

# **FORUMULATION AND STANDARDIZATION OF FUSION BITES USING “DIOSCOREA ALATA”**

## **PROJECT REPORT**

Submitted in partial fulfilment of the requirements for the award of Bachelor of  
Science degree in Clinical Nutrition & Dietetics

**By**

**KAVYA SHREE .S (41736007)**

**SATHIYA PRIYA. M (41736015)**



**DEPARTMENT OF CLINICAL NUTRITION AND DIETETICS  
SCHOOL OF ALLIED HEALTH SCIENCES**

# **SATHYABAMA**

**INSTITUTE OF SCIENCE AND TECHNOLOGY  
(DEEMED TO BE UNIVERSITY)**

**Accredited with Grade A++ by NAAC**

**JEPPIAAR NAGAR, RAJIV GANDHI SALAI. CHENNAI - 600119**

**APRIL 2024**



**SATHYABAMA**

INSTITUTE OF SCIENCE AND TECHNOLOGY

(DEEMED TO BE UNIVERSITY)

Accredited "A++" Grade by NAAC | 12B Status by UGC | Approved by AICTE

www.sathyabama.ac.in

**DEPARTMENT OF CLINICAL NUTRITION & DIETETICS**

**BONAFIDE CERTIFICATE**

This is to certify that this Project Report is the Bonafide work of SATHIYAPRIYA M (41736015) KAVYA SHREE S (41736007) who has the project work entitled **FORUMULATION AND STANDARDIZATION OF NUTRITION BITES USING "DIOSCOREA ALATA"** under my supervision from JUNE 2023 to MAY 2024.

Internal Guide  
(Ms. PRIYA S, M.Sc., NET)

External Guide  
Dr.L.STANLEY ABRAHAM

Head of the Department  
(Dr. L. INBATHAMIZH)

---

Submitted for Viva – voce Examination held on 28.04.24

Internal Examiner

External Examiner

## DECLARATION

We Sathiya priya M and Kavya Shree S hereby declare that the project entitled **"FORMULATION AND STANDARDIZATION OF FUSION BITES USING DIOSCOREA ALATA"**. Done by me under the guidance of Ms. PRIYA .S is submitted in partial fulfilment of the requirements for the award of Bachelor of Science degree in Clinical Nutrition & Dietetics.

DATE: 27.04.2024

PLACE: CHENNAI

*S. Kavya Shree*

SIGNATURE OF THE CANDIDATES

## DECLARATION

We Sathiya priya M and Kavya Shree S hereby declare that the project entitled "FORMULATION AND STANDARDIZATION OF FUSION BITES USING DIOSCOREA ALATA". Done by me under the guidance of Ms. PRIYA .S is submitted in partial fulfilment of the requirements for the award of Bachelor of Science degree in Clinical Nutrition & Dietetics.

DATE: 27.04.2024

PLACE: CHENNAI

*M. Sathiya priya*  
SIGNATURE OF THE CANDIDATES

## ACKNOWLEDGEMENT

We are pleased to acknowledge our sincere thanks to **The Board of Management of Sathyabama University** for their kind encouragement in doing this project and for completing it successfully. We are grateful to them.

We are conveying our thanks to **Dr.L.INBATHAMIZH., PHD., Head of the Department, and Department of Clinical Nutrition & Dietetics** for providing us with necessary support and details at the right time during the progressive reviews.

We would like to express our sincere and deep sense of gratitude to **our Project Internal Guide Ms.PRIYA.S and External Guide Dr.L.STANLEY ABRAHAM** for their valuable guidance, suggestions and constant encouragement paved way for the successful completion of our project work.

We wish to express our thanks to all **Teaching and Non-teaching staff members** of the Department of Clinical Nutrition and Dietetics who were helpful in many ways for the completion of the project.

## **ABSTRACT**

The study aims to comprehensively investigate *Dioscorea alata*, commonly known as purple yam, through various analytical methods and assessments. The research integrates techniques such as Thin Layer Chromatography (TLC) and Gas Chromatography-Mass Spectrometry (GC-MS) to identify bioactive compounds present in *D. alata*. Additionally, the study conducts a nutritive analysis to evaluate its nutritional composition, shedding light on its potential health benefits. Furthermore, the immunomodulatory activity of *D. alata* is explored, aiming to understand its potential in enhancing immune response. Lastly, the research extends to product development, exploring innovative ways to incorporate *D. alata* into consumer products, potentially leveraging its nutritional and immunomodulatory properties. This interdisciplinary approach provides a holistic understanding of *D. alata*'s potential applications in both health and food industries.

### **KEYWORDS:**

*Dioscorea alata*, Nutritive, purple yam, Health

## TABLE OF CONTENTS

| S.NO     | CONTENTS                                               | PAGE NO |
|----------|--------------------------------------------------------|---------|
|          | <b>ABSTRACT</b>                                        |         |
|          | <b>LIST OF FIGURES</b>                                 |         |
|          | <b>LIST OF TABLES</b>                                  |         |
| <b>1</b> | <b>INTRODUCTION</b>                                    |         |
| <b>2</b> | <b>LITERATURE SURVEY</b>                               |         |
|          | 2.1 Anti-nutritional Properties of Yam                 |         |
|          | 2.2 Nutritional and Pharmacological Properties of Yams |         |
|          | 2.3 Health beneficial potentials                       |         |
|          | 2.4 Sugars                                             |         |
| <b>3</b> | <b>AIM AND OBJECTIVES</b>                              |         |
|          | 3.1 Aim                                                |         |
|          | 3.2 Objectives                                         |         |
| <b>4</b> | <b>MATERIAL AND METHODS</b>                            |         |
|          | 4.1 Materials Ingredients used                         |         |
|          | 4.1.1 Pre-preparation                                  |         |
|          | 4.1.2 Preparation                                      |         |
|          | 4.2 Formulation                                        |         |
|          | 4.3 Thin layer Chromatography                          |         |
|          | 4.4 Phytochemical                                      |         |
|          | 4.5 total phenolic content.                            |         |
|          | 4.6 Gas Chromatography /Mass Spectrometry              |         |

4.7 Immunomodulatory Activity

4.8 Sensory evaluation

## **5 SUMMARY AND CONCLUSION**

## **REFERENCES**

## LIST OF FIGURES

|              |                                                   |
|--------------|---------------------------------------------------|
| Figure: 1.1  | Extraction of starch from yam                     |
| Figure: 1.2  | Growing stages of dioscorea alata                 |
| Figure: 1.3  | Cultivated fresh yam                              |
| Figure: 4.1  | Yam sample before drying                          |
| Figure: 4.2  | Preparation of flour                              |
| Figure: 4.3  | Preparations of bites                             |
| Figure: 4.4  | Preparation of mobilephase-1                      |
| Figure: 4.5  | Preparation of mobilephase-2                      |
| Figure: 4.6  | Preparation of mobilephase-3                      |
| Figure: 4.7  | Preparation of mobilephase-4                      |
| Figure: 4.8  | Combination of mobile phase                       |
| Figure: 4.9  | Representing rating of sensory evaluation         |
| Figure: 4.10 | Representing the rating for appearance            |
| Figure: 4.11 | Representing the rating of texture                |
| Figure: 4.12 | Representing the rating of flavour                |
| Figure: 4.13 | Representing the rating of mouthfeel              |
| Figure: 4.14 | Representing the rating for overall acceptability |
| Figure: 5.1  | Report of immunomodulatory activity               |

## **LIST OF TABLES**

Table: 4.1 Formulation episodes

Table: 4.2 Total mean and standard deviation of ratings

Table: 4.3 Rating scale and no. of responses

Table: 4.4 Range of rating and no. of response

Table: 4.5 Representing the rating scale for mouthfeel and its response

Table: 4.6 Representing the rating scale for overall acceptability and its response

Table: 5.1 Macro- nutrient composition tables

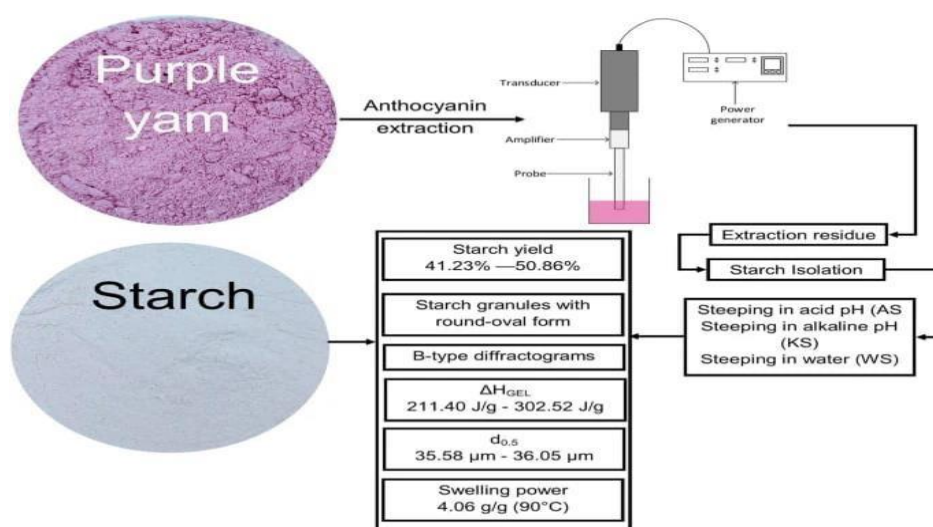
Table: 5.2 Micro nutrient composition tables

Table: 5.3 Peak report

# CHAPTER -1

## INTRODUCTION

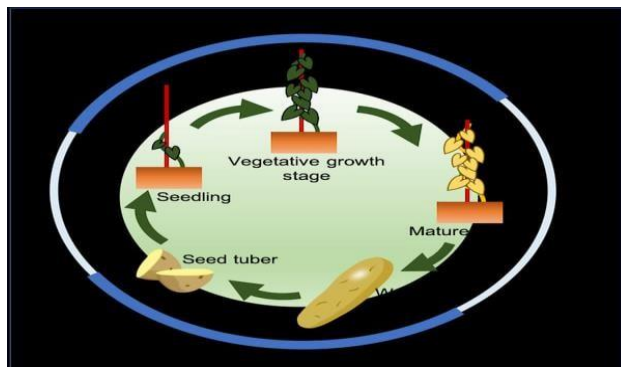
"Dioscorea alata: Nature's masterpiece, blending beauty and nutrition in a single tuber." An economical approach to put on weight-Alanine, cyaniding, and peonidin make up the bulk of its over 85% carbohydrate/starch content (Moriya, 2015). For small-scale farmers in many nations, yam is a vital crop since it can provide food security and serve as a source of revenue. For instance, about 60 million people consume yam, and West African nations grow almost 90% of the world's supply. A hydrophilic polymer that is biodegradable, starch is extracted from roots, legumes, grains, and tubers like purple yams (Adewale, 2022). The primary constituents are amylose and amylopectin, whose ratios vary depending on the source of the vegetable. Additionally, starch is a cheap substance that is frequently employed in the pharmaceutical and food industries. (Paul *et al*, 2022).



**FIG: 1.1EXTRACTION OF STARCH FROM YAM**

Planting season spans from May to June. It takes nine ten months for greater and white yams to be ready for harvest. Once collected, the outstanding nutritional characteristics of yam tubers can be stored under ambient tropical conditions for 46 months without experiencing any degradation. (Noora, 2019)The maturation period of to a lesser yam is 8–9 months. Without harming anyone, delicately remove the tubers and with an average annual temperature of 300C and a rainfall of 12002000

mm, the plant grows best in warm, humid climates with sandy loam soil that has a pH of 6.0–6.5. (Barooah, 2014).



**FIG: 1.2 GROWING STAGES OF DIOSCOREA ALATA**

The purple colouring of these yams is believed to be mostly caused by the presence of powerful antioxidants called anthocyanins, which are coloured brilliantly. (Davies, 2017)



**FIG: 1.3 COLLECTION OF PURPLE YAM FROM THENI**

Several bioactive compounds have been associated with the anticancer properties of the Dioscorea species, including dioscin, diosgenin, dioscorin, and anthocyanins (Schwinn, 2017). Our data indicate that these chemicals are not the cytotoxic agents causing the anti-proliferative action in the acetonic and ethanolic extracts of *D. alata*, despite the fact that they are abundant and anti-cancer—antiproliferative (Hong.sh, 2019). The cooked tubers are crushed in a mortar to create a thick, elastic paste, the result is pounded yam, or fufu, which is consumed

as balls with meat and sauce in other areas and Consumer sensory testing has identified several characteristics of high-quality boiled yams, including their white shade, crumbly texture that sticks to fingers, non-fibrous and simple to chew, sweet aroma, and pleasant aroma (Honfozo, 2021). But due to growing urbanisation, diets are quickly shifting, and consumption of highly processed yam-based meals is becoming popular. Examples include ready-to-use flours and ready-to-cook peeled tuber pieces in plastic bags that have been vacuum-sealed. Due to its affordability and versatility, starch finds extensive application in the food, pharmaceutical, textile, medicinal, and biotechnology sectors (Ashri, 2014) .

Starch is utilised in the food business for a variety of purposes, including thickening, gelling, emulsion stabilisation, and water binder. (Agudelo, 2014). The pigment that gives purple yams their distinctive purple colour is called anthocyanins, and purple yams are filled with these antioxidants.

Aside from their potential to prevent heart disease and stroke, anthocyanins have anti-inflammatory and anti-cancer qualities and Due to their low glycaemic index, purple yams don't raise blood sugar levels (Wallace, 2021). They are therefore a fantastic option for those who have diabetes or prediabetes and the high fibre content of purple yams is beneficial to digestive health in addition to promoting regularity, fibre can nourish the beneficial microorganisms in your stomach and Purple yams' anthocyanins have been shown in studies to enhance cognitive performance and shield brain cells from harm (Dhanabhal, 2021). And Along with potassium, manganese, and copper, purple yams are also a good source of vitamins A, C, and E. For general health and wellbeing, these nutrients are essential and also Because of their capacity to improve blood vessel function and have antiinflammatory properties, anthocyanins and other substances found in purple yams may help lower blood pressure (Chaudhuri, 2021). Salts known as oxalates are eliminated through urine because they are not metabolised by humans. Its high content in urine, however, raises the possibility of nephrolithiasis, or calcium oxalate kidney stones, since it forms insoluble compounds when it combines with calcium ions. The process of cooking encourages tissue rupture, which releases soluble oxalate and lowers the oxalate concentration in food so consuming this yam will reduces the harm occurs to our body. (Bhandari, 2003).

## CHAPTER-2

### LITERATURE REVIEW

There are not very many studies that explored the pharmacological properties of yams, although abundant information and traditional knowledge on the medicinal values of yams are prevalent in the tropical countries. Almost all *Dioscorea* Comparison Nutritional properties of yams with other root and tuber crops Chandrasekara and (Padhan, 2020); Bhattacharjee species contains high amounts of secondary metabolites that help in modulating metabolic processes in humans and animals, thus exhibiting beneficial health effects such as anti-oxidative, antihypertensive, anti-inflammatory, and antidiabetic activities as well as inhibition of receptor activities and triggering enzyme activities (Chaudhuri, 2021) ( . A wide range of other compounds with medicinal properties as well as anti-nutritional properties are also present in yams such as phenolic, flavonoids, allantoin, dioscin, dioscorin, diosgenin, polyphenols, tannins, oxalate, saponins, and alkaloids have also been reported in yams by various researchers (Wallace, 2021)

Saponins are glycosidic compounds with triterpenoid and steroidal properties, of which steroidal saponins are mainly present in different *Dioscorea* spp. (Adewale, 2022) Steroidal saponins can be further classified based on biological activities as Water-soluble polysaccharides from yams are reported to reduce blood glucose (Honfozo, 2021) and cholesterol levels especially the LDL cholesterol while improving the immune system. The presence of flavonoids has also been reported in several species of yams (Dhanabhal, 2021) spirostane, stigmastane, furostane, cholestane, ergostane, and pregnane families (agudelo, p. 2019) predominantly yams contain spirostane steroidal saponin known as diosgenin (Iachman, 2019)

#### 2.1 ANTI-NUTRITIONAL PROPERTIES OF YAMS

The presence of anti-nutritional factors influences the bioavailability of dietary nutrients such as proteins, minerals, and vitamins. Yam tubers are acrid in nature representing several anti-nutritional properties associated with skin irritation and inflammation of throat after consumption (Kumar, 2017). The presence of some

bioactive compounds such as phenolic acids (Zhao) tannins, phytates, (continued)

| Species name                         | Plant extract             | Medicinal properties                                                                                             |
|--------------------------------------|---------------------------|------------------------------------------------------------------------------------------------------------------|
| <i>D. Oppositifolia</i>              | Rhizomes/ tuber           | Relief of menopausal syndromes and rejuvenation of early mothers                                                 |
| <i>D. panthaica</i>                  | Rhizome                   | Gastric diseases, bone injuries, rheumatic arthritis, and cardiovascular diseases                                |
| <i>D. (Moriya, 2015) pentaphylla</i> | Leaf/vine                 | Treatment of paralysis                                                                                           |
|                                      | Tuber/flower/ young shoot | Rheumatism Tuber Pain relief and reduce swelling, stomach disorders                                              |
| <i>D. polystachya</i>                | Rhizome                   | Consumptive cough and dysentery, aid for digestion and gastric motility, and for restraining nocturnal emissions |
| <i>D. Septemloba</i>                 | Rhizome                   | Rheumatism, urethra, and renal infection                                                                         |
| <i>D. sylvatica</i>                  | Tuber                     | Decoction used to treat cuts, wounds, and sores                                                                  |
| <i>D. Trinervia</i>                  | Tuber                     | Chronic diarrhoea, asthma, and diabetes                                                                          |
| <i>D. villosa</i>                    | Rhizome                   | Rheumatism, menstrual complaints, and perimenopause symptoms                                                     |
| <i>D. zingiberensis</i>              | Rhizome                   | Cough, anthrax, rheumatic heart disease, rheum, arthritis, tumefaction, and sprain                               |

Designing Dioscorea Genomes for Improved Nutritional and oxalates have been reported in different species of yams, which are referred to as anti-nutritional because of their toxic properties and bitterness associated with them (Ravi Shankar, 2019)

## 2.2 NUTRITIONAL AND PHARMACOLOGICAL PROPERTIES OF YAMS

However, over the years, additional tuber characteristics targeted to consumer preferences (tuber size and shape), higher dry matter, tuber flesh colour, tuber flesh oxidation, and starch properties are being routinely measured for wider acceptance of newly developed varieties (Darkwa, 2020)

Recently, there has been some progress on application of advanced or omics tools to several species of Dioscorea. Currently based on short-reads, long reads, and chromosome conformation sequencing, good-quality reference genomes have been released for four Dioscorea species such as *D. Rotundata* (Tamiru, 2017) *D. alata* This has enabled to develop high-quality linkage maps and identification for several agronomic and quality determination systems anthracnose disease, tuber oxidative browning, tuber starch content , and postharvest hardening (Ladjeu, 2020).

## 2.3 HEALTH BENEFICIAL POTENTIALS

All the mice of the experimental sets (control and treated) did not show any sign of allergic lesion, sickness, and unusual stool and urine discharge and appeared healthy and fit throughout the feeding experimentation; hence, the treated samples are not toxigenic to the animal model. (Sartor, 2019) . Despite any significant differences in total food intake among the mice, the growth (body weight) was better in the treatment sets than in the control. (Salvan, 2019) The weight gain was maximum in the synergistic symbiotic setup comprising both the probiotics,, and FOS fractions recognizable body weight increment was also observed in the other treatment sets .The fermentable fibres in the carbohydrate fractions have been reported to influence the nutrient uptake and mineral absorption among the treated mice, as fermentable inulin-type fructans can increase the mineral absorption and positively influence endocrine function related to appetite and satiety. (Ashri, 2014). The blood sugar level in all treatment sets was average or significantly decreased, showing the FOS fractions' hypoglycaemic effects. (Respondex, 2019).

The lipid profile analysis of treated mice shows that the concentration of total serum cholesterol, serum LDL, serum VLDL, and triglyceride lowered significantly in prebiotics treated sets individually or in combination, compared to the commercial prebiotic Inulin. (Sherkat, 2017) The symbiotic combinations of FOS from hot water and 80% ethanol fractions with probiotics also showed a significantly lower cholesterol profile than untreated control mice. At the same time, serum HDL level increases significantly in all the treatment cases compared to untreated control. (Wallace, 2021)These results also prove more pronounced hypocholesterolaemia effects in symbiotic sets of the carbohydrate fractions than other sets, comparable to the inulin treated set. Therefore, our study explored that the FOS from the hot water and 80% ethanol fractions isolated from *D. Alata L.* contain some prebiotics that has a symbiotic effect on the serum lipid profile of mice. Lowering high serum cholesterol levels contributes to the prevention of atherosclerosis. Kim & Shin reported that administration of inulin, a commercially established prebiotic, for 30 days' treatment increased serum HDL in rats compared to control, which is mainly attributed to the reduced absorption of cholesterol and by the higher rate of catabolism in the liver to the hypocholesterolaemia effect. Wolthuis (Cheng, 2021)

Russo et al. proved the efficacy of inulin against gastrointestinal mobility and lipid and glucose metabolism. The blood sugar level in implicated no abrupt changes in blood sugar level in all treatment sets compared to the untreated control set when checked randomly. (Zaky, 2019)

*Dioscorea* plants have been used in traditional medicine for centuries to treat various diseases and represent an invaluable source of pharmaceuticals. Certain species of the *Dioscorea* genus have a rich history of traditional use and have been extensively studied (Tiwaeri, 2018) currently, the following species have welldocumented records and are widely employed

## **2.4 ANTI-TUMOUR ACTIVITY**

Recently, there has been a surge of interest in studying the pharmacological potential of *Dioscorea* plants as antitumor agents. Research has demonstrated their effectiveness in inhibiting the growth of various cancer cells, including those associated with liver, lung, ovary, colon, and breast cancers as well as with melanomas and leukaemia. These anti-tumour effects have been observed to occur through multiple pathways, (Jiao, 2021)

## **2.5 SUGARS**

Glucose and sucrose are the main free sugars reported in yam tubers (Kouassi, 2019) Hari Prakash and Namibian, 1996). The presence of maltose and fructose were reported after tubers were stored for some days other sugars found are pentose and mannose (Omijeh, 2018)

Hydrolysed to different sugars (especially when sprouting starts) such as maltose and fructose thus elevating the level of sugars and decreasing starch content consequently. (Harprakash, 2020)The organoleptic properties such as taste and mouthfeel are reported to improve during storage probably due to the increase in sugar content. Differences in species, variety and growth environment also influence the level of sugars in yam tubers (Mensah, 2018)

## **CHAPTER-3**

### **AIM AND OBJECTIVES**

#### **3.1 AIM**

The aim of this study is to comprehensively evaluate the phytochemical composition of *Dioscorea alata*, commonly known as purple yam, and explore its potential immunomodulatory properties. Additionally, this study seeks to conduct a thorough nutritive analysis of *D. alata* to better understand its nutritional profile. Furthermore, the research aims to utilize these findings to develop innovative products derived from *D. alata*, potentially harnessing its health-promoting properties for various applications. Through this investigation, we aim to contribute valuable insights into the medicinal and nutritional potential of *D. alata*, ultimately fostering the development of novel therapeutic and dietary interventions.

#### **3.2 OBJECTIVES**

- To evaluate the phytochemical composition from *dioscorea alata*
- To examine immunomodulatory properties from *dioscorea alata*
- To evaluate nutritive analysis from *dioscorea alata*
- Innovative product from *dioscorea alata* flour

## CHAPTER-4 METHODOLOGY

### 4.1 MATERIALS-INGREDIENTS USED:

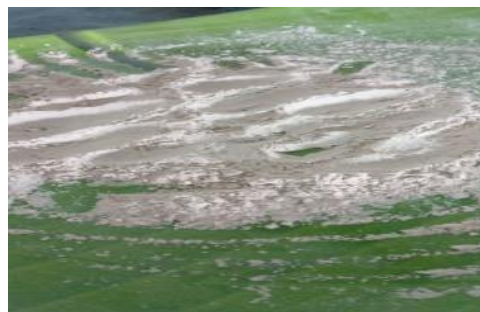
Flour [dried dioscorea alata yam], dates, sesame seeds, peanut, flaxseed.

#### 4.1.1 PRE-PREPARATION:

The fresh yam has been collected, washed and boiled and the skin of yam was peeled, chopped into fine small cubes then the cubes are kept for complete sundry.



**FIG: 4.1 (YAM SAMPLE BEFORE DRYING)**



**FIG: 4.2(PREPARATION OF FLOUR)**

#### 4.1.2 PREPARATION:

- First the yam flour has been roasted in the pan, and then transferred into cleaned container.
- Flax seeds, sesame seeds, and peanuts roasted for in low flame using pan.
- After roasting of peanuts, dates, sesame seeds and flaxseed have been blended up to half-rough textured consistency.

- The vessel which the flour is kept, this mixture has been added and mixed and made into fine balls.

This combination provides the customer with a wonderful sensory experience in addition to possible health benefits.

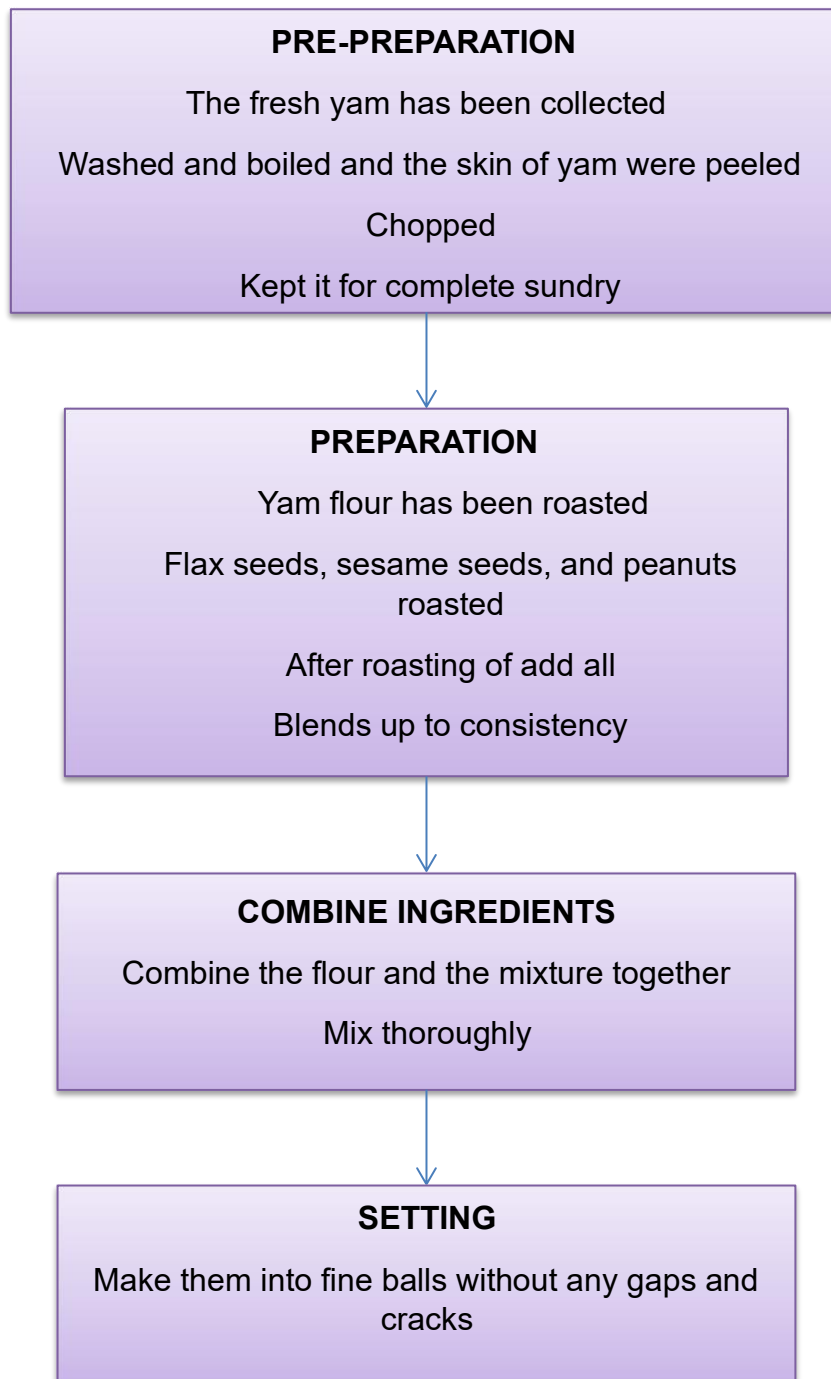
## 4.2 FORMULATION

**TABLE 4.1 FORMULATION EPISODES**

| <b>Materials used</b> | <b>Formulation<br/>- 1</b> | <b>Formulation<br/>-2</b> | <b>Formulation<br/>-3</b> | <b>Formulation<br/>-4</b> |
|-----------------------|----------------------------|---------------------------|---------------------------|---------------------------|
| Flour                 | 50g                        | 75g                       | 150g                      | 150g                      |
| Dates                 | 35g                        | 45g                       | 100g                      | 200g                      |
| Peanuts               | 70g                        | 75g                       | 100g                      | 100g                      |
| Sesame seeds          | 5g                         | 6g                        | 15g                       | 20g                       |
| Flax seeds            | 3g                         | 10g                       | 5g                        | 5g                        |

The fourth formulation, which utilizes flour, dates, peanuts, sesame seeds, and flax seeds, was selected for preparation due to the optimal consistency of the resulting balls.

## FLOW CHART OF BITES PREPARATION



**FIG: 4.3 PREPARATIONS OF BITES**

### 4.3 THIN LAYER CHROMATOGRAPHY (TLC):

Separating and analysing biomolecules from a complicated mixture is done via chromatography. An immobile phase and a stationary phase make up this separation procedure. Mixture to be separated percolates through stationary phase, forming mobile phase. Gas-liquid, liquid-liquid, or solid-liquid phases can be these two states. Polar absorbents can be used as the stationary phase in Thin Layer Chromatography (TLC), a solid-liquid chromatography technique, whereas a single solvent or a combination of solvents can be used as the mobile phase.

#### SOLVENT PHASE:

PHASE -1: hexane (75%) and acetone (25%).

PHASE-2: hexane (7:2) and ethyl acetate (3:8).

PHASE-3: chloroform (6:4), acetic acid (3:2) and methanol (1:2) and water (0:8).

PHASE-4: diethyl ether (65%), hexane (30%) and acetone (10%).



**FIG: 4.4 PREPARATION OF MOBILE PHASE-1**



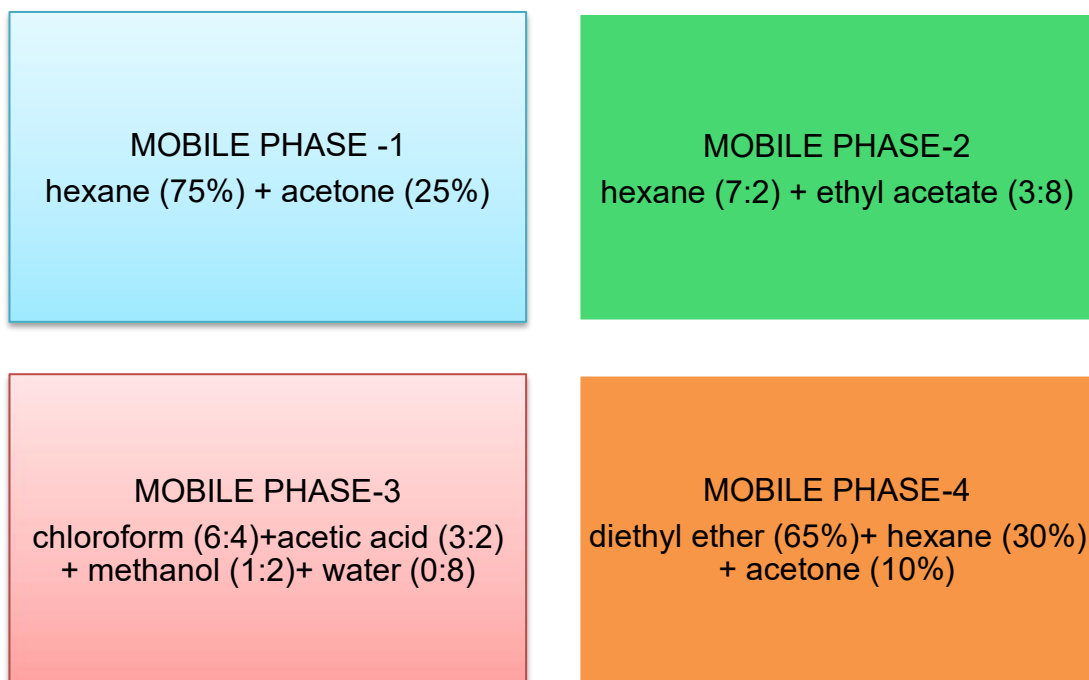
**FIG: 4.5 PREPARATION OF MOBILE PHASE -2**



**FIG: 4.6 PREPARATION OF MOBILE PHASE-3**



**FIG: 4.7 PREPARATION OF MOBILE PHASE -4**



**FIG: 4.8 COMBINATION OF MOBILE PHASE**

#### **4.4 PHYTOCHEMICAL ANALYSIS:**

The acetone and ethanolic extracts were utilised to evaluate the phytochemicals for flavonoids, alkaloids, tannins, sterols, glycosides, phenols, saponins, terpenoids, carboxylic acid, and resins. The signs for presence and absence were denoted by plus (+) and minus (–), respectively. The presence of two plusses (++) suggested strong reactivity, or a high phytochemical concentration. (lebot, 2023)

##### **4.1.3 TOTAL PHENOLIC CONTENT (TPC):**

Standards were created with 1 µg, 2 µg, 4 µg, 6 µg, 8 µg, and 10 µg of Gallic acid diluted in 200 µL of volume. A quadruplicate of 20 µL of standard was applied to each well. Each well that held 20 µL of standards and unidentified samples received around 80 µL of 7% calcium carbonate added to it. Each well received 100 µL of a 1:10 diluted Folin–Ciocalteu reagent in deionized water. Following the addition of the Folin–Ciocalteu reagent, the absorbance was measured at 700 nm. Gallic acid equivalents (GAE) per gram of sample in dry weight (mg/g) were the measure used to express the extract's total phenolic content. (lachman, 2019).

$$Y = mx + c$$

$$1.737 = 0.6736x + 0.3112$$

$$= 0.6736x + 0.3112 - 1.737$$

$$x = -1.4258 / 0.6736$$

$$= -2.1166$$

#### **4.5 GAS CHROMATOGRAPHY/MASS SPECTROMETRY:**

An electron impact mode operating at 70 eV was employed for the investigation. A 0.75-minute injectable purge was conducted while using the split less mode. The transfer line temperature was kept at 280°C while the injector temperature was adjusted to 250°C. The DB-1701 column, measuring 30 metres in length, 0.25 millimetres in inner diameter, and 0.25 millimetres in film thickness, was utilised. An 80°C start point was maintained for two minutes, after which a ramping temperature programme was implemented, increasing the temperature by 20°C per minute for ten minutes. An extra ten minutes were then spent maintaining the temperature. Helium was employed as the carrier gas, flowing at a rate of 1.2 mL min<sup>-1</sup>, with a solvent delay of three minutes. Scan mode was utilised for the operations. (Sheela, 2019).

#### **4.6 IMMUNOMODULATORY ACTIVITY:**

The ability of a drug to either stimulate or inhibit the immune system's reaction is known as immunomodulatory activity. It refers to the ability of plant extracts to either favourably or unfavourably affect immunological function. (Riaz, 2021).

#### **MATERIALS USED:**

1. Dioscorea alata extract
2. Peripheral blood mononuclear cells (PBMCs)
3. RPMI-1640 medium

4. Fetal bovine serum (FBS)
5. Penicillin-streptomycin solution
6. Phorbol 12-myristate 13-acetate (PMA)
7. Lipopolysaccharide (LPS)
8. Enzyme-linked immunosorbent assay (ELISA) kits for cytokine quantification
9. Trypan blue solution
10. Haemocytometer
11. Cell culture plates and incubator
12. Pipettes, tips, and other laboratory consumables

**Protocol:**

**1. Isolation and Culture of PBMCs:**

- a. Collect peripheral blood from healthy volunteers using standard venipuncture techniques.
- b. Isolate PBMCs using density gradient centrifugation with Ficoll-Paque solution.
- c. Wash the isolated PBMCs with RPMI-1640 medium supplemented with 10% FBS and 1% penicillin-streptomycin.
- d. Adjust the cell concentration to  $1 \times 10^6$  cells/mL.

**2. Treatment Groups:**

- a. Prepare different concentrations of *Dioscorea alata* extract in culture medium (e.g., 10 µg/mL, 50 µg/mL, 100 µg/mL).
- b. Prepare positive control groups with known immunostimulants such as PMA (10 ng/mL) and LPS (1 µg/mL).
- c. Include negative control groups with only culture medium.

### **3. Cell Stimulation and Incubation:**

- a. Plate the PBMCs in 96-well cell culture plates (100  $\mu$ L/well).
- b. Add the *Dioscorea alata* extract and control substances to appropriate wells.
- c. Incubate the plates at 37°C in a humidified atmosphere with 5% CO<sub>2</sub> for 24 to 48 hours.

### **4. Assessment of Immunomodulatory Activity:**

- a. After incubation, collect the supernatants for cytokine analysis using ELISA kits according to the manufacturer's instructions.
- b. Measure the levels of pro-inflammatory (e.g., interleukin-6, tumour necrosis factor-alpha) and anti-inflammatory (e.g., interleukin-10) cytokines.
- c. Evaluate cell viability using trypan blue exclusion assay and calculate the percentage of viable cells.

### **5. Data Analysis:**

- a. Analyse the cytokine levels and cell viability data using appropriate statistical methods (e.g., ANOVA followed by post-hoc tests).
- b. Compare the immunomodulatory effects of *Dioscorea alata* extract with positive and negative controls.

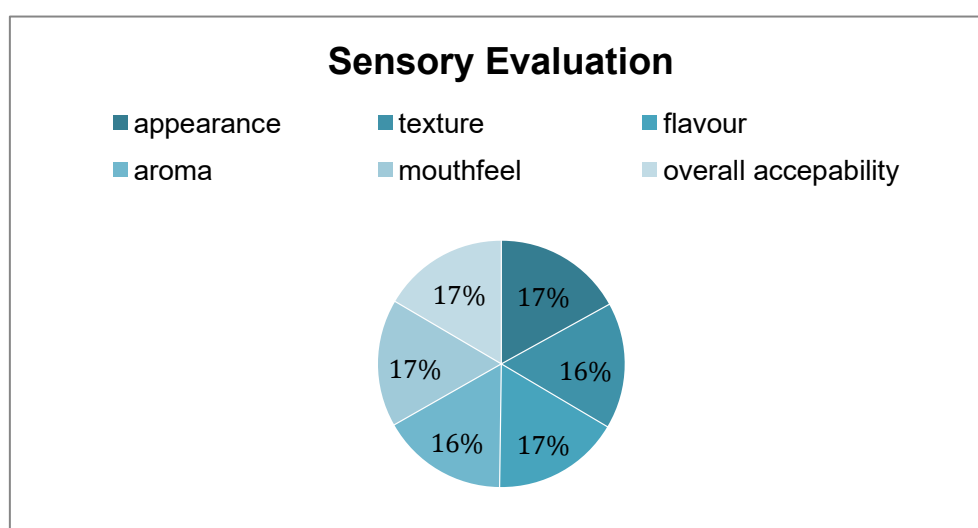
## 4.7 SENSORY EVALUATION:

### QUALITY ANALYSIS:

Via sensory evaluation with a broad group of 100 individuals, this study explores how consumers perceive yam bites. This research aims figure out the optimum yam bites features across various age demographics by assessing their comments on aspects like look, texture, flavour, aroma, and mouthfeel.

**TABLE: 4.2 TOTAL MEAN AND STANDARD DEVIATION OF RATINGS**

| CATEGORIES          | MEAN $\pm$ STANDARD DEVIATION |
|---------------------|-------------------------------|
| APPEARANCE          | 9.9 $\pm$ 0.438               |
| TEXTURE             | 9.65 $\pm$ 0.538              |
| FLAVOUR             | 9.75 $\pm$ 0.5                |
| AROMA               | 9.65 $\pm$ 0.538              |
| MOUTHFEEL           | 9.74 $\pm$ 0.504              |
| OVERALLACCEPABILITY | 9.65 $\pm$ 0.588              |



**FIG: 4.8 REPRESENTING RATING OF SENSORY EVALUATION**

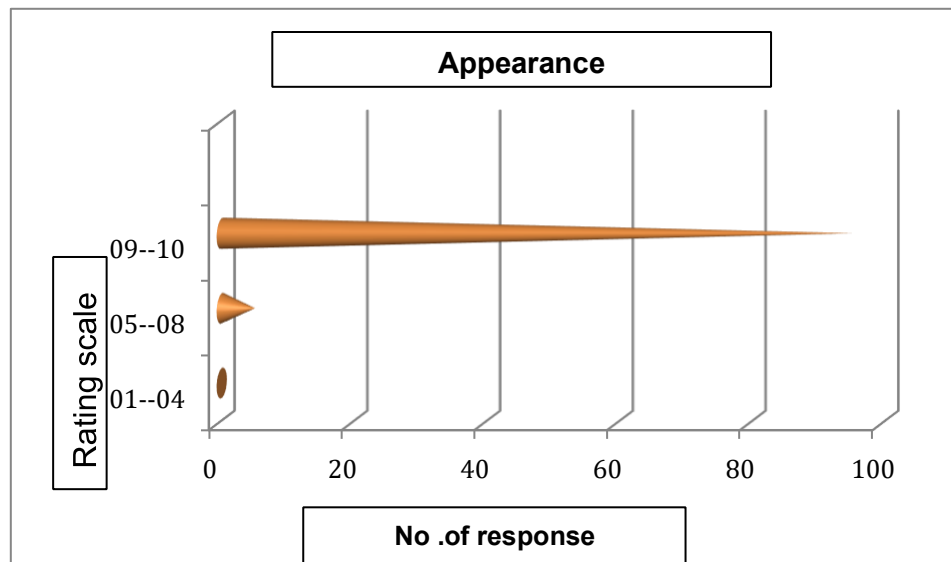
The above figure shows the sensory evaluation results from 100 individuals

## QUESTIONNAIRE:

### 1. APPEARANCE:

**TABLE: 4.3 RATING SCALE AND NO. OF RESPONSES**

| Rating scale(1-10) | No. of responses |
|--------------------|------------------|
| 1-4                | 0                |
| 5-8                | 5                |
| 9-10               | 95               |



**FIG: 4.9 REPRESENTING THE RATING FOR APPEARANCE**

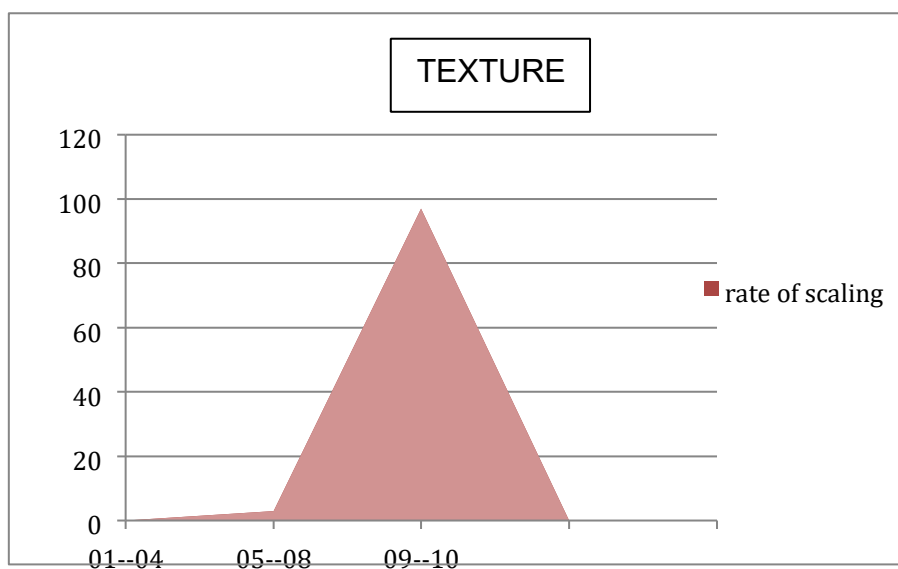
## INTERPRETATION:

It was found that majority of the people liked the appearance of the product.

## 2. TEXTURE:

**TABLE: 4.3 RANGE OF RATING AND NO. OF RESPONSE**

| RANGE OF RATING | NO.OF RESPONSES |
|-----------------|-----------------|
| 1-4             | 0               |
| 5-8             | 3               |
| 9-10            | 97              |



**FIG: 4.10 REPRESENTING THE RATING OF TEXTURE**

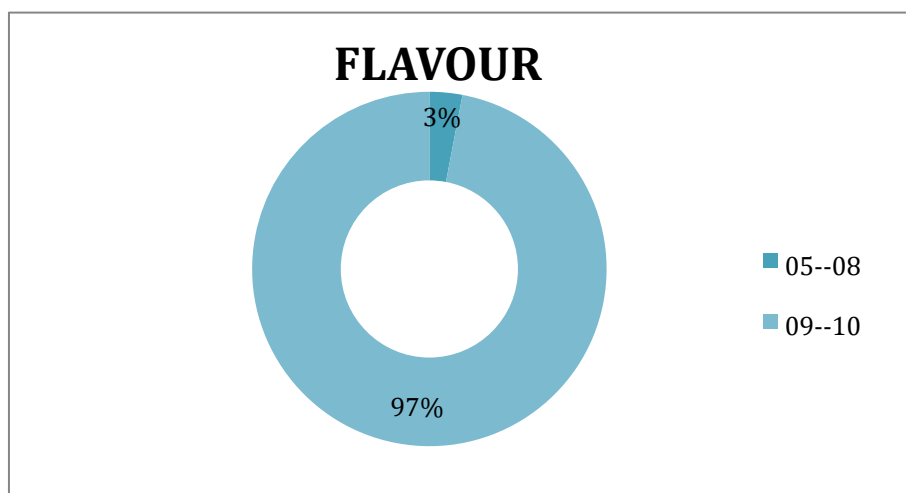
**INTERPRETATION:** It was found that majority of individual liked the texture.

### 3. FLAVOUR:

**TABLE: 4.3 RANGE OF RATING AND NO. OF RESPONSE**

| RANGE OF RATING(1-10) | NO.OF.RESPONSE |
|-----------------------|----------------|
| 0-4                   | 0              |
| 5-8                   | 3              |
| 9-10                  | 97             |

**FIG:**



**4.11**

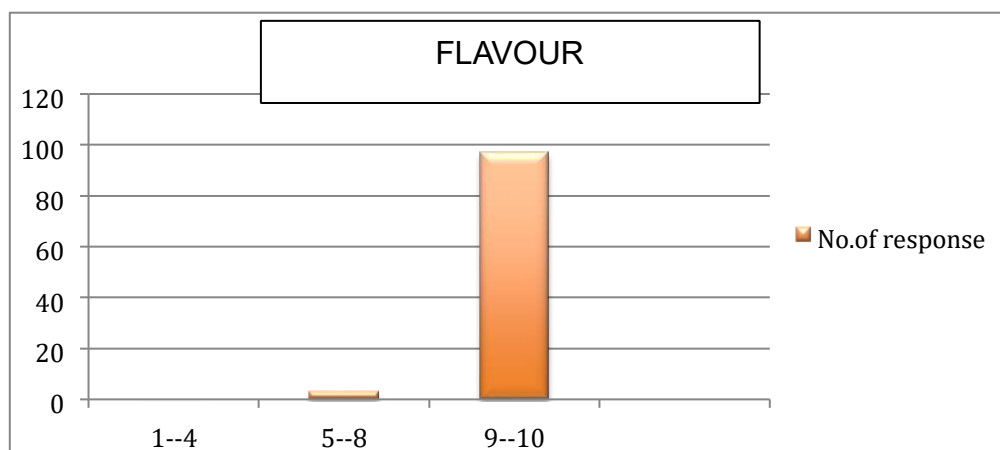
***REPRESENTING THE RATING OF FLAVOUR***

**INTERPRETATION:** It was found that majority of individual liked the flavour.

#### 4. AROMA:

**TABLE: 4.4 REPRESENTING THE RATING SCALE FOR AROMA AND ITS RESPONSE**

| Rating scale | NO.OF RESPONSES |
|--------------|-----------------|
| 1-4          | 0               |
| 4-8          | 3               |
| 9-10         | 97              |



**FIG: 4.12 REPRESENTING THE RATING OF AROMA**

#### INTERPRETATION:

It was found that majority of individuals liked the flavour of the product.

## 5. MOUTHFEEL:

**TABLE: 4.5 REPRESENTING THE RATING SCALE FOR MOUTHFEEL AND ITS RESPONSE**

| RATE OF SCALING(1-10) | NO.OF RESPONSES |
|-----------------------|-----------------|
| 1-4                   | 0               |
| 5-8                   | 3               |
| 9-10                  | 97              |



**FIG: 4.13 REPRESENTING THE RATING OF MOUTHFEEL**

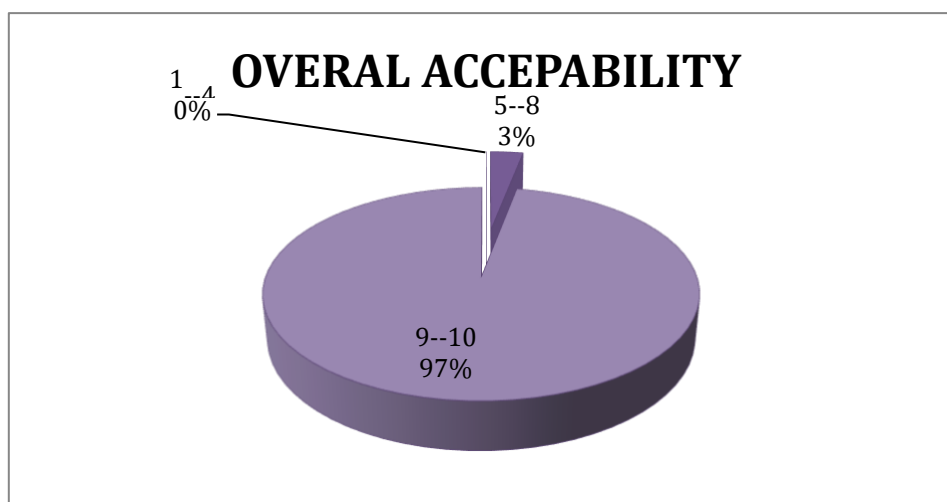
## INTERPRETATION:

It was found that the majority of individual liked the mouthfeel of the product.

## 6. OVERALL ACCEPTABILITY:

**TABLE: 4.6 REPRESENTING THE RATING SCALE FOR OVERALL ACCEPTABILITY AND ITS RESPONSES**

| RATE OF SCALING(1-10) | NO.OF RESPONSES |
|-----------------------|-----------------|
| 1-4                   | 0               |
| 5-8                   | 3               |
| 9-10                  | 97              |



**FIG: 4.14 REPRESENTING THE RATING FOR OVERALL ACCEPTABILITY**

### INTERPRETATION:

It was found that majority of individual liked the overall acceptability of the product.

## CHAPTER-5

### RESULT AND DISSCUSSION

The results and discussion pertaining to the study of nutritional analysis KASA FUSION BITES (Gluten free) using Dioscorea Alata is discussed under the following headings.

#### 5.1 MACRO –NUTRIENT COMPOSITION

**TABLE: 5.1 MACRO- NUTRIENT COMPOSITION TABLE**

| S.NO | PARAMETERS    | UNITS     | RESULTS |
|------|---------------|-----------|---------|
| 1.   | Energy        | Kcal/100g | 419     |
| 2.   | Carbohydrates | g/100g    | 67.3    |
| 3.   | Total Fat     | g/100g    | 12.5    |
| 4.   | Protein       | g/100g    | 9.22    |

From the table it is observed that the macro – nutrient composition of 100g of Nutri fusion bar using (Disocorea alata, dates, peanuts, and sesame seeds) had 419 kcals of Energy, 67.3g of Carbohydrates, and 12.5g of Protein and 9.22g of Total Fat.

#### 5.2 MICRO – NUTRIENTS COMPOSITION

**TABLE: 5.2 MICRO NUTRIENT COMPOSITION TABLES**

| S.NO | PARAMETERS    | RESULTS |
|------|---------------|---------|
| 1    | Dietary Fibre | 4.43    |
| 2    | Sodium        | 72.6    |
| 3    | Calcium       | 126     |
| 4    | Iron          | 2.65    |

Micro minerals like Dietary Fibre, Sodium, Calcium and Iron content from 100g in KASA FUSION BITES is contain 4.43g of Dietary Fibre, 72.6mg of Sodium, 126mg of Calcium and 2.65mg of Iron.

### 5.3 SENSORY EVALUATION OF FUSION BITES:

The appearance, texture, flavour, aroma, mouthfeel and the overall acceptability is very much liked by the majority of individuals.

### 5.5 GCMS ANALYSIS:

**TABLE: 5.3 PEAK REPORT**

| Peak | Reading Time | Area      | Area%  | Name                                                               |
|------|--------------|-----------|--------|--------------------------------------------------------------------|
| 1    | 41.424       | 894400    | 0.19   | Hexadecanoic acid, 1-(hydroxyethyl)-1,2-ethyl ester                |
| 2    | 44.261       | 1864209   | 0.40   | 9,12-Octadecadienoyl chloride, (Z,Z)-                              |
| 3    | 44.354       | 5585105   | 1.19   | 9-Octadecenoic acid, 1,2,3-propanetriyl ester,                     |
| 4    | 44.475       | 1016815   | 0.22   | 3-HEPTYL-1-CYCLOPENTENE                                            |
| 5    | 44.761       | 699426    | 0.15   | Glycidol stearate                                                  |
| 6    | 46.060       | 2429068   | 0.52   | OCTADECANOIC ACID, 2,3-BIS[(1-OXOTETRAHYDRO-2H-PYRAN-2-YL)METHOXY] |
| 7    | 46.551       | 28843334  | 6.16   | DODECANOIC ACID, 1,2,3-PROPANETRIYL ESTER                          |
| 8    | 46.654       | 22891935  | 4.89   | DODECANOIC ACID, 1,2,3-PROPANETRIYL ESTER                          |
| 9    | 47.167       | 359579136 | 76.83  | DODECANOIC ACID, 1,2,3-PROPANETRIYL ESTER                          |
| 10   | 48.269       | 44240962  | 9.45   | DODECANOIC ACID, 1,2,3-PROPANETRIYL ESTER                          |
|      |              | 468044390 | 100.00 |                                                                    |

The Gas Chromatography-Mass Spectrometry (GCMS) analysis revealed a significant presence of lauric acid, indicating high lauric acid content in the sample." It uses in making skincare products like soaps, lotions, and creams for its antimicrobial and moisturizing properties.

## 5.6 IMMUNOMODULATORY PROPERTY:

### Immunomodulatory Activity Testing

Name of the User: Dr. Stanley

No. of Compounds: 1 (Crude Extract)

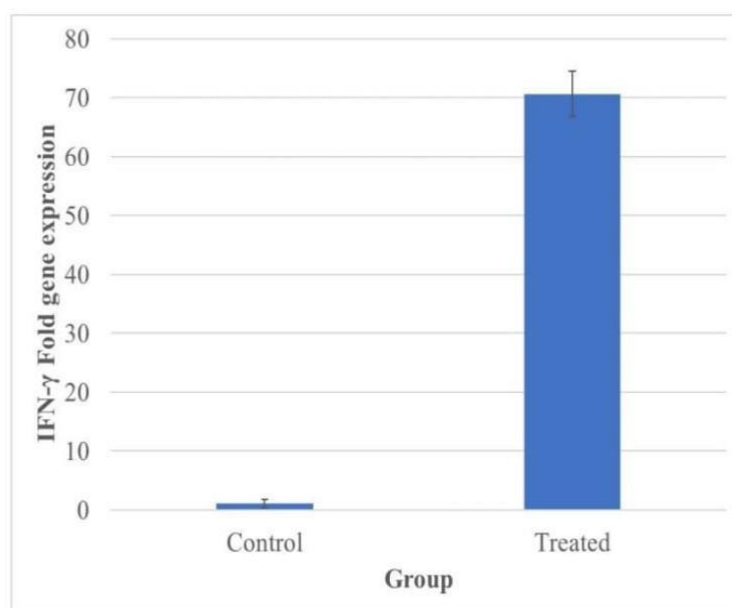
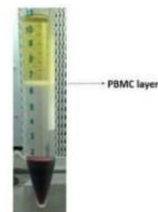
Given Concentration: 166 mg/ml

Target Gene: IFN- $\gamma$

Method: RT-PCR

Cells: PBMCs

Compound Test Concentration: 30 $\mu$ g/ml (1/2 of MIC50). RT-PCR was done in triplicates.



**Inference:** The given crude extract induces IFN- $\gamma$  expression in PBMCs in the absence of any other external stimulant.

**FIG: 5.1 REPORT OF IMMUNOMODULATORY ACTIVITY**

## CHAPTER-6

### SUMMARY AND CONCLUSION

The thesis aims to explore the phytochemical composition, immunomodulatory properties, and nutritive analysis of *Dioscorea alata*, commonly known as purple yam, and develop innovative products from it. The process involves using dried yam flour, dates, sesame seeds, peanuts, and flaxseeds to create nutritious balls. Through formulation experiments, the optimal combination of ingredients is determined. Thin-layer chromatography (TLC) is employed to separate and analyse biomolecules from the mixture, utilizing various solvent phases. Phytochemical analysis involves evaluating the presence of compounds like flavonoids, alkaloids, tannins, and more. Total phenolic content (TPC) is measured using Gallic acid equivalents. Gas chromatography/mass spectrometry (GC/MS) is utilized to analyse chemical components. Immunomodulatory activity is assessed by stimulating or inhibiting the immune system's reaction using PBMCs and ELISA kits. Sensory evaluation is conducted to assess the quality of the yam bites, with majority favorably rating appearance, texture, flavor, aroma, mouthfeel, and overall acceptability.

## REFERENCE

1. Adewale, p. (2022). *starch modifications*. A. B. . Effect of freezing methods, yam species, and storage on the quality attributes of fried yam chips. Ph.D. thesis. University of Ibadan, Nigeria
2. Agudelo. (2014). *isolation and characterization of yam*. Phenotypic and molecular assessment of genetic structure and diversity in a panel of winged yam(*Dioscorea alata*) clones and cultivars.
3. Ashri. (2014). *isolation and charterisation of yam*. Biochemical origin of browning duringthe processing of fresh yam (*Dioscorea spp.*) into dried product
4. Barooah, d. (2014). *dioscorea alata*. Physico-chemical, functional and pasting properties of native and chemically modified water yam (*Dioscorea alata*) starch and production of water yam starch-based yoghurt. Starch
5. Bhandari, M.R.; Kasai, T.; Kawabata, J. Nutritional Evaluation of Wild Yam (*Dioscorea Spp.*) Tubers of Nepal
6. Chaudhuri. (2021). *phytochemical*. Carotenoid profiling of tropical root crop chemotypes from Vanuatu, South Pacific. J. Food Comp. Anal
7. Cheng. (2021). Diversity of anthocyanins and other phenolic compounds among tropical root crops from Vanuatu, South Pacific. J. Food Comp. Anal
8. Chaudhuri, D., N. Phytochemical Analysis, Antioxidant and Anticancer Potential of Leaf Extracts from Edible Greater Yam, *Dioscorea Alata* L., from North-East India. Int. J. Phytopharm
9. Davies, k. (2017). *anthocyanins*. Nutritional and Medicinal Importance of *Dioscorea glabra* R. Baron, a Potent Wild Edible Plant Consumed
10. Dhanabal, S.P.; Mahendran, S.; Vadivelan, R. Antidiabetic Activity of Ethanolic Extract of Tubers of *Dioscorea Alata* in Alloxan Induced Diabetic Rats. Indian J. Pharmacol.
11. Harprakash. (2020). Study of antioxidant and reactive oxygenspecies scavenging activity of the edible tuber of “Greater Yam

- 12.Hhonfozo. (2021). *foo* Effects of extraction methods on yam (*Dioscorea alata*) starch characteristics. *Starch d composition analysis*
- 13.Hong.sh. (2019). *anti cancer*. Physicochemical Properties and Starch Digestibility of Autoclaved-Cooled Water Yam (*Dioscorea alata* L.) Flour.
- 14.Jiao. (2021). high performance liquid chromatography - ion trap time of flight tandem mass spectrometry Boiled yam end-user preferences and implications for trait evaluation
- 15.Kouassi 2019. Recent Highlights on Pharmacology and Analytical Methodology. Monitoring of Biochemical Parameters of Two Yam Species (*Dioscorea*
- 16.Kumar. (2017). Macronutrient Composition of Selected Traditional Yams Qualitative Phytochemical Analysis and Antibacterial Activity of *Dioscorea alata*
- 17.Ladjeu. (2020). Functional and pasting characteristics of modified water yam flour (*Dioscorea alata*
- 18.Lachman,(2019) J., Hamouz, K., Orsak, M., Pivec, V., Potato tubers as a significant source of antioxidant human nutrition. *Rostl Vyr.*, 2000, 46, 231-236.
- 19.lebot, v. (2023). *phthochemical content*. Secondary metabolites content may clarify the traditional selection process of the greater yam cultivars (*Dioscorea alata* L.)
- 20.Moriya. (2015). *nutrition composition of dioscorea alata*. Structural characterization of dioscorin, the major tuber protein of yams, by near infrared Raman spectroscopy
- 21.Moriya, C., Hosoya, T., Agawa, S., Sugiyama, Y., Kozone, I., Shin-ya, K., Terahara, N., Kumazawa, S., 2015. New acylated anthocyanins from purple yam and their antioxidant activity. *Bios. Biotech. Biochem.*
- 22.Scott, G.J., 2021. A review of root, tuber and banana crops in developing countries: past, present and future.
- 23.Padhan, A. (2020). Characterization of the Dioscorin Gene Family in *Dioscorea alata* Reveals a Role in Tuber Development and Environmental Response

24. *Ravi shankar*. (2019). Comparative analysis of proteins with stimulating activity on ovarian estradiol biosynthesis from four different *Dioscorea* species in vitro using both phenotypic and target-based approaches:
25. Respondex, S. (2019). Biological Activities and Applications of Dioscorins, the Major Tuber Storage Proteins of Yam
26. Paul (2022). *isolation and characterization of yam*. Antidiabetic activity of ethanolic extract of tubers of *Dioscorea alata* L. in alloxan induced diabetic
27. Salvan. (2019). Decrease of plasma glucose by allantoin, an active principle of yam (*Dioscorea* spp.)
28. Sartor. (2019). Changes in nutritional composition, functional, and sensory properties of yam flour as a result of pre-soaking.
29. Schwinn, k. (2017). *anthocyanins*. Effect of varieties on physicochemical and pasting characteristics of water yam flours and starches.
30. Sheela. (2019). *lauric acid*. *Dioscorea alata* L. Reduces Body Weight by Reducing Food Intake and Fasting Blood Glucose Level. British J. Med
31. Sherkat, R. (2017). Characterizing genotypic variation in biochemical composition, anti-nutritional and mineral bioavailability
32. Tamiru. (2017). Comprehensive characterization of yam tuber nutritional and medicinal quality of *Dioscorea opposita* and *D. alata* from different geographic groups
33. Wallace, K.; Asemota, H.; Gray, W. Acetone Extract of *Dioscorea Alata* Inhibits Cell Proliferation in Cancer Cells hanges in Water Yam (*Dioscorea Alata*) During Growth and Storage
34. Zaky. (2019). Ng, T.B., Lam, J.K.W., Wang, S.W., Lao, L., Zhang, K.Y.Z., Zhang, S.C.W., 2019. Research and Development of proteins and peptides with therapeutic potential from yam tubers.
35. Zhao. 2019. Isolation, Composition, Structure, Properties, Modifications, and Uses of Yam Starch.