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Optimizing Nutritional Biscuits: Enhancing Protein and Mineral Content for Improved Health Benefits

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Malnutrition is an imbalance of energy and nutrients that is shown from physical status of children and adults. According to the WHO, more than 20 million children are severely malnourished under the age of five. In Pakistan, half of all children under the age of five are stunted and one in every ten infants is wasted. Malnutrition can be improved by fortification in food items with natural ingredients. Therefore, the present study was carried out to prepare nutritionally enriched biscuits by using wheat flour, chickpea, lentil and maize flour in different concentrations. The study was performed in the lab of Food Science & Technology, Volka Foods International (R & D lab) and Central Hi-Tech lab of MNS-University of Agriculture, Multan. The raw material was analyzed to determine macro and micro components. Afterwards, biscuits was prepared and analyzed for their nutritional composition like crude fiber, crude fat, crude protein, moisture, ash content and mineral (Fe & Zn) contents. "T5" (60% wheat+20% chickpea+20% maize) was best in term of protein content, "T10" best in term of fat content. Results showed that, hardness of biscuits decreased during storage days (1st, 7th and 14th days) while moisture content of biscuits increased with the time. Treatment "T9" color values was 1* (63.85±0.937), a* (8.53±0.870) and b* (27.30±0.740) showed better result in case of color analysis. According to the result, "T6 and T9" showed better result in case of sensory characteristics like color, appearance, flavor, taste, aroma, texture and overall acceptability of biscuits. Concentration of 60% wheat flour, 20% chickpea flour and 20% lentil flour (T6) showed better results in term of color, flavor and overall acceptability while concentration of 50% wheat flour, 20% chickpea flour, 20% maize flour and 10% lentil flour (T9) was best in case of appearance, aroma, texture and taste in biscuits. Overall results showed that, T9 (Wheat flour 50%, Chickpea flour 20%, maize flour 20% and lentil flour 10%) had the best results compared to the other treatments. Sensory scores and moisture% both decreased significantly during storage study.

Keywords: Malnutrition, composite flour, fortified biscuits, compositional analysis, sensory characteristics.

INTRODUCTION

Malnutrition is a condition when the body is deficient in the nutrients which require for development, maintenance and some tasks. Protein Energy Malnutrition (PEM) a body's requirements for protein, energy or both are not met (Sheetal et al., 2013). Asian countries have severe malnutrition situation than other regions of the world, with 81.7 million stunted (55%), 33.8 million wasted (68.28%) and 18.8 million overweight (47%). Pakistan is unexpectedly one of the countries with the highest malnutrition rates, ranking 77 out of 113; it is also considered to have the worst stunting rates in the Southern Asian Countries (SAC). Stunting, wasting and

underweight are more common in rural communities than in urban ones. In terms of gender-based statistics, males are more likely than females to be stunted, wasted, underweight and overweight. One out of every ten infants waste and more than 20 million children under the age of five suffer from severe malnutrition (Ahmad *et al.*, 2020).

Cereals are Gramineae grass family edible seeds or grains that are grown for their gastronomic attributes (Art of selecting, preparing, serving and enjoying fine food). Wheat, rice, rye, barley, oats, maize, sorghum, triticale and millet are among the grains farmed in many nations. The two most important crops in the world, accounting for more than half of grain production are wheat and rice (Mckevith, 2004). People have

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been eating cereals for a long time and cereal grains are cultivated on large scale. They also contribute important part in the human nutrition as an energy source (Dias-Martins *et al.*, 2018). The FAO estimates that 2,706 million tonnes of cereal will be produced globally in 2019–2020, with a forecast of 2.741 million tonnes for the 2020–2021 season. Above 73% of the world's harvested land-dwelling is used to cultivate cereals which provide more than 60 percent of worldwide food consumption (Charalampopoulos *et al.*, 2002). Cereals contain nutrients such as carbohydrates, minerals, vitamins, proteins and fats. Some cereals also contain a lot of dietary fibre (insoluble and soluble). These grains' physiological properties support the prevention or control of diseases including hypertension, diabetes and heart risk (Baniwal *et al.*, 2021).

Wheat (Triticum aestivum) is the important cash crop for farmers and governments, as well as one of the world's most widely consumed cereals. It belongs to the Triticum genus (Tebben et al., 2018). Wheat is still recognized as the "queen of cereals" due to its versatility in producing leavened foods. Wheat is the cereal that contributes in the majority of a person's daily diet (Serna-Saldivar, 2016). Most of the grain's protein and starch are found in the inner endosperm. It is kept apart from wheat that is being milled into flour. Protein, vitamins and phytic acid are more abundant in the aleurone and outer layers of the endosperm than they are in the inner endosperm (Kumar et al., 2011). Wheat contributes 20% of the food's calories and approximately 55% of the carbohydrate. It has significant amounts of vitamins and minerals, including thiamine and vitamin B, as well as carbohydrates, protein, fat and micronutrients 78.10%, 14.70%, 2.10% and 2.10% respectively. About 72% of the protein in the seed is kept in the endosperm, which accounts for 8–15% of the total protein by grain weight (Kumar et al., 2011). The content of protein in wheat ranges between 8.7 to 19% on a dry matter basis. Cereal proteins can be categorized according to their shape, biological role, solubility or chemical make-up. Albumins and globulins are soluble in water and salt respectively but both are insoluble in water. Gliadins (known as prolamins) are soluble in 70 to 90% ethanol and glutenins are insoluble in neutral aqueous, saline and alcohol solutions. While the previous two are primarily storage proteins, the first two are cytoplasmic or metabolically active proteins. A strong relationship exists between the total amount of storage proteins and the crude protein content of grains (Gutierrez-Alamo et al., 2008).

Maize is main cereal crop that is harvest in many countries of the world and utilized as both human food and animal feed. Except in Antarctica, maize is the 3rd largest crop globally after wheat and rice. Most important for maize to thrive, it requires a specific environment, a certain amount of water and temperatures between 15 and 20 °C for germination (Ranum *et al.*, 2014). Starch, proteins and other compounds like the abundantly present anthocyanin and carotenoids

which are often associated with pigmentation, make up the majority of the ingredients in maize grains. These substances are beneficial for human health (Kuhnen *et al.*, 2022). Maize grain is a tasty and nutritious part of the plant. It includes N-p-coumaryl tryptamine, N-ferrulyl tryptamine, vitamins (C, E, K, B1, B2, B3, B5 and B6), folic acid and selenium. Because it is lacking in the usual human diet, potassium, a primary nutrient present is significant (Kumar and Jhariya, 2013).

Pulses are members of the Leguminosae family which includes species that are eaten by people and domesticated animals. Pulses have a limited number of important amino acids like methionine, tryptophan and cysteine. They nonetheless have a protein content of between 21-25%. As compared to cereals, pulses have nearly double the amount of protein. Albumin and globulin were the two main fractions used to classify pulse proteins. The primary storage proteins in pulse seeds are globulins, which account for 35 to 72% of the total protein in the seeds. Albumins make up the majority of the remaining protein component. Pulses contain a wealth of vitamins and minerals (iron, zinc, calcium and magnesium). Cardiovascular diseases and an imbalance of the biological processes were caused by different minerals being deficient in different countries of the world (Singh, 2017). Due to their exceptional benefits, pulses play a vital role in addressing the triple burden of malnutrition. A wide variety of the population will gain nutritionally from the use of pulses which offer a promising alternative source of functional and nutritious proteins and are becoming more popular among consumers of cereals (Benali et al., 2021).

Chickpea (Cicer arietinum L.) is most important legume with a lot of health and nutritional benefits. It is richest source of different phytochemicals like natural antioxidants such as flavonoid or phenolic compounds that reduce inflammation and oxidative stress associated with many chronic diseases and a rich source of non-digestible carbohydrates (Benali et al., 2021). Chickpeas is a very healthy and cheap source of protein with a protein content around 24% that varies from 15% to 30% depending on various environmental factors. Chickpeas also have 60-65% carbs, 6%S fat and are vital source of B vitamins and minerals. Desi and Kabuli are the types which have several applications (Merga and Haji, 2019). The protein content of chickpeas is thought to be superior to that of other pulses, and they are a richest source of both protein and carbohydrates. Food based solutions try to enhance the quality of food and nutrition by improving the convenience and intake of variety of foods they targeted various nutritional deficits at the same time. It may be used to create nutritious value-added goods which can be used for low-income people as nutritious food in developing nations and patients suffering from lifestyle illnesses (Hirdyani, 2014).

Lentil (Lens culinaris Medikus subsp. culinaris) was cultivated in many countries around the world and has



become an essential food crop in the agriculture and food sector. Its seed is a richest source of protein, minerals contents and vitamins for human nutrition. Its ingestion with wheat or rice offers a balance of essential amino acids for human nutrition because of its maximum content of lysine and tryptophan (Sarker and Erskine, 2006). Lentils are exceptional source of protein, carbs, minerals and vitamins contents and play an important part in human nutrition. Compared to most other grain legumes, lentils cook more quickly and are easier to decorate because of their tiny size and flat form. This might explain why worldwide per capita consumption has increased over the previous 50 years (Sharpe et al., 2013). A cup of lentils can provide 1.9 to 3.3 mg of zinc, 25 to 401 mg of selenium and 4.3 to 5.3 mg of iron (Johnson et al., 2015).

MATERIALS AND METHODS

This research was conducted in the lab of department of Food Science and Technology and Central Hi-Tech lab, MNS-University of the Agriculture, Multan and in the laboratory of Volka Food International Multan. The present study was carried out to prepare nutritionally enriched biscuits by using wheat flour, chickpea, lentil and maize flour in different concentrations. Wheat, chickpea, maize and lentil grains will be taken from local market of Multan.

Milling Process: First of all grains (wheat, chickpea, maize and lentil) were cleaned and all the undesired parts (stones, dust, husk, stones, insect damage and broken grains) were removed. The cleaned grains were milled by using the micro mill (Culatti Type MFC) to form whole meal flour (Cappelli *et al.*, 2020).

Preparation of biscuits: Recipe of El-Gohery (2021) was used with minor modifications. In a bowl, biscuit fat was measured out and heated until it melted. For creaming, ground sugar was added and constantly mixed. Smaller amounts of the flour mixture were gradually added to the cream, which was then well combined. The dry ingredients (Composite flour, skim milk powder, egg powder, and baking powder) were combined, sieved and added into the mixture. Salt was measured out and added to the mixture. A small amount of water was sprinkled to create the soft dough. After the dough was smoothed out, biscuits were cut with a biscuit cutter into small, round shapes. For consistent baking, biscuits were left in the oven for 20 to 25 minutes at 140-160°C. The freshly baked biscuits were cooled at room temperature and were packed in airtight packets for further use.

Proximate Analysis: Evaluation of proximate composition, moisture analyzer was used to determine the amount of moisture content, while dry ashing was used to determine the amount of ash. The Soxhlet method was used to determine the fat content and Kjeldahl method was used to examine protein analysis. The percentage of carbohydrates (NFE %) was

determined by subtracting the total percentage of protein, fat, ash and moisture from 100% (AACC, 2000).

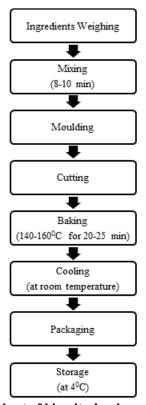


Figure 1. Flow chart of biscuits development.

Table 1. Treatment plan for biscuits development.

Treat.	Wheat	Chickpea	Maize	Lentil	Quinoa
	flour	flour	flour	flour	flour
	(%)	(%)	(%)	(%)	(%)
T_1	50	25	-	-	25
T_2	60	20	-	-	20
T_3	60	20	20	-	-
T_4	60	20	20	-	-
T_5	60	20	20	-	-
T_6	60	20	-	20	-
T_7	70	20	-	10	-
T_8	50	25	-	25	-
T ₉	50	20	20	10	-
T ₁₀	40	25	25	10	-

Mineral Analysis: The mineral profile of biscuit was measured using an atomic absorption spectrophotometer according to the AOAC (2006). Wet digestion of biscuit samples was carried out on a hot plate for two hours using a 7:3 combination of HNO₃:HCLO₄. To make the sample volume 100 ml, distilled water was used to dilute the wet digested samples. An atomic absorption spectrophotometer



was used to quantify the Fe and Zn contents in the digested samples (AOAC, 2006).

Color Analysis: The color values of biscuits were measured in triplicates using a Hunter's color measurement (USA Virginia Hunter Lab Colorimeter) analyzer. The Chroma meter produce three color parameters: a*, b* and l* which demonstrates the darkness and brightness of the samples, a* values measure redness when positive and b* values measure yellowness when positive (Wani et al., 2015).

Sensory Assessment: The trained panelists were asked to evaluate the biscuits. Biscuits' flavour, texture, and general acceptability were evaluated. The scores were provided on a scale from 1 to 9, with 9 being the level of likeliness and 1 representing the level of dislike (Lawless and Heymann, 2010).

Statistical Analysis: For statistical analysis, each sample was examined three times. The Tuckey test (beyond ANOVA) and one way ANOVA under CRD were used to examine differences between treatments according to guidelines proposed by Steel and Torrie (1980). The ANOVA and Tuckey tests were performed using Statistics 8.1 software.

RESULTS AND DISCUSSIONS

The current study was conducted in the Department of Food Science and Technology (FST) at MNS-University of Agriculture, Multan (MNS-UAM) and Volka Foods International. The planned research work was performed in Nutrient analytical lab. In current study, biscuits were analyzed for the proximate composition and other analysis. The obtained results in the research work are discussed below. Proximate Analysis of Biscuits: The proximate composition of biscuits made from flours blends are shown in Table 1. The moisture content of samples was not significantly different from each other (p>0.05). The moisture content of biscuits ranged from 3.18% to 5.37% among the treatments. The results are resembled with the finding of Van Toan and Anh (2018) who reported 4.89% to 5.96% moisture content in biscuits. The protein contents was not significantly different (P > 0.05). The protein content of biscuits ranged from 6.75% to 9.34% among the treatments. Protein content of biscuits are found similar to the result of Pasqualone et al. (2020) who reported 5.06% to 10.66% of protein content in biscuits. Ash content showed highly significant difference. The ash content of biscuits ranged from 3.79% to 9.45% among the treatments. Fibre content is varies non-significant between different treatments. Mean values regarding the Fat content results ranged from 13.68% to 37.74% among the treatments. Results for fiber content of biscuits found in the current study are similar to the result of Van Toan and Anh (2018), who reported 0.33% to 0.91% of fibre content in biscuits. NFE content ranged from 40.96% to 65.21% among the treatments.

Table 2. Mean values of proximate analysis.

Treatments	Protein %	Ash%	Fat%	Fiber%	Moisture%	NFE%	Iron (mg/100g)	Zinc (mg/100g
T1	7.60±0.71	3.79±0.83	29.04±0.99	0.52±0.16	3.45±0.59	55.23±0.50	21.4±0.05	0.13±0.04
T2	7.30 ± 0.82	4.08 ± 0.22	29.41±0.95	0.83 ± 0.15	3.18 ± 0.50	54.15 ± 0.34	19.6 ± 0.08	0.37 ± 0.04
T3	7.38 ± 0.82	3.87±0.81	26.35±0.76	0.59 ± 0.10	4.26±0.62	57.26±0.52	10.8±0.09	0.11±0.17
T4	6.75±0.93	7.64 ± 0.70	23.73±0.70	0.48 ± 0.25	4.62±0.85	56.61±0.56	8.3±0.04	0.10 ± 0.08
T5	9.34±0.95	7.11±0.96	28.65±0.84	0.43±0.23	5.37±0.71	49.03±0.52	14.6±0.04	0.07±0.05
T6	8.50±0.99	7.81±0.66	28.04±0.45	0.57±0.17	4.06±0.96	48.93±0.76	8.5±0.04	0.04 ± 0.05
T7	8.58±0.97	8.23±0.68	25.84±0.79	0.51±0.05	3.33±0.80	53.46±0.57	16.5±0.20	0.02±0.05
T8	9.14±0.99	9.45±0.53	25.12±0.57	0.44 ± 0.07	3.45±0.63	51.49±0.91	12.7±0.06	0.05 ± 0.02
Т9	7.44±0.95	8.86±0.95	13.68±0.72	0.53 ± 0.04	3.59±0.51	65.21±0.31	23.0±0.17	0.08 ± 0.14
T10	7.48 ± 0.96	7.49±0.89	37.74±0.71	0.49 ± 0.08	4.85±0.96	40.96±0.29	12.6±0.04	0.05±0.20

Table 3. Mean values of texture and color analysis.

Treatments	Hardness	l*	a*	b*
T1	19.43±0.122	42.42±0.869	5.34 ± 0.897	15.20±0.934
T2	18.48 ± 0.537	58.75 ± 0.826	7.37 ± 0.557	23.38±0.766
T3	18.18 ± 0.282	66.68 ± 0.872	7.58 ± 0.666	26.74±0.905
T4	16.89 ± 0.420	65.43 ± 0.938	7.83 ± 0.938	26.33±0.950
T5	15.22 ± 0.525	59.68±0.957	8.56 ± 0.712	27.32±0.639
T6	16.33±0.592	60.37±0.756	8.43 ± 0.643	22.29 ± 0.842
T7	20.77±1.642	58.54 ± 0.528	9.28 ± 0.767	26.05±0.583
T8	17.77±0.555	62.17±0.651	7.42 ± 0.736	25.26±0.799
Т9	14.60 ± 1.148	63.85±0.937	8.53 ± 0.870	27.30±0.740
T10	16.74±0.680	64.66±0.978	6.69 ± 0.623	30.44 ± 0.956



	Table 4.	Mean v	values	of se	ensorv	evaluatio	n.
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Treatments	Color	Appearance	Texture	Aroma	Flavor	Taste	Overall
							Acceptability
T1	4.73±0.703	5.46±0.516	5.67 ± 0.488	5.80±0.561	4.60±0.507	4.40 ± 0.507	4.67±0.617
T2	4.27 ± 0.458	4.47±0.516	4.47 ± 0.516	4.40 ± 0.507	4.53±0.516	4.40 ± 0.507	4.20 ± 0.414
T3	6.60 ± 0.507	6.27 ± 0.458	6.27 ± 0.458	5.20 ± 0.414	5.60±0.507	5.47±0.516	5.80 ± 0.414
T4	6.73±0.458	6.80±0.414	6.40±0.507	5.87±0.352	6.60±0.632	6.33±0.488	6.60±0.507
T5	6.87±0.352	6.67±0.488	6.73±0.458	6.13±0.516	5.80±0.414	5.93±0.594	6.87±0.352
T6	7.67±0.488	7.20 ± 0.414	7.27±0.458	7.40±0.508	7.47±0.516	7.27±0.458	8.07±0.594
T7	4.80±0.414	4.33±0.488	4.73±0.458	5.00±0.378	4.73±0.458	4.40±0.507	5.00±0.535
T8	5.87±0.743	7.00 ± 0.926	5.73±0.458	6.27±0.458	6.40±0.508	5.87±0.352	6.13±0.352
T9	7.67±0.488	7.27 ± 0.458	7.73±0.458	7.47±0.516	6.93±0.458	7.87±0.352	7.87 ± 0.352
T10	6.93±0.594	7.73±0.458	5.80±0.704	6.87±0.352	6.87±0.352	7.13±0.352	7.00 ± 0.000

NFE percentage of biscuits are found resembled to the findings of Farzana and Mohajan (2015) who reported 56.38% to 65.62% of NFE content of biscuits.

Mineral Analysis: Iron content of biscuits is high as compared with the study of Hemalatha et al. (2007), who reported 6.64 to 7.83 mg/100g in biscuits sample. Zinc content of biscuits are similar with the study of Doner and Ege (2004), who reported 0.58 to 2.50 mg/100g in biscuits sample. Color Analysis: Mean values of color of biscuits are given in Table 3. Parameter 1* results were noticed in the range of 42.42±0.869 to 66.68±0.872 and parameter a* results were observed in the range of 5.34±0.897 to 9.28±0.767. Maximum reading was 30.44±0.956% and minimum reading was 15.20±0.934 in parameter b*. Recent results of color analysis of biscuits are the resemblance with the findings of (Kumar et al., 2015), who reported the 31.64 to 35.39 b* value, 10.66 to 13.53 a* value and 55.00 to 67.95 L* parameter of color of crust in biscuits, respectively. When

Texture Analysis: The current findings regarding texture of biscuits are similar with the finding of Tyagi *et al.* (2007), Laguna *et al.* (2012) and Kumar *et al.* (2015), who reported 4.46N to 19.00N, 2.7N to 42.60N and 13.25N to 28.68N respectively of texture of biscuits.

Sensory Analysis: This parameter describes the success or acceptance of a food product among the client or consumer. The findings of treatments indicates that the highest value was 8.07 ± 0.516 in T_6 , while 4.20 ± 0.414 was lowest value in T_2 . Overall acceptability of biscuits are similar with the finding of Karki (2016) and Chandra *et al.* (2015) who reported 5.83 to 7.06 and 8.14 to 8.60 respectively of overall acceptability of biscuits sample.

Conclusion: Malnutrition remains a serious public health concern around the world, particularly in poor nations, due to insufficient food quantity and quality. Bakery products can play a crucial role in overcoming malnutrition in children by providing essential nutrients, improving dietary diversity, and enhancing food accessibility. Compared to other snack foods, biscuits have a larger consumer base, a reasonably long shelf

life and decent flavor. Protein and micronutrients (Fe and Zn) are necessary substances that people need in various amounts throughout their lives to coordinate a variety of physiological processes for maintaining health. According to the compositional analysis of nutritionally enriched biscuits, it can be concluded that T_5 have higher protein content, T_{10} have higher fat content, T_6 have higher ash content, T_2 have higher fiber content and T_9 have higher NFE content. According to the result, T_9 showed better results in case of color of biscuits. According to the result, T_6 and T_9 showed better results in case of color, texture, flavor, appearance, aroma, taste and overall acceptability of biscuits among all samples.

Authors contributions statement: Nasir Shabbir and Shabbir Ahmad designed, completed the experiments; Muhammad Usman, Sadaf Yaqoob and Shahrina Shakir prepared the draft; Dr. Sibt-e-Abbas, Umar Farooq reviewed and finalized the draft.

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