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ASSOCIATION OF FOOD SCIENTISTS AND TECHNOLOGISTS (INDIA) MYSORE - 570 013

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- Advancement of all the aspects of Science and Technology relating to production, processing and distribution of food, with the ultimate objective to serve humanity through better food.
- Promotion of research, development and training in the science, Technology and Engineering of Food.
- To provide a forum for exchange, discussion and dissemination of knowledge and current developments, especially among Food Scientists and Technologists as well as the Public and Society at large.

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C O N T E N T S

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Methods for Determination of Sensory Quality of Foods : A Critical Appraisal

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In a modern food corporate, sensory evaluation plays a critical role. It is one of the simplest analytical tools for monitoring quality control at all stages of food product processing, starting from the inspection of incoming raw materials to surveillance of their finished product. A successful implementation of sensory programme requires proper laboratory facilities, trained sensory panels, and adoption of precise sensory methods. A sensory panelist works as an analytical instrument, hence should be carefully selected, and rigorously trained so as to obviate inconsistency in results. Sensory tests are broadly classified into three groups, namely, discriminative, descriptive, and affective (acceptance). The discriminative test is a powerful method in terms of its sensitivity, reliability, and validity. It is more frequently used for selection, and training of panelists, and usually precedes other testing. Descriptive analysis is a sophisticated concept in the sensory evaluation of product, which has evolved from expert opinions for a more rigorous, and scientific approach to measuring perceptions. The important applications of descriptive analysis include storage stability and shelf-life studies, new product development, quality control, and establishment of correlation between sensory and physico-chemical tests. The sensory acceptance represents the third, and a final phase of test resources, which also refers to consumer studies. It provides continuity between the controlled laboratory environment, and the typical product use situation.

Keywords : Sensory evaluation, Panel selection, Discriminative method, Descriptive analysis, Scoring, Acceptance, Testing.

Definition of sensory evaluation

It has long been recognized that enjoyment of food is essential for good health. Enjoyment would mean choice, and acceptance, and not always nutrition, and wholesomeness (Solms and Hall 1981). The consumer's appreciation of food quality is, thus, all important. For consumers, the perceivable sensory attributes, colour, appearance, feel, aroma, taste, and texture are the deciding factors in food acceptance. According to the Sensory Evaluation Division of the Institute of Food Technologists (Anon 1975), the sensory evaluation is defined as a scientific discipline used to evoke, measure, analyze, and interpret results of those characteristics of foods, and materials as they are perceived by the senses of sight, smell, taste, touch, and hearing. The definition makes clear that sensory evaluation encompasses all the senses, and not taste testing alone. Sensory evaluation takes into account several different disciplines, which include experimental, social, and a working knowledge of food science and technology, and it emphasises the behavioural basis of perception. As the definition implies, sensory evaluation involves the measurement, and evaluation of the sensory properties of foods, and other materials (Stone and Sidel 1993). Therefore, sensory evaluation helps in ensuring that the consumer gets consistent, non-defective, and enjoyable foods.

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Importance of sensory evaluation

With the tremendous growth of economy, and competition, the developments in the food processing industries, the high cost of research, and development, and marketing, and the high failure rates of new products led the food scientists, and technologists hastily to turn to sensory responses, as validating tests for better prediction of success. Sensory evaluation is, therefore, considered to be an important analytical tool in the present day competitive corporate environment. Measuring the sensory properties, and determining the importance of these properties, as a basis for predicting acceptance by the consumer, represent major accomplishments for sensory evaluation (Bodyfelt et al. 1988). Though sensory evaluation is utilized by many of the companies, the methodologies adopted are not uniform, clear, and precise. The survey done by Brandt and Arnold (1977), and by the Institute of Food Technologists, Sensory Evaluation Division (Stone and Sidel 1993), revealed that sensory evaluation, as a science is not well understood, methods are not always used in appropriate ways, results are misused, and there is a dearth of qualified professionals in the field. The sensory evaluation science has, therefore, not achieved a status commensurate with its potential. This article focusses on a systematic approach to the organization, development, and operation of a sensory programme in a business, and research environment.

Applications of sensory evaluation

The modern day concept of total quality control involves application of sensory evaluation at all stages of processing, right from procurement of raw materials upto the packaging of the finished product (Reece 1979). The sensory evaluation can contribute, directly or indirectly, to a number of activities (Stone and Sidel 1993), such as (a) new product development, (b) product reformulation/cost reduction, (c) monitoring competition, (d) quality control, (e) quality assurance, (f) product sensory specification, (g) raw material specification, (h) packaging materials suitability, (i) storage stability, (j) process/ingredient/analytical/sensory relationships, and (k) advertising claims.

Organizing a sensory evaluation programme

A successful implementation of sensory evaluation programme requires three major components: proper laboratory facilities, sensory panels, and rigorous training programme (Reece 1979).

Facilities : The literature on sensory facilities has been limited. More recently, American Society for Testing and Materials (Eggert and Zook 1986) published a monograph on the design of a sensory facility, including guidelines for the size of the facility, and suggested booth dimensions, and related information. The sensory facility is as important as any instrument, and therefore, needs serious consideration. Of particular importance are ventilation, lighting, traffic patterns, and locations, product preparation, subject communications, and experimental comfort. In most cases, a facility is ideally described as a quiet area free from distraction, with controlled lighting, partitions between subjects to minimize visual contact, neutral colours for walls, and odour-free surroundings (Stone and Sidel 1993; Bennet et al. 1956).

The sensory laboratory set-up normally consists of a reception-cum-briefing room, panel booths and sample preparation room. The panel booths should be of identical design. Each booth may be 75 to 80 cm wide (ASTM 1968; Stone and Sidel 1993), having adequate space to keep samples, rinsing water, and score card. Stone and Sidel (1993) do not recommend provision of a small sink for oral rinsing in the booth. They consider that sinks are sources of odours and noise, require regular maintenance and increase the construction costs. They have suggested the use of covered, disposable containers for expectorations, as an alternative to sink. The lighting of booths should be uniform, glare-free, and arrangements should be made to provide white or coloured light. According to Stone and Sidel (1993), the lighting in the booth area should be fluorescent, except in the booths themselves, where incandescent lighting is recommended. This lighting should be sufficient to provide 100-110 foot candles of shadow-free light at the counter surface. Some workers (Laue et al. 1954; Kefford and Christie 1960) have favoured the coloured illumination to mark the differences in the colour of the samples. On the contrary, Stone and Sidel (1993) observed that the use of other than conventional lighting caused more problems than it appeared to solve and hence they did not recommend the use of coloured lights.

The ventilation for sensory room, and specially for booths is critical, and needs special attention. The entire booth area should have a slight positive pressure relative to other areas, and the air turn over in the booths should occur every 30 seconds, as a minimum, particularly for strong odours, and fragrant products (Stone and Sidel 1993). In general, odour-free environment in booths has been suggested by many authors (Amerine et al. 1965; Bodyfelt et al. 1988; Stone and Sidel 1993). A temperature of 20-25°C, and relative humidity of 62% in the testing room are considered to be ideal (Hopkins 1954).

Nakayama and Wessman (1979) observed that sensory facilities need not be, as elaborate or, as large as those in an R&D centre. The emphasis is on conducting tests that produce valid, reliable, and reproducible results. They recommended that the facilities might be divided into two distinct areas - one for sample preparation, and another for evaluation - with appropriate lighting, ventilation and temperature controls so as to create an environment conducive to preparation and evaluation. Stone and Sidel (1993) emphasized that the laboratory facility should be flexible enough to handle current and future testing activities as well as to provide a workable environment for the staff. The use of computers has been recommended in sensory evaluation by many workers (Brady 1984; Brady et al. 1985; Gordin 1987; Guinard et al. 1985; Russel 1984; Savoca 1984; Winn 1988). In that case, sensory evaluation laboratory should include space for data processing equipments, which include digitizer, personal computer (PC) with colour display, a storage peripheral, printer/plotter, and modem, if one is linked to a mainframe. If paper score cards are used for sensory evaluation, then a digitizer system is most efficient, flexible and

least costly. The digitizer converts the marks on score cards to numerical values, each value is displayed on the screen of the microcomputer, and then transmitted to storage. After all the data are entered and hard copy obtained from printer, the appropriate software is entered into the microcomputer, and the analysis is completed, and results printed (Stone and Sidel 1993). One can also use direct data entry using an electronic score card. A recent introduction is the use of score sized tablets that display a score card page (Billmeyer and Wyman 1991).

Sample size : Many factors such as inherent characteristics, preparation factors, and serving conditions influence the conduct of sensory tests (Amerine et al. 1965). The Sensory Evaluation Committee of ASTM (1968) has recommended the amount of sample for various sensory tests. In fact, the size, and the amount of the sample for one judging session vary from product to product (Amerine et al. 1965). Kefford and Christie (1960) recommended limiting the number of samples per session to three to eight, depending on the intensity of the flavour, and the judging capacity. If interest can be maintained, panel members can evaluate rather large number of samples per session especially with bland or mild flavoured foods (Kramer et al. 1961). Pangborn et al (1964) presented 100 samples of odorous material of low intensity without fatigue. For economic efficiency of sensory testing, the larger the number of samples per session, the better it will be (Pfaffmann et al. 1954).

Serving temperature : As a general rule, the samples should be served at a temperatures at which they are normally eaten (Laue et al. 1954; Larmond 1977). Serving temperatures for many foods have been suggested by various investigators (Caul 1957; Bodyfelt 1988; Stone and Sidel 1993), and the Sensory Evaluation Committee of ASTM (1968). The higher temperature than normal has been recommended for edible oil and ghee (44°C), and for hot foods (60-66°C) (Larmond 1977; ISI 1975b), and freezing temperature (–15°C) for ice cream, and hard frozen desserts (Bodyfelt 1988). The general principle must be that the temperature, used should be optimum for detecting differences that are under study (Amerine et al. 1965).

Sensory evaluation personnel

Nakayama and Wessman (1979) have recommended three levels of personnel - one at the Research, and Development level, and two at the plant levels. The details of these are given below :

(i) Sensory leader : He/she provides the expertise to the programme in terms of ensuring the application of valid sensory techniques, training the plant sensory coordinator on how to examine samples from production, and coordinating, and communicating sensory information from plant to plant, thus unifying, and standardizing the programme as much as possible.

(ii) Plant sensory coordinator : He/she has full time responsibility at the plant location for implementing and coordinating the programme. He or she is responsible for designing, and conducting sensory tests, and for analyzing, interpreting, and reporting the sensory data. The individual is regarded not as the expert taster, but rather, as the trainer of sensory methodology at the plant location.

(iii) Sensory panelists : They conduct the routine sensory evaluation. The panelists are selected, and trained by the coordinator depending on the type of the product. The actual number of panelists is dependent upon the plant size, and the situation. The Sensory Evaluation Committee of American Society for Testing and Materials (1968) has classified the sensory panels into three categories viz., trained, semi-trained, and consumer panel, which are described below :

(a) Trained panel : They should be carefully selected, and trained, and need not be expert panelists. Trained panel should be used to establish the intensity of a sensory character or overall quality of a food. The trained panels should be small in number, varying from 5 to 10 and may be used in all developmental and processing studies. A small highly trained panel will give more reliable results than a large untrained panel.

(b) Semi-trained panel : This type of panel should be constituted from persons normally familiar with quality of different classes of foods. This panel is capable of discriminating differences, and communicating their reactions, though it may not have been formally trained. In a semi-trained panel, individual variations can be balanced out by involving greater number of panelists. The panel, should normally consist of about 25 to 30 members, and should be used as a preliminary screening programme to select a few products for large scale consumer trials.

(c) Consumer panel : The members of the consumer or untrained panel should be selected at random from the potential consumers in the market area. The number of panelists should be

large enough to ensure due representation to different age, sex, race, and income level groups in the potential consumer population in the market area.

In addition to above personnel, a statistician, who helps in selecting a suitable sampling procedure, evaluation card (score card), experimental design, and statistical methods, should also be included (Amerine et al. 1965).

Selection of sensory panelists

The methods for selection, and training of the sensory panelists have been given by several workers (ISI 1976; ASTM 1968; Cross et al. 1978; Amerine et al. 1965; Bodyfelt 1988). Larmond (1977) has made the following observations regarding choosing, and training of panel.

The panel is the analytical tool in sensory evaluation. The value of this tool depends on the objectivity, precision, and reproducibility of the judgement of the panelists. Before a panel can be used with confidence, the ability of the panelists to reproduce judgements must be determined. Interest, motivation, general attitude, and emotional state of the panelists may be responsible for reliable, and valid judgements.

Amerine et al. (1965) have suggested that the persons who serve, as panelists should be in good health, and should absent themselves, when suffering from conditions that might interfere with normal functions of taste, and smell. Emotional factors, interest, and motivation appear to be more important than the age or sex of a panelist (Giradot et al. 1952; Peryam 1958). It is generally recommended that panelists refrain from smoking, chewing gum, eating or drinking for at least 30 min before the test (Larmond 1977).

According to Stone, and Sidel (1993), the motivation of the panelist affects his response to a great extent. An interested panelist is always more efficient. The panelists should be made to feel that they are an important activity, and that their contribution is very important. There must be a proper programme for panel selection so as to improve the general level of sensitivity, to match panel with problem, and to increase one's confidence in the conclusions derived from the results. A successful testing programme involves employees from all sections of an organization (Nakayama and Wessman 1979). Once individuals have indicated their willingness to participate, they are required to participate in a series of screening tests to determine their skills. This screening will include completion of a product attitude survey, and participation in a series of selected sensory tests. Once qualified, their performance needs to be monitored. The first step is to have each individual complete the survey form, which invariably includes a limited amount of demographic, and background information, such as age groupings, general job classification, sex and special product requirements, such as food liking or allergy.

A general approach for screening, as summarized stepwise by Amerine et al. (1965) is (a) use, as test materials, the same product that will be tested later, (b) prepare tests to obtain variations in the product similar to those, which will be met within the actual experiment, (c) adjust the difficulties of the test so that the group, as a whole will discriminate between samples, but some individuals will fail; (d) use score card similar to those to be employed later, (e) start with, as large a group of panelists, as is feasible, and with a selection test that is operationally simple, (f) screen on the basis of relative achievement, continuing until a top-ranking group of the size desired may be reliably selected, and (g) at each stage reject those who are inadequate and insensitive.

Screening, and selection of a subject, based on sensory skills can be achieved in several ways. The tests suggested by various workers include difference tests; ranking, scoring, descriptive preference, and sequential analysis (ISI 1976; Bradley 1953; ASTM 1968; Dawson et al. 1963; Giradot et al. 1952; Mackey and Jones 1954; Sawyer et al. 1962; Amerine and Roessler 1983; Zook and Wessman 1977; Stone and Sidel 1993).

Most investigators have suggested specific tests based on (a) discriminating differences between solutions or substances of known chemical composition, (b) ability to recognize flavours or odours, (c) performance in comparison with other panel members, and (d) ability to discriminate differences in samples to be used later in the test (Amerine et al. 1965). It was clear that an individual's sensitivity to sample aqueous solutions of sweet, sour, salty and bitter stimuli had no meaningful relationship to subsequent performance in the evaluation of a typical food, and beverages (Stone and Sidel 1993).

The individuals are provided with a tolerance range in the form of graphic illustrations, terms, definitions, and reference samples (Nakayama and

Wessman 1979) during training. The panelists are trained for desirable, and undesirable sensory attributes of the concerned food product (Bodyfelt et al. 1988). They are trained to identify, and measure the intensity of each attribute on a score sheet. Normally, a discrimination test is preferred for these activities. Many randomly selected samples of the product are evaluated by the panelists before he or she becomes qualified to make sensory checks (Nakayama and Wessman 1979). To confirm the panelists' performance, panel difference test, and just noticeable difference test have been suggested (Galanter 1962). The major point to be emphasized with regard to panelist qualifying is not the rigid adherence to one product or test procedure exclusively, but rather the provision of some diversity, and making the job enjoyable for the subject. The importance of teaching individuals to gain confidence in their ability, as sensory panelist is of paramount importance (Stone and sidel 1993).

Sensory tests

Sensory tests may be conducted to meet the five purposes (Amerine et al. 1965), such as (a) select qualified judges and study human perception of food attributes, (b) correlate sensory with chemical, and physical measurements, (c) study processing effects, maintain quality, evaluate raw material selection, establish storage stability or reduce costs, (d) evaluate quality and (e) determine consumer reaction.

Each of these purposes requires appropriate tests. There are substantial number of test methods, and new methods continue to be developed. Stone and Sidel (1993) have classified these methods into three broad categories, viz. discriminative, descriptive, and affective (Table 1).

Discriminative testing : This is one of the most useful analytical tools available to the sensory professionals (Stone and Sidel 1993). It is on the basis of a perceived difference between two products that one can justify proceeding to a descriptive test in order to identify the basis for the difference (Larmond 1977). Within this general class are a variety of specific methods, e.g., (a) paired comparison test, (b) duo-trio test, (c) triangle test, (d) multiple sample test, and (e) other test methods such as dual and multiple standard tests.

TABLE 1. CLASSIFICATION OF DIFFERENT SENSORY TESTS

Category	Test type
Discriminative	Difference : Paired comparison, duo-trio, triangle, multiple sample
Descriptive	Descriptive analysis : Flavour and texture profile, quantitative descriptive analysis
Affective	Acceptance preference : 9-point Hedonic

Adapted from Stone and Sidel (1993)

The main objective of all these methods is to answer a simple question, are these products perceived, as different? Obviously, the response to this question can have major consequences. If the conclusions from a discrimination test are to be accepted by management, as reliable, valid and believable, then it is important that each test be conducted with proper consideration for all aspects of the test design, product preparation and handling, implementation, data design analysis and interpretation (Roessler et al. 1948). Failure to appreciate all the subtleties of a test increases the risk of data misinterpretation.

Paired comparison test : The paired comparison procedure is the earliest example of the application of discrimination testing to food and beverage evaluation (Amerine et al. 1965). Cover (1936) described its use in the evaluation of meat. Since then, it has been used extensively in various foods (Peryam and Swartz 1950; Bradley 1963). It has also been used successfully for determinations of threshold for basic taste solutions (Pangborn 1959).

The paired comparison test is a two product test, and the panelist's task is to identify the one that has more of a designated characteristic such as sweetness, tenderness or skininess (Stone and Sidel 1993). In this method, a directional component could also be added which will alter the panelist to a specific type of paired test. The paired comparison test is relatively easy to organize, and implement. The two coded products (AA, BB, AB, BA) are served simultaneously, and the subject has to decide whether there is any difference or not. Requiring a difference response in all cases has been found to give better results (Gridgeman 1959).

Another version of the paired test is the A-not-A procedure (Peryam 1958; Pfaffmann et al. 1954). The subject is presented with a single sample for evaluation, which is then replaced by the second sample. The subject then makes a decision, as to whether the products are the same or different. This particular test procedure has considerable merit in those situations, where non-test variables such as a colour difference may influence results.

Duo-trio test : This is a modified paired presentation developed by Peryam and Swartz

in finding out those product variables, that are different, and from which one can establish the cause, and effect relationships.

A descriptive test involves relatively few subjects, who have been screened. Screening should be product category specific, as is the subsequent training effort. Training is primarily focussed on development of descriptive language, which is used, as a basis for scoring the product. Apart from this, the other important activities that are part of training include, the grouping of attributes by modality (i.e. appearance attributes, aroma attributes and so on), listening them by occurrence, developing a definition for each attribute, identifying helpful references for use during training, and familiarizing the subject with the scoring procedure. There are numerous applications for descriptive analysis, including monitoring competition, storage stability/ shelf life, product development, quality control, physical/chemical, and sensory correlations (Stone et al. 1974; Meilgaard et al. 1991).

Depending upon the test methods used, the training can be quite different. Some of the descriptive methods described in the literature are summarised below :

Flavour profile : This method is the only formal qualitative descriptive procedure, and is probably the most well known of sensory test methods (Stone and Sidel 1993). This method utilizes a panel of four to six screened, and selected persons who first examine, and then discuss the product in an open session (Caul 1957). Once agreement is reached on the description of the product, the panel leader summarizes the results in a report form. The method has considerable appeal because results could be obtained rapidly, and would obviate the need for statistics (Cairncross and Sjostrom 1950).

Texture profile : This method represents an advancement in descriptive analysis with respect to development of the descriptive terminology, the scales for recording intensities, and the word/ product anchors for each scale category (Stone and Sidel 1993). In developing the method, the objective was to eliminate problems of subject variability, allow direct comparison of results with known materials, and provide a relationship with instrument measures (Szczesniak et al. 1963; Brandt et al. 1963). There is a considerable appeal to the direct link between specific instrumental measures of the rheological properties of a product, and the responses of a panel of specific sensory attributes, for example, texturometer units, and hardness sensory ratings. However, separation of texture from other sensory properties of a product such as colour, aroma, taste and so forth limits the total perception of the product's sensory properties.

Quantitative descriptive analysis : This method was developed with an approach that was primarily behavioral in orientation, with a consensus approach to language development, use of replication for assessing the subject, and attribute sensitivity, and for identifying specific product differences, and defined statistical analysis (Stone et al. 1974). The development of method evolved from a number of considerations (Stone and Sidel 1993) to ensure that it would (a) be responsive to all the sensory properties of a product, (b) rely on a limited number of subjects for each test, (c) use subjects qualified before participation, (d) be able to evaluate multiple product in individual booths, (e) use a language development process free from leader influence, (f) be quantitative and use a repeated trials design, and (g) have a useful data analysis system.

The important features of the quantitative descriptive analysis methodology as suggested by Stone and Sidel (1993) are discussed now. This method provides a complete word description for all sensory properties of various ranges of products, the existing as well as new products. This methodology recommends 10 or 12 subjects. However, there have been some product categories, where 15 to 20 subjects have been used. A basic strength of this descriptive method is the ability to independently verify after each test, that individual panelists perceive differences among products on attributes in a reliable manner. This is directly measured with a one-way analysis of variance from each subject for each attribute. By establishing a formal data-based selection, the system thus becomes less dependent on a limited number of subjects.

Earlier descriptive methods, such as the flavour profile were criticized because of the difficulty in understanding the words in the description and the lack of a true numerical system, and statistical procedure for assessing product differences based on these descriptions. The quantitative descriptive analysis method has been quantified by using an appropriate measuring technique (or scale), and the reliability of individual subject established by adopting repeated judgement (Stone and Sidel 1993). A line scale has been used for product evaluation by many workers (Baten 1946; Anderson 1970), and its use in quantitative descriptive analysis was suggested by Stone et al (1974). In

this test, a line of 15 cm length is considered to be very effective. The anchors are located at approximately 1.5 cm from each end. The scale direction always goes from left to right with increasing intensity, for example, weak to strong, light to dark or some similar designated set of word anchors. The panelist's task is to make a vertical line across the horizontal line at the point that best reflects the relative intensity for that attribute (Anderson 1970). For computational purposes, the distance along the line to the mark is measured to yield a numerical value. During language development, subjects are familiarised with the scale, and its use with particular attention given to the anchor terms relative to specific products that will be evaluated.

A line scale was selected because it provided the subject with an infinite number of places in which to indicate the relative intensity for an attribute (within the constraints of the actual length of the line), numbers are not used, thus avoiding number biases, and finally each subject could mark at whatever location on the line, provided the subject was self consistent (Stone and Sidel 1993).

Empirically, this procedure was found to be very easy to use and panelists were able to complete a score card containing, as many as 40 attributes within a few minutes with high degree of reliability.

Other methods : Many more descriptive methods have been described in the literature, which are more or less on the lines of the test methods discussed above.

The spectrum descriptive analysis, for example, involves extensive training activities, reflecting the basic flavour, and texture profile procedures, with particular reliance on training the subjects with specific standards of specified intensities (Stone and Sidel 1993). Free choice profiling is another approach in which no subject screening or training is required, and the subject can use any words, they want to describe the products being evaluated. The time advantage may, however, actually not be there, since the experimenter requires to spend time, explaining the testing procedures to the subjects.

Scoring : The most frequently used of all sensory testing systems is scoring because of its diversity, apparent simplicity, and ease of statistical analysis (Amerine et al. 1965). Scoring methods have been most extensively used by the dairy industry for product development, and improvements, shelf-life studies and assessing suitability of packaging materials. Score cards based on 100 points are generally used for judging, and grading of dairy products (ISI 1975a,b; Nelson and Trout 1964). Most recently, 25 points score cards have been suggested for dairy products (Bodyfelt et al. 1988). Ellis (1961) believed that numerical rating tests gave more complete information than either ranking tests or descriptive rating tests, but the judges must be trained. Since there is no indication of liking to the test product, palatability norms should be established. The score card must be properly developed giving due weightage to all the sensory attributes.

Affective testing

Acceptance testing, a valuable, and necessary component of every sensory programme, is performed at consumer's levels. It refers to measuring liking or preference for a product. Preference can be measured directly by comparison of two or more products with each other, that is, which one of two or more products is preferred. Indirect measurement of preference is achieved by determining which product has scored significantly higher rating than another product in a multi-product test, or which product has scored higher rating than another by significantly more people (Guilford 1954). The two methods most frequently used to directly measure preference, and acceptance are the paired comparison test, and a 9-point Hedonic scale. Other methods are either modifications of these two methods or are types of quality scales; for example, excellent to poor and palatable to unpalatable.

Paired comparison : It is probably the first formal sensory method developed to assess preference, and there is extensive literature about this method (Bradley 1953; Bradley and Terry 1952; Day 1974; Gridgeman 1955, 1959).

The method requires the subject to indicate which one of the two coded products is preferred. A frequently used option allows the inclusion of no preference, as a third choice, while another option allows inclusion of a fourth choice, dislike both equally. The test is relatively easy to organise and to implement (Amerine et al. 1965). Marketing research has made the greatest use of the paired comparison preference method, either, as a two product test or as a much larger effort involving many products, which is referred to as multiple paired comparison tests. The paired preference test is, however, not very informative because the response provides no direct measure of magnitude

TABLE 2. NINE POINT HEDONIC SCALE

Attribute	Score
Like extremely	9
Like very much	8
Like moderately	7
Like slightly	6
Neither like nor dislike	5
Dislike slightly	4
Dislike moderately	3
Dislike very much	2
Dislike extremely	1

Adapted from Amerine et al. (1965)

of the preference (Stone and Sidel 1993). The test is less efficient because there is only one response for each pair of products, compared with a scoring method that yields a response per product.

Hedonic scale : The 9-point Hedonic scale has been used extensively since its development (Jones et al. 1955; Peryam and Haynes 1957) with a wide variety of products, and with considerable success. The scale is easily understood by naive consumers with minimal instruction, and the product differences are reproducible with different groups of subjects. The results from use of this scale are most informative, since computations will yield means, variance measures, and frequency distributions, all by order of presentation and magnitude of difference between products by subject, and by panel, and the data can be converted to ranks as well, yielding product preferences (Amerine et al. 1965). An example of a nine point scale is given in Table 2.

The subject task is to circle the term that best represents their attitude about the product. The responses are converted to numerical values for computational purposes : like extremely, 9; dislike extremely, 1. Vie et al (1991) have recommended calculating R-indices for 9-point Hedonic scale data. On the basis of these advantages, this method is more useful than the paired test (Stone and Sidel 1993).

Types of acceptance testing : There are three primary categories of sensory acceptance test, and the characteristics of each are given in Table 3.

The sensory acceptance test is a very cost-effective resource that has a major role to play in the development of a successful product. Properly used, it will have a significant impact on the growth, and long term development of sensory evaluation.

It is concluded that the future for sensory evaluation is very bright, judging from the number of new food industries, and many existing units that are expanding their sensory test capabilities. If sensory evaluation is to become more successful, it must move from a passive, service-oriented programme to an active, result-oriented resource.

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TABLE 3. CHARACTERISTICS OF DIFFERENT TYPES OF SENSORY ACCEPTANCE TESTS

	Laboratory	Central location	Home use
Consumer type	Employee or local resident	Public (general or selected)	Employee or public
Response per product	25-50	100	50-100
Product numbers	Maximum of 5-6 per session	Maximum 5-6	1-2
Test type	Preference, acceptance but not quality	Same as laboratory	Preference, acceptance, performance
Advantages	- Controlled conditions - Rapid data feed back - "Test-wise" subjects. - Low costs	- Large number of subjects - No company employees	- Product tested under actual use conditions. - All families' opinion obtained - Marketing information
Disadvantages	- Familiarity with product - Limited information - Limited product exposure	- Less control - Limited information - No lengthy or distasteful tasks - Limited instructions - Large number of subjects required	- Expensive

Adapted from Stone and Sidel (1993)

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SURVEY REPORT *J. Food Sci. Technol., 1995, Vol. 32, No. 5, 368-372*

Changing Profile of Food Adulteration : Perception of Food Analysts

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A survey on the perception of Indian food analysts, working in Government food laboratories on the current trends in food adulteration was carried out through questionnaire. A total of 112 analysts from 13 States, and two Union Territories perceived that adulteration of edible oils was the major problem, followed by adulteration of spices, milk, and sweets. Use of newer adulterants like ultramarine blue in dry ginger, urea in puffed rice, and aluminium in *supari* were observed. Addition of colours to potatoes, pickles, and prepared foods like *biryani* was common. The number of chemical adulterants were found to be increasing over the years.

Keywords : Food adulteration, Perception of food analysts, Survey report, Newer adulterants, Chemical adulterants.

Food, a basic need for all people, must be wholesome and safe. Food adulteration is a major public health hazard, which affects the quality of life of people (Jacob 1976). In order to ensure the quality of food available to the consumers, the Government of India had enacted several legislations, including the Prevention of Food Adulteration Act (PFA) (Bhat and Rao 1985). Various State/Union Territory laboratories, which are entrusted with implementation of the PFA act, analyze the food samples, collected by food inspectors, that are sold in the market. They report the findings to Directorate-General of Health Services (DGHS), and the information is published every year (Anon 1992a). It includes the figures on the % adulteration, number of prosecutions, and convictions. The % adulteration, has come down from 14% in 1981 to 9.3% in 1989 (Anon 1992a). This trend is similar in prosecution, and convictions also (Anon 1992a).

The nature of food adulteration may vary from State to State or there could be newer adulterants arising, as a result of changing environmental factors, like non-seasonal rains or improved production/cultivation practices. An adulterant which could be a common one in one region, may become a newer adulterant in other region. Therefore, a study was carried out to find out the perception of food analysts on adulteration, and adulterants encountered.

Materials and Methods

Survey methodology : A pre-tested questionnaire was sent to 77 State food laboratories spread all over the country. The questionnaire was aimed to elicit information on perception of food analysts about the food adulteration, in general, common adulterants which are frequently encountered, unusual adulterants observed, and suitability of available methods for their detection.

Methodology of calculation : Data were analyzed by assuming that the information provided by the analysts was based on their own experience. The % adulteration was calculated by totalling the number of analysts, reporting the encountering of particular adulterant.

Results and Discussion

A total of 112 analysts from 32 laboratories and 77 State food laboratories, who had their own experience in food analysis, adulteration detection, have responded to the questionnaire. Repeated reminders did not elicit any response from the remaining 45 laboratories. State food laboratories of Andhra Pradesh, Gujarat, Goa, Haryana, Jammu and Kashmir, Karnataka, Kerala, Madhya Pradesh, Maharashtra, Pondicherry, Rajasthan, Tamil Nadu, Tripura and West Bengal have responded, while there was no response from the laboratories of the States of Bihar, Himachal Pradesh, Nagaland, Orissa, Punjab and Uttar Pradesh.

Analysis of experience profile indicated that 65% of the analysts had more than 10 years of experience in food analysis (Table 1). According to 20 out of 112 analysts, the adulteration of edible

TABLE 1. EXPERIENCE PROFILE OF ANALYSTS

No. of years	No. of analysts
1 - 5	10
6 -10	29
11 -15	26
16 -20	17
>20	30

* *Corresponding Author*

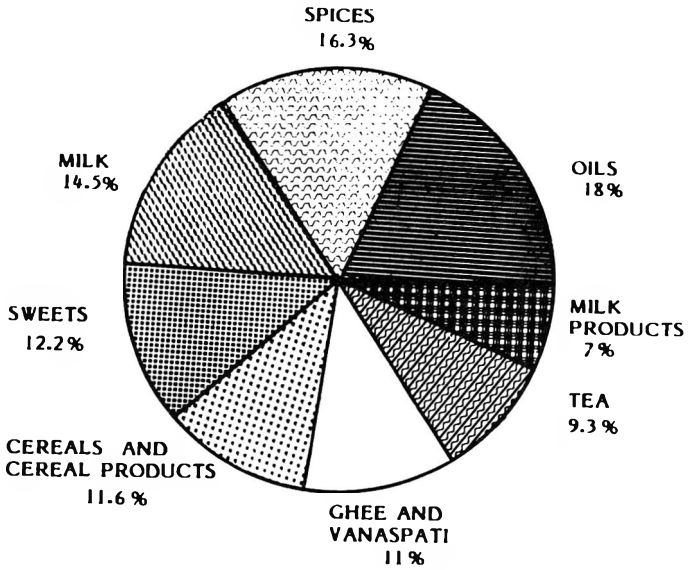

Fig. 1. Adulteration pattern of food commodities

oils was the major problem, while 18 and 16 analysts opined that spice, and milk adulteration respectively, were common (Fig 1). Analysis of adulterants found in individual food groups indicated at least 11 adulterants in edible oils (Fig 2). A total of 58 analysts reported various adulterants in edible oils. Sixteen out of 58 analysts felt that adulteration of edible oils with castor oil was a major problem, while cottonseed oil was a common adulterant in other edible oils according to 9 analysts. Castor oils seemed to be the oil of choice for adulteration in groundnut, and coconut oils. In case of mustard oil, the linseed oil was the adulterant. This observation is in line with the earlier independent survey conducted in 1100 mustard oil samples from 228 villages of Uttar Pradesh, where 150 samples were found to be

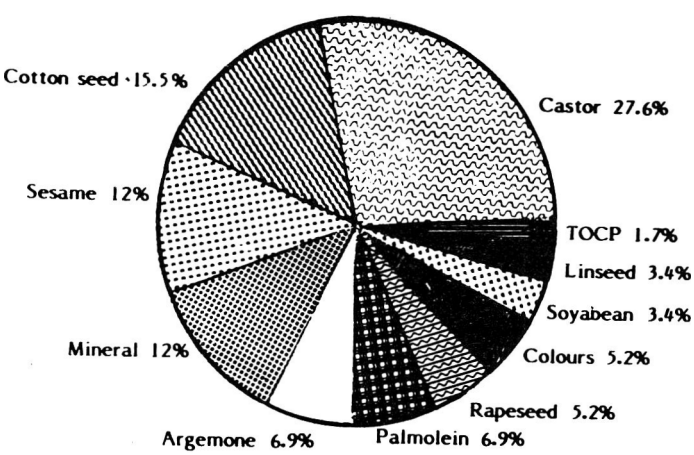

Fig. 2. Type of adulteration in edible oils TOCP - Tri ortho cresyl phosphate

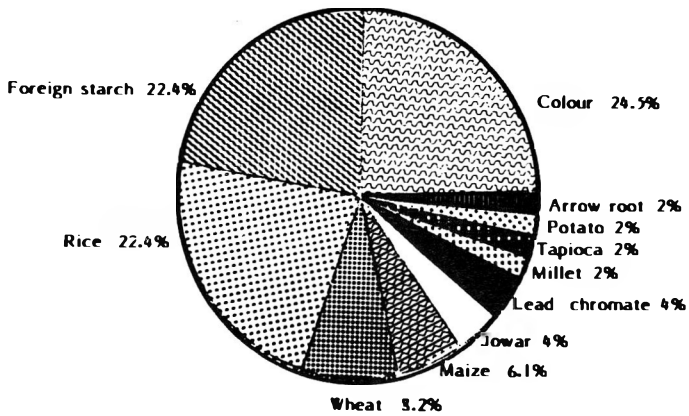

Fig. 3. Type of adulteration in turmeric

adulterated with linseed oil (Khanna et al. 1986).

It is pertinent to mention that, according to the reports of DGHS, the edible oil adulteration varied from 6.0 to 16.2% (Chadha 1989). In contrast, the survey conducted by the National Dairy Development Board (NDDB) in Rajkot, and Bangalore markets, over 90% of edible oils were adulterated (Anon 1991). Another survey, conducted in an urban slum in Hyderabad, revealed that 89% of the edible oils were adulterated (Sawma 1993). Methods for detection of adulterants and contaminants in edible oils and fats have been critically evaluated (Krishnamurthy 1993).

Analysts perceived that the second high risk adulterated group is spices, among which turmeric is the mostly adulterated item with a higher number of adulterants. Among the 49 analysts, who have reported adulterants in turmeric, 12 stated that the addition of colour was the major problem, while 11 analysts felt that foreign starch from cereals, and tuber was the major adulterant (Fig. 3). Other spices, like cardamom, cloves, and dry ginger. were not mentioned by the analysts. Perhaps, these samples were not sent to them for analysis.

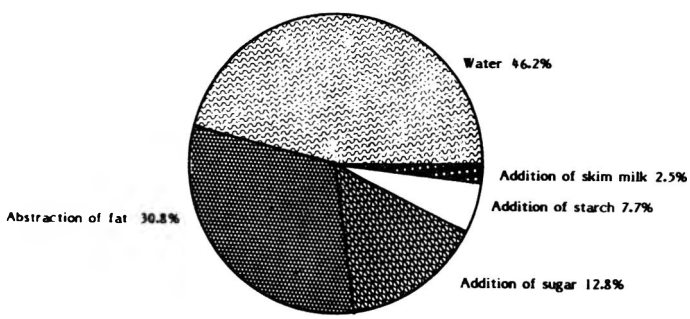

Fig. 4. Type of adulteration in milk

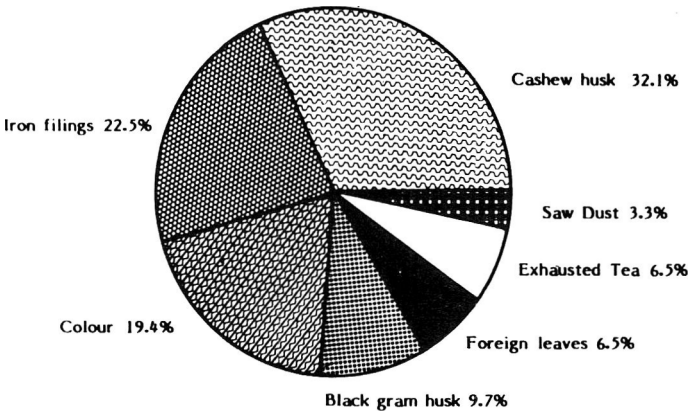

Fig. 5. Type of adulteration in powder

Among 39 analysts, who have reported adulterants in milk, 18 felt that it was water, while 12 opined that it was abstraction of fat (Fig. 4). According to the official statistics, milk is the most adulterated commodity (Chadha 1989). However, the analysts perceived it to be the third high risk group. Adulteration of milk is not taken seriously, and this is an instance of traditional compromise between the seller, and the consumer on account of economic considerations (Gopalakrishna Murthy 1985). Among tea and coffee, the first was opined to be the most adulterated, and there were 8 adulterants reported in tea (Fig. 5). Among the 31 analysts, who had reported adulterants in tea, 10 analysts felt cashew husk was the major adulterant, while 7 and 6 analysts opined iron fillings and colour, as the major adulterants, respectively. In the sweets, and confectionery, among the 46 analysts who had reported adulterants in confectionery, 17 have opined that saccharin, as major adulterant, while 16 felt addition of unpermitted colour as a major problem (Fig. 6).

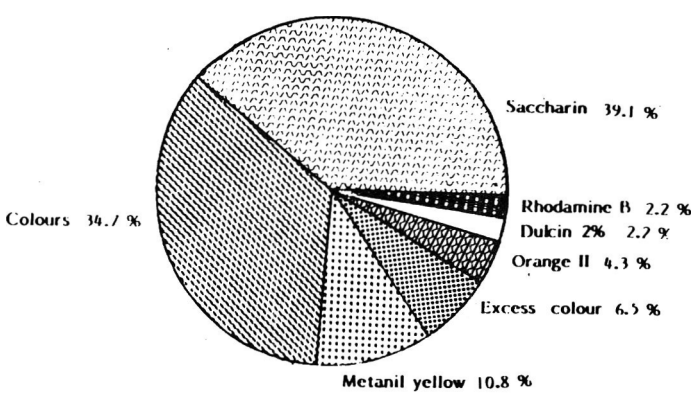

Fig. 6. Type of adulteration in confectionery

A total of 33 analysts had reported various adulterants in cereals, pulses, and their products (Fig. 7). Thirteen, out of 33 analysts, reported *Lathyrus sativus* as a major problem, while addition of colour was the common problem as per 7 analysts. *Lathyrus sativus* adulteration in *Cajanus cajan*, *Cicer arietinum* flour, and *papad* seemed to be the major problem throughout India, except in the States where the sale of *Lathyrus sativus* is not banned. Similar results were also found in independent survey conducted in Uttar Pradesh (Khanna and Mukuldas 1992).

The pattern of food adulteration is changing. The use of saccharin, and unpermitted colours in confectionery is an old problem. Colours are now being added to foods like peas, potatoes, aniseed (*Pimpinella anisum*), asafoetida, and prepared foods like chicken *biryani* (a spicy preparation with chicken and rice), and pickles. A study conducted in Calcutta revealed the fraudulent use of colours in flattened rice, fish, vegetables, cut fruits, and milk (Roy and Chakrabarti 1991). Traditionally, some sweets are decorated with silver foil in India and use of aluminium was reported in place of silver (Personal communication). Now, the use of aluminium has spread to other foods like *supari* (*Areca catechu*) and *pan masala*.

The food adulteration seems to be having a regional character. Laboratories from Assam have reported adulteration of buffalo milk with metanil yellow for facilitating its sale, as cow milk. No other State had reported such problem.

Analysis of the data on unusual adulterants indicated about 50 food items, which were found to have unusual adulterants. Many of the unusual adulterants reported by analysts were common adulterants in other region. State food laboratory,

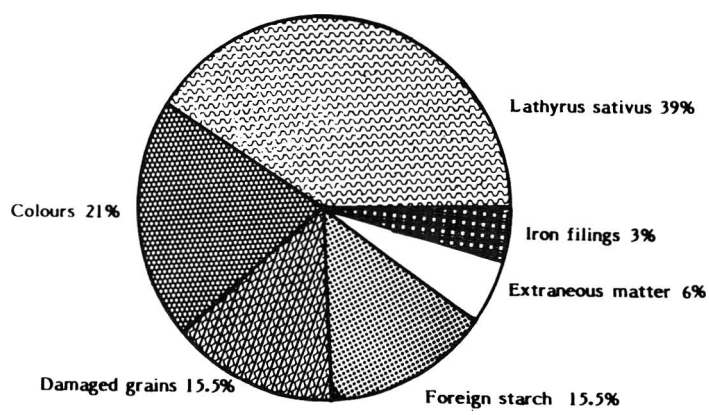

Fig. 7. Type of adulteration in cereals, pulses, and their products

TABLE 2. UNUSUAL ADULTERANTS REPORTED BY STATE FOOD LABORATORIES

Food items	Adulterants
Turmeric	Unidentified water soluble yellow colour
Supari	Aluminium foil
Pan masala	Aluminium foil
Tea	Silver oak leaf
Pan chatni	Aluminium foil
Bengalgram flour	*Pisum sativum* flour, Metanil yellow
Dry ginger	Ultramarine blue
Asafoetida	Metanil yellow
Puffed rice	Ultramarine blue, urea
Aniseed	Malachite green

Indore, has reported cashew husk in tea, as a newer adulterant. However, laboratories from Coimbatore, Thanjavur, Palayamkottai, Madurai, Bombay, Thiruvananthapuram, and Hyderabad have reported cashew husk, as a common adulterant. Table 2 reports some of the unusual adulterants.

The reported adulterants have been classified based on their nature (Table 3). It shows that the majority of the adulterants are chemicals. Some of these chemicals have been permitted under the act for use in specific foods (Anon 1992b). However, they are being added to other foods. For example, saccharin is not permitted to be used in confectionery, but it can be used in non-alcoholic beverages upto 100 ppm (Anon 1992b). In some cases, these chemicals are being used in excess of the permitted levels. For example, benzoic acid or sulphur dioxide is permitted in pickles at 250 or 100 ppm, respectively, in pickles (Anon 1992b), but were found to be at much higher levels in some samples.

TABLE 3. DIFFERENT CLASSES OF ADULTERANTS

Classification	Adulterants
Coal tar dyes	Orange, sudan, metanil yellow, auromine, orange II, malachite green, rhodamine B.
Cheaper oils	Castor oil, soybean oil, sesame oil, rapeseed oil, palmolein, mineral oil, terpentine.
Cheaper agricultural produce	Wheat starch, maize starch, *jowar* starch, rice starch, arrow root starch, amaranth seeds, date seeds.
Chemicals	Saccharin, sodium bicarbonate, sodium carbonate, acetic acid, ammonium sulphate, copper sulphate, urea, dulcin, brominated vegetable oil, monosodium stearate, ammonia, calcium oxide, ultramarine anticaking agent, benzoic acid, diezipan, ammonium chloride, chloral hydrate, triorthocresyl phosphate.
Extraneous matter	Wooden pieces, chalk, cashew husk, gram husk, silver oak leaf, fenugreek, sand, cellulose, colophany resin, tamarind husk, coffee husk, grass seeds, saw dust.
Metal contaminants	Aluminium, iron filings, lead chromate, nickel.

Regarding the adequacy of the methods available, many of the analysts have indicated the need for efficient, and specific method to detect, and identify individual oil in oil blends. Some analysts expressed the desire to have simple methods to detect food additives.

Besides DGHS statistics on food adulteration (Anon 1992a), the scientific research studies on food adulteration are limited to selected food items like sweets, and confectionery (Khanna et al. 1986), and edible oils (Anon 1991). There is a considerable difference between the official statistics of food adulteration, analysts' perception and isolated surveys conducted. To get a true picture, drawing of non-formal samples by independent authority (not through food inspectors) and its analysis for various adulterants is essential.

Acknowledgements

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SURVEY REPORT *J. Food Sci. Technol., 1995, Vol. 32, No. 5, 373-376*

Food Quality Surveillance on Colours in Eatables Sold in Rural Markets of Uttar Pradesh

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A statistically designed survey was undertaken to find out the magnitude, and usage pattern of artificial colours in sweets, savoury products (*namkins*), and powdered turmeric, and red chilli. Of 2057 samples, drawn from 224 village markets from 56 districts of Uttar Pradesh, 32% contained artificial colours of which 61.6% were non-permitted. In the artificially coloured eatables, 39% sweets, 85% *namkins*, and all the samples of powdered turmeric, as well as chilli contained non-permitted colours. Seven non-food grade dyes used include metanil yellow, orange II, Sudan I, rhodamine B, Blue VRS, auramine, and Sudan III/IV.

Keywords : Permitted colours, Non-permitted colours, Food commodities, Sweets, Savoury, Turmeric, Chilli.

Colours are used in food for centuries to increase consumer acceptibility (Walford 1980). Indiscriminate use of non-permitted colours in food preparations and the likely threat to health safety have been a matter of continued concern (Khanna et al, 1973; 1980; 1985, Khanna and Singh 1975; Babu Shenolikar 1990; Roy and Chakrabarty, 1991, Khanna and Das 1992). In an earlier survey (Khanna et al. 1985), 47.8% sweet and 61.3% *namkin* samples in rural areas were found to contain non-permitted colours. The present study was aimed to identify the usage pattern, and magnitude of artificially added synthetic colours in sweets, *namkins* (savouries), and powdered chilli, as well as turmeric sold in retail outlets of rural markets in Uttar Pradesh.

Materials and Methods

Chemicals and reagents : Permitted colours used as reference standard were the courtesy of Bush Boake Allen (India) Ltd, Madras. Green S and Fast green FCF were gifts from Williams (Hounslow) Ltd, UK. Non-permitted colours i.e., auramine, blue VRS, malachite green, and orange II were purchased from Vesco Products Co., Calcutta. Metanil yellow, and rhodamine B were from Loba Chemie Indoaustranal, Bombay and S.D. Fine Chemicals Ltd, Bombay, respectively. Fat-soluble dyes, Sudan I, butter yellow, and Sudan IV were purchased from Hartman Leddon Co., Philadelphia, USA, while Sudan II was from VEG Laborchemie, Apolda, and Sudan III from E. Merck, Germany.

Collection of food samples : Three samples each of sweets, and *namkins* and 2 samples each of turmeric, and red chilli powder were collected from 2 rural markets, out of the two randomly marked blocks in each of the 56 districts of Uttar Pradesh. These food samples were procured from 112 blocks, and 224 village markets. From Lucknow district, 20 samples from each of 8 rural blocks led to an additional collection of 160 samples. Of the 2400 samples collected in the 2 years programme, 2057 samples could be analyzed, while the remaining got spoiled during transit.

Classification of eatables : Sweet samples analyzed included (i) milk products (*khoya burfi, chenna/bengali* sweets), (ii) non-milk products (*jalebi, laddoo, khurma, sohan papri*), and (iii) sugar confectionery like *lemon drops, lollipops*, and *sugar coated saunf*. Savoury products analyzed in the survey comprised of *dalmoth, besan sev, sabudana (sago)* rolls and *sabudana* chips. In powdered spices, turmeric, and chilli samples were analyzed.

Extraction and resolution of dyes from foodstuffs : Selective extraction method based on differential solvent solubility (Khanna et al. 1985), which eliminated interference in the subsequent paper chromatographic separation, was used.

A twenty five gram portion of the sample was soaked for 3-4 h in 100 ml petroleum ether (40-60°C boiling range) with occasional shaking, and filtered. Treatment was repeated with 50 ml fresh petroleum ether. On filtration, residue was dried at room temperature, and shaken with 50 ml of 80% ethanol with occasional heating in a water bath, after which the solvent was decanted. Another 25 ml portion of fresh 80% ethanol was used for the second extraction as above. The combined filtrate was evaporated to remove ethanol, and the residue was taken up in 25 ml distilled water. The colours from water phase were then selectively extracted in 2-3 ml of acidic n-butanol (pre-acidified

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with five drops of concentrated hydrochloric acid per 100 ml water saturated n-butanol). Though small volume of acidic n-butanol serves the purpose for qualitative detection, as it gives a concentrated extract of colours, repeated extractions till acidified n-butanol layer become colourless are required for optimum extraction in cases where quantitative determination is desired. The combined butanol extracts were concentrated to a small volume under vacuum.

The butanolic coloured layer was applied on to a Whatman No. 1 chromatography paper strip, together with authentic standard colour, and run overnight in either of the following 2 solvent system (Crossby 1981).

Solvent A : Trisodium citrate : ammonia : water (2 g :15 ml : 85 ml) for pink-red, orange, and yellow samples.

Solvent B: Isopropanol : ammonia : water (7:2:1 v/v) for blue, green, and yellow samples.

Rf values of separated colours were compared, and matched with Rf values of reference dyes.

Results and Discussion

Of the 2057 samples analyzed, only 672 (32.7%) samples contained synthetic colours. Among these, 258 samples had permitted colours, while 414 samples had non-permitted colours. Thus, the percentage of samples adulterated with non-permitted colours was 20%.

Usage pattern of colours in different food commodities : The extent of addition of colours in different food commodities is given in Table 1. Analysis revealed that 394 (56.3%) of total 699 sweet samples employed externally added colours. Among coloured sweets, 240 (61%) samples contained permitted group of colours, and 154 (39%) samples had non-prescribed colours (Table 1)

Among permitted colours used in the sweets, the yellow to orange shades, viz., tartrazine, and sunset yellow were found in 144 and 71 samples, respectively. These colours along with blue, indigocarmine (12), and brilliant blue FCF (6) were used to get green shade. For red shade, only amaranth (7) was used. Among non-permitted colours, metanil yellow, was encountered in 73 samples, followed by orange II in 56 samples. The two other non-food grade colours detected in the sweets were the fluorescent pink dye (rhodamine B), and a blue colour (blue VRS) in 14 and 11 samples, respectively.

Namkins : In this group of commodity, 671 samples comprising savoury items such as, *dalmoth* (374) *besan sev* (193) *maida* products (68), *sago* rolls (21) and *sago* chips (15) were examined. Only 127 (18.9%) of the total *namkin* samples collected had artificial colours. Over 85% of these (109 samples) contained non-permitted colours (Table 1). In permitted colours, only tartrazine (11) and sunset yellow (7) were spotted. Three non-permitted colours detected included, metanil yellow (52), orange II (50) and auramine (7).

TABLE 1. PATTERN OF NON-PERMITTED COLOURS IN DIFFERENT FOOD COMMODITIES AND IN SOCIO-ECONOMIC ZONES OF THE STATE OF UTTAR PRADESH

Attribute	Number of samples			Adulteration % based on	
	Total analyzed	Artificially coloured	Containing non-permitted colours	Total samples analyzed	Artificial coloured samples
Commodity :					
Sweets	699	394	154	22.0	39.1
Namkins	671	127	109	16.2	85.8
Turmeric powder	462	106	106	22.9	100.0
Chilli powder	225	45	45	20.0	100.0
Zones :					
Bundelkhand	175	62	41	23.4	66.1
Central	452	158	107	23.6	67.7
Eastern	493	147	88	17.8	59.8
Hill	281	70	40	14.2	57.1
Western	656	235	138	21.0	58.7

Turmeric powder : Of 462 samples analyzed, 106 samples (22.9%) contained artificial colours, all of which belonged to non-permitted class (Table 1). Orange II was encountered in 49 samples, while 44 samples contained metanil yellow. In addition, 13 samples employed a blend of these two colours.

Chilli powder : In this commodity, 45 samples (20%) out of 225 samples were coloured (Table 1), and all of these contained fat-soluble, Sudan dyes.

Usage pattern of colours in 5 socio-economic zones of the State of Uttar Pradesh : The usage pattern of colours in food commodities out of 5 socio-economic zones of the State of Uttar Pradesh is shown in Table 1. In Bundelkhand zone, which comprises 5 districts, 175 samples of all 4 group of commodities were picked up. Of these, 62 (35.4%), samples were coloured, 21 with permitted, and 41 with non-permitted colours. Colour adulteration of food commodities in this zone worked out to 23.4% of total collected, and 66.1% among coloured eatables. From the Central zone,

which possessed 9 districts, a total of 452 eatables of the 4 commodities were analyzed, out of which 158 samples (34.9%) contained added synthetic colours. The numbers of permitted versus non-permitted colours were 51 and 107 samples, respectively (Table 1).

From the State capital city of Lucknow, a part of central zone of the 8 blocks, namely Bakshi Ka Talab, Chinhat, Gosaiganj, Kakori, Mal, Malihabad, Mohanlalganj and Sarojini Nagar, 149 samples were analyzed, of which 57 eatables (38.2%) contained artificial colours. Of these, permitted, and non-permitted colours were 12 and 45, respectively. These figures gave a colour adulteration value of 30 and 79%, due to use of non-permitted colours, among total, and coloured eatables analyzed, respectively.

In Eastern zone having 15 districts, 29.8% (147) samples from a total of 493 were coloured, of which 59.8% samples had non-permitted colours. The Hill zone with 8 districts showed least use of synthetic colours with only 70 (24.9%) food products out of 281, employing artificial colours. Of 656 samples analyzed from the 19 districts of Western zone, 235 samples (35.8%) were coloured. In these, 138 were dyed with non-food grade colours (Table 1).

The extent of use of non-permitted colours in the 4 groups of edible commodities among the 5 socio-economic zones is presented in Table 2. The analysis of samples having non-permitted colours, out of total collected samples of that group, revealed that the percentage adulteration ranged from a minimum of 15.1% in Bundelkhand to a maximum of 28.8% in the Eastern zone in case of sweet samples. However, in *namkins*, Eastern zone exhibited the lowest (6.2%) use of non-permitted colours, while Western zone showed the maximum use (21%) of non-food colours. In case of powdered turmeric, Hill zone showed least use of non-permitted colours with only 4.6% coloured samples, while central zone samples showed maximum use (34.6%). The frequency of non-food grade, fat soluble dyes in chilli powder showed a minimum, and maximum of 10.2% and 33.3% in Central and Bundelkhand zones, respectively. In powdered spices, addition of even permitted coal tar dyes is not permissible, as per PFA Act and Rules (PFA 1993). Hence, addition of even permitted colour would render the samples being branded as adulterated.

TABLE 2. COMMODITY WISE USE OF NON-PERMITTED COLOURS (%) IN FIVE SOCIO-ECONOMIC ZONES OF THE STATE OF UTTAR PRADESH

Zone	Non-permitted colours among total samples (%)			
	Sweet	*Namkin*	Turmeric	Chilli
Bundelkhand	15.1	20.3	33.3	33.3
Central	24.3	20.0	34.6	10.2
Eastern	28.8	6.2	18.3	14.6
Hill	18.7	14.0	4.6	22.2
Western	18.1	21.0	21.1	23.4
Total	22.0	16.2	22.9	20.0

TABLE 3. OVERALL FREQUENCY OF USE OF INDIVIDUAL COLOURS IN THE ENTIRE SURVEY SAMPLES

Colour	Total number	Percentage within the group (PC or NPC)	Percentage of total colours (PC and NPC)
Permitted (PC)	**258**	**100.0**	**38.4**
Tartrazine	155	60.1	23.1
Sunset yellow	78	30.2	11.6
Indigo carmine	12	4.6	1.8
Amaranth*	7	2.7	1.0
Brilliant blue FCF	6	2.3	0.9
Non-permitted (NPC)	**414**	**100.0**	**61.6**
Metanil yellow	169	40.8	25.1
Orange II	155	37.4	23.1
Blend Sudan I + Sudan III	30	7.2	4.5
Rhodamine B	14	3.4	2.1
Blend Metanil yellow + orange II	13	3.1	1.9
Blue VRS	11	2.6	1.6
Sudan I	9	2.2	1.4
Auramine	7	1.7	1.0
Sudan III/IV	6	1.5	0.9

* Amaranth was enlisted as a permitted colour during the conduct of present study, and has since been withdrawn (PFA 1993).

Overall use pattern of individual colours : The overall frequency of use of individual permitted, and non-permitted colours in the entire coloured survey samples is tabulated in Table 3. Tartrazine was the most used yellow permitted colour detected in 23% of total coloured samples, while sunset yellow was present in 11.6% samples. The three other permitted colours used were the indigocarmine (1.8%), and the brilliant blue FCF (0.9%), and Amaranth (1%). Amaranth (1%), besides fast red E and green S are prohibited (PFA 1993).

Among non-permitted dyes, the most commonly used colours detected in order of usage included metanil yellow (169), orange II (155), Sudan I/III (3), rhodamine B (14), blend of metanil yellow plus orange II (13), blue VRS (11), Sudan III (9), auramine (7) and Sudan I (6).

A number of dyes meant for textiles, and other uses which are easily available in cheap paper wraps, are used in foods either deliberately or through ignorance. Such an use is considered more likely in village markets than city markets, where regulatory surveillance is comparatively more organised. The data show that there is no marked difference in the pattern of use of non-prescribed colours amongst the five socio-economic zones of U.P. and especially that in the economically poor Eastern zone to that of somewhat affluent Western belt. Same is the case with the well demarcated backward districts versus non-backward districts.

In a recent survey aimed at identifying the frequency of use of added colouring matter on some foodstuffs available in and around Calcutta markets, Roy and Chakrabarti (1991) reported the fradulent use of artificial colours in some of the unsuspected food items such as flattened rice, fish, fresh vegetables, and cut-fruits. In each of these commodities, even the use of permitted food colours is non-permissible, as per the provision of the Prevention of Food Adulteration Act of India (PFA 1993). This study identified non-permitted colours such as auramine, congo red, malachite green, rhodamine B and even an inorganic pigment, copper sulphate. Use of non-permitted colours was also reported in limited surveys on eatables from Hyderabad (Babu and Shenolikar 1990), and Mysore (Nagaraja KV, Personal communication). Also, results of two separate basket surveys of edible commodities exclusively drawn from city markets of Lucknow, and Delhi showed a much less prevalence of non-permitted colours, showing a restrictive use frequency of 22.4 and 14.6% (ITRC 1993). In contrast to these above city market surveys, the rural samples in the present study showed a considerably high magnitude, and prevalance of non-food grade colours.

Animal toxicity evaluation has shown that non-permitted dyes such as auramine, blue VRS, malachite green, orange II, rhodamine B and "Sudan I/III" produce pathological lesions in vital organs like kidney, spleen and/or liver (Khanna and Das 1991). High levels of metanil yellow are known to cause testicular degeneration (Singh and Khanna 1974). Auramine, blue VRS, and rhodamine are reported to increase the incidence of tumours of lung, breast, ovary, and liver, and teratogenic abnormalities of bone, eyes, skin, and lung (Khanna et al. 1980). Hence, efforts are needed to strengthen the existing food quality monitoring, and surveillance programmes particularly in rural areas.

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Studies on Keeping Quality of Pickled Chicken in Flexible Pouches

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Storage stability of chicken pickle in vinegar, packed in two types of pouches made from (i) metallized polyester/ polyethylene and (ii) coextruded film of polyethylene/polyamide/ethylene-acrylic acid films has been studied for 90 and 180 days storage at (i) accelerated condition (38°C with 90% RH), and (ii) normal or standard condition (27°C with 65% RH). Except for a progressive increase in free fatty acid, and peroxide value, no significant changes ($P>0.05$) in other chemical parameters were observed. A decrease ($P<0.05$) in sensory quality occurred with storage. The product was found to be microbiologically safe. The experimental data indicated that both the types of pouches could be used to obtain shelf-life of 90 to 180 days under the conditions investigated.

Keywords : Chicken pickle, Storage, Flexible pouches, Chemical changes, Sensory scores.

The consumption of poultry meat has been growing, but its scientific processing, and packaging are not well developed (Sahu and Mahapatra 1992). There is also a growing demand for ready-to-eat and semi-processed meat products such as wafers and pickles. Consequently, technologies for processing poultry products such as poultry pickles, and wafers have been developed (CFTRI 1988-89). Further, packaging of these processed products is important for extending the shelf-life, and distribution over wider areas.

Several studies have been carried out on the storage aspects of pickled products based on pork (Kumar and Bachhil 1993), and quail eggs (Srivastav and Panda 1976; Appi Reddy et al. 1978, Singh and Panda 1991). Further, storage stability of egg powder in flexible pouches has also been studied (Nirmala et al. 1976), while Juhi Raikhy and Bawa (1992) studied the overall acceptability of pickled chicken eggs. But no details on the keeping quality of pickled chicken have been reported. The current investigation envisaged the evaluation of the physico-chemical properties of two newly available flexible packaging materials, their protective properties for storing the product under simulated conditions of extreme conditions as well as standard environmental conditions, and providing good keeping qualities for a minimum period of 3 to 6 months, which would be sufficient for marketing purposes.

Materials and Methods

Chicken pickle : To obtain about 12 kg of eviscerated, and deboned chicken, 23 broiler chicken were used. The meat pieces were cut to about 5 cm width, marinated, and cooked for 30 min (Sidhu 1993). Cooked pieces were deep-fat-fried in rapeseed oil, and mixed with spices paste, salt, sugar and vinegar, to make the complete pickle (Sidhu 1993).

Quality evaluation of packaging materials : The thickness, tensile strength, and heat-seal strength were determined according to test procedures of ASTM (1982). The water-vapour transmission rate (WVTR) was determined as per IS : 1060 (BIS 1982), and the oxygen transmission rate according to D1434 (ASTM 1982).

Chemical analysis : Moisture content was determined according to AOAC (1973). Acidity as pH was measured using pH-meter (Radiometer, Copenhagen). Crude fat, protein, free fatty acids (FFA), and peroxide value (PV) were determined by standard methods (Nambudiry 1985).

Microbiological analysis : The product was analyzed initially, and at the end of storage period (90 and 180 days) under the two conditions for total plate counts, total coliforms, *Staphylococcus aureus*, according to the FDA analytical procedures (1978), and yeasts plus moulds, and acid-tolerant bacteria according to BIS (1985).

Storage studies : Unit pouches filled with 200 g samples were heat-sealed, and exposed to 90% RH, 38°C (accelerated condition), and 65% RH, 27°C in humidity cabinets (Laboratory Thermal Equipment, Oldham, U.K.) as per the Standards (BIS 1060 Part 1) or normal conditions. The pouches were made of a laminate of 12 micron metallized polyester/ 100 micron co-extruded film of high density polyethylene, and low-density polyethylene; 100

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TABLE 1. PHYSICO-CHEMICAL PROPERTIES OF PACKAGING MATERIALS

Property	MPP laminate	LPE coextruded
Thickness, μm	112 (12/100)	100
Tensile strength, kN/m		
Machine direction	3.60	2.14
Transverse direction	3.30	1.84
Elongation at break, %		
Machine direction	36	> 200
Transverse direction	42	> 200
Heat-seal strength, kN/m		
Machine direction	2.90	1.93
Transverse direction	2.60	1.66
Water vapour transmission rate, g/m^2 .24 h		
38°C/92% RH condition	0.74	3.66
27°C/65% RH condition	0.30	1.17
Oxygen transmission rate ml/m^2 .24 h kPa at27°C	0.20	4.39
Yield of film, m^2/kg	8.40	9.70

MPP : Metallised polyester/high and low density polyethylene film, LPE : Linear low density polyethylene/polyamide/ polyethylene co-extruded film.

micron co-extruded white pigmented film of linear low-density polyethylene/bonding agent/polyamide/ bonding agent/ethylene-acrylic acid copolymer. The control samples packed in glass jars were stored at refrigerated condition (4-6°C).

Sensory evaluation : Sensory evaluation of pickled chicken, spread on bread pieces, was carried out by a 12 member trained panel at each interval for colour, odour, taste, and texture (mouth feel) using a 7-point Hedonic scale.

Results and Discussion

Physico-chemical properties of packaging materials : The values are indicated in Table 1.

Chicken pickle with an initial moisture content of 47.5% had an equilibrium relative humidity of 81.4% at 27°C as determined by hygrometer (Novasina water activity meter, Zurich, Switzerland). Initial microbiological loads include a total plate count of 9.0×10^4/g, while the yeasts plus moulds, and acid tolerant bacterial count were 40/g each. Coliforms, and *S. aureus* counts were absent.

Accelerated storage : Moisture content of the product during storage of 90 days has indicated a slight decrease, from the initial value of 47.53% to 45.11% in MPP laminate, and to 45.54% in coextruded film pouch (Table 2).

Initial FFA value of 6.96% increased gradually, and is in general consonance with the water vapour transmission rates of the packaging materials used. Higher rates were observed in the co-extruded film than the laminate pouches. The maximum FFA value (17.0%) was observed in pickle packed in coextruded film after 90 days storage, whereas it was only 13.45% in the case of metallized polyester pouches. Similar results were reported in pickled quail eggs by Singh and Panda (1991).

Initial pH (4.2) changed marginally to 4.3 in both types of pouches, and attained this equilibrium value after 60 days of storage. Similar results have been reported for pickled quail eggs by Tipshetti and Panda (1978) and Appi Reddy et al (1978).

There was a slight increase in the total fat content during 90 days of storage, while the protein

TABLE 2. CHANGES IN CHEMICAL AND SENSORY QUALITIES OF STORED PICKLE AT THE ACCELERATED CONDITION.

Attribute	Days of storage: 0	15		30		45		60		75		90	
		MPP	LPE	MPP	LPE	MPP	LPE	MPP	LPE	MPP	LPE	MPP	LPE
Moisture %	47.53	45.54	47.06	45.54	47.55	45.36	44.43	44.21	45.71	43.22	45.98	45.11	45.54
Acidity, pH	4.20	4.20	4.30	4.20	4.20	4.20	4.20	4.30	4.20	4.30	4.30	4.30	4.30
Fat, %	13.70	14.00	14.66	14.94	14.95	14.76	14.66	14.25	15.42	14.00	15.68	14.02	13.82
Protein %	34.50	35.14	35.63	33.98	35.12	34.58	33.08	33.73	34.15	31.45	31.80	32.97	32.07
FFA, %	6.96	7.60	7.25	8.55	8.45	9.45	10.05	10.45	12.05	12.00	15.10	13.45	17.00
PV, m.eq.	7.71	11.35	11.70	12.65	14.25	13.60	15.80	14.30	16.35	12.65	14.23	9.45	12.55
Colour	-	6.44	6.37	5.53	6.02	5.32	5.32	5.11	4.90	4.76	4.76	4.45	4.76
Texture	-	6.16	6.30	5.32	5.67	4.97	5.46	4.83	5.25	4.62	5.11	4.48	5.04
Taste	-	6.30	6.26	5.39	5.88	5.11	5.74	4.97	5.67	4.76	5.32	4.62	5.18
Overall acceptability	-	6.37	6.30	5.46	5.81	4.97	5.46	4.76	5.18	4.69	5.14	4.62	5.04

MPP and LPE : Designations same as in Table 1 ; FFA : Free fatty acids, percent as oleic acid, PV : Peroxide value, milliequivalents of oxygen/kg fat. The values are mean of three determinations.

content decreased gradually from the initial mean value of 34.50% to 32.07% in the laminate pouch, and 32.87% in the pouches made from co-extruded film (Table 2).

The peroxide value as the criterion of oxidative changes (Gopalakrishna and Prabhakar 1983) indicated significant changes. The increase was more pronounced in the co-extruded film pouch than in the metallized polyester pouches, as indicated by the value of 12.55 and 9.45, respectively, at the end of 90 days and, after reaching peak values at the end of 60 days storage.

The details of the changes in the sensory qualities, colour, texture, taste, and overall acceptability scores are presented in Table 2. There was progressive loss in sensory scores. The scores for texture, and taste were comparable to overall acceptability. Greater decrease in colour intensity was noticed. The differences in the taste, and subsequent overall acceptability may be due to the different contact layers, viz., low density polyethylene, and ethylene acrylic acid co-polymer, as the former has been shown to absorb various flavourings faster, and to a greater extent than others due to greater sorptive capacity (Baner et al. 1991).

Overall acceptability scores declined steadily with storage time, the decrease being more dependent on the packaging material. However, no detectable off-odour was perceived even upto 180 days of storage period. Penelists did not differentiate the pickle texture, but indicated undesirable changes in colour, especially for the one in the laminate pouch.

Normal storage : The moisture contents decreased gradually in both types of pouches, the decrease being more pronounced in the co-extruded film pouch sample, due to its greater water vapour transmission (Table 1). Although at the end of 90 days, the moisture contents in the laminate, and co-extruded film pouches were 46.48 and 45.80% respectively, it further decreased to 44.10 and 44.00% at the end of 180 days. The changes in the pH value were minimal, and these were similar to those of the values for accelerated storage.

The changes exhibited in the total fat, and protein contents were more or less similar to those at the accelerated condition. The FFA values showed lesser increase, as indicated by values of 11.24 to 11.30% at the end of 180 days storage, as against 13.45 and 17.00% at the end of 90 days storage at the accelerated condition in the polyester laminate and nylon-based co-extruded film made pouches. PV indicated a rapid rise initially, reaching a maximum value of 14.5 to 14.7 milliequivalents, and later decreasing gradually.

From the data, it could be concluded that enhancement in protection against oxidative changes could be attributed to the metallization web (Table 1), which was found to have an oxygen transmission rate of only 0.2 ml/m², 24 h.kPa at 27°C as against 44.5 for the polyamide-based film. The former flexible packaging material afforded good protection, due to its barrier properties, though not to the extent as indicated by its low value.

In the case of pickled quail eggs (Singh et al. 1989), it has been observed that oxidation index, the thiobarbituric acid (TBA) value, increased significantly. Higher storage temperature resulted

TABLE 3. CHANGES IN CHEMICAL AND SENSORY QUALITIES OF STORED PICKLE AT THE NORMAL CONDITION.

Attribute	Days of storage											
	0	30		60		90		120		150		180	
		MPP	LPE	MPP	LPE	MPP	LPE	MPP	LPE	MPP	LPE	MPP	LPE
Moisture %	47.53	45.48	45.63	45.67	45.28	46.48	45.80	45.45	45.32	44.12	44.10	44.10	44.00
Acidity, pH	4.20	4.20	4.30	4.20	4.20	4.30	4.30	4.20	44.30	4.20	4.30	4.20	4.30
Fat, %	13.70	11.89	12.34	13.70	13.60	14.20	14.66	14.20	14.52	14.12	14.48	14.10	14.20
Protein %	34.50	34.18	34.05	33.23	32.44	31.73	30.93	31.60	29.82	30.83	28.22	27.65	30.02
FFA, %	6.96	8.65	8.25	8.75	9.10	9.50	9.75	10.05	10.35	10.75	10.90	11.24	11.30
PV, m.eq.	7.71	14.70	14.50	14.02	14.05	10.30	13.25	8.90	11.00	7.80	9.75	7.23	8.88
Colour	-	6.16	6.30	5.39	5.81	4.97	5.32	4.34	4.97	3.92	4.41	3.85	4.06
Texture	-	5.95	5.60	4.90	5.14	4.90	4.97	4.41	4.48	4.06	4.27	3.64	4.06
Taste	-	5.88	6.02	5.46	5.39	4.76	5.25	4.34	4.76	3.92	4.41	3.61	4.06
Overall acceptability	-	6.09	6.09	5.67	5.32	5.25	4.83	4.76	4.55	4.06	4.41	3.61	3.99

MPP and LPE : Designations same as in Table 1 ; FFA : Free fatty acids, percent as oleic acid, PV : Peroxide value, milliequivalents of oxygen/kg fat. The values are mean of three determinations.

in relatively higher values. The correlation of TBA values with the acid values indicated a limited oxidation breakdown of lipids during storage. The formation of oxidative by-products has been found to be dependent on temperature, and time (Singh et al. 1989).

The values for quality parameters (Table 3) indicate that the scores decreased gradually, but to a lesser extent than for the accelerated storage. The general pattern indicated that the product packed in the co-extruded film pouches is slightly better, than that in polyethylene contact layer in the polyester-based laminate. The lower scores for this pouch may be due to the absorption of essential aromatic ingredients (scalping) by the polyethylene web (Baner et al. 1991), resulting in decreased aroma intensity. After 90 days of storage, there were noticeable decreases in the taste, and colour scores in both types of pouches. However, even upto the end of the study period, no off-flavours were observed.

The various microbial counts at the end of storage were similar to those observed in the beginning.

It may be concluded that pickled chicken could be packed, in flexible pouches made from either metallized polyester/polyethylene or polyamide (nylon) based co-extruded film pouches, as alternatives to other forms of conventional rigid containers. The product packed in flexible pouches would retain the desirable attributes well upto a minimum of 6 months, under the ambient conditions.

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Extraction of Pectin from Sugar-Beet Pulp and Intrinsic Viscosity-Molecular Weight Relationship of Pectin Solutions

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Sugar-beet pulp pectin was extracted by using HCl (pH 1.5, 80°C, 1 h), ammonium oxalate (0.25%, pH 3.5, 75°C, 1 h) and EDTA (0.5%, 90°C, 1 h). The best result was obtained with ammonium oxalate extraction in which the pectin content of the final product was 21.75%, although the efficiency among the procedure varied. The degree of methylation was higher than 50% in all pectin samples, indicating that the sugar-beet pectin is a high-methoxyl pectin. The degree of acetylation was high in all samples, indicating that the gelling capacity of sugar-beet pectin was poor. Magnitudes of intrinsic viscosity, and molecular weight of pectins obtained by extraction with HCl, ammonium oxalate, and EDTA were 110, 158, 208 ml/g and 41300, 30400, 15100, respectively. The data were fitted to Mark-Houwink-Sakurada equation as $\eta_i=0.3\ (M_{w,ave})^{0.613}$, which helped evaluate the average molecular weight of pectin solutions from sugar-beet with a knowledge of their intrinsic viscosity.

Keywords : Sugar-beet pulp, Waste, Pectin, Extraction, Intrinsic viscosity, Molecular weight.

Interest in the pectin substances has persisted because of their important role in maintaining the texture of raw, and processed fruits, and vegetables (Gee et al. 1958). Pectins exist in varying amounts in fruit cell walls, and have important nutritional, and technological properties, mainly because of their ability to form gels (Westerlund et al. 1991). Pectin is obtained mainly from the by-product or waste streams of the processing of apple, orange, and lemon juices (Kratchanova 1991). Sugar-beet pulp, a by-product of sugar extraction, is a well-known source of pectin (Phatak et al. 1988). Good quality pectin was prepared from ripe mango peel (Beerh et al. 1976) Pectin from Indian citrus peels have been characterized (Alexander and Sulebele 1980) and changes in pectin, and pectinase activity in developing guava fruits, (Pal and Selvaraj 1979) and apple fruits (Surinder Kumar et al. 1985) have also been studied.

The major use for pectins is, as a gelling agent in jams, and preserves (Kirk and Othmer 1967). For this application, a high methoxyl pectin, such as the citrus pectin with a degree of esterification (DE) greater than about 55%, is required (Zitko and Bishop 1965). Pectins of this type will only gel at low pH in the presence of high levels of sucrose or other humectants (King et al. 1986). The gelling ability of pectin depends on its solubility, and viscosity, which are a measure of its molecular weight (Rao 1993). Higher the molecular weight, the higher is its viscosity, and hence, the better is its grade (Rao 1993). Therefore, there is a need to measure the molecular weights of pectin solutions. Molecular weight distributions of pectin samples have been earlier determined by gel permeation chromatography (Phatak et al. 1988; Chou and Kokini 1987), and high performance size exclusion chromatography (HPSEC) (Fishman et al. 1991). In the present investigation, the average molecular weight was measured by light scattering technique, because of its ease, and simplicity in apparatus (Rao 1993).

The aim of the present investigation was to extract maximum pectin from sugar-beet pulp, determine some chemical characteristics of the extracted pectin, including the average molecular weight, and generate a knowledge of intrinsic viscosity of pectin solutions.

Materials and Methods

Materials : Sugar-beet pulp was obtained from Elazig Sugar Factory, Elazig (Turkey). The pulp was dried, ground and sieved to obtain 50 mesh fraction. The defatted ground beet pulp was used for pectin extraction. Experiments were performed in duplicate.

Pectin extraction : Pectin was extracted from sugar-beet pulp by three different extraction procedures, using HCl, ammonium oxalate (Phatak et al. 1988), and EDTA solvents. EDTA extraction procedure is a modification of the procedure of Phatak et al (1988). Sugar-beet pulp (1g) was mixed with 100 ml of 0.01 N HCl (pH 3.5), and 100 ml of 0.5% (w/v) of EDTA. The mixture was mechanically stirred for 60 min at 90°C, cooled to

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25°C, pH was adjusted to 7.5 by 50 ml of 0.1 M sodium phosphate, and 5 mg of protease enzyme (Sigma Chemical Co., P 5147) was added to hydrolyze the proteins present in the extract. Extract was incubated overnight at 37°C, and liquid phase separated from the solid mass by filtering. Pectin was coagulated with 4 volumes of ethanol, centrifuged for 30 min, pectin coagulate was washed twice with 45% ethanol, centrifuged for 30 min and dried.

Chemical determinations : Anhydrogalacturonic acid content of the sugar-beet pectin was determined by a colorimetric procedure, using p-hydroxydiphenyl colour reagent (Blumenkrantz and Asboe-Hansen 1973). Ash was determined by incinerating the sample overnight in a muffle furnace at 600°C (AOAC 1984). Methods given in Food Additive Codex were used for the methylation degree determination (NRC 1972). Since the amount of methoxyl in 100% methoxylated pectin is 16.32% (Gee et al. 1958), the methoxyl % was calculated from the following equation.

MeO % = (16.32/100) x (the degree of methylation)

The reaction of the ester groups in pectin with alkaline hydroxylamine at room temperature, to produce pectin hydroxamic acid, and acetohydroxamic acid, was applied to determine the degree of acetylation (McComb and McCready 1957). The amount of acetyl % was calculated by the following equation (Gee et al. 1958; McComb and McCready 1957) :

Acetyl % = (18.53/100) x (the degree of acetylation)

Viscometry : For viscosity determination, 0.5, 1.0, 1.5 and 2.0% pectin solutions, were prepared in 0.1 M sodium phosphate buffer (pH 7.0). The solutions were stirred for 10 min, and undissolved pectin particles were removed by squeezing the solution through cloth (Hwang et al. 1992). The viscosity of pectin solution, and solvent were determined at 25°C in Ubbelohde viscometer No Ic (Ubbelohde, Schott-Gerate, Hofheim, Germany). Densities of solutions were measured using pycnometer (Teknik Cam Co., Istanbul, Turkey) at 25°C. The calculation of the relative viscosity was made according to the following equation (Chen and Joslyn 1967) :

$$\eta_r = \eta/\eta_s = t_1 d_1/t_2 d_2 \quad \text{..... (1)}$$

Viscosity values were converted to specific viscosities (η_{sp}) using the following equation (Chen and Joslyn 1967) :

$$\eta_{sp} = (\eta-\eta_s)/\eta_s = \eta_r - 1 \quad \text{..... (2)}$$

η_{sp}/c known as reduced viscosity should ideally be independent of concentration, and it becomes so at the limit of zero concentration (Hwang et al. 1992). This limiting value of reduced viscosity is called the intrinsic viscosity (Tanglertpaibul and Rao 1987). Intrinsic viscosity was calculated using the following equation (Chou and Kokini 1987; Tanglertpaibul and Rao 1987) :

$$\eta_i = \lim_{c \to 0} (\eta_{sp}/c) \quad \text{..... (3)}$$

It can be shown (McMillan 1974) that reduced viscosity can be written in the form of Huggins's equation :

$$\eta_{sp}/c = \eta_i + k' \eta_i^2 c \quad \text{..... (4)}$$

The intrinsic viscosity was obtained by plotting the ratio of specific viscosity to pectin concentration (η_{sp}/c) against pectin concentration (c). The intrinsic viscosity was obtained by extrapolating the line, joining the points to zero concentration (Chou and Kokini 1987; Tanglertpaibul and Rao 1987).

Determination of molecular weight : The refractive indices of pectin solutions were measured by a refractometer (Atago Co. Ltd., Tokyo, Japan), using a mono-chromatic source of sodium vapour lamp (λ=5893 Å). The intensity of light scattered through pectin solutions was measured as the percentage of light transmitted through pectin solutions, as compared to that through 0.1 M sodium phosphate buffer (pH 7.0) by Spectronic 20 (Bausch and Lomb Inc., Rochester, NY) (Rao 1993). The experimental measurements were made at four concentrations (0.5, 1.0, 1.5 and 2.0 g/100 ml).

The average molecular weight of the ($M_{w,ave}$) was calculated by the following equation (Allock and Lampe 1981, Rao 1993).

$$1/M_{w,ave} = \lim_{c \to 0} (Hc/\tau) \quad \text{..... (5)}$$

Where H is given (Allock and Lampe 1981, Rao 1993) by

$$H = (32\pi^3 n_0^2/3\lambda^4 N_0)\ ((n-n_0)/c))^2 \quad \text{..... (6)}$$

The turbidity (τ) of the solutions was measured as the decrease in the intensity of a beam of light because of scattering. The decrease or attenuation depends on the length of the light path through the system (Allock and Lampe 1981), and by analogy to the Lambert law (Allock and Lampe 1981), it is possible to use

$$I/I_0 = e^{-\tau l} \quad \text{..... (7)}$$

The fraction of light scattered is generally very small (Allock and Lampe 1981), and it is a good approximation to express the exponential in the

above equation (Allock and Lampe 1981) as

$$e^{-\tau l} = 1-\tau l+(1/2)\ (\tau l)^2-(1/6)\ (\tau l)^3+\ldots\approx 1-\tau l \quad \ldots\ldots\ldots\ (8)$$

$$\tau l = 1-e^{-\tau l} = 1-(I/I_0) \quad \ldots\ldots\ldots\ (9)$$

$M_{w,ave}$ was measured by plotting Hc/τ versus c and interpolating to zero concentration, and noting the value of intercept (Allock and Lampe 1981).

Results and Discussion

Extraction of beet pulp with HCl, ammonium oxalate and EDTA yielded 17.71, 21.75 and 18.89% pectin, respectively, the highest yield being in case of ammonium oxalate procedure. Pectic substances as high-molecular-weight carbohydrate polymers are intimately associated with the insoluble cellular materials in plants (Gee et al. 1958). Because of their association in the three dimensional lattice of the cellular framework, it has not been possible to extract them completely from this matrix without changing their chemical, and molecular composition (Gee et al. 1958). For these reasons, most of the conclusions based upon the role of pectic substances in fruit texture have been drawn from the characterization of less than 50 to about 70% of the total pectic substances present (McCready and McComb 1954). For molecules which can exist with a variety of molecular weights, the relation between intrinsic viscosity, and molecular weights is one of the most important properties (Tanglertpaibul and Rao 1987; Gee et al. 1958). The chemical composition of pectin obtained by three different extraction procedures is shown in Table 1.

The higher galacturonic acid, and the lower ash contents of pectin are the two criteria for its purity (Hwang et al. 1992). It is evident from Table 1 that the purest pectin was produced by HCl extraction. However, the longevity of the extraction period in the HCl-procedure, compared to the other two procedures, might as well be the cause for the high galacturonic acid content of the pectin obtained by the HCl-extraction.

TABLE 1. CHEMICAL COMPOSITION OF PECTIN

Constituents, %	Extraction procedures		
	HCl, pH 1.5, 80°C 4 h	Ammonium oxalate, 0.25%, pH3.5, 75°C, 1 h	EDTA,, 0.5%, 90°C 1 h
Anhydro-galacturonic acid	68.20	64.10	53.20
Ash	4.02	9.57	12.36
Methylation degree	74.77	67.12	59.14
Methylation content	12.20	10.95	9.65
Acetylation degree	25.34	28.82	26.67
Acetyl content	4.70	5.34	4.94

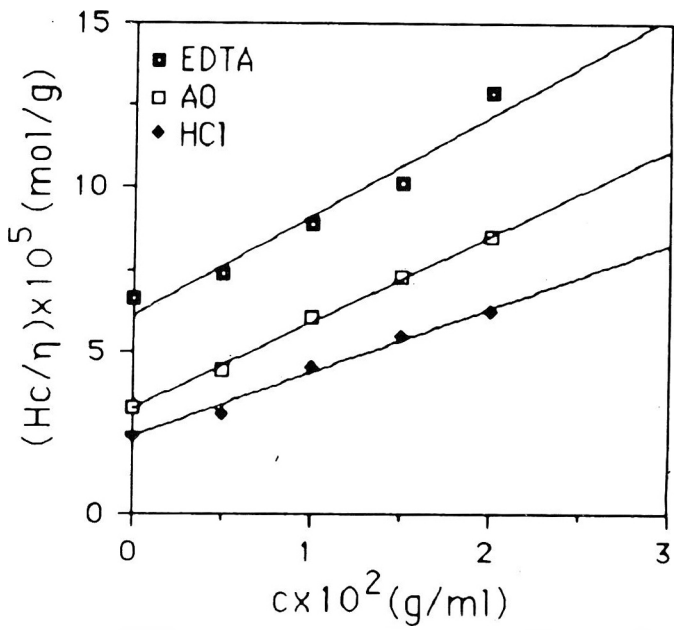

Fig. 1. Light-scattering determination of the molecular-weight of pectin in 0.1 M sodium phosphate buffer.

Since the carboxyl groups of the galacturonic acid units in pectin molecules are partially neutralized with cations (McCready 1966), the higher ash % content of pectin obtained by the EDTA extraction, as compared to other procedures, may be attributed to the complexing property of EDTA with metal ions. Phatak et al (1988) also reported that the ash % of pectin obtained by EDTA extraction was higher, as compared to that obtained by other methods.

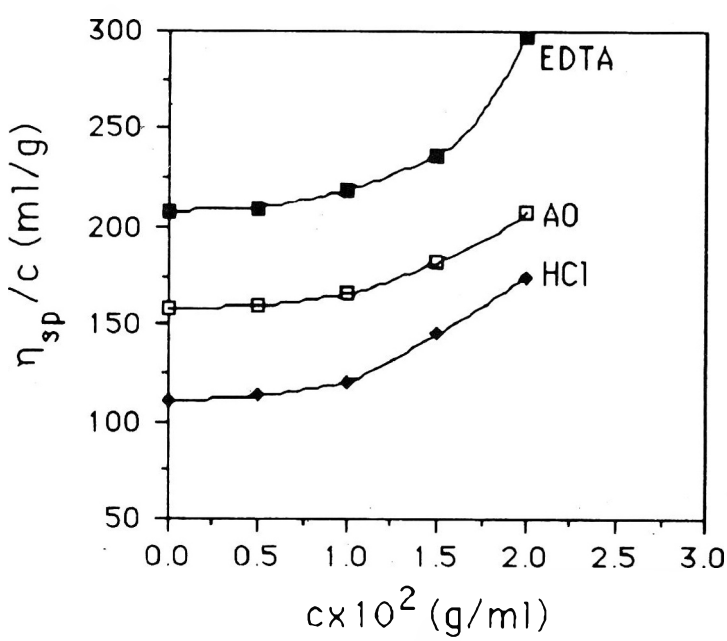

Fig. 2. Plot of η_{sp}/c versus c

The degree of methylation in all samples analyzed was found to be over 50%, indicating that the sugar-beet pectin had a high-methoxyl content. The degree of acetylation varied according to the isolation procedures. The gelling capacity of pectin decreased with an increase in the degree of acetylation (McCready 1966).

Hc/τ values were calculated and plotted against concentration of pectin solutions (Fig.1). Hc/τ versus c showed similar trends and straight lines with linear regression coefficients in the range 0.99-1.0.

The reciprocal of the intercept is the average molecular weight. Fig. 2 shows the method of determination of the magnitude of intrinsic viscosity by plotting η_{sp}/c versus the concentration of pectin solutions. Magnitudes of intrinsic viscosity, and molecular weights of pectins obtained by HCl, ammonium oxalate and EDTA extraction were 110, 158, 208 ml/g and 41300, 30400, 15100, respectively.

Intrinsic viscosity is a characteristic of macromolecules that is related directly to their ability to disturb flow, and indirectly to their size, and shape (Tanglertpaibul and Rao 1987). The average molecular weight of pectin, and its intrinsic viscosity in a solution are related by the Mark-Houwink-Sakurada equation (Brandrup and Immergut 1975) :

$$\eta_i = K (M_{w,ave})^{\alpha} \quad \text{..... (10)}$$

$$\ln \eta_i = \ln K + \alpha \ln M_{w,ave} \quad \text{..... (11)}$$

Intrinsic viscosity values obtained from plot of η_{sp}/c vs. c were used for plot of ln η_i vs. In $M_{w,ave}$. The coefficient K and exponent were calculated from the intercept, and slope of the straight line fitted to In η_i and ln $M_{w,ave}$ (K=0.3, α=0.613).

The exponent α and K are dependent on the nature of the molecule, and solvent, and on the temperature (Tanglertpaibul and Rao 1987). The values determined are applicable for pectin solutions in 0.1 M sodium phosphate buffer solvent alone, which limits the applicability of these coefficients. It is possible to determine the average molecular weight of pectin samples by measuring the intrinsic viscosity of pectin with α and K coefficients.

Nomenclature

C Concentration of the solution, g/ml

d_1 Density of solution, g/ml

d_2 Density of solvent (0.1 M sodium phosphate buffer) = 1.4353 g/ml

k' Huggins's constant

I/I_0 Fraction of light transmitted through 1 cm length of solution

I Length of the light path in the solution, cm

$M_{w,ave}$ Average molecular weight

n_0 Refractive idex of the solvent (0.1 M sodium phosphate buffer) = 1.3368

n Refractive index of the solution

N_0 Avogadro number = 6.023 x 10^{23}

t_1 Time taken by solution to flow in viscometer, s

t_2 Time taken by solvent to flow in viscometer, s

η Viscosity of solution, cps

η_s Viscosity of solvent, cps

η_{sp} Specific viscosity

η_i Intrinsic viscosity, ml/g

λ Wave length of light = 0.589x10^{-4}cm

τ Turbidity of the solution, cm^{-1}

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Metal Contents in Some of the Processed Foods and Their Effect on the Storage Stability of Pre-cooked Dehydrated Flaked *Bengal*gram *Dhal*

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Mineral composition of some convenience food products like pre-cooked, and dehydrated vegetable *pulav*, instant *halwa* mix, instant *upma* mix, preserved *chapati*, spiced potato *chapati*, instant spiced *dhals*, *alu chholay*, rice noodles, *chikki* and canned curried meat has been investigated. The levels of sodium, potassium, calcium, iron, zinc, manganese, and copper were found to vary between 42-26,000, 130-6710,17.4-725, 39-170, 6.7-36.6, 1.8-19.2 and 1.6-17.0 ppm, respectively, in these processed foods. In commercially procured, and stored canned curried meat, tin, copper and zinc were found to vary between 60-152, 1.2-12.4, and 15.3-38.8 ppm, respectively. The levels of nickel, iron and copper were found to vary between 0.40-8.70, 0.80-8.20 and 0.015-0.025 ppm, respectively, in different commercial brands of *vanaspati*, whereas nickel content was found to vary from 1.10-2.10 ppm in different commercial brands of chocolates. In dehydrated flaked *dhal*, sodium chloride exhibited slight pro-oxygenic activity, but copper, iron and cobalt exhibited strong pro-oxygenic activity. Only at 500 ppm level, nickel exhibited marginal antioxygenic activity.

Keywords : Convenience foods, Canned meat, Mineral composition, Sunflower oil, Peroxide value, *Vanaspati*, Chocolate, Metal ions.

With increasing industrialization, contamination of foods with heavy metals has become widespread (Lindsay and Sherlock 1982). Industrial effluents are often released to sea, river and even to irrigation channels or in fields, and thus some of the metallic constituents of the effluents enter the food chain (Fimreite 1971; WHO 1972). While some of the metals like sodium, potassium, calcium, and iron are of nutritional significance (Young and Scrimshaw 1979), heavy metals like lead, cadmium, chromium, nickel, and cobalt are toxic, and therefore, their concentration shall be monitored in foods (Moore et al. 1979; Grasso 1975; Jathar et al. 1981). In addition, metal contaminants may also occur, as a result of contact with processing equipment (Semwal and Arya 1994), and packaging materials (Concon 1988). Heavy metals, and their salts are also used as catalysts during processing of foods. For example, metallic nickel is used by hydrogenation industry in the manufacture of *vanaspati* (Murti and Achaya 1974). Though efficient filtration system for removal of catalysts is available, and used by the industry (Satya Prakash and Sarin 1991), retention of trace amounts of metal catalysts cannot be ruled out. Influence of copper, and iron in ghee on oxidative deterioration has been investigated (Unnikrishnan and Bhimasena Rao 1977). Traces of cupric copper have been demonstrated to accelerate nitrate induced corrosion in tinplates (Eipeson and Sastry 1984). Besides being toxic, metal contaminants affect taste, and flavour of foods, and their stability during storage (Semwal and Arya 1992). Some of the transition metal ions like iron, copper and cobalt are known to catalyze lipid peroxidation, which is a major deteriorative reaction in dehydrated convenience foods (Semwal and Arya 1992). Keeping some of these aspects in view, the mineral composition of some of the processed convenience foods, and also some popular brands of *vanaspati* and chocolates have been studied, and the data are reported in the present paper.

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Materials and Methods

Instant *halwa* mix, instant *upma* mix, *chikki* and pre-cooked dehydrated vegetable *pulav* were procured from manufacturers in Mysore, while noodles were from a manufacturer from Bombay. Instant spiced *dhals*, *alu chholay* and *chapatis* were processed in the laboratory, using methodology reported by Patki and Arya (1994), Ghosh et al (1979), and Arya et al (1977), respectively. Canned curried meat samples (*keema* and chunks) were obtained from different Army Service Corps Depots. Seven brands of *vanaspati* and six types of chocolates were procured from local market. Commercially available good quality dehusked *Bengal*gram *dhal* and refined sunflower oil were also purchased from local market.

Sample preparation : Total content of each pack was blended in a laboratory auto mix (Model 842 INT, Sumeet, Nasik, India), and 20 g sample was digested in Kjeldahl flasks, using nitric acid, and sulphuric acid mixture for oxidation of carbonaceous matter as per Jacob (1958). For each sample, blank

was prepared simultaneously by taking same amounts of nitric, and sulphuric acids mixture. Concentrations of various metals were calculated by determining the absorbance at appropriate wavelengths, using atomic absorption spectrophotometer (Model AA 670, Shimadzu, Kyoto, Japan), by aspirating the solution into the oxygen-acetylene flame (Cowley 1978). The instrument was calibrated by using standard solutions (1-6 mg/litre) of various metal salts.

Preparation of precooked dehydrated flaked Bengal*gram* dhal : *Bengal*gram *dhal* (2 kg) was soaked in 2 l water, containing required quantities of sodium chloride or sulphates of iron, copper, cobalt or nickel for 3 h at ambient temperatures (15-35°C), and 47-78% RH. The soaked *dhal* was cooked, and flaked as per the method reported by Semwal and Arya (1994). The flakes were dried to 5% moisture at 80°C in a hot air cabinet dryer (Model SDA, Type E, Kilburn, MacNeil and Magor Ltd. Calcutta, India). Required quantities (2 kg) of the flakes were treated with sunflower oil (15%, w/w) and mixed thoroughly in a stainless steel vessel, using a laddle.

Storage and evaluation : Treated and untreated *Bengal*gram *dhal* flakes (100 g) were packed in polypropylene (75 μ) pouches, and stored in an incubator maintained at 37±1°C. Initially, and at regular intervals, the samples were analyzed for peroxide value, which indicated the rate of lipid peroxidation (Arya and Thakur 1986).

Results and Discussion

The levels of sodium, potassium, calcium, iron, manganese, copper and zinc in processed convenience foods are given in Table 1. Sodium is present in largest concentrations in almost all the convenience foods, mainly due to incorporation of sodium chloride in these products during processing for taste considerations, except for *halwa* mix and rice noodles. Potassium, on the other hand, varied between 130 and 6710 ppm in different processed foods. Concentrations of potassium were also high in most of the processed foods, except for *halwa* mix and rice noodles. *Chikki* had the highest concentration of potassium, which may be ascribed to high levels of potassium in oilseeds, and pulses (Gopalan et al. 1985). Relatively, copper, and manganese were present in higher concentrations in instant pulses. Earlier also, legumes, and pulses have been reported to contain larger concentrations of these elements (Gopalan et al. 1985).

The levels of tin, zinc, and copper in 12 samples of commercially available meat chunks and *keema* samples, stored at various places for one year, are given in Table 2. In all these samples, the concentrations of tin varied between 60 and 152 ppm, and its levels were below the maximum levels permitted in foods (PFA 1954). The levels of zinc varied between 15.3 and 38.8 ppm, and those of copper between 1.2 and 12.4 ppm. Except in one sample in which the level of copper was found to be slightly higher, the levels of tin, zinc and copper were found to be below the prescribed limits in all other samples, even after one year of storage under different climatic conditions.

Till recently, there were no limits of nickel content prescribed for *vanaspati*, but there has been considerable concern about the high level of this element in some brands of *vanaspati* (Satya Prakash and Sarin 1991), which is also used by chocolate manufactures as a hardening agent (Minifie 1989). It may be seen (Table 3 and 4) that nickel content in seven brands of *vanaspati*, and six types of chocolate varied between 0.40 and 8.7 and 1.10 and 2.1 ppm, respectively. The levels of

TABLE 1. MINERAL COMPOSITION (ppm) OF PROCESSED CONVENIENCE FOODS

Products	Sodium	Potassium	Calcium	Iron	Zinc	Manganese	Copper
Instant *halwa* mix	48	375	141	70	10.7	2.6	2.2
Instant *upma* mix	12,500	2100	575	160	24.4	14.1	4.4
Vegetable *pulav*	18,500	4360	365	82	26.3	6.7	3.3
Spiced redgram *dhal*	24,500	4800	370	110	30.3	16.0	13.0
Spiced *Bengal*gram *dhal*	26,000	4560	400	141	36.6	19.2	13.5
Spiced blackgram *dhal*	21,300	4200	420	170	33.1	18.8	17.0
Chapati	6,850	2250	262	75	26.5	6.7	5.1
Spiced potato *chapati*	9,825	3200	725	118	23.3	5.1	3.2
Alu chholay	18,200	2200	200	118	24.7	6.6	2.1
Rice noodles	42	130	17	39	6.7	1.8	1.6
Chikki	1,060	6710	130	77	31.9	5.8	3.7

TABLE 2. METAL LEVELS (ppm) IN CANNED MEAT STORED AT VARIOUS PLACES FOR ONE YEAR UNDER DIFFERENT CLIMATIC CONDITIONS

Product	Place of storage	Storage temp, °C	RH, %	Tin	Zinc	Copper
Mutton chunk	Missameri	9 - 36	52 - 91	105	26.7	3.0
Mutton chunk	Missameri	9 - 36	52 - 91	92	25.8	1.3
Mutton chunk	Jabalpur	1 - 46	32 - 84	152	38.5	1.3
Mutton *keema*	Jodhpur	-2 - 49	18 - 82	100	15.6	7.8
Mutton *keema*	Jodhpur	-2 - 49	18 - 82	60	15.8	3.1
Mutton *keema*	Madras	18 - 39	61 - 88	100	20.0	3.6
Mutton *keema*	Jabalpur	1 - 46	32 - 84	96	19.6	1.2
Mutton *keema*	Jabalpur	1 - 46	32 - 84	100	21.6	1.2
Mutton *keema*	Jabalpur	1 - 46	32 - 84	80	23.8	12.4
Mutton *keema*	Jabalpur	1 - 46	32 - 84	80	38.8	4.5
Mutton *keema*	Jabalpur	1 - 46	32 - 84	92	23.5	5.5
Mutton *keema*	Bengdubi	5 - 39	56 - 89	79	15.3	7.6

iron, and copper in *vanaspati* samples varied from 0.80 to 8.20 and 0.015 to 0.025 ppm, respectively. The levels of nickel and iron were also found to vary in different batches of *vanaspati* of the same brand. The higher nickel, and iron contents in some of the brands of *vanaspati* could be due to non-adherence of quality standards during hydrogenation, and subsequent post-refining of hydrogenated oil.

Effect of metal ions on the stability of pre-cooked dehydrated Bengal*gram* dhal: The results on changes in peroxide value as a result of storage (Table 5) showed that addition of copper, cobalt and iron even at 1 ppm level considerably enhanced the rate of lipid peroxidation in dehydrated flaked *dhal*. The catalytic effect of these ions was concentration dependent, and increased with rise in concentration of metal ions from 1 to 500 ppm. Among the three metal ions, copper was the most pro-oxidant, followed by cobalt and iron. It has been claimed that transition metal ions, at higher concentrations exert antioxygenic activity, decomposing peroxides (Emmanual and Lyaskovskaya 1967). This effect has not been observed in dehydrated flaked *Bengal*gram *dhal* even at 500 ppm levels of copper, cobalt and iron. Incorporation of nickel upto 50 ppm level did not exert any significant effect, but it exerted very slight antioxygenic effect at 500 ppm level. Incorporation of salt also slightly enhanced the rate of lipid peroxidation in dehydrated flaked *Bengal*gram *dhal*, and its catalytic action increased with rise in concentration. Same pattern was observed in dehydrated rice (Semwal and Arya 1992).

From the foregoing study, it is evident that the levels of metal ions vary considerably in various processed foods, either as a result of variation in the recipe or as a result of contamination during processing or as a result of interaction with the packaging material. However, in canned curried mutton chunks and *keema*, the levels of tin, zinc and copper remained below the permitted levels even after one year of storage at various places under varying climatic conditions. Though the level of nickel in most of the samples of *vanaspati* and chocolates analyzed were also found to be within

TABLE 3. NICKEL, IRON AND COPPER CONTENTS (ppm) IN *VANASPATI* SAMPLES

	Nickel			Iron			Copper		
Brand	Mean	Range	CV	Mean	Range	CV	Mean	Range	CV
A	2.5	0.4 - 8.7	99.0	3.1	1.2 - 8.2	76.9	0.018	0.015 - 0.020	14.0
B	2.7	1.6 - 4.5	45.2	2.9	0.8 - 5.0	58.5	0.018	0.015 - 0.020	12.9
C	1.3	1.1 - 1.4	9.8	6.0	4.3 - 7.0	20.6	0.017	0.015 - 0.020	14.0
D	4.5	3.6 - 5.8	21.7	3.7	1.2 - 6.6	60.4	0.022	0.020 - 0.025	10.9
E	4.4	3.9 - 4.8	8.5	3.6	2.9 - 4.7	21.9	0.023	0.020 - 0.025	10.1
F	4.2	3.8 - 4.6	8.4	3.6	3.2 - 3.9	8.0	0.018	0.015 - 0.020	12.9
G	1.8	1.5 - 2.0	10.7	5.3	4.9 - 5.6	5.6	0.018	0.015 - 0.020	12.9

C.V. (%) - Coefficient of variation

TABLE 4. NICKEL CONTENT (ppm) IN CHOCOLATES AND CREAM BAR

Brand	Type	Mean	Range	CV
A	Milk chocolate	1.3	1.2 - 1.4	6.6
B	Milk chocolate	1.2	1.1 - 1.3	3.4
C	Milk chocolate	1.5	1.4 - 1.7	8.2
D	Milk chocolate	1.6	1.5 - 1.7	6.4
E	Cream bar	1.3	1.2 - 1.5	10.0
F	Cream bar	1.9	1.8 - 2.1	14.7

C.V. (%) - Coefficient of variation

the permissible limits, a few samples of *vanaspati* had nickel levels as high as 8.7 ppm, and therefore, there is a need to monitor the level of nickel in *vanaspati* to ensure adoption of more effective filtration technique by the industry. Besides, toxic effect of trace metal ions at higher levels, some of these metal ions (copper, iron and cobalt) catalyze lipid peroxidation and resultant off-flavours in instant cooking *dhals* and other foods. It is, therefore, essential that their maximum levels be specified in processed foods to ensure their safety and stability.

TABLE 5. EFFECT OF SODIUM CHLORIDE AND TRANSITION METAL IONS ON THE CHANGES IN PEROXIDE VALUE (meq O_2/kg fat) OF PRECOOKED DEHYDRATED FLAKED *BENGALGRAM DHAL* HAVING ADDED SUNFLOWER OIL AND STORED AT 37±1°C

Metal ions	Peroxide value (meq O_2/kg fat)			
	Storage period (days)			
	0	30	45	60
Control	8.6	29.8	50.6	67.5
Copper, ppm				
1	9.2	44.4	70.8	104.2
5	9.4	60.1	85.0	158.6
50	9.4	112.9	202.1	489.2
500	9.8	245.5	430.6	971.7
Cobalt, ppm				
1	9.0	34.8	58.2	77.6
5	9.1	38.7	65.1	91.3
50	9.3	47.2	78.3	120.1
500	9.5	100.6	192.1	454.4
Iron, ppm				
1	8.9	32.2	53.4	73.9
5	9.2	34.1	55.5	76.1
50	9.5	42.6	66.5	90.0
500	9.8	63.4	101.4	138.1
Nickel, ppm				
1	8.8	30.5	51.8	68.7
5	9.0	31.6	52.9	69.9
50	9.1	28.1	49.4	67.2
500	9.0	26.1	46.7	62.9
Sodium chloride, %				
0.5	9.2	31.7	53.2	72.3
1.0	9.1	33.9	58.5	77.6
2.0	9.1	36.2	62.5	82.9

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Loss of Moisture and Sulphur Dioxide During Air Cabinet Drying of Mango *Puree*

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Effect of addition of powdered cane sugar to mango *puree* for raising the total soluble solids to 30° Brix, and drying it as plain mango bar, or after adding 4.5% soy protein concentrate or 2% desiccated coconut powder in a cross flow air cabinet drier at 63±2°C on loss of moisture, and retention of sulphur dioxide has been investigated. Mango *puree* exhibited constant rate drying for the first 1.5 hour. During this period the drying rate (g H_2O/100 g solids/h) of plain mango bar was 35.5, as compared to 33.0 for mango-desiccated coconut bar or 30.0 for mango-soy protein concentrate bar. Their critical moisture contents (% db) were 144 and 130, respectively. During falling rate period, the drying rate of plain mango bar was higher upto the first 6.5 h, but subsequently it became lower than the drying rates of other two samples. Loss of free, and total sulphur dioxide during drying of *puree* followed first order kinetics, and free sulphur dioxide was lost more rapidly than total sulphur dioxide. Concentration of total sulphur dioxide (C) in three *puree* preparations can be represented as a function of drying time (t) by one straight line with the equation $\log_e C = 7.308-0.0363\ t$.

Keywords : Air cabinet drying, Drying rate, Fortified mango *puree*, Fruit bar, Mango bar, Sulphur dioxide.

Mango bar is an age-old product of the Indian sub-continent (Anon 1957). It is prepared traditionally by sun-drying ripe mango *puree* (Anon 1957; CFTRI 1990). But, the process is slow, unhygienic, yields a dark brown product, and the losses may be high during rainy season (Jagtiani et al. 1988). To overcome these problems, Mathur et al (1972) prepared mango bars from ripe mango *puree* by three stage drying (1 h at 80°C, followed by 2-3 h at 70°C, and 2–3 h at 65°C) in a cross flow air drier, while Nanjundaswamy et al (1976) and Rao and Roy (1980) used single stage drying at 60 or 70°C. Sugar is added to *puree* to raise the total soluble solids content, and reduce the drying period, though it lowers the drying rate (Rao and Roy 1980). Other workers (Bains et al. 1989; Chan and Caveletto 1978; Singh et al. 1989) have studied some of the drying characteristics of unsweetened or sweetened fruit *puree* in forced air circulation cabinet driers. Conversely, Khedkar and Roy (1983) studied the absorption and retention of sulphur dioxide in raw mango slices during drying and dehydration. However, Mir and Nath (1993) have fortified mango *puree* with desiccated coconut powder/soy protein concentrate, which may alter drying characteristics. Sulphur dioxide, when added to mango *puree*, as potassium metabisulphite, reduces browning, and retards microbial spoilage (Mir and Nath 1993). Losses of sulphur dioxide occur during drying (Mir 1990). In this paper, the effects of fortification of mango *puree*, added with desiccated coconut powder or soy protein concentrate, on its drying behaviour, and loss of sulphur dioxide are reported.

Drying : Mango *puree* of 'Langra' variety was adjusted to 30° Brix, and 0.6% acidity, using cane sugar powder, and citric acid, respectively. *Puree* was heated to 91-93°C for 2 min, about 1000 ppm sulphur dioxide (1734 ppm potassium meta bisulphite) added, packed in 3 L glass containers, sealed, and stored at 7±3°C till required. *Puree* was dried, as such (plain mango bar), or after fortifying with 4.5% soy protein concentrate or 2.0% desiccated coconut powder (Mir and Nath 1993). Each formulation (3.5 kg) was homogenised in a blender, and spread uniformly to a 10 mm thickness (tray loading 9.8 kg/m^2) on aluminium trays, that had been smeared with vegetable fat to facilitiate easy removal of the dried product. The trays were loaded into a forced air circulation cabinet drier, and the product was dried for 14-16 h at 63±2°C, and linear air velocity of 2.42 m/sec. During this period, the thickness of the layer reduced to about 5 mm.

Chemical analysis : During drying, 20 g samples were drawn at intervals of 1 h from the selected trays, and analyzed for moisture by vacuum oven method (temperature 70±2°C for 6 h), free and total sulphur dioxide by modified Ripper's titration method (Ranganna 1986). Samples were drawn in triplicates from three different locations of a tray, and the values were averaged.

Moisture loss during drying : Residual moisture contents of the samples were plotted against

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corresponding drying period. Drying rates were calculated by a graphical method (Perry et al. 1984), and plotted, as a function of time.

Sulphur dioxide loss during drying : Free, and total sulphur dioxide contents of samples were plotted against drying time on a semi-log paper, and the data were regressed using a first order reaction kinetics model (Perry et al. 1984).

Initial moisture content of mango *puree* was highest, and its moisture content remained higher than the other two samples (mango-soy protein concentrate or mango-desiccated coconut powder) for initial 3 h drying (Fig.1). After this period, the pattern of the curves changed, and the moisture content of the plain mango bar was 18 g/100 g bone dry product (18% db), as compared to 21% (db) for the mango-desiccated coconut powder sample, and 25% (db) for mango-soy protein concentrate.

Drying rate curves (Fig. 2) for all the three samples exhibited a constant rate period of drying for initial 1.5 h, as sufficient moisture was available at the surface for evaporation (Hawlader et al. 1991). However, the drying rate during this period, $(dM/dt)_c$, was higher for the plain mango *puree* (35.5 g H_2O/100 g solids $^{-1}$h than for mango – soy protein concentrate (30.0 g H_2O/100 g solids

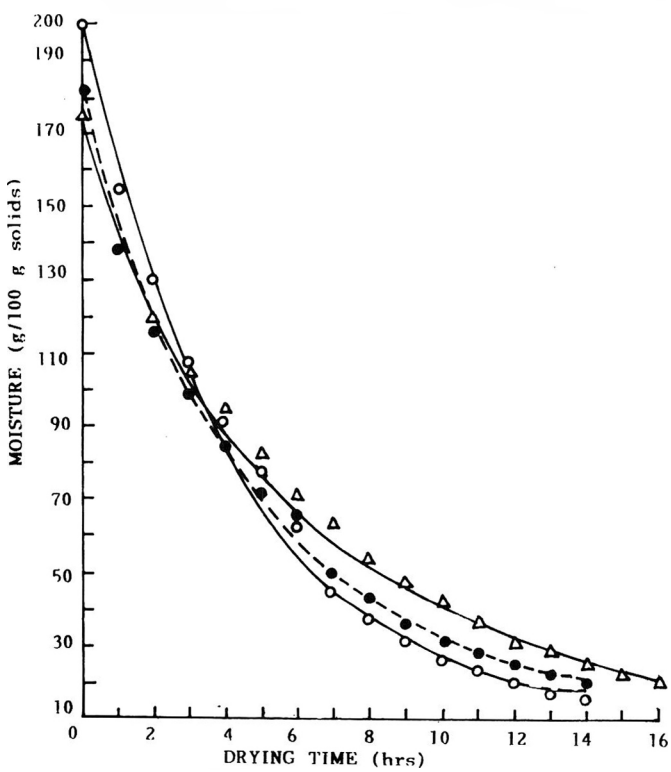

Fig. 1. Moisture content (% db) of plain mango puree (O----O), mango desiccated coconut puree (●---●) and mango-soy protein concentrate puree (Δ---Δ) during air cabinet drying at 63 ± 2°C

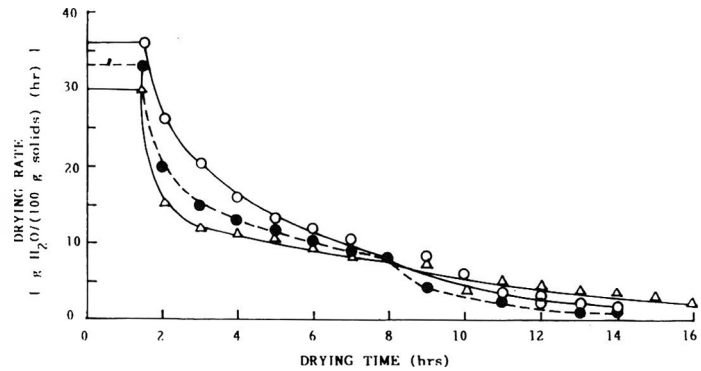

Fig. 2. Dry rate curves for plain mango puree (O----O), mango desiccated coconut puree (●---●) and mango-soy protein concentrate puree (Δ---Δ) during air cabinet drying at 63 ± 2°C

$^{-1}$h or mango–desiccated coconut powder (33.0 g H_2O/100 g solid $^{-1}$h samples. $(dM/dt)_c$, depends upon the mass transfer coefficient (K_g), drying surface area, water vapour pressure at product surface, and partial pressure of water vapour in drying air (Brennan et al. 1990). Since, the drying was carried out simultaneously using the same tray load, and surface area, the last three parameters were constant during the constant rate period. Therefore, the differences in $(dM/dt)_c$ are due to K_g only. Critical moisture content (M_c) for the plain mango bar was 144% db, against 130% db for the other two samples. As drying proceeds, the drying interface recedes to the interior, its distance from the surface increases, and the moisture movement within the solids occurs through liquid/vapour diffusion only from the upper side, which is exposed to the drying air (Hallstrom 1990).

The $(dM/dt)_f$ value for plain mango bar was higher upto the first 6.5 h, and thereafter, the drying pattern changed (Fig.2), due to differences in their chemical composition (sugars, proteins and fats). Greater water binding capacity of sugar-rich plain mango bar (Rahman and Lamb 1991) lowered its drying rate. As a result of these changes, the differences in $(dM/dt)_f$ for all the three samples became less during the final stages of drying.

Semi-log plots of residual sulphur dioxide (free or total) contents against drying times gave straight lines (Fig. 3), thereby showing highly significant ($P<0.01$) values of R^2 (0.952 to 0.997) (Table 1). It shows that the losses of free, and total sulphur dioxide during drying followed the first order kinetics, as reported earlier by Wedzicha (1987) for vegetable dehydration. The data for free sulphur dioxide were scattered, and the extent of scattering depended upon the type of sample (Fig. 3). On the

TABLE 1. REGRESSION VALUES OF CONSTANTS FOR THE MODELY Y = a e^{bx} FOR LOSS OF SULPHUR DIOXIDE DURING AIR CABINET DRYING OF MANGO *PUREE*

Sample	$\log_e a$	b	R^2	Calculated F	First order rate constant
		Residual free sulphur dioxide			
Plain mango *puree*	6.501	- 0.0074	0.957	89.9**	- 0.178
Mango-desiccated coconut powder *puree*	6.497	- 0.0687	0.997	1340.1**	- 0.158
Mango-soy protein concentrate *puree*	6.474	- 0.1080	0.952	79.8**	- 0.248
		Residual total sulphur dioxide			
Plain mango *puree*	7.321	- 0.0396	0.994	702.3**	-0.911
Mango-desiccated coconut powder *puree*	7.343	- 0.0412	0.991	423.5**	- 0.095
Mango-soy protein concentrate *puree*	7.291	- 0.0344	0.953	81.3**	- 0.079
Common tosamples	7.308	- 0.0363	0.972	179.3**	- 0.084

** Highly significant ($P < 0.01$).

other hand, variations in the total sulphur dioxide contents of the three samples were low, and it was possible to draw one common line for them. This line was found to have the following equation :

$$\log_e C = 7.308 - 0.0363\ t$$

Where C is total sulphur dioxide content (ppm) of sample at any time t (h).

Slopes of the semi-log plots in Fig. 3 represent the rate of loss sulphur dioxide, and are given, as constant 'b' in Table 1. Rate of loss of free sulphur dioxide was always greater than that of total sulphur dioxide, because some of free sulphur dioxide escapes from product to the drying air during drying. Additionally, free sulphur dioxide is known to bind with reducing sugars, pectins, and amino acids (Bolin et al. 1985). In the early stages of drying, much of the sugars are present as sucrose. But, as drying proceeds, more and more reducing sugars are produced by acid inversion of non-reducing sugars, and some of the free sulphur dioxide forms complex with them, thereby raising bound sulphur dioxide concentration in the product (McBean 1967). Total, and reducing sugar contents of the plain mango bar (71.5% and 27.8% db, respectively), and the mango-desiccated coconut powder (66.6% and 17.1% db, respectively) were higher, as compared to the mango – protein concentrate (61.4% and 18.8% db, respectively) (Mir 1990; Mir and Nath 1995). Therefore, the rate of loss of total sulphur dioxide for the former two samples were lower (Fig. 3 and Table 1).

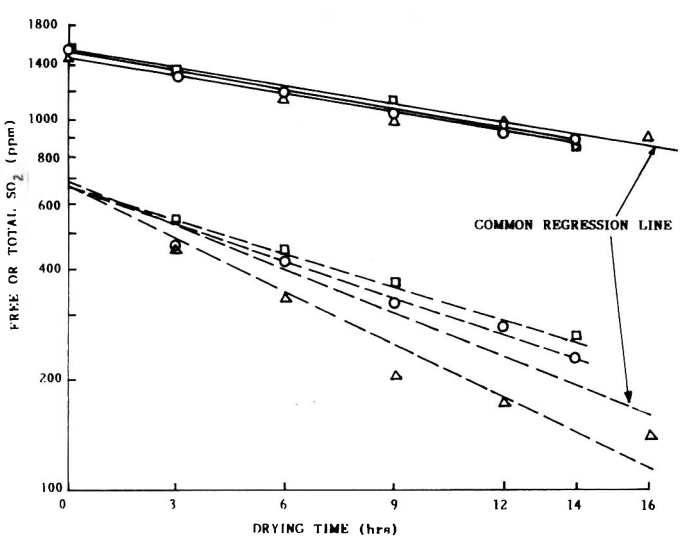

Fig. 3. Residual free and total SO_2 contents (ppm) of plain mango puree (O—O), mango desiccated coconut powder puree (●—●) and mango-soy protein concentrate puree (Δ—Δ) during air cabinet drying at 63 ± 2°C

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Physico-chemical and Nutritional Traits of Rice Bran Protein Concentrate-based Weaning Foods

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High protein weaning blends from acid stabilized rice bran protein concentrate and rice, with or without greengram have been assessed for their physico-chemical characteristics, and nutritional quality. The protein contents of weaning blends 1 and 2 were 20.6 and 21.8%, respectively. These represent calorie dense formulations with 377 and 372 calories/100 g, which conform to ISI specifications. The protein efficiency ratios of blends 1 and 2 were 2.1 and 2.2, respectively. Formulations possessed high water absorption capacity of 310 ml/100 g at 27°C, which increased to 360 ml/100 g at 60°C. The gruel viscosities of reconstituted blends were low, when compared to two rice-based commercial preparations.

Keywords : Rice bran protein concentrate, Weaning foods, Protein efficiency ratio, Water absorption, Consistency, Viscosity.

Rice is one of the major staple foods in human diets in many countries worldwide (Kaul 1975). Rice bran, a by-product of rice milling industry, has potential for utilization, as food by virtue of its nutritional quality (Barber and Barber 1980). Some of the problems encountered while utilizing rice bran for food include presence of silica, hulls, and rapid quality deterioration, due to oxidative rancidity, and enzyme activity (Barber and Barber 1980). Acid stabilization treatment can preserve full fat rice bran more effectively (Prabhakar and Venkatesh 1986). Rice bran can be incorporated in breads, muffins, breakfast foods, snack items and biscuits, as a source of dietary fibre (Barber and Barber 1974, 1980; Saunders 1986, 1990; Skurray et al. 1986; Babcock 1987; Sloan and James 1988). Besides, some workers have standardized methods for the preparation of weaning foods based on soy-whey blends (Kapoor and Gupta 1981); potato (Barinder Kaur and Gupta 1982); malted ragi and green gram (Malleshi and Desikachar 1982). Oil, and proteins are two valuable components obtained from rice bran. Defatted bran serves, as a good source of protein (14-18%), which can be incorporated into many food products (Saunders 1986). However, its use in infant foods is limited, because of high fibre content. This problem can be overcome by utilizing protein concentrate made from defatted rice bran. Full fat rice bran can also be used, but it leads to loss of valuable rice bran oil as a by-product. Earlier studies have shown that protein concentrate prepared from acid stabilized rice bran was of good nutritional quality (Prakash 1991). In the present study, two weaning foods, based on acid stabilized rice bran were prepared, and analyzed for physico-chemical, and nutritional characteristics. The results are reported in this paper.

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Materials and Methods

Fresh rice bran was procured from paddy (*Oryza sativa*) of 'Gowri Sanna' variety from local rice mill. The bran was subjected to stabilization by treating with concentrated hydrochloric acid (12 N) at the rate of 4.0 ml/100 g on v/w basis by the method described by Prabhakar and Venkatesh (1986). The stabilized bran was defatted using food grade hexane by cold extraction (Barber and Barber 1985). Protein concentrates were prepared from the defatted rice bran by alkali peptization (at pH 11.0, using 1 N sodium hydroxide), followed by isoelectric precipitation (at pH 4.0, using 1 N hydrochloric acid) of the proteins (Prakash and Ramanatham 1994). The wet protein concentrate was used to formulate the two types of weaning foods.

Weaning mix 1 consisted of rice bran protein concentrate (25%), and rice flour (75%). Weaning mix 2 consisted of rice bran protein concentrate (20%), rice flour (60%), and whole greengram flour (20%). Rice, and greengram (*Phaseolus vulgaris*) were procured from local market, cleaned, and powdered in a plate mill (Diaf make, Chandra Co., Madras). For preparation of weaning foods, rice flour and/or greengram flour was added to wet protein concentrate, and mixed well with water. The dispersion was subjected to simultaneous cooking and roller drying (Escher Wyss double drum drier, 61 cm in diam and 112 cm in length, revolving at 3-4 rpm at 3 kg/cm^2 steam pressure). The roller dried flakes were powdered in a mixer (Bajaj

supermix, Bajaj Co., Bombay) to pass through 40 mesh sieve. These blends were stored, and used for determining proximate composition, certain functional properties, and protein efficiency ratio.

Proximate analysis of samples was done according to AOAC (1984). Standard methodologies were used for determination of bulk density (Wang and Kinsella 1976), water holding capacity (Prasanappa et al. 1972), and water absorption capacity (Janicki and Walczak 1954). Consistency of reconstituted weaning foods was measured by modifying the ring test of Bookwalter et al (1968).

The calorie content of the foods was calculated, as per the method described by Gopalan et al (1989). For determining the apparent viscosity of weaning food slurries, 20 g material was suspended in 100 ml distilled water, left to equilibrate, and apparent viscosity was determined with a Synchro electric viscometer (Brookfield Engineering Laboratory Inc., Spoughton, Massachusetts, USA), using RVT spindle No. 4 at 100 rpm. The gruel viscosity was expressed in centipoise (CP). Viscosities of the samples were also measured after mixing the formulations with hot water (60°C) at the above ratio.

For the purpose of comparison, two rice-based commercial weaning foods were also evaluated for physical, and functional characteristics. Commercial weaning food A comprised of rice flour, milk solids, sugar, minerals and vitamins, whereas commercial weaning food B contained rice flour, sucrose, calcium carbonate, iron salts, and vitamins, as declared by the manufacturers on the packages.

Protein efficiency ratio (PER) was determined as per the method of Osborne et al (1919), and described by Pellet and Young (1980). 'Wistar' strain weanling male rats, 22 days old, and weighing between 37-40 g, were used for feeding standard, and test protein diets (Pellet and Young 1980). The number of rats used in case of each food was eight. All the diets contained (%) groundnut oil 10, sucrose 10, salt mixture 2 (Hubbel et al. 1937), vitamin mixture 2 (Chapman et al. 1959), test protein (at 10% level), and corn starch to make up the weight to 100 g. The results obtained were statistically analyzed, using Duncan's new multiple range test (Duncan 1955).

Results and Discussion

Proximate composition of the weaning foods is presented in Table 1. Both the weaning foods had almost similar composition, and were in conformity with the Indian standard specification for the weaning foods (ISI 1969). A minimum protein content of 14% has been prescribed for weaning

TABLE 1. PROXIMATE COMPOSITION OF WEANING FOODS*

	Formulated weaning foods		Commercial weaning foods**	
Constituent	1	2	A	B
		Composition		
Moisture, %	3.1 ± 0.40	3.9 ± 0.60	3.0 ± 0.30	-
Proteins, %	20.6 ± 1.20	21.8 ± 1.40	12.0 ± 0.90	6.0 ± 0.70
Fat, %	0.4 ± 0.12	0.4 ± 0.12	7.5 ± 0.90	0.6 ± 0.20
Ash, %	2.2 ± 0.43	2.1 ± 0.25	3.5 ± 0.30	0.7 ± 0.50
Crude fibre, %	1.0 ± 0.02	1.4 ± 0.03	1.8 ± 0.09	2.7 ± 0.01
Carbohydrates (by difference), %	73.3 ± 3.20	70.4 ± 4.50	72.2 ± 2.30	86.0 ± 4.10
Calories (KCal)	377	372	404	373
		Physico-chemical properties		
Bulk density, g/ml	0.75± 0.02	0.75± 0.03	0.43 ± 0.02	0.30 ± 0.04
Water holding capacity, ml/100 g				
at 27°C	300.0 ± 2.2	320.0 ± 1.2	200.0 ± 1.9	620.0 ± 2.6
at 60°C	340.0 ± 2.4	350.0 ± 1.9	250.0 ± 1.7	700.0 ± 2.5
Water absorption capacity, ml/100 g				
at 27°C	310.0 ± 1.7	310.0 ± 2.0	220.0 ± 1.4	640.0 ± 2.1
at 60°C	360.0 ± 2.2	360.0 ± 1.9	240.0 ± 1.6	660.0 ± 1.5
Consistency, pat spread, cm				
at 27°C	7.0 ± 0.6	7.0 ± 0.7	5.5 ± 0.3	3.0 ± 0.1
at 60°C	6.5 ± 0.4	5.5 ± 0.5	4.5 ± 0.4	3.0 ± 0.1

* values are average of three estimations ± standard deviation; ** As reported by the manufacturer.

foods by ISI (1969). Since the presently formulated foods are meant to be with high protein level, an attempt was made to bring up the protein content to 20-22%, by adding more of rice bran protein concentrate. The fat content of weaning foods was only 0.4%, since no additional fat was added to the formulations. These formulations were calorie dense, and contained 377 and 372 calories per 100 g food. For purpose of comparison, proximate composition of two commercial weaning foods, as given by the manufacturer, is also included in Table 1.

The data on bulk density, water absorption capacity, water holding capacity, consistency, and apparent viscosity of reconstituted blends are presented in Table 1 and Fig. 1. The bulk densities of both the weaning foods were high (0.75 g/ml), as compared to those of the commercial weaning blends A and B, whose bulk densities were 0.43 and 0.30 g/ml, respectively.

The water absorption capacities of weaning blends at 27°C was 310 ml/100 g, whereas commercial weaning blends exhibited lower (weaning food A), and higher (weaning food B) water absorption capacities. Since weaning blends are generally reconstituted in warm water, it is interesting to note the effect of temperature on water absorption capacity. At 60°C, the water absorption capacities of the formulated, and commercial weaning blends increased by 50 and 20 ml/100 g, respectively. It was obviously due to better gelatinization behaviour, and swelling capacity of starch granules at higher temperature (Lund and Lorenz 1990).

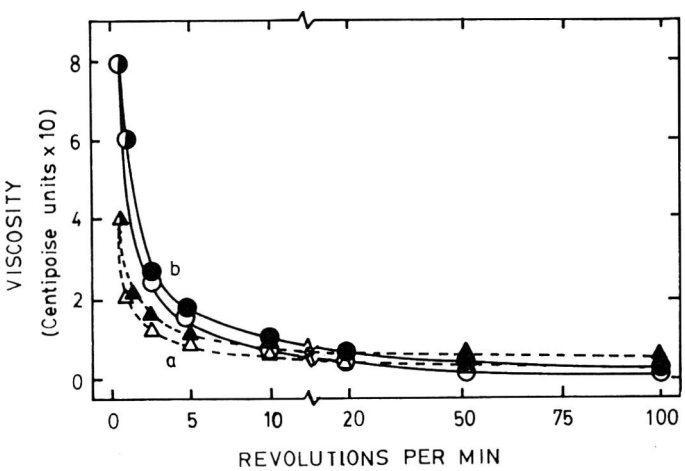

Fig. 1. Effect of temperature on apparent viscosity (100 rpm) of reconstituted weaning foods with solids : water ratio of 1:5.
a. Weaning food 1 (Δ) : 27°C, (O) : 60°C.
b. Weaning food 2 (▲) : 27°C, (●) : 60°C.

A similar profile was observed in case of water holding capacities of weaning foods. Water holding capacities of weaning foods 1 and 2 at 27°C were 300 and 320 ml/100 g sample, while the values were 340 and 350 ml/100 g sample, respectively, at 60°C.

Weaning blends 1 and 2 had pat spreads of 7.0 cm at room temperature, which decreased to 6.5, and 5.5 at 60°C, respectively (Table 1). Commercial weaning foods had lower pat spreads. Since low pat spread is related to increased viscosity (Bookwalter et al. 1968), it would lower the food consumption of child, because of its bulkiness (Ljungquist et al. 1981). Hence, a higher pat spread or low viscosity is favoured in weaning blends to increase the food intake. Considerable pre-cooking, and gelatinization of starch takes place during roller drying of weaning foods, which, in turn, affects the viscosity of reconstituted blends. Gelatinization of starch is an important functional characteristic for the formation of smooth porridge (Chandrasekhara and Ramanatham 1983). Since starch has a large water binding capacity that causes high swelling and viscosity (Lund and Lorenz 1990), it is considered as an important factor in limiting the food intake of infants, and small children (Ljungquist et al. 1981). If cereal-based weaning foods are subjected to certain processing conditions, like enzyme treatment, pre-cooking or extrusion cooking, their dietary bulk is reduced, and nutrient density is increased considerably (Malleshi 1984).

As is evident from Fig. 1, the viscosity of weaning blend 2 was higher than that of weaning blend 1. This could be attributed to the presence of greengram flour in weaning blend 2. Both the blends exhibited a slightly higher viscosities, when reconstituted in warm water (60°C) at a solid to water ratio of 1:5. These results are in conformity with the other functional characteristics of these weaning foods, like water absorption capacity, water holding capacity and pat spread, wherein an increase in temperature resulted in higher water uptake, and thickness of blends.

Data on apparent viscosities of reconstituted weaning blends, as a function of solid water ratio, along with comparison with commercial preparations are given in Fig.2. As the amount of water added to the blend increased, the gruel viscosity of blends decreased. However, there was a definite increase in viscosity of blends at higher temperature at both the ratios tested, in case of all the samples. Since

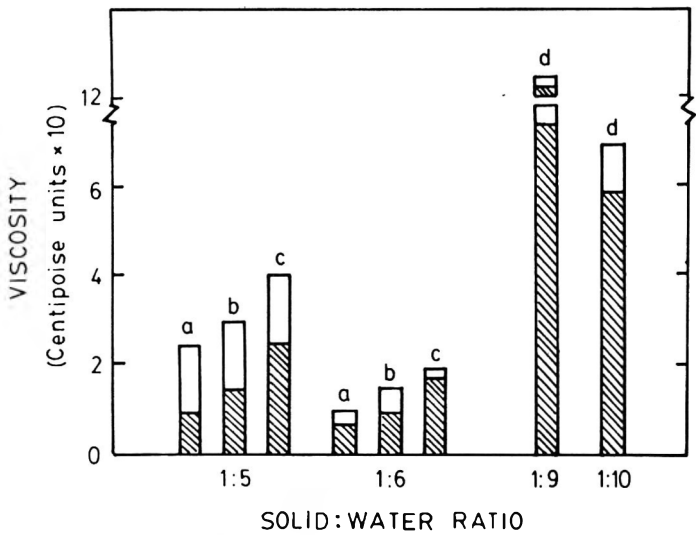

Fig 2. Effect of solids : water ratio on apparent viscosity (100 rpm) of formulated weaning foods in comparison with commercial weaning food preparations at A :27°C ☐ and B: 60°C ▨.
a. Weaning food 1, b. Weaning food 2, c. Commercial preparation A, d. Commercial preparation B.

the commercial preparation B had high water uptake, the viscosity of reconstituted material could be measured only at a solid water ratio of 1:9 and 1:10. Such observations have been reported earlier. For example, Malleshi (1984) studied the characteristics of some proprietary weaning foods, which were roller dried, and found that these exhibited significant differences in cold, and cooked paste viscosities. This could be due to different processing conditions, variation in the composition, and extent of gelatinization of starch.

The protein efficiency ratio values for weaning mixes 1 and 2 were 2.09 and 2.20, respectively (Table 2). Casein, under similar conditions, gave a protein efficiency ratio of 2.5. A slightly higher protein efficiency ratio of weaning blend 2 could be due to a better amino acid profile, since the formulation contained greengram flour, along with rice flour, and rice bran protein concentrate. However, both the formulations were not significantly different in their protein quality, as judged by Duncan's new multiple range test.

TABLE 2. PROTEIN EFFICIENCY RATIO OF WEANING FOODS

Diet	Average initial weight, g	Average protein intake, g	Average gain in weight, g	Protein efficiency ratio	
				Experimental	Adjusted
Casein	37.3	13.7	35.3	2.57[a]	2.5
Weaning food 1	37.3	13.0	27.3	2.09[b]	2.04
Weaning food 2	37.3	16.4	36.1	2.20[b]	2.15
SE_m				±0.09 (14 df)	

Randomized block design, 3 groups of eight male rats each. Means of the same column followed by different letters differ significantly according to Duncan's new multiple range test.

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An Alkali-reaction Test to Distinguish Open-Steam, Pressure-Steam and Dry-Heat Parboiled Rice

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All types of parboiled rice got degraded when immersed in very dilute alkali, but the appearance of the degraded grain differed among the three classes of parboiled rice. Open-steam parboiled rice grains remained fairly chalky white; pressure-steam parboiled grains became transparent; and dry-heat (sand heating) parboiled grains remained chalky, but got longitudinally split. Alkali reaction could thus distinguish between the three classes of product. The reaction seemed independent of starch reassociation.

Keywords : Parboiled rice, Alkali test, Kernel reaction pattern, Distinguishing type/extent of processing, Starch reassociation.

Ali and Bhattacharya (1972) showed that alkali reaction was a sensitive qualitative test to distinguish raw from parboiled rice as well as of the severity of parboiling. When rice grains were immersed in such dilute alkali solution, in which the raw rice grains remained unaffected, parboiled grains were invariably degraded, the extent increasing with increasing severity of heat treatment during processing. A study was conducted with a wide range of variously parboiled rice, with the aim to see whether the pattern of kernel reaction differed with the type, and extent of processing.

'Intan' variety of paddy (intermediate amylose content and gelatinization temperature) was steam- as well dry-heat parboiled under a wide range of processing conditions, air-dried and milled (Mahanta and Bhattacharya 1989). A brief description of the samples is given in Table 1. For the alkali-reaction test, 10 whole milled grains were immersed in 20 ml of 0.9% KOH in a 7-cm diam petridish, placed on a black polythylene sheet, and left covered overnight (Ali and Bhattacharya 1972).

A few typical results are shown in Fig.1. The raw rice grains, as observed earlier (Ali and Bhattacharya 1972), remained unaffected by the very dilute alkali solution used. But, all the 33 parboiled rice samples were attacked by alkali, the extent of degradation increasing with increasing severity of parboiling conditions. The usefulness of this reaction, as a sensitive qualitative test of parboiled rice (Bhattacharya 1979), was thus confirmed. However, the test could give only a rough estimate of the degree of parboiling treatment, as quantitative estimation of the relative degradation of the samples was difficult. But there was a distinct difference in the pattern of kernel reaction, according to the class of parboiled rice.

In parboiled rice produced by steaming under atmospheric pressure (viz., 30-0-10, 30-0-20, 30-0-60), the degraded kernels invariably remained

TABLE 1. DESCRIPTION OF EXPERIMENTAL PARBOILED RICE SAMPLES [a]

Rice code[b]	Approx. chalky centre area (%)
Steam parboiling series	
12-1-10[b]	95
12-1-20	90
12-2-10	80
12-2-20	50
12-3-10	25
12-3-20	0
17-1-10[c]	40
17-1-20	10
17-2-10	5
17-2-20	0
17-3-10	0
17-3-20	0

[a] There were 33 samples in all. Samples other than those listed above were; (a) in steam series: 22-1-10, 22-1-20, 22-2-10, 22-2-20, 22-3-10, 22-3-20, 30-0-10, 30-0-20, 30-0-60, 30-1-10, 30-1-20, 30-2-10, 30-2-20, 30-3-10, and 30-3-20 and (b) in dry-heat parboiling series[c]: 200-2.00-20, 200-4.00-16, 250-1.00-20, 250-2.00-16, 275-0.75-20 and 275-1.50-16. None of these had any chalky centre.

[b] For steam-parboiling series: First two digits indicate approximate initial paddy moisture (% w.b); the next digit indicates steam pressure (kg/cm^2); the last two digits indicate steaming time (min).

For dry-heat parboiling series: First three digits indicate sand temperature (°C); the next three digits indicate sand heating time (min); the last two digits indicate approximate final paddy moisture (% w.b.). Initial paddy moisture before sand heating. 30% (w.b). 12% moisture: original paddy:30% moisture: paddy soaked to saturation.

[c] The 12% and 17% moisture steam series and the dry-heat series, being low-moisture parboiled rice, were also moistened and tempered to promote starch reassociation (Ali and Bhattacharya 1976).

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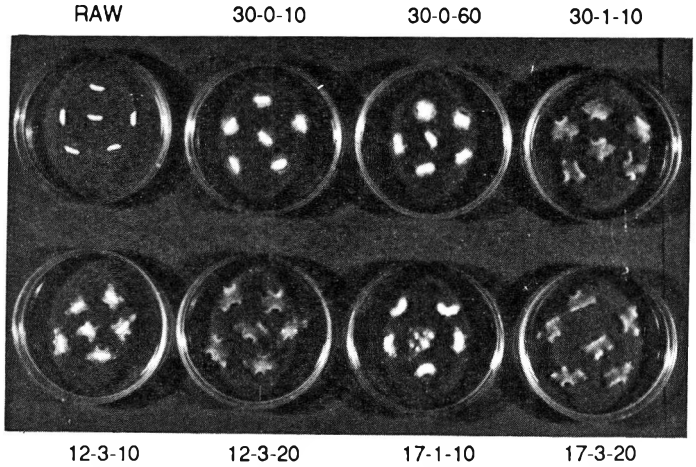

Fig. 1. Degradation of raw and steam-parboiled rice in 0.9% KOH. Explanation of code numbers is as given in Table 1.

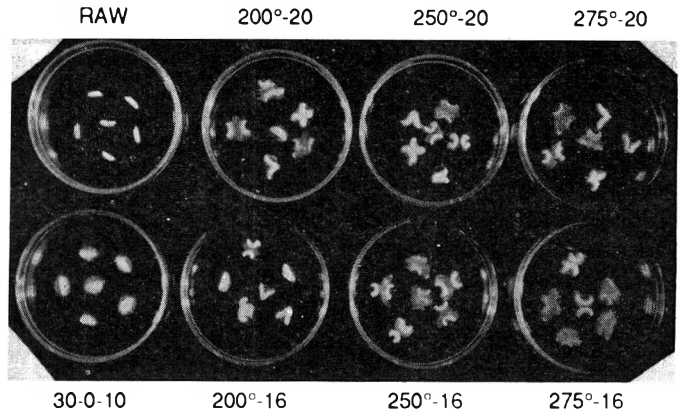

Fig. 2. Degradation of raw and dry-heat parboiled rice in 0.9% KOH. Explanation of code numbers is as given in Table 1. The 30-0-10 steam-parboiled rice was included for comparison.

chalky white, with a faint collar all around after 24 h (Fig.1). Similarly, all low-moisture paddy, pressure-parboiled by such steam pressure that left a large chalky centre in the grain (e.g., 12-1-10, 12-2-10, 17-1-10), also showed a chalky white grain mass after alkali treatment, with a faint collar around. Consequently, these samples were indistinguishable from fully soaked open-steamed samples. Low-moisture pressure-parboiled paddy, having only a small chalky centre (e.g., 17-1-20, 17-2-20), showed only a small area of opacity at the centre and a surrounding transparent mass, with no collar around.

On the other hand, all the 17 fully translucent pressure-steam parboiled rice with no chalky centre in the kernel, regardless of original paddy moisture or steaming condition employed for parboiling (viz., 12-3-20, 17-2-20 to 17-3-20, 22-1-10 to 22-3-20, 30-1-10 to 30-3-20), became fully transparent after overnight soaking in alkali (Fig. 1). The kernels also showed slight longitudinal splitting, but the two split halves did not disengage from each other.

Dry-heat parboiled paddy showed a different pattern. The extent of degradation here too increased slightly, and the opaque centre decreased in size with increasing time and temperature of sand roasting. But no sample became fully transparent. However, all the samples showed complete longitudinal splitting, with the two split halves moving appreciably apart from each other (Fig. 2).

Clearly, the alkali test, in addition to being used as an excellent qualitative test to distinghish raw from parboiled rice, and as a rough test of the degree of parboiling (Ali and Bhattacharya 1972; Bhattacharya 1979), can also be used as a test of the class of parboiled rice. It can confidently distinguish (a) fully translucent pressure-steam from open-steam parboiled rice, and (b) dry-heat from steam-heat parboiled rice. The distinguishing kernel splitting of dry-heat parboiled rice was confirmed with three other varieties ('IR8', 'IR20', 'Madhu') to show its general validity. The test can also identify pressure-steam parboiled rice having chalky centres. Even though the centre remained opaque in this case also, the surrounding grain mass, unlike in open-steam parboiled rice, became fully transparent.

Pillaiyar et al (1988) had, in a preliminary report, suggested a test to distinguish pressure-parboiled rice. A few rice grains were boiled in 5% KOH solution for 4 min. Pressure-parboiled rice grains became translucent (those with chalky centres gave translucent periphery). But rice parboiled at pressures of 0.352 kg/cm^2 or less remained opaque.

Whether or not starch reassociation in parboiled rice had any effect on the alkali reaction was tested. All low-moisture pressure-steam and dry-heat (sand heating) parboiled rice (Table 1) were moistened to 25-30% moisture to promote starch reassociation (Ali and Bhattacharya 1976), then dried back. These samples were then treated by alkali. The results seemed to remain unaffected by the moisture treatment. This suggested that alkali reaction was largely independent of starch reassociation.

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Fatty Acid Profile of Some Indian Spices

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Total fat content in seventeen spices ranged from 3.2% (cardamom) to 35.7% (nutmeg). Qualitative differences were observed in the fatty acid composition. Total saturated fatty acids ranged from 3.7-93.5%. Monounsaturated fatty acids varied from 3.8-77.8%, and polyunsaturated fatty acids ranged from 2.8-59.4%. Palmitic, oleic, linoleic and linolenic acids were major fatty acids in most of the spices, except in nutmeg, which contained 74.9% myristic acid.

Keywords : Indian spices, Lipid, Fatty acids, Monounsaturated fatty acids, Polyunsaturated fatty acids, Saturated fatty acids.

Spices and condiments, though consumed in small amounts, have distinct physiological, biochemical, and pharmacological effects (Pruthi 1976, 1980). The chemical composition, including proximate, chemical constituents, essential oils, and oleoresins have been reported (Pruthi 1980; Rao et al. 1980; Guenthen et al. 1975; Varo and Heinz 1970), and also the microbial quality of some selected spices has been studied (Kaul and Taneja 1989). Besides, bishopweed, black pepper, coriander and cumin have been surveyed for their fungal contaminants (Amita Shrivastava and Jain 1992). Little information is available on lipid and fatty acid composition of Indian spices (Hemavathy and Prabhakar 1988, 1989). In the present investigation the lipid, and fatty acid composition of 17 Indian spices are reported.

Samples of different spices, cardamom 'Mysore' and 'Malabar' (*Elettaria cardamomum*), coriander "PM', "FM', 'M' (*Coriandrum sativum*), cumin '1', 'M. C43' (*Cuminum cyminum*), fennel 'PM-PF 35', 'FM', 'PM' (*Foeniculum vulgare*), fenugreek (*Trigonella foenumgraecum*), chillies (*Capsicum annum*), bayleaves (*Laurus nobilis*), nutmeg (*Myristica fragrans*), mace (*Myristica fragrans*), Kasturi (*Curcuma zedoria*), and turmeric (*Curcuma longa*) were obtained through Spice Board, Cochin, Kerala or purchased from local markets. All samples were cleaned, kept overnight in an oven at 50°C, and ground to a fine powder in a analytical grinder (Cyclotec, Model No. 1093, Sweden).

Lipid extraction : Total lipids from 3-5 g samples of spice were extracted according to the procedure of Hubbard et al (1977), and purified following the procedure of Folsch et al (1957). The lipid extract was concentrated in a rotary evaporator (Evapotec, Model No. 668, W. Germany) at 40°C, and dried in a desiccator over concentrated sulphuric acid for 18 h. Samples were dried in an oven at 110°C to constant weight, in order to remove volatile lipid (AOAC, 1984), and total lipid was determined gravimetrically.

Fatty acid analysis : A weighed amount (20-30 mg) of spice lipid was evaporated to dryness at 40°C, in an atmosphere of nitrogen, and fatty acid methyl esters were prepared according to the procedure of Morrison and Smith (1964). The fatty acid profiles of methyl esters were determined using varian 3700 gas liquid chromatograph with flame ionization detector, coupled with integrator as described earlier (Ghafoornissa 1989). The column was packed with 10% Silar 10C, coated on chromosorb W80-100 AW.

Chromatographic conditions: The initial column temperature was 160°C, then programmed to 225°C at the rate of 3°C/min, and maintained for 20 min. The temperatures of the injector, and detector were 230 and 250°C, respectively. The flow rate of nitrogen was 25 ml/min. The individual fatty acid peaks were identified by comparing the retention times of standard fatty acids (Nu-chek prep, Elysian, MN). All determinations were performed in duplicate, and means are reported.

Total lipids: The results (Table 1) indicated that the lipid contents varied widely in different spices from 3.2% (cardamom) to 35.7% (nutmeg). It was of interest to note that mace, an aril of myristica family was as rich in lipid content as nutmeg, the kernel of myristica family. Among the seed spices, fennel and cumin were rich in lipids (20-23%). The lipid contents in three varieties of coriander ranged from 3.6-7.9%, in turmeric about 5.9%, while in chillies (27.4%).

Fatty acid composition: Palmitic, oleic, linoleic and linolenic acids were predominant in most of the spices, except in nutmeg which contained 74.9% myristic acid (Table 1). Highest amount of saturated fatty acids was found in nutmeg (93.4%). These values are similar to the reported values

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TABLE 1: FATTY ACID COMPOSITION OF SOME INDIAN SPICES

	Lipid	Fatty acids, % of total fatty acids								
	%	C 14:0	C 14:1	C 16:0	C 16:1	C 18:0	C 18:1	C 18.2	C 18:3	C 20:0
Cardamom 'Mysore'	2.9	0.5	-	21.2	-	2.9	44.1	21.4	7.8	2.1
Cardamom 'Malabar'	3.4	0.4	-	20.8	-	3.1	43.1	22.1	7.8	2.7
Coriander- 'PM'	6.7	-	-	3.8	-	0.8	74.9	17.8	2.7	-
Coriander -'FM'	3.6	-	0.1	4.0	-	0.9	77.5	16.0	1.5	-
Coriander- '2 M'	7.9	-	-	3.6	-	0.1	76.7	18.0	1.6	-
Cumin - '1'	22.9	-	-	4.0	-	1.9	66.3	25.4	2.5	-
Cumin 'MC. 43'	22.7	-	0.1	5.4	-	1.7	57.2	33.2	2.4	-
Funnel 'PM, 'PF-35'	21.4	0.2	-	4.8	-	1.0	77.3	15.1	1.6	-
Fennel 'FM'	23.0	0.2	-	5.0	-	1.0	75.5	16.5	1.7	-
Fennel 'PM'	20.0	0.1	-	4.8	-	0.1	77.8	15.3	1.8	-
Fenugreek	6.9	0.7	0.5	10.8	-	4.3	22.7	37.6	21.8	1.0
Chillies, dry	27.4	4.2	-	16.4	-	4.1	18.5	54.8	2.0	-
Nutmeg	35.7	74.9	-	9.7	-	6.9	3.8	1.8	1.0	2.0
Mace	30.4	0.3	-	31.3	-	2.9	40.3	19.8	2.9	1.8
Bay leaves	4.0	9.2	0.3	8.5	9.2	24.8	26.2	10.6	6.7	4.5
Kasturi	5.5	4.0	1.8	15.0	3.9	10.5	12.0	42.4	7.7	2.7
Turmeric	6.4	5.2	2.3	16.8	2.4	8.7	14.2	46.0	3.7	0.8

(Hollan et al. 1991). Palmitic and stearic acid contents ranged from 3.6 to 31.3% and 0.1-24.8%, respectively. Arachidonic acid was present in low quantities (0.8 to 4.5%).

The major monounsaturated fatty acid in the lipids of all spices was oleic acid (ranged from 3.8-77.5%). The highest amount was found in fennel, coriander, and cumin. The high values of 18:1 could be attributed to isomers of 18.:1 like petroselinic, and oleic acids as reported in literature (Pruthi 1980; Hemavathy and Prabhakar 1988). Fenugreek seeds showed lower amounts of oleic acid as reported by earlier workers (Hemavathy and Prabhakar 1989). Bay leaves contained good amounts of palmitoleic acid. Fatty acids like myristoleic and plamitoleic acids were found in trace amounts in turmeric varieties.

The total polyunsaturated fatty acids ranged from 2.8 to 59.4%. Linoleic acid is the major fatty acid in most of the spices. The levels in chillie, cumin, coriander and turmeric were found to be slightly higher than those reported by earlier workers (Ghafoorunissa 1989). Linolenic acid content ranged from 1-21.8%, highest being in fenugreek seeds.

There are several reports in literature, highlighting the importance of turmeric, fenugreek and cardamom on cholesterol metabolism and gastric disorders (Sharma 1984; Subba Rao et al. 1970). Essential oils from ginger, cumin, *ajowain*, coriander, basil clove and mustard showed various degrees of inhibition against microbial contamination (Meena and Vijay Sethi 1994). These observations on the fatty acid profile of some of these spices along with dietary fibre components need further studies.

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Glutamic Acid Fermentation Using *Brevibacterium* DSM 20411

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For production of glutamic acid by *Brevibacterium* DSM 20411, a number of growth media were evaluated to find out a suitable production medium for glutamic acid. Under optimized conditions with glucose as carbon source at pH 7.2 and 30°C incubation, the culture produced 1.13 mg glutamic acid/ml medium after 48 h fermentation in shake flasks. Change in ratio of fermentation medium and flask volume affected glutamic acid production.

Keywords : Glutamic acid, *Brevibacterium* DSM 20411, Ninhydrin colour reaction, Fermentation, Optimization.

Rapid increase in the demand for monosodium glutamate, as a flavour enhancing agent both for food industry, and the table use has been witnessed in recent years (Kinoshita 1963). Many L-amino acids are currently produced from cheaper carbon, and nitrogen sources by fermentation with different bacterial strains such as *Corynebacterium glutamicum, Brevibacterium lactofermentum, Serratia marcescens, Escherichia coli, and Bacillus subtilis* (Nakayama 1982). Earlier, a study has been made on the mineral requirements of a strain of *Pseudomonas aeruginosa* for the production of glutamic acid in a synthetic medium (Goswami and Majumdar 1971). Sub-sequent research resulted in an economical fermentation process for the production of L-glutamic acid (Hirose et al. 1985). Glutamate fermentation is well known as a typical type of aerobic fermentation, and still the batch/ fed batch process is the most popular fermentation method for production of glutamate (Richard Joseph and Ramachandra Rao 1973; Ishizaki et al. 1993). Much efforts have been paid to enhance the glutamate fermentation process by modifying the medium, use of newer as well as highly potent cultures, and also utilization of new raw materials (Minoda 1986). The extracellular glutamate accumulation is known to be accompained by cellular permeability (Shiio et al. 1962). In fact, the establishment of L-glutamic acid fermentation provided a significant impetus to the development of microbial production of primary metabolites (Hirose et al. 1985). The present work has been undertaken by us to develop an efficient indigenous technology. Results on production of L-glutamic acid by *Brevibacterium* DSM 20411 are reported in this communication.

Microorganism: A strain of *Brevibacterium* sp, obtained from Deutsche Sammlung Von Mikroorganismen und Zellculturen GmbH (Ajinomoto Co. Inc.) was used. Culture was maintained on agar slants containing (g/l) casein peptone 10, yeast extract 5, glucose 5, sodium chloride 5, agar powder, 15 and distilled water 1000 ml (pH 7.2-7.4). Slants were preserved at 4°C, and subcultured twice a month.

Inoculum and fermentation: Fermentation was carried out by taking 20 ml of the above medium, but without agar in 250 ml Erlenmeyer flask. After inoculating (20 h old culture, 5% v/v inoculum size), the flasks were incubated at 30±1°C for a stipulated period on a rotary shaker (MB-Orbit Environ Shaker, SK-1009R, MB Instruments, Bombay- India) at 180 rpm. Samples were withdrawn as whole flask at desired time intervals for analysis. The results are reported as the average of four sets of experiments.

The bacterial growth was determined by measuring absorbance at 610 nm in spectrophotometer (UV-160A, Shimadzu, Japan). Soluble sugars were analyzed by dinitro salicylic acid method (Miller 1959). Thin layer chromatography (Silca gel G, solvent mixture: n-butanol/glacial acetic acid/water 4:1:1, v/v) was used for the qualitative determination of glutamic acid (Brenner and Nieser 1967). Quantitatively, it was estimated by ninhydrin colour reaction by measuring the absorbance at 570 nm (Spies 1957).

Selection of production medium: The general pattern of production medium for glutamic acid was obtained from literature (Kinoshita 1963; Hirose et al. 1985; Nakayama 1982), but were modified in the present studies. Medium M5 contained 2 g glucose, 0.5 g $NaNO_3$, 0.12 g KH_2PO_4, 1 ml mineral solution ($FeSO_4.7H_2O$, $MnSO_4.4H_2O$, $MgSO_4.4H_2O$, $ZnSO_4.6H_2O$,NaCl each 1 mg), 100 µl thiamine hydrochloride, 100 µl corn steep liquor and 100 µl Tween 80 in 100 ml distilled water (pH 7.2) was used.

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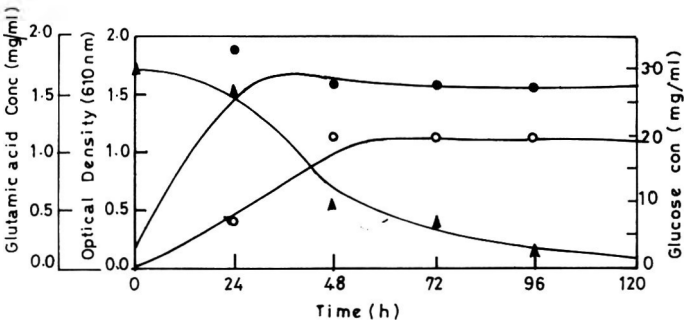

Fig. 1 Cell growth, glutamic acid production and glucose consumption by *Brevibacterim* DSM 20411 (120 h)
● Optical density (610 mm), ○ Concentration of glutamic acid (mg/ml), ▲ Glucose concentration (mg/ml)

Fig.1 shows the cell growth pattern and glutamic acid production by *Brevibacterium* sp. over a period of 120 h in M5 medium. The culture attained the maximum growth after 24 h, and the maximum glutamic acid production (1.13 mg/ml) was obtained after 48 h.

Studies on glucose consumption pattern in the medium (M5) by the culture revealed that about 70% of glucose was consumed within 48 h fermentation (Fig. 1). Efficiency of the strain to utilize the substrate (glucose), and its conversion to the desired amino acid is an important factor in fermentative production of amino acids (Amin et al. 1993).

Effect of pH: The pH of M5 was set at different initial values (pH 4, 5, 6, 7, 8, 9, 10 and 11), and the results are shown in Fig.2. Data reveal that optimum pH values for glutamic acid production are between 7.0 and 8.0.

Effect of temperature and agitation: Fermentation was carried out by taking 20 ml of M5 medium in 250 ml Erlenmeyer flasks. For studies on effect of temperature, after inoculation, the flasks were incubated at four different temperatures (20, 25, 30 and 35°C) for a stipulated period on a rotary shaker agitated at 180 rpm. For studies on effect of agitation, fermentation was carried out at five different agitation speeds (60, 120, 180, 240 and 300 rpm). The results are shown in Table 1. Data showed that optimum temperature, and agitation for glutamic acid production were 30°C and 180 rpm, respectively. Under conditions of insufficient oxygen, the production of glutamic acid is poor, and large amounts of lactic, and succinic acids accumulate, while excess oxygen increases the amount of α-ketoglutaric acid (Amin et al. 1993). It was found that both over-abundant, and meagre aeration are undesirable, the former being inhibitory to cell growth, and the latter to glutamic acid production (Hirose et al. 1966).

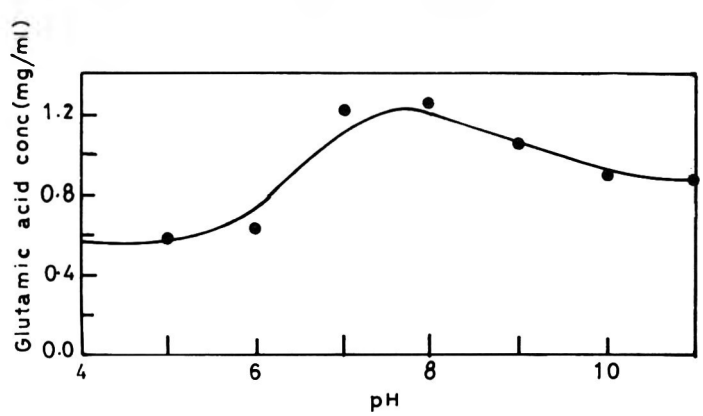

Fig. 2 Effect of pH on glutamic acid production by *Brevibacterium* DSM 20411 (48 h)

TABLE 1. EFFECT OF TEMPERATURE AND AGITATION ON GLUTAMIC ACID FERMENTATION BY *BREVIBACTERIUM* SP.

Temperature, °C	Glutamic acid, mg/ml	Agitation, rpm	Glutamic acid, mg/ml
20	0.51	60	0.40
25	0.86	120	0.71
30	1.13	180	1.13
35	1.02	240	1.05
		300	0.76

Effect of ratio of volume of the medium and flask: Two different types of sets were prepared. In one set, the volume of the medium was kept constant at 20 ml with variation in the volume of flask from 100 ml to 500 ml. In another set, the volumes of the medium were 50 and 100 ml in 250 and 500 ml flasks, respectively. It is interesting to note that glutamic acid production has increased due to changes in the ratio of volume of the medium to volume of the flask. The results are recorded in Table 2. In both cases, yields of glutamic acid were four to five fold higher than the control experiment. The balance of aerobic and anaerobic conditions is one of the important factors, which controls the yields of glutamate, as the main pathway of glutamate synthesis involves a combination of oxidative degradation of glucose and anaerobic citrate decomposition (Kinoshita 1963). It is also evident from Table 2 that it is not the ratio of the volume of the medium to flask volume is critical for the glutamic acid production, as different yields have been obtained with 1:5 ratio of volume of medium to flask volume. The data thus indicate that it is the working volume of the medium at specific medium volume to flask volume

TABLE 2. EFFECT OF RATIO OF VOLUME OF THE MEDIUM AND FLASK IN GLUTAMIC ACID PRODUCTION (48 h)

Volume of the flask, ml	Ratio of volume of medium:flask volume	Glutamic acid, production, mg/ml
500	1:25	1.168
250	1:12.5	1.175
150	1:7.5	1.315
100	1:5	3.042
500	1:5	6.868
250	1:5	5.775

ratio, which is important. Thus, the best yield was obtained, when 100 ml of the medium was taken in 500 ml flask, which resulted in production of 6.868 mg/ml glutamic acid.

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Escherichia coli in Milk, Meat and Meat Products : Isolation, Characterization, Antibiogram and Zoonotic Significance

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Seventy eight isolates of *Escherichia coli* isolated from a total of 350 samples, comprising milk, meat, and meat products, were found to be of 20 different serotypes. These isolates were grouped into various biotypes, ranging from A to U on the basis of five biochemical tests. The isolates exhibited, α, β and γ- haemolytic activities. Antibiogram revealed that majority of the isolates were sensitive to gentamicin, ampicillin, streptomycin, chloramphenicol, cotrimoxazole, bacitracin, kanamycin, but resistant to doxycycline, cloxacillin, carbenicillin, penicillin-G and polymyxin-B.

Keywords : *Escherichia coli*, Serotypes, Biotypes, Antibiogram, Haemolytic activity, Zoonotic significance.

Strains of *Escherichia coli* are known to be associated with a variety of pathological conditions (Sojka 1971). In animals, it causes colibacillosis, which is responsible for significant economic losses (Blood and Radostitis 1989) and in man, it is associated with acute gastroenteritis, particularly in infants, and older persons, urinary tract infections, and traveller's diarrhoea (Duguid et al. 1984). Various types of *E. coli* can be isolated from milk, meat, and their products (Singh et al. 1994). There may be great variations in biochemical properties within a species, and on the basis of this, the bacteria can be biotyped into several biotypes (Duguid et al. 1984), which may show some correlation with pathogenicity of the organism (Kulshrestha and Kumar 1977). Indiscriminate use of chemotherapeutic agents leads to drug resistance, which creates problems for the successful treatment of *E. coli* infections (Singh et al. 1994). Several studies have been carried out on the microbiological quality of raw meat (Bachhil and Ahluwalia 1973); occurrence of *Salmonella* in meats (Bachhil and Jaiswal 1988); also on market milks in Hisar (Gahlot et al. 1975), and milk and spray-dried skim milk powder used as ice cream ingredients (Anupam Arora and Sudarsanam 1986). Keeping in view the public health significance of this organism, the present investigation involved the incidence of *E. coli* in milk, meat, and meat products, its haemolytic activity, biochemical behaviour, serotyping, and biotyping patterns, and sensitivity to various chemotherapeutic agents.

In all, 350 samples of raw milk, meat, and meat products were collected under aseptic conditions from local market, and retail meat shops. These samples were subjected to analysis within 4 to 6 h of collection. In case of milk samples, 10 ml of aliquots were inoculated into tubes containing 10 ml double strength McConkey lactose broth, and incubated at 37°C for 24 h. After incubation, one loopful of culture broth was streaked onto MacConkey lactose agar, and eosin-methylene blue (Collee et al. 1990) agar obtained from Hi-Media Laboratories, Bombay for isolation of pure isolates (Varadaraj 1993). In case of meat samples, 25 g material was minced, and transferred into 250 ml double strength McConkey lactose broth, and incubated at 37°C for 24 h. The broth culture was streaked as above to get pure isolates.

Typical colonies resembling *E. coli* were picked up, and subjected to various morphological, cultural, and biochemical tests (Edwards and Ewing 1972; Collee et al. 1990). The suspected cultures showing typical morphological, and biochemical characteristics of *E. coli* were sent to National *Salmonella* and *Escherichia* Centre, Central Research Institute (CRI), Kasauli for serological typing and final confirmation. All the confirmed isolates were subjected to fermentation of sucrose, dulcitol, amino acid decarboxylation activity in L-lysine monohydrochloride, L-ornithine monohydrochloride, and L-arginine hydrochloride. For haemolysin production, all the isolates were streaked onto blood agar plates (Collee et al. 1990), containing 10% sheep blood (v/v), incubated at 37°C for 24 h, and examined for zone of clearance around the colonies. All the confirmed isolates were tested for antimicrobial drug sensitivity against 24 chemotherapeutic agents obtained from Span Diagnostics, Surat (India) as per modified Kirby Bauer disc diffusion method (Carter 1973).

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Out of 350 samples of milk, meat, and meat products examined, 78 (22.28%) yielded *E. coli* (Table 1). Incidence was found to be highest in chicken meat (60.87%), followed by meat products (47.5%), goat meat (21.31%), and pig meat (8.70%). Only 5 (8.33%) out of 60 milk samples were found to be contaminated with *E. coli*. The increased isolation of *E. coli* from various meat samples could be due to the fact that there are chances that gastro-intestinal tract might rupture during evisceration, resulting in contamination of wholesome meat.

Although, no relationship between the prevalence of *E. coli* and samples examined was observed, there were more samples positive for *E. coli* from meat products, which were procured locally than those manufactured by standard firms. Low incidence of *E. coli* in the latter could be due to higher salt concentration, heat treatments, and preservatives used in the manufacture of these foods.

Swines are not generally skinned during slaughter, and skin may contain the organisms, which remain sticking to it, and later gain entry in pork, and sausages prepared from such meat. The zoonotic importance of *E. coli* of porcine origin have also been reported (Lakshmanachar et al. 1985).

TABLE 1. ISOLATION AND SEROTYPING OF *ESCHERICHIA COLI* FROM MILK, MEAT AND MEAT PRODUCTS

Source	No. of samples examined	Samples positive for *E. coli*		
		No.	%	Serotypes
Milk	60	5	8.33	0:5,0:7,0:61(2) 0:106
Goat liver	61	13	21.31	0:3, 0:4 (2) 0:15,, 0:32 (2) 0:61 & untypable (6)
Goat kidney	53	9	16.98	0:4, 0:23,0:2(2) 0:61 & untypable (4)
Goat muscle	51	13	25.49	0:4,0:32,0:52 0:61, 0:62, 0:91 (3) & untypable (5)
Pig liver	23	2	8.70	0:9, 0:62
Pig muscle	39	3	7.69	0:9, 0:91, 0:164
Chicken meat	23	14	60.87	0:5, 0:62 (2) 0:158, 0:162 rough (3) & untypable (6)
Meat products (chicken sausages, pork sausages, *shahi kabab*, salami and meat pie)	40	19	47.50	0:3, 0:21, 0:32 (2) 0:60, 0:61, 0:157, 0:162 & untypable (11)

The incidence of *E. coli* in milk appears to be rather low, considering the unhygienic practices employed in milk production. There are chances that these organisms might have escaped detection owing to their small number.

The results of biochemical behaviour of 78 strains of *E. coli* revealed that 62 (79.49%) were positive for sucrose, 43 (55.13%) positive for dulcitol, and none produced hydrogen sulphide. All the isolates were positive for glucose, arabinose, lactose, indole, and methyl red (MR), while negative for urease, voges proskauer (VP), and citrate utilization tests. Out of 78 *E. coli*, 53 (67.95%) decarboxylated lysine, 60 (76.92%) arginine, and 48 (61.54%) ornithine. Such variations had been reported previously by various workers (Subbarao et al. 1975; Kulshrestha and Kumar, 1977; Sharma et al. 1992). Sharma et al (1992) and Lautrop et al (1971) have discussed that H_2S producing *E. coli* strains do appear as variants in nature.

The results of serotyping of various *E. coli* (Table 1) showed that out of 78 *E. coli* isolates, 43 which have been serotyped belonged to 20 different serotypes. The isolation of such a wide range of serotypes could be because of different sources of samples, which were procured from different retail meat shops, slaughter houses, and dairy farms. The serotype 0:32 was the most predominant, having been recovered from 7 samples, followed by 0:61 (6) and 0:62, 0:4 and 0:91 (4 each).

In the present study, all the *E. coli* strains were grouped into 20 biotypes (A to U), and the results are shown in Table 2. Majority of these belonged to A (15.38%), followed by C (11.54%), H (10.26%), B and E (6.41% each) in decreasing order (Table 2). These findings are in contrary to Samra and Sambyal (1982) and Sharma et al (1992). These variations might result owing to the different sources of isolates by various workers. Out of 78 isolates, 31 (39.74%) produced alpha, 11 (14.10%) beta, and 36 (46.15%) gamma type of haemolysin. These findings almost corraborate with findings reported earlier (Sharma et al. 1992).

The antibiogram studies of 78 isolates of *E. coli* revealed that none of the antimicrobial drug was effective against all the isolates tested. Gentamicin inhibited the growth of 97.43% of the isolates, followed by chloramphenicol (93.59%), co-trimoxazole (83.33%), ampicillin (78.20%), streptomycin (71.79%), and bacitracin (70.51%). None of the isolates was found sensitive to cloxacillin, carbenicillin, and doxycyline. The resistance towards

TABLE 2. BIOTYPING OF 78 ISOLATES OF *E. COLI*

Biotype	No. of isolates	Tests				
		S	D	LLM	LAH	LOM
A	12	+	+	+	+	+
B	5	+	+	+	+	-
C	9	+	-	+	+	+
D	4	+	+	+	-	+
E	5	+	+	-	+	+
F	4	+	-	+	-	+
G	5	+	+	-	+	-
H	8	+	-	+	+	-
I	3	-	-	+	+	+
J	4	+	-	-	+	+
K	2	-	+	+	+	-
L	2	+	+	-	-	+
M	2	-	+	-	+	+
N	1	+	+	+	-	-
O	3	+	+	-	-	-
P	3	+	-	+	-	-
R	3	+	-	-	+	-
S	1	-	-	-	+	+
T	1	-	+	-	-	+
U	1	-	-	-	-	-

S = Sucrose fermentation : 62 +, 16 -

D = Dulcitol fermentation : 35 -, 43 +

L-LM = Lysine monohydrochloride : 25 -, 53 +

L-AH = L - arginine hydrochloride : 18 -, 60 +

L-OM = Ornithine monohydrochloride : 30 -, 48 +

antibiotics by the isolates was : cephalexin, polymyxin-B, oxytetracycline, and penicillin G (98.72% each), chlortetracycline (96.15%) triple sulfa (93.59%), tetracycline (83.33%), erythromycin (90.77%), and cephaloridine (74.36%). The observed resistance pattern can be due to the indiscriminate use of these drugs in animals which encourages the incidence of drug resistance in enterotoxigenic strains of *E. coli* (Singh et al. 1994).

Some of the serotypes encountered in the present study are important from the public health point of view. Although no enterotoxigenicity or other studies were conducted, one study has documented that 0:15 is enterotoxigenic, and 0:164 is enteroinvasive, while 0:157 is enterohaemorrhagic (Wilson et al. 1984). The O-groups 4, 7 and 9 are associated with parentral septic infections particularly of urinary tract, and the 0:164 causes dysenteric type of diarrhoea in man. Serogroup 0:157 is considered to be responsible for haemorrhagic colitis with symptoms of passage of blood stained stools in children due to its ability to produce cytotoxin. Similarly, serotype 0:158 is also encountered in outbreaks of gastroenteritis in infants (Wilson et al. 1984). Out of 11 strains, which are of public health significance, 7 (63.64%) were α-haemolytic and 5, 4 and 2 belonged to biotypes A, C and H, thereby indicating that these biotypes are most pathogenic as reported earlier (Sharma et al. 1992).

Generally, meat is fully cooked before its consumption, but the use of milk, and meat products may prove hazardous for human health, as they are consumed either as such or half cooked. The possible ways of entry of various *E. coli* serotypes could be handling of milk, meat and meat products by adopting improper hygienic measures during handling, and processing. Therefore, careful processing of the milk, meat and meat products is an important public health measure. Proper hygienic measures need to be adopted for handling various food products in order to bring down the entry of these organisms in these products, thereby safeguarding the human beings from health hazards.

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Effect of Processing Differences in Carbohydrates of Cereal-legume Blends on Blood Glucose Responses in an Individual with Impaired Glucose Tolerance

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Among cereal-legume blends of malted, popped and roller dried wheat, chickpea, greengram and moth bean, the malted cereal-legume blend showed the highest glycemic response in a normal as well as in an individual with impaired glucose tolerance, as compared to popped and roller dried cereal-legume blends.

Keywords : Glycemic index, Malting, Popping, Roller drying, Cereal-legume blends.

In the beginning of 1980's, it was becoming increasingly clear that different starchy foods are digested at different rates, affecting ultimately the glycemic response (O'Dea et al. 1981). Food processing in particular can cause modifications of starch (Brand et al. 1985), and alter its physico-chemical properties such as hydration of granules, chemical nature (Snow and O'Dea 1981), and consequently can alter carbohydrate digestibility, and glycemic response (Ross et al. 1987). Since it is difficult to predict the glycemic response of starchy foods from their chemical composition due to wide variation in the digestibility of starch, Jenkins et al (1981) introduced the concept of glycemic index of foods. The degree of post-prandial response of various starchy foods depends on the degree of glucose tolerance of the individual subjects. Consequently, the most glucose tolerant subjects show the least differences in post-prandial responses, whereas the least glucose tolerant normal subjects exhibit the greatest differences (Crapo et al. 1980). Differences in post-prandial responses may, therefore, be exaggerated in subjects with impaired glucose tolerance. In this communication, the effects of malted, popped and roller dried cereal-legume based foods on post-prandial blood glucose responses in an individual with impaired glucose tolerance are reported. The study is limited to only one subject as the condition of impaired glucose tolerance is rare.

Wheat (*Triticum aestivum*), greengram (*Phaseolus aureus Roxb*), chickpea (*Cicer arietinum*), and moth bean (*Phaseolus aconitifolius*) were from local market. Malted, and popped flours from wheat, and legumes were prepared as described by Livingstone et al (1993). Malted wheat flour, and toasted greengram, chickpea, moth bean and skim milk powder were dry mixed. Popped wheat was blended with popped legumes to prepare popped food. Debranned wheat, and decorticated split legumes were toasted at 70°C, and their blend was roller dried as per Malleshi et al (1989). The blends contained (%) : wheat 75, greengram 10, chickpea 5, moth bean 5, and skimmed milk powder 5.

The glycemic index studies were conducted on a 59 year old male subject, who satisfied the criteria of impaired glucose tolerance (National Diabetes Data group 1979). Oral glucose tolerance tests were carried out, after an overnight fast, by administering 50 g glucose. *Chapatis* (unleavened flat bread) prepared from 50 g equivalent of carbohydrates of malted, popped, and roller dried blends were smeared with sugar-free jam, and fed to the subject on two separate occasions, after an overnight fast. Finger prick blood was collected at 30 min intervals over a period of 2 h and analyzed for glucose content by glucose oxidase method (Dahlqvist 1964). Simultaneously, the study was also carried out in a 43 year old normal subject on similar lines for comparison. The glycemic index (GI) of foods was calculated as follows :

$$GI = \frac{\text{Area under the curve for test food}}{\text{Area under the curve for glucose}} \times 100$$

The mean blood glucose response of the subject with impaired glucose tolerance as compared to the normal subject for the malted, popped, and roller dried foods at different time intervals is depicted in Fig. 1. At all time intervals, the blood glucose levels from any of the three blends were markedly lower, as compared to the orally administered glucose. The blood glucose response in the impaired glucose tolerant subject was much

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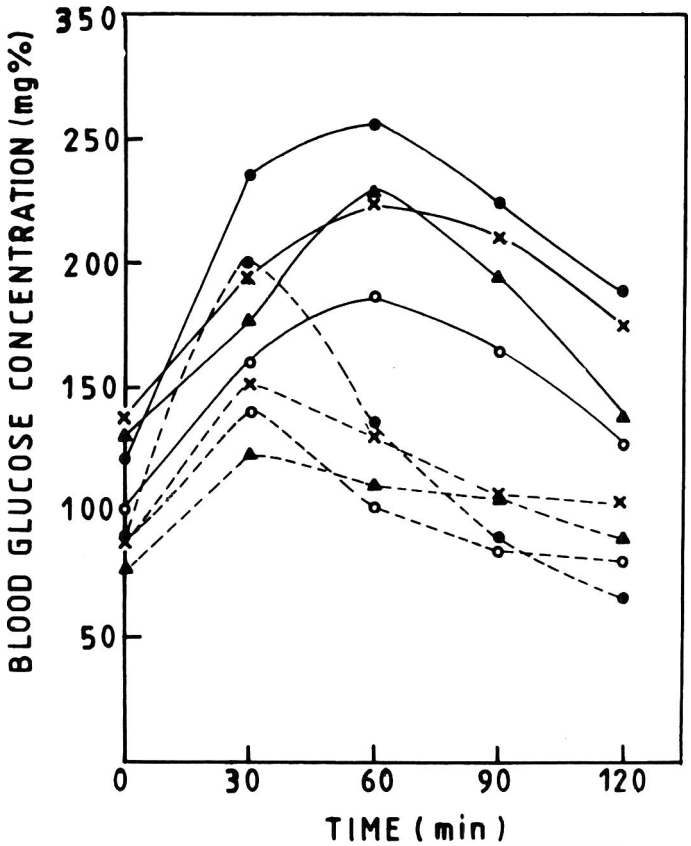

Fig. 1. Blood glucose response to wheat-legume blends in normal and impaired glucose tolerance (IGT).
—●— Control glucose —X— Malted —⊖— Popped
—▲— Roller dried - - - Normal —— IGT

higher than that in normal subject, the peak concentration reaching after 60 min in the former as compared to 30 min in the latter. At the end of 2 h, the blood glucose levels fell near to the fasting level in the normal subject, whereas the level remained above the fasting level in the impaired glucose tolerant subject (Fig. 1). The maximum blood glucose concentrations with the glucose load as well as the processed foods were much higher in impaired glucose tolerance, as compared to those in normal subject (Table 1), showing that blood glucose responses to the three processed formulations get boosted in the impaired glucose tolerant subject, as compared to those in normal subject. In impaired glucose tolerance, the maximum blood glucose concentration for the malted, and roller dried products were almost similar, while that of popped food was slightly lower. On the other hand, the maximum blood glucose concentration for the malted product was higher than that of the roller dried or popped products in the normal subject. The malted food showed a higher glycemic index than the popped, and roller dried foods in both the subjects. Interestingly, the glycemic index of roller dried food

TABLE 1. MAXIMUM BLOOD GLUCOSE CONCENTRATION AND GLYCEMIC INDEX OF MALTED, POPPED AND ROLLER DRIED CEREAL-LEGUME BLENDS IN NORMAL AND IMPAIRED GLUCOSE TOLERANT SUBJECTS*

	Maximum blood glucose concentration, mg %		Incremental area under glucose curve, mg.min/dl		Glycemic index	
Attribute	Subject		Subject		Subject	
	Normal	IGT	Normal	IGT	Normal	IGT
Glucose	199	256	4718	11760	100.0	100.0
Malted	151	223	3900	7065	82.7	60.1
Popped	137	185	1882	6435	39.9	54.7
Roller dried	123	229	2603	6099	55.2	51.9

* Average of duplicate determinations,
IGT : Impaired glucose tolerant

was least in the impaired glucose tolerant subject, in contrast to that of popped food in the normal subject.

Thus, it can be concluded that the carbohydrates of the malted food are more digestible as compared to those of popped, and roller dried foods in the impaired glucose tolerant as well as in normal subjects. The high glycemic response of the malted food may be attributed to the predigestion of starch to lower molecular weight sugars during the malting process (Lorenz 1980). The data show that the malted food may be more suitable for the normal geriatric subjects, because of its higher digestibility (Lineback and Ponpipom 1977), and low bulk characteristics (Malleshi and Desikachar 1982), while the popped food may be more suitable for geriatric diabetics.

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Proximate Composition and Protein Quality of Stabilized Rice Bran

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Fractional classification of rice bran proteins, the proximate composition, and protein efficiency ratio (PER) of untreated, and stabilized brans have been studied. Rice bran proteins are rich in albumins (32%), and globulins (26%). Protein contents of defatted, milled, and sieved bran flours ranged from 16.5 - 18.2%. PER of acid-stabilized bran was highest (2.18), followed by untreated (2.09), heat-stabilized (2.03), and parboiled rice brans (1.99).

Keywords : Rice bran, Untreated, Milled, Sieved, Acid and heat stabilization, Proeins, Protein efficiency ratio, Composition.

Possibilities of using rice bran, and its derivatives for human food have been explored, although commercial exploitation has been limited (Saunders 1986, 1990). High quality protein, and edible oil are the most valuable bran components (Saunders 1986, 1990). One of the problems in incorporating rice bran in food products is its high instability due to lipase (Akazawa 1972), which catalyzes, splitting of the oil into free fatty acids, and glycerol (Enochian et al. 1980), thus reducing shelf-life (Hunnell and Nowlin 1972). Stabilization of rice bran helps to overcome this problem (Desikachar 1974). Methods proposed to stabilize rice bran are based on altering the moisture content, temperature or pH to destroy the activity of the lipase (Prabhakar 1987). These processes involve heat treatment, low temperature storage, and chemical treatment (Cornelius 1980; Sayre et al. 1982; Prabhakar and Venkatesh 1986). Stabilized bran can be used for formulation of food products (Saunders 1990), and for extraction of oil (Cornelius 1980). Bran from parboiled paddy is more stable, and can be kept up to 15 days, without any stabilization treatment (Sayre et al. 1982). Physico-thermal properties of rice bran have also been reported (Maharaj Narain et al. 1978). David et al (1965) studied the quality of bran oil as influenced by the conditions of storage of rice bran. Narasimhan et al (1988) reported that rice bran stabilized by treatment with concentrated hydrochloric acid at 4.0 and 5.5% levels was highly unfavourable for the multiplication of red flour beetles. Flow belaviour properties of rice bran protein concentrate have also been reported (Bera and Mukherjee 1991). Since full-fat or defatted rice bran is used as a source of protein (Saunders 1990), it is of interest to study the effect of bran stabilization treatment on protein quality. Therefore, the present study was undertaken to determine the protein efficiency ratio of untreated, and stabilized rice bran.

Fresh rice bran from paddy of 'Gowri Sanna" variety was procured from local mills. Parboiled rice bran sample used was from 'Jaya' variety. Acid stabilization was effected by treating fresh rice bran with concentrated hydrochloric acid (12N) at the rate of 4.0 ml/100g (v/w) as per the method described by Prabhakar and Venkatesh (1986). Heat stabilization was achieved by wet-heat treatment of rice bran in an autoclave (Model 9143, Conrad Engelke, Krauss Maffei Co., Germany) at 1 kg/cm^2 pressure (110°C) for 10 min (Hermans 1970). The autoclaved material was dried in a cabinet dryer (Armstrong Smith, Bombay, India) at 50°C for 3 h to a final moisture content of 5%.

The raw materials, namely untreated, acid-stabilized, heat-stabilized, and parboiled rice bran were defatted individually (Barber and Barber 1985), using food grade n-hexane. Cold extraction of fat with a contact time of 10 h with the solvent was repeated four times. The defatted material was exposed to sunlight for 2 h to remove solvent traces. The defatted rice bran was finely powdered using a plate mill (Chandra Co., Madras), sieved (40 mesh), and used in all experiments. Moisture, protein (Kjeldahl), fat, total ash, and crude fibre were determined in triplicate (AOAC 1984). Carbohydrate was calculated by difference.

Proteins from untreated, defatted rice bran were fractionally classified by sequential extraction with water, sodium chloride (1 M), ethyl alcohol (75%), and sodium hydroxide (pH 11.0) (Kinsella 1976). Nitrogen extracted in each medium, and leftover residue were also analyzed (AOAC 1984).

The method described by Osborne et al (1919) was used to determine the protein efficiency ratio

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TABLE 1. PROXIMATE COMPOSITION OF RICE BRAN SAMPLES (g/100g)

Attribute	Moisture	Proteins	Fat	Carbohydrates (by difference)	Ash	Crude fibre
Rice bran ('Gowri Sanna' variety[a])						
Full Fat[a]	5.1 ± 0.55	13.5 ± 0.67	19.9 ± 0.44	41.3 ± 1.3	12.2 ± 0.40	8.0 ± 0.30
Defatted[a]	6.8 ± 0.57	17.0 ± 0.72	1.5 ± 0.05	50.6 ± 1.4	14.3 ± 0.52	9.8 ± 0.36
Parboiled rice bran ('Jaya' variety[b])						
Full fat[b]	6.4 ± 0.25	10.7 ± 0.53	19.8 ± 0.68	30.2 ± 1.32	19.9 ± 1.10	13.0 ± 0.28
Defatted[b]	3.5 ± 0.22	15.4 ± 0.62	0.50 ± 0.01	41.8 ± 1.43	24.8 ± 0.81	14.0 ± 0.32
Defatted, milled and sieved						
Untreated[a]	5.0 ± 0.42	18.2 ± 0.41	1.5 ± 0.02	56.0 ± 1.23	14.1 ± 0.31	5.2 ± 0.30
Acid stabilized[a]	4.6 ± 0.45	18.0 ± 0.65	1.6 ± 0.06	55.5 ± 1.10	14.6 ± 0.47	5.7 ± 0.25
Heat stabilized[a]	5.8 ± 0.53	17.5 ± 0.70	0.70 ± 0.02	55.6 ± 1.40	13.8 ± 0.52	6.6 ± 0.28
Parboiled[b]	3.3 ± 0.15	16.5 ± 0.52	0.48 ± 0.03	48.0 ± 1.20	22.1 ± 0.92	9.6 ± 0.29

• - Values are mean ± standard deviation of three estimations.

a. 'Gowri Sanna' variety

b. 'Jaya' variety

(PER) of unstabilized, stabilized, and parboiled rice brans. 'Wistar' strain weanling male rats, 22 days old, and weighing between 37-40 g, were used for feeding standard, and test protein diets. All the diets contained (%) groundnut oil 10, sucrose 10, salt mixture 2 (Hubbel et al. 1937), vitamin mixture 2 (Chapman et al. 1959), test protein (at 10% level), and corn starch to make up the weight to 100g. The results obtained were statistically analyzed, using Duncan's new multiple range test (Duncan 1955).

The proximate compositions of different samples of rice bran are shown in Table 1. Full-fat rice bran contained 19.9% fat, which decreased to 1.5%, while protein content increased from 13.5% to 17% on defatting. In addition, defatting resulted in an increase in other components, namely carbohydrates, ash, and crude fibre. Milling, and sieving of untreated, acid-stabilized, and heat-stabilized rice bran did not result in altering the composition. Heat-stabilized material had a slightly lower fat content, thereby indicating better extraction of oil from the cellular matrix, as a result of exposure to high temperature during heat stabilization (Graci et al. 1953). Full-fat, and defatted bran from parboiled paddy showed lower protein contents but higher contents of ash, and crude fibre, as compared to untreated rice bran sample. In the defatted sample, the proteins, carbohydrates, and other constituents proportionately increased, as a result of fat extraction. The protein content in the defatted, and milled material was higher (16.5%) with a considerable reduction in crude fibre content (9.6%), as compared to defatted samples.

As can be seen from Table 2, nearly 32% protein was extracted in water at neutral pH, after which sodium chloride extracted a further 26% of the protein. These trends indicated a high amount of albumins, and globulins in rice bran proteins, In alcohol, and alkaline media (pH 11.0), prolamines, and glutelins were extracted to the extent of 4.5 and 13%, respectively. After sequential extraction of rice bran proteins, the residue contained about 21% proteins. Similar results are reported by Cagampang et al (1966), and Betschart et al (1977). Among cereal proteins, rice proteins are considered to be of highest in quality (Lasztity 1984). Among the protein fractions of rice, albumins are reported to have the highest biological value due to a high lysine content (Tamura et al. 1952), while the values for prolamines are the lowest (Tecson et al. 1971). Globulin fraction has a high concentration of methionine (Iwasaki et al. 1982). Since rice bran is mainly made up of an aleurone layer containing high percentage of rice albumins, and globulins, the protein quality of rice bran has been reported to be higher than that of the milled rice (Lasztity 1984). Rice bran proteins are relatively rich in essential amino acids, and account for higher ratio

TABLE 2. FRACTIONATION OF RICE BRAN PROTEINS BY SEQUENTIAL EXTRACTION

Medium of extraction	Fraction	Proteins extracted, %
Water, pH 6.8	Albumins	32.20 ± 2.56
Sodium chloride, 1M	Globulins	26.11 ± 2.34
Ethyl alcohol, 75%	Prolamins	4.54 ± 1.1
Sodium hydroxide, pH 11.0	Glutelins	13.03 ± 2.03
Residue		21.12 ± 1.08

TABLE 3. PROTEIN EFFICIENCY RATIO OF DIFFERENTLY TREATED RICE BRAN FLOURS

Diet	Average initial weight, g	Average protein intake, g	Average gain in weight, g	Protein efficiency ratio, experimental	Protein efficiency ratio, adjusted
Casein	37.0	27.7	74.3	2.68[a]	2.50
Defatted rice bran, untreated	37.0	37.6	78.6	2.09[bc]	1.95
Acid stabilized rice bran	36.8	36.2	79.0	2.18[b]	2.03
Heat stabilized rice bran	36.7	37.3	75.8	2.03[c]	1.89
Parboiled rice bran	36.9	38.8	77.2	1.99[c]	1.86
SEm				± 0.05 (36 df)	

Randomized block design, 5 groups of ten rats each. Means of the same column followed by different letters differ significantly according to Duncan's new multiple range test ($p<0.5$).

of essential amino acids (g/ g total N), and essential amino acid indices (Prakash and Ramanatham 1994).

PER values of variously treated, and untreated defatted rice bran flours are presented in Table 3. PERs of acid-stabilized, and untreated rice bran were 2.18 and 2.09, as compared to PERs of 2.03 and 1.99 of heat treated, and parboiled rice bran. PER values for defatted bran, as reported in literature, range from 1.99-2.19 (Lynn 1969; Saunders 1986), and these correlated well with adjusted (as compared to casein value of 2.5) PER values of 1.95, 2.03 and 1.89 obtained for untreated, acid-stabilized, and heat-stabilized rice bran samples, respectively, in the present study. Several studies indicate that heat treatment can lower PER values, which are significantly influenced by the type of protein being heated, and the duration of heating (Wolf et al. 1979; Keyes and Hegarty 1979). These could serve to explain the lower PER values obtained for heat-stabilized, and parboiled rice bran flours. The present study indicates that acid stabilization of rice bran does not influence the protein quality adversely, and that acid stabilized bran can be used as a source of protein in food formulations.

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Organochlorine Insecticide Residues in Bovine Milk and Commercial Baby Milk Powder

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Bovine milk samples from different dairies in Bangalore city, and popular brands of baby milk powder were monitored for the residues of two organochlorine insecticides, i.e., dichlorodiphenyltrichloroethane, and hexachlorocyclohexane. Of the 30 samples of bovine milk, 83.3% samples were contaminated with dichlorodiphenyl trichloroethane, and its metabolites, while the residues of hexachlorocyclohexane, and its isomers were detected in all the samples. In five brands of baby milk powder from two manufacturing batches, dichlorodiphenyltrichloroethane, and its metabolites were not detectable, while all the samples were contaminated with α, β and γ isomers of hexachlorocyclonexane.β-hexachlorocyclohexane was predominantly present in over 60% samples above the prescribed maximum residue limit.

Keywords : Insecticide residues, Dichlorodiphenyltrichlorocthane, Hexachlorocyclohexane, Metabolites, Isomers, Bovine milk, Baby milk powder.

The buildup of pesticide residues in food chain is of concern due to health hazards to human being (Bindra 1971). Pesticide residues have been reported in foodgrains, vegetables, fruits, and animal products in market samples from time to time (Agnihotri 1983; Kalra and Chawla 1983; Kaphalia et al. 1990; Handa 1992). It is of concern that these toxic residues have invaded baby foods also (Dhaliwal and Kalra 1978; Kumar et al. 1991). Among the different organochlorine insecticides, dichloro-diphenyltrichloroethane, and hexachloro-cyclohexane have been identified as poisonous, but are still used widely in India for pest control in agriculture, and public health programmes (Mehrotra 1989). The analysis of fresh bovine milk samples from Bangalore city, and different brands of commercial milk powder was carried out in the present studies for the residues of dichlorodiphenyl-trichloroethane, and hexachlorocyclohexane insecticides.

Whole milk samples (500 ml in three replicates) from each of five main dairies (code No 1,2,3,4, and 5) in Bangalore city were collected at fortnightly intervals for three months, July-September 1991. The milk samples were collected from milk booths of different dairies, and immediately transported to laboratory in ice boxes. The samples from each of the source were processed for analysis after thorough shaking for breaking the fat globules. Five popular brands of baby milk powder, manufactured in different cities in the country, and marketed in Bangalore city were selected for analysis. The milk powder samples were collected from two manufacturing batches of each brand in two lots, in triplicates, during 1991, and 1992. Whole milk was prepared from the milk powder in measured quantity of water, based on fat content as specified on the tin.

A 20 ml milk sample, from the thoroughly homogenised milk so prepared, was extracted with 3 x 40 ml n-hexane for residues of dichlorodi-phenyltrichloroethane, hexachlorocyclohexane, and their metabolites/isomers. Insecticide residues in hexane layer were cleaned up from fat, and other co-extractives by acid digestion method (Kapoor and Kalra 1988, 1989). The quantitative determination of insecticide residues was carried out by gas liquid chromatography (Varian GC-3600 model Aerograph, Sunnyvale, California, USA), equipped with electron capture detector, and 2 m long stainless steel column packed with a mixture of 1.5% OV-17+1.95% OV 210 adsorbed on chromosorb W. Other conditions were: column temperature 190°C; inlet temperature 220°C; detector temperature 220°C, and nitrogen gas flow rate of 30 ml/min. Residues of dichlorodiphenyl-trichloroethane were identified, and analyzed for para para-dichlorodiphenyldichloroethylene, para para-tetrachlorodiphenylethane, para para-dichloro-diphenyltrichloroethane, while hexachlorocyclo-hexane residues were analyzed for α, β and γ isomers (Kalra et al. 1983).

The results of the monitoring of the extent of the residues of dichlorodiphenyltrichloroethane, and hexachlorocylohexane, along with their main metabolites/isomers, are presented in Table 1. Of the total 30 samples analyzed over a period of 3 months, 83.3% samples were found to be contaminated with dichlorodiphenlytrichloroethane,

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and its metabolites, while all the samples were contaminated with hexachlorocyclohexane in varying quantities, ranging from traces to as high as 0.225 ppm. Total dichlorodiphenyltrichloroethane residues averaged at 0.0221 ppm ± 0.0179. Among the different metabolites of dichlorodiphenyltrichloroethane, para para-dichlorodiphenyldichloroethylene accounted for 81.9%, para para-tetrachlorodiphenylethane for 10.8% and para para dichlorodiphenyltrichloroethane for 3.6% of the total dichlorodiphenyltrichloroethane residues. The presence of the residues of these metabolites was at the mean level of 0.0181 ppm in 25 samples, 0.0023 ppm in 11 samples, and 0.0008 ppm in 6 samples, respectively (Table 1). While only 10% milk samples were found contaminated with dichlorodiphenyltrichloroethane residues higher than the prescribed maximum residue limit of 0.05 ppm, 80% samples contained hexachlorocyclohexane residues above maximum residue limit of 0.01 ppm. The quantities of hexachlorocyclohexane residues were very high as compared to dichlorodiphenyltrichloroethane. Some of the samples showed 20 times high hexachlorocyclohexane residues to the prescribed maximum residue limit. This indicates the widespread, and high contamination of milk with toxic insecticide residues. The data of the status of contamination of milk with dichlorodiphenyltrichloroethane, and hexachlorocyclohexane residues across the country (Handa 1992) also concur with the present finding.

The results of residue analysis for total dichlorodiphenyltrichloroethane, and hexachlorocyclohexane in dry milk powder are detailed in Table 1. Of the 30 samples monitored, none was found to contain any detectable residues of dichlorodiphenyltrichloroethane, and its metabolites. Dhaliwal and Kalra (1978) and Kumar et al (1991) have reported contamination of dry milk powder with dichlorodiphenyltrichloroethane, and its metabolites at bioconcentration levels that may cause ill effects. On the other hand, in the present studies, all the samples were found to be

TABLE 1. RESIDUES OF ORGANOCHLORINE INSECTICIDES IN BOVINE MILK AND COMMERCIAL BRANDS OF MILK POWDER

Source • Average residues of DDT and HCH and their metabolites/isomers (μg g^{-1})

	pp-DDE	pp-TDE	pp-DDT	Total DDT	α -HCH	β-HCH	γ-HCH	Total HCH
				Bovine milk samples				
1	0.0112	0.0006	0.0008	0.0128	0.0520	0.0276	0.0540	0.1336
2	0.0146	0.0016	0.0016	0.0145	0.0331	0.0251	0.0416	0.1001
3	0.0133	0.0066	0.0000	0.0140	0.0213	0.0091	0.0216	0.0513
4	0.0308	0.0048	0.0016	0.0373	0.0331	0.0105	0.0505	0.0941
5	0.0203	0.0053	0.0012	0.0268	0.0785	0.0295	0.0586	0.1666
Mean residue	0.0181	0.0023	0.0008	0.0229	0.0436	0.0204	0.0453	0.1090
± SD	0.0133	0.0057	0.0021	0.0179	0.0322	0.0210	0.0306	0.0664
				Milk powder samples				
1A	--	--	--	--	0.050	0.100	0.050	0.200
1B	--	--	--	--	0.100	0.150	0.010	0.250
2A	--	--	--	--	0.001	0.001	0.001	0.003
2B	--	--	--	--	0.020	0.001	0.001	0.022
3A	--	--	--	--	0.030	0.020	0.001	0.051
3B	--	--	--	--	0.050	0.040	0.002	0.110
4A	--	--	--	--	0.030	0.020	0.001	0.051
4B	--	--	--	--	0.050	0.030	0.010	0.090
5A	--	--	--	--	0.005	0.0202	0.001	0.026
5B	--	--	--	--	0.005	0.0202	0.001	0.026
Mean residue	--	--	--	--	0.0341	0.0402	0.0078	0.0821
± SD	--	--	--	--	0.0200	0.0486	0.0125	0.0868

Residues of DDT and its metabolites were not detected in any of the milk powder samples

• The samples of bovine milk were collected from five dairies supplying milk to Bangalore city and the samples of milk powder were collected from different brands manufactured in Maharashtra, Gujarat, Punjab, U.P. and Karnataka States.

pp-DDE = para, para-Dichlorodiphenyldichloroethylene; pp-TDE = para, para-Tetrachlorodiphenythane; pp-DDT = para, para-Dichlorodiphenyltrichloroethane; α -HCH = Alpha-Hexachlorocyclohexane; β-HCH = Beta-Hexachlorocyclohexane; γ-HCH =Gamma-Hexachlorocyclohexane; SD = Standard Deviation.

contaminated with different isomers of hexachlorocyclohexane at varying quantities. The mean total of 0.0821 ppm hexachlorocyclohexane residues was shared by 50% b-hexachlorocyclohexane, 40% a -hexachlorocyclohexane, and 10% g-hexachlorocyclohexane isomers. The quantitative composition of various isomers was higher over the maximum residue limit of 0.05 ppm in 6 samples for a -isomer; 0.02 ppm in 10 samples for b -isomer, and 0.01 ppm in 6 samples for g -isomer of hexachlorocyclohexane, respectively. Higher persistence of b -isomer residues in milk, and milk products has also been reported by Dhaliwal and Kalra (1978), Kapoor and Kalra (1988) and Kathpal et al (1982) as compared to other isomers, probably due to its chemical stability towards degradation in dairy products (Kannan et al. 1992). The statistical analysis of residue data reflects wide difference in lower, and upper limits of confidence intervals of the residues for each of the hexachlorocyclohexane isomers, regardless of the brand. However, the samples from brand 1 were invariably found to contain exceptionally higher residues in both batches followed by brands 3, 4, 2 and 5.

This study suggests immediate review of the widespread use of hexachlorocyclohexane in pest control, as the resultant residues are hard to degrade (Kalra et al. 1983), and the persistent residues may have made their way even into the baby's system over a period of time right from birth, and may cause detrimental effects on human health.

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Proteins, Free Amino Acids and Carbohydrate Contents of Twenty-nine Almond Selections from Jammu and Kashmir

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Twenty-nine almond selections from Jammu and Kashmir, and a locally bred almond 'Hybrid-15' were analyzed for crude protein, soluble protein, free amino acid, soluble sugar, and starch contents in their matured kernels. Crude protein, soluble protein, and free amino acid contents ranged between 20.49-39.66, 13.48-23.61 and 0.19-0.64% on dry weight basis, respectively. The ranges of soluble sugar, and starch were 3.57-12.47%, and 0.73-2.63%, respectively. Selections 'JKS-47', 'JKS-50', 'JKS-65' and 'JKS-77' were superior in terms of crude protein, and starch contents, while 'JKS-189', 'JKS-190', 'JKS-266' and 'JKS-287' had higher quantities of soluble proteins, free amino acids, and soluble sugars in their matured kernels.

Keywords : Almond selections, Sub-tropics, Kernel, Crude proteins, Free amino acids, Soluble sugars, Starch.

Copious information is available on kernel composition of almonds grown in temperate regions (Barbera et al. 1987; Kester et al. 1990). However, similar information pertaining to indigenous almond selections (Kumar and Uppal 1990), growing under sub-tropical environment is limiting (Kumar 1987).

On the other hand, carbohydrate composition of mustard (*Brassica juncea*) seed meal (Sindhu Kanya and Kantharaj Urs 1983), fermented melon seeds of Nigeria (Achinewhu 1987); amino acid composition of Surabul seed kernel proteins (Azeemoddin et al. 1988), and cottonseed protein isolate (Hanumantha Rao et al. 1987) have been reported. Only recently, information on oil content, and its constituents in these selections is provided by Kumar et al (1984). In this report, quantitative data are presented on contents of crude protein, soluble protein, free amino acid, soluble sugar, and starch contents in matured kernels of almond selections from Jammu and Kashmir.

The experimental material and sampling procedure followed in the present study were as described previously (Kumar et al. 1994). Crude protein was calculated from % total nitrogen (McKenzie and Wallace 1954) by multiplication with factor 5.18 (Saura-Calixto et al. 1981). Quantitative determinations were made for the soluble proteins (Lowry et al. 1951), free amino acids (Lee and Takahashi 1966), and total soluble sugars (Yemm and Willis 1954). For the analysis of starch, free amino acids, and total soluble sugars, the extraction was done as follows: In each case, 500 mg dried sample was mixed with 5 ml of 80% ethanol on a magnetic stirrer. The extraction was repeated twice, and the extract was centrifuged at 3000 rpm for separating the supernatants. To the residue, 5 ml of 70% ethanol was added, stirred, centrifuged at 3000 rpm, and the supernatant was separated. This process of residue extraction was repeated four times. All the alcoholic extracts were pooled together and kept in a refrigerator till analyzed. The residue was processed for quantitative analysis of starch as per the procedure of Clegg (1956). The chemical analysis was done using two replicates in each, and the data reported are the average of the observations recorded in 1984 and 1985.

Crude protein : It varied from 20.49 to 39.66% (Table 1). Selections 'JKS-194', 'JKS-212' and 'JKS-287' had significantly lesser amounts of total proteins, than 'Hybrid-15'. Some of the earlier reports (Pyzhov et al. 1977; Dhaliwal et al. 1978) indicate that different almond varieties contain total proteins in the range of 14.7 to 34.9%. In another study, Saura-Calixto et al (1981) argued that almond kernels contained not below 20g protein/ 100 g almond kernel on dry matter basis.

Soluble proteins : These ranged from 13.48 to 23.61%. Selections 'JKS-57', 'JKS-77', 'JKS-168', 'JKS-185', 'JKS-189', 'JKS-190' and 'JKS-266' had higher soluble protein contents, than 'Hybrid-15'. Information on this biochemical constituent in almond kernels is completely lacking.

Free amino acids : These ranged from 0.19 to 0.64% with overall variability of 31.42%. Selections 'JKS-189', 'JKS-190', 'JKS-248' and 'JKS-266' had higher free amino acid contents than the control. Free amino acid contents of almond have also been least studied.

Soluble sugars : Thse values ranged from 3.57

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TABLE 1. CONTENTS (ON DRY WEIGHT BASIS) OF CRUDE PROTEIN, SOLUBLE PROTEIN, FREE AMINO ACID, SOLUBLE SUGAR AND STARCH IN ALMOND KERNELS UNDER SUBTROPICAL CONDITIONS

Selection No. 'JKS'	Crude proteins, %	Soluble proteins, %	Free amino acids, %	Soluble sugars, %	Starch, %
47	34.72	19.73	0.21	4.71	1.95
50	34.27	16.15	0.23	4.08	1.72
55	30.67	17.60	0.19	6.81	2.63
57	33.04	20.45	0.29	8.28	2.56
65	36.27	13.84	0.26	4.35	1.83
67	28.78	18.30	0.25	6.45	1.61
68	32.03	16.63	0.24	6.73	2.05
69	39.66	13.73	0.39	3.62	1.52
75	28.80	19.79	0.31	6.66	1.29
77	35.34	22.71	0.29	9.77	2.31
145	32.16	18.37	0.28	5.19	1.62
168	31.59	20.15	0.37	6.66	1.48
169	31.94	17.59	0.32	4.38	1.74
172	33.18	16.70	0.42	12.47	1.75
184	30.42	19.57	0.23	6.89	1.70
185	34.73	20.88	0.30	8.48	1.57
189	34.56	23.61	0.55	9.25	0.73
190	31.81	22.13	0.58	11.01	2.35
194	20.49	13.98	0.39	8.92	1.56
198	32.21	15.48	0.32	5.38	1.23
212	23.00	13.48	0.37	8.27	1.29
235	33.65	14.23	0.37	3.95	1.20
238	30.30	17.57	0.46	6.02	1.17
248	33.39	16.34	0.64	4.38	1.60
268	33.18	22.19	0.52	8.46	1.66
270	32.80	19.37	0.32	3.57	1.56
280	30.68	17.90	0.40	6.93	1.66
287	27.58	19.52	0.40	9.33	1.55
288	37.45	16.52	0.44	3.76	1.28
Hybrid-15	30.14	18.36	0.43	4.22	1.49
Mean (X)	31.96	18.09	0.35	6.63	1.65
S.E. (Mean) ±	1.38	1.01	0.04	0.86	0.15
CV (%)	11.88	15.36	31.42	35.74	24.84

to 12.47%. No selection had lower values of total soluble sugars than that in the control. In the past, many workers have reported upto 8% soluble sugars on dry weight basis in almond kernels (Kosev and Lichev 1974; Saura-Calixto et al. 1981, 1984; Lopez-Andreu et al. 1985). However, Bogdan and Molnar (1975) reported more than 10% total soluble sugars in almond kernels.

Starch : Its range extended from 0.73 to 2.63%. Selections 'JKS-189', 'JKS-198', 'JKS-235' and 'JKS-238' had lower quantities of starch in their kernels than 'Hybrid-15'. Previously, starch was considered lacking in almond kernel (Woodroof 1979). On the contrary, Kosev and Lichev (1974) found that starch content ranged between 3.06 and 4.78% in different almond kernels. Later on, Saura-Calixto et al (1983) indicated that starch varied from 0.07 to 0.17% in freshly collected kernels with corresponding lower values in samples stored for several months.

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Effect of Enzymatic Pre-treatment on Dehulling of Rapeseed

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Enzymatic pre-treatment of rapeseeds for different time intervals showed significant effect on efficiency of rapeseed dehulling. Samples treated with 51.23 mg enzyme/100g dry matter for a period of 12 h at 45°C showed increase in the dehulling fractions by 6.65 %, and reduction in the unhulled fractions by 5.53 %.

Keywords : Rapeseed, Enzymatic pre-treatment, Dehulling, Effect of pre-treatment time.

Dehulling, the process of removal of husk, is not an essential unit operation in recovery of oil from rapeseed, although it would be useful for obtaining light coloured oil, and improving the feed value of the resultant meal (Wiegand 1978). Dehulling removes a major part of the fibre, and also a group of pigments, which otherwise pass into the meal, and lower its feeding value (Niewiadomski 1990). The efficient dehulling process should fulfil three conditions i.e., complete removal of meal from coat, protection of meal from loss of oil, and avoidance of dust formation during crushing (Niewiadomski 1990). Dehulling has been reported to reduce polyphenols and phytate, thereby improving the protein quality of pearl millet (Pawar and Parlikar 1990). Dehulling could be easily carried out in a modern paddy dehulling machine (Schneider 1979). Uppal et al (1984) have carried out studies on the nutritive value of low glucosinolate variety of rapeseed meal.

Effect of enzymatic pre-treatment on dehulling of rapeseed has not yet been reported. Most of the literature reports are either on direct dehulling of seed (Niewiadomski 1990; Schneider 1979). It has been emphasized that the biodegradation of gum layer between the cotyledon, and the husk can help to loosening of the bond, thereby leading to easy removal of the husk (Verma et al. 1993). The present study was, therefore, undertaken to investigate the effect of enzymatic pre-treatment on rapeseed, dehulling at different time intervals.

Rapeseed, variety 'PT-303,' having 5.74% moisture (wet basis), was obtained from the Crop Research Centre of the University. Enzyme solution for use in the pre-treatment 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 (500 ml capacity), containing sterilized 200 ml YPSS medium. It consists of (g/L): soluble starch 15, yeast extract 4, K_2KPO_4 1, $MgSO_4$. $7H_2O$, 0.5 and distilled water 1000 ml. (pH 7.0). The flask was, then, incubated at 45°C on an incubator shaker (120 rpm for 3 days (Bhatnagar 1987). About 50 ml of the fermented medium was used to inoculate a batch fermenter (Working capacity of 1.5 L Model F-2000, New Brunswick Scientific Co. Inc., Edison, U.S.A), having 1.5 litre of sterilized wheat bran medium (wheat bran 60 g, distilled water 1.5 L, pH 7.0). Fermentation was carried out at 45°C (Bhatnagar and Johri 1987) with 1 VVM air supply, and 120 rpm agitator speed. The fermentation was continued for 72 h, the fermented material was filtered through cheese cloth, and the filtrate was centrifuged at 10,000 rpm at 10°C for 10 min. The solution, thus obtained, was used as crude enzyme for the treatment. The protein content was measured following the method of Lowry et al (1951). The filter paper activity (IU/ml) was measured, using the method of Mandels et al (1976). The crude enzyme solution had 0.3307 IU/ml filter paper activity, and 3.366 mg/ml crude protein content.

For each experiment, a sample of 20g rapeseed was taken in a conical flask (125 ml capacity), and a calculated amount of enzyme solution was mixed to increase the moisture content from 5.74% to 23.33% (wet basis), and to get the protein content of 51.53 mg/100 g dry matter of the sample. This combination was found optimum for maximum oil yield from pre-treated whole rapeseed (Sarker 1993). The flask was shaken manually, and equilibriated in a refrigerator for 8 h. The flasks were then kept on an incubator at 45°C for hydrolysis (Bhatnagar and Johri 1987). Incubation time was varied from 1 to 13 h at an interval of 1h (Sarker 1993). The hydrolyzed samples in duplicate were dried in an oven at 70°C for about 2 h to inactivate the enzyme, and also to reduce the moisture content to about 7%.

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TABLE 1. EFFECT OF CLEARANCE ON DEHULLING OF RAPESEED

Clearance, mm	Whole seed, %	Dehulled seed, %	Other,* %
0.16 ± 0.02	3.62 ± 0.11	63.05 ± 1.05	32.58 ± 0.54
0.30 ± 0.02	5.82 ± 0.17	64.85 ± 1.31	29.25 ± 0.32
0.47 ± 0.02	6.36 ± 0.32	67.17 ± 1.75	26.43 ± 0.46
0.62 ± 0.02	16.38 ± 0.35	58.22 ± 0.96	25.02 ± 0.62

* includes hulls, fragmented hulls and immature seeds, fines

The samples (20 g each) were then dehusked on a rice dehusking machine (Model Tm, Satake Engineering Co. Ltd., Tokyo, Japan). The clearance between the two rubber rolls was adjusted to 0.47±0.02 mm, based on preliminary observations (Table 1) for maximum dehulling of the grain. The dehusked, and unhusked fractions were collected in a container, and later on separated manually. The husk was collected in a separate container. Quantity of air fed through the conveyor for effective husk separation was set through preliminary trials. Untreated sample was used to compare the results. Average values of two different dehulling fractions were calculated.

Results indicate that enzymatic pre-treatment for different time durations has significant effect (r^2=0.96, n=13, p=0.01) on dehulling (Table 2). The dehulled fraction of untreated sample was 67.17%, as against the maximum value for enzyme treated sample was 73.82%, thereby indicating an increase of 6.65 %. The unhusked fractions decreased from 6.36 to 0.81%. However, the other fractions (hulls, fines, fragmented hulls, immature seeds) varied from 25.38 to 27.78%, without any specific trend with the treatment time.

TABLE 2. EFFECT OF ENZYME TREATMENT TIME ON DEHULLING OF RAPESEED

Treatment time, h	Whole seed, %	Dehulled seed, %	Other,* %
Control	6.36 ± 0.15	67.17 ± 1.20	26.43 ± 0.62
1	4.73 ± 0.12	68.05 ± 0.93	27.78 ± 0.37
2	4.48 ± 0.20	68.57 ± 0.50	26.96 ± 0.19
3	3.70 ± 0.13	69.30 ± 0.50	26.98 ± 0.18
4	3.03 ± 0.11	70.10 ± 0.60	26.87 ± 0.13
5	2.98 ± 0.10	70.24 ± 0.16	26.70 ± 0.06
6	2.65 ± 0.12	70.95 ± 0.55	26.35 ± 0.05
7	2.42 ± 0.06	71.58 ± 1.82	25.98 ± 0.01
8	1.91 ± 0.02	72.12 ± 0.78	25.96 ± 0.24
9	1.13 ± 0.05	72.87 ± 1.07	25.98 ± 0.17
10	0.89 ± 0.06	73.16 ± 0.36	25.95 ± 0.27
11	0.85 ± 0.01	73.58 ± 0.52	25.57 ± 0.18
12	0.83 ± 0.01	73.82 ± 0.58	25.38 ± 0.19
13	0.81 ± 0.02	73.21 ± 0.21	25.68 ± 0.07

* includes hulls, fragmented hulls, fragmented hulls and immature seeds, fines

Increase in dehulled fractions for treated sample has been attributed to the mixed activity enzyme action mainly carbohydrase, and pectinase (Sosulski et al. 1988) on gum like substances like pectin, galactoxyloglucan present in between the cotyledon, and seed coat (Yiu et al. 1982, Niewiadomski 1990). Enzymes degrade gummy substances into simple units, thereby improving hulling efficiency (Verma et al. 1993). They have reported improvement in hulling efficiency in pigeonpea, when pre-treated with microbial enzyme. Treatment time has significant effect ($p < 0.01$) on increase in percent dehulling, as amount of dehulled fractions increased with time to 73.82% at about 12h, and then remained nearly constant. The treatment period at which maximum increase in dehulling fractions was observed is in good agreement with the optimum treatment time reported by Smith et al (1993) in case of soybean. Enzymatic pre-treatment also enhanced the overall oil recovery upto 4.20%. The dehulled seed, and other fraction contained 54.02%, and 25.32% oil, respectively. The higher amount of oil in other fraction was due to contamination with small particles of meal, and germs. This quantity of oil represented 14.26% of the pre-treated seed oil. Oil colour of pre-treated sample was observed to be better visually, as compared to control. Free fatty acid value increased from 0.9 to 1.0 due to pre-treatment.

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Proximate Composition of Unconventional Leafy Vegetables from the Konkan Region of Maharashtra

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Analysis of ten unconventional leafy vegetables found in the forest, and culturable wasteland of Konkan revealed that drumstick leaves, *math*, *katemath*, *bharangi* and *kawala* contained comparatively higher amounts of crude proteins, crude fats, ash, crude fibres, and total carbohydrates. Drumstick leaves could be rated the best among all the leafy vegetables studied, as it also contained higher ascorbic acid, and β-carotene as well as lower oxalates. The uncoventional vegetables, in general, contained less oxalates, as compared to cultivated vegetables.

Keywords : Leafy vegetables, Unconventional, Konkan forest, Proximate composition.

Green leafy vegetables are, in general, good sources of vitamins, and minerals (Ramdasmurthy and Mohanram 1984). Certain leaves contain proteins of high quality, although the total quantity of protein is limited. (Pike and Brown 1970). There are many leafy vegetables growing wild, which can be consumed more often in larger quantities, but these are usually looked upon as poor man's food, and people from well-to-do families hesitate to eat these unconventional vegetables. Effect of cooling on the changes in ascorbic acid, and carotene content of green leafy vegetables has been reported (Ranjana Sood and Bhat 1974).

Some studies on the nutritive value of sun-dried green leafy vegetables (Sehgal et al. 1975); *Launaea Cornuta*, a wild leafy vegetable of Tanzania (Ndossi and Sreeramulu 1991); and some conventional, and non-conventional green leafy vegetables (Kausalya Gupta et al. 1992) are reported.

The agro-climatic conditions of the Konkan region are characterized by hot, and humid climate, high rainfall during monsoon, and frost-free conditions (Anon 1972). The forest, and the culturable wastelands of the region nurture certain species of green leafy vegetables during rainy season, which are locally called as *phodsi*, *dhandgi*, *kawala*, *takala*, *bharangi*, *katemath*, *shewga* (drumstick), *ghol*, and others. Majority of these are annuals, growing in *kharif* season only, and used by the poor populations in their diet. Proximate compositions of ten such leafy vegetables are reported in this communication.

The edible leafy material (Table 1) was collected from the forest, and culturable wasteland nearby Dapoli, while *math* and *palak*, the cultivated species were obtained from the experimental farm of the University. Tender leaves, and edible stems of the vegetables were washed under running tap water, rinsed with distilled water, bloated, dried at 50 to 60°C, and ground to 60 mesh in a Waring blender (Braun AG, Frankfurt. Germany). The 60 mesh powder was used for chemical analysis.

Moisture, crude fibre, and ash were estimated as per standard methods (NIN 1983), and expressed on fresh weight basis. The crude fat was estimated by extraction process of Randall (1974), using Ra-fa-tech extractor (Tecator Inc., Colorado, USA); while crude proteins were calculated from the nitrogen content by Kjeldahl method using factor 6.25 (AOAC 1965), and the carbohydrate content by difference. Ascorbic acid in the oxalic acid extract was estimated by using 2,6-dichlorophenol indophenol dye (AOAC 1955), while β-carotene was separated by column chromatography using acetone-hexane (3:7) mixture, and estimated colorimetrically (AOAC 1970). Oxalic acid was determined by titrimetric method (AOAC 1980).

Moisture : The moisture content of the leafy vegetables ranged between 76.4 and 91.9%, the highest being in *phodsi* (91.9%). *Katemath*, *kawala* and *bharangi* showed comparatively low moisture content. The lowest moisture content of 76.4% was observed in drumstick leaves. In general, the leafy vegetables containing more moisture were tender, succulent in nature, while those containing less moisture were of woody texture.

Crude proteins : The maximum crude proteins were observed in drumstick leaves (7.0%), while *dhandgi* (1.8%), and *phodsi* (1.3%) contained the lowest amount. The others were intermediate in their protein contents. Similar values have been reported for drumstick, *kangkong* and *takala* leaves (Anon 1951; Ambegaonkar et al. 1964).

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TABLE 1. PROXIMATE COMPOSITION OF LEAFY VEGETABLES (fresh weight basis)

Common name	Botanical name	Moisture, %	Crude proteins, %	Crude fat, %	Ash, %	Crude fibre, %	Total carbo-hydrates, %	Ascorbic acid, mg/100g	β-carotene, µg/100 g	Oxalic acid, mg/100g
Math	*Amaranthus tricolor*	85.1	5.3	0.8	3.4	1.8	3.7	117.7	5641.0	873.6
Spinach	*Spinacea oleracea*	90.7	3.1	0.2	2.3	0.7	3.0	88.4	3143.3	586.2
Katemath	*Amaranthus spinosus*	82.3	4.9	0.4	2.9	1.7	7.8	137.8	7764.8	1161.4
Drumstick	*Moringa olefera*	76.4	7.0	0.9	2.1	2.2	11.5	229.9	5080.3	92.5
Takala	*Cassia tora*	85.3	3.3	0.4	2.5	1.7	6.8	178.8	1873.0	619.4
Ghol	*Portulaca oleracea*	89.7	2.3	0.3	1.8	1.6	4.4	59.8	3082.9	521.8
Bharangi	*Clerodendron serratum*	81.6	3.9	0.2	1.9	2.4	10.0	45.1	1307.3	258.0
Cowpea	*Vigna unguiculata*	88.5	3.8	0.3	1.0	2.1	4.3	59.8	5652.1	26.6
Kawala	*Smithia sensitiva*	81.4	5.6	0.7	1.1	2.4	8.9	93.3	1479.4	46.8
Phodsi	*Chlorophytum tuberosum*	91.9	1.3	0.3	1.3	1.0	4.2	71.0	1000.3	16.9
Kangkong	*Ipomea aquatica*	90.3	3.2	0.3	1.3	1.4	3.5	48.8	1989.7	29.0
Dhandgi	*Tricholepis ampelxicaulis*	91.6	1.8	0.3	1.2	1.5	3.8	70.6	828.0	30.7

Crude fats : All the leafy vegetabes were found to be the poor sources of fat, which ranged between 0.2 and 0.9% (Table 1). The maximum crude fat was observed in drumstick leaves (0.9%), *math* (0.8%) and *awala* (0.7%), while the rest of the vegetables contained less than 0.4% crude fats.

Crude fibres : The leafy vegetables were found to vary in their crude fibre contents. *Bhanangi* (2.4%), *kawala* (2.4%), drumstick leaves (2.2%), and cowpea leaves (2.1%) recorded high fibres, while spinach (0.7%), and *phodsi* (10%) were low in crude fibres. The species with more succulent growth, were found to have less fibres than woody perennials. Similar results were reported by Gopalan and Balasubramanian (1980) in case of *ghol* and cowpea leaves.

Ash : Ash contents of all the vegetables were high. The unconventional sources of leafy vegetables, namely *katemath*, *takala* and *drumstick* leaves contained appreciable amounts of ash, in comparison to the cultivated species such as spinach, and *math*. In general, the mineral matter ranged between 1.0 and 3.4%.

Carbohydrates : The unconventional leafy vegetables contained more carbohydrates than the conventional sources. Drumstick leaves (11.5%), *bharangi* (10.0%), *kawala* (8.9%), *katemath* (7.8%), and *takala* (7.8%) were superior in carbohydrate contents, as compared to rest of the vegetables studied. It has been reported that drumstick leaves contain 13.4% carbohydrates (Anon 1951).

Vitamins : Ascorbic acid was found in the range of 45.1 to 229.9 mg/100 g in the vegetables studied. The highest content of ascorbic acid was in drumstick leaves, followed by *takala*, *katemath* and *math*. *Kawala*, spinach, *phodsi*, *dhandgi*, cowpea leaves, and *ghol* were intermediates, while low values of ascorbic acid were observed in *kangkong* and *bharangi*. β-carotene was maximum in *katemath* (7764.8 µg/100g), followed by cowpea leaves, *math* and drumstick leaves. *Dhandgi* had only 828.0 µg β-carotene per 100 g fresh weight.

Anti-nutritional factors : Oxalic acid in the vegetables is considered as one of the anti-nutritional factors in human diet (Singh and Saxena 1972). The lowest amount of oxalic acid (16.9 mg/ 100g) was found in *phodsi*. *Katemath* and *math* showed appreciably high amounts of oxalates. The unconventional leafy vegetables namely *phodsi*, cowpea leaves, *kangkong*, *dhandgi*, *kawala* and drumstick leaves, in general, contained lower amounts of oxalic acid, thereby indicating good palatability, and safe use of these vegetables in human diet.

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***"The Food Industries of British India"* By K.T. Achaya, Published by Neil O'Brien Oxford University Press, YMCA Library Building, Jaisingh Road, New Delhi-110 001, 1994, pp 316, Price Rs.450/-**

It was a pleasant opportunity to review the book entitled "The Food Industries of British India" by Dr. K.T. Achaya. Two things struck me simultaneously, one the key word **Food Industry** and the other key word **Dr. Achaya.** These are complementary words, and the tradition of Dr. K.T. Achaya, and his books have gone a very long way in establishing with the publishers for his meticulous, detailed, to the point as well as the critical evaluation of many aspects of such books well documented. In this particular book, through the Oxford University Press printed during 1994, the subjects covered by Dr. Achaya in the area of "The Food Industries of British India" are salt, sugarcane, beverages, dairy products, animal products, coffee and tea products, and vegetable oils along with the products of bakery, confectionery, packaging, and factors for industrial growth. Also, his favourite topics, if I may say so, which are vegetable oils, and vanaspathi is dealt with a high degree of professionality. Each chapter on the above subjects opens up with a history of the product profile, the sources, and the development of such an industry during British period in different parts of the country. The by-product recovered as well as the production, and export of this product with an ultimate analysis of the future trends in this area. These have been very well written with statistics of production in the latter British India upto 1947-1948. The entire emphasis of the book lies in important references in various subject areas on which a reader can lay hands, if he wants further details. My feeling is after going through this book which itself is a compendium of information on the various references, there is very little need for somebody to go through the original references for facts, and figures, unless he wants to have the pleasure of opening up an old reference, and enjoy reading the original. Of particular mention should be, the figures which are furnished, giving the kind of sophistication that existed even during 1920s and 1930s is worthy of documentation. I am simply amazed about the Figure 7.1 which is a Power Driven Decorticator for groundnuts which was widely used in 1930s. The most recent decorticators of course, are highly precision made, electronically driven, but fascinating enough, the principle remains the same. The other striking feature of the book is that some of the advertisement clippings from the Calcutta Gazette regarding the products especially of the beverages look so informative, and raises the value of the book to such an extent, demanding it to be preserved as a document.

Overall, if one reads a book carefully, the following aspects will become very clear. (1) the existence of advanced food industries in the British India, rather the advancement of technology even at that stage was sufficient enough to give the fillip to Food Industry a high level technology at that time also (2) the skilled labour that was available at that time to run these industries needs a special mention. (3) the leadership that was available at that time, for example with personnel like Dinshawji Cooverji Pandole, who founded Duke and Sons in 1889 in Bombay to bottle soda-lemon and a host of others who led the industry to a success (4) the linkage that was established between Food Industries of British India, and the Food Industries in the current scenario can well be seen. (5) Lastly but not leastly, the importance of Agro-based Food Industries in India has a long history, and therefore has the advantage of experience with our raw materials. Looking into other angles, the advantage of the Food Industries over a long period in the country having database, modern gadgets, skilled and experienced labour, dynamic leaders, and captains of industries and the encouraging policy from the Government to Industry in the pre-independent India have all helped today's growth of the industry. I am sure to a large extent the credit to this success lies with those industrialists from the country, who pioneered and in spite of lack of sophisticated technology during those days, were still able to carve a niche globally, even at that time. This is very clear from this book, if one reads it very carefully. The book priced at Rs. 450/-, will be a valuable addition to personnel or professional libraries, since each of its page is loaded with information of high value, and the expertise of Dr. Achaya in expressing the facts in the most simple language, such that even a student can grasp what he has conveyed through this book. The book is very well written, and will be an asset to any organization.

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***A Practical Approach to Chiral Separations By Liquid Chromatography* : Edited by G. Subramanian, Published by VCH Publishers Inc, New York, pp 405, 1994, Price DM 178**

Today, chiral chromatography has assumed great importance not only because of the current scientific interest, but also by the increasing pressure of regulatory authorities against the marketing of recemic mixtures, particularly by the pharmaceutical industry. With the impending regulation in European, and North American region on production, and use of chiral drugs, the need to monitor production, report isomeric composition of products, and vigilance study of drugs have given tremendous impetus to the development, and application of this separation technique in the past decade or so. The other industries which require enantioseparations are agro-chemical, food & drink, and petro-chemical industries. Here, the typical example of Aspartame, artificial sweetener, may be cited, while R, R- isomer has sweet taste, and the S, R- isomer is bitter. The chemical production of flavours, and fragrances is highly dependent on enantioseparation for the correct property.

This book is aimed to provide a practical approach to chiral separation by liquid chromatography without subjecting the reader to extensive amounts of theory, although it does give sufficient theoretical background to form the basis for the development of new techniques. A team of experts from academic, and industrial laboratories throughout the world have compiled their findings, and experience in this book, so that it allows a choice of methodology from the whole spectra of currently available techniques. The authors have strived to cover the literature exhaustively upto the beginning of 1994, so that the current status of the subject is reported for the benefit of researchers, analysts, and industrial chemists in the field.

This book has 12 chapters, and chapter 1 is an introduction to enantioseparation by liquid chromatography. It gives an overview of the various chiral stationary phases, and chiral mobile phase additives applicable in chiral chromatography, along with the fundamental principles of chirality. The current status of chiral chromatography in various industry is also well described.

Chapter 2 gives an idea of modelling, and enantiodifferentiation in chiral chromatography. It describes what is molecular modelling and how to use it. The various computational tools are well explained. A detailed account of modelling enantioselective binding in chromatography, that illustrates the power of molecular modelling, and its vulnerability, is also given.

Chapter 3 deals with the regulatory implications of chiral separations. The author explains the principles of licensing of medicinal products, and also emphasizes on the necessity of separation of racemic drug mixtures.

The development of various chiral stationary phases, and application in the separation of racemic mixtures, both for analytical, and preparative scale, are discussed in chapters 4-10. Chapter 4 shows enantiomer separation using tailor-made phases by molecular imprinting. Principle of molecular imprinting, its historical development, and preparation methods, and applications of various chiral stationary phases are lucidly explained. The applications of cyclodextrin-bonded chiral stationary phases are described in chapter 5, whereas polysaccharide phases are dealt with in chapter 6. Various protein-based chiral phases are discussed at length in chapter 7. Chapter 8 gives a good idea about optically active polyacrylamide/silica composites, and related packings, and their applications in chromatography. Ion-pair chromatography in enantioseparation is discussed in chapter 9. In chapter 10, separation of enantiomeric compounds by chiral selectors in the mobile or solvent phase is detailed.

The role of chiral chromatography in pharmaceutical and bioanalytical applications are revised in chapters 11 and 12, respectively.

Thus, both analytical, and preparative-scale enantioseparations by liquid chromatographic techniques are covered in a down-to-earth practical way. The theory of chiral separations is dealt with in sufficient detail to guide the practising chromatographer interested in developing new techniques. Publication of this book is timely, and of immense value to the growth of this field.

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***Plants in Nutrition* : (77 World Review of Nutrition and Dietetics) Edited by A.P. Simopoulos. Publisher - Karger S, Basel AG, Allschwilerstrasse 10, P.O. Box. Postfach, CH-4009, Basel, Switzerland, 1995, P XIV+198 Price DM 237 or $ 158.50.**

A cursory glance at the topics discussed in this

book would show that the editor has made a sincere, and systematic endeavour to discuss the plants that can enrich diet. However, these plants are not commonly grown, consumed, and known. The contributors of each paper of the book have highlighted the agro-climatic conditions favourable for the cultivation of these plants along with their nutritional, and pharmaceutical uses. In 155 pages of this book, eight papers written by different authors are included.

The first paper on Micro-algae as source of w^3 fatty acids by Cohen et al elucidates the role of w^3 fatty acids (EAP & DHP), as a prophylactic for coronary heart diseases, artery diseases, thrombosis, hypertension, cancer of the colon, etc. For meeting the increasing demand of w^3 fatty acids, the authors have suggested the exploitation of micro-algae, as an alternative source. Besides exploring the production potential of oil by these micro-algae, the authors have also suggested the future direction of research in area such as the strains of micro-algae, which can synthesize increased amounts of w^3 fatty acids, with less cost of production.

To help fight protein and β-carotene malnutrition in developing countries, the authors of the 2nd paper have highlighted new dimension of production technology through increased large scale production of *Spirulina* algae, which is rich in these nutrients. They also have exhibited its simple cultivation technology, and its favourable effect on human body. Limitations of its uses in areas, where people are not accustomed to its mild marine bouquet as well as with its colour are also discussed.

The paper by Simopoulos et al on the use of Purslane in human nutrition, and its potential for world agriculture gives concise, and additional informations especially for its use as a medicine for treating G.I tract disorders and skin infection as well as considering it as cardiac tonic, and diuretic drug. Good nutritional value of this plant has been emphasized, because it contains promising quantities of LNA, vitamin E, vitamin C, β-carotene, pectin and glutathione. They also have suggested the future direction for conducting research on its extract to be recommended as a male contraceptive.

The fourth paper on sweet lupins in human nutrition by Uauy et al has highlighted the facts that in comparison to soybean, the grains of lupins are abundant in proteins. The authors have suggested that the use of its defatted flour and isolate with cereal preparations can prove to be better than that of soybean. To make it popular, they also have advocated development of new products, which would be acceptable to consumers.

In the paper on Barley Foods, and their influence on cholesterol metabolism the authors have convincingly argued about the potential use of glucans in the lowering of blood cholesterol level. They have emphasized on the greater use of barley and its products as well as the need to increase the popularity of its products among the consumers.

The paper on Nopal epitomizes its medicinal, and nutritional values. Nopal, a succulent cactus plant, rich in Ca, Mg, K, is popular for treating gastritis, peptic ulcer and fatigue along with high serum cholesterol and sugar levels in Mexican people. In spite of its great uses and demands in Mexico, the authors are able to get only a meagre record for its production. They also have asserted that there is a wide gap in the knowledge about the effect, and mechanism of action of Nopal.

The last paper on Howthorn (*Shan Zha*) by Cheu et al has among other things, described the effect of its drink preparation in lowering blood lipids in humans and rats. The authors have emphasised that this wild fruit bearing trees grow in China and its fruits are used as food and medicine, especially for the treatment of heart diseases and obesity. The authors have observed limitations of the research on Howthorn, because in traditional medicine, the whole seeds have been used whereas modern pharmacapeia is based on isolation of active principles from the plant, as its fruits or seeds.

The editor has done a commendable work in compiling this book after putting up a lot of efforts in collections, and preparations. This book contains valuable information, and historical background. Coloured illustrations for precise identification of each plant would have added to the utility, because most of the plants are not known commonly. Subject index could be increased especially in relation to nutritional, and pharmaceutical aspects. Index for scientists, whose names were mentioned in each paper could have been added to increase the value or contributions of each scientist. Excellent quality of papers for printing, and binding have been used to add to the value of this book.

Nutritionists, food technologists, and agricultural scientists would be benefitted by the information presented in this book for their research

work. This book would also be highly useful, as a reference material.

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Control of Fish Quality : Edited by J.J. Connell, Published by Fishing News Books, A Division of Blackwell Scientific Publications, 238 Main Street, Cambridge, MA 02142, USA, 1995, pp. 245, Price : £ 25.00

This is a unique publication written by Prof. J.J. Connell, former Director of Torry Research Station, Aberdeen, Scotland. The book is organized into 10 chapters, dealing with almost all aspects of quality of fish such as intrinsic, and extrinsic factors, affecting fish quality, pathogens prevalent in fish, and measures to control them, official methods of analysis to measure physical, and chemical changes, occurring during storage of fish products, several methods of preservation including chilling, freezing, drying, salting, smoking etc. In addition, the book also contains national, and international codes of practice to maintain high quality standards for fish products.

Chapter 1 deals with terminology used in quality determination of fish, Intrinsic quality characteristics are detailed in chapter 2. Intrinsic qualities include species, size, sex, condition, and composition of fish. All these characteristics play significant roles in deciding the quality of fish. Fish also naturally harbour parasites, protozoa, platyhelminths, nematodes, crustacea, fungi and bacteria. The contamination depends upon the locality as well as species of fish. Some species of fish are naturally toxic, and others may be contaminated with pollutants such as metals, elements, organic chemicals, radioactive isotopes, and microorganisms from sewage. This information is useful for fish processing industries, which can take proper steps to avoid processing of such contaminated fish.

Chapters 3 and 4 incorporate the informaiton on the extrinsic quality of fish viz., Deterioration (1) causes and effects (2) practical measures of preventing deterioration, and factors influencing the rate of deterioration. The author has excellently dealt with this subject with many simplified flow charts for handling of fish on board, pictorial presentation of attractive packaging, which helps in augmenting sales of the products, filleting and chilling, optimum conditions of freezing method of prepared fish, canning and heat processing of fish. This chapter will certainly help food technologists to follow good manufacturing practices for obtaining better quality products. A few tables on storage life of different varieties of fish at different temperatures included in this chapter will be of much help to the students of food technology. Several methods of preservation such as salting, drying, canning, bottling, smoking, freezing, chilling etc. given in detail will help the readers to get an in-depth knowledge in the field of fish processing. Various defects observed in improper chilling, freezing, smoking etc. will warn the fishing industry to take proper note of preservation.

Safety aspects of consumption of fish are dealt with in chapter 5. Incidence of risks of diseases from consuming different products viz., raw fish, semi-preserved, and fermented fish are presented in a tabular form. Incidences, and severity of risks from different sources such as, pathogenic organisms, parasites, chemicals, scombrotoxin and naturally toxic fish are also compiled in a tabular form to facilitate readers to understand the subject.

Chapter 6 encompasses exhaustively the microbiology of fish products. The author has presented in great detail about the prevalent pathogens viz., *Aeromonas, Listeria monocytogenes, Clostridium botulinum, Vibrio cholera* and *Vibrio parahaemolyticus* in fish products. However, no mention is made of other important pathogens normally reported in shrimps i.e., *Salmonella* and *Bacillus cereus*. This information would have made this topic complete.

In the latter part of this chapter on microbiology of fish, controlling methods such as personal hygiene, disinfection, water quality, equipments etc. are described in order to obtain fish products of hygienic quality.

Several additives and preservatives can also keep the fish for longer time. However, emphasis is laid on proper packaging and labelling practices. An excellent example of good quality labelling is depicted in the book, which will be of great help to traders as well as consumers.

Chapter 7 is devoted to methods for assessing the quality of fish products during storage. The methods include sensory evaluation by sight, touch, odour and flavour. Grading of freshness based on skin, outer slime, eyes, gills, peritoneum, and internal odour is tabulated, thus, facilitating taste panelists to understand how to score the sample correctly.

Among laboratory methods, physical methods involving direct measurement of fat content of fish,

and Torrimeter to measure degree of freshness of fishery products in chilled condition are quite useful for wholesale dealers for rapid disposal of fish, if it is of doubtful quality.

Biochemical methods described include Total Volatile Bases (TVB), hypoxanthine, and trimethyl amine (TMA) changes in their concentrations with the degree of spoilage are graphically presented. Microbiological methods are also described. Thus, the chapter is of great help to quality control laboratories to assess the shelf-life of fish products. Trade in fishery products is smooth, provided quality assurance is given to consumers as well as official inspection is done. Industrial quality assurance includes end-product specification, processing specification, checking inspection of raw materials, and end product. recording, reporting and finally hazard analysis as well as critical point. Official inspection requirements for several countries in the world are given.

Chapters 9 and 10 encompass national, and international standards for fish products. Microbiological standards, standard methods of analysis of samples, national, and international codes of practice, and scoring system are dealt with in detail.

I, strongly recommend that this book should be on the shelf of every food scientist, and technologist working on fish preservation, exporters, and quality control inspectors. Graduate students will find it extremely useful in understanding the various methods of preservation, types of poisoning, and pathogenic bacteria present in fish, control measures etc. Thus, the book will be a good companion to fish traders, exporters, food inspectors, food technologists, graduate students, microbiologists. The author Prof. Connel is an authority on almost all aspects of fish technology, whose valuable experience, and views are depicted in this book.

It is needless to say that this wide range of subjects covered in this book forms the focal point, and enhances its value. The book is excellent with the printing being very clear and brought out in a professional way with good editing and quality to make the reader comfortable in terms of understanding each and every aspect.

I finally congratulate the publishers, and the author for bringing out such a valuable book.

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"Use of Irradiation to Control Infectivity of Food-borne Parasites" Proceedings of a Final Research Co-ordination Meeting held at Mexico City, Mexico from 14 to 18 June 1991. The meeting was organized by the 'Joint FAO/IAEA Division of Nuclear Techniques in Food and Agriculture. This was printed by the IAEA in Austria in December 1993. pp 140, Price 400/- (Austrian Shillings).

The proceedings include the results of the work carried out over five years (1986-1991) by twelve researchers, participating in the programme, i.e., from Argentina, Belgium, China, Japan, the Republic of Korea, Mexico, Poland, Thailand, and the United States of America.

Foodborne parasites are common throughout the world. These pose significant health problems, and cause economic losses in terms of agricultural commodities, and human productivity. The prevalence of the parasite diseases is highest in Asia especially in Japan, Thailand, China etc. where people have the habit of consuming raw sea-foods, and animal foods. Exposure of food to low dose gamma irradiation has proven to be an useful tool for the destruction of bacterial, and parasitic pathogens as well as preservation of food, in general. This proceeding provides valuable data on the use of irradiation to control infectivity of foodborne parasites.

The book starts with a summary report of the work carried out by twelve researchers, and is presented in a comprehensive manner. The brief overview has been divided in to five sections. The first section deals with fish, and invertebrate foodborne parasites, and their control by use of gamma irradiation. An extensive, and valuable information is given in minimal effective dose (MED) of irradiation needed to eliminate seven parasites.

Further, the book details on the use of irradiation to control toxoplasmosis, trichinellosis, and taeniasis/cysticercosis.

Finally, the strategies to implement food irradiation programmes in the control of these food-borne parasitic infections have been outlined.

The summary is followed by the detailed papers presented by twelve participants. The earlier available data, with the exception of data on *Trichinella spiralis*, a parasitic nematode, were insufficient for the use of irradiation technology to control foodborne parasites. The present findings, therefore, have established conclusively the potential

for application of food irradiation in the control of liver flukes, tape worms, round worms, trichinosis, toxoplasmosis, etc. This book, therefore, will certainly serve, as a valuable reference to scientists, technologists, and veterinarians working on research projects related to foodborne parasites.

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***Glyphosate* : IPCS, Environmental Health Criteria 159, Published under Joint Sponsorship of United Nations Environmental Programme, the International Labour Organisation, and World Health Organisation (WHO) as a part of the IPCS (International programme on chemical safety) World Health Organisation Geneva 1992; WHO Distribution and Sales 1211, Geneva 27, Switzerland; WHO - Regional Office for South-East Asia, World Health House, Indraprastha Estate, M.G. Road, New Delhi, 110 002. pp 177, Price SW. Fr 18.90 in developing countries.**

This report contains the collective views of an international group of experts, mainly concerned with the environment, toxicology and toxicological effects of the herbicide glyphosate. This is a small monograph with 179 pages comprising 12 chapters with the following sections 1. Summary. 2. Identity. Physical and chemical properties and analytical methods. 3. Sources of human, and environmental exposure. 4. Environmental transport, distribution and transformation. 5. Environmental levels of human exposure. 6. Kinetics and metabolism in laboratory animals. 7. Effects on laboratory animals and *in vitro* test systems. 8. Effect on humans. 9. Effect on other organisms in laboratory, and field. 10. Evaluation of human health hazards, and effects on the environment. 11. Recommendations for protection of human health. and 12. Further research followed by an exhaustive 361 references.

Glyphosate is a systemic non-selective herbicide intended to use against various deep rooted perennial species including grass, and sedges. The main biodegradative pathway appears to be splitting of C-N bond to produce amino methyl phosphonic acid (AMPA). There is also a second route by which microbes degrade the glyphosate in the soil i.e., via sarcosine and O-phosphate after which sarcosine is degraded to glycine, and finally to CO_2 via formaldehyde. Thus, the degraded compound is not toxic to mammalian systems. Glyphosate is prepared in various commercial formulations with various synonyms and concentrations in various countries. Even the recommended rates of applications vary with different countries. It is done by aerial application or by mist spraying, and there is variable timing of application.

In section 2, physical and chemical properties, and methods of analysis are discussed. This is a well written chapter with descriptions of various methods of determination, using HPLC, and GCMS. These methods being expensive, it warrants for the development of simpler methods for identification, and characterization of glyphosate. Chapter 3 deals with the sources of human, and environmental exposure. The major method of exposure is (1) occupational exposure during formulation and spraying, and this can be removed by protective clothing, and thus 96% of the protection can be obtained. Section 4 presents data on transport distribution, and transformation. It does not seem to be transported in significant amounts in soil and water. Contamination of drinking water from ground water is minimal because of the low mobility of glyphosate. In such cases, ozone can be efficiently used, and it conjugates, and removes toxicity. Besides, iron salts are known to complex with glyphosate, and thus the toxicity can be removed. Biodegradation seems to be the major source of dissipation. In the soil, most of the glyphosate is associated with the upper layer only. In the field, photodegradation has also been reported. When administered to animals it is rapidly excreted without degradation and does not bioaccumulate in tissues, in view of the high solubility, and ionic character as revealed by studies with fish, mollucs, and crustaceans. However, absorption is limited to 30-36% in rats, rabbits, goats and hens when given orally through the gastric intestinal tract. The AMPA (Aminomethyl phosphonic acid) is the only metabolite found in the urine. However, it is known to cause several symptoms, such as growth retardation increased liver weight, and degenerative changes when fed at very high levels. It has not been found to be teratogenic or carcinogenic at the level of its usage. In chattels, high levels are found to be toxic. Moderate to severe eye and dermal irritation by constant exposure have been reported. However, in humans no controlled studies have been done so far. The action of glyphosate on microorganism has been reported particularly in pseudomonas where it is degraded in the AMPA pathway. In prokarocytic cyanobacteria, enzyme systems are inhibited, indicating the herbicidal

property. Variable results have been reported with different populations of higher plants. In plants, it is known to inhibit mostly aromatic amino acid pathway and also respiration, and photosynthesis. One important aspect of glyphosate is that it does not inhibit degradation of cellulose, starch, and proteins by the microbes. However, there are conflicting reports on the phytotoxicity in aquatic plants. It is not toxic to earthworms and bees at the level of its use and does not affect nodulation. Several factors such as pH, hardness of water and temperature, seem to be involved with respect to its toxicity to fish. It also does not affect the nematode population. Thus in view of its low toxicity, low volatility, and low absorption by the body it is a safe herbicide for field application, provided the workers wear full protective clothing.

The last section deals with the future line of work. However, nothing is mentioned about its effect on the immune system. This is also one area to be pursued, apart from other areas mentioned.

This forms a useful report for toxicologists who are planning new experiments with pesticides, and herbicides, as various methods have been extensively reported. It will also be useful to agricultural scientists, policy makers of herbicide removal agencies and also useful for the Ministry of Agriculture. The book is well written and covers extensively the effect on all vertebrates, invertebrates and microbes. The references given at the end are comprehensive. I strongly recommend this book for scientists and toxicologists, who would be interested in evaluation of new herbicides and pesticides.

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"Storage of Cereal Grains and Their Products" - Fourth Edition (1992) Edited by D.B. Sauer, J.D. Miller and H.L. Trenholm. Published by the American Association of Cereal Chemists Inc., St. Paul, Minnesota, USA, pp 615, Price US $ 98/-; Other countries US $ 118/-.

This monograph is the fourth edition published by the American Association of Cereal Chemists, after an interval of 10 years. In this edition, there are 16 chapters written by 27 experts in different areas. There are four new chapters *viz.*, physical properties of cereal grains, historical perspective on development of storage techniques, integrated pest management of stored grain insects and the economics of grain storage, which make this volume a comprehensive one.

The information provided in the chapter entitled "Physical properties of grains" on structure of the grain, bulk density, angle of repose, flow of grain from a bin, hardness of grain etc. are highly useful in understanding the subsequent chapters with reference to pest attack as well as the storage system. The chapter on "History of grain storage techniques" elucidates the grain harvest and storage practices from the ancient times to the present day practices in different parts of the world. The importances of ecological factors, regulating the insect population, besides monitoring the pests are stressed under integrated pest management of stored grain insects. Under the economics of grain storage, the specific components of storage costs and the influence of government policies on the price structure are discussed.

The major problems of storage *viz.*, moisture, insects, rodents and microflora are dealt with elaborately, as in the previous edition. These chapters provide an in-depth background information for understanding and application of storage techniques discussed in the other chapters. However, this monograph deals with mainly the practical problems of storage in North America.

It would have been much more useful, if one or two chapters on practical problems of storage are included from different agroclimatic zones of the world, for example, the Asian region, where the storage practices and conditions are different.

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***Advanced Dairy Chemistry : Volume 2 : Lipids* Edited by P.F. Fox. Published by Chapman & Hall, Madras, India, 1995, pp.443 Price £ 99.**

Milk lipids contribute greatly to the organoleptic, and rheological properties of milk, and milk products. From nutritional view point also, milk fat is very important, as it contains essential fatty acids, and fat soluble vitamins, besides being a rich source of energy. In fact, the rich pleasing flavour of milk fat is not duplicated by any other fat. Consumption of milk fat is considered as a risk factor in causing atherosclerosis. However, this has been questioned by many research workers. This calls for a thorough, and clear evidence before dietary recommendations are made against milk fat. Milk lipids are subjected to chemical, and enzymatic changes, which can cause flavour defects in products, containing them

during storage. Many of these aspects have been discussed in Developments in Dairy Chemistry Vol.2 - Milk Lipids, published in 1983. Considerable progress has been made in several aspects of milk lipids during the past 11 years. Therefore, there is a need for a critical appraisal of the advances in the area of milk lipids. It is to the credit of Dr. P.F. Fox, the internationally renowned dairy chemist, that a reference book on milk lipids has been edited, and published under the series of Advanced Dairy Chemistry. After going through the book, it is evident that Dr. Fox has put forth all his efforts to provide an impressive, and much needed reference book, illustrating the advances in milk lipids, spreading over 10 valuable chapters, contributed by internationally recognised experts.

The first chapter of the book contributed by Christie, thoroughly discusses the various aspects of composition, and structure of milk lipids. The chapter gives a comparative account on the fat content, composition of lipid classes, profile, and arrangement of fatty acids of lipids from the milks of various species. The chapter is so conclusively prepared that the author rightly feels that there is no scope for further research in aspects on composition, and structure of milk lipids of cow, and human. With other species, however, relatively little is known on the composition of various lipid classes (other than triglycerides) or of their structure, and further work in this area would have both biochemical, and nutritional relevance.

Fat in milk, unlike carbohydrates, and proteins, is subjected to marked variations in both amount, and composition, due to nutritional factors, which are super imposed upon variations due to species, genetic, and lactational factors. The nutritional effects specific to fats arise principally because a considerable, but variable proportions of the fatty acids of milk lipids are derived directly from dietary lipids. Hawke and Taylor have highlighted the influence of nutritional factors on the yield, composition, and physical properties of milk fat in chapter 2. Since an appreciation of the factors, which alter the composition, and amount of milk fat secreted by the mammary gland is dependent upon a working knowledge of the biochemistry of the pathways involved, the authors have commenced the chapter with the mechanism of biosynthesis of milk fat in mammalian tissue.

The fat globule membrane is acquired from the endoplasmic reticulum during formation of the intracellular lipid droplet precursors of milk lipid globules, and from specialized apical regions of the plasma membrane during the process of cellular discharge of the lipid droplets. Molecular genetic, and technological innovations in cell biology have been applied with success to aspects of this research area. Keenan and Dylewski have reviewed the origin, growth, and secretion of lipid globules with the help of electron micrographic illustrations. The compositional aspects of the fat globule membrane are also discussed aptly.

Physical, and colloidal aspects of the milk fat globules are of importance for the stability of milk, and milk products as emulsions. This concerns creaming, aggregation, coalescence, and partial coalescence, and also some aspects of lipolysis. Foaming, and whipping are strongly affected by properties of the fat globules, as are such product properties as viscosity, and colour. The properties of high fat products may especially be dominated by those of the fat globules. Walstra has discussed these aspects in detail in the chapter on physical chemistry of milk fat globules.

The crystallization behaviour of milk fat is primarily of importance, because it greatly affects first, the consistency of high fat milk products, especially butter, and second, the occurrence, and rate of partial coalescence in oil-in-water emulsions. As a result, churning, and whipping processes are also affected. Moreover, it affects the mouthfeel of butter-like products, large crystals giving a sandy impression, and much solid fat causing a cooling effect. It also determines oiling-off in products like butter, which is promoted by low solid fat content, and large crystals. Fat crystals not only stabilise the droplets in a water-in-oil emulsion, but may also affect the appearance of some products. Fat crystallization is a complicated subject, because the wide range of triglycerides leads to a wide melting range, especially in milk fat. Nucleation, and growth of crystals are intricate because of the complicated shape of, and small differences between, the molecules. Triglycerides, like all long chain aliphatic molecules, can crystallize in various polymorphs. Consequently, milk fat crystallization never attains equilibrium, and is generally dependent on temperature history. New intricacies also occur in the formation of fat crystal networks. These aspects are discussed in a chapter by Walstra et al.

A number of economic, medical, and social factors have contributed to the decline in butter consumption. Consumers also prefer a fridge spreadable, and more nutritionally acceptable spread. The development of dairy spreads has been the response by manufacturers to these market forces. Efforts by equipment manufacturers to produce low-fat butter, using continuous churning

equipment with an additional sharing unit to incorporate the extra aqueous phase have recently met with commercial success. Chemistry, and technology of milk fat spreads have been narrated by Keogh.

Hydrolytic rancidity in milk, and milk products has been a concern to the dairy industries of most countries, and therefore calls for a constant vigilance to ensure effective control. Deeth and Fitz-Gerald have explained in detail about the lipolytic enzymes, causes, and prevention of hydrolytic rancidity, and analytical methods for estimating the degree of lipolysis. The detrimental, and beneficial effects of lipolysis have been described well in this chapter. The presence of a unique bile salt stimulated lipase in human milk, and its usefulness in the digestion of fat are also emphasized.

The other major chemical process by which lipids undergo deterioration is autooxidation. The mechanism of lipid autooxidation, products of oxidation, and off-flavours, factors affecting lipid oxidation, and antioxidants are narrated by Connor and D'Brien. Dietary cholesterol has often been blamed as a risk factor for atherosclerosis. However, evidence is mounting against this. Many studies further, have shown that pure cholestrol does not accumulate in the vascular endothelium, until lesions occur. It has been proposed that oxidation products of cholesterol may be a cause for lesion formation, after which pure cholesterol, and its esters accumulate as a secondary process leading to atherosclerosis. After a careful look on these aspects, it is observed that though a few reports indicate the presence of cholestrol oxidation products in milk, and milk products, many studies have also revealed their absence. Cholesterol is generally stable towards autooxidation, and its mechanism is similar to that of unsaturated fatty acids. Therefore, an effective control of lipid oxidation is helpful in arresting the formation of cholestrol oxidation products in milk, and milk products.

The dramatic switch from whole milk to lower fat varieties during the 1980's, and the equally dramatic fall in butter comsumption, and rise in low fat spreads have made a substantial contribution to the fall in fat intake in many industriallised countries. While cost clearly contributed to these changes in fat consumption, the increasing public interest in nutrition, and the focussing of attention by health educators on the adverse effects of fat with saturated fatty acids, and cholesterol have played a major role. The chapter on the nutritional significance of lipids by Gurr discusses the various roles of lipids in the diet, and in the body, assesses the evidence for a major role for dietary fat in the so called "diseases of affluence" and sets the nutritional significance of dairy lipids in perspective. It is summarised briefly, in the chapter, what are thought to be major implications of lipids, either as causative agents or in the dietary management of the disease. Particular reference is made to controversies, surrounding the role of milk fat in contributing to health, and disease. In the conclusion, which is a highlight of this chapter, the author advocates the need of caution, while accepting the recommendations for modification in the diets for whole populations. Since the impact of dietary cholesterol on blood cholesterol is minimal, Gurr feels that marketing of cholesterol reduced or cholesterol-free products by the dairy industry is a miscalculation by the dairy industry, and that by doing so the industry, and its scientific advisers have conceded that they have lost the battle to provide proper information on all scientific issues relating to milk, and health. While a reduction in fat intake may be a distinct advantage for many people, especially those who find difficulty in maintaining energy balance, there is a danger that, in this obsession with dietary fat, other, perhaps more important, life style factors are forgotten, including smoking, too little exercise, and raised blood pressure. The role of fat must be seen in perspective, and it should not be concluded that widespread changes in dietary fact consumption will lead inevitably to freedom from the so-called "diseases of affluence". The evidence does not point that way. The key to a good diet is variety, and milk, and milk products, both low, and full fat varieties, will continue to play an important role in contributing to that variety.

Mela, and Raats have given a description as the role of fat in the consumer acceptance of dairy products. Resolution of some of the issues raised by the authors in this last chapter of the book can have potentially important contributions to understanding the basis for, and possible industrial responses to the role of fat in the consumer acceptance of dairy, and other food products.

This book is a rich store house of information about the advances in milk lipids with latest literature citations. This book is indespensable to students, teachers, and researchers who are to know the latest developments in the area of milk lipids.

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