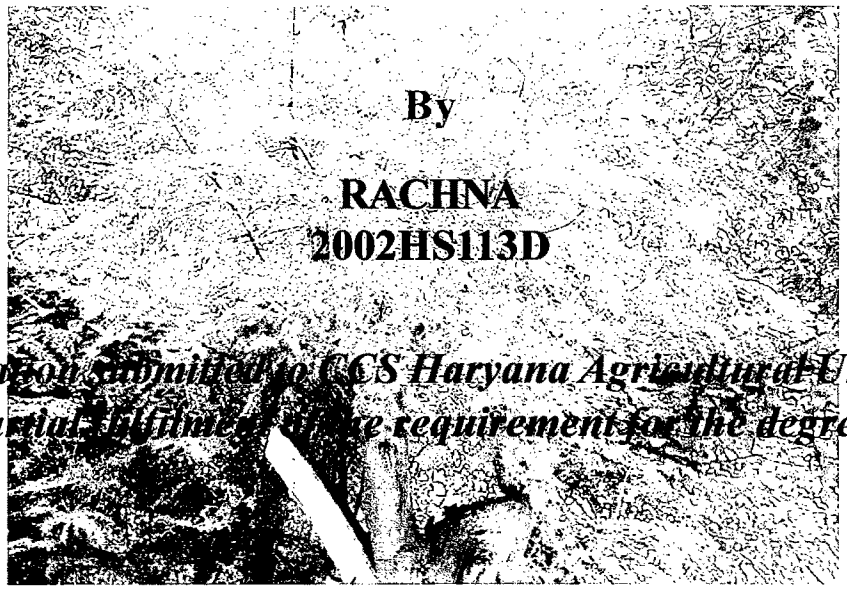
DEVELOPMENT AND NUTRIENT COMPOSITION OF VALUE ADDED PRODUCTS FROM DRUMSTICK (*Moringa oleifera*)



By

RACHNA

2002HS113D

Dissertation submitted to CCS Haryana Agricultural University in partial fulfilment of the requirement for the degree of :

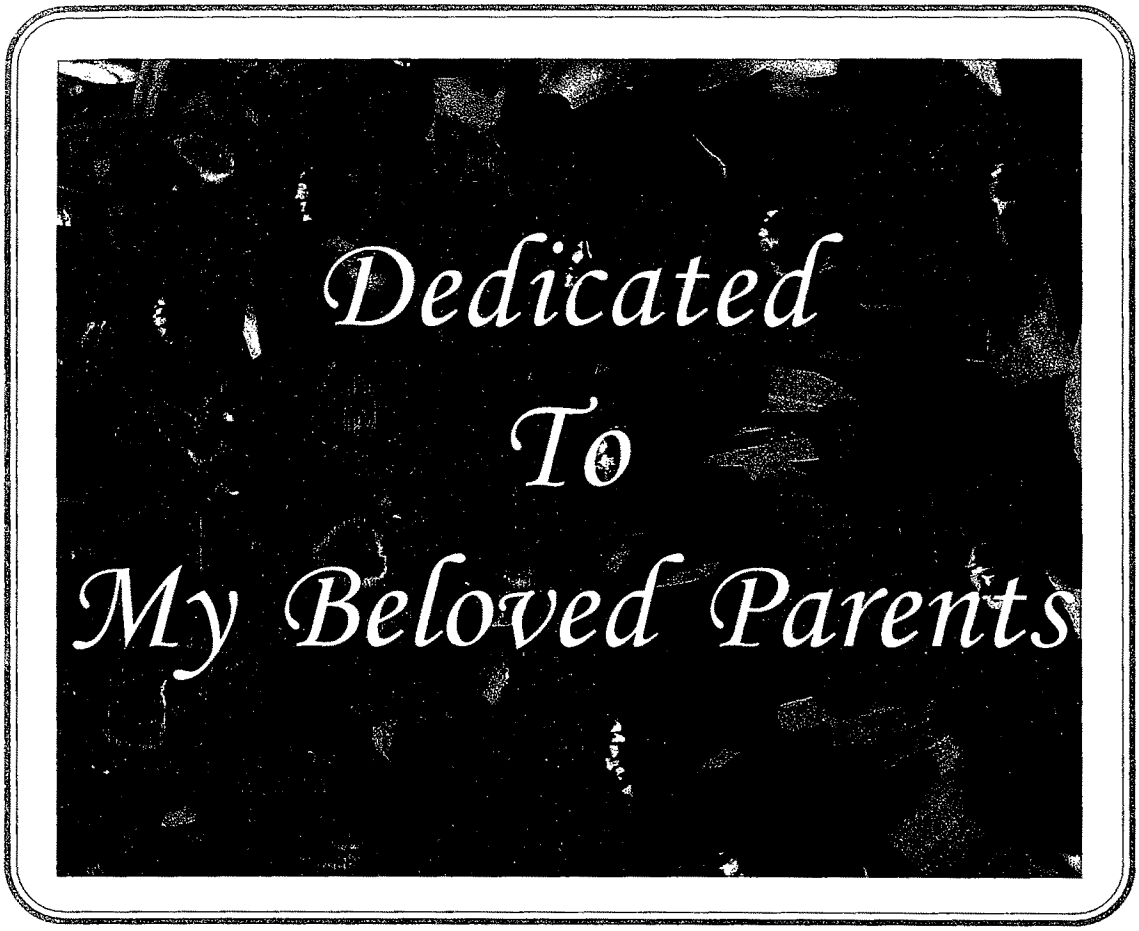
DOCTOR OF PHILOSOPHY

IN

FOODS AND NUTRITION

I.C. COLLEGE OF HOME SCIENCE
CCS HARYANA AGRICULTURAL UNIVERSITY
HISAR - 125 004

2006


Dedicated
To
My Beloved Parents

CERTIFICATE – I

This is to certify that this dissertation entitled, **"Development and Nutrient Composition of Value Added Products from Drumstick (*Moringa oleifera*)"** submitted for the degree of **Doctor of Philosophy** in the subject, **Foods and Nutrition** of the Chaudhry Charan Singh Haryana Agricultural University, Hisar, is a bonafide research work carried out by **Ms Rachna** under my supervision and guidance and that no part of this dissertation has been submitted for any other degree.

The assistance and help received during the course of investigation have been fully acknowledged.

N. Khetarpaul 15/6/06

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CERTIFICATE – II

This is to certify that this dissertation entitled **"Development and Nutrient Composition of Value Added Products from Drumstick (*Moringa oleifera*)"**, submitted by **Ms Rachna** to the Chaudhry Charan Singh Haryana Agricultural University, Hisar, in partial fulfilment of the requirement for the degree of **Doctor of Philosophy** in the subject of **Foods and Nutrition**, has been approved by the Student's Advisory Committee, after an oral examination on the same in collaboration with an External Examiner.

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Acknowledgements

I bow my head before Him, the Almighty God, without whom my present thesis would not have materialized. The very idea of this work having been completed, makes me feel grateful to all those who were incremental in the completion of this significant milestone in my academic journey.

Words are very poor substitutes to express one's emotions and feelings, there are no other alternatives to give vent to one's sentiments, particularly on an occasion like this, when one sits in acknowledging the debts of others.

I feel immense pleasure in expressing my heartfelt gratitude, obligation and indebtedness to m y Major Advisor, Dr. (Mrs.) Neelam Khetarpaul, Professor, Department of Foods & Nutrition, for her keen interest, sensible planning, inspiring guidance, close supervision throughout the pursuit of this investigation and critically going through the manuscript. Her suggestions have made this thesis a lucid writing marked by clarity of thinking and expression. Her very cordial and elegant behaviour would always remain a beacon light for me in future too.

I also express my gratitude to the members of my advisory committee Dr.(Mrs.) Pinky Boora, Dr.(Mrs.) Sushma Kaushik, Dr. R.K. Sharma and Dr. R.K. Behl for their valuable suggestions and timely assistance offered to me.

My thanks are due to both teaching and non-teaching staff members of the Department of Foods and Nutrition for their help during the course of study.

I am overwhelmed with rejoice to avail this rare opportunity to evice my profound sense of veneration and gratitude to my mother Smt. Savitri Nagpal, father Sh. Harish Chander Nagpal, sister Tamanna, and brother

Parvesh Nagpal for their inspiration, love, affection and encouragement proving to be moral force for my success.

It is my profound privilege to thank my friend Vimla, Manu, Vishakha, Navita, Sunita, Varsha and all other well wishers for their sweet association and affectionate cooperation.

The meticulous typing by Mr. Sushil Kumar is worth appreciable.

Last but not the least, I am also grateful to all those who helped me directly or indirectly to achieve the goal and to whom I have not been able to express my gratitude in individual.

Rachna

(Rachna)

Place: Hisar

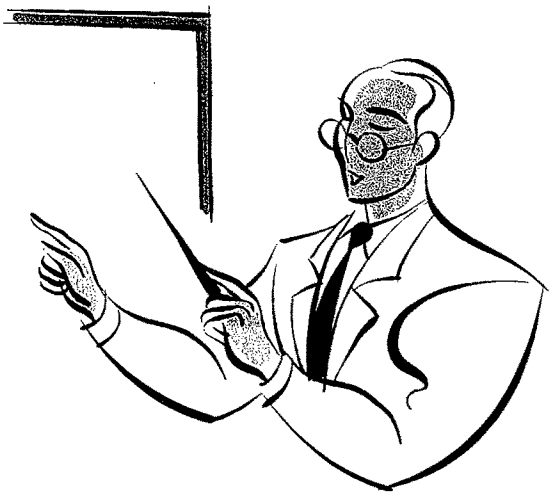
Dated: June, 2006

Contents

List of Tables

List of Figures

List of Plates



Introduction

Chapter-1

Introduction

Malnutrition has an adverse impact on the development of a nation because a workforce that is stunted both mentally and physically may have a reduced work capacity. Despite improvement in global food supplies, malnutrition and hunger remains one of the most devastating problems facing society. Much of the malnutrition has resulted due to the inability of the people to buy the costly energy rich food and to some extent lacking awareness of the essentials. Malnutrition caused by deficiencies in specific vitamins and minerals afflict some 40 percent of the world's population, especially women and children Nearly 90 percent of the pregnant women, 50 percent non-pregnant non lactating women, 30 percent men and more than 75 percent of adolescent and young children suffer from iron deficiency anaemia in our country (Kapil *et al.*,2004).

In many countries, malnutrition with significant health consequences results from deficiencies in zinc, vitamin C, folate, selenium and calcium, in addition to the three micronutrients i.e. vitamin A, iron and iodine to which so much attention is now being given. These micro nutrient deficiencies are mostly prevalent amidst poverty, environmental deprivation and social display. The survey conducted by NFHS-II in 1998-99 revealed that 47 percent

of the children under age 3 were underweight and 45.5 and 15.5 percent children were stunted and wasted, respectively in India.

Malnutrition Experts agree that the long term solution to malnutrition is the use of foods rich in the essential nutrients often lacking in people's diets. The search for novel high quality but inexpensive source of food has always remained a major concern of all agencies involved in providing adequate food and improving nutritional status of the population.

In this context, less familiar crops have a vital role to play as their economic value is beyond dispute. These crops generally are rich sources of vitamins, minerals and other nutrients and can provide a solution to the problem of malnutrition to a great extent, provided the masses are exposed to their nutritional and medicinal values through various extension strategies (Sankhla, 2005). Modern science research is proving that various parts of *Moringa oleifera* are one of the richest sources of such nutrients.

Moringa oleifera is the best known of 14 species of Moringa tree (Family Moringaceae). It is a fast growing, drought resistant tree native to sub-Himalayan tracks of Northern India, and is now growing world wide in the tropics and subtropics. Moringa goes by several names. In the Philippines, where the leaves of Moringa are cooked and fed to babies, it is called "mother's best friend" and "malunggay". Other names for it include "the horseradish tree" (Florida), "Nebeday" (Senegal) and "drumstick tree" (India). In Burmese, it is called dandalonbin; Sanskrit, *sobhanjana*; Hindi, *shajmah, shajna, segra*; Bengali, *sanjna*; Oriya, *munigha, sajina*; Punjabi, *sanjna*; Marathi, *sujna*; Tamil, *murungai* and Malayam, *moringa, muringa.*

Moringa oleifera grows well in almost all types of soils except stiff clays, but sandy loams are the best. It is strictly a tropical

plant and grows well in the plains. It is predominantly a crop of dry and arid tracts where it has been found to perform well and yield profitably. The tree can be propagated from seed or from cuttings. Cuttings which root very easily are usually preferred.

All the parts of this tree are considered useful for nutritional, medicinal and allied purposes. Two alkaloids moringine and moringinine are present in it, being responsible for many of the medicinal uses of the plant. In India, tradition maintains that the Moringa tree can cure 300 diseases and local herbalists make extensive use of Moringa products to treat a host of ailments. *Moringa oleifera* is known to have hepatoprotective, antitumor, hypoglycaemic, anti-spasmodic, anti-inflammatory, diuretic, antimicrobial activity, hypotensive, anti-ulcerogenic activity, radioprotective and hypocholesterolaemic effects.

Moringa oleifera can be an extremely valuable source of various nutrients for people of all ages. *Moringa oleifera* is full of essential disease preventing nutrients. Among the wide range of green leafy vegetables, Moringa is the richest source of beta-carotene, apart from providing other important micronutrients including β-carotene (vitamin A), vitamin C, folic acid and also calcium and potassium (Gopalan, 2000).. Small amounts of less than 10 gm of fresh Moringa leaves would meet the day's requirement of β-carotene of preschool children (Krishnaswamy, 2000).

The tree's leaves contain high amounts of vitamin A (four times more than carrots), vitamin C (seven times more than oranges), protein (twice that of milk), calcium (four times more than milk) and potassium (triple the amount in bananas).

Leaves are also good source of B-complex vitamins. The content of iron is very good. Thus, the leaves of Moringa are one of the best plant foods that can be found (Martin, 2000).

Moringa seeds contain oil that can be used for cooking. It is quite acceptable to taste and does not become rancid, a desired property in oil meant for consumption and industrial purposes. It is almost odourless and possesses a bland appreciable taste. The fatty acid composition is considered to be similar to that for olive oil. More recently, the oil has been shown to be particularly effective in the manufacture of soap with high washing efficiency.

The protective effect of fruits and vegetables against degenerative diseases such as cardiovascular disease, various cancers and neurological disease has generally been attributed to their antioxidant constituents including vitamin C and E, carotenoids, glutathine, flavonoids and phenolic acids as well as other unidentified compounds (Eberhardt *et al.*, 2000).

Moringa oleifera leaves have been reported to contain maximum amount of polyphenols followed by fenugreek and spinach leaves (Nambiar *et al.*, 2005). The flavanoids present in *Moringa oleifera* leaves include flavonols such as kaempferol and 3'-Ome quercetin, aflavone, acacetin, glycoflavone 4-Ome and vitexin. The phenolic acids identified include melilotic acid, p-coumaric acid and vanillic acid. Qurcetin has been reported to inhibit the growth of human prostate cancer cells and human breast cancer cells and has antiviral activity against several types of viruses. Besides leaves, fruits of *Moringa oleifera* i.e. pods, flowers and pods and seeds also are of valuable source of nutrients and have medicinal properties. Flowers are used as aphrodisiac and seed oil is used for treatment of rheumatism. The management of blood cholesterol level is of utmost importance in preventing

heart disease. The leaves of *Moringa oleifera* are used in the herbal medicine as a hypocholesterolaemic agent in obese patients.

The vegetable contribution of minerals and vitamins to human nutrition is, however, limited due to the presence of antinutritional factors which render some of the nutrients unavailable for human nutriture. Heat treatment on the course of cooking has been a reliable method of destroying antinutritional factors in many foods. Cooking for extended period of time at high temperatures and moisture has been effective in destroying most of the antinutritional factors in vegetables. However, these cooking conditions resulted in reduction of nutritional and organoleptic qualities of foods.

Besides, being a good source of various nutrients, seeds of *Moringa oleifera* have also been reported by various workers to be used in purification of water. The removal of organic and inorganic material from raw water is essential before it can be disinfected for human consumption. Studies have shown that this process not only removes solid contaminants but also greatly reduces amount of harmful bacteria (Mandloi *et al.*, 2004; Jeyanthi *et al.*, 2004).

Various value added food products by incorporating fresh as well as dried *Moringa oleifera* leaves, pods, flowers and seeds can be prepared by employing different methods of cooking i.e. steam cooking, sauting, frying. Such products may be acceptable and nutritionally superior to combat micronutrient deficiency. With this perspective in mind, the present study was planned to carry out work on *Moringa oleifera* with the following objectives:

i) To assess the nutrient composition of leaves, flowers, pods and seeds of *Moringa oleifera*

ii) To standardize and develop organoleptically acceptable food products from leaves, flowers, pods and seeds of *Moringa oleifera*

iii) To estimate the nutrient composition and shelf life of the most acceptable developed food products

iv) To study the hypocholesterolaemic effect of Moringa leaf powder.

Review of Literature

Chapter-2

Review of Literature

The Moringa or drumstick tree (*Moringa oleifera*) is an all-natural, inexpensive and accessible multi-vitamin. It is a multi-purpose tree of significant economic importance with several industrial and medicinal uses. Thus, it creates an interest to study its nutritional value and antinutritional factors of its various parts and various products prepared from them. The tree grows quickly, provides tasty and nutritious food, is both resilient and common in tropical areas and can even purify water, but has been overlooked by modern medicine. The literature regarding the nutritional evaluation of different parts of tree i.e. leaves, flowers, pods and seeds, development of different products, water purification, medicinal uses has been suitably reviewed under the following heads and subheads:

2.1 Nutritional analysis of pods, leaves, flowers and seeds of *Moringa oleifera*

2.2 Effect of processing and cooking on the nutrient composition of *Moringa oleifera*

2.3 Seed of *Moringa oleifera* for purification of water

2.4 Medicinal uses including hypocholesterolaemic effect of leaves of *Moringa oleifera*

2.1 Nutritional analysis of pods, leaves, flowers and seeds of *Moringa oleifera*

Duke (1983) evaluated the composition of different parts of multi-purpose drumstick tree. The study reported that per 100 g, the pod contained 86.9 g water, 2.5 g protein, 0.1 g fat, 8.5 g total carbohydrate, 4.8 g fibre, 2.0 g ash, 30 mg calcium, 110 mg phosphorus and 120 mg ascorbic acid. Leaves contained 75 g water, 6.7 g protein, 1.7 g fat, 14.3 g total carbohydrate, 0.9 g fibre, 2.3 g ash, 440 mg calcium and 70 mg phosphorus per 100 g. The seed kernel (70-74% of seed) contained 4.08 water, 38.4 g crude protein, 34.7% fatty oil, 3.5 g fibre and 3.2 g ash.

Chawla *et al.* (1988) analyzed *in vitro* availability of iron from six commonly consumed green leafy vegetables. The results of the study indicated that the *in vitro* available iron of different GLVs was neither a function of their total iron content nor related to their ascorbic acid content. However, there was some indication that oxalate might affect the availability of iron.

Awasthi and Tandon (1988) determined the nutritional attributes of 15 unconventional leafy vegetables (both cultivated and wild) consumed in different parts of rural India. The moisture, protein, fat, carbohydrate, fibre and ash contents were found to be 75.8, 6.60, 1.80, 12.60, 0.50 and 2.25 per cent, respectively. The calcium, phosphorus and iron contents were 430, 65 and 6.50 mg/100 g, respectively on fresh weight basis. The concentration of vitamin C was 226 mg/100 g.

Gupta *et al.* (1989) evaluated tender green leaves of conventional and non-conventional vegetables for nutritional and antinutritional factors, which depicted their quality. The concentration of crude protein (%), ether extractives (%), total ash (%), soluble ash (%), NDF (%), ADF (%) and hemicellulose (%) was

26.4, 6.5, 12.0, 11.4, 28.8, 13.9, 14.9, respectively. Oxalate and saponin contents were found to be 4.1 and 1.2 per cent, respectively.

Gopalan *et al.* (1989) determined the nutritional parameters of the drumstick pod. The macronutrient composition (per 100 g of edible protein) of drumstick pods was 2.5 g protein, fat 0.1 g, 2.0g minerals, 4.8 g fibre, 3.7 g carbohydrate, 30 mg calcium, 110 mg phosphorus and 0.18 mg iron.

Devadas *et al.* (1996) studied the consumption pattern of β-carotene rich foods in 500 households in Coimbatore, India and found that green leafy vegetables, particularly agathi, drumstick leaves and amaranthus were inexpensive sources of β-carotene. From data on availability, season and cost, a year calendar of β-carotene rich foods for the district was developed. The amount and cost of these foods to meet the preschool child and adult requirement of vitamin A was calculated and concluded that the year calendar could be used to select high carotene foods for increasing the β-carotene intake in the community.

Raval and Toliwal (1996) studied the drumstick seed for refining oil and to determine its chemical characteristics and fatty acid composition. The major fatty acid in the oil was oleic comprising 76.8% of the total fatty acids in the oil along with palmitic, stearic, linoleic, linolenic and arachidonic acids. At 10% level in the diet, the oil promoted a growth rate comparable to that of diet containing groundnut oil. No adverse effect of the drumstick seed oil was observed.

Freiberger *et al.* (1998) revealed that wild plants played an important role in the diet of the inhabitants of Niger. These plants tended to be drought-resistant and were gathered both in times of plenty as well as times of need. Used in everyday cooking, famine

foods might be an important source of nutrients. The goal of this study was to investigate the nutritional role of wild plants in the Nigerian diet. To this end, leaves of seven plants species i.e. *Ximenia americana, Amaranthus viridus, Corchorus tridens, Hibiscus sabdarifa, Maerua crassifolia, Moringa oleifera,* and *Leptadenia hastata* were analyzed for their mineral, amino acid and fatty acid contents. *Ximenia americana* contained large amounts of calcium. Large quantities of iron were present in *Amaranthus viridus.* All seven plants contained significant amounts of selenium and phosphorus. *Corchorus tridens* contained the maximum protein content (19-25% dry weight), and its composition compared favorably to the World Health Organization's standard for essential amino acids. *Moringa oleifera* contained 17% protein and compared favorably with the WHO standard. *Corchorus tridens* contained the largest amount of the two essential fatty acids i.e. linoleic and alpha-linolenic acids. These results reinforced the growing awareness that wild edible plant of the Western Sahel can contribute useful amounts of essential nutrients including amino acids, fatty acids and trace minerals to human diets.

Barminas *et al.* (1998) analysed the six non-conventional leafy vegetables consumed largely by the rural populace of Nigeria were analyzed for mineral composition. Mineral contents appeared to be dependent on the type of vegetables. *Amaranthus spinosus* and *Adansonia digitata* leaves contained the highest level of iron (38.4 mg/100 g and 30.6 mg/100 g, on dry weight basis, respectively). These values were low compared to those for common Nigerian vegetables but higher than those for other food sources. All the vegetables contained high levels of calcium compared to common vegetables, thus they could be a rich source of this mineral. Micronutrient content of the leaves varied appreciably. Zinc

content was the highest in *Moringa oleifera* followed by that in *Adansonia digitata* and Cassia tora leaves (25.5 mg/100 g, 22.4 mg/100 g and 20.9 mg/100 g on dry weight basis, respectively) while the manganese content was comparatively higher in *Colocasia esculenta*. The concentrations of the mineral elements in the vegetables per serving portion presented in these values indicated that the local vegetables could be valuable and important contributors in the diets of the rural and urban people of Nigeria. The mean daily intake of P, Mg, Ca, Fe, Cu and Zn were lower than their recommended dietary allowances (RDAs). However, the manganese daily intake was found not to differ significantly ($p<0.05$) from the RDA value.

Olveira *et al.* (1999) reported that *Moringa oleifera*, a multipurpose tree is cultivated for its use as a vegetable for spice, for cooking, cosmetic oil and as a medicinal plant. Owing to the use of its seeds as food and as a clarifying agent of turbid water, some nutritional and antinutritional characteristics were studied. The mature seeds contained 332.5 g crude protein, 412.0 g crude fat, 211.2 g carbohydrate and 44.3 g ash per kg dry matter. The essential amino acid profile compared with the FAO, WHO/UNU scoring pattern requirements for different age groups showed deficiency of lysine, threonine and valine. The content of methionine + cysteine (43.6 g kg^{-1} protein), however, was exceptionally higher and close to that of human milk, chicken egg and cow's milk. The seed extract agglutinated rabbit erythrocytes but did not show trypsin inhibitor and urease activities. Feeding rats with a diet containing the seed meal showed loss of appetite, impaired growth, lower NPU and enlargement of stomach, small intestine, cacecum + colon, liver, pancreas, kidneys, heart and lungs and atrophy of thymus and spleen in comparison with rats

fed on an egg white diet. The results indicated that consumption of *M. oleifera* raw mature seeds should be viewed with some caution until suitable processing methods are developed to abolish the yet unknown adverse factors.

Babu (2000) assessed a case study of *Moringa oleifera* which is a common tree in Malawi and one of the richest source of vitamin A and vitamin C compared to commonly consumed vegetable and *Moringa oleifera* is suggested as a potential solution to vitamin A deficiency.

Nambiar and Sheshadri (2001) conducted experiment on male albino rats (Charles Foster, n=40) were fed a synthetic diet deficient in vitamin A for 4 weeks. Six rats died during the depletion period. Of the 34 surviving, 5 rats were continued on the vitamin A deficient diet for 4 more weeks and 24 were repleted with vitamin A (4000 IU/kg diet) in the form of vitamin A acetate (group A, n=8), fresh drumstick leaves (group B, n=8) or dehydrated drumstick leaves (group C, n=8) for 4 weeks. The remaining 10 rats were continued on the vitamin A adequate diet for 4 (n=5) and 8 (n=5) weeks, respectively. A marked reduction in food intake, body weight, accompanied by clinical signs of vitamin A deficiency and a decline in serum vitamin A (29.2 to 19.1 μg/dl) and liver vitamin A (3.7 to 2.0 μg/dl) were seen at the end of 4 weeks of feeding a vitamin A deficient diet. On repletion significant improvements in clinical signs, food intake and body weights were noted in the three groups compared to the baseline (n=5) and at the end of 4 weeks of depletion. The gain in body weight was the highest for the group repleted with dehydrated drumstick leaves. Among the repleted groups, the serum vitamin A was the highest for group A (34.7 μg/dl) given synthetic vitamin A, compared to group B (25.8 μg/dL) and group C (28.2 μg/dl) given drumstick leaves. All these were

significantly higher than the serum vitamin A values seen at the end of 4 weeks of depletion (19.1 μg/dl). A significant improvement was also observed in the liver retinol levels on repletion for 4 weeks in the three groups, compared to the vitamin A depleted rats. These results imply that beta-carotene from drumstick leaves was effective in overcoming vitamin A deficiency although serum vitamin A levels remained some what lower compared to the group repleted serum vitamin A acetate. In terms of growth parameters, the fresh and dehydrated drumstick leaves were better than the synthetic vitamin A. It is, therefore, concluded that in the developing countries like India, sources of vitamin A such as drumstick leaves are valuable in overcoming the problem of vitamin A deficiency.

Stavros and John (2002) extracted oil from the dried seeds of the *Moringa oleifera* tree (variety of Malawi) with a mixture of chloroform/methanol (50:50). The induction period measurements demonstrated a great resistance to oxidative rancidity. After degumming, there was a reduction of 74% in induction periods. The gums produced were extracted with diethylether, n-butanol, and water, yielding four extractions: Fraction 1 (81.8% w/w), Fraction 2(0.04% w/w), Fraction 3 (0.05% w/w), and Fraction 4 (17.0% w/w). These fractions were tested for their protection of fresh sunflower oil against rancidity, at 50°C, using a UV accelerated method. The oxidation of the sunflower oil was measured using PV; absorbance E 1 cm 1% and malnondialdehyde concentration were measured by HPLC. The fraction that showed the highest antioxidant activity was further fractioned by HPLC, yielding seven fractions. Fraction HPLC 3 (present in a quantity of 330.8 and 29.11 ppm in gums and oil, respectively) showed the highest antioxidant activity. Its activity was also compared with

that of the commonly used antioxidants BHT and alpha-tocopherol on sunflower oil using the same methods. At the same level of addition (200 ppm), HPLC 3 showed higher antioxidant activity than BHT and alpha-tocopherol. The identification of HPLC 3 was done using 1 H NMR, 13 C, NMR, MS, melting point, and UV absorption spectroscopy and proved to be 3,5,7,3',4',5'-hexahydroxyflavon (myricetin).

Subadra *et al.* (2003) studied the *kanjero* and drumstick leaves' nutrient profile and potential for human consumption and found that drumstick leaves contained higher levels of beta carotene, ascorbic acid and carotenoid pigments than *kanjero* leaves.

Richter (2003) conducted a study to evaluate the suitability of freeze-dried Moringa leaf meal, *Moringa oleifera*, as an alternative source for Nil tilapia. Three experimental diets were formulated to contain *Moringa oleifera* leaf meal at levels of 10, 20 and 30% of the total dietary protein (Diet 2, 3 and 4, respectively) and one diet acting as a control (Diet 1) which included only fish meal and wheat meat as protein sources. All diets were iso nitrogenous (35% crude protein) and iso-energetic (20 kj/g). A 7-week feeding trial was carried out on triplicate groups of seven fish (9-11 g) in 45 litre aquaria connected to a recirculating system. The daily fish ration was calculated at 15 g feed per metabolic body weight (kg 0.8) per day. No feed related mortality was observed during the whole experimental period. Diets with higher inclusion level of moringa leaves (Diets 3 and 4) significantly depressed growth performance of the fish compared to Diets 1 and 2. The relatively high total phenolics, non-haemolytic saponin and phytic acid in Diets 3 and 4, respectively as well as NDF and ADF in the aforementioned diets may have contributed to the poorer growth

performance in these groups. These results suggest that up to 10% of dietary protein without significant reduction in growth.

Fifteen different kinds of leafy vegetables were investigated for their content of moisture, ash, protein, fat, carbohydrate and dietary fibre. Among all leafy vegetables, the lowest value for moisture content was observed in jute leaves (*Corchorus olitorius*) 76.52 g/100 g, whereas the highest was in pui leaves (*Basella alba*) 92.44 g/100 g. Ash content in leafy vegetables varied from 0.9 to 1.8 g/100 g. Among leafy vegetables, the lowest value for fibre content was found in bengal gram leaves (*Cicer arietinum*) 5.57 g/100 g and the highest in jute leaves 10.8 g/100 g. The results showed that protein, fat and carbohydrate contents in different leafy vegetables varied from 1.67 to 6.5, 0.09 to 1.5 and 3.3 to 12.57 g/100 g, respectively. The highest protein, fat and carbohydrate contents were observed in bengal gram 6.5 g/100 g, followed by drumstick (*Moringa oleifera*) 1.5 g/100 g and jute leaves 12.57 g/100 g. Among all the studied leafy vegetables, the highest amount of neutral detergent fibre (47.50 g/100 g) was estimated in radish leaves (*Raphanus sativus*) and the lowest (24.98 g $100g^{-1}$) in napa leaves (*Malva verticillata*). Cellulose content was the highest in jute leaves at 11.09 g/100 g, and the lowest in lettuce leaves at 4.05 g/100 g; hemicellulose content was the highest in radish leaves at 33.90 g/100 g and the lowest in napa leaves at 15.12 g/100 g. Lignin contents were the highest in jute leaves (6.01 g/100 g) and the lowest in radish leaves (0.87 g/100 g) (Islam *et al.*, 2004).

The nutritive value of stems, leaves and fruits of *Albizia lebbeck, Moringa oleifera, Pithecellobium dulce* and *Pongamia pinnata* was investigated. These were collected from two different sites: public park area (Bikaner) and junction area (Hanumangarh),

in Rajasthan, India. Crude protein (29.07%), nitrogen free extract (71.78%) and total carbohydrate (85.53%) were found maximum in *P. pinnata*. Crude fibre (29.30%) was higher in *A. lebbek*. Crude fat (16.84%) and total ash (14.57%) were higher in *M. oleifera*, while organic matter (95.38%) was found higher in *Pithecellobium dulce* (Kapoor *et al.*, 2004).

Anhwange *et al.* (2004) analysed the seeds of *Moringa oleifera* and *Detarium microcarpum* for nutritional and antinutritional contents. The chemical properties of the oils extracted from the seeds were also determined. The concentrations of the essential elements, potassium, calcium, magnesium, sodium, sulphur, phosphorus and iron were 77.4, 20.50, 1.19, 2.99, 3.75, 1.36 and 1.4, respectively for *M. oleifera* and 105.00, 23.00, 0.22, 2.36, 16.25, 1.25 and 3.12 for *D. microcarpum*, respectively. The amount of carbohydrate was the highest in *D. microcarpum* (42.20%) than in *M. oleifera* (9.11%). *Moringa oleifera* contained higher amount of proteins and lipids (40.19 and 41.58%, respectively) than *D. microcarpum* that contained 11.24 and 35.94% of protein and lipids, respectively. *Moringa oleifera* contained higher concentration of phytate (10.18 mg/100 g) and saponin (2.052%) than *D. microcarpum*. The iodine values of the oils in *M. oleifera* and *D. microcarpum* were 59.48 and 58.02, respectively. Saponification values were in the range of 179-220.66. The acid value, free fatty acid and peroxide values were low. The ester values of the oils ranged from 173.57-212.54.

Latha and Kapoor (2004) analysed gums from betelnut (*Areca catechu*), drumstick (*Moringa oleifera*) and acacia (*Acacia arabica*) [*A. nilotica*] for various chemical constituents. Analysis revealed high level of total ash, crude fibre, calcium, iron, moderate levels of phosphorus, low levels of protein and lipid contents. Gums

contained high levels of certain antinutrients, namely polyphenols and saponins and different fibres (acid detergent fibre, hemicellulose and pectin). A glycaemic response study was conducted with 24 non-insulin dependent diabetic subjects aged 40-48 years (Anantapur, India). Subjects were randomly assigned to 4 groups (A, B, C, D) and glucose tolerance test was performed on each subject. After an overnight fast, subjects in the control group (A) were given an oral dose of 50 g glucose in 100 ml water. Subjects in groups B, C and D received 15 g freshly powdered gums of betelnut, drumstick and acacia, respectively, along with 50 g glucose in 100 ml water. Blood samples were taken from the subjects at 0, 30, 60 and 120 minutes after administration of the samples and blood glucose was estimated. The inclusion of gums significantly ($P<0.01$) lowered the post prandial rise in plasma glucose levels at different intervals (30, 60, 90 and 120 minutes). The mean peak values of glucose were observed at 30 minutes in groups D and C and at 60 minutes for group B. The levels decreased significantly ($P<0.01$) after the peak in all the groups. The gums showed low values of glycaemic index.

Interprovenance variation was examined in the composition of *Moringa oleifera* oilseeds from Pakistan. The hexane-extracted oil content of *M. oleifera* seeds harvested in the vicinity of the University of Agriculture, Faisalabad (Punjab, Pakistan), Bahauddin Zakariya University (Multan, Pakistan), and the University of Sindh, Jamshoro (Sindh, Pakistan), ranged from 33.23 to 40.90%. Protein, fibre, moisture and ash contents were found to be 28.52-34.00, 6.52-7.50, 5.90-7.00 and 6.52-7.50%, respectively. The physical and chemical parameters of the extracted *M. oleifera* oils were as follows: iodine value, 67.20-71.00; refractive index (40°C), 1.4570-1.4637; density (24°C),

0.9012-0.9052 mg/ml; saponification value, 177.29-184.10; unsaponifiable matter, 0.60-0.83%; colour (1-in. cell), 1.00-1.50R + 20.00-30.00Y; smoke point, 198-202°C; and acidity (% as oleic acid), 0.50-0.74. Tocopherols (alpha, gamma, and delta) accounted for 114.50-140.42, 58.05-86.70 and 54.20-75.16 mg/kg, respectively, of the oils. The induction pericds (Rancimat, 20 l/h, 120° C) of the crude oils were 9.64-10.66 h and were reduced to 8.29-9.10 h after degumming. Specific extinctions at 232 and 270 nm were 1.80-2.50 and 0.54-1.00, respectively. The major sterol fractions of the oils were campesterol (14.13-17.00%), stigmasterol (15.88-19.00%), beta-sitosterol (45.30-53.20%), and delta 5-avensterol (8.84, 11.05%). The Moringa oils were found to contain high levels of oleic acid (up to 76.66%), followed by palmitic, stearic, behenic, and arachidic acids up to levels of 6.54, 6.00, 7.00 and 4.00%, respectively. Most of the parameters of *M. oleifera* oils indigenous to different agroclimatic regions of Pakistan were comparable to those of typical Moringa seed oils reported in the literature. The results of the present analytical study, compared with those for different vegetable oils, showed *M. oleifera* to be a potentially valuable oilseed crop (Anwar *et al.*, 2005).

Sankhala *et al.* (2005) studied proximate composition, iron, calcium, β-carotene, vitamin C and oxalic acid contents of drumstick leaves (*Moringa oleifera*). The results showed that drumstick leaves contained 75.8% moisture, 6.3% protein, 1.4% fat, 0.9% crude fibre, 13.3% carbohydrates and 91 Kcal/100 g energy. The micronutrient composition i.e. iron and calcium content was 0.8 and 430 mg/100 g, respectively. The β-carotene and ascorbic acid contents were found to be 6700 µg and 190 mg%, respectively. Oxalic acid, a non-nutrient factor was 101.2 mg%.

Nambiar *et al.* (2005) analyzed polyphenols in drumstick (*Moringa oleifera*) leaves, spinach (*Spinacea oleracea*) leaves and fenugreek (*Trigonella foenum-graecum*) leaves and were characterized using paper chromatography. Maximum polyphenols were identified in drumstick leaves, followed by fenugreek and spinach leaves. Results indicated that these green leafy vegetables are a repository of several antioxidants necessary for human health.

Aslam *et al.* (2005) studied the mineral composition of leaves and pods of *Moringa oleifera* from different agro-climatic regions of Punjab. Samples of leaves and pods were wet digested and analysed for various minerals by using Atomic Absorption Spectrophotometer (AAS) and flame photometer; the contents of K, Ca, Mg, Na in the leaves and pods of *Moringa oleifera* were found to be 19732-24397, 1839-2097; 18950-26349, 1292-1837; 98.2-109, 93.9-103.9 and 1635-2721, 1032-2105 mg kg^{-1}, respectively. The concentration of Fe, Cu, Mn and Zn was found to be 205-573, 155.2-435.9; 7.3-11.2, 20.9-32.1; 76.9-112.8, 40.2-72 and 20.9-34.1, 15.3-29 mg kg^{-1}, respectively. The level of P in the samples of leaves and pods was 1180-1450 and 1860-2125 mg kg^{-1}, respectively. The contents of different minerals in the leaves and pods of *M. oleifera* significantly varied from region to region. The results of present analysis revealed that pods and leaves of *M. oleifera* indigenous to different agro-climatic regions of Punjab contained a considerably high amount of Ca, Mg, K, Mn, P, Zn, Na, Cu and Fe and might be used as a viable supplement of dietary minerals.

2.2 Effect of processing and cooking on the nutrient composition of *Moringa oleifera*

Sensory evaluation showed that the flat noodle production with 5 per cent level of supplementation had the most comparable quality to the control on unsupplemented one, in terms of odour, texture and flavour. The 5 per cent malunggay leaves substituted pasta (MSP) also got the highest acceptability score among the substituted products followed by the flat noodle substituted at 10 per cent level. Addition of sauce, sauteing and cooking in soup of flat noodles generally improved the sensory attributes and acceptability of all the treatments. The flat noodles were preferred to be (1) light green in colour, (2) odourless, (3) with a characteristic bland or flat flavour and (4) chewy and firm in texture. The test revealed that each level of malunggay leaves substitution affected the odour, colour, texture, flavour, overall quality and acceptability of the flat noodle products. Chemical analysis indicated an increase in nutrients of flat noodles, among others. Calcium, iron and β-carotene content increased as the level of substitution was increased from 5 per cent to 15 per cent. Crude protein level was likewise significantly increased from 10.33 per cent for the control to 11.18 per cent for the flat noodle at 15 per cent level of substitution. Calcium was almost doubled (15.0 mg) in 5 per cent (M.S.P.) and a significant increase of about 10 mg at 10 per cent and 15 per cent levels of substitution, respectively was observed. The significant increase in iron content of flat noodles was observed at 10 per cent and 15 per cent level of substitution. The beta carotene increased significantly as malunggay leaves substitution increased. Nutritional contribution

of 5 per cent and 10 per cent substituted flat noodles for protein, iron and vitamin A was found to be significant (Abilgos, 1996).

Seshadri *et al.* (1997) studied the retention and storage stability of β-carotene in dehydrated drumstick leaves. The samples were analysed for total carotene, β-carotene and ascorbic acid in the fresh form, immediately after drying and after 30, 60 and 90 days of storage. Fresh leaves contained 27.1 mg of total carotene, 17.4 mg of β-carotene and 143.6 mg of ascorbic acid per 100 g. Sulfiting in addition to blanching was more effective in the retention of β-carotene immediately after dehydration (72 vs. 59%) and at the end of one month of storage (64 vs. 51%) but not at the end of 90 days of storage (53 vs. 47%) and concluded that dehydrated drumstick leaves have the potential to serve as a valuable source of β-carotene in diets of the population in India and other developing countries.

Nambiar *et al.* (2003) conducted a pilot study to assess the feasibility and acceptability of introducing dehydrated drumstick leaves, (DDL) (*Moringa oleifera*), as a source of vitamin A, into the salty recipes provided by the supplementary food (SF) component of the Integrated Child Development Scheme (ICDS) along with nutrition communication (NC). An integrated approach was adapted in this study which included comprehensive training sessions for the staff of the ICDS and Non-government organization (NGO) involved in the SF preparations. Prior to the acceptability trials, data were elicited on the socio-economic profile and knowledge about vitamin A, from 60 children of 1-5 years of age attending two anganwadi centres of the ICDS. From these, 40 children attending one anganwadi were supplemented with pre-tested DDL incorporated recipes (5-7 g DD/100 g product) along

with NC for one month. Spot observations and organoleptic evaluation results indicated high compliance of the DDL-recipes by the children. The results also indicated that the recipes were highly acceptable to the ICDS authorities as well as the NGO staff. The pilot study indicated that integration of NC along with the introduction of unconventional DDL, into the ICDS-SF, was feasible and can be endeavoured for a longer duration in the existing national programmes.

Nambiar *et al.* (2003) studied nutritional and sensory qualities of selected recipes given as supplementary food component under the Integrated Child Development Scheme (ICDS) after incorporation of β-carotene rich, blanched and shade dried drumstick leaves. Fresh, pretreated and shade dried drumstick leaves stored for 6 months at room temperature were analysed for total carotene, β-carotene and ascorbic acid at 0, 30, 60, 150 and 180 days of dehydration. Based on organoleptic evaluation of composite and hedonic scores, 6 g of dehydrated drumstick leaves were incorporated in supplementary foods (*channa, moong, poha* and *dhokla*) and nutritional evaluation was carried out. Fresh drumstick leaves contained 1656.36 µg/g dry weight of total carotene and 1090.61 µg/g dry weight of β-carotene and 201 mg/100 g fresh weight of ascorbic acid. Per cent retention of total carotene and β-carotene on 0 day was 22.7% and 25.5%, which decreased to 9% and 4.33%, respectively after 6 months of storage. On dehydration and rehydration 90% ascorbic acid was lost. Analysis of total carotene, β-carotene and ascorbic acid revealed highest retention in *dhokla* (steamed cooked) (73.52%, 69.69%, 87.89%, respectively) and lowest in *poha* (sauted) (49.39%, 54.25%, 68.55%, respectively). All 4 recipes could meet 1/3rd

Recommended Dietary Allowances of β-carotene for preschool children and 25-30% Recommended Dietary Allowances of β-carotene for pregnant and lactation mothers. These results indicate that in spite of high losses, there is enough β-carotene retained in the dry powder of drumstick leaves, which could help in eradication of several micronutrient deficiencies.

Gidami *et al.* (2004) analysed the nutrient composition and non-nutrient contents of raw and cooked *M. oleifera* leaves and immature pods. Cooking caused significant reductions ($p<0.05$) in the contents of crude protein, crude fibre, ether extracts, ash, ascorbic acid, and beta-carotene in the leaves and pods. Significant reductions ($p<0.05$) also were observed in Ca, Mg, Zn, P, Na, Cu and K except for Fe in the leaves after cooking. Similar reductions were also observed in Mg, Fe, Cu, P, Na and K except in Ca and Zn in the cooked pods. No significant reductions ($p<0.05$) were observed in total reducing sugars, phytates and trypsin inhibitor substances in both leaves and pods after cooking. On the other hand, cooking increased protein digestibility by 20.7 and 7.8% in leaves and pods, respectively. A similar increase also was observed in total carbohydrates after cooking leaves and pods. Overall, this study has established that young leaves and immature pods of *M. oleifera* are a good source of both micro- and macronutrients and have low levels of non-nutrients. Although cooking by boiling in water reduced the nutrients to some extent, considerable amounts are retained and as such servings of young leaves and immature pods, as a vegetable relish, could greatly improve the nutritional status of those at risk of malnutrition.

Bezerra *et al.* (2004) evaluated the quality of drumstick (*Moringa oleifera*) seeds subjected to ambient or cold storage for 0, 6, 12 or 24 months. Water content, germination percentage, root

length, seedling dry matter and electrical conductivity were evaluated. After 12 months of storage in plastic bottles under ambient conditions, the seeds lost their viability. Seeds subjected to cold storage for 24 months showed reduced quality.

The kinetics of ascorbic acid degradation in drumstick (*Moringa oleifera*) leaves as well as in pure ascorbic acid solutions at the initial concentrations present in drumstick leaves over a temperature range of 50-120°C (isothermal temperature process) has been studied. The degradation kinetics of ascorbic acid was also evaluated in normal open-pan cooking, pressure-cooking and a newly developed and patented fuel-efficient eco cooker (non-isothermal heating process). The ascorbic acid degradation followed first-order reaction kinetics where the rate constant increased with an increase in the temperature. The temperature dependence of degradation was adequately modelled by the Arrhenius equation. A mathematical model was developed using the isothermal kinetic parameters obtained to predict the losses of ascorbic acid from the time-temperature data of the non-isothermal heating/processing method. The results obtained indicate the ascorbic acid degradation is of similar order of magnitude in all the methods of cooking (Bineesh *et al.*, 2005).

Natural oxidants have gained considerable interest in recent years for their role in preventing the auto oxidation of fats, oils and fat containing food products. In the present study, three plant foods viz, amla (*Emblica officianalis*), drumstick leaves (*Moringa oleifera*) and raisins (*Vitis vinifera*) were used as sources of natural antioxidants. All the three extracts exhibited a high percentage of antioxidant activity which was evaluated using beta-carotene-lenoleic acid *in vitro* system, compared to synthetic antioxidants. Biscuits prepared by addition of natural extracts were subjected to

sensory studies and chemical analysis. Biscuits treated with natural antioxidants, extracted from raisins (B4) and drumstick leaves (B5) received higher ($P<0.05$) panel scores during storage period of 6 weeks, than control (B1), butylated hydroxyl anisole (BHA) (B2) and amla (B3) extract incorporated biscuits. Addition of plant extracts from the three plant foods gave an excellent antioxidant effect on the biscuit compared with the effect of BHA, as the % increase in both peroxide and acid values after 6 weeks were lower than that of the control and BHA treated samples. Extracts from drumstick leaves and amla were more effective in controlling lipid oxidation during storage (Reddy *et al.*, 2005).

2.3 Seeds of *Moringa oleifera* for purification of water

The pilot and full scale trials in Malawi have demonstrated the effectiveness of *M. oleifera* seed coagulant for the clarification of highly turbid river water. Inlet turbidities of 270-380 NTV were consistently reduced to below 4 NTV in the finished water. *M. oleifera* seed contain 40% by weight of oil, with the press cake remaining following oil extraction containing the active constituents affecting coagulation. Confirmation of the high market value of the oil make the economic case for adoption of the press cake as a coagulant overwhelming (Sutherland *et al.*, 1994).

Olayemi and Alabi (1994) investigated the efficacy of *Moringa oleifera* seeds paste for water purification. Chemical analysis found the seed to contain 34.1% protein, 15% carbohydrates and 15.5% lipids. Phytochemical tests and spectral studies led to the elucidation of a steroidal glycoside strophantidin as a bioactive agent in the seed. Comparative studies with alum showed that the seed paste was effective in the clarification and sedimentation of inorganic and organic matter in raw water. It reduced the total microbial and coliform counts by 55 and 65 per cent, respectively,

after 24 hours whereas alum achieved 65 and 83 per cent reduction under similar conditions.

Preliminary investigations into the possible use of *Moringa oleifera* seed suspension for the softening of hard water were presented. Four water sources i.e. synthetic water (distilled water spiked with calcium chloride), naturally hard surface water and groundwater from two tube wells at different locations were used for the study. Modified laboratory jar tests procedures for coagulation studies were used for the experimental runs. Water hardness from the sources varied from 300 up to 1000 mg/litre as $CaCO_3$. The mechanism for softening was found to be due to absorption with the absorption isotherm approximating to the bangnuir type, and conversion of soluble hardness causing ions to insoluble products by precipitation reactions. Removal efficiency was found to increase with increasing dosage of *Moringa oleifera.* Higher dosages were required to achieve equivalent residual hardness for water samples with the same initial hardness but higher number of hardness causing species in the water. Hardness removal was found to be independent of pH of the raw water (Muyibi and Evison, 1995).

Folkard *et al.* (1995) reported that the *Moringa oleifera* seed kernel contained significant quantities of a series of low molecular weight, water-soluble proteins which in solution, carried an overall positive charge. The proteins were considered to act similarly to synthetic, positively changed polymer coagulants. When added to raw water, the proteins bound to the predominantly negatively changed particular that made raw waters turbid (silt, clay, bacteria etc.). Under proper agitation, these bound particulates then grew in size to form the flocs which might be left to settle by gravity or be removed by filtration.

Guevara *et al.* (1996) reported that seeds of *M. oleifera* (collected from Bataan and Naga, Philippines) were extracted with distilled ethanol and concentrated under reduced pressure at 40°C. The resulting extract was partitioned between hexane, ethylacetate, butanol and water. The solvent fractions were concentrated under reduced pressure. The crude ethanol extracts (3 mg/kg) of dried and green seeds inhibited carrageenan-induced inflammation of the hind paw of mice by 85 and 77%, respectively. The hexane, butanol and water fractions of the crude ethanol extract of dried seeds inhibited inflammation by 77, 34 and 34%, respectively. The ethylacetate fraction caused a 267% increase in inflammation and exhibited toxicity, mice died after oral administration of the fraction. The crude ethanol extract of dried seeds also inhibited the formation of Epstein-Barr Virus-early antigen (EBV-EA) induced by 12-O-tetradecanoylphorbol-13-acetate. The extract (100 µg/ml) inhibited EBV-EA formation by 100% suggesting its antitumour-promoting activity.

The seeds of *Moringa oleifera* were tested as clearing and sedimentation agents in household water in Thaung Gyi Lav village (Mayamar) with 110 households. Questionnaires were completed for each household and follow-up visits were carried out to ascertain the hypothetical acceptability (attitude), initial acceptability (behaviour) and experimental acceptability. It was observed that 78.9 per cent of the people accepted to use *Moringa oleifera* seeds if these were easily available. For continuous use of *Moringa oleifera* seeds, 47.3 per cent wanted to use, 44.7 per cent could not decide and only three households (2.7 per cent) did not want to use these. It was observed that the taste and pH of water did not change after treatment with *Moringa oleifera* seeds. There was no complaint about the treated water. This study highlights

the acceptance to use *Moringa oleifera* seeds for the sedimentation of turbid water (Nyein *et al.*, 1997).

Samples of municipal and industrial wastewaters were treated by coagulation-floculation and sedimentation, using a crude water extract of dry *Moringa oleifera* seeds as a primary coagulant. The quality of the treated wastewater was analyzed and compared to that of the wastewater treated with alum. Experiments were conducted at various dosages of the crude 5% (wt/v) water extract of dry shelled and non-shelled *Moringa oleifera* seeds, using jar-test equipment. Parameters of quality of the wastewaters were measured before and after the treatment to evaluate the removal efficiency on the major pollutants of concern in wastewater treatment, such as suspended solids, chemical treatment to evaluate the removal efficiency on the major pollutants of concern in wastewater treatment, such as suspended solids, chemical oxygen demand (COD), nutrients (phosphorus and nitrogen), microorganisms and heavy metals. Results showed that *Moringa oleifera* seeds were efficient as a primary coagulant in wastewater treatment for removal of suspended solids and microorganisms, and also removal of some metals. Nutrients and COD were not successfully removed. COD and nutrients were somehow increased by coagulation using *Moringa oleifera* seeds. Compared to alum, *Moringa oleifera* seeds produced 4 to 6 times less sludge volume. Alum was found to be quite effective in phosphorus removal. The increase in COD and nutrients and nutrients observed in the case of *Moringa oleifera* seeds might be avoided by using purified proteins instead of the crude water extract (Ndabigengesere and Narasiah, 1998).

Seven plant species i.e., *Adhatoda vasica, Andrographis paniculata, Azadirachta indica, Lawsonia inermis, Moringa oleifera,*

Ocimum sanctum and *Trigonella foenum-graecum* have been screened out systematically for their effectiveness in inhibition of bacterial population of raw water (*in vitro*) at variable pH levels i.e., 6.0, 6.5,7.0, 7.5 and 8.0. Maximum percentage inhibition was observed in three plant species at 6.5 pH level while in four plant species at 7.0 pH level. Among all the studied plants, maximum inhibition was recorded in case of *L. inermis* (93.97%) which was followed by *M. oleifera* (89.72%) and *A. vasica* (81.75%). In order of maximum to minimum inhibition, individual plant species depicted their effectiveness in the following manner: *L. inermis* > *M. oleifera* > *A. vasica* > *A. indica* > *O. sanctum* > *A. paniculata* > *T. foenum-graecum*. Besides, most probable number (MPN), coliform and *E. coli* were also inhibited maximum at 7.0 pH level by the extract of *L. inermis* which was followed by *A. vasica* and *M. oleifera* at 6.5 pH level. Hence, these plant species may be used for elimination of bacterial contaminants of raw drinking water obtained directly from the sources to reduce the occurrence of different water borne diseases (Kumar and Krishna, 1999).

Jeyanthi *et al.* (2004) conducted a study to assess the quality of river Bhavani water at Bhadrakaliammankoli near Mettupalayam, Tamil Nadu, India, before and after treatment with selected medicinal plants, namely, ginger, fenugreek, *Cuminum cyminum*, vetiver (*Vetiveria indica*), drumstick seeds (*Moringa oleifera*) and the thakottai (seeds of *Strychnos potatorum*). The treatments showed different effects on the physiochemical and bacteriological characteristics of water, pH and electrical conductivity and total suspended solids and total hardness. Cumin seed treatment showed the maximum reduction (1.25 mg/l) on biochemical oxygen demand level to be within the desirable limit. The water samples treated with vetiver, drumstick seed

powder and seeds of *Strychnos potatorum* recorded chemical oxygen demand (COD) levels of 180, 72 and 176 mg/l, respectively, which were lower than the COD level of the untreated Bhavani river water. There were also variations in the acidity and alkalinity levels among the treatments. Treatments with drumstick seed powder and *S. potatorum* reduced the bacterial counts significantly to 100 and 80 x 103 cells/ml, respectively.

The 250 g of shade-dried grinded seed powder of drumstick, *Moringa oleifera* was extracted in petroleum ether through hot percolation process (Soxhlet apparatus), gave 21 g of pure active principles. The 5.0 ml of 10% aqueous solution of pure extract, when mixed with 1000 ml of contaminated water, were able to bind individual particle of the contaminants within 72 h, forming what is known as floc. Bacteria and viruses were enmeshed in the floc and after 7 days of slow-mixing all the undesirable suspended particles settled down. Ten specimens of contaminated water from different places taken during the treatment showed a very low percentage of heavy metals and other water pollutant. The water within 7 days became as clear as fresh water. This storage, cheap, indigenous natural product can easily be used for removal of water pollutants (Johri and Johri, 2004).

Mandloi *et al.* (2004) conducted a study on *Moringa oleifera* seed, maize (*Zea mays*) and chitosan in direct filtration of Bilaoli lake water and evaluated for their efficiency in removing turbidity and microorganisms from water. The experiments with these natural coagulants gave filtered water turbidity less than or almost equal to 1 NTU and thereby met the turbidity criteria for drinking water as per WHO guidelines. Bilaoli lake water had low ionic strength and low turbidity which represents one of the most difficult raw waters to treat, but natural coagulants in direct

filtration achieved good filtrate quality. The head loss development across the filter bed with chitosan was more than that of alum, while with maize it was comparable to that of alum. With *M. oleifera* seeds the head loss was much less in comparison to alum. The average most probable number (MPN) reductions obtained with *M. oleifera* seeds, maize and chitosan were 97.35%, 95.4% and 87.1%, respectively, whereas, with alum it was only 7.7%.

Sajidu *et al.* (2005) conducted a study to investigate the potential of *Moringa oleifera* polyelectrolytes for treatment of wastewater contaminated with heavy metals. The potential of *Moringa oleifera* whole seed kernels and ram press cake, in removing lead, iron and cadmium ions from synthetic contaminated water was investigated at initial metal ion concentrations of 5.00 and 7.00 ppm by means of jar tests. Metal ion removal was observed ranging from 70.86 ± 2.22% to 89.40 ± 0.00% for lead, 66.33 ± 3.38% to 92.14 ± 0.00% for iron and 44.95 ± 3.95% to 47.73 ± 6.38% for cadmium. Further experiments on optimization of the reaction conditions such as pH and coagulant dosage, method of extraction of polyelectrolytes and elucidation of the nature of interactions are being carried out.

2.4 Medicinal uses including hypocholesterolaemic effect of leaves *Moringa oleifera*

Moringa oleifera Lam (Moringaceae), commonly known as "Drumstick" is used in Indian folk medicine for the treatment of various illnesses. Geervani and Devi (1981) studied the effect of protein and fat on the utilization of carotene derived from drumstick (*Moringa oleifera*) leaves using weanling albino rats. Rats fed on 5 per cent protein diet had less vitamin A and higher carotene level in serum and liver and vice versa on 10 per cent

protein. Rats fed on animal protein diet stored more vitamin A in the liver. Fat level influenced only liver stores of vitamin A but not serum levels. Rats that received drumstick leaves stored considerably less amount of vitamin A than rats received pure β-carotene or vitamin A acetate.

The multistage model of carcinogenesis is represented as follows: initiation, promotion, conversion, progression. The skin promotion test was used to study the effect of expressed juices from some medicinal plants on the promotion stage of carcinogenesis. A combination of dimethylbenzanthracene (DMBA) as the initiator and croton oil as the promoter led to a high incidence of skin tumours in experimental mice after 10 weeks. Expressed juices from lagundi (*Vitex negundo*) leaves, ampalaya (*Momordica charantia*) fruits, tanglad (*Cymbopogon citratus*) roots and malunggay (*Moringa oleifera*) leaves reduced appreciably the incidence of skin tumours. However, expressed juice from tanglad leaves did not reduce the incidence of skin tumours initiated by dimethylbenzanthracene and promoted by croton oil (Sylianco, 1995).

Hypotensive activity of the ethanolic and aqueous extracts of *Moringa oleifera* whole pods and their parts, namely, coat, pulp, and seed was investigated. The activity of the ethanolic extract of both the pods and the seeds was equivalent at the dose of 30 mg/kg. The ethyl acetate phase of the ethanolic extract of pods was found to be the most potent fraction at the same dose. Its bioassay-directed fractionation led to the isolation of thiocarbamate and isothiocyanate glycosides which were also the hypotensive principles of the pods as observed in case of Moringa leaves. Two new compounds i.e. O-(2'-hydroxy-3'(2"-heptenyloxy))-propyl undecanoate and O-ethyl-4-(alpha-L-rhamnosyloxy)-benzyl)

carbamate along with the known substances methyl p-hydroxybenzoate and beta-sitosterol have also been isolated in the present studies. The latter two compounds and p-hydroxybenzaldehyde showed promising hypotensive activity. Structures of all these compounds have been deduced by spectroscopy and chemical reactions (Shaheen *et al.*, 1998).

Malaya *et al.* (1999) tested the methanolic extract of roots of *Moringa oleifera* for possible pharmacological effects in mice. The extract potentiated the sleeping time induced by pentobarbitone, sodium diazepam and meprobamate showed analgesic properties.

Ghasi *et al.* (2000) analysed the leaves of *Moringa oleifera* as a hypocholesterolaemic agent in obese patients. The scientific basis for their use in hypocholesterolaemia was therefore, examined. Administration of the crude leaf extract of *Moringa oleifera* (1 mg/gm) to rats with a high fat diet for 30 days decreased the high fat diet induced increases in serum, liver and kidney cholesterol levels by 14.35%, 6.40% and 11.09%, respectively, thus, having definite hypocholesterolaemic activity.

Dangi *et al.* (2002) stated that *Moringa oleifera* leaves have traditionally been used in Ayurvedic medicine for their antihypertensive activity. Preliminary studies in our laboratory indicated that a water extract of leaves of this tree is efficacious in reducing the chronotropic and inotropic effects on the isolated frog heart. The alkaloids obtained by the fractionation of the water extract of the leaves of *M. oleifera*, converted into their salt form, were tested for their activity on the isolated frog heart. The total alkaloidal salts were found to have a negative inotropic effect on the frog heart. This activity was further characterized by testing it on the isolated guinea pig ileum.

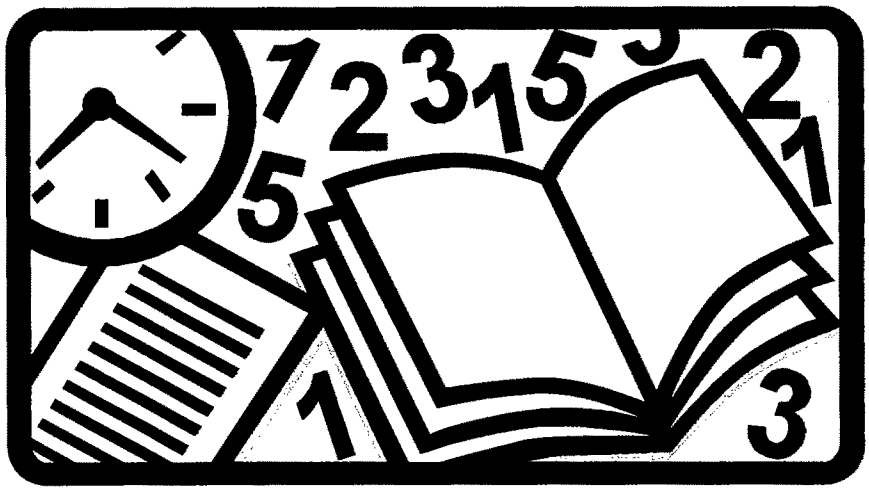
Pari (2002) evaluated the hepatoprotective effect of an ethanolic extract of *M. oleifera* leaves on liver damage induced by antitubercular drugs such as isoniazid (INH), rifampicin (RMP), and pyrazinamide (PZA) in rats. Oral administration of the extract showed a significant protective action made evident by its effect on the levels of gludtamic oxaloacetic transaminase (aspartate aminotransferase), glutamic pyruvic transaminase (alanine aminotransferase), alkaline phosphatase, and bilirubin in the serum; lipids, and lipid peroxidation levels in liver. This observation was supplemented by histopathological examination of liver sections. The results of this study showed that treatment with *M. oleifera* extracts or silymarin (as a reference) appeared to enhance the recovery from hepatic damage induced by antitubercular drugs.

Prajapati *et al.* (2003) conducted experiment with freshly lopped drumstick leaves fed *ad libitum* to goats and sheep for 44 days as sole feed, and thereafter, a 6 day metabolic trial was conducted (India). Drumstick leaves when fed *ad libitum*, not only maintained the body weight of growing goats and sheep, but also supported a moderate gain in liveweight. The digestibility of dry matter, crude protein, crude fibre and ether extract was significantly higher in sheep than in goats.

Moringa oleifera belongs to family Moringaceae. The roots are bitter, acrid, thermogenic, digestive, carminative, anthelmintics constipating, anodyne, anti-inflammatory, emmenagogue, sudrific diuretic, ophthalmic, rubefacient, expectorant, haematinic, antilithic, alxiphosmic stimulant and vesicant. They are useful in vitiated conditions of vata and kapha, dyspepsia, anorexia, verminosis, diarrhoea, colic flatulence, otalgia, paralysis, inflammations, amenorrhoea, dysmenorrhoea, fever, strangury, verical and renal calculi, ascites, ophthalmopathy, cough, asthma,

bronchitis, pectoral diseases, splenomegaly, epilepsy, hysteria, cardiopathy, abscess and pharyngodynia. The bark is acrid, bitter, thermogenic, abortifacient, antifungal and cardiac and circulatory stimulant. It is useful in ascites, vitiated conditions of vata and kapha and ringworm. The leaves are anti-inflammatory, anodyne, anthelmintic, ophthalmic and rich in vitamin A and C. They are useful in scurvy, vitiated conditions of kapha and vata, wound tumours, inflammations and helmintharis. The seeds are acrid, bitter, anodyne, anti-inflammatory, purgative, anti-pyritic and ophthalmic. They are useful in neuralgia, inflammations, intermittent fevers and ophthalmopathy (Prajapati, 2003).

Rathi *et al.* (2004) studied the effect of aqueous extract of the dried pulp and seeds of *Moringa oleifera* (collected from Belgaum, Karnataka, India) on wound healing in albino rats. The aqueous extract was studied at dose level of 300 mg/kg body weight using resutured incision, excision and dead space wound models in rats. Significant increase in would closure rate, skin-breaking strength, granuloma breaking strength, hydroxyproline content, granuloma dry weight and decrease in scar area was observed. The prohealing actions seem to be due to increased collagen deposition as well as better alignment and maturation. From the results obtained, it may be concluded that the aqueous extract of *Moringa oleifera* has significant wound healing property. Also it can be concluded that these prohealing effects may be due to their high content of crude proteins, zinc and some anti-microbial component.



Methodology

Chapter-3

Materials and Methods

The present investigation entitled, "Development and Nutrient Composition of Value Added Products from Drumstick (*Moringa oleifera*) was conducted in the Department of Foods and Nutrition, I.C. College of Home Science, Chaudhary Charan Singh Haryana Agricultural University, Hisar.

This chapter delineates information pertaining to the research design and methodological steps used for the present investigation. The research procedures have been distinctly described under the following heads and sub-heads:

3.1 Procurement of material

3.2 Standardization of recipes

3.3 Organoleptic evaluation of products

3.4 Selection of products

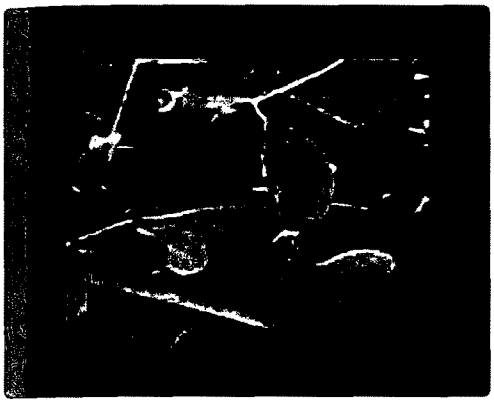
3.5 Nutritional evaluation of raw samples of pods, leaves, flowers and seeds of *Moringa oleifera* as well as its selected products

3.6 Shelf life of selected products

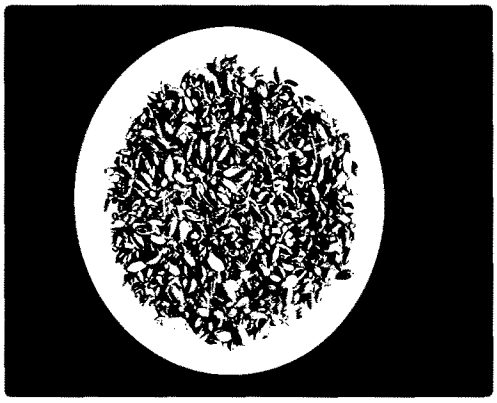
3.7 Hypocholesterolaemic effect of *Moringa oleifera* leaves

3.8 Seeds of *Moringa oleifera* for purification of water

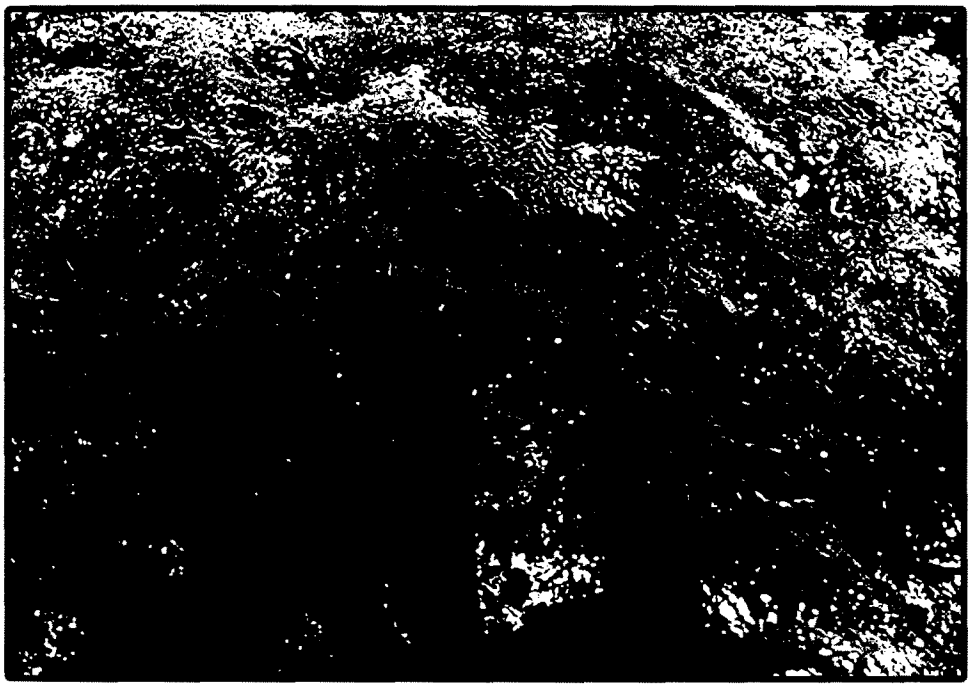
3.9 Statistical analysis



Leaves

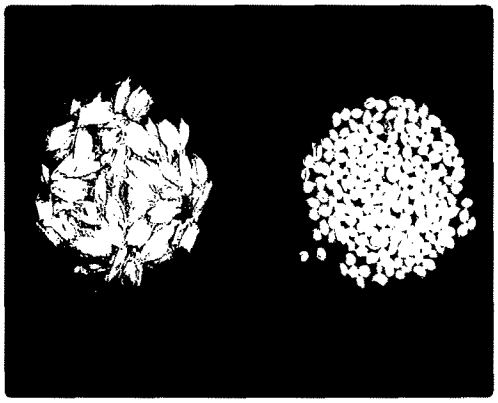


Flowers



The Miracle Tree (*Moringa oleifera*)

Pods



Seeds

3.1 Procurement of material

The samples of pods, leaves, flowers and seeds of *Moringa oleifera* (Family Moringaceae) were collected in a single lot from the field situated at Hansi-Bhiwani road.

3.2 Standardization of recipes

The following products using fresh as well as dried *Moringa* pods, flowers, leaves and seeds of *Moringa oleifera* were standardized and developed.

3.2.1 Leaves of *Moringa oleifera*

Chutney

Ingredients

M. oleifera leaves (fresh)	125 g
Onion	60 g
Tomato	30 g
Green chilli	5 g
Lemon juice	5 ml
Salt and red chilli powder	to taste

Method

1. Washed *Moringa oleifera* leaves.
2. Chopped onion, tomato and green chillies.
3. Mixed *Moringa oleifera* leaves, onion, tomato, green chillies and other spices and ground well. Added lemon juice and served.

Leaves *bhuji*

Ingredients

M. oleifera leaves (fresh)	250 g
Onion	25 g
Tomato	35 g
Salt	½ tsp
Cumin seeds	½ tsp

Products prepared from *Moringa oleifera* leaves

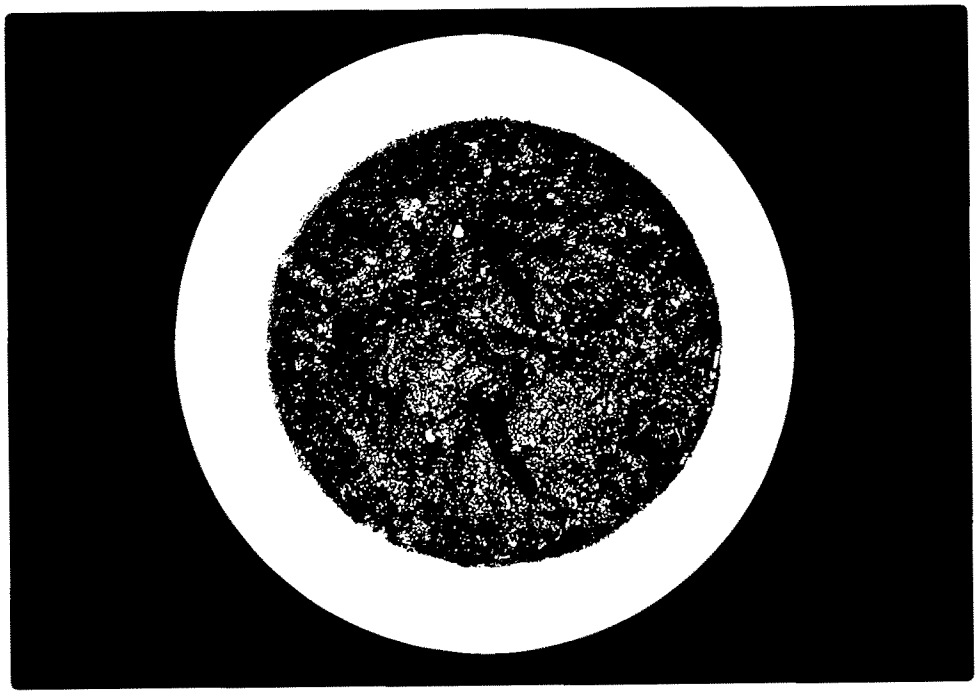


Plate 1 : Chutney

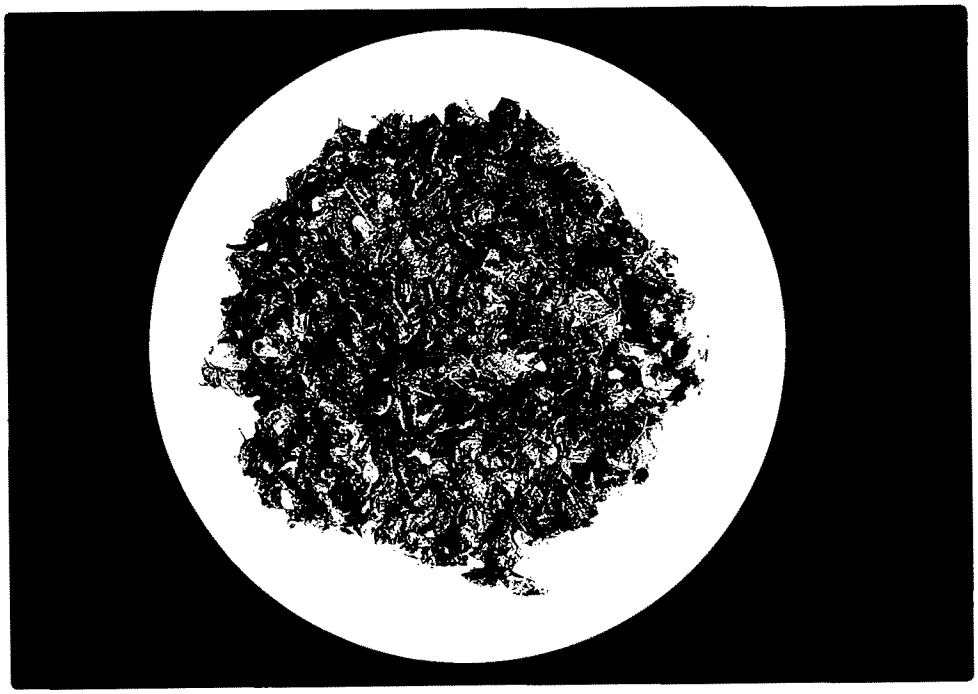


Plate 2 : Leaves *bhuji*

4. Added salt, spices, chopped leaves and potatoes and cooked on slow fire till tender.

Pakora

Ingredients

M. oleifera leaves (fresh)	100 g
Bengal gram flour	100 g
Onion	30 g
Chilli powder	½ tsp
Salt and red chilli powder	to taste
Green chillies	10 g
Vegetable oil	for frying

Method

1. Washed and chopped leaves, onion and green chillies.
2. Made thick batter of bengal gram flour with water.
3. Added all the ingredients including spices and condiments in the bengal gram batter and mixed thoroughly.
4. Took a small portion of the above batter formed in the shape of *pakora* and fried in the heated vegetable oil till golden brown in colour.

Sev

Sev made from bengal gram flour served as control. *Moringa oleifera* leaf powder was prepared by drying the fresh leaves in an oven at 60°C till moisture free. These dried leaves were ground to fine powder and incorporated in bengal gram flour at 10 and 20 per cent levels.

Types of *sev*

Ingredients	Control	I	II
Bengal gram flour (g)	100	90	80
M. oleifera leaf powder	-	10	20
Salt (g)	2	2	2
Vegetable oil	for frying		

Method

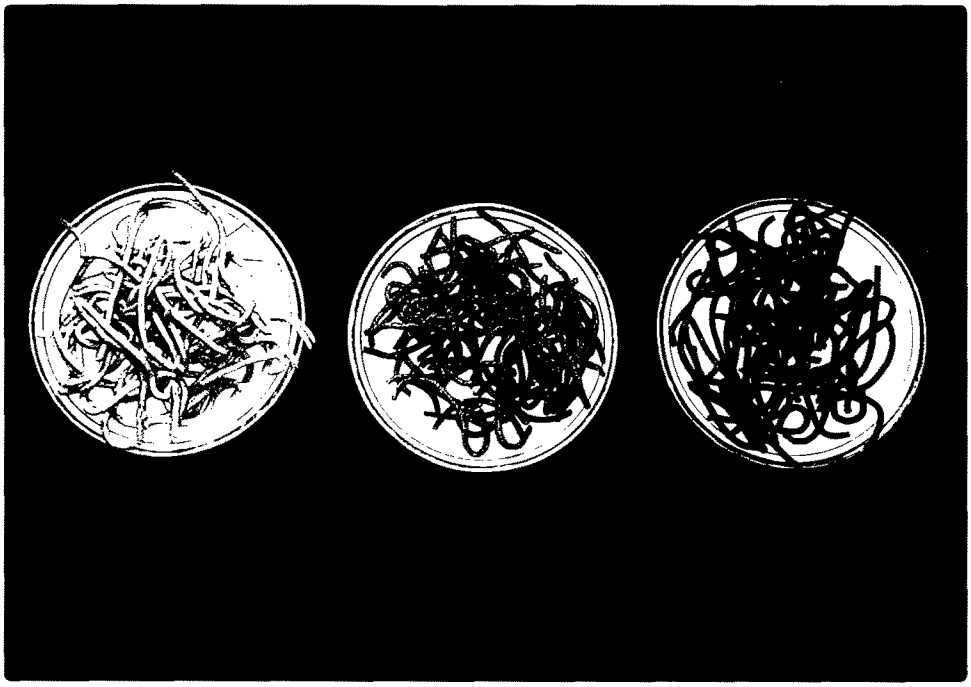
1. Mixed bengal gram flour, *M. oleifera* leaf powder and salt.
2. Added water to make a stiff dough.
3. Extruded the stiff dough through a vermicelli/*sev* machine by pressing the piston of the machine while keeping it above the *karahi* containing hot oil.
4. Fried the *sev* on low flame until golden brown.

Noodles

Noodles made from refined flour served as control. Dried *Moringa* leaf powder was incorporated in refined flour at 10 and 20 per cent level.

	Type of noodles		
Ingredients	**Control**	**I**	**II**
Refined flour	100	90	80
M. oleifera leaf powder		10	20
Water for kneading dough	30 ml	30 ml	30 ml

Noodles were made by a manual sheeting process from different flours.



I - Bengal gram flour (Control 100%)
II - Bengal gram flour (90%) + *M. oleifera* leaves (10%)
III - Bengal gram flour (80%) + *M. oleifera* leaves (20%)

Plate 5 : *Sev*

I - Refined flour (Control 100%)
II - Refined flour (90%) + *M. oleifera* leaves (10%)
III - Refined flour (80%) + *M. oleifera* leaves (20%)

Plate 6 : Noodles

1. The flour was hand mixed with a predetermined amount of water to form a stiff dough.
2. The dough was covered and kept for 30 min to permit optimum moisture equilibrium and hydration.
3. The dough was passed through the rolls to get sheet of 3 mm thickness. This was again passed through the rolls to get final sheet of 1.5 mm thickness.
4. Immediately after the dough sheet was cut into noodle strips and were taken on trays for drying.
5. The noodles were dried in hot air oven at 50°C temperature for 3 hours and packed till further use.

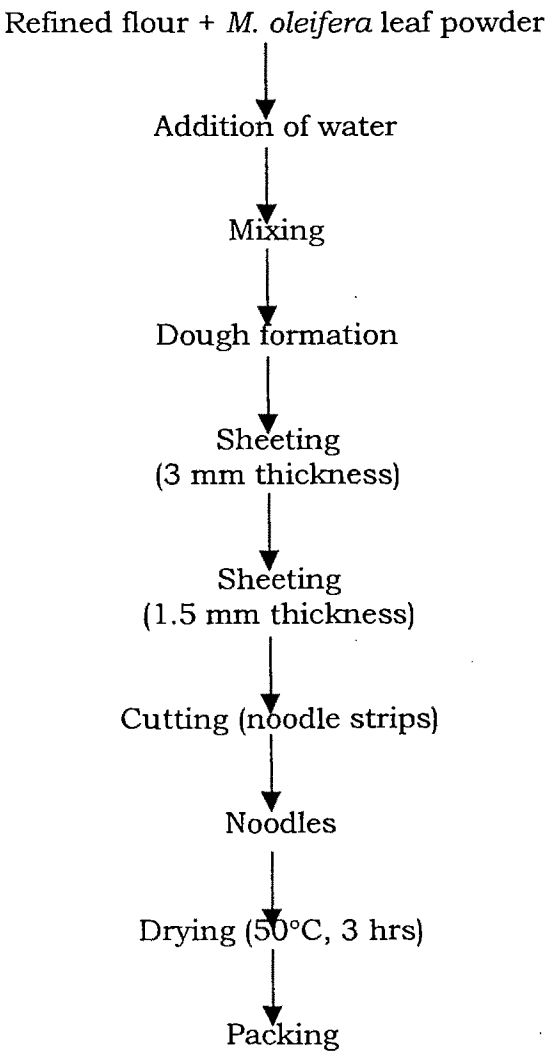


Fig. 1 Flow diagram of preparation of noodles

Preparation of cooked noodles

Noodles were boiled and seasoned with vegetables.

Ingredients

Noodles	30 g
Onion	10 g
Tomato	10 g
Capsicum	10 g
Cabbage	10 g
Carrot	10 g
Tomato sauce	1 tsp
Chilli sauce	½ tsp
Soy sauce	¼ tsp
Vinegar	¼ tsp
Oil	10 g
Salt	¼ tsp

Method

1. Water containing salt was boiled.
2. Extrudate was added into boiled water and boiled for 5 min or till soft.
3. Water was strained and kept aside.
4. Oil was heated in a pan, added chopped onion and sauted slightly. Then added chopped capsicum, cabbage and carrot and cooked till soft.
5. Salt, tomato sauce, chilli sauce, soy sauce and vinegar were added and stirred properly.
6. Boiled extrudate was mixed to vegetable mixture and cooked for 2-3 min.
7. Served hot.

3.2.2 Flowers of *Moringa oleifera*

Flower *Bhuji*

Ingredients

M. oleifera flowers	250 g
Onion	25 g
Tomatoes	35 g
Salt	½ tsp
Garam masala	½ tsp
Red chilli powder	¼ tsp
Turmeric powder	1/8 tsp
Ghee	10 g

Method

1. Washed flowers and blanched for 15 min.
2. Peeled onion and chopped it.
3. Chopped tomatoes.
4. Heated ghee, added onion and sauted till brown. Added chopped tomatoes and cooked for some time.
5. Added salt, spices, *M. oleifera* flowers and cooked on slow fire till tender.
6. Served hot.

Pea and Flower Vegetable

Ingredients

M. oleifera flowers	100 g
Peas	50 g
Tomato	15 g
Onion	10 g
Salt and Garam masala	to taste
Red chilli powder	to taste
Turmeric powder	1/8 tsp
Ghee	5 g

Products prepared from *M. oleifera* flowers

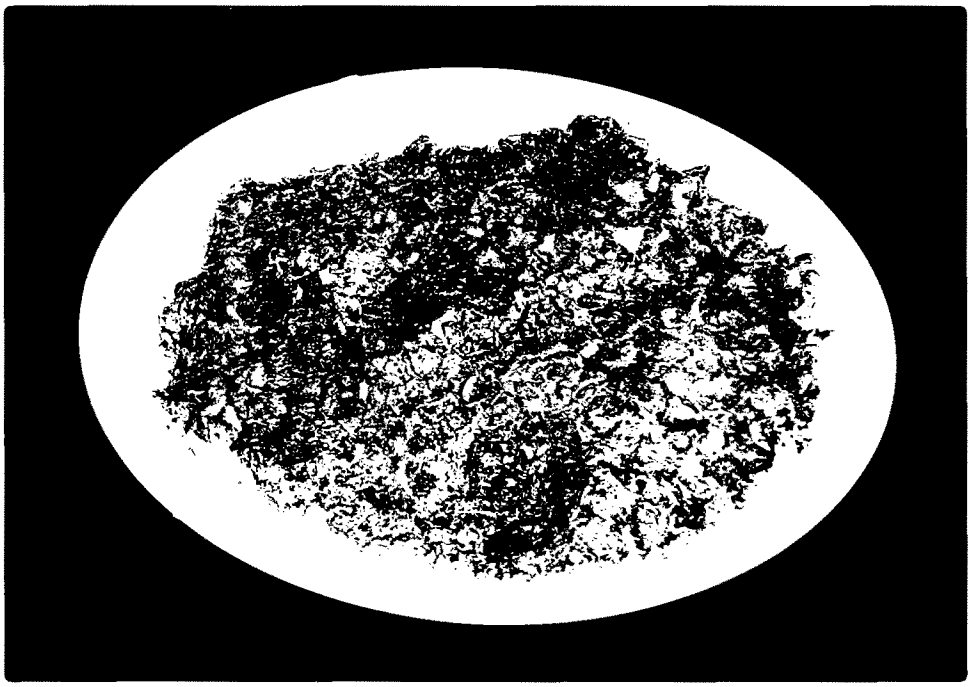


Plate 7 : Flower *bhuji*

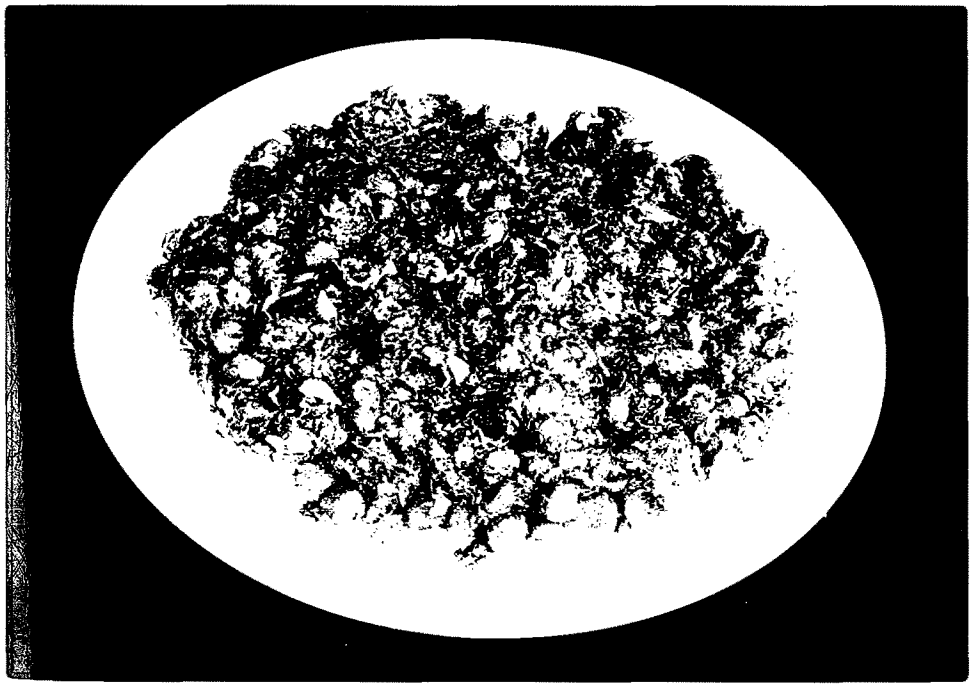


Plate 8 : Pea and flower vegetable

Method

1. Washed *Moringa oleifera* flowers and blanched for 15 min.
2. Chopped onion and tomatoes.
3. Heated ghee, added onion and sauted till light brown. Added chopped tomatoes and cooked till ghee separated.
4. Added salt, spices and peas and cooked till tender.
5. Added blanched flowers and cooked for 10 minutes.

Green Gram and Flower Vegetable

Ingredients

M. oleifera flowers	100 g
Green gram	50 g
Tomatoes	15 g
Onion	10 g
Salt	to taste
Garam masala	to taste
Red chilli powder	to taste
Turmeric powder	1/8 tsp
Ghee	5 g

Method

1. Washed *Moringa oleifera* flowers and blanched for 15 min.
2. Chopped onion and tomatoes.
3. Heated ghee, added onion and sauted till brown. Added chopped tomatoes and cooked till ghee separated.
4. Added salt, spices and green gram and cooked till tender.
5. Added blanched flowers and cooked for some time.

Raita

Ingredients

M. oleifera flowers	100 g
Curd	½ litre
Water	50 ml

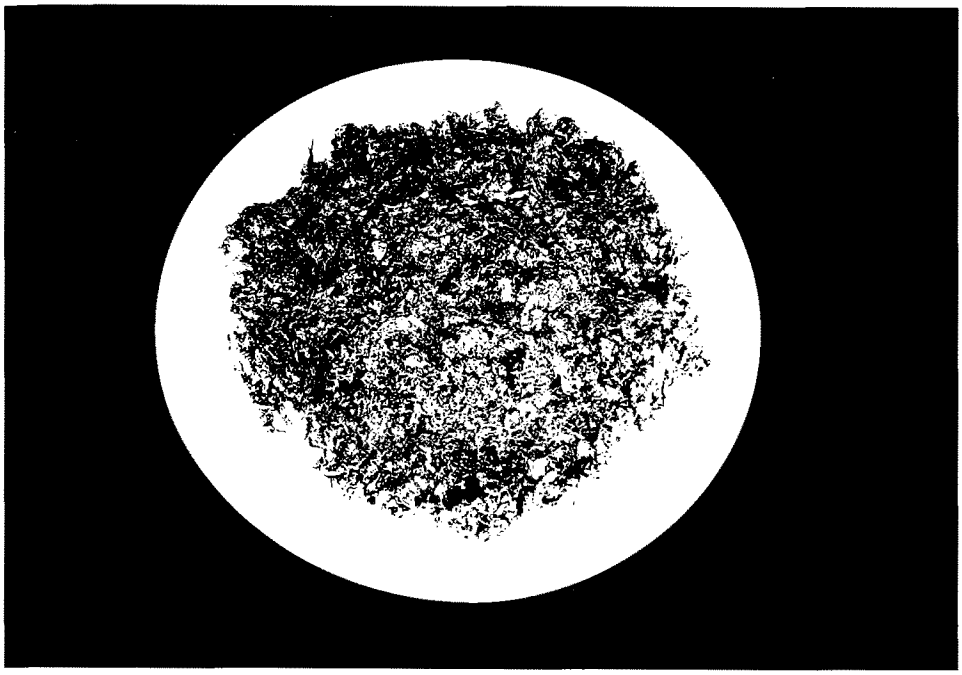


Plate 9 : Green gram and flower vegetable

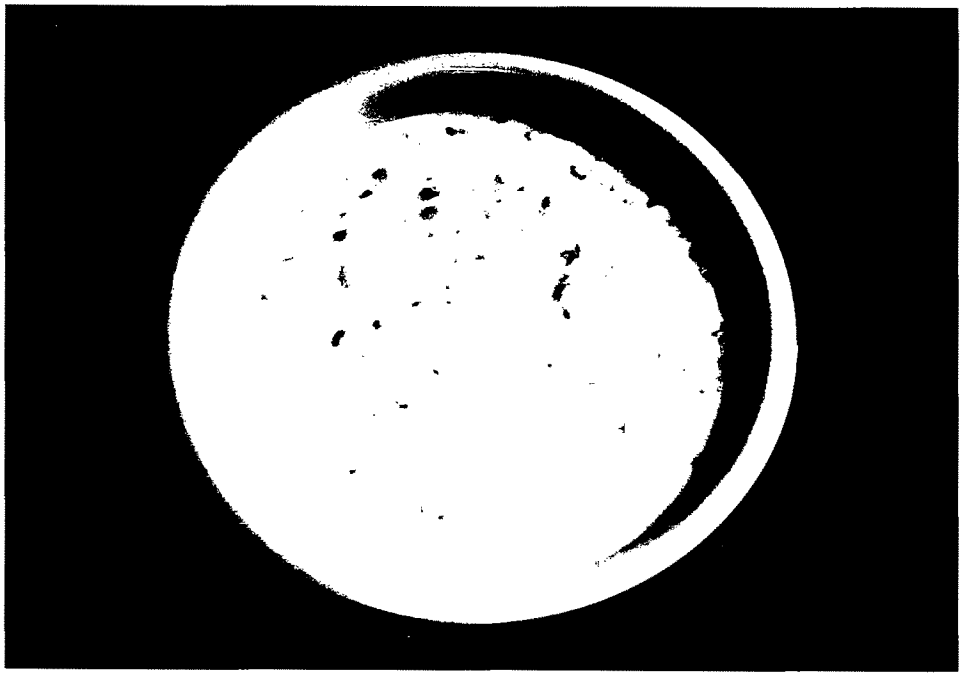


Plate 10 : *Raita*

Cumin seeds	5 g
Salt and red chilli powder	to taste

Method

1. Washed *Moringa oleifera* flowers, blanched for 15 min and mashed them.
2. Roasted cumin seeds and ground them.
3. Added mashed flowers, salt, cumin seed powder and red chilli powder in the curd and mixed well.

Cutlets

Ingredients

Boiled potatoes	75 g
Boiled green peas	10 g
M. oleifera flowers	25 g
Bread crumbs	25 g
Corn flour	15 g
Fat	for frying
Salt	to taste
Garam masala	to taste
Red chilli powder	to taste

Method

1. The *M. oleifera* flowers were washed and blanched for 15 min.
2. The potatoes were boiled, peeled and mashed.
3. Added spices and made into the shape of cutlets.
4. Prepared the batter of corn flour and water. Cutlets were dipped into corn flour batter and then rolled into bread crumbs.
5. Heated ghee and deep fried the cutlets till light brown colour.
6. Served hot.

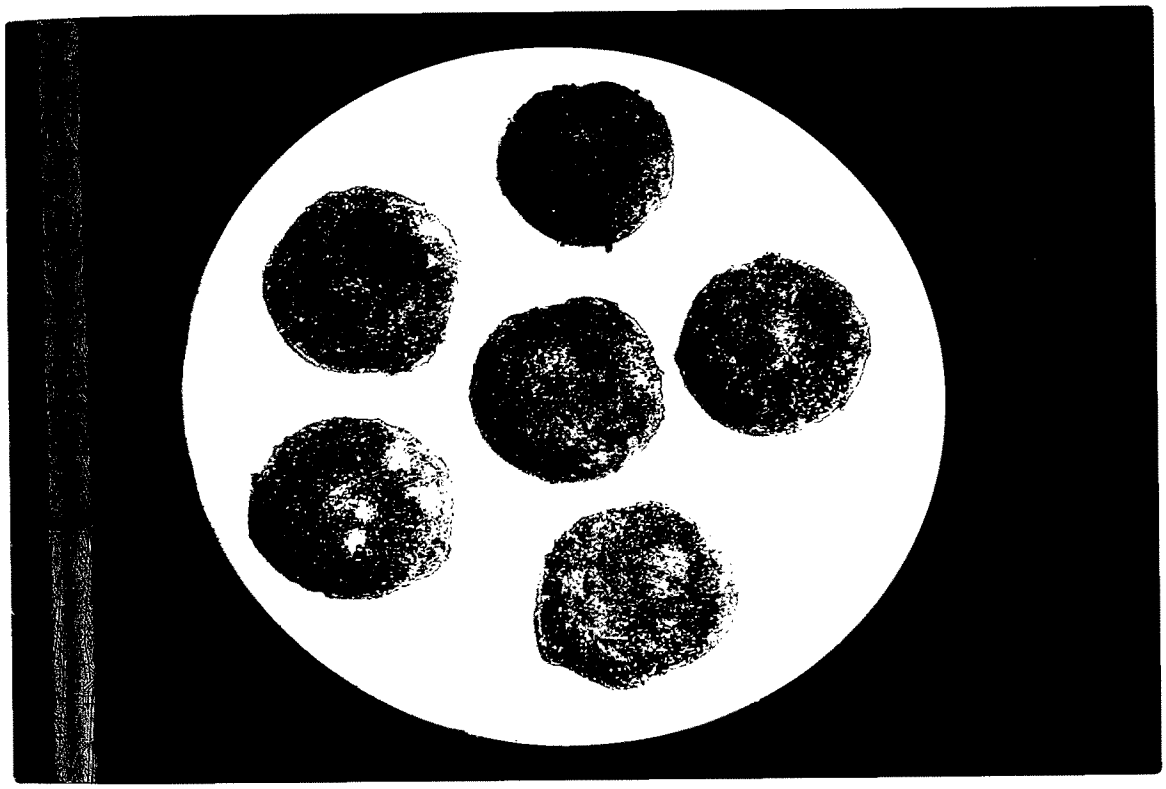


Plate 11 : Cutlets

3.2.3 Pods of *Moringa oleifera*

Pickle

Ingredients

M. oleifera pods	1 kg
Salt	125 g
Turmeric powder	25 g
Fennel seeds	50 g
Coriander seeds	50 g
Red chilli powder	40 g
Mustard oil	250 ml

Method

1. The *Moringa oleifera* pods were washed, wiped and dried in shade.
2. After 2 or 3 hours, cut the pods into small pieces of 1" in length.
3. All the spices were mixed well with mustard oil.
4. The pieces of *Moringa* pods were added to above prepared spice-oil mixture and the whole mass was mixed thoroughly and filled in air tight jar.
5. Kept the pickle in sun for 4-5 days till it was prepared.

Pod Vegetable

Ingredients

M. oleifera pods	250 g
Tomatoes	35 g
Onion	25 g
Salt and Garam masala	to taste
Cumin seeds	½ tsp
Red chilli powder	¼ tsp
Turmeric powder	1/8 tsp
Ghee	10 g

Products prepared from *M. oleifera* pods

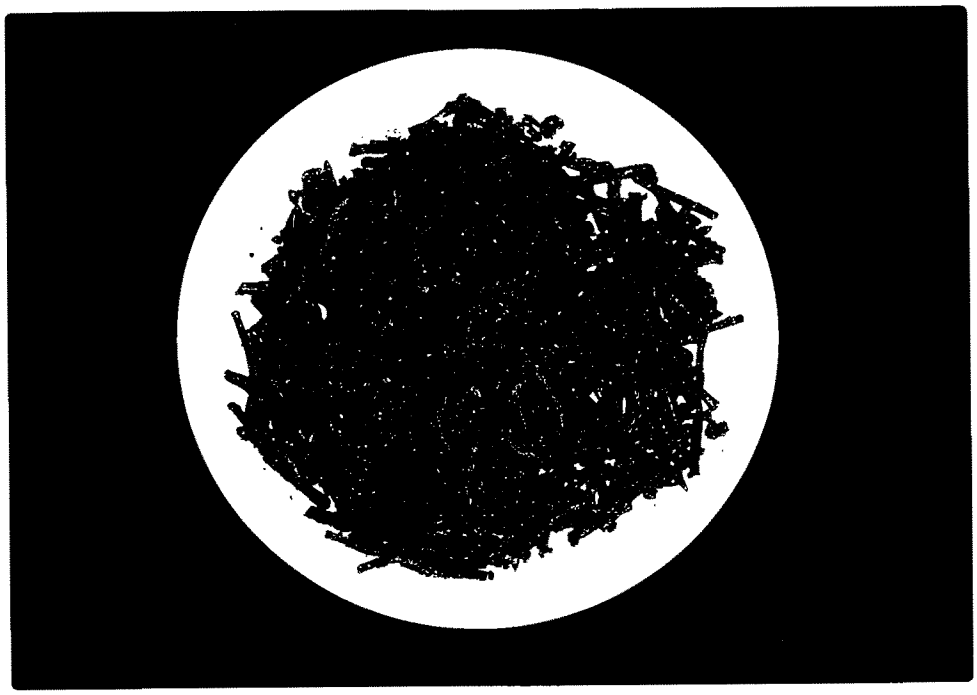


Plate 12 : Pickle

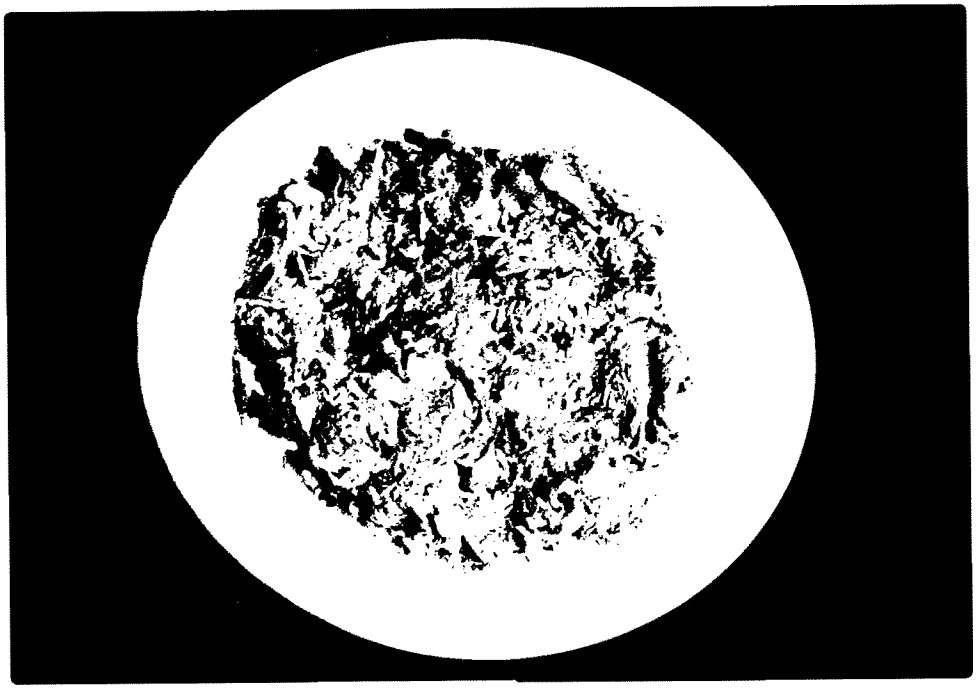


Plate 13 : Pod vegetable

Method

1. Washed *Moringa oleifera* pods and blanched till tender.
2. Peeled onion and chopped it.
3. Chopped tomatoes.
4. Heated ghee, added cumin seeds in it till crackled. Added onion and sauted till light brown. Added chopped tomatoes and cooked for some time.
5. Added salt, spices, blanched *Moringa oleifera* pods and cooked on slow fire till tender.

Potato and Pod Vegetable

Ingredients

M. oleifera pods	500 g
Potatoes	250 g
Tomatoes	75 g
Onion	50 g
Salt	1 tsp
Cumin seeds	1 tsp
Red chilli powder	½ tsp
Turmeric powder	¼ tsp
Ghee	20 g

Method

1. Washed *Moringa oleifera* pods and blanched till tender.
2. Chopped potatoes, onion and tomatoes.
3. Sauted in ghee till light brown. Added chopped tomatoes and cooked till ghee separated.
4. Added salt, spices and potatoes and cooked for 10 min till tender.
5. Added blanched *M. oleifera* pods and simmered and cooked for 10 minutes.

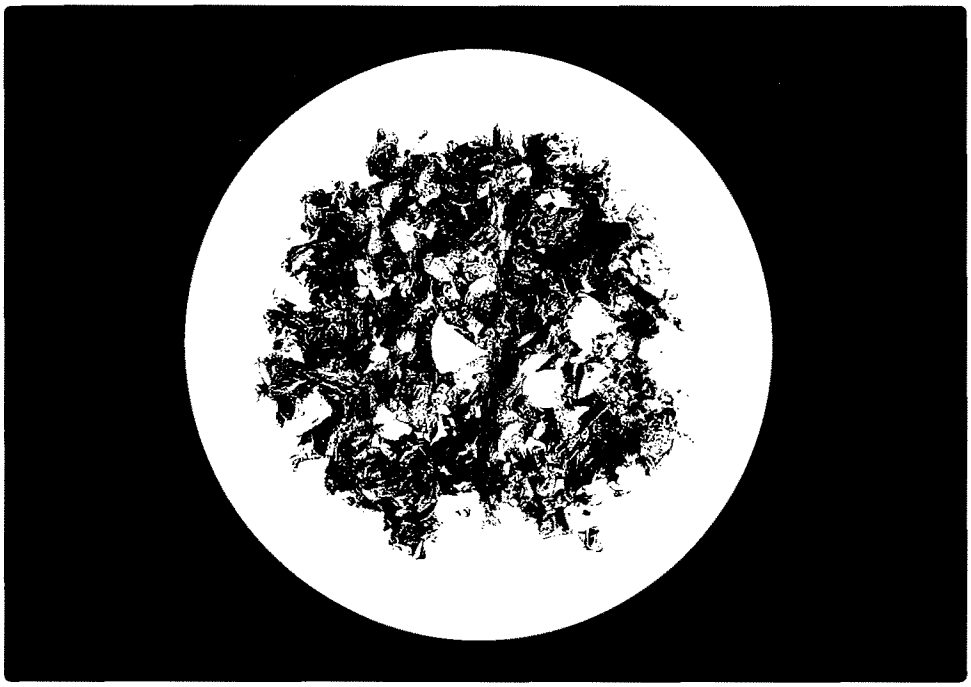


Plate 14 : Potato and pod vegetable

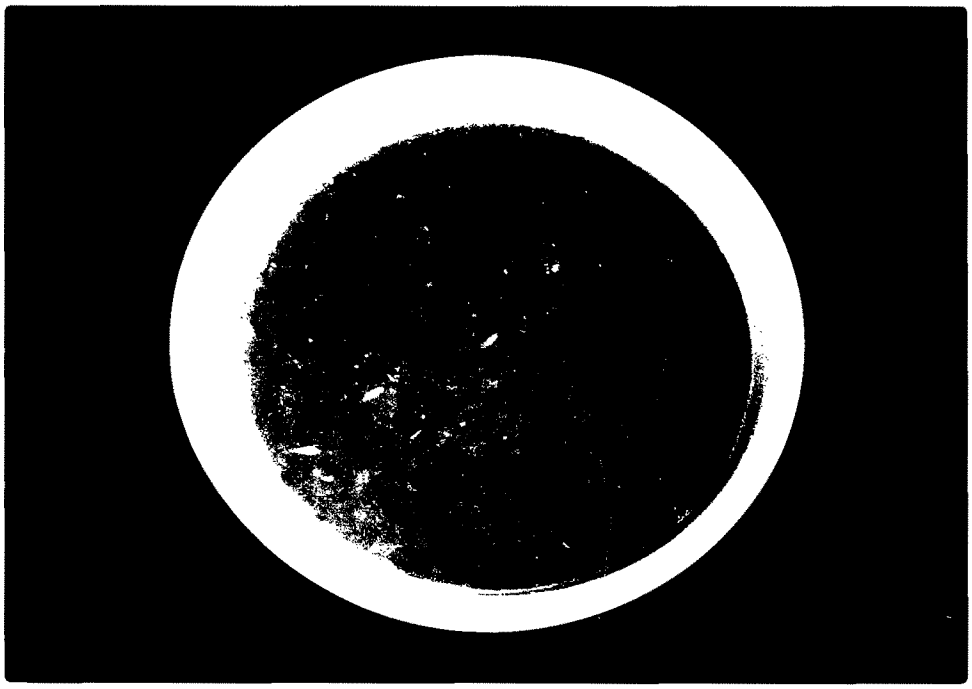


Plate 15 : *Sambhar*

Sambhar

Ingredients

M. oleifera pods	30 g
Lentil	50 g
Onion	25 g
Tomatoes	15 g
Mustard seeds	¼ tsp
Sambhar powder	½ tsp
Coriander seeds	¼ tsp
Fenugreek seeds	1/8 tsp
Dehulled bengal gram	¼ tsp
Dehulled urd bean	¼ tsp
Red chillies	1-2 No.
Curry leaves	1-2 No.
Brinjal	10 g
Capsicum	10 g
Potatoes	10 g
Ghee	10 g
Tamarind	5 g

Method

1. Washed and cut the vegetables.
2. Boiled *dal* and vegetables with salt.
3. Heated oil. Added mustard seeds and stirred till crackled. Sauted onion and tomatoes. Fried onion and tomatoes.
4. Added salt, chillies and *sambhar* powder.
5. Added boiled *dal* and vegetables to the above seasonings.
6. Soaked tamarind. Extracted the juice and added to the vegetable *dal* mixture.
7. Cooked for 15 minutes and serve hot.

Cutlets

Ingredients

M. oleifera pods	25 g
Boiled potatoes	75 g
Boiled green peas	10 g
Bread crumbs	25 g
Corn flour	15 g
Salt	to taste
Garam masala	to taste
Red chilli powder	to taste
Fat	for frying

Method

1. *M. oleifera* pods were blanched for 15 min.
2. Mixed the pods with boiled, peeled and mashed potatoes.
3. Added spices into the above mixture and made into the shape of cutlets.
4. Prepared the batter of corn flour with water. Coated the cutlets with cornflour batter and rolled into bread crumbs.
5. Heated ghee and fried the cutlets till light brown colour.

Sev

Sev made from bengal gram flour served as control. Dried *Moringa oleifera* pod powder was incorporated in bengal gram flour at 10 and 20 per cent levels. *Sev* were prepared as per procedure mentioned for dried *Moringa oleifera* leaf powder in section 3.2.1.

Noodles

Noodles made from refined flour served as control. Dried *Moringa oleifera* pod powder was incorporated in refined flour at 10 and 20 per cent level. Noodles were prepared as per procedure mentioned for dried *Moringa oleifera* leaf powder in section 3.2.1.

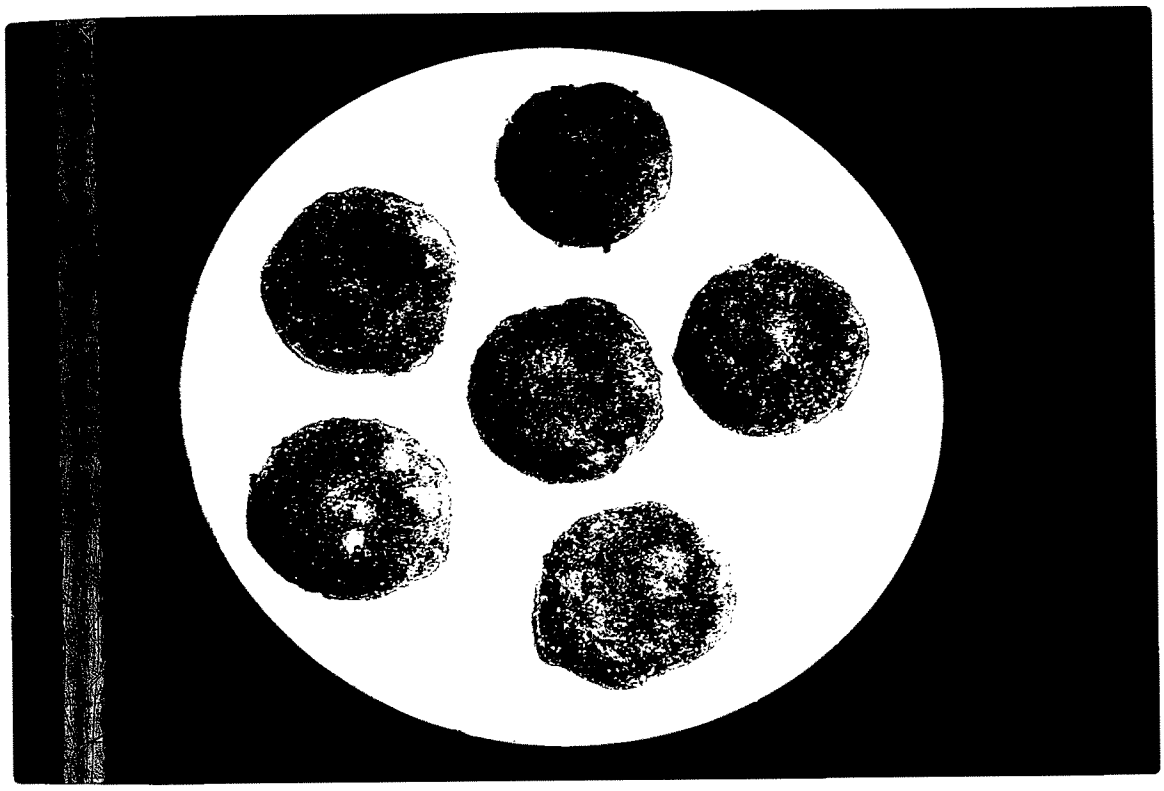


Plate 16 : Cutlets

I - Bengal gram flour (Control 100%)
II - Bengal gram flour (90%) + *M. oleifera* pods (10%)
III - Bengal gram flour (80%) + *M. oleifera* pods (20%)

Plate 17 : *Sev*

I - Refined flour (Control 100%)
II - Refined flour (90%) + *M. oleifera* pods (10%)
III - Refined flour (80%) + *M. oleifera* pods (20%)

Plate 18 : Noodles

Method

All the ingredients were mixed for the development of weaning mixtures.

Malting of wheat grain

The following main steps involved in malting process included:

i) Steeping ii) Germination iii) Roasting and iv) Milling

i) Steeping

Wheat grains were cleaned from dust and other foreign materials and steeped in equal amount of water at ambient temperature (25-30°C) and relative humidity (70%) for 12 hr.

ii) Germination

The soaked grains (whole wheat) were wrapped in damp muslin cloth and allowed to sprout for 24 hr at 30°C.

iii) Roasting

Roasted the sprouted cereal grains in hot air oven at 100 ± 5°C for two hours to develop characteristic malt aroma.

iv) Milling

Milled the malted grains and roasted pulses in grinding machine separately. Sieved the milled flour through 60 mesh sieve to get fine powder.

3.3 Organoleptic evaluation of products

For selecting the acceptable products for further study, the above mentioned products were subjected to organoleptic evaluation by a panel of ten judges from the I.C. College of Home Science, CCSHAU, Hisar. The judges were asked to record the quality characteristics i.e. colour, appearance, aroma, texture and taste by employing a nine-point Hedonic Rating Scale as given in Appendix-I. Judges were asked to rinse their mouth before and

after testing each product. Average of the scores for all these characteristics was expressed as overall acceptability.

3.4 Selection of products

On the basis of organoleptic evaluation, the most acceptable products developed from various parts of *M. oleifera* i.e. leaves, flowers, pods and seeds were selected for further nutritional and storage studies.

3.5 Nutritional evaluation of raw samples of pods, leaves, flowers and seeds of *Moringa oleifera* as well as its selected products

3.5.1 Preparation of samples for nutritional analysis

The raw samples of pods, leaves, flowers and seeds of *Moringa oleifera* as well as its selected products were analysed for proximate composition, minerals and antinutritional factors. Moisture, β-carotene and vitamin C of the samples were analysed on fresh basis whereas for all other parameters, the samples were nutritionally analysed on dry matter basis after drying the samples in an oven at 60°C till moisture free.

3.5.2 Moisture

Moisture content was estimated by employing the standard method of analysis (AOAC, 1995).

Procedure

Five gram sample was weighed in the moisture box and dried in hot air oven at 105°C for six hours. The sample was weighed after cooling it in a desiccator.

$$\text{Moisture (\%)} = \frac{\text{Loss in weight (g)}}{\text{Weight of sample (g)}} \times 100$$

3.5.3 Crude fat

Crude fat was estimated using Soxhlet method (AOAC, 1995).

Reagents

Petroleum ether (boiling range 40-60°C)

Procedure

Five gram of dried sample was weighed and transferred to an extraction thimble. The thimble was placed in a Soxhlet extractor fitted with a condenser and flask containing sufficient petroleum ether. The extraction was carried out for 6 hours and after the extraction, thimble was removed with the sample from the extraction apparatus and dried in the hot air oven to a constant weight. It was cooled in a desiccator at room temperature and weighed. The loss in the weight of the thimble was the estimate as the loss of fat from the sample and expressed as per cent of the crude fat in the sample.

3.5.4 Crude fibre

Reagents

i) Hydrochloric acid 1% (v/v)

ii) Sulphuric acid stock solution (10%, v/v): Took 55 ml conc. H_2SO_4

iii) Sulphuric acid working solution (12.5%): Diluted 125 ml of the stock solution to one litre.

iv) Sodium hydroxide stock solution (10%, w/v): Dissolved 100 g NaOH in water to dilute to one litre.

v) Sodium hydroxide working solution (1.25%): Diluted 125 ml of the stock solution to one litre.

vi) Antifoam: 2% silicon antifoam in CCl_4

Procedure

One g of fat free dried sample was weighed in one litre tall beaker. Added 200 ml 1.25 per cent H_2SO_4 and few drops of

antifoam. The solution was kept for boiling for 30 minutes under bulb condensers. Beaker was rotated occasionally to mix the contents and remove the particles from the side. Filtered the contents of the beaker through funnel. Washed the sample back into the tall beaker with 200 ml 1.25 per cent sodium hydroxide. Brought to boiling point. Boiled exactly for 30 minutes. Transferred all insoluble matter to the sintered crucible by means of boiling water till acid free. Washed twice with alcohol. Washed three times with acetone. Dried at 100°C to constant weight. Ashed in a muffle furnace at 550°C for 1 hour. Cooled the crucible in a desiccator, reweighed and the percentage of crude fibre in the sample was calculated.

3.5.5 Ash

Ash in the sample was estimated by employing the standard method of analysis (AOAC, 1995).

Procedure

Five gram of oven dried sample was weighed in the crucible. It was ignited till no charred particles remained in the crucible. The crucible was put in muffle furnace (500°C) for 5 hours or till a white ash was obtained. Then the crucible was cooled in desiccator and weighed. The loss in weight represented the organic matter and residue being the ash content.

3.5.6 Crude protein

The total nitrogen was estimated by the standard method of AOAC (1995). A factor of 6.25 was applied to convert the amount of nitrogen to crude protein.

Reagents

i) N/100 H_2SO_4

ii) Boric acid (4%)

iii) Mixed indicator solution: 0.5 g of bromocresol green and 0.1 g of methyl red was taken and dissolved in 100 ml 95 per cent ethanol and the solution was adjusted with drops of dilute NaOH to bluish purple colour.

iv) NaOH (40%)

v) Digestion mixture: 10 g K_2SO_4, 0.5 g $CuSO_4.6H_2O$ and 2 g $FeSO_4$.

Procedure

One gram sample was taken and digested with 25 ml concentrated H_2SO_4 and a pinch of the digestion mixture. The nitrogen, as ammonical salt, was distilled with 45 per cent NaOH in a kjeldahl apparatus. The ammonia, thus, liberated was absorbed in 10 ml boric acid solution containing a few drops of the mixed indicator and was titrated against standard HCl (N/100). The end point was indicated by the change of colour.

$$\text{Nitrogen (\%)} = \frac{0.00014 \times \text{Vol. of N/100 HCl used} \times \text{Vol. of digested sample made}}{\text{Wt. of sample} \times \text{Vol. of aliquot taken}}$$

3.5.7 Total carbohydrate

The total carbohydrate was calculated by the method of AOAC (1995).

Total carbohydrate (%) = 100 – (crude protein % + crude fibre % - Crude fat % + Total ash %)

3.5.8 Dietary fibre

Total, soluble and insoluble dietary fibre constituents were determined by the enzymatic method given by Furda (1981).

Reagents

i) 0.005 N HCl

ii) Phosphate buffer (pH 10)

iii) EDTA

iv) Enzymes: Alpha amylase and protease enzymes were obtained from Sigma Chemical Company, USA.

v) Ethanol (75% and absolute)

vi) Acetone

Procedure

i) **Sample preparation:** 0.5 g sample of less than 1 mm particle size food material was defatted on a Soxhlet or Goldfish apparatus.

ii) **Extraction of water-soluble material:** The prepared sample weighing about 2.0 g was dispersed in 200 ml of 0.005 N HCl and boiled for 20 min. The suspension was then cooled down to 60°C; 0.3 g of disodium EDTA was added and then adjusted to pH 5.0-6.5 with 12 ml of phosphate buffer pH 10. The extraction was continued for an additional 40 min at 60°C to ensure the extraction of pectins with minimal degradation.

iii) **Starch and protein hydrolysis:** Adjusted the pH 6.0-6.5 to bring the solution closer to the pH optimum of amylase and protease. Cooled the suspension to 20-30°C before incubation overnight with 10 mg of bacterial alpha-amylase and 10 mg of bacterial protease. The incubation was accompanied by slow stirring with a magnetic bar.

iv) **Isolation of insoluble dietary fibre:** The suspension was filtered through a coarse-tarred Gooch filtering crucible containing glass wool and the insoluble residue was washed with a small amount of water. The filtrate was saved for the

next step. The insoluble residue was then washed with water, alcohol and acetone before being dried at 70°C in a vacuum oven overnight. The dry residue constitutes insoluble dietary fibre (IDF).

v) **Precipitation and isolation of soluble dietary fibre (SDF):** The saved filtrate was acidified with a few drops of concentrated hydrochloric acid to pH 2-3; this pH tended to facilitate the rapid precipitation of polysaccharides. Slowly added four volumes of ethanol and left suspension to stand for about 1 hour. Filtered the precipitate on a tarred, coarse Gooch crucible containing glass wool, then washed with 75% ethanol, absolute ethanol, and acetone before drying at 70°C in a vacuum oven overnight. The residue was weighed in the crucible to give the soluble dietary fibre (SDF) content of the original material. The SDF fraction was corrected for ash and for-co-precipitated protein.

vi) **Total dietary fibre (TDF):** The sum of insoluble dietary fibre and soluble dietary fibre contents were calculated.

TDF = IDF + SDF

3.5.9 β-carotene

β-carotene in sample was separated by column chromatography and estimated colorimetrically (AOAC, 1995).

Reagents

i) 3% acetone in petroleum ether

ii) Alumina (aluminium oxide neutral)

iii) Sodium sulphate anhydrous

iv) β-carotene standard: β-carotene (50 mg) was dissolved in 3% acetone in petroleum ether and diluted to 50 ml (1 mg/ml).

Procedure

Preparation of chromatography column

The chromatography column was filled with aluminium oxide which had been dried at 70°C in an oven. The absorbent was gently pressed down to a depth of 10 cm by tapping or suction. The absorbent was covered with a one cm layer of anhydrous sodium sulphate. The column was wetted with 3 per cent acetone in petroleum ether. The column was not allowed to dry at any stage.

Extraction of sample

Weighed 10 g sample in a conical flask and added 30 ml 3 per cent acetone in petroleum ether to it and was allowed to stand overnight. The extract was then filtered and residue was washed with 3 per cent acetone in petroleum ether until the filtrate was of clear yellow colour. The filtrate was pooled and taken in 500 ml separating funnel. It was then shaken with 50 ml water. Washings were discarded. It was repeated 2-3 times in order to make it acetone free. The solvent was then dried over anhydrous sodium sulphate and was diluted to 100 ml by petroleum ether.

Chromatographic separation of β-carotene

Ten ml extract was taken and concentrated to 2 ml. The condensed extract was poured into adsorption column followed by 10 ml 3 per cent acetone petroleum ether. The elute containing all the β-carotene was collected and transferred to 25 ml volumetric flask. Volume was made with 3 per cent acetone petroleum ether.

For standard curve

0.2 to 2.0 ml standard solution was taken in 25 ml volumetric flask and volume was made with 3 per cent acetone in petroleum ether. The colour intensity was read at 435 nm on spectrophotometer. 0.340 O.D. corresponds to 0.32 mg β-carotene.

From the standard curve, the concentration of β-carotene in the sample can be calculated. β-carotene concentration corresponding to β-carotene (mg/100 g) was calculated as follows:

$$\beta\text{-carotene (mg/100 g)} = \frac{M \times V_1 \times 100}{V_2 \times W}$$

where,

M = Concentration of extract eluted from graph

W = Weight of sample taken

V1 = Volume of extract made

V2 = Volume of extract taken for elution

3.5.10 Ascorbic acid

Ascorbic acid in the sample was estimated by titration method of AOAC (1995).

Reagents

i) Metaphosphoric acid-acetic acid solution: 15 gm HPO_3 pellets were dissolved in 40 ml glacial acetic acid and 200 ml distilled water and diluted to 500 ml. It was filtered rapidly through filter paper into a glass stoppered bottle.

ii) Ascorbic acid standard solution (1 mg ascorbic acid/ml): Fifty mg ascorbic acid reference standard (that had been stored in desiccator away from direct sunlight) was weighed and transferred to 50 ml volumetric flask. It was diluted to volume immediately (before use) with metaphosphoric acetic acid solution.

iii) Indophenol standard solution: Fifty mg 2,6,dichloroindophenol sodium salt (that had been stored in desiccator) was dissolved in 50 ml distilled water, to which 42 mg sodium bicarbonate had been added when the dye

dissolved; it was diluted to 200 ml with distilled water and was filtered through Whatman # 1 into amber glass stoppered bottle. This was kept stoppered and away from direct sunlight in refrigerator.

Extraction

To five g of sample, 25 ml of metaphosphoric acetic acid solution was added. The sample was made to a fine pulp in pestle and mortar until the suspension appeared one, mixed well and volume was made to 100 ml with metaphosphoric acetic acid solution. Filtered rapidly through Whatman # 1.

Estimation

Two ml aliquots of ascorbic acid standard solutions were taken in triplicate in each of the three 50 ml conical flasks containing five ml metaphosphoric acetic acid solution. These standard samples were titrated rapidly with indophenol solution from a microburette until light, but distinct rose pink colour persisted at least for five seconds. Similarly, blank containing seven ml metaphosphoric acetic acid solution and distilled water equal to the reading of standard was titrated. For the sample, five ml metaphosphoric acetic acid was added to each of two ml of sample aliquots and titrated with indophenol solution as for blank and standard. Ascorbic acid content (mg/100 g) was calculated as follows:

Calculations

$$\frac{Y - B}{X - B} \times \frac{V}{W} \times 100$$

Where

Y = Volume of dye solution used against sample aliquot

B = Volume of dye solution used against blank

X = Volume of dye solution used against standard

V = Volume of aliquot made

W = Weight of the sample

3.5.11 Total minerals

One g ground sample was taken in a 150 ml conical flask. To this, 25-30 ml diacid mixture (HNO_3:$HClO_4$; 5:1 v/v) was added and kept overnight. Next day, it was digested by heating till clear white precipitates settled down at the bottom. The crystals were dissolved by diluting in double distilled water. The contents were filtered through Whatman No. 42 filter paper. The filtrate was made to 50 ml with double distilled water.

3.5.11.1 Calcium, iron and zinc

The acid digested sample was used for the determination of calcium, iron and zinc by Atomic Absorption Spectrophotometer 2380, Perkin Elmer (USA) according to the method of Lindsey and Norwell (1969).

3.5.11.2 Potassium

Potassium in acid digested sample was determined by the flame photometer.

3.5.11.3 Phosphorus

Phosphorus was determined colorimetrically by the method of Chen *et al.* (1956).

Reagents

1) Ascorbic acid (10%)
2) Ammonium molybdate (2.5%)
3) Reagent C: 6 N H_2SO_4.water, 2.5 per cent ammonium molybdate and 10 per cent ascorbic acid were mixed in the ratio of 1:2:1:1 (v/v), respectively. This reagent was prepared fresh everyday.
4) Standard phosphorus solution: 0.351 g pure and dry anhydrous monopotassium dihydrogen orthophosphate was

dissolved in a few ml water and 10 ml of 10 N H_2SO_4. The volume was made to one litre with water. This stock solution contained 80 µg P/ml. Twenty five ml stock solution was diluted to 1 litre which served as working standard solution. It contained 2 µg P/ml. Two or three drops of chloroform were added for preserving the solution.

Procedure

A suitable aliquot (1 ml) of the mineral extract was pipetted in a test tube and made the volume to 4 ml with water. Then 4 ml reagent C was added and mixed well. The content was incubated at 37°C in a water bath for 90 minutes. It was removed and allowed to cool to room temperature and absorbance was read at 820 nm against a suitable blank.

Standard curve was plotted using one to eight µg P (0.195 OD corresponded to 2 µg phosphorus).

3.5.12 HCl-extractability of minerals

Minerals including calcium, iron, sodium and potassium were extracted in 0.03 N HCl.

Added 50 ml 0.03 N HCl to one g sample. The mixture was incubated at 37°C in a water bath-cum-shaker for 3 h to simulate the conditions that occur in human stomach. The mixture was then filtered through ashless filter paper (Whatman # 42). The filtrate was oven dried, digested in the diacid mixture and proceeded for the determination of each of the minerals as given in section 3.5.11.1 for Ca, Fe and Zn and section 3.5.11.2 for sodium and potassium.

3.5.13 *In vitro* availability of calcium and iron

3.15.13.1 Calcium availability (*in vitro*):

Available calcium was extracted by the method of Kim and Zemel (1986).

Reagents

i) 0.1% Pepsin in 0.1 N HCl

ii) HCl

iii) $NaHCO_3$

iv) 0.5% pancreatin in 5% bile

Procedure

Two grams of finely ground sample was taken in a conical flask and 3 ml distilled water was added to rehydrate it. To this 20 ml of pepsin solution (0.1% pepsin in 0.1N HCl) was added. The pH was adjusted to 1.5 with dilute HCl. The contents were incubated at 37°C in a shaker-cum-water bath for one hour. After one hour, the pH contents were raised to 6.8 with sodium bicarbonate solution. Then 2.5 ml of a suspension containing 0.5% pancreatin in 5% bile was added and the contents were again incubated at 37°C for one hour. Then the contents were taken out and total volume was made to 50 ml with distilled water. The contents were then immediately centrifuged at 500 x g for 45 min at 5°C. Supernatant was collected and re-centrifuged at 25,000 x g for 45 minutes at 5°C. The supernatant was collected, oven dried, digested in the diacid mixture and proceeded for the estimation of calcium by the atomic absorption spectrophotometric method.

Iron availability (*in vitro*)

Available iron and zinc in the samples were extracted according to the procedure of Rao and Prabhavathi (1978).

Procedure

Two g sample was mixed with 25 ml pepsin HCl (0.5% pepsin in 0.1N HCl) in a conical flask. The pH of the mixture was adjusted to 1.35 with HCl and incubated at 37°C for 90 min in a water bath-cum-shaker. After incubation, pH of the contents was adjusted to 7.5 with NaOH and again incubated at 37°C in a water bath-cum-shaker for 90 min. Contents of the flasks were centrifuged at 9000 rpm for 30 min and the supernatant was filtered through Whatman # 44. The filtrate was oven dried, digested in the diacid mixture and proceeded for the determination of iron by atomic absorption spectrophotometric method.

3.5.13 Antinutritional factors

3.5.13.1 Oxalic acid

Reagents

i) HCl (6N): Added equal amounts of concentrated HCl and distilled water to prepare 6N HCl.

ii) Methyl red indicator

iii) Calcium chloride (5%)

iv) Sulphuric acid in water (1:4)

i) $KMNO_4$ (N/20)

Procedure

Weighed 2 g of sample in a 250 ml volumetric flask. Added 190 ml of water and 10 ml of 6N HCl and digested for 1 hour on boiling water bath. Allowed to cool, diluted to make volume and filtered the supernatant. Took 50 ml of filtrate in beaker and added 20 ml of 6N HCl. Evaporated the mixture to about half of its volume and filtered. Washed the precipitate several times to make the volume about 125 ml. Added 3-4 drops of methyl red to the filtrate followed by concentrated ammonia till the solution turned faint yellow. Heated at 90-100°C. Allowed to cool and filtered to

remove the precipitated ferrous impurities, if any. Boiled the filtrate, added 10 ml of 5% $CaCl_2$ with constant stirring and allowed to stand overnight. Filtered through filter paper Whatman No. 41. Washed the precipitate several times with hot water to make it free of Ca ions. Transferred the precipitate to the original beaker by washing with distilled water. Added 1:4 sulphuric acid till the precipitate was completely dissolved. Warmed the contents (70°C) and titrated with N/20 $KMNO_4$ to the near end point. Added the filter paper to the contents, stirred it thoroughly and completed the titration.

Calculations

$$\text{Oxalate (g/100 g)} = V \times 0.00225 \times \frac{V_1 \times 100}{V_2 \times W}$$

where,

V = Volume of N/20 KMNO4 used (ml)

V1 = Volume made (ml)

V2 = Aliquot (ml)

W = Weight of sample

3.5.13.2 Phytic acid

Phytic acid was determined by the method of Davies and Reid (1979).

Reagents

ii) Nitric acid (0.5M): HNO_3 69.5% (15.96 ml) (AR grade, sp. gr. 1.42) was diluted to 500 ml with distilled water.

iii) Ferric ammonium sulphate: Ferric ammonium sulphate (215 mg) was dissolved in distilled water. To it added few drops of HCl and volume was made to 500 ml with distilled water.

iv) Ammonium thiocyanate: Ammonium thiocyanate (10 g) was dissolved in distilled water and volume made to 100 ml.

v) Iso-amyl alcohol

vi) Sodium phytate: Sodium phytate (5.5% H_2O, 97% purity and containing 12 Na/mole) (30.54 mg) was dissolved in 100 ml of 0.5 M HNO_3 which gave a solution containing 20 mg phytic acid in 100 ml or 200 µg phytic acid/ml.

Extraction

To 500 mg sample, 20 ml 0.5 M HNO_3 was added in a conical flask and shaken continuously for 3 h on shaker at room temperature. The contents were centrifuged at 3000 rpm for 15 min. Supernatant was used for estimation of phytic acid.

Procedure

To a test tube, 0.5 ml HNO_3 extract was taken and volume was made to 1.4 ml with water. To it, added one ml ferric ammonium sulphate solution, the contents were thoroughly mixed and placed in a boiling water bath for 20 min. Immediately cooled the tubes to room temperature under tap water. Five ml iso-amyl alcohol was added to it, the contents were mixed vigorously and to it added 0.1 ml ammonium thiocyanate solution. The tubes were shaken well and centrifuged at 3000 rpm for 10 min. Colour intensity in the alcohol was read exactly after 15 min of addition of ammonium thiocyanate at 465 nm against iso-amyl alcohol blank.

For plotting a standard curve, 0.2 to 1.2 ml standard phytate solution containing 40-240 µg phytic acid was taken and made to 1.4 ml with water. 0.341 O.D. corresponded to 160 µg phytic acid.

3.5.13.3 Saponins

Saponins were extracted and determined by the modified method of Gestotner *et al.* (1966).

Reagents

i) Standard saponin solution: Dissolved 50 mg saponin in acetic acid and made to 100 ml with acetic acid.

ii) 1N H_2SO_4 in dioxane : water (1:3, v/v)

iii) Acetic acid 10%

iv) Conc. H_2SO_4

v) Sodium sulphate

vi) Alumina (Aluminium oxide, active acidic)

vii) Benzene

viii) Diethyl ether

ix) Methanol solution (3%) in benzene

Extraction

Five hundred mg sample was taken in an extraction flask, dispersed in 50 ml 1N H_2SO_4 in dioxane : water (1:3) and hydrolysed under reflux for 8 hr. The contents were cooled and diluted with addition of 50 ml water. Sapogenins were extracted with 25 ml and then with three successive portions of 15 ml diethyl ether. The combined ether extracts were washed with water, made moisture free by adding sodium sulphate and then dried. The dried residue was taken up in minimal amount of benzene and purified on a column of alumina.

Isolation

For preparing column, 5 g freshly activated alumina (110°C for 2 h) was suspended in 100 ml benzene and 0.2 ml 10% acetic acid was added and stirred vigorously for 30 min. Immediately it was poured into a column of 15 mm diameter and washed with 250 ml benzene. Sapogenin extract in benzene was loaded on the column. Various impurities were removed by washing the column with 100 ml benzene and the sapogenins were then eluted with 100 ml of 3% solution of methanol in benzene. The eluate was

concentrated nearly to dryness and the residue was dissolved in 10 ml acetic acid.

Estimation

To two ml acetic acid solution of sapogenins one ml glacial acetic acid followed by two ml conc. H_2SO_4 was added. The contents of tube were mixed thoroughly and were cooled to room temperature. The absorbance was read at 530 nm against a blank (containing 3 ml glacial acetic acid and 2 ml conc. H_2SO_4).

For plotting a standard curve 0.5 ml to 3.0 ml standard solution containing 0.25 to 1.5 mg saponin was taken. 0.5 mg saponin O.D. corresponded to 0.125 O.D.

3.5.13.4 Trypsin inhibitor activity

Trypsin inhibitor activity was determined by the modified method of Roy and Rao (1971).

Reagents

i) 0.1 M Phosphate buffer (pH 7.6): Sixteen ml NaH_2PO_4 (0.2 M) and 84 ml Na_2HPO_4 (0.2 M) were diluted to 200 ml with distilled water and pH adjusted to 7.6.

ii) 0.05 M phosphate buffer (pH 7.0): 0.1 M phosphate buffer (50 ml) was diluted to 100 ml with water and the pH adjusted to 7.0.

iii) Casein solution (2%): A suspension of 2 g casein was prepared with phosphate buffer (0.1 M, pH 7.6) and dissolved by warming and occasional shaking on a steam bath for about 10 minutes. The solution was cooled and made to 100 ml with phosphate buffer and stored in a refrigerator.

iv) Trypsin solution (5 mg/ml): Dissolved 125 mg trypsin (Sigma Chemical Company, USA) in 25 ml phosphate buffer (0.1 M, pH 7.6).

v) 0.001 N HCl: Conc. HCl (8.88 ml) was added to distilled water and volume made to one litre. Pipetted 1 ml of this 0.1N HCl and volume was made to one litre.

vi) Trichloroacetic acid (5%)

Extraction

One g sample was taken in a 150 ml conical flask and 25 ml 0.05 M phosphate buffer (pH 7.0) was added. The contents were shaken at room temperature for three hrs and centrifuged at 10,000 rpm for 20 minutes. The following sets of incubation mixtures were prepared.

	Test	**Control**	**Blank**
Phosphate buffer (0.1 M, pH 7.6)	1.0 ml	1.1 ml	1.0 ml
Trypsin solution (5 mg/ml)	0.5 ml	0.5 ml	0.5 ml
HCl (0.001 N)	0.4 ml	0.4 ml	0.4 ml
TCA (5%)	-	-	6.0 ml
Casein (2%)	2.0 ml	2.0 ml	2.0 ml
Extract	0.1 ml	-	0.1 ml
	Incubated at 37°C for 20 minutes		
TCA (5%)	6.0 ml	6.0 ml	-

After incubation and addition of TCA, the contents were centrifuged at 10,000 rpm for 10 minutes. TCA soluble proteins in supernatant were determined by the method of Lowry *et al.* (1951).

Reagents

i) Sodium carbonate(2%) in 0.1 N NaOH

ii) $CuSO_4.5H_2O$ (0.5%) in 1% sodium citrate

iii) Alkaline Copper sulphate: Fifty parts of solution (i) and one part of solution (ii) were mixed just before use.

iv) 1N Folin-Ciocalteau phenol reagent

v) Working casein standard solution (1 mg/ml): Diluted 5 ml casein solution (2%) to 100 ml with phosphate buffer (0.1 M, pH 7.6)

Estimation

To 0.5 ml supernatant, 5 ml alkaline copper sulphate solution was added, mixed thoroughly and allowed to stand for 10 min at room temperature. Then 0.5 ml 1N Folin-Ciocalteau phenol reagent was added and again immediately mixed. A blank by taking water was also run side by side. After 30 min, the colour intensity was read at 520 nm against a blank.

Trypsin inhibitor units: One unit of trypsin was defined as the amount of enzyme which converted one mg casein to TCA soluble components at 37°C for 20 minutes at pH 7.6. One unit of inhibitory activity is that which reduces the activity of trypsin by one unit under the assay conditions.

3.6 Shelf life of selected products

The acceptable products, suitable for storage included *sev* prepared from *Moringa oleifera* leaf as well as powder and *M. oleifera* pod pickle. *Sev* was packed in polythene bags whereas pickle was stored in plastic containers and kept at 30°C in an incubator. The stored samples were analysed at weekly interval for free fatty acids, fat acidity and organoleptic acceptability. Further, nutritional analysis of these stored samples was done for vitamin C (3.5.10), Fe (3.5.11.1) and oxalic acid (3.5.13.1).

3.6.1 Fat acidity

The fat acidity was determined by the method of AOAC (1995).

Reagents

i) Petroleum ether

ii) Benzene-alcohol-phenolphthalein solution (0.02%): To one litre benzene, one litre alcohol and 0.4 g phenolphthalein was added and mixed.

iii) Potassium hydroxide solution (0.0178N)

Procedure

Ten gram sample was extracted with petroleum ether on Soxhlet apparatus. The solvent of the extract was completely evaporated on steam bath. The residue was dissolved in extraction flask with 50 ml benzene-alcohol-phenolphthalein solution and titrated with standard potassium hydroxide (1 g/litre) to orange pink colour. Blank titration was made on 50 ml benzene-alcohol-phenolphthalein and this value was subtracted from titration value of sample.

Fat acidity was calculated as mg of potassium hydroxide required to neutralize free fatty acids of 100 g flour.

Fat acidity = 10 x (T-B)

where,

T = ml KOH required to titrate sample extract

B = ml KOH required to titrate blank.

3.6.2 Free fatty acids

Free fatty acids in the fat were determined by method of AOAC (1995)

Reagents

i) Sodium hydroxide (0.25 N): Ten g NaOH was dissolved in water and made up to 1 litre with water.

ii) Isopropyl alcohol (99%): Isopropyl alcohol was neutralized with 0.1N NaOH solution to a pink colour before adding to a sample.

iii) Phenolphthalein indicator solution: One g phenolphthalein was dissolved in 95 per cent ethyl alcohol and made up volume to 100 ml.

Procedure

Five g lipids were taken into an Erlenmeyer flask. Fifty ml neutralized isopropyl alcohol was added to it and sample was dissolved completely. Phenolphthalein indicator was added and titrated against 0.25 N NaOH to pink colour end point which persisted for 30 seconds.

$$\% \text{ FFA} = \frac{\text{ml} \times \text{N} \times \text{F} \times 100}{\text{Sample wt.} \times 1000}$$

where,

ml = ml of NaOH required

N = Normality of NaOH solution

F = Equivalent weight (282) of FFA (oleic acid)

3.6.3 Organoleptic acceptability

Products were evaluated organoleptically at weekly and monthly interval for *sev* and pickle, respectively during the storage period by the same procedure as mentioned in section 3.3.

3.7 Hypocholesterolaemic effect of *Moringa oleifera* leaves

Animal experiment

For obtaining animals (rats) application for permission for animal experiments was submitted to Institutional animal Ethics Committee (IAEC) and the meeting was held on October 7, 2005 and the objective of the study was approved by the Committee.

3.7.1 Selection of test animals

Weanling albino male rats weighing 30-40 g were obtained from the Disease Free Small Animal House, CCS Haryana Agricultural University, Hisar. The rats were randomly divided into

four groups, each consisting of seven rats fed following diets for 42 days.

Group I : Synthetic diet

Group II : Synthetic diet + cholesterol (1%)

Group III : Synthetic diet + cholesterol (1%) + *Moringa oleifera* leaves (5%)

Group IV : Synthetic diet + cholesterol (1%) + *Moringa oleifera* leaves (10%)

The rats were housed individually in polypropylene cages kept in air conditioned room maintained at 22 ± 1°C with 12 h light and dark cycle. Food and water were given *ad libitum.*

3.7.2 Composition of diets

Diets were prepared according to AIN-76 purified diets for rats (Table 3.1). The protein content of diet was adjusted after taking into account the protein content of test material. Cholesterol was added to all the diets at 1 per cent level except in group I. For the preparation of diets, the ingredients were mixed thoroughly and passed through 70 mesh sieve to ensure uniform distribution of vitamins and minerals.

Mineral and vitamin mixtures recommended by BARR Committee (1972) were used in the diet. The composition of diets has been given in Tables 3.2 and 3.3, respectively. The diets sufficient for one week's consumption were prepared at one time and stored in refrigerator.

3.7.3 Feeding and observation

The food and distilled water were given *ad libitum.* Weighed diet was given daily and unconsumed diet was collected, dried and weighed. The rats were initially weighed and then weighed on every alternate day. The feeding experiment lasted for 30 days. The rats fed on different diets were finally weighed after 30 days and gain in

Table 3.1 Composition of experimental diets (g/kg)

	Cellulose (without cholesterol)	**Cellulose (with cholesterol)**	***Moringa oleifera* leaves (5%)**	***Moringa oleifera* leaves (10%)**
Casein	153.85	153.85	132.99	112.15
Fat (hydrogenated veg. Oil)	50	50	50	50
Mineral mixture	40	40	40	40
Vitamin mixture	10	10	10	10
Sucrose	100	100	100	100
Choline chloride	2	2	2	2
Methionine	3	3	3	3
Cholesterol	-	10	10	10
Moringa oleifera leaves (5%)	-	-	50	-
Moringa oleifera leaves (10%)	-	-	-	100
Cellulose	50	50	-	-
Starch	591.15	591.15	602.1	572.85

Table 3.2 Composition of mineral mixture (AIN-76A)[a]

Ingredients	g/kg mineral mixture
Calcium phosphate (dibasic)	500.00
Sodium chloride	74.00
Potassium citrate monohydrate	220.00
Potassium sulphate	52.00
Magnesium oxide	24.00
Ferric citrate	6.00
Manganese carbonate	3.50
Zinc carbonate	1.60
Cupric carbonate	0.30
Potassium iodate	0.01
Sodium salenite, pentahydrate	0.01
Chromic potassium sulphate dodecahydrate	0.55
Starch	118.03

[a]Based on the National Academy of Science recommended levels for rats (BARR Committee on Animal Nutrition, 1972)

Table 3.3 Composition of vitamin mixture (AIN-76A)[a]

Ingredients	g/kg vitamin mixture
Thiamine hydrochloride	600.00
Riboflavin	600.00
Pyridoxin hydrochloride	700.00
Nicotinic acid	3000.00
Calcium pantothenate	1600.00
Folic acid	200.00
Biotin	20.00
Retinal acetate	1.00
Cholecalciferol	2.50
D-α-Tocopherol	To provide 500 I.U. of vitamin E activity
Menadione	5.00
Cynocobalamine	1.00
Starch (finely powdered)	To make 1000 g

[a]Based on the National Academy of Science recommended levels for rats (BARR Committee on Animal Nutrition, 1972)

weight during this period was recorded. Amount of food and protein intake during this period was calculated on dry matter basis.

3.7.4 Body fluid and organ

At the end of experiment on 30th day, the rats were anaesthetised with diethyl ether. Serum was separated by centrifuging the blood at 3000 rpm for 15 min and stored in screw capped vials kept in deep freeze until used for further analysis. Livers were excised, cleaned of adhering matter, blotted in filter paper and weighed. The organs were dried in oven at 60°C, cooled in desiccator, determined dried weight and kept for further analysis.

3.7.5 Biological evaluation of protein quality

Protein efficiency ratio (PER) was determined by the method of Chapman *et al.* (1959). The amount of food intake and protein intake during 30 days was calculated on dry matter basis. Gain in weight after 30 days was also calculated.

$$\text{PER} = \frac{\text{Gain in weight (g)}}{\text{Protein intake (g)}}$$

$$\text{FER} = \frac{\text{Gain in weight (g)}}{\text{Feed intake}}$$

3.7.6 Serum analysis

The serum was analysed for total cholesterol, HDL-cholesterol, LDL-cholesterol and triglycerides.

3.7.6.1 Total cholesterol

Total serum cholesterol was estimated with the help of fully automatic blood chemistry analyzer Polimak M-10/2 using single step reagent kit alongwith 2 water blanks and

2 cholesterol standards (100 mg/dl) and one serum control. 10 µl of serum was used for each sample.

3.7.6.2 HDL-cholesterol

Serum HDL-cholesterol was estimated with the help of fully automatic blood chemistry analyzer, Polimak M-10/2 using single step reagent kit along with 2 water blanks and 2 HDL-cholesterol. 10 µl of serum was used for each sample.

3.7.6.3 LDL-cholesterol

LDL-cholesterol in serum was estimated by using the formula of Friedewald *et al.* (1972).

LDL-cholesterol = Total cholesterol – HDL-cholesterol – $1/5^{th}$ triglycerides

3.7.6.4 VLDL-cholesterol

VLDL-cholesterol = Total cholesterol – (HDL-cholesterol + LDL-cholesterol)

3.7.6.5 Triglycerides

Serum triglycerides were estimated with the help of fully automatic blood chemistry analyzer, Polimak M-10/2 using single step reagent kit alongwith 2 water blanks and triglyceride standard (100 mg/dl) and one serum control. 10 µl of serum was used for each sample.

3.7.7 Liver analysis

3.7.7.1 Extraction and purification of lipids

The tissues were homogenized in liquid nitrogen with nitrogen with pastel and mortar and the lipids were extracted and purified by the method of Folch *et al.* (1957) as modified by Chauhan (1974).

The extracted lipid was treated with 19 volumes of cold chloroform : methanol (2:1, v/v) and kept aside for 10 minutes. After filtration through Whatman filter paper No. 41 saturated with

solvent mixture, the residue was again treated with 10 volumes of chloroform : methanol (1:2, v/v) containing five percent water. Both the filtrates were combined and an appropriate amount of chloroform was added to yield a final concentration of chloroform : methanol mixture (2:1, v/v). The filtrates contained practically all lipids of the liver and also contaminating non-lipid materials like peptides, free amino acids, sugars etc. and the residue contained proteins.

The lipids in the filtrates were freed from contaminating materials by the method of Folch *et al.* (1957) and Suzuki (1965). The filtrates in the stoppered cylinders were mixed thoroughly and agitated by adding two volumes of 0.88 percent aqueous potassium chloride. Non-lipid material was partitioned into the upper aqueous phase by keeping the cylinders undisturbed for 12 h at room temperature.

The upper phase was removed without disturbing the lower phase, by sucking it out with the help of a sharply pointed pipette attached to a vacupet. The lower phase was gently washed twice with the upper phase (chloroform : methanol : water, 3:48:47, v/v) containing no salt. The lower phase, after removing all the upper phase if turbid or cloudy, was made clear by addition of few drops of methanol. The lower phase now contained all lipids. The purified lipid extract was stored in the deep freezer for further estimation of total lipids in liver.

The purified lipid extract containing all the lipids was evaporated to dryness on a water bath at 60°C. The dried lipids were dissolved in 10 ml chloroform and stored in tightly capped vials for further estimation of various lipid components.

3.7.7.2 Total lipids

Total lipids in liver were estimated by gravimetrically by the method of Folch *et al.* (1957). A 0.5 ml of purified lipid extract was taken in a micro petriplate. The contents were dried to a constant weight by keeping the petriplate at 90° C in a hot air oven for 12hr. The weight of empty petriplate was recorded. The difference in the weight of petriplate with dried lipids and empty plate gave the amount of lipids present in 0.5ml of the lipid extract of the liver. The figure multiplied by 200 gave the amount of total lipids in mg/100g of the liver.

3.7.7.3 Total cholesterol

Total cholesterol in liver was estimated with fully automatic blood chemistry analyser as described earlier for estimation of total cholesterol of serum in section 3.7.6.1.

3.7.7.4 HDL-cholesterol

HDL-cholesterol in liver was estimated with fully automatic blood chemistry analyser as described earlier in section 3.7.6.2.

3.7.7.5 LDL-cholestrol

The LDL-cholesterol in liver was estimated by using the formula of Friedewald *et al.* (1972).

LDL-cholesterol = Total cholesterol – HDL-cholesterol – $1/5^{th}$ triglycerides

3.7.7.6 VLDL-cholesterol

VLDL-cholesterol = Total cholesterol – (HDL-cholesterol + LDL-cholesterol)

3.7.7.7 Triglycerides

Liver triglycerides were estimated with fully automatic blood chemistry analyser as described earlier in section 3.7.6.5.

3.8 Seeds of *Moringa oleifera* for purification of water

The method for purification of water using seeds of *Moringa oleifera* was standardised and samples were microbially analysed for standard plate count before and after purification. Three samples of water were taken i.e. fresh water, stored tank water and pond water.

3.8.1 Method for purification of water by seeds

i) Allowed the *Moringa* seed pods to dry naturally on the tree before harvesting them.

ii) Removed the seed husks, leaving a whitish kernel.

iii) Crushed the seed kernels to a powder with a stone or mortar.

iv) Mixed the powder with a small quantity of clean water in a small cup.

v) Poured the mixture through a tea strainer or sieved into a cup. Covered the strainer with a piece of clean cloth.

vi) Added the resulting milky fluid to the water to purify.

vii) Stirred quickly for 30 seconds, then slowly and regularly for five minutes.

viii) Covered the water and did not disturb it for at least an hour.

ix) The clean water was siphoned or poured off the top of the container.

3.8.2 Enumeration of total viable count in water

Sample diluent

i) **Ringer's solution**

Sodium chloride	2.25 g
Potassium chloride	0.11 g
Calcium chloride	0.12 g
Sodium bicarbonate	0.05 g
Distilled water	1000 ml

Dissolved the ingredients in distilled water. Distributed them in 9 ml volume in culture tubes. Sterilized in autoclave at 121°C for 15 minutes.

ii) **Normal saline solution (NSS):** It was prepared by dissolving 0.85 g sodium chloride in 100 ml distilled water. Sterilized at 121°C for 15 minutes.

b) Standard plate count agar (SPC agar)

Beef extract	5 g
Peptone	10 g
Sodium chloride	6 g
Agar-agar	15 g
Distilled water	1000 ml

Adjusted the pH to 7.2. Autoclaved at 121°C for 15 minutes.

Method

Preparations of dilutions

i) Mixed the water sample thoroughly by vigorous shaking of the container.

ii) Prepared a ten fold dilution (1:10) by transferring 1 ml water to 9 ml diluent such as Ringer's solution or NSS without touching the tip of the pipette to the next tube.

iii) Mixed the contents of 1:10 dilution with another sterile pipette and transferred 1 ml amount to 9 ml fresh diluent.

Plate inoculation

i) For this purpose pour plate method was used. After preparing appropriate dilutions, with fresh sterilize pipette transferred 1 ml quantity from each tube to duplicate petri dishes starting from the lowest dilution. Then added 15-20 ml melted SPC agar cooled to 45-48°C to each petri dish.

ii) Mixed the contents by rotating the agar plates gently in a horizontal plane and allowed to solidity.

iii) Petri dishes were incubated at 37°C for 24 h.

Observations

i) After 24 h incubation, observed the plates for development of colonies and selected the plate having 30-300 colonies for counting.

ii) Counted the colonies in the selected plate with the help of colony counter.

iii) Calculated the SPC of bacteria present per ml of water with the following formula:

SPC/ml water = $Y \times 10^Z$

where, Y = Total number of colonies counted

Z = Dilution factor

Statistical analysis

Statistical analysis of data was done using Complete Randomized Design according to standard method (Panse and Sukhatme, 1961).

Results And Discussion

Chapter-4

Results and Discussion

In the present study, efforts were made to develop some nutritious foods using various parts of *M. oleifera* (fresh as well as dried) and various cooking methods. Results of the present study have been presented and discussed under the following heads and sub-heads:

4.1 Nutritional composition of leaves, flowers pods and seeds of *M. oleifera*

4.1.1 Moisture

4.1.2 Ascorbic acid

4.1.3 β-carotene

4.1.4 Crude protein

4.1.5 Crude fat

4.1.6 Total ash

4.1.7 Crude fibre

4.1.8 Total dietary fibre

4.1.9 Total carbohydrates

4.1.10 Antinutrients

4.1.11 Total mineral composition

4.1.12 HCl-extractability of minerals

4.1.13 *In vitro* availability of Ca and Fe

4.1 Nutritional composition of leaves, flowers, pods and seeds of *Moringa oleifera*

Nutrient composition of different parts of *Moringa oleifera* i.e. leaves, flowers, pods and seeds have been presented in Tables 4.1 and 4.2.

4.1.1 Moisture

Moisture content in different parts of *Moringa oleifera* varied significantly ($P<0.05$) from each other, ranging from 6.87 to 75.65 percent on fresh weight basis. Pods had significantly ($P<0.05$) the highest (85.56%) whereas seeds (6.87%) had the lowest moisture

Table 4.1 Nutrient composition of leaves, flowers, pods and seeds of *Moringa oleifera* (on fresh weight basis)

Parts of *Moringa oleifera*	Moisture (%)	Ascorbic acid (mg/100 g)	β-carotene (mg/100 g)
Leaves	75.65 ± 0.01	219.38 ± 0.01	6.75 ± 0.01
Flowers	85.37 ± 0.01	52.83 ± 0.01	0.04 ± 0.01
Pods	85.56 ± 0.01	118.48 ± 0.01	0.09 ± 0.01
Seeds	6.87 ± 0.01	0.00 ± 0.00	0.01 ± 0.01
CD (P<0.05)	0.03	0.03	0.01

Values are mean ± SE of three independent determinations

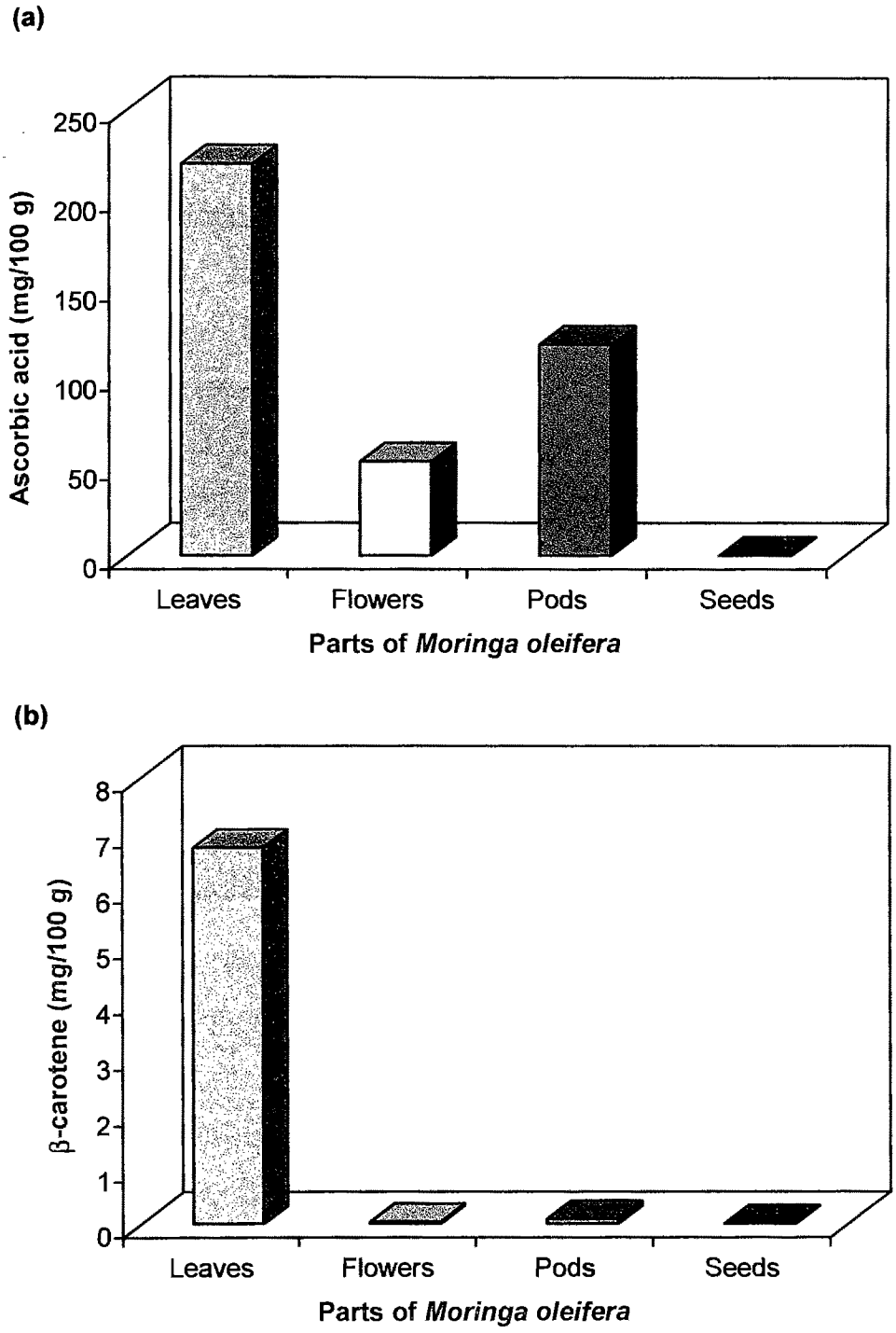


Fig. 1 : (a) Ascorbic acid and (b) β-carotene contents of leaves, flowers, pods and seeds of *Moringa oleifera*

content. Similar results have also been reported by various workers (Duke, 1983; Anhwange *et al.*, 2004; Sankhla *et al.*, 2005).

4.1.2 Ascorbic acid

The ascorbic acid content of various parts of *Moringa oleifera* has been depicted in Table 4.1 and Fig. 1. The ascorbic acid content of samples of *M. oleifera* ranged from 52.83 to 219.38 mg/100 g on fresh weight basis. The ascorbic acid content in the seeds was not detectable. The ascorbic acid content of leaves (219.38 mg/100 g) was significantly ($P<0.05$) higher than pods (118.48 mg/100 g) and flowers (52.83 mg/100 g). Leaves contained four times more ascorbic acid than flowers. Approximately, same range of ascorbic acid in *M. oleifera* pods has been reported by Duke (1983). Sankhla *et al.* (2005) quoted similar value for ascorbic acid in leaves.

4.1.3 β-carotene

The β-carotene content of leaves, flowers, pods and seeds has been depicted in Table 4.1 and Fig. 1. The β-carotene content of various parts of *M. oleifera* ranged from 0.01 to 6.75 mg/100 g. The β-carotene content of leaves was significantly ($P<0.05$) higher than pods, flowers and seeds. Further, pods contained significantly ($P<0.05$) higher β-carotene than flowers and seeds. Similar observations were recorded by some workers in the pods of *Moringa oleifera* (Gopalan *et al.*, 1989; Sankhla *et al.*, 2005).

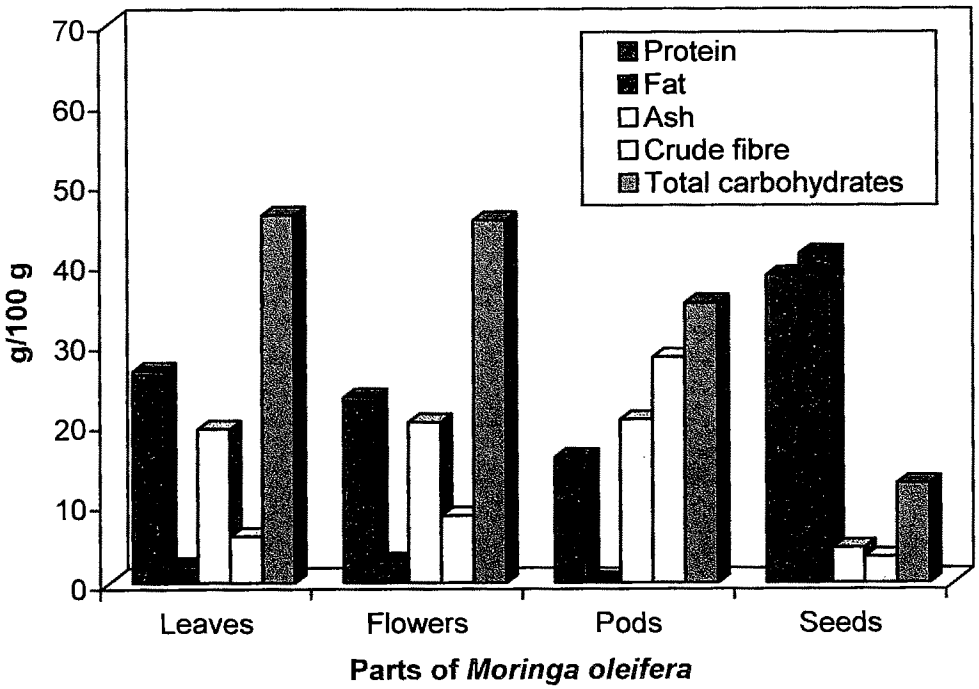
4.1.4 Crude protein

Crude protein content among the different parts of *M. oleifera* varied from 15.76 to 38.52 g/100 g; the highest being in seed and the lowest being in pods. Seeds of *Moringa oleifera* had significantly ($P<0.05$) higher protein content followed by leaves flowers and pods. Further, leaves had significantly ($P<0.05$) higher

Table 4.2 Nutrient composition of leaves, flowers, pods and seeds of *Moringa oleifera* (g/100 g, on dry weight basis)

Parts of *Moringa oleifera*	Crude protein	Crude fat	Total ash	Crude fibre	Dietary fibre			Total carbohydrates
					Total	Soluble	Insoluble	
Leaves	26.55 ± 0.01	2.13 ± 0.01	19.32 ± 0.01	5.83 ± 0.01	32.14 ± 0.01	7.28 ± 0.01	24.85 ± 0.03	46.06 ± 0.01
Flowers	23.28 ± 0.01	2.69 ± 0.01	20.14 ± 0.01	8.44 ± 0.01	36.48 ± 0.01	4.79 ± 0.01	31.69 ± 0.01	45.39 ± 0.01
Pods	15.76 ± 0.02	0.32 ± 0.01	20.48 ± 0.01	28.25 ± 0.01	59.32 ± 0.01	8.09 ± 0.01	51.17 ± 0.08	35.06 ± 0.01
Seeds	38.52 ± 0.02	41.21 ± 0.02	4.38 ± 0.02	3.26 ± 0.01	18.25 ± 0.02	3.52 ± 0.02	14.73 ± 0.01	12.50 ± 0.01
CD(P<0.05)	0.05	0.05	0.03	0.03	0.05	0.05	0.14	0.03

Values are mean ± SE of three independent determinations



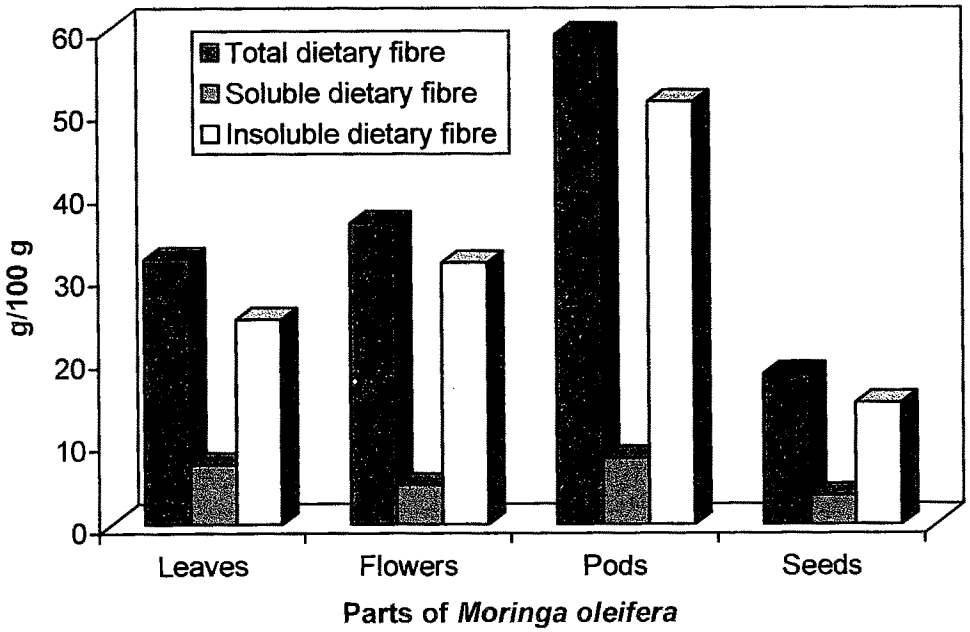


Fig. 2 : Nutrient composition of leaves, flowers, pods and seeds of *Moringa oleifera*

protein content than flowers and pods. The present findings are almost in accordance with those reported by earlier workers (Duke 1983; Awasthi and Tandon, 1988; Gopalan *et al.*, 1989; Islam *et al.*, 2004; Kapoor *et al.*, 2004; Sankhla *et al.*, 2005).

4.1.5 Crude fat

A wide range of crude fat content i.e. 0.32 to 41.21 g/100 g (DM basis) was observed in different parts of *M. oleifera*. Seeds had significantly ($P<0.05$) higher fat content followed by flowers, leaves and pods. Anhwange *et al.* (2004) reported 41.58 percent (DM basis) of fat content in *M. oleifera* seeds which was almost similar to that of the present study.

The values for fat content of *M. oleifera* leaves and pods in the present study corroborate with those reported by Duke (1983), Gopalan *et al.* (1989) and Sankhla *et al.* (2005).

4.1.6 Total ash

Total ash content of leaves, flowers, pods and seeds of *M. oleifera* ranged from 4.38 to 20.48 g/100 g (Table 4.2, Fig. 2). Seeds had the minimum whereas pods had the maximum ash content. Total ash content of pods was significantly ($P<0.05$) higher than flowers, leaves and seeds . Similar results for ash content have been reported in previous studies for various parts of *M. oleifera* (Duke, 1983; Gopalan *et al.*, 1989; Islam *et al.*, 2004).

4.1.7 Crude fibre

A wide range 3.26 to 28.25 g/100 g (DM basis) was observed in different parts of *M. oleifera*. Pods of *M. oleifera* had significantly ($P<0.05$) higher crude fibre content as compared to flowers, leaves and seeds. Seeds had the minimum amount of crude fibre. A significant difference ($P<0.05$) also existed in the crude fibre content of leaves and flowers on dry matter basis. The findings of

the present study are consistent with those reported earlier by Duke (1983), Awasthi and Tandon (1998) and Sankhla *et al.* (2005).

4.1.8 Total dietary fibre

Total dietary fibre content of different parts of *M. oleifera* ranged from 18.25 to 59.32 g/100 g and differed significantly ($P<0.05$) among themselves (Table 4.2 and Fig.2). Pods had the maximum (59.32 g/100 g) amount of total dietary fibre followed by flowers (36.48 g/100 g); the minimum total dietary fibre content was observed in seeds (18.25 g/100 g).

4.1.8.1 Soluble dietary fibre

The soluble dietary fibre content among the various parts of *M. oleifera* ranged from 3.52 to 7.28 g/100 g (Table 4.2). Pods had the maximum (8.09 g/100 g) content followed by flowers (7.79 g/100 g), leaves (7.28 g/100 g) and seeds (3.52 g/100 g) in descending order. There was a significant ($P<0.05$) difference in soluble dietary fibre content of various parts i.e. pods, leaves, flowers and seeds of *M. oleifera*.

4.1.8.2 Insoluble dietary fibre

A wide range of insoluble dietary fibre content i.e. 14.73 to 51.17 g/100 g (Table 4.2) was determined; the highest being in pods and the lowest being in seeds. The insoluble dietary fibre content of pods, flowers, leaves and seeds of *M. oleifera* varied significantly ($P<0.05$) from each other.

4.1.9 Total carbohydrates

Total carbohydrate content of samples of *M. oleifera* ranged from 12.50 to 46.06 g/100 g on dry matter basis. Leaves contained the highest content of total carbohydrates followed by flowers, pods and seeds in descending order. The total carbohydrates content of leaves was significantly ($P<0.05$) higher than flowers, pods and seeds of *M. oleifera*. Further, flowers contained significantly higher

total carbohydrate than pods and seeds. Seeds had the minimum amount of total carbohydrate i.e. 12.50 g/100 g on DM basis. Awasthi and Tandon (1988) reported 12.60 g carbohydrate in *M. oleifera* leaves on fresh weight basis.

4.1.10 Antinutrients

4.1.10.1 Oxalic acid

The oxalic acid content ranged from 264.99 to 403.80 mg/100 g among the various parts of *Moringa oleifera* (Table 4.3, Fig. 3). Pods had the maximum content (403.80 mg/100 g) followed by leaves (396.49 mg/100 g), flowers (281.52 mg/100 g) and seeds (264.99 mg/100 g). The oxalic acid content of all the parts of *Moringa oleifera* differed significantly ($P<0.05$) among themselves. Sankhla *et al.* (2005) determined oxalic acid content in *M. oleifera* leaves and value was found to be 101.2 mg/100 g on fresh weight basis.

4.1.10.2 Phytic acid

Phytic acid is known to be the major storage form of phosphorus and is considered to be an antinutritive factor. The phytic acid content ranged from 0.01 to 1.99 g/100 g among various parts of *M. oleifera* (Table 4.3); maximum being in leaves, whereas the minimum being in the seeds. The phytic acid content of flowers and pods was 1.06 and 1.83 g/100 g, respectively. The phytic acid content of various parts of *M. oleifera* differed significantly among themselves. Anhwange *et al.* (2004) analysed seeds of *M. oleifera* and phytic acid was also found to be 10.18 mg/100 g, which corroborated with the present finding.

4.1.10.3 Saponins

Range of saponins in four different parts of *M. oleifera* i.e. leaves, flowers, pods and seeds varied from 0.82 to 2.21 g/100 g (Table 4.3, Fig. 3). Seeds had the maximum (2.21 g/100 g) amount

Table 4.3 **Antinutritional factors in leaves, flowers, pods and seeds of *Moringa oleifera* pods (on dry weight basis)**

Parts of *Moringa oleifera*	Oxalic acid (mg/100 g)	Phytic acid (g/100 g)	Saponins (g/100 g)	Trypsin inhibitor activity (TIU/g)
Leaves	396.49 ± 0.02	1.99 ± 0.01	1.19 ± 0.01	0.04 ± 0.01
Flowers	281.52 ± 0.02	1.06 ± 0.01	0.82 ± 0.01	0.01 ± 0.01
Pods	403.80 ± 0.03	1.83 ± 0.02	1.04 ± 0.02	0.01 ± 0.01
Seeds	264.99 ± 0.02	0.01 ± 0.01	2.21 ± 0.01	0.01 ± 0.01
CD (P<0.05)	0.08	0.04	0.03	0.02

Values are mean ± SE of three independent determinations
Trypsin inhibitor units: One unit of trypsin was defined as the amount of enzymes which converted one mg casein in TCA soluble components at 37°C for 20 minutes at pH 7.6

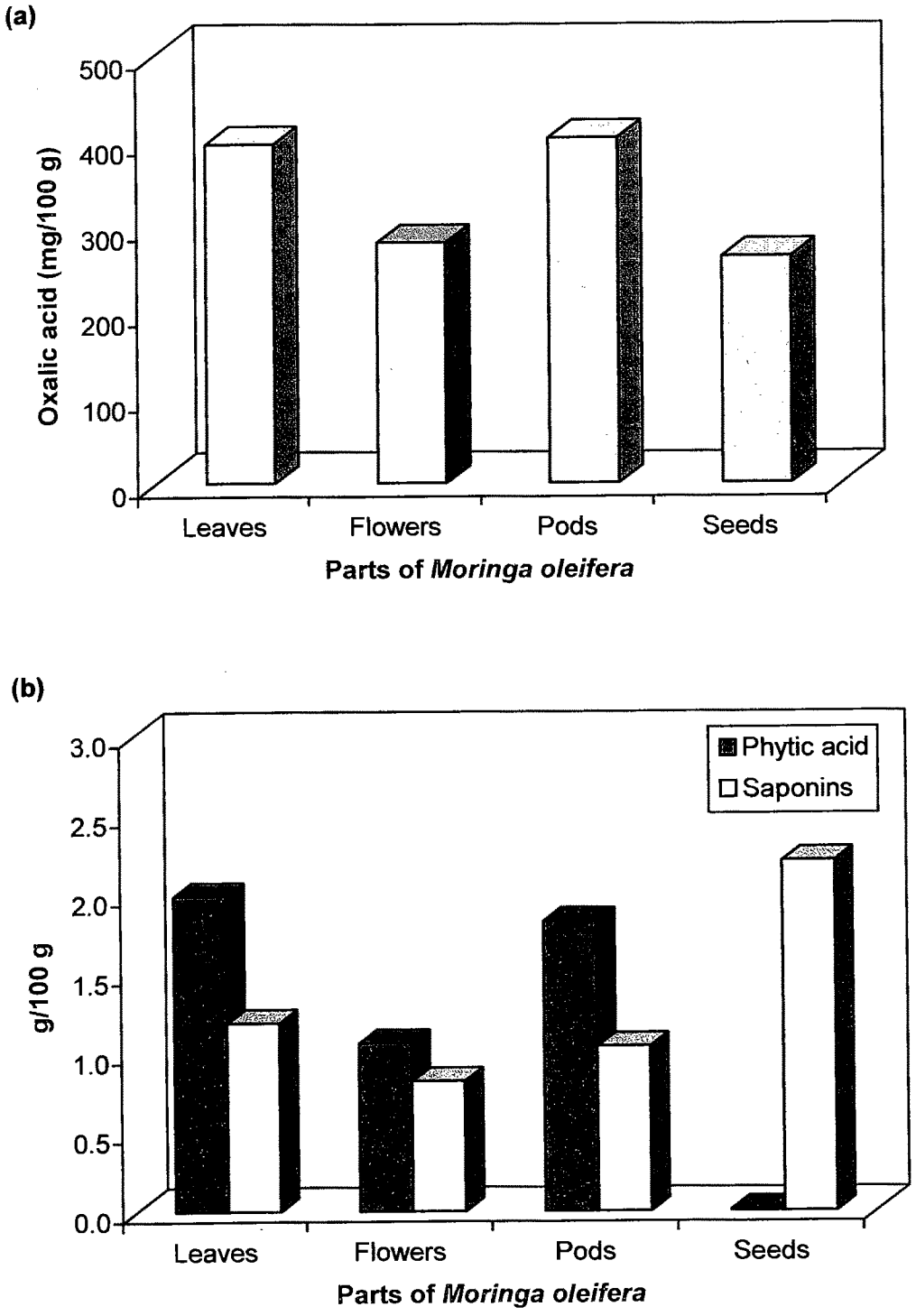


Fig. 3 : Antinutritients (a) Oxalic acid (b) Phytic acid and Saponins in leaves, flowers, pods and seeds of *Moringa oleifera*

of saponins followed by leaves (1.19 g/100 g), pods (1.04 g/100 g) and minimum (0.82 g/100 g) being in flowers. The saponin content of all the parts of *M. oleifera* differed significantly among themselves. Gupta *et al.* (1989) and Anhwange *et al.* (2004) determined similar range of saponin in leaves and seeds of *M. oleifera*.

4.1.10.4 Trypsin inhibitor activity

Trypsin inhibitors are known to affect the digestibility of proteins. The trypsin inhibitor activity of leaves was significantly ($P<0.05$) higher than that in the flowers, pods and seeds (Table 4.3). A non-significant difference was noticed in trypsin inhibitor content of flower, pods and seeds of *M. oleifera*.

4.1.11 Total mineral composition

Various parts of *M. oleifera* i.e. leaves, flowers, pods and seeds were analysed for total calcium, phosphorus, iron, zinc and potassium (Tables 4.4 and 4.5, Fig. 4) and significant ($P<0.05$) differences were noticed among the values.

4.1.11.1 Calcium

Total calcium content ranged from 136.33 mg to 1999.81 mg/100 g among different parts of *M. oleifera*. Pods had the minimum (136.33 mg/100 g) calcium content whereas leaves had the maximum (1999.81 mg/100 g). The calcium contents of seeds and flowers were 1985.33 and 232.00 mg/100 g, respectively. A significant ($P<0.05$) difference occurred in calcium content of various parts of *M. oleifera*. Awasthi and Tandon (1988) and Sankhla *et al.* (2005) reported 430 mg calcium in *M. oleifera* leaves on fresh weight basis.

Table 4.4 **Total and HCl-extractable calcium, phosphorus and iron contents (mg/100 g) of leaves, flowers, pods and seeds of *Moringa oleifera* (on dry weight basis)**

Parts of *Moringa oleifera*	Calcium			Phosphorus			Iron		
	Total	HCl-extractable	% extractability	Total	HCl-extractable	% extractability	Total	HCl-extractable	% extractability
Leaves	1999.81 ± 0.28	1637.64 ± 0.59	81.89 ± 0.02	203.53 ± 0.01	156.83 ± 0.45	77.47 ± 0.01	27.55 ± 0.02	18.10 ± 0.05	65.30 ± 0.02
Flowers	232.00 ± 0.02	187.10 ± 0.04	80.65 ± 0.01	261.31 ± 0.41	202.51 ± 0.29	77.52 ± 0.01	5.16 ± 0.02	3.30 ± 0.02	64.34 ± 0.02
Pods	136.33 ± 0.18	109.50 ± 0.16	80.32 ± 0.01	320.00 ± 0.04	244.41 ± 0.33	76.39 ± 0.02	21.24 ± 0.02	13.76 ± 0.06	65.45 ± 0.03
Seeds	1985.33 ± 0.02	1574.83 ± 0.30	79.32 ± 0.01	132.53 ± 0.01	100.07 ± 0.04	75.53 ± 0.01	75.86 ± 0.32	47.95 ± 0.05	62.73 ± 0.02
CD (P<0.05)	0.54	1.13	0.05	0.69	1.05	0.06	0.53	0.17	0.08

Values are mean ± SE of three independent determinations

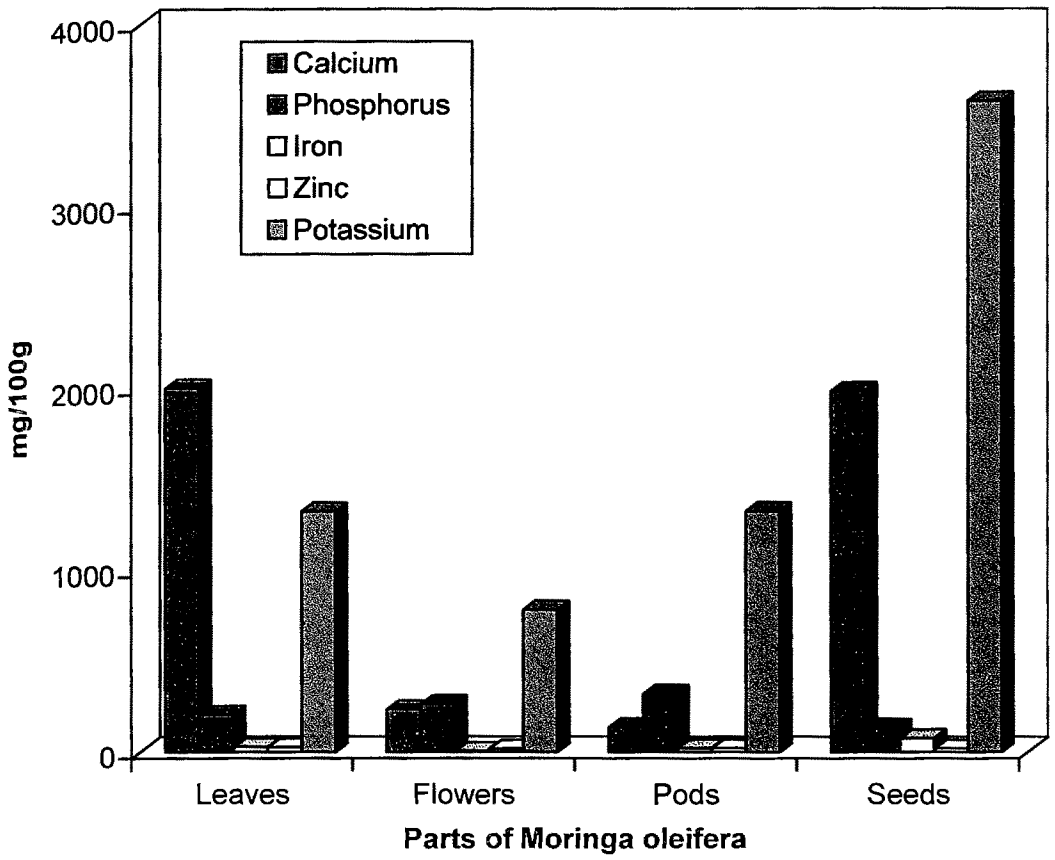


Fig. 4 : Total mineral composition of leaves, flowers, pods and seeds of *Moringa oleifera*

4.1.11.2 Phosphorus

Total phosphorus content varied from 132.53 to 320.0 mg/100 g among various parts of *M. oleifera* and these values were significantly ($P<0.05$) different among themselves (Table 4.4).

4.1.11.3 Iron

Seeds had the maximum iron content (75.86 mg/100 g) whereas flowers (5.16 mg/100 g) had the minimum (Table 4.4). A significant ($P<0.05$) difference in total iron content of all the parts of *M. oleifera* was observed. Total iron content of leaves and pods was 27.55 and 21.24 mg/100 g, respectively. Sankhla *et al.* (2005) determined micronutrient composition of *M. oleifera* leaves and they reported iron content to be 0.8 mg/100 g on fresh weight basis.

4.1.11.4 Zinc

Total zinc content ranged from 16.32 to 25.48 mg/100 g in leaves, flowers, pods and seeds of *M. oleifera*. Leaves had the maximum (25.48 mg/100 g) zinc content whereas flowers had the minimum (16.32 mg/100 g). A significant ($P<0.05$) difference was observed among various parts of *M. oleifera* (Table 4.5).

4.1.11.5 Potassium

A wide range in total potassium content (783.23 to 3850.52 mg/100 g) was determined; the highest being in seeds and the lowest being in flowers (Table 4.5). Total potassium content of all the four parts of *Moringa oleifera* i.e. leaves, flowers, pods and seeds differed significantly ($P<0.05$) among themselves.

4.1.12 HCl-extractability of minerals

Tables 4.4 and 4.5 depict the HCl-extractability of various minerals including calcium, phosphorus, iron, zinc and potassium. Extractability of minerals is generally affected by the presence of

Table 4.5 **Total and HCl-extractable zinc and potassium contents (mg/100 g) of leaves, flowers, pods and seeds of *Moringa oleifera* (on dry weight basis)**

Parts of *Moringa oleifera*	Zinc			Potassium		
	Total	HCl-extractable	Percent extractability	Total	HCl-extractable	Percent extractability
Leaves	25.48 ± 0.01	19.12 ± 0.02	75.14 ± 0.01	1323.90 ± 0.29	1004.18 ± 0.09	75.81 ± 0.01
Flowers	16.32 ± 0.01	11.64 ± 0.02	73.40 ± 0.04	783.23 ± 0.36	586.44 ± 0.06	74.82 ± 0.01
Pods	20.51 ± 0.01	14.76 ± 0.02	74.53 ± 0.02	1324.50 ± 0.62	998.83 ± 0.37	75.32 ± 0.01
Seeds	18.63 ± 0.02	12.57 ± 0.02	70.31 ± 0.02	3850.52 ± 0.04	2714.92 ± 1.51	70.54 ± 0.02
CD ($P<0.05$)	0.04	0.07	0.10	1.28	2.55	0.05

Values are mean ± SE of three independent determinations

antinutrients including phytic acid which is known to form a protein-phytate-mineral-complex (Prattley *et al.*, 1982).

4.1.12.1 Calcium

HCl-extractability of calcium ranged from 79.32 to 81.89 percent in various parts of *Moringa oleifera* i.e. leaves, flowers, pods and seeds; a significant difference ($P<0.05$) was observed in extractability of calcium among themselves (Table 4.4).

4.1.12.2 Phosphorus

A non-significant difference was observed in the extractability of phosphorus in leaves and flowers whereas pods and seeds of *M. oleifera* differed significantly ($P<0.05$) in their extractability. Seeds had minimum (75.53%) whereas flowers had maximum (77.52%) phosphorus extractability.

4.1.12.3 Iron

A significant difference ($P<0.05$) was observed for HCl-extractability of iron among various parts of *M. oleifera* i.e. leaves, pods, flowers and seeds. The range of percent HCl-extractability of iron varied from 62.73 to 65.45 percent in different parts; it was maximum in pods (65.45%) and minimum in seeds (62.73%).

4.1.12.4 Zinc

HCl-extractability of zinc varied from 70.31 to 75.14 percent; leaves had the maximum (75.14%) whereas seeds had the minimum (70.31%) HCl-extractability of zinc. A significant difference ($P<0.05$) was observed in percent extractability of zinc among various parts of *Moringa oleifera.*

4.1.12.5 Potassium

HCl-extractability of potassium ranged from 70.54 to 75.81 percent. Percent extractability of potassium in leaves was significantly higher than rest of parts of *Moringa oleifera.* Percent extractability of seed was minimum i.e. 70.54 percent.

4.1.13 *In vitro* availability of calcium and iron

Calcium and iron availability (*in vitro*) ranged from 21.52 to 25.56 and 10.27 to 13.51, respectively in various parts of *M. oleifera* (Fig. 5). Calcium availability was maximum in leaves followed by seeds, pods and flowers whereas *in vitro* availability of iron was maximum in leaves followed by pods, flowers and seeds of *M. oleifera* in descending order. A significant difference ($P<0.05$) was observed among different parts of *M. oleifera* in calcium as well as iron availability. Variation in extractabilities might be due to level of antinutrients which are known to hinder the bioavilability of minerals including calcium and iron.

4.2 Products development and their organoleptic evaluation

Different types of products were developed from various parts of *M. oleifera* i.e. leaves, pods, flowers and seeds. From the consumer point of view, the sensory characteristics were evaluated by the panelists who were quite familiar with the product quality and sensory characteristics. The data regarding sensory charactristics of various products are given in Tables 4.7 to 4.12.

4.2.1 Leaves

Various products i.e. chutney, leaves *bhuji*, potato and leaves vegetable and *pakora* were prepared by using fresh leaves. All the products prepared from fresh leaves were acceptable (Table 4.7). The colour, appearance, aroma and texture of chutney were 'liked moderately' whereas taste of chutney was 'neither liked nor disliked' by the panelists. Leaves *bhuji* and potato and leaves vegetable were 'liked moderately' for all the sensory parameters i.e. colour, appearance, aroma, texture, taste and overall acceptability.

Similar trend was followed in sensory evaluation of *pakora* i.e. which was 'liked moderately' by the panelists. The overall

Table 4.6 ***In vitro*** **availability (%) of calcium and iron of leaves, flowers, pods and seeds of Moringa** ***oleifera*** **(on dry weight basis)**

Parts of *Moringa oleifera*	Calcium		Iron	
	Total (mg/100 g)	Percent availability (*in vitro*)	Total (mg/100 g)	Percent availability (*in vitro*)
Leaves	1999.81 ± 0.28	25.56 ± 0.03	27.55 ± 0.03	13.51 ± 0.05
Flowers	232.00 ± 0.02	21.52 ± 0.38	5.16 ± 0.02	11.43 ± 0.02
Pods	136.33 ± 0.17	22.57 ± 0.12	21.24 ± 0.02	12.15 ± 0.07
Seeds	1985.33± 0.02	23.93 ± 0.67	75.85 ± 0.32	10.27 ± 0.03
CD (P<0.05)	0.54	0.07	0.53	0.05

Values are mean ± SE of three independent determinations

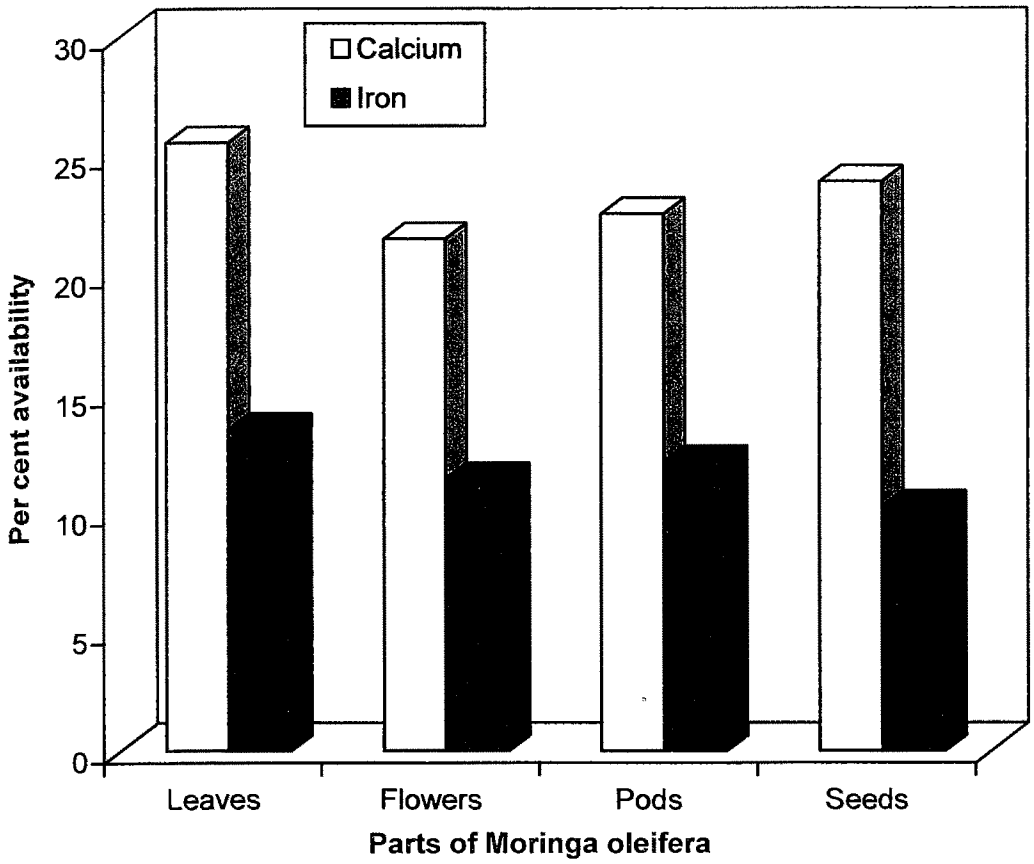


Fig. 5 : *In vitro* availability (%) of calcium and iron of leaves, flowers, pods and seeds of *Moringa oleifera*

Table 4.7 Mean scores of organoleptic acceptability of products prepared from *Moringa oleifera* leaves (on fresh weight basis)

Food products	Colour	Appearance	Aroma	Texture	Taste	Overall acceptability
Chutney	7.60 ± 0.16	7.60 ± 0.16	7.10 ± 0.31	7.30 ± 0.26	5.90 ± 0.31	7.10 ± 0.19
Leaves *bhuji*	7.80 ± 0.13	7.80 ± 0.13	7.60 ± 0.16	7.80 ± 0.13	7.50 ± 0.16	7.70 ± 0.12
Potato and leaves vegetable	7.80 ± 0.13	7.70 ± 0.15	7.80 ± 0.13	7.80 ± 0.13	7.60 ± 0.16	7.74 ± 0.13
Pakora	7.70 ± 0.15	7.70 ± 0.15	7.40 ± 0.22	7.70 ± 0.15	6.90 ± 0.10	7.48 ± 0.13

Values are mean ± SE of scores given by ten panelists on 9-point hedonic scale

acceptability of potato and leaves vegetable was the highest among all the products prepared from fresh *Moringa oleifera.*

4.2.2 Products prepared from dried *M. oleifera* leaf powder

Data on important sensory characteristics viz colour, appearance, aroma, texture, taste and overall acceptability of products i.e. *sev* and noodles prepared from *M. oleifera* leaf powder are presented in Table 4.8. The colour, appearance, aroma, texture, taste and overall acceptability of control *sev* i.e. prepared from 100% bengal gram flour and *sev* prepared from 10 percent *M. oleifera* leaf powder fell in the category of 'liked moderately'. However, the *sev* prepared from 20 percent *M. oleifera* leaf powder was 'liked moderately' in terms of colour and texture but 'liked slightly' in appearance, aroma, taste and overall acceptability.

Noodles prepared from 100% refined flour i.e. control were 'liked very much' as far as their colour, appearance, aroma, texture and taste were concerned. Noodles prepared from 10 percent *M. oleifera* leaf powder were 'liked very much' in aroma, texture and taste but 'liked moderately' in terms of colour and appearance.

The colour and appearance of the third type of noodles i.e. prepared from 20 percent *M. oleifera* leaf powder were 'liked slightly' but aroma was 'liked moderately'. The texture and taste of third type of noodles were 'neither liked nor disliked' by the judges. The overall acceptability which was the mean score of colour, appearance, aroma, texture and taste was 'liked slightly'.

4.2.3 Flowers

Various types of products prepared by using fresh flowers as given in Table 4.9 were found to be acceptable in terms of sensory evaluation. The colour, appearance, aroma and texture of the products i.e. flower *bhuji*, pea and flower vegetable, green gram and flower vegetable and *raita* were 'liked moderately' by the panelists.

Table 4.8 **Mean scores of organoleptic acceptability of products prepared from *Moringa oleifera* leaves (on dry weight basis)**

Food products	Colour	Appearance	Aroma	Texture	Taste	Overall acceptability
Sev						
Bengal gram flour (control 100%)	7.80 ± 0.13	7.60 ± 0.16	7.80 ± 0.13	7.60 ± 0.16	7.60 ± 0.16	7.68 ± 0.14
Bengal gram flour (90%) + *Moringa oleifera* leaves (10%)	7.20 ± 0.20	7.00 ± 0.15	7.20 ± 0.13	7.30 ± 0.15	7.00 ± 0.00	7.12 ± 0.10
Bengal gram flour (80%) + *Moringa oleifera* leaves (20%)	7.20 ± 0.20	6.90 ± 0.10	6.90 ± 0.10	7.00 ± 0.00	6.50 ± 0.17	6.90 ± 0.09
Noodles						
Refined flour (control 100%))	8.00 ± 0.00	8.00 ± 0.01	8.00 ± 0.01	8.00 ± 0.01	8.00 ± 0.01	8.00 ± 0.01
Refined flour + *Moringa oleifera* leaves (10%)	7.90 ± 0.28	7.90 ± 0.28	8.00 ± 0.21	8.10 ± 0.18	8.10 ± 0.18	8.00 ± 0.17
Refined flour + *Moringa cleifera* leaves (20%)	6.40 ± 0.30	6.40 ± 0.30	7.00 ± 0.29	5.90 ± 0.28	5.80 ± 0.25	6.30 ± 0.22

Values are mean ± SE of scores given by ten panelists on 9-point hedonic scale

Taste of the pea and flower vegetable was 'liked very much' whereas taste of flower *bhuji*, green gram and flower vegetable and *raita* was 'liked moderately'. The colour, aroma, texture, taste and overall acceptability of cutlets was 'liked very much' except appearance which was 'liked moderately'. Overall acceptability scores of all the developed products showed that they were acceptable to human palate but cutlet were 'liked very much' as compared to other products which were 'liked moderately'.

4.2.4 Pods

Data on important sensory characteristics viz colour, appearance, aroma, texture, taste and overall acceptability of products prepared from fresh as well as dried *M. oleifera* pods are presented in Tables 4.10 and 4.11.

The colour and appearance of pickle were 'liked very much' whereas aroma, texture, taste and overall acceptability were 'liked moderately' by the panelists. The pod vegetable, potato and pod vegetable, *sambhar* were 'liked moderately' in terms of colour, appearance, aroma, texture, taste and overall acceptability.

The appearance of cutlets prepared was 'liked very much' whereas colour, aroma, texture, taste and overall acceptability of cutlets were 'liked moderately'.

Overall acceptability which was the mean score of colour, appearance, aroma, texture and taste had shown that all the products were 'liked moderately' but pickle score was higher as compared to other products.

4.2.5 Products prepared from dried *M. oleifera* pod powder

Two different types of *sev* by using *M. oleifera* pod powder i.e. at 10 percent and 20 percent levels were prepared (Table 4.11). The sensory evaluation showed that *sev* prepared from 100% bengal gram flour i.e. control and 10 percent *M. oleifera* pod powder

Table 4.9 Mean scores of organoleptic acceptability of products prepared from *Moringa oleifera* flowers (on fresh weight basis)

Food products	Colour	Appearance	Aroma	Texture	Taste	Overall acceptability
Flower *bhuji*	7.90 ± 0.10	7.70 ± 0.15	7.90 ± 0.10	7.90 ± 0.10	7.90 ± 0.17	7.86 ± 0.10
Pea and Flower vegetable	7.80 ± 0.13	7.70 ± 0.15	7.90 ± 0.10	7.90 ± 0.10	8.00 ± 0.14	7.86 ± 0.08
Green gram and Flower vegetable	7.60 ± 0.16	7.60 ± 0.16	7.80 ± 0.13	7.80 ± 0.13	7.90 ± 0.17	7.74 ± 0.12
Raita	7.90 ± 0.10	7.70 ± 0.15	7.80 ± 0.13	7.70 ± 0.15	7.80 ± 0.13	7.78 ± 0.08
Cutlets	8.10 ± 0.18	7.90 ± 0.23	8.10 ± 0.17	8.00 ± 0.21	8.00 ± 0.21	8.02 ± 0.18

Values are mean ± SE of scores given by ten panelists on 9-point hedonic scale

Table 4.10 **Mean scores of organoleptic acceptability of products prepared from *Moringa oleifera* pods (on fresh weight basis)**

Food products	Colour	Appearance	Aroma	Texture	Taste	Overall acceptability
Pickle	8.30 ± 0.15	8.10 ± 0.23	7.70 ± 0.21	7.90 ± 0.23	7.80 ± 0.29	7.94 ± 0.18
Pod vegetable	7.50 ± 0.27	7.40 ± 0.27	7.60 ± 0.16	7.10 ± 0.31	6.60 ± 0.40	7.20 ± 0.24
Potato and pod vegetable	7.50 ± 0.17	7.40 ± 0.16	7.40 ± 0.16	7.50 ± 0.17	7.00 ± 0.21	7.28 ± 0.15
Sambhar	7.50 ± 0.17	7.80 ± 0.13	7.80 ± 0.13	7.80 ± 0.13	7.70 ± 0.15	7.78 ± 0.10
Cutlets	7.60 ± 0.16	8.00 ± 0.00	7.80 ± 0.13	7.90 ± 0.10	7.90 ± 0.10	7.90 ± 0.03

Values are mean ± SE of scores given by ten panelists on 9-point hedonic scale

were 'liked moderately' in terms of colour, appearance, aroma, texture, taste and overall acceptability. The colour and texture of *sev* prepared by using 20 percent *M. oleifera* pod powder were 'liked moderately' whereas appearance, aroma, taste and overall acceptability were 'liked slightly'.

The noodles prepared by using 100 percent refined flour i.e. control were 'liked very much' for all the sensory parameters. The noodles prepared by 10 percent *M. oleifera* pods were 'liked moderately' by panelists in terms of colour, appearance, aroma, texture and taste and overall acceptability. The noodles prepared by using 20 percent *M. oleifera* pod powder had overall acceptability score in 'liked slightly' category.

4.2.6 Seeds

Ladoo and weaning food mixture were prepared by incorporating different levels of seed powder. Sensory evaluation of *ladoo* prepared from 100 percent wheat flour (control) fell in category of 'liked moderately'. The colour, appearance, aroma and texture of *ladoo* prepared from 10 percent and 20 percent *M. oleifera* seed powder were 'liked moderately'. Taste of both *ladoo* was 'disliked moderately' whereas the overall acceptability which was in the category of 'liked slightly'.

4.2.7 Weaning food mixture

The weaning food mixture prepared from malted wheat flour, roasted bengal gram flour, skim milk powder and sugar was 'liked moderately' in terms of colour, appearance, aroma. On the other hand, weaning food mixture incorporating *M. oleifera* seed powder at 5 percent and 10 percent levels were 'disliked very much' in taste and were not acceptable to human palate.

Table 4.11 **Mean scores of organoleptic acceptability of products prepared from *Moringa oleifera* pods (on dry weight basis)**

Food products	Colour	Appearance	Aroma	Texture	Taste	Overall acceptability
Sev						
Bengal gram flour (Control 100%)	7.80 ± 0.13	7.60 ± 0.16	7.80 ± 0.13	7.60 ± 0.16	7.60 ± 0.16	7.68 ± 0.13
Bengal gram flour (90%) + *Moringa oleifera* pods (10%)	7.30 ± 0.15	7.30 ± 0.15	7.20 ± 0.13	7.30 ± 0.15	7.10 ± 0.18	7.24 ± 0.14
Bengal gram flour (80%) + *Moringa oleifera* pods (20%)	7.00 ± 0.14	6.90 ± 0.10	6.90 ± 0.10	7.00 ± 0.00	6.90 ± 0.10	6.94 ± 0.06
Noodles						
Refined flour (control 100%)	8.00 ± 0.00	8.00 ± 0.00	8.00 ± 0.00	8.00 ± 0.00	8.00 ± 0.00	8.00 ± 0.00
Refined flour (90%) + *Moringa oleifera* pods (10%)	7.50 ± 0.17	7.50 ± 0.17	7.30 ± 0.15	7.50 ± 0.15	7.20 ± 0.20	7.40 ± 0.15
Refined flour (80%) + *Moringa oleifera* pods (20%)	6.70 ± 0.15	6.20 ± 0.13	6.30 ± 0.15	6.10 ± 0.10	6.10 ± 0.10	6.26 ± 0.09

Values are mean ± SE of scores given by ten panelists on 9-point hedonic scale

Table 4.12 ***Mean scores of organoleptic acceptability of products prepared from Moringa oleifera seeds (on dry weight basis)***

Food products	Colour	Appearance	Aroma	Texture	Taste	Overall acceptability
Ladoo						
Wheat flour (control 100%)	7.30 ± 0.21	7.30 ± 0.21	7.50 ± 0.22	7.50 ± 0.22	7.50 ± 0.22	7.42 ± 0.21
Wheat flour (90%) + *Moringa oleifera* seeds (10%)	7.20 ± 0.20	7.20 ± 0.20	6.80 ± 0.29	7.00 ± 0.26	3.20 ± 0.33	6.48 ± 0.28
Wheat flour (80%) + *Moringa oleifera* seeds (20%)	7.00 ± 0.25	7.20 ± 0.20	6.80 ± 0.29	7.00 ± 0.25	3.00 ± 0.42	6.20 ± 0.24
Weaning food mixture						
Control (W+B+SM+S) (5: 2 : 2 :1)	7.60 ± 0.16	7.60 ± 0.16	7.60 ± 0.16	7.40 ± 0.16	7.60 ± 0.16	7.56 ± 0.15
WBSMS + *Moringa oleifera* seeds (10%)	7.10 ± 0.28	7.10 ± 0.28	7.00 ± 0.25	6.60 ± 0.48	2.20 ± 0.20	6.00 ± 0.25
WBSMS + *Moringa oleifera* seeds (20%)	7.00 ± 0.26	7.00 ± 0.26	6.80 ± 0.29	6.60 ± 0.48	1.60 ± 0.33	5.80 ± 0.26

Values are mean ± SE of scores given by ten panelists on 9-point hedonic scale
W = Whole wheat flour (malted) B = Bengal gram flour (roasted)
S = Skimmed milk powder S = Sugar

Nutritional composition of developed products

Various products fresh as well as those incorporating dried powder from different parts of *M. oleifera* i.e. leaves, flowers, pods using different processing treatments were nutritionally analysed for the same parameters as employed for raw leaves, flowers, pods and seeds.

4.3 Nutritional composition of products prepared from fresh leaves

Products prepared from *M. oleifera* fresh leaves were chutney, leaves *bhuji*, potato and leaves vegetable and *pakora*. The products prepared from dried *M. oleifera* leaf powder included *sev* and noodles.

4.3.1 Moisture

The moisture content of *M. oleifera* fresh leaves products have been presented in Table 4.13.

Moisture content of all fresh products was significantly ($P<0.05$) different from each other. Maximum moisture content (77.54%) was found in chutney; the minimum was in *pakora* (54.24%). Chutney had significantly ($P<0.05$) higher moisture content than rest of the products including leaves *bhuji*, potato and leaves vegetable and *pakora*. Lowest moisture content in *pakora* and vegetables might be due to evaporation of moisture in various processing treatments like frying and blanching whereas chutney was prepared only by grinding of ingredients.

4.3.2 Ascorbic acid

A wide range i.e. 22.65 to 154.72 mg/100 g was noticed in ascorbic acid content; the highest being in chutney (154.72 mg/100 g) and the lowest being in *pakora* (22.65 mg/100 g). The ascorbic acid content of all the four products differed significantly

Table 4.13 Nutrient composition of products prepared from *Moringa oleifera* leaves (on fresh weight basis)

Food products	Moisture (%)	Ascorbic acid (mg/100 g)	β-carotene (mg/100 g)
Chutney	77.54 ± 0.01	154.72 ± 0.01	3.77 ± 0.02
Leaves *bhuji*	64.98 ± 0.01	42.54 ± 0.01	2.49 ± 0.02
Potato and leaves vegetable	67.34 ± 0.01	40.33 ± 0.02	2.44 ± 0.02
Pakora	54.24 ± 0.01	22.65 ± 0.03	2.94 ± 0.02
CD (P<0.05)	0.04	0.06	0.07

Values are mean ± SE of three independent determinations

($P<0.05$) among themselves (Table 4.13). The ascorbic acid is most sensitive to heat and oxidation and was lost during various cooking treatments.

4.3.3 β-carotene

β-carotene of products prepared from fresh *M. oleifera* leaves has been depicted in Table 4.13. β-carotene of chutney was significantly ($P<0.05$) higher as compared to other products. However, there were non-significant differences in β-carotene contents of leaves *bhuji* and potato and leaves vegetable. Slight decrease in β-carotene might be due to various processing treatments.

4.3.4 Crude protein

Crude protein content among the four different products varied from 8.62 to 10.67 g/100 g; the highest being in *pakora* and the lowest being in leaves *bhuji*. A significant ($P<0.05$) difference was noticed in crude protein content of various products i.e. chutney, leaves *bhuji*, potato and leaves vegetable and *pakora*. It might be due to incorporation of higher chickpea flour in *pakora* which had higher protein content.

4.3.5 Crude fat

Crude fat content of various products ranged from 2.14 to 23.80 g/100 g. *Pakora* had significantly ($P<0.05$) higher content than potato and leaves vegetable, leaves *bhuji* and chutney. Chutney had the lowest fat content as no visible fat was added to it, while *pakora* had the highest fat content due to absorption of more visible fat used during frying.

4.3.6 Total ash

Total ash content of chutney leaves *bhuji*, potato and leaves *bhuji* and *pakora* varied from 0.80 to 0.92 g/100 g (Table 4.14).

Table 4.14 Nutrient composition of products prepared from *Moringa oleifera* leaves (g/100 g, on dry weight basis)

Food products	Crude protein	Crude fat	Total ash	Crude fibre	Dietary fibre			Total carbohy-drates
					Total	Soluble	Insoluble	
Chutney	9.56 ± 0.01	2.14 ± 0.02	0.87 ± 0.01	2.44 ± 0.02	16.74 ± 0.02	5.72 ± 0.01	11.03 ± 0.02	84.92 ± 0.02
Leaves *bhuji*	8.62 ± 0.02	18.30 ± 0.02	0.80 ± 0.01	2.56 ± 0.01	15.44 ± 0.02	5.13 ± 0.01	10.31 ± 0.01	69.59 ± 0.01
Potato and leaves vegetable	8.70 ± 0.01	18.54 ± 0.02	0.86 ± 0.01	2.61 ± 0.01	15.51 ± 0.02	5.66 ± 0.01	9.85 ± 0.01	69.20 ± 0.01
Pakora	10.67± 0.01	23.80 ± 0.03	0.92 ± 0.01	0.96 ± 0.02	14.09 ± 0.03	6.02 ± 0.02	8.07 ± 0.03	63.61 ± 0.01
CD (P<0.05)	0.05	0.07	0.03	0.04	0.07	0.04	0.07	0.04

Values are mean ± SE of three independent determinations

Pakora had the maximum whereas the leaves *bhuji* had the minimum ash content. Ash content of chutney and potato and leaves vegetable did not differ significantly ($P<0.05$). On the contrary, leaves *bhuji* had significantly ($P<0.05$) less amount of ash content when compared to other products. The increase in ash content was as a result of higher ash content of chickpea flour used in *pakora*.

4.3.7 Crude fibre

Crude fibre of products prepared from *M. oleifera* leaves has been presented in Table 4.14. Crude fibre content of products ranged from 0.96 to 2.61 g/100 g on dry matter basis. Potato and leaves vegetable had significantly ($P<0.05$) higher crude fibre content followed by leaves *bhuji*, chutney and *pakora*. *Pakora* had the minimum amount (0.06 g/100 g) of crude fibre. Higher crude fibre content was observed due to use of more amount of *M. oleifera* leaves.

4.3.8.1 Total dietary fibre

Total dietary fibre content of four different products prepared from fresh *M. oleifera* leaves varied from 14.09 to 16.74 g/100 g (Table 4.14). Chutney had the maximum (16.74 g/100 g) amount of dietary fibre followed by potato and leaves vegetable (15.51 g/100 g) and leaves *bhuji* (15.44 g/100 g); the minimum being in *pakora* (14.09 g/100 g). A significant ($P<0.05$) difference was observed when dietary fibre content of all these four products was compared with each other. It might be due to incorporation of higher amount of *M. oleifera* leaves and moreover, no processing treatments were involved in preparation of chutney.

4.3.8.2 Soluble dietary fibre

Soluble dietary fibre content among the products varied from 5.13 to 6.02 g/100 g (Table 4.14). *Pakora* had the maximum (6.02

g/100 g) soluble dietary fibre followed by that of chutney (5.72 g/100 g), potato and leaves vegetable (5.66 g/100 g) and leaves *bhuji* (5.13 g/100 g) in descending order. Effect of different processing treatments might have decreased the TDF and IDF and increased the level of SDF content.

4.3.8.3 Insoluble dietary fibre

Insoluble dietary fibre of the products varied from 8.07 to 11.03 g/100 g. Chutney had the maximum amount of insoluble dietary fibre (11.03 g/100 g) whereas *pakora* had the minimum (8.07 g/100 g). A significant difference ($P<0.05$) occurred in insoluble dietary fibre content of these products. Decrease in insoluble dietary fibre content might be due to cumulative effect of different processing treatments.

4.3.9 Total carbohydrates

Chutney had the maximum total carbohydrate content (84.92 g/100 g) whereas *pakora* (63.61 g/100 g) had the minimum (Table 4.14). A significant difference ($P<0.05$) in total carbohydrate content of the four products prepared from fresh leaves was noticed which might be due to difference in the type and amount of food ingredients used in addition to *Moringa oleifera* leaves.

4.3.10 Antinutrients

4.3.10.1Oxalic acid

Oxalic acid content in the food products ranged from 265.82 to 325.53 mg/100 g (Table 4.15). Chutney had significantly ($P<0.05$) higher oxalic acid content as compared to that of leaves *bhuji*, potato and leaves vegetable and *pakora*. Oxalic acid content of these products was less as compared to the chutney due to cumulative effect of different processing treatments.

Table 4.15 Antinutritional factors of products prepared from *Moringa oleifera* leaves (on dry weight basis)

Food products	Oxalic acid (mg/100 g)	Phytic acid (g/100 g)	Saponins (g/100 g)	Trypsin inhibitor activity (TIU/g)
Chutney	325.53 ± 0.02	1.53 ± 0.02	0.96 ± 0.01	ND
Leaves *bhuji*	272.18 ± 0.87	0.91 ± 0.02	0.59 ± 0.02	ND
Potato and leaves vegetable	270.61 ± 0.02	0.88 ± 0.01	0.63 ± 0.01	ND
Pakora	265.82 ± 0.02	1.14 ± 0.02	0.71 ± 0.01	ND
CD ($P<0.05$)	1.43	0.05	0.02	ND

Values are mean ± SE of three independent determinations
Trypsin inhibitor units: One unit of trypsin was defined as the amount of enzymes which converted one mg casein in TCA soluble components at 37°C for 20 minutes at pH 7.6

ND = Not detected

4.3.10.2 Phytic acid

The phytic acid content ranged from 0.88 to 1.53 g/100 g among the four different products (Table 4.15); maximum being in the chutney and the minimum being in potato and leaves vegetable. A non-significant difference was noticed in phytic acid content of leaves *bhuji* and potato and leaves vegetable. The phytic acid content of chutney and *pakora* was significantly ($P<0.05$) higher than that of leaves *bhuji* and potato and leaves vegetable. The decrease in phytic acid might be due to effect of different processing treatments used in the preparation of recipes .

4.3.10.3 Saponins

Saponin content of products prepared from fresh leaves i.e. chutney, leaves *bhuji*, potato and leaves vegetable and *pakora* has been presented in Table 4.15. Saponin content varied from 0.59 to 0.96 g/100 g. The highest saponin content was in chutney (0.96 g/100 g) followed by *pakora* (0.71 g/100 g), potato and leaves vegetable (0.63 g/100 g) and leaves *bhuji* (0.59 g/100 g). A significant ($P<0.05$) difference in saponin content among all the products was observed. The decrease in saponin content of products was observed as various processing methods caused considerable loss.

4.3.10.4 Trypsin inhibitor activity

No trypsin inhibitor activity was detected in chutney, leaves *bhuji*, potato and leaves vegetable and *pakora*. It might be due to negligible amount of TIA in raw *Moringa oleifera* leaves. Moreover, use of various processing methods like heating, might have caused considerable loss in trypsin inhibitor activity.

4.3.11 Total mineral composition

Tables 4.16 and 4.17 represent the total calcium, phosphorus, iron, zinc and potassium contents of the four different products prepared from fresh *M. oleifera* leaves.

4.3.11.1 Calcium

Total calcium content of products ranged from 839.64 to 1203.31 mg/100 g. Chutney had the maximum calcium content (1203.31 mg/100 g) whereas *pakora* had the minimum (839.64 mg/100 g) (Table 4.16). A significant difference in total calcium content of all the four products was observed. A wide range of calcium content was observed due to difference in method of preparation and amount of ingredients especially *M oleifera* leaves having appreciably higher calcium content i.e. 1999.81g (Table 4.4)

4.3.11.2 Phosphorus

Total phosphorus content was significantly ($P<0.05$) higher in chutney followed by potato and leaves vegetable leaves *bhuji* and *pakora*. A significant difference in total phosphorus was observed among various products prepared from fresh *M. oleifera* leaves.

4.3.11.3 Iron

Chutney had the maximum iron content (12.11 mg/100 g) whereas leaves *bhuji* (11.50 mg/100 g) had the minimum (Table 4.16). A significant ($P<0.05$) difference in total iron content of all the four products was observed. The decrease in content might be due to cumulative effect of various processing methods resulting in subsequent loss of iron content.

4.3.11.4 Zinc

A narrow range in zinc content of the products prepared from fresh *M. oleifera* leaves was observed (Table 4.17). *Pakora* had maximum (15.82 mg/100 g) zinc content whereas chutney had the

Table 4.16 Total and HCl-extractable calcium, phosphorus and iron contents (mg/100 g) of products prepared from *Moringa oleifera* leaves

Food products	Calcium			Phosphorus			Iron		
	Total	HCl-extractable	% extractability	Total	HCl-extractable	% extractability	Total	HCl-extractable	% extractability
Chutney	1203.31 ± 0.02	905.86 ± 0.37	75.30 ± 0.01	97.30 ± 0.03	71.31 ± 0.09	73.41 ± 0.01	12.11 ± 0.02	7.36 ± 0.04	61.31 ± 0.02
Leaves *bhuji*	840.14 ± 0.02	623.89 ± 0.38	74.32 ± 0.02	87.25 ± 0.02	63.49 ± 0.05	72.80 ± 0.03	11.50 ± 0.01	6.88 ± 0.04	60.34 ± 0.02
Potato and leaves vegetable	853.34 ± 0.02	633.95 ± 0.92	74.43 ± 0.04	88.53 ± 0.01	64.45 ± 0.15	72.90 ± 0.01	11.62 ± 0.01	7.04 ± 0.02	60.55 ± 0.01
Pakora	839.64 ± 0.02	598.42 ± 0.34	71.30 ± 0.03	85.72 ± 0.01	60.43 ± 0.02	70.52 ± 0.01	11.81 ± 0.01	6.91 ± 0.02	58.74 ± 0.01
CD (P<0.05)	0.07	1.82	0.09	0.07	0.30	0.06	0.05	0.11	0.06

Values are mean ± SE of three independent determinations

minimum (15.32 mg/100 g). A non-significant difference in zinc content of leaves *bhuji* and potato and leaves vegetable was observed. Increase in zinc content of *pakora* observed might be due to incorporation of chickpea flour.

4.3.11.5 Potassium

Total potassium content of the products i.e. chutney, leaves *bhuji*, potato and leaves vegetable and *pakora* varied from 952.48 to 967.31 mg/100 g.

A significant ($P<0.05$) difference was observed in potassium content of various products prepared from fresh *M. oleifera* leaves. Chutney had the maximum whereas leaves *bhuji* had the lowest potassium content due to various processing treatments.

4.3.12 HCl-extractability of minerals

4.3.12.1 Calcium

Calcium extractability in various products prepared from fresh leaves has been presented in Table 4.16. HCl-extractability of calcium in chutney was significantly ($P<0.05$) higher followed by potato and leaves vegetable, leaves *bhuji* and *pakora*. *Pakora* had minimum HCl-extractable calcium content (598.42 mg/100 g). It might be observed due to variation in type and amount of ingredients used and various types of processing treatments involved in preparation of these products. Blanching might have reduced the phytic acid content in sufficient amount as compared to frying.

4.3.12.2 Phosphorus

HCl-extractability of phosphorus in products prepared from fresh leaves ranged from 70.52 to 73.41 percent. Chutney had the maximum HCl-extractability of phosphorus. It was minimum in *pakora*. Significant ($P<0.05$) differences were noticed in phosphorus extractability of all the four products i.e. chutney, leaves *bhuji*,

potato and leaves vegetable and *pakora*. The decrease in phosphorus extractability was observed as the level of antinutrients increased. However, type and amount of ingredients also determined the extractability of phosphorus.

4.3.12.3 Iron

HCl-extractability of iron ranged from 58.74 to 61.31 percent. Chutney had significantly ($P<0.05$) higher iron extractability followed by that of potato and leaves vegetable, leaves *bhuji* and *pakora*. The decrease in iron extractability was due to variation in processing treatments including blanching, steam cooking and frying.

4.3.12.4 Zinc

Zinc-extractability was significantly ($P<0.05$) higher in potato and leaves vegetable as compared to other products prepared from fresh leaves (Table 4.17).

Pakora had the lowest zinc extractability (68.32%). Zinc extractability of chutney and leaves *bhuji* was 68.53 and 69.57 percent, respectively. All the products were significantly ($P<0.05$) different from each other in regard to their zinc extractability. Result of present study might be due to the fact that blanching improved the extractability of zinc as compared to frying.

4.3.12.5 Potassium

HCl-extractability of potassium has been depicted in Table 4.17. Potassium extractability of potato and leaves vegetable was significantly ($P<0.05$) higher followed by that of leaves *bhuji*, chutney and *pakora*. *Pakora* had the minimum potassium extractability i.e. 70.34 percent.

4.3.13 *In vitro* availability of calcium and iron

In vitro availability of calcium and iron has been presented in Table 4.18. Availability of calcium (*in vitro*) was significantly

Table 4.17 **Total and HCl-extractable zinc and potassium contents (mg/ 100 g) of products prepared from *Moringa oleifera* leaves (on dry weight basis)**

Food products	Zinc			Potassium		
	Total	HCl-extractable	Percent extractability	Total	HCl-extractable	Percent extractability
Chutney	15.32 ± 0.01	10.12 ± 0.04	68.53 ± 0.01	967.31 ± 0.02	689.63 ± 0.01	71.38 ± 0.06
Leaves *bhuji*	15.53 ± 0.01	10.32 ± 0.12	69.57 ± 0.01	952.48 ± 0.01	682.38 ± 0.04	71.64 ± 0.01
Potato and leaves vegetable.	15.57 ± 0.01	10.29 ± 0.01	69.82 ± 0.02	953.72 ± 0.02	688.33 ± 0.09	72.13 ± 0.02
Pakora	15.82 ± 0.02	10.52 ± 0.01	68.32 ± 0.02	959.61 ± 0.02	689.23 ± 0.06	70.34 ± 0.02
CD (P<0.05)	0.05	0.08	0.05	0.06	1.67	0.11

Values are mean ± SE of three independent determinations

Table 4.17 **Total and HCl-extractable zinc and potassium contents (mg/100 g) of products prepared from *Moringa oleifera* leaves (on dry weight basis)**

Food products	Zinc			Potassium		
	Total	HCl-extractable	Percent extractability	Total	HCl-extractable	Percent extractability
Chutney	15.32 ± 0.01	10.12 ± 0.04	68.53 ± 0.01	967.31 ± 0.02	689.63 ± 0.01	71.38 ± 0.06
Leaves *bhuji*	15.53 ± 0.01	10.32 ± 0.12	69.57 ± 0.01	952.48 ± 0.01	682.38 ± 0.04	71.64 ± 0.01
Potato and leaves vegetable.	15.57 ± 0.01	10.29 ± 0.01	69.82 ± 0.02	953.72 ± 0.02	688.33 ± 0.09	72.13 ± 0.02
Pakora	15.82 ± 0.02	10.52 ± 0.01	68.32 ± 0.02	959.61 ± 0.02	689.23 ± 0.06	70.34 ± 0.02
CD (P<0.05)	0.05	0.08	0.05	0.06	1.67	0.11

Values are mean ± SE of three independent determinations

Table 4.18 ***In vitro* availability (%) of calcium and iron of products prepared from *Moringa oleifera* leaves (on dry weight basis)**

Food products	Calcium		Iron	
	Total (mg/100 g)	Percent availability (*in vitro*)	Total (mg/100 g)	Percent availability (*in vitro*)
Chutney	1203.31 ± 0.02	22.35 ± 0.03	7.36 ± 0.04	13.53 ± 0.01
Leaves *bhuji*	840.14 ± 0.02	25.39 ± 0.04	6.88 ± 0.04	15.56 ± 0.02
Potato and leaves vegetable	853.34 ± 0.02	25.50 ± 0.01	7.04 ± 0.02	15.65 ± 0.01
Pakora	839.64 ± 0.02	24.35 ± 0.02	6.91 ± 0.02	14.35 ± 0.01
CD (P<0.05)	0.07	0.05	0.05	0.05

Values are mean ± SE of three independent determinations

($P<0.05$) higher in potato and leaves vegetable followed by leaves *bhuji*, *pakora* and chutney. It might be due to the reason that processing had reduced the antinutrients and consequently caused improvement in calcium availability. Chutney had the minimum calcium availability (22.35%) as all the ingredients were simply ground and no processing was noticed. However, significant differences in calcium availability were observed among all these various products prepared from *M. oleifera* fresh leaves.

Similar trend was also observed in the availability of iron in chutney, leaves *bhuji*, potato and leaves vegetable and *pakora*. Results of present study might be due to the fact that blanching improved the calcium as well as iron availability as compared to frying. Blanching might have reduced the antinutritional contents including oxalic acid, phytic acid and saponins which interfere with the mineral absorption. Similar findings have also been reported by Saharan (1994).

4.4 Nutritional composition of products prepared from dried *M. oleifera* leaves

Sev and noodles were prepared by incorporating *Moringa oleifera* leaf powder at different levels i.e. at 10 and 20 percent.

4.4.1 Moisture

Moisture content of *sev* prepared by incorporating 20 percent *M. oleifera* leaves was significantly ($P<0.05$) higher followed by *sev* prepared from 10 percent *M. oleifera* leaves and control.

Similar trend was observed in noodles i.e. noodles prepared from 20 percent *M. oleifera* leaf powder had the maximum moisture content. Increase in moisture content might have observed due to higher protein content which indicated a possible relationship between water absorption and protein content.

Table 4.19 Nutrient composition of products prepared from *Moringa oleifera* leaves (on fresh weight basis)

Food products	Moisture (%)	Ascorbic acid (mg/100 g)	β-carotene (mg/100 g)
Sev			
Bengal gram flour (control 100%)	5.42 ± 0.02	ND	0.14 ± 0.01
Bengal gram flour + *Moringa oleifera* leaves (10%)	6.00 ± 0.01	ND	1.56 ± 0.01
Bengal gram flour + *Moringa oleifera* leaves (20%)	6.10 ± 0.01	ND	3.16 ± 0.01
CD (P<0.05)	0.04	ND	0.03
Noodles			
Refined flour (control 100%)	59.41 ± 0.01	0.08 ± 0.00	0.03 ± 0.01
Refined flour + *Moringa oleifera* leaves (10%)	59.83 ± 0.01	0.10 ± 0.00	1.32 ± 0.01
Refined flour + *Moringa oleifera* leaves (20%)	60.55 ± 0.01	0.13 ± 0.01	2.75 ± 0.01
CD (P<0.05)	0.04	0.02	0.03

Values are mean ± SE of three independent determinations
ND = Not detected

4.4.2 Ascorbic acid

Ascorbic acid content could bot be detected in *sev* whereas in noodles, it was almost negligible but differed significantly among themselves in all the three types of noodles (Table 4.19). The loss in ascorbic acid might be due to cumulative effect of various processing treatments involved in the preparation of *sev* and noodles.

4.4.3 β-carotene

β-carotene content was significantly ($P<0.05$) higher in *sev* prepared from 20 percent *M. oleifera* leaf powder as compared to that prepared from 10 percent *M. oleifera* leaf powder and control (Table 4.19).

Similar trend was also observed in the noodles. It might be due to incorporation of higher amount of *M. oleifera* leaves in 20 percent supplemented *sev* and noodles as leaves had very high β-carotene content (Table 4.1).

4.4.4 Crude protein

On dry matter basis, the protein content of *sev* varied from 20.69 to 23.65 g/100 g (Table 4.20). *Sev* prepared from 20 percent *M. oleifera* leaf powder had significantly high protein as compared to that prepared from 10 percent *M. oleifera* leaf powder and control. Similar trend was also observed in noodles. Higher protein content of *sev* and noodles prepared from 20 percent *M. oleifera* leaf powder seemed to be the result of the appreciably higher content of *M. oleifera* leaf powder (Table 4.2).

4.4.5 Crude fat

On dry matter basis, the fat content of *sev* supplemented with 20 percent *M. oleifera* leaf powder was found to be significantly higher followed by *sev* having supplementation of leaf powder at 10% and control (Table 4.20).

Table 4.20 Nutrient composition of products prepared from *Moringa oleifera* leaves (g/100 g, on dry weight basis)

Foos products	Crude protein	Crude fat	Total ash	Crude fibre	Dietary fibre			Total carbohy-drates
					Total	Soluble	Insoluble	
Sev								
Bengal gram flour (control 100%)	20.69 ± 0.02	19.09 ± 0.01	2.59 ± 0.02	1.27 ± 0.01	15.64 ± 0.02	4.53 ± 0.01	11.10 ± 0.01	56.32 ± 0.02
Bengal gram flour + *Moringa oleifera* leaves (10%)	21.72 ± 0.14	20.64 ± 0.02	3.34 ± 0.01	1.47 ± 0.02	17.84 ± 0.02	4.93 ± 0.01	12.91 ± 0.02	57.16 ± 0.01
Bengal gram flour + *Moringa oleifera* leaves (20%)	23.65 ± 0.01	20.74 ± 0.01	3.95 ± 0.02	1.65 ± 0.01	20.21 ± 0.02	3.82 ± 0.66	16.38 ± 0.68	48.52 ± 0.01
CD (P<0.05)	0.06	0.05	0.06	0.04	0.08	1.32	1.36	0.04
Noodles								
Refined flour (control 100%)	9.28 ± 0.02	12.86 ± 0.01	2.54 ± 0.01	1.28 ± 0.01	12.19 ± 0.01	3.11 ± 0.02	9.08 ± 0.03	73.92 ± 0.02
Refined flour + *Moringa oleifera* leaves (10%)	11.04 ± 0.01	12.66 ± 0.01	2.74 ± 0.02	1.39 ± 0.01	15.96 ± 0.02	3.36 ± 0.01	12.60 ± 0.01	71.07 ± 0.01
Refined flour + *Moringa oleifera* leaves (20%)	13.34 ± 0.02	12.60 ± 0.02	2.80 ± 0.01	1.45 ± 0.01	17.26 ± 0.01	3.84 ± 0.01	13.43 ± 0.01	67.74 ± 0.02
CD (P<0.05)	0.06	0.06	0.06	0.04	0.04	0.04	0.06	0.05

Values are mean ± SE of three independent determinations

On the contrary, fat content was significantly (P<0.05) higher in control noodles followed by noodles incorporating *M. oleifera* leaf powder at 10 percent and 20 percent levels.

4.4.6 Total ash

On dry weight basis, ash content of *sev* was maximum in 20 percent supplemented *sev* and minimum in control (Table 4.20). Similar trend was also observed in the noodles. This increase might be attributed to higher ash content of *M. oleifera* leaf powder (Table 4.2).

4.4.7 Crude fibre

Crude fibre content of three different types of *sev* (control, 10% and 20%) was 1.27, 1.47 and 1.65, respectively, on dry weight basis. It was significantly (P<0.05) higher in *sev* prepared from 20 percent *M. oleifera* leaf powder as compared to *sev* prepared from 10 percent and control (Table 4.20).

Similarly, crude fibre content of noodles increased significantly (P<0.05) as the level of supplementation of *M. oleifera* leaf powder increased due to higher crude fibre content of *M. oleifera* leaves (Table 4.2).

4.4.8 Total dietary fibre

Results of total dietary fibre, soluble dietary fibre and insoluble dietary fibre are presented in Table 4.20.

Total dietary fibre content of control *sev* was 15.64 g/100 g. In *sev* containing 10 percent *Moringa oleifera* leaves, a significant (P<0.05) increase in TDF content was noticed. The highest TDF content was found in *sev* supplemented with 20 percent *M. oleifera* leaves.

Similarly, noodles prepared by incorporating 20 percent *M. oleifera* leaves had significantly higher content of TDF followed by *sev* prepared from 10 percent and control. Results of present study are due to appreciably higher TDF content of *M. oleifera* leaves.

4.4.8.2 Soluble dietary fibre

A non-significant difference was observed in soluble dietary fibre in *sev* prepared from *M. oleifera* leaves at different levels i.e. 10 percent and 20 percent (Table 4.20).

On the contrary, noodles prepared from 20 percent *M. oleifera* leaves had significantly ($P<0.05$) higher SDF content as compared to *sev* prepared from 10 percent *M. oleifera* leaves and control. It might be due to incorporation of higher amount of *M. oleifera* leaves (Table 4.2).

4.4.8.3 Insoluble dietary fibre

Insoluble dietary fibre (IDF) content was found to be significantly ($P<0.05$) higher in *sev* having 20 percent supplementation of leaf powder of *Moringa oleifera* followed by *sev* containing 10 percent leaf powder and control.

Similar trend in IDF content of the noodles was also observed. As the level of supplementation increased, there was increase in insoluble dietary fibre content. It might be due to addition of higher leaf powder having appreciable amount of insoluble dietary fibre (Table 4.2).

4.4.9 Total carbohydrates

A significant ($P<0.05$) difference was observed in total carbohydrate content of different types of *sev* (Table 4.20).

The maximum total carbohydrate content was found in *sev* prepared from 10 percent *M. oleifera* leaf powder followed by control and *sev* incorporating 20 percent *M. oleifera* leaf powder.

The control noodles i.e. prepared without addition of *M. oleifera* leaf powder had significantly higher total carbohydrate content as compared to noodles prepared from 10 as well as 20 percent *M. oleifera* leaf powder. It might be due to higher

carbohyddrate content of refined flour (wheat) (Gopalan *et al.*, 1989).

4.4.10 Antinutrients

Among the antinutrients, data regarding oxalic acid, phytic acid saponins and trypsin inhibitor activity of *sev* and noodles are tabulated and presented in Table 4.21.

4.4.10.1 Oxalic acid

Oxalic acid content of *sev* prepared by supplementation with 20 percent *M. oleifera* leaf powder had significantly ($P<0.05$) higher oxalic acid content followed by *sev* (10% *M. oleifera*) and control (Table 4.21).

Similar trend was observed in noodles prepared by incorporating different levels of *M. oleifera* leaf powder. Higher oxalic acid content of *sev* (20%) was due to addition of *M. oleifera* leaf powder which is considered to be rich source of oxalic acid. However, the values observed were less in products i.e. noodles and *sev* over the raw *M. oleifera* leaf powder. The reduction in oxalic acid might be due to its loss during various processing treatments used in preparation of products.

4.4.10.2 Phytic acid

The phytic acid content of *sev* (control, 10% and 20%) was 0.46, 0.47 and 0.53 g/100 g, respectively. A non-significant difference was observed in control *sev* and 10 percent *M. oleifera* leaves supplemented *sev*. Phytic acid content was maximum in 20 percent *sev*.

In noodles, a non-significant difference was observed in different types of noodles *M. oleifera* leaves contribution towards phytic acid content in the products was significant. However, phytic acid content of *sev* and noodles was less as compared to that of raw *M. oleifera* leaf powder as various processing methods

Table 4.21 Antinutritional factors of products prepared from *Moringa oleifera* leaves (on dry weight basis)

Food products	Oxalic acid (mg/100 g)	Phytic acid (g/100 g)	Saponins (g/100 g)	Trypsin inhibitor activity (TIU/g)
Sev				
Bengal gram flour (control 100%)	11.89 ± 1.01	0.46 ± 0.01	0.29 ± 0.01	98.60 ± 0.01
Bengal gram flour + *Moringa oleifera* leaves (10%)	53.57 ± 0.01	0.47 ± 0.02	0.43 ± 0.01	98.63 ± 0.01
Bengal gram flour + *Moringa oleifera* leaves (20%)	65.75 ± 0.02	0.53 ± 0.01	0.53 ± 0.02	98.64 ± 0.01
CD (P<0.05)	2.02	0.05	0.04	0.04
Noodles				
Refined flour (control 100%)	217.74 ± 0.03	0.40 ± 0.01	0.26 ± 0.01	103.32 ± 0.02
Refined flour + *Moringa oleifera* leaves (10%)	237.62 ± 0.02	0.42 ± 0.00	0.38 ± 0.01	103.34 ± 0.01
Refined flour + *Moringa oleifera* leaves (20%)	257.72 ± 0.01	0.43 ± 0.01	0.48 ± 0.01	103.34 ± 0.01
CD (P<0.05)	0.07	0.02	0.04	0.05

Values are mean ± SE of three independent determinations

Trypsin inhibitor units: One unit of trypsin was defined as the amount of enzymes which converted one mg casein in TCA soluble components at 37°C for 20 minutes at pH 7.6

involved in the preparation of *sev* and noodles caused considerable loss of this antinutrient.

4.4.10.4 Saponins

Saponin content of different types of *sev* ranged from 0.29 to 0.53 g/100 g, maximum saponin content was found in *sev* supplemented with 20 percent *M. oleifera* leaf powder.

Similarly, saponin content of different types of noodles varied from 0.26 to 0.48 g/100 g. The highest saponin content was found in noodles (20%) and the lowest in control. Higher saponin content in *sev* and noodles (20%) was due to appreciably higher amount of *M. oleifera* leaf powder (Table 4.21).

4.4.10.4 Trypsin inhibitor activity

A non-significant difference in trypsin inhibitor activity of *sev* and noodles was observed over the respective conrols (Table 4.21). It might be due to reason that trypsin inhibitor activity was not detected in raw *M. oleifera* leaves. Moreover, processing treatments might have also decreased the trypsin inhibitor activity found in other ingredients used in the preparation of *sev* and noodles.

4.4.11 Total mineral composition

Data regarding the calcium, phosphorus, iron, zinc and potassium contents of *M. oleifera* leaves supplemented *sev* and noodles are presented in Tables 4.22 and 4.23.

4.4.11.1 Calcium

A wide range of calcium content in different types of *sev* i.e. 72.78 to 358.33 mg/100 g (DM basis) was observed (Table 4.22). *Sev* prepared from 20 percent supplemented *M. oleifera* leaves powder had about 5 times more calcium content (358.33 mg/100 g) over the control (72.78 mg/100 g).

Similar trend was also observed in noodles prepared by incorporation of different levels of *M. oleifera* leaf powder. Higher

Table 4.22 **Total and HCl-extractable calcium, phosphorus and iron contents (mg/100 g) of products prepared from *Moringa oleifera* leaves**

Food products	Calcium			Phosphorus			Iron		
	Total	HCl-extractable	% extractability	Total	HCl-extractable	% extractability	Total	HCl-extractable	% extractability
Sev									
Bengal gram flour (control 100%)	72.78 ± 0.02	60.95 ± 0.23	70.30 ± 0.02	118.73 ± 0.02	84.43 ± 0.06	70.47 ± 0.02	3.81 ± 0.03	2.35 ± 0.03	62.47 ± 0.02
Bengal gram flour + *Moringa oleifera* leaves (10%)	256.54 ± 0.02	184.92 ± 0.41	72.36 ± 0.01	135.53 ± 0.01	95.24 ± 0.06	71.17 ± 0.01	5.80 ± 0.01	3.64 ± 0.05	61.37 ± 0.01
Bengal gram flour + *Moringa oleifera* leaves (20%)	358.33 ± 0.02	258.84 ± 0.40	72.42 ± 0.01	149.31 ± 0.02	105.11 ± 0.09	70.31 ± 0.02	6.84 ± 0.01	4.20 ± 0.03	61.43 ± 0.01
CD (P<0.05)	0.07	1.24	0.05	0.06	0.25	0.06	0.05	0.14	0.04
Noodles									
Refined flour (control 100%)	50.09 ± 0.01	35.75 ± 0.03	71.45 ± 0.03	244.73 ± 0.01	180.71 ± 0.03	71.34 ± 0.02	5.85 ± 0.01	3.45 ± 0.03	62.40 ± 0.05
Refined flour + *Moringa oleifera* leaves (10%)	237.52 ± 0.03	171.87 ± 0.01	72.33 ± 0.01	261.51 ± 0.01	182.62 ± 0.01	71.82 ± 0.01	6.80 ± 0.01	4.09 ± 0.04	62.31 ± 0.02
Refined flour + *Moringa oleifera* leaves (20%)	341.65 ± 0.01	247.85 ± 0.03	72.54 ± 0.02	279.31 ± 0.02	185.46 ± 0.02	71.92 ± 0.02	7.81 ± 0.02	4.82 ± 0.04	62.35 ± 0.03
CD (P<0.05)	0.08	0.10	0.07	0.06	0.08	0.06	0.05	0.12	0.12

Values are mean ± SE of three independent determinations

the level of supplementation in *M. oleifera*, more was calcium content. The main reason for increase in total calcium content was due to addition of higher amount of *M. oleifera* leaf powder. Raw *M. oleifera* leaves (DM basis) have been found to be rich source of calcium (Table 4.16). Hence, calcium content was increased with increase in the level of supplementation with leaf powder of *Moringa loleifera* in *sev and* noodles.

4.4.11.2 Phosphorus

Sev and noodles having 20 percent *M. oleifera* leaf powder had significantly ($P<0.05$) higher phosphorus content followed by *sev* and noodles (10%) and control (Table 4.22). This might be due to higher phosphorus content of *M. oleifera* leaf powder (Table 4.4).

4.4.11.3 Iron

Total iron content of different types of *sev* prepared ranged from 3.81 to 6.84 mg/100 g. Maximum iron content was found in *sev* (20%) and minimum in control *sev*.

Similarly, total iron content of noodles ranged from 5.85 to 7.81 mg/100 g. Noodles prepared by incorporating 20 percent *M. oleifera* leaf powder had the highest iron content due to higher iron content of raw *M. oleifera* leaves (on DM basis) (Table 4.4).

4.4.11.4 Zinc

Significant ($P<0.05$) differences were observed in zinc content of different types of *sev* and noodles (Table 4.23). Zinc content was the highest in *sev* and noodles having 20 percent *M. oleifera* leaf powder whereas it was the lowest in respective controls of both *sev* and noodles i.e. prepared without the addition of *M. oleifera* leaf powder.

4.4.11.5 Potassium

A wide range of potassium content was observed in different types of *sev* and noodles (Table 4.23). Potassium content of *sev* (20% *M. oleifera* leaf powder) was significantly ($P<0.05$) higher followed by *sev* (10%) and control. Similar trend was also observed in noodles. The results of present study might be due to higher potassium content of dried *M. oleifera* leaves powder (on DM basis).

4.4.12 HCl-extractability of minerals

4.4.12.1 Calcium

The values for HCl-extractability of calcium extractability of different types of *sev* varied significantly ($P<0.05$) among themselves (Table 4.22). Maximum calcium extractability was observed in *sev* supplemented with 20 percent *M. oleifera* leaves powder whereas it was minimum in control. Similar trend was also observed in noodles. Higher calcium extractability was observed due to appreciably higher amount of *M. oleifera* leaves (Table 4.4).

4.4.12.2 Phosphorus

Phosphorus extractability of *sev* and noodles supplemented with 20 percent *M. oleifera* leaf powder was significantly ($P<0.05$) higher over the control and products incorporating *sev* and noodles (10% and control) *M. oleifera* powder (Table 4.22). As the level of supplementation increased, there was increase in phosphorus extractability.

4.4.12.3 Iron

Extractability of iron was significantly higher in control *sev* followed by 20 percent and 10 percent leaf powder supplemented *sev*. However, non-significant differences were found in different leaf powder supplemented noodles.

Table 4.23 Total and HCl-extractable zinc and potassium contents (mg/100 g) of products prepared from *Moringa oleifera* leaves (on dry weight basis)

Food products	Zinc			Potassium		
	Total	HCl-extractable	Percent extractability	Total	HCl-extractable	Percent extractability
Sev						
Bengal gram flour (control 100%)	2.10 ± 0.01	1.33 ± 0.02	69.47 ± 0.02	650.35 ± 0.03	457.38 ± 0.04	70.32 ± 0.01
Bengal gram flour + *Moringa oleifera* leaves (10%)	3.84 ± 0.02	2.64 ± 0.02	69.82 ± 0.02	750.54 ± 0.01	545.24 ± 0.05	70.53 ± 0.02
Bengal gram flour + *Moringa oleifera* leaves (20%)	4.84 ± 0.01	3.30 ± 0.03	69.85 ± 0.02	837.61 ± 0.02	591.69 ± 0.04	70.71 ± 0.02
CD (P<0.05)	0.04	0.08	0.06	0.07	0.16	0.06
Noodles						
Refined flour (control 100%)	2.72 ± 0.01	1.86 ± 0.01	68.56 ± 0.01	125.45 ± 0.03	89.49 ± 0.03	71.30 ± 0.03
Refined flour + *Moringa oleifera* leaves (10%)	3.53 ± 0.02	2.42 ± 0.02	68.93 ± 0.02	241.34 ± 0.02	173.41 ± 0.01	71.82 ± 0.01
Refined flour + *Moringa oleifera* leaves (20%)	4.03 ± 0.02	2.77 ± 0.01	68.97 ± 0.01	297.53 ± 0.01	214.07 ± 0.04	71.93 ± 0.02
CD (P<0.05)	0.06	0.05	0.05	0.08	0.10	0.08

Values are mean ± SE of three independent determinations

4.4.12.4 Zinc

Zinc extractability of *sev* and noodles varied from 69.47 to 69.85% and 68.56 to 68.97%, respectively (Table 4.23). Non-significant differences were observed zinc extractability of *sev* prepared by incorporation of 10 percent and 20 percent *M. oleifera* leaf powder (Table 4.23).

Similar trend was also noticed in noodles prepared by incorporation of different levels of *M. oleifera* leaf powder i.e. 10 percent and 20 percent.

4.4.12.5 Potassium

Potassium extractability was significantly ($P<0.05$) higher in *sev* and noodles supplemented with 20 percent *M. oleifera* leaf powder over the respective controls and 10% supplemented products (Table 4.23). Increase in potassium extractability was due to addition of higher *M. oleifera* leaves.

4.4.13*In vitro* availability of calcium and iron

Calcium availability ranged from 20.65 to 21.50 percent. It was found maximum (21.50%) in control and minimum (20.56%) in supplemented with 20 percent *M. oleifera* leaf powder.

On the contrary, calcium availability was found maximum in noodles prepared by incorporating 20 percent *M. oleifera* leaf powder and minimum in control i.e. without addition of *M. oleifera* leaf powder.

Iron availability in *sev* was significantly higher in control followed by *sev* supplemented with 10 percent and 20 percent *M. oleifera* leaf powder. On the contrary, availability of iron was significantly higher in noodles containing 20% *M. oleifera* leaf powder over the control and 10% leaf powder supplemented noodles (Table 4.24).

Table 4.24 ***In vitro*** **availability (%) of calcium and iron of products prepared from *Moringa oleifera* leaves (on dry weight basis)**

Food products	Calcium		Iron	
	Total (mg/100 g)	Percent availability (*in vitro*)	Total (mg/100 g)	Percent availability (*in vitro*)
Sev				
Bengal gram flour (control 100%)	72.78 ± 0.02	21.50 ± 0.01	3.81 ± 0.03	14.39 ± 0.01
Bengal gram flour + *Moringa oleifera* leaves (10%)	256.54 ± 0.02	20.65 ± 0.01	5.80 ± 0.01	13.35 ± 0.02
Bengal gram flour + *Moringa oleifera* leaves (20%)	358.33 ± 0.02	20.56 ± 0.02	6.84 ± 0.01	13.27 ± 0.03
CD (P<0.05)	0.07	0.06	0.05	0.06
Noodles				
Refined flour (control 100%)	50.09 ± 0.01	24.36 ± 0.03	5.85 ± 0.01	13.43 ± 0.02
Refined flour + *Moringa oleifera* leaves (10%)	237.52 ± 0.03	25.43 ± 0.04	6.80 ± 0.01	13.57 ± 0.01
Refined flour + *Moringa oleifera* leaves (20%)	341.65 ± 0.01	26.35 ± 0.01	7.81 ± 0.02	13.65 ± 0.01
CD (P<0.05)	0.08	0.06	0.05	0.05

Values are mean ± Se of three independent determinations

Results of the present study may be due to the fact that blanching involved in noodles preparation increased the availability of calcium and iron as compared to frying. Similar findings have been reported by Saharan (1994) that blanching reduced the sufficient amount of antinutrients which hindered the mineral absorption and decreased the bioavailability of calcium and iron.

4.5 Nutritional composition of products prepared from *M. oleifera* flowers

Various products developed from flowers using different processing treatments viz blanching, sauting and frying were nutritionally analysed for the same parameters as the raw *M. oleifera* flowers.

4.5.1 Moisture

Moisture content of all the products prepared from flowers varied from 60.06 to 83.36 percent (Table 4.25). Highest moisture content (83.36%) was in *raita*; whereas the lowest was in cutlets (60.06%). Increase in moisture content may be due to consistency of the *raita* as compared to other products.

Overall significant ($P<0.05$) differences were found in moisture content of different products prepared by incorporation of *M. oleifera* flowers.

4.5.2 Ascorbic acid

Ascorbic acid content of various products developed from flower ranged from 2.02 to 4.19 mg/100 g. *Raita* had significantly ($P<0.05$) higher ascorbic acid content than other products. However, flower *bhuji* and green gram and flower vegetable had almost similar content of ascorbic acid. Ascorbic acid may be subsequently lost during cooking and frying involved in preparation of vegetable and cutlets, hence, these products had less ascorbic acid than *raita*.

Table 4.25 Nutrient composition of products prepared from *Moringa oleifera* flowers (on fresh weight basis)

Food products	Moisture (%)	Ascorbic acid (mg/100 g)	β-carotene (mg/100 g)
Flower *bhuji*	66.85 ± 0.01	3.29 ± 0.01	0.02 ± 0.01
Pea and Flower vegetable	67.02 ± 0.01	3.36 ± 0.01	0.02 ± 0.01
Green gram and Flower vegetable	67.32 ± 0.01	3.31 ± 0.02	0.02 ± 0.00
Raita	83.36 ± 0.01	4.19 ± 0.01	0.01 ± 0.01
Cutlets	60.06 ± 0.01	2.02 ± 0.01	0.02 ± 0.01
CD (P<0.05)	0.03	0.04	0.01

Values are mean ± SE of three independent determinations

4.5.3 β-carotene

β-carotene content of products developed from flower is presented in Table 4.25.

β-carotene content of all products except *raita* varied non-significantly among each other whereas its content in *raita* was significantly ($P<0.05$) less than the other products. Slight decrease in β-carotene content of *raita* might be due to addition of less amount of flowers in *raita* as compared to rest of products.

4.5.4 Crude Protein

Protein content among the various products varied from 3.45 to 9.56 g/100 g; the highest being in green gram and flower vegetable and the lowest being in *raita* (Table 4.26). A significant ($P<0.05$) difference was noticed in protein content of all the products developed from flowers.

Protein content of green gram and flower vegetable might be the highest due to higher protein content of green gram incorporated.

4.5.5 Crude Fat

A wide range of fat content i.e. 4.11 to 19.62 g/100 g (DM basis) was observed in different products prepared from the flowers, the lowest being in *raita* and the highest being in cutlets (Table 4.26). Cutlets had significantly ($P<0.05$) higher fat content followed by flower *bhuji*, green gram and flower vegetable and *raita*. A significant ($P<0.05$) difference was observed among various products when compared to each other. Higher fat content observed might be due to addition of higher visible fat in cutlets.

4.5.6 Total Ash

Total ash content of products developed from flowers ranged from 0.53 to 1.33 g/100 g (Table 4.26). Cutlets had the maximum whereas *raita* had the minimum ash content. Ash content of

Table 4.26 **Nutrient composition of products prepared from *Moringa oleifera* flowers (g/ 100 g, on dry weight basis)**

Food products	Crude protein	Crude fat	Total ash	Crude fibre	Dietary fibre			Total carbohydrates
					Total	Soluble	Insoluble	
Flower *bhuji*	8.53 ± 0.01	18.61 ± 0.01	0.81 ± 0.01	2.34 ± 0.01	16.53 ± 0.01	5.36 ± 0.01	11.17 ± 0.01	69.68 ± 0.01
Pea and Flower bhuji	8.85 ± 0.01	18.65 ± 0.01	0.85 ± 0.01	2.51 ± 0.01	16.61 ± 0.01	5.59 ± 0.00	11.01 ± 0.01	69.11 ± 0.01
Green gram and Flower *bhuji*	9.56 ± 0.02	18.71 ± 0.01	0.88 ± 0.01	2.58 ± 0.01	16.76 ± 0.01	5.58 ± 0.01	11.18 ± 0.00	68.22 ± 0.01
Raita	3.45 ± 0.01	4.11 ± 0.01	0.53 ± 0.01	1.39 ± 0.01	10.25 ± 0.01	3.23 ± 0.01	7.02 ± 0.01	90.12 ± 0.01
Cutlets	6.69 ± 0.01	19.62 ± 0.01	1.33 ± 0.02	1.72 ± 0.01	15.60 ± 0.01	4.52 ± 0.01	11.08 ± 0.01	70.92 ± 0.02
CD (P<0.05)	0.03	0.03	0.03	0.03	0.03	0.03	0.02	0.04

Values are mean ± SE of three independent determinations

products developed from flowers differed significantly (P<0.05) from each other. Increase in ash content might be due to variation in type and amount of ingredients used.

4.5.7 Crude fibre

On dry matter basis, the crude fibre content of the products developed from flowers ranged from 1.39 to 2.58 g/100 g (Table 4.26).

Green gram and flowers vegetable had the maximum crude fibre content whereas it was minimum in *raita*. Overall comparison indicated a significant (P<0.05) difference among various products developed from flowers. Slight increase in crude fibre content of green gram and pod vegetable might have increased due to higher incorporation of green gram.

4.5.8 Total dietary fibre

Total dietary fibre content of various products developed from flowers has been depicted in Table 4.26. Total dietary fibre content of products ranged from 10.25 to 16.76 g/100 g. Green gram and flower vegetable had the maximum (16.76 g/100 g) total dietary fibre followed by pea and flower vegetable (16.61 g/100 g), flower *bhuji* (16.53 g), cutlet (15.60 g/100 g). The minimum dietary fibre content was observed in *raita* (10.25 g/100 g). Significant (P<0.05) differences were observed in the total dietary fibre content among various products developed from flowers. Increase in total dietary fibre might have resulted due to different variation in type and amount of ingredients.

4.5.8.1 Soluble dietary fibre

Non-significant difference was observed in soluble dietary fibre content of green gram and flower vegetable and pea and flower vegetable (Table 4.26). *Raita* had the minimum SDF (3.23 g/100 g) observed. It might be due to less amount of *M. oleifera* flowers and

no other ingredient was added in it except curd which had no fibre content.

4.5.8.2 Insoluble dietary fibre

Insoluble dietary fibre content of the products ranged from 7.02 to 11.18 g/100 g. *Raita* had the minimum (7.02 g/100 g) whereas it was being maximum in green gram and flower vegetable (11.18 g/100 g). Flower *bhuji* and green gram and flower vegetable had almost similar amount of IDF as there were not much variation in IDF content of peas and green gram.

4.5.9 Total carbohydrates

A wide range of total carbohydrate content i.e. 68.22 to 90.12 g/100 g was determined; the highest being in *raita* and the lowest being in green gram and flower vegetable (Table 4.26). Significant differences in total carbohydrate content of various products developed from flowers were observed among themselves. As total carbohydrate was determined by difference method and *raita* contained less content of crude fibre, ash, carbohydrate as compared to other products.

4.5.10 Antinutrients

4.5.10.1 Oxalic acid

The oxalic acid content varied from 149.60 to 161.94 mg/100 g among the various products developed from flowers (Table 4.27). Green gram and flower vegetable had the maximum (161.94 mg/100 g) followed by pea and flower vegetable (160.43 mg/100 g) and *raita* (158.64 mg/100 g) and flower *bhuji* (158.37 mg/100 g)); the minimum being observed in cutlets (149.60 mg/100 g). The oxalic acid content of products from flower was less than that in raw *M. oleifera* flowers which may be due to various processing treatments including blanching, steam cooking, sauting, frying which resulted in lowering of oxalic acid content.

Table 4.27 Antinutritional factors of products prepared from *Moringa oleifera* flowers (on dry weight basis)

Food products	Oxalic acid (mg/100 g)	Phytic acid (g/100 g)	Saponins (g/100 g)	Trypsin inhibitor activity (TIU/g)
Flower *bhuji*	158.37 ± 0.01	0.56 ± 0.01	0.34 ± 0.01	ND
Pea and Flower vegetable	160.43 ± 0.01	0.59 ± 0.01	0.41 ± 0.01	ND
Green gram and Flower vegetable	161.94 ± 0.02	0.60 ± 0.01	0.46 ± 0.01	ND
Raita	158.64 ± 0.02	0.52 ± 0.01	0.20 ± 0.01	ND
Cutlets	149.60 ± 0.01	0.52 ± 0.01	0.29 ± 0.01	ND
CD (P<0.05)	0.06	0.02	0.02	ND

Values are mean ± SE of three independent determinations
Trypsin inhibitor units: One unit of trypsin was defined as the amount of enzymes which converted one mg casein in TCA soluble components at 37°C for 20 minutes at pH 7.6

ND = Not detected

4.5.10.2 Phytic acid

Pea and flower vegetable and green gram and flower vegetable had almost similar phytic acid content (Table 4.27). Similarly, *raita* and cutlets had almost similar phytic acid contents but significantly lower than that present in rest of the products i.e. flower *bhuji*, pea and flower vegetable and green gram and flower vegetable. This might be observed due to addition of less amount of *M. oleifera* flowers.

4.5.10.3 Saponins

Saponin content ranged from 0.20 to 0.46 g/100 g in products developed from *M. oleifera* flowers (Table 4.27). Saponin content of green gram and flower vegetable was significantly ($P<0.05$) higher followed by pea and flower vegetable (0.41 g/100 g) flower *bhuji* (0.34 g/100 g) and cutlets (0.29 g/100 g). It was the lowest in raita (0.20 g/100 g). Saponin content of *raita* was lower due to less amount of fresh *M. oleifera* flowers which had lower concentration of this antinutrient. On the other hand, green gram and peas contributed more saponins in pea and flower vegetable, green flower and flower vegetable due to higher concentration of saponin in it.

4.5.10.4 Trypsin inhibitor activity

Trypsin inhibitor activity could not be detected in the products incorporating *M. oleifera* flowers. It was due to the fact that raw *M. oleifera* did not contain any trypsin inhibitor activity and moreover, due to heating, TIA could not be detected in vegetables and cutlets which besides having *Moringa oliefera* flowers had other ingredients like peas, green gram etc. too (Table 4.27).

4.5.11 Total mineral composition

Various products prepared from *M. oleifera* flowers were analysed for total calcium, phosphorus, iron, zinc and potassium (Tables 4.28 and 4.29).

4.5.11.1 Calcium

A wide range of total calcium content i.e. 101.31 to 184.62 mg/100 g was observed in various products developed from *M. oleifera* flowers (Table 4.28). *Raita* had significantly higher calcium content (184.62 mg/100 g) followed by green gram and flower vegetable (116.54 mg/100 g), pea and flower vegetable (105.61 mg/100 g), flower *bhuji* (102.52 mg/100 g) and cutlets (101.31 mg/100 g). In addition to flowers, curd used in the preparation of *raita* might have contributed towards higher calcium content in *raita*.

Some of the calcium might have leached out during the preparation of different vegetables including flower *bhuji*, green gram and flower vegetable, pea and flower vegetable and cutlets and hence, lower calcium was present in such products.

4.5.11.2 Phosphorus

Total phosphorus content of various products developed from flowers ranged from 82.92 to 115.64 mg/100 g (Table 4.28). *Raita* had the maximum phosphorus content whereas cutlets had the minimum total phosphorus content. Significant differences ($P<0.05$) in total phosphorus content were observed in various products. It might be due to variation in type and amount of ingredients like in *raita*. Phosphorus content of *raita* was higher which might be due to more content in curd. Moreover, frying involved in preparation of cutlets might have lowered the phosphorus content.

Table 4.28 Total and HCl-extractable calcium, phosphorus and iron contents (mg/100 g) of products prepared from *Moringa oleifera* flowers

Food products	Calcium			Phosphorus			Iron		
	Total	HCl-extractable	% extractability	Total	HCl-extractable	% extractability	Total	HCl-extractable	% extractability
Flower *bhuji*	102.52 ± 0.01	74.36 ± 0.01	72.50 ± 0.01	107.55 ± 0.01	75.50 ± 0.03	71.50 ± 0.01	2.32 ± 0.01	1.50 ± 0.01	64.40 ± 0.01
Pea and Flower vegetable	105.61 ± 0.01	75.65 ± 0.01	71.32 ± 0.02	109.62 ± 0.01	76.60 ± 0.01	71.62 ± 0.01	2.41 ± 0.01	1.52 ± 0.01	64.85 ± 0.01
Green gram and Flower vegetable	116.54 ± 0.02	82.08 ± 0.02	70.43 ± 0.01	108.52 ± 0.01	75.96 ± 0.05	71.82 ± 0.01	2.47 ± 0.01	1.55 ± 0.00	64.92 ± 0.01
Raita	184.62 ± 0.01	137.22 ± 0.03	74.33 ± 0.01	115.64 ± 0.02	84.56 ± 0.03	72.35 ± 0.03	1.57 ± 0.01	0.99 ± 0.01	65.12 ± 0.01
Cutlets	101.31 ± 0.02	72.11 ± 0.02	70.36 ± 0.01	82.92 ± 0.01	61.55 ± 0.02	70.37 ± 0.01	1.96 ± 0.01	1.26 ± 0.00	63.69 ± 0.01
CD (P<0.05)	0.05	0.07	0.03	0.05	0.11	0.06	0.03	0.03	0.03

Values are mean ± SE of three independent determinations

4.5.11.3 Iron

Green gram and flower vegetable had significantly ($P<0.05$) higher iron content followed by pea and flower vegetable (2.41 mg/100 g), flower *bhuji* (2.32 mg/100 g) and cutlets (1.96 mg/100 g). The lowest total iron content was observed in *raita* due to less amount of *M. oleifera* flower and no other ingredients except curd was used which had negligible iron content (Table 4.28).

4.5.11.4 Zinc

Total zinc content of products prepared from flowers ranged from 5.41 to 7.87 mg/100 g. *Raita* had significantly ($P<0.05$) lower zinc content than the other products. However, total zinc content of products i.e. flower *bhuji*, pea and flower vegetable, green gram and flower vegetable and cutlets did not vary significantly among themselves.

4.5.11.5 Potassium

Total potassium content of products developed from flowers ranged from 349.36 to 367.92 mg/100 g (Table 4.29). *Raita* had maximum potassium content whereas cutlets had the minimum potassium content. Significant differences in total potassium content among various products were observed.

4.5.12 HCl-extractability of minerals

Tables 4.28 and 4.29 depict the HCl-extractability of various minerals including calcium, phosphorus, iron, zinc and potassium in various products developed from *M. oleifera*.

4.5.12.1 Calcium

HCl-extractability of calcium ranged from 70.36 to 74.33 percent in various products prepared from *M. oleifera* flowers. Calcium extractability was found to be maximum in *raita* whereas it was minimum in cutlet. A significant ($P<0.05$) difference in HCl-extractability of calcium was noticed among various products due

Table 4.29 **Total and HCl-extractable zinc and potassium contents (mg/ 100 g) of products prepared from *Moringa oleifera* flowers (on dry weight basis)**

Food products	Zinc			Potassium		
	Total	HCl-extractable	Percent extractability	Total	HCl-extractable	Percent extractability
Flower *bhuji*	7.84 ± 0.01	5.36 ± 0.01	68.71 ± 0.01	353.73 ± 0.32	256.99 ± 0.02	72.53 ± 0.02
Pea and Flower vegetable	7.82 ± 0.01	5.35 ± 0.02	68.88 ± 0.01	358.56 ± 0.02	257.53 ± 0.01	71.25 ± 0.01
Green gram and Flower vegetable	7.86 ± 0.01	5.41 ± 0.01	69.27 ± 0.01	360.00 ± 0.34	260.00 ± 0.01	71.43 ± 0.01
Raita	5.41 ± 0.01	3.77 ± 0.01	70.13 ± 0.02	367.92 ± 0.01	263.56 ± 0.01	72.85 ± 0.01
Cutlets	7.87 ± 0.01	5.82 ± 0.01	68.30 ± 0.01	349.36 ± 0.01	254.19 ± 0.01	71.34 ± 0.01
CD (P<0.05)	0.03	0.04	0.03	0.67	0.05	0.03

Values are mean ± SE of three independent determinations

to difference in the antinutritional contents. *Raita* had significantly lower content of oxalic acid, phytic acid and saponins and availability of minerals is more (Table 4.28).

4.5.12.2 Phosphorus

The highest HCl-extractability of phosphorus was noticed in *raita* whereas cutlets had the minimum phosphorus extractability. All the products had significantly ($P<0.05$) different extractabilities of phosphorus due to variation in type and amount of ingredients and moreover, reduction of antinutrients by various processing treatments including blanching, steam cooking and frying in the preparation of products.

4.5.12.2 Iron

Iron extractability of the *raita* was significantly ($P<0.05$) higher as compared to other products developed from *M. oleifera* flowers. Significant differences ($P<0.05$) were observed in iron extractability of various products incorporating flowers of *M.oleifera*. As the level of antinutrients reduced, it caused an improvement in iron availability as these are known to hinder the availability of various minerals.

4.5.12.3 Zinc

Zinc extractability of various products developed from *M. oleifera* flowers ranged from 68.30 to 70.13 percent (Table 4.29); the highest being in *raita* and the lowest being in cutlets. Significant ($P<0.05$) differences were observed in zinc extractability of various products due to variation in type and amount of ingredients and employment of processing treatments.

4.5.12.5 Potassium

The maximum content of potassium extractability was found in *raita* and lowest in pea and flower vegetable. Overall comparison showed significant differences in potassium extractability among

various products developed from flowers (Table 4.29). The lower contents of oxalic acid, phytic acid and saponins in *raita* might be the reason for maximum extractability of potassium in *raita* .

4.5.13 In vitro availability of calcium and iron

Calcium availability was found maximum in green gram and flower vegetable and minimum in cutlets. Iron availability was more in pea and flower vegetable and the least in cutlets. Significant differences ($P<0.05$) were observed among various products as far as calcium availability as well as iron availability were concerned. Various processing treatments, like blanching, steam cooking, frying used in the preparation of these products resulted in significant reduction of various antinutrients. As the level of antinutrients, like oxalic acid, phytic acid and saponins reduced, a significant increase occurred in bioavailability of various minerals including calcium and iron (Table 4.30). As these antinutrients bind with the minerals, form complexes and interfere in the absorption and thus, lowers the availability. The results of the present findings are in agreement with the findings of the Saharan (1994).

4.6 Nutritional composition of products prepared from fresh *M. oleifera* Pods

4.6.1 Moisture

Moisture content of products developed from fresh *M. oleifera* pods ranged from 36.69 to 80.54 percent (Table 4.31). Maximum moisture content (80.54%) was in *sambhar*; whereas minimum was in pickle. All the products had significantly different ($P<0.05$) moisture contents.

Increase in moisture content may be due to more amount of water used in *sambhar* as compared to other products. In pickle,

Table 4.30 ***In vitro* availability (%) of calcium and iron of products prepared from *Moringa oleifera* flowers (on dry weight basis)**

Food products	Calcium		Iron	
	Total (mg/100 g)	**Percent availability (*in vitro*)**	**Total (mg/100 g)**	**Percent availability (*in vitro*)**
Flower *bhuji*	102.52 ± 0.01	26.56 ± 0.01	2.32 ± 0.01	13.65 ± 0.01
Pea and Flower vgetable	105.61 ± 0.01	26.50 ± 0.02	2.41 ± 0.01	13.70 ± 0.02
Green gram and Flower vegetable	116.54 ± 0.02	27.15 ± 0.05	2.47 ± 0.01	13.60 ± 0.01
Raita	184.62 ± 0.01	25.53 ± 0.01	1.96 ± 0.01	12.68 ± 0.03
Cutlets	101.31 ± 0.02	25.05 ± 0.02	1.57 ± 0.01	12.56 ± 0.05
CD (P<0.05)	0.05	0.04	0.03	0.04

Values are mean ± SE of three independent determinations

Table 4.31 Nutrient composition of products prepared from *Moringa oleifera* pods (on fresh weight basis)

Food products	Moisture (%)	Ascorbic acid (mg/100 g)	β-carotene (mg/100 g)
Pickle	36.69 ± 0.02	89.32 ± 0.02	0.09 ± 0.01
Pod vegetable	63.73 ± 0.02	16.33 ± 0.02	0.06 ± 0.01
Potato and pod vegetable	63.86 ± 0.01	16.57 ± 0.01	0.07 ± 0.01
Sambhar	80.54 ± 0.01	14.20 ± 0.03	0.06 ± 0.01
Cutlets	61.26 ± 0.01	5.50 ± 0.01	0.05 ± 0.01
CD (P<0.05)	0.04	0.03	0.01

Values are mean ± SE of three independent determinations

addition of oil and salt might have resulted in less moisture content as these bind with water and create unaerobic conditions.

4.6.2 Ascorbic acid

A wide range of ascorbic acid content i.e. 5.50 to 89.32 mg/100 g was observed in various products; the highest being in pickle and the lowest being in cutlets (Table 4.31). Overall, significant ($P<0.05$) differences were noticed in ascorbic acid content of different developed products. Ascorbic acid subsequently decreased due to various processing treatments like, blanching, frying, steam cooking used in the preparation of various products including pod vegetable, potato and pod vegetable, *sambhar* and cutlets.

4.6.3 β-carotene

The β-carotene content ranged from 0.05 to 0.09 mg/100 g among various fresh pod products. Pickle had maximum (0.09 mg/100 g) β-content whereas cutlets had minimum β-carotene content. β-carotene content of pickle was significantly higher than potato and pod vegetable whereas pod vegetable and *sambhar* had almost similar β-carotene contents. Significant increase in the β-carotene content of pickle might have been due to addition of more amount of *M. oleifera* pods containing appreciable β-carotene.

4.6.4 Crude protein

Protein content among the various products varied from 4.93 to 11.42 g/100 g; the highest (11.42 g/100 g) being in pod pickle and the lowest being in cutlets. Protein contents of various products differed significantly ($P<0.05$) among themselves due to varying amount and type of ingredients used in the preparation of

various products. Protein content of pickle was higher which might be due to more amount of pods used in pickle (Table 4.2).

4.6.5 Crude Fat

A wide range of fat content i.e. 16.74 to 31.33 g/100 g (on DM basis) was observed in products prepared from fresh *M. oleifera* pods (Table 4.32). The fat content of pickle (31.33 g/100 g) was significantly ($P<0.05$) higher followed by cutlets (19.36 g/100 g) and potato and pod vegetable (18.41 g/100 g) in descending order. The lowest fat content was found in *sambhar* i.e. 16.74 g/100 g. Higher fat content in pickle and cutlets might be due to incorporation of higher amount of visible fat.

4.6.6 Total ash

The highest amount of total ash was found in cutlets (1.41 g/100 g) whereas it was the lowest in *sambhar* (0.65 g/100 g). The ash content of pod vegetable and potato and pod vegetable was almost similar. Slight differences in ash content might be due to variation in type and amount of ingredients.

4.6.7 Crude fibre

Crude fibre content of pickle was significantly ($P<0.05$) higher than the other products prepared from fresh *M. oleifera* pods.

Among various products, cutlets had minimum crude fibre content. Overall comparison indicated significant differences in crude fibre contents among various fresh *M. oleifera* pod products. The higher crude fibre content of pickle may be due to use of more amount of pods which had appreciably higher amount of crude fibre (Table 4.2).

4.6.8 Total dietary fibre

Total dietary fibre content of products ranged from 22.67 to 39.35 g/100 g (Table 4.32). Pickle had the maximum (39.35 g/100 g) TDF whereas cutlets had the minimum (24.42 g/100 g) TDF

Table 4.32 **Nutrient composition of products prepared from *Moringa oleifera* pods (g/100 g, on dry weight basis)**

Food products	Crude protein	Crude fat	Total ash	Crude fibre	Dietary fibre			Total carbohy-drates
					Total	Soluble	Insoluble	
Pickle	11.42 ± 0.02	31.33 ± 0.02	0.91 ± 0.02	8.43 ± 0.01	39.35 ± 0.03	9.56 ± 0.01	29.79 ± 0.00	47.81 ± 0.01
Pod vegetable	6.18 ± 0.02	18.41 ± 0.02	0.76 ± 0.01	6.72 ± 0.02	32.91 ± 0.01	7.57 ± 0.02	25.34 ± 0.01	67.48 ± 0.01
Potato and pod vegetable	6.11 ± 0.01	18.51 ± 0.01	0.77 ± 0.01	7.09 ± 0.01	30.40 ± 0.02	7.57 ± 0.01	22.83 ± 0.01	67.43 ± 0.04
Sambhar	5.85 ± 0.03	16.74 ± 0.04	0.65 ± 0.02	4.32 ± 0.02	24.42 ± 0.01	6.49 ± 0.01	17.93 ± 0.01	72.36 ± 0.01
Cutlets	4.93 ± 0.01	19.36 ± 0.01	1.41 ± 0.01	3.73 ± 0.02	22.67 ± 0.02	6.16 ± 0.01	16.50 ± 0.01	70.52 ± 0.01
CD (P<0.05)	0.03	0.06	0.03	0.04	0.03	0.02	0.02	0.03

Values are mean ± SE of three independent determinations

content. Significant (P<0.05) differences were found in TDF contents of various products. Dietary fibre content of pickle was higher due to more amount of pods which had higher TDF (Table 4.2).

4.6.8.1 Soluble dietary fibre

Soluble dietary fibre content of pickle was significantly (P<0.05) higher than the other products prepared from fresh *M. oleifera* pods.

Non-significant differences were observed among pod vegetable and potato and pod vegetable. Cutlets had the minimum (6.16 g/100 g) soluble dietary fibre. Increase in soluble dietary fibre content of pickle was observed due to addition of more amount of *M. Oleifera* pods having appreciable amount of soluble dietary fibre (Table 4.2).

4.6.8.2 Insoluble dietary fibre

Insoluble dietary fibre content ranged from 16.50 to 29.79 g/100 g; the highest being in pickle and the lowest being in cutlets (Table 4.29). Significant (P<0.05) differences in IDF were observed among various products when compared among themselves due to variation in type and amount of ingredients used in their preparation.

4.6.9 Total carbohydrates

Total carbohydrate content was maximum (72.36 g/100 g) in *sambhar* whereas it was minimum (47.81 g/100 g) in pickle (Table 4.32). All the products significantly (P<0.05) differed in carbohydrate content when compared among themselves. Besides *M. Oleifera* pods, other ingredients i.e. onion, tomato, potato and cornflour used in the preparation of various products contributed towards total carbohydrates.

4.6.10 Antinutrients

4.6.10.1 Oxalic acid

The oxalic acid content ranged from 248.51 to 300.23 mg/100 g among the various products developed from fresh *M. oleifera* pods (Table 4.33). Pickle had the maximum (300.23 mg/100 g) whereas cutlets had the minimum (248.51 mg/100 g) oxalic acid content. The oxalic acid contents of products prepared from fresh *M. oleifera* pods differed significantly ($P<0.05$) among themselves. Processing treatments e.g. blanching, steam cooking, sauting, frying resulted in lowering of oxalic acid content as observed in *sambhar* and cutlets when compared to that of pickle.

4.6.10.2 Phytic acid

There were significant ($P<0.05$) differences in phytic acid contents of various products. Pickle had the maximum (1.21 mg/100 g) while *sambhar* and potato and pod vegetable had the minimum (0.84 and 0.87 mg/100 g) phytic acid contents (Table 4.33). As there was no processing treatment like blanching, frying etc. involved in the preparation of pickle and moreover, addition of more amount of pods having appreciable amount of phytic acid might have contributed to higher phytic acid.

4.6.10.3 Saponins

Saponin content ranged from 0.40 to 0.57 g/100 g (Table 4.33). Pickle had significantly ($P<0.05$) higher saponin content than rest of the products. However, non-significant differences were observed in phytic acid contents of pod vegetable, potato and pod vegetable and cutlets due to cumulative effect of processing of various treatments. There was sufficient reduction in saponin content.

Table 4.33 Antinutritional factors of products prepared from *Moringa oleifera* pods (on dry weight basis)

Food products	Oxalic acid (mg/100 g)	Phytic acid (g/100 g)	Saponins (g/100 g)	Trypsin inhibitor activity (TIU/g)
Pickle	300.23 ± 0.37	1.21 ± 0.02	0.57 ± 0.01	ND
Pod vegetable	263.41 ± 0.01	0.91 ± 0.01	0.48 ± 0.01	ND
Potato and pod vegetable	265.56 ± 0.05	0.87 ± 0.01	0.47 ± 0.01	ND
Sambhar	260.74 ± 0.01	0.84 0.01	0.40 ± 0.01	ND
Cutlets	248.51 ± 0.01	1.02 ± 0.01	0.49 ± 0.01	ND
CD (P<0.05)	0.52	0.04	0.02	ND

Values are mean ± SE of three independent determinations
Trypsin inhibitor units: One unit of trypsin was defined as the amount of enzymes which converted one mg casein in TCA soluble components at 37°C for 20 minutes at pH 7.6

ND = Not detected

4.6.10.4 Trypsin inhibitor activity

Trypsin inhibitor activity could not be detected in any of the products developed from fresh *M. oleifera* pods. It was due to no trypsin inhibitor activity in raw *M. oleifera* pods. Moreover, if any TIA found in products was destroyed due to cooking and frying.

4.6.11 Total mineralcomposition

Various products prepared from fresh *M. oleifera* pods were analysed for total calcium, phosphorus, iron, zinc and potassium (Tables 4.34 and 4.35).

4.6.11.1 Calcium

A wide range of total calcium content i.e. 79.36 to 116.36 mg/100 g was observed in various products developed from fresh *M. oleifera* pods (Table 4.34). Pickle had significantly higher calcium content (116.36 mg/100 g) followed by potato and pod vegetable (83.56 mg/100 g), pod vegetable (81.65 mg/100 g), *sambhar* (80.85 mg/100 g) and cutlets (79.36 mg/100 g). Higher calcium content in pickle might have been observed or due to more amount of *M. Oleifera* pods used in pickle preparation. In other products calcium content was low due to it loss during various processing treatments involved in preparation.

4.6.11.2 Phosphorus

Total phosphorus content of various products developed from fresh *M. oleifera* pods ranged from 132.30 to 260.55 mg/100 g. Pickle had the maximum phosphorus content whereas cutlets had the minimum phosphorus content.

Significant ($P<0.05$) differences in total phosphorus content were observed among various products developed from fresh *M. oleifera* pods.

Table 4.34 **Total and HCl-extractable calcium, phosphorus and iron contents (mg/100 g) of products prepared from *Moringa oleifera* pods**

Food products	Calcium			Phosphorus			Iron		
	Total	HCl-extractable	% extractability	Total	HCl-extractable	% extractability	Total	HCl-extractable	% extractability
Pickle	116.36 ± 0.01	85.08 ± 0.04	72.16 ± 0.02	260.55 ± 0.01	182.49 ± 0.02	70.32 ± 0.02	18.50 ± 0.02	11.54 ± 0.02	60.33 ± 0.02
Pod vegetable	81.65 ± 0.01	59.84 ± 0.03	73.34 ± 0.02	135.72 ± 0.02	87.42 ± 0.04	70.37 ± 0.01	10.53 ± 0.02	5.86 ± 0.01	61.52 ± 0.01
Potato and pod vegetable	83.56 ± 0.01	60.62 ± 0.02	73.83 ± 0.02	136.62 ± 0.02	86.59 ± 0.01	70.85 ± 0.01	11.64 ± 0.02	6.93 ± 0.01	61.36 ± 0.03
Sambhar	80.85 ± 0.03	59.64 ± 0.01	74.3C ± 0.01	135.92 ± 0.01	98.36 ± 0.06	72.36 ± 0.01	11.82 ± 0.04	7.26 ± 0.01	61.55 ± 0.01
Cutlets	79.36 ± 0.01	57.27 ± 0.02	73.26 ± 0.01	132.30 ± 0.03	97.48 ± 0.02	70.54 ± 0.01	10.52 ± 0.01	6.27 ± 0.02	59.57 ± 0.02
CD (P<0.05)	0.02	0.07	0.04	0.06	0.08	0.04	0.05	0.04	0.04

Values are mean ± SE of three independent determinations

Total phosphorus content of pickle was higher as compared to other products which might be due to higher amount of pods used in pickle preparation (Table 4.4).

4.6.11.3 Iron

Total iron content of products developed from fresh *M. oleifera* pods has been depicted in Table 4.34. Pickle had the maximum (18.50 mg/100 g) iron content whereas it was minimum (10.52 mg/100 g) in cutlets. Iron content of cutlet differed non-significantly from pod vegetable.

Total iron content of pickle was higher as compared to other products which may be due to higher amount of pods used in pickle preparation (Table 4.4). Moreover, various processing treatments like blanching, frying, steam cooking might have resulted in loss of iron content of various products.

4.6.11.4 Zinc

Zinc content ranged from 12.51 to 16.47 mg/100 g in various products (Table 4.35). Pickle had significantly higher zinc content followed by *sambhar* and potato and pod vegetable and pod vegetable in descending order. The minimum zinc content was observed in cutlets. However, potato and pod vegetable and *sambhar* had almost similar total zinc contents. The appreciable higher amount of *M. Oleifera* pods incorporated in pickle might have higher zinc content.

4.6.11.5 Potassium

Total potassium content of products developed from fresh *M. oleifera* pods ranged from 953.83 to 1165.39 mg/100 g (Table 4.35). Pickle had the maximum whereas cutlets had minimum potassium content. Significant ($P<0.05$) differences in total potassium contents of various products might be due to

variation in type, amount and combination of various processing treatments involved in preparation of various products.

4.6.12 HCl-extractability of minerals

Tables 4.34 and 4.35 depict the HCl-extractability of various minerals and potassium in various minerals including calcium, phosphorus, iron, zinc and potassium in various products developed from fresh *M. oleifera* pods.

4.6.12.1 Calcium

HCl-extractability of calcium ranged from 72.16 to 74.33 percent in various products developed from fresh *M. oleifera* pods (Table 4.34). Calcium extractability was found maximum in *sambhar* and minimum in pickle. Significant differences were observed in calcium extractability of various products due to reduction in oxalic acid, phytic acid and saponins to varying extent (Table 4.33).

4.6.12.2 Phosphorus

The highest phosphorus extractability was observed in *sambhar* and the lowest in cutlets in pickle. Pod vegetable and cutlets had almost similar phosphorus extractability. As the level of antinutrients increases, there is decrease in the availability of phosphorus content and the same holds true for less phosphorus extractability in pickle.

4.6.12.3 Iron

Iron extractability of the *sambhar* was significantly ($P<0.05$) higher than the other products developed from fresh *M. oleifera* pods. However, pod vegetable and potato and pod vegetable had similar values for iron extractability. It might have been due to reduction in antinutrients as a result of various processing treatments which resulted in an increase in the extractability of iron.

Table 4.35 **Total and HCl-extractable zinc and potassium contents (mg/ 100 g) of products prepared from *Moringa oleifera* pods (on dry weight basis)**

Food products	Zinc			Potassium		
	Total	HCl-extractable	Percent extractability	Total	HCl-extractable	Percent extractability
Pickle	16.47 ± 0.02	11.34 ± 0.01	68.70 ± 0.03	1165.39 ± 0.35	823.57 ± 0.02	70.61 ± 0.02
Pod vegetable	12.55 ± 0.01	8.74 ± 0.01	69.77 ± 0.01	966.35 ± 0.58	691.70 ± 4.21	71.45 ± 0.53
Potato and pod vegetable	12.61 ± 0.01	8.78 ± 0.03	70.14 ± 0.02	952.56 ± 0.04	692.28 ± 0.04	72.45 ± 0.03
Sambhar	12.64 ± 0.01	8.87 ± 0.01	70.22 ± 0.02	955.30 ± 0.33	696.82 ± 0.02	72.83 ± 0.02
Cutlets	12.51 ± 0.01	8.81 ± 0.01	68.32 ± 0.02	953.83 ± 0.02	680.54 ± 0.02	71.32 ± 0.02
CD (P<0.05)	0.03	0.03	0.06	1.07	5.93	0.76

Values are mean ± SE of three independent determinations

4.6.12.4 Zinc

Zinc extractability of various products developed from fresh *M. oleifera* pods ranged from 68.32 to 70.22 percent (Table 4.35), the highest being in *sambhar* and the lowest being in cutlets. Significant ($P<0.05$) differences were observed in zinc extractability of all the products which might be due to difference in level of antinutrients present and the type of processing treatments used in the development of products.

4.6.12.5 Potassium

Potassium extractability was found to be maximum in *sambhar* and minimum in pickle. Overall various products had almost similar extractability of potassium. Difference in percent extractability was observed due to difference in level of antinutrients present and processing treatments employed in the developed products.

4.6.13 In vitro availability of calcium and iron

Calcium availability was found to be maximum in pod vegetable and minimum in cutlets. On the other hand, iron availability was found to be the highest in *sambhar* and the lowest in cutlets. Calcium availability of pod vegetable and potato and pod vegetable was almost similar. Similarly, iron availability of pod vegetable and *sambhar* was almost the same. It might have been observed due to cumulative effect of various processing treatments including blanching, steam cooking and frying which had reduced the level of antinutrients and increasing the bioavailability of calcium and iron in various products developed as reported earlier by Saharan (1994).

Table 4.36 ***In vitro*** **availability (%) of calcium and iron of products prepared from *Moringa oleifera* pods (on dry weight basis)**

Food products	Calcium		Iron	
	Total (mg/100 g)	Percent availability (*in vitro*)	Total (mg/100 g)	Percent availability (*in vitro*)
Pickle	116.36 ± 0.01	23.82 ± 0.03	18.50 ± 0.02	13.25 ± 0.03
Pod vegetable	81.65 ± 0.01	24.85 ± 0.01	10.53 ± 0.02	13.75 ± 0.01
Potato and pod vegetable	83.56 ± 0.01	24.80 ± 0.02	11.64 ± 0.02	13.70 ± 0.02
Sambhar	80.85 ± 0.03	24.70 ± 0.01	11.82 ± 0.04	13.79 ± 0.05
Cutlets	79.36 ± 0.01	22.70 ± 0.05	10.52 ± 0.01	12.83 ± 0.03
CD (P<0.05)	0.02	0.05	0.05	0.06

Values are mean ± SE of three independent determinations

4.7 Nutritional composition of products prepared from dried *M. oleifera* pods

Sev and noodles were prepared by incorporating *M. oleifera* pod powder at two different levels i.e. 10 and 20 percent. These products were analysed for various nutrients and the results are given as under:

4.7.1 Moisture

Moisture content of *sev* prepared by incorporating *M. oleifera* leaf powder at 10 percent and 20 percent was almost similar but it was significantly ($P<0.05$) higher than the control.

Noodles supplemented with 20 percent *Moringa* pod powder had significantly higher moisture content followed by 10 percent noodles and control. It might be due to high protein content of noodles supplemented with 20 percent *Moringa oleifera* pod powder which indicated a possible relationship between water absorption and protein content.

4.7.2 Ascorbic acid

Ascorbic acid was not detectable in various types of *sev* whereas different types of noodles had almost similar amount of ascorbic acid and it ranged from 0.08 to 0.13 mg/100 g (Table 4.37). The ascorbic acid is most sensitive to heat and might have been lost during cooking involved in the preparation of *sev* and noodles.

4.7.3 β-carotene

β-carotene content was significantly ($P<0.05$) higher in *sev* prepared from 20 percent *M. oleifera* pod powder as compared to *sev* prepared from 10 percent *M. oleifera* pod powder and control.

Higher the level of supplementation with *M. oleifera* pod powder, higher was the β-carotene in different types of noodles. All the different types of noodles had almost similar values for β-

Table 4.37 Nutrient composition of products prepared from *Moringa oleifera* pods (on fresh weight basis)

Food products	Moisture (%)	Ascorbic acid (mg/100 g)	β-carotene (mg/100 g)
Sev			
Bengal gram flour (control 100%)	5.43 ± 0.01	ND	0.12 ± 0.01
Bengal gram flour + *Moringa oleifera* pods (10%)	6.72 ± 0.02	ND	0.14 ± 0.02
Bengal gram flour + *Moringa oleifera* pods (20%)	6.76 ± 0.01	ND	0.16 ± 0.03
CD (P<0.05)	0.04	ND	0.01
Noodles			
Refined flour (control (100%)	59.44 ± 0.01	0.08 ± 0.01	0.03 ± 0.02
Refined flour + *Moringa oleifera* pods (10%)	59.95 ± 0.01	0.12 ± 0.02	0.04 ± 0.01
Refined flour + *Moringa oleifera* pods (20%)	60.33 ± 0.01	0.13 ± 0.02	0.06 ± 0.01
CD (P<0.05)	0.02	0.04	0.02

Values are mean ± SE of three independent determinations

carotene. As *M. oleifera* pod powder is a good source of β-carotene (Table 4.1), its incorporation led to higher content of β-carotene in the products too.

4.7.4 Crude protein

On dry matter basis, the protein content of *sev* varied from 20.74 to 22.91 g/100 g (Table 4.38). Maximum protein content was in *sev* supplemented with 20 percent *M. oleifera* pod powder.

Similarly, noodles supplemented with 20 percent *M. oleifera* pod powder had significantly higher protein content followed by *sev* (10% supplementation) and control.

Powder of *M. oleifera* pod contained 15.76 g protein/100 g (Table 4.2) and therefore, its incorporation caused an increase in protein content of *sev* and noodles supplemented with 20 percent *Moringa oleifera* pod powder.

4.7.5 Crude Fat

On dry matter basis, the fat content of *sev* supplemented with 20 percent *M. oleifera* pod powder was found to be significantly higher over the *sev* supplemented with 10 percent *M. oleifera* pod powder and control.

Fat content i.e. 12.61 to 12.86 g/100 g was found in different types of noodles, however, differences were non-significant. *M. oleifera* pod powder contained 0.32 g fat/100 g (Table 4.2) and its incorporation in the products led to increase in their crude fat content.

4.7.6 Total ash

Total ash content of *sev* supplemented with 20 percent *M. oleifera* pod powder was significantly higher as compared to *sev* having 10 percent *M. oleifera* and control (Table 4.38).

Noodles prepared with 20 percent and 10 percent *M. oleifera* pod powder had almost similar total ash contents. However,

Table 4.38 ***Nutrient composition of products prepared from *Moringa oleifera* pods (g/100 g, on dry weight basis)***

Food products	Crude protein	Crude fat	Total ash	Crude fibre	Dietary fibre			Total carbohydrates
					Total	Soluble	Insoluble	
Sev								
Bengal gram flour (control 100%)	20.74 ± 0.02	20.16 ± 0.02	2.60 ± 0.02	1.25 ± 0.03	15.69 ± 0.02	4.58 ± 0.01	11.11 ± 0.06	55.25 ± 0.01
Bengal gram flour + *Moringa oleifera* pods (10%)	21.97 ± 0.01	21.65 ± 0.01	3.56 ± 0.01	2.57 ± 0.01	19.57 ± 0.01	5.36 ± 0.01	14.21 ± 0.02	50.25 ± 0.03
Bengal gram flour + *Moringa oleifera* pods (20%)	22.91 ± 0.01	21.68 ± 0.05	4.10 ± 0.01	3.77 ± 0.05	23.25 ± 0.04	6.57 ± 0.01	16.68 ± 0.03	47.50 ± 0.02
CD (P<0.05)	0.02	0.03	0.03	0.02	0.03	0.03	0.01	0.03
Noodles								
Refined flour (control 100%)	9.32 ± 0.01	12.86 ± 0.01	2.56 ± 0.02	1.29 ± 0.01	12.18 ± 0.01	3.15 ± 0.01	9.02 ± 0.01	73.95 ± 0.02
Refined flour + *Moringa oleifera* pods (10%)	10.79 ± 0.01	12.64 ± 0.01	2.74 ± 0.01	2.64 ± 0.01	16.69 ± 0.02	4.30 ± 0.01	12.39 ± 0.01	71.16 ± 0.02
Refined flour + *Moringa oleifera* pods (20%)	11.79 ± 0.01	12.61 ± 0.01	2.77 ± 0.02	3.93 ± 0.02	20.82 ± 0.01	5.45 ± 0.02	15.37 ± 0.03	68.83 ± 0.02
CD (P<0.05)	0.02	0.02	0.03	0.04	0.04	0.03	0.02	0.02

Values are mean ± SE of three independent determinations

control had significantly (P<0.05) less content of total ash over the noodles supplemented with 10 percent and 20 percent *M. oleifera* pod powder. This might be due to incorporation of higher mineral matter present in the *M. oleifera* pod powder (Table 4.2).

4.7.7 Crude fibre

Crude fibre content of different type of *sev* varied from 1.25 to 3.77 g/100 g (Table 4.38). Maximum crude fibre was found in *sev* supplemented with 20 percent *M. oleifera* pod powder whereas it was minimum in control. Crude fibre content of both the *sev* varied significantly (P<0.05).

Similar trend was observed in the crude fibre content of noodles. As the level of supplementation of *M. oleifera* increased, crude fibre content increased due to higher crude fibre content of *M. oleifera* pods (Table 4.2).

4.7.8 Total dietary fibre

Results of total dietary fibre, soluble dietary fibre and insoluble dietary fibre are presented in Table 4.38.

Total dietary fibre content of control *sev* increased significantly (P<0.05) from 15.69 g/100 g to 19.57 and 23.25 g/100 g, *sev* supplemented with 10 and 20 percent of *Moringa oleifera* pod powder, respectively.

Similarly, noodles prepared by incorporating 20 percent *M. oleifera* pods had significantly higher TDF content followed by noodles containing 10 percent *Moringa oleifera* pod powder and control. Results of present study are due to appreciably higher TDF content of *M. oleifera* pods (Table 4.2).

4.7.8.1 Soluble dietary fibre

Soluble dietary fibre content of *sev* varied from 4.58 to 6.57 g/100 g in different types of *sev*. Maximum value of SDF was observed in *sev* supplemented with 20 percent *M. oleifera* pod

powder. Similar trend was also observed in different types of noodles. *M. oleifera* pod powder contained 7.28 g/100 g SDF content (Table 4.2) and therefore, its incorporation resulted in an increase in SDF content of the products.

4.7.8.2 Insoluble dietary fibre

As observed in data, insoluble dietary fibre content was significantly ($P<0.05$) higher in *sev* and noodles supplemented with 20 percent *M. oleifera* pod powder over the control. Due to addition of appreciably higher amount of insoluble dietary fibre in *M. oleifera* pods powder (Table 4.2), its content increased in supplemented *sev* and noodles too.

4.7.9 Total carbohydrates

A significant ($P<0.05$) difference was observed in total carbohydrate content of different types of *sev* (Table 4.38). The maximum total carbohydrate content was in control *sev* and minimum in *sev* supplemented with 20 percent *M. oleifera* pod powder.

Similar trend was also observed in noodles prepared by incorporation of different levels of *M. oleifera* pod powder. It might be due to higher carbohydrate content of refined flour and chickpea flour as compared to *M. oleifera* pod powder.

4.7.10 Antinutrients

Among the antinutritional factors, data regarding oxalic acid, phytic acid, saponins and trypsin inhibitor activity of *sev* and noodles prepared by incorporation of different levels of pods i.e. 10 percent and 20 percent are presented in Table 4.39.

4.7.10.1 Oxalic acid

A wide range of oxalic acid i.e. 11.89 to 95.53 mg/100 g (DM basis) was observed in different types of *sev*. *Sev* supplemented with 20 percent *M. oleifera* pod powder had significantly ($P<0.05$)

Table 4.39 Antinutritional factors of products prepared from *Moringa oleifera* pods (on dry weight basis)

Food products	Oxalic acid (mg/100 g)	Phytic acid (g/100 g)	Saponins (g/100 g)	Trypsin inhibitor activity (TIU/g)
Sev				
Bengal gram flour (control 100%)	11.89 ± 1.01	0.46 ± 0.01	0.29 ± 0.01	98.66 ± 0.02
Bengal gram flour + *Moringa oleifera* pods (10%)	54.62 ± 0.02	0.51 ± 0.02	0.41 ± 0.03	98.64 ± 0.03
Bengal gram flour + *Moringa oleifera* pods (20%)	95.53 ± 0.02	0.52 ± 0.01	0.52 ± 0.02	98.65 ± 0.02
CD (P<0.05)	2.02	0.02	0.03	0.03
Noodles				
Refined flour (control 100%)	217.76 ± 0.01	0.40 ± 0.01	0.26 ± 0.01	103.32 ± 0.02
Refined flour + *Moringa oleifera* pods (10%)	252.75 ± 0.02	0.42 ± 0.01	0.38 ± 0.01	103.34 ± 0.01
Refined flour + *Moringa oleifera* pods (20%)	287.62 ± 0.01	0.43 ± 0.01	0.48 ± 0.02	103.36 ± 0.02
CD (P<0.05)	0.04	0.01	0.03	0.05

Values are mean ± SE of three independent determinations

Trypsin inhibitor units: One unit of trypsin was defined as the amount of enzymes which converted one mg casein in TCA soluble components at 37°C for 20 minutes at pH 7.6

higher oxalic acid content than the *sev* supplemented with 10% *M. oleifera* pod powder and control *sev*.

Similarly, noodles supplemented with 20 percent *M. oleifera* pod had the maximum whereas control noodles had the minimum oxalic acid content. The oxalic acid content of different types of noodles varied significantly among themselves. Raw *M. oleifera* pod powder had considerable amount of oxalic acid (Table 4.3) and its incorporation led to increased amount of this antinutrient in the product. However, it was lower in products than raw pod due to reduction caused by processing treatments also which were used in preparation of noodles and *sev*.

4.7.10.2 Phytic acid

The phytic acid content of various types of *sev* (control, 10% and 20% supplementation) was 0.46, 0.51 and 0.52 mg/100 g, respectively. Phytic acid content of *sev* supplemented with 10 percent and 20 percent *M. oleifera* pod powder was almost similar. Control *sev* had significantly ($P<0.05$) lower phytic acid content as compared to both types of supplemented *sev*.

Similar trend was also observed in different types of noodles. This is due to higher phytic acid content of *M. oleifera* pod powder (Table 4.3).

4.7.10.3 Saponins

Saponin content of different types of *sev* ranged from 0.29 to 0.52 g/100 g. Maximum saponin content was found in *sev* supplemented with 20 percent *M. oleifera* pod powder.

Similarly, the highest (0.48 g) saponin content was found in noodles supplemented with 20% and the lowest (0.26 g/100 g) in control. Higher saponin content in *sev* and noodles (20%) was due to appreciably higher amount of *M. oleifera* pod powder (Table 3).

However, it was lower in products than raw pods due to cumulative effect of various processing treatments.

4.7.10.4 Trypsin inhibitor activity

Trypsin inhibitor activity in different types of *sev* was almost similar. Similarly, different types of noodles had no significant difference in TIA (Table 4.39). It might be due to the reason that trypsin inhibitor activity was not detected in raw *M. oleifera* pods. Moreover, processing treatments involved in preparation of *sev* and noodles also reduced the trypsin inhibitor activity.

4.7.11 Total mineral composition

Data regarding the calcium, phosphorus, iron, zinc and potassium contents of *M. oleifera* pods supplemented *sev* and noodles are presented in Tables 40 and 41.

4.7.11.1 Calcium

Sev prepared by incorporating 20 percent *M. oleifera* pod powder had significantly higher calcium content as compared to *sev* (10% supplementation) and control. Similar trend was also observed in noodles supplemented with different levels of *M. oleifera* pod powder.

The reason for increase in total calcium content was due to addition of higher amount of *M. oleifera* pod powder i.e. as the content of *M. oleifera* pod increased subsequently there was an increase in total calcium content of various food products.

4.7.11.2 Phosphorus

Total phosphorus contents of *sev* and noodles are presented in Table 4.40.

Sev and noodles prepared from 20 percent *M. oleifera* pod powder had significantly ($P<0.05$) higher phosphorus content followed by *sev* and noodles having 10% supplementation of *M.*

Table 4.40 **Total and HCl-extractable calcium, phosphorus and iron contents (mg/100 g) of products prepared from *Moringa oleifera* pods**

Food products	Calcium			Phosphorus			Iron		
	Total	HCl-extractable	% extractability	Total	HCl-extractable	% extractability	Total	HCl-extractable	% extractability
Sev									
Bengal gram flour (control 100%)	78.23 ± 0.01	54.99 ± 0.01	70.32 ± 0.02	118.74 ± 0.01	84.52 ± 0.02	71.17 ± 0.02	3.86 ± 0.01	2.37 ± 0.02	62.47 ± 0.02
Bengal gram flour + *Moringa oleifera* pods (10%)	87.35 ± 0.03	60.71 ± 0.05	69.50 ± 0.02	148.81 ± 0.03	104.74 ± 0.03	70.37 ± 0.01	5.86 ± 0.05	3.56 ± 0.01	61.32 ± 0.05
Bengal gram flour + *Moringa oleifera* pods (20%)	99.51 ± 0.01	69.20 ± 0.03	69.55 ± 0.02	175.35 ± 0.05	124.19 ± 0.02	70.82 ± 0.04	7.96 ± 0.05	4.91 ± 0.02	61.85 ± 0.03
CD (P<0.05)	0.03	0.02	0.04	0.05	0.07	0.05	0.02	0.04	0.04
Noodles									
Refined flour (control 100%)	50.12 ± 0.01	35.78 ± 0.03	71.48 ± 0.03	243.97 ± 0.39	174.65 ± 0.01	71.36 ± 0.01	5.42 ± 0.01	3.42 ± 0.01	63.57 ± 0.01
Refined flour + *Moringa oleifera* pods (10%)	62.74 ± 0.01	45.08 ± 0.01	71.66 ±0.01	261.50 ± 0.01	187.89 ± 0.01	71.85 ± 0.01	7.47 ± 0.01	4.72 ± 0.01	63.61 ± 0.01
Refined flour + *Moringa oleifera* pods (20%)	73.57 ±0.02	52.71 ± 0.01	71.85 ± 0.01	279.31 ± 0.02	201.20 ± 0.09	72.04 ± 0.02	9.54 ± 0.01	6.03 ± 0.01	63.63 ± 0.02
CD (P<0.05)	0.02	0.04	0.03	0.79	0.19	0.04	0.02	0.05	0.04

Values are mean ± SE of three independent determinations

oleifera pod powder, respectively (10%) and control. This is due to higher phosphorus content of *M. oleifera* pod powder (Table 4.4).

4.7.11.3 Iron

Total iron content of different types of *sev* prepared ranged from 3.86 to 7.96 mg/100 g. Minimum iron content was found in control and maximum was found in *sev* supplemented with 20 percent *M. oleifera* pod powder.

Similarly, total iron content of noodles ranged from 5.42 to 9.54 mg/100 g. Noodles supplemented with 20 percent *M. oleifera* pod powder had the highest iron content due to *M. oleifera* pod (on DM basis) powder being good source of iron (Table 4.4).

4.7.11.4 Zinc

Significant ($P<0.05$) differences were observed in zinc content of different types of *sev* and noodles. Zinc content was the highest in *sev* and noodles prepared by incorporation of 20 percent *M. oleifera* pod powder whereas it was the lowest in control (both *sev* and noodles) i.e. prepared without the addition of *M. oleifera* pod powder. The reason for increase in total zinc content was due to addition of higher amount of *M. oleifera* pod powder.

4.7.11.5 Potassium

A wide range of potassium content was observed in different types of *sev* and noodles (Table 41). Potassium content of *sev* (20% *M. oleifera* pod powder) was significantly higher over the *sev* containing 10% *M. oleifera* pod powder and control.

Similar trend was also observed in noodles. Results of present study are due to higher potassium content of raw *M. oleifera* pods (on DM basis).

4.7.12 HCl-extractability of minerals

4.7.12.1 Calcium

Calcium extractability of different types of *sev* and noodles varied significantly (P<0.05) (Table 4.40). Maximum calcium extractability was observed in control *sev*. However, it was maximum in noodles supplemented with 20 percent *M. oleifera* pod powder. Increased extractability of calcium might be due to effect of blanching as compared to frying. As blanching reduced the level of antinutrients and increased extractability of calcium. Similar findings have been resulted by Saharan (1994).

4.7.12.2 Phosphorus

A narrow range was observed in phosphorus extractability of *sev* and noodles. (Table 4.40). Phosphorus extractability was significantly (P<0.05) higher in control *sev* over the supplemented *sev*.

Noodles supplemented with 20 percent *M. oleifera* pod powder had significantly (P<0.05) higher phosphorus extractability than noodles supplemented with 10% and control.

4.7.12.3 Iron

HCl-extractability of iron was significantly (P<0.05) higher in control *sev* followed by 20 percent and 10 percent supplemented *M. oleifera* pod powder (Table 4.40). Iron extractability of different types of *sev* varied significantly (P<0.05) among themselves.

Non-significant difference was observed in noodles supplemented with 20 percent and 10 percent *M. oleifera* pod powder. Control noodles had significantly lower iron extractability as compared to supplemented noodles. It might be due to blanching process involved in preparation of noodles which improved iron extractability as compared to frying involved in preparation of *sev*.

Table 4.41 Total and HCl-extractable zinc and potassium contents (mg/100 g) of products prepared from *Moringa oleifera* pods (on dry weight basis)

Food products	Zinc			Potassium		
	Total	HCl-extractable	Percent extractability	Total	HCl-extractable	Percent extractability
Sev						
Bengal gram flour (control 100%)	2.14 ± 0.01	1.45 ± 0.01	69.42 ± 0.02	650.31 ± 0.02	457.41 ± 0.01	70.31 ± 0.03
Bengal gram flour + *Moringa oleifera* pods (10%)	3.86 ± 0.02	2.70 ± 0.05	69.32 ± 0.04	752.26 ± 0.87	531.62 ± 0.05	70.45 ± 0.07
Bengal gram flour + *Moringa oleifera* pods (20%)	4.88 ± 0.02	3.41 ± 0.02	69.47 ± 0.03	840.00 ± 0.03	595.91 ± 0.05	70.92 ± 0.02
CD (P<0.05)	0.04	0.03	0.04	1.75	0.14	0.16
Noodles						
Refined flour (control 100%)	2.72 ± 0.02	1.85 ± 0.02	68.56 ± 0.03	125.41 ± 0.04	89.54 ± 0.03	71.30 ± 0.04
Refined flour + *Moringa oleifera* pods (10%)	3.63 ± 0.02	2.42 ± 0.03	68.82 ± 0.02	241.25 ± 0.35	172.80 ± 0.03	71.38 ± 0.04
Refined flour + *Moringa oleifera* pods (20%)	4.10 ± 0.01	2.85 ± 0.01	68.87 ± 0.01	299.08 ± 0.29	214.13 ± 0.02	71.50 ± 0.03
CD (P<0.05)	0.05	0.06	0.03	0.92	0.08	0.12

Values are mean ± SE of three independent determinations

4.7.12.4 Zinc

Zinc extractability was significantly (P<0.05) higher in *sev* and noodles supplemented with 20 percent *M. oleifera* pod powder followed by noodles and *sev* supplemented with 10 percent *M. oleifera* pod and control.

Significant differences in zinc extractability were observed when different types of *sev* and noodles were compared among themselves.

4.7.12.5 Potassium

Potassium extractability was significantly (P<0.05) higher in *sev* and noodles supplemented with 20 percent *M. oleifera* pod powder over *sev* and noodles supplementing 10% and control (Table 4.41).

4.7.13 *In vitro* availability of calcium and iron

Calcium and iron availability of control *sev* was significantly higher as compared to *sev* supplemented with 10 percent and 20 percent *M. oleifera* pod powder (Table 4.42).

On the contrary, calcium and iron availability in noodles prepared by incorporating 20 percent *M. oleifera* pod powder were significantly (P<0.05) higher (25.56, 13.65%) as compared to those of noodles supplemented with 10 percent and control.

The combination of cooking methods used in the development of noodles supplemented with 20 percent *M. oleifera* pod powder showed significant improvement in calcium and iron availability. Reduction in the level of antinutrients as oxalic acid, phytic acid due to cumulative effect of various processing treatments including blanching, frying, steam cooking may also be responsible for increased bioavailability of minerals as these are known to hinder the availability of minerals. Similar findings have been reported by Saharan (1994).

Table 4.42 ***In vitro*** **availability (%) of calcium and iron of products prepared from *Moringa oleifera* pods (on dry weight basis)**

Food products	Calcium		Iron	
	Total (mg/100 g)	Percent availability (*in vitro*)	Total (mg/100 g)	Percent availability (*in vitro*)
Sev				
Bengal gram flour (control 100%)	78.23 ± 0.01	21.50 ± 0.01	3.86 ± 0.01	14.39 ± 0.01
Bengal gram flour + *Moringa oleifera* pods (10%)	87.35 ± 0.03	20.43 ± 0.01	5.86 ± 0.05	13.25 ± 0.03
Bengal gram flour + *Moringa oleifera* pods (20%)	99.51 ± 0.01	20.40 ± 0.01	7.96 ± 0.05	13.21 ± 0.01
CD (P<0.05)	0.03	0.02	0.02	0.01
Noodles				
Refined flour (control 100%)	50.12 ± 0.01	24.36 ± 0.03	5.42 ± 0.01	13.43 ± 0.02
Refined flour + *Moringa oleifera* pods (10%)	62.74 ± 0.01	25.51 ± 0.01	7.47 ± 0.01	13.60 ± 0.01
Refined flour + *Moringa oleifera* pods (20%)	73.57 ± 0.02	25.56 ± 0.01	9.54 ± 0.01	13.65 ± 0.01
CD (P<0.05)	0.02	0.04	0.02	0.04

Values are mean ± SE of three independent determinations

4.8 Shelf life of stored products

4.8.1 Organoleptic evaluation of stored *sev* and *M. oleifera* pod pickle

4.8.1.1 *Sev*

Results of the sensory scores of different types of *sev* prepared from *M. oleifera* leaves and pods are depicted in Tables 4.43 to 4.45.

Colour

Colour of *sev* (control) prepared from 100% bengal gram flour was in the category of 'liked moderately' on 0 day of storage. Colour of *sev* stored up to 8th week was also acceptable as it was in the category of 'liked slightly'. Significant decrease in colour of control *sev* was observed at the end of 8th week of storage when compared with fresh one.

Colour of *sev* (Type I) prepared from *M. oleifera* leaf powder (10%) was 'liked moderately' on zero day. At the end of 8th week of storage it was still acceptable and 'liked slightly'. Similar trend was observed in the *sev* (Type II) prepared from *M. oleifera* leaf powder (20%).

Colour of *sev* (Type III) prepared from *M. oleifera* pod powder (10%) was 'liked moderately' on zero day of storage and 'liked slightly' at the end of 8th week of storage.

Colour of *sev* (Type IV) stored up to 8th week of storage was in the category of 'neither liked nor disliked'. On mean basis, it was observed that all types of *sev* containing 10 percent and 20 percent *M. oleifera* leaf powder and pod powder were acceptable and fell in the category of 'liked slightly'. However, a significant difference was observed among *sev* prepared from 10 percent and 20 percent *M. oleifera* leaf as well as pod powder.

Table 4.43 **Changes in organoleptic scores (colour and appearance) of *sev* during storage**

Type of *sev*	Storage period (weeks)									Mean
	0	1	2	3	4	5	6	7	8	
Colour										
Control	7.80 ± 0.13	7.80 ± 0.13	7.40 ± 0.16	7.50 ± 0.16	7.40 ± 0.16	7.00 ± 0.00	7.00 ± 0.00	7.00 ± 0.00	6.70 ± 0.15	7.29
I	7.30 ± 0.15	7.10 ± 0.23	7.30 ± 0.15	6.90 ± 0.10	6.80 ± 0.20	6.60 ± 0.16	6.80 ± 0.13	6.70 ± 0.15	6.00 ± 0.00	6.83
II	7.20 ± 0.20	6.50 ± 0.22	6.40 ± 0.16	6.50 ± 0.16	6.50 ± 0.22	6.40 ± 0.16	6.20 ± 0.13	6.00 ± 0.00	6.00 ± 0.00	6.41
III	7.30 ± 0.15	7.40 ± 0.16	7.30 ± 0.15	6.80 ± 0.13	7.10 ± 0.10	6.60 ± 0.22	6.80 ± 0.13	6.50 ± 0.16	6.30 ± 0.15	6.90
IV	7.00 ± 0.14	6.90 ± 0.27	6.50 ± 0.17	6.50 ± 0.16	6.90 ± 0.17	6.30 ± 0.15	6.30 ± 0.15	5.80 ± 0.13	5.40 ± 0.16	6.40
Mean	7.32	7.14	6.98	6.94	6.84	6.58	6.52	6.40	6.08	
CD (P<0.05)	For storage = 0.15			For treatment = 0.19			For interaction = 0.43			
Appearance										
Control	7.60 ± 0.16	7.60 ± 0.16	7.40 ± 0.16	7.50 ± 0.16	7.30 ± 0.15	7.00 ± 0.00	7.00 ± 0.00	6.90 ± 0.10	6.60 ± 0.16	7.21
I	7.10 ± 0.10	7.50 ± 0.16	7.20 ± 0.13	5.90 ± 0.10	6.70 ± 0.21	6.70 ± 0.15	6.80 ± 0.13	6.70 ± 0.15	6.00 ± 0.00	6.84
II	6.90 ± 0.10	6.80 ± 0.24	6.50 ± 0.17	6.50 ± 0.16	6.30 ± 0.21	6.40 ± 0.16	6.50 ± 0.16	6.00 ± 0.00	6.00 ± 0.00	6.43
III	7.30 ± 0.15	7.40 ± 0.16	7.30 ± 0.15	6.80 ± 0.13	7.10 ± 0.10	6.70 ± 0.21	6.80 ± 0.13	6.50 ± 0.16	6.30 ± 0.15	6.91
IV	6.90 ± 0.10	6.80 ± 0.29	6.50 ± 0.17	6.50 ± 0.16	7.00 ± 0.14	6.30 ± 0.15	6.30 ± 0.15	5.80 ± 0.13	5.40 ± 0.16	6.38
Mean	7.16	7.22	6.98	6.84	6.88	6.62	6.60	6.38	6.06	
CD (P<0.05)	For storage = 0.14			For treatment = 0.19			For interaction = 0.43			

Values are mean ± SE of scores given by ten panelists on 9-point hedonic scale

I = Bengal gram flour + *Moringa oleifera* leaves (10%) II = Bengal gram flour + *Moringa oleifera* leaves (20%)

III = Bengal gram flour + *Moringa oleifera* pods (10%) IV = Bengal gram flour + *Moringa oleifera* pods (20%)

Appearance

Appearance of Type I, II, III and IV *sev* were 'liked moderately' on zero day of storage. Appearance of all types of *sev* at the end of 8th week of storage was also acceptable and fell in category 'liked slightly' except Type IV *sev* which was 'neither liked nor disliked'. Non-significant change in appearance of all types of *sev* prepared was observed up to 4th week of storage and after that its score for appearance declined significantly ($P<0.05$) during 8th week.

Overall, means for appearance of Type I and Type II differed significantly ($P<0.05$). Similarly, overall acceptability score for Type III and Type IV *sev* differed significantly ($P<0.05$). However, *sev* prepared from 10 percent *M. oleifera* leaves as well as pod powder differed non-significantly.

Aroma

A significant decrease in aroma of all types of *sev* was observed when storage period was prolonged from 0 to 8th week. During 8th week of storage (control, Type I and Type III *sev*) was in the category of 'liked slightly' and Type II and IV *sev* were 'neither liked nor disliked'. On mean basis, a significant difference was observed in aroma of Type I and Type II *sev*. Similar trend was also observed in Type III and IV *sev* i.e. *sev* prepared from *M. oleifera* pod powder at two different levels i.e. 10 percent and 20 percent.

Texture

Overall texture of *sev* was acceptable up to 8th week of storage. However, it decreased significantly with the prolongation of storage period. Texture of all types i.e. Type I, II, III and IV of *sev* differed significantly ($P<0.05$) over the control, however, non-significant difference was observed among Type I and III. Similarly Type II and Type IV *sev* had almost similar texture.

Table 4.44 **Changes in organoleptic scores (aroma and texture) of *sev* during storage**

Type of *sev*	Storage period (weeks)									Mean
	0	1	2	3	4	5	6	7	8	
Aroma										
Control	7.80 ±0.13	7.60 ±0.16	7.40 ±0.16	7.40 ±0.16	7.30 ±0.15	6.70 ±0.15	6.70 ±0.15	6.50 ±0.16	6.30 ±0.15	7.07
I	7.00 ±0.00	7.40 ±0.16	7.20 ±0.13	6.90 ±0.10	6.90 ±0.17	6.70 ±0.15	6.60 ±0.16	6.40 ±0.16	6.30 ±0.21	6.82
II	6.90 ±0.10	6.60 ±0.22	7.20 ±0.13	6.70 ±0.15	6.50 ±0.22	6.20 ±0.13	6.10 ±0.10	5.50 ±0.16	5.50 ±0.16	6.24
III	7.30 ±0.15	7.30 ±0.21	7.20 ±0.20	6.80 ±0.13	6.80 ±0.20	6.90 ±0.17	6.50 ±0.16	6.50 ±0.16	6.00 ±0.21	6.81
IV	6.90 ±0.10	7.00 ±0.25	6.80 ±0.13	6.50 ±0.16	6.70 ±0.21	6.00 ±0.00	6.00 ±0.00	5.70 ±0.15	5.30 ±0.15	6.32
Mean	7.18	7.18	6.96	6.86	6.84	6.50	6.38	6.12	5.88	
CD (P<0.05)	For storage = 0.15			For treatment = 0.20			For interaction = 0.45			
Texture										
Control	7.60 ±0.16	7.60 ±0.16	7.40 ±0.16	7.40 ±0.16	7.30 ±0.15	7.00 ±0.00	7.00 ±0.00	7.00 ±0.00	7.00 ±0.00	7.25
I	7.30 ±0.15	7.30 ±0.15	7.10 ±0.10	7.00 ±0.00	7.20 ±0.13	7.00 ±0.00	6.90 ±0.10	6.80 ±0.13	6.40 ±0.16	7.00
II	6.90 ±0.10	6.90 ±0.17	6.60 ±0.16	6.90 ±0.10	7.00 ±0.25	6.70 ±0.15	6.20 ±0.13	6.00 ±0.00	6.00 ±0.16	6.57
III	7.20 ±0.13	7.30 ±0.21	7.30 ±0.15	6.90 ±0.10	7.10 ±0.10	7.00 ±0.14	6.90 ±0.00	6.90 ±0.10	6.40 ±0.16	7.00
IV	7.00 ±0.00	6.90 ±0.17	6.90 ±0.10	6.80 ±0.13	6.90 ±0.17	6.40 ±0.16	6.40 ±0.10	5.80 ±0.24	5.10 ±0.01	6.46
Mean	7.20	7.20	7.06	7.00	7.10	6.82	6.68	6.50	6.18	
CD (P<0.05)	For storage = 0.13			For treatment = 0.17			For interaction = 0.38			

Values are mean ± SE of scores given by ten panelists on 9-point hedonic scale
I = Bengal gram flour + *Moringa oleifera* leaves (10%) II = Bengal gram flour + *Moringa oleifera* leaves (20%)
III = Bengal gram flour + *Moringa oleifera* pods (10%) IV = Bengal gram flour + *Moringa oleifera* pods (20%)

Taste

All types of *sev* were acceptable in terms of taste and fell in the category of 'liked slightly' except control which was 'liked moderately'. Period of storage had significant effect on taste score during 8th week of storage as compared to that of fresh one.

A non-significant difference was observed in taste scores of Type II and Type IV *sev*. However, both *sev* differed significantly ($P<0.05$) from other types of *sev* i.e. control, Type I and Type III.

Overall acceptability

Overall acceptability which was the mean of scores for colour, appearance, aroma, texture and taste of control, Type I and III was in 'liked moderately' category whereas Type II and IV was in 'liked slightly' category on 0 day of storage. Period of storage had significant ($P<0.05$) effect on overall acceptability and scores for overall acceptability decreased significantly during 8th week of storage over the fresh one. Overall comparison of the mean acceptability scores of all types of *sev* revealed a significant difference between Type I and Type II *sev*.

Similar trend was observed in Type III and Type IV *sev*. This showed that higher percent of *M. oleifera* leaf powder or pod powder affected the overall acceptability of *sev*.

4.8.1.2 Organoleptic evaluation of stored *M. oleifera* pod pickle

Mean scores for different organoleptic characteristics of *M. oleifera* pod pickle were in the category of 'liked moderately'. Mean scores for colour, appearance, aroma and texture of pickle stored for varying periods did not change significantly ($P<0.05$) as the storage period was increased. Improvement was observed in the taste and overall acceptability of the pickle as the storage period was prolonged (Table 4.46). however, the difference was non-significant.

Table 4.45 **Changes in organoleptic scores (taste and overall acceptability) of *sev* during storage**

Type of *sev*	Storage period (weeks)									Mean
	0	1	2	3	4	5	6	7	8	
Taste										
Control	7.60 ±0.16	7.60 ±0.16	7.40 ±0.16	7.60 ±0.16	7.30 ±0.15	6.90 ±0.10	6.90 ±0.10	6.80 ±0.13	7.00 ±0.00	7.21
I	6.90 ±0.10	7.40 ±0.16	6.80 ±0.13	6.80 ±0.13	7.00 ±0.14	6.50 ±0.16	6.70 ±0.15	6.60 ±0.16	5.90 ±0.10	6.73
II	6.60 ±0.16	6.70 ±0.21	6.40 ±0.16	6.20 ±0.13	6.40 ±0.22	6.40 ±0.22	6.00 ±0.14	5.60 ±0.16	5.90 ±0.10	6.24
III	7.10 ±0.18	7.20 ±0.24	7.10 ±0.23	6.90 ±0.10	6.90 ±0.17	7.00 ±0.00	6.80 ±0.13	6.70 ±0.15	6.30 ±0.15	6.88
IV	6.90 ±0.10	6.50 ±0.34	6.20 ±0.24	6.50 ±0.16	6.30 ±0.15	6.30 ±0.21	6.30 ±0.21	5.50 ±0.16	5.10 ±0.10	6.17
Mean	7.02	7.08	6.78	6.76	6.78	6.62	6.54	6.24	6.04	
CD (P<0.05)	For storage = 0.15			For treatment = 0.20			For interaction = 0.46			
Overall acceptability										
Control	7.68 ±0.13	7.64 ±0.14	7.40 ±0.16	7.40 ±0.16	7.32 ±0.14	6.92 ±0.04	6.72 ±0.08	6.84 ±0.04	6.92 ±0.04	7.20
I	7.14 ±0.09	7.34 ±0.14	7.16 ±0.11	6.90 ±0.08	6.90 ±0.13	6.70 ±0.08	6.12 ±0.08	6.64 ±0.11	6.76 ±0.08	6.85
II	6.88 ±0.10	6.70 ±0.18	6.40 ±0.10	6.56 ±0.11	6.54 ±0.20	6.42 ±0.12	5.76 ±0.06	5.82 ±0.05	6.20 ±0.10	6.36
III	7.24 ±0.14	7.32 ±0.18	7.26 ±0.16	6.84 ±0.11	7.00 ±0.11	6.84 ±0.13	6.26 ±0.14	6.62 ±0.10	6.76 ±0.10	6.90
IV	6.94 ±0.06	6.84 ±0.21	6.58 ±0.10	6.56 ±0.11	6.76 ±0.13	6.26 ±0.11	5.26 ±0.11	5.72 ±0.14	6.26 ±0.12	6.35
Mean	7.17	7.16	6.96	6.90	6.85	6.62	6.58	6.32	6.02	
CD (P<0.05)	For storage = 0.12			For treatment = 0.16			For interaction = 0.35			

Values are mean ± SE of scores given by ten panelists on 9-point hedonic scale

I = Bengal gram flour + *Moringa oleifera* leaves (10%) II = Bengal gram flour + *Moringa oleifera* leaves (20%)

III = Bengal gram flour + *Moringa oleifera* pods (10%) IV = Bengal gram flour + *Moringa oleifera* pods (20%)

Table 4.46 Changes in organoleptic scores of *Moringa oleifera* pod pickle during storage

Storage period (months)	Colour	Appearance	Aroma	Texture	Taste	Overall acceptability
0	8.30 ± 0.15	8.10 ± 0.23	7.70 ± 0.21	7.90 ± 0.23	7.80 ± 0.29	7.78 ± 0.12
1	8.00 ± 0.01	8.00 ± 0.15	7.70 ± 0.15	7.80 ± 0.12	7.80 ± 0.20	7.80 ± 0.09
2	7.90 ± 0.10	8.00 ± 0.10	8.10 ± 0.10	7.80 ± 0.13	7.80 ± 0.23	7.80 ± 0.09
3	7.90 ± 0.10	8.00 ± 0.13	8.00 ± 0.01	7.80 ± 0.13	7.80 ± 0.13	7.82 ± 0.09
4	8.00 ± 0.01	8.00 ± 0.15	7.70 ± 0.15	8.00 ± 0.01	7.80 ± 0.15	7.84 ± 0.05
5	8.00 ± 0.10	8.00 ± 0.16	7.70 ± 0.15	7.70 ± 0.15	7.90 ± 0.16	7.90 ± 0.10
CD (P<0.05)	0.27	0.46	0.41	0.42	0.61	0.27

Values are mean ± SE of scores given by ten panelists on 9-point hedonic scale

4.8.2 Effect of storage on fat acidity and free fatty acid content of *sev* and *M. oleifera* pod pickle

4.8.2.1 *Sev*

4.8.2.1.1 Fat acidity

Table 4.47 presents the fat acidity content of *sev* prepared from 10 percent and 20 percent of *M. oleifera* leaf powder as well as 10 and 20 percent of *M. oleifera* pod powder.

Fat acidity of Type I and Type II *sev* was 39.27 and 40.10 mg KOH/100 g, respectively on zero day of storage period (Fig. 6). Similarly, fat acidity of Type III and Type IV was 39.52 and 40.32 mg KOH/100 g, respectively. With advancement of storage period, fat acidity increased significantly.

Increase in fat acidity with advancement of storage period might be due to breakdown of triglycerides and oxidation of unsaturated fatty acids.

Dhaka (2001) also reported increase in fat acidity from 82.69 on zero day to 91.35 mg KOH/100 g on 30th day in besan *sev* prepared with 20 percent chickpea coat.

4.8.2.1.2 Free fatty acids

Free fatty acid content of Type I and II *sev* was 257.47 and 258.67 mg/100 g on zero day which increased to 450.16 and 452.17 mg/100 g, respectively during 8th week of storage (Table 4.48, Fig. 7)). Significant ($P<0.05$) difference was observed at each storage interval.

On comparing all treatments, it was found that free fatty acid content significantly varied among different types of *sev*. Free fatty acid content of *sev* increased with increase in storage period. Increase in free fatty acid with advancement of storage period might be due to breakdown of triglycerides and oxidation of unsaturated fatty acids.

Table 4.47 **Changes in fat acidity (mg KOH/100 g sample) content of *sev* during storage (on dry weight basis)**

Type of *Sev*	Storage period (weeks)									Mean
	0	1	2	3	4	5	6	7	8	
Fat acidity										
Control	38.17 ± 0.02	45.60 ± 0.01	52.93 ± 0.02	57.62 ± 0.01	63.70 ± 0.01	69.48 ± 0.01	76.72 ± 0.02	88.28 ± 0.01	115.47 ± 0.02	67.55
I	39.27 ± 0.02	46.50 ± 0.01	53.67 ± 0.02	58.92 ± 0.01	64.36 ± 0.01	70.66 ± 0.01	79.83 ± 0.01	88.63 ± 0.01	116.32 ± 0.01	68.69
II	40.10 ± 0.01	46.92 ± 0.01	54.77 ± 0.02	59.01 ± 0.01	65.08 ± 0.01	71.22 ± 0.01	78.33 ± 0.01	87.48 ± 0.01	115.32 ± 0.02	68.70
III	39.52 ± 0.01	46.70 ± 0.01	54.87 ± 0.01	60.71 ± 0.01	64.16 ± 0.01	71.67 ± 0.01	78.81 ± 0.01	90.49 ± 0.01	117.32 ± 0.02	69.36
IV	40.32 ± 0.01	47.51 ± 0.01	55.36 ± 0.01	62.87 ± 0.01	66.72 ± 0.01	72.52 ± 0.02	79.48 ± 0.01	92.31 ± 0.02	120.52 ± 0.02	70.84
Mean	39.48	46.65	54.32	59.83	64.80	71.12	78.64	89.43	116.99	
CD (P<0.05)	For storage = 0.01			For treatment = 0.02			For interaction = 0.04			

Values are mean ± SE of three independent determinations
I = Bengal gram flour + *Moringa oleifera* leaves (10%)
II = Bengal gram flour + *Moringa oleifera* leaves (20%)
III = Bengal gram flour + *Moringa oleifera* pods (10%)
IV = Bengal gram flour + *Moringa oleifera* pods (20%)

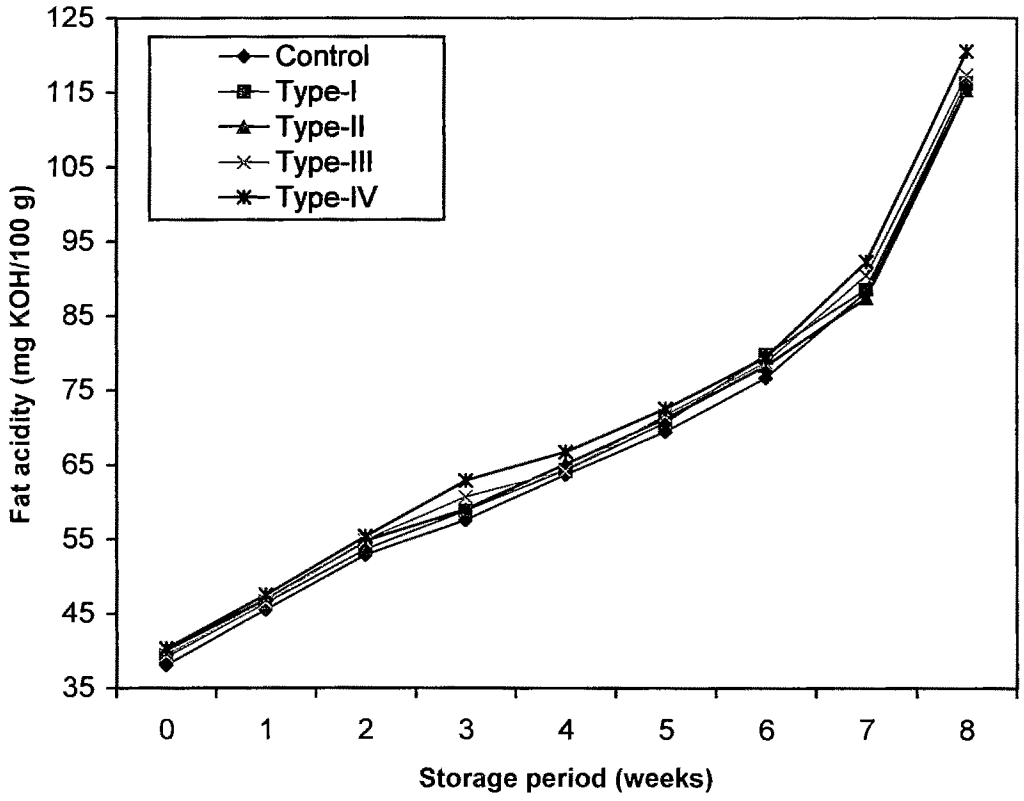


Fig. 6 : Changes in fat acidity content of *sev* during storage

Table 4.48 **Changes in free fatty acids (mg/100 g fat, as oleic acid) content of *sev* during storage (on dry weight basis)**

Type of *Sev*	Storage period (weeks)									Mean
	0	1	2	3	4	5	6	7	8	
Free fatty acids										
Control	254.66 ± 0.02	281.35 ± 0.03	305.39 ± 0.02	328.48 ± 0.01	352.94 ± 0.02	375.63 ± 0.02	398.37 ± 0.02	421.48 ± 0.01	450.16 ± 0.01	352.05
I	257.47 ± 0.02	283.64 ± 0.03	307.47 ± 0.01	330.65 ± 0.01	354.83 ± 0.02	379.48 ± 0.02	400.12 ± 0.01	425.28 ± 0.01	452.17 ± 0.01	354.57
II	258.67 ± 0.02	284.56 ± 0.04	308.28 ± 0.02	330.93 ± 0.02	354.96 ± 0.02	380.65 ± 0.01	403.17 ± 0.01	427.49 ± 0.01	453.12 ± 0.01	355.76
III	256.47 ± 0.02	283.47 ± 0.01	304.28 ± 0.02	330.64 ± 0.02	355.35 ± 0.03	377.45 ± 0.03	400.50 ± 0.03	425.27 ± 0.01	453.74 ± 0.02	354.13
IV	257.38 ± 0.02	284.35 ± 0.03	305.48 ± 0.02	332.47 ± 0.01	357.42 ± 0.01	380.35 ± 0.02	402.76 ± 0.01	427.50 ± 0.07	455.43 ± 0.04	355.90
Mean	256.93	283.47	306.18	330.64	355.64	378.71	400.98	425.41	452.93	
CD (P<0.05)	For storage = 0.02			For treatment = 0.03			For interaction = 0.06			

Values are mean ± SE of three independent determinations
I = Bengal gram flour + *Moringa oleifera* leaves (10%)
II = Bengal gram flour + *Moringa oleifera* leaves (20%)
III = Bengal gram flour + *Moringa oleifera* pods (10%)
IV = Bengal gram flour + *Moringa oleifera* pods (20%)

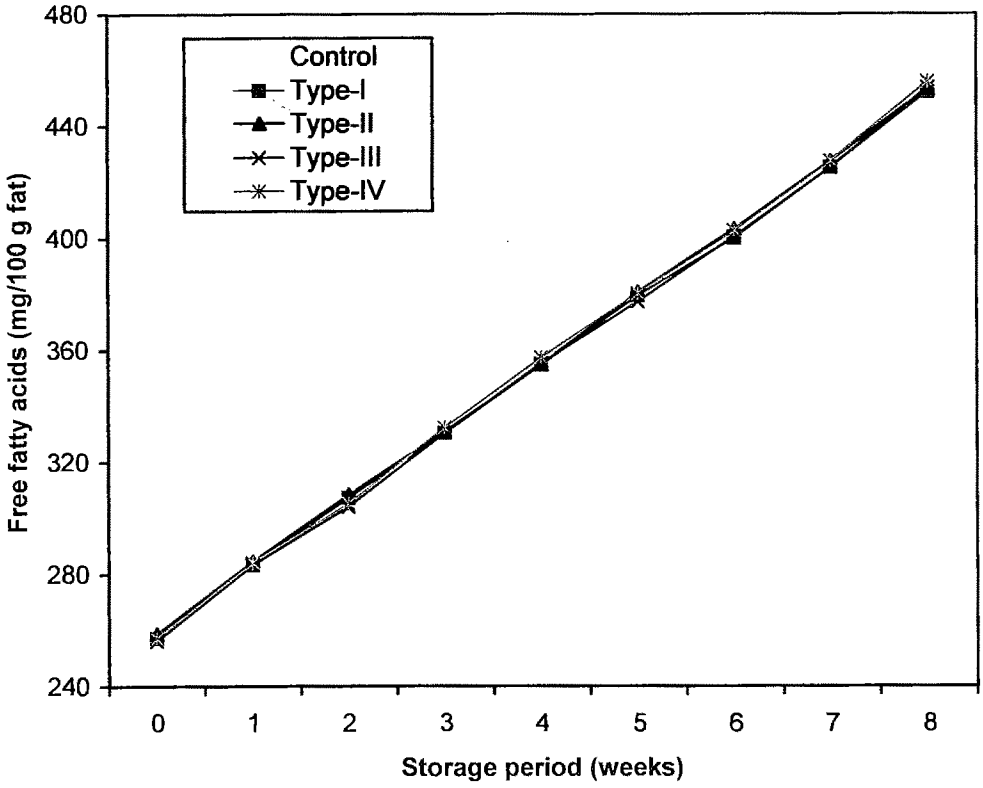


Fig. 7 : Changes in free fatty acids content of *sev* during storage

4.8.2.2 *M. oleifera* pod pickle

4.8.2.2.1 Fat acidity

Table 4.49 and Fig. 8 present the fat acidity and free fatty acid content of pickle. Fat acidity of pickle i.e. 41.02 mg KOH/100 g on zero day of storage was increased to 120.49 mg KOH/100 g during 5th month of storage. There was significant ($P<0.05$) increase in fat acidity content of pickle as the storage period advanced. It might be due to breakdown of triglycerides and oxidation of unsaturated fatty acids.

4.8.2.2.2 Free fatty acids

Free fatty acid content of pickle on 0th, 1st, 2nd, 3rd, 4th and 5th month of storage was 203.33, 245.55, 287.62, 310.27, 345.48 and 381.61 mg KOH/100 g, respectively (Fig. 8). Data revealed that there was a significant ($P<0.05$) increase in free fatty acid content of pickle with increase in storage period. It might be due to breakdown of triglycerides and oxidation of unsaturated fatty acids.

4.8.3 Nutritional changes during storage of *sev* and *M. oleifera* pod pickle

4.8.3.1 *Sev*

4.8.3.1.1 Ascorbic acid

The ascorbic acid could not be detected in different types of *sev* i.e. *sev* prepared from 10 percent and 20 percent *M. oleifera* leaf powder and *sev* prepared from 10 percent and 20 percent *M. oleifera* pod powder (Table 4.50).

4.8.3.1.2 Iron

The iron content of Type I and Type II *sev* was 5.81 and 6.84 mg, respectively on 0 day of storage which decreased slightly when subjected to extended storage for varying period (Fig. 9). Similar trend was also observed in Type III and Type IV *sev* prepared from 10 percent and 20 percent *M. oleifera* pod powder. Overall

Table 4.49 Changes in fat acidity (mg KOH/100 g sample) and free fatty acids (mg/100 g fat, as oleic acid) content of pickle during storage (on dry weight basis)

Storage period (months)	Fat acidity	Free fatty acids
0	41.02 ± 0.01	203.33 ± 0.02
1	55.27 ± 0.01	245.55 ± 0.03
2	74.36 ± 0.03	287.62 ± 0.02
3	88.47 ± 0.01	310.27 ± 0.02
4	104.64 ± 0.02	345.48 ± 0.01
5	120.49 ± 0.03	381.61 ± 0.01
CD (P<0.05)	0.04	0.05

Values are mean ± SE of three independent determinations

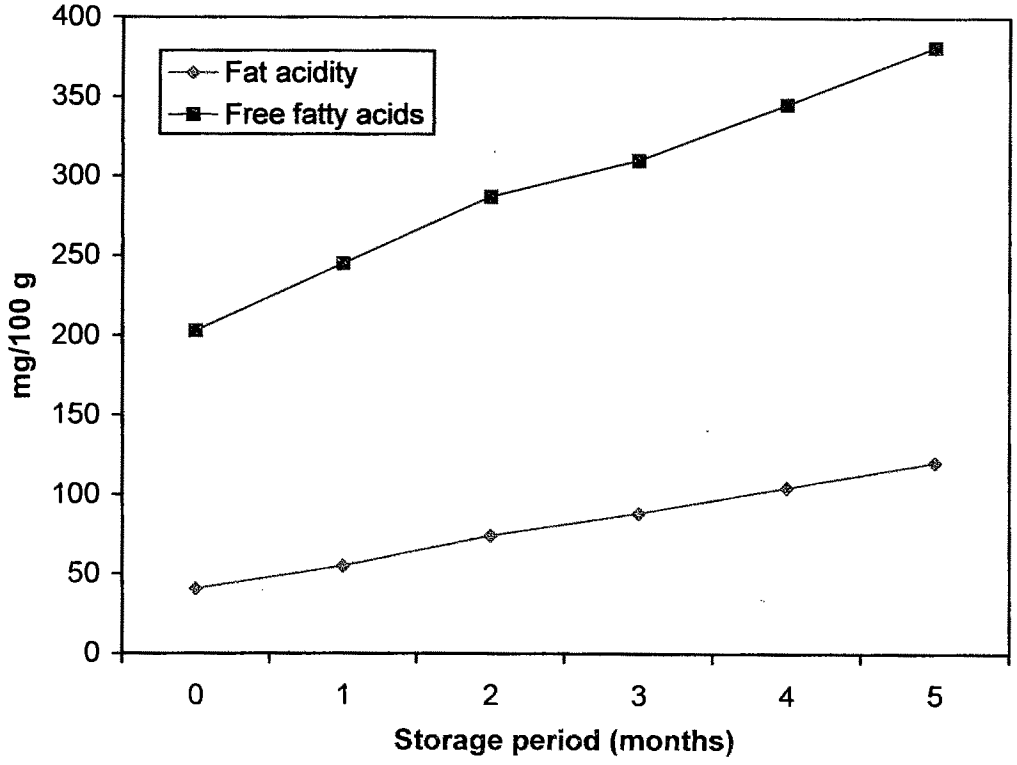


Fig. 8 : Changes in fat acidity and free fatty acids content of pickle during storage

Table 4.50 Effect of storage on ascorbic acid, iron and oxalic acid content of *sev* (on dry weight basis)

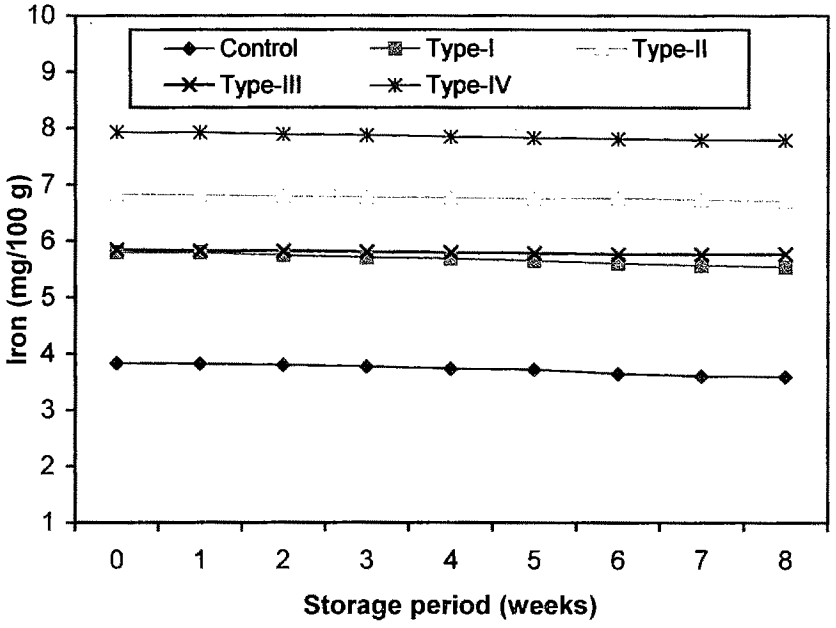
Type of *sev*	Storage period (weeks)									Mean
	0	1	2	3	4	5	6	7	8	
Ascorbic acid										
Control										
I										
II					ND					
III										
IV										
Iron										
Control	3.84 ± 0.01	3.83 ± 0.02	3.81 ± 0.02	3.78 ± 0.01	3.74 ± 0.01	3.72 ± 0.01	3.65 ± 0.01	3.61 ± 0.01	3.60 ± 0.02	3.71
I	5.81 ± 0.01	5.80 ± 0.06	5.75 ± 0.03	5.71 ± 0.01	5.69 ± 0.02	5.65 ± 0.02	5.60 ± 0.01	5.57 ± 0.03	5.54 ± 0.03	5.66
II	6.84 ± 0.01	6.82 ± 0.01	6.80 ± 0.01	6.78 ± 0.01	6.77 ± 0.01	6.75 ± 0.03	6.76 ± 0.02	6.73 ± 0.05	6.71 ± 0.03	6.76
III	5.84 ± 0.02	5.82 ± 0.01	5.82 ± 0.01	5.80 ± 0.01	5.79 ± 0.01	5.78 ± 0.03	5.76 ± 0.03	5.76 ± 0.02	5.77 ± 0.05	5.79
IV	7.94 ± 0.02	7.93 ± 0.01	7.90 ± 0.01	7.88 ± 0.01	7.85 ± 0.01	7.83 ± 0.03	7.81 ± 0.02	7.79 ± 0.03	7.79 ± 0.02	7.84
Mean	6.05	6.04	6.01	5.99	5.96	5.94	5.91	5.89	5.88	
CD (P<0.05)	For storage = 0.01			For treatment = 0.01			For interaction = 0.02			

Contd.

	Storage period (weeks)									
	0	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	
Oxalic acid										
Control	13.55 ± 0.01	13.53 ± 0.02	13.51 ± 0.00	13.47 ± 0.01	13.40 ± 0.01	13.37 ± 0.01	13.30 ± 0.01	13.25 ± 0.01	13.20 ± 0.03	13.38
I	53.57 ± 0.01	53.52 ± 0.03	53.47 ± 0.02	53.42 ± 0.02	53.34 ± 0.04	53.26 ± 0.01	53.19 ± 0.01	53.14 ± 0.01	53.09 ± 0.01	53.30
II	65.77 ± 0.03	65.73 ± 0.02	65.68 ± 0.05	65.29 ± 0.32	65.56 ± 0.01	65.51 ± 0.01	65.47 ± 0.02	65.38 ± 0.02	65.27 ± 0.02	65.58
III	54.63 ± 0.02	54.58 ± 0.01	54.85 ± 0.32	54.46 ± 0.02	54.39 ± 0.01	54.36 ± 0.03	54.28 ± 0.02	54.22 ± 0.02	54.12 ± 0.01	54.39
IV	95.53 ± 0.02	95.46 ± 0.02	95.37 ± 0.02	95.30 ± 0.01	95.23 ± 0.01	95.20 ± 0.01	95.15 ± 0.01	95.10 ± 0.02	94.93 ± 0.04	95.22
Mean	56.61	56.56	56.57	56.54	56.38	56.34	56.28	56.22	56.12	
CD (P<0.05)	For storage = 0.06			For treatment = 0.08			For interaction = 0.19			

Values are mean ± SE of three independent determinations
I = Bengal gram flour + *Moringa oleifera* leaves (10%)
II = Bengal gram flour + *Moringa oleifera* leaves (20%)
III = Bengal gram flour + *Moringa oleifera* pods (10%)
IV = Bengal gram flour + *Moringa oleifera* pods (20%)
ND = Not detected

(a)



(b)

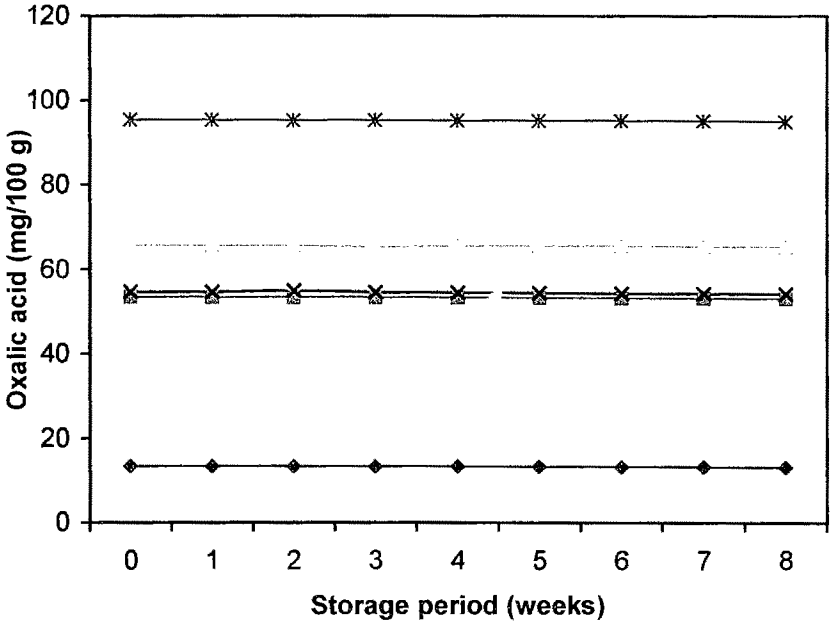


Fig. 9 : Changes in (a) iron (b) oxalic acid content of *sev* during storage

comparison of different types of *sev* showed significant differences ($P<0.05$) in iron content of *sev* during storage.

4.8.3.1.3 Oxalic acid

With an increase in storage period, the oxalic acid content of different types of *sev* decreased. Overall up to 3rd week of storage, there was non-significant difference observed in the oxalic acid of control and supplemented *sev* and after that it decreased significantly ($P<0.05$) as the storage period advanced (Fig. 9). Overall, different types of *sev* had significant variation in their oxalic acid content.

4.8.3.2 *Moringa oleifera* pod pickle

4.8.3.2.1 Ascorbic acid

A significant effect of storage upon ascorbic acid content of pod pickle could be noticed. The ascorbic acid content of fresh pod pickle was 89.33 mg/100 g on 0th day of storage (Table 4.51 and Fig. 10). After one month of storage, loss up to the extent of 15 percent in ascorbic acid over the fresh one occurred. This loss went on increasing as the period of storage prolonged. At the end of 5th month of storage, maximum loss in ascorbic acid was noticed.

Ascorbic acid content of fresh *Moringa oleifera* was higher when compared to *M. oleifera* pod pickle. The ascorbic acid is most sensitive to heat and oxidation, therefore, it might have been destroyed during various processing treatments involved in the preparation. Subsequent loss of ascorbic acid due to storage may be because of oxidation of ascorbic acid resulting in a decrease in its content in the stored products.

Similar losses due to processing and storage have been reported by several workers in guava products (Chadha, 1993),

Table 4.51 Effect of storage on ascorbic acid, iron and oxalic acid content of pickle (on dry weight basis)

Storage period (months)	Ascorbic acid (mg/100 g)	Iron (mg/100 g)	Oxalic acid (mg/100 g)
0	89.33 ± 0.02	18.50 ± 0.01	300.62 ± 0.01
1	75.63 ± 0.02	18.45 ± 0.01	300.42 ± 0.02
2	64.33 ± 0.02	18.42 ± 0.03	300.27 ± 0.02
3	56.62 ± 0.02	18.41 ± 0.01	299.24 ± 0.28
4	45.32 ± 0.02	18.36 ± 0.01	297.14 ± 0.39
5	33.16 ± 0.01	18.31 ± 0.01	295.56 ± 0.02
CD (P<0.05)	0.05	0.02	0.61

Values are mean ± SE of three independent determinations

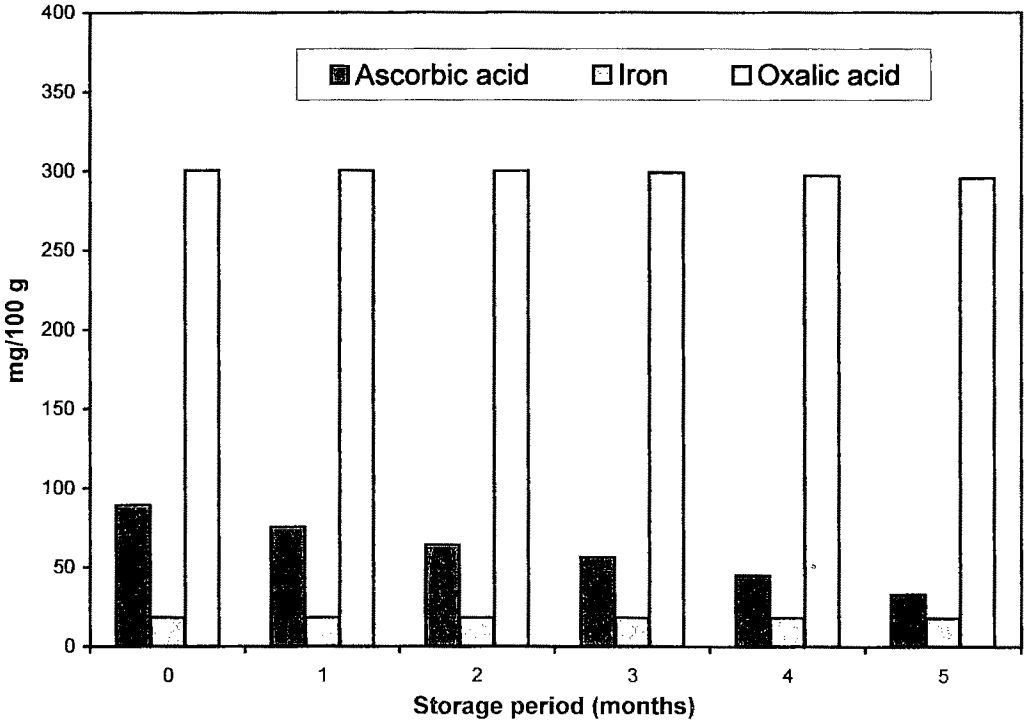


Fig. 10 : Changes in ascorbic acid, iron and oxalic acid contents of pickle during storage

mango oil pickle (Verma *et al.*, 1986), carrot pickle (Basnett, 1992) and karonda pickle (Panwar, 1996).

4.8.3.2.2 Iron

The iron content of *M. oleifera* pod pickle was 18.50 mg/100 g on 0 day of storage. After one month of storage, the iron content was 18.45 mg/100 g; it was different from the fresh one (Table 4.51, Fig. 10). At the end of 2nd month, iron content was slightly decreased, however, the difference was significant. At the end of 3rd month, iron content was found to be non-significantly different and after 5 month of storage, iron content declined to 18.31 mg/100 g which was significantly lower as compared to control.

4.8.3.2.3 Oxalic acid

Oxalic acid content of fresh pod pickle i.e. 300.62 mg/100 g decreased to 300.27 mg/100 g after 2 months of storage, however, the difference was non-significant. After 3rd month of storage, oxalic acid further decreased significantly ($P<0.05$) over the control i.e. fresh pickle. As the period of storage increased, oxalic acid content of pickle also decreased significantly. At the 5th month of storage, oxalic acid content was 295.56 mg/100 g. During storage of pickle, decrease in oxalic acid content might be due to binding of moisture by addition of salt and oil.

4.9 Biological utilization of diet containing *Moringa oleifera* leaves

4.9.1 Food consumption and weight gain

Effect of feeding *Moringa oleifera* leaves on food consumption and weight gain has been given in Table 4.52. The food consumption of control group I fed diet without cholesterol was significantly ($P<0.05$) higher than control group II fed diet containing cholesterol. Similarly, significant difference in the food

Table 4.52 Food consumption (g) and weight gain (g) of rats fed diet containing *Moringa oleifera* leaves

Dietary group	Food consumption	Weight gain
Control Group I (without cholesterol)	215.10 ± 0.23	48.14 ± 0.45
Control Group II (with cholesterol)	194.01 ± 0.72	58.25 ± 0.39
Experimental group I *Moringa oleifera* leaves (5%)	200.87 ± 0.55	56.81 ± 0.44
Experimental group II *Moringa oleifera* leaves (10%)	204.80 ± 0.31	57.67 ± 0.32
CD (P<0.05)	1.17	1.21

Values are mean ± SE of seven replicates

intake of experimental group I and II was witnessed when compared to control group II. On the other hand, the weight gain by rats in control group I was the lowest (48.14 g) and it was the highest (58.25 g) for rats fed on diet containing cholesterol (control group II). When 5 and 10 percent *Moringa oleifera* leaves were added, rats in experimental groups I and II consumed 200.87 and 204.80 g diet, respectively. The group of rats fed *M. oleifera* leaves at 5 and 10 percent gained 56.81 and 57.67 g body weight, respectively; the gain in weight of rats fed 10 percent *M. oleifera* leaves was almost similar to that of the group of rats fed 5 percent *M. oleifera* leaves. However, it was lower than the control group II (with cholesterol). Decrease in weight gain in experimental group I and II as over control group II might be due to increase in level of dietary fibre. The difference in food consumption in different groups might be because of satiety effect of diet as well as its palatability (Sosulski and Cadden, 1982).

It was observed from the Table 4.53 that when additional cholesterol was fed to rats, there was significant ($P<0.05$) increase in weight gain as compared to rats fed diet containing no additional amount of cholesterol. This might be due to accumulation of cholesterol in the body tissues of rats.

4.9.2 Feed efficiency ratio (FER) and protein efficiency ratio (PER)

FER and PER of control and experimental diet containing *Moringa oleifera* leaves has been given in Table 4.53. The feed efficiency ratio (FER) of experimental diets containing 5 and 10 percent *M. oleifera* leaves was 0.28 and 0.27, respectively; it was significantly lower for experimental diets than the control diet II (with cholesterol). PER value for control group II (with cholesterol) was significantly ($P<0.05$) higher than experimental group fed on

Table 4.53 Feed efficiency ratio (FER) and protein efficiency ratio (PER) of rats fed diet containing *Moringa oleifera* leaves

Dietary group	FER	PER
Control Group I (without cholesterol)	0.22 ± 0.01	2.25 ± 0.01
Control Group II (with cholesterol)	0.29 ± 0.01	2.92 ± 0.01
Experimental group I *Moringa oleifera* leaves (5%)	0.28 ± 0.01	2.79 ± 0.01
Experimental group II *Moringa oleifera* leaves (10%)	0.27 ± 0.01	2.77 ± 0.02
CD ($P<0.05$)	0.00	0.01

Values are mean ± SE of seven replicates

diets containing 5 and 10 percent *M. oleifera* leaves. It can be seen from data in Table 4.53 that PER of diet containing *M. oleifera* leaves was 2.79 and 2.77; these were significantly lower than that of rats fed on control diet (II) containing additional cholesterol but significantly higher than rats fed on control group I having no additional amount of cholesterol. Higher PER of cholesterol supplemented feeds might be because of deposition of cholesterol in various tissues which might have caused an increase in weight gain.

4.9.3 *Moringa oleifera* leaves and serum cholesterol

4.9.3.1 Total cholesterol

Serum total cholesterol level of rats fed on control diet and diets containing *M. oleifera* leaves at 5 and 10 percent levels has been given in Table 4.54 and Fig. 11. The serum cholesterol level of rats fed on control diet (I) containing no cholesterol was the lowest (82.19 mg/100 ml), whereas it was the highest (225.21 mg/100 ml) for the control group II fed on diet containing 1 percent additional cholesterol. This showed that feeding of additional 1 percent cholesterol in the diets of rats resulted in 63.5 percent higher serum total cholesterol concentration.

The serum total cholesterol level of experimental group was 213.10 and 198.19 mg/100 ml when the rats were fed on experimental diets containing 5 and 10 percent *M. oleifera* leaves, respectively. The group of rats fed on experimental diets had significantly lower serum cholesterol level as compared to control group (II) fed on diet containing cholesterol. The group of rats fed with higher percent (10%) of *M. oleifera* leaves had significantly lower levels of serum cholesterol as compared to group fed lower percent (5%) of *M. oleifera* leaves. The group of rats fed *M. oleifera* leaves at 5 percent level had 5.38 percent lower serum cholesterol

Table 4.54 Effect of feeding *Moringa oleifera* leaves on serum cholesterol (mg/100 ml) levels in rats

Dietary group	Total cholesterol	HDL cholesterol	LDL cholesterol	VLDL cholesterol	Total chol : HDL chol	Total chol : LDL chol
Control group I (without cholesterol)	82.19 ± 0.12 (63.50)	33.14 ± 0.18 (63.24)	19.98 ± 0.24 (76.21)	24.69 ± 0.23 (49.92)	2.48 ± 0.01	4.12 ± 0.05
Control group II (with cholesterol)	225.21 ± 0.21	90.17 ± 0.23	83.99 ± 0.14	49.31 ± 0.21	2.50 ± 0.01	2.68 ± 0.01
Experimental group I *Moringa oleifera* leaves (5%)	213.10 ± 0.15 (5.38)	89.07 ± 0.20 (1.21)	76.17 ± 0.21 (9.31)	46.68 ± 0.15 (5.33)	2.39 ± 0.01	2.79 ± 0.01
Experimental group II *Moringa oleifera* leaves (10%)	198.19 ± 0.20 (12.00)	87.45 ± 0.19 (3.01)	63.40 ± 0.18 (24.51)	42.10 ± 0.05 (14.62)	2.26 ± 0.01	3.12 ± 0.01
CD (P<0.05)	0.51	0.57	0.50	0.58	0.03	0.07

Values are mean ± SE of seven replicates
Figures in parentheses indicate per cent decrease over control group II

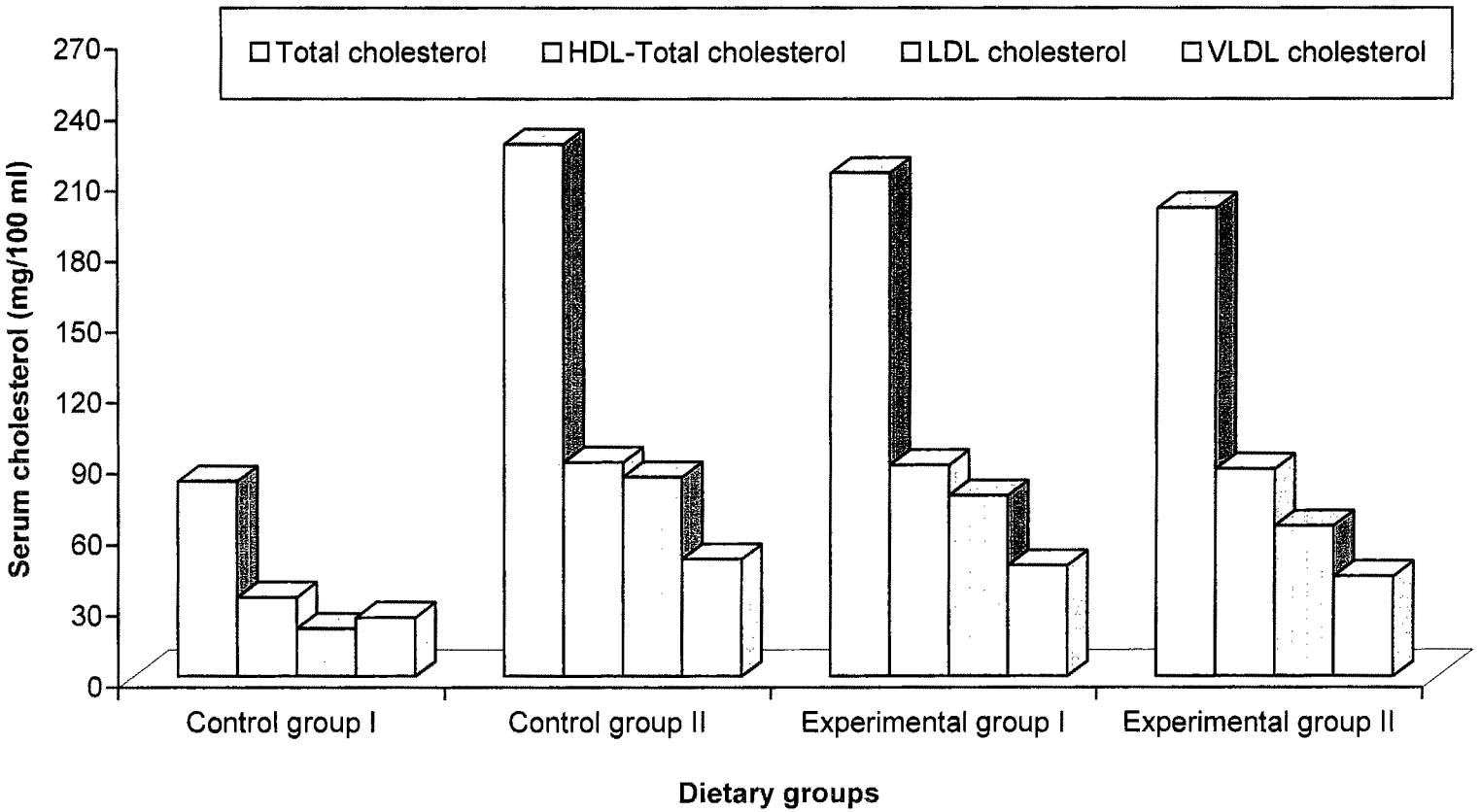


Fig. 11 : Effect of feeding *Moringa oleifera* leaves on serum cholesterol levels in rats

level whereas the group of rats fed on 10 percent *M. oleifera* leaves had 12 percent lower serum cholesterol level over the group II. The decrease in serum cholesterol levels of rats in the present experiment is in agreement with previous reports quoted by Ghasi *et al.* (2000).

A low plasma cholesterol level in rats may be due to the inhibition in cholesterol synthesis or acceleration of cholesterol metabolism by the diet containing *Moringa oleifera* leaves. Although the exact mechanism for these effects is not known, however, an appropriate dietary manipulation may reduce plasma cholesterol level (Kabir *et al.,* 1987). Cheung (1996) suggested that the significant lowering of the serum total cholesterol could be due to a reduction in cholesterol absorption.

4.9.3.2 HDL-cholesterol

Data regarding effect of feeding *M. oleifera* leaves on serum high density lipoprotein (HDL) cholesterol levels have been in given in Table 4.54. HDL-cholesterol level of rats fed control diet I was minimum (33.14 mg/100 ml) and was maximum (90.17 mg/100 ml) in rats fed control diet (II) containing 1 percent cholesterol. Rats fed experimental diet I containing *M. oleifera* leaves at 5 percent level had 89.07 mg HDL-cholesterol/100 ml whereas it was 87.45 mg/100 ml in experimental group II.

HDL-cholesterol and LDL-cholesterol levels serve as the better predictors of cardiac risk than the total cholesterol. HDL-cholesterol is known to have an antiatherogenic activity whereas LDL-cholesterol has atherogenic property (Bajaj *et al.,* 1997). Concentration of HDL is negatively correlated to heart disease because the main function of HDL is to transport the excess of cholesterol from the arterial walls to the liver where the cholesterol is uncoupled and excreted via the bile and to the other organs such

as adrenals, testes and ovaries for the production of adrenal and sex hormones (Vles and Gottenbos, 1989).

4.9.3.3 LDL-cholesterol

Low density lipoprotein (LDL) levels of the rats fed different diets has been given in Table 4.54. Data in Table revealed that LDL-cholesterol level in the group of rats fed control diet (I) containing no cholesterol was 19.98 mg/100 ml, whereas this level was 83.99 mg/100 ml when 1 percent cholesterol was added to control diet (I). The level of LDL-cholesterol in serum was significantly higher in control group II as compared to control group I. Rats fed on extra 1 percent cholesterol showed 76.21 percent increase in LDL-cholesterol.

In the groups of rats fed diet containing *M. oleifera* leaves at 5 and 10 percent, LDL-cholesterol levels were found to be 76.17 and 63.40 mg/100 ml, respectively. The rats fed on experimental diets containing higher percent (10%) of *M. oleifera* leaves had significantly ($P<0.05$) lower LDL-cholesterol levels as compared to the group of rats fed on lower percent of *M. oleifera* leaves as well as control group II. Group of rats fed *M. oleifera* leaves at 5 and 10 percent in the diet were found to have 9.31 and 24.51 percent lower level of serum cholesterol, respectively over the control group II. The results are in agreement with earlier study conducted by Ghasi *et al.* (2000). Significant lowering of LDL-cholesterol concentration could be due to reduction in cholesterol absorption . The decrease in serum LDL-cholesterol is beneficial for reducing risk of heart diseases as this class is responsible for transport of cholesterol to the arteries (Vles and Gottenbos, 1989) where it gets deposited in the lumen of the arteries resulting in blockage of arteries and increase the risk of heart attacks. Low density lipoprotein has a central role in the atherosclerotic process. It

penetrates in the walls of blood vessels where they are oxidized by free radicals and accumulate as a gruel like material that blocks the lumen of the blood vessel and this material can leak into the vessel causing thrombosis (Manson, 1992).

4.9.3.4 VLDL-cholesterol

Serum very low density lipoprotein (VLDL) cholesterol levels of rats fed on different diets has been given in Table 4.54. Group of rats fed on control diet (II) containing cholesterol had the highest serum VLDL-cholesterol concentration (49.31 mg/100 ml), whereas, the group fed on control diet (I) without cholesterol had the lowest serum VLDL-cholesterol concentration (24.69 mg/100 ml). Addition of 1 percent cholesterol in the control diet I increased the serum VLDL-cholesterol level by 49.92 percent.

The very low density lipoprotein (VLDL) cholesterol level in rats fed on 5 percent *M. oleifera* leaves was 46.68 mg/100 ml and VLDL level of rats fed on 10 percent *M. oleifera* leaves was 42.10 mg/100 ml. These values were significantly ($P<0.05$) lower as compared to control group (II). The rats fed on diets containing *M. oleifera* leaves at 5 and 10 percent levels had 5.33 percent and 14.62 percent lower serum VLDL-cholesterol as compared to control group II with cholesterol, respectively.

4.9.3.5 Ratio of serum total cholesterol to HDL-cholesterol and LDL-cholesterol

Data regarding ratio of total cholesterol to HDL-cholesterol and LDL-cholesterol have been given in Table 4.54.

The ratio of total cholesterol to HDL-cholesterol was found to be maximum i.e. 2.50 in control group II (with cholesterol) and minimum in experimental group II (*M. oleifera* leaves 10%) i.e. 2.26.

The ratio of total cholesterol to LDL-cholesterol was maximum i.e. 4.12 in control group I (without cholesterol). However, it was minimum in the control group II (with cholesterol). In the experimental group the values were 2.79 and 3.12, respectively, at 5 and 10 percent levels.

4.9.4 *Moringa oleifera* leaves and Liver cholesterol

4.9.4.1 Total cholesterol

Effect of feeding *M. oleifera* leaves on liver cholesterol levels of rats has been given in Table 4.55 and Fig. 12. Wide variation in the liver cholesterol of rats fed different diets was observed. The liver cholesterol level was found to be the lowest in the control group I fed on control diet with no cholesterol, whereas cholesterol concentration in the liver of rats fed another control diet (II) i.e. containing 1% cholesterol was the highest. Addition of 1 percent cholesterol to control diet (II) increased the level of cholesterol in liver of rats by 10 times.

The cholesterol levels of rats were found to be 1.74 and 1.42 g/100 g liver, when they were fed for 1 month on 5 and 10 percent *M. oleifera* containing diets, respectively (Table 4.55). The deposition of cholesterol in liver was found to be significantly lower in the group fed 10 percent *M. oleifera* than group fed 5 percent *M. oleifera* leaves. Both the groups of rats fed experimental diets had significantly lower cholesterol concentration in the liver than control group. Liver cholesterol decreased by 13 and 29 percent when *M. oleifera* was fed at 5 and 10 percent levels, respectively.

According to Boyd (1975), intake of plants fibre decreased the synthesis of hepatic cholesterol. Mathur *et al.* (1968) reported that direct binding of bile acids by dietary fibre in the lumen diverts cholesterol which is synthesized in the liver into *de novo* synthesis

Table 4.55 Effect of feeding *Moringa oleifera* on liver cholesterol (g/100 g) levels in rats

Dietary group	Total cholesterol	HDL cholesterol	LDL cholesterol	VLDL cholesterol	Total chol : HDL chol	Total chol : LDL chol.
Control group I (without cholesterol)	0.21 ± 0.01 (-89.50)	0.01 ± 0.01 (-90.90)	0.12 ± 0.01 (-92.30)	0.06 ± 0.01 (-87.23)	19.93 ±0.79	1.80 ±0.10
Control group II (with cholesterol)	2.00 ± 0.01	0.11 ± 0.01	1.56 ± 0.02	0.47 ± 0.01	17.84 ±0.17	1.28 ±0.01
Experimental group I *Moringa oleifera* leaves (5%)	1.74 ± 0.01 (-13.00)	0.13 ± 0.01 (18.18)	1.22 ± 0.01 (-21.79)	0.43 ± 0.01 (-8.51)	14.18 ±0.08	1.51 ±0.01
Experimental group II *Moringa oleifera* leaves (10%)	1.42 ± 0.01 (-29.00)	0.14 ± 0.01 (27.27)	0.85 ± 0.01 (-45.51)	0.40 ± 0.01 (-14.89)	10.92 ±0.42	1.65 ±0.02
CD (P<0.05)	0.03	0.01	0.02	0.02	1.48	0.14

Values are mean ± SE of seven replicates
Figures in parentheses indicate increase (+) or decrease (-) over control group II

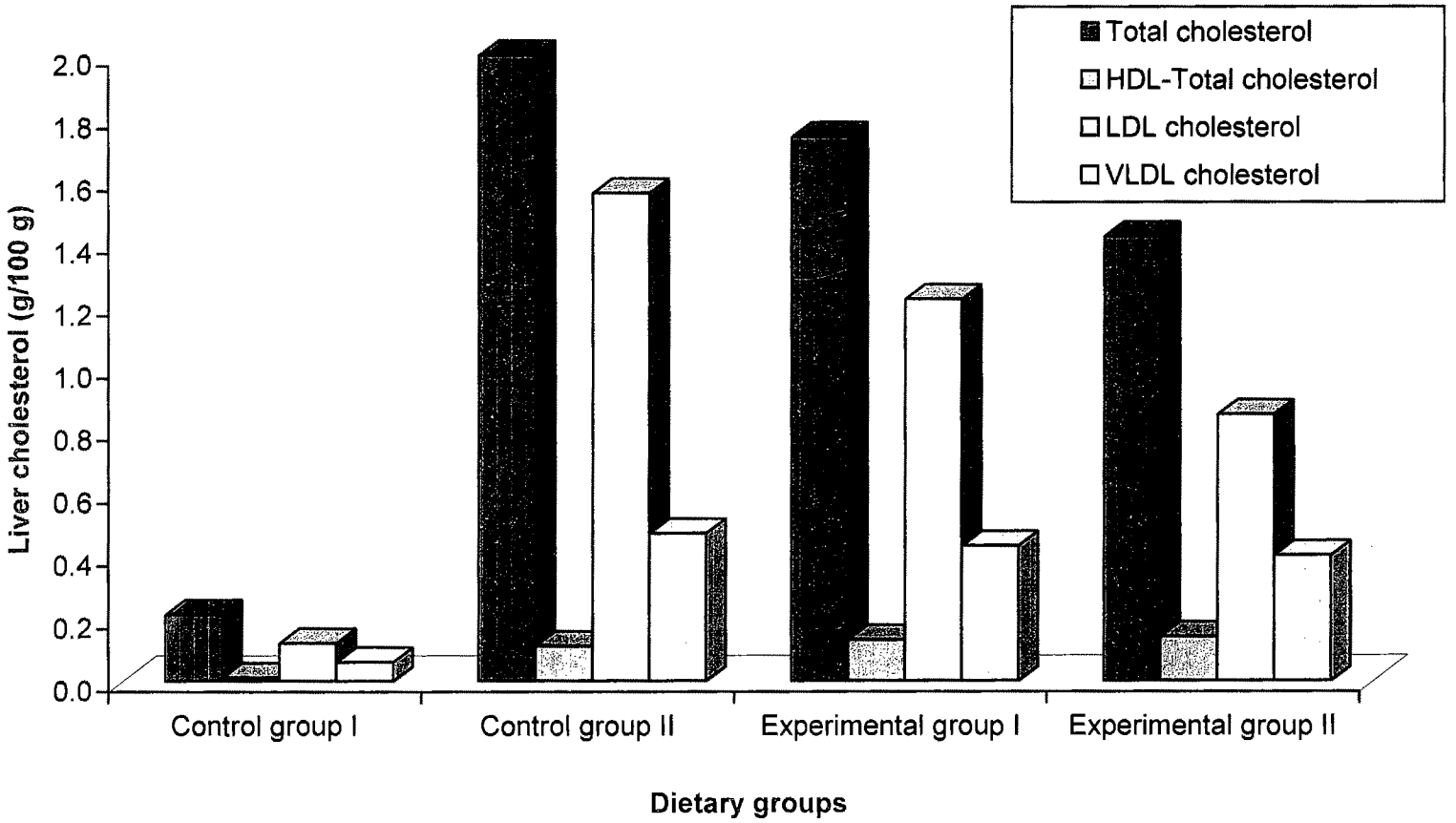


Fig. 12 : Effect of feeding *Moringa oleifera* on liver cholesterol levels in rats

of bile acid and secondly bound bile acids would be unavailable for micelle formation.

With inclusion of fibre in the diet, increase in the daily faecal output of total bile acids have been reported. Their action may be the result of chemical binding of bile acids or alteration of bacterial flora, which leads to change in the metabolism of bile acids. The net effect of a combination of these mechanisms would be the inhibition of reabsorption of bile acids in the small intestine and thus, requirement of cholesterol for bile acid synthesis is increased. This leads to interruption of entero-hepatic cycle and thus, pool of cholesterol is depleted. Therefore, less cholesterol is available for storage in liver.

4.9.4.2 Liver HDL-cholesterol

Liver HDL-cholesterol levels of rats fed control and *M. oleifera* leaves have been given in Table 4.55. HDL-cholesterol ranged between 0.01 to 0.14 g/100 g of liver. Control group I fed no additional cholesterol in diet had minimum amount of HDL-cholesterol in liver. The group of rats fed on control diet (II) containing 1 percent additional cholesterol had significantly higher level of HDL-cholesterol over the control group I.

HDL-cholesterol level of rats fed experimental diets I and II containing 5 and 10 percent *M. oleifera* leaves was 0.13 and 0.14 g/100 g liver of rats, respectively (Table 4.55), which was significantly higher than control group II fed on diet containing cholesterol. Higher the level of *M. oleifera* leaves in the diet, higher the level of HDL-cholesterol in liver.

HDL-cholesterol level in liver was 18.18 and 27.27 percent higher in rats fed experimental diet I and II containing *M. oleifera* at 5 and 10 percent, respectively.

HDL is principle acceptor of cholesterol from liver and diverts it to other organ for its disposal. HDL-cholesterol had a protective effect and act to prevent oxidation of LDL and remove cholesterol that accumulates in the walls of blood vessel (Manson, 1992).

4.9.4.3 Liver LDL-cholesterol

Data regarding LDL-cholesterol levels in the livers of rats fed different diets have been given in Table 4.55. The LDL-cholesterol concentration in the liver was found to be the lowest (0.12 g/100 g tissue) in group of rats fed control diet (I) with no cholesterol, whereas level was the highest (1.56 g/100 g tissue) in the liver of rats fed control diet (II) containing 1 percent additional cholesterol. The group of rats fed with control diet (II) deposited about twelve times more LDL-cholesterol in the liver than the group of rats fed on control diet (I).

The LDL-cholesterol concentration in the group of rats fed on experimental diet containing 5 percent *M. oleifera* leaves was 1.22 g/100 g liver tissue; the deposition of LDL-cholesterol in the liver of rats was significantly lower than the group fed control diet (II) (Table 4.55). Inclusion of *M. oleifera* leaves in the diet of rats resulted in 21.79 and 45.51 percent reduction in LDL-cholesterol in the liver as compared to liver of rats fed on control diet (II) containing additional cholesterol. The deposition of LDL-cholesterol in liver of rats fed experimental diet II containing 10 percent *M. oleifera* was significantly less than the experimental group I fed on diet containing 5 percent *M. oleifera* leaves.

4.9.4.4 Liver VLDL-cholesterol

Very low density lipoprotein (VLDL) cholesterol levels of rats fed on control diets and diets containing *M. oleifera* has been given in Table 4.55. Liver VLDL-cholesterol levels ranged between 0.06 to 0.47 g/100 g tissue. The control group I fed on control diet (I) with

no cholesterol had the lowest level whereas control diet (II) fed on diet containing 1 percent cholesterol had the highest level of VLDL-cholesterol in the liver. The cholesterol was eight times higher as compared to the level in the liver of rats fed no cholesterol.

The VLDL-cholesterol levels of rats fed on a diet containing 5 and 10 percent *M. oleifera* was 0.43 and 0.40 g/100 g liver, respectively (Table 4.55). The deposition of VLDL-cholesterol was found to be significantly less in the group of rats fed on diet containing 10 percent *M. oleifera* as over the group fed on control diet (II). VLDL-cholesterol levels in the liver were found to be 8.51 and 14.89 percent lower when the rats were fed on diet containing 5 and 10 percent *M. oleifera* than the control diet (II).

4.9.4.5 Ratio of liver total cholesterol to HDL-cholesterol and LDL-cholesterol

The ratio of total cholesterol to HDL-cholesterol was found to be maximum in control group I and minimum in experimental group II (*M. oleifera* leaves 10 percent). The ratio of total cholesterol to LDL-cholesterol was maximum in control group I and minimum in control group II.

4.9.4.6 Liver weight and deposition of lipid in liver

Effect of feeding *M. oleifera* on liver weight of rats has been given in Table 4.56. Liver weight of rats ranged between 2.36 to 2.66 g. It was the lowest in control group I fed on diet containing no cholesterol and the highest in control group II fed on diet containing additional 1 percent cholesterol. Additional cholesterol fed to rats in control group II might have increased the liver weight significantly.

The experimental groups of rats fed on diet containing *M. oleifera* leaves at 5 and 10 percent had 2.60 and 2.52 g liver weight, respectively (Table 4.56). However, the groups of rats fed

Table 4.56 Effect of feeding *Moringa oleifera* leaves on liver weight (g), total cholesterol (mg/liver), total lipid (g/liver) and triglycerides (mg/liver) in rats

Dietary group	Liver weight	Total lipid	Total cholesterol	Triglycerides
Control group I (without cholesterol)	2.36 ± 0.01	0.08 ± 0.01	5.54 ± 0.01	30.77 ± 0.49
Control group II (with cholesterol)	2.66 ± 0.01	0.23 ± 0.01	58.03 ± 0.01	63.49 ± 0.03
Experimental group I (*Moringa oleifera* leaves 5%)	2.60 ± 0.01	0.19 ± 0.01	49.51 ± 0.03	60.54 ± 0.19
Experimental group II (*Moringa oleifera* leaves 10%)	2.52 ± 0.01	0.15 ± 0.01	39.41 ± 0.01	56.01 ± 0.67
CD ($P<0.05$)	0.02	0.02	0.02	1.3

Values are mean ± SE of seven replicates

on 10 percent *M. oleifera* had significantly ($P<0.05$) lower liver weight as compared to another experimental group of rats fed on 5 percent *M. oleifera* leaves as well as control group II. The group of rats fed on experimental group I and II had significantly ($P<0.05$) lower weight of liver than the control group II.

Liver total lipids in different groups of rats ranged from 0.08 to 0.23 g/liver (Table 4.57). The control group I fed on diet containing no additional cholesterol was found to have least total lipids in the liver whereas control group II fed on diet containing 1 percent cholesterol had the highest total lipids/liver. Data suggested that addition of cholesterol in the diet of control group II had significantly increased the deposition of total lipids in the liver.

The group of rats fed on a diet containing 5 or 10 percent *M. oleifera* leaves were found to have lower total lipids per liver than the control group II.

A similar trend was observed when total cholesterol and triglyceride level of liver were calculated for the control and experimental groups. As compared to control group II, a significant lower deposition of total cholesterol and triglyceride in liver were noticed in experimental group of rats fed on diet containing *M. oleifera* leaves. This might be due to inclusion of higher fibre in the diet which resulted in less cholesterol availability for storage in liver.

4.10 Effect of *M. oleifera* seed on total viable count

Total viable count of three water samples i.e. fresh water, stored water and pond water before and after treatment differed significantly (Table 4.57 and Fig. 13).

Data on total viable count/ml of fresh water revealed that before treatment it was 228.33 and after treatment with *M. oleifera* seed it was decreased to 54.66 TVC/ml.

Table 4.57 Effect of *Moringa oleifera* seed on the purification of water in terms of total viable count

Type of water	Total viable count/ml		't' value
	Before treatment	After treatment	
Fresh water	228.33 ± 0.88	54.66 ± 0.88	139.24
Stored water (tank)	320.00 ± 5.77	79.00 ± 1.73	40.29
Pond water	350.00 ± 5.77	84.66 ± 0.88	39.98

Values are means ± SE of three independent determinations

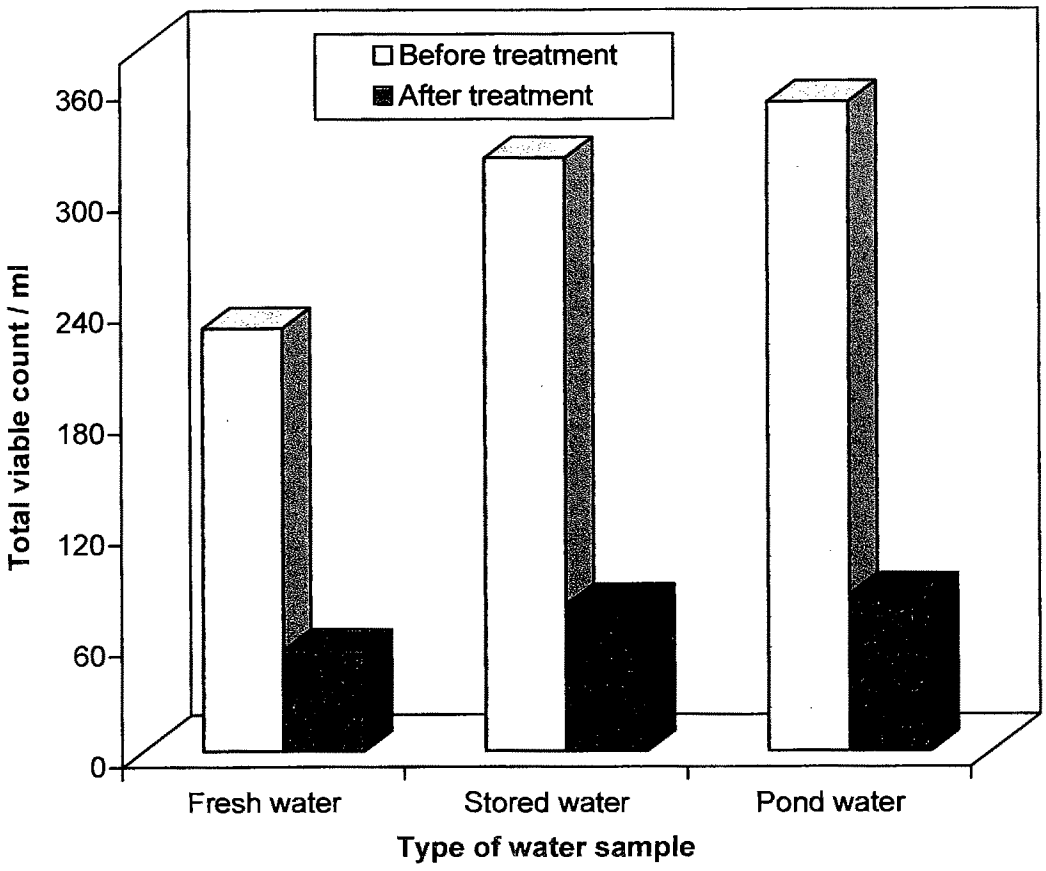


Fig. 13 : Effect of *Moringa oleife.a* seeds on the purification of water in terms of total viable count

In stored water and pond water the total viable count was 320 and 350, respectively and it was decreased to 79 and 84.61 after treating with seeds of *M. oleifera*. Thus, a significant reduction in total viable count took place. The results of the present findings are in agreement with the various workers (Jeyanthi *et al.*, 2004; Johri and Johri, 2004; Mandloi *et al.*, 2004). Olayemi and Alabi (1994) reported that the phytochemical and spectral studies led to elucidation of a steroidal glycoside strophantidin as a bioactive agent in seed. Folkard *et al.* (1995) reported that the *Moringa oleifera* seed kernel contained significant quantities of a series of low molecular weight, water-soluble proteins which in solution, carried an overall positive charge. The proteins were considered to act similarly to synthetic, positively changed polymer coagulants. When added to raw water, the proteins bound to the predominantly negatively changed particular that made raw waters turbid (silt, clay, bacteria etc.). Under proper agitation, these bound particulates then grew in size to form the flocs which might be left to settle by gravity or be removed by filtration.

Summary And Conclusion

Chapter-5

Summary and Conclusion

The emerging benefits of the plant *Moringa oleifera* indicate a great potential towards improving human nutrition. Besides a good source of nutrients, *Moringa oleifera* also has medicinal value. Various studies have shown that it can treat a number of diseases.

Hence, the present study was carried out to assess the nutrient composition of leaves, flowers, pods and seeds of *M. oleifera*, to develop organoleptically acceptable food products from various parts of *M. oleifera*, to estimate nutrient composition and shelf life of most acceptable food products and to study hypocholesterolaemic effect of *Moringa* leaf powder.

Nutritional evaluation of various parts of *M. oleifera* i.e. raw leaves, flowers, pods and seeds revealed that moisture content was the highest in pods (85.56%) whereas seeds (6.87%) had the lowest moisture content. The ascorbic acid content of leaves (219.38 mg/100 g) was significantly ($P<0.05$) higher than pods (118.48 mg/100 g) and flowers (52.83 mg/100 g). The ascorbic acid content in seed was not detected.

β-carotene content of leaves (6.75 mg/100 g) was significantly ($P<0.05$) more than pods (0.09 mg/100 g), flowers (0.04 mg/100 g) and seeds (0.01 mg/100 g).

Crude protein content among the different parts of *M. oleifera* ranged from 15.76 to 38.52 g/100 g; the highest being in seed and the lowest being in pods. Seed had the maximum fat (41.21 g/100 g) whereas pods had the minimum (0.32 g/100 g) fat content. Total ash content of pods (20.48 g/100 g) was significantly ($P<0.05$) higher than flowers (20.14 g/100 g), leaves (19.32 g/100 g) and seeds (4.38 g/100 g). Pods of *M. oleifera* had significantly ($P<0.05$) higher crude fibre content (28.25 g/100 g) as compared to flowers (8.44 g/100 g), leaves (5.83 g/100 g) and seeds (3.26 g/100 g).

Among the various parts of *M. oleifera*, significantly ($P<0.05$) higher total, soluble and insoluble dietary fibre (g/100 g) were determined in pods (59.32, 8.09 and 51.17) followed by flowers (36.48, 7.79 and 28.69), leaves (32.14, 7.28 and 24.85) and seeds (18.25, 3.52, 14.73), respectively. Leaves contained the highest amount of total carbohydrates followed by flowers, pods and seeds in descending order

Among antinutrients, oxalic acid ranged from 264.99 to 403.80 mg/100 g; the highest being in pods and the lowest being in seeds. Maximum phytic acid (1.99 g/100 g) was present in leaves followed by pods (1.83 g/100 g), flowers (1.06 g/100 g) and seeds (0.01 g/100 g) in descending order. Highest saponin content was found in seeds (2.21 g/100 g) while flowers had the lowest (0.82 g/100 g) amount. A non-significant difference was noticed in trypsin inhibitor content of flowers, pods and seeds of *M. oleifera*.

In minerals, total calcium content ranged from 136.33 mg to 1999.81 mg/100 g among different parts of *M. oleifera*; the highest being in leaves and the lowest being in pods. Phosphorus content

was maximum (320 mg/100 g) in pods whereas seed had the minimum (132.53 mg/100 g) phosphorus content.

Seeds had the maximum (75.86 mg/100 g) content of iron followed by leaves (27.55 mg/100 g), pods (21.24 mg/100 g) and flowers (5.16 mg/100 g). Total zinc content ranged from 16.32 to 25.48 mg/100 g in various parts of *M. oleifera*. Seeds had the maximum leaves of potassium (3850.52 mg/100 g) whereas flowers had the minimum (783.23 mg/100 g) level of potassium.

HCl-extractability of calcium ranged from 79.32 to 81.89 percent in various parts of *M. oleifera*. Seeds had minimum (75.53%) whereas flowers had maximum (77.52%) phosphorus extractability. The range of percent HCl-extractability of iron varied from 62.73 to 65.45 percent in different parts, it was maximum in pods (65.45%) and minimum in seeds (62.73%). Leaves had the maximum (75.14%) whereas seeds had the minimum (70.31%) HCl-extractability of zinc. Percent extractability of leaves was significantly ($P<0.05$) higher than rest of parts of *Moringa oleifera*.

In vitro calcium availability was found maximum (25.56%) in leaves followed by seeds (23.93%), pods (22.57%) and flowers (21.52%). On the other hand, *in vitro* iron availability was the highest (13.51%) in leaves while seeds had the lowest (10.27%) *in vitro* iron availability.

Different types of products were developed from various parts of *M. oleifera* i.e. leaves, pods, flowers and seeds. Chutney, leaves *bhuji*, potato and leaves vegetable and *pakora* were prepared by using fresh leaves whereas *sev* and noodles were prepared by incorporation of *M. oleifera* leaf powder at different levels i.e. at 10% percent and 20% level. The products prepared from *M. oleifera* flowers included flower *bhuji*, pea and flower vegetable, green gram and flower vegetable and *raita* and cutlets.

Similarly, various products were prepared from fresh *M. oleifera* pods as well as dried pod powder. The products prepared from fresh *M. oleifera* pods included pod vegetable, potato and pod vegetable, *sambhar* and cutlets. Products supplemented with different leaves of *M. oleifera* pod powder included *sev* and noodles. The *ladoo* and weaning mixture were prepared by incorporating different levels (10 and 20%) of *M. oleifera* seed powder.

According to sensory characteristics, all the products prepared from fresh leaves were acceptable. All the attributes of sensory characteristics i.e. colour, appearance, aroma, texture, taste and overall acceptability scores revealed that leaves *bhuji*, potato and leaves vegetable and *pakora* fell in 'liked moderately' category whereas colour, appearance, aroma and texture of chutney were liked moderately except taste which was neither liked nor disliked by the panelists. Sensory evaluation of different types of *sev* showed that control *sev* i.e. prepared from 100% bengal gram flour and *sev* prepared from 10% *M. oleifera* leaf powder fell in the category of 'liked moderately'. However, the *sev* prepared from 20 percent *M. oleifera* leaf powder was 'liked moderately' in terms of colour and texture but 'liked slightly' in appearance, aroma, taste and overall acceptability.

The overall acceptability which was the mean score of colour, appearance, aroma, texture and taste of control noodles i.e. prepared from 100 percent refined flour and noodles supplemented with 10 percent *M. oleifera* leaf powder were 'liked very much'. However, noodles prepared from 20 percent were 'liked slightly'.

Different attributes of products prepared from flowers i.e. flowers *bhuji*, pea and flower vegetable, green gram and flower vegetable *raita* and cutlets fell in 'liked moderately' to 'liked very much' categories.

Overall acceptability of products prepared from fresh *M. oleifera* pods showed that all the products i.e. pod vegetable, potato and pod vegetable, *sambhar* and cutlets, pickle were 'liked moderately'.

Sensory characteristics of products prepared from dried *M. oleifera* pod powder i.e. *sev* and noodles had shown that *sev* (control) and *sev* supplemented with 10% *M. oleifera* pod powder were 'liked moderately' in all attributes whereas colour and texture of *sev* prepared by using 20 percent *M. oleifera* pod powder were 'liked moderately' and appearance, aroma, taste and overall acceptability were 'liked slightly'.

The noodles prepared by using 100 percent refined flour i.e. control were 'liked very much' whereas noodles containing 10 percent *M. oleifera* pod were 'liked moderately' in all the sensory parameters. The overall acceptability of noodles supplemented with 20 percent *M. oleifera* pod powder had overall acceptability score in 'liked slightly' category.

Ladoo and weaning food mixture supplemented with different levels of *M. oleifera* seed powder were not acceptable to human palate and disliked.

Nutritional evaluation of products prepared with fresh *M. oleifera* leaves showed that chutney had the maximum (77.54%, 22.65 mg/100 g) whereas *pakora* had the minimum (54.24%, 52.65 mg/100 g) moisture and ascorbic acid contents, respectively. Crude protein content among the four different products varied from 8.62 to 10.67 g/100 g; the highest being in *pakora* and the lowest being in leaves *bhuji*. *Pakora* had significantly ($P<0.05$) higher fat content than potato and leaves vegetable, leaves *bhuji* and chutney. It might be due to addition of higher visible fat in *pakora* as compared to rest of products. *Pakora* had the maximum (0.92

g/100 g) whereas the leaves *bhuji* had the minimum (0.80 g/100 g) ash content. It might be due to addition of chickpea flour which had higher ash content. Potato and leaves vegetable had significantly ($P<0.05$) higher crude fibre content followed by leaves *bhuji*, chutney and *pakora*.

Total dietary fibre content of products prepared from fresh *M. oleifera* leaves varied from 14.09 to 16.74 g/100 g; the highest being in chutney (16.74 g/100 g); the lowest being in *pakora* (14.09 g/100 g). *Pakora* had the maximum (6.02 g/100 g) soluble dietary fibre content followed by chutney (5.72 g/100 g), potato and leaves vegetable (5.66 g/100 g) and leaves *bhuji* (5.13 g/100 g) in descending order.

Insoluble dietary fibre of the products prepared from fresh *M. oleifera* leaves varied from 8.07 to 11.03 g/100 g. Chutney had the maximum total carbohydrate content (84.92 g/100 g) the minimum.

Among antinutrients, oxalic acid, phytic acid and saponin contents of chutney were found to be maximum. Trypsin inhibitor activity was not detected in chutney, leaves *bhuji*, potato and leaves vegetable and *pakora* as the raw *Moringa oleifera* leaves had negligible TIA.

Among the minerals, total calcium, phosphorus, iron and potassium contents were found to be maximum in chutney whereas zinc content more in *pakora* than rest of products prepared from fresh *M. oleifera* leaves.

HCl-extractability of calcium, phosphorus and iron was found maximum in chutney whereas zinc and potassium extractability was higher in potato and leaves vegetable than the remaining products. *In vitro* availability of calcium and iron was significantly ($P<0.05$) higher in potato and leaves vegetable followed by leaves

bhuji, *pakora* and chutney. As the level of antinutrients reduced, there was increase in the bioavilability of calcium and iron. These antinutrients are known to hinder the mineral absorption.

Ascorbic acid content in *sev* and noodles prepared by incorporating *M. oleifera* leaf powder were not detected whereas in noodles it was almost negligible but differed significantly among themselves in all the three types. β-carotene and crude protein contents were significantly ($P<0.05$) higher in *sev* and noodles prepared from 20 percent *M. oleifera* leaf powder as compared to those having 10 percent leaf powder and control.

Sev prepared from 20 percent *M. oleifera* leaf powder had maximum (20.74 g/100 g) fat content whereas in case of noodles, this nutrient was found more in control (12.86 g/100 g). Total ash and crude fibre contents of *sev* and noodles were higher when supplementation with 20 percent *M. oleifera* leaf powder was made.

Among *sev* and noodles, those supplemented with 20 percent *M. oleifera* leaf powder had significantly ($P<0.05$) higher amount of total, soluble and insoluble dietary fibre than control and those having10 percent supplementation with leaf powder.

The maximum total carbohydrate content was found in *sev* prepared from 10 percent *M. oleifera* leaf powder. On the other hand, control noodles i.e. without addition of *M. oleifera* leaf powder contained the maximum content of total carbohydrates. Among the antinutrients, data regarding oxalic acid, phytic acid, saponins and trypsin inhibitors were found to be maximum in *sev* and noodles containing 20 percent *M. oleifera* leaf powder.

Total calcium, phosphorus, iron, potassium and zinc contents of *sev* and noodles supplemented with 20 percent *M. oleifera* leaves were significantly higher ($P<0.05$) than those of *sev* and noodles supplemented with 10 percent *M. oleifera* leaf powder and control.

The values for HCl-extractability of calcium, phosphorus and potassium of *sev* varied significantly ($P<0.01$) among themselves. Maximum calcium, phosphorus and potassium extractability was also observed in noodles. Iron extractability was significantly higher in control *sev* followed by 20 percent and 10 percent leaf powder supplemented *sev*. Non-significant differences were found in different types of noodles prepared from *M. oleifera* leaf powder. Zinc extractability of *sev* and noodles prepared by incorporation of 10 percent and 20 percent *M. oleifera* leaf powder differed non-significantly.

In vitro calcium and iron availability of *sev* was found to be maximum in control and minimum in *sev* supplemented with 20 percent *M. oleifera* leaf powder. On the contrary, calcium and iron availability of noodles was found to be maximum in noodles incorporating 20 percent *M. oleifera* leaf powder and minimum in control i.e. without addition of *M. oleifera* leaf powder.

Nutritional evaluation of products prepared from *M. oleifera* flowers revealed that the highest moisture content (83.36%) was in *raita*; whereas the lowest was in cutlets (60.06%).

Similarly, ascorbic acid content was maximum in *raita* (20.02 mg/100 g). β-carotene content of all the products i.e. flower *bhuji*, pea and flower vegetable, green gram and flower vegetable and cutlets varied non-significantly except *raita* which was significantly ($P<0.05$) lower as compared to other products.

Crude protein content varied from 3.45 to 9.56 g/100 g, the highest being in green gram and flower vegetable and the lowest being in *raita*. Cutlet had the maximum (19.62 g/100 g) whereas the *raita* had the minimum (4.11 g/100 g) fat content. Total ash content of products ranged from 0.53 to 1.33 g/100 g on DM basis. Cutlets had the maximum whereas *raita* had the minimum ash

content. A significant ($P<0.05$) difference in crude fibre content of products developed from flowers was observed.

Green gram and flower vegetable had the maximum (16.76 g/100 g) total dietary fibre followed by pea and flower vegetable (16.61 g/100 g); the minimum total dietary fibre content was observed in *raita*. Total carbohydrates content of products ranged from 68.22 to 90.12 g/100 g; the highest being in *raita* and the lowest being in green gram and flower vegetable.

Among antinutrients, oxalic acid, phytic acid and saponin contents of green gram and flower vegetable were found to be maximum. Trypsin inhibitor activity was not detected in products prepared from flowers of *M. oleifera*.

Total calcium, phosphorus and potassium contents were maximum in *raita*. Iron content of green gram and flower vegetable was significantly ($P<0.05$) higher as compared to rest of products developed from flowers of *M. oleifera*. Total zinc content of various products ranged from 5.41 to 7.87 mg/100 g. *Raita* had significantly lower zinc content as compared to other products.

HCl-extractability of minerals including calcium, phosphorus, iron, zinc and potassium was the highest in *raita*. *In vitro* availability of calcium and iron were found to be maximum in green gram and flower vegetable and pea vegetable and flower vegetable, respectively whereas both values were found to be minimum in cutlets. Increase in availability of calcium and iron might be observed due to blanching as compared to frying treatment.

Nutritional composition of products prepared from fresh *M. oleifera* pods revealed that the highest moisture content (80.54%) to be in *sambhar* and the lowest (36.69%) in pickle. Ascorbic acid content varied from 5.50 to 89.32 mg/100 g in various products developed containing fresh *M. oleifera* pods; the

highest being in pickle and the lowest being in cutlets. Pickle had the maximum (0.09 mg/100 g) β-carotene content whereas cutlets had minimum (0.05 mg/100 g) β-carotene content.

Protein content among the various products varied from 4.93 to 11.42 g/100 g; the highest (11.42 g/100 g) being in pod pickle and the lowest (4.93 g/100 g) being in cutlets. The fat and crude fibre content of pickle was significantly ($P<0.05$) higher as compared to rest of products prepared from *M. oleifera* pods due to addition of more amount of visible fat and *Moringa oleifera* pods, respectively. *Sambhar* had the lowest ash (0.65 g/100 g) content while cutlets had the highest amount of ash content.

Among the dietary fibre, significantly ($P<0.05$) higher total, soluble and insoluble dietary fibre (g/100 g) content was determined in pickle (39.35, 9.56 and 29.79) than rest of products prepared from fresh *M. oleifera* pods. Total carbohydrates content was maximum in *sambhar* (72.36 g/100 g) and minimum (47.81 g/100 g) was in pickle.

The contents of oxalic acid, phytic acid and saponin in pickle was found to be maximum. The products prepared from fresh pods of *M. oleifera* had no trypsin inhibitor activity.

Total calcium, phosphorus, iron, zinc and potassium contents were the highest in pickle. HCl-extractability of all the minerals i.e. calcium, phosphorus, zinc, potassium and iron were maximum in *sambhar*. Iron extractability was higher in pickle as compared to rest of products prepared from fresh *M. oleifera* pods. *In vitro* calcium and iron availability were found to be maximum in pod vegetable and *sambhar*, respectively whereas cutlets contained the minimum (22.70 and 12.83%) availability of both calcium and iron.

Nutritional evaluation of products prepared from dried *M. oleifera* pods showed that moisture content of *sev* prepared by

incorporating *M. oleifera* pod powder at 10 percent and 20 percent was almost similar but it was significantly ($P<0.05$) higher than the control. Noodles supplemented with 20 percent *M. oleifera* pod powder had significantly higher moisture content followed by 10 percent noodles and control. Ascorbic acid was not detectable in various types of *sev* whereas ascorbic acid content of noodles ranged from 0.08 to 0.13 mg/100 g and did not differ significantly ($P<0.05$) among themselves. β-carotene and protein content were found to be maximum in *sev* and noodles supplemented with 20 percent *M. oleifera* pod powder.

Sev prepared by incorporating 20 percent *M. oleifera* pod powder had significantly ($P<0.05$) higher fat content followed by *sev* (10%) and control. Ash content ranged from 12.61 to 12.86 g/100 g in different types of noodles. *Sev* and noodles supplemented with 20 percent *M. oleifera* pod powder had the highest ash and crude fibre contents. Among dietary fibre, total, soluble and insoluble dietary fibre contents were higher in *sev* and noodles supplemented with 20 percent *M. oleifera* pod powder. Total carbohydrate content of *sev* and noodles prepared without addition of *M. oleifera* pod powder i.e. control were found to be maximum.

Oxalic acid, phytic acid and saponins were significantly ($P<0.05$) higher in *sev* and noodles prepared by incorporating 20 percent *M. oleifera* pod powder. However, trypsin inhibitor activity of different types of *sev* and noodles differed non-significantly.

Total calcium, phosphorus, iron, potassium and zinc content of *sev* and noodles supplemented with 20 percent *M. oleifera* pods were significantly ($P<0.05$) higher than those of *sev* and noodles supplemented with 10 percent *M. oleifera* leaf powder and control.

Maximum calcium, phosphorus and iron extractability was observed in control *sev* i.e. prepared without the addition of

M. oleifera pod powder. On the contrary, extractability of zinc and potassium was higher in *sev* supplemented with 20 percent *M. oleifera* pod powder. It was observed that noodles supplemented with 20 percent *M. oleifera* pod powder had maximum HCl-extractability of various minerals including calcium, phosphorus, iron, zinc and potassium.

In vitro calcium and iron availability was significantly higher ($P<0.05$) in *sev* prepared without addition of *M. oleifera* pod powder whereas noodles supplemented with 20 percent *M. oleifera* pod powder had maximum calcium and iron availability.

Storage studies indicated that different types of *sev* prepared from *M. oleifera* leaf as well as pod powder were acceptable up to the end of 8th week of storage. Period of storage had significant ($P<0.05$) effect on overall acceptability of all kinds of *sev* and score for overall acceptability decreased significantly during 8th week of storage over the fresh ones.

Mean scores for different organoleptic characteristics of *M. oleifera* pod pickle were in the category of 'liked moderately'. Improvement was observed in the taste and overall acceptability of the pickle as the storage period was prolonged, however the difference was non-significant. Fat acidity and free fatty acids of different kinds of *sev* and pickle increased with increase in storage period.

Nutritional changes were observed when *sev* and pickle were stored for varying periods. The ascorbic acid could not be detected in different types of *sev* on zero day of storage. Overall different types of *sev* had significant variation in their iron and oxalic acid contents as the storage period advanced. The ascorbic acid content of fresh pod pickle was 89.33 mg/100 g on 1st day of storage. After one month of storage, loss up to the extent of 15 percent in

ascorbic acid over the fresh one had occurred. This loss went on increasing as the period of storage prolonged. Iron and oxalic acid contents of pickle also decreased significantly as the period of storage increased.

Animal studies indicated that food consumption of control group I fed on synthetic diet without cholesterol was found to be maximum (215.10 g) whereas weight gain by rats in control group I was the lowest (48.14 g) and it was the highest (58.25 g) for rats in control group II containing 1 percent cholesterol. Weight gain was significantly ($P<0.05$) lower in experimental groups containing *Moringa oleifera* leaves at different levels i.e 5% and 10% over the control group II. The feed efficiency ratio (FER) of experimental diets containing 5 and 10 percent *M. oleifera* leaves was 0.28 and 0.27, respectively. These values were significantly ($P<0.05$) lower than the control diet II (with cholesterol). PER value for control group II (with cholesterol) was significantly ($P<0.05$) higher than experimental group fed on diet containing 5 and 10 percent *M. oleifera*.

The group of rats fed on higher percent (10%) of *M. oleifera* leaves had significantly ($P<0.05$) lower levels of serum cholesterol as compared to group fed lower percent (5%) of *M. oleifera* leaves.

Rats fed experimental diet I containing *M. oleifera* leaves at 5 percent level had 89.07 mg/100 g HDL-cholesterol whereas it was 87.45 mg/100 ml in experimental group II. The rats fed on experimental diets containing higher percent (10%) of *M. oleifera* leaves had significantly ($P<0.05$) lower LDL-cholesterol levels than the group of rats fed on lower percent of *M. oleifera* leaves as well as control group II.

The very low density lipoprotein (VLDL) cholesterol level in rats fed on 5 percent *M. oleifera* leaves was 46.68 mg/100 g and

VLDL of rats fed on 10 percent *M. oleifera* leaves was 42.10 mg/100 ml. These values were significantly ($P<0.05$) lower as compared to that of control group II (with cholesterol). Rats fed experimental diet at different levels i.e. 5 percent and 10 percent had significantly lower cholesterol concentrations in the liver as compared to control group. HDL-cholesterol of the rats fed different diets ranged from 0.01 to 0.14 g/100 g of liver. HDL-cholesterol level of rats fed on experimental diets I and II containing 5 and 10 percent *M. oleifera* leaves was 0.13 and 0.14 g/100 g liver and these values were higher than that of control group II fed on diet containing cholesterol.

Inclusion of *M. oleifera* leaves in the diet of rats resulted in 21.79 and 45.57 percent reduction over the control diet II in LDL-cholesterol in the liver. The VLDL-cholesterol levels in liver were found to be 8.51 and 14.89 percent when the rats were fed on diets containing 5 and 10 percent *M. oleifera* and these values were significantly lower than those in the liver of rats fed on control diet (II).

The experimental groups of rats fed on diet containing *M. oleifera* leaves at 5 and 10 percent had significantly ($P<0.05$) lower liver weight than those fed on control diet II. Total lipids in different groups of rats ranged from 0.08 to 0.23 g/liver among experimental groups. These were significantly ($P<0.05$) lower in experimental groups than found in control group II. A similar trend was observed when total cholesterol and triglyceride levels of liver were calculated for the control and experimental groups. A significant lower deposition of total cholesterol and triglycerides in liver were noticed in rats fed on experimental diets than those fed on control diet II.

Different water samples i.e. fresh water, stored water and pond water when treated with *M. oleifera* seeds had significant ($P<0.05$) reduction in total viable count .

Overall, it may be inferred from the present study that various parts of *M. oleifera* are edible and nutritious. The products developed from leaves, flowers and pods in the present study were organoleptically acceptable and nutritionally superior. Supplementation of *M. oleifera* leaf as well as pod powder improved the level of protein, dietary fibres, β-carotene and minerals in the developed food products. These food products can be useful in bridging the nutrition gap of the people from all ages. Development and consumption of such nutritious food products can go a long way in incorporating the nutritional status of the people. In future, efforts are required to popularize these nutritious *Moringa oleifera* based food products among masses to ameliorate micronutrient deficiencies. Moreover, lowering serum as well as liver cholesterol in rats indicated therapeutic properties of the plant too.

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Appendix-I

Hedonic Scale

Name: Dated:

Product:

Test these samples and check how much you like or dislike each one. Use appropriate scale to show your attitude by assessing points that best describe your feeling about the sample. An honest expression of your feeling will help to obtain unbiased data.

Code No.	Colour	Appearance	Flavour	Texture	Taste	Overall acceptability

Rate	Organoleptic scores
Liked extremely	9
Liked very much	8
Liked moderately	7
Liked slightly	6
Neither liked nor disliked	5
Disliked slightly	4
Disliked moderately	3
Disliked very much	2
Disliked extremely	1

Note: Please rinse your mouth before and after testing

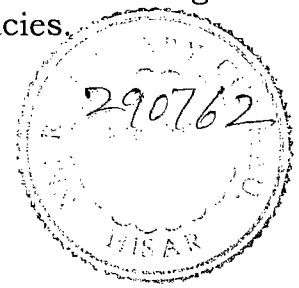
Abstract

1.	Title of thesis	:	Development and Nutrient Composition of Value Added Products from Drumstick (*Moringa oleifera*)
2.	Full name of degree holder	:	Rachna
3.	Admission No.	:	2002HS113D
4.	Title of degree	:	Doctor of Philosophy
5.	Name and address of Major Advisor	:	Dr.(Mrs.) Neelam Khetarpaul Professor Department of Foods & Nutrition CCS Haryana Agril. University Hisar-125004, India
6.	Degree awarding University/ Institute	:	CCS Haryana Agricultural University Hisar
7.	Year of award of degree	:	2006
8.	Major subject	:	Foods & Nutrition
9.	Total No. of pages in thesis	:	169
10.	No. of words in the abstract	:	356

(An abstract of the dissertation submitted to the CCS Haryana Agricultural University in partial fulfilment of the requirements for the degree of Ph.D.).

Nutritional analysis of various parts of the *Moringa oleifera* revealed that leaves had significantly ($P<0.05$) higher ascorbic acid , β-carotene and calcium contents whereas pods had the maximum amount of dietary fibre, phosphorus and potassium. Seeds were particularly rich in crude protein, fat, iron and zinc. Flowers of *Moringa oleifera* also contained these nutrients in sufficient amount. Among antinutrients, highest oxalic acid, phytic acid and saponins were found in pods, leaves and seeds, respectively. Seeds and flowers had the lowest amount of these antinutrients. Trypsin inhibitor activity was negligible in various parts of *Moringa oleifera*. Leaves had the maximum bioavailability of calcium (25.56%) and iron (13.51%). All the products prepared from leaves, flowers and pods were organoleptically acceptable as these were 'liked moderately'. *Ladoo* and weaning food mixture prepared from seeds were not acceptable. Among various fresh leaves

products, potato and leaves vegetable was organoleptically the best. Overall acceptability score was higher for cutlets containing *Moringa oleifera* flowers than that for flower *bhuji,* pea and flower vegetable and green gram and flower vegetable. Out of various products containing *Moringa oleifera* pods i.e. pod vegetable, potato and pod vegetable, pickle, *sambhar* and cutlets, pickle was liked the most. *Sev* and noodles containing 10% dried leaf powder or dried pod powder of *Moringa oleifera* leaves as well as pods were liked the most. Various products developed form fresh leaves, flowers and pods contained appreciable amount of β-carotene (0.01-3.77 mg/100 g), crude protein (3.45-11.42 g/100 g), total dietary fibre (10.25-39.35 g/100 g), iron (1.57-18.50 mg/100 g), calcium (79.36-1203.31 mg/100 g), phosphorus (82.92-260.55 mg/100 g) and potassium (349.36-1165.39). These products had less amount of oxalic acid (149.60-325.53 mg/100 g), phytic acid (0.52-1.53 g/100 g) and saponin (0.20-0.96 g/100 g) due to various processing treatments involved in preparation of products. Due to supplementation of leaf as well as pod powder in *sev* and noodles, there was significant ($P<0.05$) increase in crude protein, β-carotene, dietary fibre, calcium, phosphorus, iron, zinc and potassium content. Different types of *sev* prepared from *Moringa oleifera* leaves as well as pod powder were acceptable up to 8th week of storage. There was significant ($P<0.05$) improvement in the taste and overall acceptability of pickle when stored for 5 months. Fat acidity and free fatty acid content of different kinds of *sev* and pickle increased with increase in storage period. Overall different types of *sev* had significant ($P<0.05$) variation in their iron and oxalic acid content as the storage period prolonged. Similarly, there was significant ($P<0.05$) reduction in ascorbic acid (89.33-33.16 mg/100 g), iron (18.50-18.31 mg/100 g) and oxalic acid (300.62-295.56 mg/100 g) during storage of pickle. Animal studies revealed that there was significant ($P<0.05$) reduction in total cholesterol (5.38-12.00%, 13.00-29.00%), LDL-cholesterol (9.31-24.51%, 21.79-45.51%) and VLDL-cholesterol (5.33-14.62%, 8.51-14.89%) of the serum as well as liver when feeding synthetic diet containing different levels (5% and 10%) of *Moringa oleifera* leaf powder in experimental group of rats. Total viable counts of fresh, stored and pond water decreased significantly upon their treatment with *Moringa oleifera* seeds. Hence, consumption of these products prepared from leaves, flowers and pods of *Moringa oleifera* is very useful form nutrition point of view and shall be encouraged among masses so as to ameliorate micronutrient deficiencies.



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