NUTRITIONAL EVALUATION OF AMLA (*Phyllanthus emblica*) POMACE AND ITS VALUE ADDED PRODUCTS

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in

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BENGALURU **DECEMBER, 2021**

Affectionately dedicated to beloved Parents and My Guide

DEPARTMENT OF FOOD SCIENCE AND NUTRITION
UNIVERSITY OF AGRICULTURAL SCIENCES
BANGALORE

CERTIFICATE

This to certify that the thesis entitled **"NUTRITIONAL EVALUATION OF AMLA (*Phyllanthus emblica*) POMACE AND ITS VALUE ADDED PRODUCTS"** submitted by **Mr. RAJU, C. A., ID NO. PALB 9258,** for the award of degree of **MASTER OF SCIENCE (Agriculture)** in **FOOD SCIENCE AND NUTRITION** of the **UNIVERSITY OF AGRICULTURAL SCIENCES,** Bangalore, is a record of *bona-fide* research work carried out by this during the period of his study in this University, under my guidance and supervision and no part of the thesis has been submitted for the award of any other degree, diploma, associateship, fellowship or other similar titles.

Bengaluru
December, 2021

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Major Advisor

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1. **(K. GEETHA)**
2. **(KALPANA, B.)**
3. **(A. SATHISH)**
4. **(SRINIVASAPPA, K. N.)**

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Struggles

...any omission in this small manuscript does not lack of gratitude.

Bengaluru

December, 2021 **(RAJU, C. A.)**

NUTRITIONAL EVALUATION OF AMLA (*Phyllanthus emblica*) POMACE AND ITS VALUE ADDED PRODUCTS

RAJU, C. A.

THESIS ABSTRACT

Amla (*Phyllanthus emblica*) commonly known as Indian Gooseberry, belongs to Euphorbiaceae family is known for its cool, refreshing, pleasant taste and utilised for various products and beverages. During processing, massive amounts of amla pomace that is an abundant source of polyphenols, ascorbic acid, dietary fibre and tannins is discarded as waste. Hence, the present study was undertaken to standardize the process of dehydration, analyse the nutrients and to develop value added products from dehydrated amla pomace powder. The physico-functional properties *i.e.,* pH 3.44, colour *L** 86.47, *a** -0.74 and *b** 11.13, particle density 1.23 g/cm^3, water holding capacity 12.30 g/g, water binding capacity 12.37 g/g and swelling capacity 13 mL/g. The nutrient composition was found to have good amount of total dietary fibre (41.7g) and minerals like calcium (128mg), phosphorus (116mg) and magnesium (48mg) along with low fat (0.18g), low carbohydrate (7g) and low calorific value (36 Kcal) per 100g. The phytonutrients, *viz.*, ascorbic acid, tannins and polyphenols were 432, 524 and 677 mg per 100g, respectively. *Chikki* and *chutney* powder with amla pomace powder at 6% and 10% substitutional levels had higher sensory scores and improved the nutritional quality. The microbial counts were within the permissible limits during 45 days of storage period. The consumer acceptance was found to be acceptable. The cost of production of *Chikki* and *chutney* powder was lower since the raw material cost was nil. Hence, amla pomace powder, a nutritious by-product can be utilized effectively as a functional ingredient in value added products.

December, 2021

Department of Food Science and Nutrition
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SHAMSHAD BEGUM, S.
(Major Advisor)

ನೆಲ್ಲಿಕಾಯಿ (*ಫಿಲಾಂಥಸ್ ಎಂಬ್ಲಿಕಾ*) ಶೇಷ ಮತ್ತು ಅದರ ಮೌಲ್ಯವರ್ಧಿತ ಉತ್ಪನ್ನಗಳ ಪೌಷ್ಟಿಕಾಂಶದ ಮೌಲ್ಯಮಾಪನ

ರಾಜು, ಸಿ. ಎ.

ಪ್ರಬಂಧದ ಸಾರಾಂಶ

ನೆಲ್ಲಿಕಾಯಿಯನ್ನು (*ಫಿಲಾಂಥಸ್ ಎಂಬ್ಲಿಕಾ*) ಸಾಮಾನ್ಯವಾಗಿ ಇಂಡಿಯನ್ ಗೂಸ್‌ಬೆರ್ರಿ ಎಂದು ಕರೆಯಲಾಗುತ್ತದೆ, ಇದು ಯುಫೋರ್ಬಿಯೇಸಿ ಕುಟುಂಬಕ್ಕೆ ಸೇರಿದ ಹಳದಿ ಮಿಶ್ರಿತ ಹಸಿರು ಬಣ್ಣದ ಹಣ್ಣು. ನೆಲ್ಲಿಕಾಯಿಯು ತಂಪಾದ, ಉಲ್ಲಾಸಕರ, ಆಹ್ಲಾದಕರ ರುಚಿಗೆ ಹೆಸರುವಾಸಿಯಾಗಿರುವುದರಿಂದ ವಿವಿಧ ಉತ್ಪನಗಳು ಮತ್ತು ಪಾನೀಯಗಳನ್ನು ತಯಾರಿಸಲು ಬಳಸಲಾಗುತ್ತದೆ. ಸಂಸ್ಕರಣೆಯ ಸಮಯದಲ್ಲಿ ಪಾಲಿಫಿನಾಲ್‌ಗಳು, ಆಸ್ಕಾರ್ಬಿಕ್ ಆಮ್ಲ, ನಾರಿನಾಂಶ ಮತ್ತು ಟ್ಯಾನಿನ್‌ಗಳತಂಹ ಪೋಷಕಾಂಶಗಳ ಮೂಲವಾಗಿರುವ ನೆಲ್ಲಿಕಾಯಿಯ ಶೇಷವನ್ನು ಬೃಹತ್ ಪ್ರಮಾಣದಲ್ಲಿ ತ್ಯಾಜ್ಯವಾಗಿ ತಿರಸ್ಕರಿಸಲಾಗುತ್ತದೆ. ಆದ್ದರಿಂದ, ನಿರ್ಜಲೀಕರಣದ ಪ್ರಕ್ರಿಯೆಯನ್ನು ಪ್ರಮಾಣೀಕರಿಸಲು, ಪೋಷಕಾಂಶಗಳನ್ನು ವಿಶ್ಲೇಷಿಸಲು ಮತ್ತು ನಿರ್ಜಲೀಕರಣಗೊಂಡ ನೆಲ್ಲಿಕಾಯಿಯ ಶೇಷ ಪುಡಿಯಿಂದ ಮೌಲ್ಯವರ್ಧಿತ ಉತ್ಪನ್ನಗಳನ್ನು ಅಭಿವೃದ್ಧಿಪಡಿಸಲು ಪ್ರಸ್ತುತ ಅಧ್ಯಯನವನ್ನು ಕೈಗೊಳ್ಳಲಾಗಿದೆ. ನೆಲ್ಲಿಕಾಯಿ ಶೇಷ ಪುಡಿಯ ಭೌತಿಕ–ಕ್ರಿಯಾತ್ಮಕ ಗುಣಲಕ್ಷಣಗಳಾದ ರಸಸಾರ 3.44, ಬಣ್ಣವು ತಿಳಿ ಬಿಳುಪು ($L*$ 86.47), ತಿಳಿ ಹಸಿರು ($a*$ -0.74) ಮತ್ತು ತಿಳಿ ಹಳದಿ ($b*$ 11.13), ಕಣದ ಸಾಂದ್ರತೆ 1.23 ಗ್ರಾಂ/ಸೆ.ಮೀ^{3}, ನೀರನ್ನು ಹಿಡಿದಿಟ್ಟುಕೊಳ್ಳುವ ಸಾಮರ್ಥ್ಯ 12.30 ಗ್ರಾಂ/ಗ್ರಾಂ, ತೇವಾಂಶ ಸೇರಿಸುವಿಕೆ ಸಾಮರ್ಥ್ಯ 12.37 ಗ್ರಾಂ/ಗ್ರಾಂ ಮತ್ತು ಊತ ಸಾಮರ್ಥ್ಯ 13 ಮಿ.ಲೀ/ಗ್ರಾಂ ರಷ್ಟಿದೆ. ಪೌಷ್ಟಿಕಾಂಶಗಳ ಪ್ರಮಾಣ ಉತ್ತಮವಾಗಿದ್ದು ಒಟ್ಟು ಆಹಾರದ ನಾರಿನಾಂಶ (41.7 ಗ್ರಾಂ), ಖನಿಜಗಳಾದ ಕ್ಯಾಲ್ಸಿಯಂ (128 ಮಿ.ಗ್ರಾಂ), ರಂಜಕ (116 ಮಿ.ಗ್ರಾಂ) ಮತ್ತು ಮೆಗ್ನೀಸಿಯಮ್ (48 ಮಿ.ಗ್ರಾಂ) ರಷ್ಟಿದೆ ಹಾಗು ಕೊಬ್ಬು (0.18 ಗ್ರಾಂ), ಕಾರ್ಬೋಹೈಡ್ರೇಟ್ (7 ಗ್ರಾಂ) ಮತ್ತು ಶಕ್ತಿ (36 ಕೀ. ಕ್ಯಾಲೋರಿ) ರಷ್ಟಿದೆ. ಆಸ್ಕಾರ್ಬಿಕ್ ಆಮ್ಲ, ಟ್ಯಾನಿನ್‌ಗಳು ಮತ್ತು ಪಾಲಿಫಿನಾಲ್‌ಗಳಂತಹ ಸಸ್ಯಮೂಲ ಪೋಷಕಾಂಶಗಳು ಕ್ರಮವಾಗಿ 432, 524 ಮತ್ತು 677 ಮಿ.ಗ್ರಾಂ ರಷ್ಟಿದೆ. ಪ್ರತಿಶತ 6 ಮತ್ತು 10 ಪರ್ಯಾಯ ಮಟ್ಟಗಳಲ್ಲಿ ನೆಲ್ಲಿಕಾಯಿ ಶೇಷ ಪುಡಿಯೊಂದಿಗೆ ಚಿಕ್ಕಿ ಮತ್ತು ಚಟ್ನಿ ಪುಡಿ ಹೆಚ್ಚಿನ ಸಂವೇದನಾ ಅಂಕಗಳನ್ನು ಹೊಂದಿದ್ದು ಮತ್ತು ಪೌಷ್ಟಿಕಾಂಶದ ಗುಣಮಟ್ಟವನ್ನು ಸುಧಾರಿಸಿದೆ. 45 ದಿನಗಳ ಶೇಖರಣಾ ಅವಧಿಯಲ್ಲಿ ಚಿಕ್ಕಿ ಮತ್ತು ಚಟ್ನಿ ಪುಡಿಯ ಸೂಕ್ಷ್ಮಜೀವಿಗಳ ಎಣಿಕೆಯು ಅನುಮತಿಸುವ ಮಿತಿಯಲ್ಲಿತ್ತು. ಚಿಕ್ಕಿ ಮತ್ತು ಚಟ್ನಿ ಪುಡಿಯನ್ನು ಗ್ರಾಹಕರಿಗೆ ನೀಡಿದಾಗ ಉತ್ತಮ ಸ್ವೀಕಾರಾರ್ಹವೆಂದು ತಿಳಿಸಿರುತ್ತಾರೆ, ಕಚ್ಚಾ ಸಾಮಾಗ್ರಿಗಳ (ನೆಲ್ಲಿಯ ಶೇಷ) ಬೆಲೆ ಶೂನ್ಯವಾಗಿರುವುದರಿಂದ ಚಿಕ್ಕಿ ಮತ್ತು ಚಟ್ನಿ ಪುಡಿಯ ಉತ್ಪಾದನಾ ವೆಚ್ಚ ಕಡಿಮೆಯಾಗಿದೆ. ಆದ್ದರಿಂದ, ನೆಲ್ಲಿಯ ಉಪ–ಉತ್ಪನ್ನವಾದ ನೆಲ್ಲಿಯ ಶೇಷವನ್ನು ಅತ್ಯುತಮವಾಗಿ ವಿವಿಧ ಮೌಲ್ಯವರ್ಧಿತ ಉತ್ಪನ್ನಗಳ ತಯಾರಿಕೆಯಲ್ಲಿ ಬಳಸುವುದರಿಂದ ಪೌಷ್ಟಿಕಾಂಶಗಳನ್ನು ಹೆಚ್ಚು ಪರಿಣಾಮಕಾರಿಯಾಗಿ ಬಳಸಿಕೊಳ್ಳಬಹುದು.

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ಬೆಂಗಳೂರು–560 065

ಶಂಷಾದ್ ಬೇಗಂ, ಎಸ್.
(ಪ್ರಧಾನ ಸಲಹೆಗಾರರು)

Nutritional Evaluation of Amla (*Phyllanthus emblica*) Pomace and its Value Added Products

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Introduction

- Amla ('Aonla') (*Phyllanthus emblica or Emblica officinalis Gaertn*), also known as Indian Gooseberry is a minor sub-tropical deciduous tree belonging to the family Euphorbiaceae
- Amla has been considered as one of the best important Ayurvedic rejuvenative herbs, because it is tridosaghna (Bajracharya, 1979).
- Fruit and vegetable processing industries generate fruit pomace as a by-product in solid form, which contains many reusable substances of high value with large economic potential.
- Fruit pomace is a novel, effortlessly available, efficient, low-priced, natural and economic source of antioxidants and antimicrobial agents (Prakash *et al.,* 2013).
- Amla processing industries also generate amla pomace as a by-product which is rich dietary fibre and ascorbic acid. It also contains excellent source of minerals along with phytonutrients such as polyphenols and tannins.
- Due to the nutritional, nutraceutical and functional properties of the amla pomace powder, it can be used as a raw material with significant industrial potential and numerous applications in the food and biopharmaceutical industries.
- In the present study amla pomace was evaluated for nutritional and mineral composition and further value added products developed.

Objective

- To analyse nutritional, phytonutrients and mineral composition of amla pomace powder.
- To develop value added products from amla pomace powder.

Material and Methods

Procurement of amla and other ingredients from the local market

↓

Processing of amla fruits to extract amla pomace powder

↓

Analysis of amla pomace powder for nutritional, phytonutrients and mineral composition

↓

Formulation, sensory and nutrient computation computation of developed product (*chikki*)

Results

Table 1: Nutritional composition of amla pomace powder (Per 100g)

Nutrients	Content
Moisture (g)	4.99
Protein (g)	1.55
Ash (g)	2.60
Fat (g)	0.18
Crude fibre (g)	13.4
Total dietary fibre (g)	41.7
Insoluble dietary fibre (g)	27.4
Soluble dietary fibre (g)	14.5
Carbohydrate (g)	7.0
Energy (kcal)	36
Ascorbic acid (mg)	432

Table 2: Phytonutrient composition of amla pomace powder (Per 100g)

Phytonutrients	Content (mg)
Polyphenols	677
Tannins	524

Table 3: Mineral composition of amla pomace powder (Per 100g)

Minerals	Content (mg)
Calcium	128
Phosphorus	116
Sodium	92.2
Magnesium	48
Manganese	2
Copper	1.34
Iron	1.12
Zinc	0.92

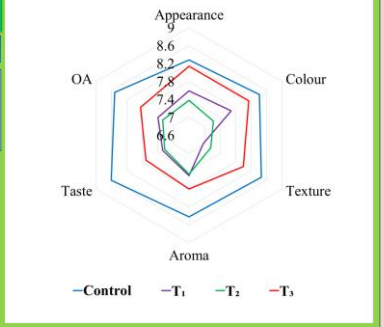

Fig. 1: Mean sensory scores of amla pomace *chikki*

Table 4: Nutritional composition of best accepted *chikki* (Per 100g)

Nutrients	Control	Best accepted product (T_3)
Protein (g)	11.6	10.2
Ash (g)	1.9	1.9
Fat (g)	17.9	15.6
Total dietary fibre (g)	4.7	6.5
Insoluble dietary fibre (g)	3.9	5
Soluble dietary fibre (g)	0.8	1.6
Carbohydrates (g)	50	50
Energy (kcal)	456	427
Ascorbic acid (mg)	00	25.9

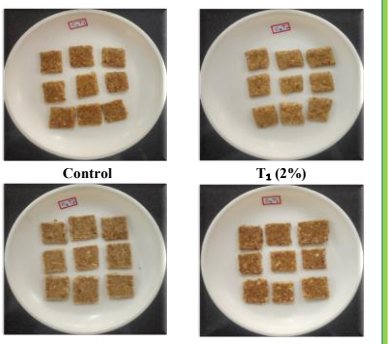

Plate 1: Amla pomace powder incorporated *chikki*

Discussion

- After dehydration 32.25g of amla pomace was obtained from 100g fresh amla pomace.
- The nutrient composition of amla pomace powder had recorded moisture 4.99 g, protein 1.55 g, ash 2.6 g, fat 0.18 g, carbohydrate 7 g and energy 36 Kcal.
- The fibre fractions of amla pomace powder indicated that the crude fibre was found to be 13.4 g/100g. The dietary fibre fractions, *viz*, total dietary fibre (TDF), insoluble dietary fibre (IDF) and soluble dietary fibre (SDF) was found to be 41.7, 27.4 and 14.5 g, respectively.
- The phytonutrients composition of amla pomace powder, *viz*, polyphenols and tannins was found to be 677 and 524 mg, respectively.
- It was noticed that amla pomace powder is a store house of minerals and calcium and phosphorous was found to be 128 and 116mg per 100g of amla pomace powder
- *Chikki* incorporated with amla pomace powder at containing 6 per cent substitutional level was found to be best accepted with good sensory attributes. And it was statistically significant at five percent level (Fig. 1)
- The nutrient composition of best accepted amla pomace *chikki* (6%) recorded 10.2 g protein, 1.9 g ash, 15.6 g fat, 6.5 g TDF, 5 g IDF, 1.6 g SDF, 427 Kcal energy, 50 g carbohydrate and 25.9 mg ascorbic acid per 100g of *chikki*.

Summary and Conclusion

- Amla pomace powder is a good source of dietary fibre and also rich in phytonutrients like ascorbic acid, total phenols and tannins.
- Amla pomace powder can be incorporated in chikki as it blends very easily without affecting the sensory parameters and hence the health benefits of amla pomace can be exploited.

Advisory committee

Chairperson : Dr. Shamshad Begum, S.

Members : Dr. K. Geetha
Dr. Kalpana, B.
Dr. A. Sathish
Dr. Srinivasappa , K. N.

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%	:	Per cent
°C	:	Degree Celsius
CD	:	Critical Difference
CHO	:	Carbohydrate
cm	:	Centimetre
et al.	:	et alibi (and associates)
g	:	Gram
hr	:	Hour
i.e.	:	That is
kcal	:	Kilo calories
kg	:	Kilogram
mg	:	Milligram
ml	:	Millilitre
µ	:	Micron
mm	:	Millimetre
No.	:	Number
pH	:	Potential of hydrogen
Rs.	:	Rupees
SE	:	Standard Error
SEM	:	Standard Error of the Mean

I INTRODUCTION

Amla or Aonla (*Phyllanthus emblica or Emblica officinalis Gaertn)* also known as Indian gooseberry or *nellikai* in kannada is a small deciduous tree and is a native of tropical Asia. It is found growing wild in India's tropical woods and on hill slopes upto 1500 metres above sea level. It is a resilient plant that grows quickly and requires little maintenance, making it an ideal tree for arid climates (Kumar *et al.,* 2014). Amla is traditionally used as an important herbal medicine in south east Asia and known for its strong antioxidant activity. Amla has been cultivated in India since time immemorial. It is described in the Vedas, Ramayan, Charaksamhita, Sushruta Samhita, Kalidas and Kadambari literature and holds a special place in Indian mythology (Nagamaniammai, 2013).

With respect to the production, India produces 1074 MT of amla. Uttarpradesh is the most important amla growing state in India, followed by Madhya Pradesh, Gujarat, Tamil Nadu and Rajasthan, accounting for 75.88 per cent of total amla output. Amla farming is widely practised in Andhra Pradesh, Karnataka and Maharashtra, and it is predicted that this crop would cover an increasing amount of land. Its economic feasibility has been demonstrated even on marginal and degraded lands (Anonymous, 2017).

Scientific classification of Amla

Kingdom	Plantae
Division	Magnoliophyte
Class	Magnoliopsida
Order	Malpighiales
Family	Phyllanthaceae
Scientific name	*Phyllanthus emblica*
Synonyms	*Emblica officinalis Gaertn.*
Genus	*Phyllanthus L*
Species	*P. emblica*

Source: https://www.scribd.com/document/12791801/Indian-Gooseberry

Amla fruits are fleshy, spherical, attractive, deeply ribbed, yellowish-green in colour and have six vague perpendicular furrows enclosing seeds. The amla fruit is well known across the world for its nutritional, commercial and medicinal benefits. Amla is a rich source of ascorbic acid, amino acids, minerals and phytochemicals such as polyphenols, tannins, emblicol, linoleic acid, corilagin, phyllemblin and rutin. It has 89 to 94 per cent pulp, 0.8 to 2 per cent fibre, 10 to 14 per cent total soluble solids, 1.4 to 2.4 acidity, 700 to 900 mg vitamin C per 100g, 2.4 to 3.1 per cent pectin and 2 to 3 per cent phenols make up the amla fruit (Parveen and Khatkar, 2015).

Amla has been utilised in Ayurveda for centuries, and its therapeutic characteristics are categorised as follows: Rasa (taste): the most prominent flavours are sour and astringent, although the fruit has five flavours, including bitter, pungent and sweet, Veeryav (nature): refreshing, Vipaka (digestive taste): sweet, Guna (characteristics): light, dry, Doshas (impact on humours): calms all three doshas: pita, kapha and vata with a specific emphasis on pitta. Amla is a staple constituent in therapies for a tingling pain throughout the body, as well as many forms of inflammation and fever; these are indications of pitta (fire) agitation (Williamson, 2002). Amla contains tridosaghna, so it is regarded as the greatest Ayurvedic rejuvenative fruit. It is the only fruit that has a natural balance of flavours like sweet, sour, pungent, astringent and bitter that is present all in one fruit, and it provoke the brain to rebalance the three fundamental components of all physiological activities, the air, fire and water elements within the body (Bajracharya, 1979).

In India, the amla fruit is prized for its medicinal properties. Fruit is acrid, cooling, refrigerant, diuretic and laxative, and as a result, it's utilised to cure a variety of ailments such as chronic dysentery, bronchitis, diabetes, fever, diarrhoea, jaundice, dyspepsia, cough and others. The dried fruits of amla, which are high in gallic acid, are a definite treatment for blood dysentery, piles and illnesses connected with piles and blood (Singh *et al.,* 2016).

Amla is a fruit that is used to make a range of unique products. The sour and astringent amla fruit is commonly used raw or cooked in the form of Murrabas, juice, squash, jam, pickles and *chutney* (Shamshad and Suresha, 2016). Many ayurvedic

medicines, such as Chyawanprash and Triphala, contain amla as one of the main ingredients (Pant *et al.*, 2004; Goyal *et al.*, 2007 and Mishra *et al.*, 2009).

In the manufacturing process of amla products, sound mature amla fruits will be washed and subjected to cutting followed by pulping with filtration, standardizing and pasteurizing for storage or otherwise can be used for making other value added food products either by boiling the pulp or sometime direct cooking of amla fruits without pulping for making products like amla preserve, candy, pickle or mouth freshener. The pulp or extract can be used in making Chyawanprash, juices, ready-to-serve beverages, fruit bar, amla sauce, cosmetic products, etc. During filtration of Amla juice lots of Amla residue or pomace will be produced.

It is also observed that fruits like amla, mango, banana, watermelon and citrus fruits all leave behind a large amount of residue in the form of peels, pulp, seeds and stones. This fruit residue or pomace has lot of potential in the culinary, pharmaceutical and cosmetic sectors because of the presence of phenolic compounds, which imparts nutraceutical properties. The extraction of bioactive compounds from less expensive or residual sources has become researcher's curiosity in the recent years (Gupta *et al.*, 2012). Fruit pomace is a novel, effortlessly available, efficient, low-priced, natural and economic source of antioxidants and antimicrobial agents (Prakash *et al.*, 2013).

The present research focuses on Amla pomace as it is having nutritional, nutraceutical and functional properties that can address significant commercial capacity and various uses as nutraceuticals in the food and biopharmaceutical sectors. Various dehydration technologies can be used to convert amla pomace into powders, which can then be used as a food supplement or as an ingredient in daily dietary or to enrich bakery products like cakes and biscuits too (Shamshad and Suresha, 2016). Also, amla pomace is known to have good antioxidant potential and functional properties like water holding capacity (WHC), swelling capacity (SC), oil holding capacity (OHC), bulk, hydrated and packed density (Ajay *et al.*, 2017).

Hence, keeping in mind to exploit the nutritional potential and to utilize amla pomace in diversified products, the present investigation had been focused for the

"Nutritional evaluation of Amla (*Phyllanthus emblica*) pomace and its value added products" with the following **objectives**, which are mostly focused on the dietary and health benefits of amla pomace.

- To study the physico-functional properties of amla pomace.
- To analyse nutritional composition of amla pomace and to develop its value added products.
- To conduct organoleptic evaluation and shelf-life study of the developed products.

II REVIEW OF LITERATURE

Fruits and vegetables contain a range of nutrients like vitamins, minerals and antioxidants. Also, the by-products of fruit and vegetable are with a good nutritional profile. Processing of fruits and vegetables leads to a build-up of high amounts of waste like peels, seeds, leaves, residues etc. On one hand, valuable nutrients, antioxidants, phytochemicals contained in agroindustry waste are lost and also contributes to environmental pollution, whereas, on the other hand the population is suffering from micronutrient deficiency and non-communicable diseases or lifestyle disorders that are usually preventable through diet.

Therefore, recycling of the by-products of fruit and vegetable processing industries will also be another added source of nutrition and also income to industries that may also help to extend the economic productivity. The literature pertaining to the production of fruits, physico-chemical characteristics of their edible waste, nutritional composition and utilization of edible waste for product development is reviewed in this chapter under the following headings:

2.1 Production of fruits

2.2 Processing and utilization of fruit and vegetable by-products

2.3 Physico-functional properties of fruit and vegetable by-products

2.4 Nutritional composition of fruit and vegetable by-products

2.5 Development of value added products and their sensory evaluation

2.6 Shelf-life studies of value added products

2.1 Production of fruits

Fruits and vegetables account for nearly 90 per cent of total horticulture production in the country. India is now the second-largest producer of fruits and vegetables in the world and is the leader in several horticultural crops, namely mango,

banana, papaya, cashew-nuts, areca nut, potato and okra. Table 1 lists the world's leading fruit-producing countries (Anonymous, 2018a).

During 2017-18, the production of horticulture crops was 311.71 million tonnes which includes 184.40 million tonnes of vegetables and 97.35 million tonnes of fruits from an area of 25.43 million hectares. The vast production base offers India tremendous opportunities for export. During 2019-20, India exported fruits and vegetables worth Rs. 9,182.88 crores which comprised of fruits worth Rs. 4,832.81 crores and vegetables worth Rs. 4,350.13 crores. Because of the short shelf life of those crops, as much as 30-35 per cent of fruits perishing throughout the stages of harvesting, storage, grading, transportation, packaging and distribution. Although India has a strong raw material base, it has been unable to tap the potential for processing and value addition to fruits. Only about two per cent of the fruits were processed into value added products that are far lower compared to other countries. Hence, there is a desire for optimum commercial utilization of fruits and to adopt production and marketing activities to the requirements of the world market to cater to domestic demand that, over the past few years, has been increasing because of various socio-economic factors. If the nutritive value of the processed food products is maintained, this sector can emerge as a significant value added food business (Anonymous, 2018b).

Table 1: Major fruits producing countries in the world (2016)

Sl. No.	Country	Area (Million Ha)	Production (MT)	Productivity (T/Ha)
1	China	16.59	272.08	16.40
2	India	6.98	90.89	13.03
3	Brazil	2.33	39.69	17.02
4	USA	1.19	27.11	22.73
5	Mexico	1.42	21.43	15.13
6	Spain	1.63	19.05	11.66
7	Indonesia	0.77	18.52	24.17
8	Philippines	1.68	16.32	9.72
9	Italy	1.14	18.00	15.79
10	Turkey	1.38	21.74	15.79
11	Others	28.60	320.62	11.21
World Total		65.24	865.88	13.27

India's position in the production of amla in the world stands at first position with the production of 1074.60 MT, followed by Philippines, Brazil, Sri Lanka and Mexico. In India, Uttar Pradesh is the highest amla producing state with the production of 384.32 MT followed by Madhya Pradesh, Maharashtra, Gujarat, Andhra Pradesh and Chhattisgarh. Table 2 indicates the state-wise area and production of amla in India (Anonymous, 2017).

Table 2: State-wise area and production of amla in India

Selected State-wise Area and Production of Amla in India (2017-2018)		
States	**Area (In ' 000 Hectare)**	**Production (In ' 000 MT)**
Andhra Pradesh	0.64	10.76
Assam	0.91	17.76
Bihar	1.59	14.92
Chhattisgarh	3.80	43.29
Gujarat	8.15	81.90
Haryana	2.24	10.75
Himachal Pradesh	2.56	1.97
Jammu and Kashmir	1.96	12.10
Jharkhand	0.30	1.49
Karnataka	0.11	0.68
Kerala	0.10	0.10
Madhya Pradesh	20.42	302.18
Maharashtra	1.60	12.25
Mizoram	0.30	1.32
Nagaland	0.27	2.88
Odisha	2.03	0.73
Punjab	0.56	7.70
Rajasthan	1.57	11.19
Tamil Nadu	7.44	152.87
Telangana	0.07	0.71
Uttar Pradesh	35.16	384.32
Uttarakhand	1.33	2.65
Others	0.20	0.10
Total	93.12	1074.60

2.2 Processing and utilization of fruits and vegetable by-products

John and Narasimham (1993) developed a technique for making a clarified juice and a ready-to-serve (RTS) beverage from the by-product of Jack fruit. Treatment of the jackfruit waste with the pectic enzyme at 0.3 per cent concentration (v/w), incubation for 2 h at 40^0 C, and subsequent filtration, giving about 60 per cent yield of clarified juice having 23° Brix and 0.15 to 0.20 per cent acidity. Sensory evaluation of ready to serve (RTS) beverages (12 % juice, 15° Brix sugars, and 0.3 % acidity) without and with carbonation at 3 levels (CO2 gas pressures 0. 775, 2.092 and 3.685 kg/cm2) by 15 trained panel members revealed that the product was highly acceptable either without or with carbonation at 0.775 kg/cm2, compared to higher levels of carbonation. They concluded that the preparation of beverages from jackfruit waste as a by-product brings about the effective utilization of jackfruit to over 80 per cent.

Shui and Leong (2006) determined antioxidant capacity, total phenolic contribution and possibilities of utilizing the star fruit residue as a valuable food ingredient. The results revealed total polyphenolic content of 33.2 ± 3.6 mg gallic acid equivalent (GAE)/g sample, total antioxidant activity of 3490 ± 310 and 3412 ± 290 mg L-ascorbic acid equivalent antioxidant capability (AEAC) and 510.3 ± 68.1 mol ferric reducing/antioxidant power (FRAP) per gram sample. The high content of phenolics and strong antioxidant activity of residue extracts indicate that residue powder might impart health benefits when utilized in functional food products and its residue extracts should even be considered nutraceutical resources in the future.

Ajila *et al.* (2007) examined valuable bioactive compounds such as polyphenols, carotenoids, dietary fibres and enzymes in the raw and ripe peels of two Indian mango varieties *viz*; Badami (Alphonso) and Raspuri. Results showed that polyphenol content in peels ranged from 55 to 110 mg/g dry peel, dietary fibre content ranged from 45 to 78 per cent of peel and was found at a higher level in ripe peels. Similarly, carotenoid content was higher in ripe fruit peels. Vitamins C and E contents ranged from 188 to 392 and 205 to 509 µg/g dry peel respectively. These were found at a higher level in ripe peels. Both raw and ripe mango peels exhibited a significant amount of protease, peroxidase, polyphenol oxidase, xylanase and amylase activities.

Ayala *et al.* (2011) reviewed the agro-industrial potential of exotic fruit by-products as a source of natural antioxidants and highlighted that tropical exotic fruits are rich in bioactive compounds, such as phenolic constituents, carotenoids, vitamins and dietary fibre. They inferred that the fruit processing industry deals with a large percentage of by-products, such as peels, seeds and unused flesh, generated in the different steps of the processing chains, and in most cases, the waste by-products can present similar or even higher contents of bioactive compounds than the final product.

Gupta *et al.* (2012) studied the nutritional constituent of six different tropical fruit residues. Proximates and mineral contents were analyzed by using standard methods. It was found that all residues contained a considerable amount of protein ranging between 1.05 per cent (*C. carandas*) and 11.25 per cent (*B. vulgaris*). Carbohydrate content was found to be the highest in *L. sinensis* seed (67.3 %). Results of the mineral analysis showed that all residues were rich in Ca, K, Mg, Na, and P content. The dietary fibre was found to be different for each residue, which ranged from 1.6 per cent (*L. sinensis*) to 39.10 per cent (*A. comosus*). This study demonstrated that all these residues possessed a significant amount of protein, carbohydrate, minerals, dietary fibre and fatty acids and have a potential to be used as functional ingredient in food formulation.

Aslam *et al.* (2014) examined the effect of Mango Peels Powder (MPP) and Mango Kernels Powder (MKP) at different replacing levels (5, 10, and 15 %) separately on rheological, proximate, physical, sensory and antioxidant properties of biscuits. The results demonstrated that mango peel powder had high contents of crude fibre and antioxidant activity whereas mango kernel powder was characterized by higher protein, total phenolic, and ash contents as compared to mango peel powder. Farinograph study of composite flour of MPP and MKP revealed an increase in water absorption (WA) from 60 to 69.8 per cent. The crude fibre contents of biscuits were improved from 0.22-16.79 per cent by the addition of mango peel and kernel powder. Their phenolic contents increased from 0.43 to 10.28 mg/g. The biscuits incorporated with MKP and MPP powder showed an increment in their antioxidant activity. Sensory performance exhibited that the biscuit acceptable with mango taste and flavour were obtained by substitution up to 10 per cent mango peel powder and up to 5 per cent with mango kernel powder.

Singh and Immanuel (2014) explored the extraction of antioxidants like phenols and flavonoids from fruit peels like pomegranate, lemon and orange peels and found that pomegranate exhibited a high percentage of antioxidant activity and phenolic content of 92.7, and 249.41 mg/g in comparison to lemon and orange-peel extract. They inferred that synthetic antioxidants like Butylated hydroxytoluene (BHT) and Butylated hydroxyanisole (BHA) are toxic and may cause health hazards. Hence, natural antioxidants extracted from fruit peels like pomegranate, lemon and orange could be a better alternative as an additive to any food products containing fat and oil to increase their shelf life by preventing rancidity.

Ferreira *et al.* (2015) assessed functional properties, proximate composition, microbiological stability and consumer acceptability of the developed products from fruit and vegetable residue (FVR) flour generated from the manufacturing of an isotonic beverages. The results revealed that FVR flour was incorporated with different levels (20 to 35per cent) into biscuits and cereal bars. The FVR flour presented a higher water holding capacity (7.4 g/100g) than oil holding capacity (1.9 g/100g) of flour, probably associated with its high levels of carbohydrates (53 %) and fibre (21.5 %). Biscuits enriched with 35 per cent of FVR flour presented significantly higher fibre ranging from 57 to 118 per cent and mineral content from 25 to 37 per cent than when only 20 per cent was added. Cereal bars presented about 75 per cent of fibre and variable mineral contents between 14 and 37 per cent.

Gowe (2015) reviewed about by-products from various fruits and vegetables, processing of banana peel and blossom and its application in food products, mango-peel and stones (45%), banana-peel (35%), citrus-peel, rag and seed (50%), pineapple-skin and core (33%), grapes- stem, skin and seeds (20%), guavas peel and core and seeds (10%), tomato skin, core and seeds (20%), potato peel (15%). The chemical composition indicated that Apple pomace contained 16 per cent fibre, mango seed kernel was rich in protein (8.5%), moisture (64.5%) and protein (6.6%) content were more in jackfruit seeds, passion fruit and banana peel were rich in moisture (81.9 and 79.2%), pumpkin seeds and watermelon seeds were protein-dense sources (29.5 and 34.1%). Because of their high nutritional value and immense functional properties, these by-products can be

used as different food additives like anti-browning additives, antimicrobial and flavouring compounds, the source of colourants, dietary fibre and protein.

Egbuonu (2015) assessed the antinutrient properties of the by-products of watermelon rind and seed. Saponin (3.0±0.03, 2.31±0.01), an alkaloid (1.39±0.00, 0.36±1.03), tannins (1.33±0.01, 0.61±0.01), phenol (0.53±0.00, 0.12±0.01), and flavonoid (2.87±0.00, 2.03±0.02) were found higher in the rind than in the seed. The content (mg/100 g) in the seed for cyanide (0.79±0.01), phytate (0.63±1.00) and oxalate (0.09±0.00) was higher than that in the rind for cyanide (0.00±0.00), phytate (0.46±0.00) and oxalate (0.08±0.01). The recorded difference in the antinutrients content in the rind and seed samples was not significant ($p>0.05$), hence negligible. The author suggested that the watermelon rind and seed may offer pharmacologic and dietary benefits at a possibly lower toxic risk and also indicated further utilization in diets and drugs are required to reduce their waste burden in the environment.

Bishnoi *et al.* (2018) assessed the active chemical constituents of Shatavari, *viz*, Sarsapogenin, Saponins A-4 to A-7, Shatavarin I to IV, Sitosterol, Glycosides of quercetin, Stigmasterol, Asparagamine A, and Sitosterol 7. Further, Shatavari roots were processed to prepare powder and herbal aonla ladoo were developed using Shatavari powder at 2, 4 and 6 per cent substitutional levels. Based on sensory analysis, herbal aonla ladoo using Shatavari powder (4%) was found most acceptable. Herbal aonla ladoo containing Shatavari powder (4%) had the moisture content (35.3%), ascorbic acid (257 mg/100 g), total phenols (1.99 mg/g) and the extent of non-enzymatic browning was recorded to be 0.13.

2.3 Physico-functional properties of fruit and vegetable by-products.

Robertson *et al.* (2000) assessed the hydration properties of resistant starches, pea hull, citrus pulp and apple pulp. Results showed swelling capacity (7 mL/g) and water retention capacity (4 g/g) were relatively low for resistant starches and pea hull. Swelling and water retention capacity of citrus (11 mL/g and 11 g/g), apple pulp (7 mL/g and 5 g/g) were lower.

Chau and Huang (2003) assessed the fibre-rich fractions (FRFs) including soluble and insoluble dietary fibres (SDF and IDF), alcohol-insoluble solid (AIS) and water-insoluble solids (WIS) from the peel of *Citrus sinensis L*. The peel was rich in insoluble FRFs (IDF, AIS, and WIS; 476−515 g/kg of peel), which were mainly composed of pectic substances and cellulose and also contained pectic polysaccharide-rich SDF (94.1 g/kg of peel). These insoluble FRFs had water-holding capacities (15.5−16.7 ml/g) and swelling properties (14.6−21.1 ml/g) much higher than those of cellulose.

Sangnark and Noomhorm (2003) conducted study about the alkaline hydrogen peroxide (AHP) treatment of sugarcane bagasse (SB) and found that it affected physical and chemical properties. The brightness, water-holding capacity (WHC) and oil-binding capacity (OBC) of SB were increased by 34, 96 and 55 per cent, respectively. Lignin was removed from SB by 53 per cent. The Colour of Solka Floc1 900, a commercial dietary fibre, was pure white (L=93.51); WHC and OBC were 8.61 g water/g sample and 7.34g oil/g sample, respectively. The results showed a highly positive correlation between particle size reduction of each dietary fibre (DF), WHC and OBC. While density showed a negative correlation, densities of AHP-SB and Solka Floc1 900 were increased, with particle size reduction from 1.06 to 1.34 and 1.09 to 1.28, respectively.

Betancur *et al.* (2004) explored the physicochemical, physiological and functional characteristics of Jack bean (*Canavalia ensiformis*) and lima bean (*Phaseolus lunatus*) fibrous residues that are obtained as a by-product during the protein and starch extraction process. The jackbean residues had higher crude fibre (22.6%), total dietary fibre (55.8%), and insoluble dietary fibre (52.4%) contents than the limabean (crude fibre 12.8%, total dietary fibre 29.4%, insoluble dietary fibre 28.6%). Water-holding capacities in both legume fibrous residues were higher for (*C. ensiformis* 39.5% and *P. lunatus* 26.5%) than their oil-holding capacities (23 and 18%, respectively). Similar antioxidant activity values were obtained for the Jack bean (39.4%) and lima bean (35.6%) residues.

Sowbhagya *et al.* (2007) evaluated the spent residue from cumin as a new source of dietary fibre and for physiochemical characteristics. Results revealed that spent residue from cumin contains 62.1 per cent TDF, 51.7 per cent IDF and 10.4 per cent SDF. The spent residue also contained 7.7 per cent starch and 5 per cent bound fat. The spent

residue exhibited 3.3 g/g water holding capacity, 4.0 g/g water retention capacity and 4.47 ml/g swelling capacity.

Mei *et al.* (2010) studied the proximate composition, chemical composition and physic-chemical properties of dietary fibre (DF) extracted from sweet potato residues after starch isolation of 10 varieties using a sieving method. The average yield and dietary fibre (DF) content of 10 sweet potato varieties were 9.97 and 75.19 per cent, respectively. Average contents of cellulose, lignin, pectin and hemicellulose were 31.19, 16.85, 15.65 and 11.38 g/100g of dry matter in dietary fibre products, respectively. Swelling capacity and water-holding capacity of the dietary fibre of sweet potato varieties had respective ranges of 8.11-12.56 ml/g and 3.54-6.15 g/g.

Sowbhagya *et al.* (2011) assessed the physicochemical and dietary fibre characteristics of celery seed spent residue(CSR) and the results revealed that it had 61 per cent total dietary fibre (TDF), 53.5 per cent insoluble dietary fibre (IDF), 7.5 per cent soluble dietary fibre (SDF), 19 per cent protein, 7.9 per cent starch and 5 per cent fat. The hydration properties of fibre increased with decrease in particle size of CSR. The CSR exhibited 6.8 g/g water-holding capacity, 6.0 g/g water-retention capacity and 5.2 ml/g swelling capacity.

Gupta and Premavalli (2010) studied the effect of particle size reduction on the physic-chemical properties of ash gourd and radish fibre. Both the fibres after extraction of juice were dehydrated and subjected to granulometry to obtain 30, 60, and 100 mesh particles. The yield was more for 60 mesh particles. The 30 mesh particles of ash gourd and radish fibre contained 66.35 per cent and 53.11 per cent total dietary fibre while the soluble fibre content was 22.76 per cent and 17.75 per cent, respectively. The fibre content decreased by 7-8 per cent with a decrease in particle sizes. The values of water holding capacity, water binding capacity, oil binding capacity, swelling power and cation exchange capacity also decreased with the decrease in particle size. However, the particle density values increased with the decrease in particle size.

Mei *et al.* (2010) investigated the physico-functional properties of dietary fibre (DF) extracted from sweet potato residues after starch isolation of 10 varieties by sieving

method. Swelling capacity, water-holding capacity, oil-holding capacity, and glucose absorption capacity determinations of the DF of sweet potato varieties had respective ranges of 8.11-12.56 ml/g, 3.54-6.15 g/g, 1.43-2.48 g/g, and 0.54-1.27 mmol/g.

Sowbhagya *et al.* (2011) assessed that celery seed had a total dietary fibre (TDF) of 56 per cent, insoluble dietary fibre (IDF) 49 per cent, soluble dietary fibre (SDF) 7 per cent, while celery spent residue (CSR) after oil and oleoresin extraction contained 61 per cent TDF, 53.5 per cent IDF, 7.5 per cent SDF, 19 per cent protein, 7.9 per cent starch and 5 per cent fat. The hydration properties of fibre increased with decrease in particle size of CSR. The CSR exhibited 6.8 g/g water-holding capacity, 6.0 g/g water-retention capacity and 5.2 ml/g swelling capacity.

Rabetafika *et al.* (2014) examined the influence of desugaring, grinding and bleaching on the compositions, physicochemical properties and the colour of dietary fibre (DF) of fibre concentrates from unusual cooked apple and pear pomaces. All the processing conditions affected the compositions and physicochemical properties of DF. The bleaching induced the greatest changes on DF producing yellow cellulose-rich fibre concentrates with improved water-holding capacity from 3.2 to 10.0 g/g and improved swelling capacity from 4.0 to 8.8 ml/g. water-holding capacity and swelling capacity tended to increase with the particle size whereas smaller granulometric sizes increased the lightness of fibres. Results showed that processing had overall positive effects on DF contents and hydration properties of pomaces from cooked fruits. Bleached fibre concentrates from apple pomace had the highest water-holding capacity (10.0 g/g) whereas that of pear had the highest fibre content (89.9%).

Huang *et al.* (2021) evaluated the pectin-rich dietary fibres of citrus peel through alkaline hydrogen peroxide treatment (AHP-CF) or homogenization treatment (H-CF) for physicochemical properties and compared with a commercial one (Fibrestar). Results showed that the AHP-CF had the highest water-holding capacity (WHC) (21.32 g/g), water-swelling capacity (WSC) (32.70 mL/g) and cation exchange capacity (CEC) (1.02 Meq/g) among the fibres. Besides, homogenization treatment (H-CF) had a similar chemical composition, oil-holding capacity, and CEC with Fibrestar but higher water-holding capacity, water-swelling capacity and thermal stability.

2.4 Nutritional composition of fruit by-products

El Kossori *et al.* (1998) investigated the proximate composition of pulp, skin and seeds of prickly pear cactus (*Opuntia ficus indica*). Results of proximate composition indicated the protein content of 5.1 per cent (pulp), 8.3 per cent (skin) and 11.8 per cent (seeds). Starch was found in each of the three parts of the fruit. The skin was remarkable for calcium (2.09%) and potassium (3.4%). Prickly pear is a neglected nutritional source that should be more widely used because of its potential nutrient contribution.

Anhwange *et al.* (2008) analyzed minerals, nutritional and anti-nutritional contents of *Musa sapientum* peels. The result of mineral content indicates the concentrations (mg/g) of potassium, calcium, sodium, iron, manganese, bromine, rubidium, strontium, zirconium and niobium to be 78.10, 19.20, 24.30, 0.61, 76.20, 0.04, 0.21, 0.03, 0.02 and 0.02, respectively. The percentage concentrations of protein, crude lipid, carbohydrate and crude fibre were 0.90, 1.70, 59.00 and 31.70, respectively. The results indicated that if the peels are properly exploited and processed, they could be a high-quality and cheap source of carbohydrates and minerals.

Ewansiha *et al.* (2011) studied the proximate and mineral composition of seed shells pericap of *Chrysophyllum albidum*. The results revealed that moisture content 12.90 per cent, ash 1.27 per cent, crude fibre 14.82 per cent, crude fats 2.38 per cent, protein 0.98 per cent and carbohydrates 67.65 per cent, while the mineral composition determined using standard analytical tools were measured in mg/100g *viz:* P = 10.80, Ca = 31.04, Mg = 0.61, Na = 0.36, K = 2.30, Fe = 35.20, Zn = 100, and Mn = 0.40.

Akpabio *et al.* (2012) analyzed the proximate composition and mineral elements in the peel, pulp and seeds of African star apple fruit. The results showed that the pulp contains a greater amount of crude fibre (3%), fat (10%), ash (3.25%), caloric values and a greater amount of moisture (47.95%) found in the peels. Carbohydrate content (83.38%) and crude protein (8.75%) were higher in the seed. Mineral elements revealed that pulp has a greater amount of sodium (69.38mg/kg) and iron (40.11mg/kg) while peel contains a greater amount of potassium (62.26mg/kg) and zinc (13.46mg/kg). Calcium (71.52mg/kg) and magnesium (29.46mg/kg) were higher in the seed.

The proximate composition and minerals contents of the most popular consumed fruit Pomegranate (*Punica granatum L*) peels indicated that the moisture content (04 ± 0.22%), ash (05 ± 0.14%), fat (9.4 ± 0.1%), pH (3.75 ± 0.2), crude fibre (21 ± 0.6%) and protein (8.719± 0.10%). The minerals like sodium, potassium, iron, manganese and zinc ppm values were 1100 ±0. 4, 10000 ±0. 6, 60.5 ± 0.2, 4.5 ± 0.8 and 4.0 ± 0.65 respectively (Ullah *et al.*, 2012).

Oikeh *et al.* (2013*)* evaluated the proximate composition and phytochemicals present in the fruit wastes (flavedo, albedo and seeds). Proximate analysis showed that the flavedo and seeds are rich sources of oil, containing 10.00 ± 0.58 and 12.00 ±1.15 per cent respectively. Ash content ranged from 0.85 ± 0.03 per cent in the albedo to 2.00 ± 0.06 per cent in the seeds. The albedo had the highest moisture and nitrogen-free extract contents (15.00 ± 0.58 per cent and 78.22 ± 0.58per cent) respectively with the lowest protein, ash, and lipid contents. The flavedo had the highest fibre content (13.43 ± 0.03%) while the seeds had the highest protein content (6.13 ± 0.51%). Tannins were highly detected in the flavedo extracts and mildly in all other extracts except the ethanolic seed extract.

Fila *et al.* (2013) evaluated the nutritional quality contents of the pulp, seeds and rind of *Citrullus lanatus*, *Nephelium lappaceum* and *Cucur bitapepo L*. The study was carried out on both fresh and dried samples. Results of the investigation revealed that the seeds of the fruits were all rich in oil, protein. Although there was a significant change ($p< 0.05$) in the nutrient contents *i.e.* protein, carbohydrate, crude fat, crude protein, moisture and ash content in the different parts of the fruits, the nutrients in the seeds and rind which are the parts always discarded, can contribute immensely to recommended daily allowance and maintenance of good nutritional status and hence good health for both man and livestock.

Gazalli *et al.* (2014) analyzed the proximate composition of Apple pomace obtained as a main by-product of the apple juice industry. It consists of 25-35 per cent of the dry mass of the apple, 7.31-8.53 per cent crude protein, 2.6-3.33 per cent fat, 19.34-20.66 per cent crude fibre, 3.85-4.7 per cent total ash and 46.4-49 per cent neutral detergent fibre.

Romelle *et al.* (2016) examined the chemical composition of peels of eight fresh fruits (orange, watermelon, apple, pomegranate, pawpaw, banana, pineapple, and mango) and the results revealed that lipid, protein, ash, crude fibre and carbohydrates contents in fruit peels varied from 3.36 ± 0.37 to 12.61 ± 0.63per cent, 2.80 ± 0.17 to 18.96 ± 0.92per cent, 1.39 ± 0.14 to 12.45 ± 0.38per cent, 11.81 ± 0.06 to 26.31 ± 0.01per cent and 32.16 ± 1.22 to 63.80 ± 0.16 per cent respectively. The minerals composition varied from 8.30 ± 0.54 to 162.03 ± 7.54 mg/100g for calcium, 0.66 ± 0.06 to 6.84 ± 0.55 mg/100g for zinc, 9.22 ± 0.63 to 45.58 ± 2.37 mg/100g for iron and 0.52 ± 0.10 to 9.05 ± 0.34 mg/100g for manganese. The phenolic content of fruit peels ranged from 0.91 ± 0.06 to 24.06 ± 0.89 per cent and can be used as good ingredients in the formulation of health benefits food products.

Morais *et al.* (2017) evaluated the proximate composition of peel, pulp and seeds of seven tropical fruits. Pulp and peel showed the highest moisture values (65.7-93.3%), while the seed ranged from 5.8 to 67.2 per cent. The drying of peels decreased moisture values (2.3-18.7%) that did not affect ash contents, total crude protein, lipids, fibre values and fatty acid composition for avocado, pineapple, banana, papaya, passion fruit, watermelon and melon. A wide range of mineral contents was noted in different parts of fruit. Calcium and potassium were found in larger quantities (25.4 to 4808 mg/100g). The largest contents of essential fatty acids like omega-6 and omega-3 were found to be 31.4 to 1970 mg/100 g in the peels and seeds.

2.5 Development of value added products and their sensory evaluation

Youssef and Mousa (2012) studied the proximate composition, physical characteristics and sensory quality attributes of wheat biscuits and wheat biscuits fortified with citrus peel powder. The sensory attributes were good and revealed that 10 per cent incorporation of citrus peel powder enhanced the nutritive value in wheat biscuits with increased crude protein, crude fat contents as well as crude fibre, moisture content and caloric value. However, it decreased carbohydrate content.

Feili *et al.* (2013) developed jackfruit-based high fibre bread by utilizing jackfruit rind flour in bread formulation and to characterize physical properties of produced high

fibre bread. Jackfruit rind flour (JRF) was incorporated into wheat flour (WF) in three different ratios (5, 10 and 15%) to produce partially substituted wheat flour (WF) with JRF. Increased level of JRF incorporated into WF caused an increase in hardness and darkness of bread samples and a decrease in their volume compared to the control. Bread samples substituted with 5 per cent JRF had the highest mean scores of overall acceptances.

Bertagnolli *et al.* (2014) developed cookies by using different amounts of guava peel flour (GPF) levels (30, 50 and 70 per cent) to evaluate the proximate composition, phenolic compound, lycopene and β-carotene levels in the cookies and flour and to evaluate the sensory acceptance. The results demonstrated low moisture, lipid and carbohydrate contents in the flour and cookies. GPF was considered rich in fibre, ash, polyphenols and β-carotene. The sensory analysis showed satisfactory acceptance of the cookies containing 30 per cent GPF regarding the aroma, flavour and texture attributes. The cookies containing 50 per cent and 70 per cent GPF received satisfactory acceptance regarding aroma only. Hence it was concluded that partial replacement of wheat flour in cookies improved the nutritional quality without affecting the products sensory quality.

Hirdyani and Charak (2015) evaluated the nutritional and sensory parameters of *chikki*, where flaxseeds are utilized to replace peanuts in the traditional peanut *chikki* at 50 per cent and 100 per cent. The incorporation of flaxseed increased the nutritional quality of *chikki* significantly especially in fat (6%), fibre (1%) and protein content (2.9%) and storage studies (30 days) revealed that the addition of flaxseed increased the peroxide value of samples during storage (~12 meq/kg), thereby making the samples more prone to rancidity. The organoleptic evaluation revealed that flaxseed incorporated *chikki* acceptable by panelists and inclusion of such fortified common daily use snacks will help the community to maintain a healthy life.

Shamshad and Suresha (2016) incorporated amla residue in the preparation of ethnic products like chapati, poushtik roti and baked products like biscuits, cookies and cakes and the sensory attributes were found to be adequate with respect to appearance, taste, texture, flavour and overall acceptability.

Zaker *et al.* (2016) incorporated orange pomace powder in various proportions *viz.*, 0, 5, 10, 15 and 20 per cent levels in cookies by replacing the maida. The results revealed that the sensory attributes of cookies prepared with 10 per cent orange pomace powder were recorded with higher acceptability as compared to other samples. The spread ratio of cookies decreased as the per cent of orange pomace was increased, with the increase in powder concentration the protein, fat content was gradually decreased and the dietary fibre increased. Hence, it was concluded that orange pomace powder can be substituted up to 10 per cent in wheat flour in cookies without adversely affecting overall quality attributes.

Ho (2016) investigated the nutritional composition and sensory quality of the control and wheat flour cookies substituted with 5, 10 and 15 per cent pitaya peel flour (PPF). The results revealed that PPF-containing cookies had significantly higher ash, fibre and carbohydrate content but lower moisture and protein than the control. Physical analyses showed that cookies incorporated with PPF had a higher diameter and spread ratio but lower crumb height than the control. Sensory evaluation revealed that PPF up to 15 per cent level did not affect nutritional quality and overall acceptability ratings of cookies by panelists.

Makanjuola *et al.* (2018) examined the proximate composition of seeds and skin of ripe matured papaya (*Carica papaya*) and the results revealed crude protein of 27.42 per cent for seeds and 14.36 per cent for skin. Ash content of 5.22 per cent (seeds), 11.03 per cent (skin), crude fibre content of 8.02 was present in the seeds while 35.23 per cent present in the skin. The carbohydrate contents for both seeds and skin were 19.71 and 37.33 per cent respectively. The selected minerals revealed 6.43mg/100g and 16.23mg/100g calcium, 720.83mg/100g and 504.33mg/100g potassium, 4.21mg/100g, and 2.73mg/100g iron while 6.42mg/100g and 1.94mg/100g zinc were present in papaya seeds and skin respectively.

Kohli *et al.* (2019) conducted a study to prepare fibre and vitamin-C rich biscuits by inclusion of amla pomace and the results revealed that the maximum fibre was obtained for biscuits prepared from 15 per cent amla pomace. The fat content of biscuits varied from 14.03-18.03 per cent. The biscuits had Vitamin C that increased with amla

pomace concentration and ranged from 26.28-49.15 mg/100 gm. The ash content of biscuits ranged from 1.8-2.27 per cent and protein content varied from 7.27-7.52 per cent. This indicated that incorporation of amla pomace powder enhanced the nutritional value of biscuits.

Surya *et al.* (2020) fortified dietary fibre into amla Ready to Serve (RTS) beverages. The amla RTS beverages were fortified with different concentration levels (0.5, 1 and 3%) of dietary fibre extracted from the amla fruit to increase the dietary fibre intake. The study showed that RTS beverage fortified with 0.5 per cent dietary fibre was found to be best in their sensory characteristics and physical properties.

The proximate analyses of rind of *Cucumis metuliferus* showed that the rind contained high concentrations of carbohydrate (54.84%) and crude fibre (11.34%); moderate amount of crude fat (8.89%) with a low concentration of ash (3.59%) and crude protein (2.95%). The rind contained varied concentrations of alkaloids, flavonoids, saponins, tannins, glycosides, terpenoids and phenol. The rind contained vitamins C, E, D, B_9 and A with appreciable concentrations of vitamins B_2, K, B_1 and β-carotene which were all above the recommended daily vitamin allowance. The high concentration of these nutrients and phytochemicals proved that the rind had higher nutritive and medicinal value (Ezekaibeya *et al.* 2020).

Ogo *et al.* (2021) assessed the functional, proximate, mineral and sensory properties of the formulated biscuit samples from blends of wheat, watermelon rind and orange pomace in the following ratio 100:0:0; 90:5:5; 80:10:10; 70:15:15 and 60:20:20, labelled samples A, B, C, D and E. The results showed that the proximate composition of samples B - E significantly increased ($p < 0.05$) except for carbohydrate content when compared with sample A used as control. There were also significant increases ($p < 0.05$) in mineral contents of supplemented samples compared to control. The sensory evaluation revealed that wheat flour can be substituted with watermelon rind and orange pomace flours up to 10 per cent without adversely affecting the overall quality attributes of the biscuits and it is an effective means of improving the nutritional quality of biscuits while contributing to waste management in the agricultural value chain.

Ashoka *et al.* (2021a) conducted a study on watermelon rind flour incorporation on nutritional and organoleptic attributes of cakes and the results revealed that cakes enriched with 10 per cent watermelon rind flour incorporation increased the nutrient composition especially fibre (2.76g per 100g) and mineral content of the cakes (Calcium-47.86mg, Phosporous-87.07mg and Iron 1.98 mg per 100g). Ashoka *et al.* (2021b) also studied the effect of watermelon rind flour incorporation on nutritional and organoleptic attributes of cookies and results showed that cookies incorporated with 30 per cent watermelon rind flour had good acceptable score (8.23). The nutrient composition of best accepted cookies had protein (4.68g), fat (30.04g), crude fibre (3.25g), carbohydrate (45.69g) and mineral content of the cookies calcium (43.11mg), iron (2.30mg) and phosphorus (71.81mg) per 100 gm. thus indicating watermelon rind flour can be utilized effectively for enhancing the nutritional composition of cakes.

2.6 Shelf-life studies of value added products

Rao *et al.* (2008) conducted storage studies of raw mango chutney powder by blending raw mango powder (RMP) with puffed chickpea (*Cicer arietinum*) powder and spices. The titrable acidity of RMP and raw mango CP were 13.7 and 8.4 per cent, respectively. Other components in the chutney powder were crude protein 7.2 per cent, crude fibre content 6.5 per cent, calcium 115.3 mg/100g and iron 31.5 mg/100g. The equilibrium moisture content - RH studies indicated that both the powders were non-hygroscopic and stable at room temperature (28±2°C) up to 6 months when packed in polyethylene pouches.

Navarro *et al.* (2009) evaluated physicochemical and microbiological properties of a high dietary fibre powder (HDFP). The storage period was 11 months and samples were stored under vacuum or air exposure and under dark or light exposure. Results revealed that HDFP is a stable product with large amounts of dietary fibre (71.62±0.24 g100/g dry sample) and polyphenols content (40.67±0.45 mg/g dry sample). The packaging material protects the HDFP only for up to 6 months. However, after six months the increased levels in moisture content decreased the quality of the product.

Pallavi *et al.* (2014) carried out storage studies of nutra *chikki* at accelerated (37±1 °C) and ambient (27±1 °C) temperatures by packing 100 g of the sample in 150-gauge polypropylene pouches for 105 days. The products were withdrawn every 15 days and the quality and stability of the product were evaluated by estimating the moisture, texture and peroxide value during storage at both temperatures. Nutra *chikki*, which had an initial moisture content of 2–3 per cent, did not alter much up to 60 days of storage at 27 °C and 37 °C. Peroxide value (PV) increased gradually at 27 °C, whereas at 37 °C, it was higher. The hardness of nutra *chikki* did not change significantly when stored at 27 °C up to 105 days, but at 37 °C a gradual increase in hardness was observed after 45 days. Nutra c*hikki* was found to be stable up to 105 days at 27 °C and up to 45 days at 37 °C.

Muttagi *et al.* (2014) evaluated the shelf life, sensory characteristics, microbial load, rancidity and peroxide value of *chikki*. The sunflower was substituted for groundnut at three levels (0, 50, and 100%). Products were stored for up to 2 months in ambient conditions (25–30 °C; RH 40–60 per cent). *Chikki* was packed in Low-density polyethylene (LDPE) and laminated pouches. Results showed that products were acceptable for sensory attributes even at the end of the storage period. Product c*hikki* stored in laminated pouches had a higher per cent free fatty acid value and peroxide value compared to that stored in Low-density polyethylene (LDPE) pouches. Stored *chikki* had a higher microbial load in the Low-density polyethylene (LDPE) when compared to that stored in laminated pouches. Products made with groundnut alone (control) were preferred over those made in combination with sunflower and groundnut (1:1) or sunflower alone. However, all products were highly acceptable at the end of the storage period.

Go *et al.* (2015) assessed the sensory evaluation and shelf-life of breadnut (*Artocarpus camansi*) cookies. The cookies were prepared in different ratios of all-purpose flour and breadnut seed flour in percent (0:100, 25:75, 50:50, 75:25, 100:0) with 100 per cent all-purpose flour served as the control. Results showed that all formulated breadnut cookies were acceptable, scoring 6.5 based on the 9-point hedonic scale and the

cookies with 50 per cent breadnut seed flour (BSF) was the most acceptable based on the general acceptability score and stored upto 6 months.

Thivani *et al.* (2016) studied the physico-chemical properties, sensory attributes and shelf-life of wheat flour biscuits incorporated with pineapple powder at the rates of 3, 5, 10 and 15 per cent (w/w basis). Results revealed that biscuits incorporated with 5 per cent pineapple powder had a slow rate of increase in moisture content (4.19 to 5%) and a decreasing trend in protein (22.15 to 22.06%), fat (18.83 to 18.79%), total sugar (58.52 to 58.34%) and mineral contents (2.212 to 1.591%) compared to other treatments during storage. Biscuits prepared with 5 per cent pineapple powder had an overall acceptability score of 7.7 on a 9-point hedonic scale. The biscuits were stored for 6 weeks at the ambient conditions of average temperature at 30±1ºC and RH at 75-80 per cent with acceptable quality. Hence, pineapple powder can be incorporated effectively at 5 percent level with good nutritional and sensory quality.

Ganapathy and Renitta (2018) analysed the biochemical composition of *Calocybe indica* chutney powder and the results indicated that the biochemical parameters decreased as compared to the initial period. A gradual increase in total plate count was observed for the product during the storage period. The sensory scores of chutney powder remained within the acceptable limit throughout the storage period. The overall acceptability of products ranged from 8.5±0.21 to 7.1±0.31 during the storage period. It was concluded that chutney powder had good sensory and nutritional profile throughout the storage period of 6 months.

III MATERIAL AND METHODS

The present investigation on **"Nutritional evaluation of Amla (*Phyllanthus emblica*) pomace and its value added products"** was carried out at Department of Food Science and Nutrition, University of Agricultural Sciences, Bangalore during the year 2020-2021. A considerable amount of waste is generated as a by-product during the processing of fruits, both at the household and industrial level. A major portion of this waste turns out to be edible and possesses a lot of nutritional and health benefits. It is essential to know the composition and nutritional characteristics of such material to study their potential for utilization. Hence, the present research work was carried out to study the physico-functional properties, analyse the nutrient composition, formulation of products, further to characterize the value added products for sensory, microbial and storage quality. The research design is depicted in Fig. 1. The described, materials and various methods used in this investigation have been given here,

3.1 Procurement of fresh amla

3.2 Processing of fresh amla to extract amla pomace

3.3 Physico-functional properties of amla pomace powder

3.4 Nutrient and phytonutrient composition of amla pomace powder

3.5 Minerals composition of amla pomace powder

3.6 Development of value added food products by incorporating amla pomace powder

3.7 Nutrient composition of the developed products

3.8 Sensory evaluation of the developed products

3.9 Shelf-life study of developed products

3.10 Consumer acceptability of the developed products

3.11 Cost estimation of the developed products

3.12 Statistical analysis

3.1 Procurement of fresh Amla

The fresh and matured amla fruits were procured from the local markets of Bengaluru, Karnataka, India.

3.2 Processing of amla

3.2.1 Processing of amla to extract amla pomace

The Amla fruits were washed under running tap water and they were wiped using a clean dry cloth. Amla fruits were cut into pieces by using a stainless-steel knife and the seeds were separated by slicing the pulp into small pieces. Then, the amla pieces were ground into pulp in the laboratory mixer, after that the juice was extracted from the pulp and the residue (pomace) was separated. The weight of seeds, pomace and juice were recorded using a digital sensitive balance and the percentages were calculated.

3.2.2 Dehydration

The amount of amla pomace obtained from fresh amla was measured as dry weight in gram from 100g amla fresh pomace sample. Dehydration was carried out by weighing fresh pomace sample and subjected to dehydration in a laboratory model ezidri ULTRAFD1000 tray dryer at 45° C for 4 hours (Fig. 2). The dried pomace was ground into powder by using a laboratory mixer and sieved. Then, the dehydrated powder was packed and used for further process (Plate 1).

3.2.3 Dehydration ratio

Dehydrated samples were weighed and per cent dry matter was calculated (Ranganna, 1986).

$$\text{Dehydration ratio} = \frac{\text{Weight of fresh sample}}{\text{Weight of dehydrated sample}}$$

3.2.4 Total edible waste of amla

The total edible waste of amla was calculated using the following formula. The per cent of edible waste was also calculated (Ashoka, 2019).

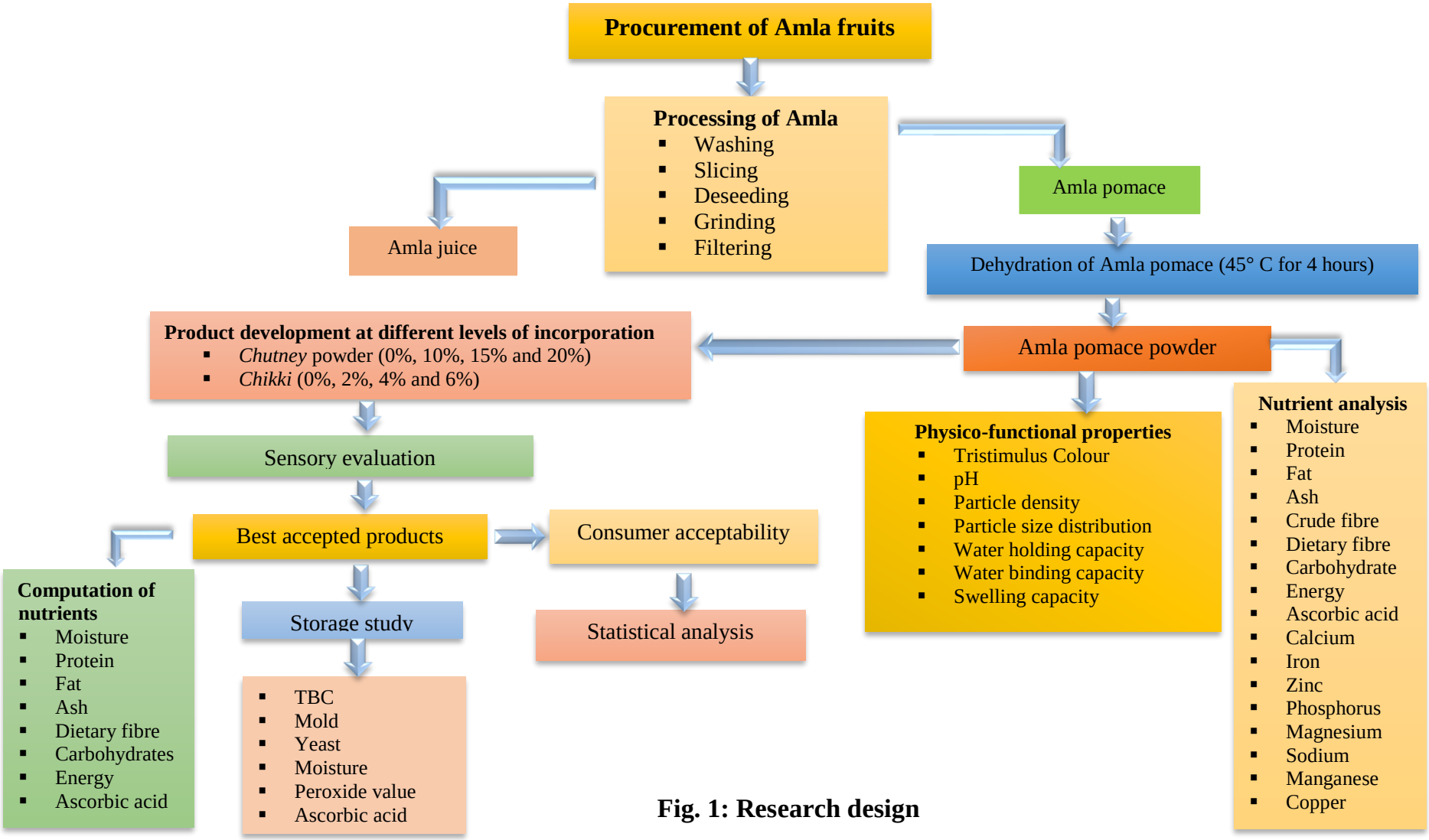

Fig. 1: Research design

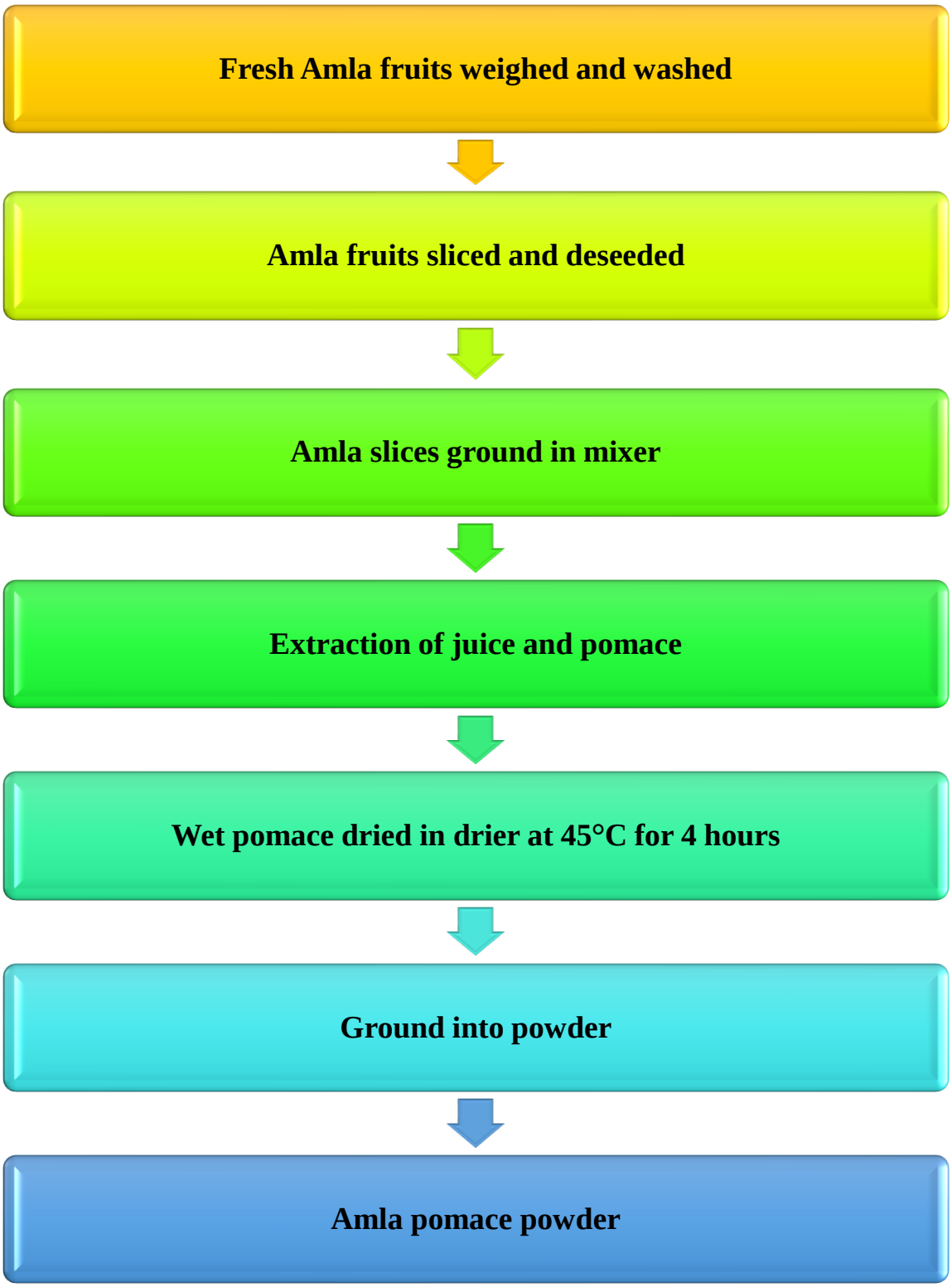

Fig. 2: Dehydration of wet amla pomace

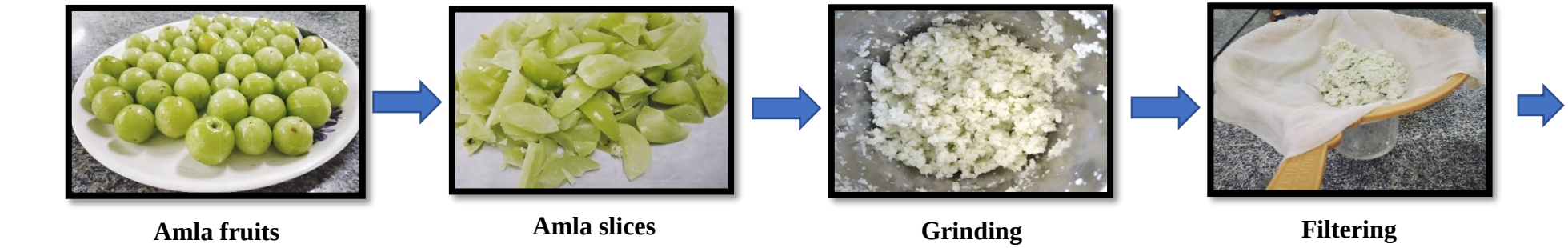

Plate 1: Extraction of amla pomace powder

Total edible waste = Weight of amla pomace

$$\text{Edible waste generated (\%)} = \frac{\text{Weight of edible waste}}{\text{Weight of whole fruit}} \times 100$$

3.2.5 Total waste generated from amla

Total waste generated from amla fruits was calculated using the following formula and percentage was calculated (Ashoka, 2019).

Total waste generated = Weight of the pomace + seed

$$\text{Total waste generated (\%)} = \frac{\text{Weight of the total waste}}{\text{Weight of whole fruit}} \times 100$$

3.3 Physico-functional properties of amla pomace powder

3.3.1 pH

pH of the amla pomace powder was measured using digital pH meter of analog model. Standard buffer solutions of pH 4.0, 7.0 and 10.0 were used to calibrate the instrument. The sample was prepared by adding 10 times distilled water to the macerated tissue in a beaker.

3.3.2 Tristimulus colour

The cieLab co-ordinates (L^*, a^*, b^*) of the amla pomace powder was directly read in a glass cuvette with a spectrophotocolorimetre, calibrated with a white tile (L^*=94.0, a^*= -1.1, b^*=0.6), at 60 °C with a D-65 illuminant source. The parameters determined were L^* [L^* = 0 (black) and L^* = 100 (white)], a* (-a^* = greenness and +a^* = redness) and b^* (-b^* = blueness and + b^* = yellowness).

3.3.3 Particle density:

Particle density was determined as described by Sangnark and Noomhorm (2003), through displacement in petroleum ether (40-60°C). Petroleum ether (40-60 °C), was

transferred into a pre-weighed (W_1) volumetric flask. The weight was noted (W_2). Repeated the similar procedure after transferring 2 g sample and noted the weight (W_3).

$$\text{Particle density (g/cm3)} = \frac{\text{Density of petroleum ether} \times \text{Weight of sample}}{\text{Weight of petroleum ether displaced}}$$

3.3.4 Water Holding Capacity:

Water holding capacity was determined using the method described by Sowbhagya *et al.* (2007). Sample (1 g) was accurately weighed into a graduate test tube and 30 ml of distilled water added. The content was allowed to hydrate for 18 h at ambient temperatures and then filtered through Whatman No.1 filter paper. The hydrated residue weight was recorded and it was dried at 105 ± 2 °C to get a constant weight. The results were expressed as gm of water held/ gm of dry sample.

$$\text{Water Holding Capacity (g/g)} = \frac{\text{Residue hydrated weight} - \text{Residue dry weight}}{\text{Residue dry weight}}$$

3.3.5 Water Binding Capacity:

Water binding capacity was determined as outlined by Sowbhagya *et al*. (2007). Sample (1 g) was accurately weighed into a graduate test tube and 30 ml of distilled water added. The content was allowed to hydrate for 18 h at ambient temperature, centrifuged (4000 rpm, 20 min) and the suspended solution was removed. A portion of wet sample was removed, weighed and dried at 100 °C to get constant weight. The results were expressed as gm of water bound per gm of sample.

$$\text{Water Binding Capacity (g/g)} = \frac{\text{Residue hydrated weight} - \text{Residue dry weight}}{\text{Residue dry weight}}$$

3.3.6 Swelling Capacity:

Swelling capacity was measured as described by Sowbhagya *et al.* (2007). Sample (1 g) was placed in a graduated test tube and the volume noted (V_1). Thereafter, 30 ml distilled water was added and the contents allowed to hydrate for 18 h. The final volume attained by the sample was measured (V_2). The result was expressed as ml of swollen sample per g of dry initial sample.

Swelling Capacity (ml/g) = Initial volume - Final volume

3.4 Nutrient and phytochemical composition of amla pomace powder

3.4.1 Moisture content (AOAC, 1980)

Samples weighing 10 g were taken and dried in an oven at 60°C. The dried samples were then weighed and the resultant value was subtracted from the fresh weight of the sample to obtain the moisture content. The moisture content of the sample was expressed in g/100 g of sample.

$$\text{Moisture \%} = \frac{\text{Initial weight (g)} - \text{final weight(g)}}{\text{Initial weight (g)}} \times 100$$

3.4.2 Estimation of protein (AOAC, 1980)

The protein content of the dried samples was estimated as per cent total nitrogen by the micro Kjeldahl procedure. Protein per cent was calculated by the multiplying the percent nitrogen by the factor 6.25.

$$\text{Protein \%} = \frac{\text{Titre value} \times \text{normality of HCl} \times 14.001 \times 6.025}{\text{Sample weight (g)}} \times 100$$

3.4.3 Estimation of fat (AOAC, 1980)

Fat was estimated as crude ether extract using moisture free samples. The solvent was removed by evaporation and the residue of fat was weighed.

$$\text{Fat \%} = \frac{\text{Weight of ether extract (g)}}{\text{Weight of sample used (g)}} \times 100$$

3.4.4 Estimation of crude fibre (AOAC, 1980)

The crude fibre of the sample was estimated by using moisture and fat-free samples and expressed as g /100g of the sample.

$$\text{Crude fibre (g/100g sample)} = \frac{[100 - (\text{moisture} + \text{fat})] \times (We - Wa)]}{\text{Wt. of a sample taken (moisture and fat free)}}$$

Where,

W_e = pre-weighed ashing

W_a = weight of the dish after ashing.

3.4.5 Estimation of ash (AOAC, 1980)

Ash content was estimated by using 5 g of sample, weighed accurately in a crucible and heated in low flame until the sample charred. Then followed by heating in a muffle furnace for about 4 to 5 hours at 600 ^{0}C, then crucibles were cooled in a desiccator and weight is recorded, it is done until getting constant weight and the ash was almost white or greyish white in colour.

$$\text{Ash content (g /100g)} = \frac{\text{Weight of Ash}}{\text{Weight of sample taken (g)}} \times 100$$

3.4.6 Estimation of tannins (AOAC, 1980)

Tannins estimation is based on the measurement of the blue colour formed by the reduction of phosphotungstomolybdic acid in alkali solution. Tannin-like compounds reduce phosphotungstomolybdic acid in alkaline solution to produce a highly coloured blue solution, the intensity of which is proportional to the amount of tannins. The intensity is measured in a spectrophptometer at 700 nm. Standard curve developed was used for the calculation of tannin content.

3.4.7 Total phenols (Ranganna, 2005)

Total phenol estimation in fibre samples was carried out using Folin Ciocalteu reagent (FCR), according to the method described by (Ranganna, 2005). Sample (0.5 g) was weighed and ground with 10 times of 80% ethanol in a pestle-mortar. The homogenate was centrifuged at 10,000 rpm for 20 min and the supernatant was decanted and kept. The procedure was repeated again with 5 volumes of ethanol and the supernatant was decanted, both the supernatant was pooled and evaporated using flash evaporator at 40 °C. The residue was dissolved in 5 ml distilled water and 0.2-2 ml aliquots were pipetted into the test-tubes. The final volume was made up to 3 ml, followed by 0.5 ml FCR. After 3 min, 2 ml 7% sodium carbonate was added and the mixture was placed in a boiling water bath for 2 min. The tubes were cooled and absorbance was measured at 650 nm, against blank. A standard curve was plotted using standard catechol graph.

3.4.8 Estimation of ascorbic acid (Ranganna, 2005)

Ascorbic acid was determined colorimetrically. Ascorbic acid was first dehydrogenated by bromination. The dehydroascorbic acid was then reacted with 2,4-dinitrophenyl hydrazine to form osazone and dissolved in sulphuric acid to give an orange-red colour solution which was measured at 540nm.

3.4.9 Estimation of soluble dietary fibre (AOAC, 1980)

The soluble fibre is estimated in the filtrate obtained after enzymatic digestion of protein and carbohydrate of defatted food. The soluble fibre is precipitated and estimated gravimetrically.

Soluble dietary fibre (SDF) = Weight of SDF residue – (Protein + Ash)

$$\textbf{SDF \%} = \frac{\text{Weight of the SDFR (g) - \{Protein (g) in SDFR + Ash (g) in SDFR\}}}{\text{Weight of the sample (g)}} \times 100$$

SDFR = Soluble dietary fibre residue

3.4.10 Estimation of insoluble dietary fibre (AOAC, 1980)

Defatted foods were gelatinized, and proteins and starch were removed by enzymatic digestion. The residue was quantified gravimetrically.

Insoluble dietary fibre (IDF) = IDF residue – (Protein + Ash)

$$\textbf{IDF \%} = \frac{\text{Weight of the IDFR (g) - \{Protein (g) in IDFR + Ash (g) in IDFR\}}}{\text{Weight of the sample (g)}} \times 100$$

IDFR = Insoluble dietary fibre residue

3.4.11 Estimation of total dietary fibre (AOAC, 1980)

The total dietary fibre was the sum of soluble and insoluble dietary fibre.

Total dietary fibre = IDF + SDF

3.4.12 Computation of carbohydrate

Carbohydrate content was calculated by the differential method.

CHO (g/100g) = 100- [Protein(g) + Fat(g) + Fibre(g) + Ash(g) + Moisture(g)]

3.4.13 Computation of energy

Energy was computed as follows.

Energy (kcal) = [Protein (g) × 4] + [Carbohydrate (g) × 4] + [Fat (g) × 9].

3.5 Minerals estimation of amla pomace powder

3.5.1 Digestion of amla pomace powder

The plant material can be digested in a di-acid mixture of a tri-acid mixture or dry ashed and dissolved in acid (Annexure, XV).

3.5.2 Estimation of calcium (AOAC, 1980)

Calcium content of the sample was estimated by preparing mineral solution (Annexure, XVII) and titrating it against 0.01N EDTA under alkaline condition.

$$\text{Ca (\%)} = \frac{\text{T.V. for Ca} \times \text{N of EDTA} \times 0.02 \times \text{Vol. of digested sample}}{\text{Aliquot taken} \times \text{weight of sample}} \times 100$$

3.5.3 Estimation of phosphorous (AOAC, 1980)

The phosphorus in the aliquot of the digested material was measured in presence of vanadium (V^{5+}) and molybdenum (Mo^{6+}). Orthophosphate forms a yellow coloured Phospho-Vanado-Molybdate complex which was read using spectrophotometer at 430nm (Annexure XVI).

$$\text{P (\%)} = \frac{\text{Graph ppm} \times \text{Vol. of digested sample} \times \text{Vol. made up}}{10^6 \times \text{weight of sample} \times \text{Aliquot taken}} \times 100$$

3.5.4 Estimation of magnesium (AOAC, 1980)

Magnesium content of the sample was calculated by preparing mineral solution and titrating it against 0.01N EDTA in the presence of buffer and then subtracting this titre value from the titre value of calcium (Annexure, XVII).

$$\text{Mg (\%)} = \frac{[\text{A} - \text{B}] \times \text{N of EDTA} \times 0.012 \times \text{Vol. of digested sample}}{\text{Aliquot taken} \times \text{weight of sample}} \times 100$$

A = Titre value of calcium, B = Titre value of magnesium.

3.5.5 Estimation of sodium (Ranganna, 2002)

Feed the digested sample solution to the Flame photometer and record the reading (if dilution is required, it should be done before feeding to the instrument). Compare the unknown sample readings with standard curve to determine the % Na in plant sample (Annexure, XVIII).

$$\text{Na (\%)} = \frac{\text{Graph ppm} \times \text{Vol. of digested sample} \times \text{Vol. made up}}{10^6 \times \text{weight of sample} \times \text{Aliquot taken for dilution}} \times 100$$

3.5.6 Estimation of Fe, Mn, Zn and Cu (AOAC, 1980)

Make suitable dilutions of di acid extract and feed standard/ sample to AAS having appropriate hallow cathode lamps. Record values and plot on graph paper (Annexure, XIX).

$$\text{Micronutrient conc. (ppm)} = \frac{\text{Graph ppm} \times \text{Vol. of digested sample}}{\text{Weight of sample}}$$

3.6 Development of value added products by incorporating amla pomace powder

Amla pomace powder based value added products were formulated as per standard procedures. Different value added products such as *chikki* and *chutney* powder were developed from pomace powder.

3.6.1 Procurement of raw materials

The raw materials required for the study were procured from the local markets of Bengaluru.

3.6.2 Amla pomace powder incorporated *chikki*

The *chikki* was prepared by using amla pomace powder with peanuts and jaggery. Three variations of *chikki* were developed by incorporating a pomace powder at different

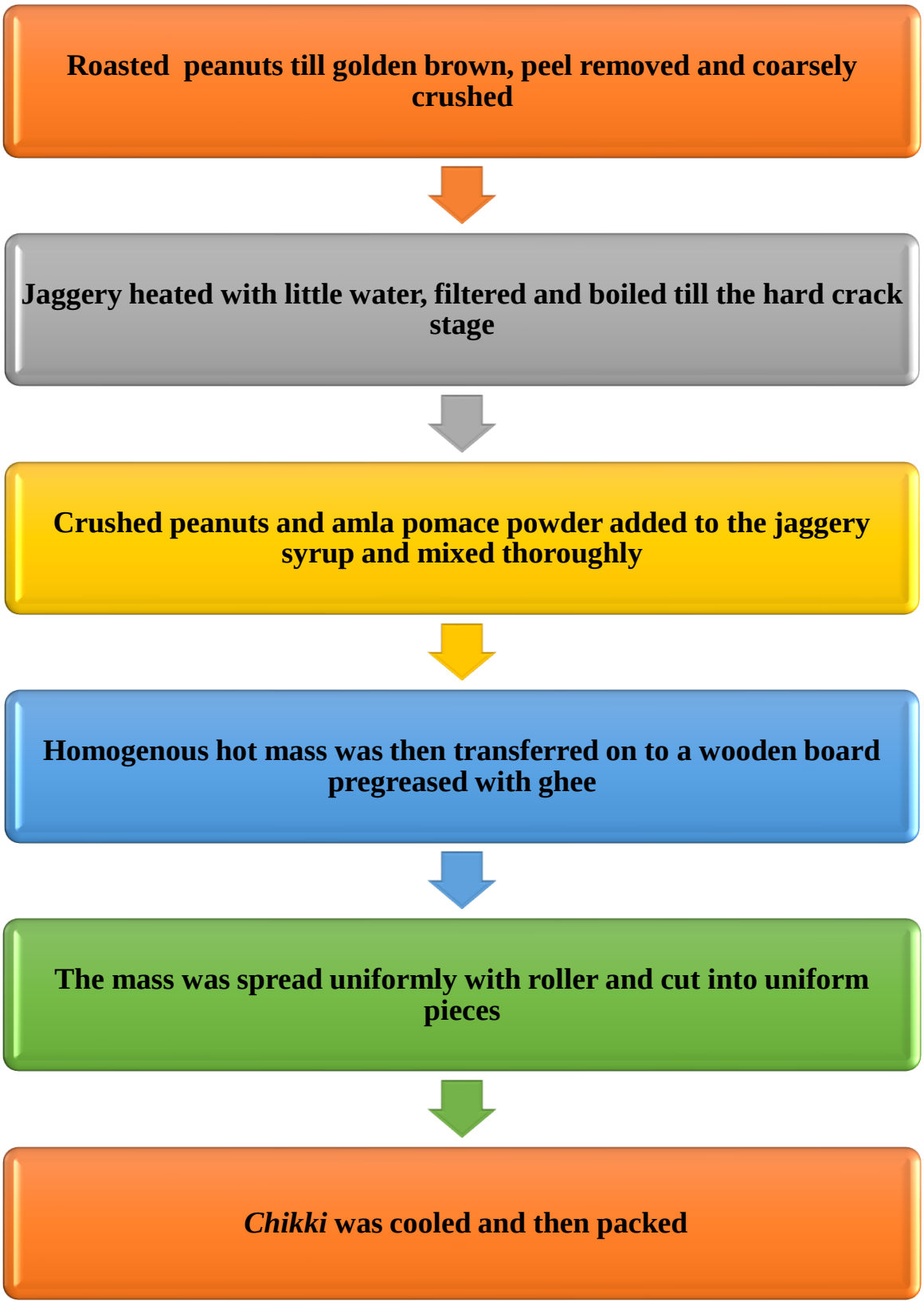

Fig. 3: Preparation of amla pomace *chikki*

Control

T_1 (2%)

T_2 (4%)

T_3 (6%)

Plate 2: Amla pomace *chikki*

variations like (2, 4 and 6%) and they were compared with control as indicated in Table 3 and Plate 2. The procedure for preparation has been mentioned in Fig. 3 (Annexure XX).

Table 3: Amla pomace powder incorporated *chikki*:

Ingredients	**Control (%)**	**T_1 (%)**	**T_2 (%)**	**T_3(%)**
Amla pomace powder	0	2	4	6
Peanut *chikki*	100	98	96	94

T_1 = 2% Amla pomace powder, T_2 = 4% Amla pomace powder and T_3 = 6% Amla pomace powder

3.6.3 Amla pomace powder incorporated *chutney* powder

The *chutney* powder was prepared by using amla pomace powder with flax seeds, black gram dhal, bengal gram dhal and other ingredients. Three variations of *chutney* powder were developed by incorporating amla pomace powder at different variations like (10, 15 and 20 %) and they were compared with control as indicated in Table 4 and Plate 3. The procedure for preparation has been mentioned in Fig. 4 (Annexure XX).

Table 4: Amla pomace powder incorporated *chutney* powder:

Ingredients	Control **(%)**	**T_1 (%)**	**T_2 (%)**	**T_3 (%)**
Amla pomace powder	00	10	15	20
Flax seed *chutney* powder	100	90	85	80

T_1 = 10% Amla pomace powder, T_2 = 15% Amla pomace powder and T_3 = 20% Amla pomace powder.

3.7 Organoleptic evaluation of the developed products

The products were subjected to sensory evaluation. Sensory attributes were evaluated by a panel of 21 semi-trained members using a nine-point hedonic scale [Amerine *et al.* (1965)] (plate 4). The products were evaluated for their appearance, colour, texture, flavour, taste and overall acceptability. The score sheet is included in (Annexure XI).

3.8 Nutrient composition of the developed products

Nutrient composition of the best accepted developed products was computed based on the nutritional composition of the ingredients using Indian Food Composition Tables, NIN, ICMR Hyderabad (Longvah *et al.*, 2017)

3.9 Shelf-life study of developed products

The shelf life study was conducted for a period of 45 days. Best accepted amla pomace *chutney* powder and *chikki* were used for storage study. The amla pomace *chutney* powder and *chikki* were prepared and stored in metallised polypropylene pouches. Then, the products were kept in both room and refrigerated conditions. The products were evaluated on initial 15^{th}, 30^{th} and 45^{th} day for sensory attributes, microbial load and biochemical parameters (moisture, peroxide value and ascorbic acid).

3.9.1 Microbial analysis

The microbial analysis of the best accepted products was carried out by standard plate count method using nutrient agar (NA) for bacteria, Martin's Rose Bengal Agar (MRBA) for mold and YEPDA for yeast. Ten grams of each sample was mixed in 100 ml water blank to give 10^{-2} dilutions. Subsequent dilutions up to 10^{-6} were made by transferring serially 1 ml of the dilution to 9 ml sterile water blanks. The population of *Escherichia coli*, bacteria and molds were estimated by transferring 1 ml of dilutions to the sterile petri plates and 20 ml of medium was poured in plates. The plates were rotated twice in a clockwise and anti-clockwise direction for uniform distribution of inoculum. After solidification of the medium, plates were incubated in an inverted position at 28 ± 2 ºC for 2 days (bacteria) and for 4 days (molds) and emerged colonies were counted [(Annexure XIV) (Tate, 1995)].

3.9.2 Sensory evaluation

All the samples were analysed for changes in sensory parameters during storage by a panel of 21 semi-trained members. A nine-point hedonic scale was used for organoleptic evaluation. The products were evaluated for their appearance, texture, colour, flavour, taste and overall acceptability.

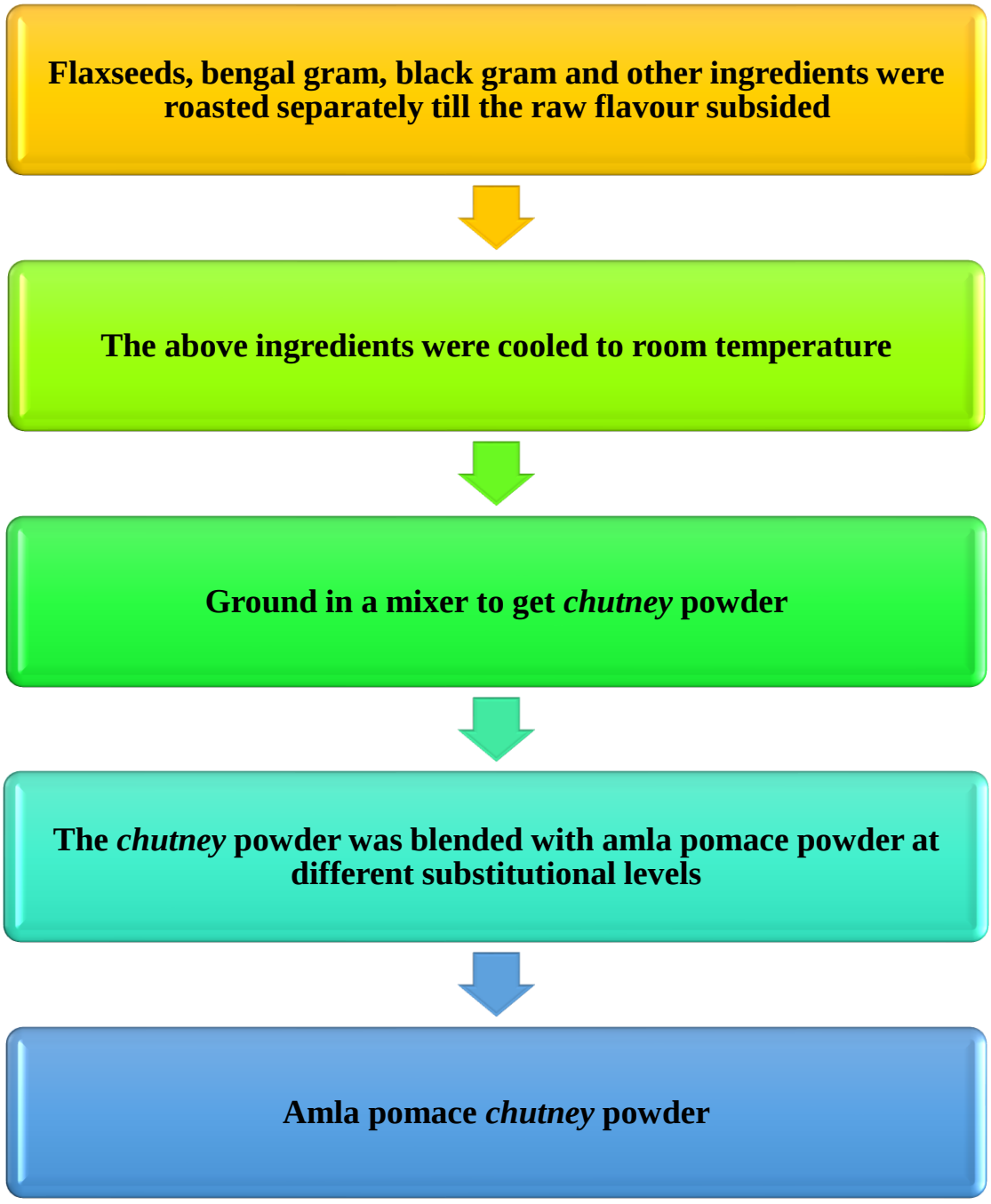

Fig. 4: Preparation of amla pomace *chutney* powder

Control

T_1 (10%)

T_2 (15%)

T_3 (20%)

Plate 3: Amla pomace *chutney* powder

3.9.3 Analysis of biochemical parameters

The samples were analyzed for biochemical parameters like moisture content, peroxide value and ascorbic acid at periodic intervals during storage study.

Peroxide Value

Clear melted fat of 0.5 to lg was weighed accurately in the boiling flask. To this 30 ml of acetic acid-chloroform mixture was added and fat was dissolved. One ml of saturated potassium iodide was added. After five minutes 100 ml of distilled water was added. The liberated iodine was titrated against N/1000ml sodium thiosulphate. When the end point approached one ml of freshly prepared starch was added and titration was completed till the blue colour disappears. Blank was carried out using all the reagents without the melted fat [(Annexure- X) **(**Raghuramulu *et al.,* 2003)]. The peroxide value was calculated as given below.

$$\text{Peroxide value of oil (meq/kg of sample)} = \frac{\text{(Titre-blank) x N x 1000}}{\text{Weight of oil (g)}}$$

Note: In case of samples, with less than 1g fat content, the peroxide value (PV) values are expressed on sample basis.

3.10 Consumer acceptability of the developed products

The consumer acceptability of amla pomace *chutney* powder and *chikki* was assessed by using the FACT scale (Annexure XII) (Deepa 2015) as well as by using a five-point hedonic scale (Annexure XIII) by the consumers (n=50).

3.11 Cost estimation of the developed products

Cost of the best-accepted *chutney* powder and *chikki* was calculated by considering the cost of the raw material purchased from the local market, 30 per cent overhead costs which included labour charge, electricity, machinery, packaging cost, etc. and 25 per cent profit. The total price was calculated for 100 g of the products (Annexure XV).

3.12 Statistical analysis

The data were subjected to Complete Randomised Design (CRD) analysis of variance for testing the significance of variation in biochemical parameters, microbial load and sensory evaluation of developed products by using the statistics *i.e.* software Statistical Package for Social Sciences (SPSS) version 12.0 (Sabine and Brian, 2004).

Plate 4: Sensory evaluation of developed products

IV RESULTS AND DISCUSSION

The present study was undertaken on the **"Nutritional evaluation of Amla (*Phyllanthus emblica*) pomace and its value added products".** The research was carried out at the Department of Food Science and Nutrition, UAS, GKVK, Bengaluru during 2020-2021. The physico-functional properties were assessed, the nutritional composition were analysed, value added products were formulated and shelf-life study was conducted. The results obtained and findings of the research are discussed in this chapter under the following headings along with tables, figures and plates.

4.1. Processing of amla

4.1.1 Fractions of amla fruit

4.1.2 Dehydration of amla pomace

4.2. Physico-functional properties of amla pomace powder

4.3. Nutritional composition of amla pomace powder

4.4. Phytonutrients of amla pomace powder

4.5. Mineral composition of amla pomace powder

4.6. Sensory evaluation of amla pomace powder

4.7. Microbial load of amla pomace powder storage

4.8. Amla pomace powder incorporated *chikki*

4.8.1 Sensory evaluation of *chikki*

4.8.2 Sensory evaluation of *chikki* on storage at room temperature

4.8.3 Sensory evaluation of *chikki* on storage at refrigerated temperature

4.8.4 Microbial load of *chikki* on storage

4.8.5 Effect of storage on moisture, peroxide value and ascorbic acid of *chikki*

4.8.6 Consumer acceptability of *chikki*

4.9. Amla pomace powder incorporated *chutney* powder

4.9.1 Sensory evaluation of *chutney* powder

4.9.2 Sensory evaluation of *chutney* powder on storage at room temperature

4.9.3 Sensory evaluation of *chutney* powder on storage at refrigerated temperature

4.9.4 Microbial load of *chutney* powder on storage

4.9.5 Effect of storage on moisture, peroxide value and ascorbic acid of *chutney* powder

4.9.6 Consumer acceptability of *chutney* powder

4.10 Nutritional composition for best accepted products by computation

4.10.1 Nutrient composition of best accepted *chikki* (per 100g)

4.10.2 Nutrient composition of best accepted *chutney* powder (per 100g)

4.11 Cost estimation of the developed products

4.1 Processing of amla fruits

4.1.1 Amla fruit fractions

The processing of amla fruit was carried out and the various amla fruit fractions is indicated in Table 5. The amla fruit fractions were categorized into three major components *viz*, juice, pomace and seeds, that constituted 72.2, 18.6 and 8.9 per cent, respectively of the total fruit weight. Also, a processing loss of 0.3 per cent of the solids was observed that can be attributed to different processing methods like washing, cutting, grinding, separation of juice, drying and sieving (Fig. 5).

It was found that the total amount of waste generated from fresh amla fruit after processing was 27.5 per cent including the weight of the pomace and seeds. Out of 27.5 per cent of total waste, the edible waste was 18.6 per cent which includes the weight of the edible amla pomace. Similar results were observed by Kohli (2019) wherein the citrus fruit pomace was found to be 25 per cent.

Table 5: Per cent fractions of amla fruit

Fruit fractions	Per cent
Juice	72.2
Pomace	18.6
Seed	8.9
Processing loss	0.3

4.1.2 Dehydration of amla pomace

The dehydration parameters of the amla pomace are indicated in Table 6. On the basis of the data obtained, the recovery amount of dehydrated amla pomace is 32.25 g per 100 g fresh pomace. It was observed that there is high moisture loss and the dehydrated ratio was found to be 3.07. The results were analysed and expressed for 100 g of residue. The dehydrated amla pomace is 32.25 g per 100 g fresh pomace. The loss of weight is due to the high moisture content in the fresh pomace.

Table 6: Dehydration of amla pomace

Parameters	Value
Duration (h)	4
Temperature (°C)	45
Weight before drying (g)	100
Weight after drying (g)	32.25
Weight loss (g)	67.75
Dehydration ratio	3.07

4.2 Physico-functional properties of amla pomace powder

The physico-functional characteristics of the dehydrated amla pomace powder such as colour, pH, particle density, water holding capacity, water binding capacity and

swelling capacity is indicated in Table 7. The pH was found to be 3.44, colour *L** = 86.47, *a** = -0.74 and *b** = 11.13, particle density 1.23 g/cm^{3,} water holding capacity was found to be 12.30 g/g, water binding capacity was 12.37 g/g and swelling capacity was 13 ml/g. Similar values are reported by Gupta *et al.* (2010) for ash gourd and radish fibre with values for particle density to be 1.21 and 1.17 g/cm^3, water holding capacity (12.26 and 13.81 g/g), and higher values for water binding capacity (14.13 and 15.61 g/g) and swelling capacity (19.49 and 12.95 ml/g) compared to the present findings. The functional properties of dragon fruit peel powder reported by Chia and Chong (2015) for water holding capacity and swelling capacity was found to be 2.52 g/g and 6.23 ml/g, respectively that is lower than the present study.

Also, the physico-functional properties of pear pomace reported by Yan *et al.* (2019) indicated values of pH to be 3.88 that is higher than the pH value obtained in the present investigation, water holding capacity was found to be 3.44 g/g and swelling capacity 5.09 g/ml that are lower than the values obtained in the present study.

Table 7: Physico-functional properties of amla pomace powder

Particulars		**Value**
pH		3.44
Colour	*L** (whiteness)	86.47
	*a** (Greenness)	- 0.74
	*b** (Yellowness)	11.13
Water Holding Capacity (g/g)		12.30
Water Binding Capacity (g/g)		12.37
Swelling Capacity (ml/g)		13
Particle Density (g/cm^3)		1.23

*L** = [*L** = 0 (black) and *L** = 100 (white)], *a** = [-*a** = greenness and +*a** = redness] and *b** [(-*b** = blueness and + *b** = yellowness].

The details regarding distribution of particle size of amla pomace powder is indicated in Table 8 and Plate 5. The distribution of particle size of amla pomace powder was 21.44, 16.93 and 61.62 per cent through the mesh sieves 53, 125 and 250 microns, respectively. The highest yield was recorded 61.62 per cent for 250 microns. The water holding capacity, swelling capacity and water binding capacity of amla pomace powder increased with increase in particle size. It exhibited 8.91 g/g, 8.5 ml/g and 7.48 g/g for 53 micron, 15.70 g/g, 12.5 ml/g and 12.11 g/g for 125 micron, 13.08 g/g, 10 ml/g and 10.55 g/g for 250 micron, respectively. As the particle size of amla pomace powder increased, the water holding capacity, swelling capacity and water binding capacity of amla pomace powder also increased. The reason can be attributed to the ability of a pomace powder to absorb and hold more water due to its texture, porosity, density and finer particle size. Similar results were observed by Gupta and Premavalli (2010) who reported distribution of particle size of ashgourd and radish fibre yields ranged from 26-30 per cent and 14-17 per cent on 30 and 100 mesh sieves, whereas in 60 mesh sieve highest yield was recorded 40.33 and 43.83 per cent, respectively.

Table 8: Distribution of particle size of amla pomace powder

Mesh size (mm)	Micron	Particle size distribution (%)	WHC (g/g)	SC (ml/g)	WBC (g/g)
0.053	53	21.44	8.91	8.5	7.48
0.125	125	16.93	13.08	10	10.55
0.250	250	61.22	15.70	12.5	12.11

WHC = Water holding capacity, SC = Swelling capacity and WBC = Water binding capacity.

4.3 Nutritional composition of amla pomace powder

The nutritional composition of amla pomace powder is indicated in Table 9. Among all nutrients ascorbic acid was found to be in higher amount of 432 mg/100g

compared to amla fruit which is having 252 mg/100g indicating that amla pomace would be good ingredient to supplement ascorbic acid. The crude fibre content of amla pomace and amla fruit was found to be 13.4g and 3.4g per 100g respectively indicating that the by-product of amla, *i.e.* amla pomace would be a good source of fibre which would otherwise go as a waste. Increase in nutritional composition of amla pomace may be due to removal of moisture content. Amla pomace has low calorific value of 36 Kcal, low carbohydrate of 7 g and low fat of 0.18 g per 100g and hence can be used to formulate low calorie, low fat, low carbohydrate, high fibre and high ascorbic acid containing functional foods. The other nutrients like moisture, protein and ash was found to be 4.99, 1.55 and 2.60 g, respectively. Hence, it can be concluded that the nutrient composition of amla pomace powder was higher than amla fruit as indicated in Indian Food Composition Tables of NIN, ICMR, Hyderabad (Longvah *et al.*, 2017) and the values being protein (0.34g), fat (0.16g), ash (0.34g) and carbohydrate (4.39g).

Further, the nutritional composition of amla pomace powder in the present study is almost similar to that reported by Nagamaniammai (2013). Also, the values obtained in the present study were lower compared to the study conducted by Fathima (2018) reported nutrient composition of banana peel powder to be moisture (6.39g), Protein (7.04g), ash (11g), fat (4.84g), crude fibre (28.30g), carbohydrate (71g) and energy (355 kcal). This difference in nutrient composition in the present study and reported studies may be due to varietal difference and agro climatic conditions.

The fibre content of amla pomace powder is presented in Table 10. The total dietary fibre (TDF), insoluble dietary fibre (IDF) and soluble dietary fibre (SDF) was found to be 41.7, 27.4 and 14.5 g, respectively. The findings were slightly higher in the present investigation than reported by Nagamaniammai (2013) who analysed dietary fibre content of amla pomace *i.e.* total dietary fibre (39.79g), insoluble dietary fibre (26.18g) and soluble dietary fibre (13.57g).

Mesh size 0.053mm (53µ)

Mesh size 0.125mm (125µ)

Mesh size 0.250mm (250µ)

Plate 5: Particle size distribution of amla pomace powder

Table 9: Nutrient composition of amla pomace powder in comparison with the amla fruit (Per 100g)

Nutrients	Amla pomace powder [1]	Amla fruit [2]
Moisture (g)	4.99	87
Protein (g)	1.55	0.34
Ash (g)	2.60	0.34
Fat (g)	0.18	0.16
Crude fibre (g)	13.4	3.4
Carbohydrate (g)	7.0	4.39
Energy (Kcal)	36	24
Ascorbic acid	432	252

1 - Analysed value as per AOAC 1980,

2 - Indian Food composition tables, ICMR, NIN, Longvah *et al.*, 2017.

Table 10: Dietary fibre composition of amla pomace powder

Parameter	Content (g/100g)
Total dietary fibre	41.7
Insoluble dietary fibre	27.4
Soluble dietary fibre	14.5

4.4 Phytonutrients of amla pomace powder

The results of phytonutrients such as polyphenols and tannins of amla pomace powder are presented in Table 11 and the values being 677 and 524 mg, respectively that are slightly lower than the study conducted by Nagamaniammai (2013) for 743 mg and 590 mg for polyphenols and tannins. The reason can be attributed to the variation in varieties, climatic condition and geographic location grown.

Table 11: Phytonutrients of amla pomace powder (Per 100g)

Phytonutrients	Content (mg)
Polyphenols	677
Tannins	524

4.5 Mineral composition of amla pomace powder

The minerals composition of amla pomace powder is presented in Table 12. It was noticed that amla pomace is the store house of minerals. The total ash content of amla pomace in the present study was 2.60 g in which the calcium content was 128 mg, phosphorus 116 mg, sodium 92.2 mg, magnesium 48 mg, manganese 2 mg, copper 1.34 mg, iron 1.12 mg and zinc 0.92 mg. It is observed that the mineral composition in the present study is lower than the results obtained by Silva *et al.* (2017) who studied the mineral content of citrus residue and found 680 mg of calcium, 11.64 mg of iron, 91.55 mg of magnesium and 7.4 of mg zinc, respectively per 100g which is higher than the present study.

Table 12: Mineral composition of amla pomace powder (Per 100g)

Minerals	Content (mg)
Calcium	128
Phosphorus	116
Sodium	92.2
Magnesium	48
Manganese	2
Copper	1.34
Iron	1.12
Zinc	0.92

4.6 Sensory evaluation of amla pomace powder

Sensory evaluation of the amla pomace powder was carried out by 21 semi-trained panelists on a nine-point hedonic scale. The panelists were given instructions either to taste amla pomace powder as such or by mixing it with water as a beverage. Sensory attributes like appearance, colour, texture, flavour, taste and overall acceptability were scored based on its intensity scale. The result of the sensory analysis is presented in Fig. 6.

The sensory scores for amla pomace powder were 7.50 for appearance, 7.68 for colour, 7.09 for texture, 6.68 for flavour, 6.77 for taste and 7.18 for overall acceptability. The sensory scores show that the amla pomace powder to be acceptable by panelists.

4.7 Microbial load of amla pomace powder on storage

Microbial study for total bacterial count, yeast and mold was carried out for amla pomace powder for 45 days at both room temperature (25±2 C°) and refrigerated temperature (4 C°) and results are presented in Table 13. The results indicated that bacterial counts of amla pomace powder on the initial day was nil, however it was observed that there was increase in on 15^{th} day, 30^{th} and 45^{th} day it was found to be 0.33×10^5 cfu/g, 1.33×10^5 cfu/g and 2×10^5 cfu/g respectively. The yeast and mold counts were found to be nil on initial and on 15^{th} day. However, it was increased to 1.33×10^2 cfu/g of yeast and 2×10^3 cfu/g of mold on 45^{th} day in room temperature conditions. In refrigerated conditions the bacterial, yeast and mold count recorded was 1.33×10^5 cfu/g, 1.33×10^2 cfu/g and 1.33×10^3 cfu/g respectively on 45^{th} day of storage period. However, it was observed that the microbial counts were within the permissible limits. Statistically significant difference was found during storage period at different time intervals.

The microbial counts in the present study were found to be lower than that reported by Akshata (2017) who conducted study on muskmelon powder where in the bacterial counts on the initial day was nil and on 15^{th} day, 30^{th} day and 45^{th} day it was found to be 1.02, 2.90 and 4.20×10^4 cfu/g respectively. Whereas mold count was found to be nil and population increased to 1.6, 2.58 and 4.65×10^4 cfu/g on 45^{th} day of storage.

Table 13: Microbial load of amla pomace powder on storage

Storage condition	Duration	TBC ($\times 10^5$cfu/g)	Yeast ($\times 10^2$ cfu/g)	Mold ($\times 10^3$cfu/g)
Room temperature	Initial	Nil	Nil	Nil
	15th day	0.33	Nil	Nil
	30th day	1.33	1	1.33
	45th day	2	1.33	2
	F value	*	*	*
	SEm±	0.23	0.16	0.33
	CD@5%	0.78	0.55	1.10
Refrigerated temperature	Initial	Nil	Nil	Nil
	15th day	Nil	Nil	Nil
	30th day	0.66	1	0.66
	45th day	1.33	1.33	1.33
	F value	*	*	*
	SEm±	0.23	0.33	0.23
	CD@5%	0.78	1.10	0.78

NS = Non-significant, * = Significant at 5% and TBC = Total bacterial count.

4.8 Amla pomace powder incorporated *chikki*

Chikki is considered as a favourite food product consumed as snacks by children and adults due to variety in sweetness, crunchiness and digestibility. A *chikki* is typically small, square in shape, flat and sweet. It usually contains peanut, jaggery and edible oil or fat. In the present investigation *chikki* was formulated with the incorporation of amla pomace powder at 2, 4 and 6 per cent levels of substitution along with control (Fig. 3).

4.8.1 Sensory evaluation of *chikki* incorporated with amla pomace powder

The result of the sensory evaluation of *chikki* incorporated with amla pomace powder is presented in Table 14 and Fig. 7. The sensory scores for appearance ranged from 7.59 to 8.14 for, 7.69 to 8.14 for colour, 6.97 to 8.00 for texture, 7.50 to 7.80 for flavour, 7.28 to 7.71 for taste and 7.40 to 7.85 for overall acceptability. Control variation had highest score for all of the sensory parameters. However, among the experimental

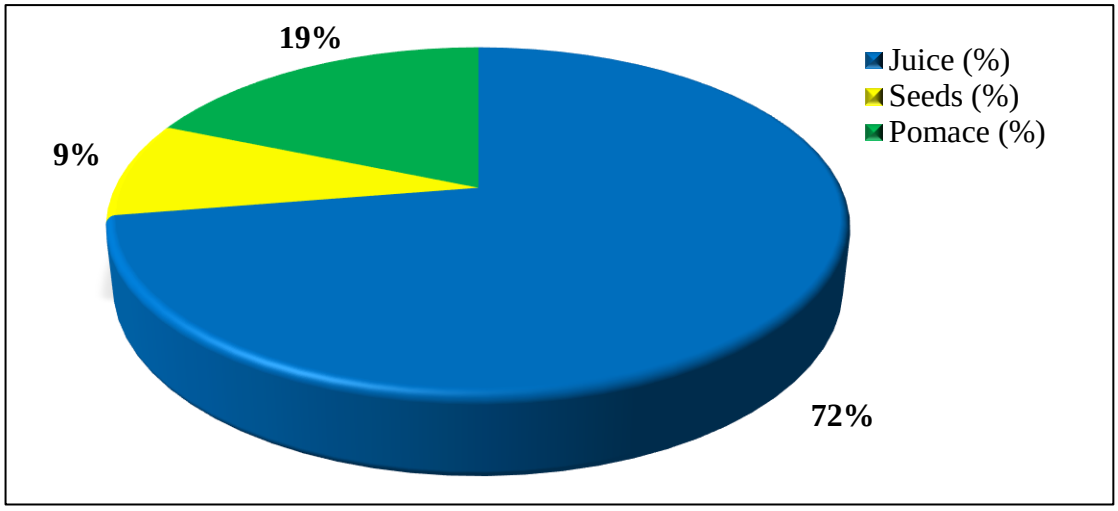

Fig. 5: Per cent fractions of amla fruits

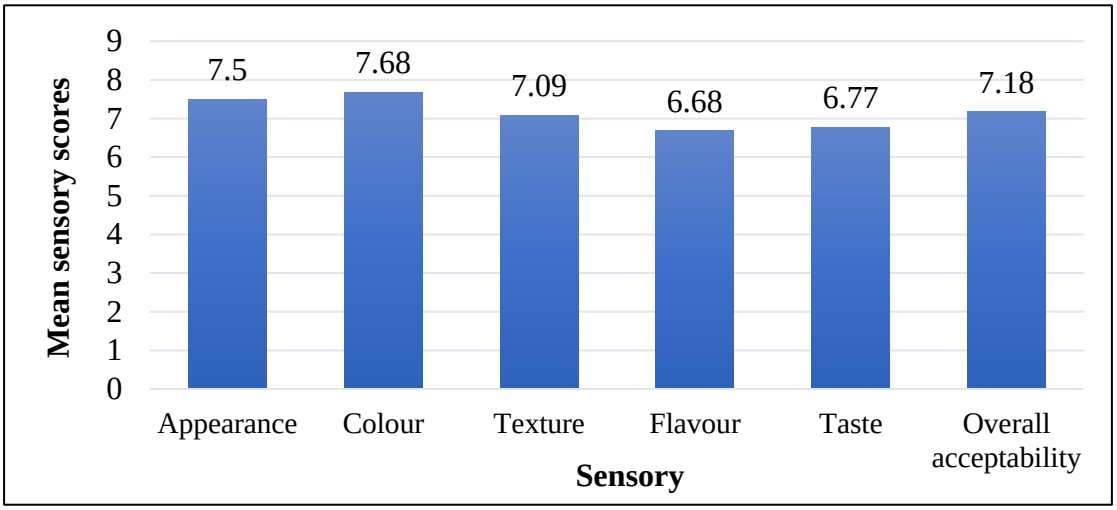

Fig. 6: Sensory scores of amla pomace powder

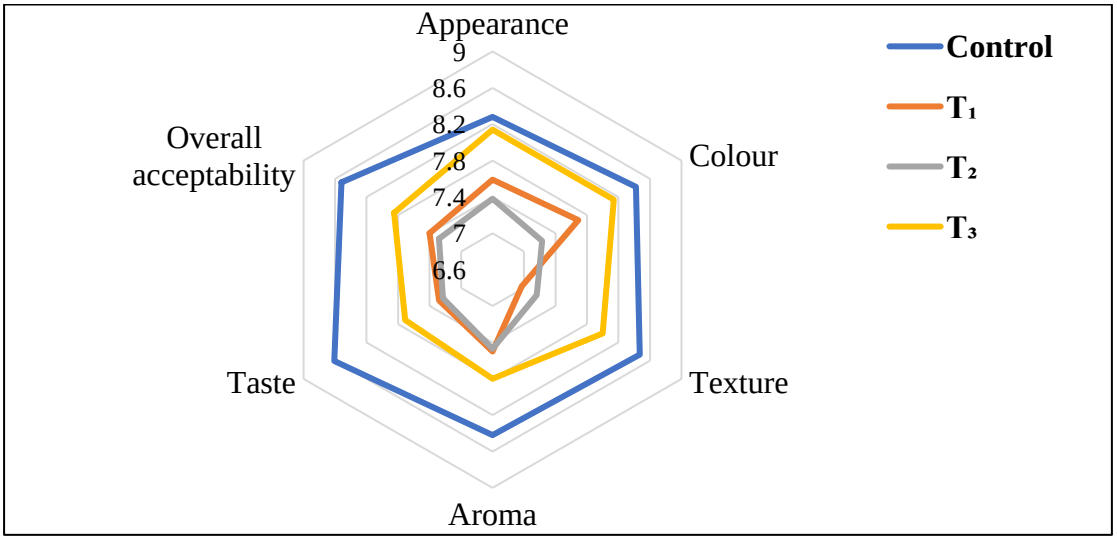

Fig. 7: Sensory scores of *chikki*

variations, T_3 with 6 per cent amla pomace powder was found to be best accepted with respect to all sensory parameters. The statistical analysis indicated difference in sensory parameters among different variations and was found to be statistically significant at 5 per cent.

Similar results were observed by Devhare *et al*. (2020) in a study on pomegranate peel powder incorporated peanut *chikki* and the results indicated that the score for the colour and appearance of *chikki* ranged from 7.30-8.50, while texture and flavour ranged from 7.20-8.47 and 7.17- 8.43, respectively, taste score ranged from 7.17-8.43 and overall acceptability ranged from 7.30-8.27.

Table 14: Sensory parameters of *chikki* incorporated with amla pomace powder

Treatments	Sensory parameters					
	Appearance	**Colour**	**Texture**	**Aroma**	**Taste**	**Overall acceptability**
Control	8.28±0.56	8.42±0.59	8.47±0.60	8.42±0.67	8.61±0.49	8.52±0.60
T_1	7.59±0.94	7.69±0.81	6.97±0.95	7.50±1.09	7.28±0.90	7.40±0.83
T_2	7.38±0.97	7.23±0.99	7.16±1.06	7.47±1.03	7.23±0.76	7.28±0.78
T_3	8.14±0.65	8.14±0.65	8.00±1.00	7.80±1.07	7.71±1.05	7.85±0.85
F value	*	*	*	*	*	*
SEm±	0.17	0.17	0.20	0.21	0.18	0.16
CD@5%	0.49	0.48	0.56	0.60	0.51	0.47

T_1 = 2% Amla pomace powder, T_2 = 4% Amla pomace powder, T3 = 6% Amla pomace powder, NS = Non-significant and * = Significant at 5%.

4.8.2 Changes in sensory scores of *chikki* incorporated with amla pomace powder during storage at room temperature

The data regarding mean sensory scores of amla pomace powder incorporated *chikki* stored at room temperature (25±2 °C) is presented in Table 15. It was observed

that a significant difference was found between control and best accepted *chikki* variation T_3 (6%). As the storage period increased, there was a gradual decrease in the sensory parameters were observed from initial period to the end of storage period. Control sample had mean sensory scores of 7.57, 7.66, 7.57, 7.61, 7.57 and 7.57 for appearance, colour, texture, flavour, taste and overall acceptability respectively. Whereas, the experimental variation T_3 had scores of 7.52 for appearance, 7.42 for colour, 7.19 for texture, 7.33 for flavour, 7.47 for taste and 7.42 for overall acceptability at 45th day of storage. Statistically there was significant difference at five per cent level for sensory attributes like appearance, colour and texture for both control and experimental (T_3) sample. However, it was observed that the sensory parameters like flavour, taste and overall acceptability was found to be non-significant for experimental T_3 and significance difference was observed for control sample. It was evident from sensory scores that even at 45th day of storage period, T_3 samples were moderately liked by the panel members when compared with control sample.

Table 15: Sensory scores of *chikki* during storage at room temperature

Product	Duration	Appearance	Colour	Texture	Flavour	Taste	Overall acceptability
Control	Initial	8.28±0.56	8.42±0.59	8.47±0.60	8.42±0.67	8.61±0.49	8.52±0.60
	15th day	7.90±0.43	8.09±0.62	8.04±0.38	7.95±0.66	8.00±0.54	7.95±0.21
	30th day	7.66±0.48	7.85±0.47	7.71±0.46	7.85±0.65	7.76±0.53	7.71±0.46
	45th day	7.57±0.50	7.66±0.48	7.57±0.50	7.61±0.49	7.57±0.50	7.57±0.50
	F value	*	*	*	*	*	*
	SEm±	0.10	0.12	0.10	0.13	0.11	0.10
	CD@5%	0.30	0.33	0.30	0.38	0.32	0.28
T_3	Initial	8.14±0.65	8.14±0.65	8.00±1.00	7.80±1.07	7.71±1.05	7.85±0.85
	15th day	7.66±0.48	7.76±0.43	7.71±0.46	7.42±0.50	7.66±0.48	7.66±0.48
	30th day	7.57±0.50	7.57±0.50	7.57±0.50	7.38±0.49	7.52±0.51	7.57±0.50
	45th day	7.52±0.51	7.42±0.50	7.19±0.40	7.33±0.48	7.47±0.51	7.42±0.50
	F value	*	*	*	NS	NS	NS
	SEm±	0.11	0.11	0.13	0.15	0.14	0.13
	CD@5%	0.33	0.32	0.39	-	-	-

T_3 = 6% Amla pomace powder, NS = Non-significant and * = Significant at 5%.

Similar results were observed by Chetana (2011) who investigated quality evaluation of peanut *chikki* incorporated with flaxseeds and their shelf-life. The sensory scores of all desirable attributes decreased slightly at both room and accelerated conditions at the end of 30 days when compared to the initial values but were still acceptable. Hence, the mean sensory scores declined with the increase in the storage period in the present study too.

4.8.3 Changes in sensory scores of *chikki* incorporated with amla pomace powder during storage at refrigerated temperature

As indicated in Table 16, it was observed that control sample had scores of 7.47, 7.57, 7.47, 7.52, 7.52 and 7.52 for appearance, colour, texture, flavour, taste and overall acceptability, respectively. Among experimental samples it was observed that, T_3 sample had scores of 7.28 for appearance, 7.04 for colour, 7.04 for texture, 7.04 for flavour, 6.95 for taste and 7.09 for overall acceptability at the end of the 45th day. It was observed that T_3 was acceptable even at 45th day of storage period when compared to control. As the storage period increased sensory scores of the amla pomace *chikki* decreased. Statistically there was a significant difference at five per cent level for all the sensory parameters of control and experimental T_3 sample.

Table 16: Sensory scores of *chikki* during storage at refrigerated temperature

Product	Duration	Appearance	Colour	Texture	Flavour	Taste	Overall acceptability
Control	Initial	8.09±0.75	8.13±0.71	7.86±1.08	8.00±0.92	7.95±0.89	7.95±0.78
	15th day	7.72±0.63	7.72±0.63	7.54±0.85	7.54±0.59	7.59±0.79	7.77±0.61
	30th day	7.50±0.51	7.59±0.50	7.40±0.66	7.50±0.59	7.54±0.67	7.59±0.50
	45th day	7.40±0.50	7.45±0.50	7.27±0.55	7.40±0.79	7.40±0.66	7.40±0.50
	F value	*	*	NS	NS	NS	*
	SEm±	0.13	0.12	0.17	0.16	0.16	0.13
	CD@5%	0.36	0.35	-	-	-	0.36
T_1	Initial	7.59±0.66	7.40±0.59	7.59±0.73	7.68±0.77	7.50±0.67	7.59±0.59
	15th day	7.50±0.51	7.36±0.49	7.45±0.50	7.50±0.51	7.36±0.65	7.40±0.50
	30th day	7.27±0.63	7.18±0.66	7.18±0.50	7.18±0.50	7.22±0.68	7.13±0.56
	45th day	7.04±0.65	7.04±0.48	6.95±0.48	7.04±0.48	7.04±0.48	7.04±0.37
	F value	*	NS	*	*	NS	*
	SEm±	0.13	0.12	0.12	0.12	0.13	0.11
	CD@5%	0.37	-	0.34	0.35	-	0.30

T3 = 6% Amla pomace powder, NS = Non-significant and * = Significant at 5%.

4.8.4 Microbial load of best accepted *chikki* on storage

The microbial load of bacteria, yeast and mold for best accepted *chikki* is presented in Table 17. The microbial load was estimated at the intervals of the initial, 15^{th}, 30^{th} and 45^{th} day of storage.

Initially, there were no microbial counts of bacteria, yeast and mold. However, as the storage period increased the bacteria, yeast and mold counts also increased simultaneously. At the 45^{th} day of storage period, there was a significant increase in all the microbial counts. The bacteria, yeast and mold count were (1.33×10^5 cfu/g), (1.66×10^2 cfu/g), (1.66×10^3 cfu/g) and (1.66×10^5 cfu/g), (1.66×10^2 cfu/g), (1.66×10^3 cfu/g) both at room temperature as well as refrigerated temperature, respectively. However, it was observed that the microbial counts were within the permissible limits.

Table 17: Microbial load of *chikki* on storage

Storage condition	Duration	TBC ($\times10^5$cfu/g)	Yeast ($\times10^2$ cfu/g)	Mold ($\times10^3$cfu/g)
Room temperature	Initial	Nil	Nil	Nil
	15^{th} day	0.33	0.66	Nil1
	30^{th} day	1.33	1.33	1
	45^{th} day	2.33	1.66	1.66
	F value	*	*	*
	SEm±	0.28	0.28	0.16
	CD@5%	0.95	0.95	0.55
Refrigerated temperature	Initial	Nil	Nil	Nil
	15^{th} day	Nil	0.33	Nil
	30^{th} day	1	1	1
	45^{th} day	1.66	1.33	1.66
	F value	*	*	*
	SEm±	0.16	0.23	0.16
	CD@5%	0.55	0.78	0.55

NS = Non-significant, * = Significant at 5% and TBC = Total bacterial count.

In line with the above results, Devhare *et al*. (2021) reported an increase in total bacterial count during storage of *chikki*. The juice mixed peanut *chikki* showed initially low microbial growth 0×10^5 cfu/g, 1×10^5 cfu/g and the count of microorganisms increased to 2×10^5 cfu/g and 3×10^5 cfu/g, respectively at 90 days of storage which is similar to the present study. Also, Muttagi *et al.* (2014) reported substantially increase in the microbial load (bacteria, yeast and mold) of stored *chikki* throughout the storage period of 60 days, which is similar to present study. Statistically, it was observed that there was significant difference at 5 per cent for total bacterial count. However, their interaction effect was non-significant.

4.8.5 Effect of storage on moisture, peroxide value and ascorbic acid content of *chikki*

The best accepted *chikki* incorporated with amla pomace powder was analyzed for moisture, ascorbic acid and peroxide value during storage period at both room temperature (25±2 °C) and refrigerated temperatures (4 °C) as indicated in Table 18.

The moisture and peroxide value was found to be increased during storage period, whereas the ascorbic acid content is decreased as the storage period extended. The moisture content in both room and refrigerated *chikki* ranged between 4.64 to 5.23 and 4.64 to 5.15 per cent, respectively and peroxide values ranged between 2.27 to 5.89 and 2.37 to 5.17 meq/kg, respectively. Whereas the ascorbic content is ranged between 23.28 to 17.36 and 23.28 to 21.75 mg, respectively during 45 days of storage. Similar results were observed by Harsimart and Dhawan (2001) was reported where in gradual decrease in ascorbic acid content of guava bar during storage period.

In par with the present study, increased trend of moisture content and peroxide value in *chikki* was also reported by Hirdyani and Charak (2015) with moisture content of 3.4 - 3.8 per cent initially and 5.2 to 5.8 per cent in all the samples after 30 days of storage. The peroxide value of the samples ranged between 2.1-4.2 meq/kg initially and addition of flaxseeds increased the peroxide value of samples during storage (~12 meq/kg), thereby making the samples more prone to rancidity. In the present study

peroxide value was lower (2.27-5.89 meq/kg) than the peroxide value of *chikki* reported by Muttagi (2014) and it ranged from 14.16 - 18.10 meq/kg.

Hence, it can be concluded that there is a tendency of increase in moisture content and peroxide value and decrease in ascorbic acid content at the end of storage period and the reason can be attributed to increase in oxidation, variation in temperature and time.

Table 18: Effect of storage on moisture, peroxide value and ascorbic acid content of *chikki*

Product	Parameters						
	Duration	Moisture (%)		Ascorbic acid (mg)		Peroxide value (meq/kg)	
		RM	RF	RM	RF	RM	RF
Chikki	Initial	4.64	4.64	23.28	23.28	2.27	2.27
	15th day	4.75	4.69	22.48	23.15	3.46	3.15
	30th day	4.92	4.87	20.42	22.59	4.69	3.99
	45th day	5.23	5.15	17.36	21.75	5.89	5.17
	F value	*	*	*	*	*	*
	SEm±	0.02	0.02	0.05	0.03	0.01	0.02
	CD@5%	0.10	0.08	0.21	0.12	0.06	0.11

NS = Non-significant, * = Significant at 5%, RM = Room temperature and RF = Refrigerated temperature.

4.8.6 Consumer acceptability of *chikki*

The best accepted amla pomace powder incorporated *chikki* (T_3) was subjected to consumer acceptance for respondents (n=50) to know the extent of likability and dislikability. Table 19, represents the consumers acceptability using FACT scale of *chikki*. Nine statements were provided to test the acceptability, the results indicated that 44 per cent of the consumers “would eat every opportunity they had”, 12 per cent of the consumers ‘would eat this very often’, and then 14 per cent of consumers ‘would frequently eat this’. Also, it was observed that 12 per cent liked the product and would eat

it now and then, and 4 per cent respondents didn't like but would eat this on an occasion and 4 per cent felt they would hardly ever eat this.

Chikki was subjected to consumer acceptability for respondents (n=50), UAS, GKVK, Bengaluru to know their extent of likability and dislikability on five-point hedonic scale Fig. 8. It was observed that for appearance out of 50 respondents, 36 per cent of them rated as excellent and 40, 16 and 8 per cent, respectively for very good, good and fair. For texture 24, 40, 24 and 12 per cent respondents quoted as excellent, very good, good and fair, respectively. With respect to taste 30, 38, 26 and 6 per cent and for colour 26, 46, 18 and 10 per cent respondents quoted it as excellent, very good, good and fair, respectively. However, for flavour 16, 34, 30 and 20 per cent respondents quoted it as excellent, very good, good and fair, respectively. Lastly for overall acceptability 28, 44, 22 and 6 respondents quoted it as excellent, very good, good and fair, respectively. Hence, the amla pomace incorporated *chikki* was found to be acceptable among consumers.

Table 19: Consumer acceptability of *chikki* by using FACT Scale

Sl. No.	Opinion	No.	Percent
1.	I would eat every opportunity that I had	22	44
2.	I would eat this very often	6	12
3.	I would frequently eat this	7	14
4.	I like this and would eat it now and then	6	12
5.	I would eat if available but would not go out of my way	5	10
6.	I don’t like this but would eat this on an occasion	2	4
7.	I would hardly ever eat this	2	4
8.	I would eat this if there were no other food choices	0	0
9.	I would eat this only if forced	0	0
Total		50	100

Results obtained from the present study is in par with study conducted by Supriya *et al.* (2020) on consumer acceptability scores for quinoa incorporated products (quinoa upma and kesaribath). Similar findings were reported by Ajila *et al.* (2008) for biscuits substituted with 20 per cent mango peel powder, Noor Aziah and Komathi (2009) in a composite flour crackers produced with unripe mango peel and Kumari and Grewal (2007) for biscuits incorporated with 20 per cent carrot pomace.

4.9 Amla pomace powder incorporated *chutney* powder

The *chutney* powder was prepared using flaxseeds, bengal gram dhal, black gram dhal, red chilli, curry leaves, garlic and onion. In the present study, amla pomace powder was incorporated at 10, 15 and 20 per cent substitutional levels with *chutney* powder (Fig.4). *Chutney* powder without incorporation of amla pomace powder was treated as control.

4.9.1 Sensory evaluation of *chutney* powder

The mean sensory scores of *chutney* powder is indicated in Table 20. and Fig. 9. The results revealed that the control *chutney* powder had high scores for all sensory attributes when compared to amla pomace *chutney* powder.

Among experimental variations T_1 had highest sensory scores for appearance (7.59), colour (7.40), texture (7.59), flavour (7.68), taste (7.50) and overall acceptability (7.59). The variation T_3 (*chutney* powder) with 20 per cent amla pomace powder incorporation had a comparatively lower score for appearance (7.22), colour (7.00), texture (7.18), flavour (7.13), taste (7.00) and overall acceptability (7.09). However, overall acceptability of amla pomace *chutney* powder revealed that among experimental variations, T_1 (10 per cent amla pomace powder incorporation) with a score of 7.59 for overall acceptability. Statistically there was significant difference at 5 per cent level between the variations and all the sensory parameters.

Similar results were obtained by Netravati (2013) for flaxseed *chutney* powder substituted with flaxseeds in different ratios, received best overall acceptability and the scores are 7.67 for 50 per cent flaxseed incorporation and 7.33 for 100 per cent flaxseed

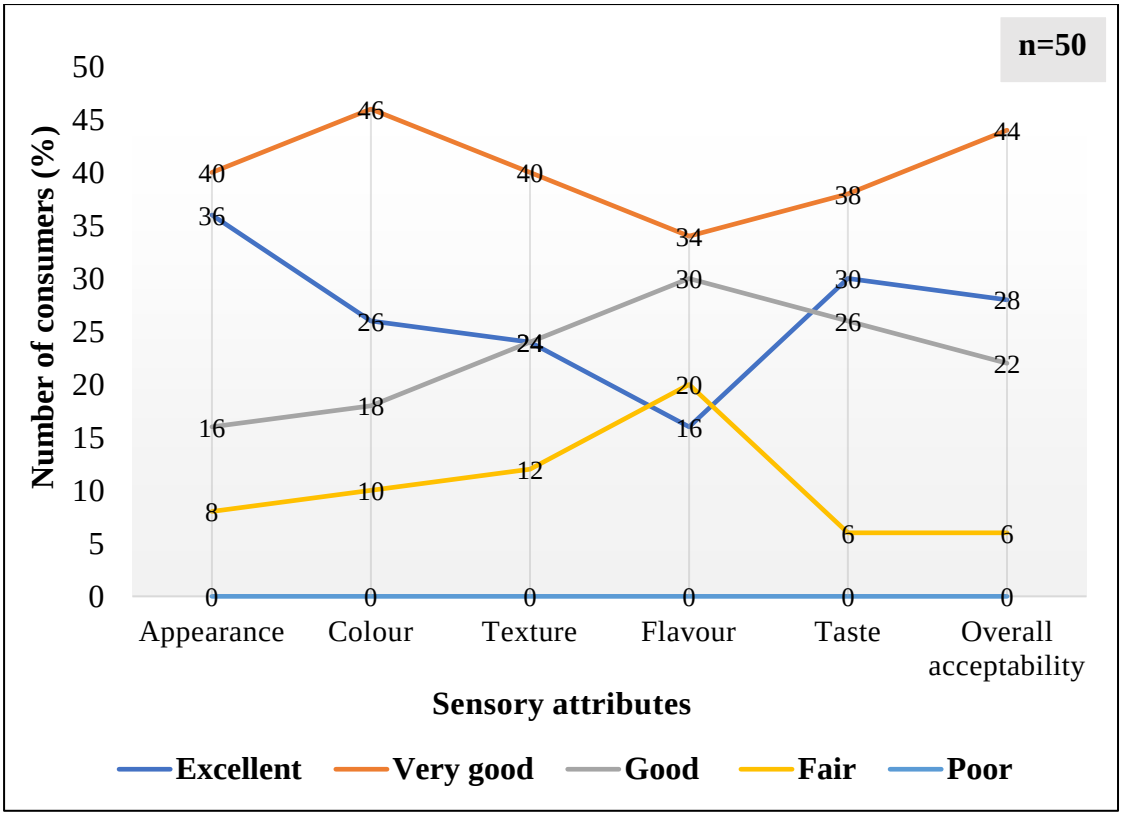

Fig. 8: Consumer acceptability of best accepted *chikki*

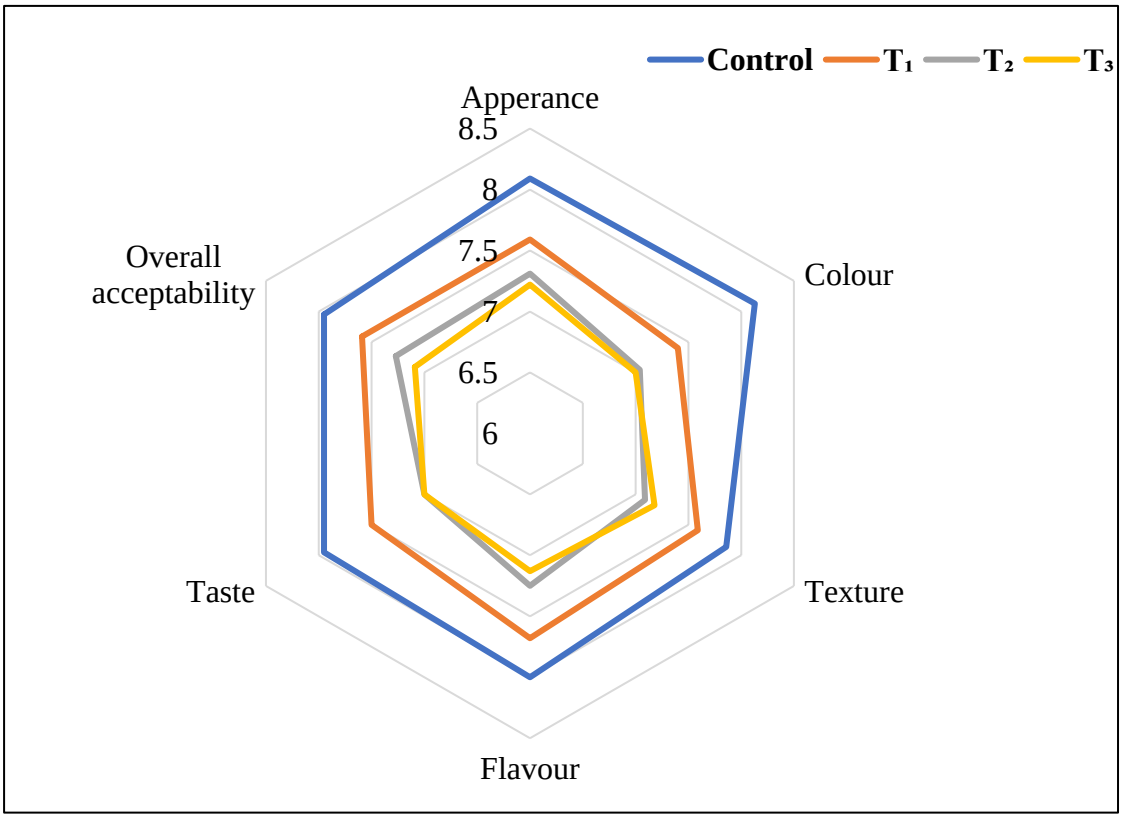

Fig. 9: Sensory scores of *chutney* powder

incorporation. Also, the results were in par with the investigation conducted by Singh and Awasthi (2003) wherein the acceptable level of green leafy vegetable powders incorporation in biscuits, *murukku*, *mathri* and *namakpare* was 15, 10, 20 and 10 per cent respectively.

Table 20: Sensory parameters of *chutney* powder incorporated with amla pomace powder

Treatments	Sensory parameters					
	Appearance	Colour	Texture	Flavour	Taste	Overall acceptability
Control	8.09±0.75	8.13±0.71	7.86±1.08	8.00±0.92	7.95±0.89	7.95±0.78
T_1	7.59±0.66	7.40±0.59	7.59±0.73	7.68±0.77	7.50±0.67	7.59±0.59
T_2	7.31±0.71	7.04±0.65	7.09±0.75	7.25±0.68	7.00±0.75	7.27±0.63
T_3	7.22±0.81	7.00±0.87	7.18±0.79	7.13±0.56	7.00±0.75	7.09±0.61
F value	*	*	*	*	*	*
SEm±	0.15	0.15	0.18	0.16	0.16	0.14
CD@5%	0.44	0.42	0.51	0.45	0.46	0.39

T_1 = 10% Amla pomace powder, T_2 = 15% Amla pomace powder, T_3 = 20% Amla pomace powder, NS = Non-significant and * = Significant at 5%.

4.9.2 Changes in sensory scores of *chutney* powder incorporated with amla pomace powder during storage at room temperature

The data regarding mean sensory scores of amla pomace powder incorporated *chutney powder* stored at room temperature (25±2 °C) is presented in Table 21. It was observed that a significant difference was found between control and best accepted *chutney* variation T_1 (10%). As the storage period increased, gradual decrease in the sensory parameters was observed from initial period to the end of storage period. Control

sample had mean sensory scores of 7.50, 7.54, 7.36, 7.50, 7.50 and 7.50 for appearance, colour, texture, flavour, taste and overall acceptability, respectively. Whereas, the experimental variation T_1 had scores of 7.09 for appearance, 7.18 for colour, 7.0 for texture, 7.31 for flavour, 7.09 for taste and 7.40 for overall acceptability at 45^{th} day of storage. Statistically there was significant difference at five per cent level for sensory attributes like texture, flavour, taste and overall acceptability for control sample and appearance, colour, flavour, taste and overall acceptability for experimental (T_1) sample. However, it was observed that the sensory parameters like appearance and colour of control sample and texture of experimental T_1 sample was found to be non-significant. It was evident from sensory scores that even at 45^{th} day of storage period, T_1 sample was moderately liked by the panel members when compared with control sample.

The findings were on par with the study investigated by Deepak (2016) on niger seed *chutney* powder incorporated with dehydrated tamarind leaf powder and the results revealed that as the number of days increased, the sensory scores of all sensory parameters decreased. It was observed that from initial day up to 30^{th} day, there was significant difference found in colour ranged between (7.60 to 6.20), taste (7.65 to 5.80) and overall acceptability ranged between (7.75 to 5.45). Similar findings were reported by Khedkar *et al.* (2019) sensory scores for curry leaf *chutney* powder 7 (excellent) to 8.2 (very good) for overall acceptance, 8.8 to 8.1 for flavour, 8.7 to 8.2 for taste, 8.8 to 8.3 for colour respectively.

The results were found in par with the study conducted by Rao *et al.* (2013) on storage of flax seed *chutney* powder. Overall sensory quality of flax seed *chutney* powder scored good (7.4) even after 6 months of storage. Hence, as per the above-mentioned studies, the mean scores were declined with the increase in the storage period in the present study too.

Table 21: Sensory scores of *chutney* powder during storage at room temperature

Product	Duration	Appearance	Colour	Texture	Flavour	Taste	Overall acceptability
Control	Initial	8.09±0.75	8.13±0.71	7.86±1.08	8.00±0.92	7.95±0.89	7.95±0.78
	15th day	7.81±0.58	7.77±0.61	7.59±0.85	7.63±0.90	7.68±0.83	7.86±0.71
	30th day	7.59±0.50	7.68±0.47	7.45±0.67	7.59±0.66	7.68±0.77	7.68±0.47
	45th day	7.50±0.51	7.54±0.50	7.36±0.58	7.50±0.67	7.50±0.74	7.50±0.51
	F value	*	*	NS	NS	NS	NS
	SEm±	0.12	0.12	0.17	0.17	0.17	0.13
	CD@5%	0.35	0.35	-	-	-	-
T_1	Initial	7.59±0.66	7.40±0.59	7.59±0.73	7.68±0.77	7.50±0.67	7.59±0.59
	15th day	7.31±0.56	7.36±0.58	7.54±0.50	7.54±0.73	7.45±0.59	7.54±0.50
	30th day	7.22±0.68	7.31±0.47	7.13±0.71	7.50±0.51	7.36±0.49	7.45±0.50
	45th day	7.09±0.68	7.18±0.50	7.00±0.61	7.31±0.56	7.09±0.68	7.40±0.59
	F value	NS	NS	*	NS	NS	NS
	SEm±	0.13	0.11	0.13	0.14	0.13	0.11
	CD@5%	-	-	0.39	-	-	-

T_1 = 10% Amla pomace powder, NS = Non-significant and * = Significant at 5%.

4.9.3 Changes in sensory scores of *chutney* powder incorporated with amla pomace powder during storage at refrigerated temperature

As indicated in Table 22, it was observed that control sample had scores of 7.40 for appearance, 7.45 for colour, 7.27 for texture, 7.40 for flavour, 7.40 for taste and 7.40 for overall acceptability. Among experimental samples it was observed that, T_1 sample had scores of 7.04 for appearance, 7.04 for colour, 6.95 for texture, 7.04 for flavour, 7.04 for taste and 7.04 for overall acceptability at the end of the 45th day. It was observed that T_1 was acceptable even at 45th day of storage period when compared to control. This amla pomace *chutney* powder which is having 10 per cent amla pomace powder can be a good functional adjunct in the daily dietary as it is good source of dietary fibre and, as the major ingredient of *chutney* powder is flax seed (45%) that can be a good source of omega-3 fatty acid and dietary fibre also.

Statistically there was significant difference at five per cent level for sensory attributes like appearance, colour and overall acceptability for control sample and appearance, texture, flavour and overall acceptability for experimental (T_1) sample. However, it was observed that the sensory parameters like texture, flavour and taste of control sample and colour, taste of experimental T_1 sample was found to be non-significant. Hence, as the storage period increased sensory scores of the amla pomace *chutney* powder decreased.

Table 22: Sensory scores of *chutney* powder during storage at refrigerated temperature

Product	Duration	Appearance	Colour	Texture	Flavour	Taste	Overall acceptability
Control	Initial	8.09±0.75	8.13±0.71	7.86±1.08	8.00±0.92	7.95±0.89	7.95±0.78
	15th day	7.72±0.63	7.72±0.63	7.54±0.85	7.54±0.59	7.59±0.79	7.77±0.61
	30th day	7.50±0.51	7.59±0.50	7.40±0.66	7.50±0.59	7.54±0.67	7.59±0.50
	45th day	7.40±0.50	7.45±0.50	7.27±0.55	7.40±0.79	7.40±0.66	7.40±0.50
	F value	*	*	NS	NS	NS	*
	SEm±	0.13	0.12	0.17	0.16	0.16	0.13
	CD@5%	0.36	0.35	-	-	-	0.36
T_1	Initial	7.59±0.66	7.40±0.59	7.59±0.73	7.68±0.77	7.50±0.67	7.59±0.59
	15th day	7.50±0.51	7.36±0.49	7.45±0.50	7.50±0.51	7.36±0.65	7.40±0.50
	30th day	7.27±0.63	7.18±0.66	7.18±0.50	7.18±0.50	7.22±0.68	7.13±0.56
	45th day	7.04±0.65	7.04±0.48	6.95±0.48	7.04±0.48	7.04±0.48	7.04±0.37
	F value	*	NS	*	*	NS	*
	SEm±	0.13	0.12	0.12	0.12	0.13	0.11
	CD@5%	0.37	-	0.34	0.35	-	0.30

T_1 = 10% Amla pomace powder, NS = Non-significant and * = Significant at 5%.

4.9.4 Microbial load of *chutney* powder on storage

The microbial load of bacteria, yeast and mold for best accepted *chutney* powder with 10 per cent amla pomace powder is presented in Table 23. The microbial load was estimated at the intervals of the initial, 15th, 30th and 45th day of storage.

Initially, there were no microbial counts of bacteria, yeast and mold. However, as the storage period increased the bacteria, yeast and mold counts also increased simultaneously. At the end of the storage period, there was a significant increase in all the microbial counts. The bacteria, yeast and mold count were (2.33×10^5 cfu/g), (1.66×10^2 cfu/g), (2.33×10^3 cfu/g) at room temperature and microbial counts were (2×10^5 cfu/g), (1.33×10^2 cfu/g) (1.66×10^3 cfu/g) refrigerated temperature, respectively. Though the microbial counts were increased, it was observed to be within the permissible limits and *chutney* powder was good for consumption. Statistically significant difference at five per cent level was observed during storage period at different time intervals.

Table 23: Microbial load of *chutney* powder on storage

Storage condition	Duration	Bacteria ($\times10^5$cfu/g)	Yeast ($\times10^2$ cfu/g)	Mold ($\times10^3$cfu/g)
Room temperature	Initial	Nil	Nil	Nil
	15th day	0.66	0.33	0.66
	30th day	1.66	1.33	1.66
	45th day	2.33	1.66	2.33
	F value	*	*	*
	SEm±	0.28	0.28	0.40
	CD@5%	0.95	0.95	1.35
Refrigerated temperature	Initial	Nil	Nil	Nil
	15th day	0.33	0.33	Nil
	30th day	1	1.33	1
	45th day	2	1.33	1.66
	F value	*	*	*
	SEm±	0.16	0.28	0.16
	CD@5%	0.55	0.95	0.52

NS = Non-significant, * = Significant at 5% and TBC = Total bacterial count.

The study conducted by Khedkar *et al.* (2019) are in line with the present results wherein there was no microbial growth was observed on initial day in curry leaf *chutney* powder. However, at the end of storage period growth of aerobic microorganisms 2.5 x 10^3 cfu/g as well as yeast and mold 2.3 x 10^3 cfu/g was observed. Another study conducted by Deepak (2016) on the storage of dehydrated tamarind leaf *chutney* powder indicated increased bacterial growth of 5.33 x 10^4 cfu/g at the end of 30^{th} day of storage study which is higher than the present study.

Statistically, it was observed that there was significant difference at 5 per cent level for total bacterial, yeast and mold count.

4.9.5 Effect of storage on moisture, peroxide value and ascorbic acid content of *chutney* powder

The best accepted *chutney* powder with 10 per cent amla pomace powder was analyzed for moisture, peroxide value and ascorbic acid during storage period at both room temperature (25±2 °C) and refrigerated temperatures (4 °C) and the results indicated in Table 24.

The moisture and peroxide value was found to be increased during storage period, whereas the ascorbic acid content is decreased as the storage period extended. The moisture content in both room and refrigerated *chutney* powder ranged between 4.50 to 5.18 and 4.50 to 5.06 per cent, respectively and peroxide values ranged between 2.38 to 6.35 and 2.38 to 5.43 meq/kg, respectively. Whereas the ascorbic content is ranged between 42.60 to 35.91 and 42.60 to 39.55 mg, respectively during 45 days of storage. There is a tendency of increase in moisture content and peroxide value and decrease in ascorbic acid content at the end of storage period and the reason can be attributed to increase in oxidation, variation in temperature and time.

Results obtained in the present study is lower than the study conducted by Netravati (2013) wherein the peroxide value in control *chutney* powder ranged from 5.89 to 22.58 meq/kg and same increasing trend was observed for flaxseed *chutney* powder and the values ranged from 4.81 to 20.08 meq/kg during 90 days of storage period.

Also, increased peroxide value (12.3 to 18.3 meq/kg) was reported by Rajiv *et al.* (2012) from the first day of storage to 90th day of storage for cookies prepared with 15 per cent incorporation of flaxseed flour with wheat flour. In present study, developed products had lower peroxide value.

Table 24: Effect of storage on moisture, peroxide value and ascorbic acid content of *chutney* powder

Product	Parameters						
	Duration	Moisture (%)		Ascorbic acid (mg)		Peroxide value (meq/kg)	
		RM	RF	RM	RF	RM	RF
Amla pomace *Chutney* Powder	Initial	4.50	4.50	42.60	42.60	2.38	2.38
	15th day	4.54	4.52	40.79	41.33	3.28	3.14
	30th day	4.95	4.81	38.21	40.74	4.84	4.16
	45th day	5.18	5.06	35.91	39.55	6.35	5.43
	F value	*	*	*	*	*	*
	SEm±	0.02	0.01	0.02	0.03	0.05	0.03
	CD@5%	0.10	0.06	0.09	0.12	0.20	0.15

NS = Non-significant, * = Significant at 5%, RM = Room temperature and RF = Refrigerated temperature.

4.9.6 Consumer acceptability of *chutney* powder

The best accepted amla pomace powder incorporated *chutney* powder (T_1) was subjected to consumer acceptance for respondents (n=50) to know the extent of likability and dislikability. Table 25, represents the consumer acceptability using FACT scale. Nine statements were provided to test the acceptability, the results indicated that 38 per cent of the consumers "would eat every opportunity they had", 16 per cent of the consumers 'would eat this very often', and then 12 per cent of consumers 'would frequently eat this'. Also, it was observed that 14 per cent liked the product and would eat it now and then, and 6 per cent respondents didn't like but would eat this on an occasion, 6 per cent would

eat if available but would not go out of their way, 4 per cent would eat this if there were no other food choices and 4 per cent felt they would hardly ever eat this.

Chutney powder was also subjected to consumer acceptability for respondents (n=50), UAS, GKVK, Bengaluru to know their extent of likability and dislikability using a five-point hedonic scale (Fig. 10). It was observed that for appearance out of 50 respondents, 32 per cent of them rated as excellent and 44, 18 and 6 per cent of the respondents rated for very good, good and fair. For texture 28, 40, 20 and 12 per cent respondents indicated amla pomace *chutney* powder as excellent, very good, good and fair, respectively. With respect to taste 24, 38, 30 and 8 per cent, respectively and for colour 20, 46, 24 and 10 per cent respondents quoted it as excellent, very good, good and fair, respectively. However, for flavour 26, 34, 30 and 10 per cent respondents rated it as excellent, very good, good and fair, respectively. Lastly for overall acceptability 30, 44, 22 and 4 per cent respondents inferred it as excellent, very good, good and fair, respectively. Hence, the amla pomace incorporated *chutney* powder was found to be accepted very well among consumers.

Table 25: Consumer acceptability of *chutney* powder by using FACT Scale

Sl. No.	Opinion	No.	Percent
1.	I would eat every opportunity that I had	19	38
2.	I would eat this very often	8	16
3.	I would frequently eat this	6	12
4.	I like this and would eat it now and then	7	14
5.	I would eat if available but would not go out of my way	3	6
6.	I don’t like this but would eat this on an occasion	3	6
7.	I would hardly ever eat this	2	4
8.	I would eat this if there were no other food choices	2	4
9.	I would eat this only if forced	0	0
Total		50	100

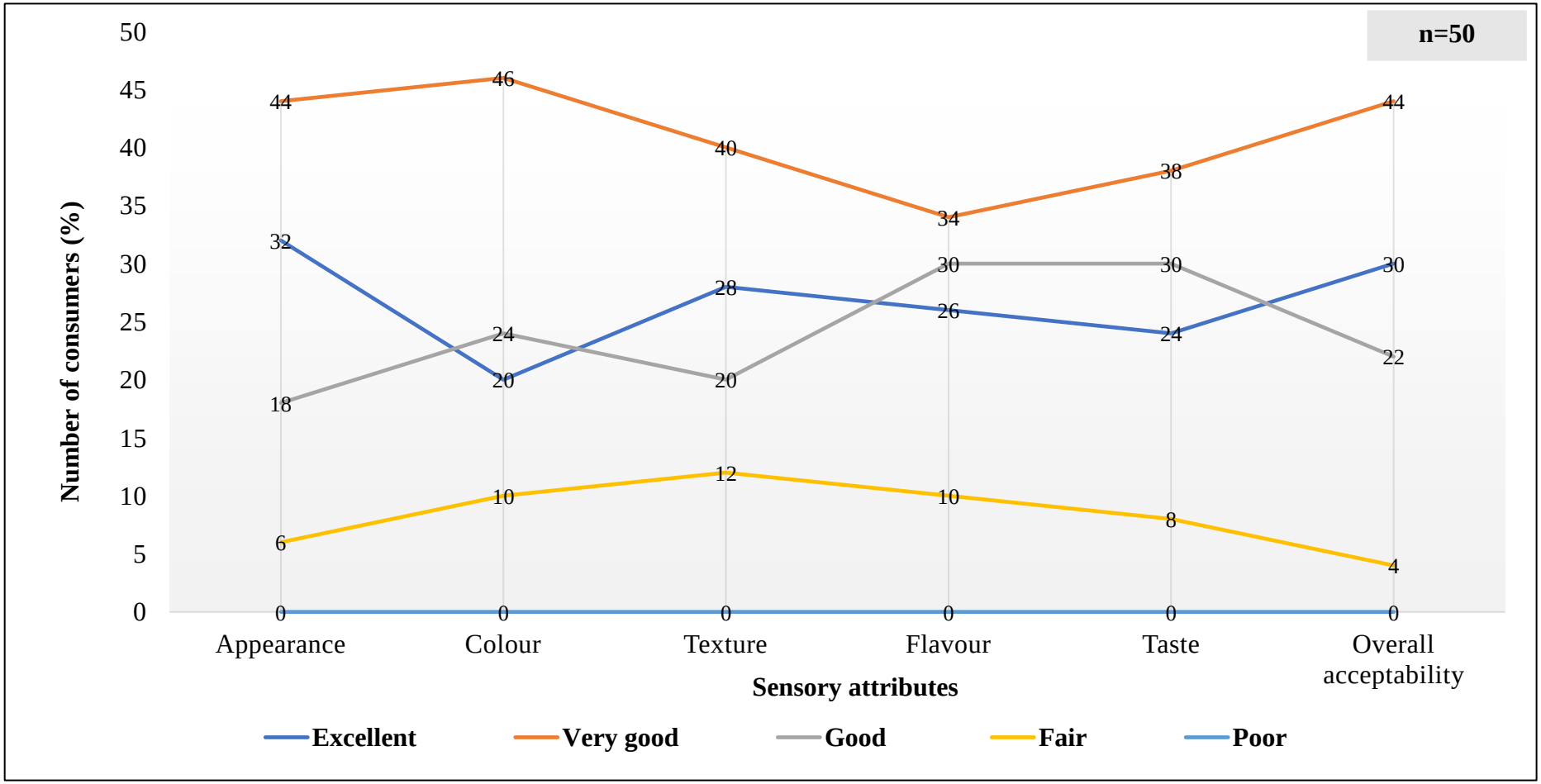

Fig. 10: Consumer acceptability of best accepted *chutney* powder

The results of the present investigation was higher than the study conducted by Singh and Kulshrestha (2008) who incorporated carrot pomace powder into different products and out of all products, enriched halwa showed (10%) liked extremely, (50%) liked very much, (20%) liked slightly. Results were in par with study conducted by Nagi *et al.* (2012) reported that biscuits containing (20%) wheat bran for mass consumer acceptability, (51%) consumers rated the product as excellent whereas (39%) and (10%) rated it as very good and good, respectively.

4.10 Nutritional content for best accepted products

4.10.1 Nutritional content of best accepted amla pomace *chikki* (Per 100g)

The nutritional composition for the best accepted products was computed as per the guidelines of Indian Food Composition Tables, NIN, ICMR Hyderabad (Longvah *et al*., 2017). The nutrient composition of best accepted *chikki* is presented in Table 26.

The best accepted *chikki* had protein, fat and calorific value of 10.2 g, 15.6 g and 427 Kcal per 100 g which is slightly lower than the control *chikki* with values being 11.6 g, 17.9g and 456 Kcal for protein, fat and calorific value respectively. The ash and carbohydrate content were found to be same 1.9 g per 100 g in both products. The dietary fibre fractions, *viz*, total dietary fibre, insoluble dietary fibre and soluble dietary fibre was found to be 6.5, 5 and 1.6 g, respectively which is higher than that of control *chikki* with values of dietary fibre fractions being total dietary fibre (4.7g), insoluble dietary fibre (3.9g) and soluble dietary fibre (0.8g) per 100 g. The ascorbic acid content was 26 mg/100g of *chikki* with 6 per cent of amla pomace powder where as it is nil in the control *chikki*.

Similar results were detected by Devhare *et al.* (2020) for nutritional composition of *chikki* containing pomegranate peel flour and results revealed that protein (14.17g), ash (1.03g), fat (23.78g), crude fibre (2.10g) and carbohydrate (51g) in control and protein (14.16g), ash (1.02g), fat (23.73g), crude fibre (2.70g) and carbohydrate (51g), respectively which was similar compared with control.

Also, Narwade (2018) prepared ragi *burfi* which contains 9.58 g protein, 1.56 g ash, 20.15 g fat and 5.06 g dietary fibre.

Hence, amla pomace *chikki* had adequate amount of ascorbic acid and dietary fibre in comparison with control *chikki.*

Table 26: Nutrient computation of best accepted *chikki* (Per 100g)

Nutrients	Control	Best accepted product (T_3)
Protein (g)	11.6	10.2
Ash (g)	1.9	1.9
Fat (g)	17.9	15.6
TDF (g)	4.7	6.5
IDF (g)	3.9	5.0
SDF (g)	0.8	1.6
Carbohydrates (g)	50	50
Energy (kcal)	456	427
Ascorbic acid (mg)	00	26

T_3 = 6% Amla pomace powder, IDF = Insoluble dietary fibre, SDF = Soluble dietary fibre and TDF = Total dietary fibre

4.10.2 Nutritional content of best accepted *chutney* powder (Per 100g)

The nutritional composition for the best accepted products was computed as per the guidelines of Indian Food Composition Tables, NIN, ICMR Hyderabad (Longvah *et al*., 2017). The nutritional composition of best accepted *chutney* powder was presented in Table 27.

The best accepted amla pomace *chutney* powder had protein and fat content of 16.5 g, and 17.6 g per 100g which is slightly lower than the control *chutney* powder with values being 17.6 g and 18 g for protein and fat, respectively. The calorific value and carbohydrate content were found to be 322 Kcal and 23 g per 100g which is lower than the control *chutney* powder with energy (347 Kcal) and carbohydrate (27g). The dietary

fibre fractions, *viz*, total dietary fibre, insoluble dietary fibre and soluble dietary fibre was found to be 22.9, 17.8 and 4.9g, respectively which is higher than that of control *chutney* powder with values of dietary fibre fractions being total dietary fibre (20.1g), insoluble dietary fibre (16.8g) and soluble dietary fibre (3.8g) per 100g. The ash was found to be similar in both the control (3g/100g) and experimental (3g/100g) variations. The ascorbic acid content was 43.6 mg /100g of *chutney* powder with 10 per cent of amla pomace powder where as it is 0.4 mg/100g in the control *chutney* powder. Hence, there is a strikable difference with respect to fibre composition and ascorbic acid content.

Shanthala and Prakash (2005) reported dried curry leaf powder (CLP) incorporated product (chapati) contained protein (12.5g), ash (9.7g), fat (5.4g), insoluble fibre (55.6g), soluble fibre (4.4g). The protein and fat content were lower, whereas total ash and insoluble fibre are higher and soluble fibre is similar, compared with present study. The results obtained in the present study was lower than that recorded by Singh *et al.* (2013) for beta-Carotene rich *laddu* using rice bran, except for beta-carotene content. The protein, fat, ash, carbohydrate, crude fibre, vitamin C and beta carotene was 7.80 g, 19.42 g, 1.18 g, 66 g, 1.14 g, 1 mg and 1.89 mg.

Table 27: Nutrient computation of best accepted *chutney* powder (Per 100g)

Nutrients	**Control**	**Best Accepted product (T_1)**
Protein (g)	17.6	16.5
Fat (g)	18	17.6
Ash (g)	3	3
TDF (g)	20.1	22.9
IDF (g)	16.8	17.8
SDF (g)	3.8	4.9
Carbohydrates (g)	27	23
Energy (kcal)	347	322
Ascorbic acid (mg)	0.4	43.6

T_1 = 10% Amla pomace powder, IDF = Insoluble dietary fibre, SDF = Soluble dietary fibre and TDF = Total dietary fibre.

4.11 Cost estimation for the developed products

The cost of production is an important consideration for commercialization and successful marketing. The cost of any product depends upon a number of variable factors like cost of raw materials, cost of processing and packaging of the product, *etc*. Here, the approximate cost of best accepted products (Per 100gm) is indicated. Overhead charges at 30 per cent of expenditure on manufacturing, which includes labour cost, depreciation cost on machinery, equipment, building *etc*., and profit at 25 per cent was included.

The production cost of *chikki* is indicated in Table 28. And the results revealed that the total production cost was found to be Rs. 16 per 100g. The cost of the product was found to be lesser than that of the cost of *chikki* calculated by Bukya *et al.* (2017).

Table 28: Production cost of *chikki*

Ingredients	**Quantity (g)**	**Rate (Rs.)**	**Cost (Rs.)**
Jaggery	50	60/kg	3
Peanuts	39	140/kg	5.46
Ghee	5	405/kg	2.02
Amla pomace	6	-	-
Total	100		10.48
Overhead charges @30%			3.14
Profit (25%)			2.62
Cost of the product			16.24
	Round off to **Rs. 16**		

The product cost of *chutney* powder incorporated with amla pomace powder is presented in Table 29.

Table 29: Production cost of *chutney* powder

Ingredients	Quantity (g)	Rate (Rs.)	Cost (Rs.)
Flax seed	45	170/kg	7.65
Black gram dhal	15	132/kg	1.98
Bengal gram dhal	15	89/kg	1.33
Red chilly	08	120/kg	0.96
Curry leaves	02	100/kg	0.2
Garlic	02	140/kg	0.28
Salt	03	18/kg	0.05
Amla pomace powder	10	-	-
Total	100		12.45
Overhead charges @30%			3.73
Profit (25%)			3.11
Cost of the product			19.29
	Round off to **Rs. 19**		

The results revealed that the cost of the product was found to be Rs. 19 per 100g which is lesser than the cost calculated by Fathima (2018) for banana blossom *chutney* powder with cost of Rs. 30 per 100g. Hence, it can be concluded that the production cost of *chikki* and *chutney* powder were found to be economical when compared to other studies.

V SUMMARY

During processing of Amla, extraction of amla juice results in lot of wet amla residue or amla pomace as a by-product that is comprised of lots of nutrients. To obtain Amla juice, amla seeds and pomace will be separated. Amla pomace contains phytonutrients such as ascorbic acid, tannins, polyphenols and dietary fibre. Also it is a store house of minerals especially calcium, phosphorus, sodium and magnesium. These bioactive compounds are known to have many health benefits and therapeutic value in human when consumed. These bioactive compounds in the amla pomace makes this by-product as a potentially viable and valuable raw material for the development of novel and functional food products which otherwise would be generally discarded as waste that can add upon to environmental pollution. Hence, the present study entitled 'Nutritional evaluation of Amla pomace and its value added products' was undertaken to exploit the nutritional potential and to utilize amla pomace as such and also to develop value added products with the objectives that focused on the determination of physico-functional properties, nutrient composition, product development, sensory evaluation and shelf-life study of the developed products.

The scientific findings of the research entitled **"Nutritional evaluation of Amla (*Phyllanthus emblica*) pomace and its value added products"** are summarized as follows:

- The amla fruits procured from a local market were cleaned, washed, sliced, ground, and filtered. During processing of Amla juice, amla seeds and pomace will be separated that constituted to 25-28 per cent of total fruit, out of which 8-9 per cent is seeds and 17-19 per cent is fresh amla pomace.

- Hundred grams of fresh amla pomace was dried and in laboratory model ezidri tray dryer at 45° C for 4 hours to get 32.25 g of dried pomace. The dried pomace was ground to get fine powder and stored in air tight zip lock covers at room temperature for further use.

- The physico-functional properties of the amla pomace powder such as tristimulus colour was found to be L^* = 86.47, a^* = -0.74 and b^* = 11.13, pH 3.44, particle

density 1.23 g/cm^3, water holding capacity 12.30 g/g, water binding capacity 12.37 g/g and swelling capacity 13 ml/g. The hydration properties of amla pomace powder increased with increase in particle size.

- The nutrient composition of amla pomace powder indicated low calorific value with low carbohydrate content and the values being, moisture 4.99 g, protein 1.55 g, ash 2.60 g, fat 0.18 g, carbohydrate 7 g and energy 36 kcal.
- Amla pomace is an abundant source of fibre. The crude fibre was found to be 13.39 g/100g. The dietary fibre fractions, *viz*, Total dietary fibre (TDF), Insoluble dietary fibre (IDF) and Soluble dietary fibre (SDF) was found to be 41.7, 27.4 and 14.5 g, respectively.
- Phytonutrients of amla pomace powder showed that 677 mg of polyphenols, 524 mg of tannins and 432 mg of ascorbic acid.
- The mineral composition indicated that amla pomace powder had 128 mg of calcium, 116 mg of phosphorous, 92.2 mg of sodium, 48 mg of magnesium, 2 mg of manganese, 1.34 mg of copper, 1.12 mg of iron and 0.92 mg of zinc.
- Amla pomace powder was subjected to sensory evaluation and it was observed to best accepted with good scores for appearance (7.50), colour (7.68), texture (7.09), aroma (6.68), taste (6.77) and overall acceptability (7.18).
- Three variations of *chikki* were prepared by incorporating amla pomace powder at different levels *i.e.* 2, 4, and 6 per cent, respectively. The variation of *chikki* with 6 per cent was found to be best accepted with good scores for appearance (8.14), colour (8.14), texture (8.00), aroma (7.80), taste (7.71) and overall acceptability (7.85).
- Three variations of *chutney* powder was prepared by incorporating amla pomace powder at different levels 10, 15 and 20 per cent. *Chutney* powder prepared at 10 per cent incorporation of amla pomace powder was found to be best accepted with good scores for appearance (7.59), colour (7.40), texture (7.59), flavour (7.68), taste (7.50) and overall acceptability (7.59).

- Three variations of *chikki* were formulated and the best accepted *chikki* was found to be the one with 6 per cent amla pomace powder that was kept for storage study at both room and refrigerated temperature for a period of 45 days. Results indicated a significant difference with respect to all sensory parameters of control *chikki* and also in best accepted variation. The sensory attributes like appearance, colour, texture were found to be significant whereas flavour, taste, overall acceptability was non-significant.

- In refrigerated condition, the sensory scores of both control and best accepted *chikki* (6% amla pomace powder) was found to be statistically significant. As the storage period increased there was a gradual decrease in the scores of sensory parameters.

- *Chutney* powder incorporated with amla pomace powder (10%) and control *chutney* powder were stored room temperature and results showed that sensory attributes like appearance, colour, flavour, taste, overall acceptability were found to be non-significant, however texture was found significant in room conditions.

- In refrigerated condition with respect to the best accepted amla pomace *chutney* powder (10%), appearance, texture, flavour, overall acceptability was found to be significant whereas colour and taste found to be non-significant. As the storage period increased, gradual decrease in the sensory scores was observed from initial stage to the 45^{th} day of storage period.

- Initially, in amla pomace powder, the microbial count of bacteria, yeast and mold were not found. As the storage period increased, the bacteria, yeast and mold counts were increased during 45 days of storage study. However, the increase in microbial counts were in the permissible limits.

- With respect to amla pomace based value added products *chikki* and *chutney* powder, it was observed that there was increase in microbial counts at the end of storage period.

- The peroxide value of best accepted products was decreased and it was within permissible limits. It was noticed that there was slight decrease in ascorbic acid

content in both products *chikki* (17.36mg and 21.75mg) and *chutney* powder (35.91mg and 39.55mg) at both room and refrigerated temperatures. Also, the moisture content of *chikki* and *chutney* powder gradually decreased at the end of the storage period.

- The computed nutritional value of *chikki* incorporated with amla pomace indicated 10.2 g protein, 15.6 g fat, 1.9 g ash, 427 Kcal energy, 50 g carbohydrate, 5 g IDF, 1.6 g SDF, 6.5 g TDF and 25.9 mg ascorbic acid for per 100 g. Amla pomace powder (10%) incorporated *chutney* powder indicated better values for protein (16.5g), fat (17.6g), ash (3g), energy (322 Kcal), carbohydrate (27g), IDF (17.8g), SDF (4.9g), TDF (22.9g) and ascorbic acid (43.6mg) per 100 g.

- The cost of production of *chikki* and *chutney* powder incorporated with amla pomace powder was found to be Rs. 16 and Rs. 19 per 100 g, respectively.

Conclusion

Amla pomace offers a tremendous potential for its applications in food and pharmaceutical industries as nutraceutical especially the dietary fibre. Amla pomace is a good source of dietary fibre and also rich in phytonutrients like ascorbic acid, total phenols and tannins. It is also a good source of minerals like calcium, phosphorus, sodium and magnesium. Amla pomace was acceptable with respect to all sensory parameters. The developed value added products such as *chikki* and *chutney* powder bt incorporating amla pomace powder enhanced nutritional value especially dietary fibre, minerals and ascorbic acid. Thus, the consumption of amla pomace and amla pomace incorporated products can be encouraged and health benefits of amla pomace can be exploited.

Future line of work

- Creating awareness about amla by-products utilization for value addition.
- Studies on nutritional composition and storage for value added products to be carried out by using different packaging materials for longer storage period.
- Scope for commercial or industrial use can be explored.
- Therapeutic use of amla pomace powder can be explored due to its high fibre and high phytonutrient content.

Limitations

- Proximate composition of the developed products could not be analysed.
- Storage studies could not be carried out for a longer duration for developed products due to time constraint.

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ANNEXURES

ANNEXURE-I

ESTIMATION OF PROTEIN (AOAC, 1980)

Principle:

Organic nitrogen digested with sulphuric acid in the presence of catalysts is converted to ammonium sulphate. Ammonium liberated by making the solution alkaline is distilled into a known volume of standard acid, which is then back titrated. Protein percent was calculated by multiplying the nitrogen percent by the factor 6.25.

Reagents

- 4% boric acid solution: 40g of boric acid was dissolved in some distilled water. The solution was then transferred to a 1000ml volumetric flask and made up to the mark.
- 40% NaOH (w/v)
- 0.1N HCl
- Mixed indicator: was made by mixing methyl red (0.2%) and Bromocresol green (02%) in a 1:2 ratios (v/v) respectively
- Concentrated sulphuric acid
- Digestion mixture: Anhydrous Sodium Sulphate and Copper Sulphate 4:1 ratio respectively.

Procedure

Digestion

0.5g of the sample was weighed into the digestion tube of the Gerhard digestion unit in duplicate and 5g of digestion mixture was added to each tube. 20ml of concentrated sulphuric acid was also added and samples were digested until the contents of the tubes became green colour. After digestion process samples were allowed to cool and transferred to the 100ml volumetric flask and made up to the mark with distilled water.

Distillation

10ml of each sample was transferred into the distillation tube of the automatic Gerhard unit and 20ml of 4% boric acid to which 3-4 drops of the mixed indicator was added and placed in the collecting conical flask to trap the liberated ammonia. The unit was furnished with 40 per cent NaOH and distilled water to facilitate operation. Distillation was done for 6 minutes and boric acid turned colour from reddish pink to green as it collects the ammonia.

Titration

The green coloured boric acid was titrated against the 0.1N HCl until its coloured turned to pink. A blank was run simultaneously. The titre value obtained was incorporated in the equation below to obtain the percent of the nitrogen present in the sample which in turn was multiplied by the factor 6.25 to obtain the protein per cent.

$$\text{Protein (g/100g)} = \frac{\text{Titre value} \times \text{Normality of HCL} \times 14.001 \times 6.25}{\text{Sample weight} \times 1000} \times 100$$

ANNEXURE-II

ESTIMATION OF FAT (AOAC, 1980)

Principle

The extraction of fat from substances is often tedious and requires thorough contact and heating with the solvent. This is done in the Soxhlet apparatus in which fresh solvent continuously comes into contact with the material to be extracted over a relatively long period of time.

Procedure

Five grams of sample was weighed into a thimble and closed with fat-free cotton wool. The thimble was placed in the Soxhlet apparatus attached to a pre-weighed flask and extracted for about 14-26 hours. Thereafter, the flask was retrieved from the apparatus with as little solvent in it as was possible. It was then transferred into an oven to evaporate the remaining solvent, leaving behind only the residue or extract. The flask was then cooled in desiccators after which it was weighed to estimate the fat.

Calculation

$$\text{Fat content (g/100g)} = \frac{\text{Weight of ether extract (g)}}{\text{Weight of sample taken (g)}} \times 100$$

ANNEXURE- III

ESTIMATION OF CRUDE FIBRE (AOAC, 1980)

Principle

During the acid and subsequent alkali treatment, oxidative hydrolytic degradation of the native cellulose and considerable degradation of lignin occurs. The residue obtained after final filtration is weighed, incinerated, cooled and weighed again. The loss in weight is the crude fibre content.

Reagents

- 0.255N standard H_2SO_4
- 0.313N Standard NaOH

Method

A weighed amount of (2.5-5g) of moisture and the fat-free sample was transferred to a fibre bag which was preheated and weighed. These bags were inserted into tubes and placed in a beaker provided in the instrument. The sample was boiled with 300ml 0.255 ± 0.005N H2SO4 for 30 minutes. Then the residue was washed with boiling water until acid-free. Then the residue was boiled with 300ml of 0.313 ± 0.005 N NaOH for 30 minutes. Again, the residue was washed with boiling water followed by alcohol wash. The residue was transferred to pre-weighed crucibles(W1) and it was dried to 2 hours at 130± 2 °C, cooled in desiccators then weighed (W2). The dried desiccators containing samples were then ignited for 30 minutes at 600 ± 15°C. Finally, the sample was cooled and weighed again

Calculation

$$\text{Crude fibre (g/100g)} = \frac{[100 - (\text{Moisture} + \text{Fat})] \times (We - Wa)}{\text{Weight of sample taken (Moisture and Fat-free)}}$$

We – Preweighed ashing dish, Wa – Weight of the dish after ashing

ANNEXURE-IV

ESTIMATION OF TOTAL ASH (AOAC, 1980)

Procedure

Total ash was estimated in the sample by weighing about 5 g of dried samples into a crucible. The crucible was placed on a wire gauge and heated over a flow till the material was completely carried and then the crucible was heated in the muffle furnace for about 4 hours. At 600 ℃. It was then cooled in a desiccator and weighted. To ensure the completion of ashing, the crucible was again heated in the furnace. For one hour cooled and weighed.

This was repeatedly done till two consecutive weights were the same and the ash was almost white or grayish colour. The estimation was done in triplicate.

Calculation

Weight of the crucible – W g

Weight of the crucible + sample – W1 g

Weight of the crucible + ash – W2 g

Weight of sample (W1 – W) g

Weight of sample (W2 – W) g

$$\% \text{ Total ash} = \frac{(W2 - W)\ g}{(W1 - W)\ g} \times 100$$

$$\text{Ash content (g /100g)} = \frac{\text{Weight of Ash}}{\text{Weight of sample taken (g)}} \times 100$$

ANNEXTURE V

ESTIMATION OF INSOLUBLE DIETARY FIBRE (AOAC, 1980)

Principle: Defatted foods are gelatinized and proteins and starch are removed by enzymatic digestion. The residue is quantitated gravimetrically.

Reagents:

- 95% Ethanol
- 78% Ethanol
- 0.08M Phosphate Buffer, pH 6.0
- 0.275 N NaOH
- 0.325 N HCl
- α- Amylase heat stable solution
- Protease solution: suspended 50 g protease in 1 ml phosphate
- Buffer pH 6.0
- Amyloglucosidase solution

Sample Preparation:

Homogenise sample and dry overnight in hot air oven at 105ºC, cool in desiccator, and dry mill portion of sample to 0.3 to 0.5mm mesh. If sample cannot be heated, freeze-dry before milling. If high fat content (75%) prevents paper milling defat with petroleum ether before milling.

Determination:

Run the blank through entire procedure along with samples to measure any contribution from reagents residue. Weigh duplicate 1g of sample, accurate to 0.1 mg, in to 500ml beakers. Sample weight should not differ 20 mg. Add 50 ml of Phosphate buffer and adjust the pH to 6.0, if necessary. Add 0.1 ml heat stable α–amylase solution.

Cover the beakers with aluminium foil and place in boiling water bath. Ensure that the contents of the beaker reach 100ºC and incubate for 15 min at this temperature and adjust pH to 7.5 with NaOH solution. Add 0.1 ml of protease solution to each beaker.

Cover beaker with aluminium foil and incubate for 30 min in 60º C with continuous agitation. Cool and adjust pH to 4.0 4.6. Add 0.3 ml amyloglucosidase, and incubate for 30ºC with continuous agitation.

Weigh crucible with a fritted disc containing 1 g celite to constant weight. The celite in the crucible is made into bed by using a stream of 78% ethanol and applying suction. Maintain suction and quantitatively transfer precipitate from enzyme digest to crucible, using filtration module.

Wash residues successively with 3 times 20 ml portions of 78%, two 10 ml portions of 95% ethanol and two 10 ml portions of acetone. Dry crucible containing residue overnight at 100ºC in hot air oven. Cool in desiccator and weigh to nearest 0.1mg. Subtract crucible and celite weight from the above to obtain the insoluble dietary fibre residue (IDF residue).

Analyse residue from one sample of set of duplicates for protein by Kjeldahl method using N ×6.25 as conversion factor and subtract from the IDF residue value.

Incinerate second residue sample of duplicate for 5 h at 525º C. cool in desiccator and weigh to nearest 0.1 mg and subtract from the IDF residue value.

Insoluble dietary fibre= IDF residue – (protein + ash)

$$\text{IDF\%} = \frac{\text{Wt. of the IDF residue (g)} - \{\text{Protein (g) in IDF residue} + \text{Ash (g) in IDF residue}\}}{\text{Wt. of the sample (g)}} \times 100$$

ANNEXURE VI

ESTIMATION OF SOLUBLE DIETARY FIBRE (AOAC, 1980)

Principle: The soluble fibre is estimated in the filtrate obtained after enzymatic digestion of protein and carbohydrates of defatted food. The soluble fibre is precipitated and estimated gravimetrically.

Reagents:

- 95% Ethanol
- 78% Ethanol
- 0.08M Phosphate Buffer, pH 6.0
- 0.275 N NaOH
- 0.325 N HCl
- α- Amylase heat stable solution
- Protease solution: suspended 50 g protease in 1 ml phosphate Buffer pH 6.0
- Amyloglucosidase solution

Sample preparation:

Homogenise sample and dry overnight in hot air oven at 105ºC, cool in desiccator, and dry mill portion of sample to 0.3 to 0.5mm mesh. If sample cannot be heated, freeze-dry before milling. If high fat content (75%) prevents paper milling defat with petroleum ether before milling.

Determination:

Follow the steps of digestion with α-amylase, protease and amyloglucosidase and quantitative transfer the digest and collect the filtrate. Add 4 volumes of preheated (60ºC) 95% ethanol. Allow the precipitation to complete for 60 min. filter through an accurately weighed crucible with celite. Follow the procedure given under insoluble fibre to obtain soluble dietary fibre (SDF) residue. Duplicate samples run similarly are analysed for protein and ash.

Soluble dietary fibre = Weight of SDF residue - (protein + ash)

$$SDF\% = \frac{\text{Wt. of the SDF residue (g)} - \{\text{Protein (g) in SDF residue} + \text{Ash (g) in SDF residue}\}}{\text{Wt. of the sample (g)}} \times 100$$

Estimation of total dietary fibre (AOAC, 1980)

The total dietary fibre is the sum of the insoluble and soluble dietary fibre, estimated as follows;

Blank % = Wt. of the blank residue (g) – {Protein (g) in the blank residue + Ash (g) in blank residue}

Total Dietary Fibre = IDF + SDF values

ANNEXURE VII

ESTIMATION OF TOTAL POLYPHENOLS (RANGANNA, 2005)

Blue colour developed by polyphenols with FCR reagent in alkaline condition was measured at 650 nm

Reagents

- 80 % Ethanol
- Folin- Ciocalltean Reagent (FCR)
- 20% $NaSO_4$
- Standard catechol

Procedure

1. Weigh exactly 0.5 to 1.0 gm of the sample and grind with a pestle and mortar in 10 times volume 80 percent ethanol
2. Centrifuge at 10000 RPM for 10 min collect supernatant.
3. Re-extract the residue with 5 times volume of 80 percent ethanol centrifuge and poor the supernatants.
4. Evaporate the supernatants to dryness over water bath.
5. Dissolve the residue in a known volume of distilled water (3ml).
6. Pipette out 0.5 and 1.0 ml into test tube.
7. Add distilled water to make up the volume to 3 ml.
8. Add 0.5 ml FCR to each test tube.
9. After 3 min, add 2 ml of 20% Na_2CO3 solution to each test tube.
10. Mix thoroughly, place the tube in boiling water bath for exactly 1 min, cool and measure OD at 650 nm.

$$\text{mg/ 100gm sample total polyphenol} = \frac{\text{Concentration of polyphenol from graph}}{\text{Aliquot taken for estimation}} \times 5 \times \frac{100}{\text{Weight of sample}} \times \frac{1}{100}$$

ANNEXURE VIII

ESTIMATION OF TANNINS (AOAC, 1980)

Principle

Tannins estimation is based on the measurement of blue colour formed by the reduction of phosphotungstomolybdic acid in alkali solution. Tannin-like compounds reduce phosphotungstomolybdic acid in alkaline solution to produce a highly coloured blue solution, the intensity of which is proportional to the amount of tannins. The intensity is measured in a spectrophotometer at 700 nm.

Chemicals and reagents

- Folin-Denis reagent
- Saturated sodium carbonate solution: 35gm of anhydrous sodium carbonate was dissolved in 100 ml of water at 70-80◦C and cooled overnight. Clear liquid was decanted.
- Tannic acid standard: Prepared using standard tannic acid from Thomas Baker chemicals
- 1% HCl
- Methanol

Preparation of standard curve

The standard curve for tannic acid solution was plotted by adding the reagents as mentioned in the table 1

Table 1: Reagents for preparation of standard curve of tannic acid

Sl. No.	Tannic acid concentration (µg)	Tannic acid (0.1 mg/ml) (µl)	H2O (µl)	Folin-Denis Reagent (µl)	Sodium carbonate solution (µl)	
Blank	0	0	75	37.5 µl	187.5 µl	OD@ 715nm
Std. 1	2	1.5	73.5			
Std. 2	4	3	72			
Std. 3	6	4.5	70.5			
Std. 4	8	6	69			
Std. 5	10	7.5	67.5			
Std. 6	12	9	66			
Std. 7	14	10.5	64.5			
Std. 8	16	12	63			
Std. 9	18	13.5	61.5			

Preparation of Sample

Accurately Weighed 0.4 g of the powdered material was transferred to a 250mL conical flask. Add 40mL Diethyl ether containing 1% Acetic acid (V/V) and mixed to remove pigment. The supernatant was carefully discarded after 5 minute and 20 mL of 70 per cent aqueous acetone was added and the flask was sealed with cotton plug covered by aluminum foil and kept in electrical shaker for 2 hours for extraction. It was then filtered through Whatman filter paper No. 1 and sample was kept in refrigerator at 4 ^{o}C until analysis.

Estimation of sample

An aliquot of the sample extract containing not more than 18 µg of tannic acid was used and the percentage of tannin will be determined.

Table 2: Reagents for estimation of tannic acid

Sl. No.	Tannic acid concentration (µg)	Sample (µl)	H2O (µl)	Folin-Denis reagent (µl)	Sodium carbonate solution (µl)	
Blank	0	-	75	37.5 µl	187.5 µl	OD@ 715nm
Sample	Unknown	75	-			

Calculation

The tannin concentration was determined by the standard graph of tannic acid solution.

Methodology

Sample extraction

Five grams of sample was extracted with 85 ml of methanol containing one per cent of HCl for 30 minutes with occasional shaking. The content was filtered using Whatman No. 1 filter paper. The filtrate was used for tannin determination.

Standard curve development

Zero to 10 ml of aliquots of the standard tannic acid solution was taken into 100 ml volumetric flask containing 75 ml of water added, three ml of folin-denis reagent and 10 ml sodium carbonate solution into each of the volumetric flasks and volume was made up to 100ml with distilled water. Solution was mixed well and colour measured after 30 minutes at 760 nm against experimental blank.

Protocol

One ml of extract was taken in 100 ml volumetric flask to which five ml of folindenis reagent and 10 ml of sodium carbonate solution was added. The contents were mixed and diluted to 100ml using distilled water and allowed to stand for 30 minutes and absorbance was measured at 760 nm. The tannin content of the samples was calculated as tannic acid equivalents from the standard graph.

Calculation

$$\text{Tannins \%} = \frac{\text{Tannic acid (mg)} \times \text{Dilution} \times 100}{\text{Sample taken for colour development (ml)} \times \text{Weight of sample (g)}} \times 100$$

ANNEXURE IX

ESTIMATION OF ASCORBIC ACID (RANGANNA, 2005)

Principle: The direct calorimetric determination is based on measurement of the extent to which a 2,6 - dichlrophenol indophenol solution is decolourised by ascorbic acid in sample extracts and in standard ascorbic acid solution.

Reagents:

- 2% metaphosphoric acid in distilled water.
- Dye solution – Dissolve 100mg 0f 2,6 - dichlrophenol indophenol dye and 84mg of sodium bicarbonate in hot distilled water (85-95 °C).
- Cool and make upto 100ml.
- Standard ascorbic acid solution – Weigh 100gm of ascorbic acid and make upto 100ml with 2% HPO_3. Dilute 4ml of this solution to 100ml with 2% HPO_3.

Preparation of sample: 1gm of the sample taken and blended with 2% HPO_3 in mortar and pestle and centrifuge.

Standard curve: To dry cuvettes or test tubes, pipette the required volume of standard ascorbic acid solution – 1, 2, 2.5, 3, 4 and 5ml and make upto 5ml with 2% HPO_3. Add 10ml of dye with a pipette, shake and take the readings within 15 to 20secs.

Set the instrument to 100% transmission using a blank consisting of 5ml of 2% HPO_3 solution and 10ml of water. Measure the red colour at 518nm or a wavelength nearest to the required wavelength using a suitable filter. Plot the absorbance against concentration.

Sample: Place 5ml of the extract (or less made to 5ml with HPO_3) in a dry cuvette, add 10ml of dye and measure as in standard.

Calculation

$$\text{Mg of ascorbic acid per 100g or ml of sample} = \frac{\text{Ascorbic acid content} \times \text{Volume make up} \times 100}{\text{ml of solution taken for estimation} \times 1000 \times \text{Wt. or volume of sample taken}}$$

ANNEXURE X

DETERMINATION OF PEROXIDE VALUE

Principle: In the oxidative rancidity, oxidation of fat due to the combination of oxygen with unsaturated fatty acids takes place and results in the formation of compounds with a peroxide structure. These are detected by the liberation of iodine from an acid solution of potassium iodide. There is another type of rancidity caused by the action of lipase. This rancidity is called hydrolytic rancidity, which is caused by the formation of low molecular weight fatty acids like butyric acid, caproic acid and caprylic acids. This can be estimated by alkali titration method mentioned under acid value of ghee and is expressed in terms of butyric acid.

Reagents:

- Acetic acid- chloroform mixture (Composed of glacial acid and chloroform in the ratio of 2:1).
- Saturated potassium iodide solution.
- N/1000 sodium thiosulphate.
- Starch indicator.

Procedure:

0.5 to 1 g of clear melted fat was weighed accurately in the boiling flask. To this 30 ml of acetic acid- chloroform mixture was added and fat was dissolved. 1 ml of saturated potassium iodide was added. After 5 min 100 ml of distilled water was added. The liberated iodine was titrated against N/1000ml sodium thiosulphate. When the end point is approached 1 ml of freshly prepared starch was added and titration was completed till the blue colour disappears. Blank was carried out using all the reagents without the oil.

Calculation:

$$\text{Peroxide value of oil (meq/kg of sample)} = \frac{\text{(Titre-blank)} \times N \times 1000}{\text{Wt of oil (g)}}$$

ANNEXURE-XI

SENSORY EVALUATION SCORECARD (PERYAM AND PILGRIM, 1957)

Name: **Date:**

Product Name:

Instruction:

- Please evaluate each of the following samples using scoring system given below.
- Write the preferred number score in the column as per evaluation.
- Rinse your mouth in between evaluating each sample.

Sample	Appearance	Texture	Colour	Flavour	Taste	Overall acceptability
1						
2						
3						
4						
5						
6						

Scoring system:

9-like extremely	8-like very much	7-like moderately
6-like slightly	5-neither like nor dislike	4-dislike slightly
3-dislike moderately	2-dislike very much	1-disliextremely

Remarks:

Signature:

ANNEXURE-XII

CONSUMER ACCEPTABILITY USING FACT SCALE

Name: FACT scale on Product

Date:

Indicate your opinion by ticking any one

Sl. No.	**Opinion**	✓
1.	I would eat every opportunity that I had	
2.	I would eat this very often	
3.	I would frequently eat this	
4.	I like this and would eat it now and then	
5.	I would eat if available but would not go out of my way	
6.	I don't like this but would eat this on an occasion	
7.	I would hardly ever eat this	
8.	I would eat this if there were no other food choices	
9.	I would eat this only if forced	
Total		

Comments:

Signature

ANNEXURE-XIII

CONSUMER ACCEPTABILITY SCALE

Name of the product: Date:

Name:

Age:

Kindly taste the product and indicate your opinion by placing a numerical score in the appropriate box given below.

Product	Appearance	Texture	Taste	Flavour	Overall acceptability

Five Point Hedonic scale

5	Excellent
4	Very good
3	Good
2	Fair
1	Poor

Comments:

Signature

ANNEXURE-XIV

MICROBIAL STUDY (STANDARD PLATE COUNT METHOD)

- Label the dilution blanks as 10^1, 10^2,10^3 and 10^4
- Prepare the initial dilution by adding 1ml or 1g of the sample into 9ml dilution blank added. Thus, diluting the original sample 10 times.
- Mix the contents and from the first dilution transfer 1ml of the suspension to dilution blank 10^2 with a sterile and fresh 1ml pipettes, diluting the original suspension to 100 times (10^2).
- From the 10^2suspension transfer 1ml to 10^3 dilutions blank with a fresh sterile pipette, thus, diluting the original ample to 1000 times (10^3) and repeat the procedure till you get the required dilutions.
- From the appropriate dilution transfer 1ml of suspension to sterile Petri dish by using a sterile pipette and pour the media, solidify and incubate the plates (Tate, 1995).

Composition of media

1. Nutrient Agar (NA) for bacteria

Beef extract	3.0g
Peptone	5.0g
Agar	15.0g
Distilled water	1000ml
pH	7.0

2. Yeast Extract Peptone Dextrose Agar (YEPDA) for Yeast

Yeast extract	10g
Peptone	20g
Dextrose	20g
Agar	18g
Distilled water	1000ml
pH	5.4

3. Martins Rose Bengal Agar (MRBA) for fungi

Dextrose	10g
Peptone	5g
K_2HPO_4	1g
$MgSo_4 7H_2o$	0.5g
Agar	15g
Rose Bengal	0.033g
Distilled water	1000ml
pH	6

ANNEXURE XV

DIGESTION OF SAMPLE

For nutrients other than N, the sample can be digested in a di-acid mixture of a tri-acid mixture or dry ashed and dissolved in acid.

The di-acid digestion is used for the determination of P, K, Ca, Mg, S, Fe, Mn, Zn and Cu. It must be followed for the determination of Ca and Mg. the tri-acid digestion is recommended only when P and K are to be estimated. Sulphur cannot be estimated from tri-acid extract. Similarly, Ca will also be underestimated. Since H_2SO_4 can contribute some micronutrients and heavy metals, di-acid digestion is normally recommended for plant analysis. Wet digestion is normally not used for the estimation of B and Mo.

Dry ashing can be used for sample preparation for the determination of Na, K, Ca, Mg, Cu, Fe, Mn, Zn, B and Mo in plant tissue. It is the preferred technique for B and Mo particularly. Dry ashing provides good precision and is an easy, rapid method requiring minimal analyst attention. It is also relatively free from reagent contamination. The main disadvantage of this procedure is that it cannot be used for elements such as N, P and S which are volatile at the ashing temperature.

Di-acid digestion: It is carried out using a 9:4 mixture of HNO_3:$HClO_4$. If the sample is high in fats/oils, pre-digestion using 25 mL HNO_3/g sample is recommended to avoid explosion. Detailed procedure is as follows:

Weigh 1 g ground plant material in 100 mL conical flask. Add 10 mL of di-acid mixture and mix the content of the flask by swirling. Placed the flask on low heat hot plate in a digestion chamber. Then, heat the flask at higher temperature until the production of red NO_2 fumes ceases, continue digestion until the volume is reduced to about 3 to 5 mL but not to dryness. The completion of digestion is confirmed by the snow white residue or when the liquid become colorless.

After cooling make up the volume with glass distilled water deionized water and filter solution through Whatman No.1 filter paper. Use aliquots of this solution for the determination of P, K, Ca, Mg, S, Fe, Mn, Zn and Cu.

ANNEXURE XVI

DETERMINATION OF PHOSPHORUS

The nutrients (except nitrogen) in plant samples is digested either with diacid/ tri acid on sand bath till digested material turns to either colour less or whitish. The phosphorus in the aliquot of the digested material in presence of vanadium (V^{5+}) and molybdenum (Mo^{6+}), orthophosphates forms a yellow coloured phospho vanado molybdate complex which can be read using spectrophotometer 430 nm.

Reagents

1. Phosphorus standard (50 ppm): Dissolve 0.2195 g of dried (40^0C potassium dihydrogen phosphate in about 400 mL of distilled water. Then add 25 mL of 7 N H_2SO_4 and make up the volume to 1 L with distilled water.

2. Solution A: Dissolve 25 g of ammonium molybdate in 400 mL of hot distilled water.

3. Solution B: Dissolve 1.25 g of ammonium metavanadate in 300 mL of boiling distilled water. Cool the content's and add 250 mL of Conc. HNO_3.

4. Vanadomolybdate reagent: By mixing the solution A and solution B and make up the volume to 1 L

Procedure

Preparation of the standard curve

1. Pipette out 50 ppm P solution to a series of 50 mL volumetric flasks to get a concentration of 0, 5, 10, 15 and 20 ppm P solution.

2. Add 10 mL of vanadomolybdate reagent, mix and make up the volume.

3. Read colour intensity at 430 nm after 30 minutes.

4. Plot the absorbance against concentration and draw the standard curve.

Plant sample

1. Pipette out 5 mL of digested sample into a 50 mL volumetric flask

2. Add 10 mL of vanadomolybdate reagent and make up the volume
3. Read colour intensity at 430 nm after 30 minutes.
4. Compare the unknown sample absorbance with standard curve

Calculations

$$P\ (\%) = \frac{\text{Graph ppm} \times \text{Vol. of digested sample} \times \text{Vol. made up}}{10^6 \times \text{weight of sample} \times \text{Aliquot taken}} \times 100$$

ANNEXURE XVII

DETERMINATION OF CALCIUM AND MAGNESIUM

Principle

Ethylene diamine tetra acetic acid is assigned the formula H_4Y, the disodium salt is therefore NaH_2Y and affords the complex-forming ion HY^{2-} in aqueous solution. The usefulness of EDTA as a titrant is due to the presence of four or six atoms which are available for coordination to a metal cation in such a way that five membered rings are produced; 1:1 complexes are formed and these are the most important ones.

The reaction with cation eg. M^{2+}, may be written as:

$M^{2+} + H_2 = MY^- + 2\ H+$

$M^4 + H_2Y^{2-} = MY + 2\ H^+$

$M^+ + H_2Y^= = (MY)^{(n-4)+} + 2H^+$

For other cations, the reactions may be expressed as :

$M^{+3} + H_2Y^= = MY^- + 2H^+$

One gram ion of the complex forming HaY^{2-} reacts in all cases with one gm ion of the metal and in each case two hydrogen ions are formed. The resulting complexes have similar structure but differ from one another in the charge that they carry.

Reagents

1. Standard EDTA solution (0.01 N): Dissolve 1.861 gm of EDTA in 900 mL distilled water and make up the volume to 1000 ml.
2. Std. Ca solution: Dissolve 0.6005 g portion of pure dried $CaCO_3$ in 0.2 N HCl. Solution is boiled to expel the CO_2 and dilute to 1 L.
3. 10 % NaOH solution: Dissolve 10 g portion of NaOH in about 90 mL distilled water and dilute to the 100 ml.
4. Murexide indicator: Mix 0.2 g of murexide with 40 g of powdered K_2SO_4.
5. Buffer solution (pH 10): Add 142 mL of NH_4OH to 17.5 g of NH_4Cl and dilute to 250 mL with distilled water.

6. Erichrome Black T indicator: Dissolve 0.2 g of the EBT powder 15 mL of triethanolamine and 5 mL of absolute ethanol.

Procedure

a) Determination of calcium

1. Measure 5 mL of digested sample onto a porcelain basin and dilute with 10 mL distilled water.
2. Measure 5 mL of 10 % NaOH (pH of sample solution would reach more than 12).
3. About 0.5 g of murexide indicator is added.
4. Titrate the contents against std. EDTA with stirring until it becomes violet in colour. Note down the burette reading as A.

b) Determination of calcium + magnesium

1. Another 5 mL of digested sample is measured onto a porcelain basin and dilute with 25 mL of distilled water.
2. Add 5 mL buffer solution. Add 3-5 drops of EBT - indicator and
3. Then titrate the contents against std. EDTA by stirring until it becomes sky blue colour. Note down the burette reading as B.
4. The titre value Ca alone is subtracted from titre value of Ca + Mg, to get the value for Mg.

$$\text{Ca (\%)} = \frac{\text{A} \times \text{N of EDTA} \times 0.02 \times \text{Vol. of digested sample}}{\text{Aliquot taken} \times \text{weight of sample}} \times 100$$

$$\text{Mg (\%)} = \frac{[\text{A} - \text{B}] \times \text{N of EDTA} \times 0.012 \times \text{Vol. of digested sample}}{\text{Aliquot taken} \times \text{weight of sample}} \times 100$$

Note: If the concentrations of interfering ions are more in the sample, then 1 or 2 drops of 1 % KCN or NaCN should be added to the sample before titration.

ANNEXURE XVIII

DETERMINATION OF SODIUM

Principle

In Flame photometry, also known as Flame emission or Flame atomic emission, the sample in solution, is sprayed into a flame to vaporize, atomize, and excite the sample. The excited atoms of the element of interest emit light at certain discreet wavelengths, which characteristic of that element. Light of the wave length of interest is separated from remander of emitted radiations and its intensity is measure. The intensity measurement can be related directly to the concentration of the element of interest usually by comparing with the measured intensities of a standard or series of standards.

Reagents

1. Std. sodium (100 ppm): Dissolve 0.191 g of NaCl in some volume of distilled water and then make up the volume to 1 L.

2. Prepare the working standards from 0 to 40 ppm of sodium

Procedure

Sample

Feed the digested sample solution to the Flame photometer and record the reading (if dilution is required, it should be done before feeding to the instrument). Compare the unknown sample readings with standard curve to determine the % Na in plant sample.

Calculations

$$\text{Na (\%)} = \frac{\text{Graph ppm} \times \text{Vol. of digested sample} \times \text{Vol. made up}}{10^6 \times \text{weight of sample} \times \text{Aliquot taken for dilution}} \times 100$$

ANNEXURE XIX

DETERMINATION OF Fe, Mn, Zn AND Cu

Make suitable dilutions of di acid extract and feed standard/ sample to AAS having appropriate hallow cathode lamps. Record values and plot on graph paper.

Calculation

$$\text{Micronutrient conc. (ppm)} = \frac{\text{Graph ppm} \times \text{Vol. of digested sample}}{\text{Weight of sample}}$$

ANNEXURE XX

FORMULATION OF FOOD PRODUCTS

Peanut *chikki*:

Ingredients	Quantity (g)
Jaggery	50
Peanuts	45
Ghee	05

Method of preparation:

- Roasting of peanuts till golden brown colour is obtained, then dehulling of peanuts seed and coarsely crushed.
- Dissolving jaggery in hot water, filtered and boiled till the hard crack stage.
- Crushed peanuts added to the jaggery syrup and mixed thoroughly.
- Pouring the mixture on pre greased surface and roll out then allow to set.
- Cutting into uniform pieces and then *chikki* was cooled and packed.

Flax seed *chutney* powder:

Ingredients	Quantity (g)
Flax seed	45
Black gram dhal	20
Bengal gram dhal	20
Red chilly	08
Curry leaves	02
Garlic	02
Salt	03

Method of preparation:

- Flaxseeds, bengal gram dhal, black gram dhal and other ingredients were roasted separately till the raw flavour subsided.
- The above ingredients were cooled to room temperature and ground in a mixer to get *chutney* powder.
- The flax seed *chutney* powder was done and packed.