

**EVALUATION OF NUTRITIONAL QUALITY OF KODO MILLET
AND ITS EXPLORATION IN COOKIES AND BISCUITS**

by

Miss. Mitkal Komal Tanaji

(Reg. No. 018/268)

A Thesis submitted to the
**MAHATMA PHULE KRISHI VIDYAPEETH
RAHURI – 413 722, DIST. AHMEDNAGAR
MAHARASHTRA, INDIA**

in partial fulfillment of the requirements for the degree

of

MASTER OF SCIENCE (AGRICULTURE)

in

FOOD TECHNOLOGY



DEPARTMENT OF FOOD SCIENCE AND TECHNOLOGY

**POST GRADUATE INSTITUTE
MAHATMA PHULE KRISHI VIDYAPEETH
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APPROVED BY

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MAHARASHTRA, INDIA.**

2021

CANDIDATE'S DECLARATION

I hereby declare that this thesis or part
there of has not been submitted
by me or other person to any
other University or Institution
for a Degree or
Diploma

Place : MPKV, Rahuri

Date : / /

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CERTIFICATE

This is to certify that the thesis entitled, “**EVALUATION OF NUTRITIONAL QUALITY OF KODO MILLET AND ITS EXPLORATION IN COOKIES AND BISCUITS**” submitted to the Faculty of Agriculture, Mahatma Phule Krishi Vidyapeeth, Rahuri Dist. Ahmednagar (M.S.) in partial fulfillment of the requirement for the award of the degree of **MASTER OF SCIENCE (AGRICULTURE)** in **FOOD TECHNOLOGY**, embodies the results of a piece of *bona fide* research work carried out by **Miss. MITKAL KOMAL TANAJI**, under my guidance and supervision and that no part of the thesis has been submitted for any other degree or diploma.

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

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Place : MPKV, Rahuri

Date : / /2021

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LIST OF ABBREVIATION

Abbreviations	Description
%	: Per cent
/	: Per
”	: Inches
⁰ C	: Degree Celsius
A.A.C.C.	: American Association of Cereal Chemists
A.O.A.C.	: Association of Official Analytical Chemists
Agril.	: Agricultural
Avg.	: Average
BHA	: Butylated Hydroxyanisole
BMKF ₀	: Biscuits 50% Maida and 0% Kodo millet Flour
BMKF ₅₀	: Biscuits 50% Maida and 50% Kodo millet Flour
C.D.	: Critical Difference
cm	: Centimeter
CMKF ₀	: Cookies 100% Maida and 0% Kodo millet Flour
CMKF ₅₀	: Cookies 50% Maida and 50% Kodo millet Flour
CRD	: Completely Randomized Design
DFWG	: Defatted Wheat Germ
<i>et al.</i>	: and others
etc.	: Etcetera
Fig.	: Figure
g	: Gram
GTR	: Gas Transmission Rate
HDL	: High-density lipoprotein
Hr	: Hour
i.e.	: That is
kg	: Kilogram
L	: Length of seed (mm)
LDL	: Low-density lipoprotein
LDPE	: Low Density Polyethylene
M	: Unit mass of seed (g)
meq	: Milli equivalent
mg	: Milligram

min	:	Minutes
ml	:	Milliliter
mm	:	Milliliter
mm ²	:	Millimeter Square
N	:	Normality
NaOH	:	Sodium hydroxide
NIDDM	:	Non-insulin dependent diabetes mellitus
Nm	:	Nanometer
No.	:	Number
OD	:	Optical Density
PP	:	Polypropylene
pp	:	Page
ppm	:	parts per million
RDA	:	Recommended Dietary Allowance
RTE	:	Ready-To-Eat
S.E.	:	Standard error
SPC	:	Standard Plate Count
Sr. No.	:	Serial number
TC	:	Total Cholesterol
Tech.	:	Technology
v/v	:	Volume by volume basis
viz.	:	Namely
VLDL	:	Very-low-density lipoprotein
vol.	:	Volume
w/v	:	Weight by volume basis
WPC	:	Whey Protein Concentrate
wt.	:	Weight
WVTR	:	Water Vapor Transmission Rate

ABSTRACT

**EVALUATION OF NUTRITIONAL QUALITY OF KODO MILLET AND ITS
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by

Miss. Mitkal Komal Tanaji

A candidate for the degree of

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in

FOOD TECHNOLOGYPost Graduate Institute,
Mahatma Phule Krishi Vidyapeeth,
Rahuri-413722
2021

Research Guide : **Dr. P. M. Kotecha**
Department : **Food Science and Technology**

The present research work on evaluation of nutritional quality of kodo millet and its exploration in cookies and biscuits was undertaken to explore the utilization of neglected but highly nutrient rich kodo millet in bakery products. The flour is a rich source of protein, dietary fiber, calcium and iron was utilized in bakery products such as cookies and biscuits. The prepared samples were analyzed for nutritional composition. The changes occurred during storage of cookies and biscuits were also studied.

Chemical composition of kodo millet showed that the moisture content was 10.90 per cent, protein 8.30 per cent, crude fat 1.30 per cent, crude fiber 8.50 per cent, carbohydrates 65.90 per cent, calcium 27.00 mg/100g and iron 1.70 mg/100g. The physical properties of kodo millet were creamish white colour, bulk density 650 kg/m³, true density 1172 kg/m³, porosity 39.68% and angle of repose 21 degree. Chemical composition of maida showed that the moisture content was 13.30 per cent, protein 12.10 per cent, crude fat 0.90 per cent, crude fiber 0.30 per cent, carbohydrates 73.90 per cent, calcium 23.00 mg/100g and iron 2.70 mg/100g.

Preliminary experiments were carried out to find out optimum level of kodo millet flour with maida for the preparation of quality cookies and biscuits. The result showed that quality cookies can be prepared from 50 % maida and 50 % kodo millet flour

(CMKF₅₀) and quality biscuits from 50 % maida and 50 % kodo millet flour (BMKF₅₀). The selected treatment of cookies and biscuits were packed in LDPE and PP packaging material and stored at ambient temperature (30 ± 4 °C) for 90 days to study their storage feasibility.

Chemical composition of the fresh cookies prepared from 50 % maida and 50 % kodo millet flour (CMKF₅₀) showed moisture content was 4.11 per cent, protein 10.20 per cent, crude fat 25.70 per cent, crude fiber 4.40 per cent, carbohydrates 69.87 per cent, calcium 25.03 mg/100g and iron 2.20 mg/100g. Chemical composition of the fresh biscuits prepared from 50 % maida and 50 % kodo millet flour (BMKF₅₀) showed moisture content was 4.07 per cent, protein 10.50 per cent, crude fat 26.62 per cent, crude fiber 4.30 per cent, carbohydrates 69.62 per cent, calcium 24.88 mg/100g and iron 2.17 mg/100g. The sensory evaluation of cookies and biscuits was carried out regularly at an interval of one month for 3 month during storage. The results on overall acceptability score of cookies and biscuits are influenced by storage. The cookies results indicate that score for overall acceptability for treatment CMKF₀ (Control) decreased from 7.30 to 7.10 in LDPE and from 7.20 to 6.90 in PP as storage period get increased. For CMKF₅₀ treatment score decreases from 7.40 to 7.15 in LDPE and 7.30 to 7.10 in PP. The CMKF₅₀ treatment is the best for the overall acceptability. The biscuits results indicate that score for overall acceptability for treatment BMKF₀ (Control) decreased from 8.35 to 8.00 in LDPE and from 8.33 to 7.90 in PP as storage period increased. For BMKF₅₀ treatment score decreases from 8.48 to 8.15 in LDPE and 8.47 to 8.13 in PP. The CMKF₅₀ treatment is the best for the overall acceptability.

Storage study of cookies and biscuits showed that the cookies and biscuits prepared by incorporation of maida and kodo millet flour can be stored up to 3 month in LDPE with minimum losses in sensory, nutritional and textural characteristics than PP. The total cost of production of cookies prepared from maida and kodo millet flour (CMKF₅₀) for 1 kg was Rs. 122/-. The total cost of production of biscuits prepared from maida and kodo millet flour (BMKF₅₀) for 1 kg was Rs. 129/-.

1. INTRODUCTION

Cereals are staple foods for a large proportion of the world population. Cereal grain contribute a significant amount of energy, protein, selected micronutrients and non-nutrients in the diet of populations all over the world in both developed and developing countries. Cereal and cereal-based food products provide more than 56 per cent of the energy and 50 per cent of the protein consumed world wide. Economically important cereals in the world are maize, rice, wheat, barley, sorghum, millets, oat and rye (FAO, 1995).

Millets contain about 8 per cent protein and 4 per cent fat. They are rich source of vitamins and minerals. Millets are especially rich in calcium. The dietary carbohydrates content of millets is also relatively high. Starch is the main carbohydrate component and they contain a higher proportion of non-starchy polysaccharides (dietary fiber) also. Prolamines and glutelins form the major portion of their proteins. The fats from millets contain a higher portion of unsaturated fatty acids and supply essential fatty acids. Although, a considerable portion of nutrients is concentrated in the seed coat, the bioavailability of the nutrients present in the endosperm is higher than the seed coat nutrients. Anti nutritional factors such as phytate and polyphenols are also present in millets but they are mostly confined to the seed coat and the milled millets are generally free from the antinutritional factors. Kodo millet (*Paspalum scrobiculatum*) and Kutki (*Panicum miliare*) are minor millets have certain specialties, which, if exploited, may yield products of superior nutritional and technological characteristics than the major cereals, but their utilization is limited.

Kodo millet (*Paspalum scrobiculatum* var. *scrobiculatum*) is grown in India as an important crop, while *Paspalum scrobiculatum* var. *commersonii* is the wild variety indigenous to Africa. The kodo millet, also known as Cow grass, Rice grass, Ditch millet, Native Paspalum or Indian crown grass originate in tropical Africa and it is estimated to have been domesticated in India 3000 years ago. In southern India, it is called varaku or koovaraku. Kodo is probably corrupt form of kodara, the Hindi name of the plant. It is grown as annual. It is a minor food crop eaten in many Asian countries, primarily in India

where in some region it is extremely important. It grows wild as a perennial in west of Africa, where it is eaten as an famine food. Often it grows as a weed in rice fields.

Food for all is a great challenge to the scientists working in the area of production, processing, storage and nutrition to combat hunger and poverty. Millets are considered as ancient grains, domesticated thousands of years ago at the beginning of human civilization. Some evidences point out about the cultivation of millets about 4000 years ago (Shahidi and Chandrashekara, 2013).

The whole grain consumption has health promoting effects like prevention of insulin resistance, heart disease, diabetes, ischemic stroke, obesity, breast cancer, childhood asthma and premature death (Balasubramanian, 2013) because of this benefits, millets can be used in functional food and as a nutraceuticals. Hence they are also called as 'nutricereals'. In addition, because of their important contribution to national food security and potential health benefits, of combating various diseases (Shahidi and Chandrashekara, 2013), millet grains are now receiving increased interest from food processors, technologist and nutritionist. Once a poor man's staple, now adorns the plates of affluent and health conscious people. Kodo millet is major food source in the deccan plateau in India (Gujrat, Karnataka and part of Tamil Nadu), some regions of Maharashtra, Odisha, West Bengal, Rajasthan, Uttar Pradesh and Himalayas and consumed traditionally as health and vitality foods in from rural India (Hegde and Chandra, 2005).

The *Paspalum* genus has more than 400 species, usually an annual crop, however many cultivars root at the nodes and grow culms after the mature plant flowers and mature their influence. Some of the species are perennial in nature (De Wet *et al.*, 1983). This crop is draught tolerant and usually grown in semi arid regions without any intercultural operations. Kodo is monocot and the seeds are very small and ellipsoidal, being approximately 1.5 mm in width and 2 mm in length; they vary in colour being light brown to dark grey. Kodo millet has a shallow root system which may be ideal for intercropping. The grain is enclosed in hard, corneous, persistent husk (FAO, 1995). The kodo millet flour has a gelatinization temperature range of 13⁰C which has less resistant to gelatinization (Shinoj *et al.*, 2006) and can be incorporated baking of bread and cakes, extrusion of cereal-based products, gravy, soup, heat set gel, porridge, instant powders

and modified flour and starches for specialty foods. As with other food grains, the nutritive value of kodo millet protein could be improved by supplementation with legume protein.

The consumption of cereals and legumes all over the world gives them an important position in international nutrition. Besides the high starch and protein content as energy source, these grains provide dietary fibre, nutritious protein and lipids rich in essential fatty acids, vitamins, minerals, antioxidants and phytochemicals. A multigrain snack helps in high intake of fibre and health enhancing components (Ragae and Abddel Aal, 2006).

Cookies are the most popular bakery items consumed nearly by all levels of society. This is mainly due to its ready-to-eat nature, good nutritional quality and availability in different varieties and affordable cost. Most of bakery products are used as a source for incorporation of different nutritionally rich ingredients for their diversification.

Cookies are made in a variety of style using an various ingredients including sugars, spices, chocolates, butter, peanut butter, nuts or dried fruits. Cookies are ideal for nutrient availability, palatability, compactness and convenience. They differ from other bakery products like bread and cakes because of having low moisture content, comparatively free from microbial spoilage and long shelf life. It is an unleavened crisp, made from wheat flour, shortening (hydrogenated fat) and sugar and is usually made light by the addition of baking powder. Wheat flour constitutes the basic ingredient for cookies production because of its gluten proteins, which are not present in flour of other cereals.

Biscuit can be defined as a small baked product made principally of flour, sugar and fat (Manley, 1998). It is different from other baked products like bread and cakes as biscuit has low moisture content. Its moisture content is usually less than 4 per cent and thus it has long shelf life, perhaps six months or longer. According to Manley (1998), biscuits can be divided into 10 major types: 1. Bread, pizza and crisp bread, 2. Water biscuits and soda crackers, 3. Cream crackers, 4. Cabin biscuits, 5. Savoury crackers, 6. Semi-sweet/hard-sweet, 7. Continental semi-sweet, 8. Short dough (moulded), 9. Wire cut types, 10. Short dough (sheeted). Wire cut types have the highest fat content in their formulation compared to other types of biscuit. Basically, there are two types of biscuit

dough; hard and soft. Hard or developed dough is used to make semi-sweet biscuit. Hard dough has high water content and relatively low fat and sugar contents. The dough is hard and extensible (it can be pulled out without immediately broken) (Manley, 1998). The principal ingredients of biscuit dough are soft wheat flour, sugar, fat, and water. They are mixed with other minor ingredients (such as baking powder, skimmed milk, emulsifier, and sodium metabisulphite) to form dough containing a well-developed gluten network. The nature and quantity of ingredients in the dough determine the quality of the biscuit. Several researchers have described the effect of major ingredients in biscuit dough system on the final product (Blanco *et al.*, 2016).

Biscuit along with bread forms major baked food accounting for over 30 and 50 per cent of total bakery products, respectively, produced in India. The production of bread and biscuit have increased from 21 to 35 lakh tone from 2017 to 2018 and 42 to 46 lakh tones during 2018–2019, respectively, recording six fold increase in a quarter of a century. In India, total number of units producing bread and biscuit belong to medium and small scale sector and their number is estimated at about 4800. The per capita consumption of biscuit and bread is increasing gradually. Baking industry is considered as one of the major segments of food processing in India. Baked products are gaining popularity because of their availability, ready to eat convenience and reasonably good shelf life. Cookies hold an important position in snack food industry due to variety in taste, crispiness and digestibility. In view of the importance of kodo millet with respect to its therapeutic, medicinal and nutritional value, the present investigation has been undertaken with following objectives:

1. To study physico-chemical properties of kodo millet.
2. To study the physical, chemical, microbial, nutritional, organoleptic and storage quality of selected kodo millet cookies and biscuits.
3. To work out the economics of kodo millet cookies and biscuits.

2. REVIEW OF LITERATURE

This chapter deals with the comprehensive review of literature, which is relevant to the study. A review of literature is an essential and important part of scientific investigation. Its main purpose is to determine the previous work done and to assist the delineation of objectives, hypothesis and research procedures to be followed.

2.1 Physical characteristics of kodo millets

Grain density is one of the important parameters which determine dehulling and milling performance. Barnyard millet was found to be relatively denser (1.80 g/ml) followed by kodo millet and little millet (1.18 and 1.46 g/ml, respectively). The densities of foxtail and little millet were recorded to range from 1.09 to 1.42 and 1.21 to 1.39 g per ml, respectively (Mandhyan *et al.*, 1987; Hadimani and Malleshi, 1993; Srivastava and Batra, 1998; Itagi 2003 and Veena *et al.*, 2005). Thus, variations in the seed characteristics were evident which were typical of genotypes.

Vijaykumar *et al.* (2013) subjected the kodo millet to heat treatment such as boiling for 25 min at 95 -100⁰C, steaming at 80- 90⁰C and pressure cooking at 9.8×10^4 Pa for 20 min. Boiling reduced the starch yield, exhibited greater porosity and water absorption capacity. Pressure cooked sample had greater oil absorption capacity and swelling power; reduced the peak and final viscosity which indicates large starch damage due to greater α -amylase activity. The resistant starch content was higher pressure cooked flour resulting in low starch digestibility index. They concluded that since, the pressure cooked flour was less viscous, high dense, rich in resistant starch, it is more suitable food in the diet for degenerative disorders.

Kumar *et al.* (2016) suggested size of kodo millet increased from 1.90 to 2.07 mm. This was due to the fact that with the increase in moisture content the size of the grain increased upon swelling, which in turn increased the diameters in all the three direction. It was observed that the length of kodo increased with increasing moisture content. The length of kodo varied from 2.61 to 2.74 mm. With respect to moisture content similar trends were observed width and thickness of kodo which varied from 1.96 to 2.23 mm and 1.33 to Length-breadth (L/B) ratio for kodo grains varied from 1.33 to 1.23 mm.

It was observed that with the increase moisture content the length-breadth ratio of kodo 1.45 mm, respectively. This was due to the fact that with the increase in moisture content the length, width and thickness of the grain increased upon swelling which in turn increased in length, width and thickness decreased. This result may be due to more expansion in breadth as compared to the length with respect to moisture content.

Muragod *et al.* (2019) stated that kodo millet is one the ancient grains of the world, In India different kinds of traditional foods are made as small millet grains as staple diet for many rural and urban households. Kodo millet is rich in carbohydrate and crude fiber. Millets are nutritionally superior to other cereals. Kodo millet grains were evaluated for physico chemical properties employing standard procedure. Kodo millet is light yellow in colour, small seeded (1.7 mm) with thousand grain weight volume and density of 2.8 g, 1.2 ml and 1.84, respectively. The mean hydration capacity of the grains was 0.54 per cent with an index of 24.52. Swelling capacity of the grains was 0.55 per cent with an index of 42.30.

2.2 Nutritional composition of kodo millet

Grains are the storehouses of many chemical components including nutrients, phytochemicals, and non-nutritive plant protective functional constituents. The nutritive value of millets is comparable to other cereals with slightly higher contents of protein and minerals (Gopalan *et al.*, 2006).

Sudharshana *et al.* (1988) stated that the kodo millet is a nutritious grain and is a good substitute to rice or wheat. The protein, fiber and mineral content are much higher than the major cereals like rice. The kodo millet grain is composed of 8 per cent protein. The major protein fraction in kodo millet is glutelin.

Studies on several varieties of millet among the different millets, kodo millet had the highest proportion of total minerals (4.9 %) barnyard millet had the total mineral content ranging between 1.5 to 4.0 per cent and lowest 1.4% was recorded in foxtail millet (Veena *et al.*, 2005). Varietal differences in nutrient composition within the species of minor millets were reported by several investigators. Studies indicated that barnyard millet recorded the lowest moisture content of 8.15 to 9.84 per cent. It was also reported that proso millet recorded a high moisture content ranging from 10.60 to 15.00 per cent, followed by little, foxtail and kodo millet with values of 10.7, 11.5 and 10.2 per cent,

respectively (Lorenz and Dilsaver, 1980; Kulkarni *et al.*, 1992; Kulkarni and Naik, 1999 and Veena *et al.*, 2005).

Chandrasekara and Shahidi (2012) study reveals that the phenolic content after digestion and colonic fermentation by gut micro flora, of cooked kodo millet had the lowered insoluble residue, which mainly consist of insoluble fiber (81 mg/g of cooked grain) as compared to finger millet (179 mg/g of cooked grain), pearl millet (132 mg/g of cooked grain), foxtail millet (157 mg/g of cooked grain) and proso millet (210 mg/g of cooked grain).

Chandrasekara *et al.* (2012) report says that the antioxidant activity of whole kodo millet decreased from 32.4 to 6.86 in case of dehulled one, whereas the dehulled boiled kodo millet has only 6.06, further the bran contains about 112 (μmol ferulic acid equiv/g defatted meal).

Annor *et al.* (2013) study revealed that expected glycine index (eGI) of whole kodo starch is lower than the rice starch. Moreover, the whole kodo grain has lower starch digestability and eGI than the decorticated grains. Therefore, they advocated use of whole kodo millets for development of acceptable product to maintain its hypoglycemic property.

Amadou *et al.* (2013) emphasized the importance of millets in providing sulphur containing essential amino acid like methionine and cysteine and the loss of germ, antioxidant containing bran and other minerals on debranning.

Chandel *et al.* (2014) reported that millet is an excellent source of fiber (9 %), as opposed to rice (0.2 %), and wheat (1.2 %). Kodo millet contains 66.6 g of carbohydrates and 353 kcal per 100 g of grain, comparable to other millets. It also contains 1.4 per cent fat and 2.6 per cent minerals. The iron content in kodo millet ranges from 25.86 to 39.60 ppm.

2.3 Health benefits of minor millets

Millets have been widely explored for health benefits owing to their chemical composition by scientific community. The health benefits of minor millets as reported by several investigators are summarized as follows:

The potential health benefits of dietary fibre have been demonstrated almost four decades ago by Burkitt *et al.* (1972). It was reported that dietary fibre reduces intestinal

transit time, provides fecal bulk and in turn prevents constipation. Thus, it helps in providing protection against duodenal ulcers and colorectal cancer (Nyman and Asp, 1982). Millets have been reported to be the rich sources of dietary fibre (Wisker *et al.*, 1985).

Resistant starch (defined as any starch that escapes digestion in small intestine) was also reported to exhibit a wide range of health benefits such as lowering caloric density and low glycemic response (Jenkins *et al.*, 1982 and Ring *et al.*, 1988). It was also reported to lower digestibility and act as a fecal bulking agent (Ranhotra *et al.*, 1991). Minor millets, with their low carbohydrate content, low digestibility and water soluble gum content (β -glucan) have been attributed to improve glucose metabolism. These grains release sugar slowly in the blood and also diminish the glucose absorption (Chen *et al.*, 1984 and Anderson *et al.*, 1991). The dietary fibre and resistant starch of minor millets have been attributed to exhibit hypoglycemic and hypolipidemic effects (Mani *et al.*, 1993, Slavin and Dwyer, 1994, Krishna kumari and Thayumanavan, 1997 and Pathak and Srivastava, 1998). Further the antioxidative properties of barnyard millet fortified cookies minor millets against hyperglycemia and oxidative stress have also been studied, which is mainly determined by their higher reserves of phytochemicals like phenolics, tannins, phytates, micro minerals etc. (Hegde *et al.*, 2004).

In addition, these grains have been demonstrated to exhibit beneficial effects on cholesterol levels, which is again attributed to their high dietary fibre and phytochemical content. It is reported that cardiovascular diseases, duodenal ulcers and hyperglycemia occur rarely in regular millet eaters (Menon, 2004). For all these superior properties of minor millets, they can be designated as “nutritious millets”.

2.4 Development of value added cookies

Ranhotra *et al.* (1991) developed high fiber chocolate chip and oat meal raisin cookies by substituting treated flour at 0, 50, 75 and 100 per cent levels with untreated wheat flour. The treated flour was prepared by exposing the flour to repeated autoclaving and cooling cycles. Complete replacement of untreated flour with treated flour adversely affected the cookies dimensions and organoleptic characteristics. The total dietary fiber content in chocolate chip cookies varied from 1.50 per cent in control cookies to 3.50 per

cent in 75.00 per cent treated flour incorporated cookies. However, in oat meal raisin cookies it ranged from 2.70 per cent to 3.90 per cent.

Singh *et al.* (1996 and 2000) developed defatted soy flour cookies and reported that with increased incorporation (from 0 to 50%) the cookie diameter, spread ratio and spread factors decreased (61.40 to 56.00 mm, 8.77 to 7.78 and 100 to 88.71 %, respectively), where as hardness increased (26.97 N to 46.25 N). It was also proposed that soy flour incorporation up to 20 per cent resulted in acceptable cookies. The level of incorporation of soy flour resulted in nutrient composition in two fold increase in protein (4.91 to 10.54 %) and four fold increase in crude fiber (0.12 to 0.46 %). Soy biscuits (20 %) contained higher amounts of calcium (41.46 mg/100g), phosphorous (181.78 mg/100g) and iron (14.53 mg/100g) than control biscuits (16.53, 147.32 and 14.53 mg/100g of calcium, phosphorous and iron, respectively). Soy biscuits exhibited higher non enzymatic browning (0.08 OD) than control biscuits (0.06 OD), which was attributed to higher levels of reducing sugars and protein in the formulation. These cookies exhibited higher *in vitro* protein digestibility (83.82 %) than control biscuits (68.46 %).

Yahya (2004) developed low fat, mungbean and chickpea (25 % each) cookies with 40 per cent sago flour as a fat replacer. It was reported that addition of fat replacer resulted in harder cookies of smaller size. Most acceptable chickpea cookies exhibited higher dietary fiber (5.38 %) and trypsin inhibitor activity (3.76 %) but low protein digestibility (70.16 %). Blend flour of soy and corn (30:20) was substituted with carrot flour (5 to 50 %) for cookie preparation by Akubor (2005). The blend flour cookies exhibited highest scores for sensory attributes. The carrot flour incorporated cookies (5 to 10 %) were comparable to blend flour cookies. However, the substitution of carrot flour beyond 10 per cent adversely affect the sensory attributes of cookies.

Sindhuja *et al.* (2005) reported that increased incorporation of grain amaranth flour in cookies (0 to 30 %) significantly reduced the spread ratio from 7.82 to 6.50, breaking strength from 4.94 to 3.39 kg but increased the thickness from 10.9 to 11.5 mm. Further, addition of lecithin (0.25 %) and glycerol monostereate (0.25 %) resulted in significantly increased the cookies spread (8.49) and decreased the breaking strength

(3.81 kg). Incorporation of grain amaranth flour was reported to yield golden brown cookies which were malty and sweet flavoured.

Muhammad *et al.* (2006) reported that addition of wheat flour with defatted wheat germ (DFWG) at levels of 0-25 per cent was investigated for nutritional properties of cookies. The crude protein content of defatted wheat germ was as high as 27.8 per cent with a highly valuable. The cookies containing 15 per cent defatted wheat germ was best regarding protein bioavailability in rats. The diets containing 15 per cent defatted wheat germ has values similar to the casein diet.

Shariff *et al.* (2009) developed fiber and mineral enriched cookies by supplementing defatted rice bran in wheat flour at 0 to 50 per cent level. It was reported that cookies with rice bran (20 %) were highly acceptable. Beyond 20 per cent level, sensory scores for all sensory attributes decreased significantly. The cookies exhibited high dietary fiber (5.94 %), potassium (18.90 mg/100g), calcium (109.22 mg/100g) and magnesium (125.98 mg/100g) than control biscuits (2.56, 44.55, 54.95 and 10.64 mg/100 g of dietary fiber, potassium, calcium, and magnesium, respectively).

Sweta *et al.* (2011) studied the utilization of alternate flours meals rice, maize, sorghum and pearl millet for the preparation of gluten free cookies as compared to conventional wheat flour cookies. The cookies prepared from rice and maize combination had highest spread ratio of 16 whereas, the lowest spread ratio was observed in rice and sorghum combination. The cookies with pearl millet and sorghum flour combination had higher fat, protein, ash and calorific values as compared to control cookies. The maximum sensory overall acceptability scores were found for cookies prepared from combination of pearl millet and sorghum flour followed by rice and sorghum, maize and sorghum, rice and maize, maize and pearl millet, rice and pearl millet and control cookies. All gluten free cookies had higher nutritional value as compared to control cookies and were acceptable by panel.

Sambavi *et al.* (2015) conducted experiment on development of cookies using a combination of foxtail millet and wheat flour and reported that composite flour of 55 per cent wheat and 45 per cent foxtail millet flour cookies were highly acceptable compared with other types of cookies. Their study further illustrated that the quality of cookies could be improved with foxtail millet flour, in terms of crude fiber and ash. The use of

foxtail millet flour in cookies making and other food products, would greatly enhance the utilization of this crop in developing countries like Sri Lanka. The nutritional quality of refined wheat flour cookies could be improved with supplementation of foxtail millet flour. Foxtail millet is preferred as it has a higher glycemic index. Further work is necessary in terms of microbiological qualities and shelf life.

Pushendra Kumar *et al.* (2015) studied development and quality evaluation of cookies incorporated by millets and cardamom powder and reported that the result of sensory (appearance, colour, flavour, texture, taste and overall acceptability) evaluation revealed that the 80:20 per cent addition of millets: wheat flour has higher overall acceptability, taste, texture and flavour and it was accepted by the panelists. However, a declining trend in acceptability was observed with increasing level of millets wheat flour for all the sensory characteristics. The nutritional value of the cookies with 70:30 per cent of millets: wheat flour fortified protein cookies was higher than other samples.

Anshula Bhat *et al.* (2015) studied evaluation of nutraceutical properties of *Amaranthus hypochondriacus* L. grains and formulation of value added cookies and reported that phytochemical analysis of the dried grains revealed the presence of good phenolics, flavonoids, alkaloids and saponins content.

Ingle *et al.* (2017) stated that fruits and vegetables are important constituents of the diet and provide significant quantities of nutrients, especially vitamins, minerals and fiber. Beetroots (*Beta vulgaris* L.) are a rich source of potent antioxidants and minerals including magnesium, sodium and potassium. It contains betaine, which is important for cardiovascular health. Beetroots are low in calories (45 Kcal per 100g) and have zero cholesterol. The study was conducted to improve the nutritional qualities of cookies with incorporation of different levels of beetroot powder i.e. 0, 5, 7, 10, 15 and 20 per cent and examined for its physical and chemical composition. The proximate composition of cookies enriched with beetroot powder from 5 to 20 per cent indicated that protein was increased from 7.39 to 9.12 per cent, crude fibre 0.95 to 1.90 per cent and ash content 0.93 to 1.89 per cent. The incorporation of beetroot powder in cookies lowered the lightness (L^*) and yellowness (b^*) but increased redness (a^*) of cookies. The hardness of the cookies was increased with increasing the level of beetroot powder. Sensory

evaluation of cookies concluded that the cookies prepared with addition of 10 per cent beetroot powder were more acceptable as compared to others.

2.5 Development of value added biscuits

Prabhavathi *et al.* (1973) developed high protein biscuits using wheat, groundnut and soybean flours in the ratio of 55:20:25. The per cent of moisture, crude fibre, ash, protein and reducing sugar were found to be 7.5, 0.52, 3.25, 15.8, and 4.9 respectively. Calcium and total phosphorus content of biscuits were 439 and 228 mg/100 g respectively. The total lysine content in unbaked and baked biscuits was 4.13 and 3.81 g/100 g protein.

Foda *et al.* (1984) evaluated the quality of biscuits supplemented with low-fat soy flour at 0, 10, 20 and 30 per cent levels. The chemical composition of biscuits was observed as moisture 2.24-6.97, fat 9.43-10.45, protein 11.43-17.14 and ash 0.49-1.90 per cent.

Patel and Rao (1995) evaluated the effect of incorporation of untreated, roasted and germinated black gram flour (0 to 25 %) on biscuit characteristics. It was reported that the incorporation of black gram flour decreased the spread ratio with concurrent increase in thickness of biscuits. Maximum reduction was indicated in biscuits incorporated with germinated black gram. The reduction in spread was attributed to altered starch properties of the flours used.

The addition of black gram flour at 25 per cent levels increased the hardness values of biscuits. Hardness of control wheat flour biscuits was reported to be 2117 g, which increased to 2442 g in untreated black gram and further higher value of 2867 g in germinated black gram dhal incorporated biscuits. Further it was inferred that black gram incorporation in any form decreased the sensory attributes of biscuits.

Onweluzo and Iwezu (1998) developed biscuits by blending fermented and unfermented soy flour with whole wheat and cassava flours in different proportions. It was reported that spread ratio (1.80) and biscuit weight (11.70 g) were highest in control samples, which also exhibited lowest breaking strength (1.80 kg). The acceptability of biscuits with whole wheat flour and fermented soy flour (1:1) was comparable to control biscuits, which contained protein (8.80 %), crude fiber (1.00 %) and fat (25.70 %) with calorific value of 486 Kcal.

Devi *et al.* (2000) developed four types of biscuits namely *nankatai*, melting moments, tricolour and salt biscuits using green gram dhal. It was reported that salt biscuits contained highest fiber (5.81 %), total minerals (11.20 %) phosphorous (30 mg/100g) and moisture (5.83 %). Fat (52.27 %) and protein (20.28 %) contents were highest in melting moments. *Nankatai* contained highest calcium (66.10 mg/100g) and carbohydrates (30.36 %), while, the tricolor biscuits contained highest iron (0.94 mg/100g). With regard to *in vitro* protein digestibility highest, (64.71 %) was recorded in salt biscuits and lowest in melting moments (57.76 %).

Singh *et al.* (2000) studied the nutritional evaluation of soy fortified biscuits and found that addition of 20 per cent defatted soy flour increases the protein content of biscuits by about 115.07 per cent and fat was found to be more or less same.

Akubor (2003) developed biscuits from cowpea and plantain flour blends (60:30). The blend flour was incorporated with wheat flour at different levels ranging from 10 to 60 per cent. It was indicated that at all levels of wheat flour substitution thickness and diameter of cookies were not affected. The biscuit thickness ranged from 3.60 to 3.80 cm, weight from 7.10 to 8.20 g and spread ratio from 3.10 to 3.40. The sensory scores for colour, flavour and texture of the blend biscuit (85:25) were comparable with those of 100 per cent wheat flour biscuits. The protein content of the cowpea plantain flour / wheat flour biscuits ranged between 15.20 to 18.96 per cent, the values increased with increased levels of inclusion of cowpea flour in the blends.

Narender *et al.* (2007) prepared biscuit from the blends containing 0, 10, 20 and 30 per cent of whey protein concentrate (WPC) and evaluated for nutritional, textural and sensory characteristics. The protein and ash contents of WPC containing biscuits were significantly higher than the control. The protein content of control sample was 5.2 per cent while it was 14.1 per cent in the 30 per cent whey protein concentrate containing biscuits.

Mridula and Gupta (2008) developed high fiber biscuits by substituting bajra flour at 10 to 50 per cent in wheat flour and fortified with 5 per cent defatted soy flour. It was reported that with increased proportion of bajra flour, the thickness and spread ratio of biscuits decreased. This was attributed to high fiber and low dough strength of bajra flour blends. Bajra flour biscuits (50 %) recorded higher crude fiber (1.08 %), fat (24.16 %)

and ash (1.41 %) than the control biscuits (0.36, 22.10 and 1.15 % of crude fiber, fat and ash, respectively). However, protein content of bajra biscuits was less (6.53 %) than control biscuits (7.11 %).

Masur *et al.* (2009) developed high protein biscuits using Bengal gram flour (10 to 25 %), altering the levels of water, fat and baking powder. Results revealed that height (5.83 cm) and diameter (58.80 mm) of biscuits were unaffected up to 20 per cent level of bengal gram dhal flour incorporation, but the spread ratio and spread factor decreased in 25 per cent bengal gram flour incorporation. The sensory quality of biscuits prepared with 16 per cent fat and 13 ml water resulted in highest scores for flavour, texture and overall acceptability. The most acceptable biscuits (20 % bengal gram dhal flour incorporation) exhibited higher protein (5.59 %), fat (0.85 %) and crude fiber (0.50 %) compared to control biscuits (4.98, 0.41 and 0.14 % of protein, fat and crude fiber, respectively).

Ranganna and Ramya (2010) reported that preparing butter biscuit, sponge cake and ajwain biscuit using kodo and wheat flour. The mixture having ratio of 20:80, 30:70 and 40:60 was used for both butter biscuit and sponge cake, whereas for ohmo biscuit, the above flour were in the ratios of 15:85, 20:80 and 25:75, respectively.

Ranganna *et al.*, (2011) used parboiled kodo millet flour for preparation of biscuit and sponge cake using different ratio of kodo millet flour and wheat flour 20:80, 30:70 and 40:60, and 15:85, 20:80 and 25:75, respectively.

2.6 Shelf life of value added biscuits

Good shelf life of foods is an important criteria in consumer acceptability and marketability of new products. Shelf life depends on chemical composition, packaging material, environmental factor and handling. The study of shelf life foods helps in understanding mechanism of extending storage quality for added value.

Goyle and Gujral (1993) studied the keeping qualities of biscuits produced from malted and raw wheat bengalgram grains (4:1) with or without 7.50 g colocassia leaf powder. The biscuits were packed in 200 gauge low density polyethylene packets, kept at accelerated conditions (37⁰C and 90 % RH) and at room temperature for one month. The study revealed that the moisture content ranged between 4.22 and 4.54 per cent. Under accelerated conditions, biscuits became soggy after 14 days; but were crisp at ambient.

The acidity of the extracted fat ranged between 0.29 and 0.58 per cent, however the peroxide value was below 1.00 meq/kg fat at the two storage environments even after 21 days of storage. Dietetic cookies and biscuits prepared using defatted soy flour (5 to 10 %) with dietetic baking powder and packed in polyethylene pouches at ambient exhibited a poor shelf life of 7- 9 days (Lakshmi, 1991). Isabgol (4, 8 and 12 %) and wheat bran (20, 30 and 40 %) incorporated biscuits at different proportion were evaluated for storage quality by Shubha (1994). It was reported that biscuits were acceptable only for 7 days without any significant changes in sensory qualities. The samples stored for 35 days at ambient, packed in low density polyethylene pouches exhibited lowered acceptability as a consequences of storage.

Rao *et al.* (1995) assessed the storage quality of flavored biscuits containing whole egg powder (10 g). Different flavours such as orange, vanillin and pineapple were used in the formula. The biscuits were packed in PEP, paper aluminum foil (0.012 mm) polyethylene laminated pouches, metalized polyester (35 μ) and biaxillary oriented polypropylene (12 μ) pouches and stored at 37⁰C at ambient temperature of 19-26⁰C and control at 4⁰C for six months. It was reported that vanillin and pineapple flavored biscuits were rated better for organoleptic characters than the orange flavored biscuits. Biscuits packed in biaxillary oriented polypropylene and metalized polyester pouches exhibited higher moisture content than those packed in polyethylene propylene pouches. The formation of peroxides (39.33 %) and thiobarbiturates (41.61 %) were found to high in biaxillary oriented polypropylene, where as free fatty acids increased in orange flavored biscuits at 37⁰C, packed in polyethylene propylene pouches.

Singh *et al.* (2000a) studied the storage quality of soy fortified biscuits. It was reported that the higher moisture uptake was recorded in biscuits packed in laminated pouches than in polypropylene material (79.19 %) during 60 days storage period. Non enzymatic browning of both control (from 0.060 to 0.110 OD) and soy biscuits (from 0.085 to 0.110 OD) increased. Soy biscuits recorded higher peroxides (7.00 meq/kg fat) than control (6.00 meq/kg fat) initially, increased to 11.24 and 11.10 meq/kg fat, respectively, during storage. However, the effect was prominent in biscuits packed in laminated pouches. Similar trend was observed with free fatty acids.

Shelf life of finger millet biscuits packed in two layered polypropylene was 75 days (at 90 % Rh, 38⁰C) and 50 days in metalized polyester poly laminated packs. The shelf life was increased over 120 days (at 65 % Rh, 27⁰C) in the two packages (Selvaraj *et al.*, 2002). Shariff *et al.* (2003) investigated shelf life of rice bran oil cookies. It was reported that the cookies prepared by replacing normal shortenings with rice bran oil were highly acceptable even after 45 days of ambient storage. Highest thiobarbiturates were recorded in cookies with 100 per cent rice bran oil (0.05). Initial values before storage were 0.03. Thus it was concluded that, by increasing the amount of rice bran oil the thiobarbiturates number decreased and the onset of rancidity delayed.

Reddy *et al.* (2005) proved that adding natural antioxidants to biscuits such as extracts of raisins (2.00 %), amla and drumstick leaves (1.00 % each) could retain much of the antioxidant activity, besides being highly acceptable when compared to biscuits containing normal fats and BHA. It was reported that antioxidant activity of biscuits decreased gradually during storage at ambient temperature in LDPE packaging material. Maximum lipid stability was reported in drumstick leaf biscuits with retention of 95 per cent antioxidant activity.

Hemalatha *et al.* (2006), evaluated little millet biscuits for storage quality. The study indicated that the biscuits packed in metalized polyester polyethylene laminated pouches stored for 120 days under ambient conditions exhibited increased acidity and moisture content. The values increased with days of storage. The millet (30.00 %) incorporated cookies though recorded higher moisture (4.04 %) than the control cookies (3.82 %), the levels were within the critical levels at the end of the storage in LDPE packaging material. These, studies indicated that the cookies and biscuits could be stored satisfactorily for several months at ambient condition using suitable packaging materials.

3. MATERIALS AND METHODS

The present investigation entitled, “Evaluation of nutritional quality of kodo millet and its exploration in cookies and biscuits” was carried out at the Department of Food Science and Technology, Post Graduate Institute, Mahatma Phule Krishi Vidyapeeth, Rahuri. This chapter deals with the experimental materials and methods adopted for conducting the experiments. The methodologies on preparation of raw material physicochemical properties of flour and cookies and biscuits, the storage studies of cookies and biscuits were performed and methodologies are reported under following heads.

3.1 Material

3.1.1 Ingredients for cookies and biscuits

The materials like maida vanaspati/ghee, sugar and other ingredients were procured from local market (Plate 3.1).

3.1.2 Kodo millet

The kodo millet was procured from Zonal Agriculture Research Station, Kolhapur.

3.1.3 Packaging material

The packaging material *viz.*, LDPE and PP bags were procured from local market and used for packaging of cookies and biscuits for storage study (Plate 3.2).

3.1.4 Equipments

The grinder, Instron texture analyzer, decorticator available in the Department of Agricultural Process Engineering, Dr. Annasaheb Shinde College of Agricultural Engineering and Technology, Mahatma Phule Krishi Vidyapeeth, Rahuri were used. The machineries available in pilot plant of bakery unit of the Department of Food Science and Technology were used for making cookies and biscuits.

3.2 Methods

3.2.1 Physical properties of kodo millet

3.2.1.1 Colour and shape

The kodo millet colour and shape was determined by visual observation.

3.2.1.2 Weight of 1000 seeds of kodo millet

The 100 seeds, were randomly drawn from the bulk sample and weighed on an electronic balance with 0.2 g accuracy and then extrapolated this weight to 1000 seeds (Vilche *et al.*, 2003; Tunde and Akintunde, 2004).

3.2.1.3 Bulk density

The bulk density, defined as the ratio of the mass sample of the seeds to its total volume, was determined using a modified standard test weight procedure (Singh and Goswami, 1996) by filling a 90 ml container with seeds poured from a height of about 150 mm at a constant rate, then the content was weighed. No separate manual compaction of seeds was done. The bulk density was calculated by dividing mass of the seeds to its volume.

3.2.1.4 True density

The true density, defined as the ratio of the mass of the sample of seeds to the solid volume occupied by the sample. This was determined using an electronic balance reading to 0.0001 g and a pycnometer (50 ± 0.1 ml) by liquid displacement method (Mohsenin 1970). Xylene (density: 0.862 ± 0.001 g cm³) was used instead of water as it is absorbed by seeds to a lesser extent. Due to the short duration of the experiment, xylene absorption was found to be negligible (Giner and Calvelo, 1987).

3.2.1.5 Porosity

The porosity (ϵ) of the bulk seed is the fraction of the space in the bulk grain that is not occupied by the grain (Thompson and Isaacs, 1970). The per cent porosity was calculated by the following formula (Mohsenin, 1970).

$$\text{Porosity (\%), } \epsilon = \frac{\text{True density} - \text{Bulk density}}{\text{True density}} \times 100$$

3.2.1.6 Angle of repose

The angle of repose was determined by using a plywood box (0.3 m of length, width and height), which had a removable front panel. The box was filled with seeds and

the front panel was quickly removed, allowing the seeds to flow to their natural slope. The angle of repose was calculated from the measurements of the horizontal displacement distance of the seeds and the height of the heap (Dutta *et al.*, 1988). Angle of repose is calculated by: $\tan \theta = h / r$.

3.2.2 Chemical characteristics of kodo millet flour

3.2.2.1 Moisture

The moisture content of the selected sample was estimated by A.A.C.C. method (2000).

Procedure

The 5 g kodo millet sample was taken in pre-weighed moisture box, dried in oven at 105⁰C for 5 hr and transferred to desiccators for cooling for 30 min. After cooling the sample was weighed. The procedure was repeated until a constant weight was obtained.

$$\text{Moisture (\%)} = \frac{(W_1 - W_2)}{W_1 - W} \times 100$$

Where,

W_1 = Weight (g) of the box with the material before drying

W_2 = Weight (g) of the box with the material after drying

W = Weight (g) of the empty box

3.2.2.2 Protein

Total protein content of the samples was estimated by determining total nitrogen content using standard Micro-kjeldhal method (A.A.C.C., 2000). Total protein content was calculated by multiplying the estimated total nitrogen content with a factor 6.25.

Reagents

1. Conc. sulphuric acid (specific gravity 1.84 g/cc, purity, 98.08 % nitrogen free).
2. Catalyst mixture: Potassium sulphate (99 g), mercuric oxide (4.1 g) and copper sulphate (0.8 g) were weighed, mixed and ground into a fine powder.
3. Sodium hydroxide (50%, w/v): Fifty gram of sodium hydroxide and 5 g of sodium thiosulphate were dissolved in distilled water separately, mixed and the volume was made up to 100 ml with distilled water.
4. Boric acid (4 %, w/v): Four g of boric acid was dissolved in distilled water and the volume was made up to 100 ml with distilled water.

5. Hydrogen peroxide (30%, v/v) commercially available in the market.
6. Mixed indicator: It was prepared by dissolving 0.1 g of bromocresol green and 0.1 g of methyl red in 100 ml of 95 % (v/v) alcohol separately. Ten parts of bromocresol green and 2 parts of methyl red indicator were mixed together and transferred to a bottle provided with a stopper.

Procedure

Powdered defatted sample (200 mg) was accurately weighed and transferred to a digestion flask. One gram of catalyst mixture was added and mixed thoroughly with the sample. Five ml of conc. sulphuric acid and five ml of hydrogen peroxide were carefully added and the sample was digested in digestion chamber. Initially the flasks were heated slowly for 10 to 15 min and then the temperature was raised gradually so that the contents boiled briskly. The digestion was continued until the sample became clear and colorless. The flasks were then cooled and after cooling the contents they were transferred to volumetric flasks. The digestion flasks were washed 3 to 4 times with distilled water. All the washings were transferred to volumetric flasks and the volume was made to 50 ml.

Ten ml of boric acid solution was pipette into a 100 ml beaker and 6 to 8 drops of mixed indicator solution were added. The beaker was placed under the condenser of the distillation unit to collect at least 50 ml distillate. At the end of distillation, the tip of condenser was washed with distilled water to collect all ammonia. The distillate was then titrated with standard hydrochloric acid solution. Before distillation, the color of boric acid plus indicator was pink which changed to blue green during distillation and finally to pink red at the end of titration. Blank titration was simultaneously carried out. The per cent of nitrogen content was calculated from the quantity of standard hydrochloric acid required for titration of the sample. The protein content was calculated by multiplying the nitrogen content by a factor of 5.46.

Total protein content of sample was calculated by formula.

$$\text{Nitrogen (\%)} = \frac{(\text{S-B}) \times \text{N} \times 14.007}{\text{Weight of sample (g)}} \times \frac{\text{Volume made (ml)}}{\text{Volume taken (ml)}} \times 100$$

Where,

S = ml of HCl required for sample titration

B = ml of HCl required for blank titration

N = Normality of HCl (0.02 N)

$$\text{Protein (\%)} = \text{Nitrogen (\%)} \times 6.25$$

3.2.2.3 Fat

The fat content of the selected sample was estimated by the Soxhlet method of A.A.C.C. method (2000).

Procedure

The sample (10 g) was taken on a filter paper and made suitable size sachets. The sachets were placed in extraction flask of Soxhlet unit and attached it to collecting flask. Poured one siphon full of hexane solvent through extraction flask into pre weighed collecting flask. Assembly condenser was connected to tap water and the flask was heated at 60⁰C. Sample with 6-8 siphoning was extracted and distilled off most of the hexane to extraction flask. The collecting flask was dried on water bath and then at 100⁰C for 1 hr in oven. The flask was cooled and weighed. Increase in weight of flask represents crude fat content.

$$\text{Fat content (\%)} = \frac{W_2 - W_1}{X} \times 100$$

Where,

W₂ = Weight of round bottom flask with fat

W₁ = Weight of empty round bottom flask

X = Weight of sample

3.2.2.4 Crude fiber

Crude fiber content of the selected samples was determined by the method of A.A.C.C. (2000).

Reagents

1. 0.255 N Sulphuric acid solution: A measured quantity of 1.25 ml of concentrated sulphuric acid was dissolved in glass distilled water and volume was made up to 100 ml.
2. 0.313 N Sodium hydroxide solution: A weighed amount of 1.25 g of sodium hydroxide

was dissolved in glass distilled water and the volume was made up to 100 ml.

Procedure

About 2 g fat free residue was taken and then transferred to the digestion flask. 200 ml boiling sulphuric acid was added and immediately the flask was connected to condenser. The flask was heated, boiled by frequently rotating for 30 min. and the volume was maintained with hot water and then filtered through filter cloth in a fluted funnel. The residue was washed on cloth with hot water or potassium sulphate solution. The residue was returned to digestion flask by washing with hot water, 200 ml boiling sodium hydroxide was added and boiled for 30 min. The volume was adjusted with boiling water, filtered it through the muslin cloth and the residue free of alkali was washed. The residue was transferred into crucible and washed with 15 ml alcohol and the crucible was dried at 110 °C for 2 hr. The crucible was cooled in desiccators and weighed. The crucible was ignited in the furnace at 550 °C for 30 min. then cooled and weighed. The loss in weight represented the crude fiber.

$$\text{Crude fiber (\%)} = \frac{(W_1 - W_2)}{\text{Weight of sample (g)}} \times 100$$

Where,

W_1 = Weight of material before ashing (g)

W_2 = Weight of material after ashing (g)

3.2.2.5 Carbohydrate

The content of carbohydrate in the selected samples were obtained by subtracting from 100, the sum of values of moisture, protein, fat and ash content per 100 g of the sample (Raghuramulu, *et al.*, 1993).

$$\text{Carbohydrate} = 100 - (\text{Moisture} + \text{protein} + \text{fat} + \text{ash})$$

3.2.2.6 Mineral

The mineral contents like magnesium (Mg), iron (Fe) and zinc (Zn) were determined by the method described in AOAC (2000). One gram of sample was digested with 10 ml of nitric acid: Perchloric acid (7:3) mixtures at temperature up to 180-200 °C till transparent contents were obtained. The contents were diluted to a volume of 100 ml with double distilled water. Concentration of mineral contents was determined by running the diluted samples through Atomic Absorption Spectrophotometer (Model: Varian, AA-

240) using air acetylene flame.

Calcium

The calcium content in the samples was determined by following procedure (A.O.A.C., 2000).

Reagents

1. Sodium acetate (20 %)
2. Oxalic acid (3 %)
3. Ammonium hydroxide (15 solutions with water)
4. Potassium permanganate (0.05 N-Dissolve 0.79 g KMnO_4 in water to make 500 ml).
5. Sulphuric acid
6. Bromocresol green indicator
7. Dilute HCl

Procedure

Five gram sample was ignited and prepared 250 ml acid extract (5 g sample burnt to ash and transferred in 150 ml beaker, added 30 ml dil. HCl- digestion, cooled and filtered through filter paper, made volume 250 ml with distilled water). About 20 ml extract was pipetted in 250 ml beaker, to which 8-10 drops of indicator and sodium acetate were added until color changed from green to blue. The beaker was covered with watch glass and heated to boiling. The calcium was precipitated by adding oxalic acid slowly to change color from blue to green. The precipitate was allowed to settle for some time, filtered through whatman No. 40/42 and washed with 50 ml ammonium hydroxide. The filter paper was punctured and washed out precipitate in beaker with mixture of 125 ml water and 5 ml conc. H_2SO_4 at 80 to 90°C. The filtrate was titrated against KMnO_4 at 70 to 90°C till permanent pink color was obtained. Filter paper was added to beaker and titration was continued (2-3 drops of KMnO_4) and the volume of KMnO_4 required was noted.

Calcium content was calculated by following formula.

$$\text{Calcium} = \frac{\text{Titre x N of KMnO}_4 \times 0.02 \times \text{Total vol. of ash solution (ml)}}{\text{ml of ash solution taken} \times \text{Weight of sample (g)}} \times 100$$

Iron content

The iron content was calculated by colorimetric method using Spectronic-20 (AACC 2000).

Reagent

1. Phennanthroline (0.1%)
2. Hydroxylamine hydrochloride (10%)
3. Acetate buffer (8.3 g unhydrous sodium acetate was dissolved in water, add 12 ml glacial acetic acid and dil. To 100 ml)
4. Std iron solution (100 ml dissolved in 0.7022 g ferrous ammonium sulphate in water by adding 2 drop conc. HCL and dil 100 ml).

Procedure

a. Standard curve: 0- 4 ml of std. iron solution was pipetted in 25 ml volumetric flask and adjusted to volume 5 ml. Then 1 ml hydroxylamine hydrochloride, mixed and rest for 5 min. Then 5 ml acetate buffer was added to this of phenanthroline reagent was added and diluted to 25 ml with water and mix. The absorbance was read at 515 nm and a graph of OD v/s iron content plotted.

b. Iron in sample: Burnt 5g sample was ashed in furnace and prepared extract as earlier. Taken 5 ml extract, develop colour as for as std. curve and read the absorbance on Spectronic-20. Iron content was calculated by following formula

$$\text{Iron (mg/100 g)} = \frac{\text{Reading (OD)}}{\text{Extract used for colour development}} \times \frac{\text{Vol. of acid extract}}{\text{Weight of sample}} \times 100$$

3.2.3 Preparation of kodo millet flour

Kodo millet grains were grinded in to flour and passed through sieve of 80 mesh to get uniform flour.

3.2.4 Preparation of kodo millet flour cookies

The cookies were prepared using following ingredients as per the traditional creaming process outlined in Fig 3.1.

Table 3.1. Recipe for making cookies

Ingredients	Quantity
Maida + kodo millet flour	100 g
Sugar	50 g
Vanaspati ghee	50 g
Sodium bicarbonate	1 g
Ammonium bicarbonate	1 g
Water	20 ml

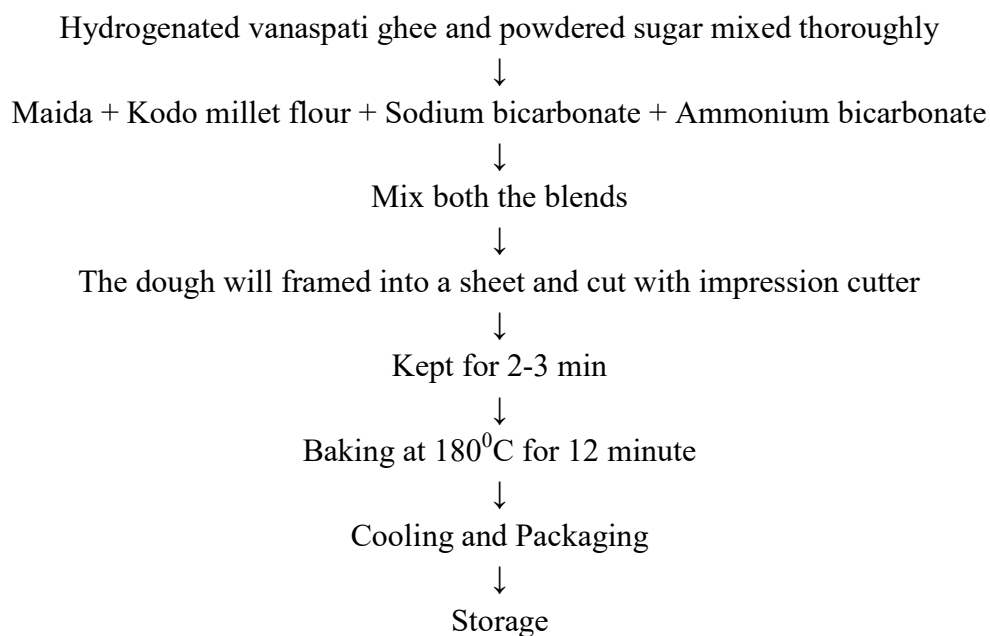


Fig 3.1. Flow chart for preparation of kodo millet cookies

3.2.5 Preparation of kodo millet flour biscuits

The biscuits were prepared using following ingredients as per the traditional creaming process outlined in Fig 3.2.

Table 3.2. Recipe for making biscuits

Ingredients	Quantity
Maida + kodo millet flour	100 g
Sugar	50 g
Vanaspati ghee	50 g
Sodium bicarbonate	1 g
Ammonium bicarbonate	1 g
Water	20 ml

Maida + Kodo millet flour+ Baking powder mix both well together



Sieve the ingredients twice



Mixed with creamed fat and sugar



Knead to soft dough



Spreading (Uniform thickness)



Cut with a fancy biscuits cutter



Bake at 160°C for 15-20 min



Cooling and packaging



Storage

Fig. 3.2. Flow chart for preparation of kodo millet biscuits

3.2.6 Treatment details

Based on review of literature and preliminary trials, the experimental work plan was prepared with details of the treatments of cookies and biscuits as given in Table 3.3 and 3.4.

Table 3.3. Incorporation of maida and kodo millet flour in cookies

Treatments	Maida (%)	Kodo millet flour (%)
CKMF ₀	100	0
CKMF ₁₀	90	10
CKMF ₂₀	80	20
CKMF ₃₀	70	30
CKMF ₄₀	60	40
CKMF ₅₀	50	50
CKMF ₆₀	40	60
CKMF ₇₀	30	70
CKMF ₈₀	20	80
CKMF ₉₀	10	90
CKMF ₁₀₀	0	100

Table 3.4. Incorporation of maida and kodo millet in biscuits

Treatments	Maida (%)	Kodo millet flour (%)
BKMF ₀	100	0
BKMF ₁₀	90	10
BKMF ₂₀	80	20
BKMF ₃₀	70	30
BKMF ₄₀	60	40
BKMF ₅₀	50	50
BKMF ₆₀	40	60
BKMF ₇₀	30	70
BKMF ₈₀	20	80
BKMF ₉₀	10	90
BKMF ₁₀₀	0	100

3.2.7 Analysis of cookies and biscuits

3.2.7.1 Sensory evaluation of kodo millet cookies and biscuits

The sensory evaluation of cookies and biscuits was carried out according to the method of Amerine *et al.* (1980) on 9 point hedonic score card as per the performa given in the Appendix I. In all, each treatment of cookies and biscuits were judged for the quality characteristics *viz.*, colour and appearance, texture, flavour, taste and overall acceptability. The average score given by all the 10 judges for different quality characteristics were recorded and mean values are presented as results.

3.2.7.2 Physical characteristic of cookies and biscuits

Physical parameters such as weight, diameter, thickness and spread ratio were determined using A.O.A.C. (2000). All parameters were determined in five replications.

3.2.7.2.1 Weight

Weight of cookies and biscuits was determined by using weighing balance and expressed in gram as average.

3.2.7.2.2 Diameter

Measured by vernier caliper and expressed in mm or diameter of cookies and biscuits was measured by laying six cookies and biscuits edge to edge with the help of a scale rotating them 90^0 and again measuring the diameter of six cookies and biscuits (mm) and then taking average value.

3.2.7.2.3 Thickness

Measured by screw gauge and expressed in mm or thickness was measured by stacking six cookies and biscuits on top of each other and taking average thickness (mm).

3.2.7.2.4 Spread ratio

Spread ratio was calculated by dividing the average value of diameter by average value of thickness of cookies and biscuits.

3.2.7.2.5 Spread factor

As per AACC (2000) method the spread factor of control sample was considered to be standard (100 %) and spread factor of other sample of cookies and biscuits was determined in comparison with the standard value. It was expressed in terms of percentage.

3.2.7.3 Chemical characteristics of cookies and biscuits

The proximate analysis of cookies and biscuits was performed for different parameters as described in 3.2.2.

3.2.7.4 Colour analysis of cookies and biscuits

The colour of cookies and biscuits was evaluated as described in 3.2.1.1.

3.2.7.5 Textural analysis of cookies and biscuits

The cookies and biscuits were analyzed for its penetration and cutting force using Texture analyzer (INSTRON). Penetration and cutting force max of the cookies and biscuits were measured by triple beam snap setup (Gaines, 1985). Load cell of 5 kg, 3 mm/s test speed and 5 mm travel distance were used. The sample was placed on two supporting beams a parted by 3 cm distance. The probe, TA-9, was programmed to travel a distance of 5 mm with a speed of 2 mm/sec with a normal cycle. Another beam connected to moving part was brought down to break down the cookies and biscuits. Five samples from each type of cookies and biscuits, fresh and stored were analyzed at 30 days interval (for 90 days). Mean value of three determinations was reported.

3.3 Storage studies of cookies and biscuits

3.3.1 Storage condition

The cookies and biscuits were packed in LDPE and PP bags and stored at ambient condition for 3 months. The samples were drawn at an interval of one month and evaluated for chemical, sensory quality, textural analysis and microbial quality.

3.4 Microbial quality of cookies and biscuits

Microbial count was recorded by using standard plate count (SPC) technique. The nutrient agar was used as growth medium and petridish were incubated at $(37 \pm 5 \text{ }^{\circ}\text{C})$ for 48 h for growth of bacterial colonies. The colonies were counted with magnifying lens. Total count was taken along with pin point colonies (Harriagan and Mccance, 1976).

3.5 Statistical analysis

All experiments were carried out by using Completely Randomized Design (CRD) and Factorial Completely Randomized Design (FCRD). The results obtained in the present investigation were analysed for the statistical significance according to the procedure given by Rangaswamy (2010).

3.6 Economics of kodo millet cookies and biscuits

The cost of production of kodo millet cookies and biscuits was calculated after consideration of the cost of raw material required, labour, processing cost and miscellaneous charges at prevailing rates during experimental period. The cost was worked out by using procedure as described by (Lal *et al.*, 1980).

4. RESULTS AND DISCUSSION

The present investigation entitled “Evaluation of nutritional quality of kodo millet and its exploration in cookies and biscuits” was carried out in Department Food Science and Technology MPKV Rahuri. During present work, physico-chemical properties, proximate composition of kodo millet were investigated. The efforts were also made to develop the technology for utilization of kodo millet as a novel ingredient in formulation of cookies and biscuits. The results obtained during present investigation are presented and discussed with the relevant information available in scientific literature under suitable headings and subheadings.

4.1 Physical properties of kodo millet

The results on physical properties of kodo millet are represented in Table 4.1. The knowledge of physical properties of seeds helps in development of processing technology and is important to minimize the effect of an inadequate use of equipment that may affect the quality of the seed and its oil, and/or lead to high operational costs (Vanesa *et al.*, 2008).

Table 4.1. Physical properties of kodo millet

Sr. No.	Physical parameter	Mean value
1.	Colour	Creamish white
2.	Weight of 1000 seeds(g)	71.20
3.	Bulk density (kg/m ³)	650
4.	True density (kg/m ³)	1172
5.	Porosity (%)	39.68
6.	Angle of repose	21 ⁰
7.	Decorticated seeds recovery (%)	60
8.	Bran recovery (%)	39
9.	Losses during decortication (%)	1
10.	Flour recovery from decorticated seeds (%)	60
11.	Flour recovery from whole seeds (%)	100

*Each value represents the average of three determinations

The seed colour was creamish white which indicated good quality. Bulk density of seeds was found to be 1.84 g/ml. The variations in density of kodo millet may be due to random harvesting of kodo millet at different maturity stages. This factor is important because it determines the capacity of storage, packaging and transport systems (Muragod, 2019).

The weight of 1000 grains was 75.23 ± 2.07 g. The shape of grain and rice were spheroid. The bulk densities of kodo millet grains, rice and husk were 653, 835 and 287 kg/m³, respectively. The true density was to be 1176, 1021 and 1029 kg/m³, respectively. The porosity was 40.0, 18.1 and 74.5 %, respectively. The angles of repose for kodo millet grain, rice and husk were found to be 18°34, 24°30 and 25°12, respectively (Chnadra kumar 2018).

4.2 Chemical composition of kodo millet

The results of chemical composition of kodo millet are represented in Table 4.2. Proximate composition generally represents the nutritional quality of product. It is necessary to analyze the proximate composition of seeds so as to judge its effect on final product as a novel ingredient.

Table 4.2. Chemical composition of kodo millet

Sr. No.	Chemical constituents	Mean value
1.	Moisture (%)	10.90
2.	Crude protein (%)	8.30
3.	Crude fat (%)	1.30
4.	Crude fiber (%)	8.50
5.	Total carbohydrate (%)	65.90
6.	Calcium (mg/100g)	27.00
7.	Iron (mg/100g)	1.70

*Each value represents the average of three determinations

The moisture content of kodo millet was observed to be 10.90 per cent is helpful for good shelf life of grains. The protein, fat and total carbohydrate content of kodo millet were found to be 8.30, 1.30 and 65.90 per cent, respectively. Crude fiber content was

observed to be 8.50 per cent, similar results were obtained by scientist Tosco (2004). These values are also comparable with Gopalan *et al.* (2006).

Kodo millet is a nutritious grain and a good substitute to rice or wheat. The protein, fiber and mineral content are much higher than the major cereals like rice. The kodo millet grain is composed of 8 per cent protein. The major protein fraction in kodo millet is glutelin (Sudharshana *et al.*, 1988).

Kodo millet is an excellent source of fiber (9 %), as opposed to rice (0.2 %), and wheat (1.2 %). Kodo millet contains 66.6 g of carbohydrates and 353 kcal per 100 g of grain, comparable to other millets. It also contains 1.4 per cent fat and 2.6 per cent minerals. The iron content in kodo millet ranges from 25.86 to 39.60 ppm. Among the millets, it has the least amount of phosphorous content (Chandel *et al.*, 2014). The values of chemical content of kodo millet reported in present investigation were more or less similar to the value reported by earlier researcher.

4.3 Chemical composition of maida

The results pertaining to chemical composition of maida are depicted in Table 4.3. Proximate composition generally represents the nutritional quality of product. It is necessary to analyze the proximate composition of seeds so as to judge its effect on final product as a novel ingredient.

Table 4.3. Chemical composition of maida

Sr. No.	Chemical constituents	Mean value
1.	Moisture (%)	13.30
2.	Crude protein (%)	12.10
3.	Crude fat (%)	0.90
4.	Crude fiber (%)	0.30
5.	Total carbohydrate (%)	73.90
6.	Calcium (mg/100g)	23.00
7.	Iron (mg/100g)	2.70

*Each value represents the average of three determinations

The moisture content of maida was observed to be 13.30 per cent is helpful for good shelf life of maida. The protein, fat and carbohydrate content of maida were found

to be 12.10, 0.90 and 73.90 per cent, respectively. Crude fiber content was observed to be 0.30 per cent, which is comparable with findings reported by other scientist Tosco (2004). These values are also comparable with Gopalan *et al.* (2006).

On the basis of observed results it may be concluded that maida contains significantly lower amount of fat (0.9 %) which defines its grain value. Higher protein content of sample justifies the suitability of utilization of maida as a nutritional ingredient in formulation of food products to improve its nutritional characteristics. Findings of present investigation are in conformity with slight differences with the values reported in earlier literature (Ixtaina *et al.*, 2010; Ayerza and Coates, 2001).

The observed differences may be due to environmental factors like climate and location. The levels of calcium, phosphorus and iron in maida are those expected for plant products (Galvao *et al.*, 1976). The high calcium content (23 mg/100g), phosphorous content (121 mg/100g) and iron content (2.7 mg/100g) of maida can be an advantage for its incorporation in food products (Blaustein *et al.*, 2006). Chemical composition especially mineral contents of maida revealed that maida is rich source of minerals. Similar conclusions have been drawn by Bushway *et al.* (1981), Mayela, *et al.* (2007) and Salazar *et al.* (2011).

4.4 Quality of cookies prepared by incorporation of maida and kodo millet flour

Cookies have been categorized as long shelf life product due to their lower moisture content. The product is liked by all segments of population and may serve as a carrier for delivering novel medicinal ingredients. Cookies are generally differentiated from biscuit by their higher fat content (Fustier *et al.*, 2007). The higher fat content of cookies is sometime responsible for deterioration due to oxidation. It was hypothesized that incorporation of maida or wheat flour may prevent the oxidation of cookies thereby enhancing its shelf life (Borneo *et al.*, 2010). The samples with codes CMKF₀, CMKF₁₀, CMKF₂₀, CMKF₃₀, CMKF₄₀, CMKF₅₀, CMKF₆₀, CMKF₇₀, CMKF₈₀, CMKF₉₀ and CMKF₁₀₀ which contained 0, 10, 20, 30, 40, 50, 60, 70, 80, 90 and 100 per cent of maida and kodo millet flour (MKF) were prepared and the results pertaining to different quality aspects of maida and kodo millet incorporated cookies are discussed under subsequent headings.

4.4.1 Sensory evaluation of cookies prepared from maida and kodo millet flour

Organoleptic quality parameters of a product assume pivotal role in anticipating the consumer response to the product (Rey, 2006). It was observed that cookies containing 50 per cent of maida and 50 per cent of kodo millet were acceptable by panel members (Table 4.4). Further it is evident that sample CMKF₅₀ combination was significantly superior in flavor and texture over other treatments carried out during experiment.

Colour and appearance uniformity are vital components of visual quality of fresh as well as processed foods and play a major role in consumer choice (Alistair, 2005). It is observed from Table 4.4 and Plate 4.1 that the sensorial score for colour improved initially with increased level of kodo millet from 8.0 in sample CMKF₁₀ to 8.8 in sample CMKF₅₀. Higher addition of kodo millet resulted in bright and light creamish cookies, which eventually resulted in increment of colour scores.

Table 4.4. Sensory evaluation of cookies fortified with maida and kodo millet flour

Sample code	Sensory attributes*					
	Colour and appearance	Flavour	Texture	Taste	Overall acceptability	Rank
CKMF ₀	8.3	7.8	8.0	7.1	7.80	6
CKMF ₁₀	8.0	7.6	7.5	8.1	7.81	5
CKMF ₂₀	7.6	7.3	7.6	8.0	7.82	4
CKMF ₃₀	8.0	8.0	7.6	8.3	7.90	3
CKMF ₄₀	8.0	8.0	7.8	8.3	8.02	2
CKMF ₅₀	8.8	8.5	8.8	8.8	8.72	1
CKMF ₆₀	6.8	7.3	6.5	7.3	6.90	7
CKMF ₇₀	6.5	7.0	6.5	7.3	6.80	8
CKMF ₈₀	6.5	7.0	6.3	7.3	6.77	9
CKMF ₉₀	6.3	7.0	6.3	7.3	6.72	10
CKMF ₁₀₀	6.5	6.5	6.5	6.5	6.50	11
Mean	7.39	7.45	7.23	7.68	7.44	-
S.E.±	0.03	0.05	0.24	0.05	0.03	-
C.D at 5%	0.11	0.16	0.72	0.16	0.11	-

*Maximum score out of 9. All results are mean value of ten determinations.

Where as,

CKMF₀: (100% maida: 0% kodo millet flour),
 CKMF₂₀: (80% maida: 20 kodo millet flour),
 CKMF₄₀: (60% maida: 40% kodo millet flour),
 CKMF₆₀: (40 % maida: 60% kodo millet flour),
 CKMF₈₀: (20% maida: 80% kodo millet flour),
 CKMF₁₀₀: (0 % maida: 100% kodo millet flour).

CKMF₁₀: (90% maida: 10% kodo millet flour),
 CKMF₃₀: (70% maida: 30% kodo millet flour),
 CKMF₅₀: (50% maida: 50% kodo millet flour),
 CKMF₇₀: (30% maida: 70% kodo millet flour),
 CKMF₉₀: (10% maida: 90% kodo millet flour),

Flavour being a combination of taste, smell and mouth feel, has multifaceted impact on sensory quality of a product (Amerine *et al.*, 1980). Incorporation of kodo millet resulted in increase of flavour characteristics up to the level of 50 per cent. The flavour score is gradually increased upto 8.5 with increase in kodo millet flour addition up to a level of 50 per cent. The panel members commented that increased level of kodo millet up to 50 per cent resulted in increased flavour of cookies.

The texture score is gradually increased with increase upto 8.8 in kodo millet flour addition up to a level of 50 per cent. The panel members commented that increased level of kodo millet up to 50 per cent resulted in increased crispiness of cookies.

With respect to taste characteristics, highest score 8.8 was observed for 50 per cent maida and 50 per cent kodo millet flour cookies while control sample scored 7.1. From consumer point of view, significant change in taste was observed at level of 50 per cent incorporation of kodo millet flour.

Overall acceptability of product is a function of various factors including colour and appearance, flavour, texture and taste. Amongst all samples containing maida 50 per cent and kodo millet flour 50 per cent combination was found to be more acceptable. Statistical analysis showed that sample CMKF₅₀ is the best sample in all sensory attributes.

Singh *et al.* (2000) reported overall acceptability of product like cookies is a function of various factors including colour and appearance, flavour, texture and taste which gives overall acceptance by considering above all attributes.

Sathe *et al.* (1981) reported similar results after replacing maida with soy and groundnut flours at 15 per cent level to prepare protein enriched crackers.

4.5 Quality of biscuits prepared by incorporation of maida and kodo millet flour

Biscuits have been categorized as long shelf life product due to their lower moisture content. The product is liked by all segments of population and may serve as a carrier for delivering novel medicinal ingredients. The samples with codes BMKF₀, BMKF₁₀, BMKF₂₀, BMKF₃₀, BMKF₄₀, BMKF₅₀, BMKF₆₀, BMKF₇₀, BMKF₈₀, BMKF₉₀ and BMKF₁₀₀ which contained 0, 10, 20, 30, 40, 50, 60, 70, 80, 90 and 100 per cent of maida and kodo millet flour (MKF) were prepared and the results pertaining to different quality aspects of maida and kodo millet incorporated biscuits are discussed under

subsequent headings.

4.5.1 Sensory evaluation of fortified biscuits prepared from maida and kodo millet flour

Organoleptic quality parameters of a product assume pivotal role in anticipating the consumer response to the product (Rey, 2006). It was observed that biscuits containing 50 per cent of maida and 50 per cent of kodo millet were acceptable by panel members plate no 4.2 (Table 4.5). Further it is evident that sample BMKF₅₀ was significantly superior in flavour and texture over BMKF₀, BMKF₁₀, BMKF₂₀, BMKF₃₀, BMKF₄₀, BMKF₅₀, BMKF₆₀, BMKF₈₀, BMKF₉₀ and BMKF₁₀₀.

Table 4.5. Sensory evaluation of biscuits fortified with maida and kodo millet flour

Sample code	Sensory attributes*					
	Colour and appearance	Flavour	Texture	Taste	Overall acceptability	Rank
BKMF ₀	8.2	8.6	8.5	8.6	8.47	6
BKMF ₁₀	8.5	8.6	8.5	8.6	8.55	5
BKMF ₂₀	8.6	8.6	8.6	8.6	8.60	4
BKMF ₃₀	8.8	8.8	8.6	8.6	8.70	3
BKMF ₄₀	8.8	8.8	8.8	8.8	8.80	2
BKMF ₅₀	8.8	8.7	8.8	8.8	8.77	1
BKMF ₆₀	8.2	8.5	8.6	8.6	8.47	7
BKMF ₇₀	8.5	8.2	8.3	8.6	8.40	8
BKMF ₈₀	8.3	8.3	8.2	8.6	8.35	9
BKMF ₉₀	8.3	8.2	8.2	8.6	8.32	10
BKMF ₁₀₀	8.0	8.0	8.0	8.0	8.00	11
Mean	8.47	8.50	8.48	8.60	8.51	-
S.E.±	0.05	0.05	0.24	0.05	0.04	-
C.D at 5%	0.16	0.16	0.72	0.16	0.13	-

*Maximum score out of 9. All results are mean value of ten determinations.

Where as,

BKMF₀: (100% maida: 0% kodo millet flour),

BKMF₂₀: (80% maida: 20% kodo millet flour),

BKMF₄₀: (60% maida: 40% kodo millet flour),

BKMF₆₀: (40% maida: 60% kodo millet flour),

BKMF₈₀: (20% maida: 80% kodo millet flour),

BKMF₁₀₀: (0% maida: 100% kodo millet flour).

BKMF₁₀: (90% maida: 10% kodo millet flour),

BKMF₃₀: (70% maida: 30% kodo millet flour),

BKMF₅₀: (50% maida: 50% kodo millet flour),

BKMF₇₀: (30% maida: 70% kodo millet flour),

BKMF₉₀: (10% maida: 90% kodo millet flour),

Colour and appearance uniformity are vital components of visual quality of fresh as well as processed foods and play a major role in consumer choice (Alistair 2005). It is observed from Table 4.5 that the sensorial score for colour improved initially with

increased level of kodo millet from 8.5 in sample BMKF₁₀ to 8.8 in sample BMKF₅₀. Higher addition of kodo millet flour resulted in bright and light brownish biscuits, which eventually resulted in increment of colour scores. More or less same effects were recorded for the appearance properties of kodo millet incorporated biscuits.

Flavour being a combination of taste, smell and mouth feel, has multifaceted impact on sensory quality of a product (Amrine *et al.*, 1980). Incorporation of kodo millet resulted in increase of flavour characteristics up to the level of 50 per cent. The panel members commented that increased level of kodo millet up to 50 per cent resulted in increased flavour of biscuits.

The texture score is gradually increased till 8.8 with increase in kodo millet flour addition up to a level of 50 per cent. With respect to taste characteristics, highest score 9.0 was observed for 50 per cent maida and 50 per cent kodo millet biscuits while control sample scored 8.6. From consumer point of view, significant change in taste was observed at level of 50 per cent incorporation of kodo millet flour.

Overall acceptability of product is a function of various factors including colour and appearance, flavour, texture and taste. Amongst all samples containing maida 50 per cent and kodo millet 50 per cent combination was found to be more acceptable. Statistical analysis showed that sample BMKF₅₀ is best sample in all sensory attributes.

Gupta and Singh (2005) reported overall acceptability of biscuits containing colour and appearance, flavour, texture and taste which gives overall acceptance by considering above all attributes.

Tyagi *et al.* (2006) prepared biscuits containing maida replaced with defatted mustard flour up to 20 per cent and reported acceptable level of defatted mustard flour of 15 per cent to yield biscuits having desirable acceptability.

Selection of best combination for preparation of kodo millet fortified cookies

On the basis of organoleptic properties (colour and appearance, flavour, texture, taste and overall acceptability) the best combination from maida and kodo millet was 50:50. For the storage study these combinations with control (100 % maida) were selected and the cookies prepared from them used for further storage study. During storage study their nutritional composition, organoleptic properties and microbial quality were analysed using standard procedures.

4.6 Physical properties of cookies prepared from best selected combination of maida and kodo millet flour

The results on effect of maida and kodo millet flour on physical properties of cookies are presented in Table 4.6.

Table 4.6. Effect of maida and kodo millet flour on physical parameter of cookies

Treatments	Weight (g)	Diameter (mm)	Thickness (mm)	Spread ratio	Spread factor (%)
CMKF ₀ (T ₀)	10.02	46.47	13.23	3.42	100.00
CMKF ₅₀ (T ₁)	10.07	46.20	13.28	3.30	97.94
Probability	0.00	0.00	0.00	0.00	0.02
T-value	2.776	2.776	2.776	2.776	2.776

*Each value represents the average of three determinations.

Where, CMKF₀: 100 % maida, CMKF₅₀: 50% maida and 50 % kodo millet flour.

Physical and textural properties affect consumer acceptance (Charles, 1992). Physical changes during baking include water evaporation, volume expansion, development of a porous structure, and alterations in dimensions of cookies (Smith, 1972; Turhan and Ozilgen, 1991).

It is seen from the Table 4.6 that weight of cookies remained more or less similar to that of control sample. Maintaining the constant weight of final product is essential to comply with the regulations and delivering uniform product to consumer. The weight of cookies ranged from CMKF₀ 10.02 to CMKF₅₀ 10.07 g. The results indicate that diameter of cookies ranged from CMKF₀ 46.47 to CMKF₅₀ 44.20 mm. Thickness of cookies ranged from CMKF₀ 13.23 to CMKF₅₀ 13.28 mm. It was observed that the spread ratio of cookies ranged from the CMKF₀ 3.42 to CMKF₅₀ 3.34. Spread factor of cookies ranged from CMKF₀ 100 to CMKF₅₀ 97.94 per cent.

The result presented in the Table 4.6 showed that as the proportion of kodo millet flour increases the physical parameters i.e. weight, diameter, thickness, spread ratio, spread factor decreases.

Faridi (1994) reported that decrease in the diameter of cookies usually occurs during baking due to CO₂ produced by leavening agents and water evaporation of cookies expand in both width and height early in baking.

Mc Watters (1978) reported a decrease in spread ratio of cookies when wheat flour was supplemented with non-wheat flours. He opined that use of composite flour

increases dough viscosity and forms aggregates by competing with limited free water available in cookie dough.

Fuhr (1962) reported that flour or any other ingredients, which absorbs water during dough mixing, decreases spread ratio. Physical characteristics of biscuits, such as thickness, diameter and spread ratio, were affected by the increased level of raw and germinated of legumes flour and mushroom flour. The changes in diameter and thickness are reflected in spread ratio which adversely affected the thickness and diameter and thus, spread ratio of the supplemented biscuits (Eiss *et al.*, 2007). The results obtained in the present study are similar to the earlier results presented by previous scientists on similar aspects.

Selection of best combination for preparation of kodo millet fortified biscuits

On the basis of organoleptic properties (colour and appearance, flavour, texture, taste and overall acceptability) the best combination from maida and kodo millet was 50:50. For the storage study these combinations with control (100% maida) were selected and the biscuits prepared from them used for further storage study. During storage study their nutritional composition, organoleptic properties and microbial quality were analysed using standard procedures.

4.7 Physical properties of biscuits prepared from best selected combination of maida and kodo millet flour

The results on effect of maida and kodo millet flour on physical properties of biscuits are presented in Table 4.7. Physical and textural properties affect consumer acceptance. Physical changes during baking include water evaporation, volume expansion, development of a porous structure, and alterations in dimensions of cookies (Smith, 1972; Turhan and Ozilgen, 1991).

It is seen from the Table 4.7 that weight of biscuits remained more or less similar to that of control sample. Maintaining the constant weight of final product is essential to comply with the regulations and delivering uniform product to consumer. The weight of biscuits ranged from BMKF₀ 8.70 to BMKF₅₀ 8.92 g. The results indicate that diameter of biscuits ranged from BMKF₀ 50.30 to BMKF₅₀ 50.15 mm. Thickness of biscuits ranged from BMKF₀ 10.70 to BMKF₅₀ 10.64 mm. It was observed that the spread ratio of biscuits ranged from BMKF₀ 4.60 to BMKF₅₀ 4.43. Spread factor of biscuits ranged from

BMKF₀ 100 to BMKF₅₀ 96.30 per cent.

Table 4.7. Effect of maida and kodo millet flour on physical parameter of biscuits

Treatments	Weight (g)	Diameter (mm)	Thickness (mm)	Spread ratio	Spread factor (%)
BMKF ₀ (T ₀)	8.70	50.30	10.70	4.60	100.00
BMKF ₅₀ (T ₁)	8.90	50.15	10.64	4.40	96.30
Probability	0.01	0.06	0.35	0.40	0.00
T-value	2.776	2.776	2.776	2.776	2.776

*Each value represents the average of three determinations.

Where, BMKF₀: 100 % maida, BMKF₅₀: 50% maida and 50 % kodo millet flour.

Chopra *et al.* (2014) reported that when fortifying biscuits with flour from buckwheat the thickness of the biscuits increases. The increase in thickness may be due to a decrease in the diameter of the biscuits. Changes in diameter and thickness of biscuits are reflected on the volume of biscuits.

Gurung *et al.* (2016) in their studies on biscuits fortified with pumpkin flour, state that fortifying the biscuits leads to an increase in the diameter and thickness of the biscuits, which on the other hand lead to increasing in volume. Good quality cookies or biscuits should have a high spread ratio (Divyashree *et al.*, 2016).

4.8 Chemical composition of cookies prepared from best selected combinations with maida and kodo millet flour

The observation recorded for moisture, protein, fat, crude fiber, carbohydrates, calcium and iron of cookies are presented in Table 4.8.

Table 4.8. Effect of maida and kodo millet flour on chemical composition of cookies

Treatments	Moisture (%)	Crude protein (%)	Crude fat (%)	Crude fiber (%)	Carbohydrate (%)	Calcium (mg/100 g)	Iron (mg/100 g)
CMKF ₀	4.15	12.40	25.91	0.31	73.91	22.99	2.71
CMKF ₅₀	4.11	10.20	25.70	4.40	69.87	25.03	2.20
Probability	0.00	0.00	0.00	0.00	0.00	0.00	0.00
T- value	2.776	2.776	2.776	2.776	2.776	2.776	2.776

*Each value represents the average of three determinations.

Where, CMKF₀: 100% maida, CMKF₅₀: 50% maida and 50% kodo millet flour.

4.8.1 Moisture content

The cookies prepared with CMKF₀ treatment was statistically superior in moisture content (4.15 %) over other treatments CMKF₅₀ (4.11 %). The highest moisture content

(4.15 %) was observed in CMKF₀ treatment with lowest moisture content in CMKF₅₀ treatment (4.11 %). Addition of kodo millet flour in the cookies reduces the moisture content and increase the shelf life of cookies.

4.8.2 Protein content

Protein content of cookies in treatments CMKF₀ was highest (12.40 %) and lowest protein content in treatments CMKF₅₀ (10.20 %). Addition of kodo millet flour in cookies reduces the protein content due to less protein content in kodo millet as compare to maida.

Mirsaeedghazi *et al.* (2008) reported that increase of protein in dough causes greater consistency of dough. The interaction including physical and chemical forces among protein molecules play key role on the rheological properties (Shiau and Yeh, 2001). The increase in protein content is acceptable for better rheological characteristics.

4.8.3 Fat content

The cookies with treatment CMKF₅₀ recorded slightly higher level of fat per cent (26.70 %) where as lowest (25.91 %) fat was recorded in CMKF₀ treatments.

In cookies production, addition of fat imparts tenderness making it more palatable; assist in texture improvements. External added fat during preparation of cookies have plasticizing effects reported by Mulvancey and Cohen (1997).

4.8.4 Crude fiber content

Treatment CMKF₅₀ (4.40 %) was statistical superior in crude fiber content than the treatment CMKF₀ (0.31 %). Crude fiber in treatment CMKF₅₀ was highest (4.40 %) with lowest value in treatment CMKF₀ (0.31 %). The kodo millet is rich in fiber content therefore as the ratio of kodo millet flour increased in the cookies the fiber content also increase. This indicated that kodo fortified cookies are rich source of fiber.

4.8.5 Carbohydrate content

CMKF₀ treatments showed statistical superior in carbohydrates (73.91 %) over CMKF₅₀ treatments. Carbohydrates content of cookies with CMKF₀ treatments was highest (73.91 %) than the treatment CMKF₅₀ (69.87 %). The decrease in carbohydrate content was observed with addition of kodo millet flour might be due to the amount of carbohydrates present in kodo millet flour was at lower level due to refinement of flour.

4.8.6 Mineral

Treatment CMKF₅₀ (25.03 mg) was statistical superior in calcium content than treatment CMKF₀ (22.99 mg). Treatment CMKF₀ (2.71 mg) was statistical superior in iron content than treatment CMKF₅₀ (2.20 mg). Wheat and maida are good source of minerals but the kodo millet flour content less amount of calcium and iron, so there incorporation in cookies lower these minerals content.

4.9 Chemical composition of biscuits prepared from best selected combinations with maida and kodo millet flour

The observation recorded for moisture, protein, fat, crude fiber, carbohydrates, calcium and iron of cookies are presented in Table 4.9.

Table 4.9. Effect of maida and kodo millet flour on chemical composition of biscuits

Treatments	Moisture (%)	Crude protein (%)	Crude fat (%)	Crude fiber (%)	Carbohydrate (%)	Calcium (mg/100 g)	Iron (mg/100 g)
BMKF ₀	4.12	11.80	25.38	0.25	73.65	22.86	2.67
BMKF ₅₀	4.07	10.05	26.62	4.30	69.62	24.88	2.17
Probability	0.00	0.00	0.00	0.00	0.00	0.00	0.00
T-value	2.776	2.776	2.776	2.776	2.776	2.776	2.776

*Each value represents the average of three determinations.

Where, BMKF₀: 100% maida, BMKF₅₀: 50% maida and 50% kodo millet flour

4.9.1 Moisture content

The biscuits prepared with BMKF₀ treatment were statistically superior in moisture content (4.12 %) over other treatments BMKF₅₀ (4.07 %). The highest moisture content (4.12 %) was observed in BMKF₀ treatment with lowest moisture content in BMKF₅₀ treatment (4.07 %). Addition of kodo millet flour in the biscuits reduces the moisture content and increase the shelf life of biscuits.

4.9.2 Protein content

Protein content of biscuits in treatments BMKF₀ was highest (11.80 %) and lowest protein content in treatments BMKF₅₀ (10.05 %). Addition of kodo millet flour in biscuits reduces the protein content due to less protein content in kodo millet as compare to maida.

Mirsaeedghazi *et al.* (2008) reported that increase of protein in dough causes greater consistency of dough. The interaction including physical and chemical forces

among protein molecules play key role on the rheological properties (Shiau and Yeh, 2001). The increase in protein content is acceptable for better rheological characteristics.

4.9.3 Fat content

The biscuits with treatment BMKF₅₀ recorded slightly higher level of fat per cent (26.62 %) where as lowest (25.38 %) fat was recorded in BMKF₀ treatments.

In biscuits production, addition of fat imparts tenderness making it more palatable; assist in texture improvements. External added fat during preparation of cookies have plasticizing effects reported by Mulvaney and Cohen (1997).

4.9.4 Crude fiber content

Treatment BMKF₅₀ (4.30 %) was statistical superior in crude fiber content than the treatment BMKF₀ (0.25 %). Crude fiber in treatment BMKF₅₀ was highest (4.30 %) with lowest value in treatment BMKF₀ (0.25 %). The kodo millet is rich in fiber content therefore as the ratio of kodo millet flour increased in the biscuits the fiber content also increase. This indicated that kodo fortified biscuits are rich source of fiber.

4.9.5 Carbohydrate content

BMKF₀ treatments showed statistical superior in carbohydrates (73.65 %) over BMKF₅₀ treatments. Carbohydrates content of cookies with BMKF₀ treatments was highest (73.65 %) than the treatment BMKF₅₀ (69.62 %). The decrease in carbohydrate content was observed with addition of kodo millet flour might be due to the amount of carbohydrates present in kodo millet flour was at lower level due to refinement of flour.

4.9.6 Mineral

Treatment BMKF₅₀ (24.88 mg) was statistical superior in calcium content than treatment BMKF₀ (22.86 mg). Treatment BMKF₀ (2.57 mg) was statistical superior in iron content than treatment BMKF₅₀ (1.42 mg). Wheat and maida are good source of minerals but the kodo millet flour content less amount of calcium and iron, so there incorporation in biscuits lower these minerals content.

4.10 Storage quality of cookies

The control cookies containing 100 % maida and standardized cookies such as maida and kodo millet flour cookies (CMKF₅₀) contain 50:50 per cent proportions were packed in LDPE and PP and stored at ambient temperature for 3 months (90 days). The

cookies were evaluated for quality evaluation for their sensory and proximate composition at an interval of 30 days.

Effect of packaging material on changes in chemical properties of maida and kodo millet flour cookies during storage period

4.10.1 Changes in moisture

The results on changes in moisture of cookies as influenced by storage are presented in Table 4.10. It was observed that moisture increased for treatment CMKF₀ from 4.13 to 4.18 per cent in LDPE and 4.15 to 4.23 per cent in PP was observed for 90 days of the storage. The sample CMKF₅₀ showed increase in the moisture content 4.07 to 4.15 per cent in LDPE and 4.09 to 4.17 per cent in PP. Significant increase in moisture was observed for treatments and significant increase was found among the packaging materials and interaction. Samples packed in PP showed maximum moisture followed by LDPE packages on the 90th day of storage. This may be due higher water vapour transmission rate (WVTR) and gas transmission rate GTR of PP than LDPE.

Sharoon *et al.* (2014) reported considerable increment the moisture content in all cookies with increasing storage duration. This increase was primarily due to packaging material (polythene bags).

Sujitha and Thirumani (2014) also reported increase in moisture content from 3.6-5.6 per cent of flaxseed cookies during the storage period of 60 days. This increase was primarily due to packaging material (polythene bags). The packaging was not airtight and lack of temperature control resulted in an increase in moisture contents of cookies. Moreover, cookies absorbed moisture from surrounding atmosphere due to hygroscopic behaviour of maida.

An increase in moisture contents of cookie samples during storage has also been reported by Leelavathi and Rao (1993), Rao *et al.* (1995) Pasha *et al.* (2002), Butt *et al.* (2004) and Sharif *et al.* (2005) either due to atmosphere or packaging materials.

The statistical analysis showed that the treatment and packaging material had significant effect on the moisture and the interaction between them was also significant. The rate of increase of moisture was rapid at ambient temperature. This may be due to the difference in temperature and packaging material during storage.

4.10.2 Changes in protein

The results on changes in protein of cookies as influenced by storage are presented in Table 4.10. It was observed that the protein decreased for CMKF₀ treatment from 12.05 to 11.99 per cent in LDPE and from 12.03 to 11.91 per cent in PP was observed for 90 days of storage. The sample CMKF₅₀ showed from 10.15 to 10.04 per cent in LDPE and from 10.13 to 10.02 per cent in PP. Significant decrease in protein was observed for treatments CMKF₀ and CMKF₅₀ in different packaging material. Samples packed in PP showed lower protein than LDPE packages for the 90th day of storage. This may be due higher WVTR and GTR of PP than LDPE.

Sharoon *et al.* (2014) reported decreased in protein content during storage (6.56 to 6.41 %) in biscuits prepared from composite flour and 7.99 to 7.80 per cent during storage period of 45 days. Reduction in protein content of cookies throughout storage might be due to increasing level of moisture which enhances proteolytic activity. Sujitha and Thirumani, (2014) also reported decrease in protein content from 6.51-5.68 % of flaxseed cookies during the storage period of 60 days. The decrease in protein content of cookie samples during storage has also been reported by Pasha *et al.* (2002), Butt *et al.* (2004) and Sharif *et al.* (2005) either due to increasing level of moisture which enhances proteolytic activity.

The statistical analysis showed that the treatment and packaging material had significant effect on the protein and the interaction between them was also significant. There was significant decrease in protein of cookies during the storage for different packaging materials. The rate of decrease of protein was rapid at ambient temperature. This may be due to the difference in temperature and packaging material during storage.

Table 4.10. Effect of packaging material on changes in chemical properties of maida and kodo millet flour cookies during storage period

Parameter	Moisture (%)	Protein (%)	Fat (%)	Crude fiber (%)	Carbohydrate (%)	Calcium (mg/100 g)	Iron (mg/100 g)
Treatment							
T ₀ : CMKF ₀	4.16	11.99	25.81	0.23	73.84	22.91	2.63
T ₁ : CMKF ₅₀	4.11	10.08	26.05	4.30	69.79	24.93	2.12
S.E. \pm	0.00	0.00	0.00	0.00	0.00	0.00	0.00
CD at 5%	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Packaging material							
P ₁ : Low Density Polyethylene	4.12	11.05	25.94	2.27	71.83	23.93	2.39
P ₂ : Polypropylene	4.16	11.01	25.92	2.25	71.80	23.91	2.37
S.E. \pm	0.00	0.00	0.00	0.00	0.00	0.00	0.00
CD at 5%	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Storage period							
C ₁ : 30 days	4.11	11.09	25.98	2.30	71.84	23.95	2.41
C ₂ : 60 days	4.14	11.03	25.93	2.26	71.82	23.92	2.38
C ₃ : 90 days	4.16	10.99	25.89	2.22	71.78	23.88	2.34
S.E. \pm	0.00	0.00	0.00	0.00	0.00	0.00	0.00
CD at 5%	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Interaction							
T ₀ P ₀ C ₁	4.13	12.05	25.87	0.27	73.88	22.96	2.68
T ₀ P ₀ C ₂	4.16	12.02	25.83	0.24	73.85	22.93	2.60
T ₀ P ₀ C ₃	4.18	11.99	25.78	0.20	73.83	22.89	2.61
T ₀ P ₁ C ₁	4.15	12.03	25.85	0.25	73.86	22.93	2.66
T ₀ P ₁ C ₂	4.19	11.96	25.80	0.21	73.84	22.91	2.62
T ₀ P ₁ C ₃	4.23	11.91	25.76	0.19	73.78	22.86	2.59
T ₁ P ₀ C ₁	4.07	10.15	26.11	4.35	69.84	24.98	2.17
T ₁ P ₀ C ₂	4.10	10.09	26.07	4.31	69.81	24.95	2.14
T ₁ P ₀ C ₃	4.15	10.04	26.03	4.26	69.77	24.91	2.11
T ₁ P ₁ C ₁	4.09	10.13	26.09	4.32	69.81	24.95	2.15
T ₁ P ₁ C ₂	4.13	10.05	26.04	4.29	69.78	24.92	2.12
T ₁ P ₁ C ₃	4.17	10.02	26.01	4.24	69.75	24.89	2.08
S.E. \pm	0.00	0.00	0.00	0.00	0.00	0.00	0.00
CD at 5%	0.01	0.01	NS	NS	0.01	NS	NS

All results are mean value of three replication.

Where, CMKF₀: Cookies with 100% maida and 0% kodo millet flour.
 CMKF₅₀: Cookies with 50% maida and 50% kodo millet flour.

4.10.3 Changes in fat

The results on changes in fat content of cookies as influenced by storage are presented in Table 4.10. It was observed that the fat decreased for treatment CMKF₀ from 25.87 to 25.78 per cent in LDPE and from 25.85 to 25.76 per cent in PP was observed for 90 days of storage. The sample CMKF₅₀ showed from 26.11 to 26.03 in LDPE and from 26.09 to 26.01 in PP. Significant decrease in fat was observed for treatments CMKF₀ and CMKF₅₀ in different packaging material. Samples packed in PP showed lower fat followed by LDPE packages on the 90th day of storage. The decline in fat content throughout storage might be due to moisture uptake by cookies from the surrounding air and break down of fats to different compounds.

Waheed *et al.* (2010) reported fat content of linseed cookies at 0 days were 24.18 per cent which decreased thereafter to 24.00 per cent after 45 days of storage.

Sujitha and Thirumaniy (2014) also reported decrease in fat content of flaxseed cookies during the storage period of 60 days.

The decrease in fat contents of cookies samples during storage has also been reported by Pasha *et al.* (2002), Butt *et al.* (2004) and Sharif *et al.* (2005) either due to moisture uptake by cookies from the surroundings air and break down of fats to different compound.

The statistical analysis showed that the treatment and packaging material had significant effect on fat and the interaction between them was non- significant. There was non-significant decrease in the fat of cookies during the storage for different packaging materials. The rate of decrease of fat was rapid at ambient temperature. This may be due to the difference in temperature and packaging material during storage.

4.10.4 Changes in crude fiber

The results on changes in crude fiber of cookies as influenced by storage are presented in Table 4.10. From the results it was observed from that the crude fiber decreased for treatment CMKF₀ from 0.27 to 0.20 per cent in LDPE and from 0.25 to 0.19 per cent in PP was observed for 90 days of storage. The sample CMKF₅₀ showed crude fibre content 4.35 to 4.26 per cent in LDPE and from 4.32 to 4.24 per cent in PP. Significant decrease in crude fiber was observed for treatments CMKF₀ and CMKF₅₀ in different packaging material. Samples packed in PP showed slightly lower crude fiber

followed by LDPE packages on the 90th day of storage. Decrease in crude fiber during storage might be due to increase in moisture content which was engrossed from air as reported by Pasha *et al.* (2002), Butt *et al.* (2004) and Sharif *et al.* (2005).

Waheed *et al.* (2010) reported that crude fibre of linseed cookies at 0 days were 1.26 per cent which decreased thereafter 1.06 per cent after 45 days of storage.

Reduction in crude fiber content of cookies throughout storage might be due to increasing level of moisture which enhances amylase activity.

The statistical analysis showed that the treatment and packaging material had significant effect on the crude fiber and the interaction between them was non-significant. There was non-significant decrease in crude fiber of cookies during the storage for different packaging materials. The rate of decrease of crude fiber was rapid at ambient temperature. This may be due to the difference in temperature and packaging material during storage.

4.10.5 Changes in carbohydrate

The results on changes in carbohydrates of cookies as influenced by storage are presented in Table 4.10. From the results it was observed that the carbohydrates decreased for CMKF₀ from 73.88 to 73.83 per cent LDPE and from 73.86 to 73.78 per cent in PP was observed for 90 days of storage. The sample CMKF₅₀ showed carbohydrate content 69.84 to 69.77 per cent in LDPE and from 69.81 to 69.75 per cent in PP. Significant decrease in carbohydrates was observed for treatments CMKF₀ and CMKF₅₀ in different packaging material. Samples packed in PP showed slight lower carbohydrates followed by LDPE packages on the 90th day of storage.

Sharif *et al.* (2009) reported that carbohydrate content reduced in defatted rice bran cookies from 68.56 to 68.23 per cent after 45 days storage.

Singh and Mohamed (2005) reported that cookies prepared with soy flour blend, with increase the protein content from 6 per cent to 17.5 per cent and reducing total carbohydrates.

Sujitha and Thirumani, (2014) also reported decrease in carbohydrates content from 66.17-69.1 per cent of flaxseed cookies during the storage period of 60 days. The decrease in carbohydrates contents of cookie samples during storage has also been reported by Butt *et al.* (2004) and Sharif *et al.* (2005).

The statistical analysis showed that the treatment and packaging material had significant effect on the carbohydrate and the interaction between them was also significant. There was significant decrease in carbohydrate of cookies during the storage for different packaging materials. The rate of decrease of carbohydrate was rapid at ambient temperature. This may be due to the difference in temperature and packaging material during storage.

4.10.6 Changes in calcium

The results on changes in calcium of cookies as influenced by storage are presented in Table 4.10. It was observed that the calcium decreased for treatment CMKF₀ from 22.96 to 22.89 mg/100g in LDPE and from 22.93 to 22.86 mg/100g in PP was observed for 90 days. The sample CMKF₅₀ showed from 24.98 to 24.91 mg/100g in LDPE and from 24.95 to 22.89 mg/100g in PP. Significant decrease in calcium was observed for treatments CMKF₀ and CMKF₅₀ in different packaging materials. Decrease in calcium during storage might be due to increase in moisture content which was engrossed from air as reported by Pasha *et al.* (2002), Butt *et al.* (2004) and Sharif *et al.* (2005).

Sujitha and Thirumani (2014) also reported decrease in calcium content of flaxseed cookies during the storage period of 60 days.

The statistical analysis showed that the treatment and packaging material had significant effect on the calcium and the interaction between them was non-significant. There was non-significant decrease in calcium of cookies during the storage for different packaging materials. The rate of decrease of calcium was rapid at ambient temperature. This may be due to the difference in temperature and packaging material during storage.

4.10.7 Changes in iron

The results on changes in iron of cookies as influenced by storage are presented in Table 4.10. It was observed from that the iron decreased for treatment CMKF₀ from 2.68 to 2.61 mg/100g in LDPE and 2.66 to 2.59 mg/100g in PP was observed for 90 days. The sample CMKF₅₀ showed from 2.17 to 2.11 mg/100g in LDPE and from 2.15 to 2.08 mg/100g in PP. Significant decrease in iron was observed for treatments CMKF₀ and CMKF₅₀ in different packaging materials.

Decrease in iron during storage might be due to increase in moisture content and as well as oxidation reaction which was engrossed from air as reported by Pasha *et al.* (2002), Butt *et al.* (2004) and Sharif *et al.* (2005).

Sujitha and Thirumani (2014) also reported decrease in iron content of flaxseed cookies during the storage period of 60 days.

The statistical analysis showed that the treatment and packaging material had significant effect on the iron and the interaction between them was non-significant. There was non-significant decrease in iron of cookies during the storage for different packaging materials. The rate of decrease of iron was rapid at ambient temperature. This may be due to the difference in temperature and packaging material during storage.

4.11 Storage quality of biscuits

The control biscuits containing 100 % maida and standardized biscuits such as maida and kodo millet flour biscuits (BMKF₅₀) contain 50:50 per cent proportions were packed in LDPE and PP and stored at ambient temperature for 3 months (90 days). The biscuits were evaluated for quality evaluation for their sensory and proximate composition at an interval of 30 days.

Effect of packaging material on changes in chemical properties of maida and kodo millet flour biscuits during storage period

4.11.1 Changes in moisture

The results on changes in moisture of biscuits as influenced by storage are presented in Table 4.11. It was observed that moisture increased for treatment BMKF₀ from 4.15 to 4.20 per cent in LDPE and 4.17 to 4.25 per cent in PP was observed for 90 days of the storage. The sample BMKF₅₀ showed increase in the moisture content 4.09 to 4.17 per cent in LDPE and 4.11 to 4.19 per cent in PP. Significant increase in moisture was observed for treatments and significant increase was found among the packaging materials and interaction. Samples packed in PP showed maximum moisture followed by LDPE packages on the 90th day of storage. This may be due higher water vapour transmission rate (WVTR) and gas transmission rate GTR of PP than LDPE.

Table 4.11. Effect of packaging material on changes chemical properties of maida and kodo millet flour biscuits during storage period

Parameter	Moisture (%)	Protein (%)	Fat (%)	Crude fiber (%)	Carbohydrate (%)	Calcium (mg/100 g)	Iron (mg/100 g)
Treatment							
T ₀ : BMKF ₀	4.19	11.78	25.31	0.20	73.63	22.81	2.58
T ₁ : BMKF ₅₀	4.13	10.02	25.56	4.27	69.59	24.83	2.08
S.E. \pm	0.00	0.00	0.00	0.00	0.00	0.00	0.00
CD at 5%	0.00	0.01	0.00	0.00	0.00	0.00	0.01
Packaging material							
P ₁ : Low Density Polyethylene	4.15	10.98	25.44	2.25	71.62	23.83	2.34
P ₂ : Polypropylene	4.18	10.82	25.42	2.23	71.60	23.81	2.32
S.E. \pm	0.00	0.00	0.00	0.00	0.00	0.00	0.00
CD at 5%	0.00	0.01	0.00	0.00	0.00	0.00	0.01
Storage period							
C ₁ : 30 days	4.13	10.80	25.48	2.27	71.64	23.85	2.36
C ₂ : 60 days	4.16	10.98	25.43	2.24	71.61	23.82	2.33
C ₃ : 90 days	4.20	10.92	25.39	2.20	71.57	23.78	2.30
S.E. \pm	0.00	0.00	0.00	0.00	0.00	0.00	0.00
CD at 5%	0.00	0.02	0.00	0.00	0.00	0.00	0.01
Interaction							
T ₀ P ₀ C ₁	4.15	12.03	25.37	0.25	73.68	22.86	2.63
T ₀ P ₀ C ₂	4.18	11.94	25.30	0.22	73.65	22.83	2.61
T ₀ P ₀ C ₃	4.20	11.88	25.28	0.18	73.61	22.79	2.56
T ₀ P ₁ C ₁	4.17	11.08	25.35	0.23	73.66	22.83	2.61
T ₀ P ₁ C ₂	4.21	11.94	25.30	0.19	73.62	22.81	2.57
T ₀ P ₁ C ₃	4.25	11.86	25.26	0.17	73.58	22.76	2.54
T ₁ P ₀ C ₁	4.09	10.04	25.61	4.33	69.64	24.88	2.12
T ₁ P ₀ C ₂	4.12	10.04	25.57	4.29	69.61	24.85	2.09
T ₁ P ₀ C ₃	4.17	9.99	25.53	4.24	69.57	24.81	2.06
T ₁ P ₁ C ₁	4.11	10.08	25.59	4.30	69.61	24.85	2.11
T ₁ P ₁ C ₂	4.15	10.00	25.55	4.27	69.58	24.82	2.08
T ₁ P ₁ C ₃	4.19	9.97	25.51	4.22	69.55	24.79	2.05
S.E. \pm	0.00	0.01	0.00	0.00	0.00	0.00	0.00
CD at 5%	NS	0.04	NS	NS	NS	NS	NS

All results are mean value of three replication.

Where, BMKF₀: Biscuits with 100% maida and 0% kodo millet flour.

BMKF₅₀: Biscuits with 50% maida and 50% kodo millet flour.

Sharoon *et al.* (2014) reported considerable increment the moisture content in all cookies with increasing storage duration. This increase was primarily due to packaging material (polythene bags).

The packaging was not airtight and lack of temperature control resulted in an increase in moisture contents of biscuits. Moreover, biscuits absorbed moisture from surrounding atmosphere due to hygroscopic behaviour of maida.

An increase in moisture contents of cookie samples during storage has also been reported by Leelavathi and Rao (1993), Rao *et al.* (1995) Pasha *et al.* (2002), Butt *et al.* (2004) and Sharif *et al.* (2005) either due to atmosphere or packaging materials.

The statistical analysis showed that the treatment and packaging material had significant effect on the moisture and the interaction between them was non-significant. The rate of increase of moisture was rapid at ambient temperature. This may be due to the difference in temperature and packaging material during storage.

4.11.2 Changes in protein

The results on changes in protein of biscuits as influenced by storage are presented in Table 4.11. It was observed that the protein decreased for BMKF₀ treatment from 12.03 to 11.88 per cent in LDPE and from 11.08 to 11.86 per cent in PP was observed for 90 days of storage. The sample BMKF₅₀ showed from 10.04 to 9.99 per cent in LDPE and from 10.08 to 9.97 per cent in PP. Significant decrease in protein was observed for treatments BMKF₀ and BMKF₅₀ in different packaging material. Samples packed in PP showed lower protein than LDPE packages for the 90th day of storage. This may be due higher WVTR and GTR of PP than LDPE.

Protein content decreased from 6.56 per cent to 6.41 per cent during storage in biscuit prepared from composite flour (Elahi, 1997).

Sharoon *et al.* (2014) reported decreased in protein content during storage (6.56 to 6.41 %) in biscuits prepared from composite flour and 7.99 to 7.80 per cent during storage period of 45 days. Reduction in protein content of cookies throughout storage might be due to increasing level of moisture which enhances proteolytic activity.

The statistical analysis showed that the treatment and packaging material had significant effect on the protein and the interaction between them was also significant. There was significant decrease in protein of biscuits during the storage for different packaging materials. The rate of decrease of protein was rapid at ambient temperature. This may be due to the difference in temperature and packaging material during storage.

4.11.3 Changes in fat

The results on changes in fat content of biscuits as influenced by storage are presented in Table 4.11. It was observed that the fat decreased for treatment BMKF₀ from 25.37 to 25.28 per cent in LDPE and from 25.35 to 25.26 per cent in PP was observed for 90 days of storage. The sample BMKF₅₀ showed from 25.61 to 25.53 in LDPE and from 25.59 to 25.51 in PP. Significant decrease in fat was observed for treatments BMKF₀ and BMKF₅₀ in different packaging material. Samples packed in PP showed lower fat followed by LDPE packages on the 90th day of storage. The decline in fat content throughout storage might be due to moisture uptake by cookies from the surrounding air and break down of fats to different compounds.

Tyagi *et al.* (2006) studied nutritional characteristics of defatted mustard flour fortified biscuits. The protein content of mustard flour biscuit increased nearly 2.5 times as a result of mustard flour incorporation coupled with reduction in fat.

Sharoon *et al.* (2014) reported decreased in fat content during storage in biscuits prepared from composite flour during storage period of 45 days.

The statistical analysis showed that the treatment and packaging material had significant effect on fat and the interaction between them was non-significant. There was non-significant decrease in the fat of biscuits during the storage for different packaging materials. The rate of decrease of fat was rapid at ambient temperature. This may be due to the difference in temperature and packaging material during storage.

4.11.4 Changes in crude fiber

The results on changes in crude fiber of biscuits as influenced by storage are presented in Table 4.11. From the results it was observed from that the crude fiber decreased for treatment BMKF₀ from 0.25 to 0.18 per cent in LDPE and from 0.23 to 0.17 per cent in PP was observed for 90 days of storage. The sample BMKF₅₀ showed crude fibre content 4.33 to 4.24 per cent in LDPE and from 4.30 to 4.22 per cent in PP. Significant decrease in crude fiber was observed for treatments BMKF₀ and BMKF₅₀ in different packaging material. Samples packed in PP showed slightly lower crude fiber followed by LDPE packages on the 90th day of storage. Decrease in crude fiber during storage might be due to increase in moisture content which was engrossed from air as reported by Pasha *et al.* (2002), Butt *et al.* (2004) and Sharif *et al.* (2005).

Narender *et al.* (2007) reported that significantly decrease in crude fiber, ash and carbohydrate content in biscuits prepared by incorporation of whey protein concentrate up to 30 per cent.

Sharoon *et al.* (2014) reported decreased in crude fiber content during storage in biscuits prepared from composite flour during storage period of 45 days. Reduction in crude fiber content of cookies throughout storage might be due to increasing level of moisture which enhances amylase activity.

The statistical analysis showed that the treatment and packaging material had significant effect on the crude fiber and the interaction between them was non-significant. There was non-significant decrease in crude fiber of biscuits during the storage for different packaging materials. The rate of decrease of crude fiber was rapid at ambient temperature. This may be due to the difference in temperature and packaging material during storage.

4.11.5 Changes in carbohydrate

The results on changes in carbohydrates of biscuits as influenced by storage are presented in Table 4.11. From the results it was observed that the carbohydrates decreased for BMKF₀ from 73.68 to 73.61 per cent LDPE and from 73.66 to 73.58 per cent in PP was observed for 90 days of storage. The sample BMKF₅₀ showed carbohydrate content 69.64 to 69.57 per cent in LDPE and from 69.61 to 69.55 per cent in PP. Significant decrease in carbohydrates was observed for treatments BMKF₀ and BMKF₅₀ in different packaging material. Samples packed in PP showed slight lower carbohydrates followed by LDPE packages on the 90th day of storage.

Narender *et al.* (2007) reported that significantly decrease in ash content and carbohydrate content in biscuits prepared by incorporation of whey protein concentrate up to 30 per cent.

Sharoon *et al.* (2014) reported decreased in carbohydrates content during storage in biscuits prepared from composite flour during storage period of 45 days.

The statistical analysis showed that the treatment and packaging material had significant effect on the carbohydrate and the interaction between them was non-significant. There was non-significant decrease in carbohydrate of biscuits during the storage for different packaging materials. The rate of decrease of carbohydrate was rapid

at ambient temperature. This may be due to the difference in temperature and packaging material during storage.

4.11.6 Changes in calcium

The results on changes in calcium of biscuits as influenced by storage are presented in Table 4.11. It was observed that the calcium decreased for treatment BMKF₀ from 22.86 to 22.79 mg/100g in LDPE and from 22.83 to 22.76 mg/100g in PP was observed for 90 days. The sample BMKF₅₀ showed from 24.88 to 24.81 mg/100g in LDPE and from 24.85 to 22.79 mg/100g in PP. Significant decrease in calcium was observed for treatments BMKF₀ and BMKF₅₀ in different packaging materials.

Decrease in calcium during storage might be due to increase in moisture content which was engrossed from air as reported by Pasha *et al.* (2002), Butt *et al.* (2004) and Sharif *et al.* (2005).

Sharoon *et al.* (2014) reported decreased in calcium content during storage in biscuits prepared from composite flour during storage period of 45 days.

Sujitha and Thirumani (2014) also reported decrease in calcium content of flaxseed cookies during the storage period of 60 days.

The statistical analysis showed that the treatment and packaging material had significant effect on the calcium and the interaction between them was non-significant. There was non-significant decrease in calcium of biscuits during the storage for different packaging materials. The rate of decrease of calcium was rapid at ambient temperature. This may be due to the difference in temperature and packaging material during storage.

4.11.7 Changes in iron

The results on changes in iron of biscuits as influenced by storage are presented in Table 4.11. It was observed from that the iron decreased for treatment BMKF₀ from 2.63 to 2.56 mg/100g in LDPE and 2.61 to 2.54 mg/100g in PP was observed for 90 days. The sample BMKF₅₀ showed from 2.12 to 2.06 mg/100g in LDPE and from 2.11 to 2.05 mg/100g in PP. Significant decrease in iron was observed for treatments BMKF₀ and BMKF₅₀ in different packaging materials.

Decrease in iron during storage might be due to increase in moisture content and as well as oxidation reaction which was engrossed from air as reported by Pasha *et al.* (2002), Butt *et al.* (2004) and Shariff *et al.* (2005).

Sharoon *et al.* (2014) reported decreased in iron content during storage in biscuits prepared from composite flour during storage period of 45 days.

The statistical analysis showed that the treatment and packaging material had significant effect on the iron and the interaction between them was non-significant. There was non-significant decrease in iron of biscuits during the storage for different packaging materials. The rate of decrease of iron was rapid at ambient temperature. This may be due to the difference in temperature and packaging material during storage.

4.12 Effect of packaging material on changes in sensory quality of maida and kodo millet flour cookies during storage period

4.12.1 Changes in colour and appearance

The results on colour score of cookies as influenced by storage are presented in Table 4.12. The results indicate that score for colour decreased for treatment CMKF₀ from 7.50 to 7.30 in LDPE and from 7.40 to 7.20 in PP was observed for 90 days of storage. The sample CMKF₅₀ showed organoleptic score for colour and appearance 7.50 to 7.20 in LDPE and 7.30 to 7.00 in PP in 90 days of storage.

A decrease in score for colour and appearance from 8.00 to 6.60 cookies during storage similar results were reported by Singh *et al.* (2000) in storage of soy fortified biscuits.

Gupta and Singh (2005) reported change in colour and appearance of biscuits containing quality protein maize during 90 days storage.

The statistical analysis showed that the treatment and packaging material had significant effect on the colour and appearance and the interaction between them was non-significant. There was non-significant decrease in colour and appearance of cookies during the storage for different packaging materials and storage temperature.

4.12.2 Changes in flavour

The results on flavour score of cookies as influenced by storage are presented in Table 4.12. The data indicate that score for flavor for treatment CMKF₀ decreased from 7.50 to 7.30 in LDPE and 7.30 to 7.20 in PP as storage period get increased. For treatment CMKF₅₀ score decreases from 7.50 to 7.20 in LDPE and 7.40 to 7.10 in PP in 90 days of storage.

Sathe *et al.* (1981) reported development of rancid flavour in crackers prepared from soy flour and ground nut flour after 60 days of storage.

Gupta and Singh (2005) reported reduced flavour score of maize fortified biscuits.

The statistical analysis showed that the treatment was non-significant and packaging material had significant effect on flavor and the interaction between them was non-significant. There was non-significant decrease in flavour of cookies during the storage for different packaging materials. The rate of decrease of flavour was rapid at ambient temperature. This may be due to the difference in temperature and packaging material during storage.

4.12.3 Changes in texture

The results on texture score of cookies as influenced by storage are presented in Table 4.12. The results indicate that score for texture decreased for treatment CMKF₀ from 7.00 to 6.90 in LDPE and 7.75 to 7.64 in PP was observed for 90 days of storage. The sample CMKF₅₀ showed organoleptic score for texture property 7.90 to 7.72 in LDPE and 7.90 to 7.87 in PP in 90 days of storage.

Singh *et al.* (2000) reported decreasing score of texture in the soy fortified biscuits storage. Gupta and Singh, (2005) reported changes in texture of biscuits containing quality protein maize, when packed in polyethylene packages and stored at ambient condition.

The statistical analysis showed that the treatment and packaging material had significant effect on the texture and the interaction between them was non-significant. There was non-significant decrease in texture of cookies during the storage for different packaging materials. The rate of decrease of texture was rapid at ambient temperature. This may be due to the difference in temperature and packaging material during storage.

Table 4.12. Effect of packaging material on changes in sensory quality of maida and kodo millet flour cookies during storage period

Parameter	Colour and appearance	Flavour	Texture	Taste	Overall acceptability
Treatment					
T ₀ : CMKF ₀	7.33	7.33	6.88	6.95	7.11
T ₁ : CMKF ₅₀	7.25	7.26	7.06	7.16	7.23
S.E. ±	0.02	0.02	0.02	0.02	0.02
CD at 5%	0.06	NS	0.06	0.06	0.06
Packaging material					
P ₁ : Low Density Polyethylene	7.36	7.36	7.05	7.13	7.23
P ₂ : Polypropylene	7.21	7.23	6.90	6.98	7.11
S.E. ±	0.02	0.02	0.02	0.02	0.02
CD at 5%	0.06	0.06	0.06	0.06	0.06
Storage period					
C ₁ : 30 days	7.42	7.42	7.05	7.25	7.30
C ₂ : 60 days	7.27	7.27	6.95	7.02	7.15
C ₃ : 90 days	7.17	7.20	6.92	6.90	7.06
S.E. ±	0.02	0.02	0.02	0.02	0.02
CD at 5%	0.08	0.08	0.08	0.08	0.07
Interaction					
T ₀ P ₀ C ₁	7.50	7.50	7.00	7.20	7.30
T ₀ P ₀ C ₂	7.40	7.40	7.00	7.00	7.20
T ₀ P ₀ C ₃	7.30	7.30	6.90	6.90	7.10
T ₀ P ₁ C ₁	7.40	7.30	6.90	7.10	7.20
T ₀ P ₁ C ₂	7.20	7.30	6.80	6.80	7.00
T ₀ P ₁ C ₃	7.20	7.20	6.70	6.70	6.90
T ₁ P ₀ C ₁	7.50	7.50	7.20	7.40	7.40
T ₁ P ₀ C ₂	7.30	7.30	7.10	7.20	7.23
T ₁ P ₀ C ₃	7.20	7.20	7.10	7.10	7.15
T ₁ P ₁ C ₁	7.30	7.40	7.10	7.30	7.30
T ₁ P ₁ C ₂	7.20	7.10	6.90	7.10	7.20
T ₁ P ₁ C ₃	7.00	7.10	7.00	6.90	7.10
S.E. ±	0.05	0.05	0.05	0.05	0.05
CD at 5%	NS	NS	NS	NS	NS

Maximum score out of 9.0

All results are mean value of three replication.

Where, CMKF₀: Cookies with 100% maida and 0% kodo millet flour.
 CMPF₅₀: Cookies with 50% maida and 50% kodo millet flour.

4.12.4 Changes in taste

The results on taste score of cookies as influenced by storage are presented in Table 4.12. The results indicate that score for taste for CMKF₀ decreased from 7.20 to 6.90 in LDPE and 7.10 to 6.70 in PP as storage period get increased. For treatment CMKF₅₀ score decreases from 7.40 to 7.10 in LDPE and 7.30 to 6.90 in PP in 90 days of storage.

Awasthi and Yadav (1998) observed similar results during storage of biscuits fortified with 15 per cent soy flour. Singh *et al.* (2000) reported decrease in taste score of biscuits containing 20 per cent defatted soy flour when stored at ambient condition.

Narender *et al.* (2007) also reported change in taste of biscuits incorporated with whey protein concentrate up to 30 per cent and stored for 60 days under ambient condition.

The statistical analysis showed that the treatment and packaging material had significant effect on the taste and the interaction between them was non-significant. There was non-significant decrease in taste of cookies during the storage for different packaging materials. The rate of decrease of taste was rapid at ambient temperature. This may be due to the difference in temperature and packaging material during storage.

4.12.5 Changes in overall acceptability

The results on overall acceptability score of cookies as influenced by storage are presented in Table 4.12. The results indicate that score for overall acceptability for treatment CMKF₀ decreased from 7.30 to 7.10 in LDPE and from 7.20 to 6.90 in PP as storage period get increased. For CMKF₅₀ treatment score decreases from 7.40 to 7.15 in LDPE and 7.30 to 7.10 in PP. The CMKF₅₀ treatment is the best for the overall acceptability.

The statistical analysis showed that the treatment and packaging material had significant effect on overall acceptability content and the interaction between them was non-significant. There was non-significant decrease in overall acceptability of cookies during the storage for different packaging materials. The rate of decrease of overall acceptability was rapid at ambient temperature. This may be due to the difference in temperature and packaging material during storage.

4.13 Effect of packaging material on changes in sensory quality of maida and kodo millet flour biscuits during storage period

4.13.1 Changes in colour and appearance

The results on colour score of biscuits as influenced by storage are presented in Table 4.13. The results indicate that score for colour decreased for treatment BMKF₀ from 8.20 to 7.90 in LDPE and from 8.10 to 8.00 in PP was observed for 90 days of storage. The sample BMKF₅₀ showed organoleptic score for colour and appearance 8.30 to 8.20 in LDPE and 8.20 to 8.00 in PP in 90 days of storage.

A decrease in score for colour and appearance from 8.00 to 6.60 cookies during storage similar results were reported by Singh *et al.* (2000) in storage of soy fortified biscuits.

Gupta and Singh (2005) reported change in colour and appearance of biscuits containing quality protein maize during 90 days storage.

The statistical analysis showed that the treatment and packaging material had significant effect on the colour and appearance and the interaction between them was non-significant. There was non-significant decrease in colour and appearance of biscuits during the storage for different packaging materials and storage temperature.

4.13.2 Changes in flavour

The results on flavour score of biscuits as influenced by storage are presented in Table 4.13. The data indicate that score for flavor for treatment BMKF₀ decreased from 8.40 to 8.10 in LDPE and 8.30 to 8.00 in PP as storage period get increased. For treatment BMKF₅₀ score decreases from 8.50 to 8.10 in LDPE and 8.40 to 7.90 in PP in 90 days of storage.

Sathe *et al.* (1981) reported development of rancid flavour in crackers prepared from soy flour and ground nut flour after 60 days of storage.

Gupta and Singh (2005) reported reduced flavour score of maize fortified biscuits.

The statistical analysis showed that the treatment was non-significant and packaging material had significant effect on flavor and the interaction between them was non-significant. There was non-significant decrease in flavour of biscuits during the storage for different packaging materials. The rate of decrease of flavour was rapid at

ambient temperature. This may be due to the difference in temperature and packaging material during storage.

Table 4.13. Effect of packaging material on changes in sensory quality of maida and kodo millet flour biscuits during storage period

Parameter	Colour and appearance	Flavour	Texture	Taste	Overall acceptability
Treatment					
T ₀ : BMKF ₀	8.00	8.16	7.96	8.13	8.14
T ₁ : BMKF ₅₀	8.18	8.21	8.01	8.26	8.29
S.E. ±	0.02	0.02	0.02	0.02	0.01
CD at 5%	0.06	NS	NS	0.06	0.02
Packaging material					
P ₁ : Low Density Polyethylene	8.15	8.26	8.23	8.26	8.23
P ₂ : Polypropylene	8.03	8.11	7.75	8.13	8.20
S.E. ±	0.02	0.02	0.02	0.02	0.01
CD at 5%	0.06	0.06	0.06	0.06	0.02
Storage period					
C ₁ : 30 days	8.20	8.40	8.32	8.40	8.40
C ₂ : 60 days	8.05	8.15	8.17	8.20	8.20
C ₃ : 90 days	8.02	8.02	7.47	8.00	8.04
S.E. ±	0.02	0.02	0.02	0.02	0.01
CD at 5%	0.08	0.08	0.08	0.08	0.03
Interaction					
T ₀ P ₀ C ₁	8.20	8.40	8.40	8.40	8.35
T ₀ P ₀ C ₂	8.00	8.20	8.20	8.20	8.15
T ₀ P ₀ C ₃	7.90	8.10	8.00	8.00	8.00
T ₀ P ₁ C ₁	8.10	8.30	8.20	8.30	8.33
T ₀ P ₁ C ₂	7.80	8.00	8.10	8.10	8.13
T ₀ P ₁ C ₃	8.00	8.00	6.90	7.80	7.90
T ₁ P ₀ C ₁	8.30	8.50	8.40	8.50	8.48
T ₁ P ₀ C ₂	8.30	8.30	8.30	8.30	8.28
T ₁ P ₀ C ₃	8.20	8.10	8.10	8.20	8.15
T ₁ P ₁ C ₁	8.20	8.40	8.30	8.40	8.47
T ₁ P ₁ C ₂	8.10	8.10	8.10	8.20	8.26
T ₁ P ₁ C ₃	8.00	7.90	7.90	8.00	8.13
S.E. ±	0.05	0.05	0.05	0.05	0.02
CD at 5%	NS	NS	NS	NS	NS

Maximum score out of 9.0

All results are mean value of three replication.

Where, BMKF₀: Biscuits with 100% maida and 0% kodo millet flour.

BMKF₅₀: Biscuits with 50% maida and 50% kodo millet flour.

4.13.3 Changes in texture

The results on texture score of biscuits as influenced by storage are presented in Table 4.13. The results indicate that score for texture decreased for treatment BMKF₀ from 8.40 to 8.00 in LDPE and 8.20 to 6.90 in PP was observed for 90 days of storage. The sample BMKF₅₀ showed organoleptic score for texture property 8.40 to 8.10 in LDPE and 8.30 to 7.90 in PP in 90 days of storage.

Singh *et al.* (2000) reported decreasing score of texture in the soy fortified biscuits storage. Gupta and Singh, (2005) reported changes in texture of biscuits containing quality protein maize, when packed in polyethylene packages and stored at ambient condition.

The statistical analysis showed that the treatment was non-significant and packaging material had significant effect on the texture and the interaction between them was non-significant. There was non-significant decrease in texture of biscuits during the storage for different packaging materials. The rate of decrease of texture was rapid at ambient temperature. This may be due to the difference in temperature and packaging material during storage.

4.13.4 Changes in taste

The results on taste score of biscuits as influenced by storage are presented in Table 4.13. The results indicate that score for taste for BMKF₀ decreased from 8.40 to 8.00 in LDPE and 8.30 to 7.80 in PP as storage period get increased. For treatment BMKF₅₀ score decreases from 8.50 to 8.20 in LDPE and 8.40 to 8.00 in PP in 90 days of storage.

Awasthi and Yadav (1998) observed similar results during storage of biscuits fortified with 15 per cent soy flour. Singh *et al.* (2000) reported decrease in taste score of biscuits containing 20 per cent defatted soy flour when stored at ambient condition.

Narender *et al.* (2007) also reported change in taste of biscuits incorporated with whey protein concentrate up to 30 per cent and stored for 60 days under ambient condition.

The statistical analysis showed that the treatment and packaging material had significant effect on the taste and the interaction between them was non-significant.

There was non-significant decrease in taste of biscuits during the storage for different packaging materials. The rate of decrease of taste was rapid at ambient temperature. This may be due to the difference in temperature and packaging material during storage.

4.13.5 Changes in overall acceptability

The results on overall acceptability score of biscuits as influenced by storage are presented in Table 4.13. The results indicate that score for overall acceptability for treatment BMKF₀ decreased from 8.35 to 8.00 in LDPE and from 8.33 to 7.90 in PP as storage period get increased. For BMKF₅₀ treatment score decreases from 8.48 to 8.15 in LDPE and 8.47 to 8.13 in PP. The BMKF₅₀ treatment is the best for the overall acceptability.

The statistical analysis showed that the treatment and packaging material had significant effect on overall acceptability content and the interaction between them was non-significant. There was non-significant decrease in overall acceptability of biscuits during the storage for different packaging materials. The rate of decrease of overall acceptability was rapid at ambient temperature. This may be due to the difference in temperature and packaging material during storage.

4.14 Microbial quality of cookies prepared from maida and kodo millet flour

The microbial count was taken at initial and 90 days, however no microbial colonies were observed on nutrient agar during initial period. The standard plate count of maida and kodo millet cookies was carried out at the end of storage.

Table 4.14. Changes in microbial count (cfu/g) of maida and kodo millet cookies during storage period

Treatment	Packaging material	Standard plate count (- X10 ⁵ cfu/g)	
		Initial (0 days)	Final (90 days)
CMKF ₀ (T ₀)	LDPE	0	2
CMKF ₅₀ (T ₁)		0	2
CMKF ₀ (T ₀)	PP	0	3
CMKF ₅₀ (T ₁)		0	4

The cookies stored in LDPE packaging material showed the count for CMKF₀ 2×10^5 , for CMKF₅₀ 2×10^5 cfu/g and in PP packaging material showed the count for CMKF₀ 3×10^5 , for CMKF₅₀ 4×10^5 cfu/g. While the standard plate count at initial stage

of storage for CMKF₀ and CMKF₅₀ stored in LDPE and PP was 0×10^5 and 0×10^5 cfu/g respectively.

The results indicated that the standard plate count directly proportional to moisture content in cookies. The acceptability of the product by the panel members after 3 months storage confirms that the minimum changes which might have occurred due to microbes were in safe limits for human consumption. Total viable counts were used as a measure of microbiological quality with respect to the levels of the general microbial contamination. Agu and Ndidiamaka (2014) reported that the growth observed could be due to post processing contamination. Aruna *et al.* (2000) reported that cookies stored at ambient temperature showed 3×10^5 CFU/g bacterial growth. Standard plate count for wheat flour cookies was reported from 6×10^3 to 8×10^3 cfu/g (Khandekar *et al.* 2005). Total plate count of guar gum cookies was 0×10^3 to 4.1×10^3 and yeast and mould count of cookies was 0×10^3 to 2.4×10^3 during 0 to 90 days of storage reported by Kukade, *et al.*, (2017).

4.15 Microbial quality of biscuits prepared from maida and kodo millet flour

The microbial count was taken at initial and 90 days, however no microbial colonies were observed on nutrient agar during initial period. The standard plate count of maida and kodo millet biscuits was carried out at the end of storage.

Table 4.15. Changes in microbial count (cfu/g) of maida and kodo millet biscuits during storage period

Treatment	Packaging material	Standard plate count (- X10 ⁵ cfu/g)	
		Initial (0 days)	Final (90 days)
BMKF ₀ (T ₀)	LDPE	0	3
BMKF ₅₀ (T ₁)		0	2
BMKF ₀ (T ₀)	PP	0	4
BMKF ₅₀ (T ₁)		0	3

The biscuits stored in LDPE packaging material showed the count for BMKF₀ 3×10^5 , for BMKF₅₀ 2×10^5 cfu/g and in PP packaging material showed the count for BMKF₀ 4×10^5 , for BMKF₅₀ 3×10^5 cfu/g. While the standard plate count at initial stage of storage for BMKF₀ and BMKF₅₀ stored in LDPE and PP was 0×10^5 and 0×10^5 cfu/g, respectively.

The results indicated that the standard plate count directly proportional to moisture content in cookies. The acceptability of the product by the panel members after 3 months storage confirms that the minimum changes which might have occurred due to microbes were in safe limits for human consumption. Total viable counts were used as a measure of microbiological quality with respect to the levels of the general microbial contamination. Agu and Ndidiamaka (2014) reported that the growth observed could be due to post processing contamination. Aruna *et al.* (2000) reported that cookies stored at ambient temperature showed 3×10^5 CFU/g bacterial growth. Standard plate count for wheat flour cookies was reported from 6×10^3 to 8×10^3 cfu/g (Khandekar *et al.*, 2005). Total plate count of guar gum cookies was 0×10^3 to 4.1×10^3 and yeast and mould count of cookies was 0×10^3 to 2.4×10^3 during 0 to 90 days of storage reported by Kukade *et al.* (2017).

4.16 Economics of cookies

Economics of cookies prepared from maida and kodo millet flour

Cost of production of maida and kodo millet flour cookies is given in Table 4.16. This cost was worked out on the basis of cost of raw material, chemical used, labor charges at prevailing rates of experimental period. The total cost of production for 1 kg of maida and kodo millet flour cookies was Rs. 122. The cost did not include rent and transport charges, sales commission, local tax, interest on capital, depreciation on equipment, etc.

Table 4.16. Economics of cookies prepared from maida and kodo millet flour

Sr. No	Item	Quantity (g)	Cost (Rs/kg)	Total cost (Rs)
1.	Maida	500	30.00	15.00
2.	Kodo millet	500	110.00	55.00
3.	Sugar	500	38.00	19.00
4.	Fat	500	80.00	40.00
5.	Sodium bicarbonate	5	50.00	00.25
6.	Ammonium bicarbonate	5	45.00	00.22
7.	LDPE			10.00
8.	PP			10.00
	Total			149.47

1.	10 % fluctuation in price	= 14.947
	Total cost (Rs)	= 164.41
2.	40 % overhead charges (Include Labor, fuel, electric charges misc.)	= 59.788
	Total cost (Rs)	= 224.198
3.	Total quantity of cookies prepared	= 2.20 Kg
4.	Cost of production of cookies (Rs/2.20 kg)	= 224.198
5.	Cost of production of cookies (Rs/kg)	=101.908
6.	20 % profit (Rs)	= 20.381
7.	Selling price of cookies (Rs/kg)	= 122.289 (Approximately 122/-)

4.17 Economics of biscuits

Economics of biscuits prepared from maida and kodo millet flour

Cost of production maida and kodo millet flour biscuits is given in Table 4.17. This cost was worked out on the basis of cost of raw material, chemical used, labor charges at prevailing rates of experimental period. The total cost of production for 1 kg of maida and kodo millet biscuits was Rs 129. The cost did not include rent and transport charges, sales commission, local tax, interest on capital, depreciation on equipment, etc.

Table 4.17. Economics of biscuits prepared from maida and kodo millet flour

Sr. No	Item	Quantity (g)	Cost (Rs/kg)	Total cost (Rs)
1.	Maida	500	30.00	15.00
2.	Kodo millet	500	110.00	55.00
3.	Sugar	500	38.00	19.00
4.	Fat	500	80.00	40.00
5.	Sodium bicarbonate	5	50.00	00.25
6.	Ammonium bicarbonate	5	45.00	00.22
7.	LDPE			10.00
8.	PP			10.00
	Total			149.47

1.	10% fluctuation in price	= 14.947
	Total cost (Rs)	= 164.417
2.	70% overhead charges (Include Labor, fuel, electric charges misc.)	= 104.629
	Total cost (Rs)	= 269.046
3.	Total quantity of biscuits prepared	= 2.50 kg
4.	Cost of production of biscuits (Rs/2.50 kg)	= 269.046
5.	Cost of production of biscuits (Rs/kg)	= 107.618
6.	20% profit (Rs)	= 21.523
7.	Selling price of biscuits (Rs/kg)	= 129.141 (Approximately 129/-)

5. SUMMARY AND CONCLUSION

The present investigation on evaluation of nutritional quality of kodo millet and its exploration in cookies and biscuit was conducted in the Department of Food Science and Technology, Mahatma Phule Krishi Vidyapeeth, Rahuri during the year 2018-2019, for the preparation of cookies and biscuits to study the changes in chemical composition, to evaluate sensory properties of cookies and biscuits during storage and to work out the economics of cookies and biscuits making.

Preliminary experiments were conducted using various levels of flour from 0 to 100 per cent maida and kodo millet flour and from that the best levels like 50:50 per cent cookies (maida: kodo millet flour), 50:50 per cent biscuits (maida: kodo millet flour) – combine 200 g flour used, 100 g of sugar, 100 g of fat, 1 g of ammonium bicarbonate and 1 g sodium bicarbonate were used for the preparation of cookies and biscuits. The cookies and biscuits samples were stored at ambient temperature ($32 \pm 2^\circ\text{C}$) for 90 days in LDPE and PP packaging material. The representative samples were drawn periodically at 30 days interval to evaluate changes in sensory and chemical parameters of the cookies and biscuits. The experiments were planned using Factorial Completely Randomized Design (FCRD). The results of this investigation are summarized and concluded below:

Composition of cookies

1. Kodo millet flour contained moisture 10.90 per cent, crude protein 8.30 per cent, crude fat 1.30 per cent, crude fiber 8.50 per cent, total carbohydrate 65.90 per cent, calcium 27.00 mg/100g and iron 1.70 mg/100g.
2. Maida contained moisture 13.3 per cent, crude protein 12.10 per cent, crude fat 0.90 per cent, crude fiber 0.30 per cent, total carbohydrate 73.90 per cent, calcium 23.00 mg/100g and iron 2.70 mg/100g.
3. Based on preliminary trials, the cookies containing flour ratio 50:50 (maida: kodo millet flour), 50 g sugar, 50 g fat, 0.5 g ammonium bicarbonate and 0.5 g sodium bicarbonate was the best over other combinations.

Composition of biscuits

1. Kodo millet flour contained moisture 10.90 per cent, crude protein 8.30 per cent, crude fat 1.30 per cent, crude fiber 8.50 per cent, total carbohydrate 65.90 per cent, calcium 27.00 mg/100g and iron 1.70 mg/100g.
2. Maida contained moisture 13.30 per cent, crude protein 12.10 per cent, crude fat 0.90 per cent, crude fiber 0.30 per cent, total carbohydrate 73.90 per cent, calcium 23.00 mg/100g and iron 2.70 mg/100g.
3. Based on preliminary trials, the biscuits containing flour ratio 50:50 (maida: kodo millet flour), 50 g sugar, 50 g fat, 0.5 g ammonium bicarbonate and 0.5 g sodium bicarbonate was the best over other combinations.

Storage studies of cookies and biscuits

1. Best treatments of cookies and biscuits were kept for three months storage. The moisture content of both cookies and biscuits increased more in PP than LDPE during storage study.
2. Protein, fat, crude fiber, carbohydrate, calcium and iron content of both cookies and biscuits decreased more in PP than LDPE during 90 days storage period.
3. The microbial count of cookies and biscuits was more in PP compared to LDPE at 90 days storage period.
4. The sensory score of all parameters decreased continuously during storage. The overall acceptability score of control cookies (CMKF₀) was decreased from 7.30 to 7.10 in LDPE and from 7.20 to 6.90 in PP. The overall acceptability score of kodo millet incorporated cookies (CMKF₅₀) decreased from 7.40 to 7.15 in LDPE and from 7.30 to 7.10 in PP. The overall acceptability score of control biscuits (BMKF₀) was decreased from 8.35 to 8.00 in LDPE and from 8.33 to 7.90 in PP. The overall acceptability score of kodo millet incorporated biscuits (BMKF₅₀) decreased from 8.48 to 8.15 in LDPE and from 8.47 to 8.13 in PP. However, better retention of colour, taste, flavour and overall acceptability of both cookies and biscuits was observed in LDPE during storage. These results are however based on laboratory studies. Further studies on preparation of kodo millet incorporated cookies and biscuits on pilot scale need to be undertaken for better utilization of maida and kodo millet flour combination for commercial cookies and biscuits making.

Conclusion

The results obtained in the present investigation indicated that better quality cookies and biscuits with maida and kodo millet flour can be prepared using 50 per cent maida, 50 per cent kodo millet flour, 50 g of sugar, 50 g of fat, 1 g of ammonium bicarbonate and 1 g sodium bicarbonate. These treatments also showed superior sensory score in respect of quality over other treatments. Cookies and biscuits stored in LDPE showed better chemical and organoleptic performance. Cookies and biscuits could be stored in good condition beyond 90 days in LDPE at ambient temperature. The cost of production of cookies (CMKF₅₀) was Rs 122/ kg cookies and that of biscuits (BMKF₅₀) was Rs 129/kg at laboratory scale.

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*Original not seen

7. APPENDICES

APPENDIX-I

Performa for organoleptic evaluation of kodo millet cookies and biscuits

Hedonic rating test for evaluating sensory quality of kodo millet cookies and biscuits

(Organoleptic Evaluation Score Card)

HEDONIC RATING TEST

Name of evaluator:

Name of Product: Kodo millet cookies and biscuits Date:

Sample No.	Colour and Appearance	Flavour	Texture	Taste	Over all acceptability
T ₀					
T ₁					
T ₂					
T ₃					
T ₄					
T ₅					
T ₆					
T ₇					
T ₈					
T ₉					
T ₁₀					

Comments (if any): _____

9-Point Hedonic Scale:

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

Signature of Evaluator

APPENDIX II

Ambient temperature during experimental period

Month	Week. No	Date	Temperature (°C)	
			Maximum	Minimum
December 2019	49	03-09	30.30	15.00
	50	10-16	28.10	11.30
	51	17-23	26.50	09.10
	52	24-31	27.80	09.00
January 2020	1	01-07	28.50	10.70
	2	08-14	28.10	12.20
	3	15-21	29.90	12.80
	4	22-28	28.40	9.50
February 2020	5	29-04	30.30	10.70
	6	05-11	31.20	13.90
	7	12-18	30.50	13.50
	8	19-25	33.70	15.00
Average	9	26-04	34.60	17.10

8. VITAE

Miss. Mitkal Komal Tanaji

Candidate for the degree of

MASTER OF SCIENCE (AGRICULTURE)

IN

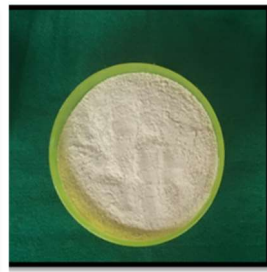
FOOD TECHNOLOGY

2021

Title of thesis		:	Evaluation of nutritional quality of kodo millet and its exploration in cookies and biscuits
Major field		:	Food Science and Technology
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Educational	Bachelor Degree Obtained	:	Received B.Sc. (Agri.) degree from College of Agriculture, Baramati.
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Kodo millet grain



Maida



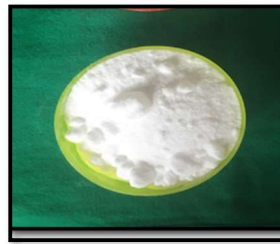
Kodo millet flour



Grinded sugar



Vanaspati ghee



Ammonium bicarbonate



Sodium bicarbonate

Plate : 3.1 Ingredient required for preparation cookies and biscuits (Raw material)



Plate : 3.2 Material used for packaging of cookies and biscuits



CMKF₀



CMKF₅₀

Plate : 4.3 Selected treatment of cookies prepared from maida and kodo millet flour



BMKF₀



BMKF₅₀

Plate : 4.4 Selected treatments of biscuits prepared from maida and kodo millet flour



CMKF₀



CMKF₁₀



CMKF₂₀



CMKF₃₀



CMKF₄₀



CMKF₅₀



CMKF₆₀



CMKF₇₀



CMKF₈₀



CMKF₉₀



CMKF₁₀₀

Plate : 4.1 Sensory evaluation of fortified cookies prepared from maida and kodo millet flour



BMKF₀



BMKF₁₀



BMKF₂₀



BMKF₃₀



BMKF₄₀



BMKF₅₀



BMKF₆₀



BMKF₇₀



BMKF₈₀



BMKF₉₀



BMKF₁₀₀

Plate : 4.2 Sensory evaluation of fortified biscuits prepared from maida and kodo millet flour