PREPARATION OF PORRIDGE FROM GERMINATED MULTIGRAIN AND ITS NUTRITIONAL EVALUATION

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Preparation of Porridge from Germinated Multigrain and its Nutritional Evaluation

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Approval Letter

This *dissertation* entitled *Preparation of Porridge from Germinated Multigrain and Its Nutritional Evaluation presented* by **Aakansha Dhital** has been accepted as the partial fulfillment of the requirement for the degree of **B.Sc. Nutrition and Dietetics.**

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Abstract

Malnutrition, a state of improper nutrient balance in the body affects health and aggravates many infectious diseases. Protein energy malnutrition is still a major problem in all the segments of population. Complementary feeding is very important component of infant feeding. After 6 months, mother's milk is not sufficient for the growing child and weaning food should be started, timely and in adequate amounts. A nutritionally rich weaning food i.e. multigrain porridge was prepared suited for children aged 1 to 3 years from germinated wheat, buckwheat and green gram. Cereals are limited in essential amino acids such as lysine even though rich in threonine and tryptophan, while most oil seeds and legumes are rich in lysine and deficient in sulphur containing amino acids. Therefore, the combination of cereals and pulses in formulation of porridge gives a nutritious food containing all the amino acids. Thirteen products were formulated varying the proportion of cereals and pulses using design expert software (D-optimal). Locally available and cheap raw materials were used where cereals were used as the staple source, legumes as a protein source. From the thirteen products, only five products having low bulk density and low swelling index were chosen. From the sensory analysis the best product was found out. The grains selected were cleaned, washed, soaked, germinated, dried, roasted and then stored in air tight containers. The roasted grains were then mixed homogenously according to the required proportion and then milled. The milled grains were sieved and the grits obtained were kept in separate air tight containers.

The formulation containing wheat, buckwheat and green gram in the ratio 1:1:1 was selected as the best sample from the sensory evaluation. The protein, fat, carbohydrate, crude fiber and total ash of the product were found to be 28.32%, 2.3%, 63.73%, 3.15% and 2.5% respectively. The food can supply 388.90 kcal/ 100 gm. The iron and calcium content of the product were found to be 3.5 mg/100 gm and 82.4 mg/100 gm. Germination also increased the vitamin C content of the grains. While analyzing the amino acid profile of the final product qualitatively, it was found that all the essential amino acids were present in the product indicating as a complete food for a weaning child and can be scaled up for industrial purpose to provide low cost weaning food to growing children.

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Abbreviations	Full form
%	Percent
ANOVA	Analysis of Variance
Cm	Centimeter
d.f.	Degree of freedom
DFTQC	Department of Food Technology and Quality Control
F pr.	F probability ratio
FAO	Food and Agriculture Organization
Gm	Gram
hL	Hecto liter
ICMR	Indian Council of Medical Research
Kg	Kilogram
LSD	Least Significant Difference
m.s.	Mean squares
Mg	Milli gram
ND	Not detectable
NDHS	Nepal Health Demographic Survey
NPU	Net Protein Utilization
°C	Degree Celsius
PER	Protein Efficiency Ratio
RF	Retention Factor
RH	Relative Humidity
S.S.	Sum of squares
UK	United Kingdom

List of Abbreviations

UNU	United Nations University
v.r.	Variance ratio
WHO	World Health Organization

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Part I

Introduction

1.1 General Introduction

Malnutrition, a state of improper nutrient balance in the body affects health and aggravates many infectious diseases. Protein energy malnutrition is still a major problem in all the segments of population. A vast majority of the population in developing and underdeveloped countries do not have adequate nutrition to sustain a healthy life (Singh, 2012). Complementary feeding is very important component of infant feeding. After 6months, mother's milk is not sufficient for the growing child and complementary feeding should be started, timely and in adequate amounts (Siwakoti, 2014).

Foods such as "dal bhat" (rice-pulse) and "dhindo" (traditional corn meal), usually consumed by adults, are given in small amounts and in diluted form to young children (6-36) months Sour and cold foods are restricted. Roasted and ground soybeans, or corn and flattened rice moistened with water, are also given as snack foods. These adult foods are very difficult for a child to digest, and therefore cause diarrhea. Infants and young children may receive food left over from an earlier meal; because of improper and inadequate storage, these leftovers are often heavily contaminated. Protein of animal origin is costly and in short supply, beans legumes or seeds can complement cereals to increase the protein available from vegetable sources. Although it is not prepared daily, "*jaulo*" is a traditional weaning food made from rice, lentils, and green vegetables and intended for convalescing young children (Alnwick et al., 1988). Dalia is a traditional food in much of Northern Europe and Russia. Dalia was a typical means of preparing cereal crops for the table. It was also commonly used as prison food for inmates in the UK prison system and so "doing porridge" became a slang term for a sentence in prison. In India, Dalia is actually the blended course granules of wheat and various types of pulses. In developing countries breakfast meals for both adults and infants are based on local staple diet made from cereals, legumes, roots, cassava and potatoes tubers (Singh, 2012).

Malnutrition is a wide spread in almost all developing countries and the condition is particularly serious in children below 3 years of age. It is estimated that 30-50% of the baby attains 4-6 months of age, milk alone is no longer sufficient to meet its nutritional requirement. It needs some calories and other nutrients as supplements to milk till it is ready to eat fully the adult foods. This is the weaning stage. It is defined as gradual transition of a baby from the mother's milk to semi solids and solid foods. The semi solid foods given to the child at this stage are generally called weaning foods. On an average, 3% children under the age of five years suffer from protein energy malnutrition in developing countries. By introducing weaning food of food quality and quantity at right time and in right proportion, the incidence of protein energy malnutrition can be prevented to a large extent (Ashturkar *et al.*, 1992). Cereals provide a major food resource for man. Cereals are consumed by both adults and children. They are eaten in large quantities and are the main sources of both major and minor nutrients. They are prepared as gruel and used in feeding children. Due to their high viscosity on cooking, a large amount of water is used during preparation to obtain the right consistency. This obviously decreases the nutrient density. Methods developed to reduce viscosity include sprouting and fermentation of the cereals before use (Gupta *et al.*, 2016).

There has been a decline in malnutrition in Nepal in the last decade, but the figures are still alarming. According to NDHS 2011, 41% of children below 5 years of age are stunted, 11% of children are wasted and 29% of children below 5 year of age are underweight. According to NDHS 2016, 36% of children below 5 years of age are stunted, 11% of the children are wasted and 27% of the children below 5 years are under weight. Malnutrition contributes to high morbidity and mortality worldwide. Strong scientific evidence suggests that insufficient quantities and inadequate quality of complementary foods, poor child-feeding practices and heavy burden of infectious illnesses have adverse impact on child survival, growth and development. Poverty, food insecurity, ignorance, poor hygiene and sanitation are some other factors responsible for high levels of child malnutrition in developing countries (Keshari and Jacob, 2019).

Cereals are limited in essential amino acids such as lysine even though rich in threonine and tryptophan, while most oil seeds and legumes are rich in lysine and deficient in sulphur containing amino acids. Therefore, the combination of cereals and pulses in formulation of *dalia* gives a nutritious food containing all the amino acids. Porridge produced from cereals is eaten in many parts of world particularly in developing countries where they are part of the basic diet (Ocheme and Chinma, 2008). Porridge is known to be a traditional and staple food in many of the countries, especially the developing ones. In modern cultures, people are enjoying porridge as a healthy breakfast food. Porridge is a convenient food not just for weaning infants, but also for the elderly and convalescents due to its easy digestibility (Rhim *et al.*, 2011). Depending on the proportion of the cereals and liquid, two types of porridges are prepared for consumption that can be easily distinguished: thick and thin porridges. Thick porridges are solid-like and can be consumed with spoon or hand whereas thin porridge or gruel is taken in by drinking as having fluid or semi-fluid consistency. The infants are usually given thin porridge as a complementary meal (Moussa *et al.*, 2011).

Cereals and pulses are the major source of protein in the diet. A judiciously mixing of pulses with cereals was in practice long before food scientists and nutritionists understood the nutritional importance of this practice. The protein quality of pulses for food is low; however, when mixed with cereals total dietary quality can exceed 70% of milk protein. In the diet, nearly 85% of protein is supplied by plant and plant products. In this context, pulses are important and cheaper source of protein when compared to animal protein besides this; they are also good source of minerals and vitamins in the daily diets of the people, especially of low-income groups. In addition to this, pulses have been reported to reduce the levels of cholesterol and blood glucose. On this context, dietary importance of pulses is having global appreciation and recognition (Eggum and Beames, 1983).

In most of the developing countries, the generality of undernutrition and micronutrient deficiencies is high among infants and young children of the age group 6 to 23 months, which has increased the risk of underweight, stunt growth, and death at these ages (UNICEF, 2009). Ideally, the children in this age range are breastfed; however, as they get older, the energy and nutrient contribution of complementary food become increasingly necessary for meeting their daily requirements. For many infants and young children, however, the small quantities of cereal based porridges commonly fed to them do not contain enough of energy and micronutrients that can meet their daily requirements (Nestel *et al.*, 2003).

Weaning in human infants is a subject of controversy in terms of its initiation and correct method of doing it. The ideal age of weaning is six months. The desirable weaning food should be rich in calories and protein with adequate amount of trace elements like iron, calcium, vitamins etc. and also inexpensive, home available, clean, easily digestible and the most importantly bio-available. Complementary feeding is another very important component of infant feeding. After 6 months, mother's milk is not sufficient for the growing child and complementary feeding should be started, timely and in adequate amounts (Siwakoti, 2014).

1.2 Statement of Problem

The period between weaning and the age of five is nutritionally regarded as the most vulnerable period of the life cycle because that is when rapid growth, loss of passive immunity and the development of the immune system against infection occur. The first two years of child's life are particularly important, as optimal nutrition during this period lowers morbidity and mortality, reduces the risk of chronic diseases, and fosters better development overall (WHO, 2016). Malnutrition among infants and young children is common in developing countries. The high price of proprietary weaning foods and of vegetable and animal proteins and the non-availability of low-priced nutritious foods, combined with faulty feeding practices and late introduction of supplementary foods, are mostly responsible for aggravating the disorder among children. Protein-energy malnutrition generally occurs during the crucial transitional phase when children are weaned from liquid to semi-solid or fully adult foods. During this period, because of their rapid growth, children need nutritionally balanced, calorie-dense supplementary foods in addition to mother's milk (Wondimu and Malleshi, 1996).

Under nutrition is one of the major problems confronting infants and young children in developing countries such as Nepal. The problem arises from two factors: an inadequacy in the supply of food needed for infants and children; and ineffective utilization of such foods available. The weaning period is a critical one in child feeding; appropriate complementation during this period is essential to the nutritional well-being of the child (Alnwick *et al.*, 1988). Poverty and food insecurity seriously constrain accessibility of nutritious diets, including high protein quality, adequate micronutrient content and bioavailability, essential fatty acids, low anti-nutrient content, and high nutrient density (Baskota, 2019).

Nepal being a developing country, malnutrition has been its major problem. The trend of malnutrition is higher in under five children. Acute malnutrition affects 11 % of children aged below five years in Nepal, where 2.6 % are severely malnourished and 8.3% are moderately malnourished (UNICEF, 2012). One of the reasons for this is inappropriate initiation and correct method of doing complementary feeding practices after 6 months of age. The desirable weaning food should be rich in calories, protein and adequate amount of trace elements like iron, calcium, vitamins etc. and also inexpensive, home available, clean, easily digestible and the most importantly bio-available (UNICEF, 1999).

The traditional child-feeding practices in many developing countries reveal that the weaning period usually starts when the infant is 4 to 6 months old and is extended to the age of two to three years (Taba, 1970). During this time, special foods are rarely available for the children. Consequently, they have to depend on the same types of foods as those eaten by adults. In the poor countries, these foods are mainly starchy tubers like cassava and sweet potato, or cereals like maize, rice, wheat, sorghum, and millet. Small children are normally given these staples in the form of gruels, i.e., boiled with water. When prepared in this way, the starch structures bind large amounts of water, which results in gruels of high viscosity. Such gruels need to be further diluted with water in order to give a consistency that is appropriate for child feeding. This dilution, however, decreases the energy and nutrient density of the gruel, and the child has to eat large amounts in order to satisfy his requirements. This high volume/viscosity characteristic of a diet is usually referred to as "dietary bulk" (Mosha and Svanberg, 1983).

Industrial manufacture of cereal-based weaning foods often includes operations intended to reduce the dietary bulk like enzyme amylase treatment, pre-cooking, or extrusion. These processes modify the starch structures and hence result in lower water-binding in the gruels. However, such sophisticated technologies make rather expensive products even when low-cost alternatives are developed (Orr, 1977).

1.3 Objectives of study

1.3.1 General objective

The general objective of the work is to prepare porridge from the germinated multigrain and its nutritional evaluation.

1.3.2 Specific objective

- To formulate and evaluate multigrain porridge using varying proportion of wheat, green gram and buckwheat.
- To determine sensory and nutritional properties of prepared multigrain porridge.
- To study the amino acid profile of the final product.

1.4 Significance of study

This formula will be beneficial especially to children of low-income group. This study will provide a basis for the preparation of weaning food from locally available raw materials using traditional pretreatment technique i.e. germination. This weaning food could be

effective in terms of digestibility and bioavailability for growing children. It is nutritious for children. It is easy to prepare and is quick, thus, saves cooking time and require few cooking skills. The weaning food protein quality is also increased by the supplementation of cereal and pulses. Thus, it could be a complete food for growing children. The formulation could be effectively scaled up for industrial preparation and this work will provide the basis for further work in this field. So, this work primarily focuses as the alternative to industrial processes for reducing the dietary bulk of weaning foods using locally available raw materials that is inexpensive, easily digestible and bioavailable which is due to the increased use of improved traditional food preparation procedures that will also modify starch structures. One such procedure that is widely known and used is germination. Germination of cereals and legumes had been done to cause increases in thiamin, riboflavin, niacin, folic acid, ascorbic acid, iron, amylase and diastase activity, ,digestibility, protein efficiency ratio, and biological value (Alnwick *et al.*, 1988). In this study, porridge is prepared which is the combination of both germinated cereal and pulses.

1.5 Limitations of work

- *Invivo* evaluation of proteins and nutrient quality using albino rats could not be done.
- Quantitative analysis of amino acid could not be performed.

Part II

Literature review

2.1 Food and Nutrition

Food is anything that we eat and which nourishes our body. It is essential because it contain substances which perform important functions in our body. Food can be defined as anything eaten or drunk which meets the needs for energy, building, regulation and protection of the body. In short, food is the raw material from which our bodies are made. Intake of the right kinds and amounts of food can ensure good nutrition and health, which may be evident in our appearance, efficiency and emotional well-being (Mudambi and V, 2007).

Nutrition is defined as the scientific study of food and its relation to health. It is the science, which deals with those processes by which body utilizes food for energy, growth and maintenance of health (Joshi, 2010). The science of nutrition entails the study of all processes of growth, maintenance and repair of the living body which depend upon the intake of food (Beauman *et al.*, 2005). Good nutrition is a goal which can be achieved by anyone who desires it. Moderation in everything that we eat is the key to success. It is necessary to select wisely from the different foods available in the market. Always remember that no single food can be the complete health food for healthy living (Joshi, 2010). Combination of cereals and pulses improve the quality of cereal as well as pulse protein (Srilakshmi, 2014).

Good Nutrition is associated with human well-being. Right from the pregnancy to birth, during the infancy and in adulthood good nutrition plays a vital role in physical and mental well-being of human being including brain functioning, immune system and physical activities, which ultimately leads to increase overall productivity of human being. Good nutrition flows throughout the life cycle and across the generations, so it plays a central role in the country's overall development (Haddad *et al.*, 2015).

2.2 Weaning

The term weaning is commonly used to refer to the termination of lactation and suckling Weaning has also been used to describe a process that involves a gradual reduction in the frequency of suckling, a reduced dependence on breast milk and an increasing reliance on non-milk foods. According to this broader definition, the weaning process is initiated with the first intake of non-milk foods and culminates in the complete cessation of suckling .The weaning process is therefore bracketed by two distinct events that may be separated by days, months or even years (Humphrey, 2010).

Healthy children are full of energy and are active. The human milk alone, even in reasonable quantities, cannot provide all the energy and protein required for maintaining an adequate velocity of growth for the infant, after the age 6 months. It is therefore necessary to introduce more concentrated energy dense nutritional and iron supplements by this age (Kambli, 2012). Weaning begins at the first introduction of anything other than mother's milk. As a breast milk substitute or solid foods begin to be offered to the baby, the weaning process has begun (Mahan and Raymond, 2016). Adequate nutrition is essential to maintain optimum health of baby at the age of 6 months. Their growth and development are according to the expected norms and show no nutritional deficiency. Weaning means addition or introduction of semi-solid foods along with continuation of breast feeding as long as possible (Kambli, 2012). The term 'weaning' comes from the word 'weiman' which means to accustom. Weaning begins from the moment supplementary food is started and continues till the child is taken off the breast completely (Srilakshmi, 2014).

The introduction of solids into an infant's diet begins the weaning process in which the infant transitions from a diet of only breast milk or formula to a more varied one. Weaning should proceed gradually and be based on the infant's rate of growth and developmental skills. Weaning foods should be chosen carefully to complement the nutrient needs of the infant, promote appropriate nutrient intake, maintain growth (Mahan and Raymond, 2016). Weaning too early may cause baby at higher risk of developing digestive disorders and adverse reactions or allergies to certain foods. On the other hand, weaning too late may deprive adequate nutrition and can result in improper growth and development (Kambli, 2012). Introduction of weaning food too late can lead to under nutrition and increased diarrheal morbidity. The child may be unwilling to accept new food (Srilakshmi, 2014).

Analysts of weaning practices vary between and within countries. Urban and rural differences, age, breast problems, societal barriers, inadequate care from family, knowledge about good lactation, method of delivery, health system practices and community or cultural beliefs have all been found to influence breastfeeding and weaning (Ibrahim *et al.*, 2017). During weaning process different types of problems are seen in

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weaning children like obesity, food allergy, choking, diarrhea, underweight, etc (Srilakshmi, 2014).

Unwillingness of the infant to eat while exerting favorite to beverage rather than eating, allergic reactions and health problems with child including vomiting, abdominal colic and diarrhea may ascend due to wrong feeding practices adopted by mothers. However, it was estimated that, there were 10 million annual deaths of under-five year old children. Over one third of under-five mortality is caused by malnourishment related to inadequate complementary feeding during weaning practice. Initiate safe and nutritionally adequate complementary foods at 6 month is vital to achieve optimal growth, development and health of the children (Ibrahim *et al.*, 2017).

Weaning is the process by which a baby slowly gets used to eating family or adult foods and relies less and less on breast milk. The process varies from culture to culture and is often regulated by the child's individual needs. Healthy babies of weaning age are growing and developing very fast, so great care has to be taken to see that they are getting enough of the right kind of food (WHO, 1989).

During weaning, babies move about more and become more independent of their mothers. They start to come into greater contact with germs in the environment. At the same time the way in which a baby's body is protected against germs changes. When babies are very small, they still have protection (immunity) received from their mothers during pregnancy. But after about 4- 5 months this protection has gone, and babies start to develop their own immunity as they come into contact with germs in the environment. Because of this change babies are very likely to get infectious illnesses from the age of about 4-5 months especially if they are not breast-fed. This is why any food prepared for babies should be stored and fed to them in very hygienic ways (WHO, 1989).

Appropriate complementary feeding during the weaning period is a complex aspect of child feeding, and is critical to nutritional well- being. In infancy, growth is more rapid than at any period of life; nutritive requirements per unit of body weight are also greater. Good food sources of energy, protein, calcium, and iron are particularly important during this time. On the basis of body weight, children require twice as much protein, calcium, and iron as do adults (Alnwick *et al.*, 1988). Proper weaning foods should supply certain vitamins and minerals not present in breast milk, while providing additional calories. However, with the addition of food other than breast milk in the developing countries, there is a marked increase in the danger of gastroenteritis as a potentially fatal disease.

During weaning the infant living in an overcrowded, unhygienic environment is at great risk of developing acute diarrheal disease with its debilitation, and nutritional hazards, directly or indirectly leading to death (Brown, 1978).

In Nepal, complementary foods have traditionally been of low caloric density and low protein content, containing little or no fat, and often limited in micronutrients. Such foods are not well suited as supplements to the breastfed infant's diet. Moreover, supplementary feeding is usually initiated too late and in quantities that are inadequate. As a result of all these factors, infants are commonly half-starved from 6-12 months of age (Alnwick *et al.*, 1988).

2.3 Nutritional Requirement of the weaning infant

A nutritionally adequate weaning diet is essential for achieving optimal growth in the first year. Growth in the first year influences both the well-being of the child and the long-term health of the adult (Sajilata *et al.*, 2002). Infancy and childhood are both critical periods of rapid physical growth and cognitive and emotional development. It is also well recognized that nutrition in these age groups is not only an important factor for the normal development but also fundamental for future health status. Infants are considered a vulnerable group because they have relatively high nutrient requirements per unit body weight. Recent research has declared infancy is the critical period in life, setting the foundation of long- term health and reduced risk for chronic diseases. Breast feeding is the preferred and recommended form of nutrition for healthy infants during the first 6 months of life providing all necessary nutrients (Grammatikaki and Huybrechts, 2016).

After 6 months, supplementary feeding has to be resorted for a baby to ensure adequate nutrient intakes in line with infant nutritional requirements to maintain the expected rate of growth, remain healthy and well nourished. The amount of nutrients requirement of a baby per kg body weight declines over the period of birth owing to decreasing growth rate, even though energy requirement for activity increases as the infant becomes older (Srilakshmi, 2014). The requirement of nutrients of infants aged 1-3years are energy 1060 kcal, protein 16.7g/day, fat 27g/day, iron 9mg/day, calcium 600mg/day (ICMR, 2010). Table 2.1 shows recommended dietary allowances for children from 6 months to 3 years.

Nutrients	6-12 months	1-3 years
Body wt.	8.4	12.9
Net calories (kcal/kg)	80 kcal/kg	1060
Proteins (gm/kg)	1.69 gm/kg	16.7
Visible fat (gm)	19	27
Calcium (mg)	500	600
Iron (mg)	5	9
Vitamin A (µg) Retinol	350	400
β- Carotene	2800	3200
Zinc (mg)	-	5
Magnesium (mg)	45	50
Thiamine (mg)	0.2	0.5
Riboflavin (mg)	0.6	0.8
Niacin (mg)	650 µg/kg	8
Pyridoxine (mg)	0.4	0.9
Vitamin B12 (µg)	0.2	02-1.0
Ascorbic acid (mg)	25	40
Dietary folate (µg)	25	80

 Table 2.1 Recommended dietary allowances for children from 6 months to 3 years

Source : ICMR (2010)

2.4 Ingredients used in porridge preparation

2.4.1 Cereals

A cereal is any grass cultivated for the edible components of its grain composed of the endosperm, germ, and bran. Cereal grains are grown in greater quantities and provide more food energy worldwide than any other type of crop and are therefore staple crops. In their natural form (as in whole grain), cereals are a rich source of vitamins, minerals, carbohydrates, fats, oils, and protein. When refined by the removal of the bran and germ, the remaining endosperm is mostly carbohydrate (Serna-Saldivar, 2016). Cereal contains about 6-12% proteins and is good sources of some B-vitamins, e.g. thiamine, niacin, pantothenic acid and vitamin B_6 and minerals, e.g. phosphorous and iron. Hence, they

provide about 70-80% of the calories, proteins and other nutrients mentioned above in the diets of the low-income groups. The limiting amino acids in different cereal proteins are lysine and threonine (Swaminathan, 1978). Additionally, cereals can be used as sources of non-digestible carbohydrates that besides promoting several beneficial physiological effects can also selectively stimulate the growth of lactobacilli and bifido bacteria present in the colon and act as prebiotics. Cereals contain water-soluble fiber, such as β -glucan , oligosaccharides, such as galacto and fructo oligosaccharides and resistant starch, which have been suggested to fulfill the prebiotics concept (Charalampopoulos *et al.*, 2002).

2.4.1.1 Wheat

Wheat is the dominant crop in temperate countries being used for human food and livestock feed. Wheat is an important source of carbohydrates. Wheat also contributes essential amino acids, minerals, and vitamins, and beneficial phytochemicals and dietary fiber components to the human diet, and these are particularly enriched in whole-grain products (Shewry, 2009). It is the most important cereal, next to rice.

Since, the proteins present in the wheat endosperm (gluten proteins) are particularly poor in lysine, white flours are more deficient in lysine compared with whole grains. Supplementation with proteins from other food sources (mainly legumes) is commonly used to compensate for this deficiency, since the limitation of a single essential amino acid causes the others to break down and become excreted, which is especially important during the period of growth. Further, wheat is a major source for natural and bio-fortified nutrient supplementation, including dietary fiber, protein and dietary mineral (Suhasini and Malleshi, 2003).

The nutrient composition of 100 gm wheat is presented in Table 2.2

Parameter	Values
Moisture (gm)	12.2
Protein (gm)	12.7
Fat (gm)	1.7
Carbohydrate (gm)	69.4
Fiber (gm)	1.9
Calcium (mg)	48
Iron (mg)	4.9

Table 2.2 Nutrient composition of 100 gm wheat

2.4.1.2 Buckwheat

Buckwheat has attracted more and more attention due to its nutritional and medicinal values in recent years. Buckwheat contains many important bioactive compounds such as protein rich in essential amino acids, oil rich in essential fatty acids, starch with a low glycemic index, polyphenols compounds (including rutin, quercetin, orientin, vitexin, isovitexin and isoorientin) and many essential minerals (Bai *et al.*, 2015).

Buckwheat is rich in dietary fiber which has a positive physiological effect in the gastrointestinal tract and also significantly influences the metabolism of other nutrients. Buckwheat seeds contain no gluten so they are safe for people with celiac disease. It influences the metabolism of other nutrients. Rutin (quercetin-3-rutinosid) is a flavonol glycoside synthesized in higher plants as a protectant against ultra violet radiation and diseases. It also decreases the permeability of the blood vessels and has an anti-oedema effect, reduces the risk of arteriosclerosis, etc (Vojtíšková *et al.*, 2020).

The nutrient composition of 100 gm buckwheat is presented in table 2.3

Parameter	Values
Moisture (gm)	11.3
Protein (gm)	10.3
Fat (gm)	2.4
Carbohydrate (gm)	65.1
Fiber (gm)	8.6
Calcium (mg)	64
Iron (mg)	15.5
Vitamin C (mg)	0

Table 2.3 Nutrient composition of 100 gm buckwheat

Source: DFTQC (2012)

2.4.2 Pulses

Legume seeds generally contain 20% to 30% protein and are lysine rich, complementing the nutritional profiles of cereals and tubers in the diet (Graham and Vance, 2003). Legumes in general are good sources of lysine and threonine. However, they are poor sources of sulphur amino acids and tryptophan. Hence, legume proteins supplement

effectively cereal proteins. Legumes are good sources of thiamine and riboflavin and fair sources of niacin. They are fair sources of calcium and iron and good sources of phosphorous. They contain fair amounts of unavailable carbohydrates (Swaminatham, 1974). Grain legumes are cultivated for their seeds. The seeds are used for human and animal consumption or for the production of oils for industrial uses. Grain legumes include beans, lentils, peas, and peanuts (Whyte *et al.*, 1953).

2.4.2.1 Green gram

Mung bean is well known as green gram or moong bean. Mung bean has been consumed as a common traditional food worldwide for more than 3500 years (Kole, 2007). Green gram is a protein rich staple food. It contains about 23-25% protein which is almost three times that of cereals (Khattak and Bibi, 2007) and are low in fat and have no cholesterol (Dostalova *et al.*, 2009).

Green gram is an excellent source of protein and is almost free from flatulence-causing factors. Because of this, green gram seeds are preferred for feeding babies and those convalescing. The seeds contain a higher proportion of lysine than any other legume seeds (Adsule *et al.*, 1986).

The nutrient composition of 100 gm green gram is presented in table 2.4

Parameter	Values
Moisture (gm)	10.4
Protein (gm)	24
Fat (gm)	1.3
Carbohydrate (gm)	56.7
Fiber (gm)	4.1
Calcium (mg)	124
Iron (mg)	7.3
Vitamin C (mg)	0

Table 2.4 Nutrient composition of 100 gm green gram

Source: DFTQC (2012)

2.5 Nutritive value of cereals and pulses

It was studied that the combination with cereals and vegetables for producing composite weaning mixes, which will prove to be of immense benefit especially for young children in developing countries, because of their low cost and ease of preparation (Ghavidel and Prakash, 2010).

It was found that the protein content was slightly decreased in the roasted weaning food because of the heatdenaturation of proteins and more decreased in the maltedweaning food because of the leaching out of the soluble protein and removal of rootlets thereby reducing the total dry matter during malting (Kshirsagar, 1994).

It was found that wheat is good source of protein, minerals, B-group vitamins and dietary fiber i.e. an excellent health building food. It contains carbohydrate 78.10%, protein 14.70%, fat 2.10%, minerals 2.10% and considerable properties of vitamins (thiamine and vitamin B) and minerals (zinc, iron). The wheat germ, which is removed in the process of refining, is also rich in essential vitamin E (Kumar *et al.*, 2011).

A research which was done on the development and sensory evaluation of low cost weaning food formulations which found that germinated and supplemented grain flour weaning food formulations were more acceptable as compared to control products made from ungerminated grain flour (Sadana and Chabra, 2004).

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A research was conducted on the production and evaluation of breakfast cereal-based porridge mixed with sesame and pigeon peas for adults. It was found that the formulated products contained 10-13% protein, 75-8-% carbohydrate and 132.2-477.8% Kcal energy. Blended porridge was found to be rich source of potassium and calcium. They recommended that these could be used to meet specific nutritional requirements of different classes of people (Kanu *et al.*, 2009).

An instant porridge based on pre-cooked wheat *dalia* and milk solids has been developed for space astronauts. A ready-to-eat breakfast cereal containing instant milk powder, wherein milk powder is uniformly distributed among the cereal pieces, resulted in a uniform proportion of cereal and liquid milk in each serving when water was added (Jha *et al.*, 2015).

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A nutritional value was found by Nutrition Collaborative Research Program during the market analysis of complementary foods in Nepal where the analysis of Sarbottam Pitho showed Moisture 3.53%, Protein 14.72%, Fat 7.4%, Ash 1.92%, Carbohydrate 73% and Energy 400 kcal (Magnani *et al.*, 2012).

2.6 Physical characteristics of the grains

The 1000 kernel weight of the wheat grain ranges from 23.2 gm to 39.7 gm and the bulk density ranges from 74 kg/hL to 53.8 kg/hL (Tabatabaeefar, 2003). The 1000 kernel weight of the buckwheat ranges from 19.98 gm to 21.74 gm and the bulk density ranges from 59.86 kg/hL to 83.37 kg/hL (Unal et al., 2017). Similarly, the 1000 kernel weight of buckwheat was found to be 23.92 gm and the bulk density was found to be 83.9 kg/hL (Subedi, 2018a). The 1000 kernel weight of the green gram ranges from 28.19gm to 35.87 gm and the bulk density ranges from 70.8 kg/hL to 80.7 kg/hL (Nimkar and Chattopadhyay, 2002).

The moisture content of the wheat was found to be 12.20%, ash 1.01%, 10.02% protein, 1.92% fat, 1.51% fiber and 87.60% carbohydrate (Kavitha and Parimalavalli, 2014). The calcium and iron content of wheat was found to be 34.8 mg/100gm and 3.3mg/100gm (L Alvarez-Jubete *et al.*, 2009). Similarly the moisture content of the buckwheat was found to be 12.8%, 2.43% ash, 3.05 % fat, 0.96% fiber, 13.59% protein and 79.96% carbohydrate (Subedi, 2018a). The calcium and iron content of buckwheat was found to be 30 mg/100 gm and 3 mg/100 gm (L Alvarez-Jubete *et al.*, 2009). The moisture content of the green gram was found to be 12.07%, 3.85% ash, 21.57% protein, 1.53% fat, 0.63% fiber and 60.35% carbohydrate. The calcium and iron content of the green gram was found to be 70.89 mg/100 gm and 5.04 mg/100 gm respectively (Paul *et al.*, 2011). Similarly, the moisture content of the green gram was found to be 7.60%, 3.42% ash, 21.9% protein, 1.40% fat, 3.80% fiber and 65.12% carbohydrate (Subedi, 2018a). The antioxidant activity i.e. vitamin C of the dry grains is almost negligible (Yang, 2001). According to DFTQC (2012) the vitamin C content of raw wheat, buckwheat and green gram was found to be at non detectable level.

The moisture content of the germinated wheat was found to be 13.56%, 0.97% ash, 14.10% protein, 1.43% fat, 1.62% fiber and 80.32% carbohydrate (Parimalavalli, 2014). The moisture content of the germinated buckwheat was found to be 7.05%, 2.51% ash, 14.44%

protein, 2.55% fat, 1.69% fiber and 78.92% carbohydrate (Subedi, 2018b). The moisture content of the germinated green gram was found to be 9.65%, 3.10% ash, 31.83% protein, 1.20% fat, 3.40% fiber and 61.24% carbohydrate (Kavitha and Parimalavalli, 2014). Upon germination the concentrations of the antioxidant vitamin i.e. vitamin C steadily increased with increasing germination time which were barely detectable in dry grains (Yang, 2001).

In case of porridge preparation, low bulk density is desirable (Nicole *et al.*, 2010). Flours with high swelling index values hold large amounts of water during their preparation into gruels and thus become voluminous with a low energy and nutrient density. Sample with the least swelling index value would provide a more nutrient-density food for an infant (Ikpeme-Emmanuel *et al.*, 2009).

2.7 Germination

Germination is the process by which an organism grows from a seed or similar structure. The most common example of germination is the sprouting of a seedling from a seed of an angiosperm or gymnosperm. In addition, the growth of a sporeling from a spore, such as the spores of hyphae from fungal spores, is also germination. Thus, in a general sense, germination can be thought of as anything expanding into greater being from a small existence or germ. Germination is usually the growth of a plant contained within a seed; it results in the formation of the seedling; it is also the process of reactivation of metabolic machinery of the seed resulting in the emergence of radical and plumule. All fully developed seeds contain an embryo and, in most plant species some store of food reserves, wrapped in a seed coat. Some plants produce varying numbers of seeds that lack embryos; these are called empty seeds and never germinate. Dormant seeds are ripe seeds that do not germinate because they are subject to external environmental conditions that prevent the initiation of metabolic processes and cell growth. Under proper conditions, the seed begins to germinate and the embryonic tissues resume growth, developing towards a seedling (Bazaz *et al.*, 2016).

Seed germination depends on both internal and external conditions. The most important external factors include right temperature, water, oxygen or air and sometimes light or darkness. Various plants require different variables for successful seed germination (Raven *et al.*, 2005). Germination has often been proposed as a simple processing method by which the nutrient composition and certain functional properties of seeds might be improved and by which the quality of a cereal can be improved for both digestibility and

physiological function. During germination, enzymatic activity and bioactive compounds increased within the grain (I. Hussain and Uddin, 2012).

Germination (an intermediate step in malting) of cereals or of legumes has been shown to cause increases in thiamin, riboflavin, niacin, folic acid, ascorbic acid, iron, amylase and diastase activity, protease activity, digestibility, protein efficiency ratio, and biological value. Germination also has been shown to increase caloric density and to reduce viscosity, with decreases in phytin and increased phosphorous and trypsin activity of hemagglutinin (Alnwick *et al.*, 1988).

2.7.1 The imbibition phase

It is an initial phase and can take place in inert, dead and viable seeds. It is attributed to the passive water uptake and absorption by the seed colloids and into the crevices and interstices of the seed cover and tissues. Towards the end of this phase and the beginning of the transition phase, the water uptake becomes an active process as it is temperature dependent there is an increase in respiration rate and in some cases, it becomes light sensitive (Subedi, 2018a).

2.7.2 The transition phase

The changeover to this lag phase is not distinguishable and can take place in the dormant and non-viable seeds. It is also known as the pause phase, since in this phase the major metabolic events take place in preparation for radicle emergence. Any condition affecting the hydration level attained during imbibition may retard or even inhibit germination (Subedi, 2018a).

2.7.3 The growth phase

It occurs only in viable, non- dormant seeds. It coincides with the radicle profusion and is thus associated with the establishment of cellular division and extension and a rapid increase in water uptake rate. Non-president cotyledons do not achieve growth phase of water, eventually their water content declines as degradation occurs (Subedi, 2018a).

2.7.4 The seeding establishment phase

It is marked by the depletion of stored reserves disintegration of cotyledons, increase in photosynthesis. The duration of each of these phases depends on certain in-herient properties, e.g. hydrable substrate levels, seed coat permeability, seed size, oxygen uptake and on prevailing condition during hydration (Subedi, 2018a).

2.8 Method of processing of raw materials

2.8.1 Wheat

Wheat should be sorted, cleaned and soaked in water for 12 hours at 22°C and 60% humidity and then drained. It should be spread on a wetted muslin cloth and covered by a wetted muslin cloth. It should be kept for germination at 22°C.Water has to be sprinkled on the layer at 3 to 4-hour interval. The grain should be germinated for 3 days. It should be then dried in cabinet drier at 55°C for 3 hours and 70°C for 1 hour until moisture sufficiently reduced to about 5%. The germinated part has to be removed, then roasted and milled into grits and packed in air tight plastic bags (Tehseen *et al.*, 2014).

2.8.2 Buckwheat

Seeds should be screened to remove impurities such as stones, strings, weed seeds, etc. The grains should be then washed with water. The cleaned seeds have to be then immersed in water in a bowl. The light material present in the sample has to be skimmed off. Steeping should be done for 20 hours in room temperature which brought moisture content to the required level of 45-50%. Steeped water has to be drained off. The steep grain has to be first collected in a muslin cloth and swirled in order to drain excess water. The grains should be spread over the germination paper and covered with the germination paper and kept for germination in open environment at temperature 26-28°C and relative humidity 85-86% in open environment. During germination, the grains should be moisturized by sprinkling water at 6-hour interval and mixed gently to equalize temperature and to aerate the mass. The buckwheat germinated in should be taken in cabinet dryer to stop further germination. Drying has to be carried out at 45°C for 24 hours. The grains should be then rubbed and sprouts have to be removed with the help of screen. The malted grains should be then milled and sieved to obtain grits. The grits should then be stored in air tight container (Subedi, 2018a).

2.8.3 Green gram

The grains have to be screened to remove impurities and then the cleaned grains should be washed and soaked in excess water. The grains almost double in volume hence the trays of bigger sizes should be taken. Soaking should be done for 10-15 hour. At the end of soaking period, the soaked grains should be spread for germination. Water has to be sprinkled during germination to keep the grains moist. The germination has to be done for 24 hours at $30\pm2^{\circ}$ C. The sprouts should be then dried to stop the germination in hot air oven at 55° C

for 20 hours. The dried grains should be dehusked and the husk and cotyledons has to be removed. To improve the taste and flavor it has to be roasted under mild heat and then dry milled (Yasmeen, 2006). Sieving should be done before standardizing the size of grits. The sample should be then packed in airtight plastic container. The container has to be stored at room temperature until further use. After that weighing, blending and cooking process should be done as required (Rana *et al.*, 2015).

2.9 Medicinal values of the raw materials used

2.9.1 Wheat

The wheat, as produced by nature, contains several medicinal virtues. Every part of the whole-wheat grain supplies element needed by the human body. Starch and gluten in wheat provide heat and energy; the inner bran coats, phosphates and other mineral salts; the outer bran, the much-needed roughage the indigestible portion which helps easy movement of bowels; the germ, vitamins B and E; and protein of wheat helps build and repair muscular tissue. The wheat germ, which is removed in the process of refining, is also rich in essential vitamin E, the lack of which can lead to heart disease. The loss of vitamins and minerals in the refined wheat flour has led to widespread prevalence of constipation and other digestive disturbances and nutritional disorders. The whole wheat, which includes bran and wheat germ, therefore, provides protection against diseases such as constipation, ischemic, heart disease, disease of the colon called diverticulum, appendicitis, obesity and diabetes (Kumar *et al.*, 2011).

Lutein is the predominant carotenoids present in wheat and the bran/germ fractions of wheat contained greater amounts of carotenoids and antioxidant activity than the endosperm fractions. Lutein, along with zeaxanthin, is important for the health of skin and eyes in humans. Wheat also lowers the level of estrogen in the blood which reduces the risk of breast and prostate cancers. Wheat germ is sodium and cholesterol free, and dense in nutrients. It is rich in vitamin E, magnesium, pantothenic acid, phosphorus, thiamin, niacin and zinc. It is also a source of coenzyme Q_{10} (ubiquinone) and PABA (para-aminobenzoic acid). Wheat germ is also high in fiber. A diet high in fiber can be useful in regulating bowel function (i.e. reducing constipation), and may be recommended for patients at risk for colon disease, heart disease, and diabetes (Kumar *et al.*, 2011).

2.9.2 Buckwheat

Buckwheat is an excellent medicinal plant as well as a nutrient-abundant crop. Its flour and leaf contain large quantity of flavonoids compounds. Buckwheat contains plentiful vitamins. Vitamin B1 can help to enhance digestive function to resist neuritis and to prevent beriberi. Vitamin B2 can enhance human body's development and is a vital element for protection against perleche, glossitis and eyeliditis. Serving to reduce lipemia and cholesterol in human body, vitamin PP is an important auxiliary medicine for the treatment of hypertension and cardiovascular diseases. It has particularly good effect on senior patients for its capacity to lower down fragility and permeability of tiny blood vessels and to restore their elasticity, thereupon is effective for protection against cerebral hemorrhage, maintenance of ocular blood circulation, and preservation and promotion of eyesight. The content of tocopherol in vitamin E is relatively higher. It is effective for prevention of oxidation and for cure of sterility and contributes to cell regeneration and to deference of aging (Gang *et al.*, 2001).

Bioflavonoids are major contributors to the therapeutic attributes of buckwheat, with rutin being the primary functional component. A major health promoting property of the bioflavonoids is their functional role as antioxidants. Rutin, quercetin -3 rhamnoglucoside, a water-soluble glycoside, has well established antioxidant properties and acts as a free radical scavenger and iron chelator. It has been shown that it could offer protection from the cytotoxic effects of oxidized low density lipoprotein (Briggs *et al.*, 2004).

2.9.3 Green gram

Green gram is well known for its detoxification activities and is used to refresh mentality, alleviate heat stroke, and reduce swelling in the summer. The mung bean was recorded to be beneficial in the regulation of gastrointestinal upset and to moisturize the skin. As a food, mung beans contain balanced nutrients, including protein and dietary fiber, and significant amounts of bioactive phytochemicals. High levels of proteins, amino acids, oligosaccharides, and polyphenols in mung beans are thought to be the main contributors to the antioxidant, antimicrobial, anti-inflammatory, and antitumor activities of this food and are involved in the regulation of lipid metabolism (Tang *et al.*, 2014).

Flavone, isoflavone, flavonoids, and isoflavonoids are the important metabolites found in the green gram. Most flavonoids have polyhydroxy substitutions and can be classified as polyphenols with obvious antioxidant activity. Flavonoids are involved in stress protection (Li *et al.*, 2012). Organic acids and lipids have also been found in green gram and sprouts. Twenty-one organic acids, including phosphoric and citric acid, and 16 lipids, including γ -tocopherol, were reported in green gram (Bowles, 1990).

2.10 Amino acids

Amino acids, often referred to as the building blocks of proteins, are compounds that play many critical roles in our body. An amino acid may be defined as a chemical compound which possesses amino acid and acid groups (Meister, 1957). The WHO/ FAO/ UNU (2007) report examines dietary protein and amino acid requirements for all age groups, protein requirements during pregnancy, lactation and catch-up growth in children, the implications of these requirements for developing countries and protein quality evaluation. Requirements were defined as the minimum dietary intake which satisfies the metabolic demand and achieves nitrogen equilibrium and maintenance of the body protein mass, plus the needs for growth in children and pregnancy and lactation in healthy women (Millward, 2012).

2.10.1 Types of amino acids

2.10.1.1 Essential amino acids

Essential amino acids cannot be made by the body. As a result, they must come from food. The nine essential amino acids perform a number of important and varied jobs in the body:

- Phenylalanine: Phenylalanine is a precursor for the neurotransmitters tyrosine, dopamine, epinephrine and nor-epinephrine. It plays an integral role in the structure and function of proteins and enzymes and the production of other amino acids.
- Valine: Valine is one of three branched-chain amino acids, meaning it has a chain branching off to one side of its molecular structure. Valine helps stimulate muscle growth and regeneration and is involved in energy production.
- Threonine: Threonine is a principal part of structural proteins such as collagen and elastin, which are important components of the skin and connective tissue. It also plays a role in fat metabolism and immune function.
- Methionine: Methionine plays an important role in metabolism and detoxification. It's also necessary for tissue growth and the absorption of zinc and selenium, minerals that are vital to human health.
- Leucine: Like valine, leucine is a branched-chain amino acid that is critical for protein synthesis and muscle repair. It also helps regulate blood sugar levels, stimulates wound healing and produces growth hormones.

- Isoleucine: The last of the three branched-chain amino acids, isoleucine is involved in muscle metabolism and is heavily concentrated in muscle tissue. It's also important for immune function, hemoglobin production and energy regulation.
- Lysine: Lysine plays major roles in protein synthesis, hormone and enzyme production and the absorption of calcium. It's also important for energy production, immune function and the production of collagen and elastin.
- Histidine: Histidine is used to produce histamine, a neurotransmitter that is vital to immune response, digestion, sexual function and sleep-wake cycles. It's critical for maintaining the myelin sheath, a protective barrier that surrounds your nerve cells (Kubala, 2018).

2.10.1.2 Non-essential amino acids

Non-essential means that the body produces amino acid, even if the body doesn't obtain it from the food eaten. Non-essential amino acids include: alanine, arginine, asparagine, aspartic acid, cysteine, glutamic acid, glutamine, glycine, proline, serine and tyrosine (Kubala, 2018).

2.10.1.3 Conditional amino acids

Conditional amino acids are usually not essential, except in times of illness and stress. Conditional amino acids include: arginine, cysteine, glutamine, tyrosine, glycine, ornithine, proline and serine (Kubala, 2018).

2.11 Chromatography

Chromatography is a separation process which depends on the differential distributions of the components of a mixture between a mobile bulk phase and an essentially thin film stationary phase (Furniss, 1989). Chromatography is the most powerful technique to separate chemically closely related substances into the individual components on the basis of their physicochemical properties. These compounds are separated on the basis of their partition coefficients between two immiscible phases. The static phase may be solid or liquid while the mobile phase may be a solid, liquid or gas. Depending upon the static and mobile phases, a variety of chromatographic techniques are available. These include chromatography on paper, thin layer gel, ion- exchange resin etc. Although modern instrument facilities such as high performance liquid chromatography (HPLC) are available for the separation of chemical substance, the classical techniques- paper chromatography and thin layer chromatography are still easy, can be set up even in an ordinary laboratory without much expenditure (Sadasivam, 1996).

2.11.1 Principle of paper chromatography

The separation of the solute (amino acids) is based on the liquid- liquid partitioning of amino acids in paper chromatography. The partitioning takes place between the water molecule (static phase) adsorbed to the cellulosic matter of the paper and the organic (mobile) phase (Sadasivam, 1996).

2.12 RF value

RF value (in chromatography) is the distance travelled by a given component divided by the distance travelled by the solvent front. For a given system at a known temperature, it is a characteristic of the component and can be used to identify components. The constant RF is indicated by:

$$RF value = \frac{Distance (cm)moved by the solute from the origin}{Distance (cm)moved by the solvent from the origin}$$

The amino acid present in the sample are then identified by comparing the RF values with that of the authentic amino acids, co-chromatographed (Sadasivam, 1996).

2.13 Technology for the preparation of multigrain porridge

Traditional treatments such as soaking, cooking, germinating have been used to improve nutritional quality of the cereals and legumes. Processing of food such as soaking, germination and fermentation leads to reduction in phytic acid and increases of the mineral solubility in foods and also improves the bioavailability of the minerals in cereals and legumes (El-Adawy, 2002).

2.13.1 Soaking or steeping

Soaking or steeping is a pretreatment for decertification of grain facilitate the removal of the husk or skin. Non- corticated grains are soaked in water for a short time lead them to easy husk removal. Soaking process increases hydration coefficient, seed weight, total protein, ash, fat, fiber of cereals and legumes (El-Adawy, 2002).

The malting process begins when the cereal grain is steeped in water. Steeping is arranged so that sufficient moisture enters the grain to initiate germination. Time period for steeping depends on temperature and degree of aeration of the steep water. A temperature of 10- 12°C is recommended with steeping times of 40- 60 hours. A temperature of 20- 25°C is recommended with steeping times of 16- 20hours for legumes (Kent, 1994).

2.13.2 Germination

Germination or sprouting of legumes and cereals increase their palatability and nutritional value. Germination also slightly increases the total essential amino acids in cereals and legumes. Dehusking, germination, cooking and roasting have been shown to produce beneficial effects on nutritional quality of legumes (Kadam *et al.*, 1985). In the recent years, consumption of cereal sprouts has become an innovation in nutrition. They have received attention as a functional food due to many benefits they express on human health. These benefits have been mostly attributed to the content of dietary fiber, essential fatty acids, vitamins and antioxidant phytochemicals, including several phenolic compounds available in the cereals. Antioxidants present in whole grain cereals act in defense to remove the reactive oxygen species (ROS), thereby preventing and curing oxidative stress-related diseases (Šaponjac *et al.*, 2016).

2.13.3 Drying

During germination, and particularly during the drying treatment, the profile of volatile compounds will change (Heiniö *et al.*, 2001). Drying produce a friable, readily milled stable product that may be stored for long periods and from which roots may be easily be removed.

2.13.4 Roasting

Roasting process render grain digestible, without the loss of nutritious component and the grains are consumed throughout the world (Srivastav *et al.*, 1994). Roasting can enhance flavor through caramelization and Maillard browning on the surface of the food (Wikipedia). Roasting reduces the moisture content, thereby concentrating the food value. Roasting also enhance acceptability by imparting a nutty flavor to the food. Most of the anti-nutritional factors or toxic effects of legumes (trypsin inhibitor, hemagglutinin, goitrogenic agents, cyanogenic glucosides, alkaloids, etc.) are partially or fully eliminated by roasting (Ndidi *et al.*, 2014). On roasting, in vitro protein and starch digestibility of foods increased by 15–21% and 16–19%, respectively. Roasting also improved in vitro iron availability by 12–19%. Roasting reduces the moisture content, thereby concentrating the food value.

(Gahlawat and Sehgal, 1994). Roasting improves color, extends shelf life, enhances flavor and reduces the anti-nutrient factors of cereals and legumes. Moreover, roasting of grains lead to denaturation of proteins, thus improving their digestibility. Roasting can improve protein digestibility (Kavitha and Parimalavalli, 2014). Heat can kill or inactivate potentially harmful organisms including bacteria and viruses. Roasting reduces the amount of aflatoxins produced by fungi (Samarajeewa *et al.*, 1990). The goal of roasting is to improve sensory qualities and achieve inactivation of destructive enzymes which improves the storage and nutritional quality of the product (Rackis *et al.*, 1986).

2.13.5 Milling and sieving

Milling is the separation of the bran and germ from the endosperm and the reduction of the endosperm to a uniform particle size. This is done by a sequence of breaking, grinding and separating operation. Milling is the process by which cereal grains are ground into flour (Bassey *et al.*, 2013). It is obvious that over milling or very high refining must be avoided, since it removes the aleuronic layers and germ rich in protein, vitamins and minerals (Viraktamath *et al.*, 1971).

2.13.6 Mixing or blending

Blending is the process of combining grain (varieties or grades) to obtain grain of a particular quality or consistency (Bassey *et al.*, 2013). It is the homogenous mixing of the entire ingredient. It is the process of combining two or more ingredients together so that they lose their individual characteristics and become smooth and uniform.

2.13.7 Packaging

Packaging has been defined as a socio scientific discipline which operates in society to ensure delivery of goods to the ultimate consumer of those goods in the best condition intended for their use. The Packaging Institute International defines packaging as the enclosure of products, items or packages in a wrapped pouch, bag, box, cup, tray, can, tube, bottle or other container form to perform one or more of the following functions: containment, protection, preservation, communication, utility and performance (Robertson, 2006).

2.14 Antinutritional factors

Antinutrients in foods are responsible for deleterious effects related to the absorption of nutrients and micronutrients. Thus, manipulation of processing conditions or removal of certain unwanted components of food may be required. There is a concern about high intake of foods that are rich in antinutrient due to their increased burden on the bodies tolerance to potentially harmful compounds (Thompson, 1993). For example, phytic acid, lectins, phenolic compounds and tannins, saponins, enzyme inhibitors, cyanogenic glycosides and glucosinolates have shown to reduce the availability of certain nutrients and impair growth. Some compounds such as phytoestrogens and lignans have also been linked to induction of infertility in humans. Therefore, it is prudent to examine all aspects related to food antinutrients, including their potential health benefits and method of analyses. Many traditional methods of food preparation such as fermentation, cooking, and malting increase the nutritive quality of plant foods through reducing certain anti nutrients such as phytic acid, polyphenols, and oxalic acid (Hotz and Gibson, 2007). Such processing methods are widely used in societies where cereals and legumes form a major part of the diet (Chavan *et al.*, 1989).

2.15 Sensory evaluation

Quality is the ultimate criterion of the desirability of any food product to the consumer. Overall quality depends on quantity, nutritional and other hidden attributes, and sensory quality. Hedonic rating relates to pleasurable or unpleasurable experiences. The hedonic rating test is used to measure the consumer acceptability of food products. Semi trained panels in smaller number are used to screen a number of products for selecting a few for consumer preference studies. The samples are served to the panelist at one session. The panelist is asked to rate the acceptability of the products on the scale, usually of 9 points, ranging from 1 point usually given for "like extremely" to 9 points given for "dislike extremely". The scores received by each samples are then averaged and compared with the average score received by other sample in the series (Ranganna, 1986).

Part III

Materials and method

3.1 Material

3.1.1 Wheat

Wheat was collected from Dharan market. It is locally known as 'gahu'. The scientific name of wheat is '*Triticum compactum*'.

3.1.2 Buckwheat

Buckwheat was collected from Dharan market. It is locally known as '*phapar*'. The scientific name of buckwheat is '*Fagopyrum esculentum*'.

3.1.3 Green gram

Green gram was collected from Dharan market. It is locally known as *'mung daal'*. The scientific name of green gram is *'Vigna radiata'*.

3.2 Chemicals, reagents and equipment used

3.2.1 Chemicals and reagents

Analytical grade reagents from different suppliers were used for all experiments.

H₂SO₄ (97-99% assay, Qualigens), NaOH (97% assay, Merck), HCl (35% assay, Merck), boric acid (99.5% assay (powder), Merck), Petroleum ether (assay > 99%, Thermo Fisher Scientific), Indicators (Phenolpthalein, Methyl red, Bromocresol green), Potassium permanganate (99% assay, SDFCL), Methanol (assay >99%, Emplura), Ethanol (assay >99%, Fischer Scientific), Oxalic acid (assay>99%, Merck), Ammonium oxalate (assay>99%, Qualigens).

3.2.2 Glasswares and equipments

Standarized and calibrated glassware and equipment were used.

- Hot air oven
- Spectrophotometer (UV-VIS single Beam Spectrophotometer MODEL NO-291
- Soxhlet apparatus (Y.P. scientific glass work)
- Centrifuge (Y.P. scientific glass work)

- Electonic balance (AMPUT Electronic Balance Model No-457B, Sensitivity ± 0.01)
- Thermometer
- Heating mantle (Burner)
- Incubator (Y.P. scientific glass work)
- Water bath (Intake serological water bath)
- Chromatography chamber
- Atomiser

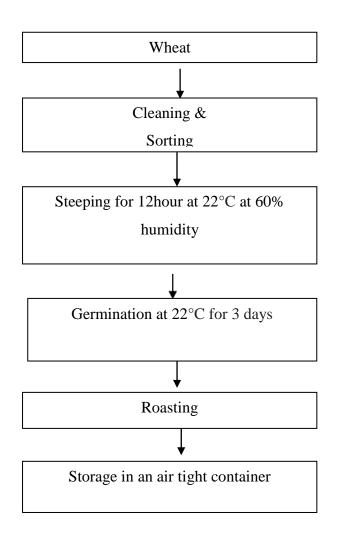
Standardized glassware such as petri dish, burette, pipette, test tubes, volumetric flask, beaker, funnel, conical flask, measuring cylinder, crucible, etc. was used.

3.3 Method

3.3.1 Processing of raw materials

3.3.1.1 Wheat

Wheat was sorted, cleaned and soaked in water for 12 hours at 22°C and 60% humidity and then drained. It was spread on a wetted muslin cloth and covered by a wetted muslin cloth. It was kept for germination at 22°C.Water was sprinkled on the layer at 3 to 4-hour interval. The grain was germinated for 3 days. It was then dried in cabinet drier at 55°C for 3 hours and 70°C for 1 hour until moisture sufficiently reduced to about 5%. The germinated part was removed, then roasted and milled into grits and packed in air tight plastic bags (Tehseen *et al.*, 2014). Fig 3.1 shows the flow diagram for the processing of wheat.



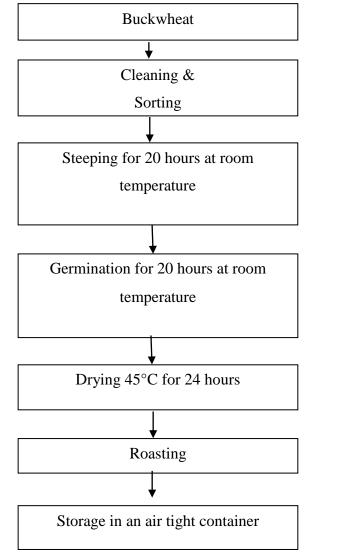
Source: Tehseen et al. (2014)

Fig 3.1 Flowchart for the processing of wheat grain

3.3.1.2 Buckwheat

Seeds were screened to remove impurities such as stones, strings, weed seeds, etc. The grains were then washed with water. The cleaned seeds were then immersed in water in a bowl. The light material present in the sample was skimmed off. Steeping was done for 20 hours in room temperature which brought moisture content to the required level of 45-50%. Steeped water was drained off. The steep grain was first collected in a muslin cloth and swirled in order to drain excess water. The grains were spread over the germination paper and covered with the germination paper and kept for germination in open environment at temperature 26-28°C and relative humidity 85-86% in open environment. During germination of the grains were moisturized by sprinkling water at 6-hour interval

and mixed gently to equalize temperature and to aerate the mass. The buckwheat germinated in was taken in cabinet dryer to stop further germination. Drying was carried out at 45°C for 24 hours. The grains were then rubbed and sprouts were removed with the help of screen. The malted grains were then milled and sieved to obtain grits. The grits were then stored in air tight container (Subedi, 2018a). Fig 3.2 shows the flowchart for processing of buckwheat grain.

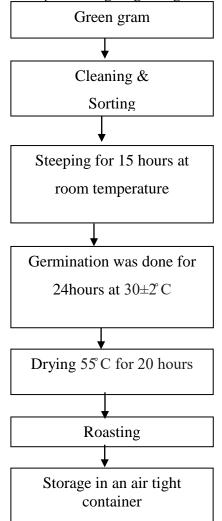


Source : Subedi (2018a)

Fig 3.2: Flowchart for the processing of buckwheat grain

3.3.1.3 Green gram

The grains were screened to remove impurities and then the cleaned grains were washed and soaked in excess water. The grains almost double in volume hence the trays of bigger sizes were taken. Soaking was done for 10-15 hour. At the end of soaking period, the soaked grains were spread for germination. Water was sprinkled during germination to keep the grains moist. The germination was done for 24 hours at $30\pm2^{\circ}$ C. The sprouts were then dried to stop the germination in hot air oven at 55° C for 20 hours. The dried grains were dehusked and the husk and cotyledons were removed. To improve the taste and flavor it was roasted under mild heat and then dry milled (Yasmeen, 2006). Sieving was done before standardizing the size of grits. The sample was then packed in airtight plastic container. The container was stored at room temperature until further use. After that weighing, blending and cooking process was done as required (Rana *et al.*, 2015). Fig 3.3 shows the flow diagram for the processing of green gram.



Source: Rana et al. (2015)

Fig 3.3: Flowchart for the processing of green gram grain

3.4 Formulation

For the formulation of multigrain porridge, the amounts of ingredients were calculated on dry weight basis. Legume was taken as the source of protein and the cereals as the staple source. The pulses and cereals were cleaned, soaked, germinated, dried and roasted well (separately) and then stored in an air tight container. The stored roasted grains were then taken in required amount as per the formulations. Then the grains were mixed and milled together. Grits were sieved and then stored in an airtight container separately and used for further work.

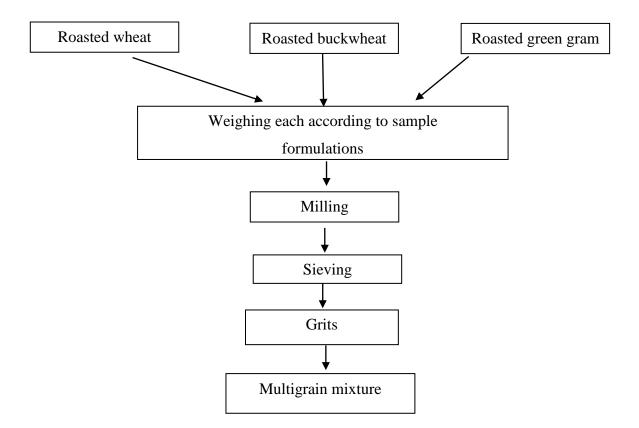


Fig.3.4 Process Flow diagram for the preparation of multigrain porridge

3.5 Research design

Using the Design expert version 10, thirteen formulations were made which are then coded alphabetically as given in the Table 3.1.

Ingredients	Wheat (%)	Buckwheat (%)	Green gram (%)
А	0	100	0
В	33	67	0
С	67	33	0
D	0	33	67
E	16.67	66.66	16.67
F	100	0	0
G	0	67	33
Н	0	0	100
Ι	16.67	16.67	66.66
J	33	0	67
K	67	0	33
L	33.33	33.33	33.33
М	66.66	16.67	16.67

 Table 3.1 Sample code for different formulations

3.6 Sensory evaluation

Sensory evaluation was performed by 9-point hedonic scoring test (9 = like extremely, 1=dislike extremely) for color, flavor, taste, texture and overall acceptance. Hedonic rating relates to pleasurable or unpleasurable experiences. The hedonic rating test is used to measure the consumer acceptability of food products. This method can be used with untrained panelist as well as with experienced ones (Ranganna, 1986). The evaluation was carried out by 14 panelists comprising of teachers and students of Central Campus of Technology, Dharan. Sensory evaluation was carried out in individual booth with adequate light and free from obnoxious odors. Each panelist was provided with samples coded random numbers and evaluation card (Appendix A). They were provided with potable water for rinsing between the samples. Verbal communication among the panelist was prohibited. They were asked to evaluate the samples individually using score card.

3.7 Analytical methods

3.7.1 Proximate analysis

3.7.1.1 Determination of moisture content

The moisture contents of the raw samples and the final product was determined by hot air oven method as described in Ranganna (1986) and that of the germinated samples by digital (Wile-55) moisture meter. The results were expressed in terms of percentage.

3.7.1.2 Determination of protein

The protein content of the raw, germinated and the final product was determined as in Ranganna (1986). The calculated data were presented per 100 g on dry basis.

3.7.1.3 Determination of crude fat

The fat content of the samples was determined as described in Ranganna (1986). The calculated data were presented as gm per 100 gm on dry basis.

3.7.1.4 Determination of total ash

The total ash of the samples was determined by incinerating the samples in muffle furnace at a temperature not exceeding 525° C for 5-6 hour, as described in Ranganna (1986). The calculated data were presented as gm per 100 gm on dry basis.

3.7.1.5 Determination of crude fiber

The crude fiber of the samples was determined as described in Ranganna (1986). The calculated data were presented as gm per 100 gm on dry basis.

3.7.1.6 Determination of carbohydrate

Total carbohydrate content of the samples was determined by the difference method.

Carbohydrate (%) = 100- [sum of protein, total ash, fiber and fat]

3.7.1.7 Determination of energy value of food

One of the methods specified by FDA was employed. This uses the general factors of 4,4 and 9 calories per gm of protein, total carbohydrate, and total fat, respectively, to calculate the calorie content of food (Bassey *et al.*, 2013)

3.7.2 Ultimate analysis

3.7.2.1 Determination of calcium

Calcium content of the sample was determined by volumetric method as described by Ranganna (1986).

3.7.2.2 Determination of iron

Iron content of the sample was determined colorimetrically as described by Ranganna (1986).

3.8 Physical analysis of raw sample

3.8.1 Bulk density

The bulk density of the raw samples was determined using a filling hopper and measuring cup as described by (Clementson *et al.*, 2010).

3.8.2 1000 kernel weight

The 1000 kernel weight of grains was calculated by counting and weighing the grains as it is an important parameter for the evaluation of variety breeding. It also helps to evaluate the grain yield and milling quality of grain (Wu *et al.*, 2018).

3.9 Physical analysis of formulated samples

3.9.1 Bulk density

Bulk density was determined by filling the sample gently in a container of known volume and weighed. The ratio between the weight and volume was calculated as bulk density (Kshirsagar, 1992).

3.9.2 Cooking time

It is the time taken by a predetermined weight of grains to cook completely. 2 gm of grains was directly dipped into boiling bath of 100 ml of distilled water. About 10 grains were taken in every 5 min and pressed between two glass plates until at least 90% of grains had no longer centers i.e. the grains were able to be smashed easily, which was considered as optimum cooking time (Vyas, 2013).

3.9.3 Swelling index

About 1gm of the sample was weighed and transferred into a clean dry test tube and then weighed. The sample was then dispersed in 50 ml of distilled water using a magnetic stirrer. The resulting slurry was heated at desired temperatures; 40°C, for 30 min in a water bath. The mixture was cooled to room temperature (25°C) and then centrifuged at 2,200 rpm for 15 min. The residue obtained was reweighed and the swelling index calculated as the ratio of the difference in weight of original flour and the residue multiplied by 100 (Ikpeme-Emmanuel *et al.*, 2009).

3.10 Data Analysis

Data were statistically processed by Gene stat version for analysis of variance (ANOVA). The scores given by the panelist were analyzed by a one-way ANOVA and the differences were considered significant at (p<0.05). LSD and interaction effects were obtained to determine whether the samples were significantly different from each other or not and also to determine which one was superior among them. Means of the data were compared by using LSD Fisher's protected method at 5% level of significance, by for analysis of variance (ANOVA).

3.11 Reconstitution of multigrain porridge mix

On the basis of preliminary study, 35 g of porridge mix gave the best results in 100 ml of milk and 5 g of sugar (A. Hussain and Kaul, 2019).

3.12 Cost evaluation

From the sensory analysis the best product was found and its cost of production was calculated per 100 gm of the product.

Part IV

Results and discussion

Multigrain porridge, a weaning food was prepared by the use of germinated wheat, buckwheat and green gram. This study focused on the formulation of multigrain porridge for the weaning infants from the cheap and locally available cereals as a staple source, legumes as a protein source followed by the household traditional method of pretreatment and germination. Germination of the cereals and legumes was done in order to improve the nutritional quality and the bioavailability. Thirteen samples were formulated and then from the physical analysis of product five samples were selected. The five samples prepared were subjected to sensory analysis on 9-point Hedonic rating and best sample was subjected to further proximate analysis.

4.1 Physical parameter of raw samples

Table 4.1 shows the various physical parameters i.e. 1000 kernel weight, bulk density of wheat, buckwheat and green gram.

 Table 4.1 The physical parameter of raw samples

Parameter	Wheat	Buckwheat	Green gram
1000 kernel weight(gm)	38.6 (0.15)	23.69 (0.19)	30.3 (0.07)
Bulk density (Kg/hL)	74.6 (0.86)	81.23 (1.11)	79.30 (0.05)

(Each value is average of three replicates and the figures in parentheses are the standard deviations.)

The 1000 kernel weight and bulk density of the wheat is similar to the result reported in (Tabatabaeefar, 2003). Similarly, the 1000 kernel weight and bulk density of buckwheat is similar to the result reported in (Unal *et al.*, 2017). Also the 1000 kernel weight and bulk density of green gram is similar to the result reported in Tabatabaeefar (2003) and Nimkar and Chattopadhyay (2002).

4.2 Proximate analysis of raw samples

Proximate analysis gives inexpensive yet very important information, particularly from the nutritional and biochemical points of views. The results normally expressed in percentage and because of the fairly general nature of test employed for the determination, the term crude is usually used as a modifier; for instant, crude protein, crude fat and crude fiber, etc.

Therefore, proximate constituent represents only a category of compounds present in biological material (Acharya and Karki, 2008).

Parameter	Wheat	Buckwheat	Green gram
Moisture (%)	11.6 (0.26)	13.8(0.041)	10(0.042)
Crude protein (%),db	11.48(0.09)	12.5(0.306)	21.29(0.004)
Crude fat (%), db	1.69(0.17)	3.01 (0.030)	1.44(0.029)
Ash content (%),db	2.9(0.017)	3.32(0.009)	3.77(0.096)
Crude fiber (%), db	1.73 (0.011)	2.33(0.022)	3.76(0.008)
Carbohydrate (%),db	82.2(0.32)	78.81(0.318)	69.74(0.026)
Iron (mg/100gm), db	3.8(0.031)	2.5(0.033)	5.01(0.191)
Calcium(mg/100gm),db	38(0.060)	52.3(0.011)	71.34(0.132)
Vitamin C	ND	ND	ND

Table 4.2 The proximate analysis of raw samples (% dry basis)

(Each value is average of three replicates and the figures in parentheses are the standard deviations.)

Table 4.2 shows the proximate and ultimate composition of the wheat, buckwheat and green gram on the dry basis. The proximate and ultimate composition of wheat is similar to that reported in Parimalavalli (2014) and L Alvarez-Jubete *et al.* (2009) respectively. The proximate and ultimate composition of the buckwheat is similar to that reported Subedi (2018a) and L Alvarez-Jubete *et al.* (2009) respectively. Also the proximate and ultimate composition of the green gram is similar to that reported in Paul *et al.* (2011). The vitamin C content of the grains is similar to the data provided by DFTQC (2012).

4.3 Proximate analysis of germinated samples

Germination affects the nutritional composition of cereal and legume flours. Germination increased moisture and protein content in cereal and pulse flours. Germination significantly affect ash, fat, fiber, carbohydrate and energy content (Parimalavalli, 2014). Table 4.3 shows the proximate analysis of germinated samples.

_		_	
Parameter	Wheat	Buckwheat	Green gram
Moisture (%)	7.2(0.1)	7.2(0.02)	3.6(0.013)
Crude protein (%),db	13.03(0.043)	14.03(0.005)	23.03(0.034)
Crude fat (%),db	1.43(0.017)	2.85(0.122)	1.24(0.021)
Ash content (%),db	2.35(0.082)	3.08(0.028)	3.28 (0.073)
Crude fiber (%),db	1.32(0.033)	1.98(0.074)	3.4(0.015)
Carbohydrate (%),db	81.87(0.014)	78.06(0.019)	69.05 (0.044)
Iron (mg/100gm),db	1.6(0.023)	1.0(0.011)	3.02(0.66)
Calcium(mg/100gm),db	47(0.15)	65.5(0.24)	80.9(0.33)
Vitamin C	4.07(0.01)	4.44(0.026)	10.2(0.039)
(mg/100gm),db			

 Table 4.3 The proximate analysis of germinated samples

The moisture content of wheat, buckwheat and green gram decreased after germination and drying process. Low moisture content of food samples is a desirable phenomenon, since the microbial activity is reduced (Oyenuga, 1968) .Low moisture content in food samples increased the storage periods of the food products (Alozie *et al.*, 2009); while high moisture content in foods encourage microbial growth; hence, food spoilage (Temple *et al.*, 1996).

The protein content of germinated wheat, buckwheat and green gram increased in comparison to the raw samples. This observation agreed with other scientific findings that processing techniques such as germination improved the nutritional quality of the food products, particularly in terms of protein content (Enujiugha *et al.*, 2003).

The fat content of germinated wheat, buckwheat and green gram decreased as compared to that of the raw samples. Similar finding has been found on the work done by Parimalavalli (2014). This decline may be because fat was used as the major source of carbon for seed growth. Also, fatty acids are oxidized to carbon dioxide and water to generate energy for germination (Bau *et al.*, 1997).

The ash content of the raw samples is greater than that of germinated sample. The decrease in ash content represents loss in minerals due to rootlet and washing in water to reduce the sour smell during the period of germination (Tatsadjieu *et al.*, 2004). The

reduction in ash content might be due to the leaching out of both macro and micro elements into the soaking water (D'souza, 2013).

The crude fiber content of the germinated wheat, buckwheat and green gram decreased in comparison to that of the raw samples. Similar finding has been found on the work done by Parimalavalli (2014).

The carbohydrate content of the raw samples is greater than that of germinated samples. The carbohydrate content of germinated sample was lower than those of raw samples. This observation could be due to the utilization of carbohydrate for biochemical activities of the germinating seeds (Wang *et al.*, 1997). Vidal-Valverde *et al.* (2003) explained that during germination, carbohydrate was used as source of energy for embryonic growth which could explain the changes of carbohydrate content after germination.

The iron content of the germinated samples is lower in comparison to the iron content of the raw samples. The reduction after soaking may be attributed to leaching of iron into the soaking medium (Saharan *et al.*, 2001). The calcium content of the grains increased after germination in comparison to the raw samples. Similar finding has been found on the work done by (Oloyo, 2004). The vitamin C content of the grains increased after germination. Since this vitamin was not found in raw grains, its production may be taken as a representative parameter in the metabolic germination process to improve the nutritional value of the grain (Yang, 2001).

4.4 Physical properties of the formulated samples

4.4.1 Bulk density

The bulk density of the formulated thirteen samples is shown in the Table 4.4.

Sample	Bulk density (Kg/hL)
А	65 (0.05)
В	65 (0.05)
С	64 (0.03)
D	66 (0.04)
Е	64 (0.05)
F	66 (0.01)
G	71 (0.03)
Н	73 (0.07)
Ι	63 (0.12)
J	69 (0.11)
Κ	67 (0.06)
L	64 (0.02)
М	62 (0.01)

Table 4.4 The bulk density of the formulated samples

From the above table five samples containing low bulk density were chosen for the sensory analysis. Low bulk density is desirable in preparation of infant and weaning foods (Nicole *et al.*, 2010). A less bulky food contains a higher nutrient content since the volume of food is low (Imtiaz *et al.*, 2011). Bulk density is an important functional property of flour and is significant for the preparation of weaning food. Germination has been reported to be a useful method for the preparation of low bulk weaning foods (I. Hussain and Uddin, 2012).

4.4.2 Swelling index

The swelling index of the formulated thirteen samples are shown in the Table 4.5

Sample	Swelling index
A	2.99 (0.11)
В	2.46 (0.12)
С	1.03 (0.10)
D	3.06 (0.06)
Е	2.09 (0.05)
F	3.29 (0.13)
G	2.52 (0.22)
Н	3.88 (0.16)
Ι	2.32 (0.07)
J	3.48 (0.19)
Κ	3.09 (0.07)
L	2.39 (0.17)
М	1.36 (0.03)

 Table 4.5 Swelling index of the formulated sample

From the above data, five samples having low swelling index were chosen for sensory analysis. The swelling index is an important parameter which ultimately determines the sample consistency (that is solid, semi-solid, or liquid). Flours with high swelling index values hold large amounts of water during their preparation into gruels and thus become voluminous with a low energy and nutrient density. Sample with the least swelling index would value provide more nutrient-density food for infant a an (Ikpeme-Emmanuel et al., 2009).

4.4.3 Cooking time

2 gm of grains was directly dipped into boiling bath of 100 ml of distilled water. About 10 grains were taken in every 5 min and pressed between two glass plates until at least 90% of grains had no longer centers i.e. the grains were able to be smashed easily, which was considered as optimum cooking time. Table 4.6 shows the cooking time of the formulated thirteen samples.

A 10.36 (0.13) B 11.50 (0.10) C 13.40 (0.24) D 12.30 (0.34) E 14.45 (0.47) F 13.30 (0.44) G 11.50 (0.26) H 10.35 (0.17) I 15.05 (0.42) J 13.15 (0.62) K 13.15 (0.53) L 15.30 (0.54) M 14.40 (0.18)	Sample	Cooking time (minutes)
C13.40 (0.24)D12.30 (0.34)E14.45 (0.47)F13.30 (0.44)G11.50 (0.26)H10.35 (0.17)I15.05 (0.42)J13.15 (0.62)K13.15 (0.53)L15.30 (0.54)	A	10.36 (0.13)
D 12.30 (0.34) E 14.45 (0.47) F 13.30 (0.44) G 11.50 (0.26) H 10.35 (0.17) I 15.05 (0.42) J 13.15 (0.62) K 13.15 (0.53) L 15.30 (0.54)	В	11.50 (0.10)
E14.45 (0.47)F13.30 (0.44)G11.50 (0.26)H10.35 (0.17)I15.05 (0.42)J13.15 (0.62)K13.15 (0.53)L15.30 (0.54)	С	13.40 (0.24)
F 13.30 (0.44) G 11.50 (0.26) H 10.35 (0.17) I 15.05 (0.42) J 13.15 (0.62) K 13.15 (0.53) L 15.30 (0.54)	D	12.30 (0.34)
G 11.50 (0.26) H 10.35 (0.17) I 15.05 (0.42) J 13.15 (0.62) K 13.15 (0.53) L 15.30 (0.54)	Е	14.45 (0.47)
H 10.35 (0.17) I 15.05 (0.42) J 13.15 (0.62) K 13.15 (0.53) L 15.30 (0.54)	F	13.30 (0.44)
I 15.05 (0.42) J 13.15 (0.62) K 13.15 (0.53) L 15.30 (0.54)	G	11.50 (0.26)
J 13.15 (0.62) K 13.15 (0.53) L 15.30 (0.54)	Н	10.35 (0.17)
K13.15 (0.53)L15.30 (0.54)	Ι	15.05 (0.42)
L 15.30 (0.54)	J	13.15 (0.62)
	K	13.15 (0.53)
M 14.40 (0.18)	L	15.30 (0.54)
	М	14.40 (0.18)

Table 4.6 Cooking time of the formulated sample

Cooking time did not show any correlation with protein, fat, ash or carbohydrate content. Cooking time was independent of all nutritional characteristics (Nielsen *et al.*, 1993).

4.5 Cooking of the porridge

The prepared five formulations selected after physical analysis i.e. samples having low bulk density and low swelling index were cooked as described by (A. Hussain and Kaul, 2019). From the physical analysis formulations C, E, I, L and M had low bulk density and low swelling index. So these five formulations i.e. C, E, I, L and M were cooked for sensory analysis. 35 gm of the different formulations of multigrain grits were taken and cooked with 100 ml milk and 5 gm sugar. The cooked products were then taken for sensory analysis.

4.6 Sensory evaluation of different formulations of multigrain porridge

So the five formulations i.e. C, E, I, L and M which was cooked was provided to 14 panelists i.e. teachers and students of Central Campus of Technology. The panelists evaluated for various parameters of the product namely color, flavor, taste, texture, mouth feel and overall acceptability. The panelists were requested to provide scores in the score

sheet as per their perception. Data were analyzed statistically and best product was found out.

4.6.1 Color

The average sensory score for color was 5.0, 5.28, 6.0, 7.36 and 6.50 for C, E, I, L and M respectively. The analysis of variance showed that there was no significant difference (p>0.05) between the formulations C and E. However, they are significantly different with sample I, L and M. Sample L was found significantly different with all the samples in terms of color and was found to score higher (7.36) among the samples. Fig 4.1 shows the histogram for mean sensory scores of color attribute.

In comparison to the other formulations, the formulation having equal amount of all the ingredients had an appealing color. This may be due to over mixing of any grains decreased the appealing color of the multigrain porridge. When all the grains were mixed in equal amount, it was eye pleasing. The color is very important parameter for selecting any food, as man eats with his eyes.

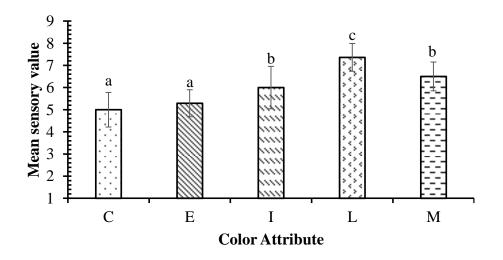


Fig.4.1 Mean sensory score of the samples for color attribute

4.6.2 Flavor

The average sensory score for flavor was 5.36, 5.36, 5.50, 7.29 and 6.64 for C, E, I, L and M respectively. The analysis of variance showed that there was no significant difference (p>0.05) between the formulations C, E and I. However, they are significantly different with sample L and M. Sample Land M were not found significantly different with each

other in terms of flavor. Sample L was found to score higher (7.36) among the samples. Fig 4.2 shows the histogram for mean sensory scores of flavor attribute.

In comparison to the other formulations, the formulation having equal amount of all the ingredients had an acceptable flavor. This may be because none of the ingredient could fade off the flavor of other ingredients. All the ingredients could contribute equally to the flavor of the porridge. The flavor was similar to that of commercial weaning food.

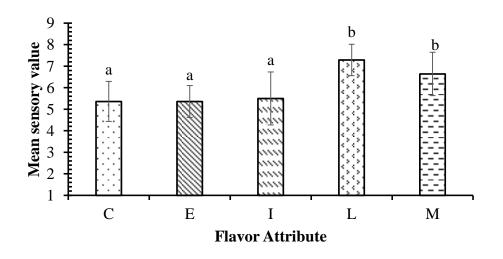


Fig.4.2 Mean sensory score of the samples for flavor attribute

4.6.3 Texture

The average sensory score for texture was 5.64, 5.29, 5.57, 7.29 and 6.86 for C, E, I, L and M respectively. The analysis of variance showed that there was no significant difference (p>0.05) between the formulations C, E and I. However, they are significantly different with sample L and M. Sample L and M were not found significantly different with each other in terms of texture. Sample L was found to score higher (7.29) among the samples. Fig 4.3 shows the histogram for mean sensory scores of texture attribute.

Like in previous case, in comparison to the other formulations, the formulation having equal amount of all the ingredients had a better texture. The formulation L had a good texture. It may be due to the softness of this formulation in comparison to the other. The sample may be less husky in comparison to the other.

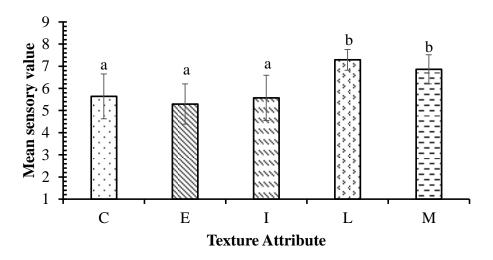


Fig.4.3 Mean sensory score of the samples for texture attribute

4.6.4 Taste

The mean sensory score for taste of the five samples C, E, I, L and M were found to be 5.64, 5.07, 5.57, 7.429 and 7.143 respectively. The taste of the sample L was reported to be higher by sensory panelist compared to other 4 samples. The statistical analysis at 5% level of significance showed that the samples C, E, I was not significantly different to each other in terms of taste. But they were significantly different with sample L and M. Similarly, sample L and M were also not significantly different with each other in terms of taste. Sample L was reported to be superior significantly from other samples. Fig. 4.4 shows the histogram for mean sensory scores of taste attribute.

Taste is the primary factor which determines the acceptability of any product, which has the highest impact as far as the market success of the product, is concerned. In this study, the formulation L has the acceptable taste in comparison to the other formulation. This may be because all the grains could contribute equally to the taste of the porridge. The other formulations had higher amount of any one ingredient. The taste of formulation L was pleasing and acceptable to the sensory panelist.

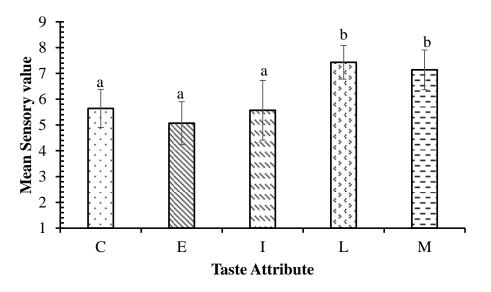


Fig.4.4 Mean sensory score of the samples for taste attribute

4.6.5 Overall acceptance

The overall acceptability of the multigrain porridge can be seen in the Fig.4.5. The mean sensory score value for the overall acceptability of the samples C, E, I, L and M were found to be 5.71, 5.00, 6.00, 7.43 and 7.07 respectively. Statistical analysis showed that the mean score value for overall acceptability of sample C and I was not found to be significantly different (p<0.05) to each other. Similarly, sample L and M was also not found to be significantly different (p<0.05) to each other. Sample E was found to be significantly different to all the remaining four samples at 5% level of significance. Sample L has got the highest value of mean sensory score.

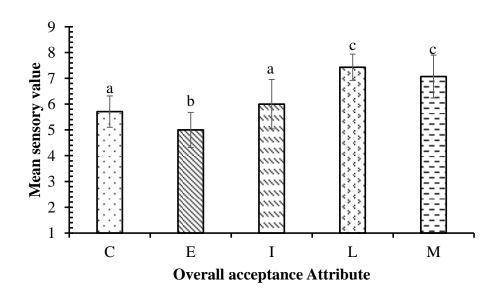


Fig.4.5 Mean sensory score of the sample for overall acceptance attribute

The sample L had the highest overall acceptability. This may be due to the better eye appealing color, better taste, flavor and texture of the formulation. The consistency and overall acceptance was highest in the sample containing equal amount of all the ingredients and having a combination of wheat, buckwheat and green gram in the ratio 1:1:1.

4.7 Analysis of the final product

Analysis of the sample L which was selected best from sensory analysis was carried out. The result is tabulated in Table 4.7.

Parameters	Amount
Moisture (%)	4.1 (0.21)
Crude protein (%), db	28.32 (0.11)
Crude fat (%), db	2.3 (0.03)
Ash content (%), db	2.5 (0.35)
Crude fiber (%), db	3.15 (0.42)
Carbohydrate (%), db	63.73 (0.25)
Iron (mg/100gm), db	3.5 (0.71)
Calcium (mg/100gm), db	82.4 (0.67)
Vitamin C (mg/100gm), db	5.2 (0.53)
Energy (Kcal/100gm)	388.9 (0.52)

Table 4.7 Analysis of the multigrain porridge

(Each value is average of three replicates and the figures in parentheses are the standard deviations.)

Similar nutritional value was found by Nutrition Collaborative Research Program during the market analysis of complementary foods in Nepal where the analysis of Sarbottam Pitho showed moisture 3.53%, Protein 14.72%, Fat 7.4%, Ash 1.92%, Carbohydrate 73% and Energy 400 kcal (Magnani *et al.*, 2012). Weaning food prepared from multipurpose flour had moisture 4.2%, protein18.6%, fat 8.40%, crude fiber 2.57%, carbohydrate 62.24%, ash content 3.2% (Ahmad *et al.*, 2013). According to Imtiaz *et al.* (2011), weaning food prepared from germinated wheat flour and mung seed flour had moisture of 5.15%, protein of 23.97%, fat of 1.33%, total ash of 2.87%, carbohydrate of 65.08%, crude fiber of 1.65% and energy of 377.16 Kcal.

4.8 Determination of amino acids in the final product

Amino acid	Solvent distance	Spot distance	RF value
Histidine	12	2.3	0.19
Isoleucine	12	9	0.75
Methionine	12	6.9	0.57
Arginine	12	3.5	0.29
Lysine	12	2.7	0.23
Phenylalanine	12	8.1	0.67
Valine	12	6.8	0.56
Tryptophan	12	6.4	0.53
Leucine	12	9.5	0.79
Threonine	12	4	0.33

4.8.1 Calculation of RF value of the known and unknown amino acids Table 4.8 RF value of the known amino acid

The best product i.e. sample L which was selected from the sensory analysis was then analyzed for the presence of essential amino acid. Thus, supposing the essential amino acids present in the sample as Q, R, S, T, U, V, W, X, Y and Z and then calculating the RF values of those amino acids.

Table 4.9 shows the RF values of different amino acid present in the sample.

Amino acid	Solvent distance	Spot distance	RF value
Q	12	2.2	0.18
R	12	8.8	0.73
S	12	6.8	0.56
Т	12	3.6	0.30
U	12	2.7	0.23
V	12	8.2	0.68
W	12	6.9	0.57
Х	12	6.3	0.53
Y	12	9.6	0.80
Z	12	4	0.33

Table 4.9 RF value of the unknown amino acid

Thus, comparing the RF values of the unknown amino acids with that of known amino acids, we found that the RF values of the unknown amino acids are similar to that of the known amino acids. From the Table 4.8 and 4.9, the RF values of unknown amino acid Q is same as that of histidine, R is same as isoleucine, S is same as methionine, T is same as arginine, U is same as lysine, V is same as phenylalanine, W is same as valine, X is same as tryptophan, Y is same as leucine and Z is same as threonine. Hence, we can conclude that all the essential amino acids are present in the best sample i.e. sample L and is a product having complete protein.

4.9 Cost evaluation

From the statistical analysis the best product was found as sample L and its cost of production was calculated per 100 g of the product. The estimated cost calculation is shown in Table 4.10.

Raw materials	Price/Kg (Rs.)	Amount of materials	Price (Rs.)
Wheat	45	3 kg	135
Buckwheat	70	3 kg	210
Green gram	100	3 kg	300
Price/Kg			215
Price/100 g			21.5

Table 4.10 Cost evaluation of the best sample L

The cost of the product was calculated to be NRs. 21.5/100 g excluding labor cost, packaging cost and taxes.

Part V

Conclusion and Recommendations

5.1 Conclusion

From the above result and discussions, it can be concluded that:

- a. Multigrain porridge was prepared from the wheat, buckwheat and green gram varying the proportion of each in different formulations.
- b. The sensory evaluation showed that the formulation L i.e. formulation having wheat, buckwheat and green gram in the ratio 1:1:1 was the best among the five formulations in terms of color, flavor, taste, texture and overall acceptability.
- c. The proximate analysis of the formulation L showed that it is a good source of protein, minerals and energy content.
- d. The proximate analysis showed that the protein, calcium and vitamin C were higher in germinated grains in comparison to the raw grains.
- e. The amino acid profile analysis of the best product showed that it contains all the essential amino acid and is a food having complete protein.

5.2 Work recommendation

This study can be further continued with the following recommendations.

- a. The formulation having wheat, buckwheat and green gram in the ratio 1:1:1 can be used and scaled up for industrial purpose to provide low cost weaning food to everyone.
- b. Invivo evaluation using albino rats could be done.

Part VI

Summary

Multigrain porridge is a weaning food that is given to weaned infants. Weaning food is generally made from the cereals and legumes in the ratio of 2:1. The cereals and legumes were germinated. Maize, buckwheat and green gram were germinated for 36, 36 and 24 hours respectively.

Thirteen different products were made from the germinated cereals and legumes varying the proportion of each grain. The raw materials were processed and the products were prepared in the laboratory. Out of the thirteen products, five products were chosen from the analysis of physical parameter i.e. bulk density and the swelling index. The sensory evaluation of the selected five samples was performed by 14 panelists who were the teachers and students from Central Campus of Technology. On the basis of sensory evaluation, the product D containing33 gm wheat, 33 gm buckwheat and 33 gm green gram was taken for further chemical analysis of the product. The protein, fat, carbohydrate, crude fiber and total ash of the product were found to be 28.32%, 2.3%, 63.73%, 3.15% and 2.5% respectively. The diet can supply 388.90 kcal/ 100 gm. The iron and calcium content of the product were found to be 3.5 mg/100 gm and 82.4 mg/100 gm.

The final product was also analyzed for its essential amino acid profile. The known standard solutions of essential amino acids were prepared separately and then the paper chromatography was done for comparing the RF values of the known standard amino acids and unknown amino acids in the product. When comparing the RF values, the RF values of known and unknown amino acids were similar. So, we concluded that the product contains complete protein.

This study where multigrain porridge has been prepared from locally available food which contains important nutrients required for weaning children could be effective in terms of digestibility, bioavailability and physiological function. If further researched the production of weaning food using different locally available nutritious food could be possible in Nepal in low cost.

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Appendices

Appendix A

A 1. Sensory analysis score card

Sensory analysis of weaning food (multigrain porridge)

Name of the panelist:Date:

Name of the product: Weaning food for children (multigrain porridge)

Type of product: Weaning food

Dear panelist, you are given five samples of multigrain porridge, please conduct the sensory analysis based on the following parameter using the table given;

Sample Code	Color	Flavor	Texture	Taste	Overall acceptance
С					
Е					
Ι					
L					
М					

Perceptions	Point
Like extremely	9
Like very much	8
Like moderately	7
Like slightly	6
Neither like nor dislike	5
Dislike slightly	4
Dislike moderately	3
Dislike very much	2
Dislike extremely	1
Comments (if any)	

Appendix B

1. Sensory evaluation of product

Panelist1311.14290.85711.820.064Residual5224.42860.4698	Source of variation	d.f.	S.S.	m.s.	v.r.	F pr.
Residual 52 24.4286 0.4698	Formulations	4	50.3714	12.5929	26.81	<.001
	Panelist	13	11.1429	0.8571	1.82	0.064
	Residual	52	24.4286	0.4698		
Total 69 85.9429	Total	69	85.9429			

Table B 1.1 One-way ANOVA for color

Since p<0.05, there is a significant difference between the samples so LSD testing is necessary.

Table B 1.2 LSD for color

Sample	Mean	Column 1	Lsd
С	5.000	А	
Е	5.286	А	
Ι	6.000	В	0.5198
L	7.357	С	
Μ	6.500	В	

Table B 1.3 One-way ANOVA for flavor

d.f.	S.S.	m.s.	v.r.	F pr.
4	43.9429	10.9857	14.55	<.001
13	10.7429	0.8264	1.09	0.384
52	39.2571	0.7549		
69	93.9429			
	4 13 52	443.94291310.74295239.2571	443.942910.98571310.74290.82645239.25710.7549	443.942910.985714.551310.74290.82641.095239.25710.7549

Since p<0.05, there is a significant difference between the samples so LSD testing is necessary.

Sample	Mean	Column 1	Lsd
С	5.357	А	
Ε	5.357	А	
Ι	5.500	А	0.659
L	7.286	В	
Μ	6.643	В	

Table B 1.4 LSD for flavor

 Table B 1.5 One-way ANOVA for texture

Source of variat	ion d.f.	S.S.	m.s.	v.r.	F pr.
Formulations	4	43.7714	10.9429	18.11	<.001
Panelist	13	14.6429	1.1264	1.86	0.057
Residual	52	31.4286	0.6044		
Total 69 89	0.8429				

Since p<0.05, there is a significant difference between the samples so LSD testing is necessary.

Mean	Column 1	Lsd
5.643	А	
5.286	А	
5.571	А	0.5896
7.286	В	
6.857	В	
	5.643 5.286 5.571 7.286	5.643 A 5.286 A 5.571 A 7.286 B

Table B 1.6 LSD for texture

Table B 1.7 One-way ANOVA for taste

Source of var	riation	d.f.	S.S.	m.s.	v.r.	F pr.
Formulations		4	61.2286	15.3071	22.89	<.001
Panelist		13	11.9429	0.9187	1.37	0.204
Residual		52	34.7714	0.6687		
Total	69	107.9429				

Since p<0.05, there is a significant difference between the samples so LSD testing is necessary.

Sample	Mean	Column 1	Lsd
С	5.643	А	
Е	5.071	А	
Ι	5.571	А	0.6202
L	7.429	В	
Μ	7.143	В	

Table B 1.8 LSD for taste

Table B 1.9 One-way ANOVA for overall acceptability

Source of variation	d.f.	S.S.	m.s.	v.r.	F pr.
Formulations	4	55.6571	13.9143	29.24	<.001
Panelist	13	10.4714	0.8055	1.69	0.090
Residual	52	24.7429	0.4758		
Total	69	90.8714			

Since p<0.05, there is a significant difference between the samples so LSD testing is necessary.

Sample	Mean	Column 1	Lsd
С	5.714	В	
Е	5.000	А	
Ι	6.000	В	0.5232
L	7.429	С	
М	7.071	С	

Table B 1.10 LSD for overall acceptance

List of Plates



P1 Protein analysis



P2 Paper chromatography



P3 Sensory analysis of the formulated sample