.21	10.86	12.25
'PCT-2'	Cured	10.92	10.65	9.59	9.12	8.78	8.53	8.50	9.42
	Uncured	11.19	10.62	9.96	9.34	8.86	8.60	8.50	9.61
'ST-323 Y'	Cured	10.20	9.90	9.52	8.94	8.19	7.96	7.83	8.93
	Uncured	10.75	10.23	9.78	9.13	8.68	8.43	8.20	9.31
'EM-321'	Cured	10.25	10.13	9.65	9.12	8.28	7.95	7.80	9.02
	Uncured	11.00	10.48	10.05	9.78	9.18	9.00	8.65	9.73
Overall mean		11.40	10.97	10.36	19.75	9.23	8.96	8.79	
CD* (cultivar) : 0.14, CD (curing/uncuring) : 0.10, CD (storage interval) : 0.18.									

* at 5% level of significance

experienced maximum loss in essential oil from 24.6 to 25.9% and 24.8 to 27.5 %, respectively. In general, the reduction in essential oil was relatively more as compared to curcumin. The rate of loss of essential oils was minimum in the last 4 months. It may be due to the lowest (10-20°C) temperature prevailing during that period (Fig.1), and the loss of more volatile fraction of essential oils in the initial stages of storage. The higher values of oil in uncured rhizomes than cured ones are, however, due to the loss during curing.

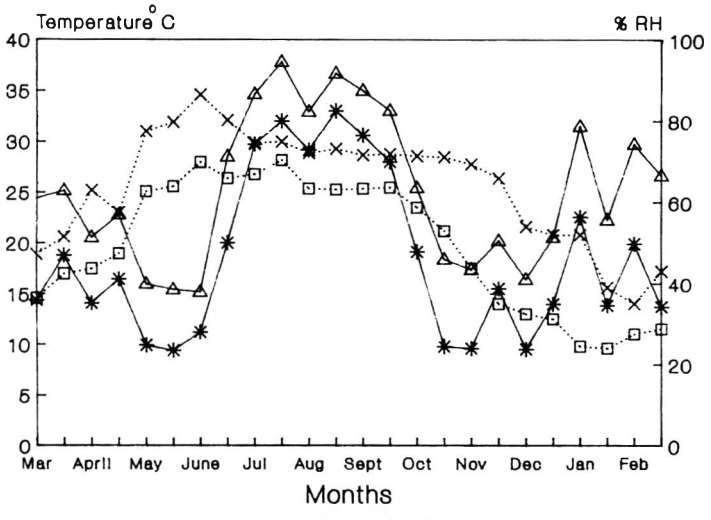

Fig.1. Minimum temperature (....□....), maximum temperature (.....X....) minimum % RH (---*---) and maximum % RH (—Δ—) during the storage period of rhizomes.

During storage, the oleoresins of all the cultivars studied were reduced and the extent of reduction increased with the increase in storage period (Table 1). Storage for 12 months resulted in depletion of oleoresins by 21.4 to 24.0% in cured and uncured rhizomes of different cultivars. The decrease in oleoresin content is more or less a reflection of decrease in curcumin and essential oils, because these two components constitute more than 80% of the total turmeric oleoresins.

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Rice Kernel Breakage Kinetics in the Process Operation for Bran Removal

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Experiments were conducted on five varieties of paddy ('Prasad', 'Manhar', 'Sita', 'UPR-239' and 'P.D-4') to study the effect of polishing time, and degree of polish on breakage behaviour. The results indicated that the % breakage increased nonlinearly with milling time, whereas breakage varied linearly with degree of polish. The breakage behaviour was found to be dependent on variety of paddy.

Keywords : Rice kernel breakage, Bran removal, Polishing, Whitening.

For improving the appearance of rice kernels, bran removal is an imperative operation in rice milling industry. Acceptable degree of whiteness of the kernels can be accomplished by 5 to 10% polishing of the brown rice (Spadaro and Mathews 1976). However, kernel breakage is the undesirable side effect of the rice polishing. In commercial scale rice milling operations, rice kernel breakage is one of the single most important factors which affects the economics of the milling process (Gariboldi 1974). Factors contributing to breakage can be classified as those related to grain properties and milling condition (Rhind 1962). The properties of grain at the time of milling are influenced by factors

TABLE 1. PHYSICAL CHARACTERISTICS OF VARIETIES AND VALUES OF COEFFICIENT OF REGRESSION EQUATION

Attribute		Variety 'Prasad'	'Manhar'	'Sita'	'UPR-239'	'PD-4'
			Paddy varieties			
Mean	length	8.87	9.13	8.99	8.65	9.25
	S.D.	0.44	0.41	0.32	0.43	0.33
Mean	width	2.15	2.18	2.28	2.50	2.51
	S.D.	0.12	0.14	0.03	0.12	0.13
Mean	thickness	1.78	1.80	1.83	1.91	1.94
	S.D.	0.02	0.22	0.08	0.10	0.09
			Brown rice			
Mean	length	6.43	6.56	6.78	6.42	8.93
	S.D.	0.29	0.20	0.22	0.30	0.42
Mean	width	1.98	2.13	2.11	2.49	2.44
	S.D.	0.13	0.15	0.14	0.20	0.11
Mean	thickness	1.56	1.74	1.68	1.84	1.87
	S.D.	0.06	0.08	0.06	0.04	0.06
			Values of coefficient of equation 1			
	m	3.29	3.37	1.38	6.99	4.44
						0.24
	n $\cdot 10^2$	1.12	0.64	1.63	1.60	0.24
	r	0.97	0.99	0.94	0.94	0.99
	SEE	0.10	0.04	0.21	0.20	0.01
			Values of coefficient of equation 2			
	C	0.09	0.03	0.14	0.24	0.60
	r	0.96	0.92	0.99	0.99	0.97
	SEE	0.08	0.06	0.12	0.17	0.26

* S. D. = Standard deviation, r = Correlation coefficient, SEE = Standard error of estimate

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such as agronomic practices, harvesting time, drying, storage, transportation, and other pre-milling operations (Rhind 1962). The factors affecting the breakage at the time of milling include temperature and moisture content of kernels, temperature and relative humidity of the environment, degree of milling and mechanical setting of the milling machinery (Raghavendra Rao et al. 1976; Bhattacharya 1980; Sharma and Kunze 1982; Mathews et al. 1981). In the review on factors influencing breakage (Spadaro and Mathews 1976), about half of the breakage of grains is reported to occur during various milling operations, such as cleaning, shelling, polishing, size separation, and in-plant handling; shelling and polishing accounting for major proportion of breakage. The other half is attributed to pre-milling conditions and past history of grain. However, it is only at the time of milling that all the effects get combined (Pandey 1991). The milling system and the operational aspects are mostly held responsible for kernel breakage, and ensuing losses (Velupillai and Pandey, 1990).

Five varieties ('Prasad', 'Manhar', 'Sita', 'UPR-239' and 'PD-4') of paddy were collected from the rice breeder of the University. Paddy samples were thoroughly cleaned to remove foreign material. Shelling was done in a laboratory model rice sheller (Model Tu-35 Satake, Japan). After separating unhusked grains, samples of 100 g of brown rice were milled in a laboratory rice whitener (Model Tm-25 Satake Japan) for 10 to 110 sec at an interval of 10 sec to attain different degrees of bran removal. All the broken grains, irrespective of their sizes in the sample, were separated from the head rice and accounted as brokens. The counting of the brokens in samples was done just after shelling, and after various times of polishing. Reported values are the average of three replicates.

The characteristic features of the rice varieties used for the experimentation are shown in Table 1.

For quantifying the process of breakage of kernels, various empirical relations were considered and based on the premise that rate of breakage could either be constant with time or a function of breakage which has already taken place or be a function of the further breakage to take place. The first two relations were discounted on the basis of the indicative inference of the past reported studies (Spardo and Mathews 1976; Kunze and Choudhary 1972) as well as on the statistical results obtained in analysis of experimental data done with a Hewlett Packard mini-computer. The following empirical equation was found to describe the process satisfactorily.

$$(B - B_0)/B_o = m \exp (n\ t) \qquad \ldots (1)$$

Where B is the breakage (%) at a polishing time t. B_0 is the % broken before polishing and m and n are the coefficients of the empirical equation. The equation was fitted after linearization (1). The reported values of coefficient m and n of the equation (1), the correlation coefficient were more than 0.94 and SEE less than 0.20, while the values of coefficients m and n varied with variety and were in range from 1.37 to 6.68 and 6.98 and 2.37×10^{3} to 1.63×10^{-2}, respectively. Fig.1 shows the breakage behaviour of different varieties with

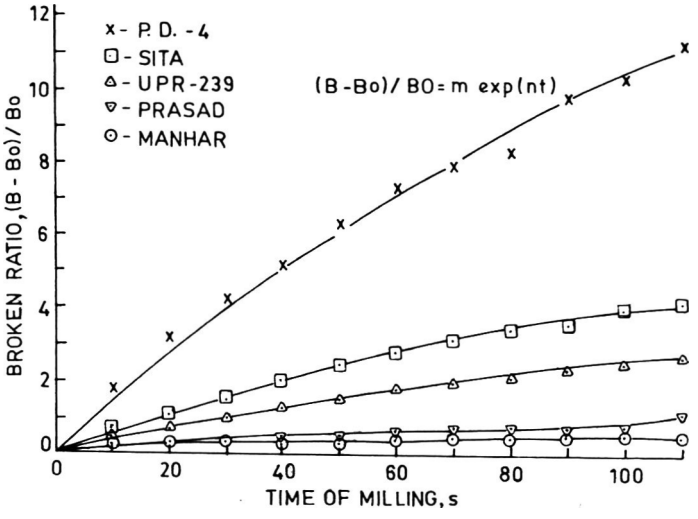

Fig. 1. Effect of time of milling on broken ratio of different varieties of paddy

the time of milling. 'P.D.–4' variety was found to be more susceptible to breakage, whereas Manhar showed the least breakage. One of the probable reasons for this could be that 'P.D.–4' was thicker in its dimension than others (Table 1). This is consistent with the observations (Pandey 1991) that the dimension of grain plays an important role in polishing, and breakage of kernels. It has been established (Pandey and Sah 1990) that whiteness of the polished rice not only depends on degree of polish, but also on whiteness of the brown rice. A compromise, therefore, has to be made on degree of polish, whiteness of kernels and the tolerance limit of brokens. Equation (1) correlates the breakage of kernel to the time of polish. However, the relationship between breakage and polish would be of more practical value, as it will provide information on degree of polish, keeping in view the breakage

tolerance of the rice variety. In case, a variety is more susceptible to breakage while polishing, remedial measures such as parboiling may be suggested for increasing the breakage resistance. Keeping this fact in view, the following breakage polish relationship has been developed.

$$(B - B_0)/B_0 = C \times P \quad \ldots (2)$$

Where P is the degree of polish (%) and C is the regression coefficient. The results are shown in Fig.2, and the values of coefficient of equation (2) are given in Table 1. along with the values of

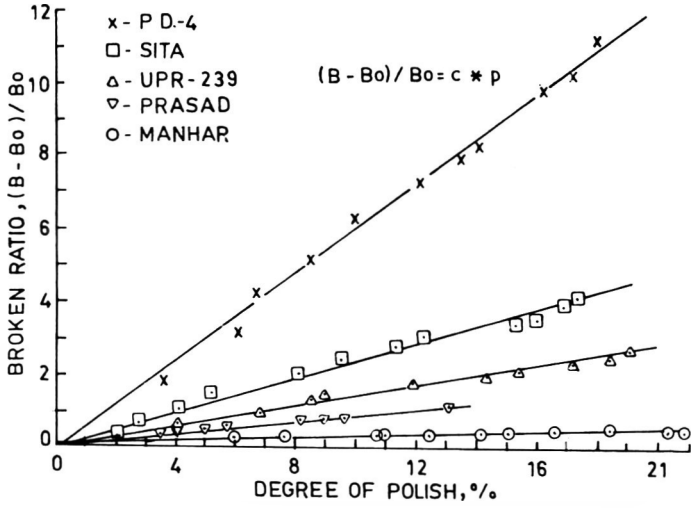

Fig. 2. Variation of broken ratio with degree of polish for various varieties of paddy

correlation coefficient and standard error of estimate. The values of coefficient C were in the range from 0.03 to 0.60. Higher values of C show higher susceptability of a variety to breakage. Variety 'P.D.4' was highly susceptible to breakage in comparison to other varieties as indicated by the values of C.

Thus, it can be concluded that the breakage - polishing time correlation for different varieties of paddy is dependable and the % breakage of grain increased linearly with degree of polish.

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Enzymatic Pretreatment of Pigeonpea (*Cajanus cajan* L.) Grain and Its Interaction with Milling

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Effect of enzymatic hydrolysis parameters i.e. incubation temperature, treatment period and moisture content of grain was optimized for dehulling efficiency. The optimum levels of these parameters were found to be 46.5°C, 12.7 h and 26.6% w. b., respectively. The enzyme concentration was varied, and maximum hulling efficiency of 88.93% was obtained at 0.08 g of total enzyme protein content, mixed in 260 g of pigeonpea grain.

Keywords : Pigeonpea grain, Milling, Enzymatic pretreatment, Dehulling, Hulling efficiency, Response surface methodology.

Pigeonpea is one of the major food legumes grown and consumed extensively in Indian sub-continent. It is mostly consumed in the form of dehusked split or *dhal*. In the traditional milling process, the recovery of *dhal* is 66-75% (Kurien and Parpia 1968) and dehusking characteristics vary with different cultivars (Ramakrishniah and Kurien 1983). Because of the presence of thick gum layer between seed coat and cotyledon, pigeonpea comes under difficult-to-mill classes of pulses. In order to loosen the husk from cotyledon, various pretreatments are employed. Influence of soaking and its effect on the dehusking efficiency was studied by Srivastava et al (1988). Among these, the application of oil and water is most common. The substitution of oil with sodium bicarbonate resulted in improved hulling efficiency (Saxena 1985). Pretreatments of oilseeds with microbial enzyme have shown increased oil availability and easy extraction rate (Sosulski et al. 1988). The same approach was tried on pigeonpea grain,as no such report is available for improving hulling efficiencies of pulses.

Pigeonpea grains ('UPAS-120' variety) were procured from the University farm, and stored after cleaning and grading. The moisture content was measured following standard method (Saxena 1985). For treatment, enzyme solution was prepared by growing *Aspergillus fumigatus* NCIM-902, obtained from National Chemical Laboratory, Pune, on wheat bran medium. Initially, 1-2 loops of *A. fumigatus* culture were transferred into a conical flask containing sterilized Y_{pss} medium. It consists, of (g/l) : soluble starch 15, yeast extact 4, K_2HPO_4 1, $MgSO_4.7H_2O$ 0.5 and pH 7.0). The flask was, then, incubated at 45°C in an incubator shaker (120 rpm) for three days (Bhatnagar 1987). About 50 ml of the fermented medium was used, to inoculate a batch fermenter having sterilized wheat bran medium (wheat bran 60 g, distilled water 1.5 l, pH 7.0). Fermentation was allowed at 45°C with 1 vvm air flow rate and 120 rpm agitator speed. The fermentation was continued for 72 h, the fermented materials were filtered through cheese cloth, and the filtrate centrifuged at 10,000 rpm for 10 min. The solution, thus, obtained was used as crude enzyme for treatment. The protein content was measured following the method of Lowry et al (1951). The filter paper activity was measured using the method of Mandels et al (1976).

Response surface methodology was adopted for experimental design to optimize hydrolysis parameters - incubation temperature, treatment period, and moisture content of grain. A second order central composite rotatable design in three variables at five levels was used (Myers 1976). Treatment combinations are given in Table 1. For each experiment, a sample of 260 g of pigeonpea grain was taken in a conical flask. Moisture content was adjusted to the desired level for hydrolysis, by adding appropriate amount of enzyme solution and distilled water, keeping the volume of enzyme solution constant (Verma 1991). The flask was plugged, shaken manually and equilibrated in a refrigerator at 4°C. A 5 g of the sample was drawn for checkup of the moisture content, and the rest was incubated at different time and temperature combinations for hydrolysis. After incubation, the samples were dried in wire mesh container at 104°C, to inactivate enzyme, and also to reduce moisture content of grain. These samples were, then, milled in a laboratory mill (Model No. Tm-05, Satake Grain Testing Mill, Satake Engineering

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TABLE 1. HULLING EFFICIENCY OF ENZYME AND WATER TREATED PIGEONPEA

Treatment combinations			Hulling efficiency (%)	
Incubation temperature, °C X_1	Incubation period, h X_2	Moisture content, %, w.b. X_3	Enzyme treated	Water treated
40.0	9.0	20.0	73.45	72.02
50.0	9.0	20.0	74.90	72.69
40.0	15.0	20.0	74.87	72.30
50.0	15.0	20.0	76.32	72.98
40.0	9.0	30.0	74.67	72.94
50.0	9.0	30.0	76.12	73.61
40.0	15.0	30.0	76.08	73.23
50.0	15.0	30.0	77.53	73.90
32.7	12.0	25.0	73.60	71.94
57.3	12.0	25.0	74.13	73.85
45.0	4.6	25.0	75.50	73.04
45.0	19.4	25.0	77.50	73.01
45.0	12.0	12.7	74.00	70.26
45.0	12.0	37.3	77.10	73.68
45.0	12.0	25.0	79.68*	73.11*

* Average of six observations based on the design. Hulling efficiency of untreated sample was 60.82%

Co. Ltd., Tokyo, Japan) at 400 rpm, 4 mm exit clearance and 12 m/s surface speed. Different fractions obtained from milling were separated by sieving, and hulling efficiency was calculated using the formula suggested by Saxena (1985).

$$\text{Hulling efficiency} = \left(1 - \frac{Un}{Tn}\right)\left(\frac{F_p}{F_p + Br + P_o}\right) \times 100$$

Where, Un = weight of unhusked grain, Tn = total weight of grain used for milling, F_p = weight of finished product (weight of dehusked grain + *dhal*), Br = Weight of broken grain, P_o = weight of powder.

Water-treated sample was used to compare the results with enzyme treatment. The hulling efficiency values for different treatment combinations are given in Table 1.

Enzyme concentration was varied from 0.02 to 0.10 g of total enzyme protein/260 g sample, keeping other hydrolysis parameters at their optimum levels. Hulling efficiency data are given in Table 2. Untreated and sodium bicarbonate-treated samples of the grains were also milled at 9.5% (wet basis) moisture content, to compare the results. Comparison of the results is given in Table 3.

TABLE 2. HULLING EFFICIENCY OBTAINED FROM MILLING OF PIGEONPEA GRAIN TREATED AT DIFFERENT ENZYME CONCENTRATIONS

Total enzyme protein, g	Hulling efficiency, %
0.02	76.48
0.04	80.60
0.06	86.74
0.08	88.93
0.10	88.68

TABLE 3. COMPARISON OF MILLED FRACTIONS OBTAINED FROM UNTREATED, WATER TREATED, $NaHCO_3$ TREATED AND ENZYME TREATED PIGEONPEA

Fractions, %	Untreated grains	Water treated	$NaHCO_3$ treated	Enzyme treated
Unhusked grains	22.51	11.03	0.83	0.65
Dehusked grains	28.52	31.99	37.92	35.89
Dhal	23.27	30.93	35.91	39.83
Finished product*	51.79	62.92	73.83	75.72
Brokens	0.22	2.68	3.39	1.66
Powder	13.97	10.15	7.20	7.21
Hulling efficiency	60.82	73.90	86.73	88.93

*Finished product = Sum of dehusked grains + *dhal*

From the dehulling efficiency data (Table 1), it is clear that enzymatic pretreatment has positive effect on hulling efficiency. Hulling efficiency of untreated grains was found to be 60.82%, while the maximum value for enzyme and water treated grains were 79.68 and 73.90%, respectively.

A response surface model for hulling efficiency, as a function of enzymatic hydrolysis parameters, was developed by applying a multiple regression technique. The fitted response function was :

$$Y = -79.5845 + 0.4906\ X_1 + 0.6599\ X_2 + 0.7383\ X_3 - 1.9332\ X_1^2 - 1.0018\ X_2^2 - 1.3376\ X_3^2$$

Where, Y = hulling efficiency (%), X_1 = temperature (°C), X_2 = incubation period (h), X_3 = moisture content (%, wet basis).

The optimum for maximum hulling efficiency was calculated by partially differentiating the above equation with respect to each parameter, and equating to zero. The optimum values were :

X_1 = 46.5°C, X_2 = 12.7 h, and X_3 = 26.6% (wet basis).

Three dimensional response for hulling efficiency of enzyme-treated pigeonpea were generated

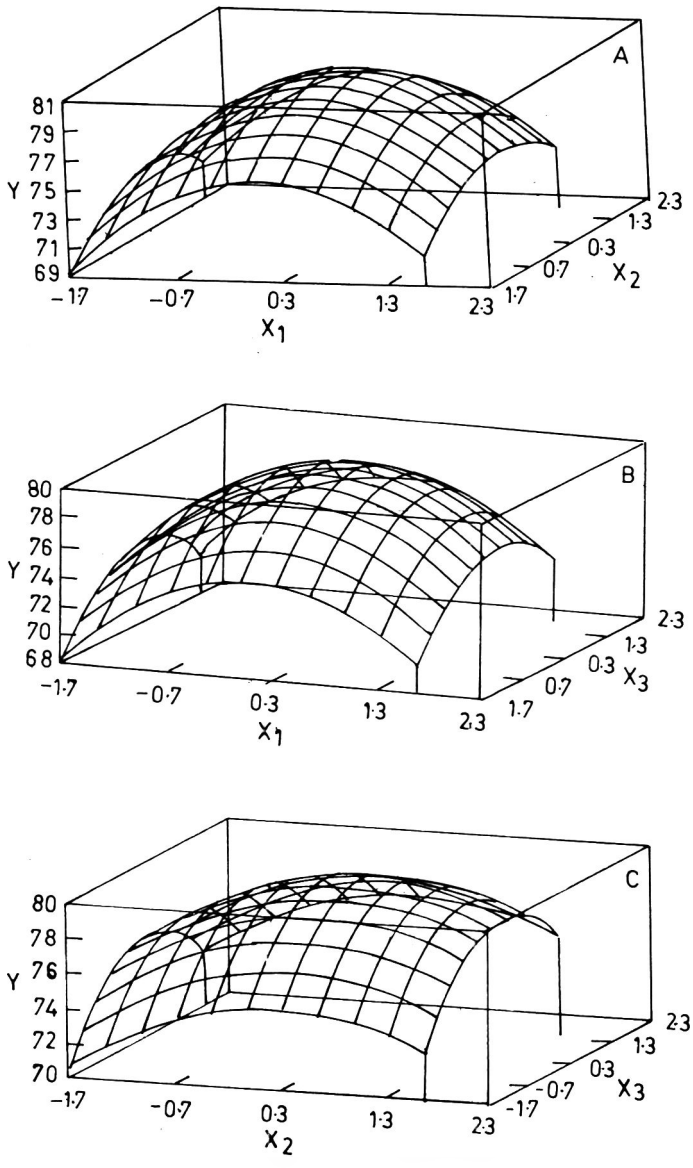

Fig. 1. Three dimensional response for hulling efficiency. Effect of (A) incubation temperature (X_1), °C and treatment time (X_2), h., (B) incubation temperature (X_1), °C and moisture content (X_3), % w.b., (C) treatment time (X_2), h. and moisture content (X_3), % w.b.

(Fig. 1). From these surfaces, it is evident that dehulling efficiency initially increases with increase in parameter value, and then started decreasing, thereby indicating the existence of optimum levels of hydrolysis parameters within the selected range. Similar behaviour was observed by Smith (1990), when soybean was treated with enzyme solution.

Enzyme concentration had shown significant effect on hulling efficiency. Results (Table 2) showed that the highest hulling efficiency of 88.93% was observed at 0.08 g enzyme/260 g sample. With further increase in enzyme content, no appreciable increase was observed. This may be due to the fact that the free reactive groups of the surface become limiting to the reaction leading to a plateau. This type of behaviour in enzyme catalysis reaction is common, and discussed by Bailey and Ollis (1986).

It is evident that the enzyme not only increased the hulling efficiency, but also reduced the amount of powder formed (Table 3). It may be mentioned that higher amount of powder produced during dehulling is a direct loss in the processing of pigeonpea. Since no work on the enzymic treatment of pigeonpea grains was available, the results could not be compared.

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Role of Citric Acid on Iron Availability in a Model System

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Role of citric acid in overcoming the effects of polyphenols and calcium phosphate salts on iron availability in a model system has been investigated. Results indicated that citric acid had the potential, as good as ascorbic acid, in counteracting the inhibitory effects of tannate and calcium phosphate on iron availability.

Keywords : Citric acid, Iron availability, Tannic acid, Calcium phosphate, Ascorbic acid, Ionisable iron.

Several chemical ligands act either as an enhancer or inhibitor of iron availability (Cook et al. 1972). Important among the enhancers is ascorbic acid, whose action is dose-dependent (Christian and Seshadri 1989; Cook and Monsen 1977). Ascorbic acid also has the ability to counteract the inhibitory effect of certain constituents, such as polyphenols on iron availability (Christian and Seshadri 1989: Siegenberg et al. 1991). Recently, citric acid has been suggested as a potential enhancer of iron bioavailability (Hazell and Johnson 1987). However, the information on dose-related dependence of the effect of citric acid and its ability in overcoming the effect of inhibitors of iron availability, such as polyphenols and calcium phosphate is not available. Therefore, in the present study, the role of citric acid in influencing non-heme iron availability, when added alone or in combination with ascorbic acid, tannic acid and calcium phosphate has been investigated in a model system. Iron availability was expressed in terms of % ionizable iron, which has been shown to correlate highly with *in vitro* available iron (Narasinga Rao and Prabhavathi 1978). Several studies have shown that the *in vitro* available iron is a good measure of bioavailable iron (Forbes et al 1991; Narasinga Rao and Prabhavathi 1978; Schricker et al. 1981).

The model system used was a ferric chloride solution (Merck, analytical reagent), providing 3 mg elemental iron in 250 ml of the solution. Different levels of citric, ascorbic and tannic acids and calcium phosphate corresponding to the entire range of daily consumption, as reported by representative diet surveys in India (NNMB 1984), were tested for their dose-effect on iron availability. For studying the combined interaction effects, the lowest and highest dose levels of each of the four constituents were selected, and all possible combinations of the constituents were generated through a computer (n=16). These combinations were added to the model system, homogenized and analyzed in four replicates, for total iron (AOAC 1984) and ionisable iron (Narasinga Rao and Prabhavathi 1978). The data were subjected to multiple regression analysis (stepwise technique) using the statistical package for social sciences (SPSS) in an IBM-PC computer assembly, while correlation coefficient for each constituent in the mixed milieu was obtained. A regression equation was also formulated to obtain the ionizable iron, when known quantities of the four constituents were present.

Results of the dose effect of the constituents are shown in Fig.1. The % ionizable iron from the ferric chloride solution *per se* was 43. Addition of citric acid at increasing dose levels (30 mg to 960 mg) did not produce any measurable effect on the ionizable iron. Increasing levels of ascorbic acid, however, brought about the dose-related increase in ionizable iron, until it reached the maximum value of 100% with 46.5 mg dose level. Both tannate and calcium phosphate exerted marked dose-dependent inhibition on ionizable iron, reducing it from 43 to 4 and 0%, respectively (Fig 1). The multiple regression analysis of the interaction effect of citric, ascorbic and tannic acids as well as calcium phosphate, in varying combinations, is shown in Fig. 2. The correlation coefficients obtained were significantly positive for citric and ascorbic acids ($r = +0.367$ and $+0.375$, respectively; $p<0.01$), indicating a strong enhancing effect. These, for tannate and calcium phosphate were significantly negative ($r = -0.255$; $p<0.05$ and -0.483; $p<0.001$,

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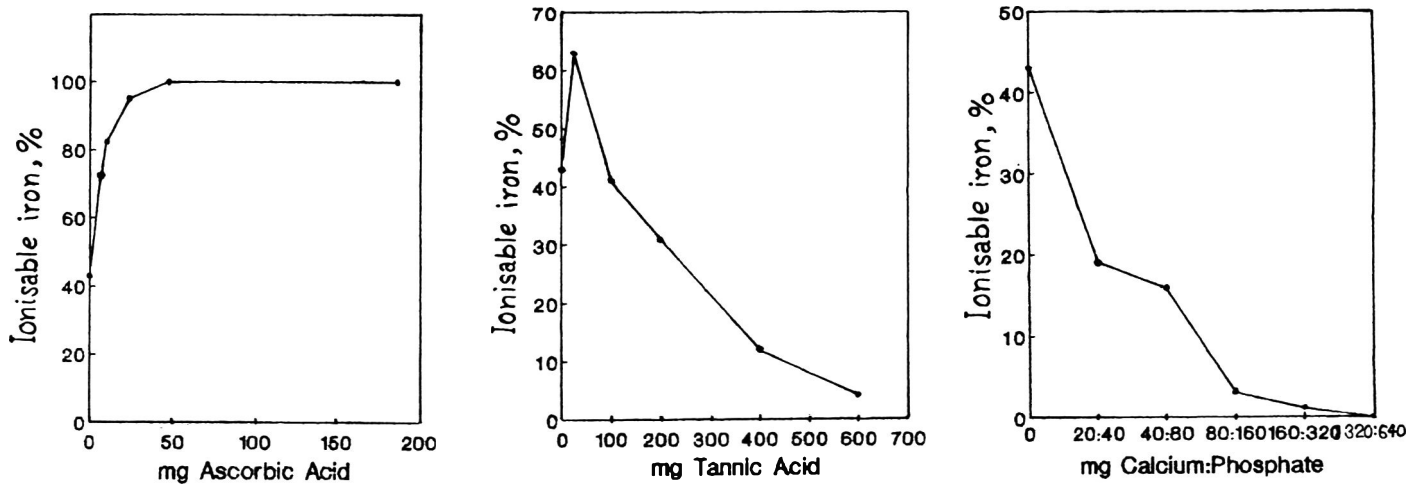
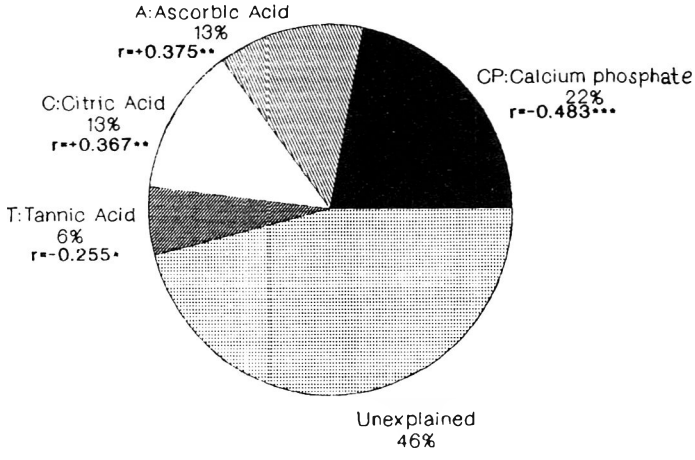


Fig. 1. Dose effect of ascorbic and tannic acids and calcium phosphate on % z ionizable iron in the model system.



• P < 0.05, •• P < 0.01, ••• P < 0.001

Fig.2. Regression analysis of the four constituents in the model system.

Y [% Ionizable Iron] = 43.4 - 0.0806 CP + 0.4299 A + 0.0198 C - 0.0255 T

respectively), indicating that they exerted an inhibitory effect on ionizable iron.

When an attempt was made to calculate the availability of iron using the highest dose levels of the four variables in the equation, it was observed that the calculated % ionizable iron (Y) increased from the basal level of 43% to 58%. Based on the dose-response curves (Fig. 1), only tannic acid and calcium phosphate at the highest dose levels were seen to reduce the availability of iron to zero. However, a net positive increase in the calculated % ionizable iron, in spite of the presence of these strong inhibitors, indicated that citric acid, when present along with ascorbic acid, was not only able to overcome the inhibitory effects of tannate and calcium phosphate, but could also increase ionizable iron beyond the basal level by 33% in the model system. Citric acid, unlike ascorbic acid, is not a reducing agent. Therefore, it cannot reduce ferric ions to ferrous ions, which is reflected in the lack of any measurable effect on ionizable iron, when citric acid is added alone. On the other hand, citric acid, by virtue of its three carboxyl and one hydroxyl group, may form loosely bound chelates with iron at neutral or alkaline pH, which can prevent the precipitation of iron at the alkaline pH by compounds like tannic acid or calcium phosphate (Kojima et al. 1981). Citric acid can also bind calcium salts, thereby preventing them from forming insoluble complexes with iron (Hazell and Johnson 1987). Citric acid, when present along with tannic acid, can also prevent formation of high molecular weight insoluble iron-polyphenol or iron-calcium phosphate complexes, thereby countering their inhibitory effect.

Studies on the effect of citrate *per se* unequivocally are very few. Studies on various fruits and vegetables have shown increased availability of iron, as a function of both citric acid and ascorbic acid (Ballot et al. 1987; Hazell and Johnson 1987). The limitation of these studies is that the effect cannot be attributed to only citric acid, as the fruits and vegetables contain many other organic acids and some undefined and unknown constituents as well. Moreover, these studies do not provide any experimental evidence to show that citric acid can overcome the inhibitory effect of tannic acid and calcium phosphate. The present study provides clear evidence that citric acid, like ascorbic acid has the ability to counter the inhibitory effects of these two constituents.

The implications of the findings for food composition tables and iron nutrition are important. In order to have a more valid estimate of the bioavailable iron from foods, one should have the food composition tables that will provide the citric

and tannic acids content. Moreover, practical recommendations for improving iron nutrition must include fruits and vegetables high in both citric and ascorbic acids. Whether other organic acids would have similar effects, need to be studied.

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Studies on *Tandoori* and Battered Quail

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The proximate composition, microbial load and acceptability of quail meat from 6, 12 and 18 week old birds as well as the processed products were determined. *Tandoori* and battered quails were found to be highly acceptable, and no statistically significant ($P \leq 0.05$) differences were observed with respect to the age of bird and proximate composition. However, slight decrease in moisture with advancement of age from 6 to 18 weeks as against higher proteins, fat, energy and methionine contents were observed. Age of the bird and method of cooking were directly related to bacterial load.

Keywords : Quail, *Tandoori* and battered products, Proximate composition, Sensory evaluation, Acceptability.

Poultry production has become an important facet of commercial development in recent years. Japanese quail (*Coturnix japonica*), a recently domesticated economic avian species, is ideally suited for commercial rearing for eggs and meat under intensive conditions. AVM Hatcheries and Poultry Breeding Research Centre (P) Limited, have franchised and Central Avian Research Institute has developed the layer and meat type Japanese quail production technology on a large scale, with about 100,000 commercial meat type quails in its farm at any given time of the year (Wahab 1991). Quail completes their juvenile stage at 5 weeks with a mean body weight in the range of 129 to 150 g (Wahab 1991). Quail meat is a high protein food of immense biological value and known for its taste and delicacy. In spite of many advantages and superiority of quail over the other species of poultry (Wahab 1991), very few studies have been carried out on quail meat products. Hence, the present investigation was undertaken to study the proximate composition, microbial count and acceptability of quail meat from 6, 12 and 18-week old birds and the processed products such as *tandoori* and battered quail.

Japanese quail of different ages were procured from the Poultry Farm of the University. The birds were bled for one min by cutting the jugular vein at the *accipito atlantus* joint, and feathers were removed manually. The birds were dressed, and washed in running tap water thoroughly. The cleaned carcasses were taken for the study. *Tandoori* and battered quail products, using 6, 12, and 18 week old birds, were made by following the method described by Beeton (1980), with some modifications with respect to additives.

The whole carcasses of quails were used for *tandoori* preparation. Earlier studies indicate that the quail meat could be stored upto 60 days after treatment with sodium tripolyphosphate without much bacterial load (Prabhakar Reddy et al. 1991) Slits were made on the dressed carcasses with the help of knife. The *tandoori masala* contained (g/1000 g dressed quails without giblets) anise seeds 5, black pepper 5, caraway 3, cardamom 3, cinnamon 2, clove 2, cumin 5, red chilli powder 10, peeled garlic 20, peeled ginger 20, peeled onion 50, table salt 35, turmeric 5, hydrogenated vegetable fat 100. *Tandoori* colour was added to the desired level. *Tandoori masala* was applied thoroughly on the surface and body cavities. The quails were kept in refrigerator for 2 h, to facilitate better absorption of *masala* by the meat tissues. They were rubbed both on inner and outer surface with melted *ghee* and baked uncovered at 150°C for 30 min.

The carcasses of quails were cut into smaller pieces. The spices and condiments used include (g/1000 g ready-to-cook quail) cinnamon 2, clove 1, corriander leaves 100, corn starch 250, curd 150, garlic 20, ginger 20, green chillies 15, pepper 2, salt 35 and oil (for frying) 250. Spices and condiments were ground to a fine paste. Corn starch was added to the paste so that the *masala* would be homogeneous and dipping was easy. The quail meat pieces consisting of both bones and muscles were dipped in the *masala* paste and mixed thoroughly. It was kept in refrigerator for 2 h. Cooking oil was heated (185°C) in a frying pan, and quail meat pieces were then fried, till the pieces turned golden brown in colour and crisp in texture.

Samples of fresh meat and products were dried in hot air oven at 80°C till constant weight. The

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dried samples were powdered and analysed for moisture, protein and fat by AOAC (1990) methods. Gross energy was determined in a Bomb-calorimeter (Gopala Krishna and Ranjhan 1980), while methionine content of the samples was estimated by the method of Koch and Hanke (1953). The standard plate counts of the samples were estimated, using standard procedure (ISI 1980). Microbial count was expressed as colony-forming units/g (Cfu/g) of the sample. The products were judged for appearance, colour, texture, flavour and overall acceptability by a panel of 15 semi-trained non-vegetarian judges selected from the University, using a 7-point Hedonic scale (Amerine et al. 1965). Results were analysed statistically (Snedecor and Cochran 1967).

The proximate composition and microbial load of quail meat of 6, 12, and 18-week old birds and their products are given in Table 1. As the age of the quail increased from 6 to 18 weeks, the moisture contents of all the samples were found to decrease. The protein and fat contents in quail meat and the products increased with advancement of age from 6 to 18 weeks. However, these increases with age were not to the extent of being statistically significant ($P \leq 0.05$). The energy content also increased from 530 to 641 Kcal in quail meat as the age increased from 6 to 18 weeks. Similarly, the energy value of products also increased with age. The differencs in energy value of quail meat and its products at different age levels were not statistically significant ($P \leq 0.05$). The battered quail samples showed lower values of methionine than those of raw meat and *tandoori* quail samples at different age levels. The results are in accordance with the findings of Strange et al. (1980). The differences in methionine contents of the samples were not statistically significant ($P \leq 0.05$).

The bacterial load of raw samples was higher than that of the quail products. Battered quail sample had lower bacterial load as compared to that of *tandoori* quail. The difference may be due to method of cooking, where the temperature of oil in deep-fat-frying of battered quail (185°C) is higher, as compared to that of *tandoori* quail (150°C). Similar findings have been reported by Panda (1980). There was a significant difference in bacterial load between the age levels and the samples i.e., raw meat and products ($P \leq 0.05$).

Quail products were found to be very much acceptable among the semi-trained taste panel members. There was not much difference in the overall mean scores for *tandoori* and battered quail, when 6, 12 and 18-week old birds were used for the preparation. In case of *tandoori* quail, the mean score for texture was highest, followed by appearance, colour and flavour. The flavour got the highest mean score in case of battered quail, followed by texture, appearance and colour. According to Kumararaja and Venkataramanujam (1980), the tenderness of meat was found to be one of the major factors, which influences the acceptability of poultry by the consumers. Singh and Srivastav (1980) have also reported that the *tandoori* quail, regardless of age, was very much acceptable, when served to the panel members.

TABLE 1. PROXIMATE COMPOSITION AND MICROBIAL COUNTS OF *TANDOORI* AND BATTERED QUAIL

Age, weeks	Samples	Moisture, %	Protein, %	Fat, %	Energy, Kcal/100g	Methionine, g/g N	Microbial count Cfu/g
6	Quail raw meat	76.3	68.0	25.8	530	0.20	3.7×10^6
	Tandoori quail	64.9	54.3	20.7	537	0.19	1.9×10^5
	Battered quail	21.6	36.8	41.0	602	0.16	1.2×10^4
12	Quail raw meat	74.5	71.8	28.7	591	0.22	6.0×10^7
	Tandoori quail	64.8	56.0	21.0	598	0.20	4.5×10^5
	Battered quail	21.3	38.5	44.2	622	0.18	1.8×10^4
18	Quail raw meat	73.2	77.0	34.6	640	0.23	2.0×10^8
	Tandoori quail	64.4	57.8	31.4	646	0.22	7.0×10^5
	Battered quail	20.8	43.8	50.4	685	0.19	2.0×10^4

The vales are mean of two replications. * $P < 0.05$.

This supports the fact that there was no difference in acceptability of *tandoori* and battered quail prepared from 6, 12 and 18-week old birds, as observed in the present study. However, Friedman's (Conover 1971) two way analysis of variance for the characteristics like appearance, colour, flavour, texture and overall acceptability of *tandoori* and battered quail at different age levels were not found to be statistically significant ($P \leq 0.05$).

It may be concluded that quail meat can be successfully used in the preparation of *tandoori* and battered products, which were found to be highly nutritious due to higher methionine content. The bacterial load gets reduced greatly on cooking. Development and marketing of delicious ready-to-eat specialities like *tandoori*, and battered quail products could offer a good avenue for profitable utilization of quail. Such products could find an important place in fast food service establishment in the country, thereby benefitting the producers, processors and consumers alike.

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Population Dynamics of Insect Pests and Damage of the White Button Mushroom in the Environment of North Eastern India

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Types of insects attacking white button mushrooms and their population fluctuation were studied during the cropping season of 1990-1991. Mean total population revealed a relationship with temperature fluctuation during the period. Insects belonging to five orders viz., Diptera, Hymenoptera, Coleoptera, Collembola and Homoptera were recorded, among which Diptera accounted for the highest population (18.5 ± 0.53/trap/day, 94.9% of the total population). Highest overall population among the Dipterans was attributed to Phoridae (8.58 ± 0.31), followed by Sciaridae (5.58 ± 0.26) and Cecidomydae (4.56 ± 0.23). In respect of the families, population fluctuation revealed a varying trend in the different months. Crop yield was considerably affected by insect pest damage (49% reduction in comparison to control).

Keywords : Insect pests; White button mushrooms, Extent of damage, Weather factors, Population dynamics.

White button mushrooms (*Agaricus bisporus*), a nutritious diet supplement, have been found to be damaged by several insect - pests (Atkins 1974; Finlay 1984; Thapa and Seth 1981) resulting in considerable deterioration of the quality and quantity. The affected fruiting bodies showed characteristic symptoms such as yellowing, shrinkage and decaying of fruit bodies, as reported elsewhere in the country and abroad (Atkins 1974; Austin and Jary 1934, 1935; Binns 1981; Shandilya et al. 1975). The product with such changes is not acceptable for consumption. An organised and scientific mushroom cultivation is relatively new in North East India and systematic studies have not been undertaken so far on insect pests attacking the crop in this region. Therefore, studies were carried out to determine the types of insects attacking the crop, their relative abundance, relationship with indoor weather parameters, and the extent of damage in a moderate size cultivation, prevalent in the North East region of the country.

The entire study was carried out during the period, commencing in mid-December and ending in mid-April of 1990-91, the usual cropping season for white button mushroom in this part of India.

For pest population studies, three rooms of 20' X 10' were used, considering each room as one replicate and, daily records were the average of the data obtained in three replicates. Mushroom cultivation was carried out, as per recommended methods (Munjal and Seth 1980) in wooden trays, keeping four trays-one on top of another-in rows 1ft apart, covering the total floor area in equidistribution. Pest population sampling was carried out regularly at 24 h interval, using a light trap with provisions for retaining daily catches without disfiguration. One light trap was placed in the centre of each room and operated for 6 h every night. Insects caught in each trap were dried, and sorted out on the succeeding day into respective orders and families, with the help of identification keys (Borror et al. 1981). Daily data obtained in respect of the different orders and families was pooled, every 10 days, every month, and at the end of the total period of investigation, for statistical treatment (Federrer 1967). Temperature and relative humidity were recorded continuously using thermohygrograph (Lawrence and Mayo, India).

Another set of experiment was carried out to determine the effect of pest infestation upon mushroom yield. For this purpose, another two rooms of similar dimensions (20' x 10') were used, where cultivation was carried out, as described above. The recommended insecticidal schedule (Munjal and Seth 1980) was applied in one room, while in the other, no control measure was adopted. Total yield/flush was recorded in both cases. Temperature and humidity conditions were similar in both the rooms. All yield values were transformed to square meter area basis, and treated statistically in randomized block design (Federrer 1967).

Data pertaining to relative abundance of insect-pests, with respect to weather parameters are expressed in Table 1. Population was the highest in the middle of February, while the period February - April showed maximum infestation. Though the population fluctuated within this period, it remained on the higher side. On the other hand, population was lowest in January. Insects belonging to five

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TABLE 1. NUMBER OF INSECTS OF DIFFERENT ORDERS AND DIPTERAN FAMILIES OCCURRING ON WHITE BUTTON MUSHROOMS *(AGARICUS BISPORUS)* IN DIFFERENT MONTHS OF THE CROPPING PERIOD.

		Mean insect counts/trap/day during				
		15 Dec - 14 Jan	15 Jan - 14 Feb	15 Feb - 14 Mar	15 Mar - 14 Apr	Average
(A)	**ORDERS; Diptera**	10.50 ± 0.76	9.16 ± 0.88	31.9 ± 0.74	22.4 ± 0.89	18.5 ± 0.53
	Hymenoptera	0.16 ± 0.11	0.08*	0.25 ± 0.12	0.50 ± 0.14	0.25 ± 0.06
	Collembola	-	0.08*	-	0.08*	0.04 ± 0.02
	Colcoptera	-	-	0.25 ± 0.11	0.50 ± 0.18	0.18 ± 0.05
	Homoptera	-	-	0.08*	1.08 ± 0.3	0.25 ± 0.07
(B)	**FAMILY; Dipteran**					
	Sciaridae	0.83 ± 0.18[a]	2.58 ± 0.25[a]	0.42 ± 0.45[a]	8.50 ± 0.57[a]	5.58 ± 0.26[a]
	Phoridae	9.50 ± 0.71[b]	5.50 ± 0.54[b]	11.42 ± 0.55[a]	7.92 ± 0.56[a]	8.58 ± 0.31[b]
	Cecidomydae	0.16 ± 0.11[a]	1.08 ± 0.22[c]	11.00 ± 0.72[a]	6.00 ± 0.46[b]	4.56 ± 0.23[c]

* Very infrequent occurrence

- For family, within a column, values followed by common letter are not significantly different at 0.05% levels of significance

orders viz., Diptera, Hymenoptera, Coleoptera, Collembola and Homoptera infested the crop. (Table 1). Diptera exhibited the highest population in different months, as well as in the entire period (18.5 ± 0.53/trap/day). Dipterans and Hymenopterans were consistent, in occurrence. Dipterans constituted, by far, the major part (94.97%) of the average total insect population. Data in respect of different Dipteran families indicate that insects belonging to three families viz., Phoridae, Sciaridae and Cecidomydae were caught in the light traps. In the first two months i.e., December-January and January-February, Phoridae constituted the highest significant population, while Cecidomydae accounted for the lowest population. In the third month (March-April), Phoridae comprised the highest population, which, however, was not significantly different from that of Sciaridae. Cecidomydae population was significantly lower than that of the above families, in this month also. Phoridae also accounted for the significantly highest population for the entire period, followed by Sciarids and Cecidomiids.

Mushroom yield figures (Table 2) indicate significantly lower yield in the absence of insecticide application, in comparison to insecticide application. Yield was considerably reduced in fourth flush and no yields were recorded from fifth flush in the absence of protective measures.

Results revealed that temperature influenced the population level of the insect pests (Fig. 1) though no such conclusion could be drawn in case of relative humidity, which did not show any appreciable variation during the experimental period. Higher population of Dipterans is in agreement with the findings of other workers (Atkins 1974; Finlay 1984; Shandilya et al. 1975). Infestation by Phoridae, Sciaridae and Cecidomydae is also reported both in India and abroad (Atkins 1974; Finlay 1984; Shandilya et al. 1975; Thapa and Seth 1981). Results indicate that higher ambient temperature conditions are suitable for multiplication of the

TABLE 2. EFFECT OF INSECT-PEST INFESTATION UPON THE YIELD OF WHITE BUTTON MUSHROOM *(AGARICUS BISPORUS)*.

Treatments	Average yield/flush (in kgs/sqm.)							Average total* yield (kgs/sqm.)
	F_1	F_2	F_3	F_4	F_5	F_6	F_7	
No treatment	0.68. ± 0.12	1.14 ± 0.12	1.12 ± 0.08	0.26 ± 0.14	-	-	-	3.04
Insecticidal treatment	0.86 ± 0.10	1.34 ± 0.12	1.34 ± 0.09	0.8 ± 0.08	0.74 ± 0.12	0.60 ± 0.15	0.28 ± 0.02	5.96[a]

* Within the column the means followed by common letters are not significantly different at 0.05% levels of significance.
F : Flush

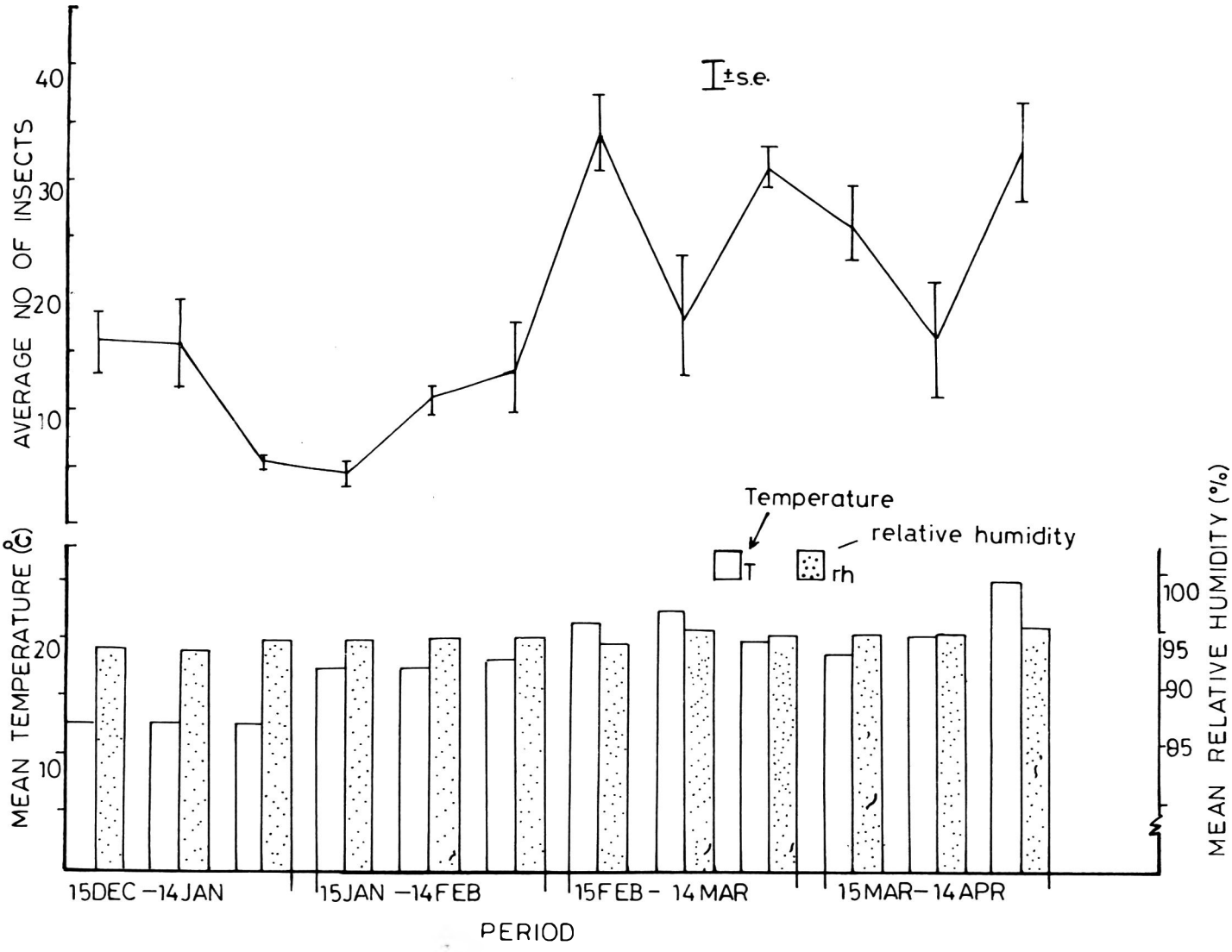

Fig. 1. Total insect population occurring on white button mushroom *(Agaricus bisporus)* in the different months.

insect pests and are most likely to cause economic damage to the crop, in this part of the country. The results also indicate that pest damage becomes more from fourth flush, resulting in serious depletion of yield and no fruiting bodies appear in the fifth flush. This suggests that end of the third flush might be the suitable time for application of pest emergence control measures.

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Effect of Incorporation of Blood Proteins into Sausage

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Effect of utilization of pig blood proteins, such as plasma, whole blood, plasma protein isolate and globin protein isolate, either individually or in combinations, as ingredients and replacements of sausage mixture in *Lecso* sausage formulation was evaluated. Shrinkage was marginal during 8 days of storage at 4 ± 1°C, protein content was higher than the control and *Escherichia coli*, *Salmonella* and *Staphylococcus aureus* were absent in all the samples. Organoleptic qualities were similar to the control, except for the least score for appearance in case of sample with 2% blood. It is evident that incorporation of blood proteins in sausage formulations upto certain levels yields acceptable product from the view point of chemical, microbial and sensory qualities.

Keywords : Blood Proteins, Sausage ingredient, Quality evaluation, Pig blood, Shrinkage, Sensory evaluation.

The availabiltiy of protein foods is becoming scarce in developing countries. The diets of the inhabitants of the developing and underdeveloped countries are sufficient in calories, but deficient in animal protein (Elboushy 1986). Animal protein consumption per person in developing countries is low at 12.8 g and in India, it is only 5.3 g in contrast to 22.4 and 58.5 g in other developing and developed countries, respectively (FAO 1986). It has been shown that about 30252.5 thousand litres of edible blood could be available from slaughtered animals in certified slaughter-houses in India, which amounts to 5779 thousand kg of high quality protein every year (Hazarika 1989). There are also reports that varieties of sausages are made from animal products such as mutton (Selvarajah et al 1974) and pork along with vegetable proteins (Padda and Kondaiah 1983). In the present study, an effort is made to use different blood proteins in *Lecso* sausage, a local Hungarian meat product, as a part replacement for sausage mix and the acceptability of the product is evaluated.

Th edible plasma and blood were collected from the Budapest pig slaughter-house by using Alfa-Laval Semi-automatic blood collecting system. Plasma protein isolate and globin protein isolate were collected from Zala Megyei Livestock Trade and Meat Industry, Zalaegerszeg, Hungary. Proximate composition of the blood protein- incorporated sausage products was studied as per standard methods (AOAC 1970). Microbial quality was determined by following the method of ICMSF (1978) and Kiss (1984). The products were evaluated for sensory qualities by a group of six trained panel members. A 9 point Hedonic Scale (Bratzler 1971) was used to evaluate the samples. For shrinkage study, the sausages were weighed at different stages i.e., stuffing, smoking, cooking, cold showering, and keeping for 20 h at cold room. The shrinkage study was conducted for a duration of 8 days at 4 ± 1°C. Statistical analysis was carried out by analysis of variance test (Steel and Torrie 1960). All the experiments were carried out in four replicates.

The basic formula for preparation of *Lecso* sausage was as specified by Meat Industry Production Norms and Guide for Meat products Ingredients (Leolkes 1980). In the basic formula, the sausage mixture was replaced individually to the same extent by (1) 5% aqueous plasma, (2) 10% aqueous plasma, (3) 1.5% blood, (4) 2% blood, (5) 1.5% plasma protein isolate (PPI), (6) 2.5% PPI, (7) 1.5% globin protein isolate (GPI), (8) 2.5% GPI, (9) 1.5% PPI + GPI (3:1), (10) 2.5% PPI + GPI (3 :1) and (11) control (without blood proteins).

After adding blood proteins, the mixtures were minced for another 30 sec. Each batch was stuffed into 34 mm pig casings. The prepared sausages were conventionally cooked for 20 min at 74°C core temperature, smoked at 72°C for 60 min, given 3 min cold shower and stored at 4 ±1°C for 20 h.

Shrinkage was found to be highest in control, followed by the samples with 10 and 5% aqueous plasma. After 8 days of storage, the shrinkages were significantly ($P < 0.05$) higher in these samples than in other blood protein - added samples. Higher water-binding properties of blood proteins may be responsible for such results. However, final shrinkage losses after 8 days of storage were very nominal in all samples.

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TABLE 1. INCORPORATION OF VARIOUS BLOOD PROTEINS INTO *LECSO* SAUSAGE AND THEIR EFFECT ON PROXIMATE CHEMICAL COMPOSITION AND pH.

Chemical composition	Blood proteins										
	1	2	3	4	5	6	7	8	9	10	11
Moisture, %	55.1 ± 3.3	57.8 ± 2.9	53.7 ± 3.3	53.0 ± 3.2	52.1 ± 2.1	50.3 ± 2.8	52.7 ± 3.0	51.5 ± 2.0	51.5 ± 2.1	51.5 ± 2.0	53.8 ± 1.9
Crude protein, %	16.4 ± 1.2	16.4 ± 1.7	17.3 ± 1.0	16.7 ± 2.0	16.3 ± 1.1	16.8 ± 1.8	17.3 ± 1.2	17.0 ± 1.0	15.4 ± 1.0	15.8 ± 1.0	15.3 ± 1.0
Fat, %	24.2 ± 1.6	24.5 ± 1.9	26.6 ± 2.0	25.6 ± 2.7	27.9 ± 1.8	26.3 ±1.7	26.3 ± 1.7	26.4 ± 1.9	26.0 ± 2.1	26.0 ± 1.9	26.2 ± 1.5
Ash, %	3.0 ± 0.5	2.8 ± 0.8	3.0 ± 0.8	3.1 ± 0.6	3.2 ± 0.3	3.3 ± 0.4	3.1 ± 0.4	3.3 ± 0.5	3.1 ± 0.5	3.3 ±0.6	3.0 ± 0.7
pH	6.5 ± 0.9	6.5 ± 1.1	6.3 ± 0.8	6.5 ± 0.9	6.5 ± 1.2	6.3 ± 1.0	6.4 ± 0.8	6.4 ± 0.9	6.4 ± 0.9	6.3 ± 0.7	6.4 ± 0.9

1 = 5% plasma, 2 = 10% plasma, 3 = 1.5% blood, 4 = 2% blood, 5 = 1.5% PPI, 6 = 2.5% PPI, 7 = 1.5% GPI, 8 = 2.5% GPI 9 = 1.5 PPI + GPI (3:1), 10 = 2.5% PPI + GPI (3:1), 11 = Control.

Results of the proximate chemical composition (Table 1) showed that the samples with added aqueous plasma had slightly higher moisture content than the control and other samples. Slightly higher protein contents, though statistically insignificant in experimental samples, indicate that addition of blood proteins at the levels used in this study has a positive effect on the protein quality. There seems to be less effect on % fat and ash contents. Addition of blood proteins did not change the pH of the sausage products significantly. Microbial analysis of blood protein-added *Lecso* sausages on first day of the storage at 4 ± 1°C showed that, except for the sample with 2% blood, all others including the control had a total count between 10^2 - 10^3/g. No samples contained bacteria of public health importance like *E. coli*, *Salmonella* and *Staph. aureus*. Analysis on 5th and 8th days also revealed that the counts were within the range of acceptable limits. No statistical differences were observed among the means of appearance, flavour, texture and acceptability of the test samples, when compared to control samples. However, lower scores were observed for sample with 2% blood for appearance, flavour and acceptability, which might be due to comparatively darker colour of the products. The panelists preferred all the blood protein-added samples equally, as they did with the control. From this study, it may be concluded that addition of 5-10% plasma, 1.5% blood, 1.5 – 2.5% PPI or GPI or their combinations in meat products would not affect the sensory properties of final product.

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Lipid Composition of *Withania somnifera*, *Phoenix sylvestris* and *Indigofera enualphylla* Seeds of Central India

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Total lipids of *Withania somnifera* (Solanacea), *Phoenix sylvestris* (Palmace) and *Indigofera enualphylla* (Leguminosae) seeds were 14.0, 10.2, and 4.4%, and consisted of neutral lipids (87.7-90.2%), glycolipids (5.8-7.3%) and phospholipids (3.6–5..0%). Neutral lipids consisted of triacylglycerols (87.0 - 88.5%) with small amounts of monoacylglycerols, diacylglycerols, free fatty acids, free sterols, sterol esters and hydrocarbons. The glycolipids were predominantly digalactosylglycerol, and acylated-sterylglucoside with small quantities of sterylglucoside and monogalactosylglycerol. The phospholipids (PL) consisted of phosphatidylcholine(PC), phosphatidylethanolamine (PE), phosphatidyl inositol (PI), and cardiolipin (CL) with trace amounts of lysophosphatidylcholine (LPC) and lysophosphatidylethanolamine (LPE). The fatty acid composition of all the lipid material showed the preponderance of palmitic and oleic acids.

Keywords : Seeds, Total lipids, Neutral lipids, Glycolipids, Phospholipids *Withania somnifera*, *Phoenix sylvestria*, *Indigofera enualphylla*.

Withania somnifera (*Aswagandh*), *Phoenix sylvestris* (*Kharjur*) and *Indigofera enualphylla* (Indigo) seeds are found scattered in Central India. The oils from these seeds are rich in palmitic, oleic and linoleic acids. Fatty acid composition of these seed oils has been reported (Bhakare et al. 1992, Kulkarni et al. 1992), but little information is available on the lipid composition of these seeds. These are reported in this communication.

The seeds were purchased locally. Authentic lipids and fatty acid methyl ester standards were procured from Sigma Chemical Co. (St. Louis, MO, USA). Only analytical grade solvents were used. The total lipids from (triplicate) 10 g samples of crushed seeds were extracted by blending with 300 ml of Folch reagent (Folch et al. 1957) consisting of chloroform and methanol (2:1, v/v) for 4 min at room temperature. The extract was concentrated by a rotary evaporator. A measured portion of the purified extract was used for gravimetric estimation of lipid classes and fatty acids analysis.

The total lipids (TL) were fractionated into neutral lipids (NL), glycolipids (GL) and phospholipids (PL) by silicic acid cclumn chromatography (Rouser et al. 1967), eluting with chloroform, acetone and methanol successively. NL were estimated gravimetrically, while GL and PL were quantitated by estimation of total sugars (Dubois et al. 1956) and total phophorus (Harris and Popat. 1954), respectively using appropriate factors (25.0, 26.2, 25.0, 29.2, 26.3, 17.5 and 16.0% of total phosphorus for PL, PC, PE, PI, CL, LPC and LPE, respectively (Williams et al. 1966).

NL were separated by TLC using hexane-diethyl ether-acetic acid (80:20:1, v/v). Individual components of NL were identified by comparison of R_f values with authentic standards and quantified by photodensitometry (Blank et al. 1954). Individual components of GL and PL were separated by TLC using chloroform-methanol-acetic acid-water (65:15:10:4, v/v) and identified by specific spray reagents and comparison of the R_f values of the standards. Individual GL and PL were extracted with chloroform-methanol-water (10:20:8, v/v) and quantified by sugar and phosphorus estimations as in GL and PL.

Fatty acid methyl esters of the lipids were prepared according to Moharil et al. (1990), and analyzed on a Perkin-Elmer 8700 GLC with a flame ionization detector at 280°C, S. S. column packed with 15% EGSS-X on Chromosorb-W (40-6-mesh) at 200°C and N_2 flow rate 60 ml/min.

The peak area and relative percentage of the methyl esters were obtained by an integrator coupled to the system and a programmed computer. The component of each peak was identified by retention time data comparing with the standards. Mean values of triplicate determinations were reported. The results are reported in Tables 1-3.

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TABLE 1. TOTAL LIPIDS AND NEUTRAL LIPIDS OF SEEDS AND THEIR FATTY ACID COMPOSITION

Seed	Lipid components	Fatty acids (Wt %) •						
		Wt.ª %	16:0	18:0	18:1	18:2	18:3	Others
Withania	TL	14.0	13.7	3.6	21.5	60.0	1.2	-
somnifera	NL	87.7	12.8	2.9	23.1	59.1	2.1	-
	TG	85.4	12.8	3.2	20.5	58.7	2.8	2.0
	MG	0.8	20.7	6.7	18.5	42.1	3.2	8.8
	DG	1.0	16.8	7.2	27.5	40.3	5.0	7.2
	FFA	6.3	18.8	6.5	24.6	38.5	5.6	6.0
	SE	2.5	17.5	5.0	28.8	37.7	4.0	7.0
Phoenix	TL	10.2	30.7	10.1	40.2	15.1	1.8	2.0
sylvestris	NL	89.8	28.7	11.2	41.3	14.8	1.2	2.8
	TG	88.5	28.5	11.2	38.8	17.1	2.0	1.4
	MG	0.9	32.7	12.2	30.4	13.2	6.5	5.0
	DG	1.5	30.7	18.7	29.5	14.3	1.2	4.6
	FFA	5.2	28.9	18.8	27.8	15.4	3.3	5.8
	SE	2.7	29.2	16.0	30.5	16.5	3.0	4.0
Indigofera	TL	4.4	18.9	1.2	8.1	56.5	8.5	6.8
enualphylla	NL	90.2	17.5	0.8	7.8	51.7	5.7	6.5
	TG	87.0	16.7	2.7	10.2	60.8	8.0	1.6
	MG	0.7	23.8	0.4	15.4	50.7	6.5	3.2
	DG	1.7	20.8	1.7	14.8	52.8	5.7	4.2
	FFA	7.1	15.8	1.8	12.8	59.7	4.8	5.1
	SE	2.3	18.4	3.6	18.7	45.8	6.7	6.8

• = Means of triplicate analysis, TL - Total lipids, TG - Triacylglycerol, MG - Monoacylglycerol, SE - Sterol esters, FFA - Free fatty acids, S - (Sterols), H (hydrocarbons) (1.0, 1.2, 1.2%), NL - Neutral lipids, DG - Diacylglycerol 'Others' means 14:0, 20:0 and 22:0 acids, a - on wt. of seeds, dry basis

TABLE 2. GLYCOLIPIDS OF SEEDS AND THEIR FATTY ACID COMPOSITION

Seed	Glycolipid components	Fatty acids (Wt %) •						
		Wt.• %	16:0	18:0	18:1	18:2	18:3	Others
	GL	7.3	20.8	2.0	40.2	30.1	1.2	6.6
Withania	MGDG	21.1	24.0	18.4	28.9	19.0	4.0	5.7
somnifera	DGDG	45.2	28.1	25.0	27.8	19.7	-	-
	ASG	23.5	26.2	18.2	27.2	29.2	-	-
Phoenix	GL	5.8	35.4	5.4	35.7	18.1	1.1	4.3
sylvestris	MGDG	24.1	27.0	8.0	28.0	36.1	0.9	-
	DGDG	40.7	30.2	11.4	27.0	30.9	-	0.5
	ASG	26.7	26.5	10.0	25.1	35.3	2.7	-
Indigofera	GL	6.2	30.1	6.1	25.7	28.7	3.8	5.6
enualphylla	MGDG	22.2	29.0	1.8	27.0	38.8	1.4	2.0
	DGDG	38.7	26.8	1.7	27.0	36.9	1.8	5.9
	ASG	27.6	22.6	1.5	26.9	37.0	2.0	-

• - Means of triplicate analysis, GL - Glycolipids, MGDG - Monogalactosyldiacylglycerol, DGDG - Digalactosyldiacylglycerol ASG - Acylatedsterylglucoside, SG - Sterylglucoside (10.2, 8.5, 11.5%), 'Others' means 14:0, 20:0 and 22:0 fatty acids

TABLE 3. PHOSPHOLIPIDS OF SEEDS AND THEIR FATTY ACID COMPOSITION

Seed	Phospholipid components	Fatty acids (Wt %) •						
		Wt.• %	16:0	18:0	18:1	18:2	18:3	Others
	PL	5.0	21.7	2.1	38.7	31.5	2.1	3.9
Withania	PC	34.0	20.0	2.1	30.0	40.1	5.0	2.8
somnifera	PE	22.4	23.1	3.0	37.4	32.2	3.0	1.3
	PI	24.8	25.5	4.0	29.4	41.1	-	-
	CL	17.8	22.6	6.3	35.7	35.4	-	-
	PL	4.4	37.6	3.8	38.8	12.9	1.0	5.9
Phoenix	PC	38.7	40.1	16.9	19.8	12.1	1.1	-
sylvestris	PE	20.7	30.8	11.2	46.3	11.7	-	-
	PI	27.2	21.2	21.8	40.3	16.7	-	-
	CL	12.7	23.1	20.9	40.3	13.4	2.1	-
	PL	3.6	28.2	1.8	28.5	28.5	8.1	4.9
Indigofera	PC	37.5	30.4	20.1	25.1	20.2	2.1	2.6
enualphylla	PE	25.7	20.8	10.7	47.8	17.9	2.8	-
	PI	29.5	14.9	20.0	43.1	22.0	-	-
	CL	8.7	18.3	17.0	45.0	19.8	-	-

• - Means of triplicate analysis, PL - Phospholipids, PC - Phosphatidylcholine, PE - Phosphatidylethanolamine PI - Phosphatidylinositol, CL - Cardiolipin, LPC - Lysophosphatidylcholine (0.7, 0.4, 0.4%), LPE - Lysophosphatidylethanolamine (0.5, 0.3, 0.2%), 'Others' means 14:0, 20:0 and 22:0 acids.

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Microflora of Fresh Cut Vegetables Stored at Refrigerated and Abuse Temperatures

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The microflora of commercially packaged spinach, cauliflower and carrots was compared after storage for 7 days at 3.3 and 15°C. Storage at 15°C produced significantly high aerobic counts in spinach, cauliflower florets and carrot sticks, but not in cauliflower heads and whole carrot. Psychrotrophic species made up a significant proportion of the microflora at both the temperatures. Little growth of coliform and lactic acid bacteria was observed at either temperature.

Keywords : Microflora, Fresh cut vegetables, Storage, Refrigerated temperature, Abuse temperature, Spinach, Cauliflower, Carrot.

Fresh cut vegetables are popular with consumers in the United States because these require little labour in their use and minimal or no waste is generated in the kitchen. However, the cutting, peeling and removal of outer leaves result in exposed surfaces, which support the growth of microorganisms (Maxcy 1982). It is critically important, therefore, that the fresh cut produce be stored at the lowest refrigeration temperatures to prevent the physiological damage to the plant tissue (King and Bolin 1989). In this study, the microflora of produce stored at 3.3°C was compared with that stored at an abuse temperature of 15°C.

Packaged fresh cut vegetables were collected from J. C. Brock Company, Buffalo, New York. These were transported to the laboratory (at Geneva, NY) over ice and stored at desired incubation temperatures. The spinach was packaged in 1.25 gauge polypropylene with 4 holes, while the carrots and cauliflower were sealed in 1.75 gauge linear low density polyethylene (3% EVA) packages. The vegetables were cultured after 1 and 7 days of storage. Approximately, 20 g were diluted (10^{-1} w/w) in water and then homogenized for 1 min in a Waring blender. Subsequent decimal dilutions were made in 0.1% peptone water. Pour plates of plate count agar (Difco, Detroit, MI) were prepared from aerobic plate counts and the enumeration of psychrotrophs. The former were incubated for 2 days at 30°C, while the latter were incubated for 10 days at 3.3°C. Coliforms were enumerated on Difco violet red-bile-agar plates incubated for one day at 35°C. Lactobacilli MRS agar (Difco), containing 0.02% sodium azide, was used to determine lactic acid bateria at 30°C for 2 days (Mundt et al. 1967).

The response of aerobic plate count to the abuse storage temperature varied with the type of produce (Table 1). Spinach stored for 7 days at 15°C yielded population, which is about 100 - folds higher than those obtained at 3.3°C. The cauliflower heads and whole carrots, on the other hand, showed somewhat lower counts, when stored at higher temperature. Contamination of these vegetables would be primarily on their surface and it is possible that dehydration of the surface at the higher temperature restricted the microbial growth (Brackett 1987). The amount of growth was proportional to the amount of exposed surface of different products. Thus spinach, carrot sticks and cauliflower florets showed higher counts when stored at both 3.2° and 15°C. A significant proportion of the aerobic population consisted of psychrotrophic bacteria. Our earlier study (Garg et al. 1990) as well as the observations of others (Brocklehurst et al. 1987; King and Bolin 1989; Magnuson et al. 1990) have shown that these are primarily the species of *Pseudomonas* and other oxidative Gram negative rods. The proportion of psychrotrophs changed little following storage at 3.3 and 15°C, thereby indicating that these organisms competed well at both storage temperatures.

With the exception of spinach, there was little increase in coliform counts on violet red-bile agar. This indicated that either these vegetables were not good growth media for coliforms even when held at the higher temperature or some sub-lethal injury has occurred to the cells during storage in ice, thereby leading to their non-resuscitation. The lactic acid bacteria can inhibit certain foodborne

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TABLE 1. MICROFLORA OF VEGETABLES STORED AT 3.3° AND 15°C

Vegetable	Temperature, °C	Storage, days	Plate count/g			
			Acrobes	Psychrotrophs	Coliforms	*Lactobacilli*
Spinach, 1		0	1.2×10^4	7×10^4	48×10^3	0
	3.3	1	30×10^4	36×10^4	58×10^3	0
		7	58×10^5	35×10^5	95×10^2	50×10^1
	15	7	52×10^7	52×10^7	5×10^4	65×10^2
Spinach, 2		0	1.4×10^4	11×10^4	38×10^3	0
	3.3	1	19×10^4	15×10^4	43×10^3	0
		7	83×10^6	40×10^6	65×10^2	15×10^1
	15	7	52×10^8	16×10^9	55×10^4	40×10^2
Cauliflower,		0	11×10^3	30×10^2	$\leq 10^1$	0
head	3.3	1	13×10^3	38×10^2	$\leq 10^1$	0
		7	54×10^4	30×10^4	$\leq 10^1$	50
	15	7	16×10^4	18×10^4	$\leq 10^2$	80×10^2
Cauliflower		0	18×10^3	45×10^2	$\leq 10^1$	0
florets	3.3	1	23×10^3	55×10^2	$\leq 10^1$	0
		7	11×10^5	99×10^4	$\leq 10^1$	50
	15	7	26×10^6	17×10^6	20×10^1	35×10^1
Carrot whole		0	56×10^3	41×10^3	19×10^2	0
	3.3	1	69×10^3	47×10^3	21×10^2	0
		7	25×10^5	15×10^5	20	22×10^2
	15	7	86×10^4	49×10^4	25×10^2	$\leq 10 \times 10^2$
Carrot sticks		0	96×10^3	44×10^3	70×10^1	0
	3.3	1	98×10^3	51×10^3	85×10^1	0
		7	94×10^5	16×10^6	30	50
	15	7	16×10^6	16×10^6	15×10^1	95×10^2

pathogens and spoilage bacteria through their ability to produce bacteriocin, hydrogen peroxide, and organic acids (Gilliland and Speck 1975; Price and Lee 1970; Spelhaug and Harlender 1989; Visser et al. 1986).

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Advances in Baking Technology : Edited by B. S. Kamel and C. E. Stauffer, Published by Blackie Academic and Professional, an imprint of Chapman and Hall, Wester Cleddens Road, Bishopbriggs, Glasgow G 64 2NZ, UK, pp 408, Hardcover 1993, Price £79.00

Wheat is the staple food for almost 35% or more of the world's population, in as many as 43 countries and is the most important foodgrain of the world. Mainly, wheat is consumed in the form of bread and other baked products. The transition of the bread making processes from the conventionally considered 'art' to a 'technology' has occurred unnoticed. The advances in baking technology, which took place in the past decades have been presented by the editors in 14 chapters contributed by professionals. It covers bread and other wheat flour-based bakery products, their quality, ingredients used, processes and processing machinery.

The chapter on wheat, presented at the very beginning, very lucidly brings out the different terms used in the wheat industry and distinguishes the different varieties grown in the world at different geographical locations and in different seasons.

Technology of bread making has been very elaborately handled by Brown, a practical bakery technologist. In this chapter, besides presenting the basic bread manufacturing processes, he goes on to describe the newer developments in machinery, processing and the ingredients recommended for use in bread manufacture to provide technological advantages and improvement in product identities. The different types of ovens and their merits and demerits have been discussed at length.

In the bread making process, the technology aspects demand certain minimum quality standards for inputs. The specific test methods to evaluate the dough characteristics are discussed in the chapter contributed by Rasper on 'Dough rheology and physical testing of dough'. The recent developments employing advanced rheometric techniques are described elaborately leaving you with the question : Will they prove to be more suitable for quality testing work than the popular empirical and imitative test?

Product quality measurements such as sensory evaluation, instrumental methods of texture measurement etc., have been covered in separate chapters.

Topics such as enzymes, dough improvers, emulsifiers, lecithin etc., are presented separately and would provide the scientific understanding of various additives, now being used by the baking industry. Benefits derived by use of these additives have been presented. The chapter on 'Emulsifiers in baking' presented by Kamel and Ponte and 'Lecithin and phospholipids in baked goods' by Silva takes the reader to the basics of emulsifiers and reveals their beneficial effects to the bakery products. Such treatises on specialized topics directed to the target group, 'the bakery technologists' can seldom be seen elsewhere.

Developing alternate technologies to conventional baking like microwave technology, extrusion technology etc., and the new product preference such as low calorie, high fibre, low fat, low sodium etc., have been included. They provide insights into the future of the baking technology with respect to processes and product quality.

In short, it is a "book written by an International team of authors from the industrial and academic sectors who review recent advances in baking industry with particular emphasis on developments in ingredients and processes" as claimed by the publishers.

This book is a must to the baking industry personnel, be it in production, research or teaching. The editors and the contributing authors deserve sincere appreciation for their efforts in bringing out this volume.

At the end of going through the book, one will be tempted to go back and read the preface by the authors. 'Fundamentally, baking may seem to be a rather static topic: mix flour, water, leavening and various flavouring materials and heat the mixture to gelatinize starch and denature the protein'. But, the dynamic developments in the past decades brought out in this volume make us change this view.

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Food Taints and Off-Flavours : Edited by M. J. Saxby, Blackie Academic Professional, Wester Cleddens Road, Bishopbriggs, Glassgow G 64 2 NZ, UK, 1993: pp. 260; Price : £69.

The problem of taints and off-flavours in foods is a major concern not only to food processing industry, but also to all other agencies involved in the production of raw materials, packaging, transportation, distribution and sale of foods. While

taints are generally caused through contamination by a malodorous substance totally alien to all foods, off-flavours result through the formation of compounds having undesirable taste and aroma through chemical or biochemical reaction of food constituents or by microbial degradation. Because of commercial importance of taints and off-flavours in foods, these have been investigated widely and a number of reviews and books have been published mainly on the causative reactions and kinetics involved in off-flavour development, major factors which influence the rate of formation of off-flavours and the nature of compounds formed during these reactions. Since a very large number of volatile and non-volatile products have been isolated and characterised, progress on identification of compounds actually reponsible for off-flavours and concentrations at which off-flavours become perceptible has been rather slow. Only recently, simultaneous anlaysis of volatile compounds by GC-MS and sniffing and other sensory tests have enabled identification of some of the character impact compounds involved in imparting taints and off-flavours to foods and packaging materials.

The present book entitled "Food taints and off-flavours' edited by M. J. Saxby has tried to present the information on the sources of taints and off-flavours, threshold concentration of compounds necessary for perception of taints and off-flavours, chemical and biochemical interaction involved and some of the methods employed in characterising taints and off-flavours and some guidelines for their prevention.

The book contains nine chapters; the first chapter providing information on definition of taints and off-flavours and various sensory tests employed for their detection and quantification. The second chapter reviews information on various chemicals and their transformations in imparting taints and off-flavours in food, water and packaging materials. The third chapter describes various techniques employed for isolation and characterisation of compounds causing taints and off-flavours. The fourth chapter describes off-flavours associated with potable water, their probable sources and the efficacy of various water purification treatments on the removal of taints. The fifth chapter describes the various types of undesirable flavours in dairy products and provides considerable information on flavour descriptions normally encountered in sensory testing and the origin of these flavours.

Autoxidation of lipids whether brought about by atmospheric oxygen or through enzymes is a major cause of off-flavours formed during processing and storage of a large number of foods. Sixth chapter describes briefly the mechanism of autoxidation of polyunsaturated fatty acids, their reaction products along with some of the off-flavours associated with aldehydes, ketones, hydrocarbons, alcohols and acids. Seventh chapter provides useful information on taints associated with the packaging materials, sensory and instrumental techniques employed to measure these taints and the causes of taints in paper and plastic packaging materials, including the role of printing inks and adhesives. This chapter provides very useful information to all those who are involved in development and quality control of packaging for food and beverage industry.

The eighth chapter describes the role of retailers in disseminating the information about outbreak of taints and off-flavours in branded products, while ninth chapter highlights the off-flavours derived as a result of microbiological and enzymic actions in foods. The information, though not exhaustive, does provide the nature of taints and off-flavour that can arise as a result of microbial action.

The book provides very extensive literature survey, which can form a basis for future research for people working in academic and research institutions, while to those involved in commerical marketing and quality control in food and beverage industry, it gives useful information on how foods can become tainted, and how to design protocols to prevent the problem of taints and off - flavours during manufacture and storage.

Though the present volume has covered taints and off-flavour problems associated with dairy products, water, packaging materials and to a certain extent in fats and oils, information on taints and off - flavours encountered in meat, fish and poultry and fruits and vegetable industries has not been provided.

Overall, the information has been presented in an expertly and precise manner and the present volume is a useful addition in the field of flavour analysis of foods.

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The Miracle of Germinated Cereal Powders : Proceedings of the International Workshops and Publications, by Tara Gopaldas and Suneeta Deshpande (Eds), Daya Publishing House, Delhi-110 035, 1992: pp. 281, Price : Rs. 600/-

This book presents a comprehensive account of work conducted in the area of development of weaning foods, utilising germinated cereal powders.

The book is organized into four sections. The first section contains the proceedings and papers presented by invited experts at the international workshops on amylase-rich foods (ARF) held at Seoul and Baroda, respectively. Section II contains twelve research publications on studies conducted on ARF from 1980 onwards at the Department of Foods and Nutrition, the Maharaja Sayajirao University of Baroda. Section III comprises abstracts of dissertation work or work in progress of four Doctoral and eighteen Masters' students of the same department on ARF. Section IV tells the reader 'How to make ARF' and 'How to make simple weaning gruels'. The authors have illustrated the book with photographs, illustrations and sketches.

Four papers presented at the Seoul workshop, review the role of traditional foods such as fermented and malted foods, for feeding weaned infants. It was reported that reduction of viscosity of cereal gruels by addition of small quantities of ARF increased energy intake and growth of children in community feeding studies. This workshop made a number of recommendations for further work on malted foods to improve their consumer acceptability and safety to the consumer. In the second workshop held at Baroda, demonstrations and discussions on the use of ARF in reducing the viscosity of weaning foods were given more emphasis than the theoretical formal lectures. The need was stressed to undertake further work on ARF technology to other nutrition and health - related applications, where high viscosity and dietary bulk might pose problems.

A major part of this book is devoted to the presentation of research publication from the Department of Foods and Nutrition, University of Baroda, emanating from investigations on ARF from 1980 onwards. Two publications present data on the superiority of malted cereal-pulse-oilseed formulations over their roasted counterparts, as observed from enhanced intake of the malted mixes by children and also their higher nutritional quality, as revealed by animal experiments. Seven other papers describe in detail, how small quantities of ARF can effectively reduce the bulk of weaning gruels based on wheat, rice, maize, jowar or soya-fortified bulgur wheat. The potential for commercial production of ARF and making it available at low cost to low income group families with weaning age children, appears promising from another study on the transfer of ARF technology from laboratory to urban slums. Well controlled feeding trials conducted over a period of six months on slum children, reported in two other papers, demonstrate how a low bulk wheat gruel containing ARF, can greatly enhance the food energy intake and growth performance during the weaning period.

This book will be of immense interest to paediatricians, R&D scientists and national/ international agencies engaged in nutrition programmes, as it contains detailed suggestions on the urgent problem of providing adequate nutrition to weaned infants and preschool children in developing countries of the world and to prevent malnutrition in this section of population. It will prove an excellent addition to any library.

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INDIAN FOOD INDUSTRY

A Publication of Association of Food Scientists and Technologists (India).

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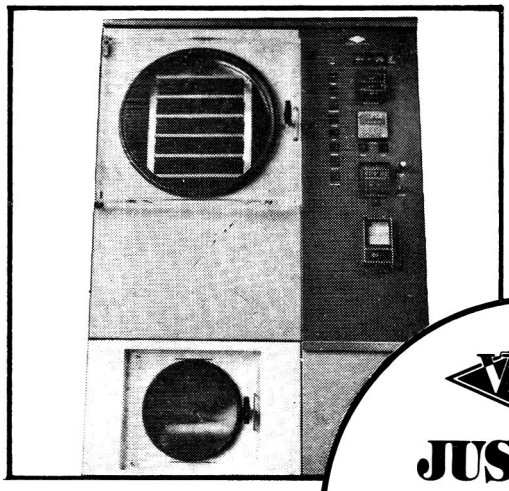

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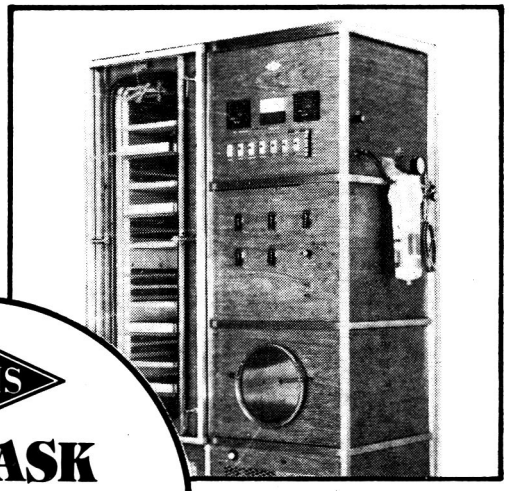

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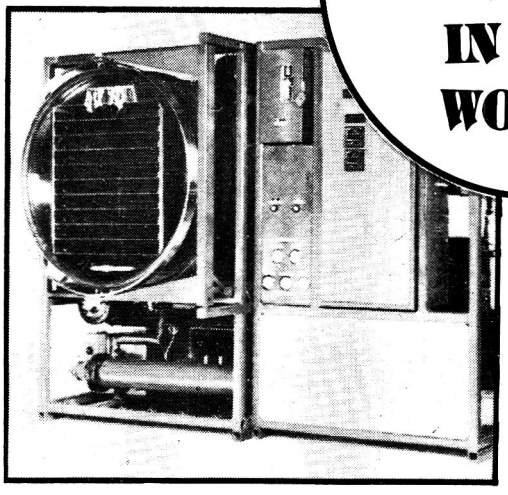

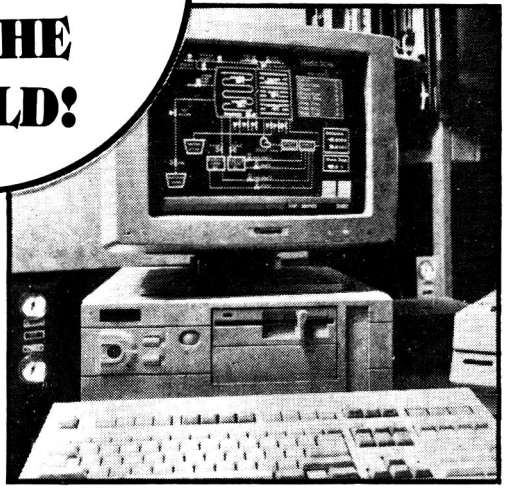

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